Aberrant distribution of T_{FH} cells in aged germinal centers reduces stromal cell function

The magnitude and quality of the germinal center response after vaccination decline with age. We found that T follicular helper (T_{FH}) cells are enriched in the dark zone of germinal centers in aged mice, which impairs the expansion of the follicular dendritic cell network upon immunization and reduces antibody responses.

This is a summary of:
Silva-Cayetano, A. et al. Spatial dysregulation of T follicular helper cells impairs vaccine responses in aging. *Nat. Immunol.* https://doi.org/10.1038/s41590-023-01519-9 (2023).

Published online: 22 May 2023

The mission

After access to clean water and sanitation, vaccines have been one of the most successful interventions for improving human health. However, vaccines do not protect us equally at all stages of our lives. Age-dependent changes in the immune system contribute to the decline in vaccine efficacy in later years. It is known that poor antibody responses are likely to be responsible for the impaired immunity in aging[1], but the causal cellular and molecular mechanisms remain elusive. We wanted to uncover these mechanisms and see whether we could use this knowledge to boost antibody responses to vaccination in older animals.

The discovery

Vaccination induces the formation of germinal centers (GCs) in lymph nodes; GCs are the major source of long-lived antibody-producing B cells that ultimately provide protection against infection. GCs are beautiful three-dimensional structures in which many different immune cell types converge to collaborate upon a network of stromal cells, including the follicular dendritic cells. GCs have two functionally distinct compartments: the light zone (supported by the follicular dendritic cells and T follicular helper (T_{FH}) cells that support B cell survival in the GC); and the dark zone (where B cells undergo cell division and mutation of the gene encoding the B cell receptor; stromal cells that support the dark zone are CXCL12-producing reticular cells). GC B cells move between the two zones to ultimately generate high-quality antibody responses. We performed quantitative imaging analyses of GCs[2] combined with in silico modeling, analysis of genetically modified mice, and human vaccination studies to understand whether and how the structure of GCs changes during aging and whether such alterations are linked to changes in GC function.

We found that the follicular dendritic cell network was smaller in aged mice. Furthermore, the T_{FH} cells that are normally localized within the light zone were instead dispersed throughout the GC in aged mice (Fig. 1), owing to enhanced expression of CXCR4 (a CXCL12 receptor) that directed the T_{FH} cells to the CXCL12-rich dark zone. Of note, increased expression of CXCR4 was also observed in our human studies. Mathematical modeling of the aged GCs indicated that these two changes could account for the defective GC size and output. We assumed that these two age-dependent changes were unrelated, but experiments with mice whose T_{FH} cells localized to either the light or the dark zone, owing to deficiency of CXCR4 or CXCR5, respectively, clearly demonstrated that stromal cell responses to vaccination require T_{FH} cells to be in the light zone. In aged mice, we were able to show that providing T_{FH} cells from young mice that could correctly localize to the light zone enabled correction of the age-dependent defect in follicular dendritic cell expansion and boost the GC response. This result demonstrated that the age-dependent defects in the GC response are reversible, and that T_{FH} cells in the GC have a previously unappreciated role in supporting the responses of stromal cell to vaccines.

The implications

This study highlights that poor humoral immunity in later years of life is not an insurmountable barrier to increasing health span. Together with other work[3,4], it demonstrates the potential to develop effective vaccines for older people if appropriate follicular dendritic cell and T_{FH} cell stimulation can be provided. Although the present study did not identify potential tractable approaches to facilitate this approach, it shows that it is possible to improve the function of the GC in aging, and generating vaccines that can facilitate this function is a key area of development.

Beyond the context of aging, these data suggest a new concept, namely that T_{FH} cells facilitate the maturation of follicular dendritic cells during GC expansion. How T_{FH} cells deliver this help to stromal cells is yet to be unraveled: possible mechanisms include direct cell contact or using B cells as intermediate mediators.

Michelle A. Linterman[1] & Alice Denton[2]
[1]Babraham Institute, Cambridge, UK.
[2]Imperial College London, London, UK.

