

ORIGINAL ARTICLE

Activating KIR genes are associated with CMV reactivation and survival after non-T-cell depleted HLA-identical sibling bone marrow transplantation for malignant disorders

C Chen^{1,4,5}, M Busson^{2,5}, V Rocha³, M-L Appert², V Lepage^{1,2}, N Dulphy², P Haas², G Socié³, A Toubert^{1,2}, D Charron^{1,2} and P Loiseau^{1,2}

¹Laboratoire d'Immunologie et d'Histocompatibilité, Hôpital Saint-Louis, CIB-HOG, AP-HP, Paris, France; ²Inserm U662, Hôpital Saint-Louis, CIB-HOG, AP-HP, Paris, France; ³Service d'Hématologie-Greffe de moelle, Hôpital Saint-Louis, CIB-HOG, AP-HP, Paris, France and ⁴Department of Pediatrics, Hospital of Zhongshan University, Guangzhou, China

Combinations of HLA and killer immunoglobulin-like receptors (KIR) may affect outcome in T-cell depleted haematopoietic stem cell transplantation (HSCT). The KIR gene family includes inhibitory (KIR2DL and 3DL) and activating receptors (KIR2DS). Ligands are HLA-C (KIR2D) and HLA-Bw4 (KIR3DL1) for inhibitory KIR and are still unknown for activating KIR. The impact of activating KIR genotypes from donor and recipient is poorly documented in HSCT outcome. Here, HLA and KIR genotypes were determined in 131 pairs from non-T-cell depleted HLA-identical sibling HSCT. No effect of 'missing KIR ligand' was detected on acute graft-versus-host disease (GVHD), relapse, survival or infections even in myeloid malignancies. However, additional activating KIR genes in the donor compared to the recipient's genotype or an identity between donor and recipient activating KIR genotypes was associated with a lower transplant-related mortality (TRM) ($P=0.005$) and in a multivariate analysis with a better survival ($P=0.02$, HR = 0.28; $P=0.013$, HR = 0.29) and a lower incidence of cytomegalovirus (CMV) reactivation ($P=0.009$, HR = 0.36). These data highlight the impact of donor-activating KIR genes on TRM, overall survival and CMV reactivation in HLA-identical sibling HSCT.

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Introduction

The Natural Killer (NK) cells are a subset of lymphocytes of the innate immune system able to recognize and mediate cytolysis of virus-infected, neoplastic and allogeneic cells, without prior sensitization.

The induction of NK cell triggering is a balance of positive or negative signals, which determines lysis or inhibition of lysis by NK cells. NK function is induced by two structurally distinct receptor families: the leucocyte receptor complex (LRC) including killer cell immunoglobulin (Ig)-like receptors (KIR) encoded by genes located on chromosome 19q and the C-type lectin receptors located on chromosome 12p within the NK gene complex (NKG).¹

Two kinds of KIR control the NK cells: (i) the inhibitory KIRs produce a signal inhibiting killing of autologous cells, (ii) activating KIRs initiate activating signals of NK cells.

The natural ligands of several KIRs have been identified as HLA class I molecules expressed by all nucleated cells. Inhibitory KIR receptors with two Ig-like domains (KIR2DL) identify HLA-C molecules as ligands: KIR2DL1 recognizes HLA-C molecules having a lysine residue at position 80 (C2 group), whereas KIR2DL2 and KIR2DL3 recognize HLA-C molecules with an asparagine residue at position 80 (C1 group). Among the KIR with three Ig-like domains, KIR3DL1 recognizes HLA-B molecules sharing the Bw4 supertypic motif.¹ Activating KIR have poorly defined ligands and functions. Only recently have some aspects of KIR2DS1 ligand interaction been unravelled.²

The impact of NK in allogeneic haematopoietic stem cell transplantation (HSCT) has been an important issue since the observation of Cudkovic and Bennett in 1971³ that lethally irradiated mice were capable of rejecting parental strain bone marrow allografts. It was subsequently shown that NK cells were responsible for this phenomenon called 'hybrid resistance'.

