Spanish Journal of Agricultural Research
21 (2), e05R01, 10 pages (2023)
eISSN: 2171-9292
https://doi.org/10.5424/sjar/2023212-19605
INIA-CSIC

REVIEW ARTICLE

OPEN ACCESS

Animal welfare of embryos and newly hatched chicks: A review

Itallo C. S. ARAUJO, Tainá S. B. LOPES, Leonardo J. C. LARA and Bruno T. A. COSTA
Universidade Federal de Minas Gerais, Veterinary School, Dept. of Animal Science, Belo Horizonte, MG, Brazil.

*Correspondence should be addressed to Italo C. S. Araujo: italloconradovet@hotmail.com

Abstract

The welfare of farm animals is a hotly debated issue today. There are few technical materials available focused on hatcheries that address and incorporate practices adopting the welfare level required by civil society. However, about 33% of the production period of a broiler chicken occurs on the hatchery. Recent studies have shown that day-old chick production has points that need to be improved. Technical standards indicate that the process of welfare should begin during egg storage such that the best conditions are provided for embryos prior to incubation. Storage until 28 days at temperatures above 12ºC can kill embryos, while exposure of eggs to 30 ºC until 14 days causes a negative impact under the hatchability. The available results regarding in ovo nutrition show that it should be considered for benefit neonate quality since inoculation of substances such vitamin E (60.4 IU) promote better incubation results. Feeding immediately after hatching is a management for welfare because animals feed-fed soon after hatch perform better up to 35 d. The most criticized point in hatcheries is the slaughter of neonates, for which some strategies are being investigated, including the use of dual-purpose aptitude strains, in ovo sexing by different methodologies, sex inversion and hormonal-based sexing. It is noted that procedures involving bird incubation require greater attention to welfare. Therefore, it is necessary to align theoretical knowledge with practical applications so that the demands of society, as well as those of industry, are met.

Additional key words: chick welfare; hatchability; hatchery; broiler embryos; in ovo injection.

Abbreviation used: FAWC (Farm Animal Welfare Council); IC (iodinated casein); RSPCA (Society for the Prevention of Cruelty to Animals)

Citation: Araujo, ICS; Lopes, TSB; Lara, LJC; Costa, BTA (2023). Animal welfare of embryos and newly hatched chicks: A review. Spanish Journal of Agricultural Research, Volume 21, Issue 2, e05R01. https://doi.org/10.5424/sjar/2023212-19605

Received: 26 May 2022. **Accepted:** 28 Apr 2023.

Funding: The authors received no specific funding for this work.

Competing interests: The authors have declared that no competing interests exist.

Introduction

High demand for animal foods has led consumers to increasingly discuss whether the practices adopted during production are compatible with animal welfare. In the poultry chain, welfare discussions center around questions about the husbandry system, where management practices, such as beak-trimming and forced molting, and the transportation and slaughter of birds, are being questioned. Discussions that were initially restricted to animal rights groups have now reached consumers. Requirements stemming from these discussions include not only nutritional and health quality, but also the opportunity to choose animal products obtained, raised and slaughtered within the criteria of animal welfare.

As a result of consumer pressure, the conventional cage-laying system has been banned in the European Union since 2012 by Directive 1999/74/EC (EC, 1999) to improve the welfare of layers, although several countries still employ the practice (Jansson, 2018).

For some time, issues regarding the welfare of commercial birds have turned to environment, management, and rearing systems. Nonetheless, the production of laying chicks is one of the questioning points regarding animal welfare in the poultry chain, mainly due to the disposal of unwanted chicks in the egg chain (Gremmen et al., 2018).

As a result, and due to the organization of the most demanding consumers, further developed countries have started to establish new production practices for hens, layers, and broiler breeders, including hatchery procedures.

Thus, the objective here is to review technical information, exemplified in two relevant manuals, and to present research involving bird embryos and exploring the management practices and techniques used in hatcheries that impact the welfare of commercial neonate birds. First, we present some key concepts used to determine hatching environment welfare.

Concepts of animal welfare applied to incubation of hatching eggs

As already mentioned, the issue of welfare in livestock production is becoming more significant every day. To properly debate and analyze this subject, it is necessary to establish a well-defined concept of welfare and methods of evaluation of welfare that apply to the routine production chain.

In 1965, a technical inquiry committee into animal welfare called the Brambell Report recommended that animals be free to stand up, lie down, turn around, groom, and stretch their limbs. Based on this report, the Farm Animal Welfare Council (FAWC) developed the so-called Five Freedoms (Webster, 2001; Underwood et al., 2021). This framework defines key aspects of animal welfare and is the basis of many of currently existing animal welfare assessment methodologies. They are: freedom from hunger and thirst; freedom from discomfort; freedom from pain, injury or disease; freedom to express normal behavior; and freedom from fear and distress (Webster, 2001).

Years later, Broom (1986) presented a broader concept of animal welfare, considering that it would be the state of an individual in relation to attempts to adapt to the environment in which it finds itself. Thus, an individual can adapt to an environment without much effort and, therefore, without excessive resource expenditure, as well as there may be major effort to adapt that requires high resource expenditure and/or even a lack of adaptability, resulting in poor welfare.

Concepts of welfare have changed over the years. Thus, Mellor & Reid (1994), considering that "welfare is the behavioral manifestation of an animal when its nutritional, environmental, health, behavioral and mental needs are met", presented the "Five Domains" model of animal welfare. This framework was based on the five freedoms and aimed to be a systematic method for assessing animal welfare, with the tool itself not being considered a definition of welfare. Unlike the model proposed by FAWC (2019), the five domains that were considered with potential for welfare compromise are thirst/hunger and malnutrition; environmental challenge; disease/injury/functional impairment; behavioral/interactive restriction; and anxiety/fear/pain/distress.

