

Radiographic Candling: A New Method To Evaluate Embryo Survival In Speckled Or Thick-Shelled Eggs

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Egg candling is essential to managing bird egg incubation, as it allows us to determine embryo growth, survival, and position within the egg. However, embryos are difficult to distinguish in birds with dark, mottled, or thick shells using a traditional light candler. Radiographs are often used at the end of incubation to ascertain the position during hatch of valuable chicks. We therefore decided to evaluate the practicality of using radiographs earlier in incubation to “candle” thick-shelled eggs. Depending on when the bones become detectable by radiographs during development, this technique could be a useful new tool to monitor embryo development and mortality in speckled or thick-shelled eggs.

To determine at which stage ossification of the cartilaginous skeleton is sufficient to be consistently detectable, embryos were euthanized at days 12 (stage 38), 12.5, 13 (stage 39), 13.5, 14 (stage 40), 14.5, 15 (stage 41), and 18 (stage 44) of incubation. In addition, embryos were fixed, cleared, and stained with Alizarin red (stain calcified bone) and Alcain blue (stain glycoproteins in cartilage) to visualize the amount of ossification at each stage of embryo development.

Our results showed that bone development was first detectable by radiographic analysis by days 12.5 to 13 (stages 38-39). This timeframe is within the early part of the rapid bone mineralization process that occurs from day 10 through 17 (stages 36-43), as previously shown by Thompson *et al.* (1989).

Our study shows that incubation managers can monitor the growth of avian embryos in thick-shelled eggs using radiographic candling from stages 38-39, which could significantly impact conservation efforts. Due to the potential harm, we envision radiographs being used at stage 39 to confirm the presence of an embryo, followed by a second radiograph several stages later to confirm continual growth and/or hatch position. This will minimize the number of radiographs and potential harm to the embryo’s health and germline. Actual detection will depend on equipment resolution and operator experience, as the early bones were difficult to distinguish from shell artifacts at stage 38.

In summary, we found that embryos in speckled or thick-shelled eggs were readily detectable using radiographic candling by stage 39.