Histologic and Molecular Microscope determinants of graft survival in the INTERHEART study: the greater importance of T cell-mediated rejection vs. antibody-mediated rejection in graft loss

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INTRODUCTION

In both heart and kidney transplants, rejection is a major cause of graft loss. In kidneys, the principal risk is antibody-mediated rejection (JCI Insight 2 (12), 201710.1172/jci.insight.94197), and molecular rejection predicts better than histologic diagnosis (JASN 26 (7):1711-1720, 2015). Similar comparisons in a heart transplant endomyocardial biopsy (EMB) population have not been performed.

METHODS

The INTERHEART population consists of 889 indication and protocol EMB single bites biopsies (455 patients) from 8 centers in Canada, USA, Australia and Europe. Affymetrix hgu219 microarray chips were used to study gene expression. Follow-up data (> 6 days post-biopsy) was available for 434 patients. One random biopsy was selected from each of these patients for analysis.

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RESULTS

There was a trend toward more failures after histologic TCMR but this was not significant.

Graft failure occurred in 55 of the 434 patients (diagnoses are shown in Table 4). The breakdown of biopsies by center is listed in Table 1. The median follow-up after biopsy (in surviving grafts) was 451 days (mean 895). The 434 biopsies were mainly for indications, at a median time post-transplant of 351 days (mean 1079, range 0-10150). Diagnoses were available in 885 out of 889 biopsies (Table 2). We analyzed rejection by unsupervised archetype analysis using kidney-derived rejection-associated transcripts (J Heart Lung Transplant 36:1192-1200, 2017) or by ISHLT histologic diagnosis. Survival analysis using one random biopsy per patient in 3-cluster versus 4-cluster is shown in Figure 1. Latest biopsy per patient was used in the survival analysis in Figure 2. The strongest short term risk for failure was TCMR, both in the 3AA model (Figure 1A) and the 4AA model (Figure 1B). The univariate factors: the failures were best predicted by the molecular TCMR score and to a lesser extent by the injury score S4injury (Figure 1B). Similar results were found with last biopsy per patient, both in MMDx (Figure 2A) and histology (Figure 2B). In univariate analysis, TCMR and injury transcripts (IRRAT) were significant (as well as LVEF) (Table 3).

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Table 1: Biopsies by center		
A Coruna		92
Bologna		201
Los Angeles - Cedars Sinai		51
Edmonton		113
Paris:		255
	France	*1*
	France - Bordeaux	*1*
	France - CHU Rouen	*9*
	France - HEPG	*94*
	France - Nantes	*11*
	France - Necker	*7*
	France - Paris	*108*
	France - Pitie	*24*
Sydney		92
Los Angeles - UCLA		7
Vienna		76
Virginia-VCU		2

Biopsy type as listed:

Cause	2
EMB	11
Follow Up	24
Follow Up (For Cause)	13
Follow Up (Protocol)	3
For Cause	117
Protocol	606
Unknown	1
NA	112

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Table 2: Diagnoses available in 885 EMBs with histology diagnoses

Diagnoses* (% of known diagnoses)		**Histology**	**MMDx**
No Rejection		334 (38%)	724 (82%)
ABMR Related	**ABMR**	51 (6%)	78 (9%)
	pABMR	63 (7%)	13 (1%)
TCMR Related	**TCMR**	84 (9%)	59 (7%)
	pTCMR	273 (31%)	5 (1%)
Other	**ABMR/TCMR (Mixed)**	9 (1%)	6 (1%)
	pABMR/pTCMR	71 (8%)	0
DSA Status at most recent biopsy‡		**(455 patients)**	
Positive		143 (31%)	
Negative		239 (53%)	
Unknown		73 (16%)	

* Translating ISHLT classes into a rejection/possible rejection (TpT/ApA) classification:
pAMR1, pAMR1I+, pAMR1H+……..Possible ABMR (pABMR);
pAMR2, pAMR3……..ABMR;
TCMR1……Possible TCMR (pTCMR);
TCMR2, TCMR3……TCMR

‡ The most recent DSA status at time of most recent biopsy was used, if known. DSA statuses dated more than 14 days after the biopsy were not considered. If the most recent DSA status at time of biopsy was not known, but the patient was most recently PRA negative, the DSA status was presumed negative.