Two manuals were identified, at least, for the poultry chain that meet the principles of five freedoms welfare. There are other manuals in the literature on welfare in the artificial incubation process; however, the two documents presented in our review present practical actions that can be implemented in the hatchery routine. In Canada, the code of practice for the care and management of hatching eggs, chickens, and turkeys was launched in 2016 by the National Farm Animal Care Council (NFACC, 2016). In the same vein, the Society for the Prevention of Cruelty to Animals (RSPCA) of the United Kingdom released a document setting standards for practices performed during routines of hatcheries (RSPCA, 2017). These documents set standards for ambiance (equipment, lighting, aeration, density, nutrition, and food), animal category (laying, breeding, broiler, etc.), and zootechnical and physiological responses of animals following the principles of the five freedoms to reach animal's welfare.

The hatchery guidelines published by RSPCA (2017) consider seven items that should be audited in hatcheries, thus establishing standard procedures to be followed in the following areas: contingencies and maintenance; building *design* as well as its facilities and equipment; competence and training for chick handling; egg management; hatchling chick management (vaccinations, sexing and selection); the humanitarian slaughter of live hatchery discards; and transport of chicks to the farm.

The other documents cited (Webster, 2001; Underwood et al., 2021) emphasize the need to control the manipulation of hatchling chicks (vaccination, sexing and transport) and euthanasia of live embryos, and refuse chicks, without other main highlights. However, what can be seen in all the technical documents is greater control of the so-called inputs and very little about the outcomes, with the exception of that published by the RSCPA (2017). RSCPA (2017) mentions behavior, neonatal chick injuries and mortality rates for chicks housed for up to seven days as factors that should be taken into account when assessing hatchery welfare. Thus, not all items cited by the RSCPA (2017) are strictly followed by hatcheries. It can also be concluded, from objects of concern with currently adopted welfare methods, that norms related to hatcheries are much less impeding than those of other poultry production sectors.

In general, the hatchery segment is very well structured to meet the requirements of chick welfare, with the main objective being the highest possible productivity without causing unnecessary suffering to produced chicks (Araújo et al., 2016; Mesquita et al., 2021; Underwood et al., 2021). Thus, if attending to the welfare of an animal can increase productivity, there is no reason to not employ methods that make it possible to meet the demands of embryo welfare. Next some potential areas for improvement are presented.

Egg storage before incubation

The length of the storage period can vary from one day to more than one week and depends on the distance between the farm and the hatchery, the incubation capacity, and the market conditions. Therefore, logistics for storage and dispatch can negatively alter incubation viability and chick quality because changes in embryo and egg components may occur during this period (Meijerhof, 1992). It is noteworthy that the strategy of egg storage before incubation is essential for breeders and hatchers as it directly reflects the logistics required within the supply chain as there may be variation in egg supply by brooders and the need for day-old chicks by farms (Fasenko et al., 2001).

In assessing the impact of storage time and temperature on fertility and hatchability of slow-growing lineage eggs, Addo et al. (2018) observed that the blastoderm and embryo were larger in eggs stored at 30ºC, with a storage period between 10 and 14 days than those stored at 18ºC for the same period. Fertile eggs and hatchability were lower in eggs stored at room temperature when compared to cooled eggs.

From this perspective, Pokhrel et al. (2018) assessed the effects of egg storage at 18ºC and 12ºC for 7 to 28 days on hatchability, chick quality and cell morphology, and found hatchability to remain higher for eggs stored for more than seven days at 12ºC compared to storage at 18ºC, demonstrating an effect mainly in the first three days of storage. In other words, eggs stored at the highest temperature had major morphological changes and death cells, since embryos stored at 18ºC continued to develop gradually until the end of the blastulation stage while those stored at 12ºC stopped the development at seven days.

These results certify that, when not properly used, both storage period and storage temperature can affect hatchery production indices and should therefore be evaluated together so that longer storage periods are achieved with lower storage temperatures. However, prolonging the storage period too much, even at temperatures below physiological zero (according to Costa et al. (2022) 18-21ºC – when the embryo's development is paralyzed), can cause cellular changes leading to delayed or decreased resumption of development (Araújo et al., 2017). Still, it is important for industry to prioritize the implementation of standard transport and storage procedures that value the incubation of eggs below physiological cooling in order to keep incubation rates within desirable values for longer periods of time (Van Roovert-Reijrink et al., 2018). Thus, considering the welfare of the embryo at the beginning of the incubation process, specific measures related to the storage period and temperature aiming at improving embryo survival, and potentially improving embryo welfare, are necessary.

Physical conditions offered by the setter

For proper development of the embryo, the incubation environment must provide a favorable microclimate with temperature, humidity and ventilation being determining factors for maintaining embryonic metabolism, since they directly influence oxygen exchange (Macari et al., 2013). Recent studies have shown that changes in these factors can impact animal metabolism and behavior during and after hatching.

In this sense, Bertin et al. (2018) assessed the influence of repeated exposure to substandard incubation temperatures on metabolism and fear behavior and found no differences in hatchability between the control group and the treatment exposed to 27ºC/1h/day. The study, however, did find a delay in hatching, decreased growth rate, increased expression of corticotropin-releasing factor in the central nervous system and greater fear reaction immediately after hatching in the treatment group compared to the control group.

In a subsequent study, to understand the influence of incubation temperature on bone mineralization of neonate broilers, Muir & Groves (2019) tested slow incubation at 37.2ºC to 37.8ºC versus the control treatment at 37.8º to 38.2ºC. The slow treatment showed better results for hatchability since more chicks were born near the end of the birth window. The percentage of bone mineralization was measured shortly after birth and the highest percentage of bone mineralization was seen both in the late birth group and in the group that hatched near the beginning of the birth window and had access to feed in less than 24 h.

The differing incubation results presented are probably due to the different methodologies applied in each study to achieve their inherent objectives. Regardless of this, they demonstrate that the impact of temperature on embryonic development can vary in its effect according to different situations, such as embryonic phase, total stress period and intensity.

Given this, it is important to develop more studies to clarify the means by which temperature acts on these responses so that incubation can be modulated in the field to improve chick quality, and potentially welfare reflected by hormone profiles and improved health, in the later stages of raising.

