EUROPEAN HEMATOLOGY ASSOCIATION

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Thalassaemia International Federation (TIF)

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ARTICLE

National prevention strategies for the control of haemoglobin disorders. A special reference to the countries of the Middle East

Michael Angastiniotis, Androulla Eleftheriou

Thalassaemia International Federation, Nicosia, Cyprus

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ABSTRACT

This is a review of the practices that have been applied for the prevention of haemoglobin disorders, mainly of thalassaemia, in several countries. It includes the motives for embarking on a programme of prevention and the need for national policies and central control to assure quality and best practices. The current situation in the countries of the Eastern Mediterranean Region of the WHO is referred to, with the conclusion that improvements are needed in many countries of the region and that collaborations and networking between countries and experts will help to promote such services.

Introduction

Haemoglobin disorders are a group of hereditary anaemias, which without optimum care, are lethal often in early childhood. In developing economies optimum care is often not provided and the result is premature death of patients, in their teens or early adulthood, who have lived the life of a chronic invalid. Such a situation is an additional burden to the health economy, since all resources offered to the patients are lost before the patient can achieve any quality of life and be able to work as an independent member of society. Optimum care on the other hand has been shown to allow for a long and productive life with patients being able to integrate and contribute to society. While developing patient care services, a programme of prevention is also essential since increasing numbers of affected births will soon overwhelm the resources required for optimum care.

The importance of prevention has been recognized as early as the 1950s, when Silvestroni and Bianco, in Italy, recommended to the High Commission for Hygiene and Health in 1955 the provision of free medical care for patients and the establishment of large scale screening and preventive counselling programs [1] and to achieve this a centre for the study of 'microcytaemia' was established. Adopting such a program in an effective way was not initially possible since warning of the consequences of carrier marriages in this autosomal recessive condition through counselling, was the only means of prevention. One of the first attempts at national scale prevention was adopted by Cyprus from 1972 [2, 3]. The eventual success of this and other national programs implemented in the Mediterranean countries, have served as an inspiration for other high prevalence countries, to formulate their own programmes.

The World Health Organization (WHO), in its resolution EB118.R1 of May 2006, urges member states to *"design, implement, and reinforce in a systematic and effective manner, comprehensive national, integrated programs for prevention and management of thalassaemia, including information and screening…Such programs being tailored to specificsocioeconomic and cultural contexts aimed at reducing incidence, morbidity and mortality"*. In a previous document WHO issued guidelines on prevention [4].

It is in the implementation of such programs that many member states have either stumbled on difficulties, or have not provided the necessary planning. On a worldwide scale few countries have actually successfully implemented the WHO resolutions.

Genetic prevention touches various sensitive issues such as marriage practices and choices in reproduction, and so it is important to have a clear understanding of the reasons for embarking upon such a programme as well as the need for a planned and regulated approach to prevention. Families have to face the death of affected children as well as the burden of chronic disease for which often they have little or no support. They experience an unbearable psychosocial burden,

Correspondence:
michael.angastiniotis@thalassaemia.org.cy

including very often social isolation. Added to all this is the financial burden which is unbearable if the support from the health services is not fully comprehensive.

Prevention is a cost-effective alternative to lifelong treatment

Cost-effectiveness has been demonstrated in various locations, mainly by comparing the cost of patient care with the cost a comprehensive prevention program. In Cyprus the cost of prevention over treatment was estimated at 0.15 *i.e.* the cost of prevention for one year was equal to the cost of treating patients for 8 weeks in 1984 [5]. This degree of cost effectiveness has increased considerably over the years with increasing survival of patients and the introduction of other modalities of treatment and monitoring. Over the years several studies have agreed that prevention is cost effective [6,7,8,9]. Despite these reports few governments have adopted policies for prevention, leaving to individual families the cost of both patient care and prevention, which results in the most educated and the most affluent of the community being able to benefit.

The prevention programme

A comprehensive prevention programme consists of the following elements [4,5]:

- Public **education and awareness**: the public must be aware of the disease and why it is asked to cooperate in a prevention programme.
- **Screening** the population to identify carriers.
- Providing carriers and at risk couples with responsible **genetic counseling**.
- The provision of services for **prenatal diagnosis** or pre-implantation diagnosis.

Such comprehensive prevention services are a public health concern, and are successful if they are part of a national plan with central coordination, budgetary support and with ethical control of practices. Programmes, should be centrally monitored and outcomes recorded.

In the Middle East several countries have now implemented prevention services and have all experienced varying degrees of reduction of affected births.

Observations from ME prevention programmes

The characteristic of most prevention programmes in the ME is that they have focused on a policy of obligatory premarital screening aiming at marriage cancellation. This is because of cultural and religious considerations relating to marriage customs as well as to attitudes concerning termination of pregnancy. With these considerations many ME countries have adopted a policy of mandatory premarital screening which aims to avoid the choice of prenatal diagnosis and termination of pregnancy. Whether such a policy can be effective in prevention, since it presupposes an acceptance of marriage cancellation, is a question that can only be answered by following the outcomes over time [10, 11]. The results so far are that no programme has achieved a 65% at-risk marriage cancellation [11].

Awareness

One of the most significant weaknesses in these programmes is the poor awareness of both public and professionals. Despite high prevalence in some countries, genetic diseases and birth defects are not given priority by health authorities and even doctors. This is partly because health policy makers have not been aware of the immense global toll of birth defects, including the thalassaemias, because of high infant mortality from infections, masking the lethal diseases of infancy [12]. There is also a widespread misconception that little can be done for these disorders and that expensive, high – technology is needed for their effective control and management [13]. In a few countries only, have good epidemiological data been recorded and few countries maintain registries. Because of this, the real burden of disease has not been quantified. Other priorities for funding, such as the infectious diseases, are thought to be more urgent. Recognizing that lack of information and understanding is a major cause of poor services, the World Health Organization passed a resolution urging member states to raise awareness and set priorities for Birth defects [14].

Raising awareness among the public is also a vital aspect since preventing a genetic disorder, like thalassaemia, requires decisions from the community members concerning screening, marriage choices and reproductive choices, all of which are personal and sensitive issues [15]. Without preparing the public, screening programmes, especially premarital screening may not yield the results that are expected from the cost-effective public health point of view [16, 17]. Awareness and education becomes even more difficult in multi-ethnic, multi-cultural and multi-lingual societies, especially where marriage practices differ among the various groups, consanguineous marriage being a striking example [18, 19].

The processes for promoting knowledge, which should involve primary care services [20] become even more difficult when barriers such as language and cultural differences require special preparation of professionals and perhaps employment of additional staff derived from the communities.

Screening

Screening to identify thalassaemia carriers is the next step following the public education programme. The availability of competent services should be ensured. Screening laboratories should be specialized and follow standard procedures with recognized algorithms for quality assurance [21]. It is important to have a reference laboratory, which has the capacity to elucidate difficult diagnosis cases. The various techniques for screening have been described and reviewed several times but the recent short guide for laboratories published by the Thalassaemia International Federation (TIF) may be a useful tool to guide laboratories and to adopting strategies [22].

Developing a strategy for targeted screening is also important. Most countries of the Middle East have adopted premarital screening [11]. This in some cultures carries the risk of stigmatization and reducing the chances of being accepted as partner [23]. Such prejudice can only be reduced through increased awareness and knowledge within the community. Other alternatives include family screening which would yield many carriers, especially in communities where consanguineous marriage is customary [24]. In communities where the carrier rate is low and ethnic groups are difficult to reach, screening in early pregnancy has been the policy adopted, despite the fact that this limits the couples' choices [25]. Screening school pupils and other groups (for example army recruits or blood donors), which are easy to reach, is a strategy that is used but this also raises questions of parental and individual consent and whether the individual will understand the implications of being a carrier. Single people may not remember or are reluctant to disclose their carrier status although in one study in Italy, the time lapse of 15 years between school screening and eventual pregnancy, the information was "well conserved" [26].

Another important question is whether screening

should be voluntary or mandated [10,11,15]. In many countries of the Middle East, the policy of mandatory premarital screening has ben adopted. The reasons for adopting this approach are first, that awareness of the disease and of the availability of carrier detection is very limited, so that making a premarital certification mandatory will inform couples, at a time when choices are more than if already married or already pregnant. Another factor is the fact that prenatal diagnosis and termination of pregnancy is not acceptable culturally and by religion in many cultures, so that early warning is expected to increase the choices concerning marriage and reproduction. It should be noted, that although screening is mandatory, the couple is free to make choices after counselling in all these programmes, although family pressures cannot be excluded [27]. Mandatory screening is not acceptable in the West but the level of education and knowledge, as well as culture must be considered [28, 29].

Genetic counselling

This is one of the most difficult aspects of prevention, particularly since trained counsellors are not clearly defined, or appointed, in most countries. It is assumed that counselling is left to the doctor, who is involved with the thalassaemia patients or the scientist involved in thalassaemia screening. Genetic counselling requires detailed knowledge of the disease in question and the solutions and choices, which can be offered for each at-risk couple. However counselling also requires communication skills as well as the adherence to principles such as not directing the couple towards solutions that the counselor favors and other ethical issues [30, 31]. One striking example is the issue of consanguinity, which many counselors in the Middle East actively discourage, in the belief that it increases the chance of bearing a thalassaemic child [32]. Added to these issues are others, such as language barriers and religious attitudes. Counseling therefore cannot be undertaken without training if couples are going to benefit and not be misinformed. Such training however, is non-existent in most countries of the region. Choosing counsellors to train is a matter that must be part of the whole planning process in a prevention programme. Specialized geneticists are few in many countries but they can be trainers of doctors, nurses, social workers or other disciplines, who can then be certified as approved counselors. Counselling should result in informed choice, which can only be based on accurate knowledge.

Prenatal diagnosis and termination of affected pregnancies

Prenatal diagnosis proved a solution for many couples detected in prevention programmes in the Mediterranean basin, where a marriage prevention approach was not accepted by the population. Cyprus is one example where any interference with the choice of partner, was rejected as a choice by 97% of the couples counselled [33]. The termination of affected pregnancies is a painful alternative and has been an issue discussed both from the legal point of view, since abortion is illegal in most countries, and the religious point of view. It was this issue that prompted the Church of Cyprus to mandate premarital screening, aiming to bring couples to counselling before the first pregnancy [34]. In the Islamic world, the issue was extensively discussed and a fatwa (a religious verdict) was pronounced by Islamic authorities in several Countries of the Middle East. The decision from various councils (but not all in Islam) was that for serious congenital conditions, termination in early pregnancy, before the first 100-120 days of gestation, is acceptable [35]. This has led to a several countries allowing the practice of prenatal diagnosis and termination of early pregnancy. Such countries are Iran [36], Pakistan [37], Tunisia [38], Bahrain [39], Egypt [40], Iraqi Kurdistan [41] UAE [42].

Pre-implantation genetic diagnosis

An alternative prenatal diagnosis is pre-implantation genetic diagnosis (PGD). This is based on assisted reproduction technology (ART), and in particular in-vitro fertilization (IVF). In this technology ova are fertilized *in vitro*, prior to implantation in the womb. In PGD single cell samples from the early embryos are used for genetic analysis and embryos are selected for implantation which are free of the disease. This technology has been used for haemoglobinopathy prevention since the early 1990s [43]. Even though this is acceptable to many couples, including in Islamic countries, there are several ethical and religious considerations, which have been extensively discussed [44, 45]. One of the main issues is that the technology leaves many embryos unused and their fate is a major concern. In Roman Catholic and Eastern Orthodox Churches, an early embryo is still regarded as loss life but others have compared the fate of the embryo with the birth of a child suffering from a serious genetic disorder. Other considerations include the high cost of the procedure, that it is burdensome to the potential mother especially since implantation often fails requiring the repetition of the whole procedure. The use of PGD has for these reasons limitations, but it remains a legitimate choice of many couples. It is available in many Middle East countries but is minor contributor to the prevention process (Table 1).

Discussion

Prevention of a genetic disease is a complex process, with essential components, which cannot be left out, if the programme is going to succeed. Success is usually measured as a reduction of affected births, but the need to save resources, which will benefit existing patients must not be forgotten. The need for voluntary blood donors may increase beyond the capacity of any system to satisfy, if the number of patients increases annually. Such donations are already inadequate in many countries of the region leaving existing patients under-transfused. The same is also true of essential drugs (mainly iron chelating agents) which 'burden' the pharmaceutical budget and already provision is limited in many countries.

The need for prevention, especially in high prevalence communities, is clear. However planning is essential, considering the need for the community to accept and collaborate as an informed partner in the process. The elements of prevention, discussed in detail in this review, need not be repeated. Convincing health authorities to adopt a national policy and not to leave the initiative to individual laboratories or other enterprises, is the duty of those who are already stakeholders in the haemoglobin disorders. These include professionals and academics in areas such as paediatrics, hematology and public health. Added support from non-governmental organizations, such as thalassaemia associations, is also very important both by lobbying health authorities, sensitizing the public and supporting high risk families and patients.

Prevention is a major component of a comprehensive programme for the control of these hereditary conditions, which will ultimately contribute to the best possible outcomes of patients.

From the information gathered so far by the Thalassaemia International Federation (TIF) much still needs to be done in many countries, within and without the Eastern Mediterranean Region. International partnerships and networking between countries, transfer of technology and staff training are actions that could achieve better results.

Table 1. National control programmes in the countries of the WHO Eastern Mediterranean Region, according to the TIF database.

Country	Expect thal birhts	Expect SCD births	Public awareness program	Screening program	Neonat. screen	PND	PGD	Nat reg
Afghanistan	283	0	no	no	no	no	no	no
Algeria	61	100	limited	no	no	no	no	planned
Bahrain	4	61	yes	yes	yes	yes	?	yes
Egypt	1423	166	limited	limited	no	limited	avail	no
Iran	486	257	yes	yes	no	yes	avail	yes
Iraq	616	183	yes	yes	no	limited	no	no
Jordan	38	40	limited	yes	no	limited	?	?
KSA	138	455	limited	yes	no	no	avail	no
Kuwait	7	18	yes	yes	yes	no	?	yes
Lebanon	9	21	yes	limited	no	yes	yes	yes
Libya	9	40	no	no	no	no	no	no
Morocco	51	164	limited	no	no	no	no	no
Oman	19	96	limited	yes	yes	no	no	yes
Pakistan	5844	455	limited	no	yes	yes	no	no
Palestine Gaza	26	14	no	yes	no	no	no	no
Palestine WB	33	31	yes	yes	no	limited	no	no
Qatar	25	37	no	limited	yes	no	no	no
Syria	338	71	-	-	-	-	-	-
Tunisia	23	55	limited	yes	no	yes	no	no
UAE	124	105	yes	yes	no	limited	?	no
Yemen	391	489	-	-	-	-	-	-
Total	9948	2858	33% yes 38% limited	52% yes 19% limit	81% no	48% avail	19% avail	19% avail

References

1. Silvestroni E, Bianco I. Screening for microcytemia in Italy: analysis of data collection in the past 30 years. Am J Hum Genet. 1975, 27: 198-212.
2. Ashiotis Th, ZachariadIs Z, Sophroniadou K, Loukopoulos D, Stamatoyannopoulos G. Thalassaemia in Cyprus. BMJ. 1973. 2: 38-41.
3. Angastiniotis MA, Hadjiminas MG. Prevention of thalassaemia in Cyprus. Lancet. 1981 Feb 14;1 (8216):369-71. PMID: 6109998.
4. WHO; Geneva: 1994. Guidelines for the control of haemoglobin disorders. WHO /HDP/HB/GL/94.1.
5. Angastiniotis M, Kyriakidou S, Hadjiminas M. The Cyprus thalassaemia control program. Birth Defects: Original Articles Series, 23(5B): 417-432.
6. Ostrowsky JT, Lippman A, Scriver CR. Cost-benefit analysis of a thalassemia disease prevention program. Am J Public Health. 1985, 75(7): 732-6.
7. Ginsberg G, Tulchinsky T, Filon D, Goldfarb A, Abramov L, Rachmilevitz EA. Cost-benefit analysis of a national thalassaemia prevention programme in Israel. J Med Screen. 1998; 5(3): 120-6.
8. Koren A, Profeta L, Zalman L, Palmor H, Levin C, Zamir RB, Shalev S, Blondheim O. Prevention of thalassemia in Northern Israel – a cost benefit analysis. Med J Hematol Infect Dis. 2014; 6(1): e20140122.
9. Leung KY, Lee CP, Tang MH, Lau ET, Ng LK, Lee YP, Chan HY, Ma ES, Chan V. Cost-effectiveness of prenatal screening for thalassaemia in Hong Kong. Prenat Diagn. 2004; 24(11): 899-907.
10. Memish ZA, Saeedi MY. Six-year outcome of the national premarital screening and genetic counselling program for sickle cell disease and -thalassaemia in Saoudi Arabia. Ann Saoudi Med. 2011; 31(3): 229-35.
11. Saffi M, Howard N. Exploring the effectiveness of mandatory premarital screening and genetic counselling programmes for -thalassaemia in the Middle East: a scoping review. Public Health Genomics. 2015; 18:193-203.
12. Christianson A, Howson CP, Modell B. March of Dimes global report on birth defects. March of Dimes Birth Defects Foundation. 2006.
13. Nacul LC, Stewart A, Alberg C, Chowdhoury S, Darlison MW, Grollman C, Hall A, Modell B, Sagoo GS, Burton H. A toolkit to assess health needs for congenital disorders in low- and middle income countries: an instrument for public health action. J Public Health. 2013; 36(2): 243-250.
14. World Health Assembly. 63rd world Health Assembly. Birth Defects. WHA63.17.
15. Old J, Angastiniotis M, Eleftheriou A, Galanello R, Harteveld CL, Petrou M, Traeger-Synodinos J. Prevention of thalassaemias and other haemoglobin disorders. Vol 1: principles. Thalassaemia International Federation publication 18. 2013.
16. Al-Azri MH, Al-Belushi R, Al-Mamari M, Davidson R, Mathew AC. Knowledge and health beliefs regarding sickle cell disease among Omanis in a primary healthcare setting: cross-sectional study. Sultan Qaboos Univ Med J. 2016; 16(4): e437-e444.
17. Okyay RA, Celenk O, Nazlican E, Albaba M. Haemoglobinopathy awareness among young students in Turkey: outcomes of a city-wide survey. Plos One. 2016; 11(7): e0159816.
18. Darr A, Small N, Ahmad WIU, Atkin K, Corry P, Modell B. Addressing the issues in consanguinity-related risk of autosomal recessive disorders in consanguineous communities: lessons from a qualitative study of British Pakistanis. J Community Genet. 2016; 7: 65-79.
19. Anwar WA, Khyatti M, Hemminki K. Consanguinity and genetic diseases in North Africa and immigrants to Europe. Eur J Public Health. 2014; 24(suppl 1): 57-63
20. Knottnerus JA. Community genetics and community medicine. Family Practice 2003; 20(5): 601-6.
21. Traeger-Synodinos J, Harteveld CL, Old JM, Petrou M, Galanello R, Giordano P, Angastiniotis M, De la Salle B, Henderson S, May A on behalf of contributors to the EMQN haemoglobinopathies best practice meeting. EMQN Best Practice Guidelines for molecular and haematology methods for carrier identification and prenatal diagnosis of the haemoglobinopathies. Eur J Human Genetics. 2015; 23, 560.
22. Old J. Prevention and diagnosis of haemoglobinopathies: a short guide for health professionals and laboratory scientists. Thalassaemia International publication 21. 2016.
23. Alkhaldi SM, Khatatbeh MM, Berggren VE, Taha HA. Knowledge and attitudes toward mandatory premarital screening among university students in North

Jordan. Hemoglobin. 2016; 40(2): 118-24.
24. Ansari SH, Baig N, Shamsi TS, Saif-ur-Rehman, Ansari ZH, Behar Z, Perveen K, Erum S, Bukhari ZR, Khan MT, Akbar M. Screening immediate family members for carrier identification and counselling: a cost-effective and practical approach. J Pak Med Assoc. 2012; 62(12): 1314-7.
25. Stirling B, Dormandy E, Roberts T, Ades A, Barton P, Juarez-Garcia A, Andronis L, Karnon J, Marteau TM. Screening for sickle cell and thalassaemia in primary care: a cost effectiveness study. Brit J Gen Practice. 2011; doi: 10.3399/bjgp11X601325.
26. Lena-Russo D, Badens C, Aubinaud M, Merono F, Paolasso C, Martini N, Mattei JF. Outcome of a school screening programme for cariers of haemoglobin disorders. J Med Screen. 2002; 9:67-69.
27. Alswaidi FM, Memish ZA, O'Brian SJ, Al-Hamdan NA, Al-Enzy FM, Alhayani OA, Al-Wadey AM. At-risk marriages after compulsory premarital testing and counselling for -thalassaemia and sickle cell disease in Saudi Arabia 2005-2006. J Genet Couns. 2012; 21(2): 243-55.
28. Cousens NE, Gaff CL, Metcalf SA, Delatycki MB. Carrier screening for beta thalassaemia: a review of international practice. Eur J Hum Genet. 2010; 18:1077-1083.
29. Cowan RC. Moving up the slippery slope: mandated genetic screening on Cyprus. Am J Med Genet. C Semin Med Genet. 2009; 151c(1): 95-103.
30. Muthuswamy V. Ethical issues in genetic counselling with special reference to haemoglobinopathies. Indian J Med Res. 2011; 134(4): 547-551.
31. Reeder R, Veach PM, MacFarlane IM, LeRoy BS. Characterizing clinical genetic counselors' countertransference experiences: an exploratory study. J Genet Couns. 2017; doi: 10.1007/s10897-016-0063-6.
32. Alnaqeb D, Hamamy H, Youssef AM, Al-Rubeaan K. Assessment of knowledge, attitude and practice towards consanguineous marriages mong a cohort of multiethnic health care providers in Saudi Arabia. J Biosoc Sci. 2016:1-18. doi: 10.1017/S0021932016000675.
33. Angastiniotis M, Modell B, Englezos P, Boulyjenkov V. Prevention and control of haemoglobinopathies. Bull World Health Organ. 1995; 73(3):375-86.
34. Kalokairinou EM. The experience of -thalassaemia and its prevention in Cyprus. Med Law. 2008; 27: 825-842.
35. Al-Matari A, Ali J. Controversies and considerations regarding the termination of pregnancy for foetal anomalies in Islam. MBC Med Ethics. 2014; 15:10 doi: 10.1186/1472-6939-15-10.
36. Deshlal MH, Namini MT, Ahmadvand A, Manshadi, M, Varnosfaderani FS, Abolghasemi H. Evaluation of the National Prevention programme with reflections on the path ahead. Hemoglobin. 2014; doi: 10.3109/03630269.2014.893530.
37. Bozdar M, Ahmed S, Jamy OH, Bin Haif T, Ali N, Khan Khattak SA. Role of genetic counselling in prenatal diagnosis of -thalassaemia in Pakistan. J Coll Phys Surg Pak. 2013; 23(8): 553-7.
38. Quali F, Siala H, Bibi A, Hadj Fredj S, Dakhlaoui B, Othmani R, Ouenniche F, Zaouri, Bouguerra B, Rezigua H, Fattoum S, Messaoud T. Prenatal diagnosis oof hemoglobinopathies in Tunisia: an 18 years experience. Int J Lab Hematol. 2016; 38(3): 223-32.
39. Almutawa F, Alqamish J. Outcome of premarital counselling of haemoglobinopathy carrier couples attending premarital services in Bahrain. J Bahrain Med Soc. 2009; 21: 217-220.
40. El-Beshlawy A(1), El-Shekha A, Momtaz M, Said F, Hamdy M, Osman O, Meshaal S, Gafaar T, Petrou M. Prenatal diagnosis for thalassaemia in Egypt: what changed parents' attitude? Prenat Diagn. 2012; 32(8):777-82. doi: 10.1002/pd.3901.
41. Al-Allawi NA1, Al-Doski AA, Markous RS, Mohamad Amin KA, Eissa AA, Badi AI, Asmaro RR, Hamamy H. Premarital screening for hemoglobinopathies: experience of a single center in Kurdistan, Iraq. Public Health Genomics. 2015; 18(2):97-103. doi: 10.1159/000368960.
42. Belhoul KM(1), Abdulrahman M, Alraei RF. Hemoglobinopathy carrier prevalence in the United Arab Emirates: first analysis of the Dubai Health Authority premarital screening program results. Hemoglobin. 2013; 37(4):359-68. doi: 10.3109/03630269.2013.791627.
43. Kuliev A(1), Rechitsky S, Verlinsky O, Ivakhnenko V, Cieslak J, Evsikov S, Wolf G, Angastiniotis M, Kalakoutis G, Strom C, Verlinsky Y. Birth of healthy children after preimplantation diagnosis of thalassemias. J Assist Reprod Genet. 1999; 16(4):207-11.
44. Harper J, Geraedts J, Borry P, Cornel MC, Dondorp WJ, Gianaroli L, Harton G, Milachich T, Kääriäinen H, Liebaers I, Morris M, Sequeiros J, Sermon K, Shenfield F, Skirton H, Soini S, Spits C, Veiga A, Vermeesch JR, Viville S, de Wert G, Macek M Jr; ESHG, ESHRE and EuroGentest2. Current issues in medically assisted reproduction and genetics in Europe: research, clinical practice, ethics, legal issues and policy. Hum Reprod. 2014; 29(8):1603-9.
45. De Wert G, Dondorp W, Shenfield F, Devroey P, Tarlatzis B, Barri P, Diedrich K, Provoost V, Pennings G. ESHRE task force on ethics and Law22: preimplantation genetic diagnosis. Hum Reprod. 2014; 29(8):1610-7.

Diagnostic dilemmas in the detection of complex haemoglobinopathies

Erol Baysal

Professor of Genomics, Consultant & Head of Molecular Genetics, Dubai Genetic and Thalassemia Center, Pathology and Genetics Dept, Dubai Health Authority, Dubai, UAE

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ABSTRACT

We have conducted molecular studies on globin genes for more than 4000 UAE nationals of all ages. The youngest case was several days old and the oldest was over 80. The prevalence of β-globin gene defects encompassing β-thal. and abnormal hemoglobins (Hb) was 8.5%. The latter included common as well as rare Hb variants *e.g.*, Hb S, Hb D-Punjab, Hb O-Arab, Hb C and Hb E. The sickle gene (S or Hb S) contributed significantly to the molecular epidemiology of the hemoglobinopathies in the UAE. Our molecular studies depicted that the majority of the β-thal mutations in the UAE are very severe. Though known as a β+ mutation, the most common IVS-I-5 (G>C) allele exhibits severe β-thal phenotypes in both homozygous and in double heterozygous states. The high frequency of moderate or severe β-thal mutations have implications in the wide spectrum of clinical manifestations seen in patients whose phenotypes vary from β-Thal Intermedia to severe, transfusion-dependent β-Thal Major. Prenatal diagnosis (PND) of hemoglobinopathies in the UAE was imminent following the establishment of mandatory premarital screening program in 2005. Direct detection of mutant genes has enabled many couples to seek DNA diagnostic services in the first trimester of pregnancy. The procedure empowers couples to consider options under informed consent. In the UAE, PND has been employed since 2005 as a principal diagnostic line of prevention through determination of fetal DNA status for β-thalassemia. For over a decade, PND has been available for pregnancies at risk for virtually all inherited hemoglobin disorders in the UAE. Hitherto, nearly 200 couples have been tested using the CVS and subsequent DNA analyses involving PCR and DNA Sequencing. The couples were predominantly from the UAE. Others were from Bahrain, Kuwait, India, Pakistan and other countries in the region. The couples with affected fetuses were counselled and given appropriate available options. Accurate molecular detection and counselling of at-risk couples is a promising way to prevent β-thalassemia in countries where it is prevalent.

Correspondence:

emerinah@eim.ae ; ebaysal@dha.gov.ae

Introduction

The United Arab Emirates (UAE) is a federation of seven emirates situated on the Eastern Arabian Peninsula bordering Oman, Saudi Arabia and Qatar. Iran and the Arabian Gulf are in the north. The population of the UAE is diverse and, like the other Gulf countries, is made up of immigrants from the Middle East, Africa, India, Pakistan, Iran, Southeast Asia and Europe. In the last two decades, the population of the UAE swelled significantly from 600,000 in 1985, to 2.5 million in 2015, and boasts slightly over 9 million people today (estimated at 9.35 million in Jan 2016). Life expectancy is one of the highest in the world; 72.7 years for males and 77.9 for females (1).

Dubai is the most populous emirate in the UAE with an estimated 2.5 million inhabitants (ca. 2015). The oil revenues in the late 60s led to a rapid economic

development that transformed Dubai into a modern metropolis. Today, Dubai is the undisputed commercial and business hub in the entire Gulf Region. The UAE nationals make up approximately 15% of the total population, while Asians (Indians and Pakistanis) make up 50%, Arabs and Iranians 23%, and Westerners and Southeast Asians account for 8%. Around 85% of the population is comprised of expatriates.

β-Thalassemia (β-thal) constitutes a major public health problem in the UAE. Not much was known about the spectrum of β-thal mutations in the UAE until the mid-1990s. Preliminary surveys by White *et al.* (2,3) based on hematological data showed that β-thal and Hb S/β-thal and other abnormal hemoglobins (Hbs) existed in the UAE at high frequencies. Owing to the unavailability of DNA methods, the findings were limited to hematological evaluations such as microcytosis, hypochromia, iron status, and Hb A2 values. The majority of Emirati families traditionally had many children, sometimes several afflicted with a hemoglobinopathy.

Previous hematology-based surveys showed that the UAE exhibited one of the highest carrier frequencies of β-thal in the Gulf region (2-6). The first molecular study on the distribution of β-thal in the UAE was reported by Quaife *et al.* (6) who showed seven β-thal alleles in 50 carriers with the most common allele being the IVS-I-5 (G>C) substitution. It was suggested that this mutation was introduced to the UAE by population migration from the Baluchistan province of Pakistan, which neighbours Iran and Afghanistan.

The Dubai Genetic and Thalassemia Center was inaugurated in 1995. Since then the Molecular Genetics Unit has been actively involved in the identification, characterization and elucidation of all types of hemoglobinopathies, predominantly in Dubai. During 1995-2015, more than 4000 patients were characterized at the molecular level. The patients' nationalities and relative percentages are depicted in Figure 1. Nearly 50% of the patients are Emiratis.

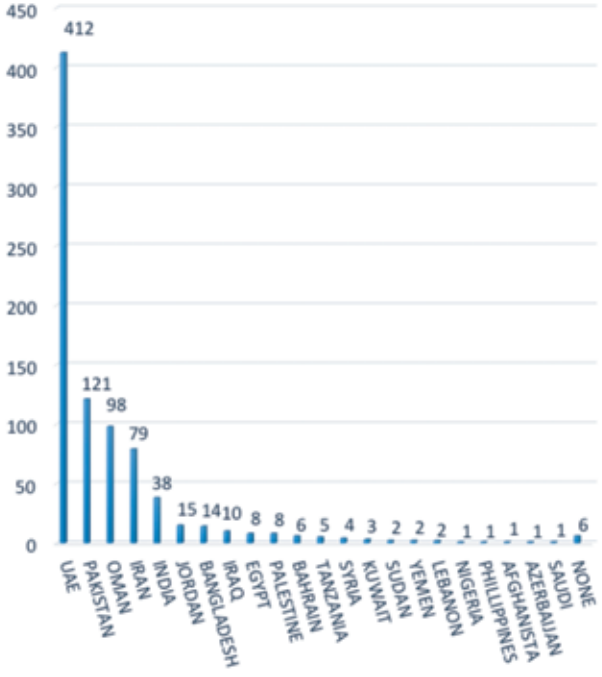

Figure 1. Nationalities of β-thalassemia patients.

Methods

The initial hematological analyses included isoelectric focusing (IEF), quantitation of Hb types by column chromatography. The current investigations used highly sophisticated high performance liquid chromatography (HPLC - Waters (Breeze) and Trinity (Ultra-2).

The molecular characterization and mutational analyses of all β-thal patients were initially carried out using current molecular techniques including amplification refractory mutation system (ARMS), restriction enzyme analysis (REA), dot-blot hybridization, β-strip hybridization, allele-specific oligonucleotide (ASO), polymerase chain reaction (PCR) and gap-PCR. Most of these techniques are now virtually obsolete. Almost all molecular characterizations are currently performed through PCR followed by DNA sequencing using a fully automated ABI PRISM™ 3130 and ABI 3500 Genetic Analyzers.

Results

Our initial study involved 2,000 randomly-selected adult UAE nationals. The results demonstrated that the incidence of β-globin gene defects in the UAE was 8.5% (7,8). The molecular characterization and mutational analyses of all β-thal patients were carried out using current molecular techniques including amplification refractory mutation system (ARMS), non isotopic dot-blot hybridization, restriction enzyme analysis (REA), reverse dot-blot hybridization, β-strip hybridization, allele-specific oligonucleotide (ASO) hybridization, direct polymerase chain reaction (PCR), gap-PCR, manual and automated DNA sequencing. All of these techniques have been described previously (8,9).

Molecular studies began with DNA extraction according to the commonly used procedures. The 5'β segment of the β-globin gene was amplified using a forward primer, located in the upstream promoter region 5' to the Cap site, and a reverse primer in the second intervening sequence. A vast majority of the β-thal mutations in the UAE were found in the 5'β segment of the β-globin gene. The 3'β segment was only amplified whenever mutation screening of the 5'β revealed no mutation.

The molecular studies showed that the majority of the β-thal mutations in the UAE are very severe; the most common allele was the IVS-I-5 (G>C). Although this allele is a β+-thal, its phenotype is very severe. Almost all the other mutations are also severe β0-thal. High frequency of moderate or severe β-thal mutations have implications in the wide spectrum of clinical manifestations seen in patients whose phenotypes vary from β-thal intermedia (βTI) to severe transfusion-dependent β-thal major (βTM).

We describe here the molecular pathology of 838 patients, of which 412 are UAE nationals among whom 249 were homozygous. The molecular pathology of the β-thal patients demonstrated that a vast majority were homozygous. Table 1 shows the distribution of β-thal mutations in 188 homozygous patients (excluding all the abnormal Hbs). The most frequent homozygous mutation was the IVS-I-5(G>C)/IVS-I-5(G>C) (53.0%) followed by –25 bp del/–25 bp del (6.8%), codons 8/9(+G)/codons 8/9(+G) (2.8%) and codon 39(C>T)/codon 39(C>T) (2.4%). These four mutations accounted for 65.0% of the homozygous patient population. Remarkably, the two most prevalent mutations, IVS-I-5 and Hb S, accounted for 77% of all the homozygous β-thal patients from the UAE. We showed 13 discrete homozygous mutations in the UAE national patients in contrast to 23 homozygous mutations in the expatriate

population (data not shown for expatriates). Since the number of homozygous mutations has a direct correlation with the degree of consanguinity, the data shown here corroborate the social tendency towards family planning. It is estimated that in the UAE, marriages are predominantly between relatives; over 50% being between first cousins.

One of the most significant observations derived from our molecular studies is that the majority of the β-thal mutations in the UAE are very severe; except for the most common allele, the IVS-I-5 (G>C). This is a β+-thal mutation while all the others are severe β0-thal alleles. High frequency of moderate or severe β-thal mutations have implications in forming a large spectrum of clinical manifestations seen in patients whose phenotypes vary from β-thal intermedia (βTI) to transfusion-dependent, severe β-thal major (βTM).

Table 2 lists the relative frequencies of the 25 different mutations found in 412 UAE national patients. The first 10 mutations account for 71.1% of the total β-thal chromosomes (excluding Hb S which occurs at 21.1%). Table 2 represents all the β-thal patients; homozygotes and compound heterozygotes. The most frequent mutation was IVS-I-5 (G>C) (44.5%) followed by –25 bp del (8.6%). This is perhaps one of the highest incidences of IVS-I-5 (G>C) allele reported in literature thus far.

The β-thal mutations among the UAE nationals showed considerable heterogeneity, similar to that found in the expatriate population. A total of 53 different compound heterozygotes were observed (data not shown). The most prevalent compound heterozygotes were the IVS-I-5/βS (31 patients), IVS-I-5/–25 bp del (17 patients) followed by IVS-I-5/IVS-I-6 (T>C) and IVS-I-5/ Cd 8 (–AA) (10 patients each). Some of the mutations were rare and some were observed only once. It is important to note that among both homozygous and compound heterozygous patients, all the mutations were β0-thal except for the IVS-I-5 (G>C) β+-thal mutation. However, the latter has a very severe phenotype in the homozygous state or when associated with another β0 allele. This is because only 5% Hb A mRNA is processed through the mutant IVS-I-5 chromosome, a level insufficient to alleviate severe β-globin chain deficiency.

During 1995-2005, our center conducted molecular characterization of 426 expatriate β-thal patients. Of these, 256 were homozygous and 171 compound heterozygotes. The number of expatriate patients in our registry was essentially equal to that of the UAE nationals; 426 expatriates and 412 UAE nationals; 50.8 and 49.2%, respectively (Figure1). The majority of the expatriate patients were from Pakistan (28.3%), Oman (23.0%), Iran (18.5%) and India (8.9%). In the expatriate patients, a total of 78 different combinations of compound heterozygote mutations were defined (data not shown). This was considerably larger than when compared to 53 compound heterozygotes in the UAE nationals. The combined UAE national and expatriate data makes the UAE by far the most heterogeneous β-thal population in the world (11, 12).

Table 3 shows similarities in the distribution of β-thal chromosomes in the UAE national patients and expatriates. The data show remarkable resemblance in the number of homozygotes, compound heterozygotes, and frequency of the most common mutation (IVS-I-5 G>C) in the two populations, thus reflecting close genetic exchanges, family planning, religious, cultural, traditional and historical traits.

Hitherto, our molecular studies revealed 66 different β-thalassemia mutations in the UAE population (Figure 2). In perspective, the total number of mutations in the UAE surpasses the combined number of mutations reported from China and India, just over 32. These are the two most populous nations with combined populations exceeding 2.5 billion. Considering Dubai's population of 2.5 million, this accounts to one thousandth of the above populations. The data reported here reflects considerable diverse molecular heterogeneity in the UAE. It must be noted that the genetic diversity of this scale poses enormous problems in establishing prevention programs and offers frightening prospects to all of us who are engaged in prenatal diagnosis programs for β-thal in the UAE. However, this problem is generally circumvented by carrying out mutational studies and genetic counselling on prospective family members and creating a comprehensive database prior to performing molecular analysis.

Complex hemoglobinopathies

Common hemoglobinopathies such as β-thal, α-thal, HPFH, δβ-thal and abnormal Hbs (HbE, S, C, Lepore, D-

Table 1. Homozygous β-thalassemia in the United arab emirates national patients.

	Mutation	*n*	Number of chromosomes	Frequency (%)[a]
1	IVS-I-5(G>C)/IVS-I-5(G>C)	132	264	53.0
2	–25 bp del/–25 bp del	17	34	6.8
3	Cd 8/9(+G)/ Cd 8/9(+G)	7	14	2.8
4	Cd 39(C>T)/ Cd 39 (C>T)	6	12	2.4
5	Cd 30(G>C)/ Cd 30(G>C)	5	10	2.0
6	IVS-II-1(G>A)/IVS-II-1(G>A)	4	8	1.6
7	Cd 5(–CT)/Cd 5(–CT)	4	8	1.6
8	–88(C>A)/–88(C>A)	3	6	1.2
9	IVS-I-1(G>A)/IVS-I-1(G>A)	3	6	1.2
10	Cd 15(G>A)/ Cd 15(G>A)	2	4	0.8
11	Cd 8(–AA)/ Cd 8(–AA)	2	4	0.8
12	IVS-I-110(G>A)/IVS-I-110(G>A)	2	4	0.8
13	Cd 82/83(–G)/ Cd 82/83(–G)	1	2	0.4
TOTAL		188	376	

[a]Excluded from the table are Hb SS (23.7%) and Hb DD (0.8%).

Punjab, O-Arab) are found in the UAE and they are detected easily. Their accurate identification can provide valuable diagnostic and prognostic options for clinicians and for couples who may consider family planning.

The diagnosis of hemoglobinopathies has become an increasing challenge in the multinational countries such as Australia, USA, Canada as well as in Europe and the Gulf Countries. This is well exemplified by Dubai which is home to around 230 different nationalities (2015 census), hence the propensity for significantly enriched thalassemia gene pool coupled with high degree of consanguinity. In an attempt to curb the hemoglobinopathy problem, the National Premarital Screening Program (PMS) became mandatory in Dubai in 2006 for all nationalities and the Prenatal Diagnosis Program (PND) has been underway successfully since 2005. These prevention programs were rendered mandatory, as hemoglobinopathies are a major public health concern in the UAE.

Dubai is arguably the most heterogeneous hemoglobinopathy nation in the world with 66 β-globin gene defects reported to date. It is anticipated that various complex hemoglobinopathies with extensive heterogeneity in genotype and variable phenotype will emerge from such admixture of genes in a small nation where first cousin marriage among the indigenous population is a norm and not an exception. In addition, α-thal and β-thal interactions occur due to relatively high frequencies of α globin and β globin gene defects; 50% and 8.3%, respectively (11, 12). This makes clinical diagnosis and laboratory evaluation much more challenging in a young nation where 26% of the population is below age 15 and 71% is between 15-64.

The laboratory diagnoses of hemoglobinopathies are often made with certain assumptions:

(a) Variation in the phenotype could be a reflection of interplay between different abnormal globin genes (α, β, γ, δ).
(b) Same genotypes may have different phenotypes in different geographical areas thus denoting the role of environment as a modulator.
(c) Complex genotypes occur rarely so no concrete conclusions must be drawn from only a few examples.

The complex hemoglobinopathies provide valuable information and guidance to clinicians, counsellors and healthcare providers. Extra care and vigilance is required to identify the causative mutations in these cases. These rare cases also highlight and emphasize the importance of accurate laboratory assessment and interpretation especially with reference to different normal ranges for MCV, MCH, HbA2 quantitation.

Prenatal diagnosis

Prenatal diagnosis (PND) of hemoglobinopathies in the UAE was imminent following the establishment of mandatory premarital screening program in 2005 and the advances made in chorionic villus sampling (CVS) and in DNA-based diagnostics. Advances in CVS aspiration have rendered the first trimester PND a standard practice. Direct detection of mutant genes has enabled many couples to seek DNA diagnostic services in the first trimester of pregnancy. The procedure empowers couples to consider options through proper genetic counselling coupled with informed consent. In the UAE, PND has been employed since 2005 as a principal diagnostic means to determine fetal DNA status for β-thal.

The ability to detect mutant globin genes in CVS has provided a rapid, safe, accurate, reliable and affordable

Table 2. β-thalassemia Gene Frequency among the Emirati patients (*n*=412).

	Mutation	No. of chromosomes	Gene frequency (%)[a]
1	IVS-I-5 (G>C)	367	44.5
2	–25 bp deletion	71	8.6
3	Codons 8/9 (+G)	25	3.0
4	IVS-II-1 (G>A)	23	2.8
5	Codon 39 (C>T)	18	2.2
6	Codon 8 (–AA)	18	2.2
7	Hb D-Punjab (GAA>CAA)	18	2.2
8	Codon 30 (G>C)	17	2.1
9	Codon 5 (–CT)	17	2.1
10	IVS-I-6 (T>C)	12	1.5
11	–88 (C>A)	9	1.1
12	Codons 82/83 (–G)	8	1.0
13	IVS-I-110 (G>A)	8	1.0
14	IVS-I-5 (G>T)	7	0.9
15	Codon 15 (G>A)	7	0.9
16	Codon 44 (–C)	6	0.7
17	Codon 110 (T>C)	3	0.4
18	IVS-II-848 (C>A)	3	0.4
19	Poly A site (AATAAA>AATAAG)	3	0.4
20	–101 (C>T)	2	0.2
21	Hb Knossos (codon 27, G>T)	1	0.1
22	Codon 37 (G>A)	1	0.1
23	Codons 36/37 (–T)	1	0.1
24	Hb E (codon 26, G>A)	1	0.1
25	δβ deletion	1	0.1

[a]Excluded from the table is Hb S (21.1%).

Table 3. Similarities in β-thalassemia alleles between emirati and expatriate patients.

Patients[a]	UAE n: 412		Expatriates n: 426	
Homozygotes	249	60.0%	264	62.0%
Compound heterozygotes	163	40.0%	162	38.0%
IVS-I-5(G>C)/IVS-I-5(G>C) (in all homozygotes)	132	54.0%	130	49.2%
IVS-I-5(G>C)/IVS-I-5(G>C) (in all patients)	132	32.0%	130	31.0%
IVS-I-5 (G>C) chromosomes	367	44.5%	336	39.4%
Homozygous mutations	15		23	
Compound heterozygous mutations	53		78	

[a]Hb S patients are excluded.

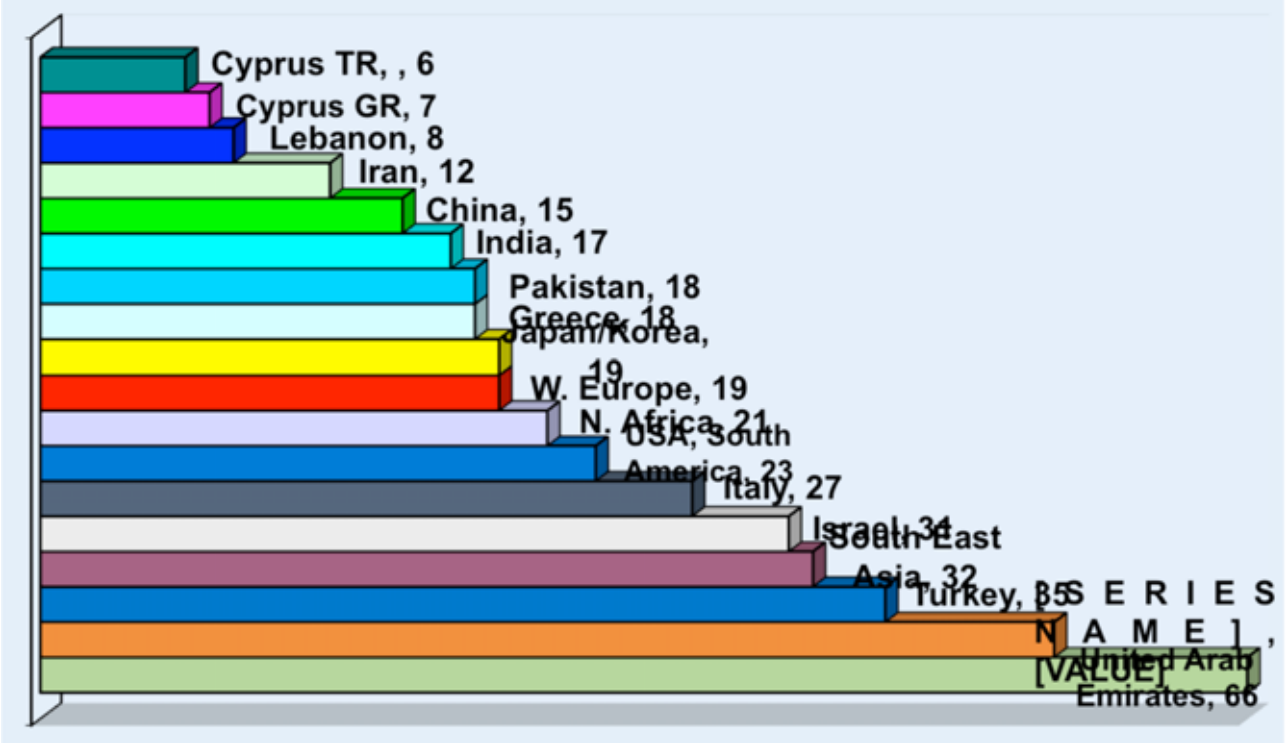

Figure 2. Heterogeneity of β-thalassemia mutations worldwide as measured by the total number of alleles in a given country or region.

methodology for the early detection of many fatal genetic diseases. The genetic information enables the couples to reach a decision compatible with their beliefs and family planning criteria.

In the Dubai Genetics Center, we employ the most advanced diagnostic tools including PCR and DNA Sequencing to diagnose many hemoglobinopathies encompassing thalassemias (α & β-thal), sickle cell disease (SCD) and abnormal hemoglobins. The DNA is extracted from CVS using Qiagen kits and is amplified by PCR using specific primers. The amplicons are sequenced on an ABI Genetic Analyzer 3130 and 3500. The results are reported within 24 hours. Without exception, the maternal contamination is excluded for each sample with STR Cofiler®.

Our results demonstrate that since 2005, PND has been available for pregnancies at risk for virtually all inherited hemoglobin disorders in the UAE. Nearly 200 couples have been tested using the CVS and subsequent DNA analyses involving PCR and DNA Sequencing. The couples were predominantly from the UAE. Others were from Bahrain, Kuwait, India, Pakistan and other countries in the region. The couples with affected fetuses were counselled and given appropriate available options. Our PND data will be not presented here.

Summary

- The IVS-I-5 (G-C) allele is the most prevalent mutation among the Emiratis and the expatriates with a frequency of >50%.
- Sickle cell gene (β-sickle) is the second most prevalent allele accounting for >20% of the chromosomes.
- Significantly high incidence of homozygous mutations demonstrate the degree of consanguinity among the UAE nationals.
- Frequency of β gene defects that encompass β-thal, β-sickle and β-variants is 8.3%.
- Frequency of various α-thal genotypes is around 50%
- UAE is the most heterogeneous population with 66 alleles at present surpassing the most populated nations like China and India put together.
- Most of the mutations identified in the UAE population are β0/+ leading to very severe phenotype.
- Premarital Screening (PMS) is mandatory since 2006.
- Prenatal diagnosis (PND) was implemented in 2005 and resulted in decreasing the β-thal births dramatically. At present there is significantly high demand for this rapid, sensitive, accurate, affordable procedure across all factions of society.

Conclusions

In conclusion, accurate detection and counselling of at-risk couples is a promising way to reduce the mortality and morbidity from β-thalassemia in countries where it is prevalent.

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References

1. Al Hosani H. Health for all in the United Arab Emirates. East Mediterr Health J. 2000;6(4):838-840.
2. White JM, Byrne M, Richards R, *et al.* Thalassaemia genes in Peninsular Arabs. Br J Haematol. 1985;60(2):269-278.
3. White JM, Byrne M, Richards R, Buchanan T, Katsoulis E, Weerasingh K. Red cell genetic abnormalities in Peninsular Arabs: sickle haemoglobin, G6PD deficiency, and α and β thalassaemia. J Med Genet. 1986; 23(3):245-251.
4. Al-Gazali LI, Bener A, Abdulrazzaq YM, Micallef R, al-Khayat AI, Gaber T. Consanguineous marriages in the United Arab Emirates. J Biosoc Sci. 1997;29(4): 491-497.
5. El Hazmi MA, Al-Swailem AR, Warsy AS. Molecular defects in β thalassaemias in the population of Saudi Arabia. Hum Hered. 1995;45(5):278-285.
6. Quaife R, al Gazali L, Abbas S, *et al.* The spectrum of β-thalassemia mutations in the UAE national population. J Med Genet. 1994;31(1):59-61.
7. Baysal E. Advances in Hemoglobinopathies Research – Hemoglobinopathies in the United Arab Emirates. Hemoglobin. 2001;25(2):247-253.
8. Baysal E. Molecular heterogeneity of β-thalassemia in the United Arab Emirates. Community Genet. 2005;8(1):35-39.
9. El Kalla S, Baysal E. Genotype-phenotype correlation of sickle cell disease in the United Arab Emirates. Pediatr Hematol Oncol. 1998;15(3):237-242.
10. Baysal E, Yousef I, Zeinali M, *et al.* Molecular basis of hemoglobinopathies in the UAE: implications for prenatal diagnosis and prevention programs. Emirates Med J. 2007;25(1):7-21.
11. Baysal, E., Molecular basis of α-thalassemia in the United Arab Emirates Hemoglobin; 35 (5-6):574-80, 2011. PMID: 22074123.
12. Baysal, E., Molecular basis of β-thalassemia in the United Arab Emirates. Hemoglobin; 35 (5-6):581-88, 2011. PMID: 22074124.

How I treat and monitor sickle cell disease

Wasil Jastaniah[1,2]

[1]Department of Pediatrics, Faculty of Medicine, Umm AlQura University, Makkah, Saudi Arabia; [2]Princess Noorah Oncology Center, King Abdulaziz Medical City, Jeddah, Saudi Arabia

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ABSTRACT

Sickle cell disease is the most common genetic hemoglobin disorder worldwide with thousands of children born yearly with this disease. Advances in our understanding of sickle cell disease management has improved over the years and it is now expected that, with early screening, monitoring, prevention, and intervention, children with sickle cell disease can live well into adulthood. However, the burden of the disease is shifting to adulthood in developed and resource-rich countries. In contrast, due to the limited access to comprehensive care programs for children with sickle cell disease in resource-poor countries, high morbidity and early mortality of patients with sickle cell disease remains a major global problem. Therefore, this review will discuss the strategy and recommendations we use in monitoring and treating patients with sickle cell disease in our center. The review will also highlight some of the challenges faced in less developed countries.

Correspondence:

jastaniahwa@ngha.med.sa

Introduction

Sickle cell disease (SCD) is an autosomal recessive disorder that results from a point mutation leading to a single nucleotide substitution (A to T) in the codon for the 6th amino acid of the beta-globin chain and, as a result, substitutes the hydrophobic amino acid valine for the normal hydrophilic amino acid glutamic acid [1]. This results in hemoglobin polymerization and precipitation leading to red cell membrane damage and deformity into sickle shaped cells. This leads to increased red cell destruction (hemolysis) and red cell clumping in vessels (vaso-occlusion). The hemolytic and vasco-occlusive phenotype lead to the spectrum of clinical manifestations observed in patients with SCD from severe anemia, leg ulcers,, and pulmonary hypertension to recurrent pain crisis, acute chest syndrome, splenic infarction, stroke, and avascular necrosis on the other end of the spectrum [2]. The clinical severity of these manifestations are influenced and modified by genetic and environmental factors that result in the variability observed between patients with SCD [1,2].

SCD is a global health problem affecting approximately 2-3% of the world population with variable geographic distribution [3]. The prevalence of sickle cell trait reaches up to 40% in some regions such as central India, the Eastern province of Saudi Arabia, and Sub-Saharan Africa and, worldwide, approximately 275,000 are born with SCD annually and need early diagnosis, monitoring, and treatment [3,4,5,6]. In developed countries, early newborn screening, penicillin prophylaxis, comprehensive care programs, institution of disease modifying therapies such as hydroxyurea, blood transfusion, and chelation therapy, in addition to, hematopoietic stem cell transplant (HSCT) as curative therapy have improved the survival and quality of life of patients with SCD and the majority of children are expected to survive well into adulthood [7]. In contrast, 90% of children in resource-poor countries do not reach adulthood [3,4,5].

In this review, the importance of early screening, penicillin prophylaxis, immunization, monitoring and screening, institution of a comprehensive care program, and transcranial Doppler (TCD) screening and stroke prevention will be reviewed. In addition, the review will highlight the indications of hydroxyurea, transfusion therapy, and HSCT in SCD. Finally, the review will focus on the current challenges faced in developing countries when dealing with patients

with SCD. The recommendations made are based on the presented evidence and the standard practice used in the authors' center.

Early screening and monitoring

Early newborn screening, initiation of penicillin prophylaxis in early infancy, and early education have been shown to reduce the risk of mortality from infections in infants and young children with SCD [8]. However, universal newborn screening is not implemented in all countries around the globe and reinforcing the importance of including SCD in newborn screening programs particularly in areas with high prevalence of the disease should be a priority to be able to initiate early preventative and screening measures to reduce the morbidity and mortality associated with SCD. This includes the early initiation of penicillin prophylaxis, immunizations, and providing access to a comprehensive SCD program.

Penicillin prophylaxis

Infection has been the leading cause of death in children with SCD under the age of 5 years. The early initiation of penicillin prophylaxis has been shown, in the randomized trial on prophylaxis with oral penicillin in children with sickle cell anemia (PROPS I) study, to reduce the risk of developing serious pneumococcal infection in SCD children under the age of 3 years by 84% and abolish mortality from this infection [9]. Subsequently, the PROPS II study showed that, in children without history of severe pneumococcal infection or surgical splenectomy, discontinuing penicillin prophylaxis at the age of 5 years may be safe [10]. Therefore, early initiation of penicillin prophylaxis between the age of 2 to 4 months at a dose of 125 mg twice daily until the age of 3 years, and 250 mg twice daily until the age of 5 years is recommended. Discontinuing penicillin prophylaxis after the age of 5 years in children with SCD who have completed their immunization schedule, had no prior invasive pneumococcal infection, or surgical splenectomy is safe. However, it is important to educate the family regarding the importance of seeking medical intervention in case of fever. Therefore, assessment of social circumstances that may lead to delayed access to care needs to be considered before discontinuing prophylactic therapy.

