REVIEW

J Anim Sci Technol 2022;64(2):187-196
https://doi.org/10.5187/jast.2022.e1

JAST
Journal of Animal Science and Technology
pISSN 2672-0191 eISSN 2055-0391

Novel zinc sources as antimicrobial growth promoters for monogastric animals: a review

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Check for updates

Received: Sep 30, 2021
Revised: Dec 17, 2021
Accepted: Jan 8, 2022

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Competing interests
No potential conflict of interest relevant to this article was reported.

Abstract

The essentiality of zinc for animals has been recognized over 80 years. Zinc is an essential trace element that is a component of many enzymes and is associated with the various hormones. Apart from the nutritional function, zinc has antimicrobial property and often be supplemented in diets in the quantities greater than which is required to meet the nutritional requirement, especially for weaning pigs. This review will focus on the application of pharmacological zinc and its mechanisms which may be responsible for the effects of zinc on performance and health of monogastric animals. Various novel sources of zinc in non-ruminant animal production will also be discussed. These should assist in more precisely formulating feed to maximize the production performance and to maintain the health condition of monogastric animals.

Keywords: Novel zinc source, Pharmacological zinc, Monogastric animals

INTRODUCTION

Zinc, an essential trace element, is a component of many metalloenzymes including DNA and RNA synthetases and many digestive enzymes [1]. The essentiality of zinc for animals has been recognized in 1933 [2]. Besides, zinc has a regulatory role in the immune system and deficiency in zinc may impair the proper immune function [3]. The concentration of zinc in body increases during the initial period of postnatal ontogenesis (2 weeks for chicks and 4 weeks for piglets), after which it remains approximately constant. Apart from the nutritional function, unlike most other minerals, copper and zinc have antimicrobial properties. Therefore, they are often supplemented in diets in the quantities greater than which is required to meet the nutritional requirement, especially for weaning pigs [4]. In practice, the use of pharmacological levels of zinc ranging from 2,000 to 4,000 mg/kg (that this "practice" is forbidden in the EU) in the form of zinc oxide is a common recommendation to reduce post-weaning diarrhea and improve growth performance in weaning pigs [5]. However, the use of high doses zinc oxide has raised serious concern related to microbial resistance development [6–8]. Moreover, due to its low absorption, high amount of zinc excretion has motivated environmental concerns [9–11]. Those provoke the question about the reasonability of pharmacological levels of zinc supplementation as a solution of the ban of antimicrobial growth promoters.

Funding sources
Not applicable.

Acknowledgements
Not applicable.

Availability of data and material
Upon reasonable request, the datasets of this study can be available from the corresponding author.

Authors' contributions
Conceptualization: Lei XJ, Kim IH.
Writing - original draft: Lei XJ, Liu ZZ.
Writing - review & editing: Lei XJ, Liu ZZ, Park JH, Kim IH.

Ethics approval and consent to participate
This article does not require IRB/IACUC approval because there are no human and animal participants.

To improve the bioavailability of zinc, several efforts have been made including: changing conventional zinc oxide powder into porous particles or nanoparticles, organic zinc (eg, zinc lactate, zinc amino acid, zinc chelate), and the application of enteric coating method [12–17]. Phytate from whole grains and some vegetables, by forming indigestible complexes with zinc in intestine strongly interferes with zinc absorption [18]. Therefore, it is suggested that supplementation of phytase in the diet can be a useful strategy to improve bioavailability of zinc [19]. This review will focus on the application of pharmacological zinc and its mechanisms which may be responsible for the effects of zinc on performance and health of monogastric animals. various novel sources of zinc in non-ruminant animal production will also be discussed. These should assist in more precisely formulating feed to maximize the production performance and to maintain the health condition of monogastric animals.

PHARMACOLOGICAL ZINC

Traditionally, antibiotics are included in the diet of young animals for the prevention of post-weaning diarrhea and enhancement of growth performance in the worldwide. However, the use of in-feed antibiotics has been fully or gradually banned by many countries including European countries, United States of America, South Korea and China due to the development of antibacterial resistance and antibiotic residues [20–23]. Supplementation of pharmacological doses of zinc in the form of zinc oxide has been proposed as one of the most effective feed additives to replace antibiotics and is already widely commercialized in several countries [24–27]. Pharmacological levels of zinc oxide have shown antimicrobial properties and are used to fight against post-weaning infections and improve growth performance [28–31].