Gases exchange during incubation

Eggs are a system that supports embryo development. The embryo development rate is directly dependent on the oxygen supply that is diffused through the pores of the shell (Araújo et al., 2017). According to Ar et al. (1974), conductance is the capacity to achieve gas exchanges (oxygen, CO_2, and metabolic water), which take place in eggs through the pores in their shells.

Due to the porous character of the eggshell, the gas composition in the incubation environment is important for the production of day-old chicks (Peebles & Brake, 1985). Thus, eggshell conductance is based on the fact that the quantity of energy available for embryo development depends on the quantity of oxygen that enters the egg through its shell, given that embryo metabolism is primarily aerobic (Araújo et al., 2017). Also, Taylor et al. (1956) suggested that CO_2 levels greater than 1.0% stimulate embryonic development in the first 48 h of incubation, and that embryos are resistant to higher CO_2 levels in the first week of incubation.

More recent studies indicate that hypoxia can advance the hatch peak by 10 to 15 h by an increase in CO_2 pressure followed by elevation in corticosterone levels, which together initiate the hatching process (De Smit et al., 2006). Bruggeman et al. (2007) suggest that in the first stage of embryonic development the initial defense barrier is the albumen, which enables the reaction between CO_2 and H_2O for the formation of protons and bicarbonate ions. These authors also suggest that this mechanism occurs at least until 96 h of incubation when the chorioallantoic complex is not yet fully formed.

After the formation of the chorioallantoic complex, embryos can increase the expression and activity of carbonic anhydrase in blood cells, transforming CO_2 in bicarbonate and subsequently reabsorbing these molecules through the chorioallantoic membrane, which is released along with calcium ions through the shell pores (Everaert et al., 2010). A study produced by Zhang & Burggren (2012) indicates that despite different adaptive strategies to the hypoxic environment when incubated with 15% of O_2 or lower levels, whether in the early stage or over long periods, embryonic development occurs more slowly, negatively impacting birth weight and mortality in the late phase.

The two hypoxia response mechanisms presented here are used by broiler and layer embryos (Everaert et al., 2011). Both strains adapt to respiratory stress by increasing oxygenation capacity, which can be visualized by an increase in the percentage of circulating hematocrit and hemoglobin (Burggren et al., 2015; Ben-Gigi et al., 2021). However, in the study conducted by Ben-Gigi et al. (2021), layer embryos showed elevated hematocrit levels only for 24 h after respiratory inducted stress, while broiler embryos maintained them for 72 h, indicating more difficulty to resume oxygenation demand, visualized by lower weight gain. The authors suggested that genetic selection for rapid growth elevated the metabolism demand of broiler embryos without balancing the cardio-vascular system, limiting their ability to respond to hypoxia.

Incubation environments with at least 15% O_2 (Zhang & Burggren, 2012) and at most 10% CO_2 (De Smit et al., 2006; Burggren et al., 2015) are those recommended to ensure acid-base control, organ development, and hatchability. Therefore, quite possibly there is greater embryonic welfare in environments with a similar gas profile.

Lighting during incubation

The influence of light on the incubation process has been studied since the mid-20th century, with current studies seeking to elucidate the effects of the light stimulus period, light spectrum, and light intensity during and after incubation (Cooper, 1972). Among the extrinsic factors necessary for embryonic development, light has a positive effect on embryo growth, and the results achieved with its use are important for industry (Shafey, 2004).

Comparing the effect of LED light on the hatching performance of eggs from white egg layers and heavy brown egg breeders, Huth & Archer (2015) found that the group of brown eggs incubated with 12:12 hours of light:dark had fewer defective chicks, higher hatchability and lower heterophil/lymphocyte rates when compared to the group incubated in the dark. These authors pointed out that differences in performance may have been due to the different eggshell colors exerting changes in the light spectrum during its passage through the eggshell. However, the results may have also occurred due to differences between the embryos of the different strains.

In a later study, Archer (2017) evaluated the use of white and red light in the first eighteen days of incubation and found better results for hatchability of fertile eggs and fear response behavior of neonate chicks in these groups compared to animals treated with blue, green and no light. Subsequently, Archer (2018) compared incubation without light to incubation with red and blue light and found no difference in early-stage mortality, which was higher in the no-light treatment and also affected hatchability and percentage of open-naval chicks, which were best in the light treatments regardless of color.

On the other hand, Tong et al. (2018) compared incubation with green light and no light, and found significant effects for embryonic growth and acceleration of birth, but no differences in weight and quality, while hatchability decreased in the green light treatment. In contrast, Yu et al. (2018) contrasted the effects of incubation with green light and no light on embryonic growth, incubation yield and hormone levels and found positive results for low intensity green light (50 lux). This group obtained greater hatchability, size, and weight at birth, as well as production.

Even contrasting results make clear the influence of luminosity on embryogenesis. However, the effects of light spectrum intensity, light period and the influence of the shell as a filter need to be evaluated together for a better understanding of the mechanisms of light and their interaction with the egg. Elucidation of these mechanisms is paramount since the light factor affects not only chick quality but other aspects such as cell differentiation and hormone production, as well as acting on chick behavior and quality parameters in the post-hatching period.

In ovo feeding

The need to induce the immune system activation as early as possible to facilitate sanitary management spurred the development of *in ovo* injections in broiler hatcheries approximately 30 years ago (Sharma & Burmester, 1982). This practice initially occurred manually, but automated technologies that can vaccinate a large volume of eggs per hour with less labor and a lower error rate are now available (Uni & Ferket, 2003). *In ovo* management, whether for the application of vaccines, stimulants, or nutrients, occurs during the transfer of eggs from setters to hatcheries around the eighth day of incubation.

According to Cardeal et al. (2015) and Givisiez et al. (2020), *in ovo* nutrition is a strategy that aims at supplementing nutrient deficiency during hatching. It can also stimulate epithelial and enzymatic development of the digestive tract, mainly for carbohydrates, which are highly required at birth. It can also provide amino acids, vitamins, minerals, and probiotics.

