

# Hatching egg quality – establishing the viability of the embryo

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The evaluation of hatching egg quality is an essential and critical part of standardised procedures for quality assessment in modern hatcheries. Typically these procedures scrutinize the eggshell and its contents, as these play an important role in supporting the needs of the growing embryo. Often little or no attention is in fact paid to the quality and viability of the embryo itself, which is surprising as these factors are the primary characteristic of a good quality hatching egg.

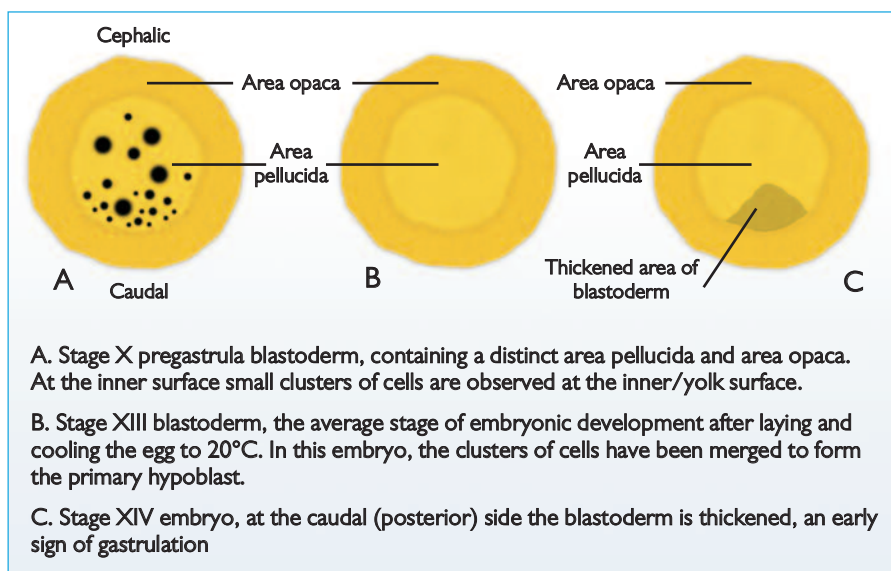
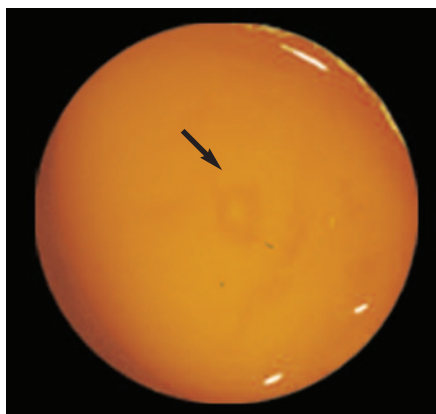
In this article, the viability of the embryo in unincubated eggs is discussed in relation to development in the hen's body and during egg handling on the farm and at the hatchery.

## Morphology

Generally at the hatchery, egg quality is evaluated by a superficial, overall description of shell quality and uniformity in the batch. Hatchery managers will break unincubated eggs only when serious fertility problems are suspected.

The fertile, unincubated egg contains an embryo (blastoderm) that developed from the fertilised oöcyte during egg formation in the oviduct. The oöcyte is the female gamete that floats on the yolk. Shortly after the release of the oöcyte in the oviduct, sev-

**Infertile egg: degenerated oöcyte (arrow).**



**Fig. 1. Schematic overview of embryos in the unincubated egg, directly after laying (A) and after cooling (B). Both embryos are in the pre-gastrula stage of development. (C) Schematic overview of a blastoderm after three hours of incubation. In this embryo, gastrulation has started and a clear thickening at the caudal (posterior) end of the embryo has formed (after *Early Embryology of the Chick*, by Bradley M. Patten, Fourth Edition).**

eral spermatozoa (male gametes) penetrate the yolk membrane, after which only one spermatozoon fuses with the oöcyte to form the single-cell embryo (zygote).

Finally, during egg formation in the oviduct and shell gland, the zygote develops into a blastoderm with a clearly recognisable Area Pellucida (AP), surrounded by an Area Opaca (AO) (Fig. 1). If for whatever reason the spermatozoa do not reach the oöcyte, the egg remains infertile and the oöcyte will degenerate to form nothing more than a small germinal disc floating on the yolk (see photo). In the unincubated egg, the infertile germinal disc is visible as a compact white spot with ruffled irregular edges.