In humans, Velardi *et al.* showed that, in T-cell depleted haploidentical grafts performed for acute myeloid leukaemia (AML), donor-versus-recipient NK alloreactivity could avoid leukaemia relapse and graft rejection and protect patients against GVHD.^{4,5}

Correspondence: Dr P Loiseau, Lab d'histocompatibilité et d'immunologie, Hôpital Saint-Louis, Inserm U662, 1 avenue Claude Vellefaux, 75010, Paris, France.

E-mail: loiseau@histo.chu-stlouis.fr

⁵Chun Chen and Marc Busson contributed equally to this study, and both should be considered the first author.

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Velardi *et al.*⁴ predicted NK alloreactivity when an inhibiting signal from donor KIR could be lacking due to an inappropriate HLA class I ligand in the recipient, mainly from HLA-C differences. Leung *et al.*⁶ also noted improved prediction of NK alloreactivity when considering the potential interaction of the HLA class I molecules of the recipient and the KIR molecules of the donor in children undergoing haplo-identical transplantation.

Following these reports, retrospective studies have looked for effects of KIR-ligand incompatibilities in patients transplanted with protocols distinct from the haploidentical setting and with variable levels of HLA mismatches. Giebel *et al.*⁷ supported the concept raised by Velardi *et al.*,^{4,5} Beelen *et al.*⁸ only confirmed the antileukaemic effect whereas others reported no benefit from mismatches that should have favoured NK-cell alloreactivity.^{9–11}

A recent study¹² reported an improved outcome in patients transplanted for AML and myelodysplastic syndrome (MDS) with T-depleted HLA-identical sibling bone marrow and lacking an HLA ligand for donor inhibitory KIR.

Moreover, Cook *et al.*¹³ reported a poorer survival for HLA-C2 homozygous patients with AML who received a transplant from a KIR2DS2-positive, HLA-identical sibling donor and more recently, they reported an impact of donor activating KIR on CMV reactivation.¹⁴ Despite the report of Cook, the role for activating KIR in HSCT remains undefined and should be explored especially if one considers their implication in responses to infections and in autoimmune diseases.^{1,15}

In order to evaluate the impact of both inhibitory and activating KIR on HLA-identical sibling HSCT, we typed 131 HLA geno-identical donor/recipient pairs for KIR genes and evaluated the following endpoints: acute GVHD, relapse, CMV reactivation, fungal and severe bacterial infections and survival.

Materials and methods

Patient, donor and transplant characteristics

One hundred and thirty one patients who received a nonmanipulated bone marrow transplant from an HLA-identical sibling donor between June 1995 and December 2002 were included in this analysis. All transplants were performed at the Saint-Louis hospital in Paris (France) for myeloid leukaemia AML, chronic myeloid leukaemia (CML) and MDS or for lymphoid disease (acute lymphoid leukaemia (ALL) and non-Hodgkin lymphoma (NHL)). The donor selection criteria included identity for HLA-A, -B, -DR, -DQ and -DP by HLA typing. Patient, donor and transplant characteristics are listed in Table 1. Outcome of the 131 transplants is summarized in Table 2.

Graft-versus-host disease prophylaxis, conditioning regimen and supportive therapy

Prophylaxis for acute graft-versus-host disease (GVHD) consisted of the standard combination of cyclosporine

Table 1 Patients, disease and transplant characteristics

Characteristics	n = 131
<i>Recipient</i>	
Age, median (range)	31.1 (3–57)
Female	59 (45%)
Children (<15 years)	31 (24%)
Positive CMV serology	81 (62%)
Previous auto/allogeneic transplant	2 (1.5%)
<i>Underlying diagnosis</i>	
Chronic myeloid leukaemia	28 (21%)
Acute leukaemia	84 (64%)
AML (n = 40)	
ALL (n = 44)	
Myelodysplasia	5 (3.8%)
Non-Hodgkin lymphoma	9 (6.8%)
Other malignancies	5 (3.8%)
<i>Stages of disease</i>	
Early	105 (81%)
Intermediate	12 (9%)
Advanced ^a	14 (10%)
<i>Donor</i>	
Age, median (range)	30.7 (4–59)
Female	59 (45%)
Sex match	55 (42%)
Donor female to recipient male	39 (30%)
ABO match	103 (79%)
ABO major mismatch	28 (21%)
Positive CMV serology	69 (53%)
<i>Transplant</i>	
GVH prophylaxis	
Cyclosporin + methotrexate	118 (90%)
Cyclosporin + methotrexate + other	7 (5.5%)
Cyclosporin + corticosteroids	6 (4.5%)
<i>Conditioning</i>	
Irradiation based	
TBI + Cy + others	35 (27%)
TBI + Mel + others	28 (21%)
Chemotherapy based	
Bu + Cy	56 (42%)
Bu + Cy + VP16	6 (5%)
Cy	0
Bu + others	6 (5%)

