

Use of Organic Acids as a Model to Study the Impact of Gut Microflora on Nutrition and Metabolism¹

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SUMMARY

Approvals for the use of nontherapeutic antibiotics in animal feed are fast disappearing worldwide. The primary effect of antibiotics is antimicrobial; all of the digestibility and performance effects can be explained by their impact on the gastrointestinal microflora. Among the candidate replacements for antibiotics are organic acids, both individual acids and blends of several acids. Like antibiotics, short-chain organic acids also have a specific antimicrobial activity. Unlike antibiotics, the antimicrobial activity of organic acids is pH dependent. Organic acids have a clear and significant benefit in weanling piglets and have been observed to benefit poultry performance. Organic acids have antimicrobial activity; however, there appear to be effects of organic acids beyond those attributed to antimicrobial activity. Reductions in bacteria are associated with feeding organic acids, which are particularly effective against acid-intolerant species such as *E. coli*, *Salmonella* and *Campylobacter*. Both antibiotics and organic acids improve protein and energy digestibilities by reducing microbial competition with the host for nutrients and endogenous nitrogen losses, by lowering the incidence of subclinical infections and secretion of immune mediators, and by reducing production of ammonia and other growth-depressing microbial metabolites. Organic acids have several additional effects that go beyond those of antibiotics. These include reduction in digesta pH, increased pancreatic secretion, and trophic effects on the gastrointestinal mucosa. Much more is known about these effects in swine than in poultry. There appears to be more variability in detecting an organic acid benefit in comparison to that observed with antibiotics. Lack of consistency in demonstrating an organic acid benefit is related to uncontrolled variables such as buffering capacity of dietary ingredients, presence of other antimicrobial compounds, cleanliness of the production environment, and heterogeneity of gut microbiota. Additional research can clarify the role of these factors and how to minimize their impact.

Key words: organic acid, gut microflora, metabolism, nutrition

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DESCRIPTION OF PROBLEM

Gastrointestinal microbial populations—ubiquitous and heterogeneous—play a complex

role in nutrition and growth that is incompletely understood despite many years of research. Competition for nutrients by the gut microflora of agricultural animals, including poultry, has

been managed in part through the use of low levels of antibiotics. This option is rapidly disappearing. As a result, we need to understand better the role of the microflora in order to manage its effects on nutrition, growth, health, and disease. Although investigators and producers are seeking a replacement for antibiotics, no single treatment or product has been successful in replicating their relatively consistent and robust effects on performance. Among the candidate replacements for antibiotics are organic acids, both individual acids and blends of several acids. These have been used in swine diets for decades and appear to provide many of the benefits of antibiotics. This review will discuss the effects of organic acids on performance and will consider whether all of these effects are solely due to antimicrobial activity or whether other mechanisms of action are involved. The lack of consistency in results, particularly in poultry, has led to uncertainty about the value of organic acids. The causes of variability in performance will be discussed.

INTRODUCTION

As a group of chemicals, organic acids are considered to be any organic carboxylic acid, including fatty acids and amino acids, of the general structure R-COOH. Not all of these acids have effects on gut microflora. In fact, the organic acids associated with specific antimicrobial activity are short-chain acids (C1–C7) and are either simple monocarboxylic acids such as formic, acetic, propionic and butyric acids, or are carboxylic acids bearing an hydroxyl group (usually on the α carbon) such as lactic, malic, tartaric, and citric acids. Salts of some of these acids have also been shown to have performance benefits. Other acids, such as sorbic and fumaric acids, have some antifungal activity and are short chain-carboxylic acids containing double bonds. Organic acids are weak acids and are only partly dissociated. Most organic acids with antimicrobial activity have a pKa—the pH at which the acid is half dissociated—between 3 and 5.

