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A bile acid could explain how calorie restriction slows ageing

Could lithocholic acid, a compound produced when gut bacteria process bile, be the missing link between a low-calorie diet and its age-defying effects? Experiments in mice, flies and nematode worms provide clues.

By David A. Sinclair

Across the ancient world, physicians from Greece to China touted the health benefits of bile and fasting. Today, the focus is on regular meals and exercise, but perhaps those ancient doctors were onto something. In two papers in *Nature*, Qu and colleagues[1,2] make a compelling case that a component of bile called lithocholic acid (LCA) triggers many of the age-defying and potentially lifespan-extending health benefits of low-calorie diets.

First, some background on calories, ageing and bile acids. Low-calorie diets were formally shown to delay ageing in the early twentieth century, when researchers fed rats a mix of food and indigestible cellulose[3]. Since then, calorie restriction has been

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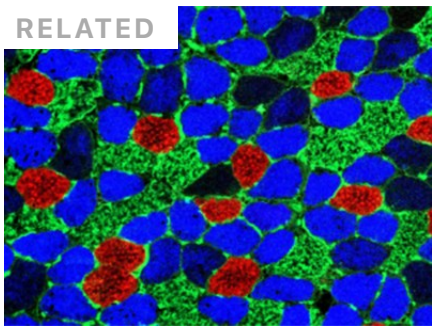

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shown to prolong lifespan in several species, although some mouse strains and wild-derived animals show minimal or even negative responses to calorie restriction[4], and in rhesus monkeys (*Macaca mulatta*) the effects have been mixed, for reasons that are debated[5].

At first, the benefits of calorie restriction were attributed to delayed development or slowed metabolism, but in the 2000s, a new paradigm emerged: that calorie restriction triggers a genetically encoded survival mechanism. This concept came to light after the discovery of alterations in single genes that extended lifespan in model organisms, ostensibly by mimicking calorie restriction and environmental threats to survival[6–8].

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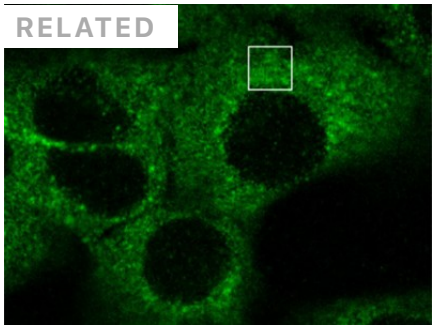

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Generally speaking, bile is less interesting than is longevity, but that might soon change. Consisting mainly of water, bilirubin (a breakdown product of haemoglobin), cholesterol and bile acids, this yellow-green fluid is synthesized in the liver, stored in the gallbladder and released into the small intestine to emulsify dietary fats and increase the absorption of fat-soluble vitamins. Gut-resident bacteria, such as species of *Clostridium* and *Lactobacillus*, convert primary bile acids into the secondary bile acids deoxycholic acid and LCA, some of which is reabsorbed into the bloodstream.

Previous work has identified bile acids as health-promoting compounds. Dafachronic acids, which are structurally related to LCA, extend the lifespans of nematode worms (*Caenorhabditis elegans*)[9] and LCA extends the lifespans of yeast (*Saccharomyces cerevisiae*) and fruit flies (*Drosophila melanogaster*; see ref. 10 and references therein). In mammals, LCA is not known to extend lifespan, but it does alter physiology in ways that are consistent with improved health, such as lowering levels of liver triglycerides, blood glucose and systemic inflammation — in part, by activating the bile-acid receptor TGR5[11]. LCA is also implicated in the lifespan-extending effects of transplanting gut microbiota from young mice into old mice, but how the bile acid might impart health benefits is unclear[12].

In mammals, a family of seven enzymes known as sirtuins (or SIRT1–7) combat numerous biological processes that contribute to ageing, including cellular senescence (in which cells stop dividing), DNA damage, decreased energy production and impaired tissue repair. Their reactions require the ubiquitous metabolite molecule NAD^+, the levels of which decrease with age and are boosted by calorie restriction. The hypothesis that sirtuins mediate some of the benefits of calorie restriction is supported by considerable data[13], but it is not without detractors.

