



**Respiratory complex in broiler chickens: predisposing factors, causative agents, and aggravating conditions**

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Avian respiratory system is structurally different with mammalian pulmonary tract. The presence of choanal cleft, complete cartilaginous rings in trachea, compact structure of syrinx, rigid lungs without alveoli, distributed air sacs throughout the body with unidirectional airflow into the lungs and thinner blood-gas barrier with a large capacity for gas uptake are some of the important distinctions between birds and mammals which affects microbiome composition and function in broiler chickens. Respiratory microbiome in chickens primarily colonized in hatcheries and quickly change during the short life of broilers. Proteobacteria has been dominant bacteria in the respiratory system of chickens which significantly upraised during the last weeks of broilers age. The Respiratory virome of broilers has also same appearing and during their short life could experience drastic increase of respiratory infective viruses such as coronaviridae and also immune depressive viruses such as circoviridae and adenoviridae. These events will be extremely affected by housing, management and environmental conditions such as biosecurity level, stocking density, heat stress, litter quality, ventilation quantity, ammonia level, vaccination programs and antimicrobial prophylactic and therapeutic approaches.

Co-infection with multiple respiratory pathogens contributes to increased mortality rates in poultry flocks of Iran, Egypt, Algeria, India, Pakistan and some other developing countries which have somewhat similar poultry production systems and control and prevention policies against poultry diseases. In a complex system such as poultry respiratory tract, especially in field conditions, viral and bacterial co-infections could be either synergistic or in some cases even antagonistic. Interaction among different viruses and bacteria even interaction of live vaccines with viral and bacterial pathogens could alter the severity of respiratory complexes. In addition, immunosuppressive viruses like the Chicken anemia virus, Infectious bursal disease virus, adenoviruses, reovirus and also mycotoxins can predispose broiler respiratory tract to secondary infections.

Infectious bronchitis and avian influenza H9N2, as common respiratory diseases of Iranian broiler farms, occurs continuously with together. The positive interaction of these two viruses in increasing mortality of broiler chicken has been shown in both field and experimental studies. In addition, these viruses have pre, simultaneous and post interactions with bacterial (*E. coli*, MG, ORT, *Staphylococcus* ...) and other viral infections (ND, AMPV ...) and also with commonly used viral live vaccines. Previous studies have revealed that IB live vaccines could enhance the severity of H9N2 AIV infection. In contrast, different results were reported about Newcastle Disease vaccines on experimental AI infection (h9n2) in broiler chickens. *E. coli* is a common ubiquitous bacteria in the poultry farms and then colibacillosis as one of the most frequent respiratory disease of broilers



could aggregate consequent of IBV and LPAI infections. Timing of the *E. coli* co-infection has different consequences on the pathogenicity of LPAI and IBV in broiler chickens which could affect antimicrobial therapy strategies in broiler's respiratory complex infections.

The management of respiratory complex infections in broilers is a multifaceted and vigorous practice with many areas of argument and several uncertain questions. Some parts of these management processes are non-controversial such biosecurity procedures for inhibition of respiratory or immunosuppressive infections. Also, the improvement of housing, management and environmental conditions such as temperature, humidity and litter related conditions along with supportive treatments could decrease the consequences of respiratory complex infections. However, we are faced with some challengeable treatments such as ND emergency vaccination, preventive antimicrobial therapy and over-the-counter medications such as anti-inflammatory drugs or expectorants, mucolytic and mucomimetic compounds. Some of these management strategies will be discussed based on experimental studies which has been investigated in Avian Disease Research Center of Shiraz University during the recent years.

**Key words:** Respiratory Co-infections, broiler chickens, Respiratory microbiome, management strategies



## **The Role of Heterologous and Homologous Vaccines in the Control of Newcastle Disease**



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Despite all efforts such as intensive vaccination conducted to control Newcastle disease (ND), the disease still is a threat to the poultry sector throughout Asia, Eastern Europe, Africa, and Central America. To be an enzootic disease and the rapid spread of recently identified sub genotype VII viruses are considered as the factors involved in the current wide-spreading of the ND.

Recent studies on the viruses in the Razi Institute gene bank revealed that up to the late 1990s, the viruses responsible for the ND epizootics in Iran are genetically classified in a group that now is called genotype XIII. Since 2009, along with the increase of ND cases in Iran, the facilities for molecular identification and detection were also upgraded. The molecular diagnosis revealed that the ND viruses (NDVs) responsible for those outbreaks belonged to genotype VII and sub-genotype VIId, that currently named VII1.1 NDV. The VII 1.1 NDV is still the cause of ND incidence in many regions of the country. Therefore, during the last 30 years, Iran's poultry breeding industry has been challenged by NDVs being heterologous in comparison to the commonly used vaccines. At this time frame, the ND has been out broken in 3 to 5 years frequencies. Administration of multiple live vaccines alongside inactive vaccine regimes, as well as more intense biosecurity measures, has been successful in temporary control of the disease. In addition, the occurrence of other diseases such as LPAI and HPAI viruses, infectious bronchitis viruses (793B, QX, and variant serotypes), and recently IBH and HHS outbreaks, reduced the attention to ND control.

The effectiveness of live and inactive homologous vaccines, as one of the strategies to control the damages associated with circulating virulent VII genotype NDVs, are still controversial. Although the imported in active and the recombinant live vaccines of the VII genotype have been licensed to be used in Iran during the last year, no formal and specific results regarding their performance against virulent homologous viruses are available. Besides, results of studies conducted during the past decade demonstrate that the common heterologous vaccines provide significant protection against circulating viruses and reduce virus shedding in Iran. To the best our knowledge, different isolates belonging to genotype VII1.1 have not necessarily same phenotypic behaviors, and may have different performances when challenged the vaccinated birds.

**Keywords:** Genotype-matched Vaccines, Challenge Efficacy test, Newcastle Disease, Iran



### **Comparative evaluation of fertility indices in Arian broiler breeder males**

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### **Introduction**

The consumption of poultry products as a main source of human diet is increasing all over the world, so that it is predicted to take the first place among different livestock's in 2023. And to achieve this goal, the production of poultry meat should be constantly increased. One of the most important factors that limit the production of day old chickens is the decrease in the fertility of broiler breeder males. The fertility rate of roosters is different among breeds and is influenced by genetic, management, nutritional and diseases, and on the other hand with increasing age, the fertility rate decreases even in good conditions. The natural pattern of fertility during the production period of a broiler breeder flock is similar to the egg production chart, at the beginning of production (25 weeks) it is low and it reaches its maximum between 30-40 weeks and it starts to decrease gradually from 45-50 weeks. At around 60-70 weeks, it decreases to such an extent that keeping the flock is not economically viable. It seems that both sexes, hens and roosters, are involved in the reduction of fertility, but the influence of roosters is more in this matter. Also, it should be noted that a rooster mates with 10 chickens on average, which is doubles the importance of fertility of roosters. (Weil S.*et al.*, 2018) Since fertility and then the number of one-day-old chicks produced is the most important factor for the economic success of broiler breeder's flock. The present study investigates and evaluates the fertility characteristics of Arian broiler breeder males and compares it with Ross 308 breed.

### **Materials and methods**

This study was conducted to investigate the semen trait (Semen volume, Sperm motility, Sperm viability and Sperm count), histomorphometric features (TDI, SPI, SCI and MI) and anti-apoptotic gens (Bcl2, Caspase3&8) of Aryan and Ross308 broiler breeder males.

**Birds:** for this study, 60 roosters (10 of each breed, at 3 ages of 35, 45, and 55 weeks) with similar body conditions and weight were evaluated.

**Semen collection:** The roosters were trained to give semen 10 days before the collection began. Semen collection was performed by abdominal massage (Hafez, 1987).

**Sperm count:** Sperm count was determined using the standard hemocytometric method.



**Sperm motility:** Semen was diluted (1:200) in modified ringer solution and one drop of the diluted semen was placed on a slide, covered with a cover slide and then sperm motility was estimated by light microscopic observation at 400× magnification.

**Sperm viability:** Eosin-Nigrosin staining method was used for sperm viability. For each sample, 200 sperms were counted and their viability percentages were recorded.

**Histological analysis:** Left testicular tissue samples which stained with Hematoxylin and Eosin (H&E) were used for Histological evaluations (Talebi A. *et al.*, 2018).

**Tubule differentiation index (TDI) and spermiation index (SPI):** one hundreds cross-sections of seminiferous tubules were randomly analyzed per sample.

**Sertoli cell index (SCI) and mitotic index (MI).** Sixty seminiferous tubules per group were randomly examined for the calculation of SCI and MI.

**Anti-apoptotic genes expression in testicular tissue:** For evaluation of the process of apoptosis in the testicular tissue, the samples prepared from the right testicle were used for the expression of Bcl2, Caspase3 and Caspase8 genes by using the RT-PCR method (Shi, L. *et al.*, 2014).

## Results

There is no significant difference ( $P \geq 0.05$ ) in the number of sperms between Ross 308 and Arian breeds in the studied ages and it decreases with increasing age in both breeds.

The sperm motility rate at the age of 35 weeks in the Ross breed is higher than that of Arian, while at the age of 45 weeks it is almost equal and at the age of 55 weeks in the Arian breed it is more than that of the Ross.

The percentage of Sperm viability in the Ross breed at 35 weeks is significantly higher than Aryan and at the age of 45 weeks, it was relatively the same, but at the age of 55 weeks in the Arian breed it is more than Ross.

No significant differences were revealed between TDI and SPI indices between Aryan and Ross males in the studied ages.

Ross and Aryan roosters had similar Bcl-2 mRNA levels at 35 and 45 weeks, whereas the mRNA level of Aryan roosters was significantly diminished by 55 weeks compared to the Ross roosters.

At all ages, no statistically significant differences were observed in caspase-3 mRNA levels between Aryan and Ross breeds. Moreover, no significant change was revealed at 35 and 45 weeks for relative expression of Caspase-3/Bcl-2 between the Ross and Aryan roosters. In contrast, it was significantly decreased in Ross breed at week 55.

Aryan breed showed lower caspase-8 mRNA levels at 35 weeks, which increased at 45 and exhibited no significant difference with Ross roosters at week 55. However, about relative caspas-8/Bcl-2 expression, the Aryan breeds showed a significantly lower relative expression at week 35, which was remarkably higher versus Ross roosters after 45 and 55 weeks.