Table 1 shows the common name, chemical name, formula, molecular weight, and first pKa of organic acids that are commonly used as dietary acidifiers for pigs or poultry. In this review, the term organic acids will be used to refer to

the group of acids, or their salts, that have been demonstrated to have animal performance benefits and antimicrobial activity. Table 1 includes a novel addition, 2-hydroxy-4- (methylthio) butanoic acid (HMB) [1]. This is a short-chain (C4) monocarboxylic acid with an hydroxyl group on the α carbon and a pKa between 3 and 4. It is a feed additive more commonly known for its activity as a methionine source (Alimet feed supplement) but is in fact an organic acid until it is converted to methionine within the body. Performance benefits beyond methionine supplementation confirm its activity as an organic acid in the feed and gut of piglets prior to absorption and conversion to methionine [2].

Many of the organic acids with beneficial effects on animal performance are also known to be effective food and feed preservatives. The magnitude of their antimicrobial effects varies from one acid to another and is dependent on concentration and pH [3]. Figure 1 shows the pH dependence of HMB, lactic and formic acids and compares their antimicrobial activity to a mineral acid, HCl [4]. There was little antimicrobial activity at pH 7.3, but at pH 4, all acids had some activity toward *E. coli* with HCl being the weakest, followed by lactic acid. Formic acid and HMB had the strongest activity in this experiment, resulting in complete bacteriolysis at 24 h.

In addition, each acid has its own spectrum of antimicrobial activity. For example, sorbic acid is better known for its antimold activity, whereas lactic acid is more effective against bacteria. Some acids, such as formic, propionic, and HMB have broader antimicrobial activities and can be effective against bacteria and fungi, including yeast [5, 6, 7]. This spectrum of activities has led to the evaluation and use of blends of organic acids in animal feed [8]. Blends of some acids have been reported to have synergistic antimicrobial activity in vitro [9].

ANIMAL PERFORMANCE

Swine

Several studies have documented the effects of organic acids on performance in young swine, particularly early-weaned piglets. A recent publication by Partanen [10] reviews the literature in this area and provides the results of a meta-analysis of existing data. Only studies using indi-

TABLE 1. List of acids and their properties

Acid	Chemical name	Formula	MW	pKa
Formic	Formic Acid	HCOOH	46.03	3.75
Acetic	Acetic Acid	CH ₃ COOH	60.05	4.76
Propionic	2-Propanoic Acid	CH ₃ CH ₂ COOH	74.08	4.88
Butyric	Butanoic Acid	CH ₃ CH ₂ CH ₂ COOH	88.12	4.82
Lactic	2-Hydroxypropanoic Acid	CH ₃ CH(OH)COOH	90.08	3.83
Sorbic	2,4-Hexadienoic Acid	CH ₃ CH:CHCH:CHCOOH	112.14	4.76
Fumaric	2-Butenedioic Acid	COOHCH:CHCOOH	116.07	3.02
HMB	2-Hydroxy-4-Methylthio Butanoic Acid	CH ₃ SCH ₂ CH ₂ CH(OH)COOH	149.00	3.86
Malic	Hydroxybutanedioic Acid	COOHCH ₂ CH(OH)COOH	134.09	3.40
Tartaric	2,3-Dihydroxy-Butanedioic Acid	COOHCH(OH)CH(OH)COOH	150.09	2.93
Citric	2-Hydroxy-1,2,3-Propanetricarboxylic Acid	COOHCH ₂ C(OH)(COOH)CH ₂ COOH	192.14	3.13

vidual acids in the absence of antibiotics and copper are considered. In the analysis of 46 weaned-piglet and 23 fattening-pig trials, significant feed-to-gain improvements were observed with formic, fumaric, and citric acids and also with potassium diformate. Weight gain and feed intake effects were significant for formic acid and potassium diformate. The author concludes that dietary acids have a beneficial effect, especially on weaned piglets, that is primarily associated with changes in the gastrointestinal microflora [10].

Poultry

There is also a body of literature for poultry, albeit much smaller. Several studies have been done using fumaric acid. An early study by Vogt and Matthes [11] reported on the effect of fumaric acid in broilers and laying hens. Fumaric acid improved feed efficiency by 3.5 to 4% in broilers. Layer feed efficiency was also improved and rate of lay was not affected. Patten and Waldroup [12] reported a significant improvement in weight gain of broilers using 0.5 and 1.0% fumaric acid, but there was no effect on feed use. Higher acid concentrations were associated with reductions in feed intake and body weights. Skinner et al. [13] reported a significant improvement of 49-d body weight and feed utilization in male broilers fed 0, 0.125, 0.25, or 0.5% fumaric acid. Mortality rates, abdominal fat percentages, and dressing percentages were not affected. A similar study was reported by Runho et al. [14] in which 0.25 to 1.0% fumaric acid was compared to an antibiotic growth promoter (Nitrovin) fed to Hubbard broilers. Growth was not affected, but feed con-

sumption was reduced, resulting in a significant improvement in feed to gain. Feed efficiencies for 0.5, 0.75, and 1% fumaric acid were comparable to the antibiotic control [14]. Associated with this performance improvement was a significant improvement in apparent metabolizable energy that was dose related. Linear regression analysis indicated an increase of 183 kcal/kg for each 1% of fumaric acid added.

There are also studies testing performance effects of feeding propionic, malic, sorbic, tartaric, lactic, and formic acids. Effects of buffered propionic acid in the presence and absence of bacitracin or Roxarsone were reported by Izat et al. [15], who found a significant increase in dressing percentage for female broilers and a significant reduction in abdominal fat for males at 49d. There were no other performance effects. Vogt et al. [16] studied malic, sorbic, and tartaric acids (0.5 to 2%) in broilers. They reported increases in weight gain, with optimal levels of 1.12 and 0.33% for sorbic and tartaric acids, respectively. Sorbic and malic acids also tended to improve feed efficiency. Versteegh and Jongbloed [17] tested the effect of dietary lactic acid on performance of broilers from 0 to 6 wk of age. Body weight gains tended to be greater, whereas feed-to-gain ratios were significantly improved when birds were fed 2% lactic acid.

DISCUSSION

The results described above serve to confirm the general impression that organic acids benefit the animal, but that the results are notably inconsistent. The remainder of this review will consider two areas of investigation. First, how does the mechanism of action of organic acids com-

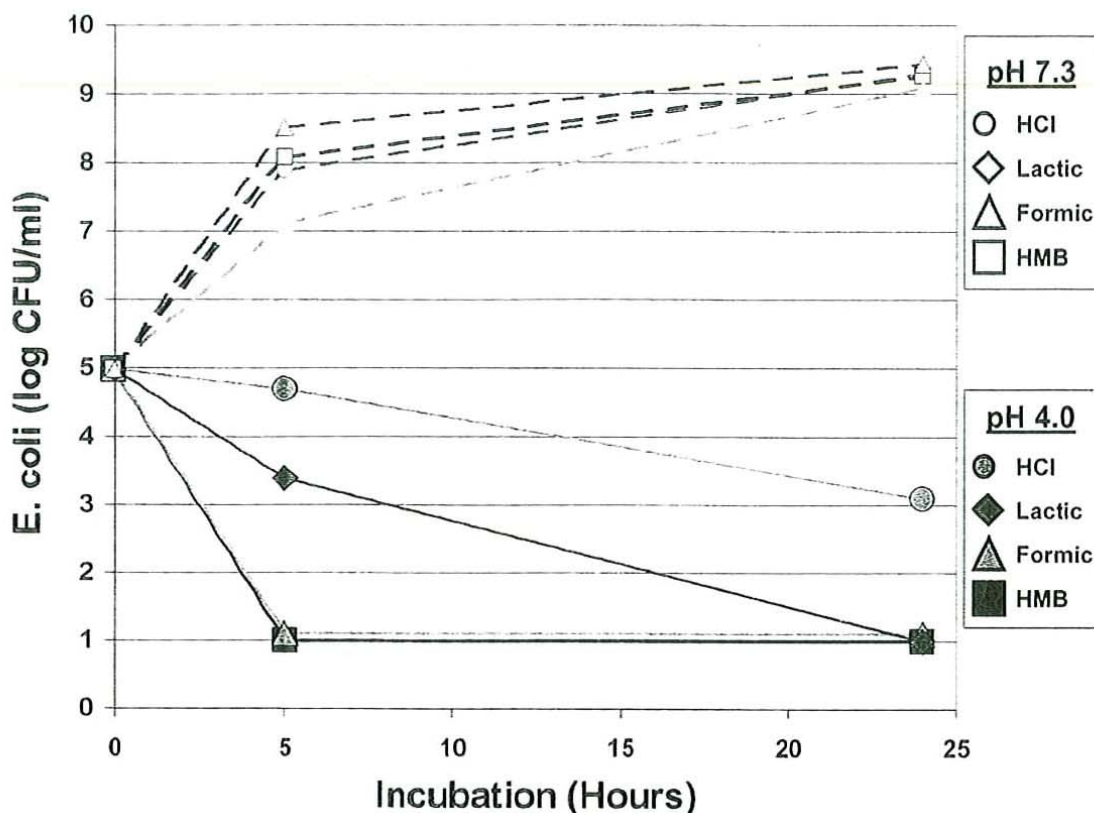


FIGURE 1. Effect of pH on the antimicrobial activity of HCl, lactic acid, formic acid and 2-hydroxy-4-(methylthio)butanoic acid. An *E. coli* inoculum (10^6 CFU) was grown in trypticase soy broth (TSB) containing either hydrochloric, lactic, formic or hydroxy(methylthio)butanoic (HMB) acids at either pH 4 or pH 7.3. Samples were taken at 5 and 24 hr for enumeration. There was little bacteriostatic activity by any of the acids at pH 7.3. Samples of *E. coli* inoculated into TSB containing formic acid or HMB at pH 4 showed total bacteriolysis.

pare to that of antibiotics? Are the benefits solely associated with antimicrobial activity, or are there other actions that contribute to the effects of organic acids? Second, why are the results inconsistent, with some studies showing multiple effects, some with one effect and others with none? What factors contribute to this inconsistency?

MECHANISM OF ACTION

Effects Related to Antimicrobial Activity

The first effect of organic acids in animal agriculture is related to feed preservation. Organic acids such as sorbic and propionic acids have long been used to control spoilage of feeds. The activity of organic acids toward gut microflora is very similar. In both cases, the acid changes the microbial populations in accordance with its antimicrobial spectrum of activity. For feeds, the activity to control fungal growth domi-

nates, whereas in the gut the populations being affected are primarily bacteria whose growth is most affected by acidic conditions. It should be emphasized, however, that the mechanism of action of organic acids is quite different from, and in addition to, that of inorganic acids such as HCl [18].

The importance of low pH on the antimicrobial activity of organic acids can be explained by its effect on the dissociation of the acid. At low pH, more of the organic acid will be in the undissociated form. Undissociated organic acids are lipophilic and can diffuse across cell membranes, including those of bacteria and molds [8, 10]. Once in the bacterial cell, the higher pH of its cytoplasm causes dissociation of the acid, and the resulting reduction in pH of the cell contents will disrupt enzymatic reactions and nutrient transport systems [19]. In addition, the process of transporting the free proton out of the cell requires energy, which will contribute

to reduced energy availability for proliferation, resulting in some degree of bacteriostasis, as is shown in Figure 1.

This direct antimicrobial activity is responsible for feed and food sanitation effects that contribute to the use of organic acids as preservatives. It also explains why a synergistic effect exists between mineral acids such as orthophosphoric acid and organic acids. The presence of orthophosphoric acid reduces the digesta pH, allowing more of the organic acid to be present in the undissociated form. Direct antimold effects are the mechanism responsible for reducing mold counts in wet litter [20].

After ingestion, direct antimicrobial activity is of greatest magnitude in the foregut, which has a very limited capability to change the digesta pH. This includes the crop and gizzard of poultry and the stomach of swine. Organic acid activity will reduce the total microbial load but will be particularly effective against *E. coli* and other acid-intolerant organisms. Many of these pathogens are opportunistic, such as *Campylobacter* and *Salmonella*. A consequent reduction in subclinical infections may contribute to improved nutrient digestibility and a reduction in nutrient demand by the gut-associated immune tissue. Reduced microbial activities in the swine stomach [21, 22] and upper small intestine [23, 24] have been reported. Similar reductions have been observed in the small intestine, cloaca, and postchill carcass in poultry [15, 25]. The relatively low pH of the upper gut tends to favor not only the antimicrobial activity of organic acids but also their absorption by diffusion into the gut epithelium [8]. Like many organic acids, absorption of HMB is predominantly by diffusion [26], but in addition, it is transported by a proton-dependent carrier system that also transports another organic acid, lactic acid, in the lower small intestine [27].

Antimicrobial action in the crop is an important part of the organic acid benefit as it is a major site of colonization for *E. coli* and *Salmonella* [28]. It should be noted, however, that it is highly desirable for organic acid activity to persist into the lower gut where many of the anaerobic opportunistic pathogens are found. Lower microbial proliferation in the ileum is also important because it reduces the competition of the microflora with the host for endogenous ni-

trogen lost into the gut lumen by pancreatic and gut epithelial secretions and by enterocyte attrition and shedding. Up to 50% of the ileal nitrogen is of endogenous origin, and reduced microbial competition for it has been shown to improve nitrogen retention in pigs fed formic acid [29].

The higher luminal pH in the lower gut would appear to favor the dissociated form of the acid, which would reduce uptake by diffusion, but an acidic microenvironment exists at the gut epithelial surface and permits diffusion of the undissociated form into the bacteria and into the enterocytes themselves [30]. Persistence of organic acid antimicrobial activity into the jejunum and ileum is also critical to another of its mechanisms of action. Lower microbial proliferation in the jejunum reduces the competition of the microflora with the host for nutrients. This reduction in competition is one of the mechanisms responsible for improved digestibility. Improved digestibility has been reported in swine by numerous researchers [10, 18].

Improved dry matter, organic matter, and nitrogen digestibility by dietary citric acid (1%) was observed in early weaned piglets by Scipioni et al. [22]. More recently, Mroz et al. [31] reported significant improvements of up to 5% in apparent ileal digestibility of crude protein and essential amino acids by dietary formic, fumaric, and n-butyric acids. Total tract digestibilities of dry matter, organic matter, calcium (up to 8.9%), and phosphorous (up to 7%) were also improved. Best results were generally observed with n-butyric acid, resulting in a higher retention of calcium and phosphorous than with the basal diet. These improvements in digestibility were associated with significant improvements in average daily gains [31].

Dietary fumaric acid (1 to 3%) was observed by Blank et al. [32] to improve ileal digestibilities of gross energy, crude protein, and the majority of amino acids in early-weaned pigs. Similar work in growing-finishing pigs fed lactic acid (3%) was reported by Kemme et al. [33, 34], who reported improvements in ileal digestibilities of amino acids and phytic acid phosphorus. They also reported improvements in total tract digestibility of ash, calcium, and magnesium. There was a synergistic effect on total tract digestibility of phosphorous in the presence of lactic acid

and microbial phytase [33]. Similar interactions between organic acids (lactic and formic acids) and microbial phytase, resulting in improved phosphorous digestibility, were reported by Jongbloed et al. [35]. In this work, the authors also report improved digestibilities of dry matter, ash, and calcium [35]. Work with dietary HMB has demonstrated nitrogen retention increases of up to 10% in young pigs [36].

Digestibility results in poultry are limited to the observation of higher metabolizable energy in broilers fed 0.5 to 1% fumaric acid by Runho et al. [14]. Clearly much remains to be done in this area.

There are other benefits resulting from the antimicrobial activity of organic acids. Reports by Eckel et al. [37] and Eidelsburger et al. [38] describe a significant reduction in ammonia in the stomach, small intestine, and cecum of weaned piglets fed 1.25% formic acid. This finding could be due to reduced microbial deamination of amino acids, which would then be available for absorption, resulting in increased nitrogen digestibility and reduced ammonia excretion as observed in swine fed organic acids. Ammonia toxicity is well documented, and a reduction in microbial synthesis of ammonia has been proposed as part of the mechanism of the growth response associated with feeding antibiotics [39]. Eckel et al. also reported reduced concentrations of biogenic amines in the small intestines of these animals fed 0.6% formic acid [37]. These and other microbial metabolites may exert a growth-depressing effect [40]. The numerous reports of antimicrobial activity in vitro and in vivo leave little doubt that organic acids exert part of their effects through reductions in gastrointestinal microbial populations, particularly among acid-sensitive species. Are these effects solely responsible for the improvements seen with organic acids? Data from studies of antibiotics, immune mediators and germ-free animals can shed some light on this question.

Orally ingested antibiotics have an antimicrobial effect that includes the gut microflora. This reduction in microflora, and its consequences, may be the underlying mechanism for beneficial effects of antibiotics [41]. The mechanism of action must be focused on the gut since some of these antibiotics are not absorbed. Anderson et al. [42] recently reviewed the evidence

that growth-promoting antibiotics act principally through alterations in gut microbial populations. For example, no growth promotion is seen when feeding antibiotics to germ-free animals. Furthermore, administration of gastrointestinal microflora to germ-free animals results in a growth depression [42]. In poultry, the magnitude of the growth-promoting effects of antibiotics is greater in less sanitary conditions, and a growth depression in germ-free broilers can be induced through injection of bacterial metabolites such as lipopolysaccharide or immune mediators such as interleukin-1 [43]. Another effect associated with antibiotics that appears to be a direct result of antimicrobial activity is the often-reported thinning of the intestinal tract [44]. This also has been observed in germ-free animals, including chickens [45].

Effects Beyond Antimicrobial Activity

Do organic acids act only via antimicrobial activity or are there other aspects to their benefits? Certainly, the effects of antibiotics include many of those reported above for organic acids, such as improved digestibility of protein and amino acids. Reduction in ammonia and production of biogenic amines are also observed with growth-promoting antibiotics [46, 47]. Other effects, however, have been reported with organic acids that suggest benefits beyond modification of the gut microbiota. These effects include other benefits associated with acidification, including improvements in digestive enzyme activity, microbial phytase activity, and increased pancreatic secretion. Finally, there is evidence of increased growth of the gastrointestinal mucosa in the presence of organic acids, particularly fatty acids such as butyric acid.

In swine, organic acids reduce the pH of digesta in the gut lumen, particularly in the foregut [38, 48]. The examples cited here were obtained with formic acid (1.25%) or lactic acid (1%). The magnitude of the pH reduction was at its maximum in the stomach and was approximately 0.5 to 1.0 pH units. Interestingly, a reduction in small intestine pH in broilers has been observed using HMB at lower levels (0.2%). In this case, the magnitude of the reduction using mixed digesta from the upper and lower small intestine was about 0.25 to 0.35 pH units below that of the nonsupplemented diet on Day 3 (5.64

vs. 5.99, respectively). The average pH over the first 10 d was 6.13 for the basal diet and 5.91 for the HMB supplemented diet, a small, but significant, difference. In the stomach, a reduction in gastric pH activates pepsinogen and other zymogens and brings the pH of the stomach closer to the optimum for pepsin activity [8].

Another benefit of lower pH is improvement in microbial phytase activity. Microbial phytase has two pH optima, 2.5 and 4.5 to 5.7, and phytic acid is much more soluble at lower pH [8]. These effects combine to improve phosphorous digestibility and retention, as described earlier. An estimate of this organic acid benefit in germ-free animals would give an estimate of the role of reduced microflora competition in this effect.

Nutrient digestibility in weaned piglets [49, 50] and hatchling poultry [51] appears to be limited by digestive enzyme secretion. Hatchling poultry have the ability to increase pancreatic secretion [52], and chickens selected for heavy body weight have higher levels of pancreatic and small intestine enzyme levels [53]. In weanling piglets [54] and 2-wk-old calves [55], organic acids have an effect on pancreatic and bile secretion [56, 57] that is mediated by their ability to diffuse into cells when in the undissociated form and then to dissociate in response to the higher pH of the cell cytoplasm [8]. This pancreas secretion effect appears to be associated with organic acids only—not with antibiotics. The proposed mechanism is the presence of a receptor in enterocytes that responds to the dissociated proton with an increase in secretin release [54]. This secretin release response by the intestinal epithelium also occurs in sheep [58]. Unfortunately, there are no data for poultry at this time to determine whether birds also respond to organic acids with increased bile and pancreatic secretion.

A final mechanism for the organic acid benefit on performance is through a direct stimulation of gastrointestinal cell proliferation. Increases in intestinal mucosa growth in rats in response to short-chain fatty acid infusion have been reported [59]. The effect is greatest with n-butyric acid and has been observed in the colon and jejunum, where it results in increased villus height, surface area, and crypt depth and is also accompanied by an increase in gastrin [60]. The effect has been reproduced in short-term pig

colonic mucosal tissue culture [61] but not in isolated rat colonocytes [62]. In work done by Sakata [63], rats fed an elemental diet containing acetic, propionic, or n-butyric acid, or a mixture of these acids showed increases in crypt cell production rate. Tests in germ-free rats indicated that the effect was independent of the presence of gut microflora. The cause of this trophic effect is not known, although a role for the autonomic nervous system and intestinal peptide hormones has been suggested [63]. It should be noted that n-butyric acid is an important respiratory fuel for the colonic mucosa [64].

In swine, Galfi and Bokori [65] observed that feeding n-butyrate (0.17%) resulted in an increase in average daily gain of 23%. The performance effect was accompanied by a decreased coliform count, an increased *Lactobacillus spp.* count, ileal villus length, and crypt depth. Many systems were affected, including the microbiota. Consequently, the role of increased intestinal tissue and crypt cell proliferation cannot be evaluated. Once again, no data were found for poultry to confirm or eliminate a trophic effect by organic acids.

What Contributes to the Lack of Consistency in Organic Acid Benefits?

There are many successful demonstrations of an antimicrobial benefit of organic acids in animal performance, but there are also reports in which no effect was found. In addition, there are instances in which performance effects are not accompanied by changes in microflora or digestibility [66]. Although no systematic review of the literature has been done, effects of organic acids seem to be less reproducible than those of antibiotics. Several factors have been identified that affect organic acid benefits. Perhaps the most frequently cited variable is the buffering capacity of the dietary ingredients [10, 31]. The buffering capacity is a measure of the amount of acid (0.1 M HCl) required to reach a given pH (usually 3 to 5) of a 10-g slurried sample of the ingredient [67]. The ingredients that contribute most to the buffering capacity are proteins and minerals. Cereals and cereal by-products tend to have a low buffering capacity. Organic acids reduce the buffering capacity of the diet, allowing more effective acidification of the digesta in the foregut, which is critical to

effective digesta enzyme activity and control of microbial proliferation. Blank et al. [32] observed that increasing the buffering capacity from 23.5 to 56.7 decreased the ileal amino acid digestibilities by up to 10%. A recommended buffering capacity value for poultry starter diets is 0 to 10 [67]. The buffering capacity of the diet is, for the most part, an uncontrolled variable in organic acid studies and may well contribute to the lack of consistency of results.

Another factor affecting the magnitude of response to organic acids is the formulation levels and the nature of the ingredients and their impact on the gut flora. It has been shown that the negative effect of withdrawal of antibiotic growth promoters from the diet is increased in diets with high indigestible protein [68]. As with antibiotics, a diet with a low level of highly digestible protein is much less likely to exhibit an acid effect. The excess of undigested protein in the gut favors the development of a proteolytic flora, with high level of production of bacterial toxins [69] or toxic metabolites such as biogenic amines [68]. Another way dietary ingredients can dampen an organic acid effect is with the fermentation in the gut of the lactose contained in whey. This important and early production of lactic acid can hide any effect of an additional organic acid, particularly in weaned piglets.

Other ingredients have impacts that are positive or negative on the flora and that are not always well understood. Wheat is an example of an ingredient favoring more gut disorders than corn, mainly when it is freshly harvested. Here, antibacterial additives can show an improvement. On the contrary, barley is considered as favorable for gut transit and often is imposed in piglet diets for that reason. Partanen [10] noted that organic acid effects tend to be greater with wheat than maize or barley. These factors can make an organic acid benefit impossible to produce experimentally. Attention must be paid to

the dietary matrix in which the organic acid is being tested.

A related variable is the presence of antimicrobial agents in the test diet. It is rare to observe an organic acid experiment in which dietary antibiotics are included, but other antimicrobial agents are sometimes present, such as elevated copper levels or anticoccidial drugs. These agents exert their own effects on the microflora and can make an organic acid effect redundant.

Another source of variability in the published literature is the range of acids, blends, and concentrations that are being used. Perhaps the most reproducible effects are with formic acid used between 0.5 and 1.5%. However, as has been described above, all of the acids in Table 1 have been associated with animal performance benefits at least once.

Another factor complicating these studies is the environment in which the study is done. Although effects have been observed in battery studies, the antimicrobial benefits would be most evident under less sanitary conditions. Organic acids, like antibiotics, are more growth permitting than growth promoting in the sense that they can only permit the animal to grow to its genetic potential given the diet it is fed. The closer the animals are to their genetic potential, the more difficult it will be to detect any effect, which would suggest that management of the environment must be a controlled variable in organic acid studies.

Perhaps the most uncontrolled of all the variables is the microflora itself. Although the dominant species are fairly consistent, the presence in conventional animals of numerous unidentified microbial populations is unavoidable and will affect the magnitude of response. Perhaps the only way to separate the role of the microflora from that of pancreatic stimulation and trophic effects on the intestinal mucosa is to use germ-free experimental models. These models have limitations, of course, when it comes to predicting a response in a commercial environment.

CONCLUSIONS AND APPLICATIONS

1. Approvals for the use of nontherapeutic antibiotics in animal feed are fast disappearing worldwide. The primary effect of antibiotics is antimicrobial; all of the digestibility and performance effects can be explained by their effect on the gastrointestinal microflora.
2. Short-chain organic acids also have a specific antimicrobial activity that is pH dependent. Organic acids have a clear and significant benefit in weaning piglets and have been observed

to benefit poultry performance. Reductions in bacteria are associated with feeding organic acids, which are particularly effective against acid-intolerant species such as *E. coli*, *Salmonella*, and *Campylobacter*.

3. Both antibiotics and organic acids improve protein and energy digestibility by reducing microbial competition with the host for nutrients and endogenous nitrogen losses, by lowering the incidence of subclinical infections and secretion of immune mediators, and by reducing production of ammonia and other growth-depressing microbial metabolites.
4. Organic acids have several additional effects that go beyond those of antibiotics. These effects include reduction in digesta pH, increased pancreatic secretion, and trophic effects on the gastrointestinal mucosa. Much more is known about these effects in swine than in poultry.
5. Lack of consistency in demonstrating an organic acid benefit is related to uncontrolled variables such as buffering capacity of dietary ingredients, presence of other antimicrobial compounds, cleanliness of the production environment, and heterogeneity of gut microbiota. Additional research can clarify the role and management of these factors.

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