

Comparative economic efficiency of using pharmacological agents for the stress prevention in the course of immunization of birds against reovirus tenosynovitis

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Abstract. This article describes the results of studies on the definition and comparative evaluation of the effectiveness of using SPAO-FA (stress protector antioxidant – feed additive) and SPAO-complex pharmacological preparation (stress protector antioxidant – complex preparation for birds) for the prevention of vaccine stress during immunization of birds against reovirus tenosynovitis. These preparations were used during 5 days (3 days before vaccination, on the day of vaccination and one day after vaccination) at the dose of 185 mg kg⁻¹ body weight: SPAO-FA was given as a part of compound feed, SPAO-complex – with water. Stress prevention in chickens of the parent flock provides equally high level of protection of birds from a field virus, a causative agent of reovirus tenosynovitis: it reduces the development of a protective level of antibodies by 7 days. Using SPAO-complex contributed to the increase in antibody titers by a factor of 2.6, and to the decrease in the coefficient of variation by the term of monitoring immunity stress by 25.7%. Using SPAO-FA increased antibody titers by 1.6 times, but had no significant effect on vaccination homogeneity. Vaccination index that should be high in the case of successful vaccination was 3.5 times higher for SPAO-complex in comparison with the basic diet; this for SPAO-FA was 1.6 times higher. Positive effect of anti-stress therapy influences the livability of chickens which was 3.5% higher with SPAO-complex, 0.79% higher – with SPAO-FA; bird live weight and the yield of day-old chicks were also higher in comparison with the basic diet by 6.44 and 0.88%, and 4.22 and 0.55%, respectively. Cost-effectiveness of using SPAO-FA per RUR of costs amounted to 180.09 RUR, for SPAO-complex, this parameter amounted to 435.86 RUR.

Key words: poultry farming, vaccination, reovirus tenosynovitis, antibody titers, coefficient of variation, vaccination index, cost-effectiveness, yield of chicks.

INTRODUCTION

Key concepts for the development of agro-industrial complex, including poultry farming today and for long term, are efficiency (Skripleva & Arseneva, 2015), quality (Zinina et al., 2018) and biosafety (Surai et al., 2017). High productivity and product quality parameters (Surai & Fisinin, 2016a) can only be obtained from healthy birds (Surai et al., 2016b). Growing chickens on an industrial basis is accompanied by a serious risk of infectious diseases of viral or bacterial origin (Zhuravel & Miftakhutdinov, 2016), vaccination is the basis for protecting poultry stock. When carrying out preventive vaccination, numerous factors should be taken into account that

may reduce the effectiveness of vaccination, including immunosuppressive conditions which can be caused by stress (Djavadov & Dmitrieva, 2012).

Reaction to physiological stress starts when brain detects a homeostatic problem and activates sympathetic nervous system which secretes catecholamines, adrenaline and norepinephrine. This is followed by a slower activation of hypothalamic-pituitary-adrenal axis. A variety of regulatory systems and mechanisms cannot lead to an entirely predictable effect; moreover, functioning of hypothalamic-pituitary-adrenal axis demonstrates significant individual variability (Sorrells et al., 2009). In regard to this study, the primary focus in theoretical and especially in practical aspects of this process should be on the effect of glucocorticoid hormones and stress on immune system and on inflammatory response (Sapolsky, 2004). Chronic exposure to glucocorticoids suppresses both innate and adaptive immunity; prolonged exposure to glucocorticoid hormones was proved to decrease the number of circulating WBC (Reichardt et al., 2001) and to reduce the production of a large number of pro-inflammatory cytokines, including IL-1 β – interleukin 1 beta and TNF- α – tumor necrosis factor-alpha (De Bosscher et al., 2003). Although vaccination of birds is essential (Anosov et al., 2015; Kuklenkova et al., 2018b; Javadov et al., 2019) it remains a stress factor in poultry industry (Fisinin & Kavtarashvili, 2015b; Surai & Fisinin, 2016b; Zhuravel et al., 2017). This leads to a decrease in the level and homogeneity of immune response (Ponomarenko, 2015). Currently, one tries to develop different methods and tools to enhance the immune response during vaccination, as demonstrated by increased titer of antibodies to avian viral pathogens (Ali et al., 2013; Xu et al., 2018, Eladl et al., 2019; Semenov et al., 2019; Walaa & Awadin, et al., 2020). There are specific studies that prove the positive effect of pharmacological anti-stress drugs during immunization of birds (Miftakhutdinov, & Amineva, 2019). Stress prevention in the vaccination process is a new scientific field, which can ensure efficiency increase of poultry industry (Miftakhutdinov & Amineva, 2019). Aside of efficiency, the economic effect of measures carried out has a significant value for the poultry industry.