Immunizations

The delivery of routine immunizations to children is considered one the most important and cost-effective preventive measures that has saved many childhood lives over the years. Patients with SCD are at increased risk of invasive bacterial infection from encapsulated organisms [11]. Therefore, in addition to the routine childhood immunizations, we give the 23-valent pneumococcal polysaccharide vaccine (PPSV23) at 2 years and a second repeated dose at 5 years. In addition, we recommend giving the meningococcal (MenACWY) vaccine 3 years after completing the primary vaccines and a booster repeated every 5 years thereafter. Furthermore, in our practice we recommend to give children above the age of 6 months the seasonal influenza vaccine annually. Patients planned for an elective splenectomy should also be planned to receive all vaccines in addition to vaccination against the pneumoccal (PCV13) and hemophillus influenza b (Hib) vaccine (if not previously received), the PPSV23, and the MenACWY with the last vaccine given at least 2 weeks prior to surgical splenectomy. These patients should continue on lifelong penicillin prophylaxis.

Monitoring and treatment of SCD

The first visit of a patient and his/her family with SCD is an important visit to conduct proper education and explain the natural history of SCD with its potential complications. The visit should include explanation regarding emergency signs that should alert the family to seek immediate medical attention such as high fever above 38.5 degrees Celsius, pain and swelling in the extremities, respiratory symptoms, abdominal pain and distension, neurological symptoms, increased pallor or fatigue, priapism, or vomiting and diarrhea. Teaching the family how to perform a spleen exam is also important and needs to be reviewed on subsequent visits. The frequency and goals of follow up and monitoring should be explained. The family should be able to understand the chronic nature of the disease and the importance of early prevention to reduce the short- and long-term morbidity and mortality associated with this disease. The monitoring methods and potential need for transfusion should be explained. Based on the age of the child, anticipatory guidance to reinforce the importance of screening and prevention is important. As the child grows to school age, communication with the school via a medical report and instructions regarding hydration and symptoms that would require immediate medical attention should be highlighted. Advice regarding screening of family members and newborns, if not routinely available, should be recommended during the first visit as well.

Comprehensive care program

During the first visit, we usually show the plan of monitoring and introduce the family to the concept of comprehensive care upfront to explain the rational for the different screening modalities as summarized in Table 1.

As shown in Table 1, the frequency of visits is once every 2 months in the first 6 months, followed by once every 3 months until 18 months of age, and then 6 monthly thereafter. During each visit, we review history of sickle cell crisis, admissions, transfusion history, home medication use, daily activity, and sleeping difficulties including snoring. In addition, monitoring vital signs including blood pressure, pain score, and growth parameters are important to record baseline values. Documentation of the spleen size, if palpable, hip and shoulder range of movement, respiratory, and neurological exam. These, together with the laboratory and imaging surveillance, can help in stratifying the severity of the SCD phenotype to adjust the frequency of visits and implement timely preventative measures. As the patient approaches 10 years of age, it is our practice to perform a cardiac evaluation, if not previously done, to assess for prolonged QTc, echocardiogram to assess tricuspid jet velocity to screen for pulmonary hypertension, and a pulmonary function test to screen for underlying lung disease. Although, the evidence to support routine screening in asymptomatic children is not strong, we believe that these baseline exams can help predict and guide in risk stratification of the severity of the sickle cell phenotype and the need for more frequent monitoring, in addition to delivering early interventions as appropriate. Similarly, we screen asymptomatic patients in our center for silent infarcts by brain MRI/MRA when the patient is old enough to get it done without any sedation. As the patient graduates to adulthood, we ensure that a full assessment including ophthalmology retinal exam, screening for avascular necrosis by x-ray or MRI based on symptoms, and urinalysis for microalbuminuria is performed.

TCD and stroke prevention

Over the past 20 years, our understanding and management of neurological complications in SCD has improved. This is reflected by the reduction in risk of overt stroke in countries utilizing Trans cranial Doppler (TCD) ultrasound screening for central nervous system (CNS) vasculopathy. Adams and colleagues have shown that starting patients with abnormally high TCD velocities (above 200 cm/second) on a strict chronic transfusion program that targets reducing the hemoglobin S level to less than 30%, can reduce the risk of development of a first stroke [12,13]. The STOP 1 and STOP 2 trial confirmed the benefit of continued chronic transfusion in children between the ages of 2 and 16 years with SCD in primary stroke prevention [12,13]. In fact, the routine use of TCD screening together with regular blood transfusion therapy has reduced the prevalence of overt stroke in developed countries from approximately 11% to 1% [14]. Recently, the TWiTCH trial has shown that bridging patients, who convert to normal TCD velocities on chronic transfusion therapy, using hydroxyurea until maximum tolerated dose is achieved then switching them to hydroxyurea alone was not inferior to continuation of chronic transfusion for primary stoke prevention [15]. This latter study, together with others, suggests a beneficial role of hydroxyurea therapy in primary stoke prevention [16,17,18]. However, optimal patient selection and monitoring is required.

In contrast, overt stroke remains a challenging and devastating problem. Chronic transfusion therapy for secondary stroke prevention can reduce the recurrence rate of overt stroke by 90% [18,19,20]. However, approximately half of patients with overt stroke on chronic transfusion continue to develop progressive vasculopathy suggesting the need for alternative approaches to therapy [21]. Switching patients with overt stroke from chronic transfusion to hydroxyurea has not been proven to be effective based on the SWiTCH trial which was terminated early [22]. However, hydroxyurea is superior to no therapy in preventing stroke recurrence [23].

Silent cerebral infarct is the most frequent neurological complication reported in patients with SCD [14,24]. The cumulative prevalence of silent cerebral infarct increases with age and it is estimated that approximately 53.3% of patients suffer from a silent cerebral infarct by the 4th decade of life [14]. Patients with silent cerebral infarct have an increased risk of developing overt stroke compared to SCD patients without silent cerebral infarct [14,24,25]. The SIT trial studied the benefit of transfusion therapy compared to standard care and showed that transfusions resulted in a relative risk reduction of 58% in the occurrence of all neurological events [25]. Based on the results of the SIT trial, patients with silent cerebral infarct may be offered transfusion therapy to prevent progression of neurological events [25]. However, as the benefit of transfusion to prevent progression was incomplete, further modalities to address the optimal management of patients with silent cerebral infarct are needed. The use of hydroxyurea or HSCT as alternative therapies to blood transfusions in patients with silent cerebral infarct requires further studies.

Hydroxyurea

Hydroxyurea (or hydroxycarbamide) is a ribonucleotide reductase inhibitor that has been used in patients with SCD for more than 20 years now [26-28]. Many experts believe that hydroxyurea has changed the lives of patients with SCD. Hydroxyurea exerts its effects by inducing fetal hemoglobin, improving laboratory parameters through its effects on hematopoiesis and thus reducing the inflammatory state, reducing hemolysis, and improving nitric oxide bioavailability [27]. Overwhelming studies have shown the effectiveness and safety of hydroxyurea in both adults and children in ameliorating the clinical complications and organ dysfunction, reducing the need for blood transfusions, decreasing the rate of hospitalizations, and attenuating mortality [26,28,29]. Based on the available evidence, the most recent published National Institute of

Table 1. Sickle cell disease comprehensive screening and monitoring.

Age	History/Exam	CBC	HPLC	RBC phenotype	Chemistry	U/A	Pulse O2%	CXR	ECG, Echo, PFT	CNS	GB U/S	Ophth Consult
2 mo												
4 mo												
6 mo												
9 mo												
12 mo												
15 mo												
18 mo												
2-9 yr												
≥10 yr												
New Pt												

2 mo visit: education, start penicillin and folic acid prophylaxis, immunization
Extra Immunizations: PPSV23 at 2 and 5 years of age
TCD start at 2 years than annually (follow TCD protocol)
MRI/MRA brain once when able to do it without sedation (around 10 years)
Hip/shoulder x-ray/MRI: If symptomatic (or upon graduation to adult practice)
For patients on transfusion:
- Screen for transfusion-related infections and liver MRI-iron overload once every 1-2 years;
- Ferritin level every 3 months.

Health Evidence Based Expert Panel Report on Management of SCD recommended that every child with SCD should be offered hydroxyurea from the age of 9 months even if asymptomatic [30]. In our practice, we offer to the family hydroxyurea with the first episode of sickle cell-related crisis if the child is 9 months of age or older. The indications, dose, and monitoring of hydroxyurea based on our practice is summarized in Table 2.

Transfusion

Red blood cell transfusion therapy is the first therapy used to target the pathophysiology of SCD. The indications of transfusion therapy have expanded over the years and more children are now being transfused chronically to prevent stroke and other complications of SCD. There are two methods of transfusion in patients with SCD: intermittent/simple transfusions and exchange transfusions. In contrast to transfusion therapy in patients with thalassemia, the goal of transfusion in SCD is to improve the anemia and reduce the sickle hemoglobin (Hb S) level to reduce the hemolysis and viscosity associated with SCD. However, due to the presence of sickle cells in the blood, the viscosity of the blood in patients with SCD is increased. Therefore, caution should be practiced when prescribing blood transfusion therapy to patients with SCD to avoid hyperviscosity and its deleterious effects on patients with sickle cell. The hemoglobin level should not exceed 11 gm/dl, and that is why exchange transfusion may be the only method to transfuse patients with SCD presenting with a hemoglobin levels above 9 gm/dl. The indications for red cell transfusions in SCD are outlined in Table 3.

The long-term use of blood transfusion has its consequences and patients with SCD receiving multiple or chronic transfusions develop iron overload that requires monitoring and chelation therapy. Chelation therapy in patients with SCD should be initiated when the total number of life time transfusions reaches 20 simple transfuisons (120 ml/kg) or when the liver iron concentration, as assessed by a quantitiative measurement (MRI measurement), exceeds 7 mg Fe/g liver dry weight [30,31]. Defiraxirox therapy has been reported to be effective and safe in treating iron overload in patients with SCD even if hydroxyurea is concomitantly used [32,33]. Other complications of transfusion therapy in patients with SCD include: alloimmunization, venous access, compliance, cost, and infection. These complications are increased in patients receiving exchange transfusion due to the increased exposure to blood with each exchange transfusion compared to simple transfusions. Therefore, efforts are needed to reduce the risk of these complications and further research is required to find alternatives to transfusion therapy.

Indication for HSCT

HSCT is the only available curative therapy in patients with SCD to date. Human leukocyte antigen (HLA)-identical related donor HSCT for children with SCD has a long-term overall survival rate of 94-97%, an event-free survival rate of 84-86%, and a graft rejection rate of approximately 10% as reported by the North American and European studies [34,35]. Despite this high success rate, HSCT remains limited by optimal patient selection criteria, infertility risk with current conditioning regimens, availability of donors, and cost.

Table 2. How I use hydroxyurea in sickle cell disease.

Indications	
1.	Dactylitis or pain crisis
2.	Acute chest syndrome
3.	Hemolytic crisis requiring transfusion therapy
4.	Conditional TCDI
5.	Parent preference in patient with abnormal TCDI or after TCDI normalization with chronic transfusion
6.	Parent preference in patient with Silent cerebral infarct (hydroxyurea vs. chronic transfusion program)
7.	Secondary stroke prevention if transfusion not possible or in combination with transfusion if progressive vasculopathy or increased frequency of exchange transfusion
8.	Parent request (age 9 months or older)
Dose	
	Start at 15 mg/kg/day and increase to maximum tolerated dose (maximum dose 30-35 mg/kg/day)
Monitoring	
1.	Before starting perform baseline CBC with differential, reticulocyte count, hemoglobin electrophoresis (HPLC), Creatinine, and LFT (ALT, bilirubin)
2.	Follow up 1 month after starting or escalating the dose to assess for toxicity with CBC and differential (hold if ANC <1x10^9/L or platelet count <100 and reduce dose if ANC=1 -1.5x10^9/L or platelet count <150. Restart at lower dose once recovered)
3.	Escalate dose monthly, by 5 mg/kg/dose, to maximum dose or maximum tolerated dose, whichever occurs first
4.	Once maximum/maximum tolerated dose reached, follow the patient every 3-4 months and assess for compliance and toxicity (CBC with differential, MCV, creatinine, ALT, reticulocyte count)
5.	Assess for efficacy by reviewing history (crisis, admissions, and blood transfusions), MCV, WBC, reticulocyte count, bilirubin, LDH, and measure Hb electrophoresis twice yearly until HbF level stable then measure as needed

The indication of HSCT in patients with SCD is summarized in Table 4. These indications are based on the selection criteria used in published clinical trials [36]. However, the timing of HSCT is important as delaying transplant after significant morbidity like stroke may affect the quality of life of such patients. Other criteria that may be considered, as indication for HSCT, is abnormal high-risk TCD velocities on screening for primary stroke prevention. However, the recent results from the TWiTCH trial showing the efficacy of hydroxyurea therapy as an alterantive to chronic blood transfusion may challenge this indication [15]. Further studies are therefore needed to study the best timing, patient selection, conditioning regimen, and donor source in patients with SCD.

Challenges in less developed countries

The majority of children with SCD are born in less developed countries. It is reported that 90% of children living in resource-poor countries do not survive to adulthood and more than half of children die before their fifth birthday [37-39]. Therefore, there is a high demand to improve early screening and intervention strategies in less developed nations. The following are some of the challenges that need to be addressed globally:

Newborn screening programs in developing countries

Newborn screening for sickle cell is not universally available at the global level. Advocating for and initiating early newborn screening will allow implementation of early antibiotic prophylaxis, optimize immunizations, and early education to prevent early death from infections. Alternatives to newborn screening such as premarital screening programs to reduce at-risk marriages, preimplantation genetic diagnosis (PGD), and school educational programs may help [40,41]. However, these programs are either associated with high cost or have shown little, if any, benefit in reducing early mortality rates from SCD, or in reducing the incidence of SCD in countries with high prevalence rates [40,41]. Therefore, there is a need to establish newborn and early screening and intervention programs in developing countries.

Standard clinical care pathways and guidelines

In developing countries, there is variation in resources and in the clinical phenotype even within the same country, in addition, there is lack of targeted guidelines. This results in lack of standardized care delivery systems and timely prevention, which impacts the disease burden and the utilization of resources. The variability in phenotype can result in a different spectrum of dis-

Table 3. Indications for red blood cell transfusion in sickle cell disease.

Acute/episodic transfusion	Chronic (long-term) transfusion
Acute stroke	Prevention of recurrent stroke Primary stroke prevention Prevention of recurrent SCI
ACS	
Splenic sequestration	Previous splenic sequestration (≤2–3 years)
Pre-operatively (select cases) Severe hemolytic anemia Severe or long-lasting aplastic crises Acute multiorgan-failure syndrome	Heart failure/renal failure/chronic PHT Chronic pain in HU non-responders Short program: pregnancy

Table 4. Indicaitons for hematopoietic stem cell transplant in patients with sickle cell disease. [36]

Children	Adult
Stroke or CNS event lasting >24 hours	Age 15 to 40 years and any of the indications below: Stroke or CNS event lasting >24 hours
Impaired neuropsychological function *with* abnormal MRI/MRA	Regular RBC transfusion therapy to prevent vaso-occlusive clinical complications (*i.e.*, pain, stroke, and acute chest syndrome)
Recurrent acute chest syndrome	Recurrent ACS in the 2-year period preceding HCT despite supportive care measures (*i.e.*, asthma therapy and/or HU)
Sickle lung disease Recurrent VOC or recurrent priapism Sickle nephropathy (GFR 30-50% of predicted normal) Alloimunization	3 VOC per year in the 2-year period preceding enrollment despite supportive care measures (*i.e.*, pain management and/or treatment with HU) ECHO TRV Jet velocity >2.7 m/s

ease. For instance, the Arab-Indian haplotype has been labeled as benign for many years but patients with this phenotype present with different patterns of morbidity [6,42]. This phenotype is predominant in certain regions of the globe such as the Eastern province of Saudi Arabia, Kuwait, Oman, and India. Patients with this haplotype suffer from more splenic complications and avascular necrosis in childhood [6,42]. In addition, as patients with this haplotype enter into adulthood, they suffer from severe sickle cell related complications including acute chest syndrome, recurrent pain, stroke, and mortality peaking in the second and third decade of life [43,44]. This suggests that regions with high prevalence of the Arab-Indian haplotype may require comprehensive care and intervention programs tailored to the natural history of the specific phenotypic spectrum of the disease. Further studies in these regions are warranted to deliver cost-effective interventions that would improve the quality of care of these patients. Developing structured and standard guidelines based on phenotype predominance and resources is required to help improve the care in different regions around the globe. This would require further clinical trials in identifying alternative and cost-effective therapeutic and preventative approaches.

Demands and resources in less developed countries

As the majority of patients with SCD live in less developed countries, the availability of resources including access to care, transfusion and chelation therapy, cost of screening programs, or availability of sufficient resources to meet the high demand in specific countries remains a big challenge [45]. This is even true in resource-rich countries. For example, a recent study from Saudi Arabia (a resource-rich developing country) estimated the number of patients that were candidates for curative HSCT in the pediatric and adult population using strict HSCT indications and found that with the current high demand, due to the high prevalence of the disease, the numbers of transplant facilities were limited to match the current demand [46]. Therefore, efforts in improving preventative measures and delivery of disease modifying therapies to improve the quality of life and reduce the morbidity and mortality of this disease remains a priority in developing nations.

Challenges in low-and middle-income countries

The variability in the resource settings in low- and middle-income countries have a major influence on the survival and quality of life of patients with SCD living in these countries. This includes access to health care, access to medications, and access to blood transfusions. In fact, death from infection remains the major cause of mortality in these countries [37-39,]. Therefore, focus on early diagnosis and antimicrobial prophylaxis to prevent infection should be the main priority. Screening and access to healthcare may also be a challenge, therefore, consideration of disease modifying therapies such as hydroxyurea for all children 9 months of age or older at a fixed dose may be appropriate in these countries, this is particularly true as blood transfusion therapy in some countries are not routinely available. These challenges highlight the need for alternative cost-effective primary and secondary preventative strategies. Therefore, further research is needed to identify resourceful strategies in treating and monitoring patients with SCD in these countries.

Transitioning to adulthood

This is a common challenge all over the globe but may be more pronounced in developing countries. Despite a steady decline in mortality rates among children with SCD in developed countries, the mortality rates in adults with SCD continues to increase [47]. The most vulnerable age group at risk of morbidity and mortality in patients with SCD in developed countries is now shifted to the 18-30 year age group [7,48]. There are multiple factors that increase the risk of morbidity and mortality in patients transitioning to adulthood [47,48]. SCD is a chronic disease with a natural progressive history, if not prevented or treated, can lead to increased morbidity and mortality as patient's grow older and the disease progresses. In addition, patient-related factors such as compliance to therapy during teenage years and the psychosocial factors upon transitioning to adulthood also contribute to the vulnerability. Furthermore, resource and practice-related factors including the burden of the disease on resources, the limited adult SCD programs/centers, limited access to specialized multidisciplinary sickle cell care in adult facilities, and the limited research in prevention and management in adult patients with SCD. Therefore, as the care and survival of children with SCD improves globally the burden of care will shift to adulthood and this should be a focus of care at the global level.

Conclusions

Significant advances have occurred over the past 20 years in caring for patients with SCD. The advances in prevention and early intervention have lead to improved survival and quality of life of children with SCD. These interventions involved the institution of simple measures such as early newborn screening and early initiation of antimicrobial prophylaxis in addition to immunizations. Early monitoring and screening has allowed for targeted preventative interventions such as hydroxyurea and transfusion therapy. These comprehensive care approaches in addition to curative therapies such as HSCT have contributed to the success in care made thus far in SCD. It is now expected that children with SCD will survive well into adulthood. However, challenges are now shifting to the adult age group and global challenges continue to exist due to the limited resources and difficulties in access to care in less developed countries. Therefore, establishing newborn screening and comprehensive care programs that are cost-effective, increasing education, developing resource-specific standard of care guidelines to allow delivery of a structured approach to care, and improving access to care needs to be prioritized at the global level.

References

1. Habara A, Steinberg MH. Minireview: Genetic basis of heterogeneity and severity in sickle cell disease. Exp Biol Med (Maywood). 2016;241:689-96.
2. Gladwin MT, Vichinsky E. Pulmonary complications of sickle cell disease. N Engl J Med. 2008;359:2254-65.
3. Modell B, Darlison M. Global epidemiology of haemoglobin disorders and derived service indicators. Bull World Health Organ. 2008;86:480-7.
4. Serjeant GR. The Natural History of Sickle Cell Disease. Cold Spring Harbor Perspectives in Medicine 2013;3: a011783.
5. Colah RB, Mukherjee MB, Martin S, Ghosh K. Sickle cell disease in tribal populations in India. Indian Journal of Medical Research 2015;141: 509-515.
6. Jastaniah W. Epidemiology of sickle cell disease in Saudi Arabia. Ann Saudi Med. 2011 May-Jun; 31(3): 289-293.
7. Quinn CT, Rogers ZR, McCavit TL, Buchanan GR. Improved survival of children and adolescents with sickle cell disease. *Blood*. 2010;115(17):3447-3452.
8. Vichinsky E, Hurst D, Earles A, Kleman K, Lubin B. Newborn screening for sickle cell disease: effect on mortality. Pediatrics. 1988;81:749-55.
9. Gaston MH, Verter JI, Woods G, Pegelow C, Kelleher J, Presbury G, *et al.* Prophylaxis with oral penicillin in children with sickle cell anemia. A randomized trial. *N Engl J Med*. 1986;314:1593-9.
10. Falletta JM, Woods GM, Verter JI, Buchanan GR, Pegelow CH, Iyer RV, *et al.* Discontinuing penicillin prophylaxis in children with sickle cell anemia. Prophylactic Penicillin Study II. *J Pediatr*. 1995;127:685-90.
11. Ramakrishnan M, Moïsi JC, Klugman KP, Iglesias JMF, Grant LR, Mpoudi-Etame M, *et al.* Increased risk of invasive bacterial infections in African people with sickle-cell disease: a systematic review and meta-analysis. Lancet Infect Dis. 2010; 10:329-37.
12. Adams RJ, McKie VC, Hsu L, Files B, Vichinsky E, Pegelow C, Abboud M, Gallagher D, Kutlar A, Nichols FT, Bonds DR, Brambilla D. Prevention of a first stroke by transfusions in children with sickle cell anemia and abnormal results on transcranial Doppler ultrasonography. N Engl J Med. 1998;339:5-11.
13. Adams RJ, Brambilla D; Optimizing Primary Stroke Prevention in Sickle Cell Anemia (STOP 2) Trial Investigators. Discontinuing prophylactic transfusions used to prevent stroke in sickle cell disease. N Engl J Med. 2005;353:2769-78.
14. Kassim AA, Pruthi S, Day M, Rodeghier M, Gindville MC, Brodsky MA, DeBaun MR, Jordan LC. Silent cerebral infarcts and cerebral aneurysms are prevalent in adults with sickle cell anemia. Blood. 2016;127:2038-2040.
15. Ware R *et al.* Hydroxycarbamine *versus* chronic transfusion for maintenance of transcranial doppler flow velocities in children with sickle cell anaemia-TCD With Transfusion Changing to Hydroxyurea (TWiTCH): a multicneter, open-label, phase 3, non-inferiority trial. The Lancet 2016:387:661-670.
16. Hankins JS, McCarville MB, Rankine-Mullings A, Reid ME, Lobo CL, Moura PG, Ali S, Soares DP, Aldred K, Jay DW, Aygun B, Bennett J, Kang G, Goldsmith JC, Smeltzer MP, Boyett JM, Ware RE. Prevention of conversion to abnormal transcranial Doppler with hydroxyurea in sickle cell anemia: A Phase III international randomized clinical trial. Am J Hematol. 2015;90:1099-105.
17. Bernaudin F, *et al.* Long-term treatment follow-up of children with sickle cell disease monitored with abnormal transcranial Doppler velocities. Blood 2016;127: 1814-1822.
18. Platt OS. Prevention and management of stroke in sickle cell anemia. Hematology Am Soc Hematol Educ Program. 2006:54-57.
19. Powars D, Wilson B, Imbus C, Pegelow C, Allen J. The natural history of stroke in sickle cell disease. Am J Med. 1978;65: 461-71.
20. Pegelow CH, Adams RJ, McKie V, Abboud M, Berman B, Miller ST, Olivieri N, Vichinsky E, Wang W, Brambilla D. Risk of recurrent stroke in patients with sickle cell disease treated with erythrocyte transfusions. J Pediatr. 1995;126: 896-9.
21. Hulbert M L *et al.* Silent cerebral infarcts occur despite regular blood transfusion therapy after first strokes in children wit sickle cell disease. Blood 2011;117: 772-779.
22. Ware RE, Helms RW; SWiTCH Investigators. Stroke With Transfusions Changing to Hydroxyurea (SWiTCH). Blood 2012;119:3925-3932.
23. Kassim AA, Galadanci NA, Pruthi S, DeBaun MR. How I treat and manage strokes in sickle cell disease. Blood 2015;125:3401-3410.
24. DeBaun MR, Kirkham FJ. Central nervous system complications and management in sickle cell disease. Blood 2016;127: 829-838.
25. DeBaun MR, Gordon M, McKinstry RC, Noetzel MJ, White DA, Sarnaik SA, *et al.* Controlled trial of transfusions for silent cerebral infarcts in sickle cell anemia. N Engl J Med. 2014;371:699-710.
26. Voskaridou E, Christoulas D, Bilalis A, Plata E, Varvagiannis K, Stamatopoulos G, Sinopoulou K, Balassopoulou A, Loukopoulos D, Terpos E. The effect of prolonged administration of hydroxyurea on morbidity and mortality in adult patients with sickle cell syndromes: results of a 17-year, single-center trial (LaSHS). Blood. 2010;115:2354-63.
27. Charache S, Terrin ML, Moore RD, Dover GJ, Barton FB, Eckert SV, McMahon RP, Bonds DR. Effect of hydroxyurea on the frequency of painful crises in sickle cell anemia. Investigators of the Multicenter Study of Hydroxyurea in Sickle Cell Anemia. N Engl J Med. 1995;332: 1317-1322.
28. Ware RE. How I use hydroxyurea to treat young patients with sickle cell anemia. Blood. 2010;115: 5300-11.
29. Strouse JJ, Heeney MM. Hydroxyurea for the treatment of sickle cell disease: efficacy, barriers, toxicity, and management in children. Pediatr Blood Cancer 2012;59: 365-371.
30. Yawn BP, Buchanan GR, Afenyi-Annan AN, Ballas SK, Hassell KL, James AH, Jordan L, Lanzkron SM, Lottenberg R, Savage WJ, Tanabe PJ, Ware RE, Murad MH, Goldsmith JC, Ortiz E, Fulwood R, Horton A, John-Sowah J. Management of sickle cell disease: summary of the 2014 evidence-based report by expert panel members. JAMA. 2014;312:1033-48.
31. Standards for the clinical care of adults with sickle cell disease in the UK. Sickle Cell Society; 2008. Available from: http://sicklecellsociety.org/wp-content/uploads/2016/02/Standards-for-the-Clinical-Care-of-Adults-with-Sickle-Cell-Disease-in-the-UK.pdf
32. Vichinsky E, Bernaudin F, Forni GL, Gardner R, Hassell K, Heeney MM, Inusa B, Kutlar A, Lane P, Mathias L, Porter J, Tebbi C, Wilson F, Griffel L, Deng W, Giannone V, Coates T. Long-term safety and efficacy of deferasirox (Exjade) for up to 5 years in transfusional iron-overloaded patients with sickle cell disease. Br J Haematol. 2011;154:387-97.
33. Vichinsky E, Torres M, Glass J, *et al.* A Randomized Phase II Study Evaluating the Efficacy and Safety of Deferasirox *Versus* Deferoxamine in Patients with Sickle Cell Disease (SCD): 2-Year Results Including Pharmacokinetics (PK) and Safety of Deferasirox with Concomitant Hydroxyurea Therapy. Blood 2011 118: 1082.
34. Walters MC. Stem cell therapy for sickle cell disease: transplantation and gene therapy. Hematology Am Soc Hematol Educ Program. 2005:66-73.
35. Bernaudin F, Socie G, Kuentz M, Chevret S, Duval M, Bertrand Y, Vannier JP, Yakouben K, Thuret I, Bordigoni P, Fischer A, Lutz P, Stephan JL, Dhedin N, Plouvier E, Margueritte G, Bories D, Verlhac S, Esperou H, Coic L, Vernant JP, Gluckman E; SFGM-TC. Long-term results of related myeloablative stem-cell transplantation to cure sickle cell disease. Blood. 2007;110: 2749-56.
36. Walters MC, De Castro LM, Sullivan KM, Krishnamurti L, Kamani N, Bredeson C, Neuberg D, Hassell KL, Farnia S, Campbell A, Petersdorf E. Indications and Results of HLA-Identical Sibling Hematopoietic Cell Transplantation for Sickle Cell Disease. Biol Blood Marrow Transplant. 2016;22:207-11.
37. Grosse SD, Odame I, Atrash HK, Amendah DD, Piel FB, Williams TN. Sickle Cell Disease in Africa: A Neglected Cause of Early Childhood Mortality. American Journal of Preventive Medicine 2011;41: S398-405.
38. Tewari S, Rees D. Morbidity pattern of sickle cell disease in India: A single centre perspective. Indian Journal of Medical Research 2013;138: 288-290.
39. Piel FB, Hay SI, Gupta S, Weatherall DJ, Williams TN. Global Burden of Sickle Cell Anaemia in Children under Five, 2010-2050: Modelling Based on Demographics, Excess Mortality, and Interventions. PLOS 2013;10: e1001484.
40. Memish ZA and Saeedi MY. Six-year outcome of the national premarital screening and genetic counseling program for sickle cell disease and β-thalassemia in Saudi Arabia. Ann Saudi Med. 2011:31:229-235.
41. Mason K, Gibson F, Gardner R, Serjeant B, Serjeant GR. Prevention of sickle cell disease: observations on females with the sickle cell trait from the Manchester project, Jamaica. J Community Genet. 2016; 7: 127-132.
42. Serjeant GR. The geography of sickle cell disease: Opportunities for understanding its diversity. Ann Saudi Med. 1994;14: 237-46.
43. Alsultan A, Alabdulaali MK, Griffin PJ, Alsuliman AM, Ghabbour HA, Sebastiani

P, Albuali WH, Al-Ali AK, Chui DH, Steinberg MH. Sickle cell disease in Saudi Arabia: the phenotype in adults with the Arab-Indian haplotype is not benign. Br J Haematol. 2014;164:597-604.
44. Al-Suliman A, Elsarraf NA, Baqishi M, Homrany H, Bousbiah J, Farouk E. Patterns of mortality in adult sickle cell disease in the Al-Hasa region of Saudi Arabia. Ann Saudi Med. 2006;26:487-8.
45. Ansong D, Akoto AO, Ocloo D, Ohene-Frempong K. Sickle cell disease: management options and challenges in developing countries. Mediterr J Hematol Infect Dis. 2013;5:e2013062.
46. Alsultan A, Jastaniah W, Al Afghani S, Al Bagshi MH, Nasserullah Z, Al-Suliman AM, Alabdulaali MK. Demands and challenges for patients with sickle-cell disease requiring hematopoietic stem cell transplantation in Saudi Arabia. Pediatr Transplant. 2016;20:831-5.
47. Lanzkron S, Carroll CP, Haywood C Jr. Mortality rates and age at death from sickle cell disease: U.S., 1979-2005. Public Health Rep. 2013;128:110-6.
48. Brousseau DC, Owens PL, Mosso AL, Panepinto JA, Steiner CA. Acute care utilization and rehospitalizations for sickle cell disease. JAMA. 2010;303:1288-94.

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How I treat and monitor non-transfusion-dependent thalassaemia

Abdul-Hamid A. Bazarbachi,[1] Hassan M. Moukhadder,[2] Rayan I. Bou Fakhredin,[2] Joseph E. Roumi,[2] Bachar F. Chaya,[1] Ali T. Taher[2]¶

[1]Faculty of Medicine, American University of Beirut Medical Center, Beirut, Lebanon; [2]Division of Hematology/Oncology, Department of Internal Medicine, American University of Beirut Medical Center, Beirut, Lebanon

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ABSTRACT

Non-transfusion-dependent thalassemia (NTDT) is a term that has been conceived to describe a group of thalassemia syndromes that fall in the gray zone of disease severity between thalassemia trait and transfusion-dependent thalassemia (TDT). Although patients with NTDT do not require regular blood transfusions for survival, the natural history of this disease entity is often complicated by a host of morbidities directly or indirectly attributable to ineffective erythropoiesis, chronic hemolytic anemia, and iron overload. Due to the seriousness of these complications, timely initiation of adequate treatment strategies is key in managing NTDT patients, while emphasizing the personalization of therapeutic plans in regards to the individual patient's symptoms, thalassemia-related and non-thalassemia-related morbidities, disease progression, and quality of life. Several strategies are at the disposal of treating clinicians whereby the mainstay of medical care includes iron chelation therapy and fetal hemoglobin induction, in addition to transfusions in the context of specific morbidities only and splenectomy in select cases. Promising treatment modalities that are currently on the rise include Janus kinase 2 (JAK2) inhibitors, hepcidin mimetics, apo-transferrin therapy, activin receptor chimeric proteins, stem cell transplantation, and gene therapy. Our ever-evolving understanding of the molecular pathophysiology of NTDT is the bedrock of uncovering yet other therapeutic targets in the NTDT realm in the future.

Correspondence:

ataher@aub.edu.lb

Introduction

Thalassemia is a group of autosomal recessive disorders of hemoglobin that includes a myriad of phenotypes stemming from defective α- and/or β-globin chain synthesis (α-thalassemia and/or β-thalassemia, respectively).[1] Transfusion dependence is the single most important determining factor in classifying the thalassemia syndromes according to clinical severity, spanning patients who are asymptomatic carriers (α- or β-thalassemia minor) to those who have transfusion-dependent thalassemia (TDT), which encompasses β-thalassemia major (TM), Bart's hemoglobin, and severe forms of HbE/β-thalassemia. A group of non-transfusion-dependent thalassemia (NTDT) patients exists in between two extreme ends. Patients in this group, which includes individuals with β-thalassemia intermedia (TI), HbH disease, and mild and moderate forms of HbE/ β-thalassemia, are not transfusion-dependent but may require transfusions during periods of stress.[2]

Epidemiology

Thalassemias are especially prevalent in low- to middle-income countries of the tropical belt, partly due to the high prevalence of consanguineous marriages in these regions. Approximately 80% of thalassemia cases worldwide are observed in the area extending from sub-Saharan Africa to the Mediterranean basin, the Middle East, and South and Southeast Asia. However, thalassemia has recently

evolved as an "emerging minority disease" in the West, in light of the increasing migration patterns in the past two decades from regions historically associated with thalassemia toward North America and Europe. α-Thalassemia remains to be the most common thalassemia, with a 5% worldwide carrier rate, in contradistinction to β-thalassemia which has a global carrier incidence of 1.5%. It remains important to stress that an accurate epidemiological registry on the incidence and prevalence of thalassemia is lacking, so any presented numbers in this regard ought to be interpreted with caution as they are not necessarily reflective of the true burden of the disease.[1]

Pathophysiology

The pathophysiology of the NTDT syndromes, similar to that of the other thalassemias, is believed to stem from hemoglobin chain imbalance and secondary oxidative damage.[3] The complications associated with NTDT are clustered under the classic triad of ineffective erythropoiesis (and resultant extramedullary hematopoiesis), chronic anemia and hemolysis, and iron overload (Figure 1).[4] In fact, chronic hemolytic anemia and related hypoxia lead to a compensatory increase in erythropoietin levels and decrease in serum hepcidin, which synergistically contribute to a state of increased iron burden.[3,5] Iron balance, in general, depends on intake and losses, and iron overload results from an overly positive iron balance. Hepcidin, which is the main regulator of iron balance, decreases iron absorption from the gut and iron release from the reticuloendothelial system.[6] In NTDT, where hepcidin levels are abnormally low, excess iron is absorbed into the system and thereafter released into the circulation, depleting macrophage iron and leading to preferential portal and hepatocyte iron loading.[7] This culminates in an inevitable increase in free iron in the bloodstream, which results in end-organ damage. Growth differentiating factor-15 (GDF-15), a member of the transforming growth factor-β (TGF-β) family which is increased with cellular stress, is normally inhibited by effective erythropoiesis.[8] It is believed that in light of ineffective erythropoiesis at the heart of NTDT progression, GDF-15 is disinhibited, thus adding to the iron overload in these patients by decreasing the levels of hepcidin in the circulation.[8] In fact, GDF-15 levels were found to have a positive linear correlation with the morbidities classically associated with NTDT, suggesting the role of this molecular player in NTDT pathogenesis.[9] On the other hand, the main culprit of iron overload in TDT is chronic transfusional iron accumulation, which contributes to a much lesser extent to the development of iron overload in NTDT.[10]

Complications

NTDT is associated with high morbidity rates as suggested by the results of the OPTIMAL CARE study, with an age-related increase in the severity of the NTDT-associated complications, manifesting as early as the age of 10 (Figure 2).[11] In fact, chronic anemia was found to be independently associated with morbidity in NTDT, where all patients with a hemoglobin level below 7 g/dL suffered from thalassemia-related complications.[12]

I. Hematological and cardiovascular

A. Hypercoagulable State

Thalassemia-attributable thromboembolic complications are 4.38 times more common in TI than in TM.[13] This multifactorial complication is due to the additive effects of the procoagulant activity of hemolyzed red blood cells (RBCs) and the concurrently increased activa-

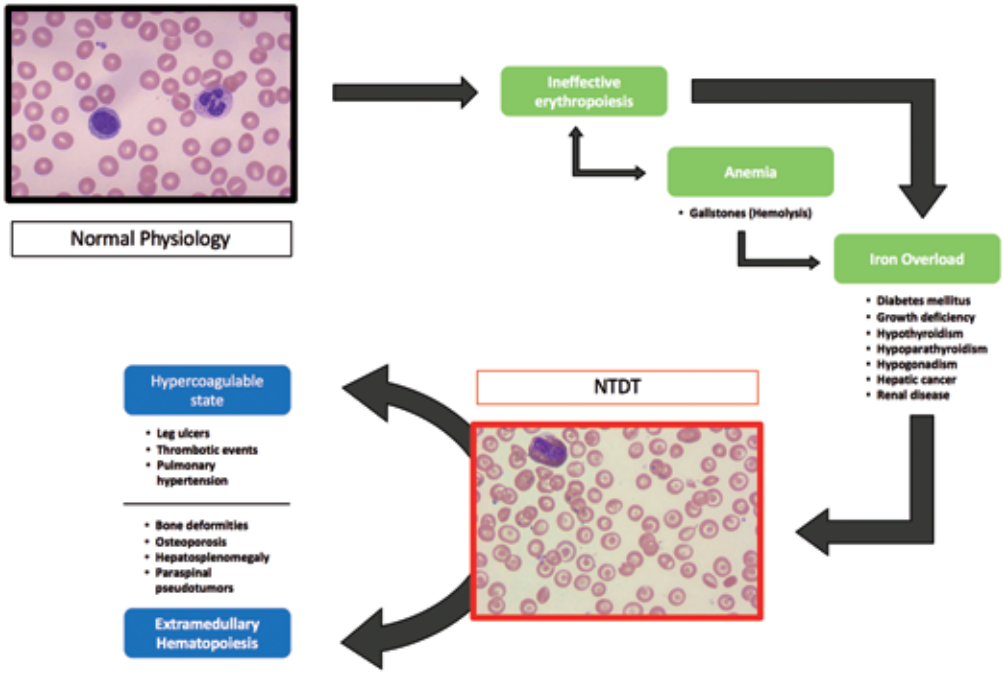

Figure 1. Pathophysiological pillars of non-transfusion-dependent thalassemia and their associated complications.
NTDT: non-transfusion-dependent thalassemia.

tion of circulating platelets, in addition to coagulation factor defects, antithrombotic factor depletion, and endothelial injury secondary to iron toxicity.[2] Furthermore, the blood transfusions NTDT patients might require can lead to the production of RBCs with an abnormally increased content of cell surface phosphatidylserine, which is thought to enhance hypercoagulability by rendering the plasma membrane negatively charged, rigid, and more prone to aggregation.[2] Splenectomy also contributes to an added risk in the occurrence of thromboembolic complications in this patient population, considering the function of the normal spleen in sequestering one-third of the total platelet burden. It was determined in a study by Taher *et al.* that splenectomized TI patients who experienced thromboembolic events secondary to a hypercoagulable state had characteristically high counts of nucleated RBCs (≥300×10^6/L) and platelets (≥500×10^9/L).[14] Nucleated RBCs, in particular, express adhesion molecules which are speculated to further contribute to the hypercoagulable state pervasive among patients with thalassemia.[15]

B. Pulmonary Hypertension

Pulmonary hypertension (PHTN) is 5 times more prevalent in NTDT than in TM.[16] The diagnosis of PHTN is established when tricuspid valve regurgitant jet velocity exceeds 2.5-2.8 m/s, the equivalent of a pulmonary arterial systolic pressure of 30-35 mmHg.[1] The physiological association between thalassemia and PHTN is still not very clear, but it is hypothesized that excessive hemolysis along with nitric oxide depletion and enhanced platelet activation are at the very core of the vasculopathy contributing to the development of PHTN in thalassemia patients, with a possible role for nucleated red blood cells in the progression of this complication.[17-19] Interestingly, transfusions were observed to reduce the incidence of PHTN in patients with TI, but a more extensive risk-to-benefit evaluation of this intervention's utility is yet to be conducted.[11]

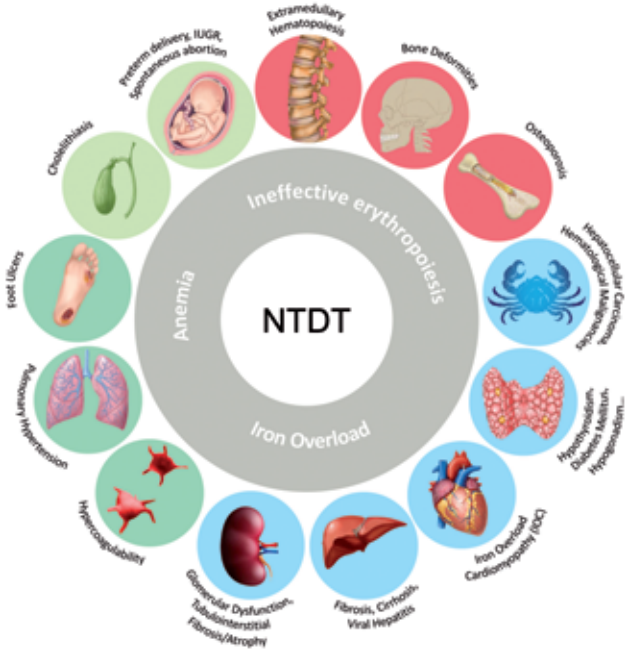

Figure 2. Complications associated with non-transfusion-dependent thalassemia. IUGR: intrauterine growth restriction; NTDT: non-transfusion-dependent thalassemia.

C. Iron overload cardiomyopathy

Iron-induced injury to cardiac myocytes causes dilatation of the left ventricular chamber and subsequent decrease of left ventricular ejection fraction, resulting in a subtype of dilated cardiomyopathy called iron overload cardiomyopathy (IOC). However, this complication is classically associated with TDT, and despite some evidence pointing towards a similar phenomenon in NTDT, the occurrence of this entity in the non-transfusion-dependent subpopulation of thalassemics remains to be speculative.[20]

D. Extramedullary hematopoietic pseudotumors

Characteristic ineffective erythropoiesis with ensuing bone marrow insufficiency is the driving force for extramedullary hematopoiesis in patients with thalassemia. This compensatory mechanism, which is physiological in fetal organs during gestation, is especially problematic in thalassemia patients as it can occur anywhere in the body.[6] It most commonly involves the liver and spleen, hence the hepatosplenomegaly frequently associated with thalassemia. While usually causing mild compression symptoms, some extramedullary hematopoietic lesions are present as pseudotumors that can cause a multitude of neurological symptoms due to spinal compression.[4] To note, extramedullary haematopoiesis is by far more commonly associated with NTDT than TDT (20% vs 1%, respectively), which is expected considering the chronic hemolytic anemia and absence of regular transfusions in the NTDT subtype.[4]

E. Leg ulcers

Reduced tissue oxygenation secondary to the combination of anemia, hypercoagulability, and ineffective erythropoiesis leads to tissue fragility and eventual trauma-induced ulceration.[1] This risk increases with age and can affect up to one-third of poorly managed NTDT patients.[21,22]

II. Hepatobiliary

Hemolysis in NTDT leads to the formation of pigmented gallstones which greatly increase the risk of complicated cholecystitis, a potentially fatal morbidity in splenectomized patients.[21] Furthermore, iron accumulation in the liver parenchyma can result in liver damage which might progress to fibrosis and eventually cirrhosis, thereby greatly increasing the risk of development of hepatocellular carcinoma (HCC).[23] Transfusion-transmitted viral hepatitis is another concern in these patients, further adding to their risk of progression to hepatic carcinogenesis.[23]

III. Endocrine and bone disease

Although more common in TDT, endocrine gland dysfunction resulting from iron toxicity is fairly prevalent in NTDT.[2] While skeletal deformities and growth delay are more commonly encountered in TDT patients, NTDT is more frequently complicated by hypothyroidism, hypoparathyroidism, adrenal insufficiency, diabetes mellitus, and hypogonadism, yet still rare overall.[11,24] Although pubertal delay is not uncommon in patients with NTDT, these patients usually have normal sexual development and are generally fertile.[25] Interestingly, iron chelation was found to be the single most important intervention in preventing endocrinopathy in NTDT patients.[11]

IV. Renal complications

Persistent hypoxia, anemia, and severe iron overload in NTDT result in both tubulointerstitial and glomerular

dysfunction manifested as glomerular hyperfiltration and proteinuria.[26,27] In addition, iron-chelating drugs might be associated with renal toxicity, hence predisposing patients to an increased risk of renal disease.[27]

V. Pregnancy-related complications

NTDT is associated with a host of complications in pregnancy, ranging from preterm delivery to intrauterine growth restriction (IUGR) and spontaneous abortion.[28] In a case series by Nassar *et al.*, up to 57.1% of pregnancies in NTDT females were found to be complicated by IUGR.[29] Data from non-thalassemic cohorts suggests that hemoglobin levels above 10 g/dL during gestation are recommended for optimal fetal growth and preclusion of preterm delivery.[30] However, targeting the 10 g/dL cutoff proved to be of clinical benefit to only 78% of the pregnant NTDT patients and their fetuses in an Italian case series, where the fetuses of the other 22% suffered from IUGR.[28] This suggests that, apart from the absolute hemoglobin concentration, transfusion therapy in pregnant women with NTDT should be tailored to the cardiac function and general condition of the mother and the growth status of the fetus.[28] However, routine transfusions are not recommended in pregnant NTDT females because blood transfusions can increase the risk of developing alloiummune antibodies that would exacerbate any preexisting hemolytic anemia, such as thalassemia.

Another pregnancy-related morbidity in NTDT patients is the possible limitation of uterine enlargement by hepatosplenomegaly, sometimes requiring splenectomy during gestation or postpartum.[29]

VI. Malignancies

Thalassemia is associated with an increased risk of developing several malignancies, most commonly HCC and hematological malignancies.[31] Interestingly, HCC seems to be more common in patients with NTDT than TDT, possibly because NTDT patients usually have improved survival compared with patients who have TDT, which enables them to live long enough to develop HCC.[23] Iron overload, transfusion-transmitted viruses, and transfusion-related immunomodulation are suggested to be incriminated in the pathophysiology of neoplastic transformation in thalassemia patients.[31]

Management

Table 1 summarizes the treatment modalities applied to the major NTDT-associated complications.

I. Splenectomy

Splenomegaly is a common sequel in NTDT patients as a result of the additive effect of extramedullary hematopoiesis and chronic passive splenic congestion.[2] The spleen is normally responsible for the sequestration of one-third of the platelets that are produced by the bone marrow and the removal of abnormal circulating RBCs. Therefore, splenic dysfunction in NTDT patients manifests in ways that range from worsening of anemia to neutropenia and thrombocytopenia (and their associated complications of bacterial infections and bleeding, respectively).[2,32] If extensive, splenomegaly can cause significant left upper quadrant pain and imminent splenic rupture.[2,32] Splenectomy, which aims at ameliorating these adverse occurrences, leads to a 1-2 g/dL increase in the total hemoglobin concentration.[2,32] Splenectomy, however, is not without complications. The OPTIMAL CARE study elucidated a high rate of long-term morbidity in relation to splenectomy, mostly attributable to ensuing hypercoagulability and increased susceptibility to infection with encapsulated bacteria.[11] More specifically, splenectomy was found to be associated with an increased incidence of venous thromboembolic events, silent cerebral infarcts, PHTN, cholelithiasis, hypothyroidism, osteoporosis, and leg ulcers, in addition to fatal overwhelming post-spelenctomy sepsis and recurrent infections.[11]

Therefore, splenectomy is reserved to select NTDT patients in whom the benefits of the intervention out-

Table 1. Morbidity-directed treatment in NTDT.

Complication	Management
Hyper-coagulable state	Aspirin[68]
Pulmonary hypertension	Phosphodiesterase-5 inhibitors Endothelin receptor antagonists Prostacyclin analogues
Iron overload cardiomyopathy	Iron chelation therapy
Extramedullary hematopoietic pseudotumors	Fetal hemoglobin inducers Novel agents (see Table 2)
Leg ulcers	Routine skin inspection[69] Keeping the legs and feet raised above the level of the heart for 1-2 hours per day[69] Occlusive dressing[69] Topical antibiotic[69]
Liver cirrhosis	Liver function and hepatitis serology testing in patients on chronic transfusion[70] MRI-based LIC measurement Biannual liver ultrasound in high-risk patients[23]
Cholelithiasis	Cholecystectomy if symptomatic
Endocrine and bone	Iron chelation therapy
Renal complications	Renal function monitoring Avoidance of nephrotoxic drugs (expert opinion)
Pregnancy-related complications	Keeping hemoglobin levels above threshold ensuring adequate fetal growth and maternal well being Splenectomy in case of splenomegaly limiting uterine enlargement

LIC: liver iron concentration; MRI: magnetic resonance imaging; NTDT: non-transfusion-dependent thalassemia.

weigh its potential complications. Examples of such situations include:[1]

- Hypersplenism resulting in symptomatic anemia, thrombocytopenia causing hemorrhages, and/or leukopenia causing recurrent bacterial infections.
- Early satiety due to gastric displacement by the spleen, or a palpable left upper quadrant abdominal mass (>20 cm in the largest diameter) that may or may not be painful but is at risk of rupture.
- Unavailability or contraindication of transfusion therapy and/or iron-chelating medicines.
- Poor growth or failure to thrive.

II. Transfusion therapy

Although NTDT patients are by definition transfusion-independent, they seldom require blood transfusions in certain clinical scenarios. The hemoglobin level per se is not an indication for blood transfusion, and transfusion therapy in patients with NTDT is guided by clinical necessity.[33] Transfusions in this subpopulation can be divided into occasional in acute settings, frequent for prolonged defined durations, and preventive in high-risk individuals.[1,32]

Occasional transfusions are warranted in serious infections, in surgical settings in anticipation of acute blood loss, and in pregnancy to decrease the burden of anemia of NTDT already accentuated by the physiologic anemia of pregnancy.[1,32]

Frequent transfusions, although not as frequent as in TM, are given for defined durations and are only indicated for children with poor growth and adults being treated for (or to prevent) specific complications.[1,32]

Preventive transfusions are given for high-risk patients, such as those at risk of thrombotic events or cerebrovascular disease, PHTN, extramedullary hematopoiesis (particularly paraspinal pseudotumors), cholelithiasis, and leg ulcers, where transfusions were shown to greatly improve the prognosis of these complications.[11,32] Conversely, blood transfusions were observed to increase the risk of endocrine disease, especially hypogonadism and osteoporosis, and worsen the overly positive iron balance, limiting their use to situations where they are absolutely indicated.[11] Another potential complication of blood transfusions is alloimmunization, which is more commonly observed in splenectomized patients, minimally or newly transfused patients of old age, and patients receiving transfusates harboring allogeneic white blood cells.[34] This complication, however, can be avoided by using matched leukoreduced blood and supplementing erythropoietin, iron, and folic acid.[34]

III. Pharmacological

A. Iron chelation therapy

As previously discussed, iron overload is associated with a host of complications in patients with NTDT, which calls for closely monitoring these patients for increased iron burden. Liver iron overload, in particular, has been found to proportionally correlate with an increased incidence of several morbities in both TDT and NTDT.[35,36] Since liver iron concentration (LIC) and total body iron are linearly related,[37] yearly non-invasive LIC quantification with R2 or R2* MRI is currently the cardinal test for estimation of total body iron in all thalassemia patients. Yet, the serum ferritin assay, which is an easy and inexpessive method compared to LIC measurement, remains to be heavily relied on in resource-poor areas where MRI technology is not available.[38] Although this marker is reflective of iron stores in patients with TDT,[39] serum ferritin level underestimates the iron burden in NTDT,[40] which can be explained by the fact that hyperabsorbed iron in NTDT is accumulated in hepatocytes, leading to usually lower serum ferritin levels.[40,41]

Iron chelation therapy in NTDT is only indicated if iron concentrations reach levels associated with increased iron-related complications, which are LIC ≥5 mg Fe/g dry weight or serum ferritin level ≥800 ng/mL in patients older than 10 years (or 15 years in hemoglobin H disease) when MRI technology is not available.[42] Iron-chelating drugs include deferoxamine, which is given parenterally, and deferiprone and deferasirox, which are administered orally.[43] In TDT, both deferoxamine and deferasirox are first line in patients >2 years old.[1] Deferiprone is not licensed for use in children younger than 6 years, and it is indicated in children >6 years old and adults only if other chelators are not tolerated or ineffective.[1] In NTDT, however, deferasirox remains to be the only chelator to have received Food and Drug Administration (FDA) and European Medicines Agency (EMA) approval based on findings from the THALASSA Trial.[2,44] One-year deferasirox treatment in NTDT patients older than 10 years was found to decrease LIC by a mean of 2.33±0.70 and 4.18±0.69 mg Fe/g dry weight at a daily dose of 5 mg/kg and 10 mg/kg, respectively, compared to placebo.[44] The overall incidence of adverse effects was comparable between the deferasirox and placebo arms, and the main side effects were nausea, gastrointestinal discomfort, and headache. These side effects, which were mild to moderate in severity, resolved spontaneously without discontinuation of the drug. The THETIS study, a phase IV, multicenter efficacy and safety study of deferasirox targeting a larger population of patients with NTDT, showed at 1-year analysis of the results that deferasirox is effective in reducing iron overload in NTDT at a starting dose of 10 mg/kg/day, with dose escalations starting at week 4 up to 30 mg/kg/day according to the LIC response.[45] In addition, the THESIS study provided more evidence about the satisfactory safety profile of the drug and reported on cases of pancreatitis and ocular toxicity possibly related to treatment with deferasirox.[45]

B. Fetal hemoglobin inducers

Increasing γ-globin chain synthesis in β-thalassemia decreases the level of free α-chains, improves both α/β-chain imbalance and erythropoiesis, and leads to the production of fetal hemoglobin (HbF).[46] Low levels of HbF in patients with the same thalassemia mutation are associated with a higher incidence of TM.[47] The extent to which HbF contributes to the clinical variation seen in NTDT was not known before the work of Musallam *et al.* in 2012, where it was demonstrated that higher blood levels of HbF are associated with a milder disease course in patients with NTDT.[48]

Among the HbF inducers, 5-azacytidine, a DNA methylation inhibitor, was found to be associated with a marked hematologic response.[46,49] However, its unfavorable safety profile has limited its use.[42] Another drug in this family is decitabine, which was shown to increase the hemoglobin level by an average of 1 g/dL.[50] The safety profile of this medicine is still under investigation, however, especially that the studies investigating its utility were done on small patient populations.[51] On the other hand, hydroxyurea, an antineoplastic cytotoxic agent commonly employed as an HbF inducer in sickle cell disease, has been used in NTDT where it is considered to be clinically effective and have satisfactory long-term safety.[52] However only small non-randomized control trials have been done to date on the effectiveness of this drug.[42] Studies have demonstrated that hydroxyurea increases γ-

chain mRNA expression by up to 9 folds in patients with β-thalassemia.[53] Importantly, hydroxyurea was shown to decrease complications such as leg ulcers, and extramedullary hematopoietic tumors and improve quality-of-life measure in patients with NTDT.[11]

The amino acid butyrate was found to be associated with increased levels of HbF in NTDT patients, with good tolerability and minimal side effects overall.[49] Other HbF-stimulating agents that are used in hydroxyurea-refractory patients with NTDT include thalidomide, which achieved a decent hematological response and was well tolerated in the limited studies done on its utilization for HbF induction in this patient population.[54]

C. Novel therapeutics

Table 2 outlines the most prominent novel agents in the thalassemia realm.

