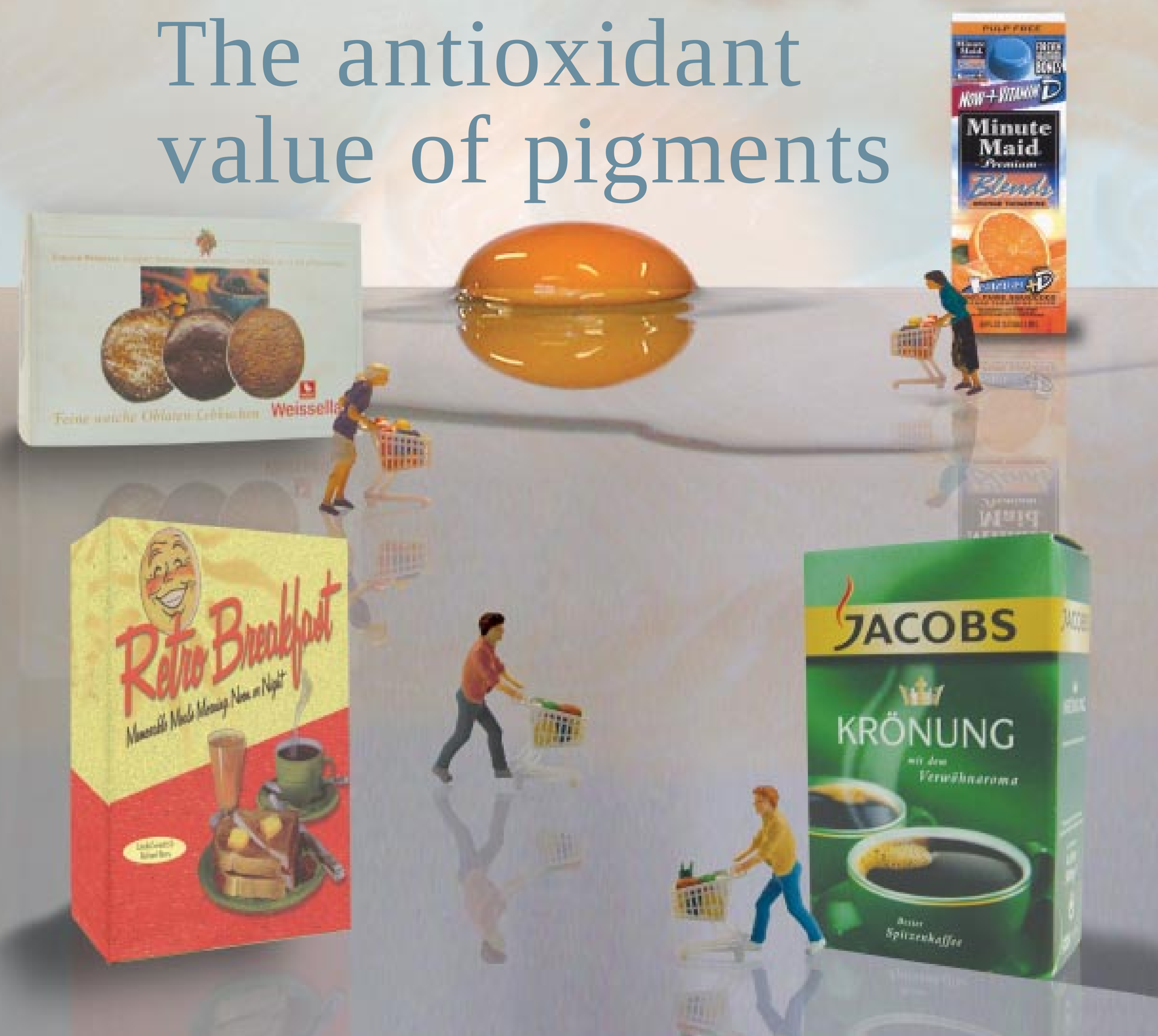
Carotenoids for all! The antioxidant value of pigments



APPEARANCE IS ONE OF THE MOST IMPORTANT ATTRIBUTES AFFECTING CONSUMER CHOICE, RANKING ALONGSIDE FRESHNESS. ADDING NATURALLY-OCCURRING CAROTENOID PIGMENTS TO FEED THUS HAS COSMETIC EFFECT, BUT, ARGUE REMI BAKER AND CHRISTOPH GÜNTHER, THEY ALSO BENEFIT ANIMALS AND HAVE CARRY OVER EFFECTS TO HUMAN HEALTH.

