haematologica
Journal of the Ferrata Storti Foundation

Clonal hematopoiesis in long-term survivors after allogeneic hematopoietic cell transplantation: associations with active chronic graft-*versus*-host disease, donor age, and transplantation for chronic myeloid leukemia

by Takashi Toya, Daichi Sadato, Chizuko Hirama, Kana Kato, Kaori Kondo, Yasutaka Sadaga, Atsushi Jinguji, Kaoru Morita, Hiroaki Shimizu, Yuho Najima, Tomomi Toubai, Kyoko Haraguchi, Hironori Harada, Yuka Harada and Noriko Doki

Received: June 2, 2026.
Accepted: November 3, 2026.

Citation: Takashi Toya, Daichi Sadato, Chizuko Hirama, Kana Kato, Kaori Kondo, Yasutaka Sadaga, Atsushi Jinguji, Kaoru Morita, Hiroaki Shimizu, Yuho Najima, Tomomi Toubai, Kyoko Haraguchi, Hironori Harada, Yuka Harada and Noriko Doki. Clonal hematopoiesis in long-term survivors after allogeneic hematopoietic cell transplantation: associations with active chronic graft-versus-host disease, donor age, and transplantation for chronic myeloid leukemia.
Haematologica. 2026 Nov 12. doi: 10.3324/haematol.2026.301340 [Epub ahead of print]

Publisher's Disclaimer.
E-publishing ahead of print is increasingly important for the rapid dissemination of science. Haematologica is, therefore, E-publishing PDF files of an early version of manuscripts that have completed a regular peer review and have been accepted for publication.
E-publishing of this PDF file has been approved by the authors.
After having E-published Ahead of Print, manuscripts will then undergo technical and English editing, typesetting, proof correction and be presented for the authors' final approval, the final version of the manuscript will then appear in a regular issue of the journal.
All legal disclaimers that apply to the journal also pertain to this production process.

Clonal hematopoiesis in long-term survivors after allogeneic hematopoietic cell transplantation: associations with active chronic graft-*versus*-host disease, donor age, and transplantation for chronic myeloid leukemia

Takashi Toya[1,2,*], Daichi Sadato[3,*], Chizuko Hirama[3], Kana Kato[1], Kaori Kondo[1], Yasutaka Sadaga[1], Atsushi Jinguji[1], Kaoru Morita[1], Hiroaki Shimizu[1], Yuho Najima[1], Tomomi Toubai[1,3], Kyoko Haraguchi[4], Hironori Harada[1,5], Yuka Harada[3], Noriko Doki[1]

[1]Hematology Division, Tokyo Metropolitan Cancer and Infectious Diseases Center, Komagome Hospital, Tokyo, Japan.

[2]Department of Hematology and Oncology, Dokkyo Medical University, Tochigi, Japan.

[3]Clinical Research and Trials Center, Tokyo Metropolitan Cancer and Infectious Diseases Center, Komagome Hospital, Tokyo, Japan.

[4]Division of Transfusion and Cell Therapy, Tokyo Metropolitan Cancer and Infectious Diseases Center, Komagome Hospital, Tokyo, Japan.

[5]Laboratory of Oncology, School of Life Sciences, Tokyo University of Pharmacy & Life Sciences, Tokyo, Japan

* These authors contributed equally to this work.

Author Contributions

T.T., D.S., H.H. and Y.H. designed the study. T.T., K.K., Y.S., C.K., M.S., A.J., H.S., Y.N., and N.D. provided medical treatment and gave informed consent to each patient. T.T., K.H., and Y.O. collected and managed clinical samples. D.S., C.H., and Y.H. underwent the sequencing analyses. T.T. and D.S. wrote the manuscript, underwent statistical analyses, and made figures and tables. H.H., Y.H., and N.D. supervised the study. All authors read and approved the manuscript.

Running heads: CH-associated features in long-term survivors after allo-HCT

Corresponding author: Takashi Toya, MD, PhD

Hematology Division, Tokyo Metropolitan Cancer and Infectious Diseases Center, Komagome Hospital. 3-18-22 Hon-komagome, Bunkyo-ku, Tokyo, Japan, 1138677

Phone: +81-3-3823-2101, Fax: +81-3-3823-5433

Email: tooya-tky@umin.ac.jp

Data availability statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Funding

This research was supported in part by JSPS KAKENHI Grant Number JP19K17848, Friends of Leukemia Research Fund, and Clinical Research Fund of the Tokyo Metropolitan Government.

Competing interests

The authors declare no conflict of interest.

Keywords: clonal hematopoiesis, allogeneic hematopoietic cell transplantation, chronic myeloid leukemia, chronic graft-versus-host disease

Acknowledgments

The authors would like to thank Editage (http://www.editage.com) for editing and reviewing this manuscript for English language.

Artificial Intelligence

During revision, the authors used ChatGPT (GPT-6-based Codex; OpenAI; a commercial proprietary artificial intelligence [AI] tool) to improve the English grammar, clarity, and readability of parts of the main text. The tool was instructed to refine the language while preserving the original scientific meaning. All AI-assisted edits were reviewed and, where necessary, modified by the authors, who take full responsibility for the accuracy, integrity, and originality of the final manuscript.

Word count**:** abstract 215/250 words, manuscript 3,284/4,000 words (method 605 words), tables: 3, figures: 3/8, supplemental tables: 8, supplemental figures: 3, references 50/50

Abstract

Clonal hematopoiesis (CH) in long-term survivors of allogeneic hematopoietic cell transplant (HCT) remains largely understudied. Using targeted sequencing, we analyzed peripheral blood samples from 101 patients who survived without relapse for more than 10 years after HCT. The median interval from HCT to sample collection was 13.8 years (range, 10.0-29.8 years). Thirty-six CHs with ≥2% variant allele frequency were detected in 29 patients (28.7%). Complete blood count did not differ according to CH status. Multivariable analysis showed donor age at sampling (odds ratio [OR] 1.06, 95% confidence interval [CI] 1.00-1.12), patients who underwent HCT for chronic myeloid leukemia (CML; OR 7.13, 95% CI 1.38-36.8), and moderate/severe chronic graft-versus-host disease at sampling (OR 5.49, 95% CI 1.05-28.7) were significantly associated with CH. Donor age at sampling was significantly higher in patients with pathogenic CH than in those with variants of unknown significance or those without CH (median 56.5 vs 48.0 vs 47.0 years old, p=0.0077). After a median follow-up of 60.0 months after sampling, one patient with CML experienced relapse. Three-year Overall survival (94.2% vs. 92.9%, p=0.800) and cumulative incidence of second cancer (18.0% vs. 14.9%, p=0.410) after CH sampling were not associated with CH status. In conclusion, CH in HCT survivors is

common and associated with donor age, transplantation for CML, and chronic graft-versus-host disease.

Introduction

Clonal hematopoiesis (CH) is characterized by genetic and/or chromosomal alterations in hematopoietic cells in individuals apparently without hematological diseases but can be associated with the subsequent development of variable hematological and non-hematological diseases.[1-5] Some studies have reported frequent CH after allogeneic hematopoietic cell transplantation (HCT).[6-9] However, the risk factors for CH in long-term HCT survivors are unclear, and their clinical impact has not been fully elucidated. Considering the occurrence of CH-associated diseases[3, 10-15] and late complications of HCT,[16, 17] some of these adverse events may be influenced by CH; however, sufficient data are lacking.

Aging is the most important trigger for CH. However, other intrinsic and extrinsic factors, such as chronic inflammation, immune reactions, and genotoxic stresses can also provoke CH,[18] and HCT recipients are exposed to various inflammatory and/or genotoxic stresses. In HCT, various factors such as recipient-derived microenvironment, donor-derived hematopoietic cells, and adaptive immunity and inflammation are important for hematopoietic stem cell integrity.[19] However, their contribution to CH is unclear, and the interplay between age-related CH and transplant outcomes, including

graft-versus-host disease (GVHD) and non-relapse mortality (NRM), is controversial.[7, 20-25]

In this study, we analyzed 101 patients who survived without relapse for >10 years after HCT using targeted sequencing and showed that CH was common in long-term survivors after HCT. Variable clinical factors such as older donors, transplantation for chronic myeloid leukemia (CML), and moderate-to-severe chronic GVHD at sampling were significantly associated with the existence of CH in long-term HCT survivors, but CH did not significantly affect later outcomes.

Methods

Patients

Patients who survived without relapse for >10 years after HCT were included in this study. Patients who had developed secondary hematological malignancies by the time of CH sampling were excluded because they received additional treatment, including subsequent HCT, precluding appropriate assessment of long-term post-transplant CH. Patients receiving maintenance anticancer therapy after HCT were also excluded. The detailed HCT procedures have been described previously.[26] None of the patients received posttransplant cyclophosphamide (PTCY)-based graft-versus-host disease (GVHD)

prophylaxis. When analyzing the association between donor age at sampling, used here as a proxy for the biological age of donor-derived hematopoietic cells, and other characteristics including CH, cord blood (CB) donors were assigned a donor age of 0 years at HCT. Donor age at sampling was then calculated as donor age at HCT plus the elapsed time from HCT to sample collection. Chronic GVHD status at the time of sampling was classified according to the National Institutes of Health (NIH) criteria,[27] but a history of chronic GVHD was classified based on the Seattle criteria[28] because most patients had been diagnosed before the NIH criteria era. The activity of cGVHD at the time of sampling was assessed using the following criteria, and cGVHD was classified as active only when both of the following criteria were met: (1) ongoing cGVHD-related manifestations documented according to the NIH criteria by multiple transplant physicians and specialist nurses at the long-term follow-up clinic; and (2) assessment of the disease as clinically active by the treating physician, with continued systemic immunosuppressive therapy.

This study was performed in accordance with the Declaration of Helsinki and approved by the Ethics Committee of Tokyo Metropolitan Komagome Hospital (approval number 2220). Written informed consent was obtained from all the patients.

DNA extraction and sequencing analysis

DNA was extracted from peripheral blood using a QIAamp DNA mini kit (Qiagen, Hilden, Germany). Targeted sequencing of 68 myeloid malignancy-associated genes was performed using AmpliSeq for the Illumina Myeloid Panel and On-demand Panel using a MiniSeq system (Illumina, San Diego, CA), as previously described (**Supplemental Table 1**).[9] A variant allele frequency (VAF) cutoff was set at 0.02 according to our validated method,[29] and 0.01 was also used to evaluate the robustness of the results. Detailed descriptions of the analytical pipeline, including error correction, noise reduction, and threshold settings, are provided in the Supplementary information. Detected variants were classified as pathogenic/likely pathogenic or variants of unknown significance (VUS), according to an expert panel.[30] In subsequent analyses, pathogenic and likely pathogenic CHs were grouped together and collectively referred to as pathogenic CH, in contrast to VUS. Copy number variants (CNVs) were evaluated using targeted sequencing data and CNVkit as previously reported.[31] Patient BM samples obtained 28 days after HCT were analyzed using the same protocol.[9]

Statistical analysis

Differences in numerical variables were compared using the Wilcoxon rank-sum test or the Kruskal-Wallis rank sum test, and those in categorical variables were evaluated using Fisher's exact test. Factors with $p < 0.1$ in univariable analyses were included in the multivariable analysis, which was performed using logistic regression analysis. Given the strong collinearity between ages at HCT and at sampling, as well as the variable interval from HCT to sample collection among patients, we used recipient and donor ages at sampling, rather than their ages at HCT, to analyze their associations with CH. Overall survival was estimated using the Kaplan–Meier method and compared using the log-rank test. The cumulative incidence of secondary cancer was evaluated using the Gray method, considering relapse and death without secondary cancer as the competing risks. The level of statistical significance was set at $p < 0.05$. Statistical analyses were performed using R version 4.4.2.

