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CAUSES OF NORMAL DEVIATIONS FROM THE INCUBATION PROCESS: MAIN PATHOLOGIES

CAUSAS DE DESVÍOS NORMALES DEL PROCESO DE INCUBACIÓN: PRINCIPALES PATOLOGÍAS

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Abstract

The objective of this review is to update the causes of deviations from normal values and the main pathologies of incubation, according to the results of embryodiagnosis. The success of artificial egg incubation depends on several factors, both on the breeding farm and in the hatchery. On the farm, factors such as nutrition, health management, reproduction, weight gain and uniformity, handling of fertile eggs, and their transport to the hatchery, with the application of good practices, are crucial to ensure an adequate hatchability rate. In the hatchery, storage and incubation must follow the basic principles of temperature, humidity, ventilation, and turning. These factors can cause deviations in the normal values of the incubation process, such as: infertile eggs, cracked eggs, specific gravity, contaminated eggs, embryonic mortality in phases I, II, and III, unhatched pips, and malformations. Additionally, the occurrence of perinatal pathologies, known as discard or second-grade chicks, affects the productivity of a hatchery. This includes not only chicks that did not hatch but also those that hatch but are not viable in the production system.

Keywords: embryodiagnosis, egg, breeding farm, embryonic mortality

Resumen

El objetivo de esta revisión es actualizar las causas del desvío de los valores normales y las principales patologías de la incubación, según los resultados de la embriodiagnosis. El éxito de la incubación artificial de los huevos depende de varios factores, tanto en la granja de reproductores como en la planta de incubación. En la granja, factores como la nutrición, el manejo sanitario, la reproducción, las ganancias de peso y la uniformidad, el manejo del huevo fértil y su traslado a la planta de incubación, con la aplicación de buenas prácticas, son cruciales para asegurar un adecuado porcentaje de incubabilidad. En la planta de incubación, el almacenamiento e incubación deben seguir los fundamentos básicos de temperatura, humedad, ventilación y volteo. Estos factores pueden ocasionar desviaciones en los valores normales del proceso de incubación, tales como: huevos infértiles, huevos cascados, gravedad específica, huevos contaminados, mortalidad embrionaria en las fases I, II y III, picados no nacidos y malformaciones. Además, la presentación de patologías perinatales, conocidas como pollitos de descarte o de segunda, afecta la productividad de una planta de incubación. Esto incluye no solo los pollitos que no lograron eclosionar, sino también aquellos que nacen, pero no son viables en el sistema de producción.

Palabras clave: embriodiagnosis, huevo, granja de reproductores, muerte embrionaria

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Introduction

The success of final chick quality during and at the end of incubation is determined by the identification of sub-optimal indicators of diagnostic techniques, candling and embryodiagnosis, process control procedures from laying at the rearing farm to hatching, collection of information on infertility, hatchability, timing and nature of embryo losses. These indicators constitute an important part of the quality control programme in hatcheries. They allow to suggest alternative management practices within the process and to adapt quality control routines to monitor the results of any changes made. In addition, they help to prevent recurrence of the same problems (Macari et al., 2015).

The increase in embryonic mortality (EM) during incubation is attributed to changes in physiological functioning and embryonic development due to genetic and environmental causes. Therefore, defining the behavior of EM allows having a basis for measuring a key parameter in incubation and exercising control in the face of evidence of abnormal changes and adverse conditions that may alter each of the stages of embryonic development. This is essential to implement corrective measures and address hatchability losses due to infertility and early, intermediate and late embryonic mortality (April, 2020).

Other elements to highlight are that within the systematization, not only the degree of embryonic development should be considered, during which chronologically the embryo presents a problem during incubation, but it is essential to categorize the causes of failure of embryonic development and its comparison with the historical records, which are available in the incubation plant for similar batches of fertile eggs (Juárez, 2014). These data referred to above, should be linked mainly with what is referred to by Ross (2010), with respect to the intrinsic aspects of the origin of the fertile egg, such as: the strain of the breeding hens, their age, the reproductive management carried out, type of feed, weight of the egg, quality of the shell, and with the extrinsic aspects such as: the time of year and the specialized handling of the eggs, such as: the collection, disinfection, transportation and storage of the fertile egg.

The present work aims to update part of the knowledge published to date on the causes of deviation from normal values of hatchability, embryonic development and incubation pathologies that would constitute a reference material for undergraduate and graduate students and professionals working in incubation plants.

Development

1. General characteristics of the production chain

It is important to highlight that throughout the production chain in a hatchery, a series of processes are carried out (Fig. 1) that begin in said plant with the storage of the eggs in the cold room at a temperature between 16 to 20 ºC for a maximum period of 7 days according to the demand of the poultry industry and the hatchery. Next, the eggs are selected and classified by size, shape and cleanliness. Subsequently, the eggs are loaded into the incubator machines in charge of simulating the environmental conditions in which hens incubate their eggs naturally (Yerpes, 2020).

Artificial incubators control the optimal parameters of temperature (37 0C), humidity (83.6 - 84 %) and turning that allow the physiological development of the embryos. Before starting the incubation profile, a maximum tempering of 12 hours at 24 -27 0C must be carried out to avoid thermal shock and condensation of the eggs (van der Pol et al., 2013).