As specified by Tona et al. (2003) some performance parameters, such as weight at seven days, may be affected by incubation factors, which indicates that early-stage performance may be proposed as a welfare indicator in hatcheries. In this sense, the *in ovo* strategy may be of interest since it has a direct action on weight gain. This zootechnical index can be considered an indicator of animal health, and besides that, it can be easily controlled in hatcheries. The substances administration occurs in the region of the amnion, commonly during the final period of embryonic development (Uni & Ferket, 2003) when amniotic fluid is ingested by the embryo and absorbed by enteric cells (Araújo et al., 2018).

In researching the *in ovo* supply of perinatal dextrin-iodinated casein (IC) on birth weight, Abousaad et al. (2017) found that the use of 240 and 720 µg IC/mL of casein favored the birth of heavier chicks compared to the other treatment groups (saline solution and 80 and 2,160 µg IC/mL). Groff-Urayama et al. (2019) also evaluated incubation parameters of chicken embryos, although subjected to amino acid inoculation by two different techniques (needle angle of 45° or 90° relative to the air tube, without exceeding it) during the final incubation phase. These authors obtained a different result than did Uni & Ferket (2003) since the embryonic mortality percentage was higher in the group with inoculation surpassing the air tube.

To determine the effect of *in ovo* inoculation of vitamin E (0.0, 27.5, 38.5, 49.5, and 60.4 IU) on incubation results and quality of newly-hatched chicks, Araújo et al. (2018) provided different levels of vitamin E supplementation on day 17.5 of incubation using a manual needle. The 60.4 IU level produced the highest hatching rate, and all supplemented groups showed better physical quality compared to the control group.

The research presented in this review has shown that there may be more than one technical standard technique for *in ovo* inoculation that is safe, and that nutrient supply can positively affect incubation performance as well as the health of neonates, likely improving chick welfare at hatch. Therefore, the development of research in the area of *in ovo* nutrition is important for determining technical guidelines aimed at improved welfare in hatcheries, that are adapted to hatchery production processes.

Handling of chicks

One-day chick quality is paramount to the profitability of the poultry chain (Araújo et al., 2018). On the other hand, management of neonates has gaps in its stages that can provide important parameters for welfare in hatcheries that are not detailed in existing welfare manuals. Therefore, it is essential to develop research around chick handling to provide operational protocols consistent with productivity and animal welfare.

When assessing the effects of early feeding on broiler performance, Abousekken et al. (2017) concluded that animals fed shortly after hatch experience greater weight gain, body weight, and better feed conversion at 35 days of life than animals receiving alternative and control feed without early feeding. Similarly, Hollemans et al. (2018) analyzed the effects of feed supply shortly after or 54 h after hatch, management, and transport on performance and behavior under stress up to 35 days. They observed that the effect of post-hatching nutrition was evident only in the first phase of rearing (21 days) while the fear response, simulated by transportation, extended to the final phase of rearing (35 days) when it was higher in late-fed chicks.

Soon after the pulling, the chicks go through steps that require intense manipulation. Given this, Hamissou Maman et al. (2018) evaluated the effect of chick body temperature at these stages on residual weight of yolk and weight of body and organs (heart, gizzard, bursa of Fabricius) as well as on incubation yield. Yolk residual weight was not affected by handling temperature, but organ weight and body weight at housing were lower in chicks that had higher body temperature during handling. The body weight showed influence over long periods, since the group with the lowest weight at housing also had the lowest weight at 35 days. It was already shown that the chick handling may affect the corticosterone levels since chicks that pass through intense handling experience significant increase in corticosterone levels that can extend until the end of the first week after hatching (Hedlund et al., 2019).

The results of this research show that newly hatched chickens are sensitive to access to water, food, variation in temperature and handling during hatchery management. Therefore, these aspects must be considered during the development of standard procedures for hatcheries aiming at improving welfare, in addition to those already established, such as density and maximum transport distance.

The most criticized point about practices that occur in hatcheries is the newly hatched chicken euthanasia. Although the regulation recommends euthanasia of animals using methods that result in the rapid loss of sensitivity and death, this management raises questions for animal welfare groups, especially regarding the disposal of males of laying lines. Thus, the industry has been working to meet this demand by seeking improvement through scientific research.

One alternative is the use of double purpose strains. For this purpose, Mueller et al. (2018) compared performance, carcass conformation, and meat quality between double suitability and commercial chicken strains. The study revealed that the performance and carcass conformation of double suitability lines are limited compared to fast-growing lines, and thus represent an unviable alternative to the conventional chicken meat production system from an economic perspective.

Another plausible strategy is to use noninvasive methods such as *in ovo* sexing. Galli et al. (2016) proposed a fluorescent spectroscopy technique using embryos on the third and fourth day of incubation. In this method, a 15-mm-long opening was made in the shell, keeping the shell membrane intact, and vessels bigger than 100 µm were measured using a Raman spectrometer. Immediately after the measurement, the shell was re-attached to the original area using a biologically compatible adhesive tape. The sex of the embryo was determined by the blood fluorescence intensity, which in this stage was higher in male than female embryos.

Using molecular methods, He et al. (2019) developed two procedures for sexing embryos based on the techniques of PCR and qPCR. About 10–20 mg of embryonic tissue from the chorion, allantois, amnion, or brain were collected on the ninth day of incubation to identify genetic differences between the sex chromosomes of males (ZZ) and females (ZW) chickens by molecular markers. There remain other techniques for sexing *in ovo*, such as odor detection as suggested by Webster et al. (2015), sex inversion as proposed by Trukhina et al. (2016), and hormone dosage as studied by Wang et al. (2019).

It seems necessary to overcome some additional obstacles, such as sexing in the earliest incubation stage or sex induction to increase the birth number of females. The sexing techniques developed so far also need to eliminate the need for tissue removal or egg-opening to perform sexing, minimize impacts on incubation yield, and especially, adapt techniques for large-scale application.

