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Review Article
Fertility and Hatchability in Goose Eggs: A Review

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Abstract

Two species of geese were domesticated from the waterfowl belonging to the *Anatidae* family, the Greylag goose (*Anser anser*) and the Swan goose (*Anser cygnoides*), which became the ancestors of most domestic geese all over the world. The rapid increase in goose production and the demand for day old goslings in the last century required improvements in breeding, nutrition, reproduction and management. This review focuses on the reproduction of geese with particular emphasis on two determining factors of artificial incubation, the fertility and hatchability of goose eggs. The first part of this review presents the factors that affect fertility and the latest ideas offering better performance in this regard. The second part of the review discusses three groups of factors influencing hatchability: breeder factors, egg factors and incubator/hatcher factors. Numerous studies were conducted in several topics with regards to goose egg hatchability in the last two decades, which are collected and discussed here. The third part discusses possibilities for future advances in relation to fertility and hatchability of goose eggs.

Key words: Egg, fertility, geese, hatchability, incubation

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INTRODUCTION

All goose species belong to the order of *Anseriformes* and the family of *Anatidae*[1-3]. The goose was one of the first avian species to be domesticated approximately 4000 years ago or even earlier[1-5]. The majority of European goose breeds are descended from the Greylag goose (*Anser anser*), while the Swan goose (*Anser cygnoides*) is the anscestor of the current Asian and African goose breeds[2-5].

Over the course of goose domestication several genetic and phenotypic traits changed and production performance improved[2,3,5]. Further, in the last 100 years waterfowl production was on an upward trend, especially since the introduction of breeding programs[2]. In the world goose production increased ten-fold between the 1960s and 2010, while in the same period a 17-fold increase occurred in Asia[2]. Because of the increased market demand for day old goslings in the goose industry and the seasonality of goose production, the improvement of reproductive performance in geese is crucial.

Fertility and hatchability are important economic factors that represent major components of reproductive performance. Fertility is affected by several factors, such as genetics, female age, sex ratio, temperature, light, sexual behavior, nutrition, housing system and health of the birds[3,6-9]. There are three groups of factors that affect hatchability: breeder factors (genetics, breeder age, season and nutrition), egg factors (egg quality, embryo survival and egg storage) and incubator/hatcher factors (temperature, relative humidity, carbon-dioxide concentration and ventilation, egg turning, cooling and hygiene)[3,9-11]. Some of the factors are interrelated and affect both fertility and hatchability and it is necessary to understand those factors at each level. Further, several studies were conducted in recent years with relevant new information regarding goose reproduction. Therefore the objective here was to provide up to date information on the factors that influence fertility and hatchability in goose eggs and also ways to improve these essential parameters.

Fertility: The fertility of the eggs are affected by genetics, female age, sex ratio, yolk size, number of production cycles, temperature, light, sexual behavior, nutrition, housing system and health of the birds[3,6-9,12]. Fertility also depends on the female's ability to ovulate, store sperm and provide an environment for fertilization and egg formation[7,8]. Further, the quality and quantity of the semen deposited by the male is crucial for fertility[3,7,11]. The fertilizing ability of ganders can be described by the Sperm Quality Factor, which includes the ejaculate volume, the sperm concentration and the percentage of live spermatozoa[13,14].

There is variation in the fertility of different goose breeds (53.8-84.72%)[15-18] but heavy breeds have lower fertility, which can be improved with crossing[3]. Fertility is also affected by the age of the birds. The fertility in the first year is usually lower, reaches its peak in the second or third year and then declines gradually[3,14,19]. The lower fertility in the first year was due to the lower quality semen[14,20], number of successful copulations[21,22], social rank and sexual experience of ganders[22] and probably the differences in sperm storage and transport of geese[23]. Thus it is suggested to use experienced (2 years or older) ganders with one year old geese to achieve higher fertility in the first laying cycle[22,23].

Seasonal factors affect fertility under natural light conditions and the seasonal changes in the reproduction of geese are controlled by light and hormones[3,24]. The reproductive season of domestic geese in temperate areas starts at early spring (8 hour day length) and finish around June (16-18 hour day length)[3,24-27] and in the same time the secretion of gonadal steroid hormones is associated with egg laying[24,28] and with sperm volume and concentration[3,29]. The synchrony of these changes in males and females is crucial in the reproductive season to reach high fertility[3]. In intensive production systems this synchrony may not always be achieved, especially in the second part of the laying season (with long day lengths) when the egg production of the females and mating activity of the males decline[25] due to the endocrinological changes. However, these problems may be overcome by artificial lighting programs[3,24,25].

The artificial lighting program suggest indoor keeping in intensive production system, however geese can also be kept in extensive production system, ie. in free-range[6,19]. Even in intensive production systems (with max. 2 birds/m^2 stocking density) the geese are allowed access to a yard during day time[3], closing them in 24 h a day may result in behavioural (e.g. nervousness) and health problems (e.g. *Mycoplasma* infection) that can affect the reproduction of geese[30]. Still the egg production of the geese can be improved 30-50% in closed, intensive systems with artificial lighting program[3]. Further, the access to open water can be beneficial in terms of mating and can reduce the possibility of phallus damage[3].

