

UDC 636.5:636.082:591.3

doi: 10.15389/agrobiology.2019.4.667eng

doi: 10.15389/agrobiology.2019.4.667rus

## REPRODUCTIVE FUNCTION IN HYBRID POULTRY. V. THE EFFECT OF EGG STORAGE PRIOR TO INCUBATION

(review)

Yu.I. ZABUDSKII

*Russian State Agrarian Correspondence University*, 1, ul. Fuchika, Balashikha, Moscow Province, 143900 Russia, e-mail zabudsky@hotmail.com

ORCID:

Zabudskii Yu.I. [orcid.org/0000-0003-1195-0266](https://orcid.org/0000-0003-1195-0266)

The author declares no conflict of interests

Received May 14, 2019

### Abstract

Storage of eggs causes the death of the blastoderm cells, including necrosis and apoptosis (S. Bloom et al., 1998). Depending on the storage conditions (duration, temperature, and air humidity), the physicochemical parameters of egg ingredients vary. Water and CO<sub>2</sub> move through the shell, leading to an increase in pH of the albumen and yolk, which changes the activity of enzymes, reduces bactericidal properties of albumen and yolk. Violation of amino acid composition and ratio is characteristic of the yolk, and lipid peroxidation is developing (M.N. Argunov et al., 2015). Destruction and increased permeability of the vitelline membrane and the internal eggshell membranes occur. The secondary sexual ratio shifts (M. McDonald, 1960; M. Tagirov, 2010; M. Boerjan, 2016). The efficiency of egg incubation decreases as well as chick yield (G. Fasenko, 1992; V. Christensen et al., 2001; K. Tona et al., 2003; P. Hristakieva, 2011; D. Terčič et al., 2016). Immunocompetence of chickens hatched from stored eggs lowers (M. Goliomytis et al., 2015). These hatchlings need elevated temperature of environment during the first weeks of life (S. Yalcin et al., 2014). Haugh units, albumen and yolk indexes, albumen pH, etc., being indicators of the stored egg quality (P. Tsarenko, 2015), vary depending on genotype, age and feeding, the stress of the parents, and the season. Each next day of storage following 2 days post egg laying needs an additional one-hour incubation period for compensation. The degree of embryo development in the freshly laid eggs is not the same in different bird species. The stage of embryogenesis depends on age, mother's type of use (for eggs, for meat, or for dual use), and the order of the egg in the cycle of egg laying. Eggs with embryos which reach the gastrula stage are less sensitive to storage (I. Reijrink et al., 2008). Reduced hatching can be partially leveled by different methods, including heating during storage which provides development to the specified stage of embryogenesis (I. Kosin, 1956; M. Petek et al, 2004; Y. Piestun et al, 2013; D. Nicholson et al., 2013; L. Dyadichkina et al., 2016). Their positive effect is limited by the egg quality, modes of storage and heating (temperature, frequency and duration of exposure), as well as incubation.

Keywords: poultry, egg hatching, storage, ontogenesis, quality assessment methods, preventive methods

The pre-incubation storage of eggs, which worsens not only the efficiency of their incubation, but also the quality of young stock [1-3], is one of the most significant factors limiting the efficiency of poultry reproduction. However, this technology is widely used at poultry enterprises providing the opportunity to limit the number of parental stock and thereby increase the profitability of production. In addition, it allows incubating large batches of eggs and, accordingly, obtaining chickens of the same age in quantities sufficient to implement the basic principle of commercial livestock: "All in — all out".

This paper describes the characteristics of the ontogeny of young stock of the main poultry species caused by pre-incubation storage during pre- and postnatal periods, as well as the prevention of negative consequences caused by pre-incubation storage.

The state of embryos in eggs laid by the hens. For ducks, tur-

keys and guinea fowls, embryos in laid eggs are mainly at development stages (DSs) 7-8 according to the classification of Eyal-Giladi et al. [4], for hens at the stage 10, for quails and geese at the stage 11 [5, 6]. The selection of poultry based on productivity characteristics caused the heterogeneity of ontogeny in genotypes, and in particular, early embryogenesis. In turkey eggs, which were selected to increase live weight gain, embryos are more likely to be at the early gastrula stage than their brothers from the divergent line [7]. In 3-4 days after egg laying by broiler hens of lines A and B, which differ in fertility and hatchability, the DS of the embryos was 10.3 and 10.7 [8], respectively, and after 7 days of incubation, it was 28.7 and 29.5 according to the classification of Hamburger et al. [9]. The area pellucida and area opaca have already formed in chicken embryos at the 10th DS, only the first signs of a hypoblast were distinguishable [5].

The condition of the embryos is also determined by the age of the mothers and the order of the egg in the oviposition cycle. The area of the blastoderm in fresh unincubated eggs expands in proportion to the aging of hens [10]. In the majority of eggs of 32- and 63-week-old broiler hens, DSs 11 and 12 are observed in embryos, respectively [11]. In the eggs laid in the first days of the cycle, the embryos are at the 10.36 DS, while in the eggs laid in the next days – at the 10.05 DS, respectively [12].

The conditions of hatching eggs with embryos at different stages of development are not the same. The hatchability of hen eggs with embryos that have not reached gastrula (<10 DS) at the time of laying is less than 55%, but if this stage has already formed (DS 12-13), then the number of chickens obtained is significantly increased [13, 14]. Based on this pattern, Fassenko et al. [15] formulated the hypothesis that eggs with embryos at the DS 12-13 have better keeping capacity in comparison with eggs with less developed embryos. Based on this hypothesis, a method has been developed to prevent the decrease in the value of stored eggs by heating them in an incubator (egg heating, EH), which will be considered below.

The laid eggs tend to be in an environment with a temperature lower than in the female body. Under the influence of hypothermia, embryogenesis ceases and the facultative diapause begins [16]. In hen eggs stored for 0, 4, and 21 days, such diapause was recorded at 14 °C, and embryos were assigned to the DSs 9.9, 10.0, and 9.9, respectively [17]. However, at ambient temperatures above the diapause threshold, genesis continues. The embryos in the hen eggs that were present in the nests for 1.5 hours (28.1 °C) were at the 10.4 DS, and within 6.5 hours (30.4 °C) they reached the 11.7 DS [18]. The presence of eggs from 59-week-old hens in the nest at 30 °C or 20 °C caused a 2.4% decrease in hatchability in the first compared to the second. The same patterns for eggs from 37-week-old hens were not found.

The blastoderm in laid hen eggs consists of 40-60 thousand cells, some of which die over time [4]. For example, in turkey eggs stored for 0 and 14 days (18 °C), the number of such cells decreased due to necrosis and apoptosis from 32 to 21 thousand, or by 34.4% [19]. The latter was found in 3.1% of blastoderm cells of eggs in newly laid hen eggs, and after 14 days of storage (12 °C) their share was already 13.9% [20]. In eggs stored for 14 days, the expression of a number of proapoptotic genes is enhanced compared with that in eggs stored for 4 days [21].

