Dunaliella salina in aquatic organisms

Fish
Carotenoids contribute to the health and reproduction of fish, as well as their pigmentation.

Pigmentation
In wild and cultured masu salmon (*Onchorhynchus macrostomus),* and in wild manchurian trout (*Brachymystax lenok)* the major carotenoids are zeaxanthin, betacarotene and isocryptoxanthin; lutein, canthaxanthin and astaxanthin are among the minor carotenoids (beta-carotene, zeaxanthin, and lutein are components of Nutra-Red™ and Algro™) (Baek and Ha, 1998).

In cultured flounder, *Paralichithys olivaceus*, colour was improved by feeding diets supplemented with beta-carotene or with lutein esters. The main carotenoids in the flesh of the flounder were lutein and zeaxanthin (Ha et al. 1993).

Marine fish larvae
In general, most marine fish larvae are cultured in tanks using 'green water' technique. Different algae such as *Nannochloropsis oculata, Tetraselmis* sp., *Isochrysis* sp. and others are added to the rearing tanks at different densities. It is unclear what is the role of the algae and what is the mechanism by which it contributes to the larvae (in most cases, the algae cell is too hard for the larvae to digest and often it will evacuate the larvae gut intact. However, it is clear that these algae are essential for larvae culture. Algae culture in hatchery is considered to be labour intensive as well as require light and in cold climate energy to heat the rearing containers (tanks, beg etc.). Recent results from preliminary studies demonstrated that barramundi (*Lates calcarifer*) larvae preforme similarly (both grow and survive) when live algae (*Nannochloropsis oculata),* green algae paste (*Chlorella or Nannochloropsis*) or *NutraPlus* was added to the rearing tanks in standard 'green water' technique.

Fish reproduction and health
The carotenoids beta-carotene, lutein, canthaxanthin and astaxanthin are found in the eggs, ovaries, testicles, and milt (sperm) of fish including salmon, trout and sole. Pigmented eggs have a higher rate of fertilisation than non-pigmented eggs. It is thought that the carotenoids may have a positive effect on sperm motility, may have a role in the respiration of the eggs, and may protect the eggs against UV light damage (Hamdorf, 1960; Hartmann, et al. 1947; Jitariu, et al. 1975)

When Japanese parrotfish (*Oplegnathus fasciatus)* and spotted parrotfish (*Oplegnathus punctatus)* were fed with rotifers which had been supplemented with beta-carotene, survival rates of larvae were higher, and production of lymphocytes in response to challenge was greater, indicating greater resistance to infections (Tachibana, et al. 1997).

In yellowtail, *(Seriola quinqueradiata)* the carotenoid content of eggs was greatly affected by the carotenoid in the broodstock diets. Egg quality was highest in eggs with a strong yellow colour and a high content of lutein and zeaxanthin (Verakunpiriya, et al. 1996).

Ornamental fish
Lutein: The colour of the native Korean bitterling (*Rhodeus uyekii*), is enhanced by addition of carotenoids to the diets; the best enhancement was with lutein supplementation (lutein is a component of *NutraPlus* and *Algro*™) (Kim et al. 1999).

Carotenoid accumulation from diet is important in sexual colouration and subsequent breeding of guppies (*Poecilia reticulata)*; the females prefer males with brighter orange carotenoid-containing spots (Grether, et al. 1999).

The skin of sailfin mollies, *Poecilia latipinna* contains the carotenoid, beta-carotene. Unlike some other poecillid fishes, sailfin mollies rely primarily on carotenoids for their pigmentation (Blanchard, et al. 1991)

Marine filter feeders
Corals, both soft and hard can grow and survive when fed *Dunaliella* paste (as Nutra-Red™). Moreover, public aquariums and coral farms / holding facilities trialed the red paste during the past two years with great success. The Aquarium of Western Australia (AQWA) is currently using *NurtaPlus* as the sole feed for both soft and hard corals as well as for sponges.

Sea urchins
Beta-carotene is a common major carotenoid found in sea-urchins, in the gonads, tests and Spines (Tsushima, and Matsuno, 1990). When *Algro*™ was added to prepared feeds, it improved the colour (both red and yellow colour value) of sea urchines. Addition of 250mg carotenoids as *Algro*™/kg feed (that is, 12.5g *Algro*™ per kg feed) was optimum, and yielded a roe product of high quality, with respect to colour, taste and size (Robinson and Castell, 2000).

Brine shrimp and rotifers
Dunaliella salina is a major component of the diet of wild harvested brine shrimp,*Artemia spp.* Addition of *Algro*™ or *NutraPlus* to the feeding regime of cultured brine shrimp enhances their natural colour and their nutritional properties (Tachibana, et al. 1997, Kolkovski, pers. comm., 2004). *Artemia* can be grown on *NutraPlus* as the sole feed or the paste can be mix with a commonly used feeds such as rice bran. The cartenoids accumulate in the *Artemia*, when feeding or enriching it for at least 24 to 48 hours prior to serving it to fish or other organisms. The result is bright red *Artemia*. The carotenoids are then pass to the target organism. Brine shrimp as well as other crustaceans (prawns *Penaeus sp*.) are capable converting carotenoids such as beta-carotene, alpha carotene and lutein to the red pigment astaxanthin (Katayama, et al. 1973). In the wild, crustacea ingest a mixture of carotenoids from algae and zooplankton. *NutraPlus* and *Algro*™ are natural replacements for that wild food source of carotenoids.

Abalone and molluscs
When added to prepared diets for farmed abalone, *Algro*™ may act as a feed attractant, and improved the colour of the shell (personal communication from Australian customer). Currently, *Algro*™ is added to commercial abalone feeds in Australia.

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