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Dr. SALIM ALI ROAD
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IMPORTANCE OF ANABOLIC AGENTS, BINDERS, ANTIOXIDANTS AND MOULD INHIBITORS IN FISH AND PRAWN FEEDS

D.C.V. EASTERSON

Research Centre of
Central Marine Fisheries Research Institute
Tuticorin-628 001

and

R. PAUL RAJ

Central Marine Fisheries Research Institute
Cochin-682 031

Binders, antioxidants, mould inhibitors and growth promoters are non-nutritional additives added in the feeds. The chemical compound choosen of the purpose should be neither toxic, nor antimetabolic, nor mutagenic, nor carcinogenic, nor teratogenic and nor bioaccumulative. Secondly must be effective at minimal concentration. Thirdly should neither retard growth, nor reduce palatability, appetite and assimilation of dietary nutrients. Fourthly should not react with the feed ingredients chemically in a way that would alter the nutritional quality of the feed adversely. Should be economical . And lastly should not reduce the quality of the meat produced by way of affecting taste, appearance, flavour and texture.

A. Binders

Binders are the substances as the name itself signifies used to bind the various feed ingredients into a compact mass and prevent the feed pellet from disintegrating

on being added to the water. This have two fold function. The popular binding substances used in aquatic feeds are as follows.

1.	Carboxymethyl cellulose (CMC)	- organic compound
2.	Polyvenyl alcohol (PVA)	"
3.	Sodium polyacrylate	"
4.	Propylene glycol alginate	- polysaccharide
5.	Algenic acid	"
6.	Na/K/Ca - alginate	"
7.	Agar	"
8.	Carrageenan (Kappa & iota)	"
9.	Natural gums - guar gum, gum arabic etc.	"
10.	Starches - corn, potatto, tapioca, rice etc.	"
11.	Wheat glutten	- protein
12.	Zein	"
13.	Gelatin	"
14.	Collagen	"
15.	Chitosan	- Amino-sugar

Items 2-3 are used 0.2 to 1.0% w/w concentration in the diet, while all others are used at 1-2 at times as high as 5% w/w levels. Algenic acid salts are the strongest among the list given. CMC is not stable in saline water and is also report to retard growth. The corn starch on boiling form hard wax-like gel contrary to that of potatto starch. Among lambda, iota and kappa varieties of carrageenan, kappa forms rigid gel, while iota forms flexible gel and lambda do not form gel. Proteinaceous binders also serve as protein sources, similarly many of the polysaccharides on digestion to varying extent serve as glucose sources. Heiner (1981) studied water stability of various binders and the finding are as given in the Table 1.

Table- 1: Evaluation of some binding agents for water stability (Heiner, 1981)

Binder	Concentration (% dry wt.)	Rank	Maximum stable time (Hrs.) Moist pellets	Dry pellets
None	-	1	6	6
None (hot water)	-	2	6	6
Corn starch	3	2	6	6
CMC	3	2	6	6
Guar gum	3	3	12	6
Collagen	3	4	12	12
Chitosan	3	3-5	18-24	9-12
Carrageenan	3	8	24	24
Agar	3	8	24	24
Sodium alginate	2	13	24	24

B. Mould Inhibitors

The formulated diet and the feed ingredients (carbohydrate and protein) on being highly nutritive under the conditions of storage and moisture come to have growth of moulds such as Penicillium spp., Aspergillus spp., Fusarium spp. etc., among fungus; Candida albicans and the like among yeasts; and bacteria like Salmonella agona. These by themselves are pathogenic, alter nutritional status, cause bad flavour and taste. Their exocrines also have been found to be toxic (mycotoxins). The most common and lethal among the mycotoxins are Aflatoxin (12 varieties), Tricothecenes (100 varieties), Ochratoxin and Zearalenone. To inhibit the growth of such above said organisms mould inhibitors need to be added to the feeds. Following is a select list of mould

inhibitors used in aquarium feeds - Table 2.

Table-2. Recommended mould inhibitors

Chemical	Recommended concentration in diet
Sorbic acid	No limit
Ca - sorbate	"
K - sorbate	"
Na - sorbate	"
Propionic acid	"
Na - propionate	"
Ca - propionate	"
Menadione	"
Sodium benzoate	less than 0.1%
Propyl-p-hydroxybenzoate	"
Methyl-p-hydroxybenzoate	"

In storing cattle feed copper sulphate at the rate of 120 mg copper per kg of feed (maximum of 240 mg Cu/kg diet) is being used. Among the two gentian violet is better and could be effective for a period upto 12 weeks (Jensen, 1977). Copper could be bioaccumulated and the toxic effects of copper could be reduced by the application of high supplementation (4 g/kg of diet) of sulphur amino acid methionine (Jensen, 1977).