Abbreviations: AML: acute myeloid leukaemia, ALL: acute lymphoblastic leukaemia, TBI = total body irradiation, Bu = busulfan, Cy = cyclophosphamide, Mel = melphalan, VP16 = etoposide.

^aAdvanced stage: CML in blastic crisis, AML/ALL in relapse or refractory disease, NHL in resistant or untreated relapse, MDS classified as refractory anaemia with excess of blast or with excess of blast in transformation and secondary acute leukaemia (modified from IBMTR classification).

(CSA) and methotrexate (MTX) in 90% of patients (n = 118) (Table 1).

Conditioning for transplantation varied according to diagnosis and stage of disease at transplantation (Table 1).

All patients were isolated in laminar airflow rooms. Irradiated and leucocyte-depleted blood products were used for all patients. Patients received transfusions of red blood cells or platelets when the haemoglobin level was lower than 8 g/dl and the platelet count was less than $20 \times 10^9/l$, respectively. Selective gut decontamination with oral antibiotics and viral/fungal/parasitic prophylaxis was performed according to local policy. All patients received

Table 2 Main clinical HSCT end points of the 131 transplants

Outcome	Number	Mean	Median	Minimum	Maximum
Follow-up	131 (100%)	30 months	17.4	0.4	97
Death	36 (27%)	7.6 months	5.3	0.4	37
aGVHD (grade 2, 3, 4)	63 (48%)	22 days	19	7	71
Severe aGVHD	11 (8%)	26 days	20	10	50
Relapse	19 (14%)	13 months	10	1	71
First bacterial infection ^a	24 (18%)	63 days	63	0	149
First CMV reactivation ^a	51 (39%)	50 days	48	18	106
First fungal infection ^a	11 (8%)	89 days	94	7	157

Abbreviations: CMV = cytomegalovirus; aGVHD = acute graft-versus-host disease; HSCT = haematopoietic stem cell transplantation.

^aOnly the first infections occurring during the first 180 days after graft are taken into account.

acyclovir as prophylaxis for herpes virus infections. The dose of acyclovir ranged from 500 to 1000 mg/m²/day according to the donor/recipient CMV serology. Pre-emptive treatment with ganciclovir or foscarnet for CMV reactivation based on CMV antigenaemia screening was used from 1994.^{16,17}

Clinical end point definitions

Acute GVHD: Grading severity of acute GVHD was performed according to published criteria.¹⁸ All patients were considered evaluable for acute GVHD at day +1 after transplantation.

Survival was calculated from the time of transplantation to death from any cause.

Transplant-related mortality (TRM) was calculated from the time of transplantation to death related to transplantation, not to relapse.

Infection definitions. First episodes of severe bacterial, viral and invasive fungal infections were analysed up to 180 days after transplant according to the criteria described by Rocha *et al.*¹⁹

We considered severe bacterial infection to have developed when sepsis, pneumonia or septic shock were diagnosed according to previously published criteria.¹⁹

CMV reactivation was defined by a positive antigenaemia level (presence of two or more positive nuclei per 200 000 leucocytes). CMV disease was diagnosed according to previously published criteria.¹⁹

Candidaemia was defined by one or more positive blood culture for *Candida* sp. Invasive candidosis was defined by clinical and/or radiological signs of fungal infection with one or more positive blood cultures for *Candida* sp. Proven invasive fungal infection was defined by histo/cytopathology evidence of fungi from a needle aspiration or biopsy with evidence of associated damaged tissue or positive culture obtained by a sterile procedure with clinical or radiological signs consistent with infection. We also considered clinical and radiological signs of invasive aspergillosis with positive antigenaemia (but without microbiological identification) as a probable invasive fungal infection.

