GPR55, a key player for B cell-mediated atheroprotection

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B cells play an essential role in regulating atherogenesis. A new study shows that the orphan receptor GPR55 is a pivotal modulator of B cell maturation and immunoglobulin production. This new finding identifies a previously uncharacterised protein in the field of atherosclerosis regulation by the immune system.

Main text

Atherosclerosis is the major cause underlying cardiovascular events, and it remains asymptomatic for long periods of time. Therefore, identifying novel diagnostic and therapeutic targets for this disease remains a burning clinical need [1]. Both the innate and the adaptive arms of the immune response have a crucial role in the development of atherosclerosis in complex and intertwined ways [2] [3]. In terms of the antibody immune response, distinct B cell subsets have been assigned pro-atherogenic or atheroprotective roles [4] [5]; likewise, some antibodies have been associated with atherosclerosis progression, whereas others provide protection [6] [5] [7] [8]. The potential for exploiting these responses for clinical purposes is increasingly being explored [6] [9]. However, important mechanistic insights regarding B cell activation and the antibody immune response in the context of atherosclerosis remain unknown. In this issue of *Nature Cardiovascular Research*, Guillamat-Prat and colleagues [10] move the field of atherosclerosis forward by showing that G protein-coupled receptor 55 (GPR55) in B cells provides atheroprotection.

GPR55 is a G protein-coupled cannabinoid receptor expressed on various immune cell types. Within the B cell lineage, plasma cells, which are responsible for secreting antibodies, have the highest GPR55 expression levels. Previous studies have shown a potential role for GPR55 in cholesterol metabolism and the development of atherosclerosis [11] [12]. However, the role of GPR55 in B cells was completely unknown. B cells differentiate in the bone marrow and populate secondary lymphoid tissues, where they can encounter antigen, often within a pathogen, and trigger a specific antibody immune response. After B cells encounter an antigen, with assistance from cognate T cells, a differentiation program is initiated known as the germinal centre reaction, in which B cells undergo high proliferation and somatic diversification of their immunoglobulin genes and thereby, clonal B cell variants with higher affinity for the initiating antigen can be generated and selected. After exiting the germinal centre, B cells differentiate into memory B cells and high affinity plasma cells [13] [14].

This work from the Steffens lab focuses on the role of GPPR55 in atherosclerosis with the use of a series of complementary approaches. In the ApoE$^{-/-}$ pro-atherogenic mouse model, the researchers show that GPR55-deficient mice (*ApoE*$^{-/-}$ *Gpr55*$^{-/-}$) developed larger atherosclerotic plaques and had alterations in the composition of advanced plaques, indicating that GPR55 exerts an atheroprotective function. This phenotype in *ApoE*$^{-/-}$*Gpr55*$^{-/-}$ mice was accompanied by increased body weight, metabolic changes and a hyperinflammatory phenotype. Interestingly, several aspects of B cell differentiation and activation were altered in *ApoE*$^{-/-}$ Gpr55$^{-/-}$ mice. For example, bone marrow differentiation was partially impaired, the proportion of marginal zone B cells was increased and the balance between B1a and B1b subsets was altered compared with *ApoE*$^{-/-}$ mice. In addition, the number of germinal centre B cells and plasma cells were drastically reduced in *ApoE*$^{-/-}$*Gpr55*$^{-/-}$ mice compared with *ApoE*$^{-/-}$ mice. Intriguingly, the titres of plasma IgG were increased in *ApoE*$^{-/-}$*Gpr55*$^{-/-}$ mice, which was unexpected given the reduction in plasma cells in these mice. In accordance with this complexity, RNAseq analysis revealed alterations in pathways involved in B cell activation, the germinal centre program, plasma cell differentiation and antibody

secretion as well as reduced expression of the surface IgE receptor CD23. The relevance of this latter finding warrants further investigation. The researchers further show that some of these effects are exacerbated by the pro-atherogenic context, possibly due to the hypercholesterolemic conditions and the on-going humoral response that occurs during atherosclerosis. Nevertheless, non-pro-atherogenic, *Gpr55*$^{-/-}$ mice also show a reduction in the number of plasma cells and increased IgG titers, meaning that GPR55 is a global regulator of the B cell immune response, that is, its function is not restricted to atherogenic situations.

Given the pleiotropic effects observed on B cell physiology in *ApoE*$^{-/-}$*Gpr55*$^{-/-}$ mice, the researchers dissected the B-cell autonomous phenotype of GPR55 deficiency on atherosclerosis development. Consequently, they used a mixed bone marrow chimera approach in which *Gpr55*$^{-/-}$ bone marrow cells were mixed with B cell deficient μMT bone marrow cells and injected into lethally irradiated *Ldlr*$^{-/-}$ proatherogenic mice. In this setting, all B cells must be derived from the *Gpr55*$^{-/-}$ bone marrow, while the μMT bone marrow provides normal non-B cell lineages, thus allowing the assessment of B cell intrinsic GPR55 functions, when compared with *Gpr55*$^{+/+}$ mixed bone marrow chimeras. In contrast to the *ApoE*$^{-/-}$*Gpr55*$^{-/-}$ complete depletion model, B cell specific deficiency of GPR55 did not increase body weight or cholesterol levels of proatherogenic *Ldlr*$^{-/-}$ mice. However, these mice still developed larger atherosclerotic plaques than control chimeras. The B cell compartment was also substantially altered in B cell *Gpr55*$^{-/-}$ chimeras, but intriguingly, the observed phenotype differed considerably from *ApoE*$^{-/-}$*Gpr55*$^{-/-}$ mice: while B1 and marginal zone B cell subsets were reduced, the number of plasma cells was increased. In addition, in B cell specific *Gpr55* deficient chimeras, antibody titres, including anti-MDA-LDL specific IgGs, were increased, but the alterations in distinct isotypes slightly differed from those observed in *ApoE*$^{-/-}$*Gpr55*$^{-/-}$ mice. Several possibilities could account for the B cell differences observed between *ApoE*$^{-/-}$*Gpr55*$^{-/-}$ mice and B cell specific Gpr55 deficient chimeras. First, GPR55 deficiency in cells other than B cells, including T cells, antigen presenting cells, or inflammatory cells can affect B cell

activation and differentiation in germinal centres and into plasma cells. Second, irradiation and subsequent bone marrow reconstitution under competitive conditions can also influence B cell differentiation programs. Accordingly, the metabolic differences observed between both models can be attributed to the contribution of cells other than B cells in ApoE$^{-/-}$Gpr55$^{-/-}$ mice, but also to the use of a different pro-atherogenic mouse model. Regardless of these considerations, which merit further analysis, the crucial finding remains that GPR55 deficiency aggravates atherosclerosis in both settings. This result is further reinforced with adoptive transfer experiments of Gpr55$^{-/-}$ B cells into ApoE$^{-/-}$ mice, which recapitulate many of the findings in ApoE$^{-/-}$Gpr55$^{-/-}$ mice including aggravated atherosclerosis, reduced numbers of germinal center and plasma cells and increased IgG titres. Although the mechanisms responsible for atherosclerosis aggravation in Gpr55 deficient conditions are not completely understood, they are probably related to an aberrant plasma cell differentiation or germinal centre response, which possibly deregulates antibody production. Indeed, these findings suggest a general role for GPR55 in the antibody immune response, which will be extremely interesting to explore under immunization and infection conditions as well as in autoimmune situations.

Together, this study unveils a key atheroprotective role for B cells that depends on GPR55 and underlies the exquisite regulation of the B cell immune response and antibody production during atherosclerosis progression.

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Competing interests

The authors declare no competing interests.