

Report Type: PROGRESS

Title: Comparison of disease resistance among heritage chicken breeds challenged with enteric pathogens – A strategy for infectious disease control in organic poultry production

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Abstract:

Organic poultry farming is uniquely threatened by infectious disease, because organic farming practices increase chicken exposure to parasites and pathogens while simultaneously restricting the use of drugs and chemicals that control infections. “Heritage” chicken breeds, which are commonly used in organic farming, may possess endogenous disease resistance. However, it is unclear how resistance to infection varies among heritage breeds. We hypothesized that heritage chicken breeds differ in susceptibility to infection by intestinal parasites and pathogens. To test our hypothesis, we compared experimental infections among six heritage chicken breeds inoculated with *Eimeria* sp. (protozoa) or *Campylobacter jejuni* (bacteria) that affect poultry health (*Eimeria*) and cause foodborne illness in people (*Campylobacter*). We observed significant differences in disease pathology (*Eimeria*) and bacteria infectivity (*Campylobacter*) among the heritage breeds, suggesting that farmers could compose flocks with breeds more robust to disease and thus enhance the resilience of the organic poultry industry.

Project Description:

Outputs

- Overview of Work Completed and in Progress: We successfully challenged chickens from seven heritage breeds with *Eimeria* sp. protozoan parasites and *Campylobacter jejuni* bacteria. For each type of infection, we completed two experimental replicates. Among breeds, there were significant and consistent differences in the tissue damage caused by *Eimeria* infection. Only one breed was consistently susceptible to infection by *Campylobacter jejuni*. Deeper examination of gut tissue damage is currently *in progress* using histology. Our data show that tissue damage (*Eimeria* sp.) and susceptibility to infection (*C. jejuni*) were significantly different among the heritage breeds tested.
- Methods: We obtained chicks (all male) at approximately 3-days of age from a single hatchery. The chicks were breeds *Rhode Island Red*, *White Rock*, *Barred Rock*, *Black Star*, *Ameraucana*, *Silver-Laced Wyandotte*, and *Black Australorp*. The chicks (30/breed) were housed in air-controlled environmental chambers. At two weeks of age, half of the chicks were each inoculated orally with 1000 oocysts of *Eimeria* sp. (two experimental replicates) or with 1×10^7 colony forming units of *C. jejuni* (two experimental replicates). Note, each infectious organism was tested separately. There were no co-infections. In

each experiment, half of the chicks were *sham* inoculated with buffer to act as uninfected controls. In each experiment, all chicks were euthanized after one-week post-infection and tissues were collected. The severity of infection by *Eimeria sp.* was determined by visual examination of gut tissue to score the relative levels of tissue damage (e.g., lesions). Infection by *C. jejuni* was determined by plating gut contents and counting bacteria colony growth.

- **Results:** All chickens challenged with *Eimeria sp.* oocysts became infected, regardless of breed. Infected birds had detectable lesions compared to uninfected controls, and there were significant differences in the lesion scores observed among the intestines of the seven breeds (Fig. 1). Breeds Black Australorp, Black Star, Rhode Island Red, and Barred Rock had higher lesion scores and more birds with detectable lesions compared to chicks in the Ameraucana and Silver-laced Wyandotte breeds (Kruskal-Wallis $P < 0.05$). Chicks in the White Rock breed had mixed results across the two experimental replicates. Following challenge with *C. jejuni*, 57% of the Silver-laced Wyandotte chicks became infected (Table 1). In contrast, only 6% of chicks in Ameraucana and Australorp breeds became infected. None of the Barred Rock, Black Star, Rhode Island Red, or White Rock breeds were infected with *C. jejuni* (GLMM $P < 0.05$).

Figure 1. Boxplots of visual lesion scores from intestines of chickens infected with *Eimeria sp.* parasites. Maximum lesion scores (y-axis) from multiple sections of intestine are shown relative to chicken breed (x-axis). Chickens from the Silver Laced Wyandotte breed (arrow) had the lowest scores across all areas of tissue (Kruskal-Wallis Test $P < 0.05$).