Thus, it can be stated that in the main species of poultry, the stage of embryogenesis at the time of egg-laying is different. The incubation properties of eggs with embryos at different stages of development are not the same. It is advisable to cool the laid eggs to ensure the stop of embryogenesis.

The effect of storage on the properties of hatching eggs. The

eggs are compressed during cooling, and water, carbon dioxide and air are transported through the shell to the outside. As a result, duck eggs stored for 8 and 15 days lose 0.53% and 0.78% of the weight, respectively [22], turkey eggs stored for 10 and 14 days lose 1.0% and 1.5% [23], hen and quail eggs stored for 10 and 20 days lose 1.8% and 3.5%; 1.9% and 4.7%, respectively [24, 25], while the size of the air chamber increases.

A decrease in the CO<sub>2</sub> content causes a rise in the protein pH from 7.6 in freshly laid eggs to 9.0 in eggs stored for 4 days, but afterward this indicator remains practically unchanged [14]. Along with alkalization, the viscosity of a dense protein decreases, which is due to the state of the ovomucin-lysozyme system formed by electrostatic bonds between its constituent molecules. The system has maximum stability at pH 7.0, and its disintegration occurs in the range of 9.0-9.5, probably due to alkaline hydrolysis [26]. The bactericidal activity of lysozyme in the protein of stored eggs decreases [27], the activity of other enzymes also changes. Due to the difference in osmotic pressure, protein water diffuses into the yolk, shifting the active reaction from pH 6.0-6.3 to pH 6.5-6.8 [14]. In proportion to the eggs aging, the protein content in the vitellin membrane changes [28], and its strength weakens [29]. Lower temperature slows down these processes, restricting the penetration of *Salmonella enteritidis* into the yolk [30].

The numerical values of a number of quality markers of stored eggs, including protein and yolk indices, as well as the number of Haugh Units (HU), are decreasing. So, in hen eggs that were stored for 14 days, a decrease was observed from 8.2% to 5.3%, from 45.5% to 42.8%, and from 79.7% to 62.2%, respectively [3]. The same occurs in pheasant eggs (a decrease from 2.4% to 1.8%; from 43.9% to 40.1%; from 83.0% to 76.5%), which correlates with a deviation in the young stock hatching (decrease from 66.7% to 41.6%) [31].

The higher the ambient temperature, the greater are abnormalities. In fresh hen eggs, HU reached 91.4, and in eggs stored for 10 days (5, 21 or 29 °C), this value decreased to 76.3, 53.7 or 40.6, respectively [32]. To a lesser extent, the egg condition is reduced due to storage at a 75-90% humidity [33].

During long-term storage in the deutoplasm, lipid peroxidation is recorded. In hens, after 21 days after laying the eggs, the amount of malondialdehyde in the yolk rises from 0.13 to 0.17 µmol/l [34]. At the same time, the antioxidant system is activated, which is expressed by an increase in the activity of catalase and glutathione peroxidase.

Thus, in the deutoplasm of stored eggs, the content of water and carbon dioxide decreases, protein and yolk are alkalized. The disintegration of the ovomucin-lysozyme system leads to the liquefaction of the dense protein. The ability of lysozyme to dissolve the wall of bacterial cells decreases, the activity of other enzymes changes. The lipid peroxidation is developing, the antioxidant defense system is activated. The properties of the vitellin membrane change. A change in the values of such quality markers of hatching eggs as the height of the air chamber, protein and yolk indices, HU and, ultimately, hatchability indicates a decrease in the value of eggs due to storage.

Embryogenesis in eggs incubated after storage. Embryogenesis after storage is slower than in freshly laid eggs. After the 42-hour incubation of eggs stored for 7 and 14 days, the lag was 5 and 12 hours, respectively, compared to freshly laid eggs [35]. Incubation until hatching 50% of the chickens was 16 hours longer in eggs stored for 18 days than in eggs stored for 3 days (502 and 486 hours, respectively) [2]. The prolongation was due to a delay in the beginning of stage I of the perinatal period and its lengthening, but not of stage II. Apparently, this is due to a delay in the increase in the blood concentration

of corticosterone which is necessary to increase the ratio of triiodothyronine and thyroxine involved in the regulation of hatching.

In general, the result of egg aging is retardation of the development of not only embryos, but also of hatched young stock. In chickens from eggs that were stored for 14 days, compared to those hatched from eggs that were stored for 3 days, the length of the jejunum, as well as the width and area of the villi in it, reduced [36]. During the first 7 days of growth, the bodyweight of chickens increased by 76% in case of 3-day pre-incubation storage of the eggs and by 64% in the case of 18-day storage [37].

The dynamics of the performance of eggs incubation varies depending on the duration of their storage. Thus, the young quail hatching averaged 86%, 88%, 84% and 82%, respectively, for the eggs after 1-, 3-, 5- and 7-day storage [38], and chicken hatching averaged 88%, 89%, 92% and 82% after 1-, 3-, 6- and 15-day storage [39]. The data presented indicate that as a result of short-term storage of eggs of both species (3 days for quails and 6 days for hens), the desired indicator increases. With an increase in the storage periods of eggs from 5 to 30 days, embryonic mortality during incubation increases in hens and ducks from 2.0% and 14.3% to 100%, respectively [16]. The presented data confirm the objectivity of the conclusion of a number of researchers [33] that the placement of eggs for incubation a few days after laying has a beneficial effect on embryogenesis and incubation performance.

The phenomenon of a shift in the secondary sexual ratio due to egg storage has been established, the manifestation of which is determined by a number of conditions. McDonald [41] found the predominance of females (54.6%) in the White Leghorn hens obtained from the eggs that were stored for 7 days at 4.4 °C, whereas 15.5 °C or 26.6 °C did not cause such an effect. In Rhode Island Red chickens hatched from eggs that were stored for 15 and 21 days at 11.5 °C, the sex ratio was 1.85♂:1♀, while in eggs hatched after storage for 3 days — 1♂:1♀ [42]. Among the chickens of the Lohmann cross (Lohmann Tierzucht, Germany) hatched from eggs that were stored for 11 days and to which EH was applied, females dominated [43]. At the same time, in the chickens of the Slobozhansky cross (National Academy of Agricultural Sciences, Ukraine) [42], as well as in the chickens of the Prelux-G cross (University of Ljubljana, Department of Animal Science, Slovenia), hatched from eggs laid by 24- and 65-week-old hens, stored from 3 to 15 days at 15 °C and then heated (EH), a similar pattern was not observed.

Consequently, the storage of hatching eggs provokes changes in the pattern of ontogenesis in both embryonic and postembryonic periods.