Sanitization of setters

The use of industrial incubators has enabled the expansion of day-old chick production, and since then, the accumulation of dust and fluff due to a large number of eggs and birds housed in a small space has been reported as one of the main factors that lead to the introduction and dissemination of microorganisms such as *Salmonella* spp. (Bains & Mackenzie, 1974; Cox et al., 1990), *Escherichia coli*, *Pseudomonas* sp., *Staphylococcus* spp. (Soucy et al., 1983), *Clostridium* sp. (Craven et al., 2001), and *Aspergillus* spp. (Wright et al., 1960) in hatcheries. The consequences can be an increase in mortality rate (Rezaee et al., 2020), increased number of sick day-old chicks (Muhammad et al., 2010), mortality in the first week, and contaminated carcasses at slaughter (Bhatia & McNabb, 1980; Christensen et al., 2021).

Disinfection of equipment and facilities with compounds such as formaldehyde, during transfer and or after chicks removal, is one of the methods often applied in hatcheries to avoid contamination (Graham et al., 2021). Despite its effective action when in organic matter, it has been reported that on contact with tracheal mucosa, formaldehyde can cause a shortening and loss of cilia, besides ciliostasis in 18-days embryos and day-old chicks (Hayretdağ & Kolankaya, 2008; Graham et al., 2018); in other words, it may increase the predisposition of embryos and neonates to respiratory injury. Therefore, other disinfectants such as polyhexamethylenebiguanide hydrochloride, hydrogen peroxide (Cox et al., 1999), and electrowettered water (Fasenko et al., 2009) have been tested as an alternative to formaldehyde.

Recently, Melo et al. (2019) evaluated ozone (5-15 ppm), hydrogen peroxide (3%), peracetic acid (0.3%), and ultraviolet radiation (8.09 mW/cm^2) as alternative disinfectants to paraformaldehyde (5.03 g/m^3) and noted that peracetic acid and ultraviolet radiation are as efficient as formaldehyde to reduce aerobic microorganisms and enterobacteria, without causing changes in hatchability and day-old chick quality. According to Zeweil et al. (2015) and Oliveira et al. (2020) paraformaldehyde affects the embryos development and is harmful to the health of the farm and hatchery professionals. In another study, Melo et al. (2020) evaluated the application of gaseous (0.9%) peracetic acid on microbial load, hatchability of fertile eggs, and day-old chick quality and found that the use of gaseous peracetic acid in environment disinfection systems, being their use therefore feasible in hatcheries to reduce microbial load and ensure hatchability and day-old chick quality.

The works presented here show that the presence of potentially pathogenic microorganisms in hatcheries is still a concern to poultry health, as is the use of disinfection methods that are potentially detrimental to the health and welfare of the embryo and day-old chicks. The peracetic acid and ultraviolet radiation have shown to be as secure alternatives to paraformaldehyde, being efficient from both production, health, and animal welfare perspectives, but still, need to be widely implemented.

Conclusions

In countries where consumer pressure on poultry production practices is high, governmental, and non-governmental organizations have established animal welfare practices that should be followed in all stages of poultry production. Procedures posing more problems from an animal welfare perspective in hatcheries are the manipulation and euthanasia of embryos and live chicks. However, other incubation-related aspects such as animal handling, and care, equipment and techniques also require attention to achieve maximum hatchability and better health conditions for newly-hatched chickens.

A gradual evolution in the animal welfare definition as well as in the indicators used to measure it have historically occurred. Current assessment tools have paid little attention to the egg incubation process and are outdated when compared to the theoretical changes in animal welfare. That is why it is necessary to have specific assessment procedures for the day-old chick production process. Theory and practice should be aligned, so that the scientific and practical tools adopted in hatcheries could be complementary and work together to improve processes during incubation stages, ensuring that society's demands are met, as well as the success of the poultry chain.

Authors' contributions

Conceptualization: I. C. S. Araujo.
Data curation: not applicable.
Formal analysis: not applicable.
Funding acquisition: not applicable.
Investigation: I. C. S. Araujo, T. S. B. Lopes, B. T. A. Costa.
Methodology: T. S. B. Lopes, B. T. A. Costa.
Project administration: I. C. S. Araujo.
Resources: not applicable.
Software: not applicable.
Supervision: I. C. S. Araujo.
Validation: I. C. S. Araujo, L. J. C. Lara.
Visualization: I. C. S. Araujo, L. J. C. Lara.
Writing – original draft: I. C. S. Araujo, T. S. B. Lopes, B. T. A. Costa.
Writing – review & editing: I. C. S. Araujo, L. J. C. Lara.

References

Abousaad S, Lassiter N, Piekarski A, Chary P, Striplin K, Christensen K, Bottje WG, 2017. Effects of in ovo feeding of dextrin-iodinated casein in broilers: I. Hatch weights and early growth performance. Poult Sci 96(5): 1473-1477. https://doi.org/10.3382/ps/pew438

Abousekken MS, Shalash SM, El-Abd NM, Essa HG, 2017. The effects of early post-hatch nutrition on broiler performance. Egypt Poult Sci J 3: 749-762.

Addo A, Hamidu JA, Ansah AY, Adomako K, 2018. Impact of egg storage duration and temperature on egg quality, fertility, hatchability, and chick quality in naked neck chickens. Int J Poult Sci 17(4): 175-183. https://doi.org/10.3923/ijps.2018.175.183

Ar A, Paganelli CV, Reeves RB, Greene DG, Rahn H, 1974. The avian egg: water vapor conductance, shell thickness, and functional pore area. The Condor 76(2): 153-158. https://doi.org/10.2307/1366725

Araújo ICS, Leandro NSM, Mesquita MA, Café MB, Mello HHC, Gonzales E, 2016. Effect of incubator type and broiler breeder age on hatchability and chick quality. Brazil J Poult Sci 18: 17-25. https://doi.org/10.1590/1806-9061-2015-0146

Araújo ICS, Leandro NSM, Mesquita MA, Café MB, Mello HHC, Gonzales E, 2017. Water vapor conductance: a technique using eggshell fragments and relations with other parameters of eggshell. Rev Bras Zootecn 46(12): 896-902. https://doi.org/10.1590/s1806-92902017001200004