The exact mechanisms regarding pharmacological doses of zinc oxide exert beneficial effects on alleviating post-weaning diarrhea and promoting growth performance are unknown, but potential theories proposed include: (1) pharmacological doses of zinc oxide may reduce intestinal permeability and improve intestinal morphology and appetite [32,33]. Carlson [34] observed that zinc oxide reduced the intestinal mucosal susceptibility to secretagogues which activate chloride secretion. In addition, it is suggested that zinc oxide may increase insulin-like growth factor-1, 2 expressions in the small intestinal mucosa [35]. (2) pharmacological doses of zinc oxide have a antimicrobial effects on gram negative bacteria and beneficial effects on the stability and diversity of gastrointestinal microbial balance, especially coliforms [36,37]. (3) pharmacological doses of zinc oxide can improve animals' defense function indicated as improved immune and anti-inflammatory responses [38]. Nevertheless, the use of high doses of zinc is criticized due to the developing bacterial resistance and the excretion of a large amount zinc which may raise environmental concerns [39,40]. Although the use of pharmacological doses of zinc oxide are allowed be in most countries and regions, the European legislation limits the use of zinc oxide in animal production to a maximum of 150 mg/kg [41]. Thus, considerable efforts have been made to improve the bioavailability of zinc, including organic zinc, nanosized zinc oxide, coated zinc oxide, and porous zinc oxide.

COATED ZINC OXIDE

Enteric coating is a common technology used to protect oral medications against the influence of stomach juices in the field of pharmacy [42]. In feed additives production, enteric coating technology also has been used for a long time [43,44]. Under the protection of outer enteric coating, the inner targeted component can safely pass through the stomach. Whereas, the coating materials

 https://doi.org/10.5187/jast.2022.e1

can be gradually degraded in the gastrointestinal tract, and the inter zinc oxide will be released slowly within the intestine [13]. Park et al. [9] suggested that the physiological level (100 mg/kg) of zinc from coated zinc oxide showed the similar growth promoting effects as pharmacological zinc oxide, but activities of maltase and sucrase in the intestinal mucosa and pancreatic tissue and small intestinal mucosal morphology were not affected by dietary treatments in weaning pigs. However, the effects of coated zinc oxide in pigs were not consistent. Byun et al. [45] indicated that inclusion of coated zinc had no effects on growth performance, intestinal mucosal morphology, and fecal consistency in weaning pigs. Song et al. [46] observed that sub-pharmacological levels (100, 200, or 400 mg/kg) zinc from coated zinc oxide had no effects on growth performance and fecal consistency but reduced villus height and the ratio of villus height to crypt depth in weaning pigs. Shen et al. [13] concluded that low concentrations (380 or 570 mg/kg) of zinc from coated zinc oxide or 2,250 mg/kg zinc from conventional zinc oxide could alleviate the occurrence of diarrhea by promoting intestinal morphology, protecting intestinal mucosal barrier from damage, stimulating mucosal immune system, and regulating microbial composition. Moreover, pigs offered diets with coated zinc oxide excreted less zinc in feces compared with those fed those fed conventional zinc oxide. In enterotoxigenic *Escherichia coli* challenged weaning pigs, Kwon et al. [47] found that both low dose (100 mg/kg) of coated zinc oxide and high dose (2,500 mg/kg) of conventional zinc oxide improved growth rate, reduced *Escherichia coli* shedding, increased goblet cell density in small intestine, and decreased gastrointestinal tract pH values in post-challenge period. In a recent study, Upadhaya et al. [48] indicated that use of coated zinc oxide in lower doses (250, 500, 750, and 1,000 mg/kg) was possible to substitute high dose of conventional zinc oxide in improving growth performance, nutrient digestibility, and intestinal microbial balance in weaning pigs. Importantly, compared with high level of conventional zinc oxide, the zinc excretion was decreased by use of low doses of coated zinc oxide. To our best knowledge, no information is available on the application of coated zinc oxide in poultry, at least for broilers and layers. Thus, researches are required to determine the effects of coated zinc oxide in poultry.