Apart from light, relatively little is known about the effects of other climatic factors on fertility. The long and cold winters delay the reproductive cycle in both sexes, while a mild and short winter may advance it. Further, the cold negatively affects the functioning of the gonads and the mating activity and sperm production of ganders decrease considerably below -2°C and above 25°C[3].

The sex ratio of the birds also has to be considered in terms of their fertilizing capacity. It is known that the heavy

breeds have lower reproducing ability; therefore it is recommended to have a sex ratio of 1 male to 3-4 females. Medium breeds can be kept with a sex ratio of 1 to 5-6, while light breeds achieve high fertilization even with a sex ratio of 1 to 6-7; however at a sex ratio of 1 to 8-10 fertility will be reduced[3]. In large scale, intensive production systems the sex ratio is generally 1:3-4 to ensure high fertility[3], though in fully closed keeping this may result in lower fertility due to low mating success as a result of intrusion (by other males or females) and high stocking density.

The provision of good quality feed (free from mycotoxins) in the rearing and laying period is necessary to maintain high fertility in geese[3]. The protein and energy content of the feed has to be high enough for the requirements for egg production but there are other nutritional factors (e.g. vitamins and microelements) that are crucial for fertility[3]. The deficiency of vitamin E in male birds causes tissue degeneration in the testes[3], affects semen volume, sperm concentration, sperm motility and viability, while in female birds the egg production, egg weight, egg composition and fertility are affected[31]. Selenium supplementation provides protection from oxidative damage and affects spermatogenesis, thus helps maintain male fertility[32]. Further, vitamin E was observed to have synergistic effects with selenium[32]. Ganders supplemented with vitamin E and organic selenium had higher ejaculate volume and sperm concentration with fewer abnormal sperms[33]. The deficiency of zinc also reduces fertility[3,34]. Zinc supplementation in domestic fowl increased sperm penetration in the egg yolk and improved fertility probably due to better semen quality and higher sexual efficiency[34].

Ultimately the fertility of geese can be improved by artificial insemination[3,14,19,35] and if done properly the level of fertility can be as high as those achieved by natural mating or even higher[3,19]. The artificial insemination is done in two steps: semen is collected from the ganders by dorso-abdominal massage and then the inseminating person inserts the index finger of his left hand into the vent of the goose locating the oviduct by palpation and using a syringe (guided along the inserted finger) the semen is deposited in the oviduct[3,14,19,35]. It is important to use fresh, good quality sperm for insemination that is free from contaminants[3,14]. The fresh semen should be inseminated as soon as possible (usually in 30 minutes)[3] but with special diluents its fertilizing capacity could be maintained up to 3-8 h[3,14]. Studies are ongoing with regards to poultry semen cryopreservation; however the fertility rates of frozen/thawed poultry semen are variable due to the unique morphological and physiological features of poultry sperm cells and due the toxic or contraceptive effects of commonly used cryoprotectants on poultry sperm[36-38]. Thus currently only the use of fresh semen is recommended for artificial insemination in the poultry industry[36,38].

Hatchability

Breeder background

Genetics: Based on their body weight, geese can be sorted into three categories: heavy (e.g. African, Toulouse and Embden), medium (e.g. Italian and Landes) and light breeds (Czech and Chinese)[3,4,39]. The hatchability shows little variability among medium and light breeds but the heavy breeds have considerably lower hatchability[3].

Based on what the geese are used for meat (e.g. Embden and Pomeranian), liver (e.g. Toulouse and Landes) and egg (e.g. Italian, Czech and Chinese) type geese are distinguished[3,4,19,39]. The reproducing capability (e.g. fertility, hatchability, egg production) of meat and liver type geese is weak or medium but it is good in egg type geese. Naturally egg type geese are used for producing meat or liver but due to their good reproduction they are used as female line in crosses[3].

Breeder age: Egg weight changes with female age in geese. It was found in several studies that the egg weight of two years old geese were greater than one year old geese[17,26,40-43]. The egg weight continued to increase up to 4-5 years of age and then declined[3,26,41,43]. This continued increase in egg weight results in increased gosling weight at hatch[3,44-46] and possibly better gosling quality, performance and survivability. Further, Merritt and Lemay[41] found an increase in hatchability from one to two years of age but then a continuous decline up to 5 years of age.

The age of the female also determines the deposition of nutrients in the egg (domestic fowl[47,48], ducks[49,50] and geese[43,51]). In domestic fowl comparative studies using eggs from younger and older birds with different internal egg characteristics (yolk content, albumen content, fatty acid composition) confirmed earlier findings that larger chicks hatched from larger eggs[44,45] but also showed a difference in yolk and energy utilization, heat production and embryo development between the eggs of different internal characteristics[47,48,52,53] that might result in differing hatchability and chick quality. The eggshell conductance of domestic fowl eggs also differed; it was higher in young and old flocks compared to peak and mature flocks[52] probably due to poorer shell quality in young and old flocks that might result in lower hatchability (for example because of excess dehydration or bacterial penetration).

