TSLP-mediated fetal B lymphopoiesis?

To the editor:

In the August 2003 issue of *Nature Immunology*[1], as well as in later studies[2], Voβhenrich *et al.* presented experimental data on the cytokine requirements of developing B cells, which led them to conclude that "TSLP [thymic stromal lymphopoietin] is the factor responsible for most of the fetal and perinatal B cell production that takes place when the IL-7–γc [interleukin 7–common γ-chain] signaling pathway is disrupted."[1] Although the data reported were technically sound and compatible with such a conclusion, the authors did not provide direct evidence to support (or exclude) the idea of a critical function for TSLP in IL-7-independent fetal B lymphopoiesis. The conclusions of Voβhenrich *et al.* were based on the demonstration that B lymphopoiesis was much more affected (tenfold more) in mice deficient in IL-7 receptor α-chain, essential for IL-7 as well as TSLP signaling, than in mice deficient in the common γ-chain (γc), required for IL-7 but not TSLP-mediated signaling[3,4]. However, these data could at best be considered strong indirect support for the idea of TSLP as the main cytokine driving IL-7-independent fetal B lymphopoiesis, as there could be other reasons for a difference in the phenotypes of γc-deficient mice and those deficient in the IL-7 receptor α-chain. Furthermore, Voβhenrich *et al.* used bone marrow of mice 4–12 weeks of age, not fetal liver, for their comparative *in vivo* analysis of B lymphopoiesis in these mice[1,2]. Instead, the extrapolation to the idea that TSLP is key to the fetal stages of B lymphopoiesis was based on the finding that fetal but not adult pro–B cells were responsive to TSLP *in vitro*[1,2]. In contrast, a lack of an important function for TSLP in adult B lymphopoiesis has been indicated by studies of TSLP receptor–deficient ($Tpte2^{-/-}$) mice[5,6].

As fetal lymphopoiesis had not been examined in singly deficient $Tslp^{-/-}$ or $Tpte2^{-/-}$ mice, we investigated B lymphopoiesis in the livers of $Tpte2^{-/-}$ mice at embryonic day 17.5 but found no deficiency in $Tpte2^{-/-}$ fetuses at any stage of B cell development (**Fig. 1a** and **Supplementary Fig. 1** online). Furthermore, when comparing B lymphopoiesis in the fetal livers of $Il7^{-/-}$ and $Il7^{-/-}Tpte2^{-/-}$ mice, we obtained no evidence for substantial involvement of TSLP in IL-7-independent regulation of fetal pro–B cells or pre–B cells, whereas we noted a slight additional reduction in the number of immature B cells in $Il7^{-/-}Tpte2^{-/-}$ fetuses relative to that in $Il7^{-/-}$ fetuses (**Fig. 1b** and **Supplementary Fig. 1**). Thus, although Voβhenrich *et al.* provided compelling evidence that fetal pro–B cells are highly responsive to TSLP[1], our studies of $Tpte2^{-/-}$ and $Il7^{-/-}Tpte2^{-/-}$ fetuses fail to support their claim that TSLP is the most important cytokine promoting IL-7-independent fetal B lymphopoiesis. Instead, although Voβhenrich *et al.* also concluded that "Flk-2 is involved, but TSLP is the main factor driving IL-7-independent fetal and perinatal lymphopoiesis,"[1] we have done additional studies of mice deficient in the cytokine Flt3L (also called Flk-2 ligand) and IL-7 ($Flt3l^{-/-}Il7^{-/-}$ mice) and of $Flt3l^{-/-}Tpte2^{-/-}$ mice and have found that the reported complete loss of B-1 as well as B-2 B lymphopoiesis in $Flt3l^{-/-}Il7r^{-/-}$ mice[7] and $Flk2^{-/-}Il7r^{-/-}$ mice[1] is entirely due to the simultaneous loss of function of IL-7 and Flt3L (C.T.J. and S.E.W.J., unpublished observations). Collectively, our findings suggest that Flt3L rather than TSLP is the key regulator of IL-7-independent B lymphopoiesis and that intact TSLP function is insufficient to restore any detectable B lymphopoiesis in the absence of these two critical regulators of B cell progenitors.

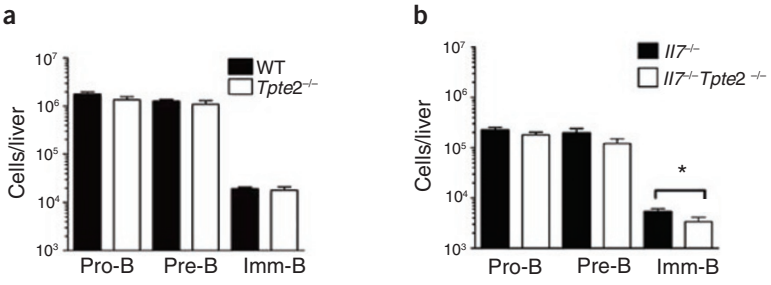

Figure 1 Critical function for IL-7 but not TSLP in the regulation of fetal B cell progenitors. Mean numbers (+ s.d.) of pro–B cells (Pro-B; $B220^+CD43^+AA4.1^+CD19^+IgM^-$), pre–B cells (Pre-B; $B220^+CD43^-AA4.1^+CD19^+IgM^-$) and immature B cells (Imm-B; $B220^+CD43^-AA4.1^+CD19^+IgM^+$) in fetal livers at embryonic day 17.5 for littermate wild-type mice (WT; $n = 8$) and $Tpte2^{-/-}$ mice ($n = 4$; each from three litters; **a**) and for littermate $Il7^{-/-}$ mice ($n = 10$) and $Il7^{-/-}Tpte2^{-/-}$ mice ($n = 10$; each from five litters; **b**). *, $P = 0.04$. Corresponding flow cytometry plots are in **Supplementary Figure 1**.

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Note: Supplementary information is available on the Nature Immunology website.

COMPETING INTERESTS STATEMENT
The authors declare no competing financial interests.

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