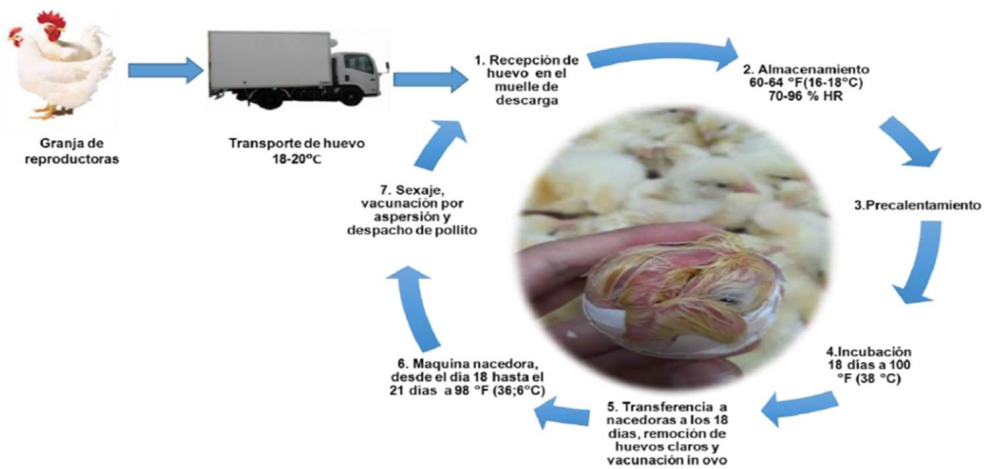

Fig. 1. Flow of the production process in the hatchery

Temperature can have a divergent or biphasic effect during the course of incubation. Increasing temperature initially accelerates embryonic growth and nutrient and energy utilization from yolk and albumen, but as incubation progresses, exposure to constant high temperatures decreases embryonic growth (Noiva et al., 2014).

It is necessary to lower the temperature level during the last 2 to 3 days of incubation, that is, this environmental indicator is adjusted according to the stages of incubation, since it is a time when the metabolic heat rate of the embryo is high, which is why the decrease in temperature stimulates the consumption of nutrients, accelerating metabolism and development in the embryos. One of the byproducts of the metabolism that occurs during incubation is water and the embryo must eliminate the excess of this nutrient in order to hatch (Wijnen et al., 2021).

During incubation, an egg loses weight due to water evaporation. This is essential to create a sufficient air chamber to allow embryonic lung ventilation after internal puncture and thus achieve successful hatching. A high hatching rate is achieved when there is a water loss of 12 to 14% of the egg weight at the time of incubation until the time of transfer (Barbosa et al., 2012).

Temperature fluctuations should be avoided during egg transport and storage. Temperature decrease should be gradual from the breeder farm to the cold room in the hatchery. The transition in egg preheating temperature (24 -27 °C “75-80 °F”) from the cold room to the incubator should be managed to avoid thermal shock of the embryo and condensation on the shell. In this way, all eggs will achieve the desired uniform temperature (van der Pol et al., 2013). Uneven preheating increases the variation in hatching time (Llamuca et al., 2024).

Schmidt (2003) points out that there are two important terms to consider in the incubation process that determine the success of the same: hatchability and embryonic development. In the commercial scenario, hatchability is the percentage of viable chicks of first quality that are suitable for breeding. It is calculated by dividing the total number of viable chicks by the total number of eggs loaded in the incubator. This percentage of hatchability is influenced by factors such as the responsibility of the breeder farms from oviposition to hatching in the incubators.

Another key element to consider, according to Sandoval et al. (2005) is the performance of the chicks during the first week on the replacement farm, especially mortality levels and body weights. Although chick performance can be influenced by on-farm management and the initial impact of hatchery procedures is often underestimated and should also be considered when problems arise.

Kuurman et al. (2003) highlight that in the case of embryonic development, avian embryogenesis is practiced in a relatively protected environment. This environment can be well manipulated in domestic birds such as Gallus gallus (chicks) where incubation has long been a commercial process. The embryonic development process begins in the oviduct and reaches the blastodermal and/or gastrulation stage of development at oviposition.

Avian embryos can be affected by “maternal effects” and by environmental conditions during the preincubation and incubation periods. In general, maternal effects on offspring include both a genetic and environmental component represented by yolk hormone deposition and embryonic nutrient utilization. These effects have a significant impact on the development of a wide range of offspring traits (Reijirink et al., 2008).

Manipulation of ambient temperature during the egg storage period, pre-warming and incubation per se significantly affects embryo development, hatching progress, chick quality at hatching and post-hatch chick development. Furthermore, they affect the acquisition of thermotolerance to the subsequent post-hatch thermal challenge (Tona et al., 2007; Tona et al., 2022).

2. Causes of deviation from normal incubation values

Among the causes of deviation from normal incubation values, infertile eggs stand out, broken eggs, specific gravity, contaminated eggs, embryonic mortality in Phase I, II and III, unborn eggs and malformations (Plano and Matteo, 2001).

2.1. Factors associated with hatchability losses due to infertility

These factors are generally related to the farm.

Infertile eggs: If as a result of embryodiagnosis it is determined that there is some fertility problem, the breeding flock at the production farm should be analyzed (Ross Tech, 2010).

Possible causes (Plano and Matteo, 2001; Moya and Bermúdez, 2017; Souza et al., 2021).

- Latrogenesis: There are many drugs that affect the fertility of flocks. For this reason, treatments must be carried out under the supervision of a veterinarian. All types of treatment carried out must be recorded in the farm's daily records in order to have precise information when infertility symptoms occur in the hatchery.
- Management of males: Infertility attributed to the management of males may be due (Galíndez and Blanco, 2016) to:
  - Excess or lack of quantity in proportion to the females.
  - Individual treatments (e.g. treatments against lice).
  - General condition: Little or excessive weight. Malformations of the lower limbs or spine (e.g. lordosis).

- The change of males: When this practice is carried out, a certain time must be considered for the formation of the harem within the batch.
- Weight loss, trauma, pododermatitis, arthritis and diseases such as cholera and parasitosis. Males may be too young or old for a given batch.
- Lack of water or its temperature, which should never be less than 7 0C or exceed 30 0C.
- Ambient temperature: Temperature extremes affect animals. In intense cold, the birds huddle together to conserve heat and the males do not work; the heat causes prostration due to imbalance.
- Inadequate nutrition: in quality and quantity.
- Nutritional imbalance and/or deficiencies: Niacin deficiency can lead to a complete lack of birth. Vitamin E deficiency significantly affects fertility.
- High animal density.
- In the case of chickens it can be due to:
- Younger or older hens cause increased embryonic mortality (Intriago et al., 2023)
- Atrophy of the ovary and the right oviduct and the presence of the very elongated left oviduct (Gairal, 2019)
- Overweight chickens (FAO, 2019)

2.2. Factors associated with hatchability losses due to early mortality

Cracked eggs: These are those that have suffered a small fissure or fracture in the shell, which is not always easy to observe. During the incubation process, they will lose moisture and at the time of embryodiagnosis they are observed to be practically empty of content or with more concentrated albumin.