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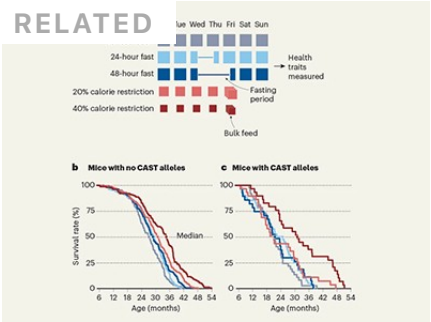

Dietary restriction can extend lifespan — but genetics matters more

This brings us to the work of Qu and colleagues. To find molecules that mediate the health benefits of calorie restriction, the researchers separated out metabolites in mouse serum (the clear liquid part of blood that remains after clotting), and tested which ones were at higher levels in calorie-restricted mice than in non-calorie-restricted mice. Among these, the authors searched for metabolites that activated the enzyme

AMP-activated kinase (AMPK), which is suspected to mediate some of the health benefits of calorie restriction[14]. Using the technique of mass spectrometry, the authors quantified 1,215 metabolites, ultimately homing in on LCA[1].

Next, they gave LCA to old mice for a month. These mice experienced health benefits reminiscent of those induced by calorie restriction, including improved muscle regeneration, grip strength and sensitivity to insulin. These effects were dependent on AMPK. Interestingly, LCA raised levels of the hormone GLP-1 without causing muscle loss, unlike today's popular weight-loss drugs that bind to the GLP-1 receptor. In nematodes and flies, LCA activated AMPK, increased stress resistance and extended lifespan — benefits that were negated when the gene encoding AMPK was deleted in the animals.

After ruling out TGR5 as the mediator of LCA's effects, the researchers turned their attention to the enzyme SIRT1. In a series of exhaustive experiments[2], the authors demonstrated that LCA stimulates SIRT1 to remove acetyl chemical groups from three crucial amino-acid residues of a large protein called vacuolar H^+-ATPase (v-ATPase), thereby inhibiting it (Fig. 1). Inhibition of v-ATPase triggers a signalling cascade involving the proteins AXIN and LKB1 (a known target of SIRT1), leading to the activation of AMPK. Qu and co-workers also showed that TULP3, a protein partner of SIRT1, binds to LCA and helps to catalyse the activation of SIRT1. Consistent with this model, TULP3 was necessary for LCA to extend lifespan in nematodes and flies.

Figure 1 | A bile acid might be the long-sought molecule that mediates the health benefits of calorie restriction. **a**, During calorie restriction, a component of bile called chenodeoxycholic acid (CDCA) is converted into lithocholic acid (LCA) by bacteria in the small intestine. LCA is absorbed into the bloodstream across the epithelium and transported to

muscle. **b**, Qu *et al*.[1,2] show that LCA binds to a protein called TULP3, which then activates SIRT1, an enzyme dependent on the metabolite molecule NAD^+. Activated SIRT1 then removes acetyl chemical groups from a large enzyme complex called v-ATPase, inhibiting it. This triggers the activation of a signalling pathway involving proteins AXIN and LKB1, which activates a key metabolic regulator, AMPK, on the surface of lysosome organelles. This signalling pathway mediates many of the health benefits of calorie restriction in mice, and the same apparent mechanism extends lifespan in fruit flies (*Drosophila melanogaster*) and nematode worms (*Caenorhabditis elegans*).

Here is where it gets even more interesting. Twenty years ago, SIRT1 was shown to be directly activated by a plant molecule called resveratrol, a chemical found in red wine that extends lifespan in yeast, nematodes and flies[15]. Later studies showed that resveratrol also activates AMPK, a downstream target of SIRT1, which prompted the development of more-potent, synthetic sirtuin-activating compounds (STACs). These potent STACs improved health and increased the lifespans of male mice through mechanisms related to calorie restriction[16]. STACs also alleviated the skin disease psoriasis in humans[17].

The ability of resveratrol to activate SIRT1 implied the existence of a naturally occurring STAC in mammals that rises during calorie restriction[15]. Over the years, STAC candidates have included the AROS protein and oleic acid from olive oil[18], but none has led to a convincing explanation of how calorie restriction works. When Qu and colleagues[2] tested a SIRT1 variant engineered to be resistant to activation by STACs, they found that LCA was no longer able to activate SIRT1. These data essentially prove that LCA works through the same activation mechanism as resveratrol and other STACs — a remarkable finding.