Results

Patient characteristics

One hundred and one patients who underwent allogeneic HCT between 1990 and 2011 were included in this study. The interval from HCT to peripheral blood sampling was 13.8 years (range, 10.0–29.8). The median patient ages at HCT and sampling were 38

(interquartile range, 31–44) and 52 (interquartile range, 45–60) years, respectively, and the median donor ages at HCT and sampling were 35 (interquartile range, 27–43) and 49 (interquartile range, 43–56) years, respectively. Among patients who underwent HCT from related donors, recipient and donor ages were strongly positively correlated both at HCT (Pearson's r=0.84, 95% confidence interval [CI], 0.70–0.91; p<0.001) and at sampling (r=0.81, 95% CI, 0.65–0.90; p<0.001). In contrast, among patients who underwent HCT from unrelated donors, no statistically significant correlations were observed either at HCT (r=−0.22, 95% CI, −0.44 to 0.02; p=0.075) or at sampling (r=−0.17, 95% CI, −0.39 to 0.08; p=0.182) (**Supplemental Figure 1**). Detailed patient characteristics are shown in **Supplemental Table 2**. Seventy-one patients had a history of cGVHD during the clinical course, and 19 patients had active cGVHD (mild, n=7; moderate, n=10; severe, n=2) at the time of sampling with involvement of the eyes (n=12), oral cavity (n=9), skin (n=6), and lungs (n=3). By definition, none of the patients had experienced molecular, cytogenetic, or hematological relapse after HCT at sampling, and no patients received chemotherapeutic agents, including molecular targeted agents after HCT, other than methotrexate for GVHD prophylaxis.

Prevalence of CH

The mean coverage depth of the targeted sequencing was 2,343 (range: 1,005–3,184). Among 101 patients, 36 CHs with ≥2% VAF were detected in 29 patients (28.7%, **Figure 1A** and **Supplemental Table 3**). Among the 29 patients with CH, 7 had two CH and 22 had one CH (**Figure 1B**), and *DNMT3A* mutations were the most common (10/36=27.8%, **Figure 1C**). The median VAF of the detected gene mutations was 4.45% (range: 2.06–38.01%, **Figure 1D**). Among the 36 CHs, 21 (58.3%) were considered pathogenic/likely pathogenic, while the others were considered VUS (**Figure 1E**); thus, 19 patients had pathogenic CH (18.8%). No CH suggestive of germline origin were identified in this cohort and mixed chimera was not evident in patients with CH (data not shown) [9].

CH and associated clinical factors

The associations between CH and clinical characteristics are shown in **Table 1 and Supplemental Table 4**. The complete blood count did not differ according to the presence of CH. The ages of recipients/donors at HCT were significantly higher in patients with CH (p=0.036 in recipients and p=0.041 in donors) and this difference remained significant at sample collection (median 54.8 vs 48.9 years old, p=0.019 in recipients; median 52.2 vs 47.7 years old, p=0.028 in donors).

When patients were classified into three groups–those with pathogenic CH, those with VUS, and those without CH–donor age at sampling differed significantly among the three groups, with patients with pathogenic CH being significantly older than those with VUS and without CH (median 56.5 vs 48.0 vs 47.0 years old, p=0.008); however, those with VUS and without CH did not differ significantly (p=0.930, **Figure 2A**). Likewise, patient age at sampling was also significantly different among the three groups (median 56.0 vs 57.0 vs 49.0 years old, p=0.031). Although patients with pathogenic CH were significantly older than those without CH (p=0.010), age did not differ significantly between pathogenic CH and VUS or between VUS and no CH (p=0.670 and p=0.260, **Figure 2B**).

Regarding the primary hematological disorder, CH and pathogenic CH were significantly more common in patients who underwent transplantation for CML than in others (CH: 62.5% vs. 22.4%, p=0.002; pathogenic CH: 37.5% vs. 14.1%, p=0.036, **Figure 2C**).

In the univariable analyses, CH was significantly or tended to be more common in patients who underwent HCT for AML (p=0.036) and CML (p=0.002), and in patients with moderate/severe cGVHD at sampling (p=0.098). In contrast, CH was less common

in patients who underwent ABO-matched HCT (p=0.033) and ABO-major-mismatched HCT (p=0.085) compared with the others.

In multivariable analysis, donor age at sampling (hazard ratio [OR] 1.06, 95% CI 1.00–1.12, p=0.032), patients who underwent HCT for CML (OR 7.13, 95% CI 1.38-36.8, p=0.019), and moderate/severe cGVHD at sample collection (OR 5.49, 95% CI 1.05–28.7, p=0.044) were significantly associated with the existence of CH (**Table 2**). Interestingly, all three patients with active pulmonary cGVHD harbored *DNMT3A*-mutated pathogenic CH. In addition, given the strong correlation between donor and recipient ages in the related-donor group, we performed an additional multivariable analysis excluding recipient age to assess the robustness of our findings. Donor age at sampling remained a significant factor in this sensitivity analysis (OR 1.07, 95% CI 1.01–1.13, p=0.012), supporting the robustness of its independent association with CH.

Pathogenic CH and associated clinical factors

We also evaluated the association between pathogenic CH and clinical factors excluding VUS. The association between pathogenic CH and the clinical characteristics was similar to that observed in CH (**Supplemental Table 5**). In multivariable analysis, donor age at sampling (OR 1.07, 95% CI 1.01–1.13, p=0.023), patients who underwent HCT for CML

(OR 4.18, 95% CI 1.00–17.4, p=0.049), ABO-matched HCT (OR 4.99, 95% CI 1.26–19.8, p=0.022), and moderate/severe cGVHD at sample collection (OR 6.92, 95% CI 1.43–33.5, p=0.016) were significantly associated with the existence of pathogenic CH (**Supplemental Table 6**).

CH and/or CNVs

Next, CH, including CNVs, were analyzed. CH and/or CNVs were detected in 32 patients (31.7%) (**Supplemental Table 7**). Although moderate/severe cGVHD at sampling was not associated with pathogenic CH in the univariable analysis (p=0.190), this factor was included in the multivariable analysis because it was a significant risk factor for CH in the previous section. In multivariable analysis, donor age at sampling (OR 1.09, 95% CI 1.03–1.17, p=0.016), patients who underwent HCT for CML (OR 10.7, 95% CI 2.22-51.7, p=0.0094), ABO-matched HCT (OR 4.47, 95% CI 1.48–13.5, p=0.0080), HLA-matched HCT (OR 5.10, 95% CI 1.24–21.0, p=0.024), and moderate/severe cGVHD at sample collection (OR 5.39, 95% CI 1.11–26.2, p=0.037) were significantly associated with the existence of CH and/or CNVs (**Supplemental Table 7**).

CHs with ≥1% VAF and associated clinical factors

Because some previous studies adopted a VAF cut-off value other than 2%,[32, 33] we also evaluated the association between clinical characteristics and CH using a 1% cut-off to further evaluate the robustness of our results.

When a 1% cutoff was adopted, CH and/or CNVs were detected in 34 of 101 long-term HCT survivors (33.7%; **Supplemental Table 3 and 8**). Multivariable analysis revealed that donor age at sampling (OR 1.06, 95% CI 1.00–1.12, p=0.043), patients who underwent HCT for CML (OR 5.06, 95% CI 1.41–18.1, p=0.013), HLA matched HCT (OR 4.94, 95% CI 1.06–22.9, p=0.041), ABO matched HCT (OR 4.15, 95% CI 1.39–12.4, p=0.011), and moderate/severe cGVHD at sample collection (OR 9.19, 95% CI 1.75–48.3, p=0.0088) as significant risk factors for the existence of CH and/or CNV (**Supplemental Table 8**).

Clinical follow-up after sampling

The clinical course of the patients after sampling was retrospectively evaluated to characterize the clinical impact of CH on long-term survivors of HCT. The median follow-up period for survivors after sampling was 60.0 months (range, 8.2–67.2). Only one patient with CML experienced relapse of the primary hematological malignancy, who had no CH at sampling, 55.1 months after sampling and 310.2 months after HCT.

Chimerism analysis and major *bcr::abl1* positivity indicated that CML was a true relapse of the primary disease rather than a donor-cell leukemia. During the follow-up period, nine patients died, and 15 patients developed a second cancer (**Table 3**). Five-year cumulative incidence of secondary cancer was 16.2%, which was comparable between the patients with and without CH (18.0% vs. 14.9%, p=0.410, **Figure 3A**). Overall survival (OS) after sampling did not differ according to the presence of CH (3-year OS: 94.2% vs. 92.9%, p=0.800; **Figure 3B**). Active moderate or severe cGVHD at sampling was significantly associated with poor OS after sampling (3-year OS, 66.7% vs. 97.6%; p<0.001; **Figure 3C**).

CH status 28 days after HCT

Next, we evaluated whether CHs emerged more than a decade after HCT or were already present during the peri-transplantation period. For 12 patients with 17 CHs more than 10 years after HCT, targeted sequencing was performed on DNA from cryopreserved BM samples collected 28 days after HCT. All patients achieved neutrophil engraftment within 28 days post-HCT. Of the 17 CH mutations identified in 12 patients at the 10-year sampling, four (23.5%), each in a different patient, were also detected in the

corresponding day-28 samples, whereas the remaining 13 (76.5%) were undetectable at day 28 (**Supplemental Figure 2**).

The 12 patients were classified into two groups based on day-28 CH status: eight patients without CH (Pt. 1–8) and four with CH (Pt. 9–12). The CH status in the day-28 sample was generally not associated with complete blood counts more than 10 years after HCT. The only significant difference in blood cell indices was that the red cell distribution width (RDW) in the >10 years sample was higher in patients with CH in the day-28 sample compared in the other (median 14.4 vs 13.1%, p=0.004, **Supplemental Figure 3A**). Neutrophil and platelet engraftment did not differ according to the day-28 CH status (**Supplemental Figure 3B, C**).

Discussion

In this study, we demonstrated that CH is common among long-term survivors of HCT. Boettcher et al. analyzed 42 long-term survivors of HCT and found that 31% had CH.[6] This prevalence was comparable to that observed in our study, despite the use of a 1% VAF cutoff. However, in age-matched healthy individuals, the frequency of CH has been reported to be < 5%,[1, 2] underscoring the high prevalence of CH among long-term HCT survivors. Older donor age, rather than older patient age, was a risk factor for CH in

multivariable analyses. Some clinical studies have indicated a worse prognosis of HCT in elderly donors[34, 35] and frequent CH might be a contributing factor, although the exact mechanisms are unclear.

In our study, CH was prevalent in the patients with moderate/severe cGVHD. Interestingly, active cGVHD at the time of sample collection, rather than a history of cGVHD, was significantly associated with CH over 10 years post-HCT. This may be a reason why the correlation between donor CH and cGVHD was controversial in the previous reports.[23, 24] As Hormaechea-Agulla et al. showed that chronic infection could drive Dnmt3a-loss-of-function CH in murine models,[36] chronic inflammation is considered an important factor in CH in humans. In contrast, transient and/or conditional CHs[37] might also affect the inflammatory status, including cGVHD. This cross-sectional study could not evaluate chronological CH status, and prospective studies are warranted to provide insights that may help clarify causal relationships.

Interestingly, in this study, patients who underwent HCT for CML were at a higher risk of developing CH, as well as pathogenic CH compared with others. Although the exact mechanisms are unclear, altered BM niche in patients with CML[38-40] might encourage CH after HCT. Reported cases with developing Ph-negative CH in molecular/cytogenetic remission in patients with CML[41, 42] also might support these

insights. Tyrosine kinase inhibitors (TKI) were also suspected of causing this phenomenon in the previous studies,[41-43] but none of the patients in this study were administered TKI after HCT, suggesting that the frequent CH was not due to TKI. Although long-term mixed chimerism has occasionally been reported in long-term survivors after allogeneic HCT for CML,[44] all CH-positive patients in our cohort showed donor-type chimerism, arguing against a recipient origin of the detected mutations.

