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EDITORIAL

The Expanding Spectrum of NFkB1 Deficiency

Vanessa L. Bryant[1,2] · **Stuart G. Tangye**[3,4]

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To the Editor,

In the last 10 years, the explosion of novel genetic etiologies of primary immunodeficiencies (PIDs) has begun an evolution from a traditional broad descriptive diagnosis to a more precise gene-based diagnosis. The most prevalent PID is common variable immunodeficiency (CVID), affecting 1:25,000 individuals. CVID is a heterogeneous collection of disorders, typically diagnosed in early adulthood, with the underlying common feature of hypogammaglobulinemia and poor antibody responses to vaccination. However, 25–30 % of patients also exhibit autoimmune and/or lymphoproliferative manifestations. A genetic defect responsible for CVID has been identified in only a small proportion (10–15 %) of all patients and include mutations in genes encoding T and B cell signaling receptor molecules, such as *ICOS*, *CD19*, *CD81*, *TNFRSF13C* (encoding BAFF-R), *MS4A1* encoding CD20 [1] and, more recently, *NFKB1* [2] and *NFKB2* [3].

Identification of the genetic basis of rare PIDs by approaches such as linkage analyses, sequencing of candidate genes and more recently via whole exome and whole genome sequencing have not only impacted diagnosis and management of patients, but also increased our understanding of the pathogenic molecular mechanisms underlying these conditions. In terms of antibody deficiencies, it is now appreciated that this is not strictly an intrinsic B cell defect that causes disease, but rather a defect in T follicular helper cell generation and/or function, as demonstrated in patients with mutations in genes encoding CD40L, ICOS, and SLAM-associated protein (SAP) [4]. Genetic disorders have also informed us of the non-redundant role of many molecular components of the immune responses to specific pathogens. For example, individuals with mutations in *SH2D1A* (encoding SAP) or *BIRC4* (encoding XIAP), causing X-linked lymphoproliferative (XLP) disorders type 1 and 2, respectively, are exquisitely susceptible to EBV-induced disease, and often develop B-cell lymphoma [1].

The NFkB signal transduction pathway comprises five family members: NFkB1 (p105/p50), NFkB2 (p100/p52), RelA (p65), RelB, and c-Rel; all of which share a common Rel homology domain (RHD) and multimerise in combinations to induce transcriptional activation of genes, including those encoding cytokines, chemokines, survival proteins, and surface receptors to direct normal cellular and humoral immune responses [5]. Loss-of-function mutations in genes encoding molecules within the NFkB signaling pathway—such as *IKBKG* (encoding NEMO), *IKK2* (encoding IKKβ), *MALT1*, *BCL10*, *CARD11*, and *NIK*—have been associated with both combined immunodeficiencies (CID) and antibody defects [1]. Recently, the first report of autosomal dominant NFkB1 deficiency was published, describing p50 haploinsufficiency in three families, each with a unique mutation in the RHD, with two mutations causing in-frame splicing and the third mutation resulting in a severely truncated protein due to the introduction of a premature stop codon [2]. The clinical features described were variable, but included adult-onset

✉ Stuart G. Tangye
s.tangye@garvan.org.au

[1] Immunology Division, Walter and Eliza Hall Institute of Medical Research, Parkville, Victoria 3052, Australia

[2] Department of Medical Biology, University of Melbourne, Parkville, Victoria 3010, Australia

[3] Immunology Division, Garvan Institute of Medical Research, Darlinghurst, NSW, Australia

[4] St Vincent's Clinical School, UNSW Australia, Sydney, Australia

hypogammaglobulinemia and sinopulmonary infections, with liver disease, lymphadenopathy, and alopecia also reported in some patients [2].

In this issue of the Journal of Clinical Immunology, Boztug et al. describe a young adult patient who presented with hypogammaglobulinemia, parapharyngeal abscesses, cervical lymphadenopathy and—most strikingly—EBV-driven lymphoproliferation [6]. Laboratory investigations revealed impaired T cell function in response to tetanus antigen and TCR stimulation and reduced memory B cells, a phenotype resembling classic CID [6]. While no mutations were identified in known EBV-associated genes, a custom next generation sequencing panel that assessed 419 known pathogenic as well as additional candidate PID genes revealed a novel heterozygous mutation in *NFKB1* that results in a severely truncated non-functional protein, similar to one described in the original report of NFkB1 deficiency by Fleigauf et al. [2]. This novel mutation affected the RHD of p105 protein, resulting in a premature stop codon. The authors demonstrated that the mutation was also present in the proband's father, who had a milder clinical phenotype, consistent with a diagnosis of probable CVID. Peripheral blood mononuclear cells from both individuals demonstrated severe reductions in expression of p105 and p50, and the complete absence of mutant p50 following mitogenic stimulation [6].