ORGANIC ZINC OXIDE

Organic mineral sources for dietary supplementation, such as proteinate and amino acid chelate, have become popular in feed industry in the past 2 decades due to their higher bioavailability [49]. In weaning pigs, Wang et al. [50] observed that low levels (50 and 100 mg/kg) of zinc from zinc glycine chelate could improve growth rate and serum alkaline phosphatase and copper/zinc superoxide dismutase activities as 3,000 mg/kg of zinc as conventional zinc oxide. But zinc excretion in feces was decreased in pigs offered zinc glycine chelate diets compared with pigs receiving diet supplemented with high level conventional zinc oxide. Case and Carlson [51] indicated that weaning pigs fed 500 mg/kg zinc in the form of an organic zinc-polysaccharide complex had comparable production performance but excreted less zinc in feces compared with pigs fed 3,000 mg/kg zinc as zinc oxide. Buff et al. [52] indicated that supplementation of 300 or 450 mg/kg zinc from zinc-polysaccharide allowed pigs obtained similar growth performance as 2,000 mg/kg zinc from conventional zinc oxide, but 300 mg/kg zinc as zinc-polysaccharide reduced 76% of fecal zinc compared with 2,000 mg/kg zinc as conventional zinc oxide. Cheng et al. [53] suggested that both 100 mg/kg of zinc from zinc lysine complex and zinc sulfate were equally effective in maintaining production performance and zinc absorption of weaning pigs. In sows, Payne et al. [54] found that supplementation of zinc from organic zinc (zinc amino acid complex) could improve the development of fetuses during gestation, thus resulting in increased piglets weight at birth and weaning. Moghaddam and Jahanian [55] replaced 75% inorganic zinc with dietary zinc-methionine and observed improvement in both cellular and humoral functions of the immune system in broilers. Ao et al. [56] reported that chelated zinc proteinate could be used as a zinc source. 12 mg/kg of zinc from chelated zinc proteinate allowed broilers obtained optimal growth performance, whereas when inclusion of phytase in the diet, the optimal dose of zinc from chelated zinc proteinate to meet the requirement of broiler was 7.4 mg/kg. Rossi et al. [57] supplemented graded levels (0, 15, 30, 45, or 60 mg/kg) of organic zinc (chelated zinc proteinate) in male chicks did not affect production performance but improved carcass quality by increasing resistance of skin to tearing. Using the same organic zinc oxide-chelated zinc proteinate, Salim et al. [58] reported that inclusion of 25 mg/kg chelated zinc proteinate had no effect on growth performance and skin quality of broiler chickens, but increased the zinc content in thigh meat and calcium content in plasma.

NANOSIZED ZINC OXIDE

Nanotechnology has revolutionized the commercial application of nanosized minerals in the areas of engineering, food, biological, and pharmacological applications [11]. Due to better antibacterial property than conventional zinc oxide, nanosized zinc oxide is the third highest produced nanosized metal after nanosized silicon dioxide and nanosized titanium dioxide [59,60]. Reducing the size of the material to the nano scale may modify the physico-chemical properties compared to the same material at larger-size scales, such as much larger surface to mass ratio, improved surface reactivity or increased ion release [61]. The nanosized zinc oxide may expose more molecules of zinc oxide to interact with the gastrointestinal tissues and microbial population. In Hendrix laying hens, Tsai et al. [62] observed that supplementation of 20 mg/kg zinc from nanosized zinc oxide (40 mg/kg zinc in basal diet) enhanced zinc retention and serum zinc, eggshell thickness, growth hormone, and carbonic anhydrase activity. Mao and Lien [63] reported that compared with conventional zinc oxide, supplementation with 100 mg/kg nanosized zinc oxide in Hendrix laying hens showed better effects on feed intake, serum zinc, ghrelin, metallothionein, immunoglobulin G (IgG), and eggshell strength. Zhao et al. [14] indicated that addition of 20 and 60 mg/kg nanosized zinc oxide improved growth performance (body weight gain) and antioxidant capacity (increased catalase activity and total antioxidant capacity in serum and reduced malondialdehyde in serum and liver) compared with 60 mg/kg conventional zinc oxide, whereas 100 mg/kg nanosized zinc oxide impaired growth rate. Although growth performance were unaffected, Li et al. [64] suggested that supplementation of 120 mg/kg both conventional zinc oxide and nanosized zinc oxide improved nutrient (crude protein, crude fat, phosphorus, and zinc) digestibility, blood IgG, and γ-globulin concentrations in weaning pigs. Moreover, pigs fed diet supplemented with nanosized zinc oxide had greater digestibility of phosphorus and zinc, blood phytohemagglutinin skin challenge, IgG, and γ-globulin values, growth hormone, carbonic anhydrase activity, and zinc concentrations in serum, compared with those fed diet with conventional zinc oxide. Milani et al. [10] suggested that low doses (15, 30, and 60 mg/kg) nanosized zinc oxide were not effective in improving production performance and controlling post-weaning diarrhea of weaning piglets, although increasing nanosized zinc oxide levels increased gain to feed ratio and reduced diarrhea occurrence during first week post-weaning. In addition, supplementation of low doses of nanosized zinc oxide increased dry matter and crude protein digestibility. More recently, Wang et al. [65] showed that supplementation of 800 mg/kg nanosized zinc oxide had comparable effects to 3,000 mg/kg conventional zinc oxide on improving growth rate, alleviating diarrhea, and improving intestinal morphology in weaning pigs. Importantly, fecal zinc was lower in pigs fed diets supplemented with low doses nanosized zinc oxide than those fed high dose of conventional zinc oxide.