SPAO-complex (stress protector antioxidant – complex preparation for birds) and SPAO.

These preparations contain an active complex of micro elements, vitamins, vitamin-like and other substances that have an effect on metabolism (Miftakhutdinov et al., 2019; Zhuravelet al., 2019a; Zhuravel et al., 2019b).

In connection with the above matter, the aim of our research was to determine and comparatively evaluate the effectiveness of using SPAO-FA feed additive and SPAO-complex pharmacological preparation for the prevention of vaccine stress during immunization of birds against reovirus tenosynovitis.

MATERIALS AND METHODS

The studies were carried out in a poultry farm of industrial type. **The object** of this study was chicken parent flock; **the subject** – parameters of humoral response to the injected live vaccine against reovirus tenosynovitis, strain 1133, with underlying giving SPAO-FA (stress protector antioxidant – feed additive) and SPAO-complex (stress protector antioxidant – complex preparation for birds) to the birds.

Drugs are developed to be used for animals and are experimentally applied in the conditions of industrial-grade poultry farms to prevent preslaughter and transportation

stresses. Preliminary tests have shown capabilities of the drugs studied to cause mediated impact on humoral immunity formation processes in vaccinations; it is proven that under the effect of SPAO-complex the protective antibodies level in terms of vaccinal prevention of rednose, infectious bronchitis, bursal disease, Newcastle disease are reliably higher than control values (Ponomarenko, 2015; Miftakhutdinov & Amineva, (2019).

Study scheme

Birds were vaccinated with a live vaccine for reovirus tenosynovitis from strain 1133 at the age of 7 and 28 days, followed by re-vaccination with an inactivated vaccine at the age of 9–11 weeks. Baseline livestock of 32,000 birds was divided into three groups. Birds of experimental group 1 received basic diet and were a control group. Birds of experimental group 2 received SPAO-FA feed additive as a part of their diet. Birds of experimental group 3 were given SPAO complex pharmacological preparation with water. Feed additive and pharmacological preparation were used for 5 days: 3 days before vaccination, day of vaccination and one day after vaccination at the dose of 185 mg kg⁻¹ of body weight.

Study results evaluation

Vaccination results were interpreted by the level of specific antibodies formation and homogeneity in titers, as well as by vaccination index (Olson,1978). Humoral immunity was evaluated 7 and 14 days after the first vaccination and 14 and 21 days after the second one.

Blood serum was used as the material studied. Antibodies generation evaluation, analysis and statistical processing were performed using BioChek (the Netherlands) test systems and BioChek II software. Results recording was done using Tecan (Austria) spectrophotometer with 405 nm wave length (The Avian Reovirus Antibody test kit). Vaccination index was set by the ratio of antibodies average titre and variation coefficient.

Cost-effectiveness was evaluated according to the method developed for poultry farming (Zhuravel & Miftakhutdinov, 2015; Fisinin et al., 2018; Zhuravel & Miftakhutdinov, 2019), thus, defining the following parameters: cost-effectiveness per RUR of expenses (ratio of benefit to veterinary expenses), benefit from veterinary measures (the difference between benefit, including prevented damage and the additional cost of production, and veterinary expenses), prevented damage (based on livability of chickens in parent flock), the cost of products obtained further by increasing its amount or quality (day-old chicks and chicken meat), veterinary expenses (material and labor costs).