Araujo ICS, Café MB, Noleto RA, Martins JMS, Ulhoa CJ, Guareschi GC, et al., 2018. Effect of vitamin E in ovo feeding to broiler embryos on hatchability, chick quality, oxidative state, and performance. Poultr Sci 98(9): 3652-3661. https://doi.org/10.3382/ps/pey439

Archer GS, 2017. Exposing broiler eggs to green, red and white light during incubation. Animal 11(7): 1203-1209. https://doi.org/10.1017/S1751731117000143

Archer GS, 2018. Effect of two different commercially available white light LED fixtures on broiler hatchability and chick quality. Brit Poult Sci 59(3): 251-255. https://doi.org/10.1080/00071668.2018.1436160

Bains BS, Mackenzie MA, 1974. Transmission of Salmonella through an integrated poultry organisation. Poult Sci 53(3): 1114-1118. https://doi.org/10.3382/ps.0531114

Ben-Gigi R, Haron A, Shinder D, Ruzal M, Druyan S, 2021. Differential physiological response of slow-and fast-growing broiler lines to hypoxic conditions during chorioallantoic membrane development. Poult Sci 100(2): 1192-1204. https://doi.org/10.1016/j.psj.2020.10.068

Bertin A, Calandreau L, Meurisse M, Georgelin M, Palme R, Lumineau S, Cornilleau, F, 2018. Incubation temperature affects the expression of young precocial birds' fear-related behaviours and neuroendocrine correlates. Scient Report 8(1): 1857. https://doi.org/10.1038/s41598-018-20319-y

Bhatia TRS, McNabb GD, 1980. Dissemination of Salmonella in broiler-chicken operations. Avian Dis 24(3): 616-624. https://doi.org/10.2307/1589797

Broom DM, 1986. Indicators of poor welfare. Brit Vet J 142(6): 524-526. https://doi.org/10.1016/0007-1935(86)90109-0

Bruggeman V, Witters A, De Smit L, Debonne M, Everaert N, Kamers B, Decuypere E, 2007. Acid-base balance in chicken embryos (Gallus domesticus) incubated under high CO2 concentrations during the first 10 days of incubation. Respir Physiol Neurobiol 159(2): 147-154. https://doi.org/10.1016/j.resp.2007.04.013

Burggren WW, Mueller CA, Tazawa H, 2015. Hypercapnic thresholds for embryonic acid-base metabolic compensation and hematological regulation during CO2 challenges in layer and broiler chicken strains. Respir Physiol Neurobiol 215: 1-12. https://doi.org/10.1016/j.resp.2015.04.008

Cardeal PC, Caldas EOL, Lara LJZ, Rocha JSR, Baiao NC, Vaz DP, Martins NDS, 2015. In ovo feeding and its effects on performance of newly-hatched chicks. Worlds Poult Sci J 71(4): 655-662. https://doi.org/10.1017/S0043933915002445

Christensen H, Bachmeier J, Bisgaard M, 2021. New strategies to prevent and control avian pathogenic Escherichia coli (APEC). Avian Pathol 50: 370-381. https://doi.org/10.1080/03079457.2020.1845300

Cooper JB, 1972. Effect of light during incubation on hatchability of turkey eggs. Poult Sci 51(4): 1105-1108. https://doi.org/10.3382/ps.0511105

Costa BTA, Lopes TSB, Sousa LS, Barbosa HJS, Gonçalves TF, Franco CMA, et al., 2022. Floor eggs: Hatchability, microbiological analysis, and effects of post-hatch use of ceftiofur on performance of broiler chicks. Res Vet Sci 152: 610-617. https://doi.org/10.1016/j.rvsc.2022.09.020

Cox NA, Bailey JS, Mauldin JM, Blankenship LC, 1990. Research note: Presence and impact of Salmonella contamination in commercial broiler hatcheries. Poult Sci 69(9): 1606-1609. https://doi.org/10.3382/ps.0691606

Cox NA, Berrang ME, Buhr RJ, Bailey JS, 1999. Bactericidal treatment of hatching eggs II. Use of chemical disinfectants with vacuum to reduce Salmonella. J Appl Poult Res 8(3): 321-326. https://doi.org/10.1093/japr/8.3.321

Craven SE, Cox NA, Stern NJ, Mauldin JM, 2001. Prevalence of Clostridium perfringens in commercial broiler hatcheries. Avian Dis 45(4): 1050-1053. https://doi.org/10.2307/1592887

De Smit L, Bruggeman V, Tona JK, Debonne M, Onagbesan O, Arckens L, Decuypere E, 2006. Embryonic developmental plasticity of the chick: Increased CO2 during early stages of incubation changes the developmental trajectories during prenatal and postnatal growth. Comp Biochem Physiol A: Mol Int Physiol 145(2): 166-175. https://doi.org/10.1016/j.cbpa.2006.06.046

EC, 1999. Council Directive 1999/74/Ec of 19 July on laying down minimum standards for the protection of laying hens. http://eurlex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:1999:203:0053:0057:EN:PDF [22 April 2021].

Everaert N, Willemsn H, Hulikova A, Brown H, Decuypere E, Swietach P, Bruggeman V, 2010. The importance of carbonic anhydrase II in red blood cells during exposure of chicken embryos to CO2. Resp Physiol Neurobio 172(3): 154-161. https://doi.org/10.1016/j.resp.2010.05.007

Everaert N, Willemsn H, Kamers B, Decuypere E, Bruggeman V, 2011. Regulatory capacities of a broiler and layer strain exposed to high CO2 levels during the second half of incubation. Comp Biochem Physiol A: Mol Int Physiol 158(2): 215-220. https://doi.org/10.1016/j.cbpa.2010.10.031

Fasenko GM, Robinson FE, Whelan AI, Kremeniuk KM, Walker JA, 2001. Prestorage incubation of long-term stored broiler breeder eggs: 1. Effects on hatchability. Poult Sci 80(10): 1406-1411. https://doi.org/10.1093/ps/80.10.1406

Fasenko GM, Christopher EOD, McMullen LM, 2009. Spraying hatching eggs with electrolyzed oxidizing water reduces eggshell microbial load without compromising broiler production parameters. Poult Sci 88(5): 1121-1127. https://doi.org/10.3382/ps.2008-00359

FAWC, 2019. https://webarchive.nationalarchives.gov.uk/20121010012427/http://www.fawc.org.uk/freedoms.htm. Farm Animal Welfare Council [19 Jul 2019].

