



Feed additive “Activo” influence on several antioxidant defense system parameters of sows and their progeny

T. Y. Prudyus

tarasvet126@gmail.com



Institute of Animal Biology NAAS, 38 V. Stus str., Lviv, 79034, Ukraine

ORCID:

T. Y. Prudyus <https://orcid.org/0000-0003-3594-7539>

Authors' Contributions:

PTY: Conceptualization; Methodology; Investigation; Data curation; Formal analysis; Validation; Visualization; Writing — original draft, review & editing.

Declaration of Conflict of Interests:

None to declare.

Ethical approval:

The research methodology was approved by the Bioethical commission in the Institute of Animal Biology (National Academy of Sciences of Ukraine), Protocol no. 167 from January 7, 2025.

Acknowledgements:

None.



Attribution 4.0 International
(CC BY 4.0)

Intensive farming technologies are developed primarily to maximise economic benefits. At the same time, productive animals are often exposed to many stress factors and excessive physiological stress, which subsequently leads to a deterioration in animal welfare, health and, ultimately, a decrease in productive performance. Oxidative stress is one of the most common negative processes that occur in the animal body in the result of various factors, namely, a violation of the pro- and antioxidant balance. This does not occur on its own, but is mainly an accompanying process under the influence of various nature stress factors, such as toxins, temperatures, critical physiological periods, infectious pathologies, excessive, unbalanced or insufficient feeding, etc. This article elucidates the results of the feed additive “Activo” effect studies, that contains secondary plant components of oregano and chilli pepper essential oils on the quantitative content of lipid peroxidation products and the activity of antioxidant defence systems in sows and piglets obtained from them. The study was carried out in the conditions of the industrial complex *Barkom LLC*, Lviv region. Based on the principle of analogues, two groups of Large White breed gestating sows were formed, 10 animals per group — control and experimental. For the experimental group, the feed additive “Activo” was additionally introduced into the diet in the amount of 0.1 kg/t of finished feed. The content of lipid hydroperoxides and TBA-active products, glutathione peroxidase and superoxide dismutase activity were determined in the blood of sows and their progeny. The results of our studies showed that the use of the feed additive “Activo” for sows and piglets obtained from them is helpful to reduce the negative effects of oxidative stress, which has a negative effect on both sows and their offspring.

Key words: sows, piglets, feed additive “Activo”, essential oils, oxidative stress, superoxide dismutase, glutathione peroxidase, TBA-active products, lipid hydroperoxides

Introduction

Modern livestock farming technologies, and pig farming in particular, are primarily aimed at producing the maximum amount of product in the shortest possible

time. Such approaches are primarily driven by economic efficiency, which is the goal of the vast majority of producers. At the same time, they often do not take into account the very high physiological stresses, sometimes on the verge of pathological disorders, that animals experience

during their industrial rearing and maintenance. An animal organism has biological limits to its growth, development, reproduction, etc. and healthy viability in general. In recent years, in addition to the emergence and spread of new diseases of various etiologies, including infectious diseases (both viral and bacterial), pigs and piglets often suffer from various physiological disorders caused by overly intensive housing and feeding technologies. It is also worth noting changes in the genetic potential of pigs, such as high fertility, multiple births, reduced fattening time, composition and properties of feed, premixes, veterinary preparations, vaccines, etc. All of this leads to excessive stress factors in pigs, which in turn often leads to a decrease in their immunity and negatively affects the health and welfare of animals, ultimately reducing the quality and quantity of their productivity.

Stressful factors in the animal bodies often result in an increase in the so-called reactive oxygen species (ROS) and free radicals formation, and a decrease in the antioxidant defence of the body [1]. There are many types of ROS, including singlet oxygen, hydrogen peroxide (H_2O_2) and oxygen radicals — superoxide anion radical ($O_2^{\cdot-}$), hydroperoxide (HO_2^{\cdot}) and hydroxyl radical (OH^{\cdot}), which are formed in the process of reducing molecular oxygen. Oxidative stress and ROS are often interrelated with a wide range of pathological processes in the body.

