

**CURRENT APPROACHES IN MAINTENANCE GUT  
HEALTH IN FARMING ANIMALS.  
PART 4. THE CURRENT PROGRESS  
IN MONOGLYCERIDES SUPPLEMENTATION TO PROVIDE  
ANTIBIOTICS RESTRICTION IN SWINE HUSBANDRY**

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## **INTRODUCTION**

Global swine production is currently in a recovery phase following a crisis caused by the spread of African swine fever, which reduced pig populations and pork production volumes in many countries, as well as the COVID-19 pandemic, which temporarily impacted market activity. Global pork production is showing growth in both volume and prices, operating within a framework of intense competition, technological transformation, and innovative progress<sup>1</sup>. The intensification of pork production has facilitated the spread of diseases, complicating the maintenance of productivity without the use of antimicrobial agents. Since the 1950s, growth promoters, including antimicrobial agents, have played a crucial role in enhancing animal productivity<sup>2</sup>. However, their use has led to a significant increase in antibiotic resistance. The European Union has banned growth promoters in 2006 (European Commission, 2006) and introduced additional restrictions on the prophylactic use of antibiotics in 2018, which took effect in 2022. These restrictions initiated an active search for antibiotic alternatives<sup>3</sup>.

In current livestock practices, there is a growing interest in using bioactive compounds as alternatives to antibiotics in animal feed<sup>4</sup>. Among these

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<sup>1</sup> Mykhalko O. H. Current state and ways of pig production in the world and Ukraine. *Bulletin of Sumy National Agrarian University. The Series: Livestock*. 2021. Iss. 3 (46). P. 61–77.

<sup>2</sup> High levels of antibiotic resistance in isolates from diseased livestock / N. A. Haulisah et al. *Frontiers in Veterinary Science*. 2021. Vol. 8. Art. 652351.

<sup>3</sup> Effects of short- and medium-chain fatty acids on production, meat quality, and microbial attributes – A Review / R. T. Szabó et al. *Molecules*. 2023. Vol. 28, No. 13. Art. 4956.

<sup>4</sup> Effects of medium-chain fatty acids as alternatives to ZnO or antibiotics in nursery pig diets / P. L. Dahmer et al. *Translational Animal Science*. 2020. Vol. 4, No. 3. P. 1–8.

compounds, monoglycerides (MGs) are of particular interest. MGs offer several advantages in combating animal infectious diseases compared to antibiotics. Firstly, they possess a broad spectrum of antimicrobial activity, inhibiting both viruses and bacteria, whereas antibiotics are only effective against bacteria. In the second place, pathogens encounter significant barriers in developing resistance to MGs, making them effective not only for treatment but also for disease prevention<sup>5</sup>. Third, MGs are more effective than fatty acids and are safer compared to other agents, such as formaldehyde<sup>6</sup>.

In present review, we provide the comprehensive views of current data on the molecular mechanisms, the potential, and effectiveness of monoglyceride application in swine production and to determine the impact of monoglycerides on the physiological state, productivity, and overall health of pigs. In addition, we also discuss about the monoglyceride effects on immune response, microbiome state and intestinal morphology.

## 1. General characteristics of monoglycerides

Monoglycerides are a class of glycerides consisting of a glycerol molecule linked to a fatty acid via an ester bond. Due to the presence of both primary and secondary hydroxyl groups in glycerol, there are two types of monoglycerides: 1-monoacylglycerols, where the fatty acid is attached to the first alcohol group, and 2-monoacylglycerols, where it is attached to the second alcohol group. Owing to their structure, monoglycerides possess both hydrophilic (water-attracting) and hydrophobic properties. Mono- and di-glycerides are generally produced through the transesterification (glycerolysis) of triglycerides with glycerol<sup>7</sup>. In industry, monoglycerides are primarily used as surfactants, typically in the form of emulsifiers. However, their most promising application appears to be as biologically active additives, providing a safe alternative to antibiotics and hormonal products.

Monoglycerides of short-chain and medium-chain fatty acids (SCFA & MCFA) have well-documented direct antimicrobial effect *in vitro* and are known to enhance immune competence in animals, improve gut health, and reduce disease transmission risk in the post-antibiotic era. SCFA  $\alpha$ -monoglycerides, such as  $\alpha$ -monopropionin and  $\alpha$ -monobutyryn (derivatives of propionic and butyric acids), are primarily active against gram-negative bacteria. In contrast,

<sup>5</sup> Churchward C. P., Alany R. G., Snyder L. A. S. Alternative antimicrobials: the properties of fatty acids and monoglycerides. *Critical Reviews in Microbiology*. 2018. Vol. 44, No. 5. P. 561–570.

<sup>6</sup> Jackman J. A., Lavergne T. A., Elrod C. C. Antimicrobial monoglycerides for swine and poultry applications. *Frontiers in Animal Science*. 2022. Vol. 3. Art. 1019320.

<sup>7</sup> Moonen H., Bas H. Mono- and Diglycerides. *Emulsifiers in Food Technology*. 2nd ed. / ed. by R. Whitehurst, C. Law. Chichester : Wiley-Blackwell, 2014. P. 73–92.