Our study had several limitations. First, this was a retrospective, single-center study with a limited sample size, potentially increasing the risk of model overfitting and imprecise estimates. Our findings should be considered exploratory and require validation in larger cohorts, given the relatively low number of events per variable in our multivariable analyses. Nevertheless, donor age at sampling, HCT for CML, and active moderate-to-severe cGVHD at sampling were consistently identified as significant factors across models using different definitions of CH. In addition, some transplantation procedures were inevitably different from the current procedures. Gibson et al. reported that HCT from a donor with *DNMT3A*-CH was associated with an increased incidence of cGVHD only when non-PTCY-based GVHD prophylaxis was adopted.[22] None of the patients in this study underwent PTCY because this procedure has not been introduced in Japan. The infusion of cytotoxic agents after stem cell infusion can induce genotoxicity

and evoke CH, although Majzner et al. reported that PTCY does not increase the risk of donor-cell leukemia.[45] In addition, because historical cGVHD diagnoses were based on pre-NIH criteria in our study, some cases of late acute GVHD may have been misclassified as cGVHD,[46] potentially affecting analyses based on a history of cGVHD. Secondly, this study did not address whether different types of CH exert distinct effects on clinical outcomes. Buscarlet et al. reported that *DNMT3A* and *TET2* CH have different phenotypes;[47] further studies with more patients are warranted. Third, a survivorship bias may have influenced the results. The effects of CH on secondary cancer, cardiovascular disease, and other critical illnesses, conditions that have been reported to be associated with CH,[48] could not be evaluated in this study because it only evaluated long-term survivors and therefore clinical consequences of CH may not be generalizable to the broader allogeneic HCT population. In addition, because only patients who achieved engraftment were included, selection bias may have led to underestimation of the adverse impact of day-28 CH on engraftment. Fourth, because this was a cross-sectional study, we could not assess longitudinal changes in CH status or evaluate patients who subsequently developed secondary hematologic malignancies. Although temporal changes in CH clone size have been reported previously,[49] prospective longitudinal studies are warranted to better characterize the diverse trajectories of CH clones in long-

term survivors after HCT, including clonal dynamics associated with the subsequent development of donor-cell leukemia. Fifth, our clinical criteria for active cGVHD may not completely distinguish active cGVHD-related inflammation from persistent, irreversible organ damage, particularly in patients with sclerotic cutaneous cGVHD or bronchiolitis obliterans syndrome.[50] Nevertheless, the association of CH with persistent cGVHD-related manifestations, but not with a history of cGVHD, suggests a potential biological link between these clinical features and CH. Prospective studies incorporating objective and reliable biomarkers of cGVHD activity are needed to validate our findings.

In conclusion, the prevalence of CH in long-term survivors after HCT was especially high despite the relatively young age of the donors and recipients, particularly among patients who underwent HCT for CML and from older donors. CH was associated with chronic GVHD status at sampling rather than a history of chronic GVHD.

References

1. Jaiswal S, Fontanillas P, Flannick J, et al. Age-related clonal hematopoiesis associated with adverse outcomes. N Engl J Med. 2014;371(26):2488-2498.
2. Genovese G, Kahler AK, Handsaker RE, et al. Clonal hematopoiesis and blood-cancer risk inferred from blood DNA sequence. N Engl J Med. 2014;371(26):2477-2487.
3. Jaiswal S, Natarajan P, Silver AJ, et al. Clonal hematopoiesis and risk of atherosclerotic cardiovascular disease. N Engl J Med. 2017;377(2):111-121.
4. Miller PG, Qiao D, Rojas-Quintero J, et al. Association of clonal hematopoiesis with chronic obstructive pulmonary disease. Blood. 2022;139(3):357-368.
5. Hecker JS, Hartmann L, Riviere J, et al. CHIP and hips: clonal hematopoiesis is common in patients undergoing hip arthroplasty and is associated with autoimmune disease. Blood. 2021;138(18):1727-1732.
6. Boettcher S, Wilk CM, Singer J, et al. Clonal hematopoiesis in donors and long-term survivors of related allogeneic hematopoietic stem cell transplantation. Blood. 2020;135(18):1548-1559.
7. Gillis N, Padron E, Wang T, et al. Pilot study of donor-engrafted clonal hematopoiesis evolution and clinical outcomes in allogeneic hematopoietic cell transplantation recipients using a National Registry. Transplant Cell Ther. 2023;29(10):640.e1-640.08.
8. Wong WH, Bhatt S, Trinkaus K, et al. Engraftment of rare, pathogenic donor hematopoietic mutations in unrelated hematopoietic stem cell transplantation. Sci Transl Med. 2020;12(526):eaax6249.
9. Adachi H, Sadato D, Toya T, et al. Donor-derived gene mutations in sex chromosome loss after stem cell transplantation. Br J Haematol. 2021;195(3):e142-e146.
10. Winter S, Gotze KS, Hecker JS, et al. Clonal hematopoiesis and its impact on the aging osteo-hematopoietic niche. Leukemia. 2024;38(5):936-946.
11. Vlasschaert C, Robinson-Cohen C, Chen J, et al. Clonal hematopoiesis of indeterminate potential is associated with acute kidney injury. Nat Med. 2024;30(3):810-817.
12. Cumbo C, Tarantini F, Zagaria A, et al. Clonal hematopoiesis at the crossroads of inflammatory bowel diseases and hematological malignancies: a biological link? Front Oncol. 2022;12:873896.

13. Singh J, Li N, Ashrafi E, et al. Clonal hematopoiesis of indeterminate potential as a prognostic factor: a systematic review and meta-analysis. Blood Adv. 2024;8(14):3771-3784.
14. Wang H, Divaris K, Pan B, et al. Clonal hematopoiesis driven by mutated DNMT3A promotes inflammatory bone loss. Cell. 2024;187(14):3690-3711.e19.
15. Kishtagari A, Corty RW, Visconte V. Clonal hematopoiesis and autoimmunity. Semin Hematol. 2024;61(1):3-8.
16. Nieder ML, McDonald GB, Kida A, et al. National Cancer Institute-National Heart, Lung and Blood Institute/pediatric Blood and Marrow Transplant Consortium First International Consensus Conference on late effects after pediatric hematopoietic cell transplantation: long-term organ damage and dysfunction. Biol Blood Marrow Transplant. 2011;17(11):1573-1584.
17. Rachner TD, Link-Rachner CS, Bornhauser M, Hofbauer LC. Skeletal health in patients following allogeneic hematopoietic cell transplantation. Bone. 2022;158:115684.
18. SanMiguel JM, Young K, Trowbridge JJ. Hand in hand: intrinsic and extrinsic drivers of aging and clonal hematopoiesis. Exp Hematol. 2020;91:1-9.
19. Prabahran A, Koldej R, Chee L, Ritchie D. Clinical features, pathophysiology, and therapy of poor graft function post-allogeneic stem cell transplantation. Blood Adv. 2022;6(6):1947-1959.
20. Frick M, Chan W, Arends CM, et al. Role of donor clonal hematopoiesis in allogeneic hematopoietic stem-cell transplantation. J Clin Oncol. 2019;37(5):375-385.
21. Nawas MT, Schetelig J, Damm F, et al. The clinical implications of clonal hematopoiesis in hematopoietic cell transplantation. Blood Rev. 2021;46:100744.
22. Gibson CJ, Kim HT, Zhao L, et al. Donor clonal hematopoiesis and recipient outcomes after transplantation. J Clin Oncol. 2022;40(2):189-201.
23. Newell LF, Williams T, Liu J, et al. Engrafted donor-derived clonal hematopoiesis after allogenic hematopoietic cell transplantation is associated with chronic graft-versus-host disease requiring immunosuppressive therapy, but no adverse impact on overall survival or relapse. Transplant Cell Ther. 2021;27(8):662.e1-662.e9.
24. Kim KH, Kim T, Novitzky-Basso I, et al. Clonal hematopoiesis in the donor does not adversely affect long-term outcomes following allogeneic hematopoietic stem cell transplantation: result from a 13-year follow-up. Haematologica. 2023;108(7):1817-1826.

25. Xie Y, Kazakova V, Weeks LD, et al. Effects of donor-engrafted clonal hematopoiesis in allogeneic and autologous stem cell transplantation: a systematic review and meta-analysis. Bone Marrow Transplant. 2024;59(11):1585-1593.
26. Najima Y, Sadato D, Harada Y, et al. Prognostic impact of TP53 mutation, monosomal karyotype, and prior myeloid disorder in nonremission acute myeloid leukemia at allo-HSCT. Bone Marrow Transplant. 2021;56(2):334-346.
27. Jagasia MH, Greinix HT, Arora M, et al. National Institutes of Health Consensus Development Project on Criteria for Clinical trials in chronic graft-versus-host disease: I. The 2014 Diagnosis and Staging Working Group report. Biol Blood Marrow Transplant. 2015;21(3):389-401.e1.
28. Shulman HM, Sullivan KM, Weiden PL, et al. Chronic graft-versus-host syndrome in man. A long-term clinicopathologic study of 20 Seattle patients. Am J Med. 1980;69(2):204-217.
29. Sadato D, Hirama C, Kaiho-Soma A, et al. Archival bone marrow smears are useful in targeted next-generation sequencing for diagnosing myeloid neoplasms. PLoS One. 2021;16(7):e0255257.
30. Kambara Y, Sadato D, Toya T, et al. Recurrent DDX41 mutation in very late relapse after allogeneic stem cell transplantation. Leukemia. 2024;38(3):667-670.
31. Talevich E, Shain AH, Botton T, Bastian BC. CNVkit: genome-wide copy number detection and visualization from targeted DNA sequencing. PLoS Comput Biol. 2016;12(4):e1004873.
32. Kapadia CD, Rosas G, Thakkar SG, et al. Incipient clonal hematopoiesis is accelerated following CD30.CAR-T therapy. Cytotherapy. 2024;26(3):261-265.
33. Panagiota V, Kerschbaum JF, Penack O, et al. Clinical Implications and Dynamics of Clonal Hematopoiesis in Anti-CD19 CAR T-cell Treated Patients. Hemasphere. 2023;7(10):e957.
34. Seo S, Usui Y, Matsuo K, et al. Impact of the combination of donor age and HLA disparity on the outcomes of unrelated bone marrow transplantation. Bone Marrow Transplant. 2021;56(10):2410-2422.
35. Shaw BE, Logan BR, Spellman SR, et al. Development of an unrelated donor selection score predictive of survival after HCT: donor age matters most. Biol Blood Marrow Transplant. 2018;24(5):1049-1056.
36. Hormaechea-Agulla D, Matatall KA, Le DT, et al. Chronic infection drives Dnmt3a-loss-of-function clonal hematopoiesis via IFNgamma signaling. Cell Stem Cell. 2021;28(8):1428-1442.e6.

37. Uryu H, Saeki K, Haeno H, et al. Clonal evolution of hematopoietic stem cells after autologous stem cell transplantation. Nat Genet. 2025;57(7):1695-1707.
38. Zhang B, Nguyen LXT, Li L, et al. Bone marrow niche trafficking of miR-126 controls the self-renewal of leukemia stem cells in chronic myelogenous leukemia. Nat Med. 2018;24(4):450-462.
39. Dawson A, Zarou MM, Prasad B, et al. Leukaemia exposure alters the transcriptional profile and function of BCR::ABL1 negative macrophages in the bone marrow niche. Nat Commun. 2024;15(1):1090.
40. Dolinska M, Cai H, Mansson A, et al. Characterization of the bone marrow niche in patients with chronic myeloid leukemia identifies CXCL14 as a new therapeutic option. Blood. 2023;142(1):73-89.
41. Tarantini F, Cumbo C, Zagaria A, et al. Clonal hematopoiesis onset in chronic myeloid leukemia patients developing an adverse cardiovascular event. Leuk Res. 2023;127:107023.
42. De Melo VA, Milojkovic D, Khorashad JS, et al. Philadelphia-negative clonal hematopoiesis is a significant feature of dasatinib therapy for chronic myeloid leukemia. Blood. 2007;110(8):3086-3087.
43. Morita K, Koya J, Toya T, Nakamura F, Kurokawa M. Philadelphia chromosome-negative acute promyelocytic leukemia manifesting after long-term imatinib treatment for chronic myeloid leukemia: a case report and literature review. Ann Hematol. 2018;97(6):1105-1109.
44. Levrat E, Roosnek E, Masouridi S, et al. Very long term stability of mixed chimerism after allogeneic hematopoietic stem cell transplantation in patients with hematologic malignancies. Bone Marrow Res. 2015;2015:176526.
45. Majzner RG, Mogri H, Varadhan R, et al. Post-transplantation cyclophosphamide after bone marrow transplantation is not associated with an increased risk of donor-derived malignancy. Biol Blood Marrow Transplant. 2017;23(4):612-617.
46. Vigorito AC, Campregher PV, Storer BE, et al. Evaluation of NIH consensus criteria for classification of late acute and chronic GVHD. Blood. 2009;114(3):702-708.
47. Buscarlet M, Provost S, Zada YF, et al. DNMT3A and TET2 dominate clonal hematopoiesis and demonstrate benign phenotypes and different genetic predispositions. Blood. 2017;130(6):753-762.
48. Zhang Q, Yim R, Lee P, Chin L, Li V, Gill H. Implications of clonal hematopoiesis in hematological and non-hematological disorders. Cancers (Basel). 2024;16(23):4118.