**In conclusion,** considering no significant changes in Ross and Aryan breeder males in germ cell differentiation and spermiogenesis (marked by TDI and SPI indices), the Ross and Aryan roosters do not exhibit a remarkable change in total sperm count in the studied ages. However, their ability to maintain the necessary sperm numbers for appropriate fertility reduces time dependently, mainly due to their age and the reduced potential of the testes in older weeks. On the other hand, we considered the pro-and-anti-apoptotic gene's sole and relative expression levels. Taking relative gene expression results into account, both roosters demonstrate similar performance at earlier weeks



(35 and 45 weeks), however, Ross roosters are able to keep this pattern longer than Aryan roosters following 55 weeks. Based on Caspase-8/Bcl-2 expression, we can conclude that Ross breeds exert better results at earlier weeks, while it decreases at older weeks. Moreover, since Ross males maintain Caspas-3/Bcl-2 expression at approximately half as low as Aryan breeds, we can suggest that they may have higher live sperms and better sperm motility at week 35. As caspase-3/Bcl-2 expression does not differ significantly in Ross and Aryan roosters at 45 weeks of age, no significant changes in sperm viability were observed, therefore no noticeable difference was observed in sperm motility when these roosters were compared. However, at the elder week of old (week 55), other factors such as testicular endocrine, antioxidant status, or other additional influencing factors may play an important role in Aryan males testicles, which they can exhibit more viable and motile sperms than Ross breeds. Because, both roosters exhibited almost the same number of sperm at week 55 while significantly higher viability and motility were revealed in the Aryan roosters.

Although there were no significant differences between the Arian and Ross 308 breeds in the parameters investigated in this study, but based on the information and results obtained from broiler breeder flocks raised in the country in recent years, the fertility rate and hatchability of Ross308 breed is much higher and better than the Arian breed, which highlights the need for more research in this field.

#### **References:**

1. Hafez, E.S.E. (1987) Reproduction in Farm Animals. 5th Edition, Lea and Febiger, Philadelphia, 315-481.
2. Talebi A, Alimehr M, Alavi MH, et al. Comparative study of semen traits and histomorphometric features of testes of broiler breeder males with different phenotypic traits. Vet Res Forum 2018; 9(1): 1-6.
3. Shi, L., H. Zhao, Y. Ren, X. Yao, R. Song & W. Yue, 2014. Effects of different levels of dietary selenium on the proliferation of spermatogonial stem cells and antioxidant status in testis of roosters. *Animal Reproduction Science*, 149(3-4), 266-272.
4. Weil S, Rozenboim I, Degen AA, et al. Fertility decline in aging roosters is related to increased testicular and plasma levels of estradiol. Gen Comp Endocrinol 1999; 115(1): 23-28.



**Effect of antioxidants on incidence of ascites syndrome and related parameters in broiler chickens**

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**Introduction**

Ascites syndrome (pulmonary hypertension syndrome) (PHS) is a major problem in broiler industry throughout the world. PHS is a metabolic defect associated with hypoxemia, heart/lung overload, venous and heart congestion, right ventricular hypertrophy, a flaccid heart, cirrhosis of the liver, and accumulation of ascitic fluid into the abdominal cavity. It is commonly accepted that PHS in fast-growing broilers is a direct effect of right atrioventricular valve insufficiency, ventricular volume overload, and right ventricular dilation and failure. Many studies have determined that different factors, such as altitude, cold, lighting, air quality, ventilation, high energy rations, and incubator environment, have all been implicated in the progressing of pulmonary hypertension. The crucial role of reactive oxygen species (ROS) in the pathogenesis of PHS has been demonstrated in the several studies. It has been confirmed that exposure to low temperatures could damage the cell antioxidant and protective systems. In addition Damage caused in the lung by lipid peroxidation events may result in oxygen insufficiency and pulmonary hypertension which would predispose the bird to heart damage and ascites. Lipid peroxidation events in heart muscle may also cause direct cellular damage and adversely affect cardiac function.

On the other hand, the change in the antioxidant status of the broiler during PHS progression is observed in conjunction with increased markers of reactive oxygen-mediated tissue injury, indicating a state of oxidative stress during this syndrome. Researchers have attempted to modulate the onset of PHS by increasing the antioxidant status of the broilers which had significantly reduced ascites-induced mortality.

**The role of some antioxidants on ascites reduction**

Antioxidants are classified as exogenous and endogenous compounds, both responsible for removal of free radicals, scavenging ROS, or their precursors, inhibiting formation of ROS and binding metal ions needed for catalysis of ROS generation. Endogenous antioxidant system (the first line of defense against reactive oxygen) is sorted in two major groups, enzymatic and non enzymatic. Enzymatic antioxidants are comprised of limited number of proteins such as catalase, glutathione peroxidase as well as superoxide dismutases along with some supporting enzymes. Non enzymatic antioxidants include selenium, ascorbic and lipoic acid, polyphenols, tocopherols and carotenoids, which are derived from dietary sources. The levels of these antioxidants are reduced in liver and lung tissues and the mitochondria of ascitic broilers. Under high stress conditions, the biological antioxidant system becomes inefficient, resulting in free radical formation exceeding the natural antioxidant capacity of the animal. Thus, there is an urgent demand for exogenous supplementation of antioxidants to counter the excess free radicals in the animal's body.



Organic Se with vitamin E supplementation decreased mortality and incidence of ascites syndrome in broilers reared under cold stress.

Broilers that received a vitamin E implant that released a total of 15 mg  $\alpha$ -tocopherol from 0 to 3 weeks of age immediately before exposure to ascites had significantly reduced ascites-induced mortality than placebo-treated broilers. Liver and lung concentrations of  $\alpha$ -tocopherol in healthy vitamin-E-treated birds were increased. Healthy vitamin E treated birds had plasma lipid peroxide values lower than placebo-treated birds in the same conditions, indicating the enhanced protection that vitamin E provides against lipid peroxidation. The vitamin E implant reduced ascites-induced mortality, probably by providing an enhanced antioxidant defence against the reactive species production that otherwise cause tissue damage and promotes ascites progression. In contrast to the results obtained with vitamin E implants, supplementing broiler diets with vitamin E did not reduce ascites-induced mortality.

The beneficial effects of vitamin C have already been confirmed in the chicken growth performance, antioxidant activity and pulmonary hypertensive response.

The synergistic effect of synthetic antioxidant blend of ethoxyquin, propyl gallate and butylated hydroxytoluene (BHT) improved the growth performance and reduced the mortality of broilers induced with ascites.

Bahadoran et al. (2021) concluded that supplementation of *Kelussia odoratissima* Mozaff alcoholic extract to broiler diet reduces ascites mortality and also improves chicken performance.

Furthermore, pomegranate peel which is naturally enriched with polyphenols had a significant reduction on mortality of ascites-induced broilers.

Melatonin is known as a scavenger of oxidants and an ameliorator of the antioxidant defense system and melatonin administration was considered to normalize the circadian rhythm of blood pressure, ameliorate ventricular function, and improve hypertension in patients with congestive heart failure.

Bahadoran et al. (2022) showed that oral supplementation of sage (*Salvia officinalis* L.) extracts could modulate pulmonary hypertensive response and decrease developmental hypertrophy and dilation of the heart and improved villus dimensions in pulmonary hypertensive broilers. These researchers showed an increase of total antioxidant and decrease of oxidant status in the hypertensive chickens fed sage, especially in concentration of 0.2%. Sage extract consists of rosmarinic and carnosic acids at high concentrations, which have antioxidant properties. Thus the strong effect of sage constituents in the oxidant and antioxidant status could justify its improvement in the developmental PHS and its other following advantages in the intestine.

## Conclusion

It is concluded that Oxidants are key factors in ascites syndrome pathogenesis and further research remains key to improving knowledge of antioxidants to prevent the PHS and reduce ascites mortality in broiler flocks.

## Reference

1. Baghbanzadeh, A and Decuypere, E (2008). Ascites syndrome in broilers: physiological and nutritional perspectives. *Avian Pathology*, 37: 117–126.
2. Bahadoran, S. Hasanpour, H and Shafiqh, Z (2016). Garlic supplement improves intestinal mucosa morphology in broiler chickens with developmental pulmonary hypertension. *Poultry Science Journal*, 4: 117-125.
3. Bahadoran, S. Teymouri, Y. Hassanpour, H. Mohebbi, A and Akbari, M. R (2022). Effect of sage (*Salvia officinalis* L.) extract in antioxidant status and intestinal morphology of pulmonary hypertensive chickens. *Veterinary Medicine and Science*, 1-9.





4. Bahadoran, S. Hassanpour, H. Arab, S. Abbasnia, S and Kiani, A (2021) Changes in the expression of cardiac genes responsive to thyroid hormones in the chickens with cold-induced pulmonary hypertension. *Poultry Science*, 100: 1-7.
5. Balog JM (2003). Ascites syndrome (pulmonary hypertension syndrome) in broiler chickens: are we seeing the light at the end of the tunnel? *Avian And Poultry Biology Reviews*, 14: 99-126
6. Hassanpour, H. Khalaji-Pirbalouty, V. Nasiri, L. Mohebbi, A and Bahadoran, S (2015). Oxidant and enzymatic antioxidant status (gene expression and activity) in the brain of chickens with cold-induced pulmonary hypertension. *International Journal of Biometeorology*, 59, 1615-1621.
7. Salamia, A.S. Majokaa, A.M. Sahaa, S. Garbera, A and Gabarroua, JF (2015) Efficacy of dietary antioxidants on broiler oxidative stress, performance and meat quality: science and market. *Avian Biology Research*, 8: 65-78.



