# News & views

Neurodegeneration

# Lipid carrier breaks barrier in Alzheimer's disease

Makoto Ishii & Costantino Iadecola

People who carry the gene variant *APOE4* are at higher-than-average risk of developing Alzheimer's disease. It emerges that this variant is linked to defects in the blood–brain barrier and subsequent cognitive decline. **See p.71**

The best-known hallmarks of Alzheimer's disease are clumps of misfolded amyloid-β (Aβ) and tau proteins, which aggregate in the brain. However, there is increasing awareness that Aβ and tau might not be the whole story – alterations in the blood–brain barrier (BBB) have also emerged as early markers of this neurodegenerative disorder[1]. The degree of disruption to the BBB correlates with the degree of cognitive dysfunction that a person experiences[2], but what causes BBB breakdown has been unknown. Montagne *et al.*[3] present evidence on page 71 that the leading genetic risk factor for Alzheimer's disease, apolipoprotein E4, is linked to BBB breakdown.

The gene *apolipoprotein E* (*APOE*) encodes a major lipid-carrier protein, ApoE, in the brain[4]. There are three predominant variants of *APOE*: *APOE2*, *APOE3* and *APOE4*. As with almost all genes, people carry two copies of *APOE*, which can be either the same or different variants. Compared with the more-common *APOE3* variant, *APOE4* markedly increases the risk of Alzheimer's disease – up to 4-fold in people with one copy of this variant, and 15-fold in people who have two copies[4]. People carrying *APOE4* who do contract Alzheimer's disease also tend to develop symptoms of the disorder earlier than those who develop the disease but do not carry the variant[4].

Proteins from blood plasma have been found in the cerebrospinal fluid (the liquid that surrounds the brain and spinal cord) of cognitively healthy people who carry *APOE4* and who subsequently go on to develop Alzheimer's disease. These proteins have presumably leaked through the BBB, indicating that the integrity of the barrier is lost before cognition declines[5]. Evidence from mouse models, and from the brains of people who have died with Alzheimer's disease, suggests that BBB breakdown is caused by the degeneration of pericytes – cells nestled in the wall of cerebral capillaries. These cells normally safeguard the BBB[5] by preventing the breakdown of junctions between endothelial cells, which make up the capillary walls.

Whether ApoE4 is responsible for early BBB dysfunction in Alzheimer's disease, by itself or in concert with Aβ and tau, was unknown. Montagne and colleagues set out to address this knowledge gap. The authors used a technique called dynamic contrast-enhanced magnetic resonance imaging to investigate the permeability of the BBB in people who had either healthy cognition or mild cognitive impairment (a prelude to Alzheimer's disease), grouped according to their *APOE* status. They found that people who were cognitively healthy and carried either one or two copies of *APOE4* had a leaky BBB in two brain regions important for memory and cognition – the hippocampus and the parahippocampal gyrus. This leakage was worse in *APOE4* carriers who exhibited mild cognitive decline.

Remarkably, these effects preceded any signs of tissue loss in the hippocampus and parahippocampal gyrus, attesting to the idea that BBB disruption is an early event in the onset of neurodegeneration. BBB leakage was independent of Aβ and tau accumulation, which the authors assessed both by studying samples of cerebrospinal fluid and through another brain-imaging technique, positron emission tomography. Montagne and co-workers found that, unlike in *APOE4* carriers, the BBB was intact in cognitively healthy *APOE3* carriers. It was, however, leaky in *APOE3* carriers who showed cognitive impairment – although less so than in *APOE4* carriers at an equivalent stage of impairment.

Next, Montagne *et al.* examined whether BBB breakdown in *APOE4* carriers was linked to pericyte degeneration. In support of this idea, they found that a biomarker of pericyte injury – a soluble form of a protein known as platelet-derived growth factor-receptor-β (sPDGFRβ) – was elevated in the cerebrospinal fluid of *APOE4* carriers compared with *APOE3* carriers. High levels of the protein in people who carried *APOE4* were associated with a

