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# USING CANDLERS TO DETERMINE THE INCUBATION STAGE OF PASSERINE EGGS

JOHN T. LOKEMOEN AND ROLF R. KOFORD[1]

*Northern Prairie Science Center*
*National Biological Service*
*8711 37th Street SE*
*Jamestown, North Dakota 58401 USA*

Abstract.—Determining the incubation stage of bird eggs can provide important information to investigators conducting nesting studies. We describe the use of candlers in the field to determine the incubation stage in eggs of Lark Buntings (*Calamospiza melanocorys*) and other small birds with an incubation period of 11–13 d. Candling was accomplished easily using simple tools and did not involve the destruction of eggs or lengthy disturbance of nests. Candling is often preferable to other methods that rely on egg mass, mass-growth curves, or immersion of eggs in water.

**UTILIZACIÓN DE ALUZADORES PARA DETERMINAR LA ETAPA DE INCUBACIÓN DE HUEVOS DE PASERINOS**

Sinopsis.—El determinar la etapa del periodo de incubación puede proveerle al investigador información sumamente importante. Describimos el uso de aluzadores, en el campo, para determinar la etapa de incubación de huevos de *Calamospiza melanocorys* y otros paserinos con periodo de incubación entre 11–13 días. El aluzado se llevó a cabo fácilmente utilizando herramientas sencillas que no conllevan la destrucción del huevo o disturbios prolongados. El aluzamiento muchas veces es preferible a otros métodos que dependen de la masa del huevo, curvas de incremento en masa o la inmersión del huevo en agua.

In nesting studies, it is often desirable to determine the incubation stage, in days, of an egg or clutch of eggs. By knowing the incubation stage, investigators can estimate the dates on which eggs were laid and incubation was initiated, as well as predict the date of hatching and fledging. This information helps investigators calculate the optimum date to determine nest fate and the best date to band or mark young (Bart 1977).

In previous studies of waterfowl, we (Lokemoen et al. 1984, Koford et al. 1992) used field candlers (Weller 1956) to age embryos in eggs because candlers are a rapid, simple, and accurate method. Eggs can be candled in the field using a simple candling device, ambient light, and a chart illustrating incubation stages. A candler can be used to view the embryos in most eggs, but not within shells that are dense or too heavily pigmented to transmit light. In comparison, other embryo-aging methods are more difficult to use in the field or require sacrificing embryos. The flotation method of aging eggs has drawbacks due to errors in aging larger eggs or later-laid eggs (Westerskov 1950). In addition, warm water must be carried in the field. Determining embryo age from egg density requires accurate field measurements of egg mass, length, and width, and development of a linear regression of egg size to egg mass from eggs of

[1] *Current address: National Biological Service, Iowa Cooperative Fish and Wildlife Research Unit, Iowa State University, Ames, Iowa 50011 USA.*

