A simple device for determination of incubation stages in eggs

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Introduction

In many field studies it is desirable to determine the incubation stage of eggs when nests of unknown age are found, e.g. to calculate laying dates, or to decide when nests should be revisited in nesting success studies. Many methods have been described, which basically fall into two categories. Firstly there are methods in which the size of the embryo is judged, and secondly those based on weight changes during incubation.

Measuring embryos in opened eggs may be the most accurate method to determine the incubation stage, but obviously this should not be done in nesting success studies, nor with rare or threatened species. Descriptions of embryos are given for Pheasant *Phasianus colchicus* (Fant 1957), Redhead *Aythya americana* (Weller 1957), Common Tern *Sterna hirundo* (Dunn *et al.* 1979), and Eider, *Somateria mollissima* (Gorman 1974). Sizes of embryos can also be satisfactorily estimated by shining a bright light through the egg (candling). Candling methods have been described for Mallard *Anas platyrhynchos,* Wood Duck *Aix sponsa,* and Bobwhite *Colinus virginianus* (Hanson 1954), Redhead (Weller 1956), and Pheasant (Westerkov 1950). Candling is now widely used in waterfowl and game bird studies, as it is easily performed in the field. However, it is impracticable with boldly marked eggs, such as those laid by waders. In such cases one has to rely on changes in weight characteristics.

It has long been known that birds' eggs lose weight during incubation, due to evaporation (Foster & Balfour 1883; Patten 1921). Weight loss is enhanced by the increasing metabolism in the embryo (Westerkov 1950), which is also demonstrated by the fact that within one clutch infertile eggs lose less weight than fertile ones (Veldman 1982). Relative evaporation speed depends on temperature and egg size (volume-surface ratio), as does the incubation period (Pringle & Barott 1937; Worth 1942; Veldman 1982). In spite of slight modifications by ambient humidity (Horton 1932; Pringle & Barott 1937), birds manage to keep evaporation speed remarkably constant (Ar & Rahn 1980). During incubation, evaporation per unit of egg surface is very similar in different species with different egg sizes (Drent 1970).

Since the shape of the egg does not change during incubation, weight loss results in a change of specific gravity (S.G.). In the Pheasant's egg the S.G. changes from 1.06 to 0.8 (Westerkov 1950). Similar values were found for eggs of Starling *Sturnus vulgaris* (Hayes & LeCroy 1971), by dividing the egg weight by the egg volume. Volumes were calculated with the formula $V=0.524LB^2$ (Romanoff & Romanoff 1949), in which V is the egg volume, L the egg length, and B the egg diameter. The formula assumes identical shapes for all eggs, which is certainly not true. Volumes tend to be over-estimated in conical eggs, and under-estimated in spherical ones (Barth 1953). Consequently, the values of the constant (0.524 in Romanoffs' formula) differ slightly between different authors (Barth 1953), depending on which species they have studied. Veldman (1982) found that a similar formula, developed by Bergtold (1929) and Westerkov (1950), which uses 11/21.5 as a constant, resulted in an average over-estimate of 7.7% in the strongly conical eggs of Lapwing *Vanellus vanellus.*

All methods which use direct weighing are impracticable in field studies, because of the accuracy needed. However, since the S.G. of the egg changes from slightly more to slightly less than the S.G. of water, floating characteristics in ordinary water give a useful and fairly accurate, indirect, clue to the weight changes. It has long been known that eggs to be taken for consumption can easily be tested for their freshness by immersing them in water. Simple water tests have been described by Patten (1921) and Barth (1953), while more elaborate tests have been worked out for eggs of Pheasant by Westerkov (1950), Common Tern by Hayes & LeCroy (1971), Western Gull *Larus occidentalis* by Schreiber (1970), and Starling by Dunn *et al.* (1979).

This paper describes simple floatation

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tests for eggs of some wader species, which can readily be carried out in the field, with the aid of a specially developed device, the 'incubometer'. The species for which tests are described are Lapwing and Black-tailed Godwit *Limosa limosa.* These species breed in high densities in Dutch meadows and pastures, and are, together with Oyster-catcher *Haematopus ostralegus,* Redshank *Tringa totanus,* Ruff *Philomachus pugnax,* and Snipe *Gallinago gallinago,* collectively known as 'meadow birds' in The Netherlands. The incubometer was developed to be used in the population studies of these six species of waders, part of the research programme of the Research Institute for Nature Management.