## The unincubated embryo

If hatching eggs are analysed before incubation on arrival at the hatchery, the viable embryo will be easily recognised as a doughnut-shaped structure, with an overall diameter of 4-5mm. The AO is clearly visible, with a white ring that stands out against the yel-

low of the yolk. In contrast, the yolk is visible in the centre of the embryo, because the AP is translucent and clear. Sometimes a whitish spot can be observed in the centre of the AP, probably because the embryo is in contact with the bottom of the subgerminal cavity, also called the nucleus of Pander.

## Embryonic development

As described, embryonic development begins after fertilisation and continues during egg formation in the oviduct. When the egg is laid, the blastoderm or 'doughnut' as it is often known, is approximately 20-22 hours old and consists of 40,000-60,000 cells. These embryonic cells have, by the time the egg is laid, differentiated into cells that are either part of the AP or of the AO.

Each of these areas pre-destined to different roles during the further development of the embryo.

The AO will mainly develop into extra-embryonic structures, including the blood

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ring (area vasculosa) and yolk sac membrane, while the cells contained in the AP will further differentiate to become the various structures that form the embryo.

The embryo's different stages of development before the egg is laid are recognised by stage-specific morphological structures.

A normal table of these first stages of chick development was published by Eyal-Giladi and Kochav in 1976 and is still used today to describe the stage of the blastoderm (embryo) at egg laying (oviposition) or at the time the hatching eggs arrive at the hatchery.

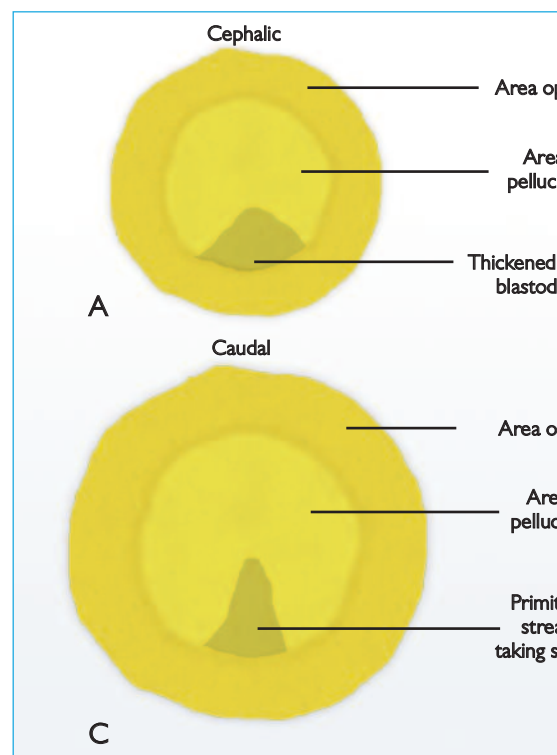
We must realise however that, firstly, the transition from one stage to the next occurs

in a very short time window. Secondly, as the eggs cool after laying, the development of the embryo continues until the temperature is below 20°C, when cell activity becomes almost zero.

The stage of embryonic development at the moment an egg is laid is influenced not only by maternal age, but also by the position of each egg in the sequence within a clutch laid by one hen.

However on average, at the point of laying, 'stage X' according to Eyal-Giladi and Kochav (1976) the blastoderm is symmetrical, with a distinct AP and AO that can be recognised by the naked eye.

In this stage of development, the AP is a thin (one-cell thick) translucent epithelium. If



**Fig. 2. Chick embryos showing four stages in the formation: (A) 3-4 hours incubation, (B) 5-6 hours incubation (based in part on the photomicrographs**

we isolate the blastoderm from the yolk and look at its inner (yolk) surface, we see that the translucent AP is not homogeneous because small clusters of cells interrupt the transparency (Fig. 1).