https://doi.org/10.5187/jast.2022.e1

TETRABASIC ZINC CHLORIDE

Tetrabasic zinc chloride is produced through a reactive crystallisation process in which zinc chloride is reacted with ammoniated zinc chloride and water. Tetrabasic zinc chloride is intended to supply zinc in final feed for all species [66]. In weaning pigs, Mavromichalis et al. [67] found that with or without antibacterial compound (carbadox), at pharmacological levels, tetrabasic zinc chloride was an effective source of zinc for enhancing growth performance. Zhang and Guo [68] conducted an experiment to evaluate pharmacological doses of zinc from tetrabasic zinc chloride (1,500, 2,250, and 3,000 mg/kg) and conventional zinc oxide (2,250 and 3,000 mg/kg) in weaning pigs. The results reported that supplementation with both tetrabasic zinc chloride and conventional zinc oxide increased daily gain, feed intake, and gain to feed ratio, but decreased fecal scores in weaning pigs. The relative bioavailability of zinc from tetrabasic zinc chloride was 159% / 125%, and 128%, 123% and 122%, respectively, compared with conventional zinc oxide in broilers [68]. In New Hampshire × Columbian female chicks, Batal et al. [69] suggested that tetrabasic zinc chloride provided same bioavailable zinc as that of analytical-grade zinc sulfate. However, to promote the growth performance, tetrabasic zinc chloride has to be included at pharmacological levels, and therefore, the problem of high zinc excretion is still present.

POROUS ZINC

High porosity zinc oxide is a novel form of zinc oxide with enhanced surface area and more importantly greater porosity (as much as ten times higher than conventional zinc oxide) and this allows it even stronger efficacy than nanosized zinc as it further increases the possible sites of the interaction among the molecules of zinc oxide, the animals, and microbial population in the gastrointestinal tract (Animine, Sillingy, France) [70]. Vahjen et al. [71] reported that porous zinc oxide exhibited higher *ex vivo* bacterial growth depressing effects than conventional zinc oxide. In weaning pigs, Morales et al. [72] suggested that pigs fed diet supplemented with 110 mg/kg zinc from porous zinc oxide increased growth rate and gain to feed ratio compared with pigs offered diet with 3,000 mg/kg conventional zinc oxide during 42 to 63 days of age. Long et al. [73] compared the effects of the zinc from nanosized zinc oxide (400 mg/kg), porous zinc oxide (400 mg/kg), and conventional zinc oxide (2,400 mg/kg) in weaning pigs. The results indicated that dietary supplementation with low dose of porous and nanosized zinc oxide had similar effects on improving production performance and intestinal morphology and reducing diarrhea and intestinal inflammatory as high level of conventional zinc oxide in weaning pigs. In addition, compared with zinc oxide, porous zinc oxide had better effect on reducing diarrhea. Also, no information is published regarding the effects of application of porous zinc oxide in poultry.

CONCLUSION

Supplementation of pharmacological doses (2,000 to 4,000 mg/kg) of zinc, especially zinc oxide, is an effective method to improve growth performance and alleviate diarrhea post-weaning. However, the use of pharmacological doses zinc is criticized due to the large amount of zinc excretion and microbial resistance. To improve the bioavailability of zinc, several novel zinc sources have been developed. However, contradictory effects of those novel zinc sources in animal production have been reported. Therefore, further studies are required to explore the exact mechanisms involved in the antibacterial functions of zinc.