The influence of the genotype, age and physiological state of parents on the resistance of embryos developing after pre-incubation storage of eggs. Published data indicate a variation in the resistance of hatching eggs of modern highly productive poultry genotypes to storage. In ISA-White cross (Institut de Sélection Animale, France) eggs stored for 10 days, the protein level decreased from 9.7 to 4.7 mm, in ISA-Brown cross eggs from 8.3 to 4.1 mm, while the egg protein pH values in these genotypes did not differ (7.4-9.3) [44]. The duration of incubation of Peking duck eggs after storage for 3 and 14 days was 27.9 and 28.2 days, respectively, of Muscovy duck eggs — 33.7 and 34.6 days [45], that is, this period increased by 1.07% and 2.67%, respectively. In meat and egg quails, the hatchability of eggs stored for 10 days reached 83% and 85%, respectively, whereas after 14-day storage this indicator reached 78% and 83% [46]. It is conspicuous that in the egg laying genotype, hatchability decreases less significantly with an increase in the storage period.

In the poultry genotypes with different egg resistance to storage, the me-

tabolism of the embryos and chickens varies. So, the glycogen concentration in the liver of 17-day-old embryos from the eggs pre-stored for 14 days and 1 day were 19.7 and 30.0 mg/g in the broiler hen line L<sup>+</sup> with higher egg resistance to storage vs. 23.5 and 24.8 mg/g in the line L<sup>-</sup> [47]. The hatchability of eggs after 14-day storage compared to 1-day storage decreased by 8% in L<sup>+</sup> and by 15% in L<sup>-</sup>.

Poultry breeding has been proven to improve the resistance of stored eggs to bacterial contamination. Jones et al. [48] used eggs from hens of freely mating lines 5, 7, and 10 the selection of which was stopped in 1950, 1959, and 1970, respectively, as well as from hens of a commercial herd genetically associated with these lines. The authors found that the surface shell contamination in fresh eggs from hens of all genotypes was the same, but, for example, eggs from the herd stock after inoculation with *Salmonella enteritidis* (SE) and *Pseudomonas fluorescens* (PF) were more contaminated on day 7 of storage, while the eggs from line 10 were less contaminated. In this case, salmonella prevailed on the shell (the SE and PF average counts were 2.7 and 0.8 log CFU/ml, respectively). Egg components, on the other hand, were more infected with pseudomonas. The SE and PF concentrations in the air chamber, shell, and deutoplasm of eggs from laying hens of older age groups exceeded this indicator in eggs from young hens. In general, eggs from line 10 were less contaminated.

Egg quality parameters change and storage resistance is getting worse in proportion to the age of the laying hens. In eggs laid by 32- and 59-week-old hens, the pH of the protein increased (8.1 and 8.3, respectively), and its height, on the contrary, decreased (7.7 and 6.3 mm) [49]. After 7 days of storage of eggs laid by 35- and 45-week-old hens, the HU decreased from 77% to 69%, hatchability from 88% to 85%, and the quality of chickens from 97% to 79% [50]. The physiological state of chickens hatched from eggs which were pre-stored for 10 days differed between 27- and 61-week-old laying hens fed a phosphorus-deficient diet [51]. In the experiment, young stock obtained from an older age group had lower ash content in bones, and rickets were more often recorded than in herd mates hatched from young laying hens.

Chickens from eggs incubated after 3 and 14 days of storage were raised under brooders at the optimum temperature (OT, 32-28 °C), at elevated temperature (ET, 34-30 °C) or low temperature (LT, 30-27 °C) [52]. On day 2 in the chickens hatched from eggs pre-stored for 3 days, the body temperature did not depend on the ambient temperature, but it turned out to be the lowest in those chickens that were obtained from eggs pre-stored for 14 days and kept at LT. For the first 7 days of growing, a greater increase in bodyweight was recorded at OT for young stock from the eggs after 3-day pre-storage and at ET after 14-day pre-storage.

Artificial molting of hens did not adversely affect eggs [53]. The hatchability and bodyweight of 7-day-old chickens from eggs that were stored for 8-18 days before incubation and were obtained from molted hens were higher than those for a batch of eggs from non-molting hens. However, it should be noted that the effect of storage on the quality of hatching eggs from hens subjected to artificial molting has not been studied enough.

Prevention of a decrease in incubation parameters of eggs during storage. There are several methods to prevent the effects of egg aging, which differ in the mechanism, for example, by the use of a controlled gas environment [54, 55] or by placing eggs with the sharp end up [56, 57] and applying a layer of propolis to the shell [58], etc. One of these methods, the EH, is considered the most effective. The experiments carried out on the main species of poultry prove its effectiveness (Table).

### Hatchability at single heating of eggs (EH) before storage

Poultry species	Age of laying hens, weeks	Conditions		Result	References
		storage	EH		
Broiler hens	32	11.5 °C; 60 %	37.50 °C; 6, 12 or 18 h	At St4, there is no effect; at St14 (for 6 h EH), hatchability is 79.0% (control — 70.5%)	[15]
	44	12.06 °C; 76 %	36.92 °C; 6 or 12 h	At St4 and St9 (for 6 h EH), hatchability is higher; at St14 (for 6 or 12 h), hatchability is lower	[66]
Turkeys	39-40	17.4 °C; 66 %	37.50 °C; 6 or 12 h	At St4, there is no effect; at St14 (for 12 h EH), hatchability is 75.3% (control — 70.6%)	[61]
Quail	20, 37	15 °C; 65 %	37.50 °C; 8 h	Hatchability in the experiment 82.6%, in the control — 70.6%; from 20-week-old laying hens — 84.7%, from 37-week-old laying hens — 77.6%. The differences between the variants St5 and St15 are minor	[62]
Guinea fowl	—	18 °C	37.50 °C; 3, 6 or 9 h	At St14 (for 6 h EH), hatchability is higher compared to the control	[67]

Note. For storage conditions, temperature and humidity are shown. St stands for pre-incubation storage of eggs (the number indicates the duration in days). Dash means that the age is not indicated [67].

The heating of freshly hatched eggs at 37.5-37.8 °C minimizes their quality deviation [59, 60]. It was established that the incubation indices correlate with storage and heating parameters. In particular, in chickens and turkeys, when the eggs were stored for 14 days before incubation, EH increased hatchability, which was not observed under 4-day storage [61]. A 6-hour heating was more favorable for the first group of eggs, while 12-hour heating for the second group.

The effectiveness of the EH depends on the age of the laying hens. In fresh quail eggs, which were processed once (8 h, 37.5 °C) and turned twice daily during storage [62], after 5 and 15 days of storage the hatchability due to EH improved by 4.4% for a batch of 20-week-old laying hens and only by 1.4% for 37-week-old laying hens, while the storage period did not affect the results of incubation. According to Gucbilmez et al. [63], the best result in batches of eggs from younger laying hens (27 and 29 weeks of age) is due to the fact that most embryos have not yet developed a hypoblast, and the EH ensures its formation, causing development to a stage more resistant to long-term aging [15].

EH effectiveness has a seasonality. Piestun et al. [64] in both the winter and summer periods used EH (at 30.2 °C for 12 hours) before 4- and 9-day storage of eggs. The hatchability of eggs in the control and experimental batches differed by 10% (84% and 94%, respectively) in the first case and only by 2% (85% and 87%) in the second one. In all cases, the bodyweight of 35-day-old young stock of both sexes increased, including due to the pectoral muscles.