The cause of a high rate of broken eggs (Ricaurte, 2005; Nideou et al., 2019; Hashemi et al., 2020) may be due to:

- Rough handling of eggs on the farm, inadequate transport (Fig. 2), poor handling in the egg room of the hatchery or during loading into the incubators.
- There are eggs that break at the time of transfer and in which a developed embryo is found, sometimes injured and with dry membranes (Fig. 3).

Poor shell quality, which predisposes to breakage and is accompanied by a high rate of contaminated eggs (due to the permeability of the shell to bacterial penetration). Poor shell quality can be due to several etiologies:

- Nutrition: Vitamin or mineral deficiency (vitamin D, calcium).

Fig. 2. Cracked eggs. Cracks are observed in hatching eggs.

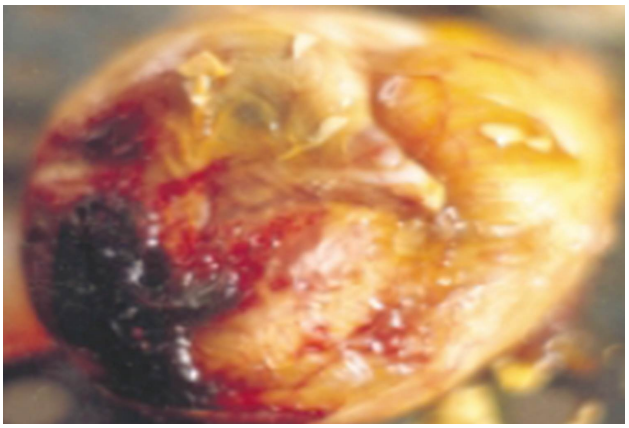

Fig. 3. Cracked eggs. Embryo trauma caused during transfer. Eggs cracked during transfer have dried membranes.

- Diseases: infectious bronchitis, postural depression syndrome.
- Ambient temperature: days of excessive heat affect the quality of the shell,
- Egg size: the larger the size, the lower the quality of the shell.
- Age of the hens: the older they are, the larger the egg.

Specific gravity: It is a way of measuring shell quality that consists of measuring the density of the eggs at different gradients of salt concentration in water (Plano and Matteo, 2001).

Procedure: In containers with a capacity of twenty liters each, water is placed at the same temperature as the eggs to be analyzed. Salt is added and measured with a densitometer (calibrated between 1.065 and 1.100), so that one container has a solution with a density of 1.065, another with 1.070, another with 1.075, and so on successively in increments of 5 until reaching a saline solution of 1.100.

50 eggs are placed in each container and the number of eggs that float for each gravimetric density is noted.

The number of floating eggs is multiplied by their density. The sum of these results is divided by the total number of eggs analyzed. This gives the specific gravity as a weighted

average. For a young batch it should be between 1.080 and 1.090. As the specific gravity decreases, the rate of cracked and contaminated eggs increases.

Contaminated eggs: At the time of laying, the egg is at body temperature, and exposure to the environment cools it down. As a result, the internal mass decreases in size and forms an air chamber, creating a vacuum that allows air to penetrate from the outside into the egg (due to pressure differences). This encourages the entry of bacteria through the pores of the shell, since the cuticle is the first barrier to prevent contamination, but since its thickness varies greatly, it does not become a good barrier.

The most common bacteria involved in egg contamination are: Pseudomona spp., Escherichia coli, Salmonella spp. (Hashemi et al., 2020; Rezaee et al., 2021; Chevalier et al., 2017). At the time of embryodiagnosis, many of the eggs have already exploded in the incubation process, but those remaining in the hatcher trays have a foul odor and a characteristic color. Contamination due to fungi gives a bluish-green color to the inside of the egg.

Plano and Matteo (2001) point out that the causes of egg contamination may be due to the specific density of the eggs, which was discussed in the quality of the shell of the previously broken eggs.

To prevent further bacterial penetration, the density should be 1.090. It is necessary to take into account the time that the egg remains in the nest and if it is a problem, it should be collected more frequently. Hygiene, cleaning and disinfection of the nests and the bedding inside them must be extreme. The above applies also to eggs and their handling, hatching egg storage and transport. It is recommended not to incubate floor eggs. handling of eggs.

Early embryonic mortality (Phase I)

It is related to the management of the incubating egg (Ross Tech, 2010), (embryo of 50,000 cells in full division). To stop the process of cell division from laying until it is placed in the incubator, it must be cooled to a temperature of 23.9 0C inside the egg in order to not affect cell vitality and in turn not stimulate it, this thermal mark is called "physiological zero" and the longer the storage time, the lower this temperature should be.

Among the main causes that are related to the management of hatching eggs (FAO, 2019; Hashemi et al., 2020, Jessen et al., 2021; Yerpes, 2020; McIlwaine et al., 2021) we can find:

- Egg storage time: If the egg is stored for more than five days, hatchability decreases by 0.5% to 1.0% per additional day.
- Storage room conditions: the temperature (18 0C) and relative humidity (between 70 and 75%) of the egg storage room must be respected; both parameters vary depending on the storage time of the eggs.
- Age of the hen: eggs from old hens should be incubated with a shorter storage time.
- If it must be older than seven days, it is better to have it from young hens, since the quality of the albumen is maintained.
- Eggs remaining in the nest for a long time: If the temperature is high, the incubation process begins there, and the embryonic structures attached to them may even form. If the egg is then cooled in the egg storage room, the embryo stops growing and inevitably dies. Embryodiagnosis shows the attached structures. If the egg is exposed to very high temperatures or solar radiation, the albumin may coagulate; clots are easily observed when examining it.
- Intense cold: leads to early embryonic mortality.
- Sudden changes in temperature and/or humidity: cause condensation of water droplets on the surface of the egg shell, which encourages bacterial contamination.
- Disinfection of eggs: If contraindicated products or high doses such as quaternary ammonium at doses greater than 1000 ppm are used, early embryonic mortality increases. When solutions are used on the surface of the egg, the temperature of the surface must be taken into account, in order not to cause sudden changes in the physical condition of the embryo.
- Too short storage time: early embryonic mortality occurs due to poor embryo position at the time of incubation. In the first moments after the egg is laid, the blastoderm is in the centre of the egg. During storage, the yolk turns towards the upper pole, leaving the blastoderm correctly positioned.
- Preheating: Precise instructions for preheating must be followed, since the correct physical conditions of the environment and uniformity must be provided to the entire egg mass.
- Incubator conditions: inadequate temperature, ventilation and turning affect embryos.
- Quality of the eggshell.
- Nutritional deficiency in the breeding ration.
- Mycotoxins.
- Breeding flock diseases: e.g. Newcastle, mycoplasmosis, diphtheria.

Intermediate embryonic mortality (Phase II).

Intermediate embryonic mortality, according to Tona et al. (2022) may be due to farm and hatchery factors:

- Very thin egg shell.
- Egg contamination.
- Poor nutrition or health status of the breeders.
- Deficiency of riboflavin, vitamin B12 and D3.
- Sudden changes in temperature or ventilation in the incubator.
- Inadequate or absent turning.
- Low temperature or high humidity in the incubator.
- Lack of oxygen or excess of carbon dioxide in the incubation room.

Late embryonic mortality (Phase III).

Sardá and Vidal (2012), van der Pol et al. (2005), Ehuwa et al. (2021), Cedeño (2022) highlight the following causes:

- High humidity or low temperature during the incubation period: the embryo is found dead in an egg with a very small air chamber, reddened tarsi (Fig. 4), bleeding beak, edematous subcutaneous tissue, a gelatinous transudate may be present in the area around the nape of the chick's neck. The yolk sac is very swollen. To complete the diagnosis of this situation, the loss of moisture in the eggs during the incubation period can be measured. To do this, the eggs are weighed when they are placed in the incubator and then again at the time of transfer. A loss of 12% of the weight of the eggs is a correct measurement.

This ensures gas exchange along with water. Excess CO2 in the embryo leads to its death by acidosis. Embryos that do not die in this period will not hatch either, as the reduced air chamber forces them to dive too high and many will die in this attempt.

- High temperature or low humidity during incubation (Cedeño et al., 2022): embryos are smaller than usual, there may be very dry and dehydrated chicks. The yolk sac is smaller than normal. Moisture loss during the first 18 days is greater than 12%. Tolerance to moisture loss is greater than to low loss.
- Embryos infected by different etiological agents.
- High humidity in the hatchery.
- Overcooled eggs.
- Deficiencies: Biotin deficiency causes embryonic mortality between 19 and 21 days of incubation. Vitamin D deficiency causes poor shell quality and leads to a large loss of moisture, as does a low calcium ration. Manganese deficiency causes short limbs and abnormal down in unhatched embryos found in this period. In Zn deficiency, birds lack a rump and some parts of the skeleton. Selenium overdose produces weak chicks.
- Very high temperature in the hatchery.

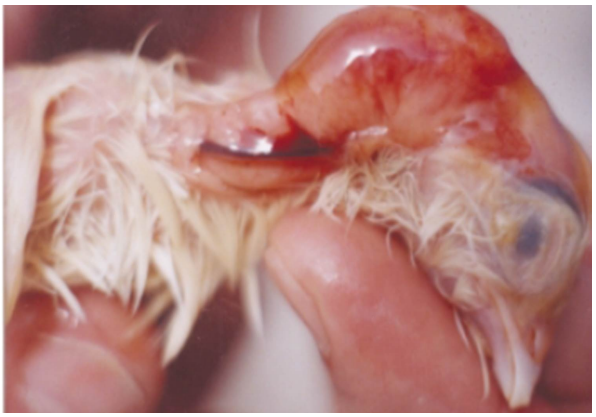

Fig. 4. PNN chick, with edema in the neck. This gelatinous transudate is found in the subcutaneous tissue due to the effort to hatch.

- Lack of ventilation.

PNN: They are chicks that are able to partially peck the eggshell and cannot hatch at birth. Among the possible causes, McIlwaine et al. (2021), Sandoval et al. (2005), Gast et al. (2020), Nolan et al. (2020) point out the following:

- Inadequate feeding of the breeders.
- Breeding disease, colibacillosis, salmonellosis, mycoplasmosis, encephalomyelitis.
- Lethal genes.
- Eggs placed upside down.
- Thin-shelled eggs.
- Trauma during transfer.
- Problems with turning during the first weeks of incubation.
- Low humidity during the twenty-first day of incubation.
- Poor air circulation or high CO2 concentration during the 20th and 21st days of incubation.
- Very late transfer.
- Fumigation with excess formalin during the chopping of eggs in the hatchery.
- High humidity or low temperature during the incubation period: see discussion of this topic in Phase III Mortality. Moisture loss during the incubation period is less than 12%, chicks peck far above the upper pole of the egg, bleeding from the strain is visible on the beak, and reddish tarsi (Fig. 5L).
- Temperature records, either high or low, for short periods.

Malformations: They are morphological defects of the embryos (Fig. 5A - 5 III), which affect 0.3% of the population and some chicks are born.

Causes of normal deviations from the incubation process: Main pathologies.