Season: The goose was one of the first birds to be domesticated[1,3,5,19], still geese display seasonal breeding patterns, as egg laying concentrates for certain months of the year[5,24]. Geese rely on seasonal changes in the daily photoperiod to determine the timing and duration of the breeding season[24,54,55].

Adult (2 years or older) domestic geese exhibit a pattern of declining egg weight over the laying season[3,26,42,51,56-58] resulting in declining day old gosling weight that may affect gosling quality in the short and long term[56,59].

Several studies showed seasonal variation in the contents of goose eggs[16,51,57,58,60]. A decline in shell weight, yolk weight, Haugh index, albumen height, thick albumen area, shell thickness and n-6 and n-3 polyunsaturated fatty acid ratio was shown over the laying season[51,57,58,60]. Thus goose egg quality declined over the laying season, which negatively affected hatchability[58]. Mazanowski *et al.*[16] found a decrease in the percentage of water, protein and ash and an increase in fat in the yolk between an early and late point in the laying season. In egg white, water and protein decreased and ash slightly increased between an early and late point in the laying season[16]. Interestingly these changes did not affect hatchability, which was relatively constant throughout the laying season[16].

Nutrition: Feeding essential nutrients to the parent stock is crucial[3,61]. Deficiencies in nutrition can negatively affect hatchability; the quality and quantity of proteins is important, as well as the continuous supply of vitamins and microelements[3]. The most important vitamins that affect hatchability due to their role in metabolism during embryo development are vitamin A, vitamin B_{12}, riboflavin and pantothenic acid[3,61]. Embryos with such deficiencies showed signs of abnormal circulatory system development, haemorrhages, poor feathering, swollen hocks, beak abnormalities and dwarfing[3,61]. According to Wilson[61] lack of vitamin D and E also reduced hatchability due to problems associated with the calcium metabolism and skeletal formation, blindness and abnormal vascular system. Several micro and macro-elements affect hatchability too[61]. Manganese is an enzyme activator in embryonic metabolism[3], while zinc deficiency caused impaired feather and bone development[3,61].

The transfer and deposition of essential nutrients in the egg for the developing embryo during egg formation is important too[61,62]. Initially the germ development is supported with nutrients from the yolk and albumen[62]. The developing embryo first utilizes carbohydrates from the albumen until the chorioallantois is developed enough to access oxygen (O_2)[11,62]. After the chorioallantois is complete the access to O_2 supports the combustion of fatty acids, which are the primary source of energy and the basis for embryo development[62] and also glycogen reserves are built from the carbohydrates[11]. In the second half of the incubation the utilization of proteins and fats increases and the rate of embryo development speed up resulting in metabolic heat production[11]. By the time of hatching reserves are concentrated in the embryo and in the yolk sac that will support the needs of the embryo for a short period of time after hatching[11,62].

Egg factors

Egg quality: The external and internal characteristics of the eggs affect embryo development and hatchability[9-11].

It is preferable to incubate eggs of average weight that show the best hatchability[9,10,18,63]. Eggs lighter or heavier than average have lower hatchability as such eggs probably have different requirements in terms of incubational parameters; for example lighter goose eggs need lower incubation temperature[3]. There is a lower weight limit for goose eggs suitable for incubation, which is 140 g for one year old geese and 150 g for two years or older geese[3,11].

Similarly, egg size is important with intermediate sized eggs showing the best hatchability[9,45]. Studies in domestic fowl, ducks and geese showed that egg shape also affected hatchability, i.e., eggs that were too round (for example spherical or oval egg shapes; see Roberts[64] or Salamon and Kent[65]) or too pointed (for example biconical or conical egg shapes; see Roberts[64] or Salamon and Kent[65]) had lower hatchability[10,66-68].

It is known from comparative studies that egg content weight increases with egg weight and that larger eggs contain proportionately more yolk[69,70]. Further, it was found using double-yolked duck eggs that larger yolks positively affected the amount of albumen secreted in the eggs[71] and also the size of the eggs[12]. This is important because there is a linear relationship between egg weight and hatchling weight, ie. the larger the egg the larger the hatchling[44-46]. Shell quality also affected hatchability in poultry[9,10,72,73]. Goose eggs with thicker shells had higher hatchability[3,10], as eggs with thinner shells were more prone to bacterial infections and excessive dehydration[3].

The quality of the eggs change during storage (domestic fowl[74,75]; ducks[76]; and geese[77]) due to the loss of carbon-dioxide (CO_2) and water[11]. The loss of CO_2 and water resulted in a reduction of egg weight, Haugh unit, albumen index and yolk index, while resulted in an increase in albumen and yolk

pH[68,74-77]. All these changes reduced the lysozyme activity limiting the antibacterial protection of the egg[11]. Further, the longer storage period affected hatchability, hatchling quality and post hatch performance[68,74-76].