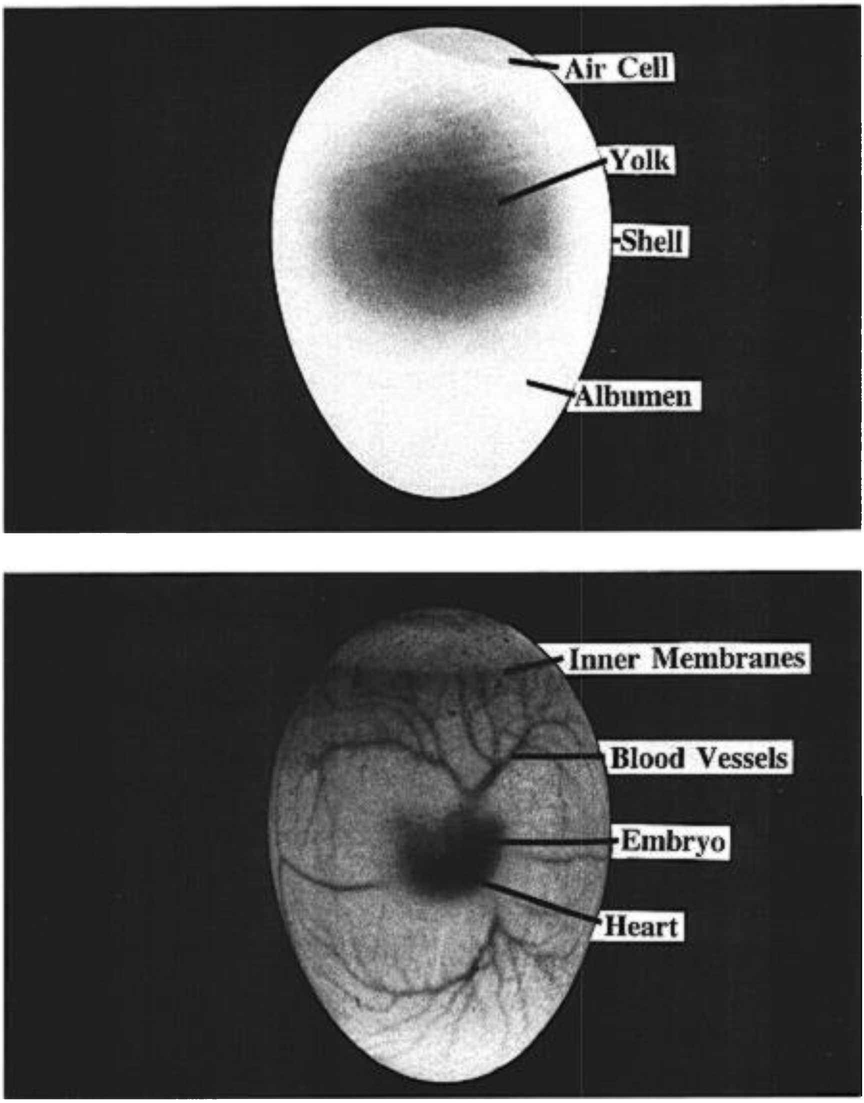
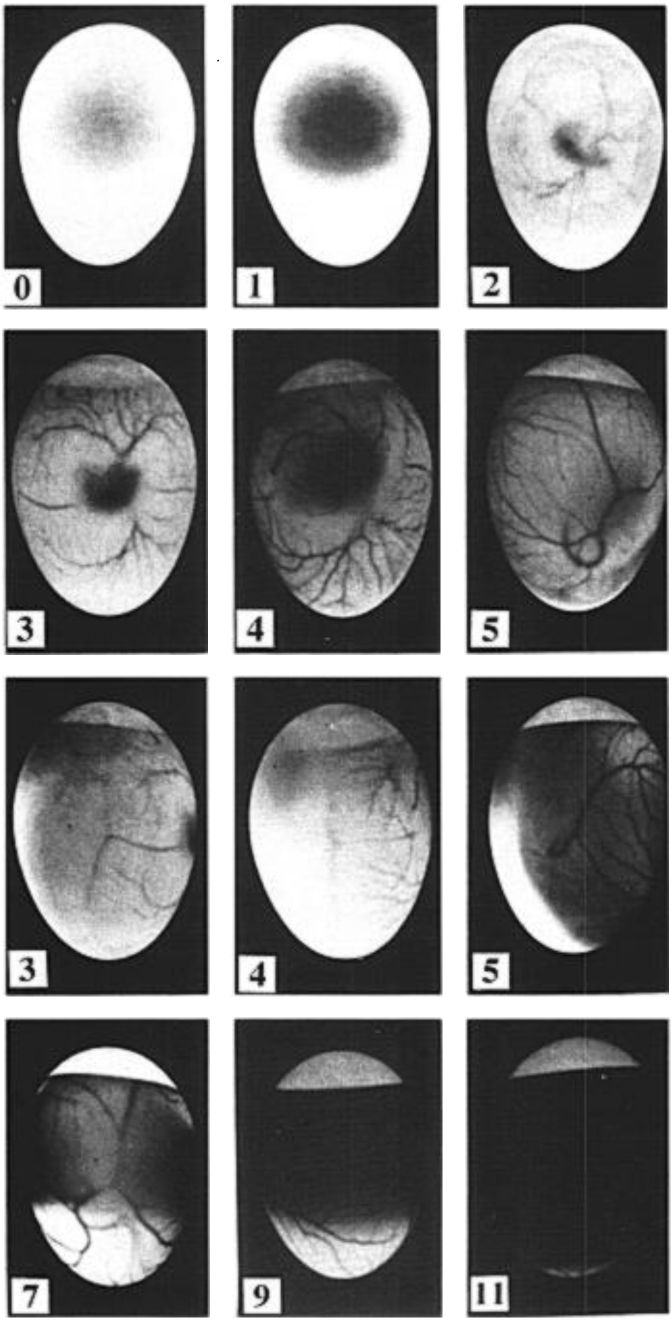