Carotenoids are amongst the most widely distributed pigments in biological systems. In nature, animals utilise colour to assess the attractiveness of a mate for breeding- good coloration in many animals signifies good foraging ability (Møller *et al.*, 2000).

Enhanced coloration through carotenoid deposition also signifies better health, as carotenoids have antioxidant functions, and therefore less colourful individuals are likely to be those with a large detoxification burden. It has even been shown that animals infested with parasites are less able to deposit carotenoids. It may be concluded therefore, that, in animals at least, colour often equals health and mate suitability.

COLOUR THERAPY

Humans are not much different in their use of colour. Food packaging is often coloured (after all, that is frequently what attracts us to buy a certain brand). The food left on supermarket shelves tends to be the discoloured produce (from vegetables to meat).

Colorants are added to many foods to make them appear healthier and more appealing. What is important is that the colours that are applied are safe; and that colours should not be used to mask unhealthy products. Beta-carotene is routinely used in soft-drinks, cheeses and butter or margarines, is well regarded as being safe and indeed is also attributed with positive health effects due to a pro-vitamin A role. In wild salmon, the carotenoid pigment ‘astaxanthin’ provides most of the colour that we have come to expect. Canthaxanthin, a carotenoid pigment abundant in plants, microbes, crustacea and birds, can also impart colour to salmon and trout flesh. In these applications, the carotenoid is referred to as a pigment rather than a colorant, the difference being that the organism selectively retains the pigment because it is biologically programmed to do so.

WILD IMITATION

Astaxanthin and canthaxanthin are added to feeds for farmed fish in order to closely reflect the characteristics of the wild product. The other argument is that, since these carotenoids are present in the natural diet of wild fish, they should also be provided in the diets of farmed fish. After all, the ability of trout and salmon to store carotenoids in their flesh indicates that these pigments play a physiological role- this has been proven to be the case. Female wild salmon mobilise carotenoid pigments from the flesh and deposit them in their eggs, where the benefits are manifold. Firstly, salmonid eggs are spawned in shallow rivers where light-induced damage may harm the delicate tissues. In this case, the carotenoids act to prevent ultraviolet instigated oxidation. Carotenoid transfer to the fry also confers benefits in the pre-feeding period. In the reproduction and larval development of salmonids, astaxanthin is essential to the survival of larvae throughout the fry stage, with levels of at least 0.11µg astaxanthin per egg necessary to prevent the appearance of M74 syndrome in Baltic salmon (Pettersson & Lignell, 1999). Furthermore, canthaxanthin has been showed to enhance the fertilisation rate of trout.

TECHNICAL PERFORMANCE

Astaxanthin and canthaxanthin may also provide stimulatory effects on growth and performance in farmed fish and shrimp.

Torrissen (1984) stated that diets supplemented with astaxanthin and canthaxanthin promoted growth during the early start-feeding phase of Atlantic salmon reared in fresh-water. In Atlantic salmon parr, Christiansen and co-workers (1994,1995) were able to show that fish fed feed containing 60 mg astaxanthin per kg grew significantly better than those on a diet free of the pigment. Essentiality was claimed for astaxanthin for growth and survival in these fish. Torrissen and Christiansen (1995) subsequently recommended a minimum supplementation of 10 mg astaxanthin or canthaxanthin per kg diet for feeds for fish and crayfish.