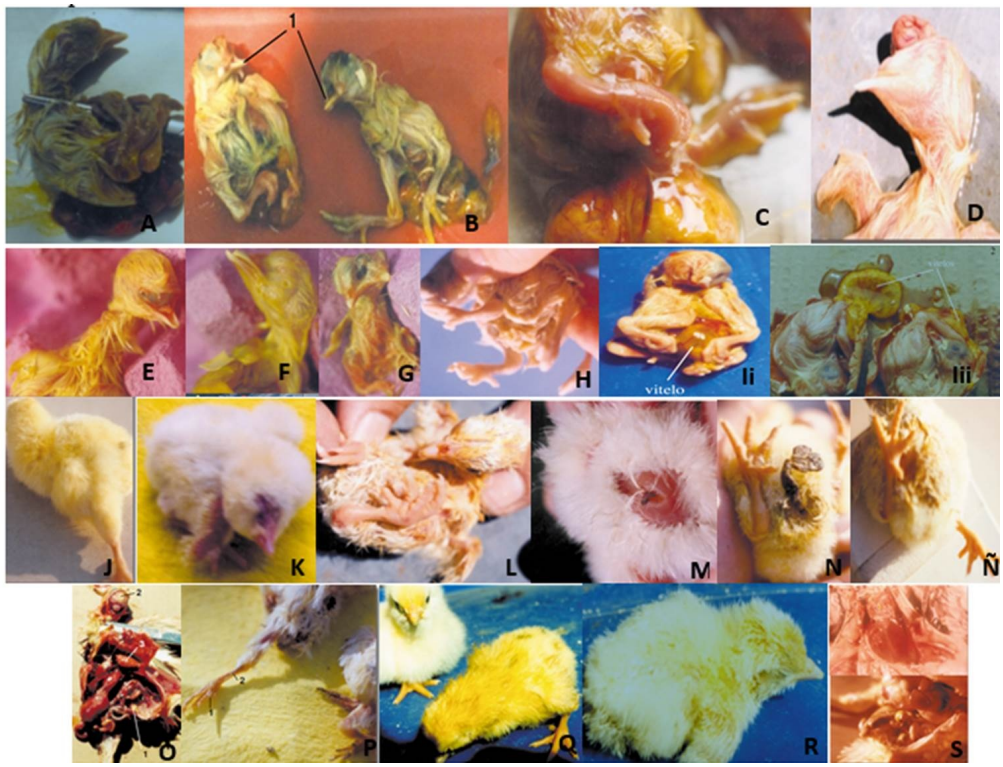

Fig. 5. Malformations

A. Three legs are observed in the same member of the embryo; it is called triplopodia. B. Duplication of the upper jaw (1). C. Malformation of the lower limb since the long bone of the tarsus has not developed, the rest of the embryo being of normal characteristics. D. Encephalocele (hernia of the brain) is the most frequent, with very diverse etiologies, commonly excess of temperature during incubation. E. Cyclops embryo, both eyes fuse into one in the middle line, a rare finding. F. Anophthalmia (total absence of eyes). G. Dicephalic embryo, the duplication is anterior or cranial. H. The duplication is posterior when the anterior parts of the body are normal, while the posterior or caudal part is double. Ii. United or fused embryos. The case shown in this photo is called cephalothoracopagus (1). Iii. When embryos come from the same egg or zygote they share the same yolk, unlike those that originate from a double-yolk egg with different zygotes and that do not have a common yolk (2). J. Malformations of the extremities due to trauma caused by the trays of the hatchers, conveyor belts, or chick boxes, with very smooth and slippery floors. K. Nervous signs at the time of birth of various etiologies. L. Chicks with the upper part of the beak bleeding due to efforts made during hatching related to little loss of moisture during the incubation period. M. Poorly healed navel. N. Black buttons on the navel, poorly healed, since parts of the yolk sac or extraembryonic tissues were not completely absorbed when the umbilical orifice closed. Ñ. Omphalitis. O. Granuloma due to Aspergillus fumigatus. Nodules in the serous membranes (1), in the brain mass (2), and in the lungs (3), caseous plugs can also be found in the bronchi. P. Dehydration and gout. The chick that suffers from dehydration usually presents a process of gout that can be articular (1) when the uric acid crystals are deposited in the joints or visceral, when it affects the internal organs. A characteristic sign of dehydration in chicks is the observation of the metatarsal vein (2). Q. Sticky chicks, this can be due to low temperature, high humidity, inadequate ventilation or egg explosion in the hatcher. A. Chicks with short, dry down and eyes stuck together; this can be due to high temperature, low humidity, excessive ventilation in the hatcher. S. Chicks with bacterial infection, the most frequent caused by E. coli, and Salmonella spp. The chicks generally present pericarditis (1). The yolk sac is found indurated or coagulated (2), this being an easier lesion to observe in chicks that are in their first week of life.

Causes of malformations (Plano and Matteo, 2001, Galíndez and Blanco, 2016):

- Hereditary factors.
- Environmental factors during incubation: Since embryonic tissues have different physiological zeros, when temperature changes occur, a certain defect will occur depending on the stage of development in which the embryo is found. Excessive heat produces encephalocele.

- Vitamin and mineral deficiencies and their effects:
  - Riboflavin: crooked fingers.
  - vitamin B2: chicks with a split upper beak.
  - Biotin: Parrot's Beak
  - vitamin B12: crooked fingers and short beak.
  - Vitamin D3: Abnormalities in the skeleton of the embryo
  - Manganese: abnormalities in the skeleton and beak of parrot.
- Storing eggs for more than seven days.

The classification referred to by Plano and Matteo (2001):

Embryos joined or fused: They are produced by incomplete division of the embryo into two parts during the period of development of the primitive streak (Fig. 1). They are generally conjoined monozygotic twins.

The suffix pago (meaning tied) is used after the term that indicates the anatomical region where the fusion occurs. For example, if they are joined at the thorax, they are called thoracopagus. If the fusion is at the abdomen, it is called abdominopagus. Those joined at the cephalic region, cephalopagus.

Duplications: Duplications of the embryo's anatomical structures occur. When the duplication affects anterior or cranial regions of the embryo, it is described as: anterior duplication. When the posterior region of the embryo is affected, it is called: posterior duplication. To name this type of malformation, the prefix di, tri, tetra is added to the anatomical region involved. For example, dicephalic, when the embryo has two heads. Posterior duplication is observed mostly in the limbs, (e.g. Triplopodia). The most frequent type of anterior duplication is the duplication of the beak in its upper valve.