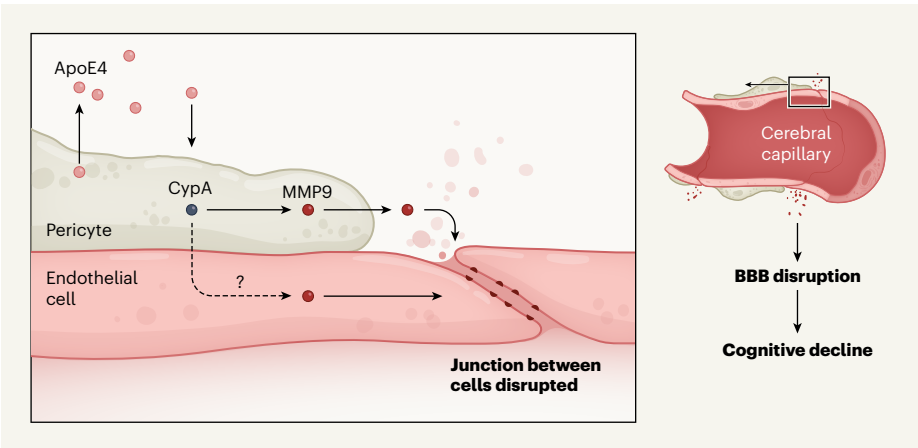


**Figure 1 | The gene variant *APOE4* and Alzheimer's disease.** People who carry *APOE4* are at heightened risk of Alzheimer's disease. Montagne *et al.*[3] provide evidence that ApoE4 protein is secreted by cells called pericytes, which abut endothelial cells that line cerebral capillaries at the blood–brain barrier (BBB). Secreted ApoE4 activates the protein cyclophilin A (CypA) in the pericytes. This triggers a downstream signalling pathway involving activation of the inflammatory protein matrix metalloproteinase-9 (MMP9) in pericytes, and possibly also in endothelial cells. This causes disruption of junctions between adjoining endothelial cells, opening the BBB in brain regions involved in learning and memory. Disruption of the BBB is associated with impaired cognition, although the mechanisms that link the two are unclear.

leaky BBB and cognitive impairment. sPDGFRβ elevation was independent of Aβ and tau.

The authors then looked for insight into the mechanisms by which pericytes might become injured. They focused on cyclophilin A (CypA) and matrix metalloproteinase-9 (MMP9), two proteins that are part of an inflammatory pathway implicated in *APOE4*-driven pericyte damage and BBB breakdown[6]. Levels of CypA and MMP9 in the cerebrospinal fluid were higher in *APOE4* carriers who had mild cognitive impairment than in cognitively healthy *APOE4* carriers or *APOE3* carriers who had comparable cognitive dysfunction. Again, this change was not related to increases in Aβ or tau.

Finally, the researchers generated pericytes *in vitro* from human induced pluripotent stem cells that expressed *APOE3* or *APOE4*. They found that *APOE4*-expressing pericytes secreted substantially more CypA and MMP9 than did *APOE3* pericytes. ApoE4 (but not ApoE3) secreted by pericytes activates the CypA–MMP9 pathway on nearby pericytes – the cells therefore cause their own demise. ApoE4 could also activate the CypA–MMP9 pathway in endothelial cells, which are susceptible to the harmful effects of *APOE4* (ref. 7). Therefore, injury to pericytes and endothelial cells might both cause BBB leakage (Fig. 1).

These observations cast new light on *APOE4* that runs contrary to the widely held idea that this gene variant contributes to Alzheimer's disease solely by promoting Aβ and tau accumulation[4]. Instead, it seems that BBB dysfunction might explain why *APOE4* carriers are susceptible to Alzheimer's disease. The authors' findings might also explain why *APOE4* carriers have worse outcomes following stroke or traumatic brain injury[8] than do people who carry other *APOE* variants. However, as Alzheimer's disease progresses, *APOE4* could also slow Aβ and tau clearance, exacerbating declines in cognition.

Even more striking is the finding that early drivers of cognitive impairment differ between *APOE4* and *APOE3* carriers. Montagne and colleagues' findings indicate that activation of the CypA pathway and pericyte damage might not be involved in cognitive impairment in people who carry the most common *APOE* variant, *APOE3*. But whether a leaky BBB caused by factors that are independent of pericytes (for example, damage to endothelial cells caused by Aβ; ref. 1) contributes to cognitive impairment in *APOE3* carriers remains unclear. The role of the BBB in *APOE2* carriers, which was not assessed in the current study, also remains unknown. Although *APOE2* is associated with a reduced risk of Alzheimer's disease compared with other *APOE* variants, this is unlikely to result from a more resilient BBB, because *APOE2* carriers have an increased risk of microhaemorrhages, suggesting vascular frailty[4].