Embryo survival: There are two 'critical' periods during incubation in avian species with mortality peaks: one occurs in early embryonic life and the other shortly before hatching[11,78,79]. The early embryonic mortality coincides with the development of the blood circulatory system and also with the diet change from simple carbohydrates to more complex proteins and lipids[11]. Further, early embryonic mortality may be the result of genetic abnormalities[80,81], insufficient availability of nutrients in the egg or the fact that the egg might be exposed to conditions, which did not match the needs of the developing embryo[82]. The peak mortality before hatching is attributed to failure to make proper transition to pulmonary respiration[11,78].

Egg storage: It would be preferable to set fresh eggs in the incubators but generally that is not possible in large hatcheries due to management factors, therefore the eggs need to be stored. The storage conditions of the eggs before incubation are critical and have significant impact on the hatchability[3,9,11].

The first important factor is the cleaning (washing and disinfecting) of the eggs after collection and before being placed into storage[3,11,19]. There are two alternatives for cleaning the eggs of waterfowl: one is to remove the cuticle[59,83,84] and the other is to keep the cuticle[3,11,85] both having advantages and disadvantages. The cuticle removal requires high concentration of chlorine, which may have consequences for people, equipment and environment[85,86]. The cuticle is a protective barrier against contamination[85,87,88] and microorganisms[11,89-91] and its removal changes (increases or decreases depending on female age) the water vapour conductance of the egg shell possibly enhancing the survival of the embryo[86,88,92], especially in those species with initially low egg shell conductance[92]. The eggshell permeability greatly increases in the absence of a cuticle, as the pores are exposed to the outside environment[89], thus such eggs intended for hatching need to be treated (with disinfectants or probiotics) to be protected against microbial and fungal infections. However, this is also true for cleaning eggs when the cuticle is not removed. Goose eggs, when dirty, need to be washed gently (making sure not to remove the cuticle) with a sponge or brush in 38-40°C water and after that dipped into a 41-42°C disinfectant solution for 1-2 min[3,11]. The egg washing should be quick[3], as it has negative impact on the hatchability (the shell membranes become rubber like making pipping difficult). Further, in the presence of a cuticle the gas exchange of the goose eggs may be impaired[92] resulting in poorer hatchability too. The eggs need to be completely dry before being transferred to storage[11]. It is important to use a wide spectrum disinfectant on the eggs that is easily soluble, effective but non-toxic and non-corrosive[3,93,94]. Goose eggs not requiring washing can be fumigated with formaldehyde or alternatively with safer products before being transferred to storage[11,93,94].

The hatching eggs need to be cooled as soon as possible below the physiological zero (20°C) and moved to storage to protect their quality[3]. Storage temperature (10-17°C) and relative humidity (55-75%) parameters vary between studies[19,68,77,95-97], however it has to be noted that up to one week storage goose eggs require 12-17°C storage temperature but for longer storage period goose eggs may need to be cooled below 10°C with 75-85% relative humidity[3,11]. The quality of the goose eggs deteriorate during storage[3,77] but it is also known from studies with broilers that during cold storage there is a delay in embryonic development[98,99] and a reduction in the number of cells in the blastoderm due to apoptosis or necrosis, which reduces embryo viability and hatchability[100-102]. To regenerate the lost cells in the blastoderm periodic warming (or sometimes called SPIDES, ie. short periods of incubation during egg storage) can be applied during egg storage (domestic fowl[98,102-104], turkeys[105,106], ducks[107] and geese[96,108]). All of the above studies reported an increase in hatchability of long stored eggs after periodic warming. Further, studies showed other benefits of the periodic warming during storage, such as advanced embryo development into a stage that is able to withstand storage better, shortened hatch window and incubation time, reduced early embryonic mortality and increased quality of the day old hatchlings[3,96,98,104-106].

The position of the eggs during storing is also important. It is known that storing eggs for longer period with pointed end up compared to the round end up resulted in higher hatchability (domestic fowl[109,110] and geese[3]). Storing the eggs pointed end up provides increased protection for the embryo against dehydration and temperature changes and also prevents the embryo from sticking to the shell membrane[3]. Alternatively eggs may be stored on setting trays and turned with automated turning system (similar to those in incubators) during storage to protect egg quality and embryo viability (domestic fowl[111] and geese[3]). However, it has to be noted that hatching eggs may be subjected to excessive turning during storage resulting in poorer hatchability[111,112].

Incubator/hatcher factors

Temperature: Most poultry species seem to have an optimal incubation temperature that falls between 37-38°C and deviations from this optimum can have major impact on hatching success[113,114]. This optimal incubation temperature starts and maintains cell multiplication and embryonic metabolic processes[3]. Below and above the optimal incubation temperature the rate of embryonic development is altered, incubation time extended, embryo vitality reduced and post hatch performance negatively affected[3,9,113-115].

