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### ► To cite this version:

Fany Blanc, Alexandre Lecoecur, David Gourichon, Nathalie Meme, Thierry Burlot, et al.. The combined effect of genetics, gut microbiota, and environment on immunity in laying hens. 8th European Veterinary Immunology Workshop, Sep 2024, Dublin, Ireland. . hal-04709617

**HAL Id: hal-04709617**

**<https://hal.inrae.fr/hal-04709617v1>**

Submitted on 25 Sep 2024

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## **The combined effect of genetics, gut microbiota, and environment on immunity in laying hens**

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Animals are vaccinated to protect them against various pathogens. Nonetheless, vaccine responses can vary greatly, with several factors potentially impacting vaccine responses and immunity. To investigate this, we studied the effects of genetics, microbiota, and rearing conditions on vaccine responses and other immune parameters. We divided 400 chicks into 8 groups to observe the influence of different laying hen lines (Rhode Island Red (RIR) and White Leghorn (LEG)), microbiota (by administering or not a cocktail of three antibiotics), and rearing conditions (whether or not they had access to an outdoor yard from 12 weeks until the end of the experiment). We monitored the humoral vaccine responses throughout the experiment by using ELISA for 5 different vaccines, and the cellular response was assessed by ELISpot (IFN- $\gamma$  secretion after restimulating splenic immune cells). Other immune parameters measured included hematocrit levels, blood cells, immunoglobulins, and natural antibodies. Our study found that the RIR line had a better vaccine response than the LEG line, which even had non-responders. We also observed differences in blood cell composition between the lines. Microbiota perturbation altered vaccine responses and had an impact mainly on the LEG line, leading to a reduction in immunoglobulins and changes in blood cell composition. Rearing conditions also moderately affected vaccine responses and cell composition, particularly in the RIR line. In summary, our study highlights the influence of genetics, microbiota, and rearing conditions on vaccine responses and immune parameters that could be exploited to improve animal health.

Keywords (3 max): immunity, genetics, microbiota