In other experiment [65], eggs from 61- and 28-week-old hens were stored at 28 °C (mode 1) or 18 °C (mode 2). On the day of collection, EH was performed (37.8 °C for 6.0 and 4.5 hours for mode 1 and mode 2, respectively). As a result, variations were noted between the modes from 11.7 DS to 13.3 DS and from 9.0 DS to 12.6 DS, respectively. The hatching of chickens from eggs stored for 12 days for the mode 1 decreased compared to the control (74% and 80%, respectively), while in batches of eggs stored for 3, 5, and 8 days, the negative effect of EH on the analyzed parameter was not found. For eggs that were stored for 11 days, a positive result was observed for mode 2: hatchability in the experiment and control was 86% and 81%, respectively.

The effect of EH at the beginning of storage can change from positive to negative depending on the duration of the temperature exposure. Thus, in broiler hens, EH (6 h, 36.9 °C) of eggs of 4- and 9-day storage caused an increase in intestinal weight in 1-day-old chickens and did not affect the morphology of the mucous membrane of the duodenum [66]. However, a longer treatment (12 h) had a depressing effect on both the development of the mucosa and the live weight of the chickens. In guinea fowl, investigation of the effect of a single EH

at 37.5 °C for 3, 6, or 9 hours before storage revealed the advantage of a 6-hour exposure, namely an increase in the hatchability of eggs stored for 14 days [67].

Dymond et al. [68] performed 4-fold EH of hens, 4 h each at 37.5 °C, followed by storage for 4 and 21 days (the control was an intact batch stored for 21 days). EH caused an increase in the number of viable blastoderm cells. Hatchability was 92% and 84%, respectively vs. 71% in the control, and incubation time was 511 and 504 hours compared to 519 h in the control [68]. In turkey, EH at 37.8-38.0 °C for 5 hours on the 3rd and 5th day at a total storage period of 10 days, under the temperature and humidity regime differentiated with regard to requirements of each incubation period, reduced embryonic mortality, increased hatchability, shortened embryogenesis, and improved offspring rearing [69].

Thus, the development of embryos at egg-laying is not the same both in poultry of different species and within the same species [4-6], as well as in lines resulted from directional selection [7, 8]. The same was observed in embryos of the same genotype at different ages of mothers [10, 11] or depending on the order in the oviposition cycle [12]. Eggs with embryos at DS 12-13 were the best in storage parameters [15].

At ambient temperatures below physiological zero, embryogenesis does not occur in laid eggs. There is no consensus on the temperature range at which development ceases [10]. According to the reports, this range is 20-21 °C, 25-27 °C or 28-29 °C [70]. Apparently, such significant fluctuations are caused not only by the factors mentioned above, but also by differences in the thermal tolerance of genotypes, even with the same direction of use [71]. Eggs should be cooled after laying. Otherwise, the division of blastoderm cells will continue, but with an increase in the frequency of apoptosis, which will negatively affect the storage resistance. It is advisable to change the storage conditions depending on the intended duration. Thus, 19 °C and 70% humidity are optimal to store broiler hen eggs for 1-2 days, 12.5 °C and 90% are optimal for 13-16 days [72]; 15-18 °C are optimal for duck eggs under 1-3-day storage, 12-15 °C for 1-8-day storage and 8-12 °C for more than 8-day storage under 78-80% humidity [73].

As already noted, depending on the storage mode, the water reserves in the egg contents decrease due to evaporation through the shell, and the air chamber becomes more voluminous [23-25, 32], the protein and yolk are alkalized, the ovomucin-lysozyme system disintegrates, the lytic activity of lysozyme decreases, the activity of other enzymes, the concentration and ratio of amino acids, including essential amino acids, change [74], the properties of under-shell membranes and the vitelline membrane are violated [28, 29], deutero-plasm is contaminated with microflora [30, 75], yolk lipids undergo peroxidation [34]. The main markers of the incubation value of such eggs are protein pH, protein and yolk indices, HU, and the air chamber height [3, 24].

During the incubation of old eggs, the embryogenesis and growth of hatched chickens lag compared to fresh eggs. Under incubation of turkey eggs pre-stored for 5, 10 and 11-15 days, 8-day-old embryos reached developmental stages 28.6, 27.7 and 27.0 [9] with a weight of 505, 437 and 406 mg, respectively [76]. One of the reasons causing a decrease in the embryo weight may be the activation of blastoderm cell death due to apoptosis during storage [21]. On average, during each next day from the laying to the beginning of incubation, its time becomes 1 hour longer, and hatchability decreases by 1% [77]. The physiological state of individuals at different hatching periods is not the same [78]. In the eggs subjected to long-term storage, a lower cardiovascular function occurs during the final period of embryogenesis, which is caused by the depletion of the energy reserves of the body, as evidenced by the activation of gluconeogenesis [47]. These circumstances must be taken into account, especially when breeding

highly productive broiler hens predisposed to the development of ascites syndrome due to their inherent chronic heart failure which is formed in embryogenesis [80].

Chickens hatched from stored eggs require elevated ambient temperature during the first week of active life [52]. Such young animals have reduced immunocompetence [81]. Considering the well-known role of lysine in the formation of immunodeficiency [82], it can be assumed that at least one of the reasons for the latter may be a decrease in the content of this essential amino acid in the protein of stored eggs [74].

The phenomenon of better incubation of eggs after short-term storage (3-6 days) compared to fresh eggs and eggs stored for a longer time is conspicuous [33, 38, 39]. It is likely that the increased hatching from the first ones is due to the fact that in them, unlike the second ones, the processes occurred before incubators are identical to those in the eggs during the nest formation in natural conditions. A characteristic feature of breeding individuals of this group, which includes most poultry species, is that during this period parents spend most of their time outside the nest. For example, ducks visit the nest only 1-2 times a day [16]. During this period, a pH gradient forms between the dorsal and basal sides of the blastoderm [40], the activity of enzymes changes, the viscosity of the protein decreases, and prerequisites are created for the formation of the embryonic fluid. During artificial incubation of fresh eggs, such a lag period is absent and the corresponding physicochemical processes ensuring the preparation of eggs for embryogenesis do not have time to occur. However, this period is short in terms of changes in the deuterooplasm that can occur under prolonged storage thus reducing quality of the hatching eggs.

With increasing age of the laying hens, the egg resistance to storage decreases [50, 51]. According to Damaziak et al. [83], the hatchability of stored eggs from 70- and 73-week-old broiler hens, as well as the quality of the obtained chickens are worse than for 49- and 52-week-old hens. Eggs from young hens can withstand longer storage at high temperature and low humidity than eggs of laying hens at the end of the productive period [84]. Embryos in stored eggs laid by hens of different ages differ in sensitivity to fluctuations in ambient temperature. There is a significant change in the embryogenesis in eggs from aging hens due to fluctuations in air temperature [85]. The authors simulated opening a warehouse door, where eggs were stored for 7 days (in poultry farms this usually happens at 09<sup>00</sup>, 13<sup>00</sup> and 17<sup>00</sup> for the scheduled movement of batches of eggs), causing temperature fluctuations in the range of 18-21 °C for 40 minutes. As a result, there was a decrease in early embryonic mortality in the offspring of 27-week-old hens and, conversely, its increase in the offspring of 50-week-old laying hens compared to eggs stored at a constant temperature.