Brain malformations: Encephalocele is the most common malformation observed in the chick embryo and consists of a herniation of brain tissue with or without meninges caused by excessive temperatures during incubation, affecting the formation of the cranial vault. It can be found in both embryos and hatched chicks.

Ocular malformations: 1. Cyclopia, which refers to embryos that have only one eye. There is only one eye socket situated in the midline, which may contain only one normal eye or two fused eyes. 2. Anophthalmia, which refers to the absence of the eyeballs caused by failures in the growth of the optic cup and may be accompanied by facial malformations (e.g. malformation of the upper beak valve).

Limb malformations: When it is due to the absence of part of the embryo's limbs, the defect is written as a prefix followed by the word melia, which means limb. If the limb is absent: amelia. If the limb is smaller than normal size, micromelia. If the fingers are shorter than normal, brachydactyly; if the fingers are fused, syndactyly.

3. Incubation pathology (perinatal)

Perinatal pathology is known as discarded or second-rate chicks that lead to a lack of productivity in a hatchery. It includes not only chicks that failed to hatch, but also those that are born but are not viable in the production system. The pathologies are listed below:

Chicks with congested navels (Sardá and Vidal, 2005): It is caused by temperature variations, most often due to very high temperatures in the stages prior to birth. It may be a case of omphalitis.

Chicks with poorly healed navels (Marvin, 2021): It occurs when the temperature between 11 and 18 days of incubation was too high or due to high humidity that does not allow the membranes to contract at the time of yolk absorption.

Chicks with black buttons on their navels (Sardá and Vidal, 2005): This is because part of the yolk sac and extra-embryonic tissues could not be properly absorbed when the umbilical opening was closed. It is the result of embryonic development that was faster than the expected maturity. It can occur due to high incubation temperature or a higher metabolic rate.

Chicks with their belly buttons open (Ross Tech, 2010): It is due to high temperature or variations in temperature. Also due to high humidity in the hatchery.

Chicks with omphalitis: It is an infection of the navel that presents signs of inflammation (Shahjada et al., 2017): It is due to the confluence of two factors:

- Due to incubation problems, both temperature and humidity in the incubator that lead to poor healing of the navel (Hussain et al., 2017, Gualpa, 2022, Ameen et al., 2017)
- Due to the presence of pathogenic bacteria that cause infection in the place, this is controlled by reducing egg contamination and with hygiene practices in the hatchery (Spackman and Stephens, 2016).

Sticky Chicks (Sardá and Vidal, 2005): It is mainly due to a low average temperature, high humidity, inadequate ventilation or egg explosion in the hatcher (due to egg contamination).

Dried chicks, with eggshells attached: It is due to low humidity in egg storage. Inadequate turning during

incubation. Low humidity in the hatchery (Plano and Matteo, 2001).

Dead chicks in the hatcher trays, dehydrated and smaller than normal: Its cause is the loss of humidity beyond what is expected (weight loss greater than 12% in the incubation period) in the first 18 days of incubation (Plano and Matteo, 2001).

Chicks panting and fresh feces on the hatcher trays: Prolonged permanence of chicks in the hatchers (Plano and Matteo, 2001).

Chicks born with defects: The causes of malformations are very varied and have already been discussed in the previous chapter; here only the defects of the hatched chicks are discussed (Plano and Matteo, 2001).

Both turning and inadequate ventilation play an important role in these pathologies (Intriago et al., 2023).

The temperature in the incubation process must be taken into account, since beyond the physiological zero of the different tissues there is a sequence in relation to the moment in which the thermal failure of the incubator occurred with respect to the appearance of malformations (Plano and Matteo, 2001).

Sprawled out chicks: It is mainly due to the slippery floor of the hatcher trays and can be caused by low humidity or nutritional deficiencies.

Chicks with nervous signs: It has varied etiology and can be caused by genetic, nutritional and environmental causes (high temperature or low humidity during incubation). If the percentage of chicks with nervous signs is very high, it can be corrected by increasing the humidity during incubation, but it is always a good idea to check the weight loss of the eggs during this period, as explained in section 5. Certain diseases can cause nervous signs in chicks (e.g. cerebral aspergillosis and encephalomyelitis, common in the first week of the chick's life on the fattening farm).

Chicks with cerebral hernia (encephalocele): There may be different causes, but it is mostly related to high temperature during the incubation period and in some cases to incorrect turning.

Chicks that can't stand up: Due to inadequate ventilation, overheating at some point during the 21 days of incubation and high humidity during the first 19 days.

Dehydrated chicks: Eggs loaded too early. Low humidity between 20 and 21 days of incubation. Chicks that have remained in the hatcher for too long. A characteristic sign of a dehydrated chick is dry skin on the leg and visible veins. Also gout or uric acid syndrome, which is rather a pathology of the chick's first week related to severe dehydration due to low humidity or high temperature in the hatcher, the chick room, due to excessive stay in the hatchers or in the plant. Uric acid precipitates in the form of urates both in the joints causing the so-called joint gout and in the internal organs (visceral gout).

Very small chicks: Light and small eggs. Low storage and incubation humidity. High incubation temperature.

Very large chicks with a swollen and soft abdomen (flabby): Low average temperature. Poor ventilation of the hatchery or incubator. High humidity, especially during the incubator period.

Weak chicks: High temperature in the hatchery. Poor ventilation. Excessive fumigation with formalin in the incubator. Poor nutritional or health status of the breeders.

Chicks with short, dry down and stuck-together eyes: High temperature, low humidity, excessive ventilation in the hatchery.

Chicks with bleeding top of beak and red tarsi: This is when the moisture loss of the egg during incubation has been low and the chick has to peck higher up (the air chamber is smaller than normal) and the lack of absorption of the yolk sac increases the effort required.

Premature births: Due to high temperature during egg storage. Incorrect preheating. High incubation temperature or low humidity at hatching.

