

## **Reply to: ‘Lack of evidence for intergenerational inheritance of immune resistance to infections’**

We read with great interest the study by Kaufmann and colleagues investigating the possibility that protection to reinfection may be transmitted intergenerationally in mice (1). The authors found no such inheritance of resistance in models using BCG,  $\beta$ -glucan or a *Candida albicans* infection as potential inducers of trained immunity. This is different from our recent findings (2) in which we show, based on independent experimental data from different laboratories, that a *C. albicans* infection or zymosan injection can induce intergenerational protection in three models of bacterial or fungal infections, and influences immunological responses to endotoxemia. We are very encouraged that this topic is gaining increased interest: it is indeed important that more information is becoming available regarding variations between different laboratories and conditions. The reason for the discrepancy between the findings in the two studies is unclear, and it likely involves local differences in mouse substrains, housing, diet, microbiome, infection models or other factors. These findings underscore the effect of the environment on the intergenerational inheritance of infection resistance. What these environmental factors are and how these factors are integrated with regards to intergenerational inheritance remains largely elusive at this time. One intriguing possibility that needs to be tested in future studies is whether such effects may be more robust in outbred wild mice, in which subtle environmental changes may have less strong impact.

A question raised by Kaufmann and colleagues is whether intergenerational transmission of resistance to infections, which is well documented in plants and invertebrate animals (3,4), has been lost once vertebrates have acquired adaptive immunity. We argue that this is not the case based on a number of arguments. First, on theoretical grounds, the evolutionary advantage to transmit resistance to infection to the next generation should be independent of whether it is mediated by innate or adaptive immunity. Darwinian evolution through genetic variability and selection is a slow process, which cannot provide protection in the short term during a severe epidemic, while intergenerational transmission of resistance based on epigenetic processes has the potential to induce rapid protection in the next generation. Second, intergenerational transmission of traits induced by metabolic stresses or toxins in

vertebrates has been demonstrated extensively, as also acknowledged by Kaufmann et al. (1). However, these are not adaptive processes, and it is highly unlikely that the epigenetic mechanisms transmitting these traits have persisted during evolution to induce deleterious effects in the progeny. It is much more likely that the capacity to transmit changes in the next generation persisted in vertebrates to confer an evolutionary advantage, and infections are the most likely target. Third, transmission of immune traits to the next generation in vertebrates has been recently demonstrated in other studies. In mice, inflammation in males induces underweight progeny (5), prenatal infection in females promotes tissue-specific immunity (6) and sepsis-induced tolerance can be transmitted intergenerationally (7). Additionally, in humans, lower overall mortality has been reported in infants whose fathers had been vaccinated with BCG (8), and maternal SARS-CoV-2 infection has been associated with increased cytokine functionality and nonspecific immune imprinting in the neonates (9). Finally, trained immunity has been shown to be transmitted in the newborn infants of hepatitis B virus-infected mothers (10).

Altogether, we believe that these new data from independent studies strengthen the conclusion that intergenerational transmission of immune traits is an important characteristic of vertebrate physiology and a function of environmental priming. However, we also acknowledge, on the basis of the data of Kaufmann and colleagues, that these mechanisms are likely influenced by the environmental conditions in which such transmission occurs, and this should stimulate more research in this important new field of investigation.

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