Whether and how BBB breakdown leads to cognitive impairment also remains to be determined. Is it a cause or a consequence of the disease process? Evidence from mice indicates that some proteins in the blood, such as fibrinogen, damage the synaptic connections between neurons[9]. But a pathogenic role for these proteins in the human brain has not yet been demonstrated.

Irrespective of these questions, Montagne *et al.* have broadened our understanding of how *APOE4* promotes cognitive impairment. They have also demonstrated that different *APOE* statuses can promote disease through different mechanisms. A deeper appreciation of how gene variants shape Alzheimer's disease might prove crucial for more-personalized approaches to treating this prevalent and incurable disease.

**Makoto Ishii** and **Costantino Iadecola** are at the Feil Family Brain and Mind Research Institute, Weill Cornell Medicine, New York, New York 10065, USA.
e-mail: coi2001@med.cornell.edu

1. Cortes-Canteli, M. & Iadecola, C. *J. Am. Coll. Cardiol.* **75**, 942–951 (2020).
2. Nation, D. A. *et al. Nature Med.* **25**, 270–276 (2019).
3. Montagne, A. *et al. Nature* **581**, 71–76 (2020).
4. Yamazaki, Y., Zhao, N., Caulfield, T. R., Liu, C.-C. & Bu, G. *Nature Rev. Neurol.* **15**, 501–518 (2019).
5. Profaci, C. P., Munji, R. N., Pulido, R. S. & Daneman, R. *J. Exp. Med.* **217**, e20190062 (2020).
6. Bell, R. D. *et al. Nature* **485**, 512–516 (2012).
7. Koizumi, K. *et al. Nature Commun.* **9**, 3816 (2018).
8. Mahley, R. W., Weisgraber, K. H. & Huang, Y. *Proc. Natl Acad. Sci. USA* **103**, 5644–5651 (2006).
9. Merlini, M. *et al. Neuron* **101**, 1099–1108 (2019).

**This article was published online on 29 April 2020.**

Atomic physics

# Exotic helium atom lit up

**Niels Madsen**

An elusive type of atom known as pionic helium has been directly excited by laser light for the first time. The work establishes a promising experimental platform for probing fundamental physics. **See p.37**

Exotic atoms are those in which one or more of the constituents of normal atoms have been replaced by an exotic particle, such as an antimatter particle. These atoms can then be probed to search for any tiny discrepancies in their properties from those predicted by models using techniques that underpin the world's most accurate timekeepers, atomic clocks – and thereby opening a window on the foundations of physics. On page 37, Hori *et al.*[1] are the first to report laser excitation of helium atoms in which one electron has been replaced by a subatomic particle called a pion.

The interest in exotic atoms arises from the fact that they often facilitate the most basic experimental strategy used in physics: changing a single parameter or component in an otherwise complex system, to observe the effect. In practice, this is not as simple as it might seem. Different particles can have different masses or charges, and might interact with their surroundings differently in other subtle ways. However, such subtleties often add to the value of exotic atoms.

As the techniques needed to study exotic atoms improve, increasing numbers of scientists are working with these atoms to investigate the fundamental properties of nature. A good example of this is the 'proton radius puzzle', which arose from a study of muonic hydrogen[2] – a hydrogen atom in which the electron has been replaced by a subatomic muon particle (muons have similar properties to electrons, but have about 200 times greater mass).

Muonic hydrogen was used to determine a key property of the proton known as the charge radius, but the value obtained was about seven standard deviations away from the expected value at the time. The value obtained using muonic hydrogen has since been independently confirmed in a study of ordinary hydrogen[3], and also in experiments in which electrons are scattered from protons[4], potentially clarifying the true value of the proton radius and thus solving the puzzle. Nevertheless, muonic hydrogen aptly illustrates how exotic, sometimes short-lived, atomic systems can be used to poke holes in seemingly well-established results.

An important feature of exotic atoms that adds to their utility as probes for fundamental physics is that they are bound systems (energy is needed to pull their components apart), with multiple internal energy states. Transitions between these states are therefore amenable to study by laser spectroscopy, the most precise measurement tool in the physics toolkit. The study of transitions in atoms – and particularly in the hydrogen atom – is an ongoing effort that has spanned more than two centuries. It inspired Niels Bohr's groundbreaking model of the atom in the early twentieth century, for example, and has driven much