Many studies have shown that the periods of sow gestation, piglet birth, lactation, and weaning are characterised by increased oxidative stress parameters [2], which subsequently negatively affects milk synthesis, reproductive functions, and shortens the period of productive use of sows [5]. In addition, numerous studies have established the fact that excessive ROS production can eventually lead to insulin resistance [11], which has a negative impact during the prenatal and lactation periods of sows, which significantly reduces animal feed intake [10, 15].

Piglets are exposed to stress during and after birth, which also increases the amount of ROS in the body. Newborn piglets often have a reduced level of passive immunity obtained with sow colostrum, which, in combination with underdeveloped active immunity, can have negative consequences for their health and survival.

In order to reduce negative stress effects, mitigate them and normalise animal homeostasis, scientists from many countries are actively working to find and develop new highly effective and at the same time safe and affordable stress-protective agents, including antioxidants. In recent years, various components of biological, microbiological and plant origin have been increasingly used for this purpose. Preparations and feed additives based on such substances should have as little toxicity and side effects as possible, unlike many artificially synthesised factors.

In this regard, the use of several plant essential oils (PEs) is promising. Essential oils are a large class of secondary plant metabolites that often have high antioxidant capacity. All (EOs) are structurally characterised by aromatic rings with several hydrogen hydroxyl groups, which

makes EOs suitable for neutralising free radicals and other reactive oxygen species. In the last decade, more and more attention has been paid to EOs as functional components of feed additives for sows and piglets. In particular, a number of studies have shown that EOs can have an anti-inflammatory effect and reduce oxidative stress in sows, increase their reproductive capacity and promote the growth and development of their progeny [3].

The aim of the study was to investigate the effect of the feed additive "Activo", which contains secondary plant components of oregano and chilli pepper essential oils, on the lipid peroxidation products quantitative content and the activity of antioxidant defence systems in sows and piglets obtained from them.

Materials and Methods

The study was conducted in the conditions of the industrial complex of *Barkom LLC*, Lviv region (Ukraine). Two groups of the Large White breed gestating sows were formed according to the principle of analogues, 10 animals per group — control and experimental. The animals were kept in the same box in individual farrowing pens under the same conditions. The control and experimental groups of animals were fed a feed balanced in terms of biologically active substances. For the experimental group, the feed additive "Activo" was additionally introduced into the diet in the amount of 0.1 kg/t of finished feed. The experiment started on the 85th day of gestation or 30 days before the expected farrowing. Blood samples were taken from the jugular vein of pregnant sows before the experiment. The sows were transferred to the maternity ward, 5 days before the planned farrowing, where they were fed lactating sow feed, and the animals in the experimental group continued to receive the specified feed additive. Lactating sows were fed in individual troughs four times a day in the amount of 7 kg. Blood samples were taken from the jugular vein for research three days before the expected farrowing, as well as on the 21st day of lactation. Piglets obtained from sows of the control and experimental groups automatically became piglets of the control and experimental groups. The piglets were fed pre-starter feed starting from the fifth day of life. The piglets of the experimental group were administered the specified feed additive in the amount of 0.2 kg/t of finished feed in the pre-start feed. The finished feed was fed from day 5 after birth until weaning. Blood samples for biochemical studies were taken from the cranial *vena cava* of newborn suckling piglets at 5th, 14th and 28th days of age.