This report by Boztug et al. [6] extends the clinical and immunological phenotype of NFkB1 deficient patients, thereby expanding the spectrum of this newly described rare PID. And while EBV-induced lymphoproliferation was not previously reported for individuals with p50 haploinsufficiency [2], the current finding suggests an important role for canonical NFkB signaling in controlling EBV infection and subsequent lymphoproliferation [6]. The susceptibility to EBV-induced lymphoproliferation exhibited by this patient may be a direct result of the impaired T cell responses described by the authors. Interestingly, mutations in *CD27* cause severe EBV-induced disease, and signaling through CD27 activates NFkB [7]. Thus, it is tempting to speculate that NFkB1 deficiency compromises CD27-mediated T cell activation, and this contributes to the EBV-induced pathology noted in the proband in this report. Similarly, given the important role of NFkB1 in lymphocyte development and effector function [5], these findings also suggest that hypogammablobulinemia in NFkB1 deficiency could result from respective intrinsic defects in B cells and T cells.

The considerable disease heterogeneity displayed by individuals with NFkB p50 haploinsufficiency highlights the requirement for appropriate gene regulation for sufficient immune responses. While there is a potential role for modifier genes and epigenetic modifications in affected/more severely affected individuals, this may simply be reflective of a gene dosage effect in different family members. In any case, the variable phenotypes observed within the four families so far described with NFkB1 deficiency [2, 6] alone highlights the context-dependent function of NFkB signaling and the need for application of tailored personalized therapies.

Finally, the findings of the study presented by Boztug et al. [6] further underscores the clinical utility of next generation sequencing approaches as part of standard clinical investigations for individuals with suspected PID. It remains to be seen if defects in NFkB signaling account for a significant proportion of predominant antibody and combined immunodeficiencies. It is clear that only with continued molecular analysis of PID patients will understand the full spectrum of disease severity and potential susceptibilities of CVID and CID. A deeper understanding of the pathogenesis of such monogenic disorders will inform our understanding of more complex immune disorders, mechanisms of immune responses, and provide opportunities for novel therapies.

References

1. Picard C, Al-Herz W, Bousfiha A, Casanova JL, Chatila T, Conley ME, et al. Primary immunodeficiency diseases: an update on the classification from the International Union of Immunological Societies Expert Committee for Primary Immunodeficiency 2015. J Clin Immunol. 2015;35(8):696–726.
2. Fliegauf M, Bryant VL, Frede N, Slade C, Woon ST, Lehnert K, et al. Haploinsufficiency of the NF-kappaB1 subunit p50 in common variable immunodeficiency. Am J Hum Genet. 2015;97(3):389–403.
3. Chen K, Coonrod EM, Kumanovics A, Franks ZF, Durtschi JD, Margraf RL, et al. Germline mutations in NFKB2 implicate the noncanonical NF-kappaB pathway in the pathogenesis of common variable immunodeficiency. Am J Hum Genet. 2013;93(5):812–24.
4. Tangye SG, Ma CS, Brink R, Deenick EK. The good, the bad and the ugly—TFH cells in human health and disease. Nat Rev Immunol. 2013;13(6):412–26.
5. Oeckinghaus A, Hayden MS, Ghosh S. Crosstalk in NF-kappaB signaling pathways. Nat Immunol. 2011;12(8):695–708.
6. Boztug H, Hirschmugl T, Holter W, Lakatos K, Kager L, Trapin D, et al. NF-kB1 haploinsufficiency causing immunodeficiency and EBV-driven lymphoproliferation. J Clin Immunol. 2016;36(6). doi:10.1007/s10875-016-0306-1
7. Alkhairy OK, Perez-Becker R, Driessen GJ, Abolhassani H, van Montfrans J, Borte S, et al. Novel mutations in TNFRSF7/CD27: clinical, immunologic, and genetic characterization of human CD27 deficiency. J Allergy Clin Immunol. 2015;136(3):703–12 e10.