Gene polymorphism typing

Peripheral blood leucocytes from patients and donors were used as a source of DNA, which was extracted by the salting-out technique.

HLA typing: HLA-A and -B typings were performed using a serological method and since October 2002 by a molecular method. Molecular techniques were used for HLA-C and HLA class II typings. Serological typing for HLA-A and -B was performed using the standard microlymphocytotoxicity method with monoclonal antibodies (One Lambda Inc., Canoga Park, CA, USA), which defines 24 HLA-A and 48 HLA-B antigens. HLA-A, -B, -C, -DRB, -DQB1 and -DPB1 medium resolution molecular typings were performed using the polymerase chain reaction (PCR)-sequence specific oligonucleotide reverse dot blot kits from Innogenetics (InnoLipa DRB, -DQB and -DPB kits, Zwijndrecht, Belgium). Molecular HLA-C typing allowed classification into HLA-C1 and C2 groups. An HLA family study (parents and siblings) was performed in most cases and allowed the definition of HLA haplotypes from DPB1 (the most centromeric) to HLA-A (the most telomeric) loci, including the HLA-C locus.

Killer Ig-like receptors (KIR) typing. Eleven KIR (KIR 2DL1, 2DL2, 2DL3, 3DL1, 3DL2, 2DS1, 2DS2, 2DS3, 2DS4, 2DS5, 3DS1) were typed using PCR-sequence specific primers method as previously described. The panel of KIR-gene-specific primers was defined by Uhrberg.²⁰

Statistical analysis

Differences in categorical variables between the two groups were evaluated by χ^2 analysis. Clinical variables were: recipient age, recipient and donor genders (female donor to a male recipient versus others), recipient and donor CMV serology, ABO compatibility, diagnosis (myeloid versus lymphoid disease) status of disease (low, intermediate or high risk), conditioning regimen (use of a radiation or busulfan-based conditioning).

Continuous variables were compared between these groups by two-tailed unpaired *t*-tests. Univariate Kaplan–Meier analysis was used to describe risk factors for death. Univariate analysis using the competing risk method as described by Fine and Gray²¹ was used for assessment of prognosis factors of GVHD and infections, with death as a competing event. Cox proportional hazards model analyses were used to identify independent risk factors for death. The proportionality assumption for each of the variables contained in the final model was checked by testing the dependency of their relative risk estimate over time. Competing risk regression, as described by Fine and Gray,

was used to identify independent risk factors for infections, acute or chronic GVHD. All tests were two sided and the type 1 error rate was fixed at 0.05. The Statistical Package for Social Scientists (SPSS 12) was used for data management and analysis by Kaplan–Meier and Cox Methods. The R Package ‘cprsk’ developed by Gray was used for competing risk analysis.

Results

No influence of ‘missing KIR ligand’ in non-T-depleted HLA-identical sibling HSCT outcome

As KIR and HLA genes segregate independently, it is possible that HLA-identical siblings may inherit different KIR genes and that the donor introduces KIR for which the corresponding ligand is lacking in the recipient leading to ‘missing KIR ligand’. KIR genotyping for donors identified the presence or absence of inhibitory KIR. Recipient HLA-C and -B types were classified into the ligand groups: C1 (HLA-C^{Asn80}), C2 (HLA-C^{Lys80}) or HLA-Bw4. Receptor-ligand binding and lysis inhibition assays have shown that Bw4-positive HLA-B molecules with an isoleucine in position 80 seem to be more effective than the others. Some HLA-A molecules are also Bw4-positive (HLA-A*23, A*24, A*25, A*32). We therefore tested the influence of KIR3DL1/Bw4 incompatibilities, taking into account Bw4 or Bw4-Ile80 and HLA-A molecules expressing the Bw4 epitope.