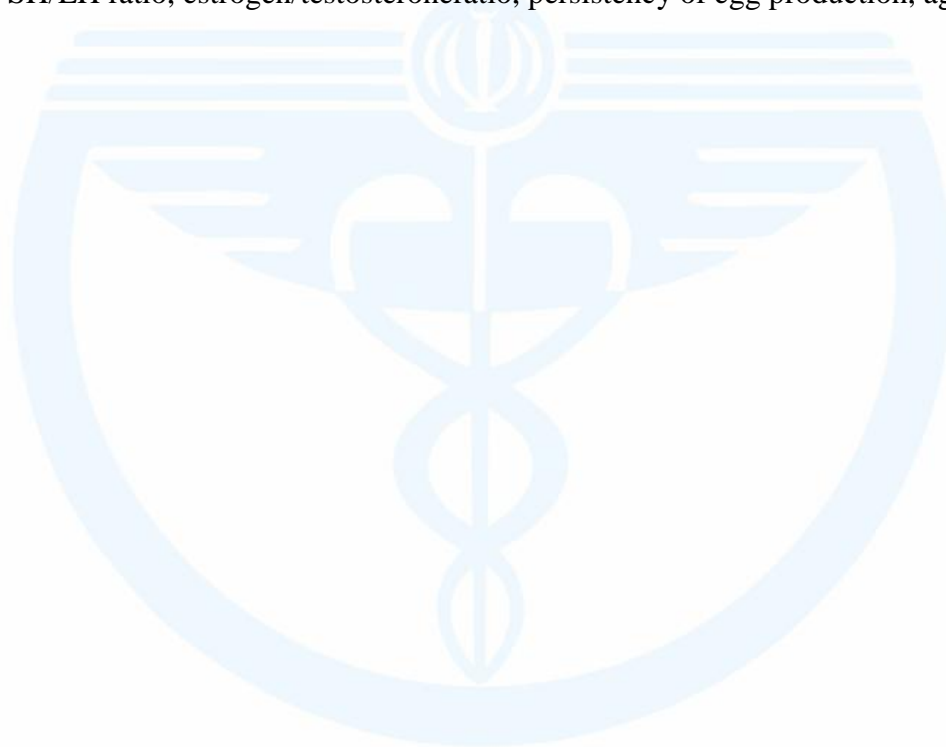
**Physiological aspects of persistency of egg production****Amjad Farzinpour***Department of Animal Science, University of Kurdistan, Sanandaj, Iran***Corresponding author's email: [Amjadfarzinpour@uok.ac.ir](mailto:Amjadfarzinpour@uok.ac.ir)**

**Abstract:** In the world, the number one priority is to increase the egg production by breeding for increased persistency in lay. It means, the long life of commercial egg production farms can be extended to 90-100 weeks to achieve 500 eggs during a production period. It has been described in the previous reports that to reach the goal of long-life layer, a multifactorial approach including genetic, nutrition and design of housing system. But it seems that one of the angels of the square of the long-life theory has been neglected and it is physiology aspect of aging in birds. To achieve this target, the main challenge lies in keeping the layer in a good body condition until the end of the flock. The ovary is a major internal organ of the female reproductive tract that produce the gametes. Over 12000 oocytes are present in the ovary of the one-day old chick after hatching but about 5000 of initial recruitment follicles which are followed by hierarchal follicles that progress toward maturity. Therefore, it can be said the ovary has a natural potential to release more gametes during the long life of layer. There are several studies that explained the age-related- disorders in layer, low percentage of egg production, follicular atresia, low rate of follicular development, decline in clutch size are the main concerns at late laying period. The process of reproductive senescence in female birds is characterized by a gradual transition from regular reproductive cycles to irregular cycles to eventual acyclicity and ultimately a low or loss of egg production at the end of production period. The causes of these disorders are summarized in two subjects; 1: decreased age-related progesterone, testosterone and LH, 2: increased estrogen that cause hormonal imbalance status which primarily in fluence reproductive organ function. During ageing, increased estrogen with negative feedback on gonadotropins and decrease in preovulatory LH surge results to reduction in hypothalamic response to gonadal steroids and consequently irregular egg laying. There is a key point about the estrogen hormone. Although estrogen plays an important role in the female reproductive performance but it practically acts like a double-edged sword. The optimum concentration of estrogen is necessary for a normal egg production rate, but a high concentration of estrogen will have several side effects. There is now significant evidence to emphasis that reproductive hormones undergo significant changes with ageing. It has been confirmed that the LH and FSH responses to GnRH are attenuated with ageing. According to several reported researches, it is concluded that hormonal imbalance for testosterone, estrogen, FSH and LH is just a major suspected etiologic factor in causing low production in aged birds. Several investigators confirmed that the estrogen/testosterone and FSH/LH ratio as the independent predictors of reproduction



performance in layer. The young birds or in peak of production that have no hormonal imbalances but the aged birds had the high imbalances in hormones during the late laying period. As age increased, average estradiol levels exhibited a linear increase and the elevated follicle-stimulating hormone/luteinizing hormone ratio is associated with low performance in aged breeder quails. Keeping the FSH/LH ratio constant at around 2.5 can be considered as a functional index for persistency of egg production in layer breeders.

**Keywords:**FSH/LH ratio, estrogen/testosteronerationo, persistency of egg production, ageing





### **Avian Mycoplasmosis: with emphasize effective medication**

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Mycoplasmosis is found in humans, animals, plants and insects. Mycoplasmosis in poultry is caused by Mycoplasma bacteria. These bacteria are transmitted vertically and horizontally. Diseases related to Avian Mycoplasmosis include: respiratory disease, synovitis, poor performance, skeletal problems and even death. Mycoplasma are very small prokaryotes with small genome and no cell wall. Mycoplasma is able to penetrate into the cells through the respiratory tract and cause systemic infections. They can spread through the blood in all the organs of the poultry body, including joints, bones, ovaries and other organs.

These bacteria are colonized on mucosal surfaces. Some species, such as Mycoplasma gallisepticum and Mycoplasma synoviae, have the ability to penetrate into cells. The most important pathogenic species in poultry are: Mycoplasma gallisepticum, Mycoplasma synoviae, Mycoplasma iowae, Mycoplasma meleagridis.

There are harmful economic effects of Mycoplasma gallisepticum in chickens and turkeys. This mycoplasma is the cause of chronic respiratory disease in chickens and turkeys, the cause of reduction in egg production, weight loss, increase in carcasses eliminated in the slaughterhouse, increase in food conversion ratio and high cost of treatment and control.

Considering the frequency of occurrence of Mycoplasma gallisepticum in Iran's poultry industry and its high economic importance, it is important to study and pay special attention to different aspects of this disease. In this study, while examining the control ways, we present effective drug programs in the control and treatment of this disease. In order to control and eradicate Mycoplasma gallisepticum infection, it is necessary to implement a biosecurity program and regular monitoring. Medication and vaccination with live attenuated or inactivated vaccines are intervention to mitigate clinical disease and production losses when maintaining MG-clean flocks is not considered feasible.

Mycoplasma gallisepticum, like other mycoplasmas, is resistant to beta-lactam antibiotics such as penicillin and cephalosporin due to the lack of a cell wall. Mycoplasma gallisepticum is sensitive to macrolides, pleuromotilins, tetracycline and fluoroquinolones. There is also antibiotic resistance in this bacterium, and it is necessary to provide a test of the minimum inhibitory concentration (MIC) of the antibiotic. Various antibiotics, including tylosin, tilmicosin, tylvalosin, tiamulin, valnemulin, oxytetracycline, chlortetracycline, enrofloxacin, danofloxacin, and lincomycin-spectinomycin have demonstrated efficacy for the treatment of MG respiratory diseases, reducing the severity of clinical signs and gross lesions, and lowering mortality and performance losses. Antibiotic treatment may



reduce MG populations in the respiratory tract, potentially reducing MG shedding and lowering the risk of horizontal transmission to neighboring flocks.

In the clinical studies conducted during 10 years, 18 positive MG broiler flocks were studied. Various medication programs were studied, and the ideal program was the combination of tiamulin and doxycycline. In this study, other antibiotics such as tilmicosin, tylosin tartrate, and lincospectin were also used, but the therapeutic effect was less compared to tiamulin and doxycycline. The results of treatment with different medication programs were evaluated by reducing losses, reducing clinical signs and necropsy, the amount of feed consumption, increasing growth, final weight, percentage of condemnation chickens and calculating the food conversion ratio (FCR).

In addition, considering the high costs of the medication program regarding the control of infection caused by MG, calculating the drug dose based on the live weight of the herd is less expensive and more accurate than prescribing the drug based on ppm. The most effective result in us regarding the drug program of tiamulin and doxycycline: 15mg/kg for 5 days for each two drugs.

However, successful antibiotic treatment can reduce disease severity, clinical signs and necropsy, and mortality caused by MG. But it will not lead to the eradication of the disease. In addition, the continuation of this method will lead to an increase in antibiotic resistance. In general, antibiotic treatment can be useful as a short-term method and under the condition of choosing an effective drug program.



**A comparative review on the control and prevention strategies of poultry mycoplasmas in the world**

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Avian mycoplasmosis, which is economically important in the poultry industry, is caused by four main pathogenic mycoplasmas, *M. gallisepticum*, *M. synoviae*, *M. meleagridis* and *M. iowae*. Many producers, especially integrations and companies that produce day-old chickens, spend a lot of time and money annually to monitor and control these microorganisms. Economic losses include weight gain reduction, decrease in feed conversion ratio, increase in cases of carcass condemnation in slaughterhouses, egg drop, fetal loss, and monitoring, prevention and treatment costs. The main complications of mycoplasma infections are chronic respiratory disease in chickens and infectious sinusitis in turkey caused by *Mycoplasma gallisepticum* and synovitis and air sacculitis caused by *Mycoplasma synoviae*. Considering the role of vertical and horizontal transmission, prevention and control measures for mycoplasmosis in birds include biosecurity, treatment and vaccination. In this review, different strategies to monitor and combat mycoplasmas in the poultry industry of different countries by using only the biosecurity or with the use of vaccines are compared. According to the results of various studies, it seems that in a country or region, preventing the occurrence of *Mycoplasma gallisepticum* infection in GGPs, GPs and breeders through the implementation of a comprehensive biosecurity program is achievable, while the control of *Mycoplasma synoviae* due to its specific characteristics can be possible by implementing biosecurity and using vaccines. Meanwhile, it seems that the control of mycoplasmas in the case of multiage commercial layers is more reasonable and achievable by using vaccines. However the use of the vaccine will overshadow the nature, level and cost of diagnostic tests due to the need to differentiate the vaccine strain from the field strain.