The growth rate of modern poultry genotypes derived through directional selection based on productivity traits varies, including at the stage of early embryogenesis. L<sup>+</sup> line turkeys, which were selected to increase bodyweight gain, lay eggs with less developed embryos than females of the L<sup>-</sup> [7]. It is known that embryos that have not reached the gastrula stage are less resistant to storage [14]. This proves the need for individualization of both storage and EH modes for almost every cross. In particular, when studying the effects of the genotype, maternal age, and duration of storage on the egg condition [86], a higher HU value was established in stored eggs from Hy-Line Brown cross hens (Hy-Line International, USA) with brown shells compared to this in eggs from the Bovan Brown cross (Hendrix Genetics, Netherlands) with the same shell color and the crosses DeKalb White (Decalb Poultry Research, USA) and Hy-Line W36 with white egg shells. The dry matter content in the deuterooplasm, the elasticity of the



vitellin membrane and its resistance to deformation also differed.

Considering the different needs of industrial poultry farming in males and females, the data on the displacement of the secondary sexual ratio compared to the standard (1:1) for different egg storage conditions is worth noticing [42, 43], especially since a shift in this ratio caused by the action of a certain temperature regime during egg incubation, fixed at the genetic level and transmitted to the next generation, was established [87].

The influence of EH as one of the most effective ways of preventing the decline in the egg quality during storage [61-69] has been tested on the main species of poultry. EH at a temperature of 37.5 °C should be carried out several days after laying, with repeating the procedure at 6-7-day intervals, while the total time of EH should not exceed 15 hours at a temperature above 32 °C [88]. Note that thermal effects on the egg occurs not only during pre-incubation storage (in the form of EH), but also during incubation through periodic changes in the temperature regime in order to train the hypothalamic-pituitary-cortico-adrenal system of embryos and to stimulate their adaptive capabilities [89-93]. These two methods are based on the use of thermal contrasts, which are similar to temperature fluctuations in natural conditions, during egg laying and hatching. EH during storage, as well as thermal training during artificial incubation, can optimize reproduction due to simulation of the processes that occur during the reproduction of birds in the natural habitat. At the same time, it is necessary to consider the unequal requirements of modern highly productive bird genotypes to environmental conditions. For example, developing embryos of the Ross 708 broiler cross (Aviagen, UK) are more sensitive to temperature conditions than the embryos of another Ross 308 broiler cross [94].

So, during the storage of eggs, changes in the physicochemical properties of the deutoplasm, vitellin membrane and shell membranes occur, the effectiveness of the protection against microflora and incubation efficiency become worse, and the conditions of the obtained young birds deteriorate. The development of embryos and the growth of chickens hatched from such eggs are slowed down. Methods have been developed to control the quality of stored eggs. In order to compensate for the decrease in the quality of hatching eggs, it is advisable to perform their heating during storage.

## REFERENCES

1. Fasenko G.M. Egg storage and the embryo. *Poultry Science*, 2007, 86(5): 1020-1024 (doi: 10.1093/ps/86.5.1020).
2. Tona K., Malheiros R.D., Bamelis F., Careghi C., Bruggeman V., Moraes V.M., Onagbesan O., Decuyper E. Effects of storage time on incubating egg gas pressure, thyroid hormones and corticosterone levels in embryos, and their hatching parameters. *Poultry Science*, 2003, 82(5): 840-845 (doi: 10.1093/ps/82.5.840).
3. Tsarenko P.P. *Ptitsevodstvo*, 2010, 4: 45-47 (in Russ.).
4. Eyal-Giladi H., Kochav S. From cleavage to primitive streak formation: a complementary normal table and a new look at the first stages of development of the chick. I. General morphology. *Developmental Biology*, 1976, 49(2): 321-337 (doi: 10.1016/0012-1606(76)90178-0).
5. Bakst M.R., Gupta S.K., Akuffo V. Comparative development of the turkey and chicken embryo from cleavage through hypoblast formation. *Poultry Science*, 1997, 76(1): 83-90 (doi: 10.1093/ps/76.1.83).
6. Sellier N., Brillard J.-P., Dupuy V., Bakst M.R. Comparative staging of embryo development in chicken, turkey, duck, goose, guinea fowl, and Japanese quail assessed from five hours after fertilization through seventy-two hours of incubation. *Journal of Applied Poultry Research*, 2006, 15(2): 219-228 (doi: 10.1093/japr/15.2.219).
7. Arora K.L., Kosin I.L. Changes in the gross morphological appearance of chicken and turkey blastoderms during preincubation storage. *Poultry Science*. 1966, 45(4): 819-825 (doi: 10.3382/ps.0450819).
8. Bakst M.R., Akuffo V., Nicholson D., French N. Comparison of blastoderm traits from 2 lines of broilers before and after egg storage and incubation. *Poultry Science*, 2012, 91(10): 2645-2648