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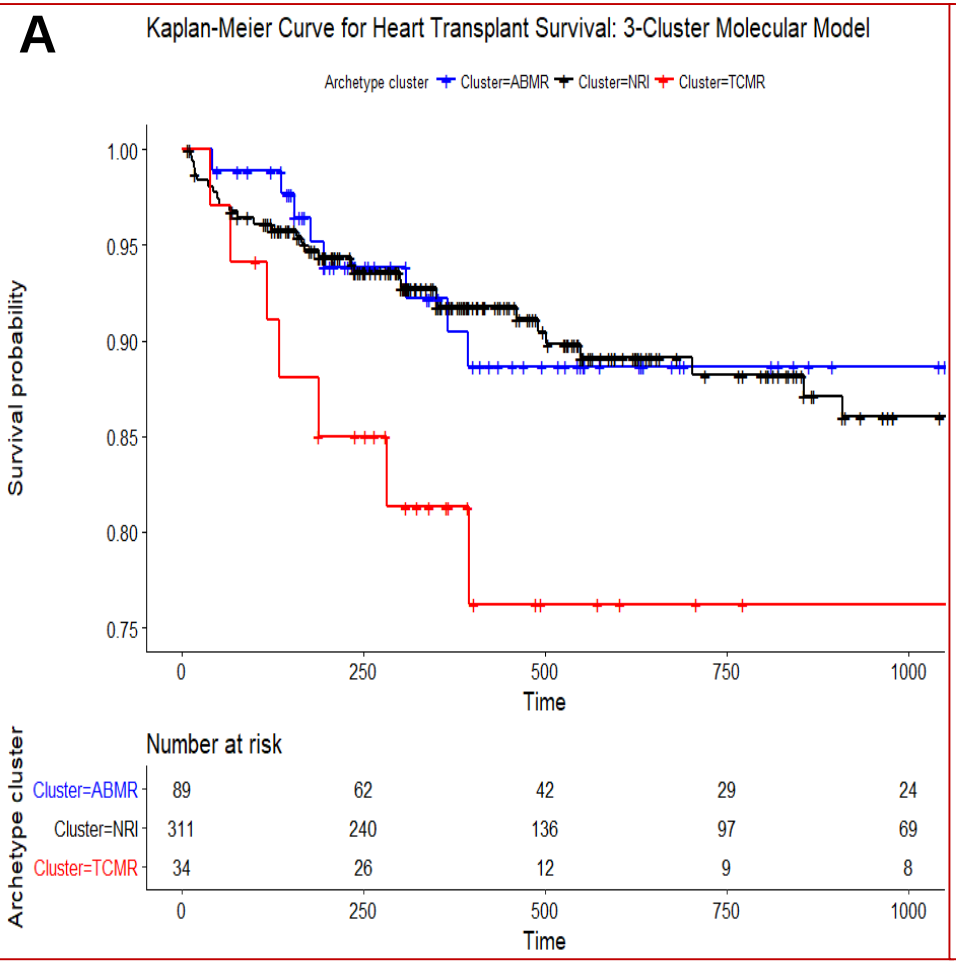