## **Spatiotemporal dependence of the repetition of diseases in broiler breeders**

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Broiler breeders` disease are important from various point of view such as spatiotemporal view. Some of them is important due to the losses caused in broiler breeder and the other due to the possibility of vertical transmission to the progeny and cause losses in them and the last group due to cause losses in both broiler breeders and their progeny with together. Biosecurity status, maternal antibodies and parent stock(PS) vaccination program, concurrent infections, age of poultry when exposed to infectious agents ,infectious agents and their pathogenicity, Immunosuppressive factors, nutritional condition of the flock and etc., have major roles in the infectious sequels. Apart from discussing the adequacy of vaccines and the route and management of implementing the vaccines and vaccination program, it should be known that some disease clinical signs occur repeatedly in a certain period of the broiler breeders` lifecycle. Getting to know the probability of the a geat which the disease occurs is more likely to give an appropriate and faster responses to the relevant possibility of the disease. The possible responses to the agents may include vaccination and a more sophisticated management of the poultry houses, and the administration of the right medicine. Both of these strategies can prevent the occurrence disease or reduction of the economic losses. According to the 78 number of broiler breeder flocks`disease data in the past four years in two provinces Golestan, and Mazandaran, the ages of the disease events is divided as up to three weeks, 4-6 weeks, 7-9 weeks,10-15 weeks, 16-21 weeks, 22-30 weeks, 31-64 weeks based on the repetition of disease and for each period of life we discussed about the main clinical signs of diseases. At the age of 0-3 weeks omphalitis and yolk sac infection, neonatal colisepticemia, Infectious Bronchitis and aspergilosis were seen. It is very important to confirm the quality of broiler breeder chicks in terms of vertically transmitted pathogens similar to Mycoplasma infection and Salmonella infection. At age of 4-6 weeks the main recommendation is to be aware of clostridiosis and coccidiosis and litter and feeding management. At the age of 7-9 weeks, litter management is very important because at age of 10-15 weeks two main identified problems are coccidiosis and staphylococcal arthritis. These phenomena, to some extent, depend on litter management, in addition to the role of Immune suppressors. At the age of 16-21 weeks, broiler breeders were vaccinated individually. At the age of 22-30 weeks we observed some individual cases of tumoric mortality and sometimes



enteritis. At this time two drug prevention recommended in some flocks based on their conditions which one of them is for mycoplasma and the other one is for ascaridial infection. After that, from 31 weeks up to the slaughtering time due to many factors occurrence of newcastle disease, avian influenza, infectious bronchitis, avian influenza, infectious bronchitis, infectious laryngotracheitis, mycoplasma infections, ORT Infection, salmonella infection, fowl cholera disease were seen and there was no clear pattern to explain the timing of these diseases at the production period. However, it should be known that most diseases occur at any age and defining such clear times is only for the purpose of transferring the events experienced in the industry in a practical way to other sand it should be known that the real situation of professional work in broiler breeder requires a dynamic mind in discovering the truth. Finally, it is important to mention two points. First, in recent years, flocks with adenovirus serotype-4 conflicts were seen among these flocks, regardless of the specific age range (and in some cases even despite the vaccination of the broiler breeder flock), it occurred in the flock. And the second point in this talk is considering the occurrence of mentioned diseases with clinical symptoms. Other diseases with no clear clinical symptoms occurred in the mentioned flocks, confirmed and proved by laboratory traces which need separate attention but could not be discussed in this short presentation.





**The application of histopathology in the diagnosis of poultry diseases:  
abandoned values of a precious technique**

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Farm investigation, history taking, necropsy, and laboratory analysis of field and clinical samples are different compartments of diagnostic puzzle in poultry industry. All poultry veterinarians are familiar with necropsy techniques. Either dead or moribund birds are usually subjected for necropsy. Even live and apparently healthy birds could be biopsied in a farm to be euthanized for necropsy procedure. Despite the high frequency of necropsies which are performed on daily basis, the application of microscopic investigations of tissue specimens are seldom implemented in veterinary laboratories of the country. Unavailability of skilled avian pathologists, lack of histotechnology equipment and relatively long running time for the procedure are the most significant causes of the abandonment of such a precious technique. Fortunately the required sampling materials including formalin solution and a simple container are often available in poultry farms. After putting the tissue samples in a vessel of preferably but not necessarily buffered 10% formalin, there is no need for either cold chain or any other special conditions while transferring the specimens to a laboratory. The collected formalinized samples can be preserved for a long time on the farm without any biosafety hazards. Frequent change and refreshing of the fixative solution is just needed to maintain the tissue quality and to prevent formaldehyde polymerization and precipitation. Medical pathology laboratories are usually well cooperated for tissue processing and slide preparation and there is no imperative need for investment on histotechnology in all veterinary laboratories.

Regardless of the diseases, histopathologic investigation of the cadaver is always informative. Small pieces of multiple tissue sections are usually studied. Immune competency of the bird and environmental state of the barn can be assessed by thorough histologic investigations of different organs. The isolation and identification of avian pathogens in clinical samples and tissues could contribute the definitive diagnosis in many infectious diseases, but there are numerous diseases which the histopathology is the only procedure for confirmed diagnosis. On the other hand, histopathologic findings can differentiate variable pathologic entities and work as pathfinder for further investigations. Some examples are presented for the right application of histopathology in poultry diseases.

Rickets or juvenile osteodystrophy is a common disease in growing birds which can be caused by different nutritional deficiencies. The etiologic diagnosis of rickets is difficult and feed analysis can give some presumption. The histologic investigation of epiphyseal plate of long bone can



differentiate the etiologic causes of rickets and it can also rule out the infectious diseases. Ionophore poisoning could be the most common toxicoses in turkey industry. While the toxicologic assessment of the feed is sometimes inconclusive, the histologic lesion of the skeletal muscle is characteristic for the diagnosis. Avian encephalomyelitis virus (AEV) is ubiquitous and its molecular detectin in the chicks is not always diagnostic. Pathognomonic lesions of AE in central nervous system are the most promising findings for its diagnosis. In addition the isolation or detection of fowl adenoviruses without demonstration of characteristic histologic lesions could not proceed to definitive diagnosis of either inclusion body hepatitis (IBH) or hepatitis-hydropericardium syndrome (HHS). Avian hepatitis E virus infection can cause characteristic coagulative and fibrinoid necrosis, vasculitis and amyloidosis in the affected liver. Other viral diseases like avian pox and infectious laryngotracheitis have pathognomonic histologic lesions to be diagnosed. Histopathology could make distinction between different lymphoid tumors. The Marek's disease (MD) and lymphoid leucosis (LL) can be differentiated from each other by histologic investigation.

The histopathology is an old but yet precious diagnostic tool in the hand of poultry veterinarians. They can apply this tool to improve their capabilities and to increase the knowledge of disease process and etiology. No other diagnostic tool like PCR, serological or microbiological assays can substitute the microscopic pathology. As poultry veterinarian, we should learn the use of histopathology in our routine practice.



### **Challenges of the country's poultry industry**

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Iran has been industrially producing poultry products for more than 70 years. However, production took off right after the Islamic revolution in 1979, and chicken meat and table eggs began to appear in supermarkets and other similar establishments. The Iranian Poultry Industry produced approximately 140 thousand metric tons of table eggs and around 200 thousand tons of chicken meat for 36 million people in the early 1980s. Per capita consumption of approximately 25 Kg and 13 Kg of chicken meat and table eggs, respectively, has happened with a yearly development pace of around 4% and 3% over the past forty or more years, despite the way that chicken meat utilization beat 30 Kg in 2020. If the hard currency exchange rate reform hadn't happened in May 2022, the nation would have produced well over 2.5 million metric tons of ready-to-cook (RTC) chicken meat and 1.3 million metric tons of table eggs by the end of 2023. However, reform in the exchange rate has negatively impacted consumption, with a 30% drop in purchasing power since May 2022, when the exchange rate corrected for imports of essential goods (from 42000 to approximately 250,000 Rials). Overall, the capacity to produce chicken meat has increased tenfold over the past 43 years, while the capacity to produce table eggs has increased approximately fourfold.

In general, the structure of the Iranian poultry market has been shaped to reflect perfectly competitive market conditions. In a fragmented configuration, more than 650 day-old chick producers (PS farms), more than 1600 table egg producers, and more than 20 thousand chicken growers compete with one another in their respective sectors. Each of these producers has a small share of the market. All of these producers are price takers, and no participant has any influence over the product's price. Nevertheless, significant integrations (about 50) have been established over the past ten years. These integrations account for about 30% of the market, with the largest accounting for 5% of all chicken meat production.



**Infectious Bronchitis virus; Current situation, Evolution, vaccination and future strategies**

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Infectious bronchitis (IB) first identified in 1937 is an acute, highly contagious respiratory disease of chickens, caused by a gamma coronavirus in the family Coronaviridae that consists of a positive-stranded RNA of 27.6 kb in length, which has been recognized as a pathogen of infectious bronchitis (IB), as well as respiratory and urogenital diseases in the commercial poultry industry worldwide. There are several different serotypes and genotype of the infectious bronchitis virus (IBV), found worldwide. All strains of IBV can replicate on most chicken epithelial surfaces, such as those of the trachea, lungs, kidney, oviduct, alimentary, and proventriculus. Damage to the epithelium predisposes the birds to secondary infections with pathogenic bacteria, especially *E. coli* and *Mycoplasma*. Several vaccines are available for prevention of the infectious bronchitis. However, since there are numerous different strains of the virus the vaccine is not always effective. Each geographical region has strains and/or variants of concern, and some are of global concern, including Massachusetts serotype. Young chicks are vaccinated with live-attenuated vaccines that are administered through sprays or via drinking water. Breeders and layers are then boosted at defined intervals with either live attenuated or inactivated vaccines. Live-attenuated vaccines are favored due to the ease of application; however, there is a risk of vaccine breakdown and reversion to virulence. The latter is compounded since the exact molecular mechanism of attenuation is unknown, and research has indicated that only a few consensus-level mutations are acquired over the passaging process, providing a short route back to virulence.