REFERENCES

1. Jensen J, Kyvsgaard NC, Battisti A, Baptiste KE. Environmental and public health related risk of veterinary zinc in pig production - using Denmark as an example. Environ Int. 2018;114:81-90. https://doi.org/10.1016/j.envint.2018.02.007
2. Todd WR, Elvehjem CA, Hart EB. Zinc in the nutrition of the rat. Am J Physiol. 1933;107:146-56. https://doi.org/10.1152/ajplegacy.1933.107.1.146
3. Maywald M, Rink L. Zinc homeostasis and immunosenescence. J Trace Elem Med Biol. 2015;29:24-30. https://doi.org/10.1016/j.jtemb.2014.06.003
4. Liu Y, Espinosa CD, Abelilla JJ, Casas GA, Lagos LV, Lee SA, et al. Non-antibiotic feed additives in diets for pigs: a review. Anim Nutr. 2018;4:113-25. https://doi.org/10.1016/j.aninu.2018.01.007
5. Gresse R, Chaucheyras-Durand F, Fleury MA, Van de Wiele T, Forano E, Blanquet-Diot S. Gut microbiota dysbiosis in postweaning piglets: understanding the keys to health. Trends Microbiol. 2017;25:851-73. https://doi.org/10.1016/j.tim.2017.05.004
6. Bednorz C, Oelgeschläger K, Kinnemann B, Hartmann S, Neumann K, Pieper R, et al. The broader context of antibiotic resistance: zinc feed supplementation of piglets increases the proportion of multi-resistant Escherichia coli in vivo. Int J Med Microbiol. 2013;303:396-403. https://doi.org/10.1016/j.ijmm.2013.06.004
7. Vahjen W, Pietruszyńska D, Starke IC, Zentek J. High dietary zinc supplementation increases the occurrence of tetracycline and sulfonamide resistance genes in the intestine of weaned pigs. Gut Pathog. 2015;7:23. https://doi.org/10.1186/s13099-015-0071-3
8. van Alen S, Kaspar U, Idelevich EA, Köck R, Becker K. Increase of zinc resistance in German human derived livestock-associated MRSA between 2000 and 2014. Vet Microbiol. 2018;214:7-12. https://doi.org/10.1016/j.vetmic.2017.11.032
9. Park BC, Jung DY, Kang SY, Ko YH, Ha DM, Kwon CH, et al. Effects of dietary supplementation of a zinc oxide product encapsulated with lipid on growth performance, intestinal morphology, and digestive enzyme activities in weanling pigs. Anim Feed Sci Technol. 2015;200:112-7. https://doi.org/10.1016/j.anifeedsci.2014.11.016
10. Milani NC, Sbardella M, Ikeda NY, Arno A, Mascarenhas BC, Miyada VS. Dietary zinc oxide nanoparticles as growth promoter for weanling pigs. Anim Feed Sci Technol. 2017;227:13-23. https://doi.org/10.1016/j.anifeedsci.2017.03.001
11. Wang C, Zhang L, Su W, Ying Z, He J, Zhang L, et al. Zinc oxide nanoparticles as a substitute for zinc oxide or colistin sulfate: effects on growth, serum enzymes, zinc deposition, intestinal morphology and epithelial barrier in weaned piglets. PLOS ONE. 2017;12:e0181136. https://doi.org/10.1371/journal.pone.0181136
12. Kołodziejczak-Radzimska A, Jesionowski T. Zinc oxide—from synthesis to application: a review. Materials. 2014;7:2833-81. https://doi.org/10.3390/ma7042833
13. Shen J, Chen Y, Wang Z, Zhou A, He M, Mao L, et al. Coated zinc oxide improves intestinal immunity function and regulates microbiota composition in weaned piglets. Br J Nutr. 2014;111:2123-34. https://doi.org/10.1017/S0007114514000300
14. Zhao CY, Tan SX, Xiao XY, Qiu XS, Pan JQ, Tang ZX. Effects of dietary zinc oxide nanoparticles on growth performance and antioxidative status in broilers. Biol Trace Elem Res. 2014;160:361-7. https://doi.org/10.1007/s12011-014-0052-2
15. Sirelkhatim A, Mahmud S, Seeni A, Kaus NHM, Ann LC, Bakhori SKM, et al. Review on zinc oxide nanoparticles: antibacterial activity and toxicity mechanism. Nano-Micro Lett. 2015;7:219-42. https://doi.org/10.1007/s40820-015-0040-x

16. Swain PS, Rao SBN, Rajendran D, Dominic G, Selvaraju S. Nano zinc, an alternative to conventional zinc as animal feed supplement: a review. Anim Nutr. 2016;2:134-41. https://doi.org/10.1016/j.aninu.2016.06.003
17. Vahjen W, Roméo A, Zentek J. Impact of zinc oxide on the immediate postweaning colonization of enterobacteria in pigs. J Anim Sci. 2016;94:359-63. https://doi.org/10.2527/jas.2015-9795
18. Revy PS, Jondreville C, Dourmad JY, Nys Y. Effect of zinc supplemented as either an organic or an inorganic source and of microbial phytase on zinc and other minerals utilisation by weanling pigs. Anim Feed Sci Technol. 2004;116:93-112. https://doi.org/10.1016/j.anifeedsci.2004.04.003
19. Blavi L, Sola-Oriol D, Perez JF, Stein HH. Effects of zinc oxide and microbial phytase on digestibility of calcium and phosphorus in maize-based diets fed to growing pigs. J Anim Sci. 2017;95:847-54. https://doi.org/10.2527/jas.2016.1149
20. Casewell M, Friis C, Marco E, McMullin P, Phillips I. The European ban on growth-promoting antibiotics and emerging consequences for human and animal health. J Antimicrob Chemother. 2003;52:159-61. https://doi.org/10.1093/jac/dkg313
21. Maron DF, Smith TJS, Nachman KE. Restrictions on antimicrobial use in food animal production: an international regulatory and economic survey. Glob Health. 2013;9:48. https://doi.org/10.1186/1744-8603-9-48
22. Levy S. Reduced antibiotic use in livestock: how Denmark tackled resistance. Environ Health Perspect. 2014;122:A160-5. https://doi.org/10.1289/ehp.122-A160
23. Dowarah R, Verma AK, Agarwal N, Singh P, Singh BR. Selection and characterization of probiotic lactic acid bacteria and its impact on growth, nutrient digestibility, health and antioxidant status in weaned piglets. PLOS ONE. 2018;13:e0192978. https://doi.org/10.1371/journal.pone.0192978
24. Ou D, Li D, Cao Y, Li X, Yin J, Qiao S, et al. Dietary supplementation with zinc oxide decreases expression of the stem cell factor in the small intestine of weanling pigs. J Nutr Biochem. 2007;18:820-6. https://doi.org/10.1016/j.jnutbio.2006.12.022
25. Yin J, Li X, Li D, Yue T, Fang Q, Ni J, et al. Dietary supplementation with zinc oxide stimulates ghrelin secretion from the stomach of young pigs. J Nutr Biochem. 2009;20:783-90. https://doi.org/10.1016/j.jnutbio.2008.07.007
26. Sargeant HR, McDowall KJ, Miller HM, Shaw MA. Dietary zinc oxide affects the expression of genes associated with inflammation: transcriptome analysis in piglets challenged with ETEC K88. Vet Immunol Immunopathol. 2010;137:120-9. https://doi.org/10.1016/j.vetimm.2010.05.001
27. Hu C, Song J, You Z, Luan Z, Li W. Zinc oxide–montmorillonite hybrid influences diarrhea, intestinal mucosal integrity, and digestive enzyme activity in weaned pigs. Biol Trace Elem Res. 2012;149:190-6. https://doi.org/10.1007/s12011-012-9422-9
28. Walk CL, Wilcock P, Magowan E. Evaluation of the effects of pharmacological zinc oxide and phosphorus source on weaned piglet growth performance, plasma minerals and mineral digestibility. Animal. 2015;9:1145-52. https://doi.org/10.1017/S175173111500035X
29. Han YK, Thacker PA. Effects of antibiotics, zinc oxide or a rare earth mineral-yeast product on performance, nutrient digestibility and serum parameters in weanling pigs. Asian-Australas J Anim Sci. 2010;23:1057-65. https://doi.org/10.5713/ajas.2010.90569
30. Heo JM, Kim JC, Hansen CF, Mullan BP, Hampson DJ, Maribo H, et al. Effects of dietary protein level and zinc oxide supplementation on the incidence of post-weaning diarrhoea in weaner pigs challenged with an enterotoxigenic strain of Escherichia coli. Livest Sci.