RESULTS AND DISCUSSION

The level of specific antibodies indicates the level of response to vaccination (Table 1).

So, 7 days after the first vaccination, that is, at the initial stage of immunity development, antibody titer was several times lower in comparison with both a protective level and expected, or baseline. At the same time, antibody titer in birds that received

SPAO-FA and SPAO-complex was 1.91 times higher. This may indicate increased priming.

Table 1. Average titer of antibodies and coefficient of variation for vaccination of chickens against reovirus tenosynovitis

Age of bird, days	Post vaccination period	Expected		Basic diet		SPAO-FA		SPAO complex	
		titer	CV	average titer	CV	average titer	CV	average titer	CV
14	7 ₁	2,000–5,000	40–80	331 ± 304.8	92	631, ± 375.7	60	621 ± 294.22	47
21	14 ₁	2,000–5,000	40–80	377 ± 305.7	81	716 ± 388.2	54	1,088 ± 420.2	39*
42	14 ₂	2,000–5,000	40–80	1,505 ± 707.8	47	2,453 ± 1,047.3	45*	4,554 ± 1,255.4	28*
49	21 ₂	2,000–5,000	40–80	1,749 ± 603.4*	35*	2,836 ± 986.2*	35*	4,603 ± 1,177.9*	26*

Note: * Level corresponding to ‘protective’ one.

14 days after immunization, values in the groups of SPAO-FA and SPAO-complex were also higher in comparison with antibody titer in the group with basic diet and also higher than the level of antibodies in an earlier period, at the initial stage of immunity development, i.e. 7 days after vaccination. In group 1, this difference was insignificant and amounted to 1.81%, in group 2 – 13.47%, in group 3 – 75.2%. This demonstrates a response to vaccination at the first stage of immunization. Positive effect of SPAO-FA and SPAO-complex on the decreasing level of immunosuppression is confirmed with the relatively high titer of antibodies on the 14th day after vaccination in comparison with experimental group 1 – in 2.12 and 3.23 times.

After the second vaccination, antibody titer in blood serum of the studied chickens changed. So, 14 days after the second vaccination, the average titer in experimental group 1 increased by 4.47 times, after 21 days – by another 1.16% and reached ‘protective’ level, although it was lower than expected. Apparently, this was due to the immunosuppression that occurred as a result of activated stress-triggering mechanisms during vaccination (Ponomarenko, 2015).

Using feed additives and drugs aimed at stress prevention contributed to a more intensive development of antibodies.

So, with adding SPAO-FA 14 days after vaccination, antibody titer was 62.99% higher than this in the blood of chickens of group 1, and reached the expected level (slightly above the minimum) and became higher than the protective one. 21 days after vaccination (term for monitoring immunity level), it increased by 16.7% and became 62.15% higher in comparison with the same parameter in the control group.

It should be noted that the level of antibodies corresponding to the protective one was not reached after the first vaccination. Immunity level that protects birds against the infection caused by a field strain of the causative agent of reovirus tenosynovitis, should be considered with an antibody titer more than 1:800 (Kuklenkova et al., 2018a). Therefore, using SPAO-complex helps to achieve this value by 21 days after the first vaccination with a live vaccine. In addition, the level of antibodies reached during an experimental test of live vaccines containing virus strains (Radyush et al., 2013) to

different extents corresponded to the parameters obtained in experimental groups 2 and 3 that demonstrated immune response.

Using SPAO-complex contributed to a more intensive immunity development. 14 days after the second vaccination, antibody titers were close to the maximum expected level and were higher than in blood serum of chickens of experimental groups 1 and 2 by 3.06 and 1.86 times, respectively. By the 21st day after the second vaccination, this value increased slightly and was also higher by 2.6 and 1.6 times, respectively. This shows that SPAO complex contributes to seroconversion and accelerates the production of antibodies.