1. Janus Kinase 2 (JAK2) inhibitors

JAK 2 is a non-receptor tyrosine kinase that, when phosphorylated, plays a role in the growth and differentiation of erythroid progenitor cells in response to erythropoietin.[2] Thalassemia patients were shown to demonstrate increased expression of phosphorylated JAK2, leading to excessive proliferation and decreased differentiation of erythroid progenitors.[25,55] Thus, JAK2 inhibitors might be effective in thalassemia.[6] Ruxolitinib, a JAK2 kinase inhibitor already used in myeloproliferative diseases characterized by aberrant JAK2 kinase activation such as myelofibrosis and polycythemia vera, was shown to decrease the spleen size in murine models with thalassemia.[56] The TRUTH study, a phase IIa study inspired by this preclinical observation, elucidated up to a 26.8% decrease of spleen volume during the 30-week study period, in addition to slightly improved pre-transfusion hemoglobin levels, with a benign safety profile overall.[56] Although all study subjects had TM, the results of this clinical trial might be promising for NTDT patients with enlarged spleens.

2. Hepcidin mimetics

As discussed above, low hepcidin levels are among the most significant contributors to iron overload in NTDT.[57] Minihepcidins are hepcidin analogues that increase the levels of hepcidin, therefore decreasing iron absorption from the gastrointestinal tract, increasing the redistribution of iron to macrophages, and limiting end-organ toxicity.[58] In addition, experiments on mice revealed that minihepcidin therapy increases hemoglobin concentrations, decreases reticulocyte counts, and reduces spleen size.[58,59]

3. Apo-transferrin therapy

Transferrin, the body's main iron transporter, delivers iron to different tissues by receptor-mediated endocytosis.[10] The low hepcidin state in NTDT causes saturation of blood transferrin with circulating iron, resulting in the accumulation of toxic non-transferrin bound iron.[46] Transferrin mainly circulates in the blood in three major forms depending on iron levels: monoferric transferrin, dimeric transferrin, and apo-transferrin.[60] In experiments on thalassemic mice, daily apo-transferrin injections increased hemoglobin levels, decreased apoptosis of erythroid precursors and improved their maturation, and decreased the size of the spleen.[60] These findings have promising clinical implications in NTDT patients.[46]

4. Activin receptor fusion proteins

Sotatercept (ACE-011) is an activin type IIA receptor (ActRIIA) fusion protein that blocks the activity of numerous TGF-β cytokines.[61] It acts mainly on late-stage erythropoiesis leading to increased hemoglobin production. Phase I clinical data have confirmed that sotatercept therapy increases RBC counts and hemoglobin concentrations.[62] In NTDT murine models, sotatercept therapy caused a decrease in ineffective erythropoiesis and bilirubin levels and markedly improved anemia.[61] A phase IIa study by Porter *et al.* concluded that the subcutaneous administration of ACE-011 every three weeks might improve anemia in NTDT patients with a good safety profile.[61] Interim results from this study showed exposure- (or dose-) related mean hemoglobin increases for NTDT patients and reduced transfusion burden for TDT patients, with a favorable safety profile in general.[63] Other fusion proteins of the same family include luspatercept (ACE-536), which is currently undergoing extensive research, especially in TDT.[51]

5. Stem cell transplantation

Stem cell transplantation remains to be the only curative therapy in thalassemia, both TDT and NTDT.[64] A ret-

Table 2. Novel therapeutic modalities in NTDT treatment.

Modality	Mechanism of action	Treatment outcomes
JAK2 inhibitors	Inhibition of JAK2, a signaling molecule that regulates erythropoietin-mediated proliferation and differentiation of erythrocyte progenitors	Reduction of splenomegaly and avoidance of splenectomy (and associated sequelae) due to ineffective erythropoiesis
Hepcidin mimetics (*e.g.*, minihepcidins)	Behaving as agonists at the hepcidin receptor, therefore regulating iron homeostasis	Limitation of intestinal absorption of iron, increase of iron sequestration by macrophages, and suppression of ineffective and extramedullary hematopoiesis
Apo-transferrin therapy	Transporting iron between sites of uptake, storage, and utilization	Improvement of anemia and ineffective erythropoiesis (and corresponding reversal of splenomegaly), reduction of iron overload, and increase of hepcidin expression
ActRIIB/IgG1 Fc recombinant protein	Trapping of TGF-β superfamily ligands and inhibition of Smad2/3 signaling	Treatment of anemia due to ineffective erythropoiesis through promotion of red blood cell precursor differentiation
Molecules targeting BCL11A, MYB, KLF1	Induction of fetal hemoglobin through inactivation of gene products or interference with epigenetic mechanisms, ultimately improving the α/β globin chain imbalance	Improvement of anemia and possibly other NTDT complications

NTDT: non-transfusion-dependent thalassemia.

rospective study on the natural history of thalassemia patients who received allogeneic hematopoietic stem cell transplantation (HSCT) showed a 2-year overall survival rate of 88±1% and a 2-year event-free survival incidence of 81±1%.[64] Patients who received a transplant from an HLA-identical sibling had the best results, with 2-year overall and event-free survival rates of 91±1% and 83±1%, respectively. However, these data were from patients with TDT, and the literature remains to be lacking in reports of HSCT in NTDT.

6. Gene therapy

Recent studies described long-term correction of murine models of human β-thalassemia and sickle cell anemia by lentivirus-mediated gene transfer.[65,66] Evidence of high gene transfer and expression in transduced hematopoietic cells in humans has also been noted. Although promising, this therapeutic modality was applied on knock-out models of thalassemia that might not necessarily reflect the phenotypic variation of the disease in humans.[66] No solid data from systematic studies on gene therapy in human thalassemia patients are available to date.

Conclusions

The pathophysiology of NTDT and its complications can be attributed to the well-known triad of ineffective hematopoiesis, chronic anemia, and iron overload.[57] While previously considered a disorder of the developing world, thalassemia is increasingly establishing itself in the Western world as an epidemiologically expanding disease that one might speak of a "thalassemia pandemic" in the next few years.

Classically, thalassemia patients have been grossly categorized into TDT and NTDT and have received treatment accordingly. Because quite often the fine line between TDT and NTDT might be blurry, a disease severity scoring system specific for NTDT could be of great clinical utility, directly influencing the management schemes of this subpopulation of thalassemia patients, enhancing their survival, and bettering their quality of life. The scoring system developed by Cappellini *et al.* is one example that yet awaits validation.[67] Such scoring systems can help draw an association between the ascribed score and elements of treatment, allowing for individualized management plans tailored to the individual patient's clinical status. Finally, it remains important to underline that multicenter studies investigating into the clinical utility of the novel drugs discussed above is an absolute need, provided the very promising preclinical and early-phase clinical data on these medicines/therapies.

References

1. Taher A, Vichinsky E, Musallam K, Cappellini MD, Viprakasit V. Guidelines for the management of non transfusion dependent thalassaemia (NTDT): Thalassaemia International Federation, Nicosia, Cyprus; 2013.
2. Musallam KM, Rivella S, Vichinsky E, Rachmilewitz EA. Non-transfusion-dependent thalassemias. haematologica 2013;98: 833-44.
3. Olivieri NF. The β-thalassemias. New England journal of medicine 1999;341:99-109.
4. Haddad A, Tyan P, Radwan A, Mallat N, Taher A. β-Thalassemia Intermedia: A Bird's-Eye View. Turkish Journal of Hematology 2014;31.
5. Musallam KM, Cappellini MD, Taher AT. Iron overload in beta-thalassemia intermedia: an emerging concern. Curr Opin Hematol 2013;20:187-92.
6. Rivella S. The role of ineffective erythropoiesis in non-transfusion-dependent thalassemia. Blood reviews 2012;26:S12-S5.
7. Nemeth E. Hepcidin in β thalassemia. Annals of the New York Academy of Sciences 2010;1202:31-5.
8. Tanno T, Noel P, Miller JL. Growth differentiation factor 15 in erythroid health and disease. Curr Opin Hematol 2010;17:184-90.
9. Musallam KM, Taher AT, Duca L, Cesaretti C, Halawi R, Cappellini MD. Levels of growth differentiation factor-15 are high and correlate with clinical severity in transfusion-independent patients with beta thalassemia intermedia. Blood Cells Mol Dis 2011;47:232-4.
10. Ginzburg Y, Rivella S. β-thalassemia: a model for elucidating the dynamic regulation of ineffective erythropoiesis and iron metabolism. Blood 2011;118:4321-30.
11. Taher AT, Musallam KM, Karimi M, et al. Overview on practices in thalassemia intermedia management aiming for lowering complication rates across a region of endemicity: the OPTIMAL CARE study. Blood 2010;115:1886-92.
12. Taher AT, Musallam KM, Saliba AN, Graziadei G, Cappellini MD. Hemoglobin level and morbidity in non-transfusion-dependent thalassemia. Blood cells, molecules & diseases 2015;55:108.
13. Taher A, Isma'eel H, Mehio G, et al. Prevalence of thromboembolic events among 8,860 patients with thalassaemia major and intermedia in the Mediterranean area and Iran. Thromb Haemost 2006;96: 488-91.
14. Taher AT, Musallam KM, Karimi M, et al. Splenectomy and thrombosis: the case of thalassemia intermedia. J Thromb Haemost 2010;8:2152-8.
15. Garozzo G, Distefano R, Miceli A, Bonomo P. Detection of ICAM-1, ICAM-2, ICAM-3, PECAM-1 and VCAM-1, evaluation of hypercoagulable state and platelet aggregation in hemoglobinopathy patients with erythroblasts. Haematologica 2001; 86:778-9.
16. Derchi G, Galanello R, Bina P, et al. Prevalence and risk factors for pulmonary arterial hypertension in a large group of beta-thalassemia patients using right heart catheterization: a Webthal study. Circulation 2014;129:338-45.
17. Gladwin MT, Lancaster JR, Jr., Freeman BA, Schechter AN. Nitric oxide's reactions with hemoglobin: a view through the SNO-storm. Nat Med 2003;9:496-500.
18. Gladwin MT, Vichinsky E. Pulmonary complications of sickle cell disease. N Engl J Med 2008;359:2254-65.
19. Phrommintikul A, Sukonthasarn A, Kanjanavanit R, Nawarawong W. Splenectomy: a strong risk factor for pulmonary hypertension in patients with thalassaemia. Heart 2006;92:1467-72.
20. Pennell DJ PJ, Piga A, Lai Y, El-Beshlawy A, Belhoul KM, Elalfy M, Yesilipek A, Kilinç Y, Lawniczek T, Habr D, Weisskopf M, Zhang Y, Aydinok Y; CORDELIA study investigators. A 1-year randomized controlled trial of deferasirox vs deferoxamine for myocardial iron removal in β-thalassemia major (CORDELIA). Blood 2014; 123:1447-54.
21. Musallam KM, Taher AT, Rachmilewitz EA. beta-thalassemia intermedia: a clinical perspective. Cold Spring Harb Perspect Med 2012;2:a013482.
22. Olivieri NF, Muraca GM, O'Donnell A, Premawardhena A, Fisher C, Weatherall DJ. Studies in haemoglobin E beta-thalassaemia. Br J Haematol 2008;141:388-97.
23. Moukhadder HM, Halawi R, Cappellini MD, Taher AT. Hepatocellular carcinoma as an emerging morbidity in the thalassemia syndromes: A comprehensive review. Cancer 2016.
24. Kurtoglu AU, Kurtoglu E, Temizkan AK. Effect of iron overload on endocrinopathies in patients with beta-thalassaemia major and intermedia. Endokrynol Pol 2012;63: 260-3.
25. Taher A, Isma'eel H, Cappellini MD. Thalassemia intermedia: revisited. Blood Cells Mol Dis 2006;37:12-20.
26. Quinn CT, Johnson VL, Kim HY, et al. Renal dysfunction in patients with thalassaemia. Br J Haematol 2011;153:111-7.
27. Mallat NS, Mallat SG, Musallam KM, Taher AT. Potential mechanisms for renal damage in beta-thalassemia. J Nephrol 2013;26:821-8.
28. Origa R, Piga A, Quarta G, et al. Pregnancy and beta-thalassemia: an Italian multicenter experience. Haematologica 2010;95: 376-81.
29. Nassar AH, Usta IM, Rechdan JB, Koussa S, Inati A, Taher AT. Pregnancy in patients with beta-thalassemia intermedia: outcome of mothers and newborns. Am J Hematol 2006;81:499-502.
30. Levy A, Fraser D, Katz M, Mazor M, Sheiner E. Maternal anemia during pregnancy is an independent risk factor for low birthweight and preterm delivery. European Journal of Obstetrics & Gynecology and Reproductive Biology 2005;122:182-6.
31. Zanella S, Garani MC, Borgna Pignatti C. Malignancies and thalassemia: a review of the literature. Annals of the New York Academy of Sciences 2016.
32. Taher AT, Musallam KM, Cappellini MD, Weatherall DJ. Optimal management of

beta thalassaemia intermedia. Br J Haematol 2011;152:512-23.
33. Allen A, Fisher C, Premawardhena A, et al. Adaptation to anemia in hemoglobin E-ss thalassemia. Blood 2010;116:5368-70.
34. Chou ST, Liem RI, Thompson AA. Challenges of alloimmunization in patients with haemoglobinopathies. Br J Haematol 2012;159:394-404.
35. Musallam KM, Cappellini MD, Wood JC, et al. Elevated liver iron concentration is a marker of increased morbidity in patients with β thalassemia intermedia. Haematologica 2011:haematol. 2011.047852.
36. Borgna-Pignatti C, Marsella M. Iron chelation in thalassemia major. Clinical therapeutics 2015;37:2866-77.
37. Angelucci E, Brittenham GM, Mclaren CE, et al. Hepatic iron concentration and total body iron stores in thalassemia major. New England Journal of Medicine 2000; 343:327-31.
38. Puliyel M, Sposto R, Berdoukas VA, et al. Ferritin trends do not predict changes in total body iron in patients with transfusional iron overload. American journal of hematology 2014;89:391-4.
39. Fischer R, Harmatz PR. Non-invasive assessment of tissue iron overload. ASH Education Program Book 2009;2009: 215-21.
40. Taher A, El Rassi F, Isma'eel H, Koussa S, Inati A, Cappellini MD. Correlation of liver iron concentration determined by R2 magnetic resonance imaging with serum ferritin in patients with thalassemia intermedia. Haematologica 2008;93:1584-6.
41. Pakbaz Z, Fischer R, Fung E, Nielsen P, Harmatz P, Vichinsky E. Serum ferritin underestimates liver iron concentration in transfusion independent thalassemia patients as compared to regularly transfused thalassemia and sickle cell patients. Pediatric blood & cancer 2007;49:329-32.
42. Vichinsky E. Non-transfusion-dependent thalassemia and thalassemia intermedia: epidemiology, complications, and management. Current medical research and opinion 2016;32:191-204.
43. Cappellini MD, Cohen A, Piga A, et al. A phase 3 study of deferasirox (ICL670), a once-daily oral iron chelator, in patients with β-thalassemia. Blood 2006;107:3455-62.
44. Taher AT, Porter JB, Viprakasit V, et al. Deferasirox effectively reduces iron overload in non-transfusion-dependent thalassemia (NTDT) patients: 1-year extension results from the THALASSA study. Annals of hematology 2013;92:1485-93.
45. Taher AT, Cappellini MD, Aydinok Y, et al. Optimising iron chelation therapy with deferasirox for non-transfusion-dependent thalassaemia patients: 1-year results from the THETIS study. Blood Cells, Molecules, and Diseases 2016;57:23-9.
46. Musallam KM, Taher AT, Cappellini MD, Sankaran VG. Clinical experience with fetal hemoglobin induction therapy in patients with β-thalassemia. Blood 2013; 121:2199-212.
47. Galanello R, Sanna S, Perseu L, et al. Amelioration of Sardinian β0 thalassemia by genetic modifiers. Blood 2009;114: 3935-7.
48. Musallam KM, Sankaran VG, Cappellini MD, Duca L, Nathan DG, Taher AT. Fetal hemoglobin levels and morbidity in untransfused patients with β-thalassemia intermedia. Blood 2012;119:364-7.
49. Perrine SP, Ginder GD, Faller DV, et al. A short-term trial of butyrate to stimulate fetal-globin-gene expression in the β-globin disorders. New England Journal of Medicine 1993;328:81-6.
50. Olivieri NF, Saunthararajah Y, Thayalasuthan V, et al. A pilot study of subcutaneous decitabine in β-thalassemia intermedia. Blood 2011;118:2708-11.
51. Taher AT, Cappellini MD. Management of non-transfusion-dependent thalassemia: a practical guide. Drugs 2014;74:1719-29.
52. Fung EB, Kwiatkowski JL, Huang JN, Gildengorin G, King JC, Vichinsky EP. Zinc supplementation improves bone density in patients with thalassemia: a double-blind, randomized, placebo-controlled trial. The American journal of clinical nutrition 2013;98:960-71.
53. Italia KY, Jijina FJ, Merchant R, et al. Response to hydroxyurea in β thalassemia major and intermedia: experience in western India. Clinica Chimica Acta 2009;407: 10-5.
54. Aerbajinai W, Zhu J, Gao Z, Chin K, Rodgers GP. Thalidomide induces γ-globin gene expression through increased reactive oxygen species–mediated p38 MAPK signaling and histone H4 acetylation in adult erythropoiesis. Blood 2007;110:2864-71.
55. Libani IV, Guy EC, Melchiori L, et al. Decreased differentiation of erythroid cells exacerbates ineffective erythropoiesis in β-thalassemia. Blood 2008;112:875-85.
56. Aydinok Y, Karakas Z, Cassinerio E, et al. Efficacy and Safety of Ruxolitinib in Regularly Transfused Patients with Thalassemia: Results from Single-Arm, Multicenter, Phase 2a Truth Study. Am Soc Hematology; 2016.
57. Bazarbachi A-Ha, Chaya BF, Moukhadder HM, Taher AT. Non-transfusion-dependent thalassaemia: a panoramic survey from pathophysiology to treatment. EMJ 2016;1:53-61.
58. Gardenghi S, Ramos P, Roy CN, et al. Hepcidin as a Therapeutic Tool to Limit Iron Overload and Improve Anemia In β-Thalassemia. Blood 2010;116:1009-.
59. Ramos E, Ruchala P, Goodnough JB, et al. Minihepcidins prevent iron overload in a hepcidin-deficient mouse model of severe hemochromatosis. Blood 2012;120:3829-36.
60. Li H, Rybicki AC, Suzuka SM, et al. Transferrin therapy ameliorates disease in [beta]-thalassemic mice. Nature medicine 2010;16:177-82.
61. Porter J, Origa R, Forni GL, et al. A phase 2a, open-label, dose-finding study to determine the safety and tolerability of sotatercept (ACE-011) in adults with beta (β)-thalassemia: interim results. Blood 2013;122: 3448-.
62. Ruckle J, Jacobs M, Kramer W, et al. Single dose, randomized, double blind, placebo controlled study of ACE 011 (ActRIIA IgG1) in postmenopausal women. Journal of Bone and Mineral Research 2009;24:744-52.
63. Cappellini M, Porter J, Origa R, et al. Interim results from a phase 2a, open-label, dose-finding study of sotatercept (ACE-011) in adult patients (PTS) with Beta-thalassemia. Haematologica; 2015: Ferrata Storti Foundation Via Giuseppe Belli 4, 27100 Pavia, Italy. p. 17-8.
64. Baronciani D, Angelucci E, Potschger U, et al. Hemopoietic stem cell transplantation in thalassemia: a report from the European Society for Blood and Bone Marrow Transplantation Hemoglobinopathy Registry, 2000–2010. Bone marrow transplantation 2016;51:536-41.
65. Bank A, Dorazio R, Leboulch P. A Phase I/II Clinical Trial of β Globin Gene Therapy for β Thalassemia. Annals of the New York Academy of Sciences 2005;1054:308-16.
66. Raja J, Rachchh M, Gokani R. Recent advances in gene therapy for thalassemia. Journal of pharmacy & bioallied sciences 2012;4:194.
67. Cappellini MD, Porter JB, Musallam KM, et al. Development of a new disease severity scoring system for patients with non-transfusion-dependent thalassemia. Eur J Intern Med 2016;28:91-6.
68. Succar J, Musallam KM, Taher AT. Thalassemia and venous thromboembolism. Mediterranean journal of hematology and infectious diseases 2011;3: 2011025.
69. Taher A, Vichinsky E, Musallam K, Cappellini M-D, Viprakasit V. Guidelines for the management of non transfusion dependent thalassaemia (NTDT): Thalassaemia International Federation, Nicosia, Cyprus; 2013.
70. AL-Sudani RS, Liu J, Wu J. Estimation of Co-Integration of the Relationship between Blood Transfusion and Iron Deposits in Thalassemia Patients and Study of the Effects of Some Physiological Factors. International Mathematical Forum; 2016. p. 1089-102.

Thalassaemia major emergency cases

Perla Eleftheriou

Consultant Haematologist, University College London Hospital, UK

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Correspondence:

perla.eleftheriou@uclh.nhs.uk

Case #1

45yrs male with Thalassaemia intermedia on regular blood transfusions for last 15 years and very poor adherence to iron chelation, presents with a cardiac T2* 3.9 ms and severe hepatic siderosis. Admitted to the hospital and switched Exjade to intensive intravenous Desferrioxamine and oral Deferiprone. Cardiac T2* increased to 6.4 ms after 5 months on combination chelation, but patient was re-hospitalised acutely unwell after contracting pneumonia. Despite best supportive treatment, he developed septiciaemia, multiorgan failure and decompensated cardiac failure as a consequence and died.
Cardiac iron accumulation is the single greatest risk factor for cardiac dysfunction in thalassaemia. Continuous careful and meticulous clinical monitoring of iron overload and chelation is vital.

Aim

In the presence of severe myocardial iron (T2*<10 ms) to de- iron promptly to prevent cardiac failure. The urgency of escalation of chelation was under recognised for long at the previous non specialist treating centre. Patient's non adherence was a huge limiting factor. Despite improvement in cardiac T2* with acute intensive chelation , high risk of cardiac function decompensation in the presence of trigger factors persists.Intensive rescue chelation (continuous iv) best to initiate as inpatient and can lead to lengthy stay. Very close outpatient follow up after discharge needed.

Case #2

30yrs, male. Significant Iron overload- very suboptimal adherence to iron chelation. Also, history of Ulcerative colitis (bowel resection), previous portacath thrombosis (right atrium)- anti coagulated with Warfarin , however self-discontinued 18 months prior to presentation. Currently presents with several month history of progressive bilateral arm swelling and mild ankle oedema.
Venogram at presentation showed complete occlusive thrombus of the left distal brachiocephalic vein and non-occlusive thrombus of the right and left subclavian veins. Warfarin was restarted with a target INR of 3-4 , significance of full adherence emphasised. Compliance remained suboptimal,therefore following a repeat DEXA scan which showed improved BMD in the osteopenia range, warfarin was switched to LMWH (Dalteparin 5000 U bd sc).
After 12 months od LMWH treatment, clinical signs of thrombosis were resolving and upper arms no longer swollen. Review on admission for sub acute bowel obstruction (required dilatation of his anal rectal pouch) revealed he had discontinued therapeutic LMWH for about 1 year, arms swollen again but no objective detection of acute thrombosis on the CT venogram. LMWH restarted (Fragmin 12500 U od sc) with the view to consider switching to Rivaroxaban. Hyper coagulable state in thalassaemia is considered multifactorial and the risk is recognised to be increased in NTDT compared to TDT patients. Splenectomy and transfusion naivety are increasingly highlighted as important risk factors for VTE in NTDT and in TDT patients the risk is significantly increased in the presence of indwelling catheters used for intravenous chelation for example. An individualised approach is recommended to establish an optimal strategy for preventing the occurrence of this complication and the role of DOACs such as Rivaroxaban in patients with TDT and VTE and on chelation needs to be further explored.

Case #3

19yrs, female presented with 24hr history of pyrexia, rigors, headache, diarrhoea ,vomiting and abdominal pain. She had been on DFO 2.5 g 5x/week ,was splenectomised in childhood and was on regular penicillin. She had previous history of unexplained sepsis and has been on erythomycin for 24hrs prior to presentation. At presentation the antibiotics were changed to amoxicillin for treatment of presumed sinusitis but symptoms did not resolve and the diarrhoea worsened . Ciprofloxacin was added and a few days later gram negative rods were identified in her blood cultures. She was switched to Amikacin and Azlocillin and the US abdomen was suggestive of mesenteric adenitis. The pyrexia and diarrhoea

improved. The identity of the Gram negative rods in the blood was revealed to be Yersinia enterocolitica. The patient was sent home on Septrin. 3 months later, reported diarrhoea at the end of each 5 days of DFO usage and also intermittent abdominal colicky pains. Stool cultures were positive for Yersinia. Yersinia antibody titres 1/320. Was treated with oral septrin bd for 1 month and improved. 8 months later, abdominal pain and diarrhoea recurred. Again, Yersinia was cultured in stool. Was recommenced on Septrin and treated for 2 months, since then no further relapse.

Yersinia enterocolitica can present with localised infection (terminal ileitis, mesenteric adenitis, gastroenteritis, peritonitis) and is usually benign and self limiting or the presentation can be generalised with septicaemia. The latter is associated with high mortality if not diagnosed and treated promptly. Predisposing factors: raw pork,cirrhosis, maligancy, diabetes, immunosuppression, iron overload, DFO use. Yersinia does not make its own siderophore but has a high requirement for iron and therefore uses siderophores of other microorganisms. (*e.g.* DFO).

Challenges in the management of silent cerebral infarct in sickle cell disease

Wasil Jastaniah[1,2]

[1]Department of Pediatrics, College of Medicine, Umm AlQura University, Makkah, Saudi Arabia; [2]Princess Noorah Oncology Center, King Abdulaziz Medical City, Jeddah, Saudi Arabia

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ABSTRACT

This report presents a case of a child with sickle cell disease who, during the course of her disease, was found to have magnetic resonance imaging (MRI) changes consistent with silent cerebral infarct (SCI). Despite chronic transfusion therapy, she continued to develop progressive cerebral vasculopathy. This case study discusses the challenges in the management of SCI in patients with sickle cell disease and presents the management approach used in treating this patient. This report highlights the need for further studies to identify effective preventative and curative therapies for this common complication in patients with sickle cell disease.

Correspondence:

jastaniahwa@ngha.med.sa

Introduction

Silent cerebral infarct (SCI) is the most common permanent and progressive brain injury reported in patients with sickle cell anemia [1,2]. SCI is defined both radiologically and clinically by the presence of a three-millimeter or greater signal abnormality on T2-weighted brain magnetic resonance images (MRI) and seen in two image planes in the absence of clinical neurological impairment [3]. The distribution of SCI on brain MRI correlates with small vessel disease in watershed areas [4]. This suggests that the pathogenesis of SCI may be related to defective regulation of cerebral perfusion pressure, which could be aggravated by low baseline hemoglobin levels and high baseline systolic blood pressure measurements [4,5]. The cumulative prevalence of SCI is reported with increasing frequency by age with no evidence of a plateau at a certain age group [1,2,6]. In fact, SCI is common in infants and preschool children with an approximate cumulative incidence of 25% before 6-years and 39% by 18-years of age [1,2,6]. By the 4th decade of life, it is estimated that approximately half of patients with sickle cell disease suffer from SCI [1]. SCI is associated with an approximate 15-fold increased risk of developing ischemic overt stroke compared to sickle cell diseased patients without SCI [1,2,7]. In addition, SCI is associated with decline in cognitive function, work memory, and processing [1,2,7,8]. Despite the above, there are no clear recommendations to support routine screening of SCI in patients with sickle cell anemia and the best preventative and management approach remains to be defined. Therefore, the present case study presents the challenges in the management of SCI in patients with sickle cell anemia and describes the treatment approach utilized to manage this common and progressive complication.

Case presentation and Discussion

A 12-year old Saudi girl known case of sickle cell anemia presents to our practice at seven years of age with past history of osteomyelitis of the left arm at the age of three years and history of hospitalization three times for pain crisis. Her transfusion history was positive for receiving red blood cell transfusion once during her episode of osteomyelitis. Her physical exam was unremarkable and she was pain free. Laboratory workup revealed hemoglobin of 8.2 g/dL, mean corpuscular volume (MCV) of 80, and hemoglobin electrophoresis measured by HPLC showed hemoglobin S level of 90%, hemoglobin F of 8%, and hemoglobin A2 of 2% consistent with hemoglobin SS disease. Based on her history, hydroxyurea therapy was started to help reduce the recurrent pain crisis. One year later she was pain-free with no history of hospitalizations or pain crisis. Her systemic review was unremarkable and her clinical exam was normal. Laboratory investigations showed hemoglobin of 9 g/dL, MCV 98, and normal liver and renal profile. However, transcranial Doppler

ultrasound image (TCDI) showed a maximum velocity of 180 cm/sec in the right middle cerebral artery. Therefore, a repeat TCDI was requested and an MRI/MRA study of the brain was also requested to rule out stenosis as the patient had a conditional TCDI despite being on hydroxyurea. The child's compliance to hydroxyurea was reviewed and the dose was increased to 30 mg per kilogram per day. The repeated TCDI was normal, however, the MRI/MRA brain image showed signal abnormalities in the right fronto-parietal and left frontal areas consistent with SCI. Her neurological examination was intact with no evidence of neurological deficit. The child and family were counseled regarding the diagnosis of SCI. The risk of developing overt stroke, cognitive dysfunction, and progression was explained [1,2,7,8]. The management options available for SCI were explained [7]. The SIT trial evaluated the benefit of transfusion therapy compared to standard care and showed that transfusions resulted in a relative risk reduction of 58% in the occurrence of all neurological events [7]. Based on these results, patients with SCI may benefit from transfusion therapy to prevent progression of neurological events [7]. The child developed SCI while on hydroxyurea, however, she had no previous MRI images and was started on hydroxyurea therapy only one year before identifying SCI on brain MRI images and thus may have had the changes before commencing hydroxyurea therapy. Whether continuing hydroxyurea therapy at maximum tolerated dose would prevent progression of SCI or not is unclear but was deemed unlikely in this patient. This reflects the lack of routine screening and prevention guidelines for SCI. It may therefore be reasonable to screen all patients early or at least before implementing disease-modifying therapies such as hydroxyurea or transfusion therapy for any sickle cell related complications. No studies have been conducted to evaluate the effectiveness of hydroxyurea therapy in preventing progression of SCI compared to transfusion therapy. Therefore, hydroxyurea therapy was discontinued and the child was started on a chronic transfusion program to prevent progression of her SCI. Three years later, while on chronic transfusion therapy, she presented at the age of 11-years with an episode of headache and transient loss of vision. An urgent MRI/MRA with diffusion-weighted images (DWI) of the brain was requested to rule out ischemic stroke and she was planned for an exchange transfusion to reduce the hemoglobin S level to less than 30%. The results of the MRI/DWI showed no evidence of ischemic infarcts and stable appearance of the SCI compared to previous images. Based on these findings she was continued on a strict blood exchange transfusion program to keep her hemoglobin S level less than 30% and a repeat MRI/MRA study in one-year time was requested.

One year later, at 12-years of age, while on strict chronic exchange transfusion, she had no history of repeated headaches or neurological deficit. However, the repeat MRI/MRA images showed increased signal abnormalities in watershed areas involving the right fronto-parietal, left fronto-parietal, and occipital areas of the brain, in addition to evidence of right middle cerebral artery stenosis on MRA images. Therefore, despite strict transfusion therapy, she developed progressive cerebral vasculopathy. Although, the SIT trial showed benefit of transfusion therapy in reducing the risk of neurological events, this benefit was incomplete in preventing further progression [7]. This highlights the need to identify other modalities of preventative and curative therapy to optimize the management of patients with SCI. The use of hydroxyurea or hematopoietic stem cell transplant (HSCT) as alternative therapies to blood transfusions requires further studies. Nevertheless, HSCT was considered the best option for our patient as she developed SCI while on hydroxyurea and had evidence of progressive cerebral vasculpathy despite transfusion therapy. HSCT is considered the only curative therapy for sickle cell disease and SCI with cognitive deficit is one of the indications for HSCT in patients with sickle cell anemia [9]. However, one of the challenges in providing this therapy is the availability of a suitable related donor. Unfortunately, no HLA-matched related donor was identified for our patient. Therefore, the option of alternative donor HSCT was discussed with the family. However, the use of alternative donor HSCT in sickle cell disease should only be offered under a clinical trial as the benefits and risks from this form of therapy still needs to be addressed. The family was offered to be transferred to a center with an active clinical trial utilizing alternative donor HSCT but they did not consent for this form of therapy. Based on this, the child was started on hydroxyurea in combination with chronic transfusion therapy in an attempt to reduce the rate of progressive cerebral vasculopathy. The use of hydroxyurea and transfusion as combined therapy has not been formerly studied. However, reports have shown that this combination is safe [10]. In fact, a recent report on seven children with progressive cerebral vasculopathy received combination therapy with no observed toxicity suggesting that this approach may be reasonable in the absence of other options [10]. However, further studies are warranted.

Conclusions

SCI is the most common neurological complication observed in patients with sickle cell disease and is associated with a progressive natural history. Therefore, there is need for optimizing early screening and prevention measures. Although transfusion therapy may reduce the risk of progressive neurological events, the benefit of transfusion therapy for SCI is incomplete. Therefore, alternative therapies such as HSCT or the combined use of hydroxyurea with blood transfusion may help halt the development of progressive cerebral vasculopathy in these patients. The present case study highlights the challenges in the management of sickle cell disease patients with SCI and addresses the need for further studies.

References

1. Kassim AA, Pruthi S, Day M, Rodeghier M, Gindville MC, Brodsky MA, DeBaun MR, Jordan LC. Silent cerebral infarcts and cerebral aneurysms are prevalent in adults with sickle cell anemia. Blood. 2016;127:2038-2040.
2. DeBaun MR, Kirkham FJ. Central nervous system complications and management in sickle cell disease. Blood 2016;127:829-838.
3. Casella JF, King AA, Barton B *et al*. Design of the silent cerebral infarct transfusion (SIT) trial. Pediatr. Hematol. Oncol. 2010;27:69-89.
4. DeBaun MR, Armstrong D, McKinstry RC, Ware RE, Vichinsky E, Kirkham FJ. Silent cerebral infarcts: a review on a prevalent and progressive cause of neurologic injury in sickle cell anemia. Blood. 2012;119:4587-4596.
5. Dowling MM, Quinn CT, Rogers ZR, Buchanan GR. Acute silent cerebral infarction in children with sickle cell anemia. Pediatr. Blood Cancer. 2010;54:461-464.
6. Kassim AA, Galadanci NA, Pruthi S, DeBaun MR. How I treat and manage strokes in sickle cell disease. Blood 2015;125:3401-3410.
7. DeBaun MR, Gordon M, McKinstry RC, Noetzel MJ, White DA, Sarnaik SA, *et al.* Controlled trial of transfusions for silent cerebral infarcts in sickle cell anemia. N Engl J Med. 2014;371:699-710.
8. Vichinsky EP, Neumayr LD, Gold JI *et al.*; Neuropsychological Dysfunction and Neuroimaging Adult Sickle Cell Anemia Study Group. Neuropsychological dysfunction and neuroimaging abnormali- ties in neurologically intact adults with sickle cell anemia. JAMA. 2010;303:1823-1831.
9. Walters MC, De Castro LM, Sullivan KM, Krishnamurti L, Kamani N, Bredeson C, Neuberg D, Hassell KL, Farnia S, Campbell A, Petersdorf E. Indications and Results of HLA-Identical Sibling Hematopoietic Cell Transplantation for Sickle Cell Disease. Biol Blood Marrow Transplant. 2016;22:207-11.
10. Brousse V, Gandhi S, de Montalembert M, *et al.* Combined blood transfusion and hydroxycarbamide in children with sickle cell anaemia. British Journal of Haematology. 2013;160:255-272.

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Thalassaemia and other haemoglobinopathies in the WHO Eastern Mediterranean region

Asmus Hammerich,[1] Slim Slama,[2] Karima Gholbzouri[3]

[1]Acting Director, Department of Noncommunicable Diseases and Mental Health (NMH), WHO EMRO, Cairo/Egypt; [2]Medical Officer, NCD Management Unit, Department of Noncommunicable Diseases & Mental Health (NMH), WHO EMRO, Cairo/Egypt; [3]Medical Officer, Women's and Reproductive Health Unit, Department of Health Promotion and Protection (DHP), WHO EMRO, Cairo/Egypt

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Correspondence:

hammericha@who.int

ABSTRACT

Introduction: Thalassemia and other haemoglobinopathies constitute a serious health problem and a major cause of death and disability around the world and particularly in the Eastern Mediterranean Region (EMR), placing a high burden on individuals and their families as well as on our health systems. In the Region, these conditions, categorized as autosomal recessive single gene disorders, remain among the most common congenital and genetic disorders. *Burden of thalassaemia and other haemoglobinopathies:* Haemoglobinopathies population carrier rates in the EMR range from: 2-50% for α-thalassaemia; 2-7% for β-thalassaemia; 0.3-30% for sickle cell and 2.5 to 27% for G6PD deficiency. *The global response:* Acknowledging the high burden that thalassaemia and other haemoglobinopathies pose on families and health systems, WHO has since the early 1970s, and with two endorsed global resolutions in 2006 and 2010 (WHA59.20 on sickle cell disease and resolution WHA63.17 on birth defects), advocated and promoted the development and implementation of comprehensive national, integrated programs for the prevention and management of those disorders, including surveillance, dissemination of information, awareness-raising and screening, tailored to specific socioeconomics and cultural contexts, with the aim to reduce the related incidence, morbidity and mortality. *Main challenges are:* • Consanguineous marriage account for 20-50% of all marriages, with first cousin unions amounting to 20-30% of all marriages in the EMR; • Young age of marriage and low educational level bringing high fertility; • Religious and social reservations regarding interventions during pregnancy; • The fact that haemoglobinopathies are not always recognized as priorities in national health plan; • Limited preventive programs and lack of accurate data; • Lack of public awareness about genetic risks and the possibilities of prevention; • Poor genetic education of health professionals both at undergraduate and postgraduate levels; • Political instability & related humanitarian crises affecting health system delivery in several countries of the Region; and • Poor data and surveillance system to monitor and assess health outcomes and health system performance. *WHO EMR initiatives:* There have been important WHO EMRO initiatives in response to congenital and genetic disorders (CGD) including thalassaemia and other haemoglobinopathies such as promoting an inclusive preconception care (PCC) core package for the Region recommending interventions to reduce burden of CGD at the preconception/premarital stage. *Conclusions:* Evidence suggests that successful programs where those that combined the following elements: 1) Public awareness raising and education to improve genetic literacy and address common misconceptions which represent key barriers to the successful implementation of community genetic programs; 2) Adequate dialogue with legal, cultur-

al and religious leaders in order to establish acceptability of policies according to the specific context of the country and its population; 3) Availability of population screening programs with provision of genetic counselling for individuals and at-risk couples, including at the primary health care level; and 4) The availability of choices for prevention, including pre-natal diagnosis and pre-implantation genetic diagnosis, linked with legal therapeutic abortion.

Introduction

Thalassemia and other haemoglobinopathies constitute a serious health problem and a major cause of death and disability around the world and particularly in the Eastern Mediterranean Region (EMR), placing a high burden on individuals and their families as well as on our health systems. In the Region, these conditions, categorized as autosomal recessive single gene disorders, remain among the most common congenital and genetic disorders. Haemoglobinopathies population carrier rates in the EMR range from: 2-50% for α-thalassaemia; 2-7% for β-thalassaemia; 0.3-30% for sickle cell and 2.5 to 27% for G6PD deficiency.

Leading causes of thalassaemia and other haemoglobinopathies are well known with the main one being consanguinity (20-50% in the EMR). Increasing demand for services required to prevent, early detect and manage them, require more systemic and integrated approaches to reduce the burden of these conditions. Many EMR countries face major challenges in providing comprehensive, high-quality and up-to-date health care including genetic services. Costs related to the treatment of severe forms of haemoglobinopathies remain high and quality services may be inaccessible.

Methodology and hypothesis

Available epidemiological data on haemoglobinopathies published in the Region point to the high population carrier rates of these disorders. The disorders occur in EMR countries in rates similar to or exceeding those in industrialized countries. This phenomenon might be explained by the following factors:

First, previous presence of malaria in the Region has conferred a selective advantage to carriers against falciparum malaria allowing them to live longer and to give birth to more children presenting these defects.

The second critical factor is cultural and relates to high consanguinity rates, especially among first cousins, which, coupled with large family size, represents one of the predictors for the expression of autosomal recessive genetic disorders and haemoglobinopathies, including thalassemia and sickle-cell disease. Despite some recent changes, consanguinity rates in the Region remain the highest in the world, reaching 20-50% in most EMR countries. Social, cultural, political and economic factors all play a role in favoring consanguineous marriages among the young generation just as much as they did among earlier generations, particularly in rural areas.

Finally, the Region like many parts of the world is undergoing an important epidemiological and demographic transition. As neonatal and childhood mortality rates decline due to improved social and public health conditions, infants who would have previously died from these conditions before being diagnosed, are now surviving to present for diagnosis and treatment.

Results and response

Haemoglobinopathies, such as the most severe forms of thalassemia, were in the past a synonym of an early death. Bone marrow transplantation, the only radical curative treatment for the most severe homozygote forms, poses challenges in terms of donor compatibility and remains out of reach for most affected individuals in our Region, due to the lack of infrastructure, and the cost and capacity required performing this complex intervention.

However, improved access to blood transfusions and iron chelation therapy mean that patients with thalassemia can today expect to live longer, provided they comply with their therapy. As survival improves, these conditions become chronic, thus requiring regular follow up including the management of recurrent exacerbations and psychological support.

The frequency of haemoglobinopathic disorders in the Region and the increasing demand for services that are required to prevent, early detect and manage them, require more systemic and integrated approaches to progress towards reducing the burden of these conditions.

Acknowledging the high burden that thalassaemia and other haemoglobinopathies pose on families and health systems, WHO has since the early 1970s, and with two endorsed global resolutions in 2006 and 2010 (WHA59.20 on sickle cell disease and resolution WHA63.17 on birth defects), advocated and promoted the development and implementation of comprehensive national, integrated programs for the prevention and management of those disorders, including surveillance, dissemination of information, awareness-raising and screening, tailored to specific socioeconomics and cultural contexts, with the aim to reduce the related incidence, morbidity and mortality.

Despite some success stories, such as premarital screening programs in Northern Iraq, the carrier screening program in Iran or extensive community awareness campaigns on sickle cell anemia implemented in Bahrain over the past 20 years, national programs and services offered in our Region remain patchy, selective and ultimately inadequate for providing the range of preventive and curative services needed. As it stands, most EMR countries still face major challenges in providing comprehensive, quality health services including genetic services required to address these disorders.

For instance, recently published reviews of the efficacy of mandatory premarital screening and genetic counselling programs in selected countries of the EMR demonstrated mixed results, showing that these programs were often unsuccessful in discouraging at-risk marriages, primarily due to poor timing of the screening, a lack of knowledge regarding inherited diseases, and sociocultural and religious concerns.

By contrast, success stories in the Region and globally were recorded when programs adopted a more comprehensive "life cycle" approach to prevention, covering the preconception period, pregnancy and interventions after birth such as newborn screening.

Discussion

The following challenges remain:

- Consanguineous marriage account for 20-50% of all

marriages, with first cousin unions amounting to 20-30% of all marriages in the EMR.
- Young age of marriage and low educational level bringing high fertility.
- Religious and social reservations regarding interventions during pregnancy.
- The fact that haemoglobinopathies are not always recognized as priorities in national health plan.
- Limited preventive programs and lack of accurate data.
- Lack of public awareness about genetic risks and the possibilities of prevention.
- Poor genetic education of health professionals both at undergraduate and postgraduate levels.
- Political instability and related humanitarian crises affecting health system delivery in several countries.
- Poor data and surveillance system to monitor and assess health outcomes and health system performance.

At the same time there have been important WHO EMRO initiatives in response to CGD including thalassaemia and other haemoglobinopathies, such as promoting an inclusive preconception care (PCC) core package for the Region recommending interventions to reduce burden of CGDs at the preconception/premarital stage.

Moreover, a WHO EMRO landmark publication on community-based control of genetic and congenital disorders of 1997 (WHO EMRO Technical Publications Series 24) is currently revised. It includes recommendations on premarital prenatal, newborn and school screening, genetic counselling, community education, genetic laboratories, and congenital abnormalities and hereditary diseases registries.

On the global level, world leaders have last year agreed on a new, bold development roadmap with the adoption of the Sustainable Developments Goals (SDGs) and the pledge "to leave no one behind". Health occupies a prominent place on this new agenda as SDG 3, notably with the Universal Health Coverage (UHC) framework attached.

Achieving UHC to reach the new health-related SDG will require addressing a broader range of health challenges including a growing burden of noncommunicable diseases (esp. cardiovascular and chronic lung diseases, diabetes, cancer and mental health) as well as the unfinished agenda related to maternal and child health and communicable diseases. The UHC framework along its three dimensions, namely access to quality essential health services, coverage of services, and financial protection, offer an opportunity for health systems and programs to revisit the set of population and individual health interventions to be prioritized.

Revisiting past work accomplished in the field of congenital disorders, WHO EMRO has over the last two years undertaken a situation analysis of congenital disorders in the Region. This work has informed a recent expert consultation which, in line with the UHC imperative, has started to identify a core package of evidence-based high impact interventions to be included as part of an essential set of community genetic services.

Recommendations were made for the inclusion of specific interventions related to thalassemia and other haemoglobinopathies, such as population carrier screening, prenatal diagnosis with termination of pregnancy, in accordance with the country's specific health system, cultural, religious and legal context.

This core package of high impact interventions needs to be tailored also to country specific epidemiology and health system capacity. Given the phenotypic diversity of haemoglobin disorders, country specific profiles combining reliable data on the magnitude and characteristics of these disorders as well as an assessment of the health system requirements (*e.g.* reference genetic laboratory services) to implement this core package of interventions, will be required.

The WHO EMR Office is currently finalizing core interventions along the continuum of care with guiding programmatic steps to support our Member States in reviewing their epidemiological status, implementing preventive and management measures to reduce the burden of congenital and genetic disorders, and particularly thalassemia and other haemoglobinopathies, as well as monitoring the effectiveness and progress made.

Conclusions

Evidence suggests that successful programs where those that combined the following elements:

1) Public awareness raising and education to improve genetic literacy and address common misconceptions which represent key barriers to the successful implementation of community genetic programs;
2) Adequate dialogue with legal, cultural and religious leaders in order to establish acceptability of policies according to the specific context of the country and its population;
3) Availability of population screening programs with provision of genetic counselling for individuals and at-risk couples, including at the primary health care level; and
4) The availability of choices for prevention, including pre-natal diagnosis and pre-implantation genetic diagnosis, linked with legal therapeutic abortion.

There is hope that the described renewed interest and effort in the field of congenital and genetic disorders will contribute to raising the profile and further strengthen country capacity in preventing and managing the most prevalent haemoglobinopathies in the EM Region.

This is work in progress, where the continuous support of a wide range of concerned partners is very much required and appreciated.

References

WHO HQ: World Health Statistics, 2014.
WHO EMRO: Technical publications Series 24 – Community control of genetic and congenital disorders, 1997.

ARTICLE

Endocrine complications in thalassaemia syndromes

Farrukh T. Shah

Consultant haematologist, King Khalid University Hospital, Riyadh, Saudi Arabia; Whittington hospital, London

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ABSTRACT

The outcomes for patients with thalassaemia syndromes has improved significantly in the last 4 decades with improvements in blood transfusion, chelation therapy and monitoring. As a consequence of this success, patients are now expected to live a nearly normal life expectancy and therefore aspire to lead as normal a life as possible. Endocrine complications and in particular their impact on quality of life and fertility are of paramount concern for patients and the clinicians involved in their care. Endocrinopathies are preventable provided that iron burden is well controlled from an early age and maintained at safe levels lifelong. Recent data suggests that endocrinopathies may be reversed in some patients with high intensity chelation regimes and very low target serum ferritins, but there remain significant concerns of the feasibility of this in routine patients, many of whom already struggle with maintaining even reasonable levels of iron burden.

Prevalence of endocrinopathies

Endocrine organs are particularly susceptible to iron related damage and certainly pituitary damage and its direct impact on transition to puberty is known to occur early in childhood. Data from cohort studies show that in some populations endocrinopathies remain highly prevalent especially in those patients who have had poor chelation either due to lack of adherence or insufficient access to chelation due to either financial factors or clinician factors. More recently MRI assessments undertaken of target organs such as the pituitary, pancreas etc.. are adding to our understanding of the pathophysiology iron related endocrinopathies in young children.

Hypogonadism

Hypogonadism is the most prevalent of all iron overload related endocrinopathies and is defined as patients having failure of initiation and development of secondary sexual characteristics by the age of 13 in females or 14 years in males or the presence of primary or secondary amenorrhea in females over the age of 16 or the need for testosterone replacement.

Registry data from centres shows that hypogonadism is the most prevalent endocrinopathy in patients with transfusion dependant thalassaemia and ranges from 32-50% (1-7). What is striking in these publications is that even in centres where the population is relatively young there is a high prevalence of hypogonadism.

If hypogonadism is diagnosed, then initiation of the appropriate hormone therapy to help correct growth potential in a timely fashion is critical to ensure growth potential is achieved. In situations where the epiphysis have fused and growth is not the issue it remains important to ensure adequate and appropriate hormone replacement therapy to ensure development of secondary sexual characteristics and prevent osteoporosis. The psychosocial burden of being visibly different to their peers is significant for patients and all efforts should be made to prevent this.

Short stature

Short stature is defined as height which is two standard deviations below the mean height for age and sex or below the 3rd percentile or more than two standard deviations below the mid parental height. Short stature is relatively common in thalassaemia patients with incidences ranging from 20-40% (8) (9). The reasons

Correspondence:
farrukh.shah@nhs.net

for short stature are multifactorial in thalassaemia patients. Under transfusion can result in short stature especially if the anaemia is significant. Hypogonadism by virtue of failure to transition through puberty may result in short stature particularly if the induction of puberty is delayed after the closure of epiphysis. Short stature may also be due to the impact of high doses of desferrioxamine given to children which resulted in the classical short truncal height seen in many of the older patients with thalassaemia (10) (11). Aside from all of the thalassemia specific issues nutritional issues should also be reviewed.

In patients with short stature all the above issues should be excluded and although growth hormone deficiency the role of Growth hormone deficiency as confirmed by a low IgF 1Beta and failure to increase levels following a growth hormone stimulation test. Growth hormone replacement therapy can correct the deficiency state. More commonly in adults some degree of subclinical/ clinical degrees of GH deficiency are being diagnosed (9).

Hypothyroidism

Hypothyroidism is a relatively common side effect generally ranging from 5 to 7% with a higher prevalence in older patients and those with multiple endocrinopathies (9, 12) Predominantly this occurs due to the thyroid gland failing to respond to the pituitary signal. It is relatively uncommon for hypothyroidism to be the only endocrinopathy is a thalassaemia patient and is seen more often in patients with multiple endocrinopathies often a reflection of long standing poor iron chelation.

Hypoparathyroidism

Hypoparathyroidism is again rarely seen as an individual endocrinopathy and more frequently seen in patients with multiple endocrinopathies. The prevalence of hypoparathyroidism ranges between 1 and 8% in various studies and is a relatively rare endocrinopathy and criteria used to confirm a diagnosis of hypoparathyroidism in the various studies is not uniform. It is important when diagnosing hypoparathyroidism that the Ca/Vitamin D levels are as optimised as possible before confirming low PTH levels.

Diabetes Mellitus/Impaired glucose tolerance

The frequency of diabetes is quite variable with rates fluctuating between 5-25% across the studies. However the frequency of diabetes is relatively high in the older thalassaemia major patients where rates are around 40% (9, 12, 13). This is probably because most older patients started chelation therapy at an older age and were more likely to have developed significant iron overload. There does appear to be a relationship between cardiac T2* and the risk of developing diabetes and this is most likely due to periods of poor chelation.

Monitoring for impaired glucose tolerance should start in patients from around the age of 10-15 years.

Adrenal insufficiency

It is difficult to be clear about the precise frequency of adrenal deficiency in the thalassaemia population as this was not previously well studied but recent publications are suggesting rates of adrenocortical insufficiency at 2-60% (14-16). Many patients received and still do receive hydrocortisone as part of the blood transfusion to reduce reactions and where possible this should be limited as much as possible to avoid iatrogenic

The role of multiple endocrinopathies

Although multiple endocrinopathies are more prevalent in older patients, even in young patients can develop multiple endocrinopathies as this reflects the impact of poor chelation (12, 17, 18) .

Reversibility of endocrinopathies

Recent publications have suggested that endocrinopathies can be reversed (19, 20). These studies raise questions about what threshold of iron burden is needed to reverse endocrinopathies as both studies recorded improvement in endocrinopathies only in those patients who had very low levels of liver iron and ferritin (Poggi: Median ferritin 185 ug/l (160-649ug/l) and Farmaki: mean serum ferritin 103±60 ug/l.) However, both studies has very small numbers of patients who were considered to have reversed an endocrinopathy and certainly in Poggi's study patients developed new endocrinopathies despite very low ferritins.

In Farmakis study the major endocrinopathy that improved was diabetes with improvement in 1 patient with NIDDM to an impaired glucose tolerance and the improvement of 2 patients with impaired GTT to normal GTT. Reversibility of impaired glucose tolerance has been previously described (18, 21, 22).

The Poggi's study did define what ferritin was associat-

Table 1.

Measurement	Frequency
Assessments of weight and height – sitting and standing	6 monthly from diagnosis until final adult height
Assessment of puberty, using Tanner staging	Yearly from age 10 years
Plain X ray of left wrist	1-2 yearly from age 10 or if concern about fall in height velocity
Growth hormone stimulation test	If declining growth velocity from age 8
Oral glucose tolerance test	Yearly from puberty, or from age 10 years
Full annual diabetes review, including glycaemic control, cardiovascular risk factors, diabetic complications and sexual health	Yearly if a diagnosis of diabetes is established
TSH, fT4	Yearly from age 12
Ca, PO4, ALP, PTH	6 monthly from age 12 If calcium level is low
Vitamin D level	At least every year from age 2
Morning cortisol level	Yearly from age 14
LH/FSH and oestradiol	If menstrual disturbance develops
testosterone level	Yearly from age 14
LH/FSH and sex hormone binding globulin level	In men if morning testosterone level is low

ed with an increased risk of developing an endocrinopathy in patients who did not previously have one. In addition, the analysis suggests for reversal of an endocrinopathy a target ferritin of below 200ug/l was needed.

Diagnostic work up

See Table 1.

Management and prevention of endocrinopathies

Literature from times when good chelation was not readily available and the modern era confirm that endocrinopathies are inextricably linked to iron overload in thalassaemia patients. Therefore, the key principle that underpins management of these disorders is prevention. Appropriate screening of patients to identify abnormalities at an early stage where they may be reversible by increasing the intensity of chelation is critical. In addition, the optimisation of patient concordance with therapy is needed. Management of heavily iron overloaded patients' needs a multidisciplinary approach with Endocrinologists, haematologists, and clinical psychologists all playing a critical role. Once an endocrinopathy has been established optimised management of the relevant endocrinopathy as well as iron overload is essential to prevent the development of further complications.

Specific management issues in Hypogonadal male patients

It is essential that patients are managed by a centre with expertise in male factor infertility but also have active involvement of the thalassaemia specialist. In order to have a good response to spermatogenesis iron burden must be well controlled and kept at optimal levels for the duration of spermatogenesis induction. In addition any co-existing endocrinopathies should be optimally managed, in particular diabetes should be optimised as much as possible and the HbA1c kept well within the target limit as per national guidance for Insulin dependent diabetes. The clinical work up for hypogonadism should be overseen by the fertility team. By and large for patients who have not completed puberty spontaneously the testicular volumes will frequently be small and they will require spermatogenesis induction. Conversely in patients who completed puberty with simple testosterone priming and did not require testosterone supplementation long term or those who completed puberty naturally will mostly have normal testicular volumes and spermatogenesis; provided they have not at a later post pubertal stage developed hypogonadism. The goal should therefore be for all male patients to complete puberty naturally or with minimal testosterone priming. There are newer studies which suggest that induction of puberty in hypogonadal adolescents can be successfully undertaken with human chorionic gonadotropin and recombinant FSH (23) which would then hopefully reduce the need for spermatogenesis induction in the adult patients.

Spermatogenesis induction should be undertaken in a stepwise manner in patients who require it. In our centre the practice is to stop testosterone supplementation and initiate.

Therapy with HCG (750-2000iu sc 3 times a week). In patients with testicular volumes greater than 8mls it is likely to be all that is needed. This is generally given for 6 months with monthly to 3 monthly sperm samples. In thalassaemia patients this might be needed for longer if there is some evidence of improvement in sperm counts or endogenous testosterone levels.

In patients who initially had lower testicular volumes or those who are not showing improvement in sperm counts with HCG alone the addition of FSH (human menopausal gonadotrophin) generally at a dose of 75 iu sc on alternate days is recommended. This combination therapy is continued until adequate sperm is produced for sperm banking or conception occurs. We do not recommend that treatment is continued much beyond the completion of the 3rd year. Patients should continue on the normal chelation regime until there is adequate spermatogenesis and once the count is sufficient for spontaneous conception they should be switched over to desferrioxamine for 2 to 3 months prior to planned conception. At this time point it is our practice to undertake sperm banking for future pregnancies. In patients who fail spermatogenesis induction they should then proceed to testicular biopsy and if possible Intracytoplasmic Sperm injection (ICSI).