MCFA  $\alpha$ -monoglycerides, such as  $\alpha$ -monocaprylin,  $\alpha$ -monocaprin, and  $\alpha$ -monolaurin, are more effective against gram-positive bacteria. Numerous studies have confirmed the antibacterial effects of  $\alpha$ -monoglycerides in vitro and in vivo. Additionally, recent findings indicate that MCFA  $\alpha$ -monoglycerides, particularly  $\alpha$ -monolaurin, exhibit antiviral properties. The biological activity of butyric acid is associated with the modulation of certain cellular responses, including histone deacetylase inhibition and G-protein-coupled receptor activation in various cell types<sup>8</sup>.

Medium-chain fatty acids (MCFAs) represent a class of saturated fatty acids containing from 6 to 12 carbon atoms. This class includes caproic acid (C6), caprylic acid (C8), capric acid (C10), and lauric acid (C12), which found in natural products such as coconut oil, palm kernel oil, and milk<sup>9</sup>. Mono-, di-, and triglycerides of MCFAs are formed via esterification with glycerol as a rule. Apart from serving as an energy source, MCFAs demonstrated potential for improving gut morphology, stimulating growth, preventing infections, regulating immune response, and functioning as an alternative to antibiotics<sup>10, 11</sup>. Several results demonstrated that both mono- and tri-acylglycerol of DL-3-hydroxybutyrate is non-toxic, stimulate metabolic energy production upon intravenous administration as well as they can be used as nutrients for farming animal feeding<sup>12</sup>.

The primary structure that provides the biological activity of MGs are micelles – self-organized particles that form stable spheres at the content above the critical micelle concentration (CMC). MGs with longer hydrocarbon chains have lower CMC values due to stronger hydrophobic interactions, that promote micelle self-assembly<sup>13</sup>. The non-ionic nature of MGs allows them to form micelles at lower concentrations in compare with MCFAs, which are anionic and prone to electrostatic repulsion. This accounts for their greater efficacy, as MGs can form micelles at lower concentrations.

<sup>8</sup> Salvi P. S., Cowles R. A. Butyrate and the Intestinal Epithelium: Modulation of Proliferation and Inflammation in Homeostasis and Disease. *Cells*. 2021. Vol. 10, No. 7. Art. 1775.

<sup>9</sup> Short- and medium-chain fatty acids exhibit antimicrobial activity for oral microorganisms / C. B. Huang et al. *Archives of Oral Biology*. 2011. Vol. 56, No. 7. P. 650–654.

<sup>10</sup> Effects of medium-chain fatty acids as alternatives to ZnO or antibiotics in nursery pig diets / P. L. Dahmer et al. *Translational Animal Science*. 2020. Vol. 4, No. 3. P. 1–8.

<sup>11</sup> Hanczakowska E., Świątkiewicz M., Natonek-Wisniewska M., Okoń K. Medium chain fatty acids (MCFA) and/or probiotic *Enterococcus faecium* as a feed supplement for piglets. *Livestock Science*. 2016. Vol. 192. P. 1–7.

<sup>12</sup> Birkhahn R. H., McCombs C., Clemens R., Hubbs J. Potential of the monoglyceride and triglyceride of dl-3-hydroxybutyrate for parenteral nutrition: Synthesis and preliminary biological testing in the rat. *Nutrition*. 1997. Vol. 13, No. 3. P. vi–219.

<sup>13</sup> Competing Interactions of Fatty Acids and Monoglycerides Trigger Synergistic Phospholipid Membrane Remodeling / B. K. Yoon et al. *The Journal of Physical Chemistry Letters*. 2020. Vol. 11, No. 13. P. 4951–4957.

MGs can inhibit a wide range of membrane-containing viruses and bacteria via damaging their membranes. Fatty acids and monoglycerides exhibit significantly higher activity in micellar form, whereas their activity in monomeric form is considerably lower or absent<sup>14</sup>. The antibacterial effect of MGs accompanied by increased bacterial membrane permeability, local damage, and disruption of membrane functions, including electron transport, oxidative phosphorylation, and enzyme activity disturbance<sup>15</sup>. MGs are also effective against membrane-bound viruses, where membrane disruption is a key factor of their antiviral activity.

## 2. Effect of monoglycerides on performance and feed conversion in pigs

The supplementation of monoglycerides in pig feeding attracts special attention due to their potential to improve productivity and feed conversion efficiency. This section examines the effects of monoglycerides on growth, feed conversion, and the overall health of pigs based on recent scientific research. There is a significant amount of conflicting data regarding the lack of effects of SCFA- or MCFA-containing monoglycerides on the performance of weaned piglets<sup>16, 17, 18</sup>. However, recent studies indicate that adding a monoglyceride blend to piglet diets reduces the growth-to-feed ratio in Enterotoxigenic *Escherichia coli* (ETEC) infected pigs. The addition of capric acid to piglets' diets increased their body weight gain and exhibited moderate antibacterial activity against *E. coli*<sup>19</sup>. At the same time, studies have shown that a blend of monoglycerides (butyric, caprylic, and capric acids) did not affect the performance of broilers infected with necrotic enteritis<sup>20</sup>. Conversely, feed additives containing  $\alpha$ -glycerol monolaurate ( $\alpha$ -GML) improved the growth performance of broilers,

<sup>14</sup> Spectrum of Membrane Morphological Responses to Antibacterial Fatty Acids and Related Surfactants / B. K. Yoon et al. *Langmuir*. 2015. Vol. 31, No. 37. P. 10223–10232.