Galli R, Preusse G, Uckermann O, Bartels T, Krautwald-Junghanns ME, Koch E, Steiner G, 2016. In ovo sexing of chicken eggs by fluorescence spectroscopy. Anal Bioanal Chem 409(5): 1185-1194. https://doi.org/10.1007/s00216-016-0116-6

Givisiez PE, Moreira Filho AL, Santos MR, Oliveira HB, Ferket PR, Oliveira CJ, Malheiros RD, 2020. Chicken embryo development: Metabolic and morphological basis for in ovo feeding technology. Poult Sci 99(12): 6774-6782. https://doi.org/10.1016/j.psj.2020.09.074

Graham BD, Selby CM, Graham LA, Teague LE, Tellez-Isaias G, Hargis BM, Vuong CV, 2021. Development of a wild-type Escherichia coli environmental bloom model to evaluate alternatives to formaldehyde fumigation in broiler chicken hatch cabinets. Poult Sci 100(3): 100975. https://doi.org/10.1016/j.psj.2020.12.072

Graham BD, Teague LE, Latorre JD, Yang Y, Baxter MFA, Mahaffey BD, Tellez-Isaias G, 2018. Use of probiotics as an alternative to formaldehyde fumigation in commercial broiler chicken hatch cabinets. J Applied Pout Res 27(3): 371-379. https://doi.org/10.3382/japr/pfy008

Gremmen B, Bruijnis MRN, Blok V, Stassen EN, 2018. A public survey on handling male chicks in the Dutch egg sector. J Agric Environ Ethics 31(1): 93-107. https://doi.org/10.1007/s10806-018-9712-0

Groff-Urayama P, Padilha J, Einsfeld S, Pertile S, Gorges M, De Andrade M, Takahashi S, 2019. Performance, intestinal morphometry and incubation parameters of broiler chickens submitted to in ovo feeding with different techniques and amino acids. Can J Anim Sci 0: 1-9. https://doi.org/10.1139/cjas-2018-0131

Hamissou Maman A, Özlu S, Uçar A, Elibol O, 2018. Effect of chick body temperature during post-hatch handling on broiler live performance. Poult Sci 98(1): 244-250. https://doi.org/10.3382/ps/pey395

Hayretdag S, Kolankaya D, 2008. Investigation of the effects of pre-incubation formaldehyde fumigation on the tracheal epithelium of chicken embryos and chicks. Turk J Vet Anim Sci 32(4): 263-267.

He L, Martins P, Huguenin J, Manso T, Galindo T, Gregoire F, Espeut J, 2019. Simple, sensitive and robust chicken specific sexing assays, compliant with large scale analysis. PloS One 14(3): e0213033. https://doi.org/10.1371/journal.pone.0213033

Hedlund L, Whittle R, Jensen P, 2019. Effects of commercial hatchery processing on short-and long-term stress responses in laying hens. Scient Report 9(1): 2367. https://doi.org/10.1038/s41598-019-38817-y

Hollemans MS, De Vries S, Lammers A, Clouard C, 2018. Effects of early nutrition and transport of 1-day-old chickens on production performance and fear response. Poult Sci 97(7): 2534-2542. https://doi.org/10.3382/ps/pey106

Huth JC, Archer GS, 2015. Effects of LED lighting during incubation on layer and broiler hatchability, chick quality, stress susceptibility and post-hatch growth. Poult Sci 94 (12): 3052-3058. https://doi.org/10.3382/ps/pev298

Jansson DS, 2018. Gaining insights into the health of non-caged layer hens. Vet Record 182(12): 347-349. https://doi.org/10.1136/vr.k1271

Macari M, Gonzales E, Patrício IS, Nääs IA, Martins PC (Eds), 2013. Manejo da incubação, 3th ed. Jaboticabal, FACTA, SP, Brazil. 536 pp.

Meijerhof R, 1992. Pre-incubation holding of hatching eggs. Worlds Poult Sci J 48(1): 57-68. https://doi.org/10.1079/WPS19920006

Mellor DJ, Reid CSW, 1994. Concepts of animal well-being and predicting the impact of procedures on experimental animals. Improving the well-being of animals in the research environment, 3-18.

Melo EF, Clímaco WLS, Triginelli MV, Vaz DP, De Souza MR, Baiao NC, Lara LJC, 2019. An evaluation of alternative methods for sanitizing hatching eggs. Poult Sci 98(6): 2466-2473. https://doi.org/10.3382/ps/pez022

Melo EF, McElreath JS, Wilson JL, Lara LJC, Cox NA, Jordan BJ, 2020. Effects of a dry hydrogen peroxide disinfection system used in an egg cooler on hatchability and chick quality. Poult Sci 99(11): 5487-5490. https://doi.org/10.1016/j.psj.2020.05.050

Mesquita MA, Araújo ICS, Café MB, Arnhold E, Mascarenhas AG, Carvalho FB, et al., 2021. Results of hatching and rearing broiler chickens in different incubation systems. Poult Sci 100(1): 94-102. https://doi.org/10.1016/j.psj.2020.09.028

Mueller S, Kreuzer M, Siegrist M, Mannale K, Messikommer RE, Gangnat IDM, 2018. Carcass and meat quality of dual-purpose chickens (Lohmann Dual, Belgian Malines, Schweizerhuhn) in comparison to broiler and layer chicken types. Poult Sci 97(9): 3325-3336. https://doi.org/10.3382/ps/pey172

Muhammad M, Muhammad L, Ambali AG, Mani AU, Azard S, Barco L, 2010. Prevalence of Salmonella associated with chick mortality at hatching and their susceptibility to antimicrobial agents. Vet Microb 140(1-2): 131-135. https://doi.org/10.1016/j.vetmic.2009.07.009

Muir WI, Groves PJ, 2019. The leg strength of two commercial strains of meat chicken subjected to different incubation profiles. Animal 13(7): 1489-1497. https://doi.org/10.1017/S1751731118002999

NFACC, 2016. Code of practice: For the care and handling of hatching eggs, breeders, chickens, and turkeys. National Farm Animal Care Council, Ottawa, Canada.