- (doi: 10.3382/ps.2011-02118).
9. Hamburger V., Hamilton H.L. A series of normal stages in the development of the chick embryo. *Journal of Morphology*, 1951, 88(1): 49-92 (doi: 10.1002/jmor.1050880104).
  10. Meijerhof R. Pre-incubation holding of hatching eggs. *World's Poultry Science Journal*, 1992, 48(1): 57-68 (doi: 10.1079/WPS19920006).
  11. Pokhrel N., Ben-Tal Cohen E., Genin O., Sela-Donenfeld D., Cinnamon Y. Cellular and morphological characterization of blastoderms from freshly laid broiler eggs. *Poultry Science*, 2017, 96(12): 4399-4408 (doi: 10.3382/ps/pex242).
  12. Fasenko G.M., Hardin R.T., Robinson F.E., Wilson J.L. Relationship of hen age and egg sequence position with fertility, hatchability, viability, and preincubation embryonic development in broiler breeders. *Poultry Science*, 1992, 71(8): 1374-1383 (doi: 10.3382/ps.0711374).
  13. Steinke L. Keimscheibeuntersuchungen an hühnereiern unter besonderer berücksichtigung des entwicklungszustandes. *Archiv für Geflügelkunde*, 1972, 36(1): 5-10.
  14. Reijrink I.A.M., Meijerhof R., Kemp B., van Den Brand H. The chicken embryo and its micro environment during egg storage and early incubation. *World's Poultry Science Journal*, 2008, 64(4): 581-598 (doi: 10.1017/S0043933908000214).
  15. Fasenko G.M., Robinson F.E., Whelan A.L., Kremeniuk K.M., Walker J.A. Prestorage incubation of long-term stored broiler breeder eggs. 1. Effects on hatchability. *Poultry Science*, 2001, 80(10): 1406-1411 (doi: 10.1093/ps/80.10.1406).
  16. Bolotnikov A.M., Shurakov A.I., Kamenskii Yu.N., Dobrinskii L.N. *Ekologiya rannego ontogeneza ptits* [Ecology of early ontogenesis in birds]. Sverdlovsk, 1985 (in Russ.).
  17. Fasenko G.M., Robinson F.E., Hardin R.T. Research note: variability in preincubation embryonic development in domestic fowl. 2. Effect of duration of egg storage period. *Poultry Science*, 1992, 71(12): 2129-2132 (doi: 10.3382/ps.0712129).
  18. Fasenko G.M., Wilson J.L., Robinson F.E., Hardin R.T. Effects of length of egg nest holding time and high environmental temperatures on pre-storage embryonic development, survival, and hatchability of broiler breeders. *Journal of Applied Poultry Research*, 1999, 8(4): 488-492 (doi: 10.1093/japr/8.4.488).
  19. Bakst M.R., Akuffo V. Impact of egg storage on embryonic development. *Avian and Poultry Biology Reviews*, 2002, 13(7): 125-131 (doi: 10.3184/147020602783698520).
  20. Bloom S.E., Muscarella D.E., Lee M.Y., Rachlinski M. Cell death in the avian blastoderm: resistance to stress-induced apoptosis and expression of anti-apoptotic genes. *Cell Death and Differentiation*, 1998, 5(6): 529-538 (doi: 10.1038/sj.cdd.4400381).
  21. Hamidu J.A., Uddin Z., Li M., Fasenko G.M., Guan L.L., Barreda D.R. Broiler egg storage induces cell death and influences embryo quality. *Poultry Science*, 2011, 90(8): 1749-1757 (doi: 10.3382/ps.2011-0136).
  22. Dyadichkina L.F., Pozdnyakova N.S., Melekhina T.A., Goldin Yu.S., Yukhacheva N.A., Minaikhina V.A. *Ptitsa i ptitseproduktuy*, 2017, 2: 60-62 (in Russ.).
  23. Hristakieva P., Lalev M., Oblakova M., Mincheva N., Ivanova I. Effect of storage duration on the quality of hatching turkey eggs. *Archiva Zootechnica*, 2011, 14(3): 57-65.
  24. Tsarenko P.P., Kuleshova L.A. *Izvestiya Sankt-Peterburgskogo gosudarstvennogo agrarnogo universiteta*, 2015, 40: 112-117 (in Russ.).
  25. Tsarenko P.P., Vasil'eva L.T. *Ptitsevodstvo*, 2016, 11: 29-34 (in Russ.).
  26. Belitz H.-D., Grosch W., Schieberle P. Eggs. In: *Food chemistry*. Springer, Berlin, Heidelberg, 2009: 546-562 (doi: 10.1007/978-3-540-69934-7\_12).
  27. Baron F., Nau F., Guerin-Dubiard C., Bonnassie S., Gautier M., Andrews S.C., Jan S. Egg white versus *Salmonella Enteritidis*! A harsh medium meets a resilient pathogen. *Food Microbiology*, 2016, 53(B): 82-93 (doi: 10.1016/j.fm.2015.09.009).
  28. Mori M., Masuda N. Proteins of the vitelline membrane of quail (*Coturnix coturnix japonica*) eggs. *Poultry Science*, 1993, 72(8): 1566-1572 (doi: 10.3382/ps.0721566).
  29. Brake J., Walsh T.J., Benton C.E., Jr., Petite J.N., Meijerhof R., Penalva G. Egg handling and storage. *Poultry Science*, 1997, 76(1): 144-151 (doi: 10.1093/ps/76.1.144).
  30. Chen J., Thesmar S.H., Kerr W.L. Outgrowth of *Salmonellae* and the physical property of albumen and vitelline membrane as influenced by egg storage conditions. *Journal of Food Protection*, 2005, 68(12): 2553-2558 (doi: 10.4315/0362-028X-68.12.2553).
  31. Demirel Ş., Kırıkçı K. Effect of different egg storage times on some egg quality characteristics and hatchability of pheasants (*Phasianus colchicus*). *Poultry Science*, 2009, 88(2): 440-444 (doi: 10.3382/ps.2008-00131).
  32. Samli H.E., Agma A., Senkoğlu N. Effects of storage time and temperature on egg quality in old laying hens. *Journal of Applied Poultry Research*, 2005, 14(3): 548-553 (doi: 10.1093/japr/14.3.548).
  33. Christensen V. Factors associated with early embryonic mortality. *World's Poultry Science Journal*, 2001, 57(4): 359-372 (doi: 10.1079/WPS20010025).
  34. Argunov M.N., Stepanov V.A., Petunina K.V. *Vestnik Voronezhskogo gosudarstvennogo agrarnogo universiteta*, 2015, 4(47/2): 66-75 (in Russ.).
  35. Mather C.M., Laughlin K.F. Storage of hatching eggs: The effect on early embryonic devel-