<sup>15</sup> Casillas-Vargas G., Ocasio-Malavé C., Medina S., Morales-Guzmán C., Del Valle R. G., Carballeira N. M., Sanabria-Ríos D. J. Antibacterial fatty acids: An update of possible mechanisms of action and implications in the development of the next-generation of antibacterial agents. *Progress in Lipid Research*. 2021. Vol. 82. Art. 101093.

<sup>16</sup> Low-Protein Diet Supplemented with Medium-Chain Fatty Acid Glycerides Improves the Growth Performance and Intestinal Function in Post-Weaning Piglets / Z. Cui et al. *Animals*. 2020. Vol. 10, No. 10. Art. 1852.

<sup>17</sup> Dietary Supplementation with Mono-Lactate Glyceride Enhances Intestinal Function of Weaned Piglets / H. Li et al. *Animals*. 2023. Vol. 13, No. 8. Art. 1303.

<sup>18</sup> Evaluation of different blends of medium-chain fatty acids, lactic acid, and monolaurin on nursery pig growth performance / L. L. Thomas et al. *Translational Animal Science*. 2020. Vol. 4, No. 2. P. 548–557.

<sup>19</sup> Hanczakowska E., Świątkiewicz M., Natonek-Wisniewska M., Okoń K. Medium chain fatty acids (MCFA) and/or probiotic *Enterococcus faecium* as a feed supplement for piglets. *Livestock Science*. 2016. Vol. 192. P. 1–7.

<sup>20</sup> Monoglyceride Blend Reduces Mortality, Improves Nutrient Digestibility, and Intestinal Health in Broilers Subjected to Clinical Necrotic Enteritis Challenge / A. Kumar et al. *Animals*. 2021. Vol. 11, No. 5. Art. 1432.

that attributed to increased feed intake and enhanced gut health<sup>21</sup>.  $\alpha$ -GML application demonstrated potential for enhancing antimicrobial activity in sows and promoting the growth of suckling piglets. The addition of  $\alpha$ -GML to sow diets increases ( $P < 0.05$ ) piglet body weight at weaning and overall digestibility of crude fat in sows<sup>22</sup>.

Recent data have shown that dose of 0.1%  $\alpha$ -GML improves the performance and growth of weaned piglets and reduces post-weaning diarrhea, which the authors attribute to improved nutrient digestibility, reduced systemic inflammation and intestinal permeability, and stable gut microbiota in piglets<sup>23</sup>.

The addition of glycerol monolaurate to sow diets improves gut health in suckling piglets. Specifically, the results of GML supplementation demonstrated a significant increase in fat, lactose, and protein content in colostrum of sow, while piglets exhibited a reduction in IL-12 concentration in the duodenum, TNF- $\alpha$ , IL-1 $\beta$ , and IL-12 in the jejunum, and IL-1 $\beta$  in the ileum. These changes accompanied by increase in villus height, crypt depth, and the villus-to-crypt ratio (V/C) in the jejunum and V/C in the ileum in the experimental animals in compare with control group<sup>24</sup>. Together, these factors contribute to the growth and development of piglets.

The other results suggest that the addition of MCFA to pig diets enhances growth performance, provides residual mitigating activity against PEDV, and does not significantly alter fecal microbial composition<sup>25</sup>.

The application of monoglycerides with lysolecithin in finishing pig diets reduces metabolic energy costs, improves pig weight uniformity within groups, and shortens the time to reach the target slaughter weight, although it worsens feed conversion<sup>26</sup>. Similar results in implementing feeding strategies to minimize feed costs and improve pig performance have been obtained by

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<sup>21</sup> Effects of alpha-glycerol monolaurate on growth, immune function, volatile fatty acids, and gut microbiota in broiler chickens / J. Lan et al. *Poultry Science*. 2021. Vol. 100, No. 3. Art. 100918.

<sup>22</sup> Supplementation with alpha-glycerol monolaurate during late gestation and lactation enhances sow performance, ameliorates milk composition, and improves growth of suckling piglets / L. Li et al. *Journal of Animal Science and Biotechnology*. 2023. Vol. 14, No. 1. Art. 38.

<sup>23</sup> Effects of combined application of benzoic acid and 1-monolaurin on growth performance, nutrient digestibility, gut microflora and systemic inflammation levels of weaned piglets / K. Wei et al. *Porc Health Manag*. 2023. 9. P. 46.