C. Antioxidants

Lipids especially unsaturated fatty acids on exposure to air, light, heat and moisture go bad and come to acquire bad taste and odour. This process is known as rancidity. Polyunsaturated fatty acids are highly prove for oxidation and whereby come to loose its essentiality. On oxidation

they become saturated and yield organic acid by the following reaction.

unsaturated fatty acid + O_2 Peroxide + H_2O

Unsaturated fatty acid + Peracid Aldehyde

Antioxidation

Ordinary organic acid (Eg: Butyric acid) Cu, Fe, EDTA, cyanide and chlorophyll enhance the said process of oxidation and are known as prooxidants. Antioxidants on the otherside absorb oxygen available for oxidation and whereby prevent oxidation. Their role can be illustrated by way of the below given equations, where A - oxidant, AO_2 - peroxide and B - antioxidant.

AO_2 + B AO+BO A + B + O_2

The antioxidants are capable of functioning at minimum levels. Thus one molecule of hydroquinone can protect 40,000 molecules of acrolein from oxidation. Apart from the antioxidants given in Table 3, gallic acid and phosphoric acid are some of the important antioxidants.

The quantity of unsaturated fatty acids in a lipid is measured in terms of iodine number, saponification value, Acetyl number and Reichert-Meissel number.

Table 3 : Antioxidants used in diets

Antioxidant	Recommended concentration in the diet in terms of lipid content
Citric acid	No limit
Ascorbic acid	"
Lecithin	"
S and -tocopherol	"
Butylated hydroxyanisole (BHA)	Less than 0.2%
Butylated hydroxytoluene (BHT)	"
Nordihydroquaiaretic acid	"
Propylgallate	"
Thiodipropionic acid	"
Resin guaiac	"
Ethoxyquin	Less than 0.015%
Santoquin	"

D. Anabolic Agents

The use of growth promoting substances in the rearing of cattle, sheep, pig and chicken is a common practice. The growth promoting agents ie anabolic substances can be grouped under the following categories: 1. Hormones 2. Antibiotics and 3. Organics. Anabolic substances should not be confused with essential nutrients. When certain essential nutrients on being deficient, supplementation of them would result in higher growth rate. These essentials are not called as anabolic agents. In this case growth retardation is due to deficiency of such essential nutrient. Anabolic agents are, chemical substances which on trace quantities capable of enhancing protein synthesis well above the normal.

The use of anabolic agents is beneficial (1) by way of fast growth reduction in growth out period is made possible. Thus either a hatchery or in a grow out system with the available facility production turn over could be increased. (2) Production of oversized ('giant') aquacultural product is possible (3) By way of increased utilisation higher food conversion efficiency is obtained, whereby better utilization of offered diet is possible.

I. Hormones

The anabolic hormones can be grouped under 4 categories:

i. Growth hormones (natural source - pituitary)
ii. Anabolic steroid hormones (Nat. sour. - Gonad)
iii. Thyroid hormones
iv. Insulins

i. Growth Hormones: Donaldson et al. (1979) have made a very detailed review on the subject. The usual methods of administration of growth hormone (GH) is by way of intramuscular injection (im), intraperitonial (ip) injection and by implanting the pellet in the muscle whereby permitting slow release of the hormone over a period. Administration of GH through the medium was not successful while, bathing the gills with GH marginally enhanced growth. With the higher dosage (200 µg/g) and increasing the frequency of injection the growth increase was elevated but the increase was not proportional. Thus at about 3.5 µg/g in one per week seems to give optimal growth in Oncorhynchus kisutch (Cocho salmon).

Among the use of human, ovine and bovine GH, bovine gave better result and the human least. Slight elevation of temperature along with improved the growth.

GH increased the growth by way of increasing the appetite and conversion efficiency. Mobilisation of fat and oxiation of lipids was increased. Fat come to be used as energy source and proteins conserved for growth. An increase in protein/g the body weight too was observed. Further synthesis and release of insulin too was elevated.

ii. Steroid hormones:

The following steroids have been tested (Donaldson et al., 1979) in fishes.