Late births: Low temperature during incubation. Lack of preheating. Eggs stored for long periods at low temperatures. Eggs that are too large.

Eggshells stained with blood inside: The chicks hatched before their navels had healed due to the high temperature work of the hatchers.

Aspergillosis: It is a disease of the newborn chick caused by Aspergillus fumigatus. Contamination can come from the egg, from all the places with which it has come into contact and from the incubation plant. Aspergillosis can occur in different forms:

- Bronchial aspergillosis: characterized by gasping, cyanosis of mucous membranes, nails and beak. At necropsy, a yellowish-caseous plug is found blocking the lumen of the bronchi.
- Pulmonary aspergillosis: yellow nodules are found in the lung parenchyma or in the serosa. In the most severe cases of this disease, they can appear in the brain mass.

Chicks with pericarditis, yolk coagulation and liver congestion: This is a bacterial infection. A laboratory diagnosis is recommended to identify enterobacteria (Salmonella spp., E. coli) (Spackman and Stephens, 2016).

Conclusions

In this review, the importance of the causes of deviation from normal values of hatchability, embryonic development and incubation pathologies. The main factors associated with egg infertility and early, intermediate and late embryonic mortality were identified at farm, storage and

basic process levels of the hatchery regime. Malformations and incubation pathologies determined by biological control and embryodiagnosis were highlighted as key techniques in their assessment with the aim of taking corrective actions in the breeder flock and in the hatchery.

References

Abril, G. H. (2020). Determinación de pérdidas de incubabilidad mediante ovoscopía y embriodiagnosis. Universidad de Cundinamarca. Facultad de Ciencias Agropecuarias. Programa de Zootecnia Fusagasugá. 1-20.

Ameen, A. M., Mustafa, S. O., Hassan, D. R. & Rajab, D. A. (2017). The Effect of Aspergillus Fungi, Other Fungus, and Isolated Salmonella and *E. coli* Bacteria on Poultry Farms and Poultry Hatcheries at Veterinary Laboratory in Veterinary Directorate in Duhok, Kurdistan Region of Iraq. *EC Microbiology* 5.2, 52-58.

Barbosa, V. M., Rocha, J.S.R., Pompeu M. A., Martins, N. R. S., Baiâ, N. C., Lara, L. J. C., Batista, J.V. M. S.P. & Leite, R.C. (2012). The effects of relative humidity and turning in incubators machines on the incubation yield and chick performance. *World's Poultry Science Journal,* 69, 89-98. ISSN: 0043-9339 DOI: https://doi.org/10.1017/S0043933913000081.

Cedeño, O. (2022). Efecto del manejo en edad de reproductoras y temperatura de incubación sobre ventana de nacimiento y calidad del pollito BB. *Repositorio Digital ESPAM,* 1 (1), 68.

Chevalier, S., Bouffartigues, E., Bodilis, J., Maillot, O., Lesouhaitier, O., Feuilloley, M.G. J., et al. (2017). Structure, function and regulation of Pseudomonas aeruginosa porins. *FEMS Microbiol. Rev*., 41, 698-722. doi:10.1093/femsre/fux020

Ehuwa, O., Jaiswal, A. K. & Jaiswal, S. (2021). Salmonella, food safety and food handling practices. *Foods,* 10, 907.

FAO. (2019). Función de las aves de corral en la nutrición humana. *Revisión de desarrollo avícola*, 2 (1), 136.

Gairal M., N. (2019). Fisiología de la puesta de la gallina. Veterinaria Digital. 3(1): Disponible en: https://www.veterinariadigital.com/articulos/fisiologia-de-la-puesta-de-la-gallina/ [Consultado 20.10.2024].

Galíndez, R y Blanco, F. (2016). Eclosión, muerte embrionaria y calidad de pollitos en cuatro razas de gallinas reproductoras venezolanas. *Revista Científica*, 27 (1), 56-61.

Gast, R. K. & Porter, R. E. Jr. (2020). Salmonella infections. In: Swayne DE, Boulianne M, Logue SM, et al, eds. *Diseases of Poultry*. 14th ed. Wiley; 719-753.

Gualpa, Á. (2022). Parámetros básicos de incubación para asegurar la eclosión en los huevos de gallina. *Repositorio Universidad Técnica de Babahoyo*, 1 (1): Disponible en: http://dspace.utb.edu.ec/handle/49000/13262. [Consultado 24.10.2024].

Hashemi, S., Ghalyanchilangeroudi, A., Hosseini, S. M. & Sheikhi, N. (2020). Comparative trachea transcriptome analysis between avian infectious bronchitis virus and avian pathogenic *E. coli* individual infection and co-infection in SPF chickens. *Acta Virol.,* 64, 457-469.

Hussain, A., Bilal, M., Habib, F., Gola, B.A., Muhammad, P., Kaker, A., Yousaf, A. & Khalil, R. (2019). Effects of low temperature upon hatchability and chick quality of ross-308 broiler breeder eggs during transportation. *Online Journal of Animal Feed Research*, 9 (2), 59-67.

Intriago, M. V. A., Cedeño, R. O. A., Hurtado, E. A. & Rivera, F. R. D. (2023). Manejo en edad de reproductoras y temperatura de incubación sobre ventana de nacimiento y calidad del pollito BB. *Ciencia UNEMI*, 16 (43), 24-34.

Jessen, C. T., Foldager, L. & Riber, A. B. (2021). Effects of hatching on-farm on behaviour, first week performance, fear level and ran geuse of organic broilers. *Appl. Anim. Behav. Sci.*, 238:105319

Juárez EMA. Embriodiagnóstico. Evaluación casuística del fracaso en el desarrollo embrionario durante el proceso de incubación. Parte I. Los Avicultores y su entorno Nº 84, BM Editores. Departamento de Producción Animal: Aves FMVZ-UNAM. [http://www.produccion-animal.com.ar] 2014. [Consultado 21 septiembre de 2024].

Kuurman, W. W., Bailey, B.A., Koops, W. J. and Grossman, M. (2003). A model for failure of a chicken embryo to survive incubation. *Poultry Science*, 82 (2), 214-222.