In the blood of sows and piglets we determined the parameters: the lipid hydroperoxides and TBA-active products content, the activity of glutathione peroxidase and superoxide dismutase, by the methods described [11, 14]. Namely, the lipid hydroperoxides content in blood plasma was determined by a method based on the optical density spectrophotometric measurement of reaction products with ammonium thiocyanate, Mohr's salt and hydrochloric

acid. The concentration of TBA-active products, which characterises the lipid peroxidation rate, was determined by a method based on the reaction between MDA and thiobarbituric acid (TBA), which at high temperature and acidic environment proceeds with the formation of a coloured trimethyl complex containing one MDA molecule and two TBA molecules. Glutathione peroxidase activity was determined by a method based on a comparative study of the degree of reduced glutathione oxidation (GSH) by tertiary butyl peroxide. Superoxide dismutase (SOD) activity was determined by the method based on the reduction of nitroblue tetrazolium to nitroformazone by superoxide anion radicals formed in the reaction between phenazine methanesulfate and the reduced form of NADPH.

The results were processed by standard methods of mathematical statistics using *Microsoft Excel* software. Statistical significance was determined by the Student's test.

The study is fully complied with the ethical requirements for the use of animals in experimental research (Strasbourg, 1986; Kyiv, 2002), and the study methodology is approved by the Bioethics Committee of the Institute of Animal Biology of the National Academy of Sciences of Ukraine.

Results and Discussion

Sows are exposed to various stressors, including pregnancy, lactation, weaning, estrus and insemination throughout their productive lives. One of the most stressful periods is the farrowing period, during which sows undergo significant physiological changes associated with zygote formation, embryo growth, and subsequently farrowing and lactation. The most significant oxidative stress occurs during pregnancy, as the placenta is the site of active oxygen metabolism [16]. Scientists have proved that oxidative stress and lipid peroxidation are stronger and more intense in pregnant animals compared to non-pregnant animals [6]. Excessive formation of reactive nitrogen species and reactive oxygen species disrupts the normal physiological function of the mammalian placenta and affects most other physiological processes [16].

According to the results of our studies with the use of the feed additive "Activo" in gestating and lactating sows, the lipid peroxide products (LPP) and TBA-active products content in their blood compared to the control was significantly lower in the experimental group animals (table 1). Starting from the first blood sampling period and the beginning of the experimental feed additive introduce, it can be seen that with the systematic consumption of mixed fodder in the blood of the experimental group sows, the indicators of TBA-active products significantly decreased. TBA-active products indicators in the control group slightly decreased at the beginning of the 85th day of farrowing, but increased on the 112th day and decreased on the 21st day of lactation. The increase of

parameters on day 112 of gestation can be explained by a abrupt increase in fetal development. But, as can be seen from the results obtained in the experimental group, the indicators of oxidative processes were systematically statistically significantly reduced: on the 85th, 112th day of pregnancy, and 21st day of lactation by 16.78 % ($P<0.05$), 22.65 % ($P<0.001$), 24.89 % ($P<0.001$) compared to the control group, respectively.

Thus, analysing the feed additive "Activo" use, which contains components from oregano, chilli, cinnamon and rosemary, in particular, such as carvacrol, thymol, we see that these components have a great potential to enhance antioxidant protection and the ability to reduce oxidative stress in sows. Analysing similar works by other authors, we can say that this, in turn, has a positive effect on the embryonic and post-embryonic development of piglets [4, 8], development and function of the gastrointestinal tract [9], body formation [4] and acid-base balance [14], etc. A number of studies also show that the addition of oregano, chilli, carvacrol, and thymol to sow feed has an effect on the antioxidant function of suckling piglets [3].

An interesting natural tendency in the presence of lipid peroxidation products — hydroperoxides (HPO) and TBA-active products, was noted in the study of young piglets blood. According to the estimation of total lipid peroxidation (LPP), we see a tendency to their gradual decrease (table 2).

In particular, it was admitted that piglets of the experimental group had significantly lower level of TBA-active products on the 14th day of life of 6.28 % ($P<0.01$) compared to the control group. Regarding the content of lipid hydroperoxides, in piglets of the experimental group, statistically lower values were found on the 5th and 14th day of life 26.42 % ($P<0.05$), 19.23 % ($P<0.01$), respectively, compared to the control group. Since piglets born from sows of the experimental group consumed only colostrum and milk from birth to 5 days of life and showed lower HPL and TBARS, this may indicate the transfer of active components of essential oils to piglets through colostrum. We can also see that piglets' consumption of sows milk in the experimental group and starter feed containing oregano, chilli, carvacrol, thymol led to an even lower reduction in LPPs. The obtained results indicate the inhibitory effect of the studied feed additive on the intensity of the lipid peroxidation processes.