49. Fabre MA, de Almeida JG, Fiorillo E, et al. The longitudinal dynamics and natural history of clonal haematopoiesis. Nature. 2022;606(7913):335-342.
50. Lee SJ, Wolff D, Kitko C, et al. Measuring therapeutic response in chronic graft-versus-host disease. National Institutes of Health consensus development project on criteria for clinical trials in chronic graft-versus-host disease: IV. The 2014 Response Criteria Working Group report. Biol Blood Marrow Transplant. 2015;21(6):984-999.

Table 1. The association between patient characterisitcs and clonal hematopoiesis

Characteristics	Patients with CH (n=29)	Patients without CH (n=72)	p value
Patient age at HCT, years, median	42.0	36.0	0.036
Donor age at HCT, years, median	37.0	34.0	0.041
Patient age at sampling, years, median	54.8	48.9	0.019
Donor age at sampling, years, median	52.2	47.7	0.028
Interval between HCT and sample collection, years, median	14.8	13.2	0.47
Underlying disease			0.034
Acute myeloid leukemia	5 (14.7%)	29 (85.3%)	
Chronic myeloid leukemia	10 (62.5%)	6 (37.5%)	
Acute lymphoblastic leukemia	4 (26.7%)	11 (73.3%)	
Aplastic anemia	3 (20.0%)	12 (80.0%)	
Myelodysplastic syndromes	4 (30.8%)	9 (69.2%)	
Non-Hodgkin lymphoma	2 (40.0%)	3 (60.0%)	
Multiple myeloma	1 (33.3%)	2 (66.7%)	
Graft source			0.68
Bone marrow	24 (30.0%)	56 (70.0%)	
Peripheral blood	5 (29.4%)	12 (70.6%)	
Cord blood	0 (0.0%)	4 (100.0%)	
Neutrophil engraftment, day, madian (range)	18.0	18.0	0.99
Platelet engraftment, day, madian (range)	30.0	28.0	0.62
History of acute GVHD, grade			0.64
0-1	19 (27.1%)	51 (72.9%)	
2-4	10 (32.3%)	21 (67.7%)	
History of chronic GVHD			0.51
None	11 (36.7%)	19 (63.3%)	
Limited	10 (27.0%)	27 (73.0%)	
Extensive	8 (23.5%)	26 (76.5%)	
Chronic GVHD at sampling			0.28
None	22 (26.8%)	60 (73.2%)	
Mild	1 (14.3%)	6 (85.7%)	
Moderate	5 (50.0%)	5 (50.0%)	
Severe	1 (50.0%)	1 (50.0%)	
WBC count at sampling, /μL, median	6,200	7,200	0.22
Neutrophil count at sampling, /μL, median	3,590	3,758	0.52
Lymphocyte count at sampling, /μL, median	2,052	2,220	0.20
Hemoglobin level at sampling, g/dL, median	13.7	13.8	0.94
Platelet count at sampling, /L, median	22.4	22.3	0.95
RDW-CV, %, median	13.7	13.5	0.61

GVHD, graft-versus-host disease; HCT, hematopoietic stem cell transplantation; WBC, white blood cell.

Table 2. Multivariable analysis for clonal hematopoiesis

Characteristics	Odds ratio (95% confidence interval)	p value
Patient age at sampling	1.05 (0.994-1.11)	0.082
Donor age at sampling	1.06 (1.00-1.12)	0.032
Underlying disease		
Acute myeloid leukemia	0.407 (0.108-1.53)	0.18
Chronic myeloid leukemia	7.13 (1.38-36.8)	0.019
Others	1	
ABO Incompatibility		
Matched	2.50 (0.729-8.59)	0.15
Major mismatch	0.161 (0.0172-1.51)	0.11
Others	1	
Chronic GVHD at sampling		
Moderate or severe	5.49 (1.05-28.7)	0.044
Others	1	

Table 3. Type of second cancer after sampling

Affected organ	Total (n=101)	Patients with CH (n=29)	Patients without CH (n=72)
Appendix	1	0	1
Esophagus	2	0	2
Lung	1	1	0
Oral cavity	6	2	4
Ovary	1	1	0
Prostate	1	1	0
Skin	3	1	2
Total	15	6	9

Figure legends

Figure 1. Prevalence and characteristics of CH. (A) Pie chart showing the proportion of patients with and without CH. (B) Number of patients stratified by the number of CH mutations. (C) Bar chart of patients with each identified CH mutations. (D) Distribution of variant allele frequencies (VAFs) for the detected CH mutations. (E) Pie chart showing the functional annotations of the CH mutations. CH indicates clonal hematopoiesis; VAF, variant allele frequency; and VUS, variant of unknown significance.

Figure 2. Association between CH and clinical factors. (A) Donor age and (B) patient age at the time of sampling, stratified by CH status (no CH, VUS, or pathogenic CH). (C) Prevalence of CH stratified by disease history. CH indicates clonal hematopoiesis, and VUS, variant of unknown significance.

Figure 3. Prognostic impact of CH. (A) Cumulative incidence of secondary malignancies stratified by CH status. (B) OS from the time of sampling, stratified by CH status. (C) OS after sampling, stratified by active cGVHD at the time of sampling.

CH indicates clonal hematopoiesis; mode-severe cGVHD, moderate or severe chronic graft-versus-host disease; and OS, overall survival.

Figure 1

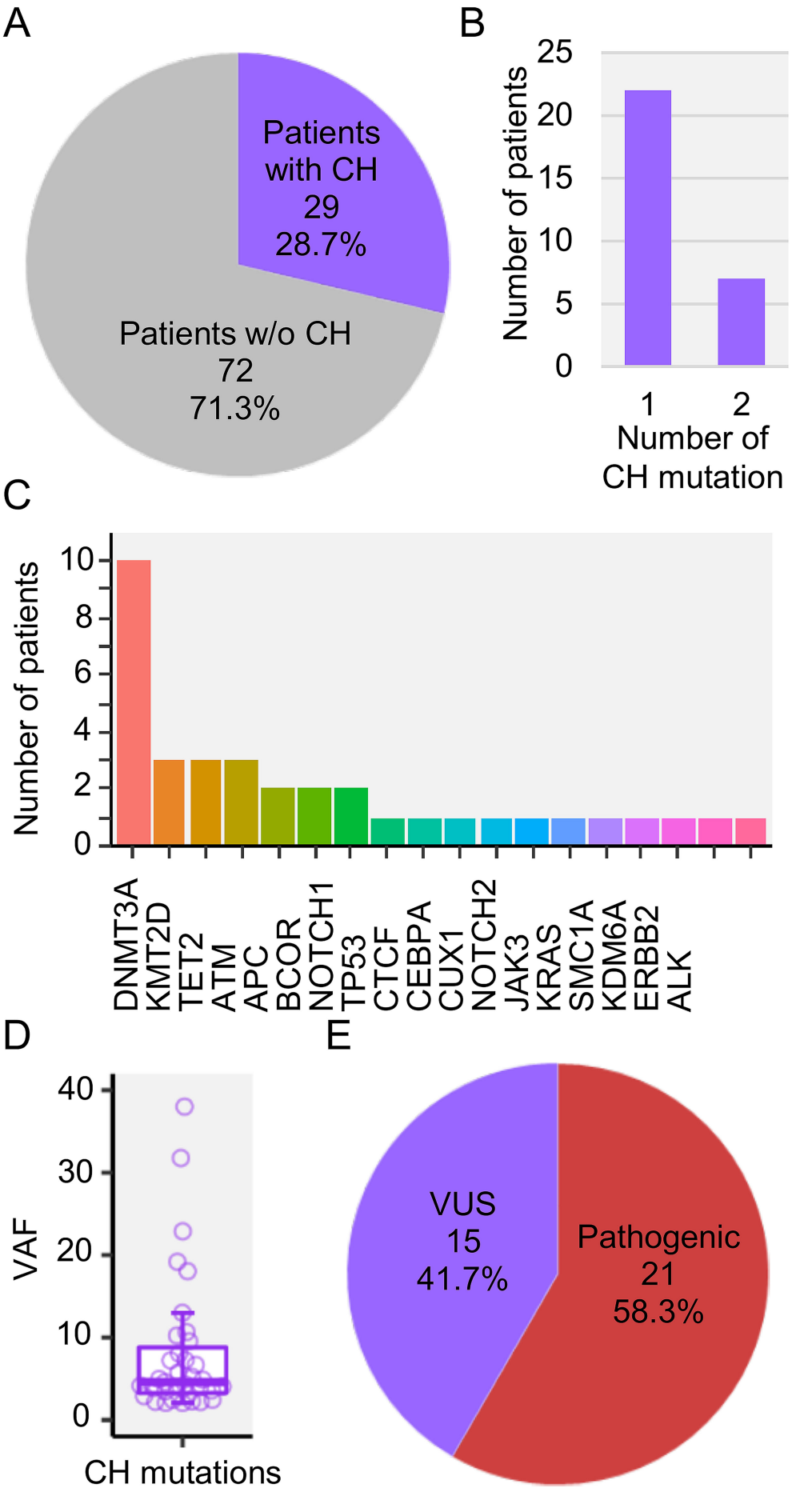
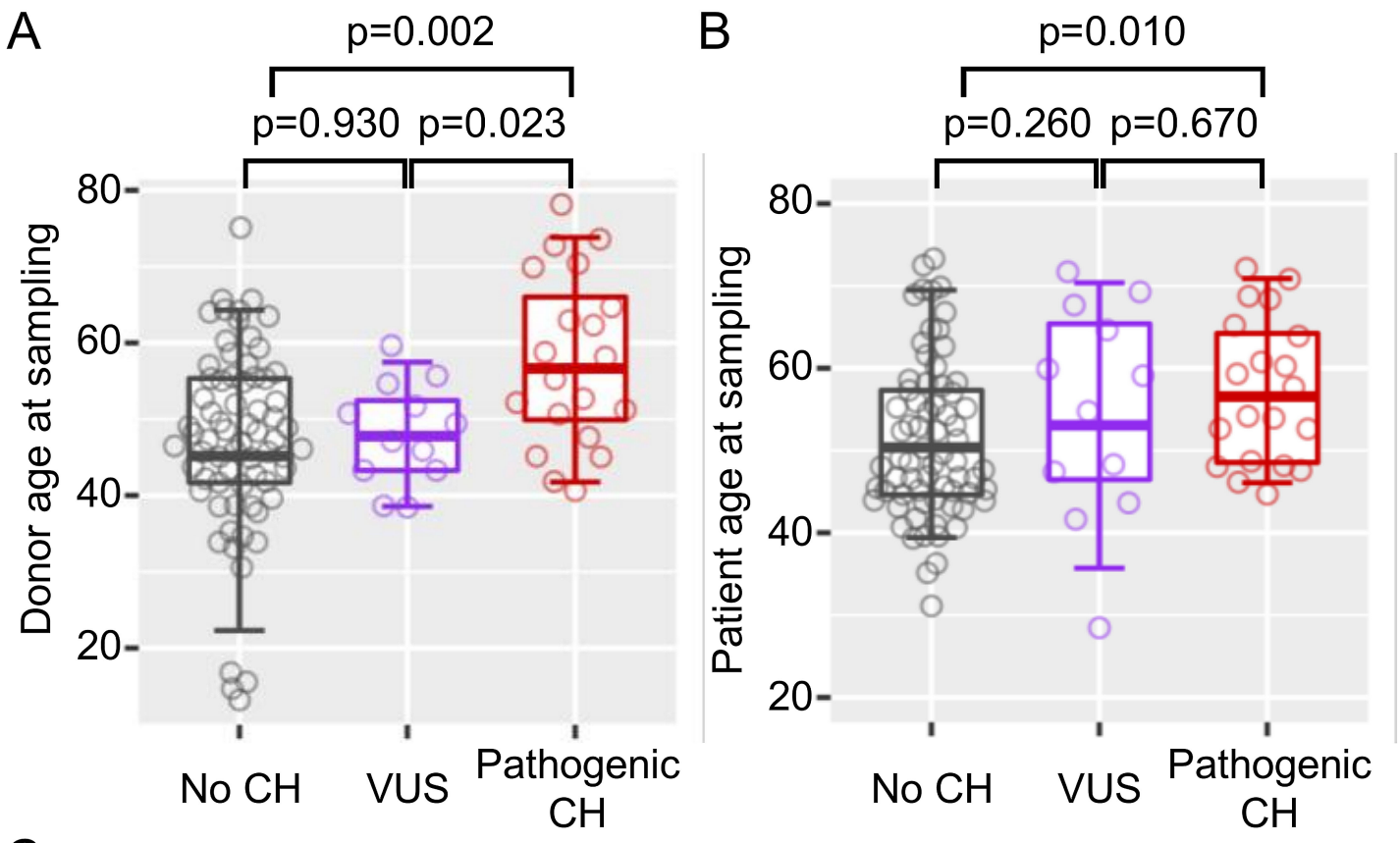
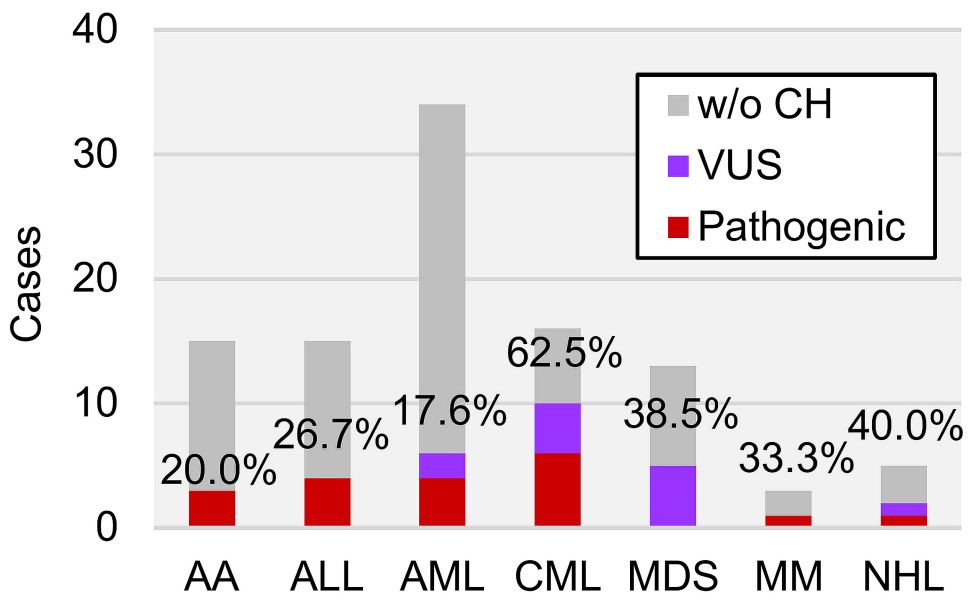