In our centre we have had 16 long term adult Thalassaemia major patients with children, 3 needed induction of puberty and 13 went through puberty spontaneously. 2 patients who had completed puberty and were not on testosterone supplementation required spermatogenesis induction. Of the 5 who required spermatogenesis induction the duration of therapy was between 3 months to 3 years and with one patient failed to have any spermatogenesis. The reasons for the spermatogenesis failure remain unclear. So far in our 19 patients 27 children have been born (range: 0-5, average 1.7 children) 7 children in 4 patients with spermatogenesis induction. In the 4 patients post spermatogenesis induction one has 3 children and the another 2 children without further treatment, one has IVF with banked sperm for subsequent children, one no further children by choice.

Patients who have undergone spontaneous puberty generally have few problems with fertility however it is important to discuss plans for initiation of a family with the patients so that the treatment can be optimised to ensure optimal sperm production and reduce any potential of teratogenicity from chelation therapy (Figure 1).

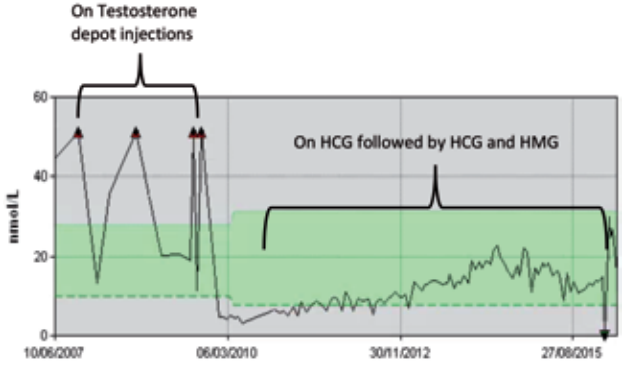

Figure 1. Testosterone levels in a patient undergoing spermatogenesis induction. Graph of patient undergoing spermatogenesis induction showing endogenous serum testosterone levels improving on HCG very slowly and then more significant improvement on HMG.

Conclusions

Prevention of endocrine complications is critical to improving long term outcomes in thalassaemia patients. This is only possible by good monitoring to identify problems early in childhood and adolescence in order to ensure optimal growth, prevention of hypogonadism and other endocrinopathies. It appears from recent data that target ferritins should be considerably lower than previ-

ously acceptable values for prevention of new endocrinopathies (<1300ug/l). The data on reversibility of endocrine complications remains sparse and requires more clarity from larger multicentre studies. All patients with endocrinopathies should be managed by multidisciplinary teams with endocrinologists playing a key role in preventing long term sequelae of poorly managed endocrine complications.

References

1. Cunningham MJ, Macklin EA, Neufeld EJ, Cohen AR. Complications of beta-thalassemia major in North America. Blood. 2004;104(1):34-9.
2. Jensen CE, Tuck SM, Old J, Morris RW, Yardumian A, De Sanctis V, et al. Incidence of endocrine complications and clinical disease severity related to genotype analysis and iron overload in patients with beta-thalassaemia. Eur J Haematol. 1997;59(2): 76-81.
3. Kattamis CA, Kattamis AC. Management of thalassemias: growth and development, hormone substitution, vitamin supplementation, and vaccination. Seminars in hematology. 1995;32(4):269-79.
4. Toumba M, Sergis A, Kanaris C, Skordis N. Endocrine complications in patients with Thalassaemia Major. Pediatric endocrinology reviews : PER. 2007;5(2):642-8.
5. De Sanctis V, Eleftheriou A, Malaventura C. Prevalence of endocrine complications and short stature in patients with thalassaemia major: a multicenter study by the Thalassaemia International Federation (TIF). Pediatric endocrinology reviews : PER. 2004;2 Suppl 2:249-55.
6. Mula-Abed WA, Al Hashmi H, Al Muslahi M, Al Muslahi H, Al Lamki M. Prevalence of endocrinopathies in patients with Beta-thalassaemia major - a cross-sectional study in oman. Oman medical journal. 2008;23(4):257-62.
7. Belhoul KM, Bakir ML, Kadhim AM, Dewedar HE, Eldin MS, Alkhaja FA. Prevalence of iron overload complications among patients with b-thalassemia major treated at Dubai Thalassemia Centre. Annals of Saudi medicine. 2013;33(1):18-21.
8. Altincik A, Akin M. Prevalence of Endocrinopathies in Turkish Children With beta-Thalassemia Major: A Single-Center Study. Journal of pediatric hematology/oncology. 2016;38(5):389-93.
9. De Sanctis V, Elsedfy H, Soliman AT, Elhakim IZ, Kattamis C, Soliman NA, et al. Clinical and Biochemical Data of Adult Thalassemia Major patients (TM) with Multiple Endocrine Complications (MEC) versus TM Patients with Normal Endocrine Functions: A long-term Retrospective Study (40 years) in a Tertiary Care Center in Italy. Mediterr J Hematol Infect Dis. 2016;8(1):e2016022.
10. De Sanctis V, Soliman AT, Elsedfy H, Skordis N, Kattamis C, Angastiniotis M, et al. Growth and endocrine disorders in thalassemia: The international network on endocrine complications in thalassemia (I-CET) position statement and guidelines. Indian journal of endocrinology and metabolism. 2013;17(1):8-18.
11. De Sanctis V, Pinamonti A, Di Palma A, Sprocati M, Atti G, Gamberini MR, et al. Growth and development in thalassaemia major patients with severe bone lesions due to desferrioxamine. Eur J Pediatr. 1996;155(5):368-72.
12. Ang AL, Tzoulis P, Prescott E, Davis BA, Barnard M, Shah FT. History of myocardial iron loading is a strong risk factor for diabetes mellitus and hypogonadism in adults with beta thalassemia major. Eur J Haematol. 2014;92(3):229-36.
13. De Sanctis V, Soliman AT, Elsedfy H, Pepe A, Kattamis C, El Kholy M, et al. Diabetes and Glucose Metabolism in Thalassemia Major: An Update. Expert review of hematology. 2016:1-8.
14. Huang KE, Mittelman SD, Coates TD, Geffner ME, Wood JC. A significant proportion of thalassemia major patients have adrenal insufficiency detectable on provocative testing. Journal of pediatric hematology/oncology. 2015;37(1):54-9.
15. Matin S, Jahromi MG, Karemizadeh Z, Haghpanah S, De Sanctis V, Soliman A, et al. The Frequency of Adrenal Insufficiency in Adolescents and Young Adults with Thalassemia Major versus Thalassemia Intermedia in Iran. Mediterr J Hematol Infect Dis. 2015;7(1):e2015005.
16. Ucar A, Oner N, Ozek G, Cetincakmak MG, Abuhandan M, Yildirim A, et al. Evaluation of the glucocorticoid, mineralocorticoid, and adrenal androgen secretion dynamics in a large cohort of patients aged 6-18 years with transfusion-dependent beta-thalassemia major, with an emphasis on the impact of cardiac iron load. Endocrine. 2016;53(1):240-8.
17. Sharma R, Seth A, Chandra J, Gohain S, Kapoor S, Singh P, et al. Endocrinopathies in adolescents with thalassaemia major receiving oral iron chelation therapy. Paediatrics and international child health. 2016;36(1):22-7.
18. Gamberini MR, De Sanctis V, Gilli G. Hypogonadism, diabetes mellitus, hypothyroidism, hypoparathyroidism: incidence and prevalence related to iron overload and chelation therapy in patients with thalassaemia major followed from 1980 to 2007 in the Ferrara Centre. Pediatric endocrinology reviews : PER. 2008;6 Suppl 1:158-69.
19. Poggi M, Sorrentino F, Pugliese P, Smacchia MP, Daniele C, Equitani F, et al. Longitudinal changes of endocrine and bone disease in adults with beta-thalassemia major receiving different iron chelators over 5 years. Ann Hematol. 2016;95(5):757-63.
20. Farmaki K, Tzoumari I, Pappa C. Oral chelators in transfusion-dependent thalassemia major patients may prevent or reverse iron overload complications. Blood cells, molecules & diseases. 2011;47 (1):33-40.
21. Farmaki K, Angelopoulos N, Anagnostopoulos G, Gotsis E, Rombopoulos G, Tolis G. Effect of enhanced iron chelation therapy on glucose metabolism in patients with beta-thalassaemia major. British journal of haematology. 2006;134(4):438-44.
22. Christoforidis A, Perifanis V, Tsatra I, Vlachaki E, Athanassiou-Metaxa M. Evolution of OGTT in patients with beta-thalassaemia major in relation to chelation therapy. Diabetes research and clinical practice. 2007;76(1):6-11.
23. Rohayem J, Hauffa BP, Zacharin M, Kliesch S, Zitzmann M. Testicular growth and spermatogenesis: new goals for pubertal hormone replacement in boys with hypogonadotropic hypogonadism♀ -a multicentre prospective study of hCG/rFSH treatment outcomes during adolescence. Clinical endocrinology. 2017;86(1):75-87.

ARTICLE

Cardiac complications in haemoglobinopathies

Dimitrios Farmakis

Department of Internal Medicine, "Laiko" University Hospital, National and Kapodistrian University of Athens, Athens, Greece

EUROPEAN HEMATOLOGY ASSOCIATION

Ferrata Storti Foundation

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Cardiac complications represent a leading cause of morbidity and mortality in patients with inherited hemoglobinopathies, including -thalassemia and sickle cell disease (SCD). In the current era, given the global distribution of hemoglobinopathies due to the immigration of populations with high prevalence of the disorders on one hand and the prolonged survival of patients by virtue of modern therapy on the other, both the cardiovascular spectrum and the need for long-term cardiac follow-up and care of patients are continuously growing. Therefore, there is a need for increased awareness and better understanding of the cardiovascular complications in hemoglobinopathies so as healthcare professionals are able to diagnose and treat those complications in a proper and timely manner.

Chronic hemolysis is the key pathogenetic mechanism underlying the hemoglobinopathies. Chronic hemolysis results in anemia that in turn leads to compensatory elevation of cardiac output and, when severe, a high cardiac output state. On the other hand, anemia treatment with repetitive blood transfusions, unless combined with proper iron chelation therapy, results in myocardial iron overload. Chronic hemolysis has further been associated with a complex vasculopathy, caused by NO depletion, endothelial dysfunction and elastic tissue abnormalities, while in the case of SCD, the abnormal hemoglobin S may cause recurrent episodes of microvascular obstruction that are followed by ischemia and reperfusion injury. The above mechanisms, which have a differential expression in the different hemoglobinopathies, may ultimately lead to left ventricular (LV) dysfunction, pulmonary hypertension (PH) and/or right ventricular (RV) dysfunction.

The main **phenotypes of cardiac disease** seen in patients with hemoglobinopathies include:

- Iron overload cardiomyopathy seen mainly in regularly transfused patients with thalassemia major who are not properly treated with iron chelation regimens. Left ventricular diastolic dysfunction is the early-stage condition. In advanced stages, two distinct cardiomyopathy phenotypes are recognized, the most frequent dilated cardiomyopathy phenotype, characterized by LV dilatation and reduced contractility, and the less frequent restrictive cardiomyopathy phenotype with restrictive LV filling, preserved systolic function and secondary pulmonary hypertension.
- Pulmonary hypertension is the main cause of heart disease and failure in non-regularly transfused patients with thalassemia intermedia, while it also seen in one third of patients with SCD, in whom it carries an adverse prognosis.
- High output failure represented the main cause of death in thalassemia major patients before the introduction of regular transfusion therapy and it may also be seen in patients with severe thalassemia intermedia not receiving blood transfusions.
- Acute pericarditis used to be a frequent complication in poorly treated thalassemia major patients, but only sporadic cases are seen nowadays in regularly transfused and chelated subjects. Acute myocarditis has been considered a potential cause of heart failure in thalassemia major, but it is also sporadically encountered today.
- Myocardial ischemia, mainly in the form of stress-induced myocardial perfusion abnormalities, has been encountered in patients with SCD.
- Arrhythmias, including a wide spectrum of supraventricular and ventricular arrhythmias and conduction abnormalities, are usually seen in patients with coexistent significant cardiac dysfunction, regardless of the underlying cause. In addition, myocardial iron overload has been recognized as a risk factor for the development of arrhythmias.

The basic **cardiac workup** of patients with haemoglobinopathies includes history taking, physical examination, resting electrocardiogram, chest radiogram and resting echocardiography. This workup should be performed regularly on an annual basis when no cardiac abnormalities are present, in shorter intervals in the presence of cardiac complications or when new symptoms suggestive of heart disease develop. In addition to basic workup, cardiac magnetic resonance imaging with determination of myocardial T2* relaxation time represents the gold standard to assess myocardial iron overload and guide iron chelation therapy in patients on regular transfusion therapy. Additional modalities including ambulatory electro-

Correspondence:
dimitrios_farmakis@yahoo.com

cardiography, stress testing and cardiac catheterization may be required in selected cases.

The **prevention and treatment** of cardiac complications in patients with hemoglobinopathies lies primarily on the proper management of the main disease itself with hemoglobinopathy-specific modalities that include regular blood transfusions and iron chelation in the case of -thalassemia and hydroxyurea or exchange transfusions in SCD. Cardioactive medications or other cardiac modalities are required in the presence of specific forms of heart disease. When heart failure with reduced LV ejection fraction develops, treatment with neurohormonal inhibitors, including angiotensin enzyme inhibitors or angiotensin receptor blockers, beta blockers and mineralocorticoid receptor antagonists, is required in combination with the intensification of the hemoglobinopathy-specific therapy.

References

1. Kremastinos DT, Farmakis D, Aessopos A, Hahalis G, Hamodraka E, Tsiapras D, Keren A. Beta-thalassemia cardiomyopathy: history, present considerations, and future perspectives. Circ Heart Fail 2010; 3:451-8.
2. Kremastinos DT, Farmakis D. Iron overload cardiomyopathy in clinical practice. Circulation 2011; 124:2253-63.
3. Farmakis D, Aessopos A. Pulmonary hypertension associated with hemoglobinopathies: Prevalent but overlooked. Circulation 2011; 123:1227-32.
4. Gladwin MT, Sachdev V, Jison ML, et al. Pulmonary hypertension as a risk factor for death in patients with sickle cell disease. N Engl J Med 2004; 350:886-95.
5. Gladwin MT, Vichinsky E. Pulmonary complications of sickle cell disease. N Engl J Med 2008; 359:2254-65.
6. Aessopos A, Farmakis D, Tsironi M, et al. Endothelial function and arterial stiffness in sickle-thalassemia patients. Atherosclerosis 2006; 191:427-32.
7. Aessopos A, Farmakis D, Karagiorga M, et al. Cardiac Involvement in Thalassemia Intermedia: A Multicenter Study. Blood 2001; 97:3411-6.
8. Aessopos A, Farmakis D, Deftereos S, et al. Thalassemia heart disease: a comparative evaluation of thalassemia major and thalassemia intermedia. Chest 2005;127: 1523-30.
9. Aessopos A, Kati M, Farmakis D. Heart disease in thalassemia intermedia: a review of the underlying pathophysiology. Haematologica 2007; 92:658-65.
10. Aessopos A, Farmakis D, Trompoukis C, et al. Cardiac involvement in sickle beta-thalassemia. Ann Hematol 2009; 88:557-64.
11. Anderson LJ, Holden S, Davis B, et al. Cardiovascular T2-star (T2*) magnetic resonance for the early diagnosis of myocardial iron overload. Eur Heart J 2001; 22:2171-9.
12. Aessopos A, Tsironi M, Vassiliadis I, et al. Exercise-induced myocardial perfusion abnormalities in sickle beta-thalassemia: Tc-99m tetrofosmin gated SPECT imaging study. Am J Med 2001; 111:355-60.
13. Kremastinos DT, Tiniakos G, Theodorakis GN, Katritsis DG, Toutouzas PK. Myocarditis in β-Thalassemia Major: A Cause of Heart Failure. Circulation 1995; 91:66-71.
14. Engle MA, Erlandson M, Smith CH. Late cardiac complications of chronic, refractory anemia with hemochromatosis. Circulation 1964; 30:698–705.
15. Kirk P, Roughton M, Porter JB, et al. Cardiac T2* magnetic resonance for prediction of cardiac complications in thalassemia major. Circulation 2009;120: 1961-8.

ARTICLE

Liver complications in thalassaemia syndromes

Farrukh T. Shah

Consultant haematologist, King Khalid University Hospital, Riyadh, Saudi Arabia; Whittington hospital, London

EUROPEAN HEMATOLOGY ASSOCIATION

Ferrata Storti Foundation

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Introduction

In the last few years there have been increasing reports of malignancies in in thalassaemia major (TDT) and non-transfusion dependant thalassaemia (NTDT), the majority of which have been liver tumours and this has prompted a renewed interest in iron overload and its impact on cancer risk (1-3). Liver disease is multifactorial in thalassaemia syndromes with iron overload, concurrent infection or past infection with Hepatitis C (HCV) , Hepatitis B (HBV), metabolic syndrome, lifestyle issues such as alcohol use and the possible immune modulation by blood transfusion potentially all playing a role (4-7).

Studies suggest an incidence of around 2% for development of Hepatocellular Carcinoma (HCC) in prospective studies in patients with thalassaemia (3) however, these are in the Italian population where there is a high prevalence of HCV; therefore the true risk of developing HCC is still unclear, particularly in the population without co-infection.

The role of iron in health and disease

Iron is an essential component of cytochrome enzymes, myoglobin and haemoglobin (8). Its role as an electron transporter is critical for cell homeostasis and it has an essential role in cell proliferation (9). Excess iron has a pro-oxidant effect that has been implicated in the development of liver cirrhosis, heart disease and diabetes (10, 11). More recent publications have shown a potential relationship between breast cancer prognosis and iron overload (12). In TDT iron overload develops rapidly as a consequence of blood transfusions, this iron is predominantly stored in the reticuloendothelial system (RE) and as iron overload worsens may spill into the hepatocytes. Recent histological studies have shown iron to also be distributed according to a periportal-to-pericentral pattern with a decreasing gradient in a large population of TDT patients suggesting that either there is an element of increased gastrointestinal iron absorption due to insufficient suppression of endogenous erythropoiesis or there is some degree of iron redistribution with chelation therapy (13). Conversely in NTDT, iron overload occurs more slowly and is predominantly via increased iron absorption through the gastrointestinal tract. The resultant iron is predominantly stored in the hepatocytes (14). This differential in both the rate and route of iron loading results in significant differences in the distribution of iron in end organs and ultimately affects the type of complications seen in patients with thalassaemia syndromes. In TDT endocrine and cardiac complications occur as a consequence of the iron overload at a relatively young age and long term liver cirrhosis is less commonly seen, presumably as a consequence of the predominantly RE site of iron storage which prevents more significant damage to the hepatocytes. In NTDT, cirrhosis is more commonly seen as a consequence of iron overload as well as HCC in a similar manner and frequency to Hereditary haemochromatosis (HH) (14).

The role of iron in liver damage and carcinogenesis

The liver is the main site for storage iron. In health, iron is transported around the body bound to transferrin and stored in the body either as hemosiderin or ferritin. Iron is present in the serum as transferrin bound ferric iron, however once transferrin becomes fully saturated then free iron is found in the form of non-transferrin bound iron (NTBI/LPI). Intracellular NTBI species can cause oxidative damage by generation of free radicals by the Haber- Weiss reaction. Reactive oxygen species (ROS) such as superoxide and hydroxyl radicals can be derived from electron leakage from the mitochondrial electron transport chain (15). The generation of ROS in the presence of iron overload is thought to be the primary way by which iron is likely to be carcinogenic.

There are a number of mechanisms by which iron may promote carcinogenesis including up-regulation of oxidative and inflammatory responses and potentially acting at either the promotion or progression stages of the carcinogenic process (10). Iron via ROS is therefore thought to have both an apoptotic and an anti-apoptotic effect in carcinogenesis.

Correspondence:

farrukh.shah@nhs.net

ROS once generated can result in genomic instability because it is thought to be the primary cause of iron induced base oxidation (16) and is known to alter the methylation pattern of a number of genes and cause a variety of DNA lesions (17). In particular iron is known to cause a higher frequency of G:C to T:A transversions at codon 249 of the p53 gene in liver of patients with HH (18). 8-Hydroxy-guanine (8-OHdG) is one of the most common lesions resulting from ROS and is increased in healthy individuals with higher iron stores (19) and also increased in amount in hepatocytes when iron loaded(20).

Apoptotic effects are primarily thought to be due to ROS induced lipid peroxidation and the formation of very reactive , unsaturated aldehydes (21, 22), which cause further oxidative damage by protein alkylation, mitochondrial dysfunction, altered nuclear signalling, inhibition of membrane transporters and further generation of ROS (23). This results in organelle damage, lysosomal leakage and ultimately cell death. The anti-apoptotic effects of ROS are thought to be partly due to its role in upregulating the NF- B signalling pathway, resulting in increased expression of cytokines, chemokines, cellular adhesion molecules, inflammatory enzymes and ROS induced DNA damage.

Liver complications in thalassaemia syndromes

Liver iron overload and cancer

In thalassaemia patients liver fibrosis is known to occur at liver iron values above 15 mg/g/dw,(24). The presence of fibrosis appears to be related to the degree of liver injury as measured by the transaminase levels and the presence of iron overload as documented by the ferritin and was found to be unrelated to HCV co-infection in multivariate analysis (13). However, the same study found using a mathematical model that progression to cirrhosis was more rapid in HCV positive patients (49 years) compared to non-infected patients (67 years). A later study confirmed that more severe fibrosis is seen in patients with HCV infection (25).

Liver iron therefore alone, can promote fibrosis and cirrhosis but the co-infection with HCV results in a more rapid progression to cirrhosis. Data from the Italian registry has shown an increasing incidence of liver cancer in thalassaemia patients; however of the 62 patients reported in the registry data only 4 patients, all of whom were NTDT, had never had an infection with HCV or hepatitis B. In addition, the median age at diagnosis of HCC was 48 years of age which suggested that HCC occurs in thalassaemia patients at a younger age then the general population rate of HCC (75 and 79 years for males and females, respectively). However, the Italian rate of HCC in HCV infected populations peaks in the 34-64 age cohort (26) and therefore in a similar population, the risk of HCC developing is probably in the correct age range and not occurring earlier than expected. The exact mechanism by which HCV causes HCC is not clear, but there is a known association with alcohol use, smoking and obesity. In addition, most HCC is known to occur in the presence of cirrhosis (26). Therefore, in TDT patients many years of iron overload resulting in significant transaminitis, would result in ROS mediated damage and development of fibrosis which may then may act as fertile ground for the progression to carcinogenesis, however the incidence of HCC is still not clearly defined in the HCV negative TDT population.

In NTDT the incidence of HCC is increased and from the Italian registry suggests a rate of around 1.75% compared to the general population rate of 0.1% (10/100,000); even if all HCV and HBV co-infected patients NTDT are excluded the rate remains high at 0.25% (4/1607) (1).

Hepatitis C

Infection with HCV appears to be a major risk factor in the development of HCC in TDT and NTDT. Many patients who were infected have not been treated for HCV or failed HCV eradication using interferon and ribavirin. The predominant genotype in Europe is genotype 1, middle east genotype 4 and in South Asia genotype 3 (27). The prevalence for HCV infection globally is 2% but there are wide variations according to regions with 31 countries accounting for around 80% of the HCV infections globally. The incidence in the high risk populations is therefore significantly higher and ranges between 4.4-87% (1, 28, 29)

Recent EASL guidelines recommend eradication of HCV in all patients with compensated or non-compensated chronic liver disease whether they have previously been treated or not (30).

Monitoring

Good monitoring is critical in identifying complications early and ensuring better outcomes.

In particular liver iron levels should be monitored regularly in both NTDT and TDT. Patients who are known to be cirrhotic should be regularly reviewed by a hepatologist and have liver ultrasound or screening by MRI or CT every 6 months along with alpha fetoprotein levels. They should also undergo surveillance for oesophageal varices at the time of diagnosis of cirrhosis and then every couple of years (31).

Conclusions

Hepatic iron overload is a serious complication associated with increased risk of fibrosis and liver cirrhosis. In the presence of HCV or HBV there is an increased association with HCC. In all patients the liver iron should be kept at the lowest possible levels (with normal transaminases) to reduce the risk of progression to fibrosis or cirrhosis. Patients should be advised about lifestyle factors that impact on progression to cirrhosis such as alcohol, obesity and smoking. All patients with HCV, HBV, documented fibrosis or cirrhosis should be reviewed regularly by a hepatologist.

References

1. Borgna-Pignatti C, Garani MC, Forni GL, Cappellini MD, Cassinerio E, Fidone C, et al. Hepatocellular carcinoma in thalassaemia: an update of the Italian Registry. British journal of haematology. 2014;167(1):121-6.
2. Voskaridou E, Ladis V, Kattamis A, Hassapopoulou E, Economou M, Kourakli A, et al. A national registry of haemoglobinopathies in Greece: deducted demographics, trends in mortality and affected births. Ann Hematol. 2012;91(9):1451-8.
3. Mancuso A, Sciarrino E, Renda MC, Maggio A. A prospective study of hepatocellular carcinoma incidence in thalassemia. Hemoglobin. 2006;30(1):119-24.
4. Burrows L, Tartter P, Aufses A. Increased recurrence rates in perioperatively transfused colorectal malignancy patients. Cancer detection and prevention. 1987;10 (5-6):361-9.
5. Aquina CT, Blumberg N, Becerra AZ, Boscoe FP, Schymura MJ, Noyes K, et al. Association Among Blood Transfusion, Sepsis, and Decreased Long-term Survival After Colon Cancer Resection. Annals of surgery. 2016.
6. Lannan KL, Sahler J, Spinelli SL, Phipps RP, Blumberg N. Transfusion immunomodulation--the case for leukoreduced and (perhaps) washed transfusions. Blood cells, molecules & diseases. 2013;50(1):61-8.
7. Kew MC. Hepatocellular carcinoma: epidemiology and risk factors. Journal of hepatocellular carcinoma. 2014;1:115-25.
8. Andrews NC. Iron metabolism: iron deficiency and iron overload. Annual review of genomics and human genetics. 2000;1:75-98.
9. Andrews NC. Disorders of iron metabolism. The New England journal of medicine. 1999;341(26):1986-95.
10. Okada S. Iron-induced tissue damage and cancer: the role of reactive oxygen species-free radicals. Pathology international. 1996;46(5):311-32.
11. Hanson EH, Imperatore G, Burke W. HFE gene and hereditary hemochromatosis: a HuGE review. Human Genome Epidemiology. American journal of epidemiology. 2001;154(3):193-206.
12. Pinnix ZK, Miller LD, Wang W, D'Agostino R, Jr., Kute T, Willingham MC, et al. Ferroportin and iron regulation in breast cancer progression and prognosis. Science translational medicine. 2010;2(43):43ra56.
13. Prati D, Maggioni M, Milani S, Cerino M, Cianciulli P, Coggi G, et al. Clinical and histological characterization of liver disease in patients with transfusion-dependent beta-thalassemia. A multicenter study of 117 cases. Haematologica. 2004;89(10):1179-86.
14. Origa R, Galanello R, Ganz T, Giagu N, Maccioni L, Faa G, et al. Liver iron concentrations and urinary hepcidin in beta-thalassemia. Haematologica. 2007;92(5):583-8.
15. Halliwell B, Gutteridge JM. Role of free radicals and catalytic metal ions in human disease: an overview. Methods Enzymol. 1990;186:1-85.
16. Puppo A, Halliwell B. Formation of hydroxyl radicals from hydrogen peroxide in the presence of iron. Is haemoglobin a biological Fenton reagent? The Biochemical journal. 1988;249(1):185-90.
17. Pra D, Franke SI, Henriques JA, Fenech M. Iron and genome stability: an update. Mutation research. 2012;733(1-2):92-9.
18. Hussain SP, Raja K, Amstad PA, Sawyer M, Trudel LJ, Wogan GN, et al. Increased p53 mutation load in nontumorous human liver of wilson disease and hemochromatosis: oxyradical overload diseases. Proceedings of the National Academy of Sciences of the United States of America. 2000;97(23):12770-5.
19. Nakano M, Kawanishi Y, Kamohara S, Uchida Y, Shiota M, Inatomi Y, et al. Oxidative DNA damage (8-hydroxydeoxyguanosine) and body iron status: a study on 2507 healthy people. Free radical biology & medicine. 2003;35(7):826-32.
20. Morel I, Hamon-Bouer C, Abalea V, Cillard P, Cillard J. Comparison of oxidative damage of DNA and lipids in normal and tumor rat hepatocyte cultures treated with ferric nitrilotriacetate. Cancer letters. 1997;119(1):31-6.
21. Halliwell B, Chirico S. Lipid peroxidation: its mechanism, measurement, and significance. Am J Clin Nutr. 1993;57(5 Suppl):715S-24S; discussion 24S-25S.
22. Picklo SMJ, Montine TJ, Amarnath V, Neely MD. Carbonyl Toxicology and Alzheimer's Disease. Toxicology and Applied Pharmacology. 2002;184(3):187-97.
23. Del Rio D, Stewart AJ, Pellegrini N. A review of recent studies on malondialdehyde as toxic molecule and biological marker of oxidative stress. Nutrition, Metabolism and Cardiovascular Diseases. 2005;15(4):316-28.
24. Angelucci E, Muretto P, Nicolucci A, Baronciani D, Erer B, Gaziev J, et al. Effects of iron overload and hepatitis C virus positivity in determining progression of liver fibrosis in thalassemia following bone marrow transplantation. Blood. 2002;100 (1):17-21.
25. Di Marco V, Capra M, Gagliardotto F, Borsellino Z, Cabibi D, Barbaria F, et al. Liver disease in chelated transfusion-dependent thalassemics: the role of iron overload and chronic hepatitis C. Haematologica. 2008;93(8):1243-6.
26. Bosetti C, Turati F, La Vecchia C. Hepatocellular carcinoma epidemiology. Best practice & research Clinical gastroenterology. 2014;28(5):753-70.
27. Gower E, Estes C, Blach S, Razavi-Shearer K, Razavi H. Global epidemiology and genotype distribution of the hepatitis C virus infection. Journal of hepatology. 2014;61(1 Suppl):S45-57.
28. Mirmomen S, Alavian SM, Hajarizadeh B, Kafaee J, Yektaparast B, Zahedi MJ, et al. Epidemiology of hepatitis B, hepatitis C, and human immunodeficiency virus infecions in patients with beta-thalassemia in Iran: a multicenter study. Archives of Iranian medicine. 2006;9(4):319-23.
29. Wonke B, Hoffbrand AV, Brown D, Dusheiko G. Antibody to hepatitis C virus in multiply transfused patients with thalassaemia major. Journal of clinical pathology. 1990;43(8):638-40.
30. EASL Recommendations on Treatment of Hepatitis C 2016. Journal of hepatology. 2017;66(1):153-94.
31. Yardumian A, Telfer PT, Shah F, Ryan K, Darlison M, Miller E, et al. Standards for the clinical care of children and adults with thalassaemia in th UK. 3rd edition ed: UKTS; 2016 2016.

The use of e-Registries in building and upgrading services

Michael Angastiniotis,[1] Zinonas Antoniou,[2] E.C. Schiza,[2] Costas P. Pattichis,[2] Christos N. Schizas[2]

[1]Thalassaemia International Federation; [2]University of Cyprus, Department of Information Technology, Cyprus

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ABSTRACT

This paper presents an electronic registry system for the purposes of the rare congenital conditions that require lifelong follow up and treatment. The main objective of the eRegistry focusses on the prevention of major rare anaemias (RAs) by facilitating the access, at a European level, to the best genetic counselling, diagnosis and clinical management of the patients with RA independently of their country of origin. This can be achieved by promoting an extension of the full Electronic Health Record system and specifically the electronic registries for RAs, across Europe for the purposes stated hence promoting service development for the benefit of patients. The eRegistry will serve as an epidemiological tool to improve the management of patient services and ultimately improve patient care.

Correspondence:

michael.angastiniotis@thalassaemia.org.cy

Introduction

Thalassaemia and Sickle cell disorders are complex lifelong haematological disorders which are complicated over time with multi-organ involvement. Effective management requires many resources which the health services of each country are called upon to provide, if patient survival and wellbeing are to be safeguarded. The elements of management include services such as those listed in Table 1.

The need for complex services make it essential to understand the real burden of disease and this is an important issue for budgetary and public health planning [1,2]. Registries, health records and other databases are essential tools for gathering information, which help to define the epidemiology, clinical outcomes and the natural history of these rare conditions. Such information will help to improve quality of care and to plan services, as well as to assist in research projects including clinical trials and the recruitment of volunteer patients. Policies concerning reference centres, networking and cross-border health, make the development of registries at healthcare facility, national and international level, necessary tools to facilitate the creation and implementation of these policies.

The Thalassaemia International Federation, through a European project with the title eEnerca, has developed a rare anaemias registry. In its development, EU directives were studied, including the current state of the art for standards and legal issues which concern data ownership and confidentiality, incorporating the necessary safeguards for privacy in the electronic registry.

It's important to mention that Health, Demographic Change and Wellbeing is under the Horizon 2020 ambitious Work Programme and focus on the improvement of healthcare for the benefit of the citizen. This approach relies on the philosophy that citizens are the owner of their own medical records. Thus eHealth vision relies on the patient-centred philosophy, meaning the Electronic Health Record (EHR) of the citizen needs to be interoperable with the eRegistry and other systems. An integrated and structured EHR environment yields many benefits, such as better management of resources, improved care coordination, chronic disease management, national and worldwide access of medical data and the resolution of interoperability issues, elimination of medical errors and delays, reduced operational costs, personalized prescription, and patient involvement in their treatment. Interoperability is a very important functionality that an EHR management system should offer to the healthcare providers and the citizen. Hence guar-

antee instantaneous updating of the content of EHR and eRegistries when a change takes place. This will secure that medical professionals will have always readily available the latest data concerning the citizen under examination. The eRegistries need to rely on eHealth EU Interoperability Framework (EIF) and this will help expand the market for healthcare [3].

Having an interoperable eRegistry system, a doctor can share information in a harmonized way with colleagues and other experts in red cell haematology in Europe and other countries. This will contribute to the national or European registry and have access to relevant clinical information in a timely manner.

eRegistries: a by-product of the electronic health record

A registry is "an organised system that collects and stores information in the form of unified data, to evaluate specified outcomes for a population defined by a particular disease".

A registry is different from an index or a simple list, since it contains more extensive information. However, it does not collect all clinical information, and so it is not an electronic health record. An electronic registry, compared to paper based documents, allows complex data to be analysed and shared by several institutions. Its quality depends on the quality of data collected. Such eRegistries can be very easily derived from an eHealth repository or built independently if eHealth environments are not fully developed in a country or region as can be seen in Figure 1.

A registry should also follow legal and ethical requirements, which must be adopted by each national agency. Such requirements include the respect for privacy and confidentiality of patient information. EU Directives 96/9/EC and 91/250/EEC state the legal requirements for building and operating medical databases.

In short, a registry is understood to be a collection of

Table 1. Services required for patient care in red blood disorders.

Blood adequacy and safety which involves the blood transfusion services.
Essential drugs.
Technology is essential (*e.g.* MRI to assess iron overload) and its development and implementation requires health technology assessment (HTA) to guide policy decisions.
Day care units for blood transfusions and outpatient clinics.
Multidisciplinary care and Psychosocial support.
Reference centres and networking are policies still under development and eHealth support for networking is still not offered in many centres.
There is need for educational activities such as ePlatforms, publications and workshops which need financial support.
Epidemiological studies (geomapping, micromapping and, disease surveillance) are still not used to support policy and service development.

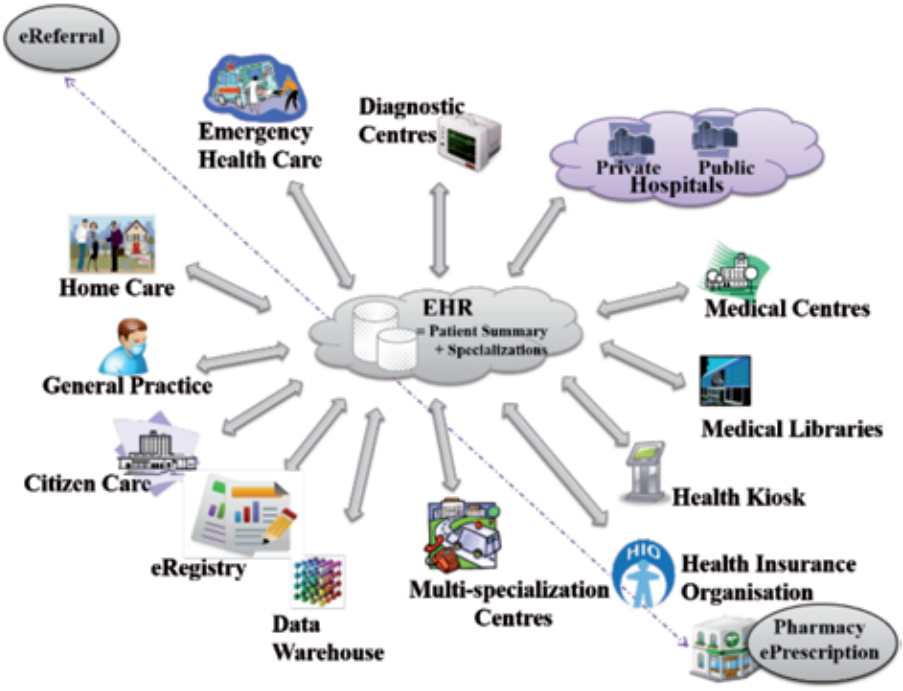

Figure 1. eRegistry system communication in an eHealth environment.

standardised data and information concerning patients with a common disorder [2].

Uses of patient registries in public health

- The first need is to know the number of patients affected by a particular disorder. The number of patients recorded should be as near to the total number in existence in a particular population, allowing the collection of the most accurate data possible. Such data may lead to the creation of a broad geomap, country by country. However, in order to serve need for service development, finer locating of patients, recording distance from the nearest reference centre, will eventually assist in developing both centres of excellence and a network linking reference and secondary centres, as described in the Enerca White Book [1].
- A registry should monitor incidence over time as well as prevalence.
- Outcomes such as death rates, complication frequencies, and survival figures, should also be derived from any registry designed for epidemiological purposes. Other examples of data analysis, include the age distribution of the patient population (or cohort), and patient reported outcomes.
- The support for networking between centres, is particularly important in the case of rare diseases for which multi-centre collaborations for clinical research but also for diagnostic and clinical support by experts, is essential because of the often reduced experience of peripheral centres in such diseases and are essential for quality care. A disease is defined as rare according to the European definition is one that occurs in 5 individuals in 10000 of the population. There are many such conditions and in many countries, the haemoglobin disorders fall into this category. For the purposes of providing optimum care and adopting prevention policies, electronic registries are essential tools for public health services.
- Facilitate teleconsultation, network of experts operation, intelligent systems applications (diagnosis, prognosis, monitoring, and prevention).
- Can be easily adopted by countries that have no electronic systems for patient care implemented.
- When properly anonymised the data can be integrated in a cohort and used for research purposes and demographic analyses.

Such data are important not only for academic recording but also for health planning, such as manpower planning, locating services and budgeting.

Registries can also be designed to support auditing a service, for clinical trials and other research. In this respect patient sub-groups can be identified.

In the case of haemoglobin disorders for which complex services are required (see Table 1), the organisation of data electronically is a major tool in making sense of the clinical data collected over many years.

Developing a registry for rare diseases

For the purpose of networking, it is useful for registries to be designed in a manner that facilitates communication between centres. This requires standardisation, which has been studied by various agencies in Europe and in North America. There has been agreement on the general content of a rare disease registry. In a joint declaration by rare disease organisations, EURORDIS-NORD-CORD, 10 key principles for the development of rare disease registries have been defined, and in developing the Enerca-TIF registry, it was decided to adopt and to include these principles in the design:

1. The registry is a global priority for 'increasing knowledge on rare diseases, pooling data for epidemiological research and clinical research'. This means that enough information must be gathered to satisfy these requirements.
2. The registry should encompass the widest geographic scope as possible. This is recognised to go beyond the boundaries of any single country, as expressed in the International Rare Disease Research Consortium (IRDiRC), which advocates worldwide sharing of information, data and samples by robust and harmonised registries. The registry could, therefore, be offered to as many countries possible, especially to those who wish to network.
3. The registry is centred on a group of diseases – the rare congenital anaemias, including thalassaemia and sickle cell disease, will benefit from the implementation of a national as well as local registries.
4. Interoperability is a basic principle. To facilitate this process, interoperability standards recognised internationally are being utilised.
5. A minimum set of Common Data Elements of demographic information: These elements should be consistently used in all rare disease patient registries, so that patient identification and location can be shared. There are several examples of common data elements, developed independently or in collaboration by various agencies and projects. One dataset agreed in the European setting can be found in the following document: "Guidelines on patient summary set of data for electronic exchange under the cross-border directive" - This list of data is based on the minimal dataset recommended by the European Health Ministerial Conference which was held in Dublin in 2013 with similar data adopted by the EPI-RARE project [4, 5].
6. Linking with a corresponding biobank data.
   Linking with a biobank will not be a difficult undertaking but following international standards requires expertise and practical experience.
7. Patient reported data.
   Patient reports on the experiences and quality of life are important components of registry. NORD (National Organization of Rare Diseases) in the US is developing a system for patients and their families to enter their own data.
8. Public-private partnerships to ensure sustainability, which is a major concern for any registry which hopes to be used over many years.
9. Patients as stakeholders in the governance of the registry: In the West, the patient is the owner of his/her health data, which can only be used with his consent.
10. Registries must also be instruments to empower patient communities.

The data included in the registry for the anaemias

For the registry design and development, the current standards and legal issues which concern data ownership and confidentiality have been taken into consideration,

incorporating the necessary safeguards in the electronic registry.

As a result, a registry was developed, primarily to aim of epidemiological surveillance and the secondary to facilitate clinical research. The registry includes the following fields, arranged in a modular manner:

- A minimal dataset based on the needs of networking (cross border health) as well as the needs of collecting epidemiological data on rare anaemias. This includes mostly demographic data,
- An emphasis on the diagnosis and the accuracy of the diagnosis. The diagnosis is coded using an autocomplete mechanism for both ICD10 code and the Orphacode.
- Annual clinical summaries with patient data from patients with transfusion dependent and milder anaemias and from patients with sickle cell syndromes are collected separately because of the different manifestations and complications.
- Patient outcome measures are encountered in order to assess the effectiveness of the service,
- Aggregate data to support overall European or international epidemiological information,
- Statistical assistance for data analysis. This module facilitates the research, both epidemiological and clinical including an extensible set of queries.

The principles of interoperability and extendibility were considered in the design.

In order to gather data from several centres, nationally and internationally if required, the registry is designed with respect to interoperability guidelines. This means facilitating each local registry to share their data with other centres, if of course they so wish. This would serve the primary purpose of the registry, which is epidemiological mapping of the rare anaemias across countries. This makes it necessary to use a generalized approach in order to represent every conceivable kind of data structure in a consistent way. The Institute of Electrical and Electronics Engineers defines interoperability as the "ability of two or more components to exchange information and to use the information that has been exchanged" [6]. Interoperability for health information systems requires accurate and consistent data exchange and use of the information that has been exchanged. This includes syntactic interoperability as the ability to exchange data and semantic interoperability as the ability to understand exchanged data. These are the core constructs of interoperability and must be present in order for registries to share data successfully [7, 8]. Additionally, care must be taken to ensure that integration efforts comply with legal and regulatory requirements for the protection of patient privacy.

The reason for making the registry extendible by means of adding modules, especially to capture clinical data, is to increase the possibility of the registry being used for research purposes and for recruitment for clinical trials. A simple registry may be expanded by adopting a modular architecture so that it may be further developed, even to become a full electronic patient clinical record

Functionality of the registry takes into consideration the primary aims (epidemiology, service development) but also offers the option of adding data collection on clinical aspects for secondary aims. Basic clinical outcome information is part of epidemiology, but by expansion, it is understood to collect longitudinal data on the progress of the condition in individual patients.

It is observed that in many cases, data entry may be difficult because of physician reluctance or legal objections to data sharing. This is a common difficulty encountered by all registries.

Ethical and legal considerations in creating the database

Medical information exchange has always been a sensitive subject due to the highly confidential nature of health information. Besides having means to identify a patient, facilities to identify a health professional or health care provider organisation is also a requisite. There is a need for maintaining confidentiality of medical information and protecting the patients' privacy. Information should be exchanged in a secure manner when shared between health professionals and health care provider organisations. This is achieved by creating a patient, but also a health professional/health care provider organisation identifier, which is coupled to a digital identity, and is issued by a certified authority. This identifier provides a base to create a trust circle between health professionals/health care provider organisations and is also a precondition for electronic signing by the health professional/health care provider organisation. Regarding the role of each user of the eRegistry, different authentication accesses is provided, having different functionalities.

According to the British Medical Association: "The physician must maintain secrecy on all he knows. In this general principle, however, there are a few exceptions, releasing the doctor from the duty of confidentiality. These are:

- when the patient gives his consent,
- when it is in the interest of patients, it overrides the doctor's duty to secrecy,
- for research purposes but only approval by the Ethics Committee for Clinical Research and
- the information required for legal procedures" [9, 10].

Violations of this right can destroy the trust that is a fundamental condition between doctor and patient.

The patients should be well informed on the purpose of their registration and give written consent for their inclusion. Also they should be informed of any additional uses of the registry, if they change from the original use.

Conclusions

A registry of patients suffering from a rare disease, is an essential tool in knowing and understanding the burden of disease. This in turn allows a knowledge based planning for patient care, such as locating services and manpower development, based on facts. Registries are designed for a specific purpose, such as epidemiology, but can be modified if required. Another important use is the networking and collaboration between centres so that interoperability is aspect.

Ethical and legal principles should be respected, mainly with respect to privacy, and safeguards should be incorporated in the design of the registry.

References

1. ENERCA recommendations for centres of expertise in rare anaemias: a White book. Published by the Enerca project. 2014.
2. Gliklich R, Leavy MB. Patient registries for understanding the natural history of diseases. iHealth Connections, 2011;1(1):12-15.
3. Schiza EC, Neokleous KC, Petkov N, Schizas CN. A patient centered electronic health: eHealth system development. Technology and Health Care. Technol Health Care. 2015;23(4):509-22. doi: 10.3233/THC-150919.
4. www.epsos.eu
5. Taruscio D, Mollo E, Gainotti S, Posada MP, Bianchi F. The EPIRARE proposal of a set of indicators and common data elements for the European platform for rare disease registration. Archives of Publ Health. 2014; 72:35.
6. Institute of Electrical and Electronics Engineers (IEEE). IEEE Standard Computer Dictionary: A compilation of IEEE Standard Computer Glossaries. New York, NY, 1990.
7. Vittozzi L, Gainotti S, Mollo E, Donati C, Taruscio D. A model for the European platform for rare disease registries. Publ Health Genomics. 2013; 16(6): 299-304.
8. Santoro M, Coi A, Lipucci Di Paola M, Bianucci AM, Gainotti S, et al. Rare disease registries classification and characterisation: a data mining approach. Publ Health Genomics. 2015; 18(2): 113-22.
9. Moskop JC, Marco CA, Larkin GL, et al.: From hippocrates to HIPAA: privacy and confidentiality in emergency medicine: Part I. Conceptual, moral and legal foundations. Ann Emerg Med 2005, 45:53-9.
10. Geiderman JM, Moskop JC, Derse AR: Privacy and confidentiality in emergency medicine: obligations and challenges. Emerg Med Clin North Am 2006, 24:633-56.

Further reading

1. Glicklich RE, Dreyer NA, Leavy MB. Registries for evaluating patient outcomes: a user's guide. AHRQ Publication no. 07-EHC001-1 (www.effectivehealthcare.ahrq.gov)
2. Richesson RL, Shereff D, Andrews JE. Standardisation of questions in rare disease registries: the PRISM library. Interact J Med Res. 2012; 1(2): e10.
3. Thompson R, Johnston L, Taruscio D, Monaco L, Beroud C et al. RD-Connect: an integrated platform connecting databases, registries, biobanks and clinical bioinformatics for rare disease research. J Gen Intern Med. 2014; suppl 3: s780-7.
4 Muschol M, Lablans M, Wagner OFT, Uckert F. OSSE – open source registry software solution. Orphanet J Rare Dis. 2014; 9(suppl 1): 09.
5. EUCERD Minimum data set for rare disease registries January 2015 www.eucerd.eu
6. Coding of rare diseases in health information systems with Orphacodes. Joint Research Centre & EUCERD Joint Action Workshop Report. Ispra, 1-2 October 2014 www.eucerd.eu
7. Summar ML. Registries for Rare Diseases: involve the patient. www.medscape.com/viewarticle/837851

ARTICLE

Experience of thalassaemia care in Saudi Arabia

Zakaria M. Al Hawsawi,[1] Mohammed A. Alaidros,[2] Ahmed M. Tarawah,[3] Waheed A. Turkistani,[3] Mohamed A. Zolaly,[4] Abdulhadi M. Habib[5]

[1]Consultant Pediatric Haematology Oncology, Clinical Assistant Professor, College of Medicine, Taiba University, Madina Munawara, Kingdom of Saudi Arabia; [2]Pediatric Haematology Oncology Registrar, Pediatric Department, Madina Maternity and Children's Hospital, AlMadina AlMunawara, Kingdom of Saudi Arabia; [3]Consultant Pediatric Haematology Oncology, Pediatric Department, Madina Maternity and Children's Hospital, AlMadina AlMunawara, Kingdom of Saudi Arabia; [4]Consultant Pediatric Haematology Oncology, Associate Professor, College of Medicine, Taiba University, AlMadina AlMunawara, Kingdom of Saudi Arabia; [5]Consultant Pediatric Endocrinology, National Guard Hospital, AlMadina AlMunawara, Kingdom of Saudi Arabia

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Introduction

Thalassemia is a hereditary blood disease characterized by the absence or reduction in synthesis of -globins chains of the normal hemoglobin resulting in an imbalance between alpha and beta chains and consequent ineffective erythropoiesis and hemolysis.

The molecular basis of -thalassemia in various Arab countries reveals the presence of 52 mutations (1) , which are mostly of Mediterranean and Asian origin. In Saudi Arabia 14 mutations were identified and the commonest of which is IVS-1-110 (G-A), condons 39 (C-T) and IVS11-1 (G-A) (2)

Al Madinah Thalassemia Center was established in 1992 (3) at Madina Maternity and Children's Hospital.

Madinah Almunawara is a holy city at KSA with estimated population of 1.5 million with estimated gene frequency of -thalassemia of 0.1% (4) . The center provides comprehensive Thalassemia care to all Thalassemia and other transfusion dependent hemoglobin disorders in the region with age limit up to 20 years.

The aim of this study is to demonstrate our experience in managing Thalassemia patients and to describe the outcome of comprehensive Thalassemia care in improving adherence of patients to iron chelation therapy.

Methods

The medical records of 135 transfusion dependent patients with Thalassemia and sickle cell disease followed at Madinah Thalassemia Center were retrospectively reviewed from 2008 to 2013, 110 Thalassemia patients and 25 patients with sickle cell disease were enrolled in the study, the age range was 6 months to 20 years. The comprehensive care includes regular blood transfusion at 2-4 weeks intervals with the use of desferrioxamine, deferiprone and defrasirox, it also includes multidisciplinary care through joint clinic with endocrinologist and other specialties.

In 2007 general survey for all patients was done through questionnaire to evaluate patients medical knowledge and their adherence to iron chelation therapy.

Steps for improvement of adherence to iron chelation therapy was started on 2008 which includes:

1. Patients and parents education
2. To provide iron chelator to all patients
3. To minimize drugs adverse reaction
4. Social support

Results

135 patients were included in the study, 80% of the patients had severe iron overload with serum ferritin level more than 3000 along with very poor adherence to iron chelator, also the medical knowledge about the disease was very poor at 2007.

Result of implementation of the program from 2008 to 2013 showed 65% of patients had serum ferritin less than 2500 with improvement in adherence to iron chelator therapy.

Discussion

Our previous study in the Madinah region which reviewed the care over 10 years from 1992 to 2002 showed poor Thalassemia care with lack of comprehen-

Correspondence:
zhawsawi@hayoo.com

sive Thalassemia and poor adherence to iron chelation therapy. Our study demonstrated the importance of patients education and social support in improving adherence to iron chelation therapy.

With implementation of improved adherence program, significant improvement of patients compliance occurs and patients with serum ferritin less than 2500 increase from 30% to 65%.

Also this study highlight our experience with combined hematology and endocrine clinic indicated the, this model of care has potential benefits.

Patients were assessed by endocrinologist, hematologist, Gynecologist and social worker on the same day in the environment where they receive their regular blood transfusion, from this joint clinic we were able to evaluate endocrine complication among our patients (5) .

Detail improvement adherence program for iron chelation and its outcome will be discussed in the conference presentation.

Conclusions

Improvement of adherence to iron chelation therapy can be achieved with:

1. Comprehensive thalassemia care
2. Multidisciplinary care through joint clinic
3. Continuous patients and parents education
4. Continuous social support.

References

1. Zahed L. The spectrum of -thalassemia mutation in Arab population. J Biomed Biotech 2001; 1: 129-132
2. Hasounah FH, Sejency SA, Omer JA, Old JM, Oliver RWA. Spectrum of -thalassemia mutation in the population of Saudi Arabia. Hum hered 1995; 45:
3. Alhawawi ZM, Ismail GA, Al Harbi HA, Alsubhi ZR. -thalassemia major . Saudi medj 2003; 24:1027 – 1029
4. Elhazmi MAF. Hemoglobin disorders, a pattern for thalassemia and hemoglobinopathie in Saudi Arabia. Acta Hematol 1982; 68: 43-51.
5. Habib AM, Alhawsawi ZM, Marsy MM, Alharbi AM, Osilan AS, Almagamsi MS, Zolaly MA. Endocrinopathies in beta Thalassemia major. Saudi medj 2013; 34: 67-73.

ARTICLE

Reference centres and networking. Experiences from the region: Pakistan

Mohammed Adil Akhter

Member, Medical Advisory Board, Thalassaemia Federation of Pakistan, Executive Member, Pakistan Pediatric Association, Punjab, Associate Professor of Pediatrics, Fatima Jinnah Medical University, Sir Ganga Ram Hospital, Lahore, Punjab, Pakistan

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The history of thalassaemia care before the 1990's in Pakistan was mainly based on transfusing blood and not much else. True holistic care in Pakistan started in the Pediatric department of Sir Ganga Ram Hospital, Lahore under the supervision of Prof. Dr. Jovaria Mannan in 1991. As thalassaemics flocked to this department, it was soon realized that they would need a center where they can feel safe and get the multi-disciplinary care they deserve. In 1994 along with Prof. Dr. Yasmin Raashid, a gynecologist, the Thalassaemia Society of Pakistan was formed.

With a daunting population of over 200 million and a gene frequency of Beta-thalassaemia of 5-8%, the enormity of the healthcare burden was too large to even imagine. Multiple satellite thalassaemia care centers, individually catering for these patient's blood transfusion requirements was less than ideal. But the Thalassaemia Society of Pakistan networked with Sir Ganga Ram Hospital, Lahore was able to provide quality multi-disciplinary care to them.

As years went by and thalassaemics were able to scale to the base camp of K2, at a height of 5650meters/18,645feet, it was realized that optimal care for thalassaemics in Pakistan can be possible, especially in the form of centers associated with tertiary care teaching hospitals across the country.

As it was soon found, there are many satellite Non-Governmental Organizations (NGO's) catering for thalassaemia across the country with varying quality of care. The need to consolidate care and bring these NGO's under one umbrella was felt to be essential as this would ensure that children with thalassaemia across the country would get quality care no matter where they lived. This led to the formation of the Thalassaemia Federation of Pakistan http://tfp.org.pk in 2005, a platform of now more than 43 NGO's across Pakistan.

The Thalassaemia Federation of Pakistan acted as a catalyst of change in the mindset of healthcare providers towards thalassaemics. Awareness was created, networks were formed with private NGO's as well as Government Healthcare centers. These partnerships across the country led to NGO's creating a voice for thalassaemic patients, healthcare providers and parents on one platform.

Prevention programs started to bloom in different provinces because of the voice of the Thalassaemia Federation of Pakistan and its provincial chapters. This led to the formation of the largest prevention program in Asia as the Punjab Thalassaemia Prevention Programme http://ptpp.punjab.gov.pk/.

The Sir Ganga Ram Hospital, Lahore is currently the main reference center for the Management and Prevention of Thalassaemia in Punjab. This center in coordination with the Thalassaemia Federation of Pakistan has conducted numerous training sessions across the country. It houses the Punjab Thalassaemia Prevention Programme and thus offers management and preventative services under one roof. It coordinates activities across the province and the country.