According to the results of glutathione peroxidase and superoxide dismutase determination in sows' erythrocytes, it was found increased activity of these enzymes (table 3). During the period of the "Activo" feed additive use, a significantly higher level of COD activity was found in animals of the experimental group on the 21st day of lactation by 21 % ($P<0.05$) compared to the control.

Furthermore, in piglet blood, superoxide dismutase (SOD) is the primary antioxidant, which maintains and controls the free radicals level and creates conditions for the normal use of the oxygen environment of the body. This enzyme deactivates reactive oxygen species, which degrade to hydrogen peroxide [13].

Table 1. The content of lipid peroxidation products in the blood plasma of sows (M±m, n=5)

Indexes	Groups	Periods of experiment		
		85 days in gestation	112 days in gestation	21 days in lactation
TBARS, µmol/ml	C	4.31±0.103	4.37±0.111	4.26±0.143
	E	3.5868±0.0304*	3.38±0.068***	3.1998±0.091***
HPL, Units. E/ml	C	2.58±0.110	2.60±0.045	2.42±0.035
	E	2.261±0.054**	2.241±0.057***	2.150±0.041***

Note. Here and further * — P<0.05, ** — P<0.01, *** — P<0.001 compared to the control group.

Table 2. The content of lipid peroxidation products in the blood plasma of suckling piglets (M±m, n=5)

Indexes	Groups	Periods of experiment		
		5 th day of life	14 th day of life	28 th day of life
TBARS, µmol/ml	C	4.8167±0.054	4.3323±0.03	3.9424±0.126
	E	4.4501±0.128	4.0599±0.061**	3.480±0.184
HPL, Units. E/ml	C	0.53±0.02	0.52±0.01	0.489±0.02
	E	0.39±0.03*	0.42±0.03*	0.436±0.019

Table 3. Activity of glutathione peroxidase and superoxide dismutase in blood erythrocytes of sows (M±m, n=5)

Indexes	Groups	Periods of experiment		
		85 days in gestation	112 days in gestation	21 days in lactation
Glutathione peroxidase, nM NADPH/min./mg Hb RBS	C	10.55±0.21	7.39±0.25	9.73±0.09
	E	11.18±0.40	6.32 ±0.53	10.23±0.23
Superoxide dismutase, units Act/mg protein×min	C	21.54±3.61	19.91±1.84	22.15±1.57
	E	22.86±1.64	23.81±1.41	26.81±1.20*

Table 4. Activity of glutathione peroxidase and superoxide dismutase in blood erythrocytes of suckling piglets (M±m, n=5)

Indexes	Group	Periods of experiment		
		5 days of life	14 days of life	28 days of life
Glutathione peroxidase, nM NADPH/min./mg Hb RBS	C	15.1±0.31	13.45±0.19	13.86±0.17
	E	15.5±0.81	16.1±0.68**	17.0±0.72**
Superoxide dismutase, units Act/mg protein×min	C	22.40±1.18	23.36±1.07	26.30±1.25
	E	24.43±0.69	25.56±0.75	31.12±1.03*

From the table 4 data, we can see that the feed additive "Activo" exhibits antioxidant properties, which led to a decrease in the free radical oxidation activity degree, which led to a decrease in the use of GSH (glutathione peroxidase) and a statistical increase in its use in erythrocytes in the experimental group on the 14th and 28th day by 19.7 % (P<0.01), 22.7 % (P<0.01), respectively, com-

pared to the control. This explains the increase in protective and detoxifying properties in the body.

