The importance of betaine and some attractive substances as fish feed additives

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SUMMARY - Betaine, L-alanine, L-glutamic acid, L-arginine, glycine and inosine are known as dietary feeding attractants for many fish species. However, these additives also have very important physiological functions within the animal body. For example, betaine given singly or mixed with other attractants has been found to have a positive effect on fish growth and survival rate. Betaine is also a very important substance for methyl donation and osmoregulation during the transfer of salmon and trout from freshwater to seawater. Similarly, L-alanine and L-glutamic acid can readily enter the citric acid cycle and be used for energy supply in fish and fish larvae. Moreover, L-arginine is an essential amino acid for fish, glycine is an important constituent of collagen and elastin, and inosine is a nucleoside and has vitamin activity in many fish species.

Key words: Attractants, amino acids, betaine, glutamic acid, arginine, glycine, inosine.

RESUME - *"Importance de la bétaïne et de certaines substances attractives en tant qu'additifs dans les aliments poisson". La bétaïne, L-alanine, acide L-glutamique, L-arginine, glycine et inosine, sont connus comme attractifs alimentaires pour plusieurs espèces de poissons. Cependant, ces additifs ont également des fonctions physiologiques très importantes dans l'organisme animal. Par exemple, la bétaïne distribuée seule ou mélangée avec d'autres attractifs s'est avérée avoir un effet positif sur la croissance et le taux de survie des poissons. La bétaïne est également une substance très importante pour l'apport de méthyle et la régulation osmotique pendant le transfert du saumon et de la truite depuis des eaux douces à des eaux salées. De la même façon, la L-alanine et l'acide L-glutamique peuvent d'emblée entrer dans le cycle de l'acide citrique et être utilisés comme source d'énergie chez les poissons et les larves. De plus, la L-arginine est un acide aminé essentiel pour les poissons, la glycine est un constituant important du collagène et de l'élastine, et l'inosine est un nucléoside et apporte une activité de vitamine chez beaucoup d'espèces de poissons.*

Mots-clés : *Attractifs, acides aminés, bétaïne, acide glutamique, arginine, glycine, inosine.*

Introduction

The use of dietary feeding attractants within compound aquafeeds has received considerable attention in recent years. The rationale behind their use has been to improve dietary food intake, and at the same time by promoting quicker food intake, minimizing the time the feed remains in water and thereby the leaching of water soluble nutrients, and at the same time providing additional nutrients for protein and energy metabolism. It follows therefore that if aquafeed are ingested with minimum wastage and feed efficiency therefore maximized, that feed wastage and water pollution will be minimized. This paper therefore presents a mini-review concerning the use of dietary feeding attractants within aquafeeds.

Betaine

Betaine (glycine betaine, trimethylglycine) is a highly water soluble and therefore diffusable compound which has the ability to stimulate the olfactory bulb of fish. It is found in high quantities within marine invertebrates (Meyers, 1987), micro-organisms and some plants, and as such constitutes an important part of the natural diet of marine carnivorous fish and crustacean species. However, under culture conditions farmed fish usually have little or no chance of obtaining sufficient quantities of betaine from aquafeeds composed of conventional feed ingredients unless the diet is specifically supplemented with exogenous betaine.

The main physiological or metabolic functions of betaine are related osmoregulation and methyl donation. For example, betaine has the ability to protect cells against dramatic changes in osmotic pressure in fish. It has been found that within marine invertebrates, an increase in salinity and temperature stimulates mitochondrial betaine synthesis; the net result being that betaine is accumulated at high levels and prevents abnormal water loss from the cells. Moreover, as a result of its osmolarity, betaine also helps to prevent enzyme inhibition. This is very important when transporting fish such as salmon from a freshwater to a seawater environment, since osmotic stress may result in a reduction in betaine synthesis within the fish liver mitochondria while the uptake of betaine to mitochondria increases (Virtanen and Soivio, 1985). It is perhaps not surprising therefore that research studies have shown that feeding salmon smolts with betaine supplemented diets for a 5-8 week prior to seawater transfer resulted in improved showed fish adaptation and increased growth performance.