Methods

For his own use, Van Paassen (1980, 1981) developed a method of visual inspection, without taking any measurements, and without any equipment. Tests were simply carried out on the hand, in a nearby ditch. He described six clearly distinguishable stages, which help to estimate the hatching date with an accuracy usually of less than two days. The method is especially useful in the first half of the incubation period, when eggs do not yet float, and the behaviour of an egg in water changes very rapidly with the progress of incubation. The six stages are described in Table 1.

In order to standardize the method, also making it applicable for untrained persons, and also to investigate the second half of the incubation period (floating stage), we developed a simple apparatus, the 'incubometer'. It is basically a perspex vessel, internally 7 cm long, 4 cm wide, and 7 cm high. The outer part (Fig. 1) can be shifted up and down, and the sides of the vessel are calibrated in millimetres. The vessel is filled with water and the egg is carefully placed in it. To reduce breaking risks, the bottom is lined with a thin piece of rubber.

With this incubometer, Veldman (1982) followed the incubation stages in 45 Lapwing nests and 11 Black-tailed Godwit nests. Eggs were individually marked, and were tested daily, with the exception of those in two Lapwing nests, which were used to investigate the influence of daily immersion. In all eggs the following measurements were taken:

1. Sinking stage.
   a) the angle between egg axis and the horizon,
   b) the relative increase of the height to which the blunt end of the egg is raised.
2. Floating stage.
   a) the angle between egg axis and the horizon,
   b) the percentage of the egg diameter visible above the surface,
   c) the height of the part above the surface.

Care was taken that visits to the nests were as short as possible. Nests were not disturbed during adverse weather. A record was kept on survival and hatching success of all nests.

Not all the eggs could be followed during the entire incubation period. Some eggs were lost (due to predation or agricultural activities), and many nests were found which were already completed. Therefore, data had to be combined from eggs which had been followed through the sinking stage, and eggs which had been followed through the floating stage. Sixteen Lapwing

Table 1. Distinguishing features of stages of incubation in eggs of Lapwing ***Vanellus vanellus*** **and Black-tailed Godwit** *Limosa limosa.* (from Van Paassen 1981).

Stage	Description	Number of days incubated	
		Lapwing	B-t Godwit
1	egg lays flat in the hand	0 – 2	0 – 5
2	blunt end is raised noticeably, egg axis makes angle with hand	2 – 4	5 – 8
3	egg is raised to complete vertical position, but still sinks	4 – 7	8 – 10
4	egg starts to float but refuses to come to the surface	8	11
5	egg comes rapidly to the surface, but hardly emerges	9 – 11	12 – 15
6	egg emerges for great part (up to a fifth) above surface	11 – 26	15 – 24

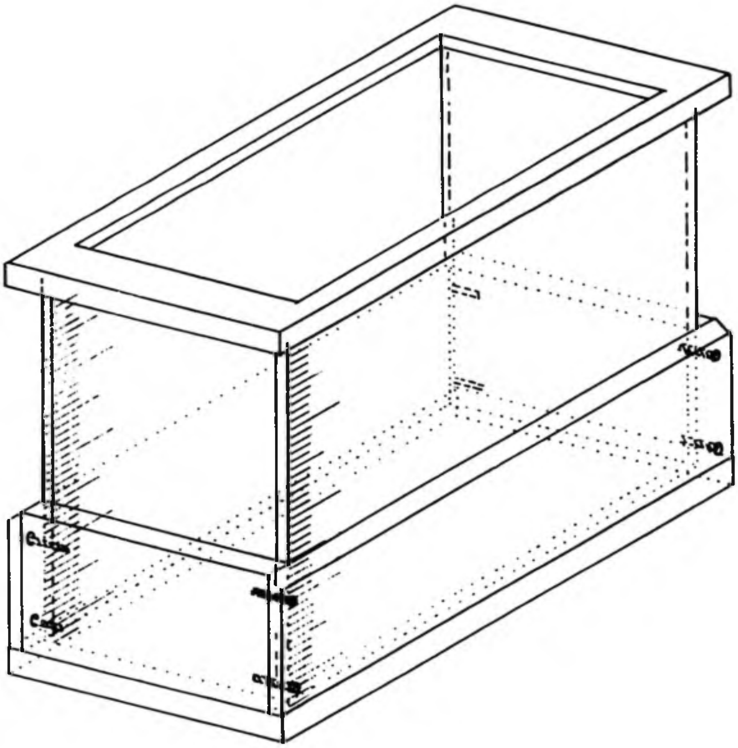

Figure 1. Design of the incubometer. The outer part can be shifted up and down, which enables very accurate readings of the height above the surface in floating eggs.

eggs and five Godwit eggs were followed throughout the entire incubation period.