According to Bogenfurst[3] the optimal incubation temperature for geese is 37.8°C between 1-12 days of incubation, 37.5°C between 13-23 days of incubation, 37.2°C between 24-27 days of incubation and 37-37.2°C during hatching. The gradually declining temperature is applied to compensate for the embryo's metabolic heat production in the second half of the incubation (see Cooling section). However, many studies maintained one standard temperature throughout the incubation period and another one (usually 0.3-0.5°C lower) during hatching[58,80,95-97], which was within the 37-38°C optimum incubation temperature suggested by Visschedijk[113] and French[114]. It is important to note that in the application of one standard incubation temperature the frequency and length of the cooling has to be modified to compensate for the metabolic heat production of the embryo compared to the method that uses gradually decreasing temperature.

Relative humidity: Gases pass through the pores and membranes of the egg shell by diffusion[116,117] and the rate of diffusion is related to the functional porosity of the egg shell and the pressure difference across the egg shell[118,119]. Avian eggs lose weight during incubation, which is mainly due to the loss of water vapour[120,121], as the mass gain of oxygen equals the mass loss of carbon-dioxide[117,120]. This weight loss is necessary for normal embryonic development[3,122,123] but excess water loss results in the drying of the shell membranes, while too little water loss enhances the growth of microbes and also results in the swelling of the shell membranes that plug the pores causing embryonic death[3]. The humidity during incubation also affects the bone development of the embryo and the volume of the air chamber increases (necessary for the transition to pulmonary breathing at internal pipping) due to the weight loss of the egg[3], which is optimally around 12-13% in geese[3,95,124].

The rate of weight loss is determined by the relative humidity of the air around the egg. The relative humidity can be determined by the comparison of the dry and wet bulbs in the incubator. However, regular weighing of the eggs and the examination of the growth of the air chamber during incubation gives more precise information[3]. According to Bogenfurst[3,11] for goose eggs during incubation the optimal relative humidity (and wet bulb temperature) is 63% (31.1°C) for day 1-4 of incubation, 54% (29.5°C) for day 5-12 of incubation, 56% (29.5°C) for day 13-23 of incubation and 57% (29.5°C) for day 24-27 of incubation. During hatching 77-80% relative humidity (with 33-34°C wet bulb temperature) is necessary, which makes movement and cracking through the shell easier for the goose embryo. The relative humidity should increase gradually during hatching and reach its peak at the start of external pipping. If relative humidity is too low, the goslings will be sticky (and their feathers stick to the shell); if it is too high the goslings will be covered in egg contents and their navels will not close properly[3].

Several studies used constant relative humidity settings during incubation ranging between 40-65% and during hatching ranging between 65-75%[58,68,80,95-97,124] (Table 1). The hatchability of fertile in these studies ranged between 58.5-87.8%[58,68,95-97], which is relatively good considering the wide range of relative humidity setting used in the studies.

Carbon-dioxide concentration and ventilation: Carbon-dioxide (CO_2) is produced during incubation due to the metabolism[117,125] and in low concentration it is necessary for embryo development through the utilization of the calcium content of the egg shell[3,11]. The carbon-dioxide concentration can reach up to 0.5% in the incubator without affecting embryo development but it can greatly reduce hatchability above 1.5%[3,11].

Table 1: Goose egg hatchability results of studies presenting temperature and relative humidity settings during incubation and hatching. Overall hatchability of fertile (%) was calculated from results originating from multiple experiments in the mentioned studies

	Incubator settings		Hatcher settings		
Study	Temperature (°C)	Relative humidity (%)	Temperature (°C)	Relative humidity (%)	Hatchability of fertile (%)
Amantai *et al.*[68]	37.5-38.1	day 1-8: 60-65, day 9-25: 45-50	not presented	not presented	65 (overall)
Biesiada-Drzazga *et al.*[58]	37.8	52	37.2-37.3	65-75	76.6
Kucharska-Gaca *et al.*[96]	37.8	57	37	75	78.2
Kucharska-Gaca *et al.*[97]	37.7	55	37.4	75	76.5 (overall)
Meir and Ar[95]	37.5	40	37.2	68.5	77.1

In geese it is advised to have relatively higher (around 1%) carbon-dioxide concentration before pipping, because the lack of oxygen encourages the embryo to hatch and from the reaction of carbon-dioxide and water vapour we get carbonic acid that dissolves the egg shell and makes the hatching easier for the embryo[3,11]. However after hatching the ventilation should be increased to achieve normal oxygen concentration[3] otherwise mortality will occur.

The ventilation settings for goose eggs should be carefully managed during incubation to achieve the required carbon-dioxide concentration but also to achieve maximum hatchability without unnecessary embryo mortality during incubation and hatching[3]. In the first few days the vents should be closed on the incubator to reach the required temperature as soon as possible but after that gradually the vents can be opened up to 50% by day 12 of incubation. Then between day 12 and 24 of incubation the vents can be open up to 75-80% and after that vents can be open 80-100% until transferring to the hatcher[3,11]. Similarly in the hatcher the vents can be open 80-100% depending on carbon-dioxide concentration and temperature requirements[3,11].