The degree of titer homogeneity is proved by the coefficient of variation; we can judge about the quality of vaccination in the batch of birds by its level. So, vaccination being a technological stress reduces the activity of specific humoral immunity, as demonstrated by the value of the coefficient of variation. 7 days after the first vaccination, this value was maximal, in control group – above the expected level. Using SPAO-FA and SPAO-complex allowed achieving a higher level of homogeneity – coefficient of variation in groups 2 and 3 was lower than in group 1 by 65.22% and 48.91%, respectively, and was within the expected range.

14 days after the first vaccination, a decrease in the coefficient of variation in each group was noted.

The smallest value was in the group of SPAO-complex; it was lower in comparison with vaccination according to the scheme adopted at the farm by 2.08 times; coefficient of variation in the group of SPAO-FA was 1.5 times lower than in experimental group 1. Moreover, vaccination homogeneity was achieved only in group 3 (coefficient of variation was lower than 45%).

Therefore, SPAO-FA and SPAO-complex contribute to vaccination homogeneity to a greater extent. At the same time, the effect of SPAO-FA is less pronounced than that of SPAO-complex. Both drugs shorten the time to reach the level of coefficient of variation that indicates homogeneity (< 45) – to 14th day after the second vaccination. In addition, using SPAO-complex helps to reduce the coefficient by the term of monitoring immunity level, i.e. to 21st day after the second vaccination, by 25.7%. Using SPAO-KD in comparison with basic diet has no effect on the coefficient of variation by 21st day after the second vaccination.

A sign of successful vaccination is the ratio of analyzed parameters: average antibody titer and coefficient of variation, i.e. vaccination index (van Lirdam & Bosman, 2011).

Vaccination index is a logical parameter that demonstrates high values with a successful vaccination. Data in Table 2 allow concluding that immunity level gradually increases reaching its maximum to the checkpoint – 21 days after the second vaccination (Table 2).

At the same time, better seroconversion was noted in the group of SPAO-complex; vaccination index in the blood of chickens of experimental group 3 is 3.5 times higher than in experimental group 1 and 2.18 times higher than in experimental group 2.

Table 2. Reovirus tenosynovitis vaccination index

Post vaccination period	Vaccination	Vaccine + SPAO-FA	Vaccine + SPAO complex
7 ₁	3.60	10.52	13.21
14 ₁	4.16	13.26	27.90
14 ₂	32.02	54.51	162.64
21 ₂	49.97	81.03	177.04

Study results – titer level and homogeneity – confirmed by vaccination index allow concluding that the development of antibodies occurred as a result of the action of vaccine; no information for the development of infectious process was found.

With strict compliance with the requirements for immunization and adequate immune status of birds, coefficient of variation will be low within normal limits. A high value of the coefficient of variation is due to both a low antibody titer, for example, if treatment plan was not followed, and high antibody titer that may be associated with the circulation of a field virus (Miftakhutdinov & Amineva, 2019).

Considering specific features of the technological process in poultry farming, a contradiction should be noted. On the one hand, vaccination is aimed at creating the immunity of poultry flocks to infectious diseases including reovirus tenosynovitis and on the other hand, it, as a stress factor, triggers the inhibition of antibody production what leads to decreased activity of specific humoral immunity. Negative association between stress and antibody response that confirms these studies is described in the number of works (Burns et al., 2003; Segerstrom & Miller, 2004; Auerbach et al., 2014). Effect of antistress drugs on achieving a high immune response during vaccination was also proven (Ponomarenko, 2015; Miftakhutdinov & Amineva, 2019).

Production parameters allow concluding about positive effects of the pharmacological complex and the feed additive that were used for anti-stress treatment.

So, by the time of reaching 140 days of age, that is, during the period when birds undergo intensive immunization, also against reovirus theosinovitis, the livability of birds in experimental group 1 was 97.2%, in experimental group 2–97.6%, in experimental group 3–97.8%. In the period of egg production (from 140 days of age to slaughtering at the age of 61 weeks) characterized with a relatively lower immunological load, the livability of laying hens also differed and amounted to 90.9, 91.2 and 93.5%, respectively. In general, over the period of growing and egg production, the livability of laying hens was higher with using the feed additive and the pharmacological complex. So, livability in experimental group 2 was 89.1% what is higher than in experimental group 1 by 0.79%. In experimental group 3, livability amounted to 91.5% what is higher than in experimental group 1 by 3.5% (Table 3).