<sup>24</sup> Maternal supplementation with glycerol monolaurate improves the intestinal health of suckling piglets by inhibiting the NF-kppB/MAPK pathways and improving oxidative stability / H. Zhao et al. *Food & Function*. 2023. Vol. 14, No. 7. P. 3290–3303.

<sup>25</sup> Effect of dietary medium-chain fatty acids on nursery pig growth performance, fecal microbial composition, and mitigation properties against porcine epidemic diarrhea virus following storage / J. T. Gebhardt et al. *Journal of Animal Science*. 2020. Vol. 98, No. 1. Art. skz358.

<sup>26</sup> Gonzalez Sanchez D., Toth S., Desbruslais A. P30. Supplementing lysolecithin with emulsifier and monoglycerides to diets reformulated to lower energy on performance and lean-fat deposition in fattening pigs. *Animal – Science Proceedings*. 2022. Vol. 13, No. 2. P. 166.

other researchers<sup>27</sup>. At the same time, the supplementation complex mixture containing lysolecithins, monoglycerides, and synthetic emulsifier in a dose 750 g/t to post-weaning piglet diet significantly improves their performance, enhances the digestibility of protein, fat, and fiber without increase in feed intake<sup>28</sup>.

Thus, the use of monoglycerides and their derivatives in pig feeding has a beneficial effect on animal performance and health, especially in animals challenged with infectious diseases. The inclusion of monoglycerides in pig diets demonstrates antimicrobial properties and enhances resistance to infectious diseases via reducing diarrhea incidence and improving intestinal wall morphology.

### **3. Effectiveness of monoglycerides in the treatment and prevention of swine diseases**

Nowadays, monoglycerides are widely proposed as feed additive in pig production to improve performance and feed conversion efficiency. They have the potential to positively impact animal health due to their antimicrobial properties, which help reduce disease incidence and improve metabolic processes. Experimental studies have shown that MCFAs exhibit high bacteriostatic activity against various pathogens, including gram-positive and gram-negative bacteria, viruses, fungi, algae, and protozoa.

The study of fatty acids in respect with its antimicrobial properties is well-documented in the literature. One of the most common diseases in piglets is post-weaning diarrhea (PWD), often caused by the adhesion and proliferation of ETEC F18 or F4 in the small intestine. Clinical signs include profuse diarrhea, growth retardation, dehydration, and lethargy<sup>29</sup>. It has been shown that butyrate and valerate glycerides alleviate diarrhea symptoms and modulate immune response in piglets co-infected with ETEC<sup>30</sup>. Specifically, pigs fed with glycerides exhibited higher body weight, reduced diarrhea severity, and lower levels of neutrophils and TNF- $\alpha$  in serum on day 4 post-inoculation. The use of

<sup>27</sup> Wealleans A., Vasanthakumari B., Desbruslais A. P5. Improvements in digestibility and growth of growing-finishing pigs following supplementation with a combination of lysolecithins, monoglycerides and a synthetic emulsifier. *Animal – Science Proceedings*. 2022. Vol. 13, No. 2. P. 158.

<sup>28</sup> A combination of lysolecithins, a synthetic emulsifier, and monoglycerides, supports piglet performance, nutrient digestibility, and faecal consistency / A. Desbruslais et al. *Animal – Science Proceedings*. 2023. Vol. 14, No. 6. P. 810.

<sup>29</sup> Effects of a blend of essential oils, medium-chain fatty acids, and a toxin-adsorbing mineral on diarrhea and gut microbiome of weanling pigs experimentally infected with a pathogenic *Escherichia coli* / Y. He et al. *Journal of Animal Science*. 2022. Vol. 100, No. 1. Art. skab365.

<sup>30</sup> Dietary butyrate and valerate glycerides impact diarrhea severity and immune response of weaned piglets under ETEC F4-ETEC F18 coinfection conditions / L. Kovanda et al. *Journal of Animal Science*. 2023. Vol. 101. Art. skad401.

an antimicrobial MCFAs blend (0.5%) reduced the incidence of severe diarrhea in weaned piglets infected with F18 ETEC and improved their feed efficiency at the final stage of the experiment. This blend also altered the microbiota diversity in feces and ileal mucosa<sup>31</sup>. Comparative analyses have shown that adding monoglyceride mixtures can enhance disease resistance in weaned piglets by reducing diarrhea severity and mitigating intestinal and systemic inflammation<sup>32</sup>. However, monoglyceride efficiency remains not comparable to high doses of zinc oxide to treat diarrhea symptoms.

The inclusion of glycerol butyrate in pig diets reduces release of pro-inflammatory factors (TNF $\alpha$ , IL6, and IL1B) in the jejunum and ileum prior to ETEC infection by inhibiting the NF- $\kappa$ B/MAPK pathway<sup>33</sup>. The results of the other study demonstrated that monoglycerides reduce the viability of porcine epidemic diarrhea virus (PEDV) in feed and prevent the disease's transmission to post-weaning piglets<sup>34</sup>. Recent studies have shown that benzoic acid, combined with probiotics or plant essential oils, can more effectively alleviate diarrhea and stimulate growth in weaned piglets than benzoic acid alone<sup>35, 36, 37</sup>.