Llamuca, L., Llamuca, M. & Intriago, V. (2024). Efecto de la edad de gallinas reproductoras en la incubabilidad y calidad del pollito bb: Una revisión. *Revista Científica Arbitrada en Investigaciones de la Salud. "GESTAR",* 7 (14), 184-204.

Macari, M., González, E., Sales, Patricio I, Alencas Nääs I, Martins P. (2015). Manejo de la incubación. Capitulo: Embriodiagnóstico y patología perinatal. 3ed. FACTA (Brasil). p 265.

Marvin, A. (2021). Absorción de saco vitelino. Avinews. Disponible en: https://avinews.com/absorcion-de-saco-vitelino/. [Consultado 18 septiembre de 2024].

McIlwaine, K., Law, C. J., Lemon, K., Grant, I. R. & Smyth, V. J. (2021). A Review of the Emerging White Chick Hatchery Disease. *Viruses,* 13 (12), 2435.

Nideou, D., N`nanle, O., Kouame, Y. A. E., Chrysostom, C., Gbeassor, M., Decuypere, E. *et al*. (2019). Effect of High Temperature during First and Second Halves of Incubation on Layer Chicken Embryo Physiology. Int. J. *Poult. Sci.,* 18 (12), 626-633. doi: 10.3923/ijps.2019.626.633

Noiva, R., Menezes, A. & Peleteiro, M. (2014). Influence of temperature and humidity manipulation on chicken embryonic development. *BMC Vet Res*., 10 (234), 1-10. doi: 10.1186 / s12917-014-0234-3

Nolan, L. K., Villancourt, J. & Barbieri, N.L. (2020). Colibacillosis. In: Swayne DE, Boulianne M, Logue SM, eds. *Diseases of Poultry*. 14th ed. Wiley. 770-830.

Reijirink, I. A. M., Meijerhof, R., Kemp, B., Van Den Brand, H., Arora, K. L., Kosin, I. L. *et al.* (2008). The chicken embryo and its micro environment during egg storage and early incubation. *Worlds Poult Sci. J.*, 64 (04), 581-598.

Rezaee, M. S., Liebhart, D., Hess, C., Hess, M. & Paudel, S. (2021). Bacterial Infection in Chicken Embryos and Consequences of Yolk Sac Constitution for Embryo Survival. *Infectious Disease*, 58 (1), 71-79.

Ricaurte, G. S. L. (2005). Embriodiagnosis y ovoscopia. Análisis y control de calidad de los huevos incubables. *Revista Electrónica de Veterinaria. REDVET*, VI (3): Disponible en: http://www.veterinaria.org/revistas/redvet [Consultado 24.10.2024].

Moya R., J. y Bermúdez C., A. I. (2017). Factores que afectan la incubabilidad de huevo fértil en aves de corral. *Nutrición Animal Tropical*, 11 (1), 16-37.

Ross Tech**. (**2010). Investigación de las prácticas de incubación. Aviagen. 1-39.

Sandoval, A., Yuño, M., Bakker, M. L., Rodríguez, E. & Beretta A. (2005). Aplicación de la embriodiagnosis para evaluar la eficiencia de la planta de incubación de parrilleros en una empresa avícola comercial en la Argentina. *RIA*, 34 (2), 75-89.

Sardá J., R. & Vidal P., A. (2012). Patología de la incubación. Instituto de Investigaciones Avícolas, La Habana, Cuba. Disponible en: https://www.produccion-animal.com.ar/produccion_aves/produccion_avicola/22-patologia_de_la_incubacion.pdf [Consultado 18 septiembre de 2024].

Sardá, R. y Vidal A. (2005). Pérdidas durante el proceso de incubación de los huevos. Portal Veterinaria. Disponible en: https://www.portalveterinaria.com/avicultura/articulos/2806/patologia-de-la-incubacion.html [Consultado 18 septiembre de 2024].

Shahjada, Z., Khalid, H., Monowarul, I., Majumder, S., Hasan, I., Rahman, M. et al. (2017). Bacteria causing omphalitis in newly hatched chicks from broiler and layer flocks and their antibiotic profiles. *Int J Nat Soc Sci.*, 4 (2), 73-81.

Spackman, E. & Stephens, C. (2016). Virus isolation and propagation in embryonating eggs. In: Eilliams SM, ed. A Laboratory Manual for the Isolation, Identification and Characterization of Avian Pathogens. 6th ed. American Association of Avian Pathologist. 361-367.

Tona, K., Voemesse, K., N'nanlé, O., Oke, O. E., Kouame, Y. A. E., Bilalissi, A. *et al.* (2022). Chicken Incubation Conditions: Role in Embryo Development, Physiology and Adaptation to the Post-Hatch Environment. *Front. Physiol.*, 13, 895854.

Tona, K., Onagbesan O., Bruggeman, V., De Smit, L., Figueiredo, D. & Decuypere, E. (2007). E.Non-ventilation during early incubation in combination with dexamethasone administration during late incubation: 1. Effects on physiological hormone levels, incubation duration and hatching events. *Domestic Animal Endocrinology*, 33 (1), 32-46.

van der Pol C. W., van Roovert-Reijirink, I. A. M., Maatjens, C. M., van den Brand, H. & Molenaar R. (2013). Effect of relative humidity during incubation at a set eggshell temperatura and brooding temperature posthatch on embryonic mortality and chick quality. *Poultry Science*, 92, 2145-2155.

Wijnen, H. J., van der Pol, C. W., van Roovert-Reijirink, I. A. M., De Smet, J., Lammers, A., Kemp, B. *et al.* (2021). Low incubation temperature during late incubation and early feeding affect broiler resilience to necrotic enteritis in later life. *Front. Vet. Sci.*, 8, 1-16.

Yerpes R., M. (2020). Factores de riesgo asociados con la mortalidad de los pollos de engorde durante la primera semana de vida. Repositorio Universitat Autónoma de Barcelona. 1 (1), 137.