Authors have also reported slight growth benefits in the larvae and juveniles of non-salmonids, for example carp and tilapia. Segner *et al.* (1989) demonstrated that astaxanthin supplementation was beneficial to liver structure and also hepatic glycogen storage in tilapia and *Colisa labiosa*, the thick-lipped gourami. A precise mechanism for these effects is not clear.

Due to the demonstrated essentiality; and the similarity in action of astaxanthin and canthaxanthin to vitamins A and E; these carotenoids have been considered as vitamins (Torrissen & Christiansen, 1995). In a crustacean model, growth benefits of feeding astaxanthin to the prawn *Penaeus japonicus* have been recorded by workers including Chien and Jeng (1992) and Nègre-Sadargues *et al.* (1993). Nègre-Sadargues *et al.* (1993) showed an increase in growth rate of at least 25% through the addition of astaxanthin to a non-pigmented basal feed.

ANTIOXIDANTS IN FOOD…

The role of carotenoids in human and animal health is now widely accepted. Through their antioxidant role, carotenoids may modulate many free radical mediated diseases and conditions. Under certain conditions, astaxanthin has been seen to be a more powerful antioxidant than vitamin E. *In vivo* studies on mammals and fish have demonstrated a clear antioxidant effect (Nishigaki *et al.*, 1994; Nakano *et al.*, 1995; Christiansen *et al.*, 1995).

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This article has been adapted from the original: Baker and Günther (2004). “The role of carotenoids in consumer choice and the likely benefits from their inclusion into products for human consumption.” *Trends in Food Science & Technology* 15, 484-488; It is reproduced in this edited form by kind permission of the authors.

...AND IN FEED

Overall, the growing body of literature clearly points to that fact that astaxanthin (circulating bound to blood lipoproteins) is able to suppress free radical initiation or propagation, and thus improve the animal's general health. Recent evidence indicates that astaxanthin also functions as an antioxidant in membranes isolated from salmon previously fed astaxanthin (Bell *et al*., 2000). It is likely that the same is true of canthaxanthin given the evidence of its antioxidative effects in model systems (Palozza & Krinsky, 1992; Packer, 1993; Woodall *et al*., 1997). Recently, canthaxanthin ingestion has been linked to the antioxidant status of chicks (Surai *et al*., 2003), where increased feed inclusion was able to improve the chick's antioxidant defences either directly, or by a vitamin E sparing action.

COLOUR AWARENESS- EGG YOLK

Canthaxanthin is commonly used in the poultry sector. In diets for laying hens, it can be incorporated into the rations from where the pigment is digested and transferred to the yolk, enabling egg producers to satisfy consumer expectations for yolk colour. Upon finding two yolks of different colours within the same box, consumers are likely to suspect that the paler yolk is less fresh or comes from a less healthy hen. So, uniformity of colour is important. Herein lies the next problem. At the lower end of the colour spectrum, it is more difficult to assure evenness of colour. This is because at lower feed carotenoid contents, deposition is linearly related to dose. Many other factors, including genetic background, feed intake and health status, can also affect deposition efficiency. At higher carotenoid doses, however, colour saturation is approached, meaning that small differences in carotenoid content of feed (or finally of yolk) manifest as almost insignificant changes in perceived colour. This holds true for egg and for fish flesh. Thus, at higher carotenoid doses, the safety margin is much larger, reducing the probability of having off-spec products. However, not all markets appreciate highly coloured egg yolks or salmon fillets. In some countries, golden/orange yolks are considered normal because in former times, birds were fed with grass meal or yellow-corn, and the natural xanthophyll intake was therefore fairly high. In countries where these raw materials were not utilised, paler yolks are considered the norm. The ability to include nature-identical carotenoids into hen rations enables producers to offer reliable flexibility of choice.

HUMAN HEALTH GOALS

There is another potential advantage to the inclusion of carotenoids in feed for food-producing animals: our health. As with animals, we have a situation where colour equates to general health. The direct influences of carotenoids (beta-catotene, lycopene and lutein) on human health are much promoted. Canthaxanthin and astaxanthin have not received quite the same exposure but there is considerable evidence to suggest that they would exert positive influences on the human immune system and both are viewed as anti-carcinogens. However, as with any compound (even vitamins), overdoses are to be strictly avoided. The retinopathy arising in a small number of individuals caused by direct consumption of very high doses of canthaxanthin over a prolonged period has led to imposition of maximum inclusion limits for canthaxanthin in animal feed.

As a potential source of vitamin A (Schiedt *et al*. 1985), astaxanthin has been implicated in immune function. Vitamin A and retinoid metabolites are known to influence the immune response by any one of a number of proposed mechanisms (Bendich & Shapiro, 1986).

ANTIOXIDANT CONSUMPTION

It has been observed that canthaxanthin has antioxidant capabilities *in vitro* (Packer, 1993). Furthermore, this effect

Factors affecting yolk pigmentation

Dietary factors
- -> Dose and type of product applied;
- -> Natural xanthophyll content of the feed (consider variability of feed stuffs);
- -> Fat content and quality (oxidation);
- -> Energy level;
- -> Presence of emulsifiers;
- -> Antioxidants (vitamin E, ethoxyquin, etc.);
- -> Competing substances (e.g. when vitamin A >25000 IU/kg);
- -> Calcium level;
- -> Mycotoxins (reduce plasma carotenoids);
- -> Gossypol in cottonseed meal causing browning;
- -> Piperazine as a treatment for worms (induces brown/green colour);
- -> Nicarbazine against coccidia (induces brown/green colour).

Bird physiology
- -> Feed intake;
- -> Age of bird (stage in laying cycle);
- -> Breed (genetics);
- -> Temperature and humidity (climate/weather).

Bird health
- -> Adequate bile quantities in the digestive tract;
- -> Condition of surface area of digestive tract;
- -> Enzymic digestion efficiency;
- -> Diseases;
- -> Parasites.

Feed production
- -> Mixing standards;
- -> Abrasion/ aggressiveness of carrier;
- -> Pelleting and extrusion conditions;
- -> Storage duration and conditions.

Carotenoid product characteristics
- -> Physical form, crystal size;
- -> Storage and premix stability;
- -> Vitamin A activity.

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has been recorded in membrane model systems (Palozza & Krinsky, 1992), in liposomes (Woodall *et al*., 1997), and in isolated liver-cells from rats previously fed canthaxanthin and then subjected to aflatoxin induced damage (Gradelet *et al*., 1998). Epidemiological studies suggest that consumption of foods containing beta-carotene, lycopene and other carotenoids, including astaxanthin and canthoxanthin may be effective against certain types of cancer (Peto *et al*., 1981). Astaxanthin's and canthaxanthin's cancer suppressive role (Gradelet *et al*., 1998) may also be explained in relation to vitamin A bio-conversion, where vitamin A is important in cell differentiation control, but a further explanation of the tumour-preventive effect is due to the molecules' antioxidant properties.

ENHANCE FOOD QUALITY

The practice of enhancing foods is not restricted to these applications. Most foods on the supermarket shelves are in some way standardised with respect to presentation, shape, weight, flavour, and of course, colour. We have all come to associate certain physical characteristics to important quality attributes. Desirable attributes often equate to brand or product loyalty. Manufacturers of processed foods use this knowledge in the design of the food and the packaging. In salmon, one of the distinguishing features is the orange-pink flesh colour. This is often the first sign of quality, particularly today when a majority is sold as fillets, steaks or smoked sides. It is most likely that the ability of salmon products to successfully enter non-traditional markets is affected by its year round supply, flavour, affordability and ease of preparation; but very importantly, the luxurious appearance of salmon. Trying to market a grey product would have almost certainly been less successful. So what have we achieved?

KNOCK-ON HEALTH EFFECTS

Salmon is a good source of omega-3 fatty acids (n-3 fatty acids), known to exert a beneficial effect on human health. A good intake of omega-3 fatty acids has