Egg turning: Egg turning has many functions that include prevention of the embryo adhering to the egg shell[9,126-130], reduction of embryo malpositioning[112,131,132] and proper and timely closure of the chorioallantoic membrane[132-134]. Egg turning is also necessary for embryonic growth and for the proper utilization of albumen by the developing embryo[132-135]. Several studies were conducted in domestic fowl about egg turning[112,126-128,136] and now it is generally accepted that turning more than 24 times a day is unnecessary[129]. Further, now commercial incubators rotate eggs 90° (45° from either side of the vertical) at hourly intervals using automatic turning systems[3,11,135].

Goose eggs are placed on the trays and incubated horizontally, ie. lying on their sides[3,11,19,84,137], with the air chamber facing the fans in the incubator[3]. Turning is crucial for the goose eggs due to the egg size, as the chorioallantoic membrane needs to cover a large amount of nutrients necessary for the goose embryo development[3]. In old incubators the long axis of the goose egg is parallel with the tray at setting (see Bogenfurst[3,11] or Salamon and Kent[84]) and for this reason additional manual turning is needed apart from the hourly automatic 90° turning[3,11]. Generally this means manual turning by 180° once a day between day 5 and 10 of incubation and twice a day between day 11 and 20 of incubation[3,11].

Bogenfurst[3,11] solved this problem with his new setting technique, as the goose eggs are placed on the setting trays horizontally but the long axis of the goose eggs is in a 45-60° angle relative to the setting tray (same technique was also used by Milojevic[137]). This way the eggs are more stable, more eggs fit on the trays and additional manual turning is not needed, apart from the automatic turning through 90° (45° from either side of the vertical) every two hours[3,11]. Milojevic[137] achieved 89.77% hatchability of fertile goose eggs with this technique.

Salamon and Kent[84] conducted a study testing the above technique but also comparing two methods of additional manual turning (ie. turning by hand 180°) in the experiments. However, Salamon and Kent[84] encountered some challenges to replicate the technique used by Bogenfurst[3,11]. The average sized goose eggs (175-190 g) laid by adult geese (2-5 years old) could not be set in the 45-60° angle relative to the setting tray, only in a 20° angle at best but mainly the long axis of the goose eggs was parallel to the setting tray[84], which was not satisfactory for proper turning according to Bogenfurst[3,11]. Therefore Salamon and Kent[84] used the eggs of one year old geese that tend to be smaller and lighter than the eggs of adult geese[26,41,42,60,138]. Salamon and Kent[84] found that additional manual turning (apart from the automatic hourly 90° turning) once a day between day 10 and 26 of incubation increased the hatchability of goose eggs by around 17% (eggs without additional manual turning had 44.12% hatchability). Thus simply setting the goose eggs in the 45-60° angle relative to the long axis of the setting tray does not provide the turning requirements of goose eggs and additional manual turning is necessary for better hatchability[84] in contrast to the suggestion of Bogenfurst[3,11]. It has to be noted that the additional manual turning has to be done over and back (ie. one day to one direction, the following day to the opposite direction), not over and over, as turning in one direction could cause the rupture of the blood vessels or the yolk sac resulting in embryo death[11]. Interestingly, the technique of additional manual turning did not matter, as no statistical difference was found in hatchability between goose eggs turned along the long axis and along the short axis (63.77% vs. 61.94%, respectively)[84]. However, from a practical point of view Salamon and Kent[84] noted that turning along the long axis is faster based on their experience but turning along the short axis ensures a full 180° turn.

The hatchability results of the above studies are contradictory and it may not be necessary to use additional manual turning apart from the automatic turning of the incubators. However, it is not possible to determine the reasons for the differences in hatchability between the above studies, as the exact details of storage and incubation parameters were not presented. Thus further investigation is needed to clarify the above findings.

Cooling: In natural incubation the broody goose leaves the nest for a certain period of time every day in the second half of the incubation period and the cooling in artificial incubation mimics this behavior by the broody goose[3]. The cooling is particularly important from day 15 of incubation when the embryo starts to produce heat due to its metabolism, thus the temperature of the egg is continuously higher than the temperature inside the incubator[3,11]. Cooling is normally applied once a day from day 5-8 of incubation (8-15 min), while from day 16 of incubation (20-30 min) eggs are cooled twice a day but hatchability may decrease by 20% if cooling is missed[3,11]. In incubators requiring manual egg turning the cooling is done during the manual egg turning[11]. The cooling requires fresh air with a temperature of 20°C and normally the trolleys are pulled out from the incubators to achieve the proper effect on the eggs[3,11]. The cooling has to last until the egg shell temperature of the goose eggs reach 29°C[3,11]. Then the goose eggs need to be sprayed with warm (40°C) water to compensate for water loss[3,11] but it is advised to mix some kind of disinfectant in the spraying water to prevent microbes entering through the pores, as the cooling effect of the spraying creates a vacuum in the egg[11]. After the cooling process the incubators need to heat up the eggs to the incubation temperature within 30-40 min, because the lower than desired incubation temperature increases the length of incubation, the frequency of malpositions and affect post hatch performance[3,11,114].

