Ishida *et al.* reply:

We thank Ferguson and Stuart for their comments and appreciate the opportunity to respond. Our data show that leukocytes mediate physiological retinal vascular pruning and pathological vaso-obliteration. These effects were demonstrated in both CD18-deficient mice and in rats treated with antibody to CD18 (ref. 1). The results, therefore, transcend a single species.

Our data also identify a role for FasL as an apoptosis-inducing molecule in vascular pruning and vaso-obliteration. We generated the data by antibody-based blockade[1] and did not rely on *gld* mice. In the discussion of our paper[1], we noted the possibility of the existence of redundant molecular or cellular mechanisms in the processes of pruning and vaso-obliteration.

Although our data and the data obtained from *gld* mice[2,3] are seemingly contradictory, they may in fact be complementary. Additional studies are required to confirm this. The phenomena that Ferguson and Stuart describe in *gld* mice are similar to those observed in our experiments. In our CD18-deficient mice, leukocyte adhesion and vascular pruning were delayed by approximately two days, but were not indefinitely suppressed. Thus, the retinal vasculature of the adult CD18-deficient mice was essentially normal[1], as was the neural retina. We have learned that the timing of the analyses is critical, as vascular modeling occurs quickly. When placed in a hyperoxic environment for 72 hours, the CD18-deficient mice showed vaso-obliteration similar to that seen in wild-type mice (S.I., K.Y., T.U. and A.P.A., unpublished data), but significantly less vaso-obliteration was seen in CD18-deficient mice at 48 hours[1].

Both vascular pruning and vaso-obliteration may represent defense mechanisms aimed at protecting the sensory retina from oxidative stress. As noted in our paper, compensatory systems may be used *in vivo* to preserve this important defense mechanism. We hypothesize that the *gld* mice data[2,3] reflect this biological redundancy. FasL blockade did not suppress leukocyte adhesion in our experiments[1]. Under prolonged oxidative stress, we speculate that known leukocyte products, such as perforin and TRAIL, have a similar role in endothelial cell apoptosis in *gld* mice. We also noted[1] that FasL-bearing blood-borne cells of nonleukocytic origin may have a role in pruning and vaso-obliteration[4,5]. It is therefore possible that leukocyte cytotoxicity does not depend on a single molecular pathway, and that FasL expression in the retina is not from a single cell type. A temporal examination of the role of blood-borne cells in pruning and vaso-obliteration is required in the genetically altered mice that Ferguson and Stuart describe.

The fact that *gld* mice show increased pathological neovascularization after hyperoxia-induced retinal ischemia[2,3] is also consistent with our published data[6]. In our model of oxygen-induced retinopathy, T lymphocytes served as negative regulators of pathological neovascularization.

COMPETING INTERESTS STATEMENT

The authors declare competing financial interests (see the *Nature Medicine* website for details).

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