- opment. *British Poultry Science*, 1977, 18(5): 597-603 (doi: 10.1080/00071667708416408).
36. Yalcin S., Gursel I., Bilgen G., Izzetoglu G.T., Horuluoglu B.H., Gucluer G. Egg storage duration and hatch window affect gene expression of nutrient transporters and intestine morphological parameters of early hatched broiler chicks. *Animal*, 2015, 1: 1-7 (doi: 10.1017/S175173111500261X).
  37. Tona K., Bamelis F., De Ketelaere B., Bruggeman V., Moraes V.M.B., Buyse J., Onagbesan O., Decuyper E. Effects of egg storage time on spread of hatch, chick quality, and chick juvenile growth. *Poultry Science*, 2003, 82(5): 736-774 (doi: 10.1093/ps/82.5.736).
  38. Petek M., Baspinar H., Ogan M., Balci F. Effects of egg weight and length of storage period on hatchability and subsequent laying performance of quail. *Turkish Journal of Veterinary and Animal Sciences*, 2005, 29(2): 537-542.
  39. Terčič D., Pestotnik M. Effects of flock age, pre-storage heating of eggs, egg position during storage and storage duration on hatchability parameters in layer parent stock. *Acta agriculturae Slovenica*, 2016, 5: 138-142.
  40. Stern C.D. The sub-embryonic fluid of the egg of the domestic fowl and its relationship to the early development of the embryo. In: *Avian incubation*. S.G. Tullett (ed.). Butterworth-Heinemann, London, 1991: 81-90.
  41. McDonald M.W. Effect of temperature of storage and age of fowl eggs on hatchability and sex ratio, growth, and viability of the chickens. *Australian Journal of Agricultural Research*, 1960, 11(4): 664-672 (doi: 10.1071/AR9600664).
  42. Tagirov M.T. *Biotechnologia*, 2010, 3(3): 84-91 (in Russ.).
  43. Boerjan M. *Pre-storage incubation and SPIDES: New procedures in hatching egg storage*. Pas Reform, 2014. Available <https://www.pasreform.com/en/knowledge/25/pre-storage-incubation-and-spides-new-procedures-in-hatching-egg-storage>. Accessed 24.07.2019.
  44. Silversides F.G., Scott T.A. Effect of storage and layer age on quality of eggs from two lines of hens. *Poultry Science*, 2001, 80(8): 1240-1245 (doi: 10.1093/ps/80.8.1240).
  45. Bagliacca M., Paci G., Marzoni M. Effect of egg weight categories, storage time and storage temperature on incubation length in duck eggs (*Cairina moschata* L. and *Anas platyrhynchos domestica* L.). *The Journal of Poultry Science*, 2005, 42(3): 205-214 (doi: 10.2141/jpsa.42.205).
  46. Romao J.M., Moraes T.G.V., Teixeira R.S.C., Cardoso W.M., Buxade C.C. Effect of egg storage length on hatchability and weight loss in incubation of egg and meat type Japanese quails. *Brazilian Journal of Poultry Science*, 2008, 10(3): 143-147 (doi: 10.1590/S1516-635X2008000300001).
  47. Christensen V.L., Wineland M.J., Fassenko G.M., Donaldson W.E. Egg storage effects on plasma glucose and supply and demand tissue glycogen concentrations of broiler embryos. *Poultry Science*, 2001, 80(12): 1729-1735 (doi: 10.1093/ps/80.12.1729).
  48. Jones D.R., Curtis P.A., Anderson K.E., Jones F.T. Microbial contamination in inoculated shell eggs: II. Effects of layer strain and egg storage. *Poultry Science*, 2004, 83(1): 95-100 (doi: 10.1093/ps/83.1.95).
  49. Lapro C., Gama L.T., Chaveiro Soares M. Effects of broiler breeder age and length of egg storage on albumen characteristics and hatchability. *Poultry Science*, 1999, 78(5): 640-645 (doi: 10.1093/ps/78.5.640).
  50. Tona K., Onagbesan O., De Ketelaere D., Decuyper E., Bruggeman V. Effects of age of broiler breeders and egg storage on egg quality, hatchability, chick quality, chick weight, and chick posthatch growth to forty-two days. *Journal of Applied Poultry Research*, 2004, 13(1): 10-18 (doi: 10.1093/japr/13.1.10).
  51. Shim M.Y., Pesti G.M., Bakalli R.I., Edwards H.M. Jr. The effect of breeder age and egg storage time on phosphorus utilization by broiler progeny fed a phosphorus deficiency diet with 1-OH vitamin D<sub>3</sub>. *Poultry Science*, 2008, 87(6): 1138-1145 (doi: 10.3382/ps.2007-00378).
  52. Yalcin S., Bilgen G., Gursel I. Effect of egg storage duration on brooding temperature requirements of broilers. *Proc. XIVth European Poultry Conference. Stavanger, Norway, June 23-27, 2014*. Stavanger, 2014: 608.
  53. Tona K., Bamelis F., De Ketelaere B., Bruggeman V., Decuyper E. Effect of induced molting on albumen quality, hatchability, and chick body weight from broiler breeders. *Poultry Science*, 2002, 81(3): 327-332 (doi: 10.1093/ps/81.3.327).
  54. Krivopishin I.P. *Ozon v promyshlennom pitsevodstve* [Ozone in industrial poultry farming]. Moscow, 1988 (in Russ.).
  55. Reijrink I.A.M., van Duijvendijk L.A.G., Meijerhof R., Kemp B., van den Brand H. Influence of air composition during egg storage on egg characteristics, embryonic development, hatchability, and chick quality. *Poultry Science*, 2010, 89(9): 1992-2000 (doi: 10.3382/ps.2009-00610).
  56. Elibol O., Brake J. Effect of egg position during three and fourteen days of storage and turning frequency during subsequent incubation on hatchability of broiler hatching eggs. *Poultry Science*, 2008, 87(6): 1237-1241 (doi: 10.3382/ps.2007-00469).
  57. Popova L.A., Komarchev A.S. *Pitsevodstvo*, 2014, 2: 10-13 (in Russ.).
  58. Akpınar G., Canogullari S., Baylan M., Alasahan S., Aygun A. The use of propolis extract for the storage of quail eggs. *Journal Applied Poultry Research*, 2015, 24(4): 427-435 (doi: 10.3382/japr/pfv043).