Results

Fresh eggs sink, and lie as flat as their shape permits. In both Lapwing and Godwit eggs, the angle between egg axis and horizon is 25–35°. In Lapwing eggs, there is a noticeable difference between relatively small eggs ($V < 24.5$ cm^3 according to the formula of Bergtold and Westerkov), and larger ones. In small Lapwing eggs, large Lapwing eggs, and Godwit eggs, the average initial angle is 27°, 32°, and 28° respectively. This angle quickly increases when the egg is incubated, and the air chamber grows. The increase is especially rapid between the fourth and the seventh day. After nine days the egg stands almost vertically in the water, and is very close to floating. The length of the sinking period is 11 days for small Lapwing eggs, and 12 days for larger ones and Godwit eggs. Since a small Lapwing egg starts with a smaller angle, it rises more quickly than a large one. The floating period takes 16 days in a small Lapwing egg, and 15 days in a large one, so both have the same average incubation period of 27 days. Despite its much larger size, the Godwit's egg has an incubation period of only 24 days, and, consequently, it has a floating stage of only 12 days. The increase of the height to which the floating egg is raised above the surface is almost exactly linear. Shortly before hatching, both Lapwing and Godwit egg emerge 6 – 7 mm above the surface. Since small Lapwing eggs start to float one day earlier than large ones, they keep ahead by about 0.5 mm throughout the floating stage.

Lapwing eggs float in an almost vertical position, Godwit eggs make an angle of 80° with the surface. This angle hardly changes during the first week of the floating period. In the last few days before hatching, the chick changes its position within the egg, which results in a second, very rapid change in the angle between egg axis and horizon.

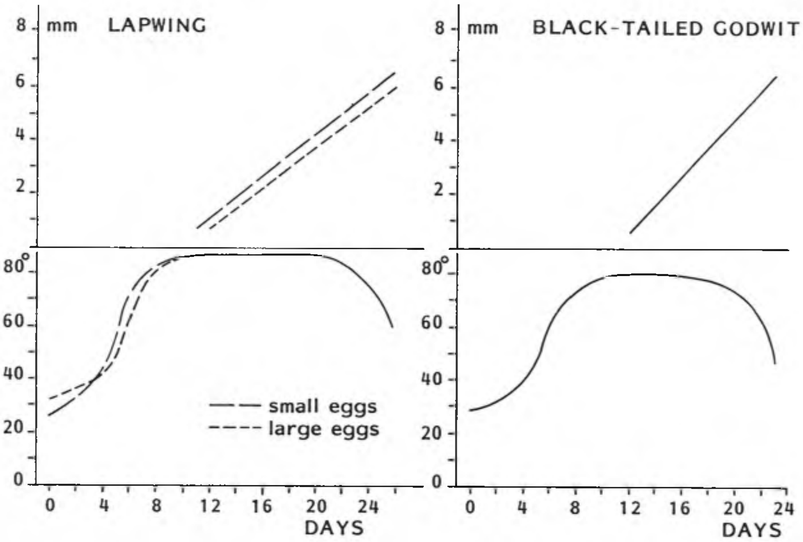

Figure 2. Calibration curves for Lapwing *(Vanellus vanellus)* **and Black-tailed Godwit** *(Limosa limosa).* The lower half of the figure shows the angle between egg axis and horizon, as a function of the number of days eggs have been incubated. The upper half shows the height above the surface during the floating stage.

The angle decreases to 65° in Lapwing eggs, and 55° in Godwit eggs.

During the first week of the sinking period, the relative increase of the height to which the blunt end of the egg is lifted is as good a parameter as the angle between egg axis and horizon. However, when this angle exceeds 75°, it further increases in height so little that it no longer gives any clue. Therefore, we only present the results based on the angle of the egg axis.