(a) Androgens (Nat. source - Testis)

Testosterone
17 -Methyltestosterone
11 - Ketotestosterone
4 - chloro-testosterone acetate
Ethylestrenol
Methenolone acetate
Dimethazine
Oxymetholone
Testosterone propionate
Methylandrostenediol
Stanozolol
1-Dehydrotytosterone
17 -Ethyenyltestosterone

(b) Estrogens (Nat. Source - ovary)

Estradiol
Estrone
Estradiol
Diethylstilbestol
Estradiol benzoate
Estradiol monopalmitate

(c) Progestogens (Nat. source corpus lutium of ovary)

Progestrone
Pregnanediol
Melengestrol acetate

(d) Corticosteroids (Nat. source - Interrenal gland in fishes, Adranal cortex in mammals)

Cortisol

Deoxycorticosterone acetate

a. Andogens: The usual route of administration is by way of moist diet. Im and Ip injections need handling of fishes and in the process of handling the fish by all possibility could get injured. Mixing of hormone in the watery medium has proved unsuccessful and retarded growth. The successful results have been obtained in the order of lesser value are - 17 methyltestosterone; ethylestrenol; oxymetholone, stanzolol and 1 - dihydrotestosterone acetate. Androgens supressed the growth in females and also resulted in impotent males.

b. Estrogens: Diethylstilbestol only shows slight improvent in growth in fishes even here it is only marginal. Further the results are not consistant with different species.

c & d. Progestrons and corticosteroids: Here too the results are not promising in fishes. It is of interest that many of the above give hormones have enhanced the growth in mammals and in aves but have proved unpromising in fishes. The study in crustaceans is very scarce.

iii. Thyroid hormones: 3,5,3' - triiodothyroxine (T_3) and 3,5,3',5' - tetraiodothyroxin (T_4) are the most common mammalin a hormone used. The addition of the hormone to the watery medium has resulted in growth enhancement in more cases than when either injected or give through diet. Often growth

abnormalities and physiological syndromes have been observed to result on the use of TH. In the water the following concentrations were beneficial $1:1 \times 10^7$; and $1:5 \times 10^5$. The administration of thyroxin resulted in the elevation of SDA and food uptake.

iv. Insulin: Evidences show that insulin play a specific role in the incorporation of plasma amino acids into muscle protein. But the administration insulin to fishes has produced contradicting results.

II. Organics

In the higher vertebrates arsenicals and tranquilisers have been found to increase growth but not much work have been conducted in aquatic organisms. Further arsenic is a cumulatory to poison and so caution need to be observed in the actual application in aquaculture. The common chemical used are as follows:

i. Arsenical

3-nitro-4-hydroxyphenylarsonic acid
p-amino-phenylarsonic acid
Na-arsanilate (Na salt of the second above given acid)

ii. Tranquillisers

Chlor promazine
Reserpine
Natural alkaloid Ranwolfa

How far tranquillisers are anabolic is not clear. It is supposed that by way of their action of reducing excitability and environmental stress the organism is able to conserve energy from metabolism and use them for body building (McDonald et al., 1977).

III. Antibiotics

The antibiotics permitted to be used in diet (0.001 to 0.005%) in USA are as follows: Bacitracin and derivatives, Chlortetracyclin, Lincomycin, Erythromycin thiocyanate, Oxytetracycline, Oleandomycin, Procaine penicillin, and Tylosin. In the rearing of birds, swine, cattle, sheep and rabbits the results have been encouraging while it is negative in case of hamster, guinea pig, cockroack, bugs, aphids and protozoans. The known study is aquatic organisms are as given below.

Animal	Dose	Result
Lebistes reticulatus (fish, guppy)	Chlor. Strep., 1 mg/ml water	Death
Salmo gairdneri (fish, rainbow trout)	Chlortet 1:10,000-1:6,000	Inhibition of metamorphosis and death.
Venus mercenaria (Bivalve)	Chloram 0.1-0.2 ppm	Increased larval length
Astralorbis glabratus	Pleocidin, Nystatin, Patulin	growth retardation

In case of animals in which growth promotion has been effected the reasons given are as follows.

1. Change of intestinal microflora to increase the number (of vitamin synthesisers) useful symbiots, ie preferential anihilation of microflora.

2. Inhibition of gut micro-organisms which utilise or destroy nutrients (better conservation of nutrients)

3. Inhibition of microorganisms that produce toxins (reduction of pathogens)

4. Prevention of thickening of gut walls whereby facilitating increased absorption of nutrients, especially lysine, lipids and calcium (Mc Donald, et al. 1977).

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