Previous studies have confirmed the antiviral activity of free MCFAs, including capric, lauric, and myristic acids, as well as long-chain unsaturated fatty acids including like oleic, linoleic, and linolenic acids, against vesicular stomatitis virus (VSV), herpes simplex virus (HSV), and Visna virus<sup>38, 39</sup>.

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<sup>31</sup> Effects of a blend of essential oils, medium-chain fatty acids, and a toxin-adsorbing mineral on diarrhea and gut microbiome of weanling pigs experimentally infected with a pathogenic *Escherichia coli* / Y. He et al. *Journal of Animal Science*. 2022. Vol. 100, No. 1. Art. skab365

<sup>32</sup> Effects of monoglyceride blend on systemic and intestinal immune responses, and gut health of weaned pigs experimentally infected with a pathogenic *Escherichia coli* / S. Park et al. *Journal of Animal Science and Biotechnology*. 2024. Vol. 15, No. 1. Art. 12.

<sup>33</sup> Glyceryl butyrate attenuates enterotoxigenic *Escherichia coli*-induced intestinal inflammation in piglets by inhibiting the NF- $\kappa$ B/MAPK pathways and modulating the gut microbiota / M. Tian et al. *Food & Function*. 2022. Vol. 13, No. 11. P. 6282–6292.

<sup>34</sup> Monoglyceride reduces viability of porcine epidemic diarrhoea virus in feed and prevents disease transmission to post-weaned piglets / F. C. Phillips et al. *Transboundary and Emerging Diseases*. 2022. Vol. 69, No. 1. P. 121–127.

<sup>35</sup> Protective Effects of Benzoic Acid, *Bacillus coagulans*, and Oregano Oil on Intestinal Injury Caused by Enterotoxigenic *Escherichia coli* in Weaned Piglets / J. Pu et al. *BioMed Research International*. 2018. Vol. 2018. Art. 1829632.

<sup>36</sup> Benzoic Acid Combined with Essential Oils Can Be an Alternative to the Use of Antibiotic Growth Promoters for Piglets Challenged with *E. coli* F4 / L. M. Rodrigues et al. *Animals*. 2020. Vol. 10, No. 11. Art. 1978.

<sup>37</sup> The use of an alternative feed additive, containing benzoic acid, thymol, eugenol, and piperine, improved growth performance, nutrient and energy digestibility, and gut health in weaned piglets / C. D. Silva Júnior et al. *Journal of Animal Science*. 2020. Vol. 98, No. 5. Art. skaa119.

<sup>38</sup> Hilmarsson H., Kristmundsdóttir T., Thormar H. Virucidal activities of medium- and long-chain fatty alcohols, fatty acids and monoglycerides against herpes simplex virus types 1 and 2: comparison at different pH levels. *APMIS*. 2005. Vol. 113, No. 1. P. 58–65.

<sup>39</sup> Inactivation of enveloped viruses and killing of cells by fatty acids and monoglycerides / H. Thormar et al. *Antimicrobial Agents and Chemotherapy*. 1987. Vol. 31, No. 1. P. 27–31

Furthermore, other studies have demonstrated similar antiviral activity of MCFAs and their derivatives, including alcohols and monoglycerides, against herpes simplex viruses 1 and 2.

Currently, there are no vaccines or effective drugs for treating Seneca Valley virus (SVV) infection. In vitro studies have shown that monolaurin inhibits SVV replication by 80%, while in vivo studies have demonstrated that monolaurin reduces clinical manifestations, viral load, and organ damage in SVV-infected piglets<sup>40</sup>. Monolaurin significantly reduced inflammatory cytokine release and promoted interferon- $\gamma$  production, enhancing viral clearance activity. Researchers concluded that monolaurin is a potentially effective alternative for treating SVV infection in pigs.

The antiviral activity of caprylic, capric, and lauric acids, as well as glycerol monolaurate (GML), has been investigated concerning ASFV inhibition in liquids and feed. Compounds and its dosages that can effectively inhibit ASFV proliferation have been identified<sup>41</sup>. Low GML concentrations had a minimal effect on virulence, while sufficiently high GML concentrations caused >99% virulence reduction. The onset concentration of antiviral activity corresponded to the critical micelle concentration of GML, highlighting the essential role of formulation in stimulating protective responses against ASFV<sup>42</sup>.

The use of modern nanotechnology enables the creation of more effective formulations without altering the chemical composition of bioactive preparations. For example, natural GML has low water solubility and limited efficacy against gram-negative bacteria. It deserves to be mentioned that nanoencapsulation not only improves bioavailability but also reduces ecotoxicity<sup>43</sup>.

Fatty acids and monoglycerides have proven effective against *Neisseria gonorrhoeae*. Microemulsions with  $\alpha$ -linolenic acid are effective in treating neonatal gonococcal ophthalmia<sup>44</sup>. In the absence of vaccines, monocaprin may serve as a novel prophylactic agent to prevent *N. gonorrhoeae* growth and colonization, to which the bacterium cannot develop resistance<sup>45</sup>.