To sum up, the results of our studies have shown that the use of the "Activo" feed additive for sows and piglets obtained from them is advisable to reduce the negative effects of oxidative stress, which has a negative effect on both sows and their progeny.

References

1. Agarwal A, Makker K, Sharma R. Clinical relevance of oxidative stress in male factor infertility: An update. *Am J Reprod Immunol*. 2008; 59 (1): 2–11. DOI: 10.1111/j.1600-0897.2007.00559.x.
2. Berchieri-Ronchi CB, Kim SW, Zhao Y, Correa CR, Yeum KJ, Ferreira ALA. Oxidative stress status of highly prolific sows during gestation and lactation. *Animal*. 2011; 5 (11): 1774–1779. DOI: 10.1017/S1751731111000772.
3. Chen J, Huang Z, Cao X, Zou T, You J, Guan W. Plant-derived polyphenols in sow nutrition. *Anim Nutr*. 2023; 12: 96–107. DOI: 10.1016/j.aninu.2022.08.015.
4. Garcia-Contreras C, Vazquez-Gomez M, Barbero A, Pesantez JL, Zinellu A, Berlinguer F, Gonzalez-Añover P, Gonzalez J, Encinas T, Torres-Rovira L, Nuñez Y, Ballesteros J, Ayuso M, Astiz S, Isabel B, Ovilo C, Gonzalez-Bulnes A. Polyphenols and IUGR pregnancies: Effects of maternal hydroxytyrosol supplementation on placental gene expression and fetal antioxidant status, DNA-methylation and phenotype *Int J Mol Sci*. 2019; 20 (5): 1187. DOI: 10.3390/ijms20051187.
5. Lapointe J. Mitochondria as promising targets for nutritional interventions aiming to improve performance and longevity of sows. *J Anim Physiol Anim Nutr*. 2014; 98 (5): 809–821. DOI: 10.1111/jpn.12160.
6. Mihu D, Sabău L, Costin N, Ciortea R, Măluțan A, Mihu CM. Implications of maternal systemic oxidative stress in normal pregnancy and in pregnancy complicated by preeclampsia. *J Matern Fetal Neonatal Med*. 2012; 25 (7): 944–951. DOI: 10.3109/14767058.2011.600796.
7. Mosnier E, le Floch N, Etienne M, Ramaekers P, Sève B, Père MC. Reduced feed intake of lactating primiparous sows is associated with increased insulin resistance during the peripartum period and is not modified through supplementation with dietary tryptophan. *J Anim Sci*. 2010; 88 (2): 612–625. DOI: 10.2527/jas.2008-1768.
8. Parraguez VH, Sales F, Peralta OA, De los Reyes M, Campos A, González J, Peralta W, Cabezon C, González-Bulnes A. Maternal supplementation with herbal antioxidants during pregnancy in swine. *Antiox*. 2021; 10 (5): 658. DOI: 10.3390/antiox10050658.
9. Prudyus T. Morphological characteristics of the duodenum of piglets fed with various feed additives. *Reg Mech Biosys*. 2023; 14 (2): 266–272. DOI: 10.15421/022339.
10. Rains JL, Jain SK. Oxidative stress, insulin signaling, and diabetes. *Free Rad Biol Med*. 2011; 50 (5): 567–575. DOI: 10.1016/j.freeradbiomed.2010.12.006.
11. Rosalovsky VP, Grabovska SV, Salyha YT. Changes in glutathione system and lipid peroxidation in rat blood during the first hour after chlorpyrifos exposure. *Ukr Biochem J*. 2015; 87 (5): 124–132. DOI: 10.15407/ubj87.05.124.
12. Salyha N. Effects of L-glutamic acid and pyridoxine on glutathione depletion and lipid peroxidation generated by epinephrine-induced stress in rats. *Ukr Biochem J*. 2018; 90 (4): 102–110. DOI: 10.15407/ubj90.04.102.
13. Vyslotska LV, Gutty BV, Kozenko OV, Khalak VI, Chornyj MV, Martyshev TV, Krempa NY, Vozna OY, Todoruk VB. System of antioxidant protection of the body of piglets under the action of feed additive "Sylimevit". *Sci Mess LNUVMBT Ser Vet Sci*. 2021; 23 (104): 10–17. DOI: 10.32718/nvvet10402.
14. Wang T, Yao W, Xia J, Li J, Shao Y, Huang F. Dietary supplementation with garcinol during late gestation and lactation facilitates acid-base balance and improves the performance of sows and newborn piglets. *J Anim Sci*. 2019; 97 (11): 4557–4566. DOI: 10.1093/jas/skz292.
15. Weldon WC, Lewis AJ, Louis GF, Kovar JL, Giesemann MA, Miller PS. Postpartum hypophagia in primiparous sows: I. Effects of gestation feeding level on feed intake, feeding behavior, and plasma metabolite concentrations during lactation. *J Anim Sci*. 1994; 72 (2): 387–394. DOI: 10.2527/1994.722387x.
16. Wu F, Tian FJ, Lin Y, Xu WM. Oxidative stress: Placenta function and dysfunction. *Am J Reprod Immunol*. 2016; 76 (4): 258–271. DOI: 10.1111/aji.12454.
17. Zhao Y, Flowers WL, Saraiva A, Yeum KJ, Kim SW. Effect of social ranks and gestation housing systems on oxidative stress status, reproductive performance, and immune status of sows. *J Anim Sci*. 2013; 91 (12): 5848–5858. DOI: 10.2527/jas.2013-6388.