The Federation did not stop there, it further developed networks with the Safe Blood Transfusion Programme, Government of Pakistan http://stbp.gov.pk. This would help ensure that even thalassaemics in the rural parts of the country would have access to safe blood.

Healthcare in Pakistan is primarily consistent with a pay out of pocket model. Over the years the Federation has approached the governments to provide iron chelation medication free of cost to patients across the country. Although some Government hospitals do provide the drugs, the number of patients surpasses the capability of the government to cater for a majority of thalassaemic patients.

When the Pakistan Bait-ul-Mal http://pbm.gov.pk , an autonomous institiution set up to provide social protection to the poor marginalized segments of the society, planned a thalassaemia center, they approached the Federation. Experts from Lahore, Peshawar, and Rawalpindi advised them to follow the Sir Ganga Ram Hospital, Lahore – Thalassaemia Society of Pakistan model. This would ensure optimal care for thalassaemics as well as help develop a multi-disciplinary team of experts to cater for these patients.

Over all in Pakistan this is an ongoing process of developing reference centers across the country and further networking between thalassaemics, parents, healthcare providers, NGO's, government institutions, blood transfusion authorities, health regulatory authorities and autonomous institutions to improve the lives of thalassaemics.

Correspondence:
dradilakhter@gmail.com

Patient's rights: legislation globally and in the region

Mahmoud Hadipour Dehshal, Mehdi Tabrizi Namini

Blood Transfusion research center, High Institute for Education and Research in Transfusion Medicine, Tehran, Iran

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ABSTRACT

Formulated in 1948, the human rights declaration insists on the equity of all mankind. The concept has been developed into different areas such as patient rights. The concept of patient rights cannot be unanimously understood in different cultures. "Patient rights" is attached importance to in all countries located in the Middle East. However, there are reports showing the fact that respecting patient rights is not acceptable in the region. In the article we aim to have an overlook at the concept of patient rights and find a way towards the concept through sharia law.

Introduction

The human rights declaration ratified in 1948 emphasizes on equality of all mankind.[1] The concept of human rights has developed much since then affecting the rights of the more vulnerable walks of people including patients. The understanding and perception of the patient rights differs across countries and cultures strongly intertwined with social norms.[2] Whatever the perceptions are, the models of patient rights stress on the effective relations between physicians and health care providers. The best model to ensure patients' rights is the one which provides patients with adequate and complete information about their condition and treatment options and ensure their rights in decision making. In this model, the patient is considered to be the consumer of the health services that deserves most to know about what they should. The patients' rights are not limited to the decision making faculty; it entitles the patient with the rights of privacy and the treatment refusal. The consensus on the patients' rights paves the ground for moral and legal wise treatment whereby the human virtue of patients is recognized.[3]Knowledge about the variations and delicacies in different cultures can lead to the adoption of constructive decisions in the improvement of patients' rights.

Overview of patients' rights

The general notion of patient rights is depicted at Figure 1.

The step-one essentials of the patient rights

A) The patient shall be entitled the access to correct and complete information on their condition and health record;
B) The patient in case finding a part of health information or the application of a certain means of treatment partially ambiguous shall be entitled to question accordingly;
C) The patient shall have the right to inform caregivers on the treatment denial;
D) The patient shall sign the informed consent in which the type of care, other alternative options, the positive treatment outcomes, and the care side effects are emphasized;
E) Care providers shall respect the patients' rights.[4]

The step-two essentials of the patient rights

A) The health condition of the patient shall be assessed and the full treatment be provided;
B) The patient shall be entitled the supportive care including the adequate pain relief if deemed appropriate;
C) The care process shall abide by the scientific based standards;

Correspondence:
mehregan_hadipour@yahoo.com

D) During the course of treatment, the communication between the care givers and patients shall be in plain and clear language;
E) During the course of treatment, any changes in the care process shall be communicated with the patients with their subsequent consent;
F) The patient is entitled to know about the care expenses.[5]

The step-three essentials of the patient rights

A) The patient information shall be kept confidential and not to be disclosed without authorization;
B) The access of other care givers to the patient information for better care management is authorized after the patient notification;
C) The communication of the patient information is authorized just among the care givers involved in treatment;
D) The use of the patient information for research purposes is not allowed unless authorized otherwise by the patient.[6]

The step-four essentials of the patient rights

A) Care should not be aborted incomplete for non-medical reasons;
B) The end of the care period shall not differ from the previously informed time limit;
C) Before the care attempt, all care necessities shall be available;
D) The patient is entitled to refuse the medical treatment at any time during the process;
E) The medical practitioner is obliged to follow up patients medically appropriately long enough after the end of procedures like surgeries;
F) The physician feeling unable to continue their practice shall relegate the treating position to a new substitute or to refer the patient to an eligible alternative doctor.[7]

The patients' rights from the perspective of the Islamic laws

The Islamic Sharia law emphasizes on the right to properties and persons.[8] Based on the sharia law, man has the freedom of choice to decide on their own care which is rendered null and void in the absence of access to adequate information. In Islam context of law, the right to decision making about the care is assigned to the relatives just in case of immature persons or the ones suffering from mental incompetency or retardedness or the altered mental status and the ones lacking discretion like the alcoholic addicts or mentally damaged patients. This list of exclusion is similar to the ones ratified in the western countries.[9]Confidentiality has been firmly emphasized on in Islam religion. Imam Ali has denounced the disclosure of people secrets as disloyalty and has condemned it[10]; just in rare cases and based on the law mandate or the patient consent could it be allowed and again it is not different from the laws and statutes in the free world.[11]

The only exception in the Sharia law that is prohibitive pertains to suicide that is haram (forbidden by Islamic law).[12] The main question is if the patient at risk of death or serious harm has the right to care refusal. This is a subtle issue that requires more judicious scrutiny.

The status of the patient rights in the Middle-East and the North Africa

"Patient rights" is attached importance to in almost all the countries in the Middle-East. The law of the patient rights and liabilities has been formulated in Iran aiming to provide appropriate and accessible care.[13] In 2012, the new Iraqi patient rights charter was drafted.[14] What happens in Jordan in regard to patient rights is not dissimilar to the current practice in the western countries.[15] The Lebanon hospitals have also approved of a set of laws and regulations for the protection of patient rights.[16]Egypt, Saudi Arabia, Tunisia, Kuwait, Bahrain, Qatar, UAE, and Oman follow a set of laws for patient rights that is akin to that in the west, though it seems to be a translation of the universal declaration of the patient rights.However, the study conducted in one of the hospitals in Saudi Arabia estimates the patients' awareness about their rights to be very low. Another study in Iran emphasizes on the immediacy of change in the process of obtaining the informed consent prior to surgeries.

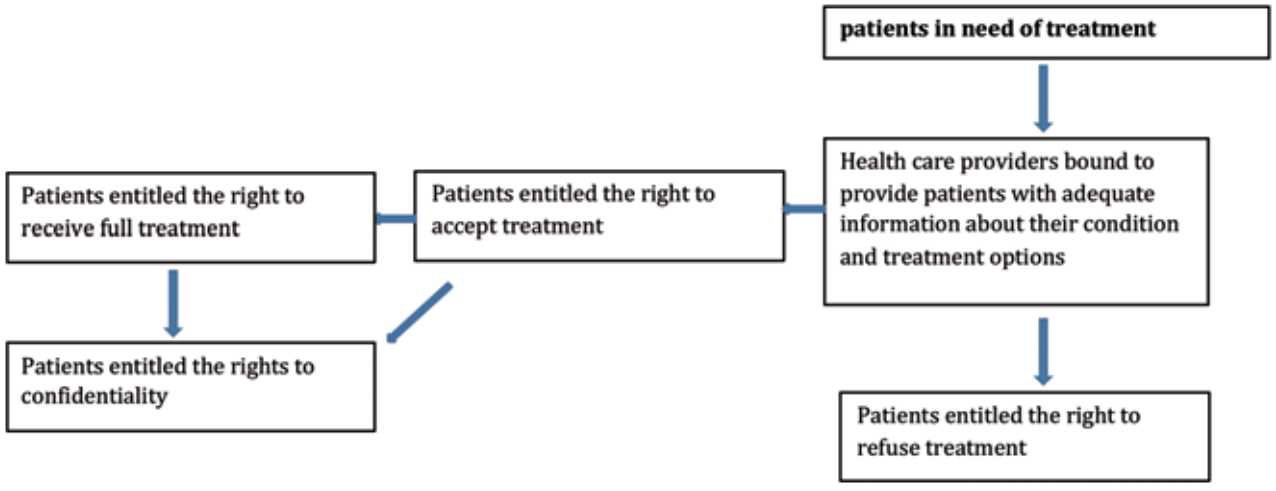

Chart 1. The general sketch of the patients' rights.

Conclusions

The patient rights are of prime importance in then contemporary world. The abidance by the patient rights can ensure access to more qualified care. Since "the patient rights" in different cultures is perceived differently, it should be addressed contingent on the relevant cultural norms and criteria. David Land in the prelude of his book "The Wealth and Poverty of Nations" emphasizes that the Third World countries should seek their way of development in conformity with their own social realities.[17] It is then imperative that Islamic countries by scrutiny in the Sharia laws strive to elicit and formulate their appropriately tailored patient rights. More importantly, it is the role of education in raising the awareness of patients and care providers about the patient rights without which the protocols and laws would be issued but to no avail.

References

1. Universal Declaration of Human Rights, united nations, available from http://www.un.org/en/universal-declaration-human-rights/
2. Patients' rights, World Health Organization, available from http://www.who.int/genomics/public/patientrights/en/
3. Advancing Human Rights in Patient Care: Practitioner Guides, open society publication, available from https://www.opensocietyfoundations.org/publications/advancing-human-rights-patient-care-practitioner-guides
4. Appelbaum PS. Assessment of patient's competence to consent to treatment. New England Journal of Medicine. 2007; 357: 1834-1840
5. Fong Ha. J, Longnecker. N, Doctor-Patient Communication: A Review, Ochsner J. 2010 Spring; 10(1): 38–43
6. CONFIDENTIALITY, PATIENT RIGHTS & RESPONSIBILITIES, the university of Arizona, available from https://www.health.arizona.edu/confidentiality-patient-rights-responsibilities
7. The Right to Treatment, USlegal, available from https://healthcare. uslegal.com/patientrights/the-right-to-treatment/
8. Allama Majlisi. Bihar Al-Anwar (Sea of Lights). 1694-1698. 722/2, Ketabolelm . Vol 33. Hadith (narration) 7: "People dominate over their own lives and properties".
9. Habiba MA (2000) Examining consent within the patient-doctor-relationship. Journal of Medical Ethics 26:183-87
10. Allama Majlisi. Bihar Al-Anwar (Sea of Lights). 1694-1698. Vol 74. Hadith (narration): "Do not divulge the other one's secrets even though they have done so and do not breach their trust even though they have betrayed you".
11. Merideth, P. The Five C's of Confidentiality and How to DEAL with Them, Psychiatry (Edgmont). 2007 Feb; 4(2): 28–29.
12. Maede Surah, Verse 32: " that if anyone killed a person not in retaliation of murder, or (and) to spread mischief in the land - it would be as if he killed all mankind, and if anyone saved a life, it would be as if he saved the life of all mankind".
13. Parsapoor A, Bagheri A, Larijani B. Patient's rights charter in Iran, Acta Med Iran. 2014; 52(1):24-8.
14. New Iraqi Patients' Rights Charter Is Major Step for Human Rights, available from http://www.msiworldwide.com/2012/05/new-iraqi-patients-rights-charter-is-major-step-for-human-rights
15. Patient Rights, The Hashemite Kingdom of Jordan, The Official Site of the Jordanian e-Government, http://www.jordan.gov.jo/wps/portal
16. Your Rights and Responsibilities, http://www.hopital-libanais. com/hospitalizations/your_rights_and_responsibilities
17. Almoajel. A.M, Hospitalized Patients Awareness of Their Rights in Saudi Governmental Hospital, Middle-East Journal of Scientific Research 11 (3): 329-335, 2012
18. Nasiriany, KH , Salemi, S, Salman Yazdy, N, Hosseini, F, HOW NURSES RESPECT PATIENT'S RIGHTS, Iran Journal of Nursing, Volume 15, Number 32 and 33 (2002)
19. Lands. D, The Wealth and Poverty of Nations, (ISBN 0-393-04017-8), published in 1998.

ARTICLE

Challenges of migrations in the Middle East region

Walid Slim,[1] Galev Aleksandar[2]

[1]Migration Health Officer, IOM Iraq; [2]Regional Migration Health Coordinator, IOM Jordan

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Introduction

The International Organization for Migration (IOM) thanks the Thalassemia International Federation (TIF) for the invitation to, and is delighted to contribute to the discussion on health challenges related to the migration and displacement in the region, to the 2nd Middle East (ME), Gulf (G), Maghreb (M) and African (A) Conference on Thalassemia and Other Hemoglobinopathies, organized by the TIF in Amman, Jordan in November 2016.

The TIF raised valid concerns regarding the delivery of, and access to health care services to refugees, migrants, and internally displaced persons affected by the Syrian war.

IOM's Migration Health Division (MHD) advocates for equitable access to health care for all migrants and displaced populations globally; MHD will bring to the audience of the scientific program of the MEGMA Conference the following points:

- Briefing on the ongoing humanitarian crisis in the Middle East,
- The dynamics, trends and challenges of migration taking place in the context of crisis: including irregular migration, human smuggling and trafficking, and regulated migration,
- Introduction of the United Nations' cluster approach for the coordination of the humanitarian crisis response,
- Presentation of IOM's health services provided to the affected population and hosting communities in the Middle East region,
- Presentation of some of the preliminary results of the IOM's work on the Health Profile of the
- Syrian refugees in the Middle East region, examined by the IOM for resettlement/migration.

Context of the crisis in the middle east

The humanitarian situation in the Middle East continues to worsen as the Syrian crisis reaches its 5th year and intensified violence. While the Syrian crisis is considered one the greatest refugee crisis witnessed in generations (1), there are other countries in the region where forced migration, internal displacement and border crossings are taking place, mainly in Yemen and Iraq. Other countries in the region like Jordan, Lebanon and Turkey are affected by the influx of migrants and refugees fleeing the violence. As the conflict intensifies across the region, refugees and internally displaced persons (IDP) are increasingly exposed to risks to their physical safety, mental health and wellbeing due to the lack and inadequate access to essential health and essential social and public services. Furthermore, they are increasingly exposed to greater risk of trafficking and abuse during their hopeless trip to safer places, including countries in the region, as well as the European countries.

The total number of Syrian population in need of humanitarian assistance is estimated to 13.5 million persons, including 6.5 million of internally displaced persons and minimum of 4.7 million of refugees (1).

In Iraq, the humanitarian situation continues to deteriorate as the ongoing military operations are intensifying. The total number of affected population is estimated to 10 million including 3.3 million of internally displaced persons, 250,000 Syrian refugees and millions of affected host communities (2).

One year of conflict in Yemen has resulted an estimated total number of 22.5 million civilians who urgently need some form of humanitarian assistance. This includes 14.4 million people unable to meet their food needs, 19.4 million who lack clean water and sanitation, 14.1 million without adequate healthcare, and at least 2.7 million who have fled their homes within Yemen or sought asylum in the neighboring countries (3).

Health facilities in war-torn countries are becoming a regular and deliberate target, as stated by the international humanitarian groups. In 2015, there were 94 aerial and shelling attacks on 63 Doctors Without Borders (MSF) supported facilities in Syria, resulting in the total destruction of 12 of them; and the death or injury of

Correspondence:
wslim@iom.int

81 MSF supported medical staff. Also, medical facilities in Yemen were target to Airstrikes bombing toward the end of 2015 (7).

Dynamics and challenges of migration in the middle east region

There are multiple patterns of migration taking place in the Middle East region: regular and irregular labor migration; forced migration; and mixed migration flows. Forced migration to, from and through the region is the result of people fleeing conflict. Forced migration constitutes the second predominant migration pattern in the region. Movements are large scale and result in temporary or protracted displacement of migrants outside or within countries of origin. Secondary displacement or onward migration are also common. (4)

1. Mixed migration flows

Mixed migration flows in the Middle East region are motivated by:

- flight from conflict,
- generalized violence,
- persecution,
- breakdowns in public order,
- famine or drought,
- the desire to join family members abroad; and
- escape from economic hardship or the search for a better livelihood and lifestyle opportunities.

Mixed migration is therefore commonly defined as complex population movements including refugees and displaced people, asylum-seekers, migrant workers and other migrants. (4)

Migrants and refugees in mixed migration are using same routes and sharing transport means to their destinations, where human traffickers and migrant smugglers often play an important role. This involves persons without the requisite documentation who cross borders and arrive at their destination in an unauthorized manner. Such a journey renders the migrants and refugees vulnerable under the full power and abuse of smugglers and traffickers. Mixed migration flows, migrants smuggling and human trafficking lead to limited possibilities of the humanitarian community and the governmental authorities of origin, transit and destination countries to manage the flows of migrants and provide adequate protection and health services.

In the context of mixed migration flows in crises, the humanitarian community and local authorities in destination countries work in partnership to achieve equitable access to treatment and preventive care for the refugee, IDPs, and other displaced populations. This is achievable through opening existing health-care services to displaced populations (the case of urban migrants) or by setting up parallel health facilities (the case of camps or transit centers). It is often extremely difficult to impossible to provide such services before and during the movement of migrants undertaking irregular journeys.

2. Regular migration and refugee resettlement programs

The governance of international migration has been defined as all "policies and programmes of individual countries, inter-State discussions and agreements [at bilateral, regional and international level], multilateral forums and consultative processes, the activities of international organizations, as well as relevant laws and norms" that affect migration directly or indirectly (4). Some traditional migration-receiving nations have developed health components within their immigration regulatory processes; while many countries who do not have long histories of immigration often lack immigration health policies and practices.

The modern day immigration health regulations include pre-departure and post-arrival health assessment for transmissible and non-transmissible diseases. These programs provide pre-immigration screening, treatment and immunization services aiming to protect the public health and the migrants by identifying their medical needs.

The United Nation's cluster system for humanitarian response

Following an independent review of the Humanitarian Response System, the United Nations' Inter- Agency Standing Committee (IASC) adopted the Cluster approach in 2005, to address gaps and to increase the effectiveness of humanitarian response through implementation of an improved inter- agency coordination mechanism. The Cluster System ensures that international responses to humanitarian emergencies are predictable and accountable, by defining clear leadership and division of roles and responsibilities of organizations in eleven different areas (5): the 11 Clusters (Figure 1). The 11 Clusters are:

1. Camp management and camp coordination - led by the United Nations High Commissioner for Refugees (UNHCR) and the IOM, depending on the type of crisis.
2. Early recovery - led by the United Nations Development Programme (UNDP).
3. Education - led by the United Nations International Children's Emergency Fund (UNICEF) and the Save the Children organization.
4. Emergency communications - led by the United Nations World Food Programme (WFP).
5 Food security - led by the United Nations World Food Programme (WFP) and the Food and Agriculture Organization of the United Nations (FAO).
6. Health - led by the World Health Organization (WHO).
7. Logistics - led by the United Nations World Food Programme (WFP).
8. Nutrition - led by the United Nations International Children's Emergency Fund (UNICEF).

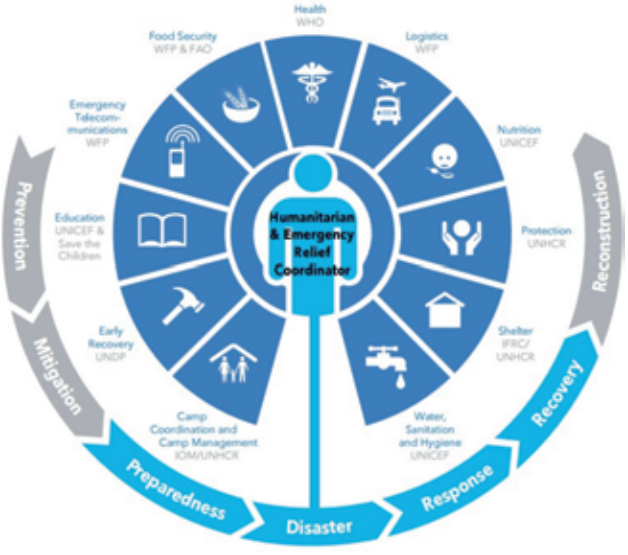

Figure 1.

9. Protection - led by the UNHCR.
10. Shelter led by the UNHCR and the International Federation of Red Cross and Red Crescent Societies (IFRC).
11. Water, sanitation and hygiene - led by the UNICEF.

In response to humanitarian crisis some or all of the above clusters can be activated to coordinate the response of all UN and non-UN agencies in coordination and leadership of the country's government authorities. The cluster system have comprehensive mechanism of activation and deactivation of clusters based on the assessment of the needs.

The Cluster has six core functions at country level (5):

1. To support service delivery: the cluster provides a platform of coordination that ensures service delivery is driven by the Humanitarian Response Plan and strategic priorities, and eliminates duplication of service delivery.
2. To assist in strategic decision making by preparing needs assessments and analysis of gaps (across and within clusters), and setting of priorities.
3. To plan and implement cluster strategies by developing sectoral plans, objectives and indicators of the overall response's strategic objectives; adhering to common standards; and clarifying funding requirements.
4. To monitor and evaluate performance by monitoring and reporting on activities and needs; measuring progress against the cluster strategy and agreed results; and recommending corrective action where necessary.
5. To build national capacity in preparedness and contingency planning.
6. To support advocacy by identifying concerns, and contributing key information and messages; and undertaking advocacy on behalf of the cluster, cluster members, and affected people.

IOM's health services

Since its establishment in 1951, the International Organization for Migration has been committed to the principle that humane and orderly migration benefits migrants and society. As an intergovernmental organization, IOM acts with its partners in the international community to:

- assist in meeting the operational challenges of migration,
- advance understanding of migration issues,
- encourage social and economic development through migration, and
- uphold the human dignity and well-being of migrants.

According to the IOM's vision on Migration Health, migrants and mobile populations benefit from an improved standard of physical, mental and social wellbeing, which enables them to substantially contribute towards the social and economic development of their home communities and host societies.

The Migration Health Division (MHD) of IOM is implementing all IOM's health activities and services. The role of IOM's Migration Health Division in emergency response is driven by the dual objective to:

- advance the public healthcare agenda aimed to support local health systems and
- establish a competent operational approach to provide direct interventions to crisis-affected populations, while identifying gaps related to health and human mobility.

Prioritizing health-care support leads to the reduction of morbidity and mortality among vulnerable crisis-affected migrant populations. (6)

IOM's MHD is implementing activities in three programmatic areas:

- Health assessment and travel health assistance (through the refugee resettlement and regular immigration programs in partnership with member state Governments),
- Health promotion for migrants: activities in this programme area promote the equitable access to quality health services for migrants and mobile populations, including migrants in irregular situations such as trafficked persons and stranded migrants, as well as labor migrants and migrant hosting communities. Advancing the understanding through relevant health indicators monitoring for migrant populations is done through this service area as well,
- Health services for crisis affected populations is a service area which provides direct health care services to migrants, refugees and displaced persons affected with humanitarian crisis and natural deserters, including primary health care services, tuberculosis detection and management, and mental health and psychosocial support programmes, amongst others.

Syrian refugee's health profile study (preview)

IOM conducted a descriptive study on the health profile of the Syrian refugees in MENA region. It tries to describe the health profile of Syrian refugees based on the analysis of secondary data of the refugee health assessment programs conducted in Jordan, Lebanon, Turkey, Iraq, Egypt, and few other countries from 1st of January 2015 to 30th of June 2016. Disease groups and specific conditions were encoded according to the International Classification of Diseases and Related Health Problems, 10th Revision (ICD-10). Data was entered into IOM's Migration Management Operational Systems Application (MiMOSA) database. Medical conditions reported by refugees to IOM Physicians during clinical examination were recorded, and relevant medical records examined. The study population consisted of 67,026 Syrian refugees located in Jordan (42.3%), Lebanon (49.2%), Iraq (1.4%), Turkey (3.6%), Egypt (3.1%) and other countries (0.5%). The mean age was 37.4 years; 48.4% were female; 16% examined were children below 5 years, and 45% were under 15 years or age.

The overall prevalence rate of morbidity among the study population was 321/1000, and about 21% of the assessed refugees have one morbid condition, or more. In comparison with other refugee groups assessed by IOM, about 29% of Iraqi refugees, 27.4% of Myanmar refugees, about 31% of Somali refugees, and 22.6% of Eritrean refugees were diagnosed with one or more morbid conditions.

The top eight groups of diseases that have the highest prevalence were:

1. diseases of the circulatory system,
2. endocrine, nutritional and metabolic diseases,
3. diseases of the musculoskeletal system and connective tissue,
4. mental and behavioral disorders,
5. diseases of the eye and adnexa,
6. diseases of the respiratory system,
7. diseases of the digestive system, and
8. diseases of the nervous system.

Diseases of the blood and blood-forming organs and certain disorders involving the immune mechanism according to ICD-10 coding blocks rank 14th in prevalence among the morbidity groups of diseases with a prevalence rate of 7.5 per 1000 of the study population. Hemolytic and nutritional anemias formed more than

three quarters of the reported cases of blood diseases with a prevalence rates of 2.9/1000 and 2.7/1000 respectively. The prevalence of blood diseases among females was higher than among males (8.7/1000 compared to 6.3/1000). The two age groups mostly affected were; 25-34 and 15-24 (prevalence rates of 9.1and 8.2/1000 respectively). By combining the age groups, the prevalence was highest in the age 15-44 with a rate of 8.5/1000 and becomes lowest when the age grow longer, the rate becomes 4.1/1000 for the age group 45 and more, and it is in between for under 15 years old children(rate of 7.4/1000). This probably can be interpreted on the basis that most of these diseases are anemias and since women in the child bearing age lie in this age group (15-44) and knowing that anemias specially the nutritional one are so common among women in the fertile age in this region. The prevalence of all types of thalassemia combined is 1.5 per 1000 (Table 1).

Table 1. Prevalence rate/1000 of thalassemia.

Variables	Frequency	Percent
Gender		
Males	53	43.8
Females	50	56.2
Age		
0 - 4	20	13.2
5 - 14	46	32
15 - 24	14	14.2
25 - 34	14	19.4
35 - 44	9	14.6
45 - 54	1	4.8
Location country		
Lebanon	24	23%
Jordan	58	56%
Turkey	8	8%
Egypt	4	4%
Iraq	9	
ICD-10 classification		
Thalassaemia D56	39	38%
Beta thalassaemia D56.1	22	21%
Thalassaemia trait D56.3	15	15%
Other thalassaemias D56.8	3	3%
Thalassaemia, unspecified D56.9	24	23%
Total	103	100%

References

1. Overview: 2016 syria humanitarian response plan and 2016-2017 regional refugee and resilience plan; London 4th February 2016. https://www.humanitarianresponse.info/en/operations/whole-of-syria/document/overview-2016-syria-humanitarian-response-plan-2016-2017-regional
2. IRAQ: Humanitarian Dashboard, May to August 2016. United Nations Office for the Coordination of Humanitarian Affairs; 12th October 2016. https://www.humanitarianresponse.info/en/operations/iraq/infographic/iraq-humanitarian-dashboard-may-august-2016.
3. 2016 Humanitarian Response plan, January – December 2016; Yemen. https://www.humanitarianresponse.info/en/programme-cycle/space/document/2016-yemen- humanitarian-response-plan
4. 2015 Situation Report on International Migration: Migration, Displacement and Development in a Changing Arab Region; International Organization for Migration and United Nations Economic and Social Commission for Western Asia. http://publications.iom.int/system/files/pdf/sit_rep_en.pdf
5. Reference module for cluster coordination at country level; revision July 2015; IASC Working Group.
6. Migration health annual review 2014; International Organization for Migration. 7) International activity report, second edition 2015. Doctors Without Borders (MSF International).

ARTICLE

Clinical outcomes of gene therapy with BB305 lentiviral vector for sickle cell disease and β-thalassaemia

Jean-Antoine Ribeil,[1] Marina Cavazzana,[1] Fabien Touzot,[1] Emmanuel Payen,[2] Bénédicte Neven,[1] Francois Lefrere,[1] Felipe Suarez,[3] Elisa Magrin,[1] Yves Beuzard,[2] Resy Cavallesco,[4] Stany Chretien,[3] Philippe Bourget,[1] Fabrice Monpoux,[5] Corinne Pondarré,[6] Pablo Bartolucci,[6] Manfred Schmidt,[7] Christof von Kalle,[7] Frans A. Kuypers,[8] Laura Sandler,[9] Sandeep Soni,[9] Michaela Semeraro,[1] Wassim El Nemer,[10] Olivier Hermine,[1] Mariane De Montalembert,[1] Stéphane Blanche,[1] Salima Hacein-Bey-Abina,[1] Philippe Leboulch[2,4,11]

[1]Hôpital Universitaire Necker-Enfants Malades, Paris, France; [2]CEA (iMETI) and University of Paris-Sud, Fontenay-aux-Roses, France; [3]Paris Descartes Sorbonne Paris Cite University, Paris, France; [4]Brigham & Women's Hospital, Boston, MA; [5]Centre Hospitalier de Nice Sophia-Antipolis, Nice, France; [6]Université Paris Est Créteil, Créteil, France; [7]National Center for Tumor Diseases, Heidelberg, Germany; [8]Children's Hospital Oakland Research Institute, Oakland, CA; [9]Bluebird bio, Cambridge, MA; [10]INSERM, Paris, France; [11]Universitaire Paris-Sud, Paris, France

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Purpose of the study

In patients with hemoglobinopathies, transduction of hematopoietic stem cell (HSC) with β-globin gene may induce production of functional β-globin, potentially reducing or eliminating the symptoms of disease. LentiGlobin BB305 Drug Product (DP), an investigational gene therapy product for the treatment of sickle cell disease (SCD) and β-thalassemia (βT), consists of autologous CD34+ cells transduced with the BB305 lentiviral vector. BB305 is a replication defective, self-inactivating vector using erythroid-specific globin gene regulatory elements. The vector carries an HBB gene with an anti-sickling amino acid substitution found in γ globin (T87Q) that also allows for HPLC monitoring of transgene globin levels in the subjects' blood.

Materials and methods

HGB-205 is a phase 1/2 clinical study of safety and efficacy of LentiGlobin BB305 DP in severe SCD and transfusion-dependent βT. Subjects undergo HSC collection via bone marrow harvest (SCD) or mobilization and apheresis (βT). CD34+ cells are selected and transduced with BB305 vector to produce the DP. Subjects undergo myeloablation with IV busulfan followed by DP infusion. As of 10 November 2015, 5 subjects had been treated: 1 with SCD and 4 with βT. No replication competent lentivirus has been detected and integration site analysis shows polyclonal reconstitution without clonal dominance at all time points to date (follow-up between 2 and 23 months). No subjects have experienced drug product-related adverse events; safety observations to date are consistent with myeloablative conditioning.

Results

The subject with SCD received 5.6 x 10e6 CD34+ cells/per kg from two DP lots with VCN of 1.0 and 1.2. He achieved neutrophil engraftment on Day +37 and platelet engraftment on Day +91. Prior to study treatment, he had been on prophylactic transfusions to manage severe SCD symptoms including multiple vaso-occlusive crises. He stopped transfusions at Day +88 and at 12 months post DP infusion had a total Hb of 11.7 g/dL, of which approximately 49% is anti-sickling hemoglobin (47% HbA^{T87Q}, 2% HbF). This subject has had no post-infusion pain crises or SCD-related hospitalizations. For the βT subjects the cell dose infused was 8.8 to 13.6 x 10e6 CD34+ cells/per kg, with VCN between 0.8 and 2.1, with neutrophil engraftment at Day +13 to +28 and platelet engraftment at Day +17 to +24. The first two βT subjects treated had genotype β^0/β^E. They have been transfusion independent since ~2 weeks after DP infusion, with consistent levels of total Hb (>10g/dL) and HBAT87Q (7-8 g/dL and 9-10 g/dL). The third βT subject is homozygous for the IVS1 nt 110 G>A mutation. This subject at 4.5 months of follow-up had total Hb 8.0 g/dL and had gone ~2 months without transfusion. The fourth βT subject (β^0/β^E) has less than 2 months follow-up.

Conclusions

These interim data suggest that gene therapy with LentiGlobin DP is a promising potential treatment for severe SCD and transfusion-dependent βT.

Correspondence:
jaribeil@gmail.com

Blood and blood products: adequacy and safety

Hassan Abolghasemi,[1] Vahid Abolghasemi[2]

[1]Pediatric Congenital Hematologic Disordes, Research Center, Mofid children hospital, Shahid Beheshti University of Medical Sciences, [2]Baqiatallah University of Medical Science, Teheran, Iran

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ABSTRACT

Regular blood transfusions have been a crucial aspect of the treatment of thalassemia since the 1960s.thalassemic patients are the largest community of lifelong users of donated bloods and between 6000,000-12000,000 transfusions are needed each year for around 500,000 patients with beta thalassemia[1]. Ensuring the safety and availability of blood and blood components is an essential public health responsibility. Measures to ensure blood safety play a major role in preventing the transmission of HIV, hepatitis B and C viruses and other emerging blood borne pathogens in health care system. The World Health Organization (WHO) supports that providing safe and adequate blood should be an integral part of every country's national health care policy and infrastructure. According to WHO report specific legislation covering the safety and quality of blood transfusion are only implemented in 62% of countries worldwide (of which 81% are high-income countries, 60% are middle-income countries and 44% are low-income countries). Of the 112.5 million blood donations collected globally, approximately half of these are collected in the high-income countries, with 19% of the world's population. There is a marked difference in the level of access to blood between low- and high-income countries. The whole blood donation rate is an indicator for the general availability of blood in a country. The median blood donation rate in high-income countries is 33.1 donations per 1000 people. This compares with 11.7 donations per 1000 people in middle-income countries, and 4.6 donations per 1000 people in low-income countries.70 countries report collecting fewer than 10 donations per 1000 people. Of these, 38 countries are in WHO's African Region, 6 in the Americas, 6 in the Eastern Mediterranean, 5 in Europe, 6 in South-Eastern Asia and 9 in the Western Pacific. All are low- or middle-income countries. Thus there is a significant difference in the level of access to safe blood between low- and high-income countries, especially considering that up to 65% of blood transfusions are given to children under five years of age in low-income countries. To safeguard the health of the transfusion recipient, including patients with thalassaemia, blood should be obtain from carefully selected regular voluntary, non-remunerated donors and should be collected, tested processed, stored and distributed, in the context of dedicated, quality assured national blood transfusion centers. Blood screening should be performed according to the quality system requirements. According to WHO report16 countries are not able to screen all donated blood for 1 or more of the above infections [2]. WHO recommends that all blood donations should be screened for transfusion transmitted infections prior to use. Irregular supply of test kits is one of the most commonly reported barriers to screening. Haemovigilance is required to identify and prevent occurrence or recurrence of transfusion related adverse effect, to increase the safety, efficacy and efficiency of blood transfusion. in this review we discuss some aspects of blood safety from vein to vein with focus on information provided from countries in the region.

Correspondence:

h.abolghasemi@ibto.ir

Introduction

Blood transfusion saves lives and improves health in significant numbers of people specially patients with congenital blood disorders, but many patients requiring transfusion do not have timely access to safe and adequate blood. Providing safe and adequate blood and blood products should be an integral part of every country's national health care policy and infrastructure. Regular blood transfusions have been a crucial aspect of the treatment of thalassemia since the 1960s.thalassemic patients are the largest community of lifelong users of donated bloods and between 6000,000 – 12000,000 transfusions are needed each year for around 500,000 patients with beta thalassemia[1].

WHO recommends that all activities related to blood collection, testing, processing, storage and distribution be coordinated at the national level through effective organization and integrated blood supply networks. The national blood system should be governed by national blood policy and legislative framework to promote uniform implementation of standards and consistency in the quality and safety of blood and blood products. The capacity to provide patients with the different blood components they require is still limited in low-income countries: 43% of the blood collected in low-income countries is separated into components, 78% in middle-income countries and 96% in high-income countries (Figure 1).[2]

In 2013, 73%, or 122 out of 167 countries, had a national blood policy. Overall, 65%, or 108 out of 167 countries, have specific legislation covering the safety and quality of blood transfusion, including:79% of high-income countries; 64% of middle-income countries; and 41% of low-income countries. About 112.5 million blood donations are collected worldwide. More than half of these are collected in high-income countries, with 19% of the world's population. About 13 000 blood centres in 176 countries report collecting a total of 110 million donations. Collections at blood centres vary according to income group. The median annual donations per blood centre is 5400 in the low- and middle-income countries, as compared to 16 000 in the high-income countries.

Based on these data from WHO blood centers are more efficient in developed country compared with developing countries. so there is a marked difference in the level of access to blood between low- and high-income countries. The whole blood donation rate is an indicator for the general availability of blood in a country. The median blood donation rate in high-income countries is 33.1 donations per 1000 people. This compares with 11.7 donations per 1000 people in middle-income countries, and 4.6 donations per 1000 people in low-income countries. 70 countries report collecting fewer than 10 donations per 1000 people. Of these, 38 countries are in WHO's African Region, 6 in the Americas, 6 in the Eastern Mediterranean, 5 in Europe, 6 in South-Eastern Asia and 9 in the Western Pacific. All are low- or middle-income countries.[2]

Data about the gender profile of blood donors show that globally 28% of blood donations are given by women, although this ranges widely. In 16 of the 119 reporting countries, less than 10% of donations are given by female donors. The study by Khadir and coworkers[7] in 2001 on 12,121 females in eight provinces of iran showed that the more common reasons for not donating blood are concern for contacting infectious diseases or becoming anemic (40.4%). A lack of information about the importance of donated blood (37.7%), misconceptions about the consequences of blood donation (31.3%),

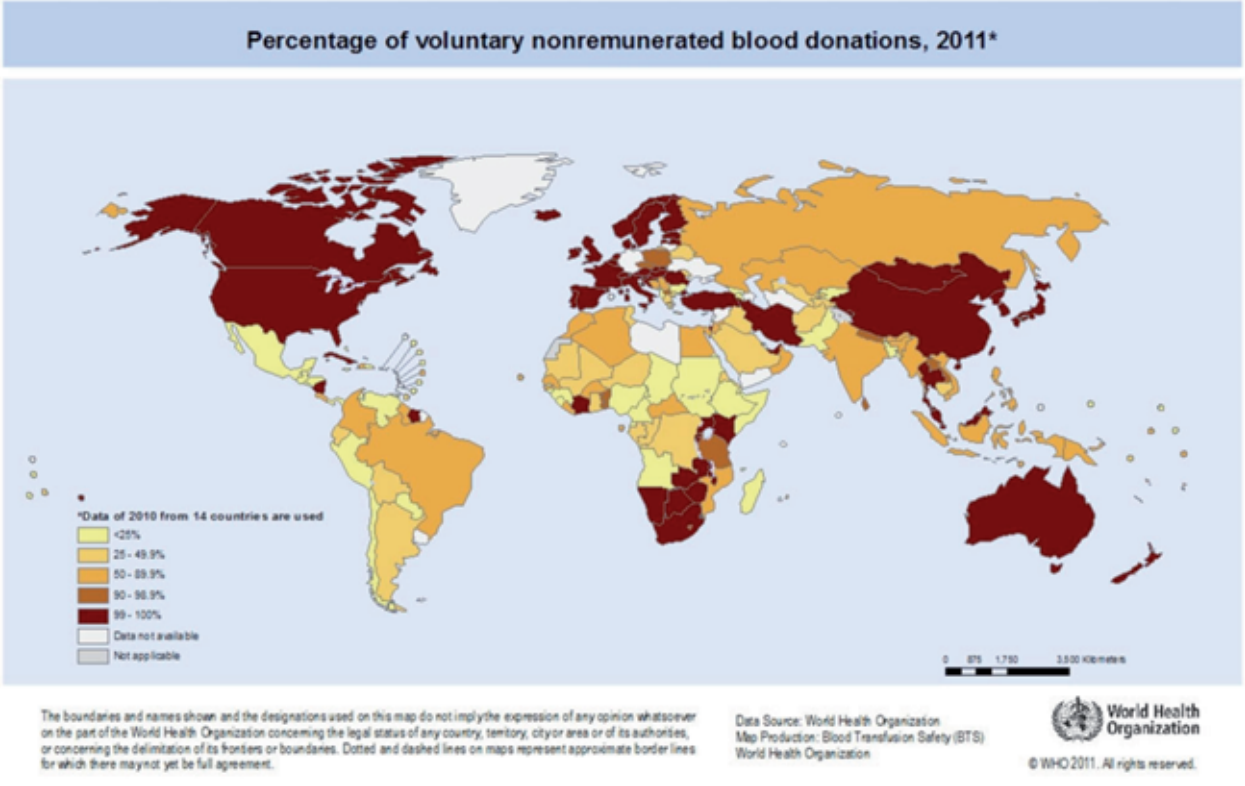

Figure 1. Percentage of voluntary nonremunerated blood donations, 2011.

and finally a fear of the needle were other reasons mentioned[10].

The age profile of blood donors shows that, proportionally, more young people donate blood in low- and middle-income countries than in high-income countries. Demographic information of blood donors is important for formulating and monitoring recruitment strategies. all blood center should provide a complete demographic feature of blood donors in order to find the best altruism approach for recruiting and retention more blood donor

Types of blood donors

voluntary unpaid,family/replacement and paid are 3 types of blood donors.Two studies in the late 1959 showed a higher incidence of post-transfusion hepatitis in paid and professional donors. In the early 1970s, all organizations participating in blood transfusion procedures in United States issued position statements supporting the concept of voluntary donation. In 1978, the FDA requested that all blood and blood products to be labeled as "paid" or "volunteer." In accordance with this trend, voluntary blood donation was promoted in European countries too. This kind of donation has been accepted for over 50 years in France, Luxembourg, Northern Ireland, and also it was introduced in Spain in the 1980s, Italy, Portugal, Greece, and Estonia in the 1990s.

An adequate and reliable supply of safe blood can be assured by a stable base of regular, voluntary, unpaid blood donors. These donors are also the safest group of donors as the prevalence of blood borne infections is lowest among this group. World Health Assembly resolution WHA63.12 urges all Member States to develop national blood systems based on voluntary unpaid donations and to work towards the goal of self-sufficiency. Donor selection is a critical process to identify high risk volunteers and defer them from donating blood. Despite viral screening test on all donated blood, one cannot rely on screening tests alone to ensure a safe blood supply. Monitoring and assessment of the deferral procedure is of utmost importance to balance blood availability and safety. A study by razjou and etal compared the prevalence of HIV, HCV, and HBV markers between deferred donors and accepted blood donors in order to evaluate the effectiveness of the current donor selection process in Iran.The prevalence of HIV, Hepatitis B, and Hepatitis C was 120 (CI 95%; 90-150), 1280 (CI 95%; 1170-1390), and 580 (CI 95%; 510-650) in 100,000 deferred donors respectively. A significant increase exists in the prevalence of HBV (1.7 times), HIV (24 times) and HCV (15 times) in deferred donors as compared to accepted blood donors[3]. Another study was done by amini ethal showed the frequency of HBV infection entering the blood supply has decreased as a result of improvement in donor recruitment and selection, usage of software in transfusion services and possibly decreasing HBV infection prevalence in general population[5].

In a retrospectively designed study, yenicisu ethal evaluated the influence of the permanence and qualifications of health-care professionals on blood disposal rates due to hepatitis seropositivity. They observed a decrease of 44.2% in the number of blood units being rejected due to the donor's hepatitis B seropositivity in the second study period in which self-exclusion forms and where blood donation candidates were evaluated by a family physician[7]. in another study by cokac N ethal in turkey the decreasing trends observed in TTI from the 17-year period studied indicated the value of safety measures taken, in particular the implementation of donor screening procedures in 1997[8].

Data reported to WHO shows significant increases of voluntary unpaid blood donations in low- and middle-income countries:

- An increase of 10.7 million blood donations from voluntary unpaid donors from 2008 to 2013 has been reported by 159 countries. The highest increase of voluntary unpaid blood donations is in the African (85%) and South-East Asian (74%) Regions. The maximum increase in absolute numbers was reported the South-East Asia region (5.3 million donations), followed by the Western Pacific Region (2.8 million donations).
- 74 countries collect more than 90% of their blood supply from voluntary unpaid blood donations (39 high-income countries, 26 middle-income countries and 9 low-income countries). This includes 62 countries with 100% (or more than 99%) of their blood supply from voluntary unpaid blood donors.
- In 72 countries, more than 50% of the blood supply is still dependent on family/replacement and paid blood donors (11 high-income countries, 45 middle-income countries and 16 low-income countries).
- 24 countries still report collecting paid donations in 2013, around 1 650 000 donations in total (Figure 2). [2]

Quality systems for blood safety

Blood transfusion is a multi-step chain with risk of error

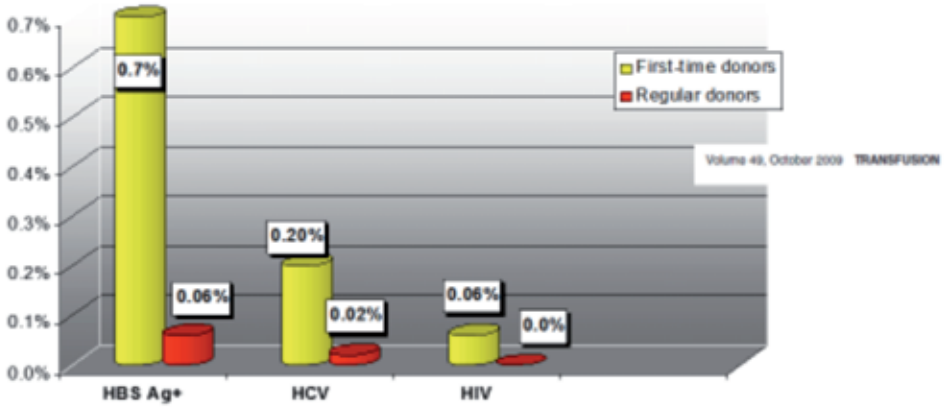

Figure 2. Prevalence of positive HBS Ag, HCV and in blood donors in IRAN, 2007.

in each process from donor selection, blood collection and processing, testing of donor and patient samples, issue of compatible blood, to transfusing the patient. An effective quality system provides a framework within which activities are established, performed in a quality-focused way and continuously monitored to improve outcomes. The risk associated with blood transfusion risk can be dramatically reduced by the introduction of quality systems, external quality control and education and training of staff.

A quality system should cover all aspects of its activities from vein to vein and ensure traceability, from the recruitment and selection of blood donors to the transfusion of blood and blood products to patients. It should also reflect the structure, needs and capabilities of the blood transfusion service, as well as the needs of the hospitals and patients that it serves. Organizational management; Standards; Documentation; Training and Assessment are key elements of a quality system. Management commitment and support are essential for the development, implementation and monitoring of a national quality system in order to ensure continuous quality improvement. All staff should understand the importance of quality and the consequences of failure in the quality system[2].

Blood screening

WHO recommends that all blood donations should be screened for transfusion transmited infections prior to use. Screening of donated blood should be mandatory for HIV, hepatitis B, hepatitis C and syphilis. other tests should be added based on epidemiologic data which is available in health care system for instance malaria or west Nile virus. Blood screening should be performed according to the quality system requirements. based on WHO report16 countries are not able to screen all donated blood for 1 or more of the above infections. Irregular supply of test kits is one of the most commonly reported barriers to screening. 81% blood screening laboratories in high-income countries are monitored through external quality assessment schemes, as compared to 55% in middle-income countries and 34% in low-income countries. The prevalence of transfusion-transmissible infections (TTI) in blood donations in high-income countries is considerably lower than in low- and middle-income countries[2] (Table 1).

These differences reflects the variation in prevalence of TTI among general population and population who are eligible to donate blood, the type of donors (such as voluntary unpaid blood donors from lower risk populations) and the effectiveness of the system of educating and selecting donors. There are a lot of subject need to be discussed in this regards, including the sensitivity and specificity of the tests, using rapid test, nucleic acid amplification test (NAT) and automation system and all of these elements have impact on affordability of screening test and consequently the level of blood safety in different countries.

Supply of plasma-derived medicinal products

Plasma contains several therapeutically important proteins, the most important of them are factor 8 and 9, gamma globulin and albumin. Currently more than 25 of them are commercially available to treat life-threatening diseases such as hemophilia, immune deficiency disorders and sever kidney and liver diseases. Some of these medicines already included in the WHO Model List of Essential Medicines indicating their importance from a global perspective. However, unfortunately due to very high cost of these medicines, they are not available for a majority of the patients living in low income countries. The capacity to provide patients with the different blood components they require is still limited in low-income countries: 43% of the blood collected in low-income countries is separated into components, 78% in middle-income countries and 96% in high-income countries[2]

There are some options available for securing accessibility to these medicines. These include local production, importation and contract fractionation of locally produced plasma. Although local production of plasma-derived medicines and/or importation of these medicines might be a practical approach to respond to the needs for these medicines, in recent years several countries in the world and some countries in the region have used contract fractionation of locally produced plasma as a very effective approach for improving availability and affordability of plasma-derived medicines in their national market [11].

World Health Assembly resolution WHA63.12 urges Member States to establish, implement and support nationally-coordinated, efficiently-managed and sustainable blood and plasma programs according to the availability of resources, with the aim of achieving self-sufficiency. It is the responsibility of all governments to ensure sufficient supply of plasma-derived medicinal products namely immunoglobulin and coagulation factors, which are needed to prevent and treat a variety of serious diseases. 43 countries (26 high-income, 16 middle-income, 1 low-income) of the 175 reporting countries reported producing all or part of the PDMP through the fractionation (for example, domestic or/and contract fractionation) of plasma collected in the country.106 countries report that all PDMP are imported: 18 countries report that no PDMP were used during the reporting period; 8 countries report that plasma collected in the country was sold to the manufacturers of plasma-derived medicinal products and products purchased from PMDP suppliers in the market. Around 14.3 million liters of plasma from 43 reporting countries (22 high-income countries, 12 middle-income countries and 1 low-income countries, covering a population of 2.76 billion) was fractionated for the production of

Table 1. Prevalence of TTIs in blood donations (Median, Interquartile range (IQR)), by income groups.

	HIV	HBV	HCV
High-income countries	0.003% (0.001%-0.040%)	0.030% (0.008%-0.180%)	0.020% (0.003%-0.160%)
Middle-income countries	0.120% (0.020%-0.340%)	0.910% (0.280%-2.460%)	0.320% (0.090%-0.690%)
Low-income countries	1.080% (0.560%-2.690%)	3.700% (3.340%-8.470%)	1.030% (0.670%-1.800%)

plasma derived medicines during the year. This includes around 50% plasma recovered from the whole blood donations [11,6].

Haemovigilance

Haemovigilance is required to identify and prevent occurrence or recurrence of transfusion related unwanted events, to increase the safety, efficacy and efficiency of blood transfusion. haemovigilance covering all activities of the transfusion chain from vein to vein. The system should include monitoring, identification, reporting, investigation and analysis of adverse events near-misses and reactions related to transfusion and manufacturing. The pioneering work on hemovigilance started in France in 1994 with the setup of monitoring systems by Blood Transfusion Committees and establishing a national hemovigilance system. Later, in 1995 with an objective to improve public confidence in safe blood supply, the European Council published a resolution.

The data provided from the investigations and analyses facilitate corrective and preventive actions to be taken to minimize the potential risks associated with safety and quality in blood processing and transfusion for donors, recipient and medical personel. Such information is also crusial to introduce required changes in the applicable policies, improve standards, systems and processes, assist in the formulation of guidelines, and increase the safety and quality of the entire process from donation to transfusion.

The haemovigilance system should involve all relevant stakeholders, and should be coordinated between the blood transfusion service, hospital clinical staff and transfusion laboratories, hospital transfusion committees, regulatory agency and national health authorities. Extension of the haemovigilance system to regional and global sharing of information will further enhance the process of learning for improvement.

The WHO draft guidelines on adverse event reporting and learning systems: from information to action emphasize the fundamental role of reporting systems in enhancing patient safety by learning from failures of health care systems. Effectiveness of such systems should be measured not only by data reporting and analysis but by the use of such systems to improve patient safety. Among countries around Persian Gulf a few countries such as Saudi Arabia and iran reported their activity around haemovigilance. In one center in Saudi Arabia with population of 23,132 donors, One hundred and forty-eight donor reactions were reported, resulting in a rate of 0.6%. Eighty-four transfusion reactions were reported and most were allergic reactions (79.7%). Errors or incidents were reported with approximately 0.3% of the total number of submitted samples/request forms

Status of blood safety in the region

The Eastern Mediterranean region consists of a heterogeneous group of countries with various levels of development. In the 1990s, blood transfusions was reported to be one of the main causes of HIV contamination in the Middle East. A cumulative total of 3,745 AIDS cases were reported from the countries of the region through the end of 1995. Information about the mode of transmission was available in 3,461 cases (92.4%). Of that percentage, 368 (10.6%) cases were due to the receipt of blood or blood products, the largest numbers of which were reported in Iran, Morocco, Saudi Arabia, Egypt, and Iraq. However, it was later proven that the majority of the HIV cases in Iranian patients who received plasma products were attributed to imported concentrated coagulation factors from the France[4]. Nonetheless, owing to improvements in transfusion safety in the region, blood transfusion is no longer the main route of HIV transmission and even hepatitis .

Although there is confirmed data that the prevalence of transfusion transmited infections are lowest among unpaid blood donors who give blood voluntarily, several countries in the region still rely heavily on paid donors and/or replacement donors. Despite the presence of a centralized blood transfusion service in some countries of the region, such as Iran, several countries have fragmented blood transfusion services.[9]

A limited number published data is available that describes blood safety and availability statuses in the eastern Mediterranean region. This type of data is even scarcer with regard to Arab countries of the Persian Gulf coasts. According to a published survey from 17 Arab countries, 14 had thus far formulated a national policy but only nine had national regulation. In 2004, a total of 2,400,000 units of blood were collected in these 17 countries. The proportion of blood collected on the basis of voluntary blood donation in these countries varies drastically and is reported to be between 3% and 100%. Donation rates vary from 0.2% (Syria) to 2.7% (Kuwait). In Saudi blood banks, 47% of donated blood is from the relatives, friends, or colleagues of patients, and voluntary donation constitutes 40% [12] of donated blood. However, all this donated blood is tested against hepatitis B and C and HIV in these countries. Multi-transfused patients, including thalassemia patients, are among the most vulnerable to TTIs. Therefore, the prevalence of infectious agents in this group of patients could be considered an indirect index of blood safety. The prevalence of HCV infection in thalassemia patients in the Eastern Mediterranean region has been recently reviewed.

Although up to 80% of adult thalassemia patients are infected with HCV in the world, there is substantial variation among the countries in the region. For example, in Iran, HCV infection rates range from 2% to 32% and in Saudi Arabia, from 33% to 93%. In Kuwait and Jordan, all the HCV-infected thalassemia cases were transfused before the onset of blood donor screening. However, in this regard, there is no data from most of the countries in the region. In Egypt, sexual contact is the main cause (71%) of HIV transmission. However, cumulatively and until 2009, blood and blood products were identified as the cause of infection in 5% of the cases. To prevent HIV transmission through blood and blood products, a blood safety policy applies to all blood banks in Egypt, which includes the screening of donated blood for HBV, HCV, HIV, and syphilis. From 2008 and 2009, 52 HIV-positive blood bags were detected and discarded. In 2009, a total number of 1,280,000 blood units were screened and 44 HIV-positive blood bags were detected and discarded. An analysis of data from 211,772 blood samples collected from 2000 to 2008 in Egypt has shown that the overall HBsAg and anti-HCV prevalence were 1.65% and 9.02%, respectively. Anti-HCV and HBsAg prevalence has dropped from 11.06% (in 2000) to 6.3% (in 2008) and from 1.24% (in 2000) to 1.17% (in 2008), respectively. The prevalence of anti-HCV and HBsAg was significantly higher among males donors (10% and 1.74%, respectively) compared with females (3% and 1.07%, respectively). It was also reported that there was a significantly higher occurrence of both anti-HCV and HBsAg prevalence among donors from rural area (11.3% and 2.27%, respectively) compared with urban donors. Kuwait's central

blood bank is the only public supplier of blood and blood components in Kuwait that performs blood and blood component collection, processing, testing, distribution, and transfusion services for all governmental and private hospitals in the country. As a reflection of the country's multinational residences, over the past decades only 40-43% of blood donors in Kuwait were Kuwaiti nationals. On the basis of a study conducted in 2002 among 26,874 blood donors to Kuwait's central blood bank, 51.2% were Kuwaiti nationals and 48.8% were non-Kuwaiti Arabs. However, only 15.3% of donations were volunteers, while 84.7% were replacement donors.

The prevalence of anti-HCV among Kuwaiti national and non-Kuwaiti Arab first-time donors was 0.8 and 5.4%, respectively, whereas the prevalence of HBsAg was 1.1 and 3.5%, respectively. Among the first-time donors, 13.7 percent were positive for the presence of anti-HBs. These figures show that the heterogeneity of the population living in Kuwait, and the primary reliance on replacement blood donors, might have a significant impact on the prevalence of hepatitis infection among blood donors.