<sup>40</sup> In vitro and in vivo antiviral activity of monolaurin against Seneca Valley virus / B. Su et al. *Frontiers in Veterinary Science*. 2023. Vol. 10. Art. 980187.

<sup>41</sup> Inhibition of African swine fever virus in liquid and feed by medium-chain fatty acids and glycerol monolaurate / J. A. Jackman et al. *Journal of Animal Science and Biotechnology*. 2020. Vol. 11, No. 1. P. 1–10.

<sup>42</sup> Glycerol Monolaurate Inhibits Wild-Type African Swine Fever Virus Infection in Porcine Macrophages / J. A. Jackman et al. *Pathogens*. 2023. Vol. 12, No. 10. Art. 1193.

<sup>43</sup> Ecotoxicology of Glycerol Monolaurate nanocapsules / L. Q. Lopes et al. *Ecotoxicology and Environmental Safety*. 2017. Vol. 139. P. 73–77.

<sup>44</sup> Belagal P. Current alternative therapies for treating drug-resistant *Neisseria gonorrhoeae* causing ophthalmia neonatorum. *Future Microbiology*. 2024. Vol. 19, No. 7. P. 631–647.

<sup>45</sup> Churchward C. P., Calder A., Snyder L. A. S. Mutations in *Neisseria gonorrhoeae* grown in sub-lethal concentrations of monocaprin do not confer resistance. *PloS One*. 2018. Vol. 13, No. 4. Art. e0195453.

Stress associated with piglet weaning suppresses digestion, absorptive capacity, and intestinal energy provision. MCFAs can effectively improve nutrient absorption and lipid metabolism in piglets to meet their energy needs, thereby supporting the growth and development of weaned piglets<sup>46</sup>. MCFAs, such as hexanoic and octanoic acids, have shown significant anticandidal activity, as have long-chain fatty acids, including lauric acid<sup>47</sup>.

Thus, in a view of intense competition and increasing food safety requirements, the supplementation of monoglyceride blends could be prospective strategy to remove antibiotics in productive animals farming. Besides, both antimicrobial and metabolic effect of monoglycerides can be important factors for reducing the risk of antibiotic resistance and enhancing swine production profitability.

Monoglycerides, known for their antimicrobial and anti-inflammatory properties, are emerging as promising additives to enhance immune and antioxidant systems in pigs. The effects of monoglycerides are directed at strengthening immunity, reducing oxidative stress, and increasing the overall resilience of animals against enteric infections. Administering  $\alpha$ -GML to piglets at a dose of 1000 mg/kg increases cytokine concentrations, particularly IL-10, and the activity of antioxidant enzymes (GSH-Px) in the serum of weaned piglets. Under these conditions, serum MDA and TNF- $\alpha$  concentrations decrease. However, no differences were found in T-AOC and SOD activity or IL-6 concentration in serum<sup>48</sup>. It is suggested that  $\alpha$ -GML enhances antioxidant capacity by modulating intestinal barrier function.

In pigs, administering 0.6%  $\alpha$ -GML reduces malondialdehyde and hydrogen peroxide levels in the ileum and significantly increases ( $p < 0.05$ ) superoxide dismutase and catalase activities in the ileum and colon<sup>49</sup>.

Mechanisms of antioxidant capacity transfer from the sow to the fetus through the gut-placenta axis have also been studied, using  $\alpha$ -GML glycerol monolaurate feed supplements during pregnancy<sup>50</sup>. Correlational analysis demonstrated a significant negative association between plasma lipopolysaccharide levels and placental weight, oxidative stress, and inflammation.

<sup>46</sup> Effects of Medium-Chain Fatty Acid Glycerides on Nutrient Metabolism and Energy Utilization in Weaned Piglets / Z. Cui et al. *Frontiers in Veterinary Science*. 2022. Vol. 9. Art. 938888.

<sup>47</sup> Short- and medium-chain fatty acids exhibit antimicrobial activity for oral microorganisms / C. B. Huang et al. *Archives of Oral Biology*. 2011. Vol. 56, No. 7. P. 650–654.

<sup>48</sup> Effects of alpha-glycerol monolaurate on intestinal morphology, nutrient digestibility, serum profiles, and gut microbiota in weaned piglets / L. Li et al. *Journal of Animal Science*. 2022. Vol. 100, No. 3. Art. skac046.

<sup>49</sup> Supplementation with alpha-glycerol monolaurate during late gestation and lactation enhances sow performance, ameliorates milk composition, and improves growth of suckling piglets / L. Li et al. *Journal of Animal Science and Biotechnology*. 2023. Vol. 14, No. 1. Art. 38.

<sup>50</sup> Enhanced Antioxidative Capacity Transfer between Sow and Fetus via the Gut-Placenta Axis with Dietary Selenium Yeast and Glycerol Monolaurate Supplementation during Pregnancy / J. Zhang et al. *Antioxidants*. 2024. Vol. 13, No. 2. Art. 177.