In fact IBV is able to evolve rapidly and recombine, leading to the emergence of a remarkable genetic and phenotypic variation over time. This heterogeneity has noteworthy points to understanding of IBV epidemiology and its control. Currently, vaccination is the most effective and applied strategy to limit the disease impact. Nevertheless, the antigenic variability leads to poor cross-protection, requiring the use of different vaccine combinations in order to broaden the protection spectrum or the development of new vaccines against recently emerged or introduced genotypes. Unfortunately, even closely related vaccines can fall into episodes of incomplete protection or vaccine immune-escape because of amino acid substitution in specific antigenic sites. A high mutation rate does not automatically lead to a comparably elevated heterogeneity in biological features: the persistence and spread of new phenotypic variants implies that they must be

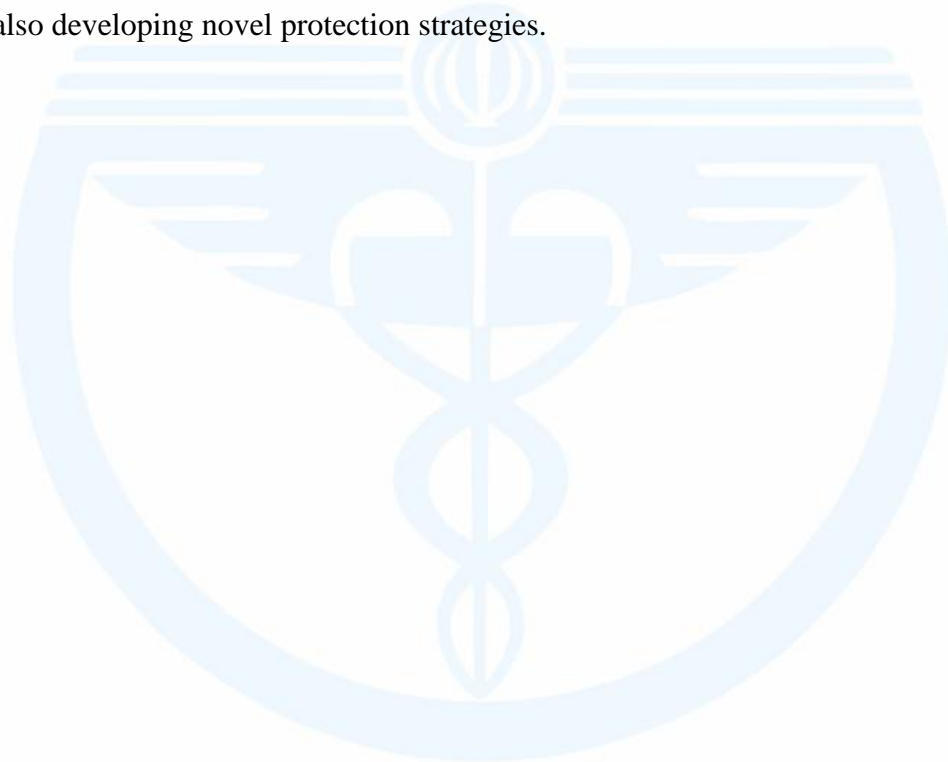


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favorably selected by the environment. In this sense, the host immunity represents one of the most obvious forces that can promote viral diversification, especially in antigenic regions. Besides natural immunity, vaccine administration could significantly contribute to this process. When immunity is not sterilizing, wild strains are able to circulate in a new and more challenging environment, potentially adapting to it. In conclusion before engaging in battle against IB disease you must first prepared by enough knowledge around the various virus aspects. In addition, there is a vital importance in continuous monitoring of the virus evolution, different vaccine protection studies and also developing novel protection strategies.



سازمان نظام دامپزشکی جمهوری اسلامی ایران  
Veterinary Council I.R. IRAN



**Isolation, identification and antibiogram of *Mycobacterium* from poultry in Iran**



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*Mycobacteria*, which have long been known as one of the most important zoonosis diseases, cause dangerous diseases in humans and animals. Atypical or non-tuberculosis *Mycobacteria* are among the large members of *Mycobacteria* found in soil and water. The *Mycobacterium avium* complex includes *Mycobacterium avium* subsp. *avium*, *Mycobacterium avium* subsp. *intracellulare*, and *Mycobacterium avium* subsp. *paratuberculosis*. According to the report of the World Health Organization, tuberculosis is the second infectious cause of death after Corona and the second cause of death of patients with AIDS, therefore, the presence of *Mycobacterium avium* complex in the birds and personnel and subsequent, antibiogram, helps to the health of society, especially in patients with AIDS.

The goals of this research are to isolate *Mycobacterium* from birds and personnel of Alborz Bird Park, and identification of *Mycobacteria* and the antibacterial susceptibility pattern of isolated *Mycobacteria*.

In this research, 90 samples of bird feces and 10 samples of human sputum were taken from Alborz Bird Park and transferred to the Tuberculosis Reference Laboratory of Razi Vaccine and Serum Research Institute.

The specimens were decontaminated and cultured on glycerinated and pyruvate Lowenstein-Jensen's medium as well as Herrold's medium.

Genomic DNA of all isolates were extracted by phenol-chloroform method and identification of all isolates were done using 16SrRNA, IS6110, P90-91, IS901, IS1245 primers.

42 suspected cultures to *Mycobacterium* were grown out of 100 samples, and the genus of *Mycobacterium* were confirmed in 33 isolates using PCR by 16SrRNA primers.

After performing PCR by IS6110 and P90-91 primers, no *Mycobacterium tuberculosis* complex and *Mycobacterium avium* subsp. *paratuberculosis* were observed. After performing PCR by IS1245 primers, which indicates the presence of *Mycobacterium avium* complex, 21 isolates belonged to *Mycobacterium avium* complex, and using IS901 primers, 16 isolates were confirmed to belong to *Mycobacterium avium* subsp. *avium*. Among these, 12 isolates were also reported as other NTM.

All 33 isolates were tested with six antibiotics: streptomycin, rifampin, etambutol, kanamycin, isoniazid and ethionamide. A higher percentage of isolates were resistant to the above antibiotics.

**Key Words:** *Mycobacterium avium* complex, *Mycobacterium avium* subsp. *avium*, birds, antibiogram



### The role of nutrition in poultry gut health and immunity

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Developing a concept of “gut health” has been a challenge amongst nutritionists, veterinarians and scientists, worldwide. It can be defined as the state of resilient equilibrium or homeostasis of the intricate intestinal neuro-endocrine-immune-microbial systems that allows full functionality of the intestinal tract, overcoming challenges to guarantee animal health, well-being, and productive performance. As it represents the largest mucosal surface continuously exposed to potential aggressors, the intestinal tract is also responsible for protecting the bird, as a complex barrier and an active immunological organ with more resident immune cells than anywhere else in the body. Evidence from humans and murine models has clearly established the causal role of the gut microbiota in shaping, educating, and developing the immune system. The interface between intestinal immune system and microbiota controls many aspects of the intestinal health of animals and therefore must be considered as an additional organ that fulfills important functions with specific nutritional requirements.

The perihatch period is a vital time for the development of the gut microbiome and immune system. Providing chicks immediate access to feed has been shown to improve intestinal mucosal function, improve gut associated lymphoid tissue function, improve early growth, and yields. It has been demonstrated that supplying nutrients and additives to the growing embryo through in ovo feeding may enhance the development of the intestinal tract. Early microbial communities' modulation toward beneficial bacterial colonization also holds a great promise for improving health and productivity in poultry.

A wide range of factors associated with diet can affect the delicate balance among the components of the chicken gut and, as a result, affect health status and production performance of birds in commercial poultry operations. It has been extensively shown that the presence of anti-nutritional factors and mycotoxins have a significant detrimental effect on intestinal health, compromising its functionality through oxidative stress and inflammation and promoting the occurrence of enteric pathogens and diseases. Feed composition, feed form and physical texture can be further explored as opportunities beyond just simply supplying nutrients, but also to manage intestinal health in poultry production. In addition, feed deprivation and withdrawal also influence gut health and microbiota composition. Strategies to modulate the intestinal health through probiotics, prebiotics, enzymes, phytochemicals, or microbiota transplantation are showing promises in enhancing disease resistance and production efficiency in poultry.

**Key words:** intestinal health, poultry, microbiome, feeding, feed additives

**Avian Influenza H9N2, Is this subtype the next pandemic virus?****Hasan Nili***Zeitoon Isfahan Vaccine Innovators Company  
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There are an increasing number of countries in different continents reporting avian influenza H9N2 subtype infection in their commercial and wild birds. Also, inter-species transmission among birds, mammals and human infections with the viruses becoming more frequent than before. Interestingly several new emerging human-infecting avian influenza viruses, such as H7N9, H10N8 and recently isolated H3N8 have obtained at least some segments of their genome from H9N2 viruses. The first highly pathogenic avian influenza virus (H5N1) isolates from Hong Kong in 1997 with fatal bird flu transmission to human, shared some of its genome components from H9N2 of a quail in the same region. Recent Research shows efficient replication of H9N2 avian influenza virus in new host which indicate that the host range of the virus is expanding without adaptation being needed. New findings also show that these viruses circulating in the commercial birds have more chance to exchange their genome segments with other avian influenza subtypes, and show more affinity to human receptors, facilitating inter-species transmission. Although until late last year more than 100 cases of human infections with H9N2 avian influenza virus have been reported, it is assumed that the number is most likely to be much higher. Since the great number of the isolated viruses have shown more affinity in binding to human-type sialic acid receptors.

Although nationwide vaccination programs are being conducted in different countries, the efficacy of vaccines and vaccination programs should be constantly monitored, since H9N2 viruses have experienced significant genetic reassortment in recent years and shedding and circulation of the virus have been continued and not prevented despite vaccination programs of commercial flocks. Recent studies show that there has been evolution in new avian influenza H9N2 subtypes in Iran and other parts of the world.

The surveillance programs have to be intensified and vaccination strategies should be revised based on genomic and antigenic characteristics of circulating viruses. Also, more preventive measures should be applied to prevent inter-species transmission of H9N2 avian influenza virus specially exposure of veterinary personals and poultry workers with the virus.

**Key words:** Avian influenza virus, H9N2 subtype, pandemic potentials





**Using of EggYolk Specific Immunoglobulin (IgY) in Prevention and Treatment of Diseases in Humans and Animals**

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Polyclonal antibodies, widely used in research and diagnostics, are conventionally isolated from the blood of immunized mammals. The fact that antibodies can also be detected in the yolk of eggs laid by immunized hens, led to the development of the yolk antibody technology as an alternative method less stressful to animals. Hens can be kept under nearly natural conditions and antibodies be isolated from the eggs. Avian antibodies have been shown to be in some applications even more effective than mammalian antibodies, especially when phylogenetically highly conserved antigens have been used for immunization. Further, the IgY-technology has the advantage that avian antibodies can be produced against several antigens which do not induce an immune response in mammals.