59. Kosin I.L. Studies on pre-incubation warming of chicken and turkey eggs. *Poultry Science*, 1956, 35(6): 1384-1392 (doi: 10.3382/ps.0351384).
60. Coleman J.W., Siegel P.B. Selection for body weight at eight weeks of age: 5. Embryonic stage at oviposition and its relationship to hatchability. *Poultry Science*, 1966, 45(5): 1008-1011 (doi: 10.3382/ps.0451008).
61. Fassenko G.M., Christensen V.L., Wineland M.J., Petitte J.N. Examining the effects of pre-storage incubation of turkey breeder eggs on embryonic development and hatchability of eggs stored for four or fourteen days. *Poultry Science*, 2001, 80(2): 132-138 (doi: 10.1093/ps/80.2.132).
62. Petek M., Dikmen S. The effects of pre-storage incubation of quail breeder eggs on hatchability and subsequent growth performance of progeny. *Animal Research*, 2004, 53(6): 527-534 (doi: 10.1051/animres:2004035).
63. Gucbilmez M., Ozlu S., Shiranjang R., Elibol O., Brake J. Effects of pre-incubation heating of broiler hatching eggs during storage, flock age, and length of storage period on hatchability. *Poultry Science*, 2013, 92(12): 3310-3313 (doi: 10.3382/ps.2013-03133).
64. Piestun Y., Druyan S., Brake J., Yahav S. Thermal treatments prior to and during the beginning of incubation affect phenotypic characteristics of broiler chickens post hatching. *Poultry Science*, 2013, 92(4): 882-889 (doi: 10.3382/ps.2012-02568).
65. Silva F.H.A., De Faria D.E., Torres K.A.A., De Faria Filho D.E., Coelho A.A.D., Savino V.J.M. Influence of egg pre-storage heating period and storage length on the digestive tract of newly-hatched broiler chicks. *Brazilian Journal of Poultry Science*, 2008, 10(1): 17-22 (doi: 10.1590/S1516-635X2008000100004).
66. Kgwatalala P.M., Faki O., Nsoso S.J. Influence of pre-storage incubation on the hatchability of guinea fowl eggs stored for fourteen days. *Journal of Animal Science Advances*, 2013, 3(6): 304-309 (doi: 10.5455/jasa.20130627052724).
67. Reijrink I.A.M., Meijerhof R., Kemp B., Graat E.A.M., van Den Brand H. Influence of pre storage incubation on embryonic development, hatchability, and chick quality. *Poultry Science*, 2009, 88(12): 2649-2660 (doi: 10.3382/ps.2008-00523).
68. Dymond J., Vinyard B., Nicholson A.D., French N.A., Bakst M.R. Short periods of incubation during egg storage increase hatchability and chick quality in long-stored broiler eggs. *Poultry Science*, 2013, 92(11): 2977-2987 (doi: 10.3382/ps.2012-02816).
69. Dyadichkina L.F., Pozdnyakova N.S., Melekhina T.A., Goldin Yu.S., Kolomeitsev V.A., Minaikhina V.A., Yukhacheva N.A. *Pitisevodstvo*, 2016, 9: 2-6 (in Russ.).
70. Decuyper E., Michels H. Incubation temperature as a management tool: a review. *World's Poultry Science Journal*, 1992, 48(1): 28-38 (doi: 10.1079/WPS19920004).
71. Zabudskii Yu.I., Kiselev L.Yu., Delyan A.S., Kamalov R.A., Golikova A.P., Fedoseeva N.A., Myshkina M.S. *Problemy biologii produktivnykh zhivotnykh*, 2012, 1: 5-16 (in Russ.).
72. *Rukovodstvo po inkubatsii Hubbard Balt Iza, 2013*. Available <http://www.isabalt.com/uploads/technical/2016/11/04/rukovodstvo-po-inkubatsii.pdf>. No date.
73. Fisinin V.I., Dyadichkina L.F., Goldin Yu.S. et al. *Tekhnologiya inkubatsii yaits sel'skokhozyaistvennoi ptitsy: metodicheskie nastaveniya* [The technology of poultry eggs incubation: instructions]. Sergiev Posad, 2011 (in Russ.).
74. Sarkisyan S., Abramyan V., Mkhchyan E. *Pitisevodstvo*, 2009, 4: 39-40 (in Russ.).
75. Gast R.K., Holt P.S. Influence of the level and location of contamination on the multiplication of *Salmonella enteritidis* at different storage temperatures in experimentally inoculated eggs. *Poultry Science*, 2000, 79(4): 559-563 (doi: 10.1093/ps/79.4.559).
76. Bakst M.R., Welch G.R., Camp M.J. Observations of turkey eggs stored up to 27 days and incubated for 8 days: embryo developmental stage and weight differences and the differentiation of fertilized from unfertilized germinal discs. *Poultry Science*, 2016, 95 (5): 1165-1172 (doi: 10.3382/ps/pew010).
77. Mailyan E. *Agrorynok*, 2009, 5: 16-18 (in Russ.).
78. Grikhina N.V., Galkina Yu.Yu. *Materialy Mezhdunarodnoi nauchno-prakticheskoi konferentsii «Problemy i perspektivy razvitiya nauki i obrazovaniya» (Kishinev, 06 iyunya 2017 goda)* [Proc. Int. Conf. «Problems and prospects for the development of science and education» (Kishinev, June 06, 2017)]. Kishinev, 2017: 62-66 (in Russ.).
79. Haque M.A., Pearson J.T., Hou P.-D.L., Tazawa H. Effects of pre-incubating egg storage on embryonic functions and growth. *Respiration Physiology*, 1996, 103(1): 89-98 (doi: 10.1016/0034-5687(95)00062-3).
80. Zabudskii Yu.I. Reproductive function in hybrid poultry. III. An impact of breeder flock age (review). *Sel'skokhozyaistvennaya biologiya [Agricultural Biology]*, 2015, 50(4): 444-457 (doi: 10.15389/agrobiologiya.2015.4.444eng).
81. Goliomytis M., Tsipouzian T., Hager-Theodorides A.L. Effects of egg storage on hatchability, chick quality, performance and immunocompetence parameters of broiler chickens. *Poultry Science*, 2015, 94(9): 2257-2265 (doi: 10.3382/ps/pev200).
82. Chen C., Sander J.E., Dale N.M. The effect of dietary lysine deficiency on the immune response to Newcastle disease vaccination in chickens. *Avian Diseases*, 2003, 47(4): 1346-1351 (doi:

- 10.1637/7008).
83. Damaziak K., Pawęska M., Gozdowski D., Niemiec J. Short periods of incubation, egg turning during storage and broiler breeder hens age for early development of embryos, hatching results, chicks quality and juvenile growth. *Poultry Science*, 2018, 97(9): 3264-3276 (doi: 10.3382/ps/pey163).
  84. Rocha J.S.R., Baiao N.C., Barbosa V.M., Pompeu M.A., Fernabdes M.N.S., Lara L.J.C., Matias C.F.O., Batista J.V.M.S.P. Negative effects of fertile storage on the egg and the embryo and suggested hatchery management to minimize such problems. *World's Poultry Science Journal*, 2013, 69(1): 35-44 (doi: 10.1017/S0043933913000044).
  85. Özlü S., Elibol O., Brake J. Effect of storage temperature fluctuation on embryonic development and mortality, and hatchability of broiler hatching eggs. *Poultry Science*, 2018, 97(11): 3878-3883 (doi: 10.3382/ps/pey253).
  86. Jones D.R.D., Karcher M., Regmi P., Robison C.O., Gast R.K. Hen genetic strain and extended cold storage influence on physical egg quality from cage-free aviary housing system. *Poultry Science*, 2018, 97(7): 2347-2355 (doi: 10.3382/ps/pex052).
  87. Yılmaz A., Tepeli C., Garip M., Çağlayan T. The effects of incubation temperature on the sex of Japanese quail chicks. *Poultry Science*, 2011, 90(10): 2402-2406 (doi: 10.3382/ps.2011-01471).
  88. Nicholson D., French N., Tullett S., van Lierde E., Jun G. Short periods of incubation during egg storage — SPIDES. *Lohmann Information*, 2013, 48(2): 51-61.
  89. Zabudskii Yu.I. Povyshenie adaptatsii broilerov k intensivnoi tekhnologii otkorma posredstvom okhlazhdeniya inkubiruemykh yaits s progressivno uvelichivayushcheysya ekspozitsiei. *Sel'skokhozyaistvennaya biologiya* [*Agricultural Biology*], 1993, 28(4): 69-74 (in Russ.).
  90. Zabudskii Yu.I., Shuvalova M.V. *Materialy XVII Mezhdunaranoi konferentsii Vsemirnoi nauchnoi assotsiatsii po pitsevodstvu (15-17 maya 2012 goda, Sergiev Posad)* [Proc. XVII Int. Conf. of the World's Poultry Science Association (May 15-17, 2012, Sergiev Posad)]. Sergiev Posad, 2012: 240-242 (in Russ.).
  91. Zabudskii Yu.I., Golikova A.P., Fedoseeva N.A. Heat training for prenatal period of ontogenesis as a method to increase the thermotolerance in poultry (review). *Sel'skokhozyaistvennaya biologiya* [*Agricultural Biology*], 2012, 47(4): 14-21 (doi: 10.15389/agrobiol.2012.4.14rus) (in Russ.).
  92. Artemov A.D. *Problemy biologii produktivnykh zhivotnykh*, 2014, 1: 5-20 (in Russ.).
  93. Zhuchkova N.A. *Vestnik Orlovskogo GAU*, 2017, 1(64): 81-85 (doi: 10.15217/48484) (in Russ.).
  94. Hamidu J.A., Torres C.A., Johnson-Dahl M.L., Korver D.R. Physiological response of broiler embryos to different incubator temperature profiles and maternal flock age during incubation. 1. Embryonic metabolism and day-old chick quality. *Poultry Science*, 2018, 97 (8): 2934-2946 (doi: 10.3382/ps/pey089).