Oliveira GDS, Dos Santos VM, Nascimento ST, Rodrigues JC, 2020. Alternative sanitizers to paraformaldehyde for incubation of fertile eggs. Poult Sci 99(4): 2001-2006. https://doi.org/10.1016/j.psj.2019.11.032

Peebles ED, Brake J, 1985. Relationship of eggshell porosity to stage of embryonic development in broiler breeders. Poult Sci 64(12): 2388-2391. https://doi.org/10.3382/ps.0642388

Pokhrel N, Cohen EBT, Genin O, Ruzal M, Sela-Donenfeld D, Cinnamon Y, 2018. Effects of storage conditions on hatchability, embryonic survival and cytoarchitectural properties in broiler from young and old flocks. Poult Sci 97(4): 1429-1440. https://doi.org/10.3382/ps/pex393

Rezaee MS, Liebhart D, Hess C, Hess M, Paudel S, 2020. Bacterial infection in chicken embryos and consequences of yolk sac constitution for embryo survival. Vet Pathol 58: 71-79. https://doi.org/10.1177/0300985820960127

RSPCA, 2017. Welfare standards for hatcheries. Chicks, poults and ducklings. Society for the Prevention of Cruelty to Animals, UK, 17 pp.

Shafey TM, 2004. Effect of lighted incubation on embryonic growth and hatchability performance of two strains of layer breeder eggs. Brit Poult Sci 45(2): 223-229. https://doi.org/10.1080/00071660410001715821

Sharma J, Burmester B, 1982. Resistance of Marek's disease at hatching in chickens vaccinated as embryos with the Turkey Herpesvirus. Avian Dis 26(1): 134-149. https://doi.org/10.2307/1590032

Soucy K, Randall CJ, Holley RA, 1983. Microbiological monitoring of hatchery sanitation. Poult Sci 62(2): 298-309. https://doi.org/10.3382/ps.0620298

Taylor LW, Sjodin RA, Gunns CA, 1956. The gaseous environment of the chick embryo in relation to its development and hatchability: 1. Effect of carbon dioxide and oxygen levels during the first four days of incubation upon hatchability. Poult Sci 35(6): 1206-1215. https://doi.org/10.3382/ps.0351206

Tona K, Bamelis F, De Ketelaere B, Bruggeman V, Moraes VM, Buyse J, Decuypere E, 2003. Effects of egg storage time on spread of hatch, chick quality, and chick juvenile growth. Poult Sci 82(5): 736-741. https://doi.org/10.1093/ps/82.5.736

Tong Q, McGonnell IM, Demmers TGM, Roulston N, Bergoug H, Romanini CE, Exadaktylos V, 2018. Effect of a photoperiodic green light programme during incubation on embryo development and hatch process. Animal 12(4): 765-773. https://doi.org/10.1017/S1751731117002117

Trukhina AV, Lukina NA, Smirnov AF, 2016. Experimental sex inversion of chicken embryos at aromatase inhibition, estrogen receptor modulation, DNA demethylation and progesterone treatment. Natur Sci 8(11): 451. https://doi.org/10.4236/ns.2016.811047

Underwood G, Andrews D, Phung T, Edwards LE, 2021. Incubation, hatchery practice and the welfare of layer hens. Anim Product Sci 61(10): 867-875. https://doi.org/10.1071/AN20391

Uni Z, Ferket PR, 2003. US Patent No. 6592878, Washington DC, US Patent and Trademark Office.

Van Roovert-Reijrink IAM, Van der Pol CM, Molenaar R, Van den Brand H, 2018. Effect of warming profile at the onset of incubation on early embryonic mortality in long stored broiler eggs. Poult Sci 97(11): 4083-4092. https://doi.org/10.3382/ps/pey261

Wang Y, Jin G, Ma M, Xiang X, 2019. Sex differences in serum steroid hormone levels during embryonic development in hen eggs. Poult Sci 98(11): 6053-6062. https://doi.org/10.3382/ps/pez270

Webster AJ, 2001. Farm animal welfare: the five freedoms and the free market. The Vet J 161(3): 229-237. https://doi.org/10.1053/tvjl.2000.0563

Webster B, Hayes W, Pike TM, 2015. Avian egg odour encodes information on embryo sex, fertility and development. PloS One 10(1): e0116345. https://doi.org/10.1371/journal.pone.0116345

Wright ML, Anderson GW, Epps NA, 1960. Hatchery sanitation as a control measure for aspergillosis in fowl. Avian Dis 4(4): 369-379. https://doi.org/10.2307/1587686

Yu Y, Li Z, Zhong Z, Jin S, Pan J, Rao X, Yu Y, 2018. Effect of monochromatic green LED light stimuli during incubation on embryo growth, hatching performance, and hormone levels. T ASABE 61(2): 661-669. https://doi.org/10.13031/trans.12485

Zeweil HS, Rizk RE, Bekhet GM, Ahmed MR, 2015. Comparing the effectiveness of egg disinfectants against bacteria and mitotic indices of developing chick embryos. The J Basic Appl Zool 70: 1-15. https://doi.org/10.1016/j.jobaz.2014.12.005

Zhang H, Burggren WW, 2012. Hypoxic level and duration differentially affect embryonic organ system development of the chicken (Gallus gallus). Poult Sci 91(12): 3191-3201. https://doi.org/10.3382/ps.2012-02449