Changes in neurovascular function in brain microvessels during aging

Age-related decline in brain health is associated with poor blood flow and limitations in energy supply, although the vascular mechanisms are poorly understood. We report an age-related decrease in responsivity of brain microvessels, accompanied by a decrease in vessel density and loss of vascular mural cell processes.

This is a summary of:
Cai, C. et al. Impaired dynamics of precapillary sphincters and pericytes at first-order capillaries predict reduced neurovascular function in the aging mouse brain. *Nat. Aging*, https://doi.org/10.1038/s43587-022-00354-1 (2023).

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The question

Neurovascular coupling (NVC) — the rise in cerebral blood flow that accompanies increased brain activity — is impaired in the aging brain. Despite reduced perfusion, however, oxygen consumption during brain activation is relatively preserved or increased[1,2]. This mismatch between blood supply and metabolic activity is partially attributed to the failure of brain vascular mural cells (VMCs) to control key neurovascular functions in aged mice, and is believed to challenge how well the aged brain can cope with brain diseases, potentially contributing towards neurodegeneration.

The microvascular inflow tract (MIT) comprises arterioles, precapillary sphincters and arteriolar capillaries that have key roles in NVC[3]. Notably, precapillary sphincters at the junction of the penetrating arteriole and its initial branch (first-order capillary) are crucial in maintaining capillary perfusion and are bottlenecks for brain energy supply[4]. However, how neurovascular functions change with brain aging is incompletely understood. Understanding the mechanisms of NVC is an important step towards developing strategies aimed at rebalancing blood supply and metabolic need with age.

The observation

We used 4D two-photon imaging to examine NVC at the MIT in the cortex of living mice. To study microvessel reactivity, we induced vasodilation using whisker electrical stimulation or, alternatively, by inserting a glass micropipette filled with vasoactive signaling molecules in proximity to the MIT and ejecting a small volume locally. Our findings revealed that at the MIT, vascular responsivity decreases with age, contributing to the reduction in NVC responses in aged mice (Fig. 1a,b). Mechanistically, reduced vascular reactivity with aging might be ascribed to an impaired ability of VMCs, especially precapillary sphincters, to relax. By comparison, we found that the ability of vessels to constrict was largely preserved with age.

To characterize aging-related changes in mural cells, we assessed the coverage of VMCs and examined their content of α-smooth muscle actin (αSMA), a contractile protein (Fig. 1c). We reconstructed the aged angiotome on the basis of two-photon image stacks and found that the coverage of vessels of the MIT with VMCs was reduced with age, whereas VMC cell-body density and αSMA density at VMCs were preserved. Capillary density decreased in proximity to the penetrating arteriole, but vessel diameters increased at all locations, reducing the overall vascular resistance.

The implications

Our data reveal crucial roles for precapillary sphincters and first-order capillaries for cerebrovascular resistance in healthy brain aging. Prevention or amelioration of reduced vascular responsivity might restore cerebral blood flow in the aged brain and prevent aging-related brain frailty.

Although we describe in some detail the mechanisms that underpin age-related microvascular dysfunction, our experiments were conducted in anesthetized mice, meaning that the results will need to be addressed in awake, aged and freely moving mice. Furthermore, how to mitigate the impairment of NVC with age is currently unknown and will also need to be addressed in further studies.

In the future, we aim to use genetically encoded indicators and modulators within vascular cells of awake aged mice during natural locomotion and whisker shaking, with and without exposure to drugs that rescue neurovascular functions.

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EXPERT OPINION

"Cerebrovascular function is imperative for brain health and its dysfunction is associated with many neurological diseases. This study demonstrates that vascular dysfunction is also associated with normal aging, including the pathological effects of aging on neurovascular coupling as well as potential mechanisms by which the cerebrovasculature might be compensating for such changes. It further illustrates that this dysfunction occurs at multiple vascular segments, with the most pronounced dysfunction occurring at precapillary sphincters." **Anusha Mishra, Oregon Health & Science University, Portland, OR, USA.**

FIGURE

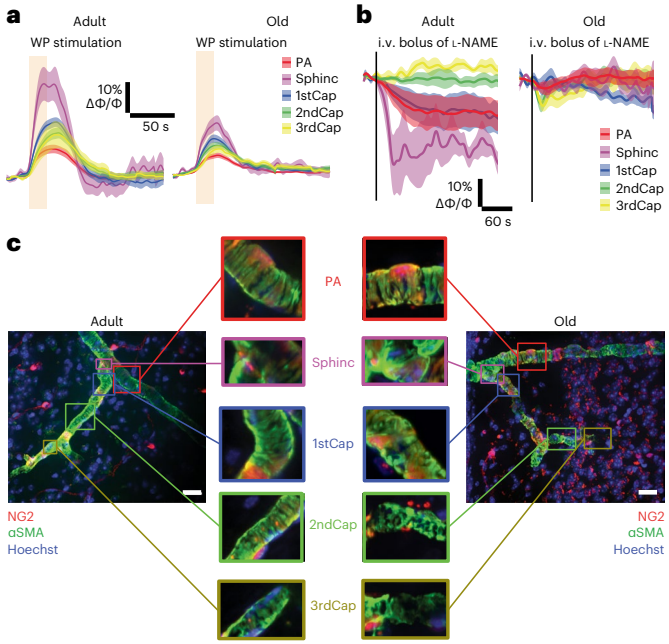

Fig. 1 | Age-related change in vascular reactivity and VMC structure at the MIT. a, Vessel diameter change upon whisker pad (WP) stimulation in adult and aged mice. PA, penetrating arteriole; sphinc, precapillary sphincter; 1stCap, first-order capillaries; 2ndCap, second-order capillaries; 3rdCap, third-order capillaries. **b**, Vessel diameter change upon intravenous infusion of NO synthesis inhibitor L-NAME in adult and aged mice. i.v., intravenous. **c**, Maximum intensity projection of an adult (left) and an aged (right) brain image stack. Red, NG2; green, αSMA; blue, Hoechst. Enlarged insets of each vessel segment are presented in the middle for comparison. Scale bar, 10 μm. The coverage of VMCs on the vessel surface at the MIT was reduced with age.

BEHIND THE PAPER

Over years of studying brain microvessels, we noticed the importance of first-order capillaries and precapillary sphincters for the regulation of capillary blood flow, and how vulnerable they are in brain diseases. Therefore, we became interested in the roles of the MIT during aging. Most of the in vivo experiments were performed before and during COVID-19 lockdown. When we noticed the prominent change of NVC at the aged MIT by puffing multiple vasoactive compounds, we decided to further examine the vascular structure by immunohistochemical staining. We were amazed with the fact that, similar to neuron number preservation with reduced dendrite lengths and numbers during healthy aging, VMCs follow the same pattern of reduction of complexity. **C.C.**

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FROM THE EDITOR

"Precapillary sphincters recently emerged as important regulatory structures of vascular flow in the brain. Using multi-photon imaging in vivo, this study finds that cerebrovascular responsivity decreases with age, and importantly, that this phenomenon is most pronounced at the level of precapillary sphincters. These changes in brain capillary dynamics were accompanied by a reduction in vessel density and mural cell processes that together could lead to cerebral blood flow dysregulation." **Sebastien Thuault, Chief Editor, *Nature Aging*.**