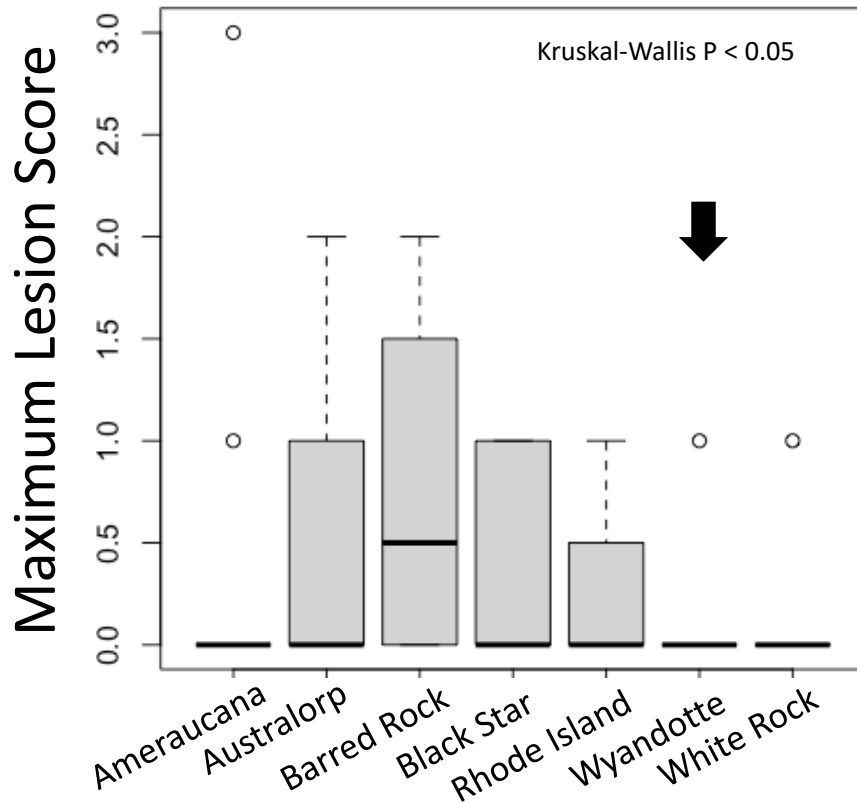


Table 1. Percentages of chickens (~24 birds/breed) colonized by *Campylobacter jejuni* bacteria following oral inoculation.

BREED	% Chicks Infected with <i>C. jejuni</i>
Silver-laced Wyandotte	57
Black Australorp	6
Ameraucana	6
Barred Rock	0
Black Star	0
Rhode Island Red	0

- **Discussion:** Chickens on organic and open-environment (e.g., pastured) poultry farms experience high frequencies of infections by enteric parasites (e.g., *Eimeria sp.*) that can harm chicken health and lower productivity (Cornell et al., 2021). In addition, chickens on 85% of these poultry farms are positive for *C. jejuni* infection, which poses an important health risk for people that consume contaminated meat or eggs (Smith et al., 2023). Organic and open-environment poultry farms often use “heritage” chicken breeds that are not used by conventional poultry producers (Cornell et al., 2021). We tested the hypothesis that heritage breeds would differ in susceptibility to infection and parasite-induced damage. We found that all breeds were equally susceptible to

infection by *Eimeria sp.*, but the breeds Silver-laced Wyandotte and Ameraucana were relatively resistant to damage caused by the parasite. We observed that the Silver-laced Wyandotte breed was more easily infected by *C. jejuni*, which was notable given the apparent resistance of the breed to pathology caused by *Eimeria sp.* infection. The *C. jejuni* bacteria do not typically cause pathology in host birds and we did not observe any evidence of pathology, morbidity, or mortality associated with the experimental challenges with these bacteria.

Currently there is little known about disease resistance among heritage chicken breeds. This is problematic, given that these breeds are used in production systems with high disease pressure and farmers are often prevented from using parasiticides and antibiotics (Cornell et al., 2021). Our results demonstrate that heritage chicken breeds have different disease resistance phenotypes. These data suggest that farmers could select breeds to construct flocks with higher “natural” resistance to infection, or lower pathological effects when infected. As a result, farmers may be able to reduce the number of infected birds in a flock (i.e., greater resistance) or reduce the level of damage caused by infections (i.e., greater resilience).