Egg yolk constitutes a relevant alternative source of antibodies. It presents some advantages over mammalian serum immunoglobulins regarding productivity, animal welfare and specificity. The main immunoglobulin present in avian blood (IgY) is transmitted to their offspring and accumulates in egg yolks, which enables the non-invasive harvesting of high amounts of antibodies. Moreover, due to structural differences and phylogenetic distance, IgY is more suitable for diagnostic purposes than mammalian antibodies, since it does not react with certain components of the human immune system and displays greater avidity for mammalian conserved proteins. IgY has been extensively used in health researches, as both therapeutic and diagnostic tool.

The avian egg contains all of the necessary nutrients and growth factors required for the developing embryo, including antibodies that are transported from the blood of the hen into the egg yolk to provide immunity to the chick. Since the discovery of egg yolk antibodies, now called immunoglobulin Y (IgY), in the late 1800s, this process has been harnessed to produce antigen-specific yolk antibodies for numerous applications in the medical and research fields, including in areas such as diagnostics and proteomics. However, one of the most valuable and promising areas of IgY research is its use for passive immunization to treat and prevent human and animal diseases. The following review covers the key features and advantages of IgY and the production and purification of IgY from the egg yolk, as well as highlights some of the most promising applications of egg yolk antibodies in human and veterinary medicine. This presentation aims to review IgY applications in both human and veterinary health.



### Conclusion

The latest findings using IgY have clearly demonstrated the versatility of this technology. Obtaining IgY from birds presents several technical and economical advantages over mammalian IgG, and as described in this review, IgY technology has a broad spectrum of applications in human and veterinary health. It can be used in multiple types of therapies; it is useful in the prevention of various types of diseases and detects, by means of different techniques, several classes of antigens, such as microorganisms, tumor markers and substances.

Among the advantages of this technology, the replacement of invasive antibody collection by its extraction from eggs is one of the most interesting, considering the animal welfare benefits, with this technology it is possible to achieve great quantities of antibodies with a lower cost of production and less damage to animal welfare.

Due to its structural differences and phylogenetic distance, IgY is more specific for diagnostic use and displays greater avidity for mammalian conserved proteins than IgG, being, thus, an important alternative in the search for more effective diagnostics and therapies. In addition, in view of its proven ability to neutralize microorganisms, IgY represents an important therapeutic resource in times of increasing resistance to antibiotics and emergence of viral diseases for which there is no treatment.

**Keywords:** Chicken antibodies, immunoglobulin Y, IgY production and purification, antibody therapy, antimicrobial, passive immunization

### **Reference**

[BOOK] Chicken egg yolk antibodies, production and application: IgY-Technology

R Schade, I Behn, M Erhard, A Hlinak, C Staak - 2000 - books.google.com

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Veterinary Council I.R. IRAN



### **Avian Immunosuppressive Diseases**

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The avian immune system provides an extremely useful model for studies of basic immunology. Birds and mammals evolved from a common reptilian ancestor more than 200 million years ago and have inherited many common immunological systems. They have also developed a number of very different and, in some cases, remarkable strategies. Because of their economic importance, and the ready availability of inbred lines, most avian immunology research has involved the domestic chicken, *Gallus gallus domesticus*.

- Avian Immune systems consist of:
  - Anatomical
  - Innate
  - Humoral
  - Cell mediate
  - Mucosal

Control of infectious diseases is essential for the production of healthy poultry flocks, and this is generally achieved by extensive vaccination programs in combination with good management practices, including biosecurity measures to reduce the risk of infection. The success of vaccination programs depends on

The ability of the birds to mount a vigorous immune response after vaccination. In addition to the innate

ability of a particular bird to mount an immune response to a vaccine or an infection, there are numerous

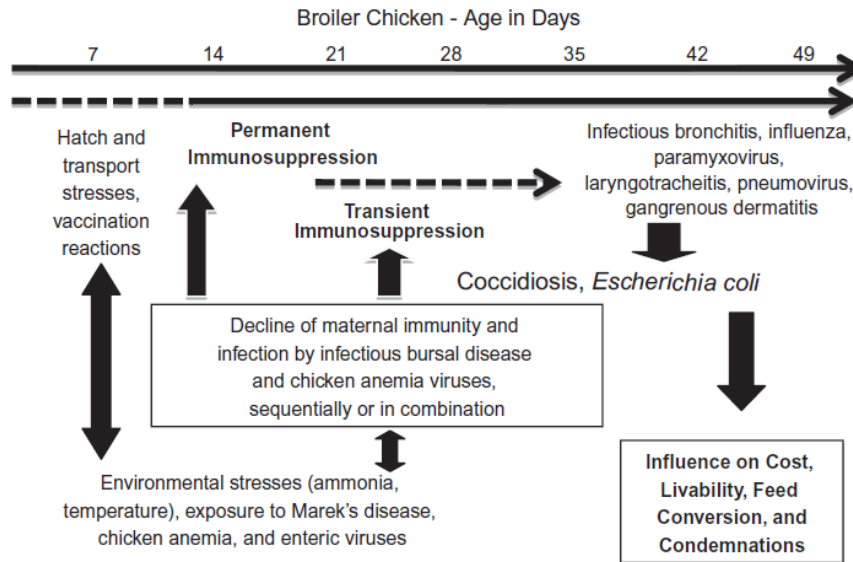
external factors influencing the level of protective immunity.

immunosuppression is defined as “A state of temporary or permanent dysfunction of the immune response resulting from insults to the immune system and leading to increased susceptibility to disease,

and often a suboptimal antibody response and suboptimal innate and cell-mediated responses.”

Such dysfunction often results from infection of cells of the immune system, leading to their impaired function against the primary and subsequent infections in a non-specific manner.

Immunosuppression is a major problem for the poultry industry, but actual figures indicating the scale of the problem are difficult to find. Infection with pathogens and/or environmental factors, including management errors, can result in immunosuppression, and interactions between the two usually exacerbate the problem.



Environmental stressors include the later period of incubation, hatching and chick handling. Other stressors include suboptimal housing conditions and mycotoxins. Any infection causing clinical disease may result in immunosuppression, but the focus here is on pathogen-induced immunosuppression

in the absence of clinical disease. Viruses causing immunosuppression and/or clinical disease at any age include MDV, reticuloendotheliosis virus (REV), reovirus and, although controversial, avian leukosis virus (ALV). Other pathogens—for example, chicken infectious anemia virus (CIAV)—may cause

clinical disease in young chicks, but the major damage is caused by subclinical infection resulting in immunosuppression. Infectious bursal disease virus (IBDV) also causes clinical disease in young chicks, but results in damage to immune tissue, particularly the bursa of Fabricius and bursa-derived lymphocytes, compromising the host's ability to mount effective responses upon subsequent infection by other pathogens.

**Poultry industry and its role on food Supply****Ghasem Rezaeianzadeh***Iran Veterinary Organization, Tehran-Iran*

Poultry farming is a significant pillar of Secure Food Supply Plan in each country and nowadays the poultry industry role in supplying animal derived proteins of various communities is more indispensable than any other time.

Low production cost, high productivity, advantages of white meat over red meat beholden to its nature, short rearing period and finally rapid growth rate in comparison with livestock and aquatics grant unique position to poultry farming in the field of animal derived protein production.

Poultry feeding easiness (no need for pasture and rearing using concentrate feed), high reproduction capacity (short generation gap and abundance of progeny), high growth rate, low post slaughter carcass loss, control over animal premise rearing conditions and high economical productivity are other important reasons for poultry farming development granting an special role to chicken meat and other poultry products in Secure Food Supply Plan. To this end, different countries invest in this industry given the importance of this industry. The same applies to our country and we witness the ever-increasing progress of this industry in Iran granting us a noticeable rank among poultry and poultry products producer countries.

**2-Poultry diseases and mortalities in poultry industry**

Given the poultry diseases and mortalities role in production reduction, irreparable damages might be imposed to the industry particularly due to newly emerging or re-emerging diseases.

Poultry industry development in different countries and rise of global communications namely import and export poultry and poultry products caused us to witness poultry diseases increment which exacerbates and intensifies the status owing to poultry farms over condensation.

Taking into account the current poultry industry status in Iran i.e. abundance of low capacity poultry farms with separated management, poultry disease in one farm imposes economic losses including mortalities, low growth and production rates, treatment incurred expenses threatens other farms in the area which is more likely to spread to other farms all over the country which challenges national poultry industry. In case of newly emerging or re-emerging diseases, this could impose trade restrictions on export of poultry and poultry products facing poultry industry imposing huge economic losses to the country.

**3- Iran Veterinary Organization solutions to face with poultry industry current challenges**

Veterinary medicine plays an important role in health of human communities and improving animal health aiming at keeping animal wealth and prevention of zoonotic diseases. According to WHO, 25% of animal product added value derives from special veterinary services. To this end,



following measures have been adopted in the field of poultry health along with some approaches to improve the current status:

A-Adopted measures:

- 1-Correcting breeder and layer farms structure in terms of hygienic aspect relying on farms audit plans
- 2-Updating and reviewing poultry diseases control programs
- 3-Paving the way for exporting and removing imposed trade restrictions by other countries
- 4-Using domestic and science -based companies in production of required ingredients including vaccines and antigens
- 5-Offering executive approach in order to conduct placement unauthorized farms
- 6-Conducting placement of production in high risk and poultry farms condensed area
- 7-Using specialized system for poultry disease surveillance and monitoring for rapid access to poultry diseases foci
- 8- Vaccinating back yard and rural areas poultry population
- 9-Conducting salmonella monitoring project in grandparentand broilerbreeders farms

B- Proposed approaches to improve current status

- 1-Correcting poultry farms structures in terms of hygienic aspects
- 2-Managing age conditions for keeping poultry flocks in different farms
- 3-Establishing and strengthening integrated poultry farms
- 4- Developing poultry and poultry products transport system qualitatively and quantitatively
- 5-Planning production relying on information and appropriate statistics abstracted from poultry industry realities
- 6-Conducting placement of unauthorized farms
- 7- Developing and correcting training plans for all beneficiaries using state of art know-how and modern and updated methods
- 8-Improving relationships between educational and research sections and governmental and nongovernmental executive departments in order to develop applied researches with participation of industry owners
- 9-Reviewing regulations and directives

**Iran's Poultry Industry in past, present and future****Mostafa Seyed Mostafavi**

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Although it is said that the origin of chicken was from India, but the age of domestic and rural chicken keeping (backyard poultry) in Iran is more than 2500 years. In the years before these years prominent civilizations were China, Iran and Egypt, and Greece and Rome were being formed. Maybe India was a part of Iran's great civilization.