KIR genotyping of 131 donors revealed that 96% of individuals were positive for KIR2DL1 and 85% for KIR3DL1. Forty nine percentage of individuals were positive for KIR2DL2 and 91.1% for KIR2DL3 with 100% donors positive for one or both C1 receptors. These KIR gene frequencies are in line with frequencies published elsewhere in Caucasoid populations.^{22–24} HLA genotyping showed that 83 donor–recipient pairs (63%) could be characterized by the lack of a recipient HLA ligand for donor KIR. Among the 83 pairs with a ‘missing KIR ligand’, 45 pairs had no C2 group allele in the recipient for donor KIR2DL1, 22 pairs no C1 group allele for donor KIR2DL2 and/or 2DL3 and 39 pairs no HLA-Bw4 in the recipient for donor KIR3DL1 (Table 3).

‘Missing KIR ligand’, in this cohort of patients, was not associated with any deleterious or beneficial effect on acute GVHD, relapse or overall survival, or with CMV reactivation, fungal or severe bacterial infections even if only taking into account the population of myeloid diseases (data not shown).

Association of activating KIR genes with CMV reactivation and survival in non-T-depleted HLA-identical sibling HSCT: univariate and multivariate analyses

Influence of donor KIR gene repertoire. No association was found between any individual activating KIR gene and CMV reactivation, fungal or severe bacterial infections, acute GVHD, relapse or overall survival (data not shown).

Similarly, the number of activating KIR genes present in the donor genotype without considering recipient genotype

Table 3 Characteristics of ‘missing KIR-ligand’

<i>‘missing KIR-ligand’</i>	<i>Number of pairs with KIR-ligand incompatibilities n = 83^a</i>	<i>%</i>
C2 group absent in the recipient for donor KIR2DL1	45	54
C1 group absent in the recipient for donor KIR2DL2 and/or 2DL3	22	26.5
HLA-Bw4 absent in the recipient for donor KIR3DL1	39	47

Abbreviation: KIR = killer cell immunoglobulin (Ig)-like receptors.
^a63% of total 131 transplants pairs.

was not associated with any end point of the study even for CMV reactivation.

Two broad haplotypes of KIR genes have been defined. The haplotype A carries in addition to the so-called ‘framework loci’ common to both haplotypes, a single activating KIR gene, KIR2DS4. The haplotype B is characterized by additional activating KIR loci. Therefore, haplotype B carriers potentially express multiple activating KIR.

At variance with Cook *et al.*,¹⁴ transplants performed with an AA haplotype donor had a similar CMV reactivation rate (38.2%) to those where the donor was not homozygous for haplotype A (genotype AB or BB) (39.2%). This observation held true when taking into account only the transplants where donor and recipient were seropositive for CMV.

Influence of donor KIR gene repertoire in combination with the presence or absence of ligands in the recipient. The impact of an activating KIR gene presence in the donor combined with absence of the HLA ligand for its homologous inhibitory receptor in the recipient was tested. That is, donor 2DS1 in absence of C2 ligand, donor 2DS2/3 in absence of C1 ligand or donor 3DS1 in absence of Bw4 motif in the recipient. Indeed, one combination, donor KIR2DS2 and homozygous C2C2 ligand in the recipient resulted in a higher risk of CMV reactivation (donor2DS2/recipientC2C2 versus others $P=0.012$, Fisher exact test). The two factors considered separately were not associated with an increased rate of CMV infection, emphasizing the importance of considering the combination of donor and recipient genotypes.

