Effect of rituximab on anti-donor T cell responses
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Dear Editors,

We read with interest the paper by Schachtner et al in the March issue of Transplant International [1], reporting that a) the presence of anti-donor IFNγ producing T cells pre-transplantation, measured by ELISPOT, was predictive of T cell mediated rejection (TCMR) particularly after ABOi transplantation, but less so after ABOc transplantation and b) a reduced proportion of patients developed anti-donor IFNγ producing T cells after ABOi compared to after ABOc. They postulated several reasons for their findings, including the impact of rituximab used during their ABOi induction protocol.

We believe our published observations using ELISPOT are pertinent to help interpreting Schachtner's data. We've documented anti-donor specific IFNγ production by CD4+ T cells by ELISPOT, in 2 cohorts of transplant recipients with chronic rejection [2-4]. By adding whole donor protein preparations into CD8-depleted PBMC, the ELISPOT assays in our studies measured the activity solely of the sensitized indirect pathway of allorecognition. Although Schachtner et al used whole donor PBMC, so were potentially measuring a combination of direct and indirectly reactive anti-donor cells, the data they present suggest that they too were mostly detecting indirect alloreactivity. TCMR involves T cells with both direct and indirect allospecificity [5]. We also compared the IFNγ produced when CD25+ regulatory T cells or CD19+ B cells were depleted from PBMC, to infer the functional impact of these cells on anti-donor responses.

We showed that B lymphocytes play an important role as antigen presenting cells for IFNγ production by indirect pathway anti-donor T cells [2]. However, responses were complex, with evidence in some patients that IFNγ production was regulated either by CD4+CD25+CD39hi regulatory T cells or CD10+CD24+CD38+ transitional B cells. We could associate these patterns of anti-donor responsiveness with rate of deterioration in graft function in both cohorts [3, 4], suggesting that the ELISPOT was measuring activity of immune cells involved in damaging the graft. Importantly we showed that optimising immunosuppression could stabilise graft function, in association with either increased regulation of anti-donor activity by Tregs or transitional cells or complete suppression of activity. In contrast, we found that rituximab, which depleted >95% of all circulating B cells for a prolonged period, led to a prolonged reduction in the relative proportion of transitional B cells but maintained the relative proportion of memory B cells. Thus, it abolished all evidence of B cell regulation in ELISPOTS, but did not reduce the proportion of ELISPOTS showing B cell-dependent anti-donor IFNγ production.

We believe these findings are highly pertinent and provide a context that helps understand those reported by Schachtner et al. By depleting most B cells for a prolonged period, rituximab removes an important antigen presenting cell population for indirect alloreactive T cells, explaining the reduction in de novo anti-donor activity post ABOi transplantation. But by selectively reducing the relative proportion of transitional B cells but not memory B cells, rituximab also potentially enhances the relative strength of pre-existing anti-donor IFNg indirect pathway alloresponses capable of mediating TCMR.

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