#### 4. Effects of monoglycerides on intestinal morphology and microbiome in pigs

Gut health is a key factor in ensuring optimal productivity in pigs and resilience to infectious diseases. Monoglycerides have beneficial effects on the morphology of intestinal structures and can modulate the composition of the gut microbiota, thereby supporting intestinal barrier function<sup>51</sup>. Studies have shown that supplementation  $\alpha$ -GML to piglets at a dose of 1000 mg/kg increases the villus height-to-crypt depth ratio in the duodenum and villus height in the jejunum and ileum compared to the control group, although it did not affect crypt depth in any intestinal section<sup>52</sup>. Additionally, GML promotes an increase in goblet cell numbers<sup>53</sup>.

MCFA-containing glycerides influence intestinal morphology, immune and goblet cells in post-weaning piglets. *E. coli* infection is associated with reduced villus height and crypt depth. However, in piglets exposed to LPS, MCFA-derived glycerides restore villus morphology and increase IgA levels in blood. A similar effect was observed in the ileum, albeit with a less pronounced impact<sup>54</sup>.

The application of  $\alpha$ -GML to piglets in a dose 1000 mg/kg increases the relative abundance of Firmicutes by 17.87% ( $P < 0.05$ ) and decreases the proportions of Bacteroidota and Campylobacterota ( $P < 0.05$ ). In the cecum, the levels of unclassified Lactobacillus and Blautia species significantly increased ( $P < 0.05$ ) while Eubacterium rectale, unclassified Campylobacter, and uncultured\_bacterium Alloprevotella decreased ( $P < 0.05$ )<sup>55</sup>.

Gene expression analysis indicated that linoleic acid significantly enhances the expression of genes involved in adipogenesis and pathways dependent on PPAR $\gamma$ , C/EBP $\alpha$ , ap2, and NRF1 signaling. Cells incubated with LNA showed reduced phosphorylation of Ser 112 in PPAR $\gamma$  compared to cells treated with palmitic or oleic acid. The lack of significant difference in triacylglycerol levels upon inhibition of PPAR $\gamma$  Ser 112 phosphorylation suggests varied metabolic

<sup>51</sup> Fibronectin measurement as a potential molecular marker for barrier function assessment of piglet intestine / D. M. Masiuk et al. *Theoretical and Applied Veterinary Medicine*. 2023. Vol. 11, No. 2. P. 3–8.

<sup>52</sup> Supplementation with alpha-glycerol monolaurate during late gestation and lactation enhances sow performance, ameliorates milk composition, and improves growth of suckling piglets / L. Li et al. *Journal of Animal Science and Biotechnology*. 2023. Vol. 14, No. 1. Art. 38.

<sup>53</sup> Low-Protein Diet Supplemented with Medium-Chain Fatty Acid Glycerides Improves the Growth Performance and Intestinal Function in Post-Weaning Piglets / Z. Cui et al. *Animals*. 2020. Vol. 10, No. 10. Art. 1852.

<sup>54</sup> Medium-chain glycerides affect gut morphology, immune- and goblet cells in post-weaning piglets: In vitro fatty acid screening with Escherichia coli and in vivo consolidation with LPS challenge / K. De Keyser et al. *Journal of Animal Physiology and Animal Nutrition*. 2019. Vol. 103, No. 1. P. 221–230.

<sup>55</sup> Supplementation with alpha-glycerol monolaurate during late gestation and lactation enhances sow performance, ameliorates milk composition, and improves growth of suckling piglets / L. Li et al. *Journal of Animal Science and Biotechnology*. 2023. Vol. 14, No. 1. Art. 38.

pathways of dietary fatty acids in porcine adipocytes. LNA significantly promotes lipid accumulation, likely by regulating PPAR $\gamma$  phosphorylation<sup>56</sup>.

The supplementation of GML increases the expression of claudin-1, occludin, and ZO-1 in the jejunum and ZO-1 in the ileum, indicating modulation of the intestinal barrier<sup>57</sup>. Administering 0.6% glycerol monolactate in pigs significantly ( $p < 0.05$ ) increases occludin expression in the intestine (H. Li et al., 2023). Additionally, GML applying to sows enhances the levels of zonula occludens-1, occludin, and claudin-1 proteins in the small intestine ( $P < 0.05$ ), along with increased mRNA expression of mucins (MUC-1, MUC-3, and MUC-4) and porcine beta-defensins<sup>58</sup>.

Capric acid reduces total intestinal length, while probiotics do not affect intestinal mass or length but increase villus width and crypt depth in the epithelial cells of the small intestine. MCFAs do not impact these parameters. Both additives exhibit strong antibacterial action: capric acid against *E. coli* and probiotics against *C. perfringens*<sup>59</sup>. Sequencing of 16S rDNA demonstrated that supplementing sows with GML alters the colon microbiome of piglets and reduces the relative abundance of *Escherichia shigella*<sup>60</sup>.

