Acidifiers in Aqua Feeds: A Solution for Antibiotic-Free Feeding of Fish and Shrimp

Christian Lückstädt BIOMIN GmbH Industriestrasse 21, 3130 Herzogenburg, Austria christian.lueckstaedt@biomin.net

Routine use of antibiotics as growth promoters is a matter of debate in the animal farming industry. The EU has banned all antibiotic growth promoters (AGP) from livestock production with effect of January 2006. Alternatives to AGP are sought in a variety of forms. Acidifiers consisting of organic acids and their salts present a promising alternative. In animal nutrition, acidifiers exert their effects on performance via three different ways (Freitag 2007): (a) in the feed; (b) in the gastro-intestinal tract of the animal; and (c) due to effects on the animal's metabolism (Table 1).

Table 1: Effects of organic acids and salts in animal nutrition (Kirchgessner and Roth 1988)

	Effective Form	Effects
Feed	H^{*} H^{+} and Anion	-pH reduction -reduction of acid binding capacity -reduction of microbial growth -antibacterial effects
Intestinal tract	H⁺	-pH reduction in stomach and duodenum -improved pepsin activity
	Anion	 -complexing agents for cations -(Ca⁺⁺, Mg⁺⁺, Fe⁺⁺, Cu⁺⁺, Zn⁺⁺)
	H [∗] and Anion	-antibacterial effects -change in microbial concentrations
Metabolism		-Energy supply

Role in feed hygiene

A certain amount of fungi, bacteria or yeast is unavoidable in feeds. Under favourable conditions such microbes multiply rapidly during storage, especially at higher moisture levels (>14%) in a warm environment. Acidifiers function as conserving agents by reducing the pH of the feed, and thereby inhibiting microbial growth and thus lower the uptake of possibly pathogenic organisms and their toxic metabolites by the farmed animals.

Role in intestinal tract

The mode of action of organic acids in the intestinal tract involves two different ways: on one hand they reduce the pH-level in the stomach and particularly in the small intestine, and on the other hand inhibit growth of gramnegative bacteria through the dissociation of the acids and production of anions in the bacterial cells.

During periods of high feed intake such as when the animals are young, or when the feeds are high in protein, free hydrochloric acid levels in the stomach are reduced. This reduction negatively impacts pepsin activation and

> pancreatic enzyme secretion and impairs digestion. Providing acidifiers in the feed tackles this problem and aid in feed digestion (Eidelsburger 1997). Positive effects of organic acids on protein hydrolysis have been demonstrated (Mroz et al. 2000). Likewise feed supplementation with organic acids has been shown to lead to lower duodenal pH, improved nitrogen retention and overall increased nutrient digestibility (Øverland et al. 2000; Kluge et al. 2004).

> Growth rates of many gram-negative bacteria, like *E. coli* or *Salmonella* spp. are reduced below pH 5. Low pH

also forms a natural barrier against ascending microbes from the ileum and large intestine. Moreover, small acids are lipophilic and can diffuse across the cell membrane of gramnegative bacteria. In the more alkaline cytoplasm they dissociate and cause pH reduction. This reduction alters cell metabolism and enzyme activity thus inhibiting growth of intraluminal microbes, especially pathogens. Several investigations demonstrate a reduction in bacterial count in the stomach (Kluge et al. *Aqua Feeds: Formulation & Beyond, Volume 3 Issue 4 2006* 2004) and the duodenum (Kirchgessner and Roth 1991; Hebeler et al. 2000; Hellweg et al. 2006), while acid tolerant beneficial *Lactobacillus* spp. seem to be unaffected or may even be enhanced in number (Hellweg et al. 2006).

Role in metabolism

Most organic acids have a considerable amount of energy (Table 2). Organic acids are generally absorbed through the intestinal epithelia by passive diffusion. Short chain acids can be used in various metabolic pathways for energy generation, for instance for ATP generation in the citric cycle. As the energy content of organic acids is completely used in metabolism it should be considered in the energy

calculation of feed rations. For example, propionic acid contains one to five times more energy than wheat (Diebold and Eidelsburger 2006).

Testing organic acids in aqua feeds

Acid preservation of fish and fish viscera to produce fish silage has been a common practice and its final product has been widely used in fish feeds with reported beneficial effects (Gildbert and Raa, 1977; Åsgård and Austreng, 1981). These beneficial effects of acid preserved products caught

the attention of the scientific community to investigate the effects of these short-chain acids onto the fish feed directly. Several studies have been conducted with different species including carnivores like rainbow trout *Oncorhynchus mykiss*, Atlantic salmon *Salmo salar* and artic charr *Salvelinus alpinus*, herbivorous filter feeders (tilapia), omnivorous fish (catfish) and shrimp.

The effect of supplementation of commercial diets with sodium salts of lactic and propionic acid (10 kg/tonne of feed) was tested in Arctic charr in brackishwater at 8°C (Ringø, 1991). Fish fed the diet with added sodium lactate increased in weight from about 310 g to about 630 g in 84 days, while fish fed diets without either salts reached a final weight of only 520 g. Inclusion of sodium propionate in the diet however had a growth depressing effect

compared to the control. The gut content from Arctic charr fed the sodium lactate supplemented diet contained lower amounts of water, energy, lipid, protein and free amino acids. It has been observed that charr feeding on high doses of commercial feeds, as it often appears under aquaculture conditions, have a tendency for diarrhoea. When charr was feeding on diets containing sodium lactate, diarrhoea did not occur, probably indicating much lower amounts of remaining nutrients and water in the gut. Furthermore, it was proposed that the growth promoting effect of dietary lactate in Arctic charr is caused by the relatively slow gastric emptying rate (Gislason et al., 1996). An increased holding

Table 2: Gross energy content of selected organic acids and theirsalts (Freitag 2007)

Organic acid/salt	Solubility in Water	Gross energy (kcal/kg)	
Formic acid	Very good	1385	
Acetic acid	very good	3535	
Propionic acid	very good	4968	
Lactic acid	Good	3607	
Fumaric acid	Low	2747	
Citric acid	Good	2460	
Calcium formate	Low	931	
Sodium formate	very good	931	
Calcium propionate	Good	3965	
Calcium lactate	Low	2436	

time in the stomach augments the antibacterial potential of the lactic acid salt and can have therefore a larger inhibition effect against possible pathogenic bacteria (Sissons, 1989). The improved growth of the Artic charr did not affect the chemical composition of the fish (Ringø et al., 1994).

Rainbow trout fingerlings were fed five experimental diets, which consisted of a control diet (AquaNutro, South Africa), three diets containing 0.5, 1 and 1.5% of an organic acid blend (Bolifor FA 2400 S Aqua, KEMIRA GrowHow[®], Sweden) and a diet containing an AGP. At the end of the trial, improvement in growth was observed with increasing level of organic acid inclusion. Inclusion levels of 1 and 1.5% resulted in significant improvement in specific growth rate of the fish when compared to the control. The improvement was nearly similar Table 3: Effects of potassium diformate supplementation in diets onperformance of tilapia challenged with V. anguillarum (Ramli et al. 2005)

Acidifier inclusion in diet (kg/tonne)							
	0	2	3	5			
Initial weight (g)	16.7	16.7	16.7	16.7			
Final weight (g)	218 ^ª	258 [°]	246 ^b	252 ^{bc}			
FCR	1.34ª	1.23⁵	1.25⁵	1.22 ^b			
Mortality (%), day 10-85	33.0ª	20.8 ^b	18.4 ^b	11.0 ^c			

^{abc}within rows, means without common superscripts are significantly different (p<0.05)

to what was achieved with AGP inclusion. Details of this trial (de Wet 2005) are available in a previous issue of *Aqua Feeds: Formulation & Beyond* (Volume 2, Issue 3). In Turkey's rainbow trout farms, an acidifier consisting of formic and propionic acid and their salts on a sequential release medium (Biotronic[®]) has been tested and is currently used.

Ramli et al. (2005) tested potassium diformate (potassium salt of formic acid; Formi[®]) as a growth promoter in tilapia grow-out in Indonesia (Table 3). In this study, fish were fed over a period of 85 days 6 times a day diets containing different concentrations of potassium-diformate (0, 2, 3 and 5 kg/tonne feed). The fish were challenged orally starting day 10 of the culture period with Vibrio anguillarum at 10^5 CFU per day over a period of 20 days.

Over the entire feeding period from day 1 to 85, potassium diformate improved the weight gain and feed efficiency of tilapia. Survival rates of fish after the challenge with *V. anguillarum* on day 10 were also significantly higher compared to the negative control and the effect was dose dependent.

In a recent trial, Owen et al. (2006) tested the sodium salt of butyric acid as a feed additive in the omnivorous tropical catfish *Clarias gariepinus* at 2 kg/tonne in two diets: the main protein source in one diet was fishmeal and in the other diet was defatted soya. Slightly higher growth and concomitant reduction in FCR were observed in catfish fed fishmeal diet supplemented with sodium butyrate when compared with the control diet. Sodium butyrate supplementation did appear to increase the proportion of gram positive bacteria in the hindgut of *C*.

gariepinus, though this increase was not statistically significant.

Tung et al. (2006) reported that 5 kg/tonne sodium citrate boosted the growth of the Kuruma shrimp *Masurpenaeus japonicus*. I have done some work that suggests that a dosage of 2.5 kg/tonne calcium formate can enhance shrimp survival in brackishwater farms in Taiwan. These results are to be evaluated again in more than just one grow-out season.

Though there are only a limited number of published studies on the use of acidifiers in growth promotion and disease prevention in aquaculture, results from the studies indicate promising potential and compel aqua feed manufacturers to consider using acidifiers. The use of acidifiers can be an efficient tool to achieve sustainable, economical, and safe fish and shrimp production.

References

References are available from the author.



Dr Christian Lückstädt received his M.Sc. in Fisheries and Aquaculture from Humboldt University in Berlin, Germany and his Ph.D. in Aquaculture from the University of Hohenheim, Germany and SEAFDEC-Aquaculture Department, Iloilo, Philippines. His Ph.D. thesis was on feed intake and feed utilization in farm-raised milkfish. Since 2003, he is the Product Manager of Acidifiers for Biomin GmbH.