2010;133:210-3. https://doi.org/10.1016/j.livsci.2010.06.066

31. Sales J. Effects of pharmacological concentrations of dietary zinc oxide on growth of post-weaning pigs: a meta-analysis. Biol Trace Elem Res. 2013;152:343-9. https://doi.org/10.1007/s12011-013-9638-3
32. Zhang B, Guo Y. Supplemental zinc reduced intestinal permeability by enhancing occludin and zonula occludens protein-1 (ZO-1) expression in weaning piglets. Br J Nutr. 2009;102:687-93. https://doi.org/10.1017/S0007114509289033
33. Pearce SC, Sanz Fernandez MV, Torrison J, Wilson ME, Baumgard LH, Gabler NK. Dietary organic zinc attenuates heat stress–induced changes in pig intestinal integrity and metabolism. J Anim Sci. 2015;93:4702-13. https://doi.org/10.2527/jas.2015-9018
34. Carlson D, Poulsen HD, Sehested J. Influence of weaning and effect of post weaning dietary zinc and copper on electrophysiological response to glucose, theophylline and 5-HT in piglet small intestinal mucosa. Comp Biochem Physiol A Mol Integr Physiol. 2004;137:757-65. https://doi.org/10.1016/j.cbpb.2004.02.011
35. Li X, Yin J, Li D, Chen X, Zang J, Zhou X. Dietary supplementation with zinc oxide increases IGF-I and IGF-I receptor gene expression in the small intestine of weanling piglets. J Nutr. 2006;136:786-91. https://doi.org/10.1093/jn/136.7.1786
36. Jensen-Waern M, Melin L, Lindberg R, Johannisson A, Petersson L, Wallgren P. Dietary zinc oxide in weaned pigs — effects on performance, tissue concentrations, morphology, neutrophil functions and faecal microflora. Res Vet Sci. 1998;64:225-31. https://doi.org/10.1016/S0034-5288(98)90130-8
37. Katouli M, Melin L, Jensen-Waern M, Wallgren P, Möllby R. The effect of zinc oxide supplementation on the stability of the intestinal flora with special reference to composition of coliforms in weaned pigs. J Appl Microbiol. 1999;87:564-73. https://doi.org/10.1046/j.1365-2672.1999.00853.x
38. Wang YZ, Xu ZR, Lin WX, Huang HQ, Wang ZQ. Developmental gene expression of antimicrobial peptide PR-39 and effect of zinc oxide on gene regulation of PR-39 in piglets. Asian-Australas J Anim Sci. 2004;17:1635-40. https://doi.org/10.5713/ajas.2004.1635
39. Lichten LA, Cousins RJ. Mammalian zinc transporters: nutritional and physiologic regulation. Annu Rev Nutr. 2009;29:153-76. https://doi.org/10.1146/annurev-nutr-033009-083312
40. Brugger D, Windisch WM. Strategies and challenges to increase the precision in feeding zinc to monogastric livestock. Anim Nutr. 2017;3:103-8. https://doi.org/10.1016/j.aninu.2017.03.002
41. Starke IC, Pieper R, Neumann K, Zentek J, Vahjen W. The impact of high dietary zinc oxide on the development of the intestinal microbiota in weaned piglets. FEMS Microbiol Ecol. 2014;87:416-27. https://doi.org/10.1111/1574-6941.12233
42. Yang Q, Ma Y, Zhu J. Applying a novel electrostatic dry powder coating technology to pellets. Eur J Pharm Biopharm. 2015;97:118-24. https://doi.org/10.1016/j.ejpb.2015.10.006
43. Martín MJ, Lara-Villoslada F, Ruiz MA, Morales ME. Microencapsulation of bacteria: a review of different technologies and their impact on the probiotic effects. Innov Food Sci Emerg Technol. 2015;27:15-25. https://doi.org/10.1016/j.ifset.2014.09.010
44. Pan L, Zhao PF, Yang ZY, Long SF, Wang HL, Tian QY, et al. Effects of coated compound proteases on apparent total tract digestibility of nutrients and apparent ileal digestibility of amino acids for pigs. Asian-Australas J Anim Sci. 2016;29:1761-7. https://doi.org/10.5713/ajas.16.0041
45. Byun YJ, Lee CY, Kim MH, Jung DY, Han JH, Jang I, et al. Effects of dietary supplementation of a lipid-coated zinc oxide product on the fecal consistency, growth, and morphology of the intestinal mucosa of weanling pigs. J Anim Sci Technol. 2017;59:29. https://doi.org/10.1186/