Thus, monoglycerides can have beneficial effects on intestinal morphology, particularly by increasing villus height and improving the villus-to-crypt ratio, which promotes more efficient nutrient absorption. Additionally, monoglycerides can modulate the composition of the gut microbiota, reducing pathogenic bacteria and supporting the balance of beneficial microbiota.

## CONCLUSIONS

Monoglycerides exhibit significant potential in swine production maintenance due to their antimicrobial, antiviral, and anti-inflammatory properties. They not only contribute to improved animal productivity but also provide essential protection against infectious diseases. The use of monoglycerides in pig feeding has demonstrated high effectiveness in reducing the incidence of diarrhea

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<sup>56</sup> PAM, OLA, and LNA are Differentially Taken Up and Trafficked Via Different Metabolic Pathways in Porcine Adipocytes / C. Yu et al. *Lipids*. 2017. Vol. 52, No. 11. P. 929–938.

<sup>57</sup> Low-Protein Diet Supplemented with Medium-Chain Fatty Acid Glycerides Improves the Growth Performance and Intestinal Function in Post-Weaning Piglets / Z. Cui et al. *Animals*. 2020. Vol. 10, No. 10. Art. 1852.

<sup>58</sup> Maternal supplementation with glycerol monolaurate improves the intestinal health of suckling piglets by inhibiting the NF-kappaB/MAPK pathways and improving oxidative stability / H. Zhao et al. *Food & Function*. 2023. Vol. 14, No. 7. P. 3290–3303.

<sup>59</sup> Effects of monoglyceride blend on systemic and intestinal immune responses, and gut health of weaned pigs experimentally infected with a pathogenic *Escherichia coli* / S. Park et al. *Journal of Animal Science and Biotechnology*. 2024. Vol. 15, No. 1. Art. 12.

<sup>60</sup> Maternal supplementation with glycerol monolaurate improves the intestinal health of suckling piglets by inhibiting the NF-kappaB/MAPK pathways and improving oxidative stability / H. Zhao et al. *Food & Function*. 2023. Vol. 14, No. 7. P. 3290–3303.

caused by bacterial infections, particularly *Escherichia coli*, by stabilizing gut microbiota and strengthening the immune system. Additionally, they can improve intestinal morphological parameters, such as villus height and the villus-to-crypt ratio, which enhance nutrient absorption and overall pig health.

Monoglycerides also play a crucial role in stimulating immune and antioxidant systems, reducing oxidative stress, and helping to maintain animal health under stressful conditions, such as weaning or intensive rearing. Their antioxidant properties protect cells from damage and reduce inflammation, which is critical for supporting healthy metabolism and increasing disease resilience in animals. Given the current requirements to reduce antibiotics supplementation in animal husbandry, monoglycerides serve as an effective alternative capable of replacing traditional antimicrobial agents.

Thus, monoglycerides represent a promising and safe feed additive for enhancing the productivity, health, and overall resilience of pigs. Their application helps reduce dependence on antibiotics, supporting stable productivity and competitiveness in swine production amid global challenges, such as combating antibiotic resistance and meeting the growing demand for environmentally sustainable products.

## SUMMARY

The article examines the properties and prospects of using monoglycerides in swine production as an alternative to antibiotics, aligning with modern demands for environmental safety and sustainable livestock farming. Monoglycerides are bioactive compounds which develop antimicrobial, antiviral, and anti-inflammatory effects. Furthermore, monoglycerides application confirmed to be effective tool to support the animal health, immune status, and productivity of pig farming, especially in current intensive production requirements.

The use of monoglycerides enhances the overall resistance of the organism to infections, which is crucial for minimizing disease incidence and ensuring stable productivity in scenarios with limited antibiotic use. Recent research findings indicate that monoglycerides positively impact intestinal morphology, particularly by increasing villus height and improving the villus-to-crypt ratio, which boosts nutrient absorption efficiency. In addition, monoglyceride blends can modulate the composition of the gut microbiota via the pathogenic bacteria reduction and regulation of beneficial microbial taxa balance. This effect lead to the improvement in digestion, metabolism, and overall animal health that is especially valuable during stress periods, including post-weaning complications.

Besides, monoglycerides demonstrated an ability to stimulate the immune system and enhance antioxidant defense. They reduce oxidative stress progress,

that is an essential factor to maintain cell viability and organ functionality, and promote cytokine synthesis, which is involved in regulating immune responses. Furthermore, due to their antimicrobial properties, monoglycerides can serve as an effective alternative to antibiotics in new strategy for treatment bacterial and viral infections.

Taking into the account an absent of environmental toxicity, reduction of antibiotic dependence, and positive effects on pig health, monoglycerides could become an efficient tool to build the strategy of sustainable swine production. Their application contributes to improved productivity, quality, and product safety while supporting livestock resilience to global challenges, such as the rising demand for environmentally friendly products and control over infectious diseases. Thus, monoglycerides are a promising feed additive which capable to enhance the competitiveness and efficiency of swine farms in line with contemporary standards.

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