EXPERT OPINION

"The authors demonstrates that T_{FH} cell localization within GCs is important for high affinity B cell responses via canonical B cell helper function, and for the expansion of follicular dendritic cell networks in GC light zones. Moreover, they suggest that this localization is defective in aging owing to CXCR4-mediated mechanisms. These findings provide insights into GC biology and identify potential targets for amplifying vaccine responses in older individuals." **Claire E. Gustafson, Allen Institute for Immunology, Seattle, WA, USA.**

FIGURE

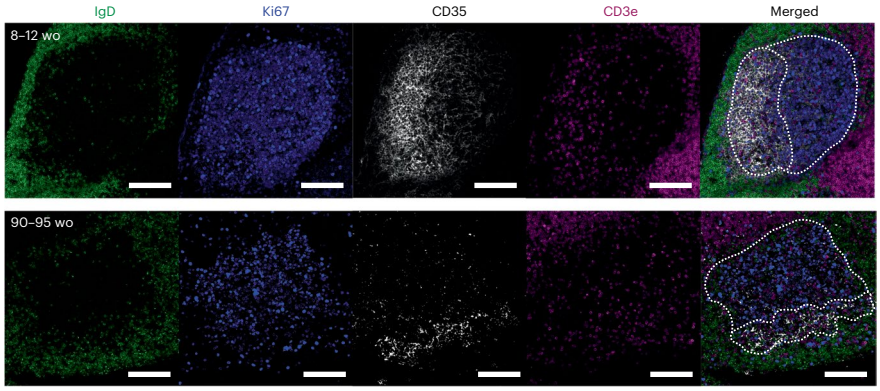

Fig. 1 | The spatial organization of the GC is altered in aged mice. Representative confocal images of GCs from adult (8–12-week-old (wo)) and aged (90–95 wo) BALB/c mice 14 days after immunization with the model antigen NP-KLH in alum, which enables direct assessment of the magnitude and quality of the response to vaccines. Lymph node sections were stained for IgD (green), the follicular dendritic cell marker CD35 (white), proliferation marker protein Ki67 (blue), which identifies proliferating GC B cells, and T cell surface glycoprotein CD3e (magenta). The white dotted lines identify the GC and its light and dark zones. Scale bars, 100 µm

BEHIND THE PAPER

This work was seven years in the making, and it was paused in March 2020, just after we made some of the most exciting discoveries. We paused this project to use the expertise and models set up for this study to run pre-clinical testing for the SARS-CoV-2 vaccine candidate ChAdOx1 nCoV-19 (AZD-1222), created by the University of Oxford and AstraZeneca, in the context of aging[5]. Although putting this work on hold was not ideal, the synergies of our pre-clinical study of AZD-1222 with the clinical trial data that emerged from the phase 1/2 trials demonstrated that aged mice are a relevant and useful system for understanding how aging influences the response to vaccination. **M.A.L.**

REFERENCES

1. Lee, J. L. & Linterman, M. A. Mechanisms underpinning poor antibody responses to vaccines in ageing. *Immunol. Lett.* **241**, 1 (2022).
A review article summarizing age-dependent changes in the response to vaccination.
2. Fra-Bido, S. et al. Optimized immunofluorescence staining protocol for imaging germinal centers in secondary lymphoid tissues of vaccinated mice. *STAR Protoc.* **18**, 100499 (2021).
A protocol paper for confocal imaging of GCs.
3. Yao, Y. et al. Selenium–GPX4 axis protects follicular helper T cells from ferroptosis. *Nat. Immunol.* **22**, 1127–1139 (2021).
This paper reports that selenium supplementation can boost T_{FH} cell responses to vaccines.
4. Almanan, M. et al. IL-10-producing T_{fh} cells accumulate with age and link inflammation with age-related immune suppression. *Sci. Adv.* **6**, eabb0806 (2020).
This paper reports that blocking IL-10 receptor can enhance T_{FH} cell-dependent vaccine responses in aged mice.
5. Silva-Cayetano, A. et al. A booster dose enhances immunogenicity of the COVID-19 vaccine candidate ChAdOx1 nCoV-19 in aged mice. *Med* **2**, 243–262.e8 (2021).
This paper reports that a booster dose of AZ-1222 is required to support humoral immunity in aged mice.

FROM THE EDITOR

"It is known that as individuals age, their ability to mount efficient antibody responses begins to wane. Here, the authors use high-tech imaging analysis to identify defects in the migration of $CD4^+$ T cell to sites within lymph nodes where these cells provide requisite help to the antibody-producing B cells. Their findings might eventually point to ways to 'rejuvenate' T cell positioning to boost immunity in older individuals." **Laurie A. Dempsey, Senior Editor, *Nature Immunology*.**