Candling (testing for fertility): It is practical to candle the goose eggs twice during incubation[3] but necessary at least once. Candling is important to remove eggs that are infertile or contain a dead embryo and it is also a good time to check embryo development[3,11]. There are some good photographic guides on goose embryo development[139,140].

The first candling of goose eggs can be done between day 6-10 of incubation[3,11,19,58,84,96,97,141]. This is the time when clear eggs showing no development, eggs with blood rings or other irregularities (rotten eggs, cracked eggs, eggs with moving air chamber) are removed[3,11]. Clear eggs should be cracked open and the germinal disc examined to determine true fertility[3,11,141,142]. The fertilized germinal disc of a goose egg is larger (compared to an unfertilized germinal disc) due to the division of cells and contains a visible white ring that surround a lighter appearing central area[3,11,141].

The second candling of goose eggs can be done on day 26-27 of incubation, when the eggs are transferred to the hatcher[3,11,58,96,97]; however it may or may not be done. The candling of goose eggs at this stage is difficult and needs a good eye. One sign of embryo death is if we can see inside the egg (due to the unused proteins) with the candling light at transferring to the hatcher. Further signs of embryo death are the lack of movement (that would be mainly visible in the air chamber) and no blood circulation[3,11].

Hygiene: In the hatchery the goal is to produce a healthy hatchling. Therefore the hatchery must follow a strict cleaning (removal of dirt) and disinfecting protocol to eliminate possible pathogen microorganisms that affect chick quality[3,143]. To achieve this it is important to follow the correct procedures: removing all visible debris manually, high pressure washing with foaming detergent, rinsing, allow drying and finally disinfecting[144,145].

In single stage incubators a thorough cleaning and disinfecting is possible after every batch[145-147], however an incubator room with single stage incubators is still a multi stage operation due to eggs originating from different flocks and embryos being at different stage of development[145]. In multi stage incubators cleaning and disinfection is more difficult due to the continuous running of the incubators where the growth of bacteria is uninterrupted[145,146]. As fumigating with formaldehyde cannot be done between 24 and 96 hours of embryo development[3,11,93,145,148], regular spray or mist disinfection and the removal of exploders or their debris is necessary in multi stage incubators[145,146]. For the floors, walls and incubators a universal cleaning agent should be used that is suitable for foaming enabling better adhesion and longer time for the chemical to work[11,145,146]. A thorough disinfection can be achieved by fogging in the incubator room, as this method allows the product to enter into the incubators through the air inlets and disinfect the machines at the same time[145].

In the hatcher rooms, chick rooms and washing rooms stronger chemicals are needed for cleaning[144,145] and any dirt in these areas should be removed when still wet[3]. It is advised to use an alkaline foaming detergent or a high viscosity alkaline gel for cleaning these rooms and then use a disinfectant with residual action for long lasting effect[144-146].

Equipment, trolleys, trays, baskets and crates also need to be cleaned with high pressure washing using an alkaline detergent to avoid transmission of infection from one batch of eggs or hatchlings to the other[3,11,145,146]. Then spraying with disinfectant or fumigation should be applied[3].

It is crucial that the cleaning detergents, gels or foams and the disinfectants are chemically compatible[143,144,147], ie. if the cleaning agent contains positively charged ions, then the disinfectant should not contain negatively charged ions[143]. It is known that phenols and cresols are not compatible with non-ionic surfactants and cationic ones like quaternary

ammonia[143]. Further it is also important that apart from being effective the chemicals used in the hatchery should be non-irritant and non-toxic[11,147]. Formaldehyde is a widely used chemical in hatcheries; however it is carcinogenic and toxic[3,11,145,146] posing a threat to both embryos and hatchery personnel[148]. Due to these reasons efforts have been made to find alternative chemicals that are effective but not affecting hatchability or personnel health[93]. Some of these are: quaternary ammonia compounds (QACs), multiple phenolic compounds, iodine compounds (iodophors), glutaraldehyde, chlorine, ozone, hydrogen peroxide (6% concentration), electrolyzed oxidizing water, sodium dichlorocyanurate and sodium perborate[11,93,94,149,150]. Interestingly, a recent study showed that spray application of probiotics in hatch cabinets can also be an alternative to formaldehyde fumigation with similar positive results[151].

Another important point is that only authorized personnel should be allowed into the hatchery[147], who must take shower, change clothes and use shoe/boot washers/dippers before entering into the premises[3,11,146]. Also, it is important in a modern hatchery that the hatchery manager creates good hygiene awareness among all personnel, provide training and ensure that procedures are fully and correctly implemented[147]. The tires of vehicles (transporting hatching eggs, day-old chicks, etc.) entering into the hatchery premises need to be sprayed with disinfectant too[3,145].

