were alive on day 14 or day 21 had a marrow examined then. Third, we did not examine marrow cellularity, feeling that this might be particularly susceptible to inter-observer variability. Finally, delaying until day 21 in patients with 20-59% blasts on day 14 might not affect CR rate, but might shorten CR duration. However, this risk has to be weighed against the competing risk of giving a second induction course, particularly to older patients. Specifically, while there will be myelosuppression on either a second induction course or a first post-remission course, the risk of infection at any given neutrophil count is less when a patient is in CR than when not, and duration of neutropenia is often less in patients in CR. Hence in older patients physicians might prefer to wait until CR to re-treat. Our data make the option of delay more plausible and suggest the need to revisit the NCCN recommendations in patients given 3+7.

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Lack of prognostic value of *FCGR3A*-V158F polymorphism in non-Hodgkin's lymphoma

Recently it was shown that the therapeutic efficacy of the anti-CD20 monoclonal antibody rituximab might be influenced by single nucleotide polymorphisms in the Fcγ receptor IIIa gene (*FCGR3A*).[1-3] Binding of the Fc (constant) region of immunoglobulin G1 (IgG1) to the FcGRIIIa on the surface of natural killer (NK) cells or macrophages triggers antibody-dependent cellular cytotoxicity (ADCC), inducing B-cell elimination. The *FCGR3A*-158 valine (V) allele has a higher affinity for IgG1 than the phenylalanine allele (F) and mediates ADCC more effectively.[4] Homozygous 158V follicular lymphoma patients were found to have better responses to single agent rituximab[1,3] and longer progression free survival.[2,3]

Fcγ receptor polymorphisms can also influence the immune response to auto-antibodies and have been shown to be risk factors in autoimmune disease.[5] In lymphoma patients, auto-antibodies to antigens expressed on lymphoma cells have been identified[6] and effector cells that express polymorphic *FCGR3A*-158, may also have altered binding to these antibodies that could influence host response and disease progression independent of rituximab therapy. In addition, a recent investigation has demonstrated that individuals expressing *FCGR3A*-158VV and VF show greater NK cell surface expression of *FcGRIIIa* receptors than the FF types.[7]

Few studies have examined a statistically large enough group of non-Hodgkin's lymphoma (NHL) patients to determine if VV patients have a biologically different disease or survival advantage when treated only with chemotherapy or radiation. We selected patients from 291 newly diagnosed NHL patients who were entered into our prospective biological prognostic factor study between 1990-1995. The Human Subjects Review Committee at the University of Toronto and the appropriate committees at Sunnybrook Health Science Centre approved the study. All patients provided informed consent. We studied the 194 patients who had sufficient information for a detailed multivariate analysis with five factors including grade, tumor bulk, International Progostic Index (IPI) score, B symptoms and *FCGR3A*-158 genotype. DNA sequencing of a 162 bp PCR product amplified specifically from the *FCGR3A* gene, determined genotype.[8]

There were 69 patients with indolent lymphoma (12 with International Working Formula (IWF) grade A, and 57 with grade B or C) and 125 patients with aggressive lymphoma (IWF grade D, E, F, G, H, I or J-known T-cell phenotypes were excluded). The IPI scores for all 194 patients were predictive of progression free and overall survival by Kaplan-Meier analysis as expected.

For the entire 194 patients, frequencies of the VV, VF, and FF polymorphisms, were 13%, 46% and 43% (see *Online Supplementary Table S1* for subgroup frequencies). The χ^2 test showed that there were no significant differences in the genotype frequencies (p=0.8752) between the two disease subgroups.

The population was in Hardy-Weinberg equilibrium. We calculated that our analysis had 89% power to detect differences attributed to genotype in 194 patients. We recognize that our cohort includes a somewhat heterogeneous group of indolent and aggressive NHL. A limitation of our analysis is that the power to detect small differences in outcomes in these smaller groups is reduced.