Similarly, during the floating stage, the relative diameter of the emerging part is rapidly increasing during the first days, but only very little later in the stage. Furthermore, this diameter is not always easy to judge, and the variation is great, due to different egg shapes. Therefore, we only present the results of the increase in height above the surface. The curves, which can be used to calibrate the incubometer, are given in Fig. 2.

Discussion

Table 2 gives the numerical distribution of the deviation (in days) between estimated and actual incubation day, when only the angle of the egg axis is used in the sinking stage, and the height above the surface in the floating stage. It can be seen that mistakes up to nine days are possible in floating Lapwing eggs, and up to five days in other cases. In spite of this, over 90% of all estimates are accurate within two days in the sinking period, and three days in the floating stage. Therefore, the method seems to be reliable enough for many practical purposes in the field.

When comparing Fig. 2 with Table 1, there is a discrepancy in the judgement of the day at which eggs start floating. This is probably caused by the fact that, when the egg has an S.G. close to 1.0, even the slightest vibration will cause it to lose contact, when it is held in the hand, and then submerged.

Surprisingly enough, the incubometer method is not more accurate than simple visual inspection by an experienced observer. Judging the angle of the egg axis with the naked eye is comparable to estimating minutes on a clock with only the quarters of the hour marked. The great advantage of the use of a calibrated incubometer, however, is that the results of different observers can be directly compared. The individual user will quickly learn to make his estimates with the naked eye, at least in the sinking stage. For each new species the user will have first to develop calibration curves. We do not believe that a general method can be developed, applicable to many bird species,

Table 2. Accuracy of estimates of incubation stage in eggs of Lapwing and Black-tailed Godwit.
dev = mean deviation in days
abs = mean of absolute values of deviation in days
N = number of observations

accuracy in days	percentage of observations that fall within accuracy			
	Lapwing		Black-tailed Godwit	
	sink	float	sink	float
0	27.6	27.2	34.4	29.9
1	68.7	59.9	75.0	60.6
2	90.9	79.5	92.5	81.9
3	97.3	90.3	97.5	94.5
4	99.0	96.1	98.1	99.2
5	100.0	98.4	100.0	100.0
6		99.4		
7		99.8		
8		99.9		
9		100.0		
dev	.0	.0	.1	.3
abs	1.2	1.5	1.0	1.3
N	608	830	160	127

as Schreiber (1970) suggests.

For convenient field use, we suggest that it is sufficient to use two eggs in a clutch, and to take a third only when these two show a large difference. Eggs within one clutch should show little difference in their development, as the first laid eggs are kept slightly warm until the clutch is completed, and incubation starts (Stiefel 1961; Dunn *et al.* 1979). Of course, in species which start incubating when the first egg is laid, all eggs should be tested individually.

When two eggs show a very large difference, one should consider the possibility that either the heavier egg is not fertilized, or the lighter one is slightly damaged. Weight loss in infertile eggs can, however, resemble the development in a normal egg. Infertile Lapwing eggs tend to float after 13 days, and rise up to 4 mm above the surface after 15 days in the floating stage. Evaporation depends on temperature, as abandoned eggs hardly changed over a period of fourteen days in cold weather.

Eggs which show even barely visible cracks lose weight much more rapidly. Damaged Lapwing eggs float within a week, and rise up to 10 mm above the surface two weeks later. Most embryos do not survive dehydration under these circumstances, although some of these eggs hatch normally.

Daily immersion did not influence the incubation period. In two nests only part of the clutch was treated daily. Other eggs were immersed with intervals of three, five, and ten days respectively. In both nests four chicks hatched normally, at the same time. The average incubation period in 22 Lapwing nests in which eggs were immersed daily, was 26.9 days, which is not different from the normal average (Glutz, Bauer & Bezzel 1975).

Summary

Many methods to determine the stage of incubation in birds' eggs are available. In field studies of species which have boldly marked eggs, like waders, only those methods using floating characteristics are practicable. In order to standardize methods, and to obtain comparable results from different researchers, we developed an 'incubometer', and calibrated it for the Lapwing *Vanellus vanellus* (based on 45 nests), and the Black-tailed Godwit *Limosa limosa* (based on 11 nests).

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