The prevalence of HBsAg has been evaluated in blood donations in Iran over the period from 1998-2007. A total of 14,599,783 donations were collected during these 10 years. The overall HBsAg prevalence rates declined from 1.79% in 1998 to 0.41% in 2007 as a result of improvements in donor recruitment and selection, implementation of automation in transfusion services, and a possibly decreasing HBV infection prevalence in the general population. A separate report has also examined the results from the viral screening of 6,499,851 donations from 2004 through 2007 for HBV, HCV, HIV, and syphilis. The overall prevalence of these viral markers was 0.56% for HBV, 0.004% for HIV, and 0.13% for HCV. There was a significant decrease in HBsAg prevalence from 0.73% in 2004 to 0.41% in 2007. Despite an increasing HIV prevalence in the general population of Iran, the prevalence of HIV decreased from 0.005% in 2004 to 0.004% in 2007. HCV prevalence showed a slight decline in blood donations from 0.14% in 2005 to 0.12% in 2007.

The situation in Pakistan with regard to blood transfusion has remained far from satisfactory over the years. There is extreme fragmentation and rampant commercialism with questionable transfusion practice quality in the majority of blood establishments throughout the country. The blood transfusion services in Pakistan are mostly hospital-based, and majority of blood donors are usually first-time replacement donors or paid and directed donors. About 75% of blood donations in Pakistan come from replacement donors, and around 15% of the blood is still donated by professional donors. Only 10% of blood donors in Pakistan make voluntary unpaid donations. It seems the cultural and socioeconomic factors are associated with a reluctance to donate blood at all, especially without reward. The annual estimated requirement of blood is approximately 1.5 million units, with 40% of the demand being met by the public sector. Although there is about a 40% shortage of blood and blood components in this country, about 85% of blood used as whole blood products.

In Pakistan, crude low-sensitivity kits, available at very low prices, are being used for screening. Commercial blood donors with infections like HIV, HCV, and HBV are not stringently screened before a blood donation. Published reports indicate discouraging trends in appropriate screenings of potential donors for viral infections, unsafe sexual behavior, and drug abuse. Data on the serologic testing of blood donors for HBV, HCV, HIV, and syphilis in a 10-year period (1996-2005) in Lahore has been reviewed. The frequency of serologic markers ranged from 1.46-2.99% for HBV, with a downward trend over time: 3.01-4.99% for HCV, 0-0.06% for HIV, and 0.19-0.57% for syphilis. However, volunteer donors (6.98% of all the donors) had the lowest seroprevalence for the diseases.

The statuses of blood transfusion services in Afghanistan have also been reviewed recently. Owing to the lack of appropriate infrastructure and resources, the national blood supply system is damaged and heavily disrupted. Afghanistan, with an estimated population of 30 million, needs at least 300,000 units of blood. Presently, more than 95% of blood donation in Afghanistan comes from family replacements, which are often paid by the patient's family, and less than 5%are from voluntary donation [11].

IRAN experience

Iranian Blood Transfusion Organization (IBTO) is the only nationally accredited organization in Iran that performs blood transfusion procedures ranging from blood donor recruitment as well as blood distribution. IBTO was established in May 1974. This government-based organization provides its services free of charge. Before its establishment, blood services were provided through hospital-based systems. IBTO is managed by the Supreme Council, which consists of five experts in hematology and related fields appointed by the Minister of Health. The Managing Director of IBTO, elected by the Supreme Council, ensures proper implementation of the decisions adopted. The financial resources of IBTO are covered by a government-approved budget.[9]

The mission of IBTO is to provide and ensure a safe and adequate blood supply in Iran. IBTO fulfils its goals through 30 regional blood centers, which are located in 30 different provinces with more than 200 blood donation sites throughout the country to meet the demands of the Iranian community for blood. It also aims at promoting transfusion medicine in Iran. The trend in yearly blood donations has significantly increased from 1,183,630 blood units in 1998 to 1,735,008 by the end of 2007. During this period the overall growth rate was 59.8% ($p < 0.005$). Yearly blood donation in the northwest region of the country was 13 per 1000 in contrast to 39 per 1000 in the central region. There was a significant decrease in the number of donations during the months of April, September, and January whereas in May, August, and the religious month of Muharram a significant increase was noted. Voluntary donations increased from 77% in 1998 to 100% by the end of 2007.

Continuous donor recruitment efforts in Iran have resulted in a significant increase in blood donation rates during the past decade as well as achieving 100% voluntary, nonremunerated blood donors [9]. Nevertheless some provinces will have to put more effort into donor recruitment and retention so as to ensure self sufficiency in their blood supply.. Since 2004, IBTO has initiated a programme to enter into a contract fractionation agreement for the surplus of recovered plasma produced in its blood collecting centres. Although IBTO has used this project as a mean to improve national transfusion system through upgrading its quality assurance systems, IBTO fractionation project has played a major role in improving availability of plasma-derived medicines in Iran. During 2006-2007, this project furnished the Iran market with 44% and 14% of its needs to the intravenous immunoglobulin and albumin, respectively. Iranian experience showed that contract fractionation of plasma in

countries with organized centralized transfusion system, which lack national plasma fractionation facility, in addition to substantial saving on national health resource and enhancing availability of plasma-derived medicines, could serve as a useful means to improve national blood safety profile[6].

The prevalence of HBsAg has been evaluated in blood donations in Iran over the period from 1998-2007. A total of 14,599,783 donations were collected during these 10 years (Figure 3). The overall HBsAg prevalence rates declined from 1.79% in 1998 to 0.41% in 2007 as a result of improvements in donor recruitment and selection, implementation of automation in transfusion services, and a possibly decreasing HBV infection prevalence in the general population. A separate report has also examined the results from the viral screening of 6,499,851 donations from 2004 through 2007 for HBV, HCV, HIV, and syphilis. The overall prevalence of these viral markers was 0.56% for HBV, 0.004% for HIV, and 0.13% for HCV. There was a significant decrease in HBsAg prevalence from 0.73% in 2004 to 0.41% in 2007. Despite an increasing HIV prevalence in the general population of Iran, the prevalence of HIV decreased from 0.005% in 2004 to 0.004% in 2007. HCV prevalence showed a slight decline in blood donations from 0.14% in 2005 to 0.12% in 2007[5].

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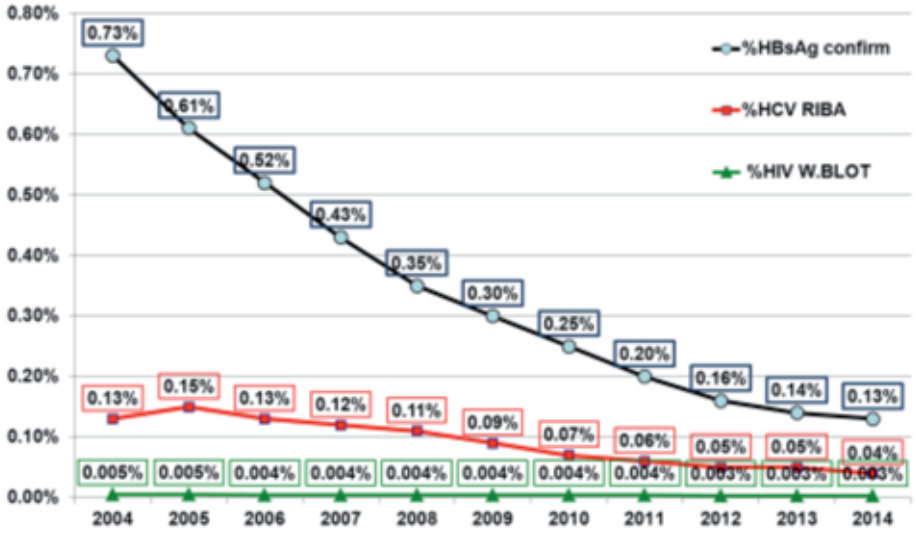

Figure 3. Prevalence of HBV, HCV and HIV in Blood Donation, year 2004-2014.

References

1. http://www.thalassaemia.org.cy/policy-issues/policy-issues/blood-safty-donation
2. Blood safety and availability WHO Fact sheet Updated July 2016.
3. Razjou F1, Maghsudlu M, Nasizadeh S, Zadsar; M,Transfus Apher Sci. 2012 Aug;47(1):13-6. doi: 10.1016/j.transci.2012.03.001. Epub 2012 Apr 20. The impact of donor selection on blood safety in Iran.
4. Cheraghali AM1, Eshghi P, Abolghasemi H; East Mediterr Health J. 2011 Jun;17(6):552-6. Social consequences of infected haemophilia cases in the Islamic Republic of Iran.
5. Kafi-abad SA1, Rezvan H, Abolghasemi H.; Transfus Med. 2009 Aug; 19(4):189-94. doi: 10.1111/j.1365-3148.2009.00935.x. Trends in prevalence of hepatitis B virus infection among Iranian blood donors, 1998-2007.
6. Cheraghali AM1, Abolghasemi H.; Haemophilia. 2009 Mar; 15(2):487-93. Plasma fractionation, a useful means to improve national transfusion system and blood safety: Iran experience.
7. Yenicesu I1, Dilsiz G, Oztürk G. J Int Med Res. 2004 Nov-Dec;32(6):671-5. Trop Doct. 2008 Apr;38(2):112-3. doi: 10.1258/td.2007.060187. Reduction of hepatitis B seroprevalence in blood banking units by combined utilization of self-exclusion forms and clinical evaluation of blood donation candidates in Turkey.
8. Hepgul S, Ozbayburtlu S, Altunay H, Ozsoy MF, Kosan E, Aksu Y, Yilmaz G, Pahsa A. J Int Med Res. 2004 Nov-Dec;32(6):671-5. Trends in major transfusion-transmissible infections among blood donors over 17 years in Istanbul, Turkey 1.
9. Mahtab Maghsudlu, Soheila Nasizadeh, Hassan Abolghasemi, and Shima Ahmadyar; Transfusion. 2009 Nov;49(11): 2346-51. doi: 10.1111/j.1537-2995.2009.02309.x. Epub 2009 Jul 14.Blood donation and donor recruitment in Iran from 1998 through 2007: ten years' experience_ 0230923462351.
10. Khadir M, Maghsudlu M, Gharehbaghian A. Evaluation of the attitude of Iranian women towards blood donation in eight provinces. Vox San 2004;87 Suppl 3:s93-145.
11. Hepat Mon. 2011 Jun 1; 11(6): 422–426. Published online 2011 Jun 1. PMCID: PMC3212795 Blood safety concerns in the Eastern Mediterranean region Abdol Majid Cheraghali
12. Hindawi SI1, Badawi MA, Raj ET, Gholam KA, Al-Weail SO, Azher F, Saudi Med J. 2016 May;37(5):538-43. doi: 10.15537/smj.2016.5.15084. The use of transfusion quality indicators as a tool for hemovigilance system implementation at a tertiary care center in Saudi Arabia.
13. Saudi Arabian Blood Services (2011) Are we on the right track? Available: https://www.transfusionmedicine.ca/sites/transfusionmedicine/files/events/Saudi%20Arabian%20Blood%20Service.pdf. Accessed 10 October 2014.

ARTICLE

Approach to haemovigilance in transfusion dependent patients

Salwa Hindawi,[1] Maha Badawi[2]

[1]Associate Professor in Haematology and Transfusion Medicine, King Abdulaziz University, Jeddah, Saudi Arabia; [2]Assistant Professor in Haematology and Transfusion Medicine, King Abdulaziz University, Jeddah, Saudi Arabia

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ABSTRACT

Transfusion of blood and blood components is a critical element for the delivery of healthcare services to patients. Tools to help improve the safety of the blood supply of a country include:

i) Clinical Transfusion Guidelines to improve transfusion practices.
ii) An Audit System to monitor adherence to the guidelines as well as the effects of adjustments to the guidelines.
iii) A Haemovigilance Program, which monitors the entire blood supply chain, develops measures and solutions to problems that might threaten the safety of blood component recipients, and monitors the implementation of these corrective actions.

Transfusion dependent patients are those who require frequent and long-term transfusion support to sustain life. Most of these patients have been diagnosed with one of the following conditions:

√ Thalassemia syndromes
√ Sickle cell anaemia
√ Chronic hemolytic anaemias
√ Bone marrow failure syndrome, aplastic anaemia, and myelodysplastic syndrome.

In additions to the potential complications of red blood cell (RBC) transfusion common to all recipients, there are special problems that are unique in transfusion dependent patients who are on chronic transfusion support. The most common of these complications are alloimmunization and iron overload. Specific measures must be considered to decrease the burden of these complications on patients' outcomes.

Introduction

Transfusion of blood and blood components is an integral part of healthcare services to patients with many haematologic conditions, such as thalassemia major (TM), sickle cell anaemia (SCA), and bone marrow failure syndromes. While blood transfusion is considered a lifesaving intervention in these patients, it is associated with certain risks. The risk of transfusion-transmitted infections has decreased significantly in many parts of the world given improved donor screening and introduction of pathogen reduction technologies. However, the non-infectious complications of transfusion are still a cause of concern around the world. These include errors and near-misses, in addition to acute or delayed transfusion reactions. These potential complications carry significant risk that affects the safety and well-being of transfusion dependent patients (TDP). Given how integral blood transfusion is to the outcomes of these patients, it is essential to follow good transfusion practices to:

- reduce the risk of alloimmunization to red blood cell (RBC) antigens.
- ensure maximum possible survival of transfused red cells.
- ensure the presence of optimal number of RBCs per unit transfused.

Haemovigilance (HV) is a relatively recent development in transfusion safety. Haemovigilance is now universally recognized as an essential safety and quality process along the blood transfusion chain. According to the definition by the

Correspondence:
sihindawi@yahoo.com

International Haemovigilance Network, Haemovigilance is a set of surveillance procedures covering the whole transfusion chain (from the collection of the blood and its components to the follow up of recipients), intended to collect and assess information on unexpected or undesirable effects resulting from the therapeutic use of labile products, and to prevent their occurrence or recurrence. The concept of Haemovigilance within the blood transfusion services encompasses many activities such as setting standards of quality and safety for the collection, testing, processing, storage, and distribution of human blood and blood components, in addition to surveillance of adverse transfusion events.

Health authorities are expected to develop and implement a Haemovigilance program to monitor and track adverse events, which can include processes to improve safety of transfusion, such as:

- Development of clinical transfusion guidelines, in addition to development of inter-professional educational activities to ensure competency of staff in use of blood and blood components.
- Appropriate labelling for accurate identification of samples and blood components.
- Implantation of hospital standards, clinical guidelines and protocols for safe blood transfusion, investigation and reporting of adverse events.
- Maintain traceability and documentation of transfused blood products.
- Notification and response to product recalls and look-backs.
- Active participation in a hospital transfusion committee
- Integration of Haemovigilance in the hospital quality system and implementation of processes to take corrective and preventive actions and for monitoring.
- Training and assessment of staff involved in all steps of clinical transfusion, blood sampling, laboratory practice, handling of blood units in clinical areas, bedside administration of transfusion and patient monitoring.
- Regular audits of clinical practices.
- Implementation of effective communication strategies between blood transfusion services and clinical departments within the hospital, and blood transfusion services within other hospitals.

Haemovigilance Approach to TDP

All patients who receive transfusion are at risk of developing transfusion reactions and are also at risk of transfusion-transmitted infections. TDP are at higher risk of development of an additional number of complications, including alloimmunization and iron overload.

To minimize the risks of such complications, Haemovigilance processes should be followed by all health care professionals involved in the transfusion chain in all hospitals. Assigning a Hemovigilance officer to support the education of staff involved in transfusion chain is encouraged.

Hemovigilance processes may involve:

- Promoting the appropriate use of blood and blood products.
- Provision and organization of education and training activities relevant to staff involved in transfusion process.
- Coordination of efforts toward collection and reporting of serious adverse reaction and events related to blood transfusion.
- Tracing and recall of blood component transfusion as needed.
- Participation in an active hospital transfusion committee.
- Reviewing quality indicators and audits.

In TDP, developing a Haemovigilance system (HV) aims to improve their quality of life through developing measures and solutions to problems that might threaten the safety of these patients, and monitor the implementation of these corrective actions.

An effective HV in TDP encompasses early application of preventive measures starting at the time of diagnosis, in addition to appropriate medical care and close follow up to minimize adverse events related to blood transfusion therapy. It is crucial to monitor iron overload through documenting units of packed red blood cell transfused, and through assessment of ferritin level and transferrin saturation. Such data must be recorded in a patient's transfusion history and made accessible to treating physicians. To reduce the risk of alloimmunization, TDP should ideally receive antigen matched RBC units. This is facilitated by wide-scale phenotyping (and/or) genotyping. In addition, it is advisable that blood components be leukodepleted. They can also can be washed and irradiated if needed.

Reducing donor exposure and improving availability of blood from rare group donors can be achieved through the use of apheresis procedures to collect blood form specific donors and also support treatment of complicated cases.

It is highly recommended to have national patient registries and patient-specific identification cards. In addition, planning and implementation of a national donor registry to maintain a list of volunteer phenotyped blood donors must be considered. Communication and collaboration between hospitals is an essential step towards improvement of patient care.

Iron Overload

Regular blood transfusion can lead to iron overload, which is a serious complication in TDP. Significant morbidity and mortality may result from iron overload in inadequately chelated patients. Chelation should be considered after one to two years of transfusion therapy, when the serum ferritin is greater than 1000 ng/dL, or when the hepatic iron is approximately 7mg/gm dry weight. The oldest available therapy is deferoxamine, a safe and effective treatment. However; it has to be administered parenterally which results in poor compliance. Oral chelating agents are also available, which are safe, effective agents with a positive benefit to risk ratio. Determination of liver iron by biopsy is recommended prior to initiation of iron chelation therapy as well as every 12 to 24 months (or as clinically indicated). Through not routinely available, liver iron can be also determined by superconducting quantum interference device (SQUID) or T2-star magnetic resonance imaging (MRI). Evaluation of ferritin level quarterly is an available option for follow up in most hospitals.

Alloimmunization

Development of antibodies against red cell antigens that the person lacks is called alloimmunization. A number of factors contribute to increasing the risk of alloimmunization, such as the RBC antigenic differences between the blood donor and the recipient, the recipient's immune status and the immunmodulatory effect of the allogeneic blood transfusion on the recipient's immune system. The transfusion dependent patient is at significantly high risk of alloimmunization, although the rates vary in the literature. In SCA, the rates range from 18 to 47%. Locally the rate is around 13%, while 11.5% of thalassemia patients have alloantibodies.

Patients who develop alloantibodies must receive group compatible packed red blood cell units that are negative for the antigen towards which the patient has an antibody and compatible on serologic cross-match.

Alloimmunization in general may be mitigated by a number of methods, including transfusion of phenotypically matched RBCs units, and universal leukodepletion.

Conclusions

Haemovigilance in TDP can lead to improvements in quality and safety of medical care provided to patients. This is facilitated by implementation of specific clinical guidelines for management of TDP and to have a national policy for extended red blood cell typing of patients including typing for C, E, e, c and K antigens before the first transfusion. Screening for new antibodies and matching for Rh including C, E, c, e and K antigens must be done for new patients. Using leukoreduced blood with pre-storage filtration, reporting of any adverse reaction, and follow up of corrective and preventive actions are essential. To improve survival in TDP, special attention must be paid for prevention, early diagnosis, and treatment of iron overload. The combination of transfusion and chelation therapy has dramatically extended the life expectancy of these patients.

References

Aide-memoire, National Haemovigilance System. World Health Organization. 2015.

Alboog AS, Tayeb TM, Alsager MO, et al. Red blood cell alloimmunization in sickle cell disease patients in Jeddah, Saudi Arabia: a pilot study. JKAU Med Sci. 2015;22(3): 17-22.

Ameen R, Al-Shemmari S, Al-Humood S, Chowdhury R, Al-Eyaadi O, Al-Bashir A. RBC alloimmunization and autoimmunization among transfusion-dependent Arab thalassemia patients. *Transfusion*. 2003;43 (11):1604-1610. doi:10.1046/j.1537-2995. 2003.00549.x.

Brittenham G, Griffith P, Nienhuis A et al. Efficacy of Deferoxamine in Preventing Complications of Iron Overload in Patients with Thalassemia Major. *New England Journal of Medicine*. 1994;331(9):567-573. doi:10.1056/nejm199409013310902.

Faber J. Worldwide overview of existing haemovigilance systems. *Transfusion and Apheresis Science*. 2004;31(2):99-110. doi:10. 1016/j.transci.2004.07.004.

Gader A, Al Ghumlas A., Al-Momen A. Transfusion medicine in a developing country-alloantibodies to red blood cells in multi-transfused patients in Saudi Arabia. *Transfus and Apher Sci.* 2008; 39:199–204.

Hendrickson J, Tormey C, Shaz B. Red Blood Cell Alloimmunization Mitigation Strategies. *Transfusion Medicine Reviews*. 2014;28(3):137-144. doi:10.1016/j.tmrv. 2014.04.008.

Langhi D, Jr., Ubiali EM, Marques JF, Jr., et al.Guidelines on Beta-thalassemia major - regular blood transfusion therapy: Associacao Brasileira de Hematologia, Hemoterapia e Terapia Celular: project guidelines: Associacao Medica Brasileira - 2016. Rev Bras Hematol Hemoter. 2016;38 (4):341-5.

Telen M, Afenyi-Annan A, Garrett M, Combs M, Orringer E, Ashley-Koch A. Alloimmunization in sickle cell disease: changing antibody specificities and association with chronic pain and decreased survival. *Transfusion*. 2014;55(6pt2):1378-1387. doi:10.1111/trf.12940.

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Medicinal products; accessibility and availability; experience of Egypt

Amal El-Beshlawy

Cairo University, Egypt

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Correspondence:

amalelbeshlawy@yahoo.com

In Egypt B-Thalassemia has been described since 1944. It is the most common hereditary chronic hemolytic anemia with a high carrier rate (5.5 to >9%). As there is no national prevention program, the number of patients are increasing (1000/1.5 million live birth) The registered cases in the big thalassemia centers in 2013 was more than nine thousands (Figures 1, 2 and 3).[1]

How we treat?

The corner stone of management is blood transfusion and iron chelation therapy. Supportive treatment Vit. D. folic acid, Hb F inducers L-Carnitine (Phenylbutyric acid derivative) and Hydroxycarbamide (Hydroxyurea) are common medications offered to our patients.

Chelation therapy in Egypt:

It was mainly Desferrioxamine (DFO), though it has a well-established impact on the survival of our patients its inconvenient mode of administration renders the compliance to be difficult among our patients with the increase of morbidity and mortality from iron overload.[2]

The era of oral iron chelators

Deferriprone (DFP) was the first oral iron chelator (1994) Followed by Deferasirox (DFX) in 2005. The compliance of our patients to chelation remarkably improved after the oral therapy. In a Thalassemia International Federation (TIF) survey in 2001 the compliance to DFO was 48% in the Egyptian patients. Our studies reported 80% of the patients are compliant to oral iron chelators.[3]

Availability of oral iron chelators

DFO& oral iron chelators (DFP & DFX) are available in Egypt with the license of the Ministry of Health. Egypt was a partner in the international studies for the safety and efficacy of oral iron chelators.[4]

The accessibility of the chelation therapy to the patients is through the thalassemia centers in the university hospitals, school medical insurance, the Egyptian Thalassemia Association (ETA), and the ministry of Health specialized committees (Figure 4).

Challenges

The high cost of oral iron chelators limit its availability to all patients and motivated the school insurance to limit its use to patients below the age of 10 years. Patient advocacy groups supported by the concerned physicians, ETA and TIF let the governmental institutes and the school medical insurance to support the availability of the oral iron chelators to all the thalassemic patients. This increases the expenditure for DFX from 19,000,000 in 2013 to 30,284,875 EP in 2016 (Figure 5).

Supportive medications

Vit. D and L-Carnitine are medications which our patients receive regularly from the time of the disease confirmed diagnosis.

L-Carnitine is a butyric acid derivative essential in myocardial energy production, it stabilize the red cell membrane, antioxidants and HbF inducer.[5,6]

The studies on L-Carnitine in our thalassemia patients showed improvement in cardiac function[7,8] Growth parameters and Physical fitness[9-11].

L-Carnitine being a butyric acid derivative our study reported an increase in the fetal hemoglobin production.[12] Previous studies reported the potential positive effect of L-Carnitine in thalassemia patients.[11]

Hydroxycarbamide (Hydroxyurea): It is the main medications for the treatment of thalassemia intermedia in Egypt based on our study and different international

published studies conforming the positive effect in adult and pediatric patients. It can decrease or stop the need for blood transfusion with the concomitant decrease of iron overload and the blood born viral transmission.[13-14]

All supportive medications are available and accessible to most of our patients through the caring centers (Figure 6).

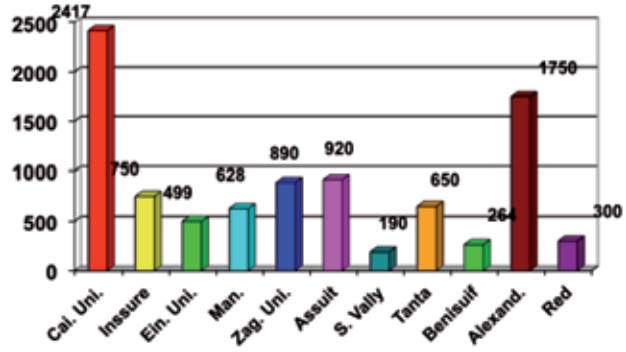


Figure 1. Registered cases of homozygous beta thalassemia in big centers of Egypt in 2013 (n=9258).

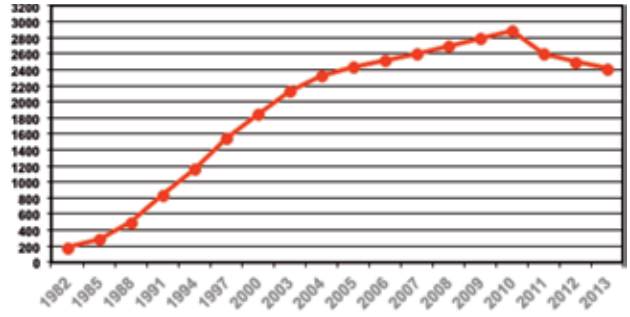


Figure 2. Number of thalassemia patients in the pediatric hematology clinic-Cairo university from 1982 to 2013.

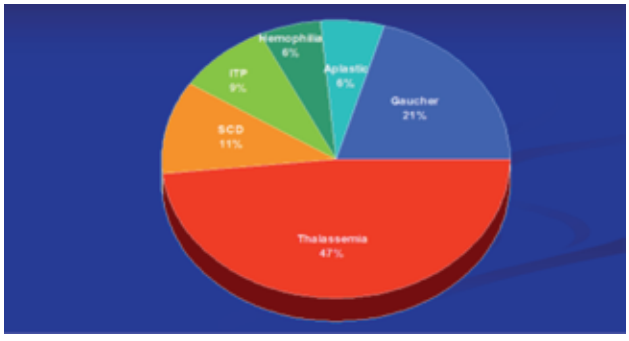


Figure 3. Follow up cases in hematology clinic of pediatric hospital of CU (n=10968).

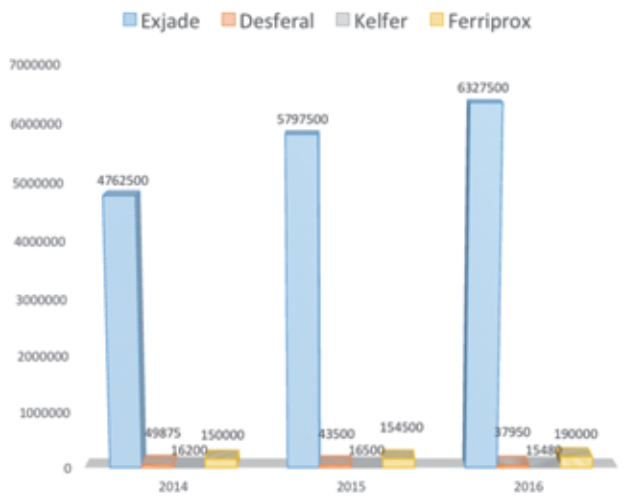
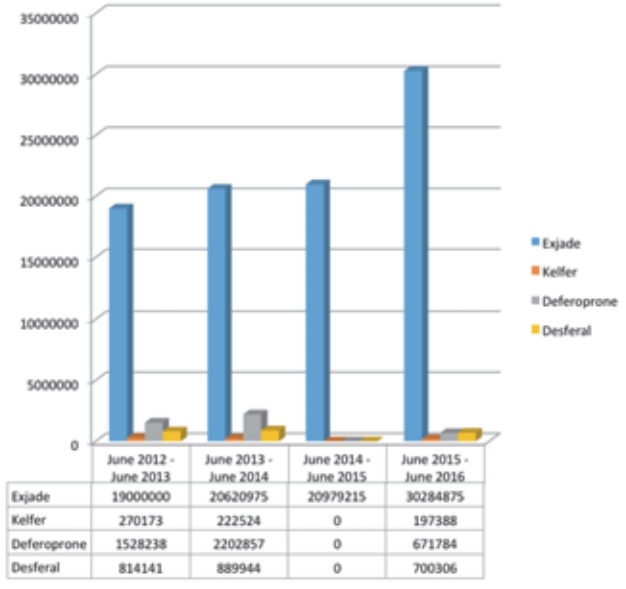


Figure 4. Health insurance and universities expenditure on iron chelation.



	June 2012 - June 2013	June 2013 - June 2014	June 2014 - June 2015	June 2015 - June 2016
Exjade	19000000	20620975	20979215	30284875
Kelfer	270173	222524	0	197388
Deferoprone	1528238	2202857	0	671784
Desferal	814141	889944	0	700306

Figure 5. Ministry of health (Schhool Insurance) yearly expenditure on chelating agents.

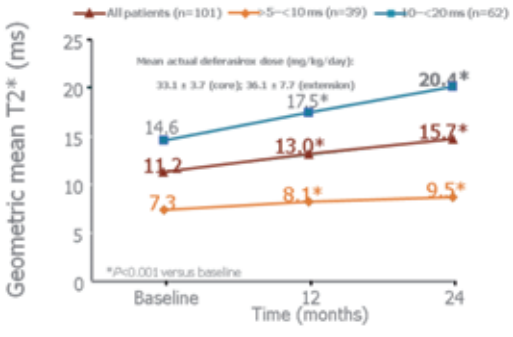


Figure 6. At 24 months, myocardial T2* improved significantly from baseline in patients with baseline T2* >5–<10 and 10–<20 ms (*P*<0.001).[15]

References

1. El-Beshlawy A1, Youssry I. Prevention of hemoglobinopathies in Egypt. Hemoglobin. 2009;33 Suppl 1:S14-20.
2. John B Porter, Michael Evangeli and *Amal El-Beshalwy:* The challenges of adherence and persistence with iron chelation therapy. International Journal of Hematology 2011, Nov. 94(5): 453-60. Epub 2011 Oct. 13.
3. El Beshlawy A . The Egyptian experience with oral iron chelators. Hematology. 2005;10 Suppl 1:174-5.
4. Ali Taher, Amal El-Beshlawy, Mohsen S Elalfy, Kusai Al Zir, Shahina Daar, Dany Habr, Ulrike Kriemler-Krahn, Abdel Hmissi, Abdullah Al Jefri Efficacy and safety of deferasirox, an oral iron chelator, in heavily iron-overloaded patients with beta-thalassaemia: the ESCALATOR study. European journal of haematology. 2009 Jun. 82 (6): 458-465.
5. Palmieri L, Ronca F, Malengo S, Bertelli A: Protection of beta-thalassaemic erythrocytes from oxidative stress by propionyl carnitine. Int J Tissue React. 1994;16(3):121-9.
6. Perrine SP et al. A short-term trial of butyrate to stimulate fetal-globin-gene expression in the beta-globin disorders. N Engl J Med. 1993 Jan 14;328(2):81-6.
7. A El-Beshlawy, L. Ragab, A. Abd El Fatah, E. Youssry, A. Makhloof and A. Taher. Improvement of Cardiac Function in Thalassemia Major Treated with L-Carnitine. Acta Haematologica 2004; 111:143-148.
8. A. El Beshlawy, E. Abdel Raouf, F. Mostafa, M. Talaat, H. Ismaeel, E. Aoun, A. V. Hoffbrand and A. Taher: Diastolic Dysfunction and Pulmonary Hypertension in Sickle Cell Anemia: Is There a Role for L-Carnitine Treatment? Acta Haematologica 2005; 557.
9. A El Beshlawy, SMA El Dayem, F El Mougy: Screening of growth hormone deficiency in short thalassaemic patients and effect of L-carnitine treatment- Arch Med Sci, 2010
10. Ye ilipek M.A. Hazar V. Yegin O :*L*-Carnitine Treatment in Beta Thalassemia Major. Acta Haematol 1998;100:162-163.
11. El-Beshlawy A, El Accaoui R, Abd El-Sattar M,Gamal El-Deen M H, Youssry I, Shaheen N, Hamdy M, El-Ghamrawy M :Effect of L-carnitine on the physical Fitness of thalassemic patients. Ann Hematology 2006; 86:31-34.
12. *Amal El-Beshlawy* (2), Mona Hamdy, Mona El Ghamrawy: Fetal globin induction in beta-thalassemia. *Hemoglobin.* 2009; 33 Suppl 1:S197-203. (ISSN: 0363-0269, IF: 1.304, Total citation: 7).
13. A El-Beshlawy, M El-Ghamrawy, MA EL-Ela, F Said Response to hydroxycarbamide in pediatric β-thalassemia intermedia: 8 years' follow-up in Egypt: Annals of Hematology December 2014, Volume 93, Issue 12, pp 2045.2050.
14. Karimi M, Musallam KM, Cappellini MD, Daar S, *El-Beshlawy A*, et al: Risk factors for pulmonary hypertension in patients with thalassemia intermedia: *Eur J Intern Med.* 2011 Dec;22(6):607-10. doi: 10.1016/j.ejim.2011.05.013. Epub 2011 Jun 29.

ARTICLE

The use of safe and effective medicinal products in the region

Mahmoud Hadipour Dehshal

Blood Transfusion Research Centre, High Institute for Research and Education in Transfusion Medicine, Tehran, Iran

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Introduction

Generics and Bio-similars are options to increase the access to treatment and balance the price of medicine in the contemporary market. They are copies of pioneer medicines that should be of the same efficacy and safety.[1] Health care professional and consumers ought to be assured about the quality of the copied drugs and their similarity to original ones by the national health authority. In 1984 two senators of the United States proposed an act called Hatch-Waxman based on which the distribution of generic drugs become easier and generic drugs producers just need to prove their products enjoy the similar bioequivalence of original brand-names.[2] According to the United States Food and Drug Administration (FDA) nearly 8 in 10 prescriptions in America are for generic drugs.[3]

In 2010 FDA imposed a guideline to allow the distribution of bio-similars (follow-on biologics) into the market. According to FDA it is mandatory for bio-similars' producers to conduct clinical trials to support the demonstration of similarity to original drug.[4]

The Middle East and North Africa (MENA) region is the next growth engine of global pharmaceutical sales. Medical awareness has caused growing demands for medicines and medical devices.[5] Recently the production of generics and bio-similars either manufactured by local companies or imported by multinational companieshas played a more effective role in the regional health system[6] with more access to necessary drugs. However, the lack of regulation, surveillance, and tracking over drugs production and distributions has increased consumer concerns over the quality of the drugs.

Drug quality

According to World Health Organization (WHO) "Quality assurance is a wide-ranging concept covering all matters that individually or collectively influence the quality of a product."[7] Thus, the goal of medicine quality assurance is both to access safe and effective medicines through the structured and valid methods and to maintain quality of medicine through the appropriate storage, distribution, monitoring, and use. To assure consumers about quality of drugs, the national authority have to develop norms and standards and enforce the organizations to continuously evaluate products' quality. WHO is considering the development of an international pharmacopeia in order to secure equal access to effective and safe care throughout the world. In addition medical professionals and consumers are able to play an effective role in securing quality of drugs because it is unquestionably accepted that part of the process of quality assurance needs to happen in post-marketing study.

Drug quality in MENA

MENA is an English acronym referring to the Middle East and North Africa region which extends from Iran to Morocco. The population of the region is estimated to be roughly about 381 million people about 6 percent of the total world population.[8] Multinational pharmaceutical companies have an eye on the attractive market while local pharmaceutical companies such as Hikma from Jordan and SPIMACO from Saudi Arabia aim to be the leaders in the region.[9] A study was conducted on Egypt pharmaceutical market as an example of MENA market, by which Quality and Safety control, Pharmaceutical legislation, GMP, and Pharmacological were proven to be lower than what it should have been in a standard system. Therefore, it can be inferred that a political movement is inevitable to push fresh legislation aimed at dealing especially with the companies which produce substandard medicines. In addition, there is an ambiguous area between producers and regulators in MENA market which ought to be modified and the independent regulator should restore consumers' confidence about quality of drugs. Regarding GMP, all countries located on MENA should revise their legislation and impose GMP standards on domestic companies and active pharmaceuti-

Correspondence:
mehregan_hadipour@yahoo.com

cal ingredient producers. In this case receiving advice from and making close collaboration with international organizations such as WHO can be an efffectivestrategy. Our findings show that some domestic producers have switched from approved active ingredients into cheaper and more inferior ones. To prevent such devastating strategy, inhibiting penalties should be imposed on the pharmaceutical producers which easily break the law. Furthermore, disagreement over the information in local products inserts versus brand-name inserts is another concern about pharmaceutical companies in the region. The national authority should implement appropriate measures to remove misleading data from pharmaceutical products and make consumers ensured that they can receive useful information about their prescribed medicine. Since MENA lies into growing markets, competition for penetrating into it would make deviation from the ethical pharmacy; hence, self-regulation and national regulation should be implemented so that domestic and foreign companies market their products based on the pharmaceutical ethical codes. To fulfill the goal, NGOs should start with them and accordingly push governments and producers to impose ethical codes.

Conclusions

Nowadays MENA is an attractive market for pharmaceutical companies especially in the field of thalassaemia. Due to the lack of financial resources, decision makers support generic and domestic products with the strategy addressing the concern of consumers about the quality and safety of the products. To make societies confident about the standardization of medicine, the better legislation and well-organized pharmaceutical authority is required.

References

1. Understanding Generic Drug, available fromhttp://www.fda.gov/Drugs/ResourcesForYou/Consumers/BuyingUsingMedicineSafely/UnderstandingGenericDrugs
2. Fact Sheet: What's Involved in Reviewing and Approving Generic Drug Applications? available fromhttp://www.fda.gov/Drugs/ResourcesForYou/Consumers/BuyingUsingMedicineSafely/UnderstandingGenericDrugs/ucm506040.htm
3. Fact about Generic Drugs. Available from: http://www.fda.gov/Drugs/ResourcesForYou/Consumers/BuyingUsingMedicineSafely/UnderstandingGenericDrugs/ucm167991.htm
4. Information on Biosimilars. Available from http://www.fda.gov/Drugs/DevelopmentApprovalProcess/HowDrugsareDevelopedandApproved/ApprovalApplications/TherapeuticBiologicApplications/Biosimilars/
5. Report from: Middle East and North Africa. Jill E. Sackman. Pharmaceutical Technology. Oct 02, 2013
6. Industry Trend Analysis - Advances To Medicine Dispensing Will Improve Market Attractiveness - NOV 2016. Available from http://www.pharmaceuticalsinsight.com/industry-trend-analysis-advances-medicine-dispensing-will-improve-market-attractiveness-nov-2016
7. Medicines quality assurance. Available from http://www.who.int/medicines/areas/quality_safety/quality_assurance/en/
8. Middle East and North Africa. Availble from http://www.worldbank.org/en/region/mena
9. The Outlook for Pharmaceuticals in the Middle East and North Africa. Available from http://www.reportlinker.com/p05703 64/The-Outlook-for-Pharmaceuticals-in-the-Middle-East-and-North-Africa.html #utm_source=prnewswire&utm_medium=pr&utm_campaign=Managed_care

2nd MEGMA Conference on Thalassaemia & other Haemoglobinopathies
Amman - Jordan, 11-12 November 2016

SCIENTIFIC PROGRAM - POSTER ABSTRACTS

BLOOD TRANSFUSION ADEQUACY AND SAFETY

ASSESSMENT OF A COHORT OF β-THALASSAEMIA PATIENTS: NEW CHALLENGES

Mohamed Bradai[1], Nouria Benmouaffek[2]

[1]Hématology Department; [2]Pediatric Department, CHU of Algiers Algeria

Background: The combination of transfusion and chelation therapy has dramatically extended the life expectancy of Beta-thalassemic patients who can now survive into their fourth and fifth decades of life. The goal of long-term transfusional support is to maintain the patient's hemoglobin level at 9-10g/dL. However, complications are still frequent and affect the patients' quality of life, particularly in countries with limited resources. The main objective of this study is to determine the prevalence of prominent complications in a cohort of Algerian thalassemia patients. *Materials and Methods:* This study was conducted to assess 81 polytransfused Beta-thalassemic patients, mean age 16 years (6-31), sex ratio: 37M/44G: We assess blood transfusion regime, pre-transfusion hemoglobin, iron overload, by serum ferritin level, hepatic iron concentration. Bone mineral density (BMD) measurements at lumbar and femoral regions have been done using dual x-ray absorptiometry. We assess Growth and pubertal maturation (Taner stage), and dosing gonadic hormone dosage, calcium, vitamin D, Parathyroid, and thyroid hormone. *Results:* The pretransfusion hemoglobin level was maintained between 6 and 9 g/dL in 90% of patients. Additionally, 5% had levels less than 6 g/dL, 5% from 9 to 11 g/dL. Fifty-five percent of patients had undergone splenectomy at a median age of 9 years (range, 1-31 years). Ferritin levels ranged from 147 to 13500 ng/mL (median, 4992 ng/mL). Among 44 patients assessed, 77% had values of 15 mg/g dry weight or higher, 50% LCI>43 mg/gdw, 20% had moderate LCI 15 to 7 mg/gdw, and only 1 patient <7mg/gdw. Hepatic biopsy, performed in 26 patients (assessed for bone morrow transplantation), all had hepatic fibrosis moderate to severe (Metavir score). BMD performed in 49 patients, prevalence of lumbar osteoporosis and osteopenia were 38% and 62%. Dosage of vitamin D performed in 34 patients noted: 5 <10ng/ml, 17 [10-20ng/ml], 8 [20-30ng ml], 4 normal >30ng/ml. Twelve patients receive Biphosphonate treatment. Short stature was seen in 65% of our patients . Hypogonadism was diagnosed in 22,9% of 74 patients who had reached pubertal age: 50% of hypogonadic females and males were receiving hormonal substitution. No patient had Hypoparathyroidism and primary hypothyroidism Three cases of primary amenorrhea, and 2 diabetes aged 17 and 31 years. Despite high ferritine serum no patients had heart disease requiring medication. *Discussion:* The goal of long-term hypertransfusional support is to maintain the patient's hemoglobin level at 9-10 g/d. This threefold is rarely reached in our patients, for mixture of reasons (compliance, limited blood pack). Iron chelation has been formerly limited, reason of the high prevalence of hemochromatosis in the majority of our patients. Intensification of iron therapy is now assessed to obtain reversal tissues lesions. Well treated 90% of thalassemics patients reached normal puberty; in contrast, in our group of patients, only 20% achieved normal pubertal status after 16 years. Poor pubertal growth and impaired sexual maturation, and endocrine abnormalities in children, adolescents and young adults have been observed because of conventional treatment deficiency, substitutive hormonal therapy is indicated. BMD is a good index of bone status in patients with Thalassemia and should be done in these patients annual. *Summary:* High prevalence of complications among our thalassemics signifies the importance of more detailed studies along with therapeutic interventions. In conclusion, the survival of patients with thalassemia major improving, but the prevalence of severe complications is still high.

BONE DISEASE

EVALUATION OF BONE MINERAL DENSITY IN PATIENTS WITH HAEMOGLOBIN H DISEASE

Mehran Karimi[1],Tahereh Zarei,[1] Sezaneh Haghpanah,[1]
Shirin Parand[1], Hossein Moravej[2],
Mohammad Hossein Dabbaghmanesh[3],
Gholamhossein Ranjbar Omrani,[3] Zohre Zahedi[1*]

[1]Hematology research center, Shiraz University of medical science, Shiraz, Iran; [2]Department of pediatrics, School of Medicine, Shiraz University of medical science, Shiraz, Iran; [3]Endocrine and Metabolism Research Center,Nemazee Hospital, Shiraz University of Medical Sciences, Shiraz, IR Iran

Background: This study was conducted to assess bone mineral density (BMD) and bone mineral content (BMC) of patients with hemoglobin H (HbH) disease. *Methods:* In this cross sectional study, we evaluated BMD and BMC in 21 patients over age 10 out of 37 Iranian patients with HbH disease, from November 2014 to August 2015. BMD and BMC were measured by dual energy X-ray absorptiometry of lumbar spines and femur neck. Also, association of BMD with sex, age, hemoglobin, calcium, phosphorus and serum ferritin level was evaluated. All patients were non-transfusion dependent and took folic acid 5mg/kg orally once a day. An informed written consent form was filled by those who accepted to take part in the study. *Results:* Mean age of the patients was 20.23±9.87 years and ranged from 11-56 years old. Prevalence of patients with BMD in the lumbar spine below the expected range for age was 33.3%. Also, 14.3% of our patients suffered from BMD with below the expected range for age in the femur neck region. There was no significant association in any of the evaluated variables with BMD in the lumbar and femur neck (P value>0.05). A total of 5 (13.5%) patients were splenectomized.Bone mineral density showed no significant association with age, sex and splenectomy (P value>0.05). *Conclusions:* Data regarding bone density in HbH disease is limited, osteoporosis as a common complication of thalassemia inter-

mediate syndrome should be considered even in HbH which shows its prevalence is less than beta thalassemia intermedia. *Keywords:* Evaluation, Hemoglobin H, Bone mineral density.

WHICH PAMIDRONATE PROTOCOL IS THE BEST FOR TREATING OSTEOPOROSIS IN β-THALASSAEMIA MAJOR?

Mandana Zafari[1], Mehrnoush Kowsariyan[2*]

[1]Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [2]Professor of Pediatrics, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran

Background and Purpose: One of the most common complications in thalassemia major patients is osteopenia and osteoporosis. In this study, we compare the therapeutic effect of two treatment protocols involving infusion of 45 mg of Pamidronate injection every 6 weeks (P45) and 90 mg Pamidronate infusion every 4 weeks (P90). *Methods:* Bone mineral density was measured using dual energy X-ray absorptiometry (DEXA). Z-score of lumbar vertebra (as L total) and the femoral head (as F total) were compared before and after administration of two protocols. Bone density between the two groups was compared by Student t test and by the paired t test before and after the intervention. Data were analyzed using SPSS (18). *Results:* Ninety-one patients were treated with Pamidronate 45 mg (P45), and 36 patients were treated with Pamidronate 90 mg (P90). Ninety-one and 36 patients received P45 and P90 protocol, respectively. Mean age was 29.4±8.1 and 30.9±8.0 years old in P45 and P90 groups, respectively. Sixty-two and 58% of P45 and P90 group were female. The means of F total were –1.73±1.11 and –1.47±0.92 before and after treatment in patients P45 (P=0.01) and were –1.83±0.75 and –1.57±0.99 in group P90 (P=0.005), respectively. Before treatment, the means of L total were –2.95±0.81 and –2.92±0.66 (P=0.8) and after treatment were –2.53±1.13 and 2.81±0.98 (P=0.1) in P45 and P90 groups, respectively. In P45, between the mean of L total was statistically significant difference before and after treatment (effectiveness of both protocols). *Conclusions:* As the medication is expensive and should be administrated parenterally, we recommend P45 protocol which is less expensive with fewer injections. *Keywords:* Beta (β)-thalassemia major, Osteopenia, Osteoporosis, Bone mineral density, Pamidronate, DXA.

OSTEOPOROSIS AMONG β-THALASSAEMIA PATIENTS IN THE WEST BANK OF PALESTINE

Bashar Karmi[1], Akram Kharroubi[2], Walaa Shamasna[1], Elias Saba[3]

[1]Medicare, Ramallah; [2]Al-Quds University, Faculty of Health Professions, Abu Deis; [3]Palestinian Osteoporosis Prevention Society [POPS], Bethlehem, Palestine

Purpose: This study was carried out to evaluate the bone mineral density (BMD) in thalassemia major by DEXA and calculate the hemoglobin threshold value in patients with thalassemia that develop osteoporosis. *Background:* Hemoglobinopathy in thalassemia patients is associated with bone disorders like deformities, bone pain, fractures, osteopenia and osteoporosis. Apart from disease process per se, high-dose iron chelating therapy with desferrioxaminemay also contribute to osteopenia and osteoporosis, which is a significant cause of morbidity in these patients. It is characterized by low BMD and disruption of bone architecture, resulting in reduced bone strength and increased risk of fractures. *Methods:* Thalassemia patients from different governorates of the West Bank of Palestine. From a total of 551 thalassemia patients in the West Bank, 135 agreed to participate and singed a consent form. BMD was estimated as well as hemoglobin levels (114 of them were below the age of 30 years). Subjects were classified as having osteoporosis if the z score was less than -2 at any of the three sites (lumber spine, femoral neck and total hip). *Results:* In thalassemia patients, there was no difference in mean age, BMI and hemoglobin levels of males and females, BMD of femoral neck and total hip was statistically significantly lower in females compared to males. However, lumber spine BMD was not statistically different. Chi square analysis showed no significant difference between males and females in the number of subjects having osteoporosis at the three sites or any site. Osteoporosis subjects have statistically lower weight but have no difference in BMI. Hemoglobin was statistically significantly lower mean values. Pearson correlation showed significant positive correlation between hemoglobin and BMD, but the correlation coefficient was higher between hemoglobin and lumber spine BMD (r=0.444) compared to femoral neck (r=0.291) and total hip (r=0.224). Weight was also positively correlated with BMD at the three sites. Analysis of the ROC curves for hemoglobin using BMD as a reference standard at any of the three sites showed that hemoglobin threshold for thalassemia subjects to have osteoporosis was ≤9.3 g/dL with area under the ROC curve (AUC) equals 0.699 and the percentage of osteoporosis among thalassemia subjects of the study was 67.4%. The sensitivity and specificity were 87.9% and 47.7%, respectively. Only ROC curve for hemoglobin using lumber spine BMD as a reference standard was valid (or using osteoporosis at any of the three sites). When lumber spine was used to identify subjects having osteoporosis, the AUC was 0.727, the sensitivity and specificity were 82.8% and 54.4%, respectively. The AUC of hemoglobin using BMD for total hip and femoral neck were not significant from random (0.50). *Conclusions and Recommendations:* Two-third of thalassemia patients had osteoporosis which is close to the international prevalence of 50%. The ROC curve analysis revealed that the threshold of hemoglobin associated with osteoporosis was ≤9.3 g/dL. It is recommended that hemoglobin levels be kept above 9.3 g/dL for the purpose of bone health.

DIAGNOSTIC AND MONITORING TECHNIQUES

EVALUATION OF RENAL IRON DEPOSITION USING T2* MAGNETIC RESONANCE IMAGING IN THALASSAEMIA (LARGE COHORT STUDY)

Shirkavand Afshan[1*], Mokhtari Hesari Parisa[2], Akhlaghpoor Shahram[3*], Mansoori Bahar[4], Azarkeivan Azita[5], Hashemieh Mozhgan[6]

[1]PhD, Medical physicist, Pardis Noor Medical Imaging center, Tehran, Iran; [2]Biostatistician, Cancer Quality of Life Dept., Breast

Cancer Research center, ACECR, Tehran, Iran; [3]Professor of Radiology, Tehran University of Medical Sciences, Pardis Noor Medical Imaging center, Tehran, Iran; [4]Radiology resident. Radiology department, University Hospitals Case Medical center. Cleveland. Ohio, USA; [5]Associate professor of Pediatrics Hematology Oncology, Blood Transfusion Research center, Institute for Research and Education in Transfusion Medicine, Thalassemia Clinic, Tehran, Iran; [6]Associate professor of Pediatrics Hematology Oncology, Imam Hossein Medical center, Shahid Beheshti University of Medical Sciences, Tehran, Iran

Background: Patients with thalassemia require chronic blood transfusion for survival. Multi-organ hemosiderosis is a known complication.T2-Star (T2*)MRI has been introduced as a non-invasive tool for detecting iron overload in liver and heart in these patients. *Objectives:* The purpose of this study is twofold: 1) Determining of kidneyT2* MRI values and prevalence of iron overload in Iranian thalassemia patients. 2) Investigating the correlation between serum ferritin and renal, myocardial and hepatic hemosiderosis. *Methods:* From July 2012 to June 2014, 821transfusion dependent Major and intermedia tethalassaemia patients (age range 10-50 years) were enrolled and underwent MRI. Iron values were calculated and averaged in different region of interest ROIs using fast-gradient-echo multi-echo T2*sequences. The results were analyzed statistically. *Results:* 19.6% patients with mean age (27.98.6years) had pathological renal iron content less than 36 ms while mean kidney T2* of the total population was 50.26ms. There wasa weakrelationship between age and kidney T2*relaxation time (r=0.14, p-value=0.005). A weaknegative correlation between kidney T2*relaxation time and serum ferritin (r=-0.40, p-value<0.0001) was noted. For liver and heart, T2*relaxation time correlated weakly by renal T2*relaxation time (r=0.31, 0.36 respectively, p-value<0.0001). *Conclusions:* Renal hemosiderosis was seen in some of the thalassemia patients. A weak correlation between hemosiderosis of liver, heart and, serum ferritin level with kidney T2* was noted. Monitoring renal iron overload in thalasseamic patients using MRI T2* method could be beneficial for optimizing iron chelating regimen of patients and preventing its toxicity effects in kidney. *Key words:* Iron overload, Thalassaemia, Kidney, MRI T2*.

SERUM FERRITIN LEVEL AS A SURROGATE TO LIVER IRON CONCENTRATION AND CARDIAC T2* FOR DETERMINATION OF HEPATIC AND CARDIAC IRON OVERLOAD IN PATIENTS WITH THALASSAEMIA MAJOR

Hassan M. Moukhadder[1,2], Rayan Bou Fakhredin[1], Joseph Roumi[1,2], Hassan Beydoun[1], Therese Abou Nasr[2], Suzanne Koussa[2], Ali T. Taher[1,2]

[1]American University of Beirut Medical Center, Department of Internal Medicine, Beirut, Lebanon; [2]Chronic Care Center, Hazmieh, Lebanon

Background: It is very well established that iron toxicity is the hallmark of life-threatening complications in thalassemia. The utility of serum ferritin level, a marker of total body iron stores, as a less costly and more readily available tool in place of liver iron concentration (LIC) and cardiac T2* has been investigated in the literature. Our goal is to study the association between serum ferritin levels and LIC on one hand and serum ferritin level and cardiac T2* on the other hand in thalassemia major (TM) patients who are chelated with deferasirox, deferoxamine, or deferiprone. *Methods:* A total of 70 patients with TM, aged 12 to 51 years, were included and classified into 3 subgroups according to the chelating agent used. Serum ferritin level, LIC, and cardiac T2* were determined for each patient. SPSS was used for determination of statistical significance of the correlations noted in each subgroup using the Pearson correlation coefficient. *Results:* In the total 70 patients, LIC and serum ferritin level were found to be positively correlated (r=0.429, p=0.002). Only in the subgroup of patients receiving deferasirox therapy, however, LIC and serum ferritin level were also found to positively correlate (r=0.324, p=0.034). No other statistically significant associations were determined. *Conclusions:* Our findings echo those reported in the literature, where serum ferritin level was found to be positively correlated with LIC. While all chelating drugs decrease systemic iron burden, deferasirox is able to specifically target liver iron, as reflected by our analysis. This is particularly relevant in our practice where deferasirox is the most commonly prescribed iron chelator in TM. Other well-established associations such as the negative relationship between serum ferritin level and cardiac T2*, especially in deferiprone users, could have been reported had the corresponding sample size been larger.