A
Patients with CH
29
28.7%
Patients w/o CH
72
71.3%
B
Number of patients
25
20
15
10
5
0
1
2
Number of CH mutation
C
Number of patients
10
8
6
4
2
0
DNMT3A
KMT2D
TET2
ATM
APC
BCOR
NOTCH1
TP53
CTCF
CEBPA
CUX1
NOTCH2
JAK3
KRAS
SMC1A
KDM6A
ERBB2
ALK
D
VAF
40
30
20
10
0
CH mutations
E
VUS
15
41.7%
Pathogenic
21
58.3%

Figure 2

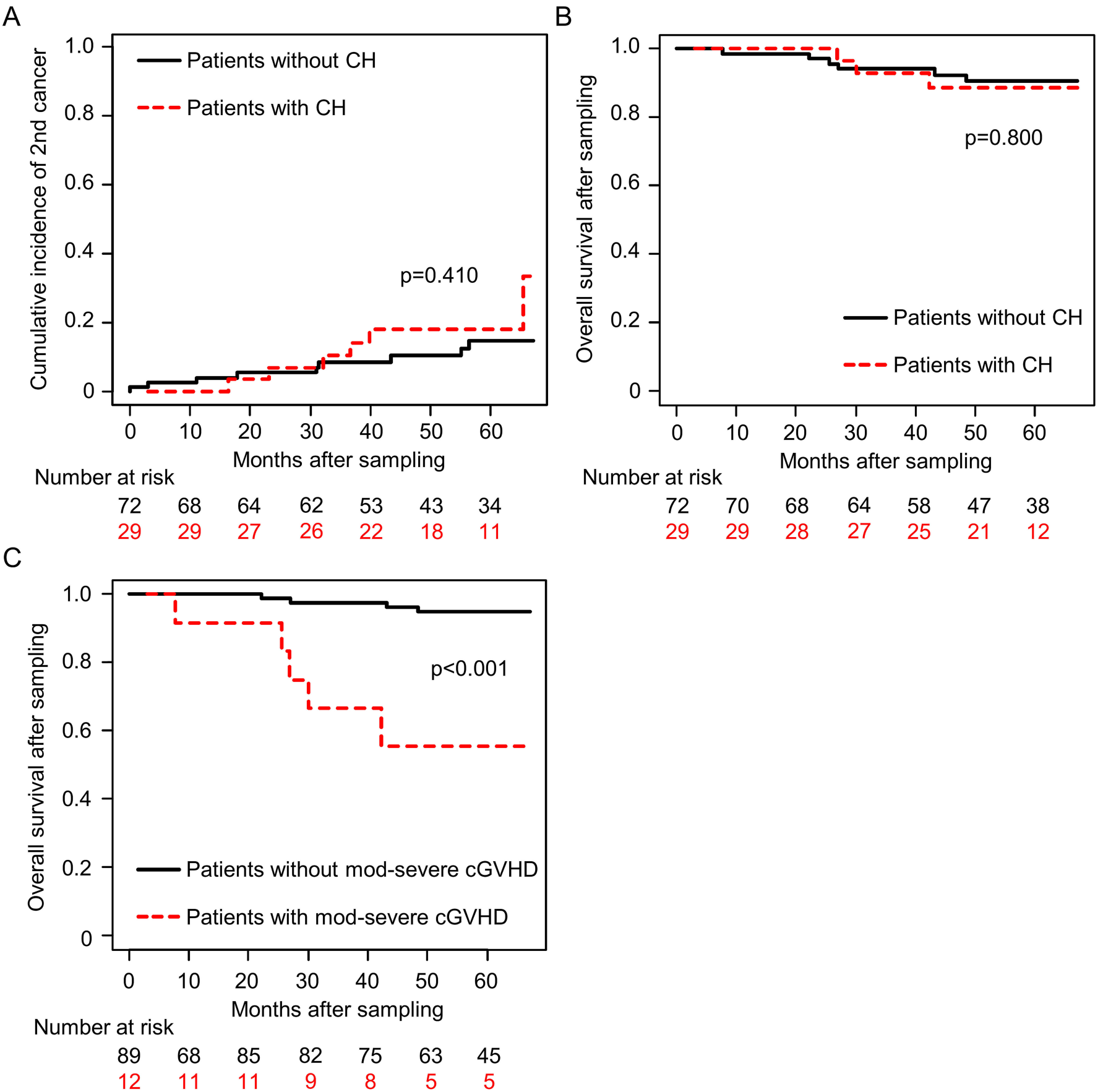


C



	AA	ALL	AML	CML	MDS	MM	NHL
Total :	15	15	34	16	13	3	5
VUS CH:	0	0	2	4	5	0	1
Pathogenic CH:	3	4	4	6	0	1	1

Figure 3


A
Cumulative incidence of 2nd cancer
1.0
0.8
0.6
0.4
0.2
0
Patients without CH
Patients with CH
p=0.410
0
10
20
30
40
50
60
Months after sampling
Number at risk
72 68 64 62 53 43 34
29 29 27 26 22 18 11
B
Overall survival after sampling
1.0
0.8
0.6
0.4
0.2
0
p=0.800
Patients without CH
Patients with CH
0
10
20
30
40
50
60
Months after sampling
Number at risk
72 70 68 64 58 47 38
29 29 28 27 25 21 12
C
Overall survival after sampling
1.0
0.8
0.6
0.4
0.2
0
p<0.001
Patients without mod-severe cGVHD
Patients with mod-severe cGVHD
0
10
20
30
40
50
60
Months after sampling
Number at risk
89 68 85 82 75 63 45
12 11 11 9 8 5 5

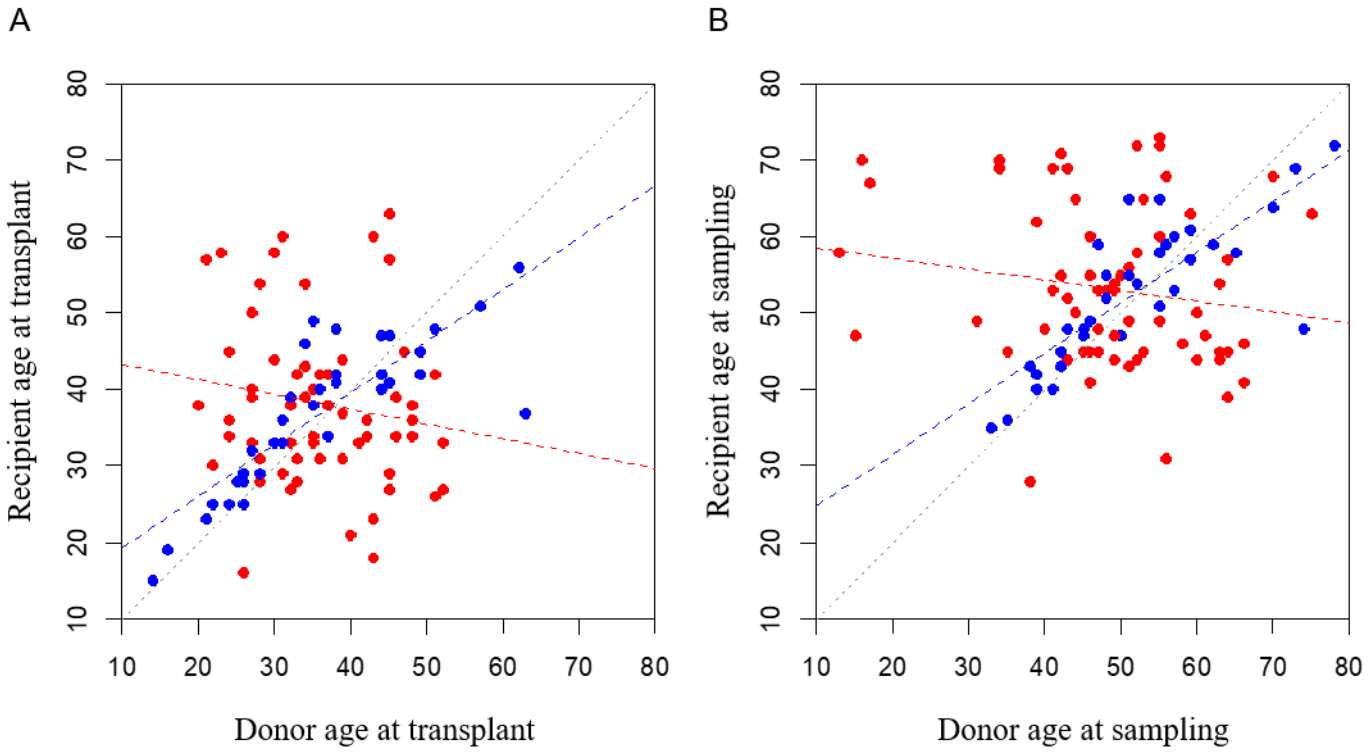
Clonal hematopoiesis in long-term survivors after allogeneic hematopoietic cell transplantation: associations with active chronic graft-versus-host disease, donor age, and transplantation for chronic myeloid leukemia