In the group that received SPAO-complex, production of hatching eggs per hen increased by 3.24%, the number of day-old chicks – by 4.22%. In the group of SPAO-FA, the increase in production parameters was less significant: production of hatching eggs was higher by 0.22%, of day-old chicks – by 0.55%

Therefore, using feed additive and pharmacological preparation as an anti-stress treatment helps to reduce mortality and to increase the livability of livestock, as well as to increase final production parameters – hatching eggs, and, consequently, the number of day-old chicks.

In second-order breeding units, birds after using are slaughtered; the meat is used for preparing ground meat, or carcasses are sold, therefore, the weight of birds before slaughtering and meat yield are important parameters. Using feed additive and preparation had no effect on meat yield but at the same time, the live weight of chickens that received SPAO-FA was higher than the live weight of chickens received basic diet by 23 g, or 0.88%; and the live weight of chickens received SPAO-complex – by 167 g, or 6.44%. It was proved that live weight of birds immunized against reovirus tenosynovitis was significantly lower than that of non-immunized ones (Lazovskaya & Prudnikov, 2015). Vaccination being a technological stress reduces the gain in live

weight while using feed additive and pharmacological complex reduces the negative impact of stress factor and contributes to an increase in production parameters. In this case, the feed additive has less effect than the pharmacological complex.

Table 3. Production parameters of chickens with using SPAO-complex and SPAO-FA

Parameter	Basic diet	SPAO-FA	SPAO complex
Baseline number of housed chickens, birds	32,000	32,000	32,000
Number of laying hens (140 days), birds	31,110	31,250	31,310
Number of laying hens at the end of technological cycle, birds	28,279	28,500	29,275
Gross egg production, thousand pcs.	5,913	5,959	6,119
including hatching eggs, thousand pcs.	5,644	5,697	5,879
Yield of hatching egg, %	95.5	95.6	96.1
Eggs obtained from laying hen, total, pcs.	190.2	190.7	195.4
including hatching eggs, pcs.	181.9	182.3	187.8
Chickens obtained from laying hen, birds	144.3	145.1	150.4
Hatching rate, %	79.3	79.6	80.1
Livability of laying hens during egg production, %	90.9	91.2	93.5
Livability of laying hens during technological cycle, %	88.4	89.1	91.5
Weight of a bird at the end of the technological cycle, g	2,590	2,613	2,757
Meat yield, %	75.1	75.1	75.2
Feed consumption for the production of hatching eggs, kg/10 pcs.	2.68	2.67	2.62

Using SPAO-FA feed additive and SPAO-complex pharmacological preparation for chickens of the parent flock in the course of their vaccination against reovirus infection reduces the negative effect of vaccination as a stress factor and stimulates immune response.

During this experiment, 10.09 kg of SPAO-FA and 10.09 kg of SPAO-complex were consumed. Considering the cost of preparations, material expenses for SPAO-FA (experimental group 2) amounted to 18,168.48 RUR, for SPAO-complex (experimental group 3) – 37,346.32 RUR. SPAO-complex was given with water, with the help of a medicator; time spent on preparing the drug for use (weighing, putting in a drinking system) was about 10 minutes per day, i.e. 100 minutes, or 1.67 hours for the whole study period. SPAO-FA was given with mixed feed; time spent was slightly less and amounted to 90 minutes, or 1.5 hours for the whole study period. Average salary of a veterinarian at the poultry farm was 50,000 RUR per month; taking into account its size per year (600,000 RUR) and annual working hours (1761.4 hours), salary per hour amounted to 340.64 RUR. Therefore, the cost of labor when using SPAO-complex amounted to 568.87 RUR, SPAO-FA – 510.96 RUR, the charges were 154.31 and 171.80 RUR, respectively. Thus, veterinary expenses for experimental group 2 amounted to 18,833.75 RUR, for experimental group 3 – 38,086.99 RUR.