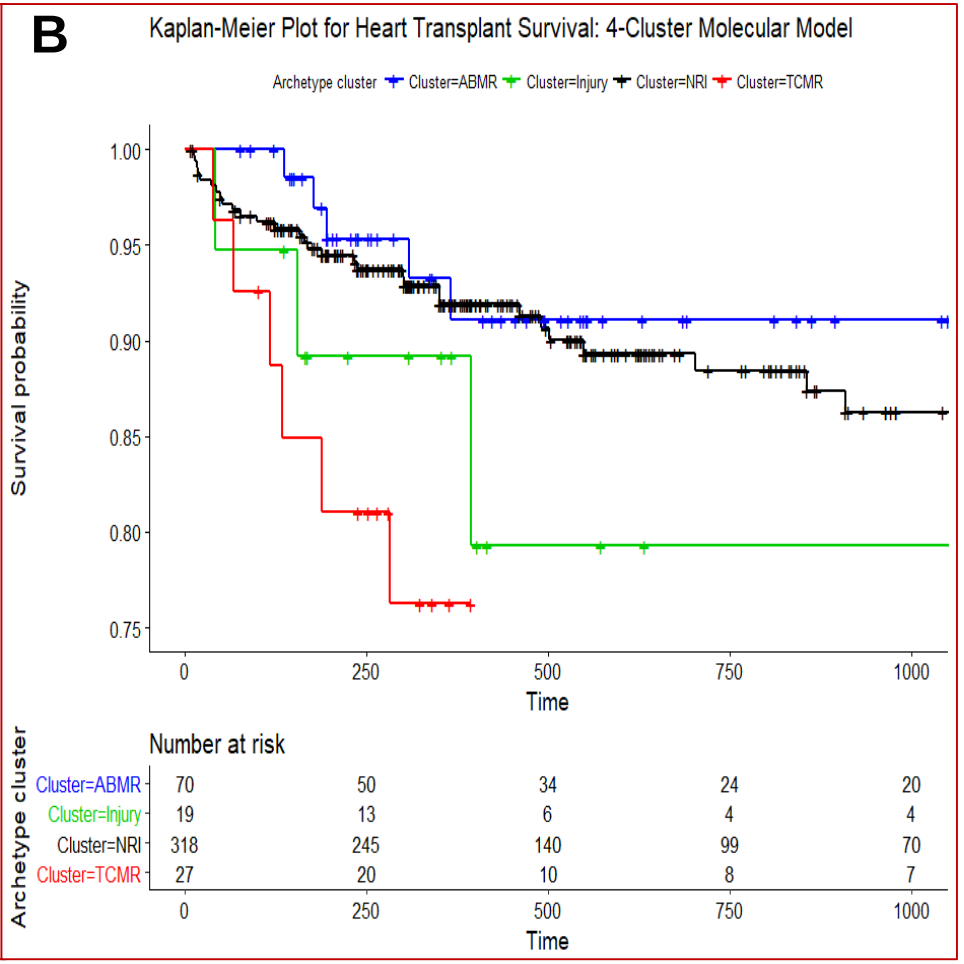

Figure 1. Kaplan-Meier survival curves for 434 patients. One biopsy per patient is selected at random for use in the analyses (N=342), and is categorized according to its highest molecular score: $S1_{\text{No Rejection}}$(RATCluster=1), $S2_{\text{TCMR}}$(RATCluster=2), and $S3_{\text{ABMR}}$(RATCluster=3).

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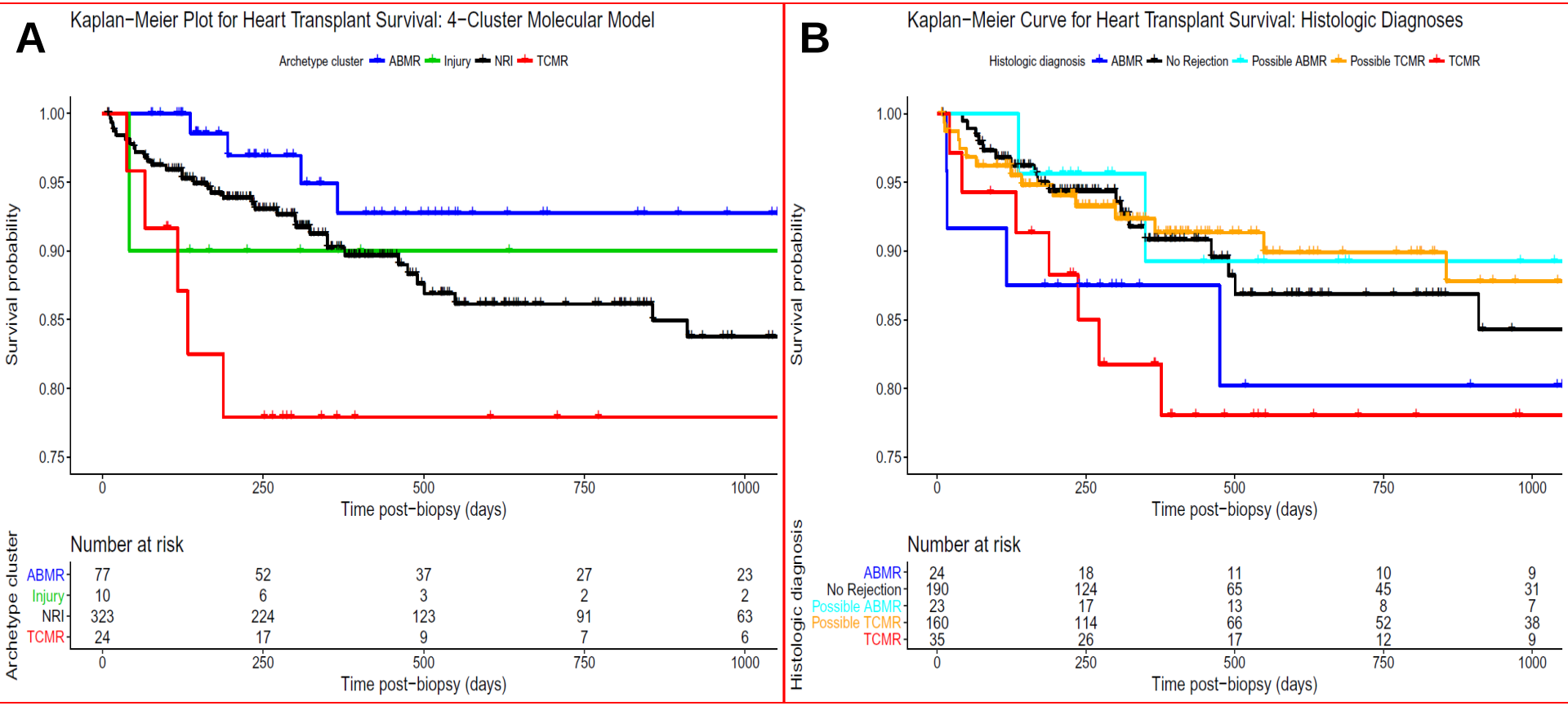