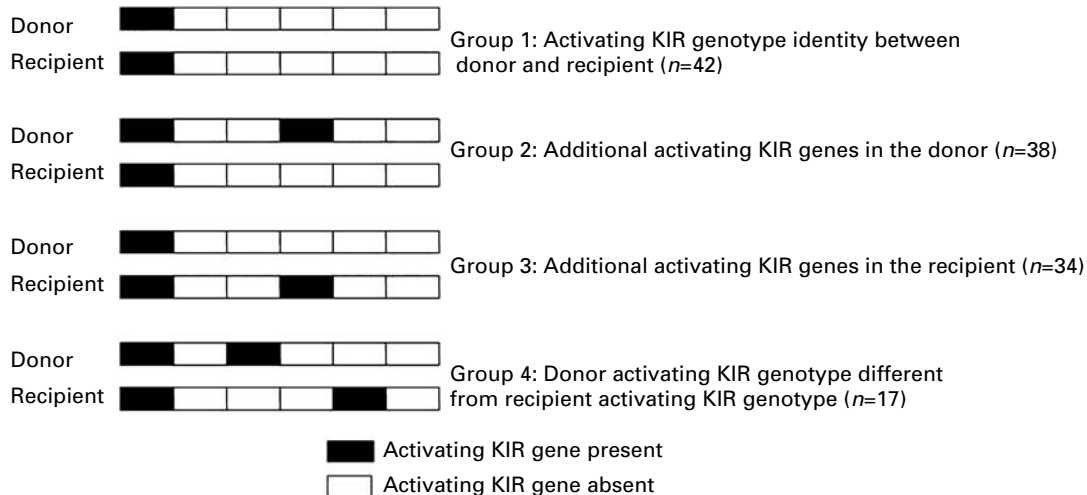
Influence of combined donor and recipient KIR gene repertoires. Considering donor and recipient repertoires together, we observed a worse survival when the donor lacked an activating KIR present in the recipient. We therefore performed an extensive evaluation of the impact of the differential expression between donors and recipients of all activating KIR on acute GVHD, survival and infections. We classified the pairs according to activating KIR genotypes of donors and recipients into four groups

(Figure 1): identical donor and recipient activating KIR genotypes (group 1, $n=42$, Figure 1), additional activating KIR genes in the donor compared to the recipient (group 2, $n=38$, Figure 1) or in the recipient compared to the donor (group 3, $n=34$, Figure 1), differences between donor and recipient activating KIR genotypes (i.e., donor and recipient genotypes with activating KIR genes absent in each other) (group 4, $n=17$, Figure 1).

No statistically significant association was found between any group of donor/recipient activating KIR genotypes and acute GVHD or relapse. However, overall survival was significantly better and TRM significantly lower in the cases with identity between donor and recipient activating KIR genotypes (group 1, $n=42$) or with additional activating KIR genes in the donor (group 2, $n=38$) (Figure 2, $P=0.015$ and Figure 3, $P=0.005$, respectively). Group 4 ($n=17$) with differences between donor and recipient activating genotypes had the worse outcome in terms of survival and TRM.

We then investigated the impact of activating KIR genotypes on infections. No impact on bacterial or fungal infections was detected. However, the cases with additional activating KIR genes in the donor (group 2, $n=38$) had a lower risk of CMV reactivation (Figure 4, $P=0.036$).

In a multivariate Cox analysis, five factors were associated with survival: ABO incompatibility ($P=0.003$, HR = 2.89), stage of disease (advanced/intermediate versus early) ($P<0.0001$, HR = 4.40), age of recipient (> 15 years versus < 15 years) ($P=0.01$, HR = 4.72), additional activating KIR genes in the donor or identity between donor and recipient activating KIR genotypes ($P=0.02$, HR = 0.28; $P=0.013$, HR = 0.29, respectively). A competing risk regression showed that 'additional activating KIR genes in the donor' was also associated with a lower rate of CMV reactivation: ($P=0.009$, HR = 0.36). These results support the concept that the combination of donor/recipient activating KIR genotypes are by themselves risk factors for survival and CMV reactivation.



Schematic representation of activating KIR genotype: each box represents an activating KIR gene

Figure 1 Mode of classification of the pairs according to the activating KIR gene contents of donor and recipient genotype.