Ways for improving fertility and hatchability: The suggestion by Gumulka and Rozenboim[22,23] to use experienced (2 years or older) ganders with one year old geese is impractical in large scale production because the geese are kept for 4-5 years in production. In order to have experienced ganders for one year old geese a new breeding flock needs to be raised every year (or every second year) and these different age groups need to be managed on the same breeding farm. This is costly and requires a lot of housing. However, the use of experienced ganders may have an effect on the applied sex ratio in large scale production. As the experienced ganders produce higher quality semen[14,20] and the number of successful copulations is higher[21,22], a smaller number of ganders may be needed in the breeding flocks (for example with a sex ratio of 1:4-6). Further, over the years of production a gradually declining sex ratio may be applied (which is generally inevitable due to the mortality of females over the years of production, unless ganders are culled to maintain constant sex ratio). It is known that with age ganders become less fertile and uninterested in mating, while fertility of females decline and their mortality increases[3,19]. Therefore a starting sex ratio of 1 male to 4-6 females may be applied in the first laying season, which could change to 1 male to 3-4 females by the third or fourth laying season. This way less ganders may be needed in the start, which is economically beneficial, as birds in grand-parent or parent flocks are expensive and costly to keep.

To improve fertility further studies should be conducted with regards to artificial insemination and cryopreservation of gander semen. The artificial insemination of geese using frozen/thawed semen yielded around 60% fertility on average[36,37]; however higher fertility may be achieved with the improvement of cryopreservation methods and/or application of different cryoprotectants[37,38]. Further, it is not enough to achieve high fertility of goose eggs using frozen/thawed semen; the hatchability of those eggs should be high as well. There are also opportunities to improve the hatchability of goose eggs. The periodic warming during egg storage is relatively well studied in domestic fowl, however only a few studies were conducted using goose eggs with the application of different warming techniques[96,108]. Bogenfurst[108] warmed long stored goose eggs for 5 hours every five days on 37.8°C, while Kucharska-Gaca *et al.*[96] used goose eggs (stored for three days) that were warmed once or twice for 6 hours on 37.8°C. The hatchability was around 80% in both studies, however it has to be noted that the technique of Bogenfurst[108] was able to maintain this hatchability up to 17 days of storage. Thus further experiments are necessary to determine how often, how long, how many times and on what temperature should goose eggs be warmed during storage to achieve the best possible hatchability for short or long stored eggs.

The incubation of eggs in light offers another possibility to improve hatchability. Several studies incubating domestic fowl and duck eggs in light showed a positive effect on hatchability[152-156], though others did not find any difference between the hatchability of eggs incubated in dark or light[152,157]. Light exposure during incubation also improved chick and duckling quality[154,156], decreased stress and reduced fear responses[152,154,157]. Thus light exposure during incubation may have an important role in poultry welfare. Further, Archer[155] showed that white and red light had positive effects on hatchability, while eggs incubated in green light or dark had similar hatchability. Thus hatchability of eggs can be increased by incubating them in white or red light, or in a combination of both[153-156] that could also be applied in the goose industry after conducting some experiments (especially on the long term effects of light exposure during incubation with regards to the sensitivity of geese to photostimulation).

The use of probiotics in animal nutrition has been studied for long time[158-163]. Studies demonstrated several benefits of probiotics in poultry production, such as better growth and

production performance, better feed consumption and absorption of nutrients, improved immune function, decreased ammonia excretion, reduced inflammatory reactions and prevention of pathogen colonization through modification of the intestinal microbiota[158-163]. However, the application of probiotics could start at the hatchery, as shown by the study of Graham *et al.*[151]. It is known that hygiene is very important in a hatchery and thorough cleaning is followed by disinfection in the buildings and equipment but after disinfection probiotics could be used on the incubators[151] to create a 'safer' environment for the eggs during incubation. Eggs are also disinfected[3,11]; still harmful bacteria may grow on them in the favorable conditions during incubation. Eggs may be sprayed with probiotics during incubation to prevent the growth of harmful bacteria by competitive exclusion[158,160,163]. Same procedure could be applied on the eggs in the hatchers, where the birds are hatched and are more exposed to bacterial infection. Further, the day old chicks in the hatchery may be sprayed with probiotics too to ensure the early benefits of improved gut health and immunity[159,160,162,163].

CONCLUSION

Fertility and hatchability are two important components of reproductive performance. Both traits are sensitive to genetic and environmental influents. Ultimately fertility can be improved by artificial insemination if influencing the other factors (such as sex ratio, nutrition, temperature or light) did not work. However, there are still ways to improve artificial insemination by using fine-tuned methods and/or different cryoprotectants. Egg quality (including its treatment and storage) affect hatchability, however the setting and hatching parameters (temperature, relative humidity, ventilation, turning, cooling) also have major impact on the outcome of the incubation process. There seem to be no standard incubational parameters for geese that work for everyone, as seen from the wide range of settings used (Table 1) due to certain factors (e.g. genetics, climatical conditions, equipment) in different parts of the world with more or less success depending on the applied techniques. Still there are methods such as the periodic warming during egg storage or the application of probiotics in the hatchery that can positively influence hatchability, though further research is needed to determine the best possible way of using them.

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