Supplementary information

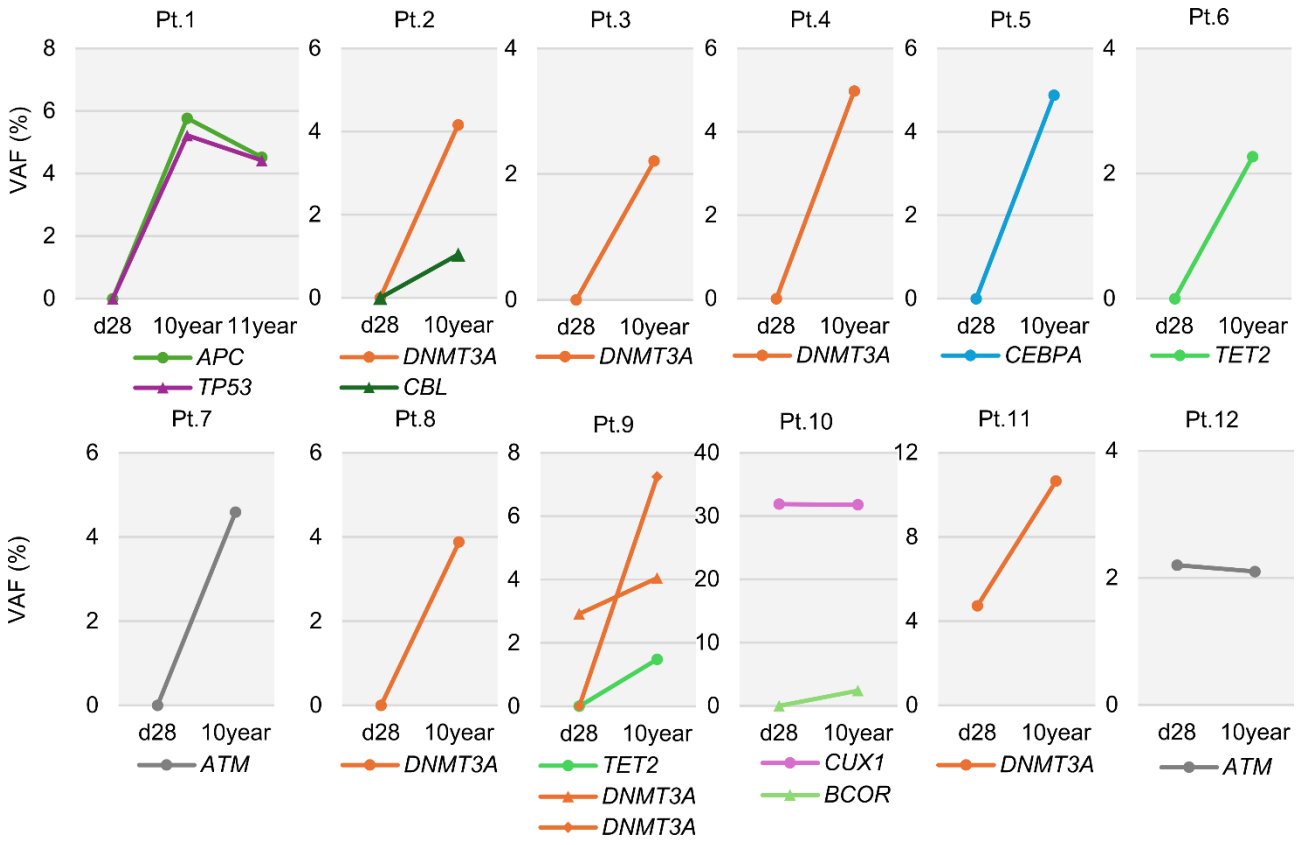
Supplemental Methods

Gene variant discovery

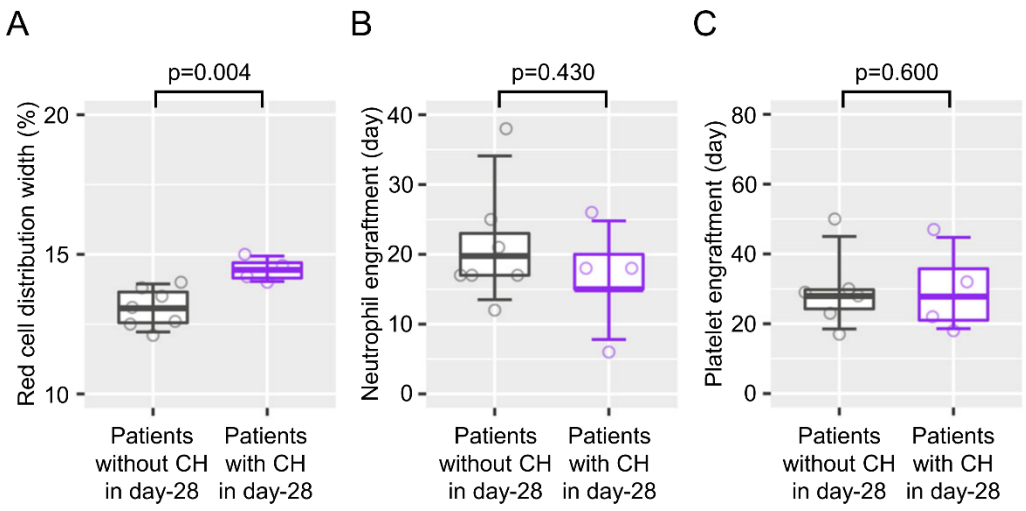
Fastq files from next-generation sequencing were pre-processing using Trimmomatic,[1] and the resulting reads were aligned to the human reference genome (hg19) using Burrows–Wheeler Alignment (BWA).[2] Qualimap[3] was used to evaluate the coverages of mapped reads. Variant calling was performed using Mutect2 (from the GATK suite).[4] To exclude sequencing artifacts and errors, we applied a Panel of Normals constructed from 13 individuals without hematological disorders, followed by the FilterMutectCalls, as described previously.[5] Based on validation experiments from our prior study demonstrating the successful removal of artificial/noise variants at this threshold, variants with a frequency of ≥0.02 were considered positive. Additionally, variants with a VAF of ≥0.01 were retained specifically for multi-timepoint tracking analyses to ensure the robustness of longitudinal results.



Supplementary Figure 1. Correlation between donor and recipient ages (A) at hematopoietic stem cell transplantation and (B) at sampling. Each point represents an individual donor–recipient pair. Blue and red points indicate related and unrelated donors, respectively. The corresponding dashed lines represent group-specific linear regression lines, and the gray dotted line indicates the line of identity (recipient age = donor age).



Supplementary Figure 2. Longitudinal dynamics of CH clones from day 28 to >10 years after HCT in 12 patients. CH indicates clonal hematopoiesis, and HCT, hematopoietic cell transplantation.



Supplementary Figure 3. Association between CH status at day 28 post-HCT and clinical characteristics >10 years after HCT. (A) RDW, (B) neutrophil engraftment, and (C) platelet engraftment, stratified by CH status. CH indicates clonal hematopoiesis; HCT, hematopoietic cell transplantation; and RDW, red cell distribution width.

Supplemental Table 1. List of analyzed 68 genes.

Gene	Target exons	Gene	Target exons	Gene	Target exons
ABL1	4–6	*GATA2*	2–6	*PHF6*	full
ALK	full	*HRAS*	2,3	*PRPF8*	full
APC	full	*IDH1*	4	*PTEN*	full
ASXL1	full	*IDH2*	4	*PTPN11*	3,13
ATM	full	*IKZF1*	full	*RAD21*	full
ATRX	full	*JAK2*	12,14	*RB1*	full
BCOR	full	*JAK3*	full	*RET*	full
BCORL1	full	*KDM6A*	full	*RUNX1*	full
BRAF	15	*KDR*	full	*SETBP1*	4
CALR	full	*KIT*	2,8–11,13,17	*SF3B1*	13-16
CBL	8,9	*KMT2A*	full	*SH2B3*	full
CDKN2A	full	*KMT2D*	full	*SMC1A*	full
CEBPA	full	*KRAS*	2,3	*SMC3*	full
CSF3R	14–17	*LRP1B*	full	*SPEN*	full
CUX1	full	*MPL*	10	*SRSF2*	1
DDX41	full	*MTOR*	full	*STAG2*	full
DNMT3A	full	*MYD88*	3–5	*TET1*	full
ERBB2	full	*NF1*	full	*TET2*	full
ERBB3	full	*NOTCH1*	full	*TP53*	full
ETV6	full	*NOTCH2*	full	*U2AF1*	2+6
EZH2	full	*NPM1*	12	*WT1*	7+9
FBXW7	full	*NRAS*	2,3	*ZRSR2*	full
FLT3	14,15,20	*NTRK3*	full		

Supplemental Table 2. Patient characterisitcs (n=101)

Characteristics	Value
Patient age at HCT, years, median (range)	38.0 (15-63)
Donor age at HCT, years, median (range)	35.0 (0-63)
Patient age at sampling, years, median (range)	52.4 (28-73)
Donor age at sampling, years, median (range)	49.0 (13-78)
Transplant year	
1990-2004	61
2005-2011	40
Interval between HCT and sample collection, years, median (range)	13.8 (10.0-29.6)
Patient sex (male/female)	60/41
Donor sex (male/female)	63/38
Sex mismatch	53
Female to male	25
Underlying disease	
Acute myeloid leukemia	34
Chronic myeloid leukemia	16
Acute lymphoblastic leukemia	15
Aplastic anemia	15
Myelodysplastic syndromes	13
Non-Hodgkin lymphoma	5
Multiple myeloma	3
Graft source	
Bone marrow	80
Peripheral blood	17
Cord blood	4
HLA	
Matched	72
Mismatched	29
Relationship	
Related	35
Unrelated	66
GVHD prophylaxis	
Cyclosporin + MTX	59
Tacrolimus + MTX	38
Cyclosporin	2
Tacrolimus	1
MTX	1
Conditioning intensity	
Myeloablative	81
Reduced intensity	20
Disease status at HCT	
CR	61
Non-CR	25
N/A	15
Neutrophil engraftment, day, median (range)	18 (6-46)
Platelet engraftment, day, median (range)	29 (2-202)
History of acute GVHD, grade	
0-1	70
2-4	31
History of chronic GVHD	

None	30
Limited	37
Extensive	34
Chronic GVHD at sampling	
None	82
Mild	7
Moderate	10
Severe	2
Immunosuppressant at sampling	
None	86
Steroid	13
Calcineurin inhibitor	9
Other	1
WBC count at sampling, /μL, median (range)	6,900 (3,500-19,400)
Neutrophil count at sampling, /μL, median (range)	3,726 (1,452.5-15,908)
Lymphocyte count at sampling, /μL, median (range)	2,200 (876-4,995)
Monocyte count at sampling, /μL, median (range)	399 (36-1,455)
Hemoglobin level at sampling, g/dL, median (range)	13.8 (7.8-17.4)
Platelet count at sampling, /L, median (range)	22.4 (9.4-44.8)
MCV at sampling, median (range)	94 (82-109)
RDW-CV, %, median (range)	13.5 (12.1-21.3)
Reticulocyte count at sampling, median (range)	14 (5-32)
LDH at sampling, median (range)	186 (126-373)

CR, complete remission; GVHD, graft-versus-host disease; HCT, hematopoietic stem cell transplantation; LDH, lactate dehydrogenase; MCV, mean corpuscular volume; MTX, methotrexate; RDW-CV, red blood cell distribution width-coefficient of variation; WBC, white blood cell. N/A in disease status indicates aplastic anemia as a primary disease.