FIGURE 1. Major components of a Lark Bunting egg early in development. The heart is distinguishable as the darkened area within the embryo.

known age for each species of interest (Green 1984) Examining the embryo directly to determine age is a problem because one or more eggs in each clutch must be destroyed (Caldwell and Snart 1974).

During nesting studies in 1991–1993, we adapted and developed a candling method to age embryos in eggs of passerines and other small birds.

## Lark Bunting



METHODS

*Study profile.*—In 1991, we estimated the age of embryos in passerine eggs in the field using information provided for waterfowl eggs by Weller (1956) and for Mourning Dove (*Zenaida macroura*) eggs by Hanson and Kossack (1963). The success of these efforts prompted us to rate the ability of our field assistants to age embryos in 1992. We compared the estimates of embryo age by six field assistants with the actual date of hatching for 40 passerine nests. The sample included Lark Bunting (*Calamospiza melanocorys*), Horned Lark (*Eremophila alpestris*), and Vesper Sparrow (*Pooecetes gramineus*) clutches that were found during the egg-laying stage and that ultimately hatched. Although we had no pictures of passerine egg development in 1992, we described the technique to the field assistants and provided pictures of waterfowl egg development (Weller 1956) scaled for passerine eggs.

The 1992 tests were encouraging so we developed a technique to photograph incubation stages in eggs in 1993 and photographed the daily stages of egg development in 1994. Photographs were made from known-age eggs collected in the wild and incubated in incubators until hatching. Egg photographs were mainly of species of small passerines with incubation periods of about 11–13 d.

*Field candling technique.*—For candling eggs in the field we used only ambient light and portable candling tubes. The tubes were made of 15-cm lengths of foam pipe insulation. Foam insulation with inside diameters of 1.3 cm and 1.6 cm is suitable for candling most small bird eggs. The foam is easily shaped around the egg to screen out light from the side when viewing.

Eggs are held at the distal end of the candling tube but are not placed entirely inside the tube. The egg is held vertically, with the blunt end or air cell of the egg upward and the pointed end of the egg downward. Generally, eggs incubated fewer than 3 d are best viewed while looking toward the ground, and eggs incubated 3 d or more are best viewed against a bright sky. Embryonic development is not symmetrical so the egg must be turned on its long axis to view the air cell, embryo, and other details from various positions. In the field, we candled at least 3 eggs in each clutch, because some eggs may be dead or infertile. The eggs of all species, especially passerines, require careful handling because they are quite fragile.