s40781-017-0159-z
46. Song YM, Kim MH, Kim HN, Jang I, Han JH, Fontamillas GA, et al. Effects of dietary supplementation of lipid-coated zinc oxide on intestinal mucosal morphology and expression of the genes associated with growth and immune function in weanling pigs. Asian-Australas J Anim Sci. 2018;31:403-9. https://doi.org/10.5713/ajas.17.0718
47. Kwon CH, Lee CY, Han SJ, Kim SJ, Park BC, Jang I, et al. Effects of dietary supplementation of lipid-encapsulated zinc oxide on colibacillosis, growth and intestinal morphology in weaned piglets challenged with enterotoxigenic Escherichia coli. Anim Sci J. 2014;85:805-13. https://doi.org/10.1111/asj.12215
48. Upadhaya SD, Ki YM, Lee KY, Kim IH. Use of protected zinc oxide in lower doses in weaned pigs in substitution for the conventional high dose zinc oxide. Anim Feed Sci Technol. 2018;240:1-10. https://doi.org/10.1016/j.anifeedsci.2018.03.012
49. Hill GM, Mahan DC, Jolliff JS. Comparison of organic and inorganic zinc sources to maximize growth and meet the zinc needs of the nursery pig. J Anim Sci. 2014;92:1582-94. https://doi.org/10.2527/jas.2013-6744
50. Wang Y, Tang JW, Ma WQ, Feng J, Feng J. Dietary zinc glycine chelate on growth performance, tissue mineral concentrations, and serum enzyme activity in weanling piglets. Biol Trace Elem Res. 2010;133:325-34. https://doi.org/10.1007/s12011-009-8437-3
51. Case CL, Carlson MS. Effect of feeding organic and inorganic sources of additional zinc on growth performance and zinc balance in nursery pigs. J Anim Sci. 2002;80:1917-24. https://doi.org/10.2527/2002.8071917x
52. Buff CE, Bollinger DW, Ellersieck MR, Brommelsiek WA, Veum TL. Comparison of growth performance and zinc absorption, retention, and excretion in weanling pigs fed diets supplemented with zinc-polysaccharide or zinc oxide. J Anim Sci. 2005;83:2380-6. https://doi.org/10.2527/2005.83102380x
53. Cheng J, Kornegay ET, Schell, T. Influence of dietary lysine on the utilization of zinc from zinc sulfate and a zinc-lysine complex by young pigs. J Anim Sci. 1998;76:1064-74. https://doi.org/10.2527/1998.7641064x
54. Payne RL, Bidner TD, Fakler TM, Southern LL. Growth and intestinal morphology of pigs from sows fed two zinc sources during gestation and lactation. J Anim Sci. 2006;84:2141-9. https://doi.org/10.2527/jas.2005-627
55. Moghaddam HN, Jahanian R. Immunological responses of broiler chicks can be modulated by dietary supplementation of zinc-methionine in place of inorganic zinc sources. Asian-Australas J Anim Sci. 2009;22:396-403. https://doi.org/10.5713/ajas.2009.80473
56. Ao T, Pierce JL, Pescatore AJ, Cantor AH, Dawson KA, Ford MJ, et al. Effects of organic zinc and phytase supplementation in a maize–soybean meal diet on the performance and tissue zinc content of broiler chicks. Br Poult Sci. 2007;48:690-5. https://doi.org/10.1080/00071660701694072
57. Rossi P, Rutz F, Anciuti MA, Rech JL, Zauk NHF. Influence of graded levels of organic zinc on growth performance and carcass traits of broilers. J Appl Poult Res. 2007;16:219-25. https://doi.org/10.1093/japr/16.2.219
58. Salim HM, Lee HR, Jo C, Lee SK, Lee BD. Effect of sex and dietary organic zinc on growth performance, carcass traits, tissue mineral content, and blood parameters of broiler chickens. Biol Trace Elem Res. 2012;147:120-9. https://doi.org/10.1007/s12011-011-9282-8
59. Padmavathy N, Vijayaraghavan R. Enhanced bioactivity of ZnO nanoparticles—an antimicrobial study. Sci Technol Adv Mater. 2008;9:035004. https://doi.org/10.1088/1468-6996/9/3/035004
60. Piccinno F, Gottschalk F, Seeger S, Nowack B. Industrial production quantities and uses of ten