Вплив кормової добавки «Активо» на деякі показники системи антиоксидантного захисту свиноматок та отриманих від них поросят

Т. Я. Прудиус

tarasvet126@gmail.com

Інститут біології тварин НААН, вул. В. Стуса, 38, м. Львів, 79034, Україна

Інтенсивні технології вирощування сільськогосподарських тварин розробляються переважно для отримання максимального економічного ефекту. При цьому продуктивні тварини часто зазнають дії багатьох стресових чинників і надмірних фізіологічних навантажень, що призводить до погіршення добробуту тварин, їх здоров'я і, врешті-решт, до зниження продуктивних показників. Одним з найпоширеніших негативних процесів, які виникають у тваринному організмі внаслідок дії різноманітних чинників, є оксидативний стрес, тобто порушення про- і антиоксидантної рівноваги. Оксидативний стрес не виникає сам по собі, а переважно є супровідним процесом за дії стрес-факторів різної природи — наприклад, токсинів, температур, критичних фізіологічних періодів, інфекційних патологій, надмірної, чи незбалансованої, або недостатньої годівлі тощо. У роботі показано результати досліджень впливу кормової добавки «Активо», до складу якої входять вторинні рослинні компоненти ефірних олій орегано, перцю чилі, на кількісний вміст продуктів ПОЛ та активність систем антиоксидантного захисту у свиноматок та отриманих від них поросят. Дослідження проведено в умовах промислового комплексу ТОВ «Барком» (Львівська обл.). За принципом аналогів було сформовано дві групи порослих свиноматок великої білої породи по 10 тварин у кожній — контрольну та дослідну. Для дослідної групи в раціон додатково було введено кормову добавку «Активо» у кількості 0,1 кг/т готового корму. У крові свиноматок та поросят від них отриманих визначали вміст гідропероксидів ліпідів і ТБК-активних продуктів, активність глутатіонпероксидази та супероксиддисмутази. Результати проведених нами досліджень показали, що застосування для свиноматок та отриманих від них поросят кормової добавки «Активо» є доцільним з метою зменшення негативної дії оксидативного стресу, який чинить негативну дію як на свиноматок, так і на їхнє потомство.

Ключові слова: свиноматки, поросята, кормова добавка «Активо», ефірні олії, оксидативний стрес, супероксиддисмутаза, глутатіонпероксидаза, ТБК-активні продукти, гідропероксида ліпідів