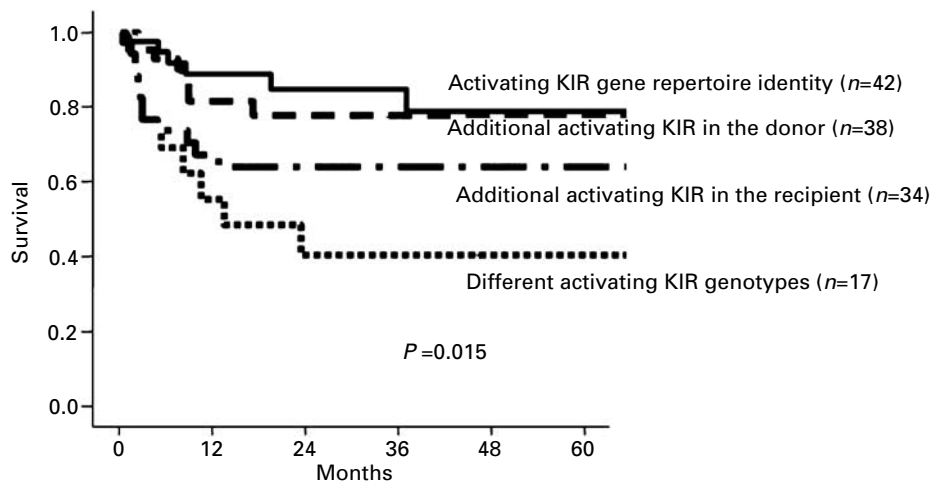


Figure 2 Overall survival according to donor/recipient activating KIR genotype classification.

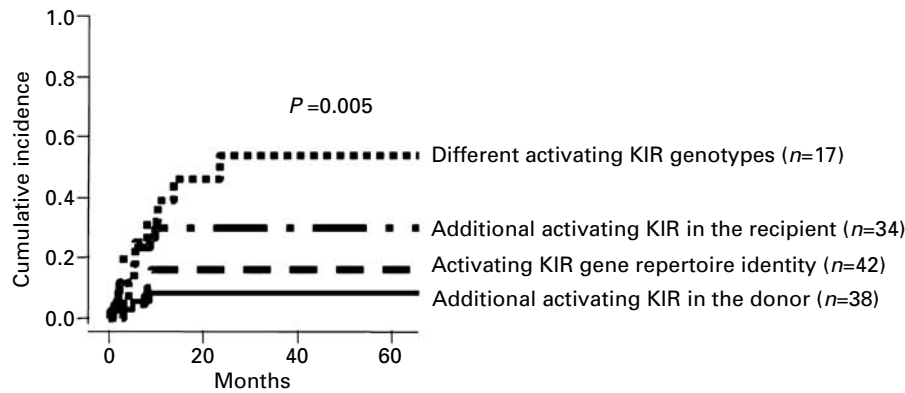


Figure 3 TRM according to donor/recipient activating KIR genotype classification.

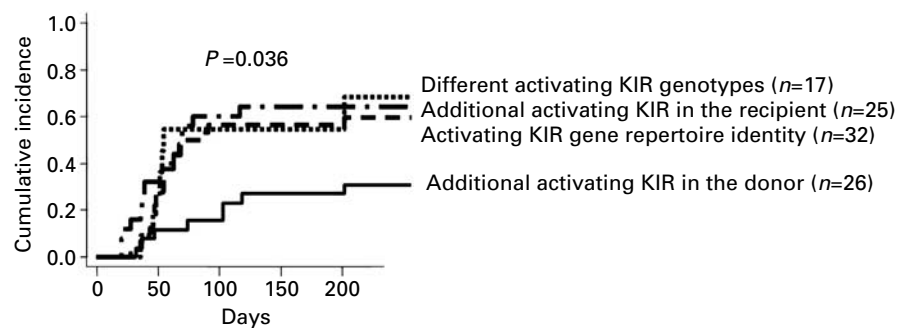


Figure 4 CMV reactivation according to donor/recipient activating KIR genotype classification during the first 180 days after graft (CMV seronegative pairs at the time of the graft were excluded).

Discussion

We describe here a comprehensive analysis of combined HLA and KIR genotypes in a homogenous group of intrafamilial geno-identical sibling HSCTs.

First, we examined the influence of donor KIR gene repertoire on acute GVHD, relapse, survival, CMV reactivation, fungal or severe bacterial diseases alone or in combination with patient HLA-C or Bw4 genotypes following a non-T-depleted HLA-identical sibling HSCT.

In contrast to previous studies,^{4,6,7,12} ‘missing KIR ligand’ was not found to be associated with any deleterious or beneficial effect on acute GVHD, relapse or overall survival even taking into account only the population of myeloid diseases.