DETERMINATION OF CARDIAC FUNCTION BY SERUM FERRITIN LEVEL, CARDIAC T2*, AND LIVER IRON CONCENTRATION IN PATIENTS WITH THALASSAEMIA MAJOR

Hassan M. Moukhadder[1,2], Joseph Roumi[1,2], Rayan Bou Fakhredin[1], Hassan Beydoun[1], Therese Abou Nasr[2], Suzanne Koussa[2], Ali T. Taher[1,2]

[1]American University of Beirut Medical Center, Department of Internal Medicine, Beirut, Lebanon; [2]Chronic Care Center, Hazmieh, Lebanon

Background: Cardiotoxicity due to iron overload remains to be the leading cause of death in patients with thalassemia major (TM). While serum ferritin levels mirror systemic iron overload, iron burden in the heart and liver is measured using a non-invasive MRI technology that detects myocardial and hepatic iron concentrations, respectively. The aim of this study is to determine the correlation between left ventricular ejection fraction (LVEF) and serum ferritin level, cardiac T2*, and liver iron concentration (LIC) in patients with TM. *Methods:* A total of 70 TM patients, 38 males and 32 females aged 12 to 51 years, were studied. All patients were receiving iron chelation therapy with deferasirox, deferoxamine, or deferiprone. Serum ferritin level was classified as (500-1000 ng/mL), (1000-1500 ng/mL), or (>1500 ng/mL). LIC was categorized as (<3 mg/g dry weight [dw]), (3-7 mg/g dw), or (>7 mg/g dw). Finally, cardiac T2* was stratified as (>20 ms), (10-20 ms), or (<10 ms). IBM SPSS was used for statistical analysis and for determination of potential correlations between the investigated variables. *Results:* LIC was found to negatively correlate with LVEF (r= 0.477, p=0.025) in patients with LIC >7 mg/g dw, while cardiac T2* was found to positively correlate with LVEF (r=0.999, p=0.024) in patients with T2* <10 ms. No other statistically significant associations were found. *Conclusions:* Our results echo synonymous data in the literature, which advocates the predictability of cardiac function deterioration by severe myocardial siderosis. However, the negative correlation

between severe hepatic iron overload and cardiac function in our analysis does not mirror analogous findings in the literature, which speak against a significant association between the aforementioned variables. This might be accounted for by our small population size (2 patients), which is one of our principal study limitations.

ENDOCRINE COMPLICATIONS

THE SAUDI EXPERIENCE ON THE EFFECTS OF ENDOCRINE COMPLICATIONS AND ITS MORBIDITY IN THALASSAEMIA

Soad Khalil Al Jaouni

Department of Hematology, King Abdulaziz University Hospital, Faculty of Medicine, King Abdulaziz University, Jeddah, Kingdom of Saudi Arabia

Background: Endocrine dysfunction in thalassemia is a common complication due to excessive iron overload and suboptimal chelation. Disturbances in growth pubertal development, abnormal gonadal functions, impaired thyroid, parathyroid, adrenal functions, diabetes. Bone complication are commonly encountered. *Methods:* Summary of the two retrospective studies. 1990-2004: Survival and disease complication of thalassemia major (TM) in single institute at King Abdulaziz University Hospital (KAUH), Jeddah, Saudi Arabia. *Results:* A total of 278 patients were enrolled, the endocrine complication were increased with age and iron overload. Four patient (1%) died due to endocrine dysfunction. 2006-2010: Endocrinopathies in children and adolescents with β thalassemia major at KAUH total of 143 patients enrolled in the study. *Results:* Vitamin D3 deficiency was the commonest (56%) endocrinopathy in both children and adolescents with β-TM, followed by puberal delay (29.37%) and hypothroidism (21%); 7.6% of the patients had no endocrinopathhies, and 45.5% had 3 or more endocrinopathies. Growth hormone deficiency was observed in 12.58% of the patients. The overall mean and SD serum ferritin levels were 3400.86 and 3067.43 ng/mL, respectively. Iron overload worsened as the children grew older; the mean and SD serum ferritin levels were 2893 and 1919 ng/mL, respectively for pre-adolescents and 4299 and 4276 ng/mL, respectively for adolescents (p=0.0368). *Conclusions:* Comprehensive care with early detection and recognition of disease complication and appropriate transfusion regimen and chelation therapy are the keys to managing, preventing disease complication improve quality of life on thalassemia.

THE ASSOCIATION OF PANCREATIC MRI R2* WITH FASTING AND 2-HOUR POSTPRANDIAL BLOOD GLUCOSE AMONG THALASSAEMIA MAJOR PATIENTS IN INDONESIA

Pustika Amalia Wahidiyat, Siti Ayu Putriasih, Nitish Basant Adnani

Pediatric Hematology-Oncology Division, Department of Child Health, Faculty of Medicine Universitas Indonesia, Dr. Cipto Mangunkusumo Hospital, Jakarta, Indonesia

Background: Iron overload assessment is of pertinent importance among thalassemia major (TM) patients. While the use of MRI to estimate hepatic and cardiac iron stores has largely replaced other conventional methods, there are limited studies that evaluate pancreatic siderosis and its correlation with blood glucose results in TM patients. *Objectives:* This study aims to identify the relationship between pancreatic MRI R2* and blood glucose results among TM patients in Indonesia. *Methods:* This study evaluated TM patients with pancreatic MRI R2* and fasting and 2-hour postprandial blood glucose examinations results, who were grouped into normal (pancreatic R2* <30 Hz), mild (pancreatic R2* 30-100 Hz), or moderate (pancreatic R2* >100 Hz) pancreatic siderosis. The fasting and 2-hour postprandial blood glucose results for the three groups were then compared. *Results:* Sixty-three TM subjects were included, with an age range of 7-18 years old. Among them 35 (55.5%) subjects were categorized as normal, while 25 (39.7%) had mild, and 3 (4.8%) had moderate pancreatic siderosis. One patient had an impaired fasting glucose (127 mg/dL), and 2 others had an impaired glucose tolerance (141 mg/dL and 147 mg/dL). Data analysis revealed no significant differences in the fasting and 2-hour postprandial blood glucose levels between the three groups. *Conclusions:* The results of this study is in accordance with previous studies which demonstrate that while pancreatic R2* may be sensitive for glucose dysregulation, most patients may still have normal blood glucose results. Blood glucose testing and MRI are recommended for TM patients after age 10. However, in this study, pancreatic siderosis was already seen in 44.5% of the patients, the youngest being 7 years old. Further rigorous trials are warranted to identify whether pancreatic R2* should be conducted at an earlier age for TM patients. *Keywords:* MRI R2*, pancreatic siderosis, blood glucose, thalassemia.

EPIDEMIOLOGY AND PREVENTION

CARRIER FREQUENCY OF α-THALASSAEMIA MUTATIONS AMONG NEWBORNS IN NORTHERN IRAN

Mohammad Reza Mahdavi[1], Mernoush Kosaryan[2], Hossein Karami[3], Merhad Mahdavi[4], Hossein Jaladi[5], Payam Roshan[6*]

[1]Department of Laboratory Medicine, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [2]Department of Pediatrics, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [3]Pediatrics Hematology and Oncology, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [4]Veterinary Medicine, Sina Mehr Research Center, Sari Iran; [5]Research in Thalassemia, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [6]Immunology, Sina Mehr Research Center, Sari, Iran

Background and Purpose: Alpha Thalassemia is one of the most prevalent hemaglobinophaties worldwide. Alpha thalasseima patients may represent wide spectrum of symptoms ranging from asymptomatic to severe life threatening anemia. This study was done to assess the carrier frequency of alpha globin gene mutations among newborns in north of Iran. *Methods:* In this descriptive study, 412 cord blood samples of neonate from Amir Mazandari Hospitali were randomly selected during 2012. Genomic DNA was extracted using phenol-chloroform method. Multiplex Gap- PCR and PCR-RFLP methods were applied in order to detect three common gene deletions, one

triplication and one point mutation. *Results:* Total allelic frequency of investigated mutations was 0.0825. The -α3.7 deletion with allelic frequency of 0.0485 was the most prevalent mutation among 412 neonates. Allelic frequencies of -α4.2, αααanti3.7 triplication and α-5nt mutations were 0.0206, 0.0109 and 0.0024; respectively and -Med double gene deletion was not detected. *Conclusions:* Most mutated cases had single gene deletion that is asymptomatic while -Med double gene deletion was not detected among the neonates. Therefore, there is low probability of a child birth with Hb H disorder in the region. *Keywords:* Alpha Thalassemia, Alpha globin, Gene Mutation, Newborn, Iran.

NON-INVASIVE PRENATAL DIAGNOSIS OF β-THALASSAEMIA BY DETECTION OF THE CELL FREE FETAL DNA IN MATERNAL CIRCULATION; A SYSTEMATIC REVIEW STUDY AND META-ANALYSIS

Mandana Zafari[1], Mehrnoush Kowsaryan[1*], Pooria Gill[1], Abbass Alipour[1], Mohammad Reza Shiran[2], Hossein Jalalli[1], Ali Banihashemi[3], Fatemeh Fatahi[4]

[1]Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [2]Department of Physiology & Pharmacology, School of Medicine, Mazandaran University of Medical Sciences; [3]Amir Kola Genetic laboratory, Babol University of Medical Sciences, Sari, Iran; [4]Mazandaran University of Medical Sciences, Sari, Iran

Introduction: The discovery of cell free fetal DNA (cffetal-DNA) in mother circulation has opened up new opportunities in non- invasive prenatal diagnosis. Since 2002 many studies have been examined the sensitivity and specificity of this method for NIPD of Thalassemia in couples at risk of having affected baby. Here we report a review and meta-analysis of the published papers to evaluate the use of this method for early NIPD of this disease. *Methods:* We applied a sensitive search of multiple databases including; PubMed, SID, ProQuest, Springer, Ovid, Biomedical Center, Clinical Key, Global Health, JAMA, and Science Direct. *Results:* 9 studies, including 250 pregnancies and 250 conventional prenatal diagnosis results met our inclusion criteria. Overall mean sensitivity was 99% (95% confidence interval 69% to 100) and mean specificity was 99% (95% confidence interval 89% to 100). *Conclusions:* Based on this review and meta-analysis we concluded that prenatal diagnosis of Thalassemia, in suitable cases can be determined with high level of accuracy by analyzing cffDNA. Using cffDNA in prenatal diagnosis to replace existing conventional and invasive methods can remove or reduce the risk of miscarriages. *Key words:* Non invasive prenatal diagnosis, Thalassemia, cffDNA, maternal plasma.

HIGH RESOLUTION MELTING ANALYSIS FOR NON-INVASIVE PRENATAL DIAGNOSIS OF IVS-II-1 (G-A) FETAL DNA IN MINOR β-THALASSAEMIA MOTHERS

Mandana Zafari[1], Pooria Gill[1,2*], Mehrnoush Kowsaryan[1], Abbass Alipour[1,2], Ali Banihashemi[3]

[1-] Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [2]Immunogenetics Research Center, Mazandaran University of Medical Sciences, Sari, Iran; [3]Amir Kola Genetic Laboratory, Babol University of Medical Sciences, Sari, Iran

Background and Purpose: β- thaslassemia carrier couples are at risk of having affected baby; prenatal diagnosis (PND) plays an essential role in obstetrical care of them. Routine prenatal diagnosis such as; chorionic villus sampling (CVS) and amniocentesis are invasive methods. Detection of cell free fetal DNA in maternal plasma is a non invasive method for determination of paternally inherited mutation in fetus. The aim of this study is determine the sensitivity and specificity of this method. *Methods:* Genomic DNA from 35 β-thalassemia minor couples whose pregnancy was at risk for homozygouse β- Thalassemia was used. We focused exclusively on samples in which the father was carrier for IVS II-I G→A mutation and the mother had been genotyped to carry another β-globin gene mutation. Ten milliliters of maternal blood from each pregnant woman were collected, plasma was separated, and it stored at -80°C until analysis. We were masked as to the identity the samples with numerical coding system, so, these were examined in a blinded manner. Extracted DNA analyzed by HRM-Real time PCR for detection of paternally inherited mutation. The CVS sample was obtained Trans abdominal puncture with a 23-gauge needle under ultrasound guidance. The samples were taken from CVS analyzed by reveres dot blot analysis. *Results:* The sensitivity and specificity of this method were 92.6%, 82.6% respectively. The positive and negative predict value were 86.2%, 90.47% respectively. *Conclusions:* Non invasive prenatal diagnosis is a sensitive and specific method for determination of paternally inherited mutation in fetus at risk of thalassemia major. *Key words:* Non Invasive Prenatal Diagnosis, High Resolution Real Time PCR, IVS II-I, B Thalassemia.

NATIONAL PROGRAMME FOR BLOOD GENETIC DISORDERS – REVISITED

Mohsen A.F. El-Hazmi

Ash Shura Council, Riyadh, Saudi Arabia

The first indication for the presence of Blood Genetic Disorders in Saudi Arabia was the finding of Sickle Cell gene in an applicant for employment to Aramco, the Saudi Oil Company in the Eastern Province. Consequently, several studies, including a National Screening Programme of various regions of the Kingdom were carried out. As a result, the presence of Sickle Cell and the Thalassaemia genes were documented in all regions, but at a variable frequency. Therefore, Community and official health care Committee Members worked toward a comprehensive prevention and care programmes, where clinics and specialised centres were established. The Charity societies played a central role in these efforts, including awareness, counseling and community acceptance of pre-marital prgramme. Currently, there exists six societies located in the major regions. Several regions of the vast country lack the availability of the professional and hence the services. To ensure more appropriate service coverage of various regions of the country, this paper present a proposal for extending the service umbrella of the charity societies and the National programme to all regions of the country. The proposal was presented and discussed in the societies meetings. It is hoped that excutive plan will be drawn for the implementation of the programme in due course.

PRENATAL GENETIC IMPLANTATION AS A POSSIBLE ALTERNATIVE TO ELECTIVE ABORTION: THE LEBANESE EXPERIENCE

Rayan Bou Fakhredin[1], Hassan M. Moukhadder[1,2], Joseph Roumi[1,2], Rita Aoun[2], Therese Abou Nasr[2], Ali T. Taher[1,2]

[1]American University of Beirut Medical Center, Department of Internal Medicine, Beirut, Lebanon; [2]Chronic Care Center, Hazmieh, Lebanon

Background: Preimplantation genetic diagnosis (PGD) has recently been employed as a means of identifying embryos obtained by *in vitro* fertilization (IVF) which are afflicted with autosomal recessive disorders. In a country like Lebanon where elective abortion remains to be illegal, PGD has established itself as an ethically acceptable alternative to such prenatal diagnostic procedures as chorionic villus sampling (CVS) and amniocentesis for those who advocate against termination of pregnancy. The aim of this report is to highlight the increasing role of PGD in detecting cases of β-thalassemia born to couples seeking prenatal genetic counseling at the Chronic Care Center, a hemoglobinopathy center in Lebanon. *Methods:* Retrospective data was collected from the geneticist at our center, which included all PGD procedures performed during the period extending from 2011 till 2015 for autosomal recessive heritable diseases. Our primary endpoint was tracking the cases of β-thalassemia detected by PGD during the aforementioned timeframe. *Results:* A total of 15 PGD procedures were performed from 2011 to 2015, 5 of which were targeted at preimplantation detection of β-thalassemia: 1 case was detected in 2013, 2 cases in 2014, and 2 cases in 2015. *Conclusions:* Compared to other approaches for prenatal diagnosis of β-thalassemia, PGD is a convenient tool that is able to identify the disease prior to gestation. This serves to reduce the rate of elective abortion and the moral dilemma associated with it. Endorsement of PGD by governmental agencies as a legal obligation for at-risk couples can serve as a preventive measure to elective abortion in a country where the latter is still illegal. However, this remains to be limited by both financial and religious obstacles.

IMPACT OF PRENATAL DIAGNOSIS OF THALASSAEMIA ON DISEASE PREVENTION: A SINGLE-CENTER EXPERIENCE FROM LEBANON

Michele Abi Saad[1], Hassan M. Moukhadder[1,2], Rita Aoun[1], Joseph Roumi[1,2], Rayan Bou Fakhredin[2], Sanaa Aoun[1], Ali T. Taher[1,2]

[1]Chronic Care Center, Hazmieh, Lebanon; [2]American University of Beirut Medical Center, Department of Internal Medicine, Beirut, Lebanon

Background: The Chronic Care Center is the only center in Lebanon specialized in the treatment and prevention of thalassemia. In 1994, the center launched a national thalassemia prevention program, and in 1999, prenatal diagnosis for thalassemia was introduced at the Genetics Laboratory of the center. The aim of this report is to highlight the impact of prenatal diagnosis on decreasing the incidence of thalassemia in Lebanon. *Methods:* Data on the yearly count of prenatal diagnostic procedures performed from 1999 to 2015 was obtained from the Genetics Laboratory at the Chronic Care Center. Registered data also included the prevalence of carrier, normal, and affected cases in each year. *Results:* From 1999 till 2015, 262 prenatal diagnostic procedures targeted at the detection of thalassemia were performed. 67 fetuses were diagnosed with thalassemia, corresponding to 25.57% and therefore echoing the mode of inheritance of the disease as an autosomal recessive disorder. On another level, 14 cases of thalassemia could be prevented yearly from 2000 till 2012 (20 being the expected number of cases per year in accordance with 8-year data before 1994, and 6 being the actual average annual incidence), which denotes a 70% reduction rate. 46 thalassemia cases were detected during the aforementioned period, translating into an average number of 3.5 cases per year (17.5% of the prevented cases). In fact, the percentage of cases prevented due to prenatal diagnosis was 28% in the last 5 years during the same time period, *i.e.*, from 2008 till 2012, reflecting an annual decrease of 5.6 cases by prenatal diagnosis. *Conclusions:* Our analysis clearly demonstrates that the demand for prenatal diagnosis for the detection of thalassemia before birth is on the rise in Lebanon and that prenatal diagnosis has been playing an important role in decreasing the incidence of the disease in the country.

INSIGHT INTO THE INCIDENCE AND INHERITANCE PATTERN OF β-THALASSAEMIA IN LEBANON

Joseph Roumi[1,2], Hassan M. Moukhadder[1,2], Rayan Bou Fakhredin[1], Rita Aoun[2], Ali T. Taher[1,2]

[1]American University of Beirut Medical Center, Department of Internal Medicine, Beirut, Lebanon; [2]Chronic Care Center, Hazmieh, Lebanon

Background: Thalassemia is prevalent in Lebanon, with a carrier rate of 2.3% in the general population and 4-41% in high-risk groups. The incidence of β-thalassemia has dropped due to the implementation of prenatal screening and diagnostic tools. It is far well known that the inheritance of β-thalassemia follows simple Mendelian genetics with an autosomal recessive mode of inheritance. However, some mutations in β-thalassemia behave in a dominant negative fashion whereby the mutated allele can decrease the production of normal β-globin encoded by the normal allele, thereby variably modifying the mode of inheritance of the disease. The aim of this study is to reflect on the incidence of β-thalassemia major over a 6-year period (2010-2015) in Lebanon. *Methods:* Data on 94 heterozygous couples was retrieved from our center's registry on cases of β-thalassemia detected by prenatal diagnosis (chorionic villus sampling or amniocentesis) from 2010 to 2015. The annual incidence of β-thalassemia was then calculated and compared to the rates expected based on standard Mendelian laws. *Results:* The incidence of β-thalassemia major was found to be 34.3%, which corresponds to 6 cases per year, with a pronounced year-to-year variegation and absence of a well-defined decreasing pattern over the years. 4 cases per year are implied by simple Mendelian genetics (an incidence of 25%). *Conclusions:* The incidence of β-thalassemia did not follow a steady decreasing pattern over the studied 6-year period, which might infer the presence of pitfalls in implementation of and/or compliance with the premarital law in Lebanon, or reflect constant socio-demographic changes in the Lebanese pop-

ulation. In addition, the higher than expected incidence of β-thalassemia in the Lebanese population could be explained by the presence of genetic modifiers which deserve to be further investigated. Our relatively small sample size, however, might be a major limitation to making such a conclusion.

EPIDEMIOLOGY OF β-THALASSAEMIA MAJOR IN THE RABAT REFERENCE CENTER

Mohammed Khattab*, Zineb Isfaoun, Maria El Kababri, Amina Kili, Mohammed El Khorassani, Laila Hessissen

University Children's Hospital of Rabat, Morocco

Background: Since the thalassemia national program was launched by the Ministry of health in 2011, Blood transfusion and iron chelation are offered for free of charge to all patients without health insurance (85%). Rabat center is the main reference center for hemoglobin disorders where patients have been followed for 25 years. The aim of this study is to evaluate our experience. *Methods:* All patients with major thalassemia followed regularly in the Rabat center were examined and their medical records reviewed and updated during 2015. *Results:* With a median follow-up of 15 years (1-25), 105 patients with major thalassemia are currently treated in our center. Sex-ratio was 1.1 (56 males and 49 females). The median age was 12 years (9m-31y). The consanguinity was very high at 50%. The mean age of onset of transfusion was 2y (1m-16y), and of iron chelation started at the mean age of 6y (1-20). Facial abnormalities were observed in only 15%. Splenomegaly and hepatomegaly were present in 63% and 18% respectively. Splenectomy was done in 19%. The median pré-transfusionnel Hb was 9.8g/dl (6.6-11.3). The median serum ferritin was 5106ng/ml (225-22.300). Alloimmunization and autoimmunisation were observed in 8% and 5% respectively. Short stature was observed in 50%, puberty delay in 21% and diabetes mellitus in 6%. *Conclusions:* The Moroccan thalassemia program had a positive impact on the main aspects of outcome this disease. We are working on implementation an efficient monitoring tests for evaluation of iron chelation and recruitment of specialists taking in charge complications of major thalassemia.

FERTILITY AND PREGNANCY

EVALUATION OF MARRIAGE AND CHILDBIRTH IN PATIENTS WITH NON-TRANSFUSION DEPENDENT β-THALASSAEMIA MAJOR AT THALASSEMIA RESEARCH CENTER OF SARI, IRAN

Mandana Zafari[1], Mehrnoush Kowsariyan[2*]

[1]*Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran;* [2]*Professor of Pediatrics, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran*

Background and Purpose: Patients with non-transfusion-dependent beta thalassemia major (NTDTM) could reach old age, marry and have children with appropriate care. This study aimed to review the marital status and maternal-fetal outcomes of NTDTM patients at Thalassemia Research Center (TRC) of Sari, Iran. *Methods:* In this study, medical records of patients with β-thalassemia major were reviewed from July 2014 to December 2014. All the patients were interviewed, and a questionnaire was designated by research methodology experts. Reliability of the questionnaires was measured through a pilot study on 12 patients using the test-retest method (r=0.9). In addition, epidemiological characteristics and pregnancy outcomes of the patients were recorded. Data analysis was performed using descriptive statistics in SPSS. *Results:* In total, 419 records were reviewed, and 74 cases (17.6%) had NTDTM. During a 25-year marriage period, 23 pregnancies were reported with 18 childbirths. Low birth weight was observed in three neonates (23.1%), and there was one assisted pregnancy. In addition, one female NTDTM patient was married to a β-thalassemia carrier and had two abortions (one after prenatal diagnosis). In this study, 24 (32.4%) and 14 (58.3%) male NTDTM patients were married, and only one case had a child. Mean age of marriage in male NTDTM patients was 25.3±4.2 years. *Conclusions:* According to the results of this study, proper management of NTDTM patients will help them reach the reproductive age. It also seems that fertility is higher among female NTDTM patients. *Keywords:* Childbirth, Fertility, Infertility, Intermedia β-thalassemia, Marriage, Non-transfusion-dependent thalassemia major, Pregnancy.

GENE REGULATION AND THERAPY

β-GLOBIN GENE CLUSTER HAPLOTYPES OF HB D-LOS ANGELES IN MAZANDARAN PROVINCE, IRAN

Mohammad Reza Mahdavi[1], Hossein Jalali[1], Mehrnoush Kosaryan[1], Payam Roshan[2], Mehrad Mahdavi[2*]

[1]*Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran;* [2]*Sina Mehr Research Center, Sari, Iran*

Background and Purpose: Several types of hemoglobin D (Hb D) are distinguishable by DNA analysis, and the aim of this study was to identify the types of Hb D variant and β-globin gene haplotypes linked to Hb D in Mazandaran Province, northern Iran. *Methods:* Fifty five individuals were identified as Hb D carriers, and PCR-RFLP analysis revealed that all 55 had the Hb D-Los Angeles type. To identify haplotypes associated with the βD allele, family linkage analysis, using the PCR-RFLP method for seven polymorphisms in the β-globin gene cluster, was carried out on families of 23 of these 55 individuals. *Results:* We observed three different haplotypes in association with Hb D-Los Angeles. In most cases (91.4%) βD alleles were linked to haplotype I [+ – – – – + +]. Haplotype II [– + + – + + +] and an atypical haplotype [– + + – – + –] were each in association with the βD allele in only one case (4.3%). This is the first report worldwide of the [– + + – – + –] haplotype in association with Hb D-Los Angeles. *Conclusions:* We conclude that more than 90% of the evaluated Hb D-Los Angeles alleles in Mazandaran has the same origin, and the two rare haplotypes may represent different genetic origins and/or other molecular events, such as gene conversion or recombination, in the region. *Keywords:* β-Globin, Hb D-Los Angeles, Mazandaran, Iran.

β-GLOBIN GENE HAPLOTYPES ASSOCIATED WITH HAEMOGLOBIN D-PUNJAB IN NORTHERN IRAN

Hossein Jalali[1], Mohammad Reza Mahdavi[2], Mehrnohsh Kosaryan[3*], Hossein Karami[4], Payam Roshan[5], Fatemeh Maddahian[6]

[1]Research Student in Thalassemia, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [2]Department of Laboratory Medicine, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [3]Department of Pediatrics, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [4]Hematology and Oncology, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [5]Immunology, Sina Mehr Research Center, Sari, Iran; [6]Genetic, Sina Mehr Research Center, Sari, Iran

Background and Purpose: Hemoglobin D-Punjab is one of the variant of hemoglobin caused by a mutation on position 121 of beta globin gene which is frequent in India, Pakistan and Iran. Heterozygote form of this variant is mainly asymptomatic while in combination with hemoglobin S, severe form of anemia occurs. This study was carried out to determine the beta globin gene haplotypes associated with hemoglobin D-Punjab in Northern Iran. *Methods:* This descriptive study was carried out on families of 18 individuals whom were carriers of hemoglobin D-Punjab in Sari in Northern Iran. Genomic DNA was extracted from peripheral blood samples using Phenol-chloroform standard protocol. In order to identify different haplotypes associated with hemoglobin D-Punjab, PCR-RFLP method and family linkage analysis were used. *Results:* In 17 subjects hemoglobin D-Punjab was linked to [+ - - - - + +] haplotype and in one case association with [- + + - + + +] haplotype was observed. *Conclusions:* The hemoglobin D-Punjab alleles have mainly unicentric origin and [- + + - + + +] rare haplotype may have different genetic origin or is created as a result of gene recombination. *Keywords:* Hemoglobin D-Punjab, Haplotype, PCR-RFLP, Iran.

HEART AND VASCULAR ABNORMALITIES

PREVALENCE OF PULMONARY HYPERTENSION IN PATIENTS WITH THALASSAEMIA INTERMEDIA; A SINGLE CENTER EXPERIENCE

Zahra Badiei*, Hassan Mottaghi Moghaddam, Hamid Farhangi, Reza Shakeri

Pediatric Hematology & Oncology, Faculty of Medicine, Mashhad University of Medical Sciences, Mashhad, Iran

Background: One of the most significant complications in patients with thalassaemia intermedia is pulmonary arterial hypertension (PAH) syndrome. PAH is more common in TI than major form, and it may cause cardiac complications in patients who are older than 30y. The aim of this study was to estimate the prevalence of PAH in TI patients so that they can be referred expeditiously for treatment, thereby preventing the complications that occur later. *Methods:* Forty-one thalassaemia intermedia patients were examined at the Hemophilia-Thalassaemia center of Mashhad (Sarvar Clinic). Electrocardiography, chest-X Ray, and echocardiography tests were performed for all of the patients by the same pediatric cardiologist. The data were processed by SPSS software, version 11.5, and the results were analyzed using chi-squared, Student's t, and Mann-Whitney tests. *Results:* The mean age of the patients was 21.93±8.34 y. They had been under pediatric heart specialists' constant examination and treatment since their childhood when they were diagnosed with TI, and continue to receive regular follow-up care. The prevalence of pulmonary hypertension was 24% in our study population. In patients with thalassaemia intermedia, the left ventricular (LV) mass indices were about 3-5 times higher than would be expected in a normal population. Patients with higher LV mass indices have a greater risk of developing pulmonary hypertension, and those with serum ferritin levels below 1000 ng/ml are less susceptible to diastolic dysfunction. *Conclusions:* Pulmonary hypertension is common in patients with thalassaemia intermedia. Irregular chelation therapy or absence of this treatment might lead to diastolic dysfunction, and serum ferritin levels below 1000 ng/ml could be an important factor in preventing the development of diastolic dysfunction or slowing down its progression. *Keywords:* Thalassaemia intermedia, pulmonary hypertension, echocardiography, ferritin.

KIDNEY INJURY MOLECULE-1 AND HEART-TYPE FATTY ACID BINDING PROTEIN AS NOVEL EARLY MARKERS OF PROXIMAL AND DISTAL RENAL TUBULAR DYSFUNCTION IN CHILDREN WITH β-THALASSAEMIA MAJOR

Laila M. Sherief[1],Osama R. El-Safy[1], Amal Zidan[2], Doaa M. Youssef[1], Manar El-Sayed

[1]Departments of Pediatrics; [2]Clinical Pathology, Faculty of Medicine, Zagazig University, Egypt

Background: Survival of the patients with B-thalassemia has improved recently. This success has allowed previously unrecognized complication to emerge, including renal abnormalities. *Aim:* Was to investigate urinary kidney injury molecule-1(KIM-1) and urinary heart type fatty acid binding protein (H-FABP) as novel early markers of renal tubular dysfunction in children with β-thalassemia major. *Patients and Methods:* This cross-sectional study, case control study included a total of 124 children; as sixty-two children with beta thalassemia major (B-TM) aged 5-18 years, on regular blood transfusion regimen represented the patient group and 62 healthy children, with comparable age and gender, were assigned as control group. All participants were subjected to history taking, thorough clinical examination and laboratory investigations including; complete blood count, liver and kidney function tests, serum ferritin, Hepatitis markers including HBs Ag and HCV antibody and tubular markers, urinary (KIM-1), urinary (H-FABP) determined by ELISA immunoassay. *Results:* Our B-TM patients have been transfusion-dependent for as long as 9.51±2.96 years with significantly higher serum ferritin levels, liver enzymes and bilirubin when compared to controls. On the contrary, hemoglobin and glomerular filtration rate (eGFR), were significantly lowered in patients *versus* normal peers. Urinary KIM-1 and H-FABP were significantly increased for patients when compared with healthy controls. Of particular interest is the positive correlation between urinary kIM-1 and H-FABP and, age, duration of transfusion, blood urea, serum creatinine, and serum ferritin levels. *Conclusions:* We concluded that renal dysfunction is not rare in children with beta thalassemia major and that renal tubular dysfunction may not be detected by routine tests so the use of early mark-

ers KIM -1 and H-FABP is recommended in children with beta thalassemia.

THE RELATIONSHIP BETWEEN SERUM VASCULAR ENDOTHELIAL GROWTH FACTOR AND β-THALASSAEMIA MAJOR

Farzane Farokhi[1*], Javad Razaviyan[2,3], Mehrnoush Kosaryan[4], Mostafa Roudbari[5], Samira Esmaeili Reykande[3,6], Aily Aliasghariyan[7], Maryam Dehghani[8]

[1]Department of Biology, Azad University of Sari Branch, Mazandaran, Iran; [2]Clinical Biochemistry, Medical School, Tehran University of Medical Sciences, Tehran, Iran; [3]Scientific Research Center, School of Allied Medicine, Tehran University of Medical Sciences, Tehran, Iran; [4]Pediatrics, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [5]Department of Laboratory Medicine, Faculty of Allied Medicine, Mazandaran University of Medical Sciences, Mazandaran, Iran; [6]Department of Hematology, Allied Medical School, Tehran University of Medical Sciences, Tehran, Iran; [7]Medical Microbiology, Thalassemia Research center, Hemoglobinopathy Institute, Mazandaran University of medical sciences, Sari, Iran; [8]Department of Epidemiology and Biostatistics, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran

Background and Purpose: Beta thalassemia is an inherited disorder characterized by absent or reduced amounts of beta globin chains. Vascular Endothelial Growth Factor (VEGF) is a significant regulator of hemangioblast differentiation. This study was aimed to assess serum VEGF levels in patients with beta thalassemia major in comparison with control group. *Methods:* This historical cohort study was conducted on 36 patients with β-thalassemia major who had received regular blood transfusion and 26 healthy people which were referred for checkup in a general hospital, Sari, north of Iran, during March to May 2015. Demographic characterization and laboratory tests such as Complete Blood Count (CBC), and evaluation of levels of serum ferritin, serum VEGF, hepatitis B virus antibody and hepatitis C virus antibody were carried out for our patients. The statistical analyses were performed by SPSS (16) software. The Pearson correlation coefficient test was used to test the significant correlations for quantitative parameters. A value of $P<0.05$ was considered statistically significant. *Results:* Mean serum VEGF level in case and control groups was 153.8±77.5 and 120.2±45.4 pg/ml, respectively. Serum VEGF level was higher in beta thalassemia major ($p=0.037$). Serum VEGF level was significantly higher in splenectomized patients ($P=0.006$). There was not any significant correlation between serum VEGF levels and Hemoglobin, WBC and platelet count and neither was with serum ferritin level ($p>0.05$). *Conclusions:* Serum VEGF level was higher in thalassemic patients. Splenectomized patients had higher serum VEGF levels than others. *Keywords:* Thalassemia major, Vascular Endothelial Growth Factor, Angiogenesis, Splenectomy.

DIGITAL THERMOGRAPHY AND VASCULAR INVOLVEMENT IN THALASSAEMIA INTERMEDIA

Marwan Refaat[1], Patrick Zakka[2], Mostafa Hotait[3], Samir Arnaout[1], Hussein Isma'eel[1], Ali Taher[4]

[1]American University of Beirut Faculty of Medicine, Division of Cardiology; [2]American University of Beirut Faculty of Medicine, Department of Internal Medicine; [3]American University of Beirut Faculty of Medicine, Division of Neurology; [4]American University of Beirut Faculty of Medicine, Division of Hematology/Oncology

Background: Physical properties of thalassemic cells as being deformed, rigid and aggregable, and increased levels of endothelial adhesion proteins in these patients all contribute to vascular complications. Digital thermal monitoring (DTM) is a novel non-invasive tool that evaluates vascular function based on changes in fingertip temperature during and after cuff occlusion. In this study, we applied this test on Thalassemia Intermedia (TI) patients. *Methods:* 15 patients with TI were recruited from a thalassemia chronic care facility. Exclusion criteria included any factors that are known to be associated with vascular damage. Patients underwent DTM and results were extracted as vascular reactivity index (VRI), a measure of how well the circulatory system responds to physiologic and pharmacologic stimuli that require adjustments of blood flow. The DTM machine used defines 3 categories: poor (VRI <1), intermediate (1<VRI<2) and good (VRI>2). *Results:* 15 patients were recruited: 9 females (age 32.44±11.26) and 6 males (age 30.67±8.43). The average VRI in males was 2.05±0.41 and in females 2.32±0.45, but the difference was not significant ($p=0.250$). The patient's age ($R^2=0.017$, $p=0.458$) and BMI ($R^2=0.003$, $p=0.460$) showed poor correlation with vascular reactivity. Ferritin levels were not shown to be correlated with VRI ($R^2=0.011$, $p=0.478$). 33.3% (5/15) of patients VRI's fell in the intermediate zone of vascular reactivity. *Conclusions:* A significant proportion of patients had vascular reactivity indices in the intermediate zone, suggesting that thalassemia intermedia in itself may be associated with a decrease in VRI. This sample should be compared to a control group to look for any significant differences in VRI. If any differences are found, early detection of VRI changes in these patients may help in earlier intervention to prevent or delay incidence of vascular disease-related symptoms.

HEPATOLOGICAL COMPLICATIONS

DECREASE OF HEPATITIS C BURDEN IN PATIENTS WITH TRANSFUSION DEPENDENT β-THALASSAEMIA MAJOR, THALASSEMIA RESEARCH CENTER, 1995-2014

Mehrnoush Kosaryan, Aily Aliasgharian*

Professor of Pediatrics, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran

Background and Purpose: Chronic hepatitis C infection used to be one of the most important burdens on patients with transfusion-dependent beta thalassemia major (TDTM). Chronic active hepatitis reduces quality of life, and liver cirrhosis and cancer shorten life expectancy in many cases. We compared the characteristics of our patients at the Thalassemia Research Center (TRC) regarding hepatitis C infection at two time points. *Methods:* A review was conducted in a cohort of 390 TDTM patients with a history of at least one blood transfusion in 2014. Type of treatment protocol for hepatitis C virus (HCV) and the number of courses were defined. Descriptive statistics were performed using SPSS software (V16). *Results:* Screening for HCV started in 1995 at the TRC. Seventy-seven (15%) patients were antibody-positive in 1995. Tests for virus detection were not available at

the time. Patients have been examined using serum AST, ALT, bilirubin, PT, PTT, and liver biopsy, and 45 were treated using alpha interferon alone. A second liver biopsy was performed at the end of treatment for 21 patients, and a blinded pathologist compared the histology according to the Knodell score. According to normalization of liver enzymes, the treatment was successful (McNemar test, P <0.02). Based on the Knodell score, 54%, 31%, and 11% had complete, partial, and no response, respectively. A quantitative test for viremia became available thereafter. Thirteen patients who were resistant to alpha interferon have been treated using "Pegasys"™±ribavirin. Ten patients responded; however, three have been resistant and are still viremic. Twenty-seven patients received no treatment. Twenty-two (81.4%) had negative PCR tests. Five viremic patients refused treatment. A second screening test for HCV antibody was introduced in 2001, and, since then, annual screening for HCV antibody has been performed for all patients. No new case has been found since 2001. During the follow-up period, two deaths have been recorded in the cohort; none was a direct consequence of liver disease. Both patients had negative PCR tests for viremia. In 2014, there were 72 patients (52% men) with positive antibody tests, with a mean age of 30.5±5.7 years. They mean age at the first blood transfusion was 2.8±2.5 years. At the time of publishing, 15 patients (3.8%; 95% confidence interval 2-5.6) had viremia. Five patients had documented liver cirrhosis. *Conclusions:* The prevalence of hepatitis C virus has decreased dramatically owing to primary prevention (donor blood screening and discarding infected blood) and antiviral treatment of affected patients. Better clinical management with iron chelating agents and supportive therapy for cirrhotic patients is also in place. *Keywords:* Hepatitis C Virus; Treatment; Iran.

IRON LIVER TOXICITY PRESENTING AS HEPATIC NODULE IN MAJOR β-THALASSAEMIA PATIENT: A CASE REPORT

Anna Mira Lubis

Hematology and Medical Oncology Division, Department of Internal Medicine, Faculty of Medicine, Universitas Indonesia

Iron overload is still the biggest issue in major thalassemia patients. Not only viral hepatitis, liver iron overload secondary to long life blood transfusion support lead to Kuppfer iron loading and considerably elevated liver iron concentration promoting liver fibrogenesis. A 20 years old was female diagnosed with major Beta Thalassemia since she was 8 months old. She has been having regular blood transfusion since she was 1 year old. She also has been given deferiprone in combination with deferioxamine as iron chelator agent. Her average ferritin level in the last 2 years was 8000 ng/dL. The recorded iron toxicity found was endocrinopathy manifested as hypothyroidism. She has chronic liver disease with portal hypertension assessed by ultrasound. There was no hepatitis B nor hepatitis C infection and her liver dan cardiac 2015 MRI T2*was 3,98 ms and 18,72 ms respectively. In the last 1 year she had been having complaints of abdominal enlargement. She had recurrent transudate type ascites and umbilical hernia. A repetitive ultrasound evaluation was performed and a hepatic nodule was found. A multiphase abdominal Computed Tomography revealed hepatosplenomegaly, a heterogenous mass (3,3x2,7x2,95 cm) with prominent contras enhancement at vein and delayed phase, in the third segmen of left hepatic lobus and prominent Portal Vein and Lienalis Vein suggesting portal hypertension. A Liver MRI T2* was significantly worsening (1,26 ms) with normal liver function tests and Alfa Fetoprotein (AFP) value. An ultrasound- guided biopsy was performed on her hepatic nodule and the histopathology result revealed chronic hepatitis with iron accumulation grade 4, consistent with Metavir Grade 2-3, stage F2, without malignant cell. She received an intensive iron chelation ever since with close monitoring of liver MRI T2*. This case ilustrates the liver iron toxicity in iron overloaded major thalassemia patient, even under iron chelation. Although the impact in quality of life is devastating, it is a predictable and preventable complication. Recognition and monitoring of iron toxicity are critical to optimize iron chelation therapy to improve survival and quality of life.

HAEMATOPOIETIC STEM CELLS TRANSPLANTATION

A RETROSPECTIVE LONG-TERM STUDY FROM ALLOGENEIC HAEMATOPOIETIC STEM CELLS TRANSPLANTATION IN MAJOR β-THALASSAEMIA: THE ALGERIAN EXPERIENCE

Malek Benakli, Ahmed Nacer Redhouane, Rachida Belhadj, Farih Mehdid, Farida Tensaout, Nadia Rahmoune, Dina Ait Ouali, Mounira Baazizi, Hanane Bouarab, Sara Zerkout, Nadia Boukhenfouf, Rose-Marie Hamladji

Hematology and BMT Department, Pierre and Marie Curie Center, Algiers, Algeria

Introduction: Allogeneic hematopoietic stem cell transplantation (Allo-HSCT) remains the only curative approach for children with major β-thalassemia. HLA-matched related donor allo-HSCT should be performed as early as possible if donor is available. This retrospective study analyzes the outcome of 47 consecutive pediatric patients (pts) with β-thalassemia who underwent Allo-HSCT with HLA-identical donors in a our center. *Materials and Methods:* From June 1999 until May 2015, 47 consecutive pts affected by transfusion-dependent major β-thalassemia underwent allo-HSCT from matched related donors in our institution (Sibling HLA identical: 38, pheno-identical: 6, cord blood: 3). The median age: 7,6 years (3-15) within 28 (59,7%) older than 7 years. Sex-ratio: 1,47. The median red blood cell transfusions was 69,5 units/pt (20-177). Irregular iron chelation in 39 pts (82,9%) and median serum ferritin at transplant was 2238 ng/ml (250-7424). Liver biopsy was performed in all pts and Risk class Pesaro was determined in all pts: 6 (13%) were assigned to risk class 1, 19 (41,3%) to class 2 and 22 (45,7%) to class 3. Nineteen pts (41,3%) had splenectomy. The median time diagnosis-graft was 80,7 months (27-174). The preparative regimen consist on Misulban 500 mg/m^2 per os or Busilvex doses adapted, Ciclophosphamide 200 mg/kg and Thymoglobuline 10mg/kg (Pesaro 1-2); Misulban 14 mg/kg or Busilvex dose adapted, Ciclophosphamide 120 mg/kg and Thymoglobuline 10 mg/kg (Pesaro 3); Busilvex dose adapted, Thiotepa 10mg/kg and Fludarabine 160 mg/m^2 (Cord blood). Prevention of GVHD consisted of the association Ciclosporine and Methotrexate short cycle (Seattle) or Ciclosporine alone (cord

blood). The grafts used are peripheral blood stem cells in 41 pts (84%) with an average rate of CD34+ cell: 10,3x10^6/kg (4,47-37,90), bone marrow in 4 pts with an average rate of Nuclear Cells (NC): 5.12 10^8/kg and cord blood in 3 pts with a rate of NC: 4.8 10^7/kg. At November 2015, the minimal follow-up was 7 months and the maximal 198 months. *Results:* Median time of aplasia, observed in all pts, was 17 days (6-71). Neutrophil engraftment was observed at day 16 (9-68). Transfusions were required in all pts with an average of red blood cells: 5,4 units/pt (2-15) and Platelets concentrates: 5,3 units/pt (0-34). Seven pts (5 with Pesaro score 3) presented an early rejection and received boost without any benefit. Veinous occlusive disease observed in 4 pts (8,7%). Acute GVHD grade II-IV was seen in 13 cases (28%) and extensive chronic GVHD in 4 pts (8,7%). Thirty-five pts (74,4%) are alive with a median follow up of 71 months (7-185) within 29 pts (68%) with total donor chimerism. Twelve pts (25,5%) died (graft rejection: 4, earlier infection: 2, VOD: 1, GVH: 4, hydrocephalus: 1). Overall survival (OS) and event-free survival (EFS) at 16 years are 75,7% and 66,8% respectively. According to Pesaro score, OS and EFS are 76,7% and 70,5% (Pesaro 1-2) and 74,6% and 62,8% (Pesaro 3). *Conclusions:* In our series, most of pts have higher Pesaro score and are older than 7 years. Allo-HSCT should be performed early to have a best results.

IRON OVERLOAD AND MANAGEMENT

SAFETY OF DEFERASIROX IN β-THALASSAEMIA PATIENTS WITH SERUM FERRITIN LESS THAN 1000

Mehran Karimi[1*], Zahra Barati Shourijeh[2], Sezaneh Haghpanah[1], Zohre Zahedi[1]

[1]Hematology Research Center, Shiraz University of Medical Sciences, Shiraz, Iran; [2]Department of Biochemistry, Islamic Azad University, Fars Science and Research Branch, Shiraz, Iran

Background: In patients suffering from anemia with blood transfusion, organ dysfunction caused by iron overload is the most important cause of mortality in both pediatrics and adolescent. The aim of this study was to survey the safety and efficacy of deferosirox (Exjade) in patients with serum ferritin level below 1000 ng/ml. *Study design and Methods:* This was a historical-cohort study of 53 beta thalassemia major and intermedia patients who had lower serum ferritin level and have been on deferosirox 10-38 mg/Kg/day for up to 12 months. Serum ferritin level was categorized into three groups: 300ng/ml, 300-500 ng/ml and 500-1000 ng/ml. It was checked at 3, 6 and 12 months after treatment. Any adverse effect was evaluated every 3 months. *Results:* Fifty-three patients participated in this study. A reduction in serum ferritin levels was observed overall, however, it was significant only in patients whose baseline ferritin was 500-1000 mg/dl. Some mild drug dependent adverse effects were seen during the study which were not serious enough to be the cause of discontinuing therapy. *Conclusions:* This study confirmed that deferosirox (Exjade) is an effective drug with no significant adverse effects in both beta thalassemia major and intermediate patients even when serum ferritin is less than 1000 ng/ml. *Key words:* Deferosirox, Ferritin, beta thalassemia, adverse effects.

EFFICACY OF ORAL DEFERASIROX BY TWICE-DAILY DOSING IN PATIENTS WITH TRANSFUSION-DEPENDENT β-THALASSAEMIA

Ebrahim Salehifar[1], Hosein Karami[2], Mehrnoosh Kosaryan[3], Hosein Masoudi[4], Aily Aliasgharian[5], Masoumeh Mousavi[6], Razieh Avan[7]

[1]Department of Clinical Pharmacy, Thalassemia Research Center, Hemoglobinopathy Institute, Faculty of Pharmacy, Mazandaran University of Medical Sciences, Sari, Iran; [2]Department of Pediatrics, Thalassemia Research Center, Hemoglobinopathy Institute, Faculty of Medicine, Mazandaran University of Medical Sciences, Sari, Iran; [3]Department of Pediatrics, Thalassemia Research Center, Hemoglobinopathy Institute, Faculty of Medicine, Mazandaran University of Medical Sciences, Sari, Iran;[4]Student Research Committee, Faculty of Pharmacy, Mazandaran University of Medical Sciences, Sari, Iran; [5]Medical Microbiology, Thalassemia Research Center, Hemoglobinopathy Institute, Faculty of Medicine, Mazandaran University of Medical Sciences, Sari, Iran; [6]Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [7]Resident in Clinical Pharmacy, Student Research Committee, Faculty of Pharmacy, Mazandaran University of Medical Sciences, Sari, Iran

Background and Purpose: Patients with Beta thalassemia major need consistent blood transfusion from early years of life. Deferasirox is used as an oral chelating agent (once daily) to excrete excess iron. This study aimed to compare the efficacy of deferasirox twice daily and the usual once daily dosing. *Materials and Methods:* This before after clinical trial was performed in 2013-2014 in patients who were at least 2 years of age and received only deferasirox as the chelating agent. All patients had received deferasirox for at least six month once daily. The last ferritin before entering the study and the mean deferasirox daily dose during the previous six months were considered as baseline ferritin and deferasirox dose, respectively. Laboratory tests were performed including CBC-diff, serum ferritin, Creatinine (Cr), Aspartate Aminotransferase (AST) and Alanine Aminotransferase (ALT) to evaluate the efficacy and safety of deferasirox. *Results:* A total of 21 transfusion-dependent patients (mean age: 21±6 years old) were included of whom 67% were male. The mean ferritin level decreased significantly from 1814±922 ng/ml to 1472±907 ng/ml (P=0.02). There were no any significant changes in AST, ALT and Cr levels compared to baseline values. *Conclusions:* Twice daily dosing of deferasirox was associated with more decrease in ferritin level compared to baseline single daily dose values without any hepatic or renal adverse effects. *Keywords:* Beta thalassemia major, deferasirox, ferritin, twice-daily, efficacy.

IN VIVO IRON-CHELATING ACTIVITY AND PHENOLIC PROFILES OF THE ANGEL'S WINGS MUSHROOM, *PLEUROTUS PORRIGENS* (HIGHER *BASIDIOMYCETES*)

Masoumeh Khalili[1], Mohammad Ali Ebrahimzadeh[2], Mehrnoush Kosaryan[3]

[1]Pharmaceutical Sciences Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; Student Research Committee, School of Pharmacy, Mazandaran University of Medical Sciences, Sari, Iran; [2]Pharmaceutical Sciences Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [3]Professor of Pediatrics, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran

Background and Purpose: Pleurotus porrigens is a culinary-medicinal mushroom. It is locally called sadafi and is found in the northern regions of Iran, especially in Mazandaran. This mushroom is used to prepare a variety of local and specialty foods. Because of the phenol and flavonoid contents and the strong iron-chelating activity of this mushroom, it was selected for an assay of *in vivo* iron-chelating activity. *Methods:* Methanolic extract was administered intraperitoneally to iron-overloaded mice at two dosages (200 and 400 mg/kg/24 hours) for a total of 20 days, with a frequency of 5 times a week for 4 successive weeks. The total iron content was determined by atomic absorption spectroscopy. Plasma Fe^{3+} content was determined using a kit. Liver sections were stained by hematoxylin and eosin and Perls stain. *Results:* A significant decrease in the plasma concentration of iron was observed in mice treated with extracts (P <0.001). The animals showed a dramatic decrease in plasma Fe^{3+} content when compared with the control group (P <0.001). Also, Perls stain improved the smaller amount of deposited iron in the liver of iron-overloaded mice treated with the extract. Liver sections revealed a marked reduction in the extent of necrotic hepatocytes, fibrous tissues, and pseudo-lobules. A high-performance liquid chromatography method was developed to simultaneously separate 7 phenolic acids in extract. Rutin (1.784±0.052 mg g^{-1} of extract) and p-coumaric acid (1.026±0.043 mg g^{-1} of extract) were detected as the main flavonoid and phenolic acids in extract, respectively. *Conclusions:* The extract exhibited satisfactory potency to chelate excessive iron in mice, potentially offering new natural alternatives to treat patients with iron overload. More studies are needed to determine which compounds are responsible for these biological activities. *Keywords:* Medicinal mushrooms, Hematoxylin, Eosin, Iron-Chelating, P-Coumaric Acid, Pleurotus, Porrigens, Rutin.

IRON CHELATION AND LIVER DISEASE HEALING ACTIVITY OF EDIBLE MUSHROOM (*CANTHARELLUS CIBARIUS*), *IN VITRO* AND *IN VIVO* ASSAYS

Masoumeh Khalili[1,2], Mohammad Ali Ebrahimzadeh[1*], Mehrnoush Kosaryan[3], Ali Abbasid[4], Mohammad Azadbakhta[5]

[1]Department of Medicinal Chemistry, Pharmaceutical Sciences Research Center, School of Pharmacy, Mazandaran University of Medical Sciences, Sari, Iran; [2]Student Research Committee, School of Pharmacy, Mazandaran University of Medical Sciences, Sari, Iran; [3]Thalassemia Research Centre, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [4]Legal Medicine Research Center, Legal Medicine Organization, Tehran, Iran

Abstract: Cantharellus cibarius, an edible mushroom, has been reported to display a wide variety of biological properties, including iron-chelation activity. In the current study we evaluated the chelating capacity of ethyl acetate and methanolic extracts of *Cantharellus cibarius* on iron-overloaded mice. Extracts and defroxamine were injected for a frequency of 5 times a week for 4 weeks. Total iron and Fe^{3+} content of plasma was determined by atomic absorption spectroscopy and kit respectively. Liver sections were stained by haematoxylin and eosin and Perls' stain. Iron-overloaded animals treated with the extract, showed a dramatic decrease in plasma iron content when compared with the control group. The highest activity was observed in the methanolic extract. High-performance liquid chromatography was performed to simultaneously separate 5 phenolic acids and 2 flavonoids in extracts. p-Coumaric acid and ferulic acid were discovered to be major phenolic acids in ethyl acetate extract and methanol extract, respectively. Both methanolic and ethyl acetate derived extracts of mushroom *Cantharellus cibarius* exhibit satisfactory potency to chelate excessive iron in mice. *Keywords:* Iron chelation, liver disease, mushroom, *Cantharellus cibarius*, *in vitro*, *in vivo*.

EFFICACY OF THREE IRON-CHELATING AGENTS IN THALASSAEMIA PATIENTS FROM OUR PRACTICE

Joseph Roumi[1,2], Rayan Bou Fakhredin[1], Hassan M. Moukhadder[1,2], Hassan Beydoun[1], Therese Abou Nasr[2], Suzanne Koussa[2], Ali T. Taher[1,2]

[1]American University of Beirut Medical Center, Department of Internal Medicine, Beirut, Lebanon; [2]Chronic Care Center, Hazmieh, Lebanon

Background: Iron chelation therapy (ICT) remains one of the pillars in the treatment of thalassemia. Regular blood transfusions in transfusion-dependent patients and increased iron absorption in the gut eventually lead to iron deposition in multiple organs, including the liver and the heart. The aim of this study is to determine the efficacy of three iron-chelating drugs – deferasirox, deferoxamine, and deferiprone – in terms of serum ferritin level, liver iron concentration (LIC), and cardiac T2*. *Methods:* Using SPSS, we calculated the mean serum ferritin level, mean LIC, and mean cardiac T2* in patients taking deferasirox, deferoxamine, or deferiprone. We then compared the aforementioned means across the three drug categories for any statistically significant differences. *Results:* The mean serum ferritin level in patients chelated with deferasirox, deferoxamine, and deferiprone, was 1,518.1860, 4,326.1818, and 1,490.9375 ng/mL, respectively. The mean LIC in the three patient populations was 5.4026, 7.6182, and 8.2175 mg/g dry weight, respectively, and the mean cardiac T2* was 28.8047, 24.8000, and 27.3688 ms, respectively. The superiority of deferasirox in lowering LIC was statistically significant (p=0.023). However, the supremacy of deferiprone in chelating cardiac tissue, reflected by the highest mean cardiac T2*, was not statistically significant (p=0.6). *Conclusions:* Deferasirox is the iron-chelating drug of choice for reducing liver iron overload in patients with thalassemia. This finding is commensurate with data previously published in the literature, which also advocates the advantage of deferasirox over other iron chelators in ameliorating the burden of iron overload in the liver.

URINARY IRON EXAMINATION TO EVALUATE IRON OVERLOAD IN CHILDREN WITH THALASSAEMIA MAJOR

Pustika Amalia Wahidiyat[1], Ellen Wijaya[2]

[1]Pediatric Hematology-Oncology Division, Department of Child Health, Faculty of Medicine, Universitas Indonesia, Dr. Cipto Mangunkusumo Hospital, Jakarta, Indonesia; [2]Department of Child Health, Faculty of Medicine Universitas Indonesia, Dr. Cipto Mangunkusumo Hospital, Jakarta, Indonesia

Background: Iron overload evaluation is particularly important in thalassemia major (TM) patients. The current gold standard

examination is MRI T2*; however, it is only available in one centre in Jakarta. Serum ferritin (SF) is widely available but may not reflect the true amount of free iron. Urinary iron examination shows a promising prospect as an efficient alternative measure of iron overload. *Objectives:* To evaluate the appropriateness of urinary iron examination as an indicator of iron overload. *Methods:* A preliminary cohort study was conducted at the Thalassemia Centre of Dr. Cipto Mangunkusumo Hospital, Jakarta, Indonesia, with TM patients aged 7-17 receiving deferiprone therapy as the subjects. A 24-hour urinary iron examination was serially performed using inductively coupled plasma – mass spectrometry (ICP-MS) on the day prior to transfusion, the day of transfusion, the 1st and 7th day following transfusion, and the day prior to the next transfusion. These were then correlated to the time of transfusion, transferrin saturation (TS), SF, and MRI T2* results. *Results:* Among the 20 subjects, 15 showed higher urinary iron excretion prior to transfusion (mean: 16,142.0 ng/mL, SD: 14,312.2) compared to that following transfusion (mean: 12,116.6 ng/mL, SD: 6,957.0). An increase was also noted prior to the next transfusion (mean: 15,560.9 ng/mL, SD: 11,599.0). Urinary iron was found to significantly correlate to TS (r=0.55, p=0.012), but not to SF (r=0.145, p=0.542), heart MRI T2* (r=0.228, p=0.333), or liver MRI T2* (r=-0.38, p=0.875). The heart MRI T2* showed normal iron stores on all subjects (100%), while liver MRI T2* showed mild, moderate, and severe iron overload in 10 (50%), 5 (25%), and 5 (25%) subjects respectively. *Conclusions:* Urinary iron is an appropriate measure to evaluate iron overload. Increasing the dose of deferiprone or using a combination therapy may resolve liver iron overload. *Keywords:* Urinary iron examination, thalassemia, iron overload.