So, have Qu and colleagues identified the elusive native SIRT1 activator? Possibly —

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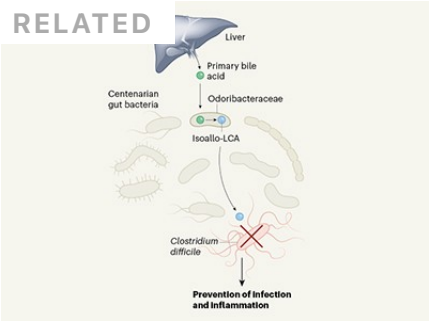

Role of bile acids and gut bacteria in healthy ageing of centenarians

but confounding results and key questions remain. For example, it is unclear whether TULP3, LCA or both directly interact with the part of the SIRT1 protein that flexes to mediate its activation. Despite improving health in the three species tested, there was no statistically significant lifespan extension in the LCA-treated mice. And although LCA extended the lifespans of nematodes and flies, those animals don't produce LCA[10], suggesting that a related molecule, such as a dafachronic acid, directly activates SIRT1 in those species. Testing the effects of calorie restriction and LCA on nematodes, flies and mice that lack SIRT1 activation would help to resolve which health benefits are due to SIRT1 activation and which are not.

The known effects of bile acids are only partially consistent with the authors' model. In support of the model, feeding pigs (*Sus scrofa domesticus*) chenodeoxycholic acid (CDCA), an LCA precursor that raises LCA levels, boosts growth rates and improves health[19]. But in humans, CDCA has been prescribed as a medicine since the 1970s to shrink gallstones, and there are no reports that the medicine has other health benefits. Nor is there evidence that gallbladder removal, which can disrupt bile flow, increases susceptibility to age-related diseases. Although carefully designed clinical trials could resolve these apparent contradictions, caution is essential. LCA has the potential to cause liver toxicity and, when combined with DNA-damaging agents, could increase the risk of cancer[20].

The involvement of gut microbiota in the production of LCA and the benefits of calorie restriction might explain why faecal transplants from young animals improve

the health and increase the lifespans of older animals, and why some mice do not respond to calorie restriction. The work also raises questions about the potential long-term metabolic effects of oral antibiotics and the removal of the gallbladder and the appendix, the body's reservoir of gut microorganisms. Ultimately, if Qu and colleagues' model holds up, these findings could be remembered as a milestone linking caloric intake to age-related diseases, and as a catalyst for the use of bile acids to treat diseases in ways that physicians from the ancient world would have certainly approved of.

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References

1. Qu, Q. *et al. Nature* https://doi.org/10.1038/s41586-024-08329-5 (2024).

2. Qu, Q. *et al. Nature* https://doi.org/10.1038/s41586-024-08348-2 (2024).

3. McCay, C. M., Crowell, M. F. & Maynard, L. A. *J. Nutrition* **10**, 63–79 (1935).

4. Liao, C.-Y., Rikke, B. A., Johnson, T. E., Diaz, V. & Nelson, J. F. *Aging Cell* **9**, 92–95 (2010).

5. Mattison, J. A. *et al. Nature Commun.* **8**, 14063 (2017).

6. Kimura, K. D., Tissenbaum, H. A., Liu, Y. & Ruvkun, G. *Science* **277**, 942–946 (1997).

7. Lin, S.-J., Defossez, P.-A. & Guarente, L. *Science* **289**, 2126–2128 (2000).

8. Anderson, R. M., Bitterman, K. J., Wood, J. G., Medvedik, O. & Sinclair, D. A. *Nature* **423**, 181–185 (2003).

9. Gerisch, B. *et al. Proc. Natl Acad. Sci. USA* **104**, 5014–5019 (2007).

10. Staats, S. *et al. Mol. Nutr. Food Res.* **62**, e1800424 (2018).

11. Thomas, C., Pellicciari, R., Pruzanski, M., Auwerx, J. & Schoonjans, K. *Nature Rev. Drug Discov.* **7**, 678–693 (2008).

12. Bárcena, C. *et al. Nature Med.* **25**, 1234–1242 (2019).

13. Chen, D. & Guarente, L. *Trends Mol. Med.* **13**, 64–71 (2007).

14. Martin-Montalvo, A. *et al. Nature Commun.* **4**, 2192 (2013).

15. Howitz, K. T. *et al. Nature* **425**, 191–196 (2003).

16. Mercken, E. M. *et al. Aging Cell* **13**, 787–796 (2014).

17. Krueger, J. G. *et al. PLoS ONE* **10**, e0142081 (2015).

18. Najt, C. P. *et al. Mol. Cell* **77**, 810–824 (2020).

19. Song, M. *et al. Anim. Nutr.* **7**, 365–375 (2021).

20. Kawasumi, H. *et al. Oncology* **45**, 192–196 (1988).

COMPETING INTERESTS

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