*Photographing embryonic development.*—Most of our photographic work

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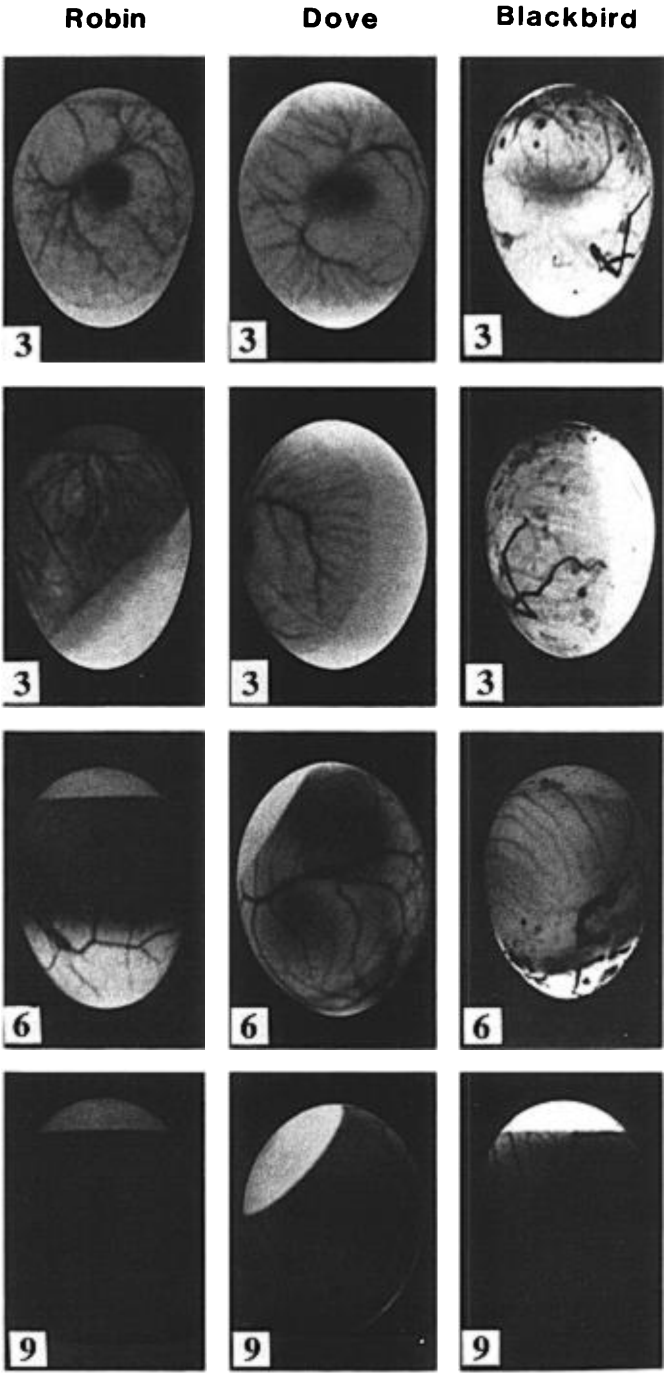
FIGURE 2. The incubation stages of a Lark Bunting egg. The top two rows of pictures show developmental stages from day 0 through day 5 with the yolk or embryo in the center of the egg. The third row of pictures shows the same developmental stage as row two, but the egg is turned 90° and the embryo is at the side of the egg. The fourth row shows egg development stages of day 7, day 9, and day 11 with the embryo having grown so that it fills most of the egg interior.


Robin
Dove
Blackbird
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9

on embryonic development was done with Lark Bunting eggs ($n$ = 6), which photographed well because the egg shells are translucent and unmarked. For comparative purposes, we also examined and photographed the incubation stages of eggs from Mourning Doves ($n$ = 3), House Wrens (*Troglodytes aedon,* $n$ = 2), Eastern Bluebirds (*Sialia sialis,* $n$ = 1), American Robins (*Turdus migratorius,* $n$ = 2), Red-winged Blackbirds (*Agelaius phoeniceus,* $n$ = 5), Brown-headed Cowbirds (*Molothrus ater,* $n$ = 2), and Common Grackles (*Quiscalus quiscula,* $n$ = 1).

Eggs were placed in an incubator that automatically controlled temperature (38 C), relative humidity (50–55%), and turned the eggs each hour. Incubation periods of eggs in the incubator were compared with those reported by Harrison (1979) to ensure that we were quantifying natural embryonic development.