Prevented damage amounted to 163,540 RUR for experimental group 2, and 737,040 RUR for experimental group 3. The cost of additionally obtained chicks in experimental group 2 was 3,164,140 RUR, in experimental group 3 – 15,389,570 RUR.

Taking into account the livestock by the end of hatching egg production cycle for one batch, the average weight of birds before slaughter, as well as meat yield, 55,005.2 kg of meat were obtained in experimental group 1, 55,927.3 kg – in experimental group 2, and 60,694.8 kg – in experimental group 3. Therefore, the cost of

additionally obtained poultry meat amounted to 82,989 RUR in experimental group 2, and 512,064 RUR in experimental group 3.

In general, the additional value in experimental group 2 was 3,247,129.0 RUR, in experimental group 3 – 15,901,634.0 RUR.

Benefit in experimental groups 2 and 3 amounted to 3,391,835.25 RUR and 16,600,587.01 RUR. Cost-effectiveness per RUR of costs was 180.09 RUR and 435.86 RUR, respectively. Therefore, the effectiveness of SPAO-complex is 2.4 times higher in comparison with SPAO-FA.

As a result of the positive influence of SPAO-FA and SPAO-complex on bird organism during exposure to technological stresses, a high economic benefit was achieved due to the livability of the livestock and increased productivity. Therefore, the pronounced immunotropic effect of SPAO-complex and the less pronounced immunotropic effect of SPAO-FA during the development of stress is confirmed by the high level of production parameters and, as a result, level of cost-effectiveness of introducing anti-stress treatment into the hatching egg production cycle.

To prevent post-vaccination stress in order to ensure a homogeneous and high level of bird protection from the causative agent of reovirus tenosynovitis and to increase production parameters, we recommend using SPAO-complex (stress protector antioxidant – complex preparation for birds) and SPAO-FA (stress protector antioxidant – feed additive) for 5 days: 3 days before vaccination, on the day of vaccination and one day after vaccination at the dose of 185 mg kg⁻¹ of body weight.

CONCLUSION

1. Stress prophylaxis in chickens of parent flock through the use of SPAO-FA and SPAO-complex during vaccination against reovirus infection provides a homogeneous and high level of bird protection against field virus: it reduces the time required for development of protective antibody level, contributes to more intensive achievement of vaccination homogeneity, as demonstrated by an increase in production parameters. A more significant effect was achieved when using SPAO-complex.

2. During using SPAO-FA and SPAO-complex, antibody titers reached the expected level and were higher than the protective level by the 14th day after the second vaccination, in the birds that received basic diet – by the 21st day after the second vaccination. By the 21st day after the second vaccination, antibody titers in animal that were given SPAO-FA were 1.6 times higher than in animal that received basic diet; and in group of SPAO-complex – 2.6 times higher.

3. SPAO-FA and SPAO-complex used during vaccination shorten the time of reaching the level of coefficient of variation that indicates homogeneity (< 45) by the 14th day after the second vaccination. Using SPAO-complex helps to reduce coefficient of variation by the time of monitoring immunity level, i.e. by 21st day after the second vaccination, by 25.7%. Using SPAO-FA in comparison with basic diet had no effect on the coefficient of variation by this time.

4. Vaccination index that should be of high values during successful vaccination was 3.5 times higher when using SPAO-complex in comparison with basic diet; that of SPAO-FA was 1.6 times higher.

5. When using SPAO-complex, livability of chickens was higher by 3.5%, live weight of poultry – by 6.44%, number of day-old chicks – by 4.22%; these parameters with using SPAO-FA were higher by 0.79%, 0.88% and 0.55%, respectively.

6. Cost-effectiveness of using SPAO-FA per 1 RUR of expenses amounted to 180.09 RUR, for SPAO-complex it amounted to 435.86 RUR.

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