Figure 2. Kaplan-Meier survival curves for 434 patients. Last biopsy per patient is selected. Survival curves are categorized according to its highest molecular score: $S1_{NRI}$(RATCluster=1), $S2_{TCMR}$(RATCluster=2), and $S3_{ABMR}$(RATCluster=3) and $S4_{injury}$(RATCluster=4) and by histologic diagnoses: antibody-mediated rejection (ABMR), T cell-mediated rejection (TCMR), no rejection, possible ABMR and possible TCMR.

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Table 3: Significant univariate associations of future failure (last biopsy)

Variable	Mechanism	HR	p-value
Ejection fraction (LVEF)	injury	0.95	0.00015
NK cell burden (ABMR)*	ABMR*	0.14*	0.001
3AA $S2_{TCMR}$ score	TCMR	4.59	0.01
4AA $S2_{TCMR}$ score	TCMR	5.26	0.02
Injury transcripts (IRRAT)	Injury	1.94	0.045
Time of biopsy (log10 TxBx)	time of biopsy	1.51	0.047

* Note the effect of ABMR-related variables is slightly protective

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Table 4: PFH diagnoses compared to histology in N=55 failures in last biopsy per patient

	PFH diagnoses (N=55)	**Histology (N=55)**
No rejection	44	20
TCMR	7	7
pTCMR	1	14
ABMR	2	5*
pABMR	1	6
ABMR/TCMR (Mixed)	-	-
pABMR/pTCMR	-	3

* Some histologic ABMR were not confirmed by MMDx.

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CONCLUSION

1. In the latest analysis of graft loss during follow-up after INTERHEART biopsies, there are 55 failures in 455 patients. Because these were mainly protocol biopsies, most losses followed biopsies showing no rejection.
2. Low LVEF is an important determinant of risk.
3. The principal rejection finding in biopsies followed by early loss was TCMR, particularly molecular but also histologic.
4. Multivariable analysis is weak because number of failures is too small, but indicates the hazard of molecular TCMR outweighs histology TCMR.
5. In one random biopsy per patient, the univariate analysis indicates the principal risk factors related to molecular TCMR and injury; ABMR is weakly protective.
6. When only the last biopsy is considered, molecular TCMR remains the principal risk factor for failure in the follow-up period (plus LVEF) but long term follow-up is necessary to show the risk of factors such as ABMR.

DISCLOSURE

Dr Jeff Reeve has no relevant disclosures
Dr Phil Halloran has shares in TSI, University of Alberta research company