Supplemental Table 3. Detected clonal hematopoiesis (>1% variant allele frequency)

ID	Gene	Mutation_type	cDNA_change	AA_change	VAF	Read_Depth	Classification
TKLfF	*NOTCH1*	missense_variant	c.4834G>A	p.Gly1612Ser	38.01	3107	Likely pathogenic
TrRzd	*CUX1*	missense_variant	c.2008C>T	p.Arg670Trp	31.77	2436	VUS
TT5tb	*ATM*	missense_variant	c.779C>A	p.Pro260His	22.88	1757	VUS
UeGKz	*SMC1A*	splice_donor_variant	c.2562+1G>A		19.19	396	Pathogenic
T97wR	*TET2*	frameshift_variant	c.1741_1745delCGTAA	p.Arg581Ter	18.04	1463	Pathogenic
TVQsE	*ERBB2*	missense_variant	c.1214A>G	p.Glu405Gly	12.99	1370	VUS
TL3us	*DNMT3A*	missense_variant	c.2644C>T	p.Arg882Cys	10.66	2241	Pathogenic
TBdC8	*DNMT3A*	splice_acceptor_variant	c.1475-2A>G		10.23	1945	Pathogenic
TBdC8	*TET2*	missense_variant	c.4144C>A	p.His1382Asn	9.54	818	Likely pathogenic
TP4k4	*KDM6A*	stop_gained	c.439C>T	p.Gln147Ter	8.04	696	Pathogenic
YHToo	*DNMT3A*	missense_variant	c.1915C>T	p.Leu639Phe	7.24	2418	VUS
TQmZU	*NOTCH2*	missense_variant	c.4369C>G	p.Leu1457Val	7.22	2022	VUS
T97wR	*KRAS*	missense_variant	c.442A>G	p.Thr148Ala	6.68	1063	Likely pathogenic
D2Boz	*APC*	missense_variant	c.8003A>G	p.Asn2668Ser	5.77	2147	VUS
D2Boz	*TP53*	missense_variant	c.713G>A	p.Cys238Tyr	5.22	3219	Pathogenic
KwUXU	*DNMT3A*	missense_variant	c.2645G>A	p.Arg882His	4.98	1286	Pathogenic
TWZtL	*CEBPA*	missense_variant	c.566C>A	p.Pro189His	4.88	471	VUS
Tc6n5	*ATM*	missense_variant	c.560T>A	p.Val187Glu	4.59	631	VUS
TrGP7	*NOTCH1*	frameshift_variant	c.3422delA	p.Tyr1141SerfsTer38	4.3	1140	Pathogenic
TTc8L	*DNMT3A*	missense_variant	c.2645G>A	p.Arg882His	4.16	1513	Pathogenic
YHToo	*DNMT3A*	missense_variant	c.2711C>T	p.Pro904Leu	4.05	1852	Pathogenic
TVQsE	*DNMT3A*	missense_variant	c.1919T>G	p.Phe640Cys	4	2149	VUS
T4ioY	*CTCF*	missense_variant	c.987G>T	p.Met329Ile	3.97	780	Pathogenic
TvR9E	*DNMT3A*	splice_donor_variant	c.2478+2T>C		3.88	1521	Pathogenic

TT5tb	*KMT2D*	inframe_deletion	c.11217_11222delGCAGCA	p.Gln3744_Gln3745del	3.6	1889	VUS
T4X2v	*KMT2D*	inframe_deletion	c.11217_11222delGCAGCA	p.Gln3744_Gln3745del	3.45	4291	VUS
TVisQ	*ALK*	missense_variant	c.4433T>C	p.Met1478Thr	3.09	1584	VUS
ZX4hf	*DNMT3A*	frameshift_variant	c.2250delC	p.Phe751SerfsTer28	2.89	1281	Pathogenic
TUUzw	*BCOR*	stop_gained	c.3643A>T	p.Arg1215Ter	2.44	739	Pathogenic
TrRzd	*BCOR*	missense_variant	c.812C>T	p.Ser271Leu	2.42	744	VUS
WxeLi	*TET2*	frameshift_variant	c.3521delC	p.Ala1174ValfsTer52	2.27	616	Pathogenic
T6uO1	*DNMT3A*	frameshift_variant	c.1356_1357delAC	p.Pro453SerfsTer19	2.21	3348	Pathogenic
T3oUw	*JAK3*	frameshift_variant	c.115delC	p.Gln39SerfsTer108	2.18	2109	VUS
LSDGs	*ATM*	missense_variant	c.8666A>G	p.Asp2889Gly	2.1	1475	VUS
TfdiW	*KMT2D*	missense_variant	c.1424C>A	p.Pro475His	2.06	583	VUS
TUY25	*APC*	missense_variant	c.4507T>C	p.Ser1503Pro	2.06	874	VUS
TKLfF	*JAK3*	frameshift_variant	c.115delC	p.Gln39SerfsTer108	1.95	2406	VUS
YHToo	*TET2*	missense_variant	c.5650A>G	p.Thr1884Ala	1.48	2158	VUS
UeGKz	*DNMT3A*	missense_variant	c.2711C>T	p.Pro904Leu	1.45	1239	Pathogenic
TuzQY	*ATM*	missense_variant	c.3755A>G	p.Tyr1252Cys	1.34	1419	VUS
TT5tb	*KMT2D*	frameshift_variant	c.9265delG	p.Val3089TrpfsTer30	1.28	2880	Pathogenic
NMHxW	*SMC1A*	missense_variant	c.577G>T	p.Ala193Ser	1.24	2103	Likely pathogenic
TTc8L	*CBL*	missense_variant	c.1133T>C	p.Phe378Ser	1.04	2793	Likely pathogenic
T4X2v	*DNMT3A*	missense_variant	c.1969G>A	p.Val657Met	1.03	1750	VUS

Supplemental Table 4. The detailed association between patient characterisitcs and clonal hematopoiesis

Characteristics	Patients with CH (n=29)	Patients without CH (n=72)	Univariable analysis	Multivariable analysis	
			p value	OR (95% CI)	p value
Patient age at HCT, years, median	42.0	36.0	0.036		
Donor age at HCT, years, median	37.0	34.0	0.041		
Patient age at sampling, years, median	54.8	48.9	0.019	1.05 (0.994-1.11)	0.082
Donor age at sampling, years, median	52.2	47.7	0.028	1.06 (1.00-1.12)	0.032
Interval between HCT and sample collection, years, median	14.8	13.2	0.47		
Transplant year			0.51		
1990-2004	13 (32.5%)	27 (67.5%)			
2005-2011	16 (26.2%)	45 (73.8%)			
Sex mismatch			0.19		
Yes	12 (22.6%)	41 (77.4%)			
No	17 (35.4%)	31 (64.6%)			
Underlying disease			0.034		
Acute myeloid leukemia	5 (14.7%)	29 (85.3%)		0.407 (0.108-1.53)	0.18
Chronic myeloid leukemia	10 (62.5%)	6 (37.5%)		7.13 (1.38-36.8)	0.019
Acute lymphoblastic leukemia	4 (26.7%)	11 (73.3%)			
Aplastic anemia	3 (20.0%)	12 (80.0%)			
Myelodysplastic syndromes	4 (30.8%)	9 (69.2%)			
Non-Hodgkin lymphoma	2 (40.0%)	3 (60.0%)			
Multiple myeloma	1 (33.3%)	2 (66.7%)			
Graft source			0.68		
Bone marrow	24 (30.0%)	56 (70.0%)			
Peripheral blood	5 (29.4%)	12 (70.6%)			
Cord blood	0 (0.0%)	4 (100.0%)			
HLA			0.15		
Matched	24 (33.3%)	48 (66.7%)			
Mismatched	5 (17.2%)	24 (82.8%)			
Relationship			0.25		
Related	13 (37.1%)	22 (62.9%)			
Unrelated	16 (24.2%)	50 (75.8%)			
ABO Incompatibility			0.20		
Matched	20 (37.7%)	33 (62.3%)		2.50 (0.729-8.59)	0.15
Major mismatch	2 (12.5%)	14 (87.5%)		0.161 (0.0172-1.51)	0.11
Minor mismatch	4 (22.2%)	14 (77.8%)			

Major-minor mismatch	3 (21.4%)	11 (78.6%)			
GVHD prophylaxis			0.55		
Cyclosporin-based regimen	20 (32.8%)	41 (67.2%)			
Tacrolimus-based regimen	9 (23.1%)	30 (76.9%)			
Other	0 (0.0%)	1 (100.0%)			
Conditioning intensity			0.17		
Myeloablative	26 (32.1%)	55 (67.9%)			
Reduced intensity	3 (15.0%)	17 (85.0%)			
Disease status at HCT			1		
CR	19 (31.1%)	42 (68.9%)			
Non-CR	7 (28.0%)	18 (72.0%)			
Neutrophil engraftment, day, madian (range)	18.0	18.0	0.99		
Platelet engraftment, day, madian (range)	30.0	28.0	0.62		
History of acute GVHD, grade			0.64		
0-1	19 (27.1%)	51 (72.9%)			
2-4	10 (32.3%)	21 (67.7%)			
History of chronic GVHD			0.51		
None	11 (36.7%)	19 (63.3%)			
Limited	10 (27.0%)	27 (73.0%)			
Extensive	8 (23.5%)	26 (76.5%)			
Chronic GVHD at sampling			0.28		
None	22 (26.8%)	60 (73.2%)			
Mild	1 (14.3%)	6 (85.7%)			
Moderate	5 (50.0%)	5 (50.0%)			
Severe	1 (50.0%)	1 (50.0%)		5.49 (1.05-28.7)	0.044
Calcineurin inhibitor use at sampling			0.27		
Yes	4 (44.4%)	5 (55.6%)			
No	25 (27.2%)	67 (72.8%)			
Steroid use at sampling			0.51		
Yes	5 (38.5%)	8 (61.5%)			
No	24 (27.3%)	64 (72.7%)			
WBC count at sampling, /μL, median	6,200	7,200	0.22		
Neutrophil count at sampling, /μL, median	3,590	3,758	0.52		
Lymphocyte count at sampling, /μL, median	2,052	2,220	0.20		
Monocyte count at sampling, /μL, median	338	426	0.34		
Hemoglobin level at sampling, g/dL, median	13.7	13.8	0.94		
Platelet count at sampling, /L, median	22.4	22.3	0.95		

MCV at sampling, median	95.0	93.5	0.28
RDW-CV, %, median	13.7	13.5	0.61
Reticulocyte count at sampling, median	15.0	14.0	0.78
LDH at sampling, U/L, median	188	186	0.79

CI, confidence interval; CR, complete remission; GVHD, graft-versus-host disease; HCT, hematopoietic stem cell transplantation; LDH, lactate dehydrogenase; MCV, mean corpuscular volume; MTX, methotrexate; OR, odds ratio; WBC, white blood cell. N/A in disease status indicates aplastic anemia as a primary disease.

Supplemental Table 5. The association between patient characteristics and pathogenic clonal hematopoiesis

Characteristics	Patients with pathogenic CH (n=19)	Patients with VUS (n=10)	Patients without CH (n=72)	p value
Patient age at HCT, years, median	42.0	42.0	36.0	0.108
Donor age at HCT, years, median	39.0	32.5	34.0	0.026
Patient age at sampling, years, median	56.0	57.0	49.0	0.054
Donor age at sampling, years, median	55.0	48.5	47.5	0.014
Interval between HCT and sample collection, years, median	15.0	14.7	13.2	0.766
Transplant year				0.721
1990-2004	9	4	27	
2005-2011	10	6	45	
Sex mismatch				0.016
Yes	11	1	41	
No	8	9	31	
Underlying disease[#]				
Acute myeloid leukemia	4	1	29	
Chronic myeloid leukemia	6	4	6	
Acute lymphoblastic leukemia	4	0	11	
Aplastic anemia	3	0	12	
Myelodysplastic syndromes	0	4	9	
Non-Hodgkin lymphoma	1	1	3	
Multiple myeloma	1	0	2	
Graft source				0.953
Bone marrow	16	8	56	
Peripheral blood	3	2	12	
Cord blood	0	0	4	
HLA				0.252
Matched	15	9	48	
Mismatched	4	1	24	
Relationship				0.346
Related	9	4	22	
Unrelated	10	6	50	
ABO Incompatibility				0.552
Matched	14	6	33	
Major mismatch	1	1	14	
Minor mismatch	2	2	14	
Major-minor mismatch	2	1	11	
GVHD prophylaxis				0.614
Cyclosporin-based regimen	12	8	41	
Tacrolimus-based regimen	7	2	30	
Other	0	0	1	
Conditioning intensity				0.239
Myeloablative	16	10	55	
Reduced intensity	3	0	17	
Disease status at HCT				0.127
CR	14	5	42	
Non-CR	2	5	18	
Neutrophil engraftment, day, madian	18.0	18.5	18.0	0.921
Platelet engraftment, day, madian	30.0	29.0	29.0	0.832

History of acute GVHD, grade				0.784
0-1	13	6	51	
2-4	6	4	21	
History of chronic GVHD				0.389
None	7	4	19	
Limited	5	5	27	
Extensive	7	1	26	
Chronic GVHD at sampling				0.353
None	13	9	60	
Mild	1	0	6	
Moderate	4	1	5	
Severe	1	0	1	
Calcineurin inhibitor use at sampling				0.436
Yes	3	1	5	
No	16	9	67	
Steroid use at sampling				0.445
Yes	4	1	8	
No	15	9	64	
WBC count at sampling, /μL, median	6,700	5,600	7,200	0.214
Neutrophil count at sampling, /μL, median	3,761	2,892	3,758	0.065
Lymphocyte count at sampling, /μL, median	1,773	2,349	2,220	0.221
Monocyte count at sampling, /μL, median	372	320	426	0.391
Hemoglobin level at sampling, g/dL, median	13.7	14.3	13.8	0.531
Platelet count at sampling, /L, median	24.0	20.2	22.3	0.514
MCV at sampling, median	96.0	93.5	93.5	0.404
RDW-CV, %, median	13.8	13.5	13.5	0.343
Reticulocyte count at sampling, median	15.0	15.0	14.0	0.956
LDH at sampling, U/L, median	193	181	186	0.722

CR, complete remission; GVHD, graft-versus-host disease; HCT, hematopoietic stem cell transplantation; LDH, lactate dehydrogenase; MCV, mean corpuscular volume; MTX, methotrexate; WBC, white blood cell. N/A in disease status indicates aplastic anemia as a primary disease. #P values could not be computed using Fisher's exact test due to the large number of categories, which resulted in sparse cell counts.