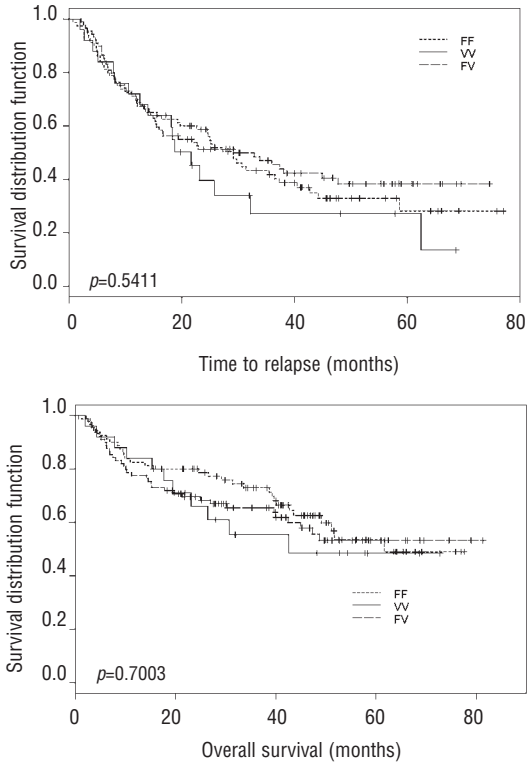

Figure 1. Kaplan-Meier progression free survival (PFS) and overall survival (OS) according to FCGR3A-V158F for 194 patients. (A) From log-rank test, there was no significant difference in relapse times between genotypes (p=0.5411). PFS at five years was 28.2% for FF, 38.3% for FV and 27.2% for VV. Additional testing at 24 months of follow-up, FF 58.7%, FV 51.3% and VV 39.6% was not statistically significant (p=0.1958). (B) From log-rank test, there was no significant difference between genotypes (p=0.7003). OS at five years was 53.5% for FF, 53.3% for FV and 48.5% for VV. Additional testing at two years of follow-up showed no significant difference among 3 genotypes, FF 80%, FV 69.6%, VV 66.0% (p=0.5995). (Censored data is indicated with bars: |).

Figure 1 shows progression free survival (PFS) (a) and overall survival (OS) (b) according to genotype for all 194 patients. There was no statistical difference in relapse times for the three genotypes (p=0.5411). Although the VV patients unexpectedly appear to relapse earlier, the values were not statistically different (VV *vs.* FF, p=0.1958). OS survival at five years was not significantly different for the genotypes, p=0.5411.

Table 1 shows the results for VV *vs.* VF *vs.* FF genotypes compared with patients' characteristics. None of the adverse factors contributing to IPI scores, or presence of B symptoms were related to *FCGR3A*-V158F genotype. However, less bulky disease was significantly associated with the VV genotype in the indolent patients (p=0.0086).

Uninvariate or Multiple Cox Proportional Hazard model of relapse for 194 patients demonstrated that only B symptoms and high IPI scores predicted relapse. Overall survival analysis by Univariate or Multiple Cox Proportional Hazard model showed that only B-symptoms, IPI score and grade were statistically prognostic.

Weng *et al.*[9] examined PFS in 158 follicular lymphoma (FL) patients who received only chemotherapy and not immunotherapy and also found that the VV group had the lowest 2-year PFS at 32% *vs.* 40% for FF and 38% for F carriers, but their results were not statistically significant. The subtypes chronic lymphocytic leukemia (CLL) and mantle cell lymphoma (MCL) may respond to monoclonal antibodies in pathways other than ADCC. This has been suggested because in studies of CLL patients receiving rituximab or alemtuzumab monoclonal antibody therapy[10,11] the VV genotype actually had the lowest response to treatment. In mantle cell patients receiving rituximab in the Ghelmini study,[2] the VV genotype did not have better survival. Our indolent group included only[12] patients (IWF A) who would now be classified as CLL or MCL types and it is unlikely that this small subset of patients could have significantly influenced the overall conclusion.

Our cohort includes 125 aggressive lymphomas that include mostly diffuse large B-cell lymphoma (DLBCL). To our knowledge, no evaluation of genotype versus response to monotherapy rituximab has been made for DLBCL. The response to treatment in DLBCL with R-CHOP has been found to be favorably associated with *FCGR3A*-158VV in one report by Kim *et al.*[12] but Mitrovic *et al.*[13] found no such association. Kim's report included an analysis of 85 patients treated with CHOP only, but showed an unusually high frequency (57%) of 158VV genotype and very low frequency (12%) of 158FF. The VV type did not influence PFS or OS in DLBCL treated with R-CHOP in these studies. Also, as reported by Carlotti *et al.*, FCGRIIIA polymorphism did not predict clinical outcome of FL patients treated with sequential CHOP and rituximab.[14] ADCC via FcGRIIIa receptors may not be the major mechanism of elimination of lymphoma cells in patients treated with immunochemotherapy.