Eggs were removed from the incubator each day to photograph and document embryo development. Each egg was placed horizontally into a cradle that held the egg firmly on its side, but allowed artificial light to be projected from below. The cradle was lined with plasticine to make a light-proof seal. A C-700 Super Chromega enlarger colorhead was used as the source of light. We found that green or cyan light projected from the enlarger gave the most natural portrayal of blood vessels and embryo within the yellowish-hued egg interior. A single-lens reflex camera, equipped with a 50-mm macro lens, was held above the egg on a copy stand, with the lens pointed vertically at the side of the egg. We used shutter speeds of 0.5–12 s and F-stops of 4.0 to 16.0 and three types of black-and-white film: Kodak Plus-X Pan (125 ASA), T-Max (100 ASA), and Tri-X Pan (400 ASA). Use of brand names does not constitute endorsement by the U.S. Government.

## RESULTS

Candling proved to be a useful technique for determining embryo ages in eggs of Lark Bunting and species with similar incubation periods. Most passerines have egg shells that are sufficiently translucent to be viewed easily with field candlers. Some species that have heavy shell markings, such as the Red-winged Blackbird, can be aged by candling, but other species, such as the House Wren, are more difficult to view. Also, in developing our technique, we found that it was not possible to age eggs of most shorebirds, such as Killdeer (*Charadrius vociferus*), Marbled Godwit (*Limosa fedoa*), and Willet (*Catoptrophorus semipalmatus*), because the dense shell blocks light penetration.

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FIGURE 3. The incubation stages of the American Robin, Mourning Dove, and Red-winged Blackbird. The top two rows of pictures show development during the third day of incubation for the three species. The embryo is in the center of the egg in row one, but the egg is turned 90° in row two and the embryo is at the side of the egg. The third and fourth rows of pictures show egg incubation during the sixth and ninth days of development when the embryo begins to fill the center of the egg.

*Egg development.*—Development of eggs in the incubator paralleled that we observed in the field. For the focal species in this study, the period of egg incubation that we observed in the incubator was similar to that listed by Harrison (1979). For eggs artificially incubated in 1994, the two Lark Bunting eggs hatched in 12 d, the American Robin egg in 12 d, the Mourning Dove egg in 14 d, and one Red-winged Blackbird egg in 11 d and the other in 12 d. Major egg features are depicted in Figure 1.

The general stage of incubation is immediately apparent as soon as the egg is viewed through the candler. Eggs that are relatively clear are either infertile, unincubated, or in the first 4 d of incubation. Eggs that show dark patterns mainly in the center of the egg are in mid-incubation, about 5–8 d, and eggs that are quite dark are in late incubation, about 9–11 d for passerines or 10—13 d for Mourning Doves. Development of embryos in Lark Bunting eggs is shown in Figure 2 and the comparative development of embryos in Mourning Dove, American Robin, and Red-winged Blackbird eggs is shown in Figure 3. A description of the daily stages of embryonic development is presented in Table 1.

For the 40 nests in our test of field assistants, the mean error rate between the hatch date estimated when the nest was located and the actual date of hatch was 1.27 d (SD = 1.037, range = 0–4 d). Given the inherent variability involved in egg incubation, it is probably not possible to estimate hatching with much greater accuracy.

DISCUSSION

Based on our results, trained field assistants can routinely age embryos in eggs of Lark Buntings and similar species to within a day or two by candling. However, estimates of hatching date can vary by two or more days for a variety of reasons, including: (1) environment-induced changes in the length of the incubation period, (2) inherent variation among individual embryos, (3) investigator error in age estimates, and (4) embryo development continues throughout the day.

The progression through developmental stages appeared to be the same for many species of bird eggs that we examined in the field. By scaling for longer incubation periods, we were able to obtain reasonably accurate ages for eggs of Mourning Doves and Western Meadowlarks (*Sturnella neglecta*). Some features are recognizable across a broad range of taxonomic groups. For example, the distinct line seen separating blood vessels from clear albumen on days 4–6 for passerines is similar to that seen on days 8–12 in Mallard (*Anas platyrhynchos*) eggs (24-d incubation, Weller 1956). Some features seem to be species or genus specific. For example, the air cell tips well down the side of the egg in the Mourning Dove and to a lesser extent in waterfowl, but tips little in passerines we examined.