It is important to note that no breed tested in these experiments was universally resistant. All birds became infected with *Eimeria sp.* parasites, regardless of breed. The breed with the lowest pathology associated with *Eimeria sp.* – Silver-laced Wyandotte – was also the most likely to become infected with *C. jejuni*. Conversely, breeds that were resistant to *C. jejuni* infection were also the breeds with higher levels of pathology from *Eimeria sp.* (e.g., Barred Rock and Rhode Island Red). This suggests possible trade-offs in the defenses that are effective against these two organisms. Future work will need to identify how different breeds respond to the diversity of infectious organisms found on open-environment farms (Cornell et al., 2021). In addition, these data underscore the importance of parasite/pathogen surveillance on farms, so that farmers can make informed decisions about management approaches that may include breed selection.

Finally, it is important to acknowledge that this work was done with young birds (2 weeks of age). Young birds are raised on farms and may become infected with these organisms. However, adult birds are also part of the disease cycles and are arguably more important economically. It is unclear if the results observed in this study forecast the responses of adult chickens. Moreover, it remains unknown how these breeds respond to infection in terms of economic metrics such as growth rate, egg production, and feed conversion. Those economic factors will be the focus of a grant proposal that will follow this work.

References:

Cornell, K.A., Smith, O.M., Crespo, R., Jones, M.S., Snyder, W.E., Owen, J.P. (2021) Prevalence patterns for enteric parasites of chickens managed in open environments of the Western United States. *Avian Diseases*. doi.org/10.1637/21-00079

Smith, O.M., Cornell, K.A., Crossley, M.S., Crespo, R., Jones, M.S., Snyder, W.E., Owen, J.P. (2023) Wind speed and landscape context mediate *Campylobacter* risk among poultry reared in open environments. *Animals*. doi.org/10.3390/ani13030492

- Publications, Handouts, Other Text & Web Products: Currently, there are two manuscripts in preparation from this work. These manuscripts will be submitted to the journal *Poultry Science*. The first will focus on *Eimeria* and the second will focus on *C. jejuni*.
- Outreach & Education Activities: An arts-based teaching module is in development to help young learners explore the relationships between genetic variation and disease. This curriculum is a spin-off from a larger project titled “HEAL: Health Education through Arts-Based Learning” that has been funded by the National Institutes of Health. The module associated with this BioAg research will task learners with creating different chicken breeds based on colors and feather patterns. After the students compose flocks of the different chicken breeds, they will be told that certain colors/patterns associate with disease resistance. The learners will then “test” the spread of disease in their flocks with simulations, based on the frequencies of their chicken’s visual patterns in the flock. We anticipate testing this curriculum in summer 2024.

Impacts

- **Short-Term**: This work has provided robust, definitive evidence that heritage chicken breeds differ in the severity of pathology associated with infection by *Eimeria sp.* parasites and differ in susceptibility to colonization by *Campylobacter jejuni* bacteria. These data form a strong rationale for continued work to identify what breeds provide effective defenses against infectious diseases observed on open-environment farms.
- **Intermediate-Term**: In a 3- to 5-year timeframe, we expect to characterize both disease resistance traits and economic metrics (e.g., feed conversion efficiency) for heritage chicken breeds commonly used on open-environment farms.
- **Long-Term**: In a timeframe beyond 5 years, heritage breed selection may become a key strategy for disease management in organic poultry production. This will enhance the resilience and sustainability of organic farming practices which do not have the tools for disease management available to conventional producers.

Additional funding applied for/secured: A grant is currently in preparation for submission to the USDA-NIFA *animal health* program.

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Recommendations for future research: Future work should address how adult chickens from different heritage breeds respond to infections with parasites and pathogens common on open-

environment farms. In addition, future work should address how breeds respond to co-infections (i.e., simultaneous infections by different organisms). Co-infections were commonly observed on open-environment farms (Cornell et al., 2021), but they have rarely been studied in research on infectious disease.