We found that in indolent subtypes, VV patients presented with less bulky disease. Less bulky disease was also found in the VV type patients in a study of 144 FL patients reported by Ghelmini *et al.*[2] The study by Carlotti *et al.*[14] was unable to determine the same association, however, this may be explained by the fact that they defined bulk as greater than 10 cm and accordingly had far fewer patients with bulky disease. It is possible that there may be a biological difference in the VV genotype that influences the bulk detected at first diagnosis.

In conclusion, we found that *FCGR3A*-158V was not prognostic in the absence of monoclonal antibody therapy.

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The online version of this article contains a supplemental appendix.

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A possible role of ^{18}F-FDG positron-emission tomography scanning in the early detection of rituximab-induced pneumonitis in patients with non-Hodgkin's lymphoma

Rituximab is safe and effective in the treatment of patients with non-Hodgkin's lymphoma (NHL). Rituximab is generally well-tolerated. Its major adverse effects are infusion related and include fever, chills, dyspnea and hypotension. Dyspnea is a frequent complaint in NHL patients, and is often related to anemia or general fatique. However, it may also be the first sign of a severe underlying disease. Recently, rituximab-induced pneumonitis (RP) has been reported as side-effect of rituximab, often presenting with complaints of dyspnea.[1-11] [^{18}F]-fluorodeoxyglucose positron-emission tomography (^{18}F-FDG PET) is currently a routine modality in the early diagnosis and follow-up of NHL patients. ^{18}F-FDG PET may show abnormalities other than lymphoma activity.

We describe 4 patients with dyspnea related to RP, in which ^{18}F-FDG PET proved to be of diagnostic value. We performed a single center, retrospective case-control study of NHL patients treated with C[H]OP-rituximab for the period January 1, 2003 - April 30, 2007 (51 months) to investigate variables associated with RP and to investigate the abnormalities found on ^{18}F-FDG PET. For this case-control study, we included patients with a documented ^{18}F-FDG PET before rituximab therapy and in whom *a priori* the lymphoma response was evaluated by ^{18}F-FDG PET within six weeks after finishing the C[H]OP-rituximab therapy. Patients who received simultaneously, or in the past, chemotherapy-regimens other than C[H]OP were excluded.

RP was defined as the presence of characteristic clinical findings such as dyspnea, fever, cough, and the presence of diffuse unilateral or bilateral pulmonary activity detected by ^{18}F-FDG PET during the treatment with rituximab. Consolidations or ground-glass opacities on chest x-ray or high-resolution computed tomography (HRCT) were considered as supportive findings for the diagnosis of RP. The diagnosis RP was only made after exclusion of other causes of diffuse lung disease.

All subjects were reviewed for clinical, laboratory, and radiological characteristics. Treatment schedules were reviewed for the body surface-adjusted and cumulative dose of rituximab and for the dose-interval. In patients with an RP, an extensive search for other diseases was performed. This included analysis of blood, sputum and bronchoalveolar lavages (BAL). BAL was cultured for a broad panel of respiratory pathogens, i.e. common bacteria, *Legionella*, *Chlamydia*, *Pneumocystis*, mycobacteria, fungi and viruses such as *Adenovirus*, *Para-*, and Influenza virus. Polymerase chain reaction was performed for common viruses, *Chlamydia*, *Legionella* and mycoplasma. In addition, immunophenotyping of white blood cells in the BAL was performed. Pulmonary function and carbon monoxide diffusion tests were carried out. Chest X-ray and HRCT were reviewed for other abnormalities. ^{18}F-FDG PET were analyzed for activity pattern and maximum Standardized Uptake Values (SUV_{max}). SUV_{max} was corrected for body weight. Serial ^{18}F-FDG PET were performed to detect the time to disappearance of the increased FDG-uptake. Patients with RP were considered as cases (case group) and were compared with patients without RP (control group).

Statistical analysis was performed with SPSS 15.0 software (SPSS Inc, Chicago, Illinois, USA). The number of