Supplemental Table 6. Multivariable analysis for pathogenic clonal hematopoiesis

Characteristics	Odds ratio (95% confidence interval)	p value
Patient age at sampling	1.02 (0.955-1.08)	0.60
Donor age at sampling	1.07 (1.01-1.13)	0.023
Underlying disease		
Chronic myeloid leukemia	4.18 (1.00-17.4)	0.049
Others	1	
ABO Incompatibility		
Matched	4.99 (1.26-19.8)	0.022
Others	1	
Chronic GVHD at sampling		
Moderate or severe	6.92 (1.43-33.5)	0.016
Others	1	

GVHD, graft-versus-host disease.

Supplemental Table 7. The association between patient characterisitcs and clonal hematopoiesis

Characteristics	Patients with CH (n=32)	Patients without CH (n=69)	Univariable analysis	Multivariable analysis	
			p value	OR (95% CI)	p value
Patient age at HCT, years, median	42.0	36.0	0.017		
Donor age at HCT, years, median	36.5	33.0	0.027		
Patient age at sampling, years, median	56.2	48.7	0.011	1.03 (0.980-1.09)	0.22
Donor age at sampling, years	51.9	47.3	0.021	1.09 (1.03-1.17)	0.016
Interval between HCT and sample collection, years, median	14.5	13.2	0.48		
Transplant year			0.89		
1990-2004	13 (32.5%)	27 (67.5%)			
2005-2011	19 (31.1%)	42 (68.9%)			
Sex mismatch			0.29		
Yes	14 (26.4%)	39 (73.6%)			
No	18 (37.5%)	30 (62.5%)			
Underlying disease			0.094		
Acute myeloid leukemia	7 (20.6%)	27 (79.4%)			
Chronic myeloid leukemia	10 (62.5%)	6 (37.5%)		10.7 (2.22-51.7)	0.0094
Acute lymphoblastic leukemia	4 (26.7%)	11 (73.3%)			
Aplastic anemia	3 (20.0%)	12 (80.0%)			
Myelodysplastic syndromes	5 (38.5%)	8 (61.5%)			
Non-Hodgkin lymphoma	2 (40.0%)	3 (60.0%)			
Multiple myeloma	1 (33.3%)	2 (66.7%)			
Graft source			0.52		
Bone marrow	26 (32.5%)	54 (67.5%)			
Peripheral blood	6 (35.3%)	11 (64.7%)			
Cord blood	0 (0.0%)	4 (100.0%)			
HLA			0.060		
Matched	27 (37.5%)	45 (62.5%)		5.10 (1.24-21.0)	0.024
Mismatched	5 (17.2%)	24 (82.8%)			
Relationship			0.11		
Related	15 (42.9%)	20 (57.1%)			
Unrelated	17 (25.8%)	49 (74.2%)			
ABO Incompatibility			0.12		

Matched	22 (41.5%)	31 (58.5%)		4.47 (1.48-13.5)	0.0080
Major mismatch	2 (12.5%)	14 (87.5%)			
Minor mismatch	4 (22.2%)	14 (77.8%)			
Major-minor mismatch	4 (28.6%)	10 (71.4%)			
GVHD prophylaxis			0.77		
Cyclosporin-based regimen	21 (34.4%)	40 (65.6%)			
Tacrolimus-based regimen	11 (28.2%)	28 (71.8%)			
Other	0 (0.0%)	1 (100.0%)			
Conditioning intensity			0.11		
Myeloablative	29 (35.8%)	52 (64.2%)			
Reduced intensity	3 (15.0%)	17 (85.0%)			
Disease status at HCT			1		
CR	21 (34.4%)	40 (65.6%)			
Non-CR	8 (32.0%)	17 (68.0%)			
Neutrophil engraftment, day, madian	18.0	18.0	0.73		
Platelet engraftment, day, madian	30.0	28.0	0.91		
History of acute GVHD, grade			0.36		
0-1	20 (28.6%)	50 (71.4%)			
2-4	12 (38.7%)	19 (61.3%)			
History of chronic GVHD			0.51		
None	12 (40.0%)	18 (60.0%)			
Limited	11 (29.7%)	26 (70.3%)			
Extensive	9 (26.5%)	25 (73.5%)			
Chronic GVHD at sampling			0.38		
None	25 (30.5%)	57 (69.5%)			
Mild	1 (14.3%)	6 (85.7%)			
Moderate	5 (50.0%)	5 (50.0%)			
Severe	1 (50.0%)	1 (50.0%)		5.39 (1.11-26.2)	0.037
Calcineurin inhibitor use at sampling			0.14		
Yes	5 (55.6%)	4 (44.4%)			
No	27 (29.3%)	65 (70.7%)			
Steroid use at sampling			0.54		
Yes	5 (38.5%)	8 (61.5%)			
No	27 (30.7%)	61 (69.3%)			
WBC count at sampling, /μL, median	6,200	7,200	0.37		
Neutrophil count at sampling, /μL, median	3,726	3,695	0.85		

Lymphocyte count at sampling, /μL, median	2,232	2,177	0.19
Monocyte count at sampling, /μL, median	345	425	0.66
Hemoglobin level at sampling, g/dL, median	13.9	13.8	0.68
Platelet count at sampling, /L, median	23.0	22.1	0.84
MCV at sampling, median	94.5	94.0	0.35
RDW-CV, %, median	13.7	13.5	0.63
Reticulocyte count at sampling, median	15.0	13.0	0.45
LDH at sampling, U/L, median	182.5	186	0.37

CI, confidence interval; CR, complete remission; GVHD, graft-versus-host disease; HCT, hematopoietic stem cell transplantation; LDH, lactate dehydrogenase; MCV, mean corpuscular volume; MTX, methotrexate; OR, odds ratio; WBC, white blood cell. N/A in disease status indicates aplastic anemia as a primary disease.

Supplemental Table 8. The association between patient characterisitcs and clonal hematopoiesis with 1% cutoff

Characteristics	Patients with CH (n=34)	Patients without CH (n=67)	Univariable analysis	Multivariable analysis	
			p value	OR (95% CI)	p value
Patient age at HCT, years, median	41.0	36.0	0.026		
Donor age at HCT, years, median	35.5	33.0	0.049		
Patient age at sampling, years, median	55.3	48.7	0.019	1.04 (0.983-1.10)	0.18
Donor age at sampling, years	51.5	47.3	0.042	1.06 (1.00-1.12)	0.043
Interval between HCT and sample collection, years, median	14.5	13.2	0.53		
Transplant year			0.83		
1990-2004	14	26			
2005-2011	20	41			
Sex mismatch			0.14		
Yes	14	39			
No	20	28			
Underlying disease			0.11		
Acute myeloid leukemia	8	26			
Chronic myeloid leukemia	10	6		5.06 (1.41-18.1)	0.013
Acute lymphoblastic leukemia	4	11			
Aplastic anemia	3	12			
Myelodysplastic syndromes	6	7			
Non-Hodgkin lymphoma	2	3			
Multiple myeloma	1	2			
Graft source			0.48		
Bone marrow	28	52			
Peripheral blood	6	11			
Cord blood	0	4			
HLA			0.036		
matched	29	43		4.94 (1.06-22.9)	0.041
mismatched	5	24			
Relationship			0.078		
Related	16	19			
Unrelated	18	48		0.59 (0.19-1.87)	0.37
ABO Incompatibility			0.19		
Matched	23	30		4.15 (1.39-12.4)	0.011

Major mismatch	3	13			
Minor mismatch	4	14			
Major-minor mismatch	4	10			
GVHD prophylaxis			0.60		
Cyclosporin-based regimen	23	38			
Tacrolimus-based regimen	11	28			
Other	0	1			
Conditioning intensity			0.19		
Myeloablative	30	51			
Reduced intensity	4	16			
Disease status at HCT			1		
CR	22	39			
Non-CR	9	16			
Neutrophil engraftment, day, madian (range)	18.0	18.0	0.77		
Platelet engraftment, day, madian (range)	30.0	29.0	0.85		
History of acute GVHD, grade			0.50		
0-1	22	48			
2-4	12	19			
History of chronic GVHD			0.42		
None	13	17			
Limited	11	26			
Extensive	10	24			
Chronic GVHD at sampling			0.18		
None	26	56			
Mild	1	6			
Moderate	6	4			
Severe	1	1			
Calcineurin inhibitor use at sampling			0.16	9.19 (1.75-48.3)	0.0088
Yes	5	4			
No	29	63			
Steroid use at sampling			0.76		
Yes	5	8			
No	29	59			
WBC count at sampling, /μL, median	6,200	7,200	0.35		
Neutrophil count at sampling, /μL, median	3,726	3,694.80	0.88		
Lymphocyte count at sampling, /μL, median	2,001	2,096	0.15		

Monocyte count at sampling, /μL, median	345.0	430.0	0.60
Hemoglobin level at sampling, g/dL, median	13.9	13.8	0.66
Platelet count at sampling, /L, median	22.2	22.4	0.81
MCV at sampling, median	94.0	94.0	0.38
RDW-CV, %, median	13.7	13.5	0.67
Reticulocyte count at sampling, median	15.0	13.5	0.92
LDH at sampling, U/L, median	177	186	0.17

CI, confidence interval; CR, complete remission; GVHD, graft-versus-host disease; HCT, hematopoietic stem cell transplantation; LDH, lactate dehydrogenase; MCV, mean corpuscular volume; MTX, methotrexate; OR, odds ratio; WBC, white blood cell. N/A in disease status indicates aplastic anemia as a primary disease.

Supplementary references

1. Bolger AM, Lohse M, Usadel B. Trimmomatic: a flexible trimmer for Illumina sequence data. Bioinformatics. 2014;30(15):2114-2120.
2. Li H, Durbin R. Fast and accurate short read alignment with Burrows-Wheeler transform. Bioinformatics. 2009;25(14):1754-1760.
3. Okonechnikov K, Conesa A, Garcia-Alcalde F. Qualimap 2: advanced multi-sample quality control for high-throughput sequencing data. Bioinformatics. 2016;32(2):292-294.
4. McKenna A, Hanna M, Banks E, et al. The Genome Analysis Toolkit: a MapReduce framework for analyzing next-generation DNA sequencing data. Genome Res. 2010;20(9):1297-1303.
5. Sadato D, Hirama C, Kaiho-Soma A, et al. Archival bone marrow smears are useful in targeted next-generation sequencing for diagnosing myeloid neoplasms. PLoS One. 2021;16(7):e0255257.