Domestic and rural poultry farming continued until the 1900s, but from now on, it gradually became traditional chicken farms on the edge of gardens and agricultural fields, and was further developed, and changed from small nests to domed roof houses known as Sayerd Ziya Tabatabaei ( who brought this design from Egypt) chicken farms.

In the 1950s, poultry farming was further developed with the arrival of 60,000 purebred chickens from New Hampshire and Rhode Island Red breed, and a number of hatching eggs and incubators under the name Truman Principle 4. Of course, with this import, the Newcastle disease spread and almost the native Iranian poultry generation was destroyed. In the same decade, feed mills and hatcheries were built and served small poultry farmers.

But in the 1960s, it was the time of the establishment of large industrial chicken farms, in this decade, the first flock of layer grandparents was imported into the country by the Morghak Company, and in the same decade, the first export of hatching eggs to Syria took place.

Now in the country there are nearly 50 units of broiler breeders, one unit of layer grandparent and 4 units of layer parent farms, as well as a large number of broiler and layer farms. Chicken meat is still for consumption by the rich and sometimes recommended to the sick people. For this reason, chicken meat is imported to solve the shortage.

After the Islamic revolution in 1978 and the adoption of the self-sufficiency policy, the development of poultry farms increased with the support of the government.

Creation of a day-old chicken distribution center, which was an institution in the form of a horizontal chain, to control the production and distribution of chicken and imported feed ingredients and distribution of the final product, namely meat and eggs, using leafy goods that all people can buy and consume at an approved price.



During these days, which coincided with the beginning of the imposed war, the government started importing broiler grandparents and at the same time started creating a pure line center at babolkenar. The import of pure line flocks from the Netherlands took place in 1991, in order to be able to produce the required broiler breeders in the country in crisis conditions. But a few years later, in 2000, due to the economic importance of chicken meat production and production efficiency, the importation of grandparents from world-renowned breeds was allowed. In addition to being self-sufficient, Iran also had an export demand and annually exported about 40,000 tons of eggs and 80,000 tons of chicken meat and thousands day-old chicks and pullet to neighboring countries.

Within 40 years after the Islamic revolution and at the end of the fifth economic development program, the poultry industry increased the amount of chicken meat production to 2300000 tons with an average annual growth of 6.3% and the egg production to 950000 tons with an average annual growth of 4%. At the end of the 6th economic development plan of 2021, it predicted 2,500,000 tons of chicken meat and 1,100,000 tons of eggs, which were realized.

However, the spread of the global corona disease in the world and in Iran had a negative impact on consumption, and the turmoil caused by the market, along with the change in political conditions and the establishment of serious sanctions against Iran, caused a very heavy inflation in the economy and devaluation of the national currency severely decreased people's purchasing power. These issues caused the adoption of several policies in the production and consumption of these products.

While there are built capacities for the production of 3.4 million tons of chicken meat and 1.8 million tons of table eggs, including grandparent farms, broiler breeders and layer breeder farms, as well as hatchery feed mills, supplements and concentrates production plants, and industrial poultry slaughterhouses. But only 60-70% of the built capacities are used for rotational production.

There are solutions to get rid of the challenges of the industry, which I will discuss in the main article.



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**8<sup>th</sup> International Veterinary Poultry Congress****هشتمین کنگره بین المللی دامپزشکی طیور****Interpretation of serological results in poultry flocks****Nariman Sheikhi***Department of Clinical Science, Faculty of Veterinary Medicine, Science and Research branch, Islamic Azad University, Tehran, Iran***Corresponding author's email: [Pasteurlab78@yahoo.com](mailto:Pasteurlab78@yahoo.com)**

For good and reliable serological results, we need good blood samples. Good samples have some characters such as: the sample size (at least 15 -25 samples are needed in different situations), selection of suitable birds, choosing the appropriate site for blood sampling, the appropriate blood volume (between 1 to 3 milliliters is usually enough), appropriate ways of keeping samples, choosing the correct time of sampling after challenging with pathogens, vaccination, ..., choosing the right time of the day for sampling, and the correct ways of sample transporting (sample paper is a simple and effective way to send serum samples to laboratories).

When rapid serum agglutination test is selected, the following points should be considered: choosing the right time after performing killed vaccination, sensitivity and specificity of the test, and not freezing the samples. We should notice that other infections may cause incorrect positive results. In case of chickens younger than one week old it has many false positive. If the result is positive, the test must be repeated after one week. Hemagglutination Inhibition test has good specificity and low sensitivity. Specific antigen is used for AIV, ND, EDS and some other pathogen in HI tests. Usually, titer higher than  $\log_3$  is considered positive. ELISA tests may detect antibody or antigen. There are different kinds of ELISA tests. Direct ELISA can be used to detect antibody or antigen. Indirect ELISA is often used for detecting antibodies. In Sandwich ELISA, monoclonal antibody can be used. In Competitive ELISA, different kinds of birds' samples can be tested with the same kit. Titer, mean titer, geometric mean titer, CV, baseline, and vaccination index are used for interpreting the results.

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## **The Structure, Evolution and Pathogenesis of influenza A viruses: at a Glance**

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Influenza A viruses are the most important in Orthomyxoviridae family, as these viruses are known to infect a wide variety of birds and mammals, while the other type have more restricted host ranges. They have 8 single-stranded-negative-sense, RNA segments. These encode 10 main proteins including 3 polymerase proteins—PB1, PB2 and PA, two surface glycoproteins—hemagglutinin (HA) and neuraminidase (NA), Nucleocapsid protein—nucleoproteins (NP), Matrix proteins—M1, Ion pore protein M2 and 2 nonstructural proteins (NS1 and NS2) respectively. On the basis of 2 surface glycoproteins this genus is classified into subtypes. 16 HA and 9 NA subtypes of influenza A viruses have been isolated from birds (H1 to H16 and N1 to N9). More recent studies have shown expression of a set of accessory proteins like PA-X, PB1-F2, etc, which varies between virus strains and is nonessential for virus replication in cell culture, but in many cases has been shown to affect virulence and/or transmission in vivo.

The major functions of the HA are the receptor-binding and fusion activities. The HA has essential roles in antigenic drift (antigenicity) and shift. Host cell attachment by influenza A viruses is mediated by receptor-binding domain (RBD) of HA. The recognition of specific types of sialic acid contained in glycan receptors constitutes one of the major determinants of viral host range and transmission properties. The RBD of HA is formed by the 190-helix at the top of HA, the 220-loop at the edge of the globular head, and the 130-loop at the other edge of the globular head. The HAs of human influenza virus strains preferentially bind to sialic acid linked to galactose by  $\alpha$ 2,6-linkages, whereas the HAs of avian influenza virus strains prefer a sialic acid linked to galactose by  $\alpha$ 2,3-linkages. As genetic factors that play a role in the adaptation of influenza viruses to new hosts continue to be defined, it becomes increasingly clear that numerous factors (often interrelated) and selective pressures are involved in driving the evolution of host range variants and partially adapted strains. The second major function of the HA is acid pH-triggered fusion, which is required for the uncoating process. Proteolytic cleavage of HA is a prerequisite for membrane fusion.

The HA proteins of highly pathogenic H5 and H7 viruses contain multiple basic amino acids at the cleavage site which are recognized by ubiquitous proteases such as furin. For this reason, these viruses can cause systemic infections in poultry. In contrast, the HA proteins of avirulent avian and non-avian influenza A viruses. These viruses, therefore, produce localized infection of the respiratory and/or intestinal tract that is usually asymptomatic or mild. The NA is the 2<sup>nd</sup> major glycoprotein of influenza A viruses. Generally speaking, NA is responsible for viral mobility, allowing the virus to reach the site of receptor and leave after viral replication. NA inhibitors can restrict the normal spread of the infection. Since HA and NA have opposing functions, it is likely that balanced activities of these two proteins is critical for efficient IAV replication.



After un coating, the viral ribonucleo proteins (vRNPs) are transported into the nucleus. as all viral RNA synthesis occurs in the nucleus, one that is unusual for an RNA virus. The trafficking of the vRNPs into and out of the nucleus is a tightly regulated process. When a virus infects a cell, it has to contend with the rapid on set of the host innate immune response, whose mission it is to establish an antiviral state within the cell and prevent virus replication. NS1 protein interacts with a variety of proteins to inhibit host cell immune responses and promote viral replication. Influenza viruses assemble and bud from the apical plasma membrane of polarized cells.

Influenza viruses' evolution is a complex process that includes the accumulation of mutations over time and the reassortment of viral RNA segments in cells infected with two or even more different viruses. In wild aquatic birds, avian influenza viruses evolve slowly; despite occurrence of mutations, most are not sustained in viral populations since they do not provide an evolutionary advantage. However avian viruses in terrestrial poultry, including highly pathogenic H5N1 viruses evolve rapidly. In contrast to most avian influenza viruses, human influenza viruses show detectable net evolution over time.

Here, a brief look on virological and evolutionary aspects of influenza A viruses insisting on Iranian isolates especially H9N2 viruses will be present.



**Toxicity and Bioactivity of Silver Nano Particles: Chick Embryo Model and Molecular Dynamic simulation**

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**Objectives**

The poultry industry is mainly affected by infections due to microorganisms which reduce the growth rate and cause economic losses. Currently, vaccines and antibiotics are utilized to combat these infectious microorganisms, but extended and improper use of vaccines and antibiotics may pose risks for poultry raising and public health, and there is a need for drug alternatives. Nanotechnology could reduce such risks and can improve the poultry products. Various studies in the literature focus on the toxicity and side effects following nano particles applications. The present study was conducted to investigate the vascular-toxicity of silver nano particles (AgNPs) for chick embryonic vessels. The side effects of AgNPs on the tear film and some aspects of AgNPs bioactivity were also evaluated.