engineered nanomaterials in Europe and the world. J Nanopart Res. 2012;14:1109. https://doi.org/10.1007/s11051-012-1109-9
61. Peters RJB, Bouwmeester H, Gottardo S, Amenta V, Arena M, Brandhoff P, et al. Nanomaterials for products and application in agriculture, feed and food. Trends Food Sci Technol. 2016;54:155-64. https://doi.org/10.1016/j.tifs.2016.06.008
62. Tsai YH, Mao SY, Li MZ, Huang JT, Lien TF. Effects of nanosize zinc oxide on zinc retention, eggshell quality, immune response and serum parameters of aged laying hens. Anim Feed Sci Technol. 2016;213:99-107. https://doi.org/10.1016/j.anifeedsci.2016.01.009
63. Mao SY, Lien TF. Effects of nanosized zinc oxide and γ-polyglutamic acid on eggshell quality and serum parameters of aged laying hens. Arch Anim Nutr. 2017;71:373-83. https://doi.org/10.1080/1745039X.2017.1355600
64. Li MZ, Huang JT, Tsai YH, Mao SY, Fu CM, Lien TF. Nanosize of zinc oxide and the effects on zinc digestibility, growth performances, immune response and serum parameters of weanling piglets. Anim Sci J. 2016;87:1379-85. https://doi.org/10.1111/asj.12579
65. Wang C, Zhang L, Ying Z, He J, Zhou L, Zhang L, et al. Effects of dietary zinc oxide nanoparticles on growth, diarrhea, mineral deposition, intestinal morphology, and barrier of weaned piglets. Biol Trace Elem Res. 2018;185:364-74. https://doi.org/10.1007/s12011-018-1266-5
66. FEEDAP [EFSA Panel on Additives and Products or Substances used in Animal Feed]. Scientific opinion on the safety and efficacy of tetra-basic zinc chloride for all animal species. EFSA J. 2012;10:2672.
67. Mavromichalis I, Webel DM, Parr EN, Baker DH. Growth-promoting efficacy of pharmacological doses of tetrabasic zinc chloride in diets for nursery pigs. Can J Anim Sci. 2001;81:387-91. https://doi.org/10.4141/A01-005
68. Zhang B, Guo Y. Beneficial effects of tetrabasic zinc chloride for weanling piglets and the bioavailability of zinc in tetrabasic form relative to ZnO. Anim Feed Sci Technol. 2007;135:75-85. https://doi.org/10.1016/j.anifeedsci.2006.06.006
69. Batal AB, Parr TM, Baker DH. Zinc bioavailability in tetrabasic zinc chloride and the dietary zinc requirement of young chicks fed a soy concentrate diet. Poult Sci. 2001;80:87-90. https://doi.org/10.1093/ps/80.1.87
70. Campbell JM, Crenshaw JD, Polo J. The biological stress of early weaned piglets. J Anim Sci Biotechnol. 2013;4:19. https://doi.org/10.1186/2049-1891-4-19
71. Vahjen W, Zentek J, Durosoy S. Inhibitory action of two zinc oxide sources on the ex vivo growth of porcine small intestine bacteria. J Anim Sci. 2012;90:334-6. https://doi.org/10.2527/jas.52921
72. Morales J, Cordero G, Piñeiro C, Durosoy S. Zinc oxide at low supplementation level improves productive performance and health status of piglets. J Anim Sci. 2012;90:436-8. https://doi.org/10.2527/jas.53833
73. Long L, Chen J, Zhang Y, Liang X, Ni H, Zhang B, et al. Comparison of porous and nano zinc oxide for replacing high-dose dietary regular zinc oxide in weaning piglets. PLOS ONE. 2017;12:e0182550. https://doi.org/10.1371/journal.pone.0182550