Welty (1982:356) noted that most passerine species lay eggs at 1-d intervals until the clutch is completed. The day of clutch initiation can be estimated by counting back one day for each day of incubation and one day for each egg present in the clutch, assuming that incubation began

TABLE 1. Descriptions of the daily incubation stages of American Robin, Lark Bunting, and Red-winged Blackbird eggs with comparative information for Mourning Dove eggs.[a]

| Day | Yolk or embryo | Air cell | Blood vessels |
|---|---|---|---|
| 0 | Interior is uniformly light and clear; yolk is indistinct located equidistant from sides of egg. | Small or possibly not visible if egg is still warm. | None |
| 1 | Yolk becomes more clearly defined and more orange in color. | Identifiable, but remains small. | None |
| 2 | Small embryo visible, perhaps late in day. Embryo appears as small dark spot (red) in center of egg. Yolk rotates freely and floats toward upper side with embryo on top. | Remains small. | A few narrow vessels attached to embryo. |
| 3 | Embryo is now discernible as a curved dark expanse in the center of the egg. With good conditions the beating heart can be seen, but embryo moves little. | Quite apparent in all species. | Extend from embryo to form a network across one side of the egg below air cell. |
| 4 | Embryo expands in size; in good light embryo can be seen moving quite rapidly. | Similar to day 3. | Dark vesseled areas separated from clear areas by line which usually extends vertically midegg. |
| 5 | Egg interior is largely obscured by vessels, but embryo can be seen and it moves regularly. | Increase in size for all species. | Vesseled areas near air cell expand and clear areas decrease. |
| 6 | Embryo indistinct often appearing as two dark lobes. | Larger and easily seen. | Completely encircle the egg interior. |
| 7 | Embryo seen as a dark shape below air cell. Embryo still shows some movement. | May tip slightly to side in passerines. | Continue to expand within the shell. |
| 8 | Development similar to day 7; embryos of passerines show little movement. | Similar to day 7. | Similar to day 7. |
| 9 | Due to embryo growth, details become difficult to observe. | Prominent for all species. | Vessels can be seen at each end of egg. |
| 10–11 | Egg is almost totally opaque; only light at air cell. Embryo does not move until just before hatching, when bill breaks inner shell membranes. | At greatest extent. | Mainly obscured. |
| 12 | Passerine embryos chip through egg shell and hatch. | | |

[a] Egg development for all three passerine species is similar throughout incubation period and for Mourning Doves through day 7. However, air cells of Mourning Doves tilt down the side of shell, whereas passerine air cells extend across the top of shell. After about 7 days, embryo development in Mourning Doves lags behind embryo development in passerines because Mourning Doves require two additional days to hatch. As a result, passerine embryos stop movement on day 9, whereas Mourning Dove embryos stop movement on day 11. Also, it is possible to view details in passerine eggs through day 9, but eggs become mostly opaque on days 10 and 11, whereas it is possible to view details in Mourning Dove eggs through day 11, but eggs become mostly opaque on days 12 and 13.

on the day that the last egg was laid. If incubation in a given species typically begins before the last egg is laid, appropriate adjustments can be made.

Candling eggs can increase accuracy of nesting data collected in field studies of passerines because the age of the embryo is known. Candling also increases the efficiency of field work because fewer nest visits are required to determine egg hatching success. Reducing the number of visits at a nest can reduce bias such as increased nest abandonment and increased predation associated with nesting studies (Bart 1977, Westmoreland and Best 1985). Knowing the age of the embryos would be doubly important for researchers wanting to mark or study young at hatching. The candlers we employed were easy to use and easy to transport. Young (1988) described another type of portable egg candler made from a plastic jug, but this style is bulkier and more difficult to carry.

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