COMBINATION OF DEFEROXAMINE/DEFERIPRONE IN IMPROVING CARDIAC, LIVER AND PANCREATIC T2* IN TWINS WITH β-THALASSAEMIA MAJOR AND SEVERE IRON OVERLOAD: A CASE REPORT

Ludi Dhyani Rahmartani, Pustika Amalia Wahidiyat, Teny Tjitra Sari

Thalassemia Centre, Department of Child Health, Faculty of Medicine Universitas Indonesia / Cipto Mangunkusumo Hospital, Jakarta, Indonesia

Background: Adequate chelating agent is important to manage iron overload in thalassemia patients. The combination of deferoxamine/deferiprone (DFO/DFP) as iron chelator agents for thalassemia patients has been reported since many years, particularly in patients who do not respond adequately to a single chelating agent. The aim of this case report is to investigate the efficacy and safety of combined DFO/DFP in Thalassemia patients with severe iron overload. *Case:* These twins were diagnosed beta thalassemia major since 20 years ago. They were diagnosed cardiomyopathy due to lack of efficacy in removing liver/cardiac iron with monotherapy, diabetes mellitus and hepatitis C. They received combination therapy with DFO (30 mg/kg/day) and DFP (100 mg/kg/day). After 1 year, the improvement in cardiac and liver T2* value was detected. Twin #1: cardiac T2* from 9,43 ms to 10,83 ms, liver T2* from 1,18 ms to 1,70 ms. Twin #2: cardiac T2* from 13,45 ms to 15,89 ms; liver T2* from 1,57 ms to 2,19 ms. The pancreatic T2* #1 was 7,32 ms and 9,53 in #2. The ferritin level was also reduced, from 5,992 mg/dL to 2,310 mg/dL in #1 and 6,039 mg/dL to 2639 mg/dL in #2. No adverse effect was found in these twins until now. *Conclusions:* Combined DFO/DFP can be considered when monotherapy fails to remove iron overload in cardiac and liver tissues. *Keywords:* Combination therapy; deferoxamine; deferiprone; cardiac liver iron overload; thalassemia.

IS THERE A DIFFERENCE IN NEUTROPHIL PHAGOCYTOSIS AMONG DIFFERENT IRON CHELATORS?

T.T. Sari, A.A.P. Akib, D. Gatot, S. Bardosono, S.R.H. Hadinegoro, A.R. Harahap, P. Idjradinata

Thalassaemia Centre, Department of Child Health, Dr. Cipto Mangunkusumo Hospital, Faculty of Medicine Universitas Indonesia

Background: Iron overload in thalassemia patients may lead to the occurrence of many complications, one of which is an impaired immune response. Neutrophil phagocytosis remains as a controversy among thalassemia patients, which can be mainly attributed to difference in transfusion requirements, splenectomy status, iron overload, or iron chelator agents used. Iron chelators are primarily used to decrease the burden of iron overload and improve immune response. This study aims to evaluate neutrophil phagocytosis following the administration of different iron chelators. *Methods:* This cross sectional study was conducted at Dr. Cipto Mangunkusumo Hospital. Subjects were healthy thalassemia major patients above 12 years old who were seronegative-HIV and receiving iron chelation therapy for at least 1 year. Neutrophil phagocytosis was measured using the Phagotest® kit, with a reference value of 96.8-99.6%. *Results:* Among the 116 subjects, 68 (58.6%) were female and 48 (41.4%) were male. The median age of subjects was 21 years old (range 12-38). Fifty-percent of all subjects had undergone splenectomy. The iron chelators used included deferiprone (55%), desferoxamine (22%), deferasirox (14%), and combination therapy (9%). The mean ferritin level was 5,256.50 ng/mL (range 645-21,835). Mean percentage of neutrophil phagocytosis (%) for the subjects receiving desferoxamine, deferiprone, deferasirox, and combination therapy were 46.75 (±29.77), 43.21 (±26.0), 48.10 (±30.67), and 39.58 (±24.43) respectively (p=0.868). *Conclusions:* There were no significant differences observed in neutrophil phagocytosis receiving various iron chelators. Neutrophil phagocytosis in thalassemia patients was comparatively lower to that in healthy individuals. It needs other strategy may be implemented to improve neutrophil phagocytosis. *Keywords:* Neutrophil phagocytosis, thalassemia, iron chelator.

MISCELLANEOUS

REFRACTIVE ERRORS AND OCULAR BIOMETRY COMPONENTS IN THALASSAEMIA MAJOR PATIENTS

Samira Heydarian, Reza Jafari, Hosein Karami

Background and Purpose: The aim of this study is to determine and compare biometric and refractive characteristics of thalassemia major patients and normal individuals. *Methods:* In this cross-sectional study, 54 thalassemia major patients were selected randomly as case group, and 54 age- and sex-

matched healthy subjects were regarded as control group. Refractive errors, corneal curvature and ocular components were measured by autokerato-refractometery and A-scan ultrasonography, respectively. *Results:* Mean spherical equivalent was -0.0093±0.86 D in thalassemia patients and -0.22±1.33 D in the normal group. The prevalence of myopia, Hyperopia, and emmetropia among thalassemia patients was 16.7, 19.4, and 63.9%, respectively. While in the control group, 26.9% were myopic, 25% were hyperopic, and 48.1% were emmetropic. The prevalence of astigmatism in case group was 22.2%, which was not significantly different from that in control group, (27.8%, p=0.346). Mean axial length in thalassemia patients was 22.89±0.70 which was significantly lower than that in normal group (23.37±0.91, p=0.000). The flattest meridian of the cornea (R1) was significantly steeper in thalassemia patients (7.77±0.24) in comparison to normal individuals (7.85±0.28). Although thalassemic patients had significantly smaller axial length and vitreous chamber depth in comparison to normal group, which could be due to their abnormal physical growth, there was no significant difference between the mean of spherical equivalent among two groups. *Conclusions:* This can be due to their steeper corneal curvature that overcomes the refractive disadvantage of their shorter axial length. *Keywords:* Thalassemia major, Ocular biometry, Refractive error, Corneal curvature, Axial length In.

OCULAR ABNORMALITIES IN MULTI-TRANSFUSED β-THALASSAEMIA PATIENTS

Reza Jafari, Samira Heydarian, Hosein Karami, Mohammad Momeni Shektaei, Kiumars Noruzpour Dailami, Ahmad Ahmadzadeh Amiri, Majid Reza Sheikh Rezaee, Asad Allah Farrokh Far

Background and Purpose: The aim of this study was to assess ocular changes in thalassemia patients who have received multiple transfusions and chelate binding therapy in order to avoid iron accumulation. *Methods:* A cross-sectional study. Subjects and Methods: A total of 54 thalassemia major patients were selected as case group, and 54 age- and sex-matched healthy subjects were regarded as a control group. Ocular examination included visual acuity, refraction testing, slit lamp examination, funduscopy, tonometry, perimetry, tear break-up time test, and color vision testing were performed for all the participants. We computed the frequency and duration of blood transfusion, the mean serum ferritin level, pre-transfusion hemoglobin concentration, and type, duration, and daily dose of chelation therapy for thalassemia patients based on their records. Statistical Analysis Used: All data analysis was performed using SPSS, version 19. *Results:* All the thalassemic patients were asymptomatic, but abnormal ocular findings (dry eye (33.3%), cataract (10.2%), retinal pigment epithelium degeneration (16.7%), color vision deficiency (3.7%), and visual field defects (33.7%)) were seen in 68.5% of thalassemic group. The prevalence of ocular abnormalities in normal group was 19.4%, which was significantly lower than that in thalassemia patients (P=0.000). No significant correlation was found between ocular abnormalities and mean serum ferritin level (P=0.627) and mean hemoglobin concentration (P=0.143). Correlation of number of blood transfusion with the presence of ocular abnormalities was found to be statistically significant (P=0.005). *Conclusions:* As life expectancy for beta-thalassemia patients extends, regular ophthalmological evaluation to detect early changes in their ocular system is recommended. *Key words:* Beta-thalassemia major, Blood transfusion, Chelation therapy, Ferritin, Ocular abnormality.

DOES β-THALASSAEMIA INCREASE THE INCIDENCE OF BELLS PALSY?

Shahrazad S. Al Jebori

FICMS, Kerbala Medical College Department of Family and Community Medicine, Iraq

Background: A five year old Iraqi patient with beta thalassemia major (βTM) was presented with recurrent attack of Bell's palsy (BP) on 2 successive years being a milder one in the second year. This is a rare occurrence in βTM patients and needs to be further explored. This case report is presented to find out whether βTM patients are at higher risk for developing BP. *Methods:* This is a case report of a nine year old Iraqi child who is the only child born to parents of secondary consanguinity relatives from Babil Province was diagnosed as βTM on the age of 6 months on regular blood transfusion and iron chelation therapy of deferasirox tablets, his medical follow up takes place at Babil Thalassemia center in Babil Maternity and Child Hospital. On the age of 5 years, He was presented with a sudden onset of right sided BP within 24 hours of swimming in a cool swimming pool at his home, July 2011. A neurosurgeon examined him, no investigations were done, the diagnosis was purely clinical, steroid & neurotonics were recommended, antiviral agents were not prescribed , lacrimation & taste were not affected, there was a slow improvement, 2 weeks later physiotherapy sessions including facial massage and electric stimulation were recommended after which the child improved. The next year, another attach of BP developed at a similar time preceded by the same sequel of events, this time the attach was milder with residual effects seen in the eyes & mouth. *Results:* This coincidence of βTM & BP can be further studied by observational epidemiologic studies to prove or disprove the hypothesis that βTM patients especially children are at higher risk of developing BP than healthy children. *Conclusions:* β-thalassemia major patients may be at higher risk of developing BP. *Key words:* Beta Thalassemia major, Bell's Palsy, Babil Thalassemia center.

THALASSAEMIA, NEW CHALLENGES/PALESTINE

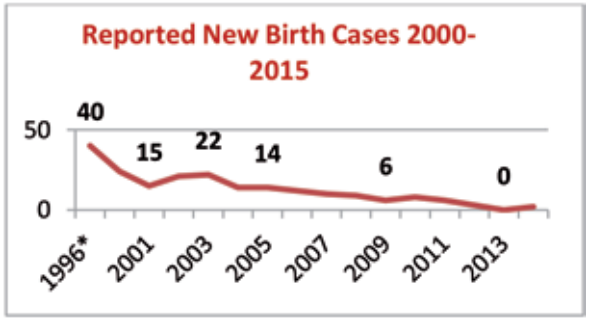
Bashar Karmi

Thalassemia Patients' Friends Society (TPFS), Palestine

Background: Mediterranean region is considered an endemic area for Thalassemia. A lot of efforts were made since 1990 to handle thalassemia disease. These works included both prevention and improving health services. The breaking point was establishing Thalassemia patients' Friends Society on 1996. Through the last twenty years TPFS efforts were directed to achieve these two goals, preventing new thalassemia cases and improving the health situation and welfare of thalassemia patients and their families. The first challenge was dealing with some foreign word like thalassemia in the Palestinian community. Finding our way

out with such preventable disease was also a promising one "no new thalassemia cases by year 2013". Now, we are standing in a position to assess our achievement and to reconsider other challenges and opportunities. Nevertheless, we at TPFS are looking for effective control of thalassemia complications, and the promotion of equal access to quality health care for every patient with thalassemia in Palestine. Still we can see the positive changes taking place in the daily life of our patients during the last years. The growing interest in this group of patients and the inspiring hard work done by patients themselves are our main tool in this mission. Major observations from our data show that: a) 3-4% of Palestinian populations are carriers of β Thalassemia. b) Reduce the number of new patients to less than 10 cases per year since 2002. c) 863 patients registered in Palestine. d) The average age of patients was raised from 7-8 years in 1996 to 19-20 years in 2015, their undergraduate studies and got married and formed their Owen family. e) More than 100 patients graduated from colleges and universities from different sectors and specialties. f) Nearly 30-35 patient are working within. g) Palestinian organization and companies. *Results:* The number of patients born every year has dropped from 50-70 each year before 1996, to less. Than 10 since 2001, the reason for the new cases after the instructions of 2000, it has been observed That the new cases are mostly concentrated in the north parts of the west bank, especially after Obligation the circulation of Court Judge for the premarital examination in 2000. Launching campaigns since more than ten years to Raise awareness and knowledge about the disease and the importance of prevention, also creating of a social dialogue that has yielded and promoted the development of laws and regulations, resulting in: issuing binding instructions to attach the pre- marriage certificate with a blood test that indicates that one of partners is free from the thalassemia disease as of 2001. Partnering with different institutions and sectors (governmental and non- governmental organizations) (Table 1). *Conclusions:* The first twenty years of the march of TPFS is considered to be a very rich of dedication, with the participation and help of hundreds of parents from all districts and thousands of volunteers from schools, universities and academic intuitions, in addition to activists in the medical and youth sectors. We hope thalassemia one day will be something from the past, and that our work will succeed in protecting our community from thalassemia. There is no doubt that thalassemia has become a predictable disease, the risk exists only with two couple- carriers.

Table 1. No new thalassemia cases of 2015.



NEW DEVELOPMENTS IN THALASSAEMIA

POTENTIAL EFFECTS OF SILYMARIN AND ITS FLAVONOLIGNAN COMPONENTS IN PATIENTS WITH β-THALASSAEMIA MAJOR: A COMPREHENSIVE REVIEW IN 2015

Hadi Darvishi Khezri[1], Ebrahim Salehifar[2*], Mehrnoush Kosaryan[3], Aily Aliasgharian[4], Hossein Jalali[1], Arash Hadian Amree[1]

[1]Research Student, Thalassemia Research Center, Mazandaran University of Medical Sciences, Sari, Iran; [2]Clinical Pharmacology, Thalassemia Research Center, Mazandaran University of Medical Sciences, Sari, Iran; [3]Pediatrics, Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran; [4]Microbiology, Faculty of Medicine, Thalassemia Research Center, Mazandaran University of Medical Sciences, Sari, Iran

 Major β-thalassemia (β-TM) is one of the most common inherited hemolytic types of anemia which is caused as a result of absent or reduced synthesis of β-globin chains of hemoglobin. This defect results in red blood cells lysis and chronic anemia that can be treated by multiple blood transfusions and iron chelation therapy. Without iron chelation therapy, iron overload will cause lots of complications in patients. Antioxidant components play an important role in the treatment of the disease. Silymarin is an antioxidant flavonoid isolated from Silybum marianum plant. *Methods:* In the present study, we reviewed clinical and experimental studies investigating the use of silymarin prior to September 1, 2015, using PubMed, ISI Web of Knowledge, Science Direct, Scopus, Ovid, and Cochrane Library databases and we evaluated the potential effects of silymarin on controlling the complications induced by iron overload in patients with β-TM. *Results and Conclusions:* Based on the results of the present study, we can conclude that silymarin may be useful as an adjuvant for improving multiple organ dysfunctions. *Keywords:* Silymarin, Flavonolignan, β-Thalassemia Major.

LUSPATERCEPT DECREASES TRANSFUSION BURDEN AND LIVER IRON CONCENTRATION IN REGULARLY TRANSFUSED ADULTS WITH β-THALASSAEMIA

Antonio G. Piga[1], Silverio Perrotta[2], Angela Melpignano[3], Caterina Borgna-Pignatti[4], M. Rita Gamberini[4], Ersi Voskaridou[5], Vincenzo Caruso[6], Paolo Ricchi[7], Antonello Pietrangelo[8], Xiaosha Zhang[9], Dawn M. Wilson[9], Ashley Bellevue[9], Abderrahmane Laadem[10], Matthew L. Sherman[9], Kenneth M. Attie[9]

[1]Turin University, Turin, Italy; [2]Second University of Naples, Naples, Italy; [3]Ospedale "A. Perrino", Brindisi, Italy; [4]Arcispedale S. Anna, Cona, Ferrara, Italy; [5]Laiko General Hospital, Athens, Greece; [6]ARNAS Garibaldi, Catania, Italy; [7]AORN "A. Cardarelli", Naples, Italy; [8]CEMEF, Medicina 2, Modena, Italy; [9]Acceleron Pharma, Cambridge, MA, USA; [10]Celgene Corporation, Summit, NJ, USA

Background: Luspatercept (ACE-536) is a fusion protein containing a modified activin receptor type IIB that promotes late-stage erythroid differentiation. Luspatercept corrected

the effects of ineffective erythropoiesis in a thalassemia mouse model and was well tolerated and increased hemoglobin in a phase 1 clinical study. *Methods:* In this phase 2 open-label study, luspatercept was administered to adults with β-thalassemia SC every 3 wks for 12 weeks in the completed base study and an additional 2 years in the ongoing long-term extension study. Six cohorts (n=35) were treated at dose levels from 0.2-1.25 mg/kg. Patients in the expansion cohort (n=29) and extension study (n=51) were treated at 0.8 mg/kg with escalation up to 1.25 mg/kg. *Results:* 30 regularly transfused pts (≥4 units RBC/8 wks) completed the base study and 24 enrolled in the extension study (data as of 11 March 2016). Data summarized are for the long-term extension study; results from the base study will also be presented. Median age was 38 yr (range 22-55 yr), 67% had prior splenectomy. Median baseline transfusion burden was 8 units/12 wks (range 4-15 units). Mean (±SD) LIC was 5.1±5.3 mg/g dw. 16/24 (67%) pts achieved a ≥33% and ≥50% decrease in transfusion burden over any 12-weeks compared to baseline, respectively. Duration of response ranged from 12-48+ weeks. 2/3 (67%) pts with baseline LIC ≥5 mg/g dw had a decrease in LIC ≥2 mg/g dw after at least 6 months of treatment. Luspatercept was generally well tolerated, with no related SAEs. AEs were mostly mild-moderate; the most frequent related AEs (≥10%) were bone pain, myalgia, arthralgia, headache, asthenia, and musculoskeletal pain. *Conclusions:* Luspatercept treatment was well-tolerated and led to decreased RBC transfusion requirements and LIC in TD β-thalassemia patients. A Phase 3 study of luspatercept in regularly transfused adults with β-thalassemia is ongoing (NCT02604433).

LUSPATERCEPT INCREASES HAEMOGLOBIN AND IMPROVES QUALITY OF LIFE IN NON-TRANSFUSION DEPENDENT ADULTS WITH β-THALASSAEMIA

Antonio G. Piga[1], Silverio Perrotta[2], Angela Melpignano[3], Caterina Borgna-Pignatti[4], M. Rita Gamberini[4], Ersi Voskaridou[5], Vincenzo Caruso[6], Paolo Ricchi[7], Antonello Pietrangelo[8], Xiaosha Zhang[9], Dawn M. Wilson[9], Ashley Bellevue[9], Abderrahmane Laadem[10], Matthew L. Sherman[9], Kenneth M. Attie[9]

[1]Turin University, Turin, Italy; [2]Second University of Naples, Naples, Italy; [3]Ospedale "A. Perrino", Brindisi, Italy; [4]Arcispedale S. Anna, Cona, Ferrara, Italy; [5]Laiko General Hospital, Athens, Greece; [6]ARNAS Garibaldi, Catania, Italy; [7]AORN "A. Cardarelli", Naples, Italy; [8]CEMEF, Medicina 2, Modena, Italy; [9]Acceleron Pharma, Cambridge, MA, USA; [10]Celgene Corporation, Summit, NJ, USA

Background: Luspatercept (ACE-536) is a fusion protein containing a modified activin receptor type IIB that promotes late-stage erythroid differentiation. Luspatercept corrected the effects of ineffective erythropoiesis in a thalassemia mouse model and was well tolerated and increased hemoglobin in a phase 1 clinical study. *Methods:* In this phase 2 open-label study, luspatercept was administered to adults with β-thalassemia SC every 3 wks for 12 weeks in the completed base study and an additional 2 years in the ongoing long-term extension study. Six cohorts (n=35) were treated at dose levels from 0.2-1.25 mg/kg. Patients in the expansion cohort (n=29) and extension study (n=51) were treated at 0.8 mg/kg with escalation up to 1.25 mg/kg. *Results:* 34 non-transfusion dependent pts (<4 units RBC/8 wks) completed the base study and 27 enrolled in the extension study (data as of 11 March 2016). Data summarized are for the long-term extension study; results from the base study will also be presented. Median age was 37 yr (range 23-62 yr); 67% had prior splenectomy. Median baseline Hgb was 8.7 g/dL (range 7.6-9.8 g/dL). Mean (±SD) LIC was 4.9±3.4 mg/g dw. 21/27 (78%) and 15/27 (56%) pts achieved ≥1.0 g/dL and ≥1.5 g/dL increase in mean Hgb over any 12-weeks, respectively. Duration of response ranged from 113-505+ days. Increases in mean hemoglobin over a 12-week period correlated with increases in patient reported quality of life questionnaire, FACIT-F (r=0.67, p=0.001). Decreases in measures of iron overload were also observed in some patients. Luspatercept was generally well tolerated, with no related SAEs. AEs were mostly mild-moderate; the most frequent related AEs (≥10%) were bone pain, headache, musculoskeletal pain, and arthralgia. *Conclusions:* Luspatercept treatment was well-tolerated and led to increased hemoglobin levels, decreased LIC, and improved quality of life in NTD adults with β-thalassemia.

NON TRANSFUSION-DEPENDENT THALASSAEMIA

UTILITY OF SERUM FERRITIN LEVELS BETWEEN 300 AND 800 NG/ML IN CLINICAL DECISION MAKING IN NON TRANSFUSION-DEPENDENT THALASSAEMIA WHEN MAGNETIC RESONANCE IMAGING FOR LIVER IRON CONCENTRATION MEASUREMENT IS NOT AVAILABLE

Antoine N. Saliba[1], Khaled M. Musallam[2], Maria D. Cappellini[3], Vip Viprakasit[4], Shahina Daar[5], Hassan M. Moukhadder[2,6], Ali T. Taher[2,6]

[1]Indiana University School of Medicine, Department of Medicine, Indianapolis, Indiana, United States; [2]American University of Beirut Medical Center, Department of Internal Medicine, Beirut, Lebanon; [3]University of Milan, IRCCS Ca' Granda Foundation Maggiore Policlinico Hospital, Department of Clinical Sciences and Community, Milan, Italy; [4]Mahidol University, Siriraj Hospital, Department of Pediatrics and Internal Medicine, Bangkok, Thailand; [5]Sultan Qaboos University, College of Medicine & Health Sciences, Department of Hematology, Muscat, Oman; [6]Chronic Care Center, Hazmieh, Lebanon

Background: The assessment of liver iron concentration (LIC) by magnetic resonance imaging (MRI) remains to be the recommended means of quantifying iron overload in patients with non-transfusion-dependent thalassemia (NTDT) to guide iron chelation therapy (ICT). On the other hand, serum ferritin measurement, a simple and more affordable tool, has a significantly positive correlation with LIC and therefore may be utilized in lieu of the latter in countries or centers where MRI is not available or affordable, in light of the high prevalence of NTDT in developing countries. While serum ferritin levels of >800 ng/mL were shown in previous studies to most certainly predict an LIC ≥5 mg/g dry weight (dw), which corresponds to the concentration heralding iron-related morbidity and therefore prompts initiation of ICT, almost 50% of patients with levels <800 ng/mL were demonstrated to have LIC levels ≥5 mg/g dw. To this purpose, we evaluated the utility of serum ferritin in predicting

the need for ICT for levels ranging between 300 and 800 ng/mL, 300 being the upper limit of normal serum ferritin level. *Methods:* We looked at 71 patients (mean age 33.3±13.9 years) with serum ferritin levels ranging between 300 and 800 ng/mL attending centers in Italy, Lebanon, Oman, and Thailand. 53.5% were female, and 45.1% were splenectomized. *Results:* Using a logistic regression analysis with LIC ≥5 mg/g dw as the desired outcome, we were able to construct a probability curve for serum ferritin level in predicting an LIC ≥5 mg/g dw. *Conclusions:* Instead of categorically assuming patients with serum ferritin levels <800 ng/mL not to be eligible for ICT, our probability curve helps assign a percentage risk of having an LIC ≥5 mg/g dw to such patients, which, in tandem with the patients' general clinical picture, can better guide decision making towards implementation of ICT.

ARAP-536 (MURINE ANALOG OF ACE-536/LUSPATERCEPT) INHIBITS SMAD2/3 SIGNALING AND PROMOTES ERYTHROID DIFFERENTIATION BY RESTORING GATA-1 FUNCTION IN A MURINE MODEL OF β-THALASSAEMIA

Pedro Martinez[1*], Manoj Bhasin[2], Robert Li[1], Scott Pearsall[1], Ravindra Kumar[1], Rajasekhar Suragani[1]

[1]*Preclinical Department, Acceleron Pharma, Cambridge;* [2]*Genomics and Proteomics Center, Beth Israel Deaconess Medical Center, Harvard Medical School, Boston, United States*

Background: Luspatercept (modified ActRIIB receptor-Fc fusion protein), a Smad 2/3 ligand trap, has demonstrated efficacy in correcting ineffective erythropoiesis (IE) and anemia in murine models of MDS and β-thalassemia. *Methods:* β-thalassemic mice ($Hbb^{th3/+}$) were administered a single bolus of vehicle (VEH) or RAP-536. Splenic basophilic erythroblasts were sorted for genome wide transcriptome profiling. Also, mouse erythroid leukemic (MEL) cells, primary fetal liver erythroid and β-thalassemic erythroid precursors were treated with GDF11 in the presence or absence of ACE-536. *Results:* Transcriptome analysis of β-thalassemic erythroblasts identified 74 genes that were differentially expressed in RAP-536 treated samples *vs.* VEH. Gene set enrichment analysis (GSEA) depicted significant upregulation of 158 activated genes downstream of GATA-1 (erythroid master regulator) such as those involved in heme biosynthesis and terminal erythroid differentiation. ACE-536 binds and inhibits Smad2/3 ligands such as GDF8, GDF11 and Activin B. Treatment of MEL and fetal liver erythroid cells with GDF11 induced Smad2/3 phosphorylation and ACE-536 co-treatment inhibited the increase in pSmad2/3. In differentiating erythroid cells, GDF11 treatment displayed reduced nuclear GATA-1 protein levels. Reactive oxygen species (ROS) and Caspase 3/7 were elevated in MEL and primary fetal liver cells following GDF11 treatment. Importantly, treatment of erythroid cells with ACE-536 and GDF11 decreased ROS, and restored GATA-1 to control levels. *Conclusions:* These data provide a potential mechanistic role for luspatercept as a novel treatment of β-thalassemia. By inhibiting pSmad2/3 signaling, RAP-536 treatment decreases ROS, caspase3/7 activation and GATA-1 cleavage. Thus by restoring GATA-1 availability and functional activity, RAP-536 treatment causes upregulation of genes involved in promoting terminal erythroid maturation, and consequently corrects anemia in β-thalassemia. Luspatercept has initiated phase 3 studies in patients with MDS and β-thalassemia.

QUALITATIVE RESEARCH ON THE OBSTACLES IN MARRIAGE AND REPRODUCTIVE PROCESS OF PEOPLE WITH TRANSFUSION DEPENDENT THALASSAEMIA

Sachiko Hosoya[1*], Gholam Reza Jamali[2], Mohsen Shahriari[3]

[1]*Research Fellow, Research Institute of Language and Cultures in Asia and Africa, Tokyo University of Foreign Studies;* [2]*Nursing, School of Nursing and Midwifery, Isfahan University of Medical Sciences;* [3]*School of Nursing and Midwifery, Isfahan University of Medical Sciences*

Background: In Iran, the largest population age of the people with transfusion dependent Thalassemia (PTDT) now has reached late teens to 20's. In other words, most of the PTDT are in their reproductive age. However, many PTDT have difficulties going through the marriage and reproductive process. Considering the recent increase of life expectancy of PTDT in Iran, it is an urgent task now to create a better support program for PTDT's marriage/reproductive life from the nursing care perspective, and a basic study for this purpose is needed. *Methods:* Semi-structured interview with 60 married/single PTDT who came to the Thalassemia Center in Seyed-al-Shoahad hospital in Isfahan, and qualitative analysis were conducted in 2016. *Results:* There are some differences between the difficulties which male and female PTDT experience. For male PTDT, unemployment were recognized as the hardest obstacles, while female PTDT found that the fertility problems are more difficult matters. Prejudice towards PTDT is a serious problem for both men and women. Many of them had a thought to choose a PTDT as a marriage partner at least once, but only some of them have married with PTDT, because of their families' disagreement. The negative episodes such as unsuccessful marriage proposal, quarrel with their family over their marriage choice, or pressure to have a baby, tend to result in depression and inappropriate self-care. *Conclusions:* Pre-marital educational programs for PTDT about the fertility should be created based on this research, as well as the personal counseling programs for their own family and future partner/family about the PTDT's capabilities and health situations. To solve their unemployment problem, which could be recognized as a health issue equally as a social issue, the staff in Thalassemia word can approach through collaborative programs with local community.

KNOWLEDGE ATTITUDE AND PRACTICES AMONG PATIENTS ON TREATMENT OF THALASSAEMIA IN MALDIVES

Fathmath Jeehan Saleem

Maldivian Thalassaemia Society, Republic of Maldives

Background: Maldives has the highest prevalence of beta Thalassaemia in the world with 18% of its population being carriers for Thalassaemia (SHE 1992). To date over 800 patients have been registered. Adherence to the management protocol followed by patients is inadequate resulting in premature mortality among the young adults. *Aims and Objectives:* The aim of the study was to assess awareness and knowledge among young patients on standard treatment

protocols that they require and if the knowledge translates into practices on factors that make them susceptible to and prevent them from chronic organ damage and other complications. It further explores availability and access to standardised treatment in the country. *Methods:* A questionnaire based KAP survey among Thalassaemia patients (above 12 years) registered at National Thalassaemia centre (NTC/MBS). Survey was conducted betweeen July 2014 to October 2014. Questionannaire was designed to gather information on transfusion regime, chelation therapy and growth and development, treatment facilties and sevices. *Results:* Results of the survey show that standard treatment with blood transfusion and iron chelation is the primary focus of treatment for patients despite their age. Patients are knowledgeable about transfusion, but not so much on chelation and less aware and knowledgeable on aspects related to growth and development. 100% of the patients strictly adhered to transfusion regime while with chelation all patients have had lapses at some point of their life. The average serum ferritin levels of patients are over 4000 µg/l. Over 90% patients are aware excess iron in their body will have cardiac, hepatic and endocrine problems which can be life threatening. However patients are not fully concerned that iron overload could have an immediate impact on their own health. Lack of motivation to adhere to treatment has been reported as the main reason for poor compliance. Majority patients reported they are blamed for low compliance rather then encouraged or provided support toward establishing a routine chelating regime. Patient reported that their inputs, discussion into decisions reagrding their health and wellbeing are not encouraged. Treatment regime has a meticulous focus on transfusion and adult patients are still being treated and managed with teatment adequate for children. All patients recognised the importance of professional support as they face daily challenges growing up with a chronic condition. Psychosocial issues are a very important yet a neglected area to integrate into standard treatment protocols in the country. *Conclusions:* Improved patient knowledge and their positive attitude is very important in shaping up their compliance and healthy progression to adulthood. Both medical community and patients need to reshape their thinking and improve knowledge. Information from the study needs to translate to targeted awareness programs, identify and fill gaps in treatment protocols, which will have behavioral modifications to prevent complications and potentially saving young lives. A patient centred approach in management and treatment is required to ensure patients an equal chance in life. *Key words:* Transfusion, Chelation, Thalassaemia, Growth and development, Compliance.

ATTRIBUTIONAL STYLES IN ADOLESCENTS WITH TRANSFUSION-DEPENDENT THALASSAEMIA

Lerlugsn Suwanthol, Tuangrath Sangpaypan, Phetcharat Naknum, Kleebsabai Sanpakit

Department of Pediatrics, Faculty of Medicine Siriraj Hospital, Mahidol University, Bangkok ,Thailand

Background: Adolescents with Transfusion- dependent thalassemia must cope with the ongoing anxiety and negative attributional styles due to treatment management, but few measured broad categories of causal explanations for this population. *Objectives:* To examine empirically the relation between anxiety symptoms and three dimensions of attributions : internal-external, stable-unstable, and global-specific in Adolescents with Transfusion- dependent thalassemia. Methods: With cross-sectional research design and consisted of 102 adolescents (48 males, 54 females) diagnosed with Transfusion- dependent thalassemia (more than 50 times for blood transfusions) completed measure of Attributional Styles and Anxiety Questionnaires. For correlations in the predicted directions among variables were examined with Pearson product- moment correlation coefficients, t-test and One way ANOVA were conducted to ascertain significant between group differences on attributional factors and levels of anxiety symptoms. *Results:* Adolescent samples with higher levels of anxiety revealed statistically significant relationship among three negative attributional dimensions (overall composite $F=4.5$, $p<.05$; negative composite $F=4.99$, $p<.01$; negative internality $F=4.50$, $p<.01$; negative stability $F=3.42$, $p<.05$ and negative globality $F=3.77$, $p<.05$). In addition, significant age- group differences were found for the total negative globality ($t=-2.05$, $p<.05$) and negative globality ($t=-2.22$, $p<.05$). *Conclusions:* This work reveals consistent support for the reformulated learned helplessness theory of depression. In finding that individuals who attribute negative life events to internal, stable, and global causes will be more vulnerable to anxiety than those who make external, unstable, and specific attributions. Moreover, those adolescents more than 17 years evidenced a more negative globality attributional style than their groups less than 16 years, and this pattern may be influenced by female adolescents. Finally, examining adolescent's attributions as a risk factor for poor adjustment will help pediatric professionals identify adolescent at risk for mental health problems, while investigating the effect of negative globality attributional style will help clarify whether cognitive interventions should be target on early, middle or late adolescence. *Keywords:* Attributional styles, anxiety, adolescents with transfusion- dependent thalassemia.

QUALITY CARE FOR QUALITY OF LIFE

MULTIFACETED APPROACH TO THALASSAEMIA CARE AND MANAGEMENT – OUR EXPERIENCE AND LESSONS LEARNED

Shashikant Patil, Rajendra Kulkarni, Vaishali Kale, Snehal Patil, Nandkishor Tated, Atul Jain

Arpan Thalassemia Society, Arpan Blood Bank, Nashik, Maharashtra, India

Background: If managed properly with adequate overall medical care and good quality & safe blood transfusion support, the quality of life of thalassemia major patients can be improved significantly. *Aim:* To implement a comprehensive, multifaceted thalassemia care and management programme under one roof. *Methods:* Thalassemia major patients within the expanse of north Maharashtra were registered with our organisation. A programme for medical management including iron chelation therapy, haematologic, cardiologic, paediatric, orthopedic, pathological, radiological evaluation was defined. A monitoring programme was formulated using guidelines given by Indian Academy of Paediatrics. All

patients were offered ID-NAT tested, inline-leucodepleted blood free of cost with the help of corporate support. A dedicated donor programme was implemented for the transfusion support of the study group. All the patients were followed up for febrile non-hemolytic reactions, sero-conversion for TTI, alloimmunization, growth-disorders, iron overload for a period of 3 years. *Results:* Total 181 patients with age ranging from 2 years to 30 years were enrolled in this programme. 4662 transfusions have been given over the period of 3 years to 181 patients. 3 patients (1.6%) have developed alloimunization. 1 patient (0.5%) had experienced febrile non-hemolytic transfusion reaction. Severe iron overload (according to T2* MRI) was seen in 2 out of 12 patients (16.66%). Growth disorders/stunted growth have been observed in 25 (13.81%) patients. None of the above patients were found to have sero-conversion for HIV, HBV or HCV. *Conclusions:* Implementing a comprehensive transfusion therapy and health monitoring programme for thalassemia patients under one roof is feasible with support from corporate & society. Careful periodic monitoring of these patients & implementation of NAT tested, leucodepleted & phenotype matched blood transfusion can go a long way to improve the quality of life and overall life expectancy of thalassemia major patients.

PRESENTATION OF THALASSAEMIA WARD IN EAST MEDICAL SPECIAL CENTER IN TEHRAN AND CLINICAL CHALLENGES IN MANAGEMENT OF THALASSAEMIC PATIENTS IN ALL FIELDS

M. Sabzechian[1], R. Fallah[2], M. Gholami[3]

[1]Pediatric Hematologist Oncologist in East Medical Special Center; [2]General Physician in East Medical Special Center; [3]Neurse of Thalassemic Patients in East Medical Special Center, Iran

East Medical Centre with the most advanced and specialized medical facilities based on international standards have been established by the Charity Foundation for Special Diseases. The East medical special center is located in the south east of Tehran and ready to deliver health care in different parts of the public tariff. East Medical Centre by the Charity Foundation for Special Diseases and assistance of the people and the central government set up a fourth center in the country launched by the Charity Foundation for Special Diseases. This center was started its activity on 2005. Thalssemia ward is very active in last 5 years. About 60 thalassemic patient confer in this center regularly. During the last 3-4 years the number of patients referred with regard to the facilities of this center is to this sector increased by 50%. At this time about 60 thalassemic patients is in health care coverage. Health care provided to patients with thalassemia include: regular visits to patients by medical professionals, complete tests of hematology, kidney, liver, virology, endocrinology (every 6 months in own laboratory), psychiatric counseling, family counseling, marriage counseling is done, regular visit by cardiologist and endocrinologist, blood transfusion, intra venuos desferal therapy, intravenous zolendronic acid in osteoporotic thalassemic patients, intravenous anti biotic therapy for splenectomized patients, and other services. In this center Ab screening is done in laboratory. Thalassemia patents refer for liver and heart T2 and R2 star MRI, bone mineral density assay, abdominal sonography, opyometry, oudiology, echocardiography annually. Patient information and results is recorded in file for each patient. The services provided patient satisfaction was very good during recent years . At this time 50 cases of major thalassemia, 6case of intermedia thalassemia, one case ofsickle celldisease, one case of alpha thalassemia, one case of lepor thalassemia is confer in this center. Two cases of major thalassemia refer for bone marrow transplantation ward. One case of two patient that refer for BMT have very good result.

EFFECT OF DEFERASIROX USE ON THE EDUCATIONAL STATUS OF PATIENTS WITH THALASSAEMIA: A 20-YEAR, SINGLE-CENTER EXPERIENCE

Rayan Bou Fakhredin[1], Joseph Roumi[1,2], Josephine Ajouz[2], Hassan M. Moukhadder[1,2], Rita Aoun[2], Suzanne Koussa[2], Ali T. Taher[1,2]

[1]American University of Beirut Medical Center, Department of Internal Medicine, Beirut, Lebanon; [2]Chronic Care Center, Hazmieh, Lebanon

Background: Very few reports in the literature have focused on the psychosocial status of patients with thalassemia. Indeed, the multitude of medical morbidities that have been historically associated with thalassemia, namely those stemming from iron overload and its complications, used to get in the way of patients wanting to seek education. However, an improved health-related quality of life following the movement towards deferasirox use in the Chronic Care Center in Lebanon in 2006, enabled many patients to actualize themselves through the attainment of higher education levels. The aim of this study is to evaluate the impact of deferasirox use on the educational status of patients with thalassemia from our practice. *Methods:* A total of 60 thalassemia patients (30 males and 30 females) were included in this study. Patients were divided into 3 age groups: 12-18, 19-25, and 26-40. The educational status of the patients in each group was compared between two time periods – 1996-2005 and 2006-2016. Educational status was categorized as university degree, high school degree, or less than high school degree. *Results:* In the 12-18 age group, the number of patients achieving a high school degree increased from 30% before the implementation of deferasirox use to 90% after it. Similarly, the number of patients achieving a university degree increased in both 18-25 and 25-40 age groups after the introduction of deferasirox treatment, from 20% to 50% in the former and from 30% to 70% in the latter. *Conclusions:* Individuals with thalassemia are now able to seek and achieve higher levels of education after the introduction of deferasirox use for iron chelation, which has translated into a health-related quality of life more permissive of their quest to better educate themselves. This might be very helpful in creating effective psychosocial plans for thalassemia patients.

DEVELOPMENT OF PATIENT-REPORTED OUTCOMES SYMPTOM MEASURE FOR PATIENTS WITH NON-TRANSFUSION-DEPENDENT THALASSAEMIA (NTDT-PRO©)

Ali Taher[1], Antoine Saliba[1], Antonis Kattamis[2], Richard Ward[3], Gale Harding[4], Xiaohan Hu[5], Dalia Mahmoud[5]

[1]American University Beirut, Beirut, Lebanon; [2]Aghia Sophia Children's Hospital, Athens, Greece; [3]University Health Network,

Toronto, Canada; [4]Evidera, Inc., Bethesda, USA; [5]Celgene Corp., Summit, USA

Background: Currently, there is no disease-specific Patient-Reported Outcomes (PRO) measure available to assess key symptoms experienced by patients with non-transfusion-dependent thalassemia (NTDT). *Aim:* To develop a symptom measure for NTDT patients for use in clinical trials. *Methods:* A qualitative study was conducted to develop the NTDT-PRO© symptom measure consistent with regulatory requirements. The instrument was developed based on concept elicitation interviews, input from clinical experts, and refined through a process of cognitive interviews. Concept elicitation interviews were conducted among a total of 25 NTDT patients recruited from Lebanon, Greece, and Canada. Findings from these interviews were used to develop the draft measure. Cognitive interviews were subsequently conducted among subjects in Greece and Lebanon (N=21) to examine the relevance and understanding of the items among the target population. All interviews were conducted in person in the local language with written informed consent obtained. *Results:* Based on findings from concept elicitation interviews, a total of nine symptoms were included in the NTDT-PRO© Version 1. An 11-point numeric rating scale was used for the response options and a daily recall period was selected based on the day-to-day variability of symptoms. Findings from the cognitive interviews indicate that subjects understood the items as intended, with a few minor exceptions. Four items were deleted as they were not considered core symptoms, and one item was added to address the symptom both with and without physical activity. The NTDT-PRO© Version 2 includes six items that address three key symptoms of tiredness, weakness, and shortness of breath both during physical activity and at rest. *Conclusions:* The NTDT-PRO© Version 2 consists of six items to assess the key symptoms of NTDT using a 24-hour recall period, with plans to further evaluate the measures in an ongoing observational study and for eventual inclusion into a randomized phase 3 study.

SICKLE CELL DISEASE

CLINICAL AND HAEMATOLOGICAL PROFILES AMONG BAHRAINI CHILDREN WITH SICKLE CELL DISEASE

Husain AlMukharraq

Consultant Pediatrician-Hematologist & Oncologist, Manama, Bahrain

Aim: To determines the clinical and hematological parameter and manifestation of children with sickle cell disease in Bahrain. *Patients and Methods:* Total number of admission over one year in the pediatric department were 4552, sickle cell disease patients were 552 (12.1%) percentage. *Results:* A total of 552 files reviewed and 52 were excluded and the remaining 500 enrolled in the study. The study population divided into 2 group according to age: Group I: 1-7 years and group II: 8-15 years. The male is predominating in both groups 68% and 52% respectively, there is no difference in the mean hemoglobin. Although the MCV and MCH is lower in both groups yet it is lower in group I. The retics counts is higher in group II (4.9% compared 3.9%). The mean WBC and platelets counts were both high in group I. The HBS is higher in group II (78.1% *VS* 65.8%), the HBF higher in group I (25.9% *VS* 17.7%). The G6PD deficiency was higher in group I (42.1% *VS* 35.2%). The jaundice and splenomegaly were higher in group II (84% and 56% respectively). Vaso-occlusive (painful) crises represent 69.3%, fever 11.7%, hyper hemolysis/severe anemia 9.1%, acute chest/pneumonia 6.5%, gastroenteritis 2.7%, osteomyelitis 2.2%. The cerebrovascular accident, sequestration and hand food syndrome were each less than 1%. *Conclusions:* The low MCV and MCH in both group could be explained by the co-inheritance of alpha thalassemia or iron deficiency anemia; HbS, jaundice and splenomegaly is higher in older age children; the G6PD deficiency is a common inherent among patients with sickle cell disease. The Vaso-occlusive crisis is the commonest manifestation of sickle cell disease where other complications were relatively less common in comparison to other SCD haplotypes.

ASSOCIATION OF METHYLENETETRAHYDROFOLATE REDUCTASE A1298C AND C677T POLYMORPHISMS WITH VASO-OCCLUSIVE CRISIS IN SICKLE CELL DISEASE

Ahmed A. Raouf[1], Mona M. Hamdy[2], Ahmed M. Badr[2], Osama F. Shalaan[3], Moustafa A. Sakr[3], Abdel Rahman A. Abdel Rahman[3]

[1]Clinical Biochemistry Department, Liver Institute , Menoufia University, Egypt; [2]Pediatrics Department, Faculty of Medicine, Cairo University, Egypt; [3]Molecular Diagnostics and Therapeutics Department, Genetic Engineering and Biotechnology Research Institute, University Of Sadat City, Egypt

Background: Vaso-occlusion is a determinant for most signs and symptoms of sickle-cell anemia (SCA). Elevated Homocysteine concentration contribute to thrombosis, a frequent event in sickle cell anemia. Methylenetetrahydrofolate reductase (MTHFR) is an important enzyme, which regulates homocysteine metabolism, and some polymorphisms of gene encoding this enzyme are associated with a decreased activity of the enzyme. The aim of the study was to assess an association between the C677T and A1298C polymorphisms and the frequency of vasoocclusive crisis. *Methods:* A case- control study was carried over a period of one year from January to December 2014 including 50 sickle cell disease patients collected together with 30 age, and sex matched apparently healthy controls. Venous blood sample were aspirated from both groups to estimate serum homocysteine, folic acid, C677T and A1298C polymorphism identification through tetra primer ARMS PCR. Statistical analysis was done, using the student T-test, Pearson correlation analysis and χ2test. *Results:* Homocysteine level was significantly higher in the patients group compared with control group with P value >0.01. Moreover, a strong positive correlation between homocysteine level and the frequency of vaso-occlusive crisis was found (×2 4.836 and P value 0.04). Association between vaso-occlusive events and polymorphism frequency showed no significant difference for both the C677T and the A1298C polymorphisms (χ2=0.206; p=0.9020) and (χ2=1. 720; p=0.4231) respectively. *Conclusions:* We concluded that hyperhomocysteinaemia is positively correlated with the frequency of vaso-occlusive crisis and neither the presence of C677T nor A1298C MTHFR gene polymorphism are risk factors for vaso-occlusive crisis in the SCD patients evaluated. *Keywords:* Vaso-occlusive crisis. Hyperhomocysteinemia, C677T, A1298C.

CLINICAL PRESENTATION OF CHILDHOOD SICKLE CELL DISEASE AND ITS CORRELATION WITH DISEASE PROGNOSIS: A SINGLE CENTRE STUDY

Abdullah A. Bukhari[1], Abdulmalik A. Mulla[1], Abdullah M. Sonbol[1], Anas M. Makhdoum[1], Assem A. Alblowi[1], Badr Z. Mahmoud[1], Zakaria Alhawsawi[2], Abeer Abd Elmoneim[1,3]

[1]Pediatric Department, Taibah University, Almadinah Almounourah, Saudi Arabia; [2]Hematology/oncology center, Maternity and Children Hospital (MCH), Almadinah Almounourah, Saudi Arabia; [3]Pediatric Department, Sohag University, Sohag, Egypt

Background and Aim: Sickle cell disease (SCD) is an autosomal recessive disorder characterized by production of abnormal hemoglobin S and is associated with high morbidity and mortality. The prevalence of SCD in Saudi Arabia varies significantly in different parts of the country, with the highest prevalence is in the eastern province, followed by the south-western provinces. In Madinah, the estimated prevalence of sickle cell homozygosity (Hb SS) is 0.01, and that of the carrier state (Hb AS) is 0.087. In this study we aim to evaluate the clinical presentation of SCD children in Madinah Governorate, Saudi Arabia and its correlation to the disease prognosis. *Patients and Methods:* In a retrospective analysis the study included all SCD children admitted to Maternity and Children hospital (MCH) in Almadinah Almounourah, Saudi Arabia from year 2000 to 2015. Descriptive analysis and chi-square test were used to identify the main factors using SPSS (Version 20, USA). Backward logistic regression was used to find the association between factors. *Results:* SCD is commonly diagnosed in infants aged 6-11 months, with hand-foot swelling and jaundice being the commonest symptoms at presentation. Anaemic and vaso-occlusive crises were seen more common in children aged 1-5 years. The over-all morbidity pattern is the same in both sexes with diseases such as bronchopneumonia, malaria, osteomyelitis, urinary tract infections, septicaemia and septic arthritis being common. Age and presence of complications such as acute chest syndrome and strock at first presentation have been found to influence morbidity pattern in our studied patients. *Conclusions:* There is need for early diagnosis and counseling, so that mothers or caregivers will be able to assist in prompt identification of these morbidities and to seek for prompt and appropriate treatment in the health facilities.

THE OUTCOME OF PREOPERATIVE TRANSFUSION GUIDELINE ON SICKLE CELL DISEASE PATIENTS AT KING FAHD HOSPITAL-JEDDAH

Sameera M.R. Felemban[1,2], Rekha Bajoria[2], Amani Alsawaf[1], Ratna Chatterjee[2], Abdulelah I. Qadi[1]

[1]King Fahd Hospital, Jeddah, Saudi Arabia; [2]Institute For Women's Health, UCL, UK

Background: We developed a local hospital preoperative transfusion guideline for sickle cell disease (SCD) patients to reduce the perioperative and the postoperative complications. This study was conducted to evaluate the outcome of clinical practice on SCD patients undergoing surgeries in our institution. *Methods:* A retrospective review of 75 SCD patients undergoing surgery at King Fahd Hospital, Jeddah, Saudi Arabia was conducted between April 2005 and May 2010. The medical records were reviewed to define the perioperative risks and the postoperative complications in relation to the type of transfusion modality selected. *Results:* The medical records of 75 SCD patients who underwent surgeries were reviewed. Preoperatively, 25.3% had complete exchange transfusion (CETX), 17.3% had partial exchange transfusion (PETX), 26.7% had simple top up transfusion (STX) and 30.7% did not require transfusion (NTX). The postoperative complications included vaso-occlusive crises (VOC) in 20%, acute chest syndrome (ACS) in 2.7%, and fever in 16% cases. 33.3% patients required the prolonged period of the hospital stay. In the patients of our study, postoperative fever, VOC, ACS, and the length of hospital stay did not show any difference regardless of types of transfusion modalities. However, the correlation was highly significant between the pre-operative haemoglobin (Hb) level and postoperative fever ($P < 0.01$) and VOC ($P < 0.01$). Interestingly, SCD patients who received hydroxyurea had less postoperative complications such as fever ($P < 0.05$) and vaso-occlusive crises ($P < 0.05$), while those who received prophylactic heparin in the postoperative period had a reduced length of hospital stay ($P < 0.01$) and vaso-occlusive crises ($P < 0.01$). *Conclusions:* The guidelines for preoperative transfusion in SCD patients were effective in reducing the postoperative morbidity and mortality. Moreover, this guideline emphasises the surgical situations where preoperative transfusion is needed and optimum regimen is required for different surgical sub-types.

DEMANDS AND CHALLENGES FOR PATIENTS WITH SICKLE CELL DISEASE REQUIRING HAEMATOPOIETIC STEM CELL TRANSPLANTATION IN SAUDI ARABIA

Abdulrahman Alsultan[1,2], Wasil Jastaniah[3,4], Sameera Al Afghani[5], Muneer H. Al Bagshi[6], Zaki Nasserullah[7], Ahmed M. Al-Suliman[8], Mohammed K. Alabdulaali[8]

[1]Department of Pediatrics and Sickle Cell Disease Research Center, College of Medicine, King Saud University, Riyadh, Saudi Arabia; [2]Department of Pediatric Hematology/Oncology, King Abdullah Specialist Children's Hospital, Riyadh, Saudi Arabia; [3]King Abdulaziz Medical City, Jeddah, Saudi Arabia; [4]Department of Pediatrics, College of Medicine, Umm alQura University, Makkah; [5]King Fahad Specialist Hospital, Dammam, Saudi Arabia; [6]Maternity and Children's Hospital, Hofuf, Saudi Arabia; [7]Maternity & Children's Hospital, Dammam, Saudi Arabia; [8]King Fahad Hospital, Hofuf, Saudi Arabia

Background: Allogeneic hematopoietic stem cell transplantation (HSCT) is the only curative treatment for sickle cell disease (SCD). *Methods:* In this study, we estimated the number of Saudi SCD patients who are candidates for HSCT. We used presence of overt stroke, recurrent acute chest syndrome (ACS), and frequent severe pain crisis as indications for HSCT. *Results:* We enrolled 376 SCD patients. A total of 250 patients were from the Southwestern (SW) provinces; the mean age of this group was 20 years old (range, 4-56 years old) and 48% of these subjects were female. The total number of potential transplant candidates from the SW was 59 (23.6%). Among individuals from the Eastern (E) province, 126 participated in the study. These subjects had a mean age of 25 years old (range, 4-55 years old) and 58% were females. There were a total of 22 (17.4%) transplant candidates from E province. Based on the reported preva-

lence of SCD in Saudi Arabia and the size of the Saudi population in 2010, it is estimated that nearly 61,000 SCD patients reside in Saudi Arabia. Of these, approximately 18,000 are children under 14 years of age. The projected number of Saudi SCD patients who are candidates for HSCT is 10,536 patients. Of those, 2,148 are children. *Conclusions:* The burden of SCD on HSCT centers in Saudi Arabia is substantial and is difficult currently to meet the demand. We recommend recruiting/training more transplant physicians and nurses, expand current capacity of centers if feasible, and open new transplant centers to make HSCT a practical therapeutic option for patients with severe SCD in Saudi Arabia.

HYDROXYUREA, AN EFFECTIVE THERAPY TO AVOID SPLENECTOMY IN SICKLE CELL DISEASE AND THALASSAEMIA: SINGLE INSTITUTE EXPERIENCE IN SAUDI ARABIA

Soad Khalil Al Jaouni

Hematology Department, King Abdulaziz University Hospital, Faculty of Medicine, King Abdulaziz University, Jeddah, Saudi Arabia

Background and Purpose: Spleen plays an important part in the immune system which helps the body fight infection and have a major role in children. Splenectomy have been decline in hemoglobin disorders in recent years. To assess the role of hydroxyurea if can eliminate the risk of splenectomy in sickle cell disease (SCD) and thalassemia in our medical center. *Methods:* Total of 54 patients enrolled in the study between 2004 to 2016 at King Abdulaziz University Hospital, Faculty of Medicine, King Abdulaziz University, Jeddah, Kingdom of Saudi Arabia, 25 sickle cell disease (SCD), 17 thalassemia major (TM) were suboptimal transfusion dependents and hypersplenism, 12 thalassemia inter media (TI). *Results:* Risk of splenectomy have been avoided in 24/25 SCD, 15/17 in TM and 12/12 in TI. Recent studies and meta analysis showed that hydroxyurea is safe and non- carcinogenic. *Conclusions:* Hydroxyurea is an effective and safe therapy can eliminate risk of splenectomy in SCD and thalassemia. *Keywords:* Hydroxyurea, SCD, Thalassemia, splenectomy.

MY PERSONAL JOURNEY THROUGH HYDROXYUREA THERAPY IN SICKLE CELL DISEASE DURING PREGNANCY AND LACTATION

Aishath Shifneez Shakir

Maldivian Thalassaemia Society, Maldives

Hydroxyurea (HU), a drug that is known to reduce the frequency of sickle crisis episodes and hospitalizations in SCD patients. However, it is believed that if HU is used during pregnancy or if the patient becomes pregnant while taking this drug, the patient should be told of the potential harm to the fetus as animal testing shows that the drug crosses to the placenta and causes embryo toxicity, fetal malformations, growth retardation. Therefore, patients are advised to avoid becoming pregnant while taking HU. This leaves pregnant sickle women with a real dilemma: Take a drug with unknown effects on fetal development or give up a helpful, necessary medication. I am Shifneez, 33 yrs old from Maldives. As a sickle beta-thalassaemia patient, I dedicate my efforts to create sickle cell awareness throughout my country. Past 11 years, I have had 3 pregnancies each experience different with regard to HU therapy. Today I am a mother of 2. I had the courage to go through my last pregnancy while taking HU. I believe that my daughter is the hope everyone is looking for. Many women are in fear, when it comes to pregnancy and raising a family, since we are reminded of how risky it is to become a mother. In my opinion, doctors who are unaware that HU can actually improve the health of sickle cell patients during pregnancy is taking a huge risk by stopping it since it would mean the recurrence of sickle complications that could be life threatening for mother and baby. It's really important that doctors find it in themselves to empower patients and encourage them to be the best they can be. I hope the story of my personal journey can open up the possibility for others like me to have their dreams come true.