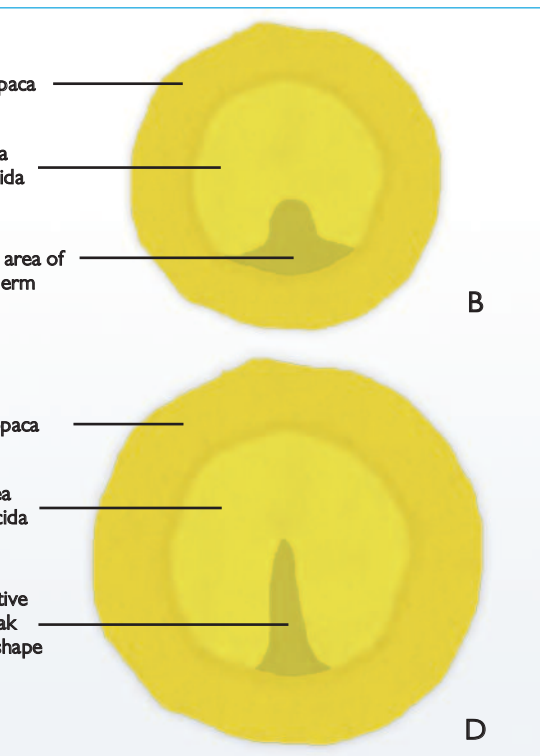
These clusters are not distributed homogeneously over the AP, but more crowded towards one side of the embryo: the future tail of the posterior structure of the blastoderm. It is thought that clusters of cells observed at the lower surface of the stage X blastoderm take part in the development of the primary hypo-blast, which is completely formed at stage XIII.

The primary hypoblast is a second embryonic layer developed below the epithelial AP. So in the freshly laid egg, the dorsal-ventral axis and the anterior-posterior (head-to-tail) axis of the future embryo and chick have already been determined (Fig. 1).

## Resistant stage

The stage X blastoderm in the freshly laid egg develops further to the pre-gastrula stage XIII in the nest and during egg handling at the farm. It has been suggested that the pre-gastrula stage XIII, when the primary hypoblast has been completed, is the most resistant stage for further egg handling and egg storage (Fasenko et al, 2001; Reyriink et al, 2008).

In stage XIII, the AP has become less translucent because the primary hypoblast covers its ventral surface. With the completion of the primary hypoblast, the chicken embryo is prepared for the next step in embryonic development: gastrulation.

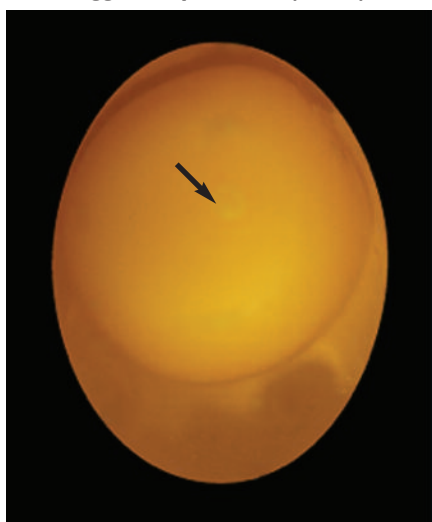


**Formation of the primitive streak during early gastrulation, (C) 7-8 hours incubation, (D) 10-12 hours of Spratt, I. Exptl. Zool., 103, 1946).**

Gastrulation is a well known and extensively studied embryonic process, characterised by (temperature dependent) cell movements and the formation of a multilayered body plan for the future chicken (Fig. 2 above).

Gastrulation-related movement of cells starts when the embryo temperature increases during incubation. The pre-gastrula stage XIII embryo has a completed primary hypoblast and is more resistant to egg handling and storage than the younger stages. As soon as gastrulation-related cell movements begin, the embryo is very vulnerable to egg handling and temperature variations. In batches of good quality eggs, the embryos will be uniformly developed to

**Fertile egg: embryonic disc (arrow).**



the pre-gastrula stage XIII, with a clear AO and two layers to the AP.

## Summary

The stages of development and their consequences for egg handling and storage are:

- In a freshly laid egg, the embryo is at stage X of embryonic development – preceding the formation of the primary hypoblast.
- Post-laying conditions may or may not support the embryo to develop to the storage resistant stage XIII chicken embryo – when formation of the primary hypoblast has completed.
- Post-laying, egg handling procedures

should allow the uninterrupted development of the embryo to the resistant stage XIII. Disruption of the developing embryo by cooling the eggs too rapidly should be prevented.

- Development beyond stage XIII should be prevented by optimum egg handling and storage temperatures below 20°C.
- In batches of good quality eggs, the embryos will have been uniformly developed to stage XIII on arrival at the hatchery.
- New discussion about the benefits of pre-storage incubation support the idea that for optimum hatching egg quality, embryos need to develop from stage X at oviposition to stage XIII for optimum storage prior to incubation. ■