**Materials & Methods**

Firstly, the vascular-toxicity of AgNPs as well as its interaction with apoptotic regulator proteins was predicted via in silico approach. In the next step, the apoptotic-signaling pathway in embryonic vasculature was evaluated using a chick's yolk sac membrane model. The interaction of AgNPs with a tear film lipid layer was also assessed through molecular dynamics simulation. Finally, the effect of AgNPs on the tumor cell line was investigated via chick's chorioallantoic membrane model.

**Results**

In silico simulation confirmed vascular-toxicity of AgNPs. There was an accurate affinity between AgNPs, Bax and Bcl-2 (-7.7 kcal/mol). Molecular dynamics assay revealed complex stability in the chick body conditions. Furthermore, AgNPs is suggested to alter Bcl-2 more than BAX. Morphometric quantification of the vessels showed that the apoptotic activity of AgNPs in embryonic vasculature was related to a marked reduction in vessel area, vessel diameter and mean capillary area. Based on the qPCR and immunohistochemistry assays, enhanced expression level of BAX and reduced expression level of Bcl-2 confirmed apoptotic responses in the vessels of the YSM. Furthermore, by employing molecular dynamics simulations, we demonstrate that AgNPs, due to its small size, is able to penetrate the tear lipid film and alters the tear components. Lastly,



current research demonstrates that multifunctional cytotoxic activity of AgNPs can be used as an anti-cancer agent against human breast cancer cell lines (MCF7).

### **Conclusion**

We observed that induction of an apoptotic signal can cause the embryonic defect of the vascular system following AgNPs treatment. The acquired data also raised suspicions that alteration in apoptotic genes and proteins in the vasculature are two critical pathways in vascular-toxicity of AgNPs. As far as the authors are aware, the current investigation is the first to assess the impact of AgNPs on tear lipid film by employing an in silico assay. This assay offer promising techniques for investigating the toxic effects of various agents. In addition, AgNPs are of great interest due to their biological property against tumor cells. The proper knowledge of this characteristic is essential to maximize their potential applications in many areas while minimizing their hazards to poultry, humans and the environment. In this regard, the chick embryo can consider as an appropriate model for investigating the anti-cancer activity of nano particles.

**Keywords:** Apoptosis, Cancer, Chick embryo, Molecular dynamics simulation, Nano particles, Silver, Tear



**How hepatitis-hydropericardium syndrome as a viral exotic disease is getting to be an endemic disease in northeast Iran**

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There have been many changes in the pathogenicity of fowl adenoviruses (FAdVs) during the last 30 years. Historically, FAdVs were known as inclusion body hepatitis (IBH), but now two emerging diseases, hepatitis-hydropericardium syndrome (HHS) and adenoviral gizzard erosion have been introduced in the field. Regular monitoring of FAdV infections has been done in Khorasan Razavi province for many years. The first occurrence of IBH was observed in some commercial broiler flocks 2–4 wk old with 3-5% mortality in 2009. The sporadic occurrences of IBH were the prominent features of fowl adenovirus infections for 12 years. The first occurrence of HHS in Iran was observed in a broiler flock in Bajestan. Our investigations showed that the FAdV-4 might be accidentally transmitted by horizontal infection in this flock. The stamping out program was successfully implemented for this case. For this reason, there was no HHS outbreak report for three months in the province. Unfortunately, the second wave of the disease started simultaneously in four broiler flocks through the progeny of a broiler breeder flock located in northern Iran. Although stamping out program was also implemented for these flocks, subsequent occurrences of the disease were reported from other parts of the province. Then epidemiological evidence revealed the horizontal transmission of the FAdV4 among the broiler flocks after the second wave of the disease. During the last two years, the number of HHS occurrences is increasing and the disease is spreading in a focal but creeping pattern in northeast Iran. It seems that HHS has become endemic in northeast Iran despite the intensive vaccination of broiler breeder flocks and the implementation of control and preventive measures at the provincial level.

**Keywords:** Hepatitis-hydropericardium syndrome, FAdV-4, control measures, Iran



**Mitigating the effects of ascites syndrome in a susceptible broiler strain**

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**Objectives:** Ascites accounts for a significant share of broiler mortality. The average loss of ascites in the world is estimated to be 4.2%, which causes more than one billion dollars of economic loss to the poultry industry annually. Ascites syndrome, also known as pulmonary hypertension syndrome, is mainly caused by pulmonary vascular volume insufficiency, and metabolic and or environmental hypoxia. The morphological changes of cardiovascular decrease pulmonary vascular compliance and increase pulmonary vascular resistance, which leads to pulmonary circulation disorders, right ventricular failure and hypertrophy. The vascular volume insufficiency is mainly caused by pulmonary vasoconstriction and vascular remodeling. Research results indicated that acetylsalicylic acid attenuates pulmonary arterial hypertension in rats by reducing plasma 5-hydroxytryptamine levels. L-Arginine inhibiting pulmonary vascular remodeling is associated with promotion of apoptosis in pulmonary arterioles smooth muscle cells in broilers. Furthermore, the addition of L-arginine to broiler diets results in a linear increase in broiler plasma nitric oxide concentration, induces vasodilation and reduces the incidence of ascites. Glycyrrhizin can ameliorates fibrosis, vasculopathy, and inflammation in animal models of systemic sclerosis. Dietary nitrate ameliorates pulmonary hypertension and has acytoprotective role for endothelial nitric oxide synthase and xanthine oxidoreductase. Therefore this study focuses on regulating vasoactive substances by using pharmacological and active metabolites on a genetically ascites susceptible broiler strain.

**Materials & Methods:** Three hundred forty-eight, one-day-old genetically susceptible broiler strains that have been selected for generations in a hyperbaric atmosphere and an ascites resistance modern broiler chickens were grown for 42 days. Standard three phases of pelleted feed and ad-libitum water were accessible during the 22-h light schedule day. The experiment was carried out at 1340m altitude in an environmentally controlled house. Four experimental treatments included groups without or with a combined dose of a feed therapeutic premix. The therapeutic premix supplied 1.8, 36.7, 7.3 and 46 mg/kg body weight acetylsalicylic acid, L-arginine, glycyrrhizin and nitric oxide, respectively. Treatments were replicated ten times. Performance traits were measured weekly. The occurrence of ascites was evaluated by measuring the ratio of the right ventricle to the total ventricle. At 42 days, blood viscosity was assessed by measurement of hematocrit and blood nitrite and nitrate levels were measured as indicators of nitric oxide metabolites. Because nitric oxide is a free radical, it is oxidized quickly into  $\text{NO}_2^-$  and  $\text{NO}_3^-$  in aqueous solutions and blood, allowing the measurement of total nitrite to equal total nitric oxide.

**Results & Conclusion:** Modern resistance broiler chickens performed better than susceptible strain ( $P < 0.001$ ). The incidence of ascites and relevance mortality was higher in the susceptible genotype compared to resistance birds ( $P < 0.01$ ). Feed administration of acetylsalicylic acid, L-arginine, glycyrrhizin and nitric oxide significantly increased the body weight of ascites-susceptible chickens compared to no administrated control group ( $P < 0.0001$ ). Plasma total nitrite concentration of treated and non-treated susceptible and resistance chickens were 68.39 vs 55.95 and 63.39 vs 73.08



$\mu M$  ( $P < 0.15$ ). Blood hematocrit significantly reduced in treating resistance strain ( $P < 0.0002$ ). Obtained results showed significant improvement in the physiological and growth performance of 42-d susceptible broiler chickens, which received acetylsalicylic acid, L-arginine, glycyrrhizin and nitric oxide in the feed. The appropriate dosage and a deeper understanding of the underlying mechanisms need more investigation.

**Keywords:** pulmonary hypertension, vasoactive, L-arginine, nitric oxide, broiler







**Prevalence and phylogenetic analysis of Fowl Adenovirus Serotype 4 in broiler chickens with Hydropericardium Hepatitis Syndrome (HHS) by Real-Time PCR HRM methods in Iran**

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**Introduction and Objectives:**

Fowl adenoviruses (FAdVs) are members of the Aviadenovirus genus belong to Adenoviridae family. The Fowl adenoviruses are divided into five species (FAdV-A to FAdV-E) and 12 serotypes (FAdV-1 to FAdV-8a and FAdV-8b to FAdV-11). FAdV-4 is the main causative agent of hydropericardium hepatitis syndrome (HHS) and IBH in chickens, which yield severe economic losses in the poultry industries for more than 30 years. HHS was first observed in 1987 at Angara Goth, Pakistan and therefore it was named as Angara disease. Later, the disease was reported in other countries including India where the disease was first detected in Jammu followed by Punjab and Delhi in 1994. Angara disease was emerged in Iran in 2020 for the first time and spread rapidly. The aim of this study was determination of the prevalence rate of Angara disease in broiler farms and finding the origin of FAdv by using the genomic sequence and phylogenetic analysis of positive samples.

**Materials & methods:** A total of 85 samples from 17 broiler farms that clinically suspected to HHS were tested in Isfahan province, Iranian 2022. Clinical signs and symptoms were a sudden increase in mortality with a swollen liver containing multiple pale or hemorrhagic foci and hydropericardium. Samples were collected from liver of suspected cases. Liver samples were stored in -20°C for detection of viral genome by real-time PCR, and confirmation by PCR HRM using universal and specific primers. The nucleotide sequence of PCR product was found by Sanger method and compare with consensus nucleotide sequence using Basic Local Alignment Tools (BLAST) in website of the National Center for Biotechnology Information of USA. Phylogenetic tree drawn by MEGA software version 10.

**Results and discussion:** The results of Real Time PCR of liver samples showed that 8 out of 17 broiler farms (47%)

.were positive for HHS adenovirus. The result of molecular diagnosis of adenovirus by DNA sequencing revealed that the pathogenic adenovirus serotype 4 (FAdV-4) is the main causative agent of HHS in this area. Also, phylogenetic analysis showed that the predominant virus has the highest similarities with Chinese strains.

**Key words:** Adenovirus, Angara disease, Real-time PCR, HRM, Broiler, Iran