A key issue in explaining differences between the KIR-ligand incompatibility studies in HSCT could be donor T-cell depletion of the graft or the use of antithymocyte globulin (ATG) during conditioning, or GVHD prophylaxis. All these treatments lead to *in vivo* depletion of donor T-cells during the early post-transplantation period, thus making apparent NK-cell alloreactivity. In fact, all reported studies showing a beneficial impact of KIR-ligand incompatibilities on relapse and survival are those studying transplants with donor T-cell depletion by CD34 + selection, CD3 + T-cell depletion^{4,6,12} or to the use of ATG.⁷

We focused this study on the impact of activating KIR genotypes on HSCT outcome. NK cells mediate innate

immunity against viruses, bacteria and parasites by using their inhibitory or activating cell surface receptors that regulate NK responses, but the functions and natural ligands of the activating KIR receptors are still poorly understood. However, activating KIR have been associated with several autoimmune diseases as well as with infectious diseases. Activating KIR2DS2 was found to be a risk factor for vasculitis in patients suffering from rheumatoid arthritis,²⁵ the genetic combination – presence of KIR2DS2 and absence of 2DL2 – a risk factor in scleroderma²⁶ and KIR2DS1 and/or 2DS2 in absence of the HLA ligands for their homologous inhibitory receptors (KIR2DL1 and 2DL2/3), a risk factor in psoriatic arthritis.^{27,28} KIR2DS2/KIR2DL2 was also associated with type I diabetes.²⁹

NK cells mediate innate immunity against viruses, bacteria and parasites by using their inhibitory or activating cell surface receptors that regulate NK responses. On entering infected tissue, NK cells are activated through surface receptors that sense microbial products or cellular stress. In contrast, in healthy tissue the activation pathways are kept in check by signals coming from inhibitory receptors. Recent work has demonstrated that a murine analog of activating KIR, Ly49H, recognizes the gene product m157 from murine cytomegalovirus (CMV) and that inheritance of Ly49H is the major determinant of susceptibility or resistance to murine CMV infection in different mouse strains.³⁰ In humans, it has been reported that the activating KIR3DS1, in combination with HLA-B

Bw4-80Ile + could be associated with delayed progression to AIDS³¹ and that genes encoding the inhibitory KIR 2DL3 receptor and its HLA-C1 ligand could directly influence the resolution of low-dose hepatitis C virus (HCV) infection.³² The hypothesis for the latter association is that the inhibitory control conferred by KIR2DL3 and HLA-C1 interaction is weaker than others, and therefore this inhibitory control would be more easily overwhelmed by activating signals. Therefore, it may well be that the true ligands for activating KIRs may include virally encoded products.

Two aspects have been considered in this study. First, the analysis of KIR/ligand interactions showed that donor KIR2DS2 and homozygous C2 typing in the recipient could be associated with an increased risk of CMV reactivation. This is in agreement with the data from Cook *et al.*¹³ showing an increased TRM and a lower survival rate in such a KIR/ligand combination. Second, we analysed the influence of combined activating KIRs from donor and recipient on HSCT outcome. Recently, Cook *et al.*¹⁴ observed that sibling transplants, where both donor and recipient were CMV seropositive and where donors had more than one activating KIR gene, were associated with a 65% reduction in CMV reactivation. In our cohort of patients, we also evidenced a role for donor-activating KIR in genetic susceptibility to CMV reactivations but only when considered in combination with recipient activating genotypes. Additionally, we evidenced the impact of donor/recipient KIR genotypes on TRM and survival.

A beneficial effect of a higher number of KIR genes in the donor¹⁴ and the importance of taking into account donor and recipient KIR genotype²³ have been previously emphasized. These observations are consistent with the data presented here, which, in addition, emphasize the role of activating KIR in the control of CMV reactivation, a major complication after allogeneic HSCT.

In fact, the impact of donor/recipient KIR genotypes was restricted to activating genes and not observed with the inhibitory donor/recipient KIR genes. As inhibitory and activating KIR genes are in frequent linkage disequilibrium, this would suggest a direct functional role for activating KIR. Data recently obtained for KIR2DS1 ligand recognition² support that activating and inhibitory KIR recognize the same sets of self-MHC class I molecules, differing by their binding affinities. KIR2DS2-HLA-C interaction appears of very low affinity and rather peptide independent. Nonetheless, the data reported here and in autoimmune diseases, suggest that this receptor could be of importance in the fine control of NK activation.

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