As mentioned previously, since carnivorous marine fish species normally consume invertebrate animals containing high levels of betaine, then it is reasonable to suppose that the addition of supplemental betaine to formulated aquafeeds will help to mimic the smell and taste of the diet to that of their natural prey organisms. Beklevik and Polat (unpublished data) investigated the effects of using betaine and alanine as dietary feeding attractants (at 1.5% of the total diet) on the growth and carcass composition of rainbow trout (*O. mykiss*) and found that the addition of betaine resulted in increased feed consumption and growth, and that betaine was superior to alanine as a feeding attractant and growth promoter. Similar results were also reported by Can and Sener (1992) and Murai *et al.* (1983) with trout fingerlings. Other studies where betaine has been shown to act as a dietary feeding attractant include those of Goh and Tamura (1980) with red seabream (*Chrysophyrys. major),* Mackie and Mitchell (1982) with dover sole (*Solea solea),* and Mackie and Mitchell (1983) with the European eel (*Anguilla anguilla).*

Betaine also plays an important role within tissues as a methyl donor, which in turn may be used for the synthesis of methionine, carnitine, phosphatidyl choline and creatine; these substances playing a key role in protein and energy metabolism. Although betaine can be synthesized from choline in the mitochondria, synthesis is not usually sufficient to meet tissue demands within rapidly growing animals (Stekol *et al.*, 1953). Moreover, dietary betaine has been shown to spare the dietary requirement for choline and methionine in rainbow trout (Rumsey, 1991). It follows therefore that the metabolism of choline, betaine and methionine are closely related.

L-amino acids

Although the use of conventional and unconventional plant protein sources as dietary fishmeal replacers within aquafeeds is nutritionally feasible, the resultant feeds are generally much less palatable to the farmed species. It follows therefore that to overcome these difficulties (at least in terms of palatability) that these diets be supplemented with dietary feeding attractants and stimulants.

In this respect, like betaine, free amino acids are also high water soluble and easily diffused in water. In particular, L-alanine, L-glutamic acid, L-arginine and glycine have been reported to have dietary attractant properties; alanine, glutamic acid and glycine being non-essential amino acids, and L-arginine being an essential dietary amino acid for fish. For example, Polat (1996) studied the early amino acid metabolism of African catfish (*C. gariepinus*) and reported that alanine was a very important energy source in the fish, together with valine, serine, leucine and isoleucine. It follows therefore that the addition of dietary free amino acids as feeding stimulants for fish (and especially fish larvae) could also act as an important dietary energy source. Moreover, glycine is also an important constituent of collagen and elastic tissues. However, it is important to note that individually L-alanine, L-glutamic acid, L-arginine and glycine are not very effective in terms of attractive properties, but are very effective attractants when mixed together with glycine betaine or inosine. For example, Mackie and Mitchell (1985) summarized the results of various studies using dietary feeding attractants, and reported the positive effect of mixtures of dietary free amino acids as feeding stimulants in rainbow trout, *O. mykiss* (Adron and Mackie, 1978), European eel, *Anguilla anguilla* (Mackie and Mitchell, 1983), Japanese eel, *A. japonicus* (Takeda *et al.*, 1983), sea bass, *Dicentrarchus labrax* (Mackie, 1982), red seabream, *Chrysophyrys major* (glycine betaine plus L-amino acids; Goh and Tamura,

1980), Dover sole *Solea solea* (glycine betaine plus L-amino acids; Mackie and Mitchell, 1982), and puffer, *Fugu pardalis* (Glycine betaine plus L-amino acids; Ohsugi *et al.*, 1978).

Inosine

Inosine, a nucleoside, has also been found to be a very important attractant by itself or in combination with certain amino acids. For example, inosine and inosine-5-monophosphate have been reported as specific feeding stimulants for turbot fry, *Scohthalmus maximus* (Mackie and Adron, 1978), Dover sole, *Solea solea* (with betaine and glycine; Metailler *et al.*, 1983), and yellowtail, *Seriola quinqueradiata* (Takeda *et al.*, 1984). However, the main problems of using inosine and/or inosine-5-monophosphate as feeding attractants are their high cost. However, their use may be economically justified within larval feeds for marine fish larvae during the early weaning period since the total quantity of feed consumed is relatively low.

Conclusion

Betaine and certain free amino acids give the opportunity to fish nutritionists in preparing more palatable feed for fish during the different growth stages. As a mixture of attractive substances, they can usually beneficially affect both feed consumption and fish growth. It is generally accepted that the use of dietary feeding attractants and stimulants within aquafeeds will increase in the future as the industry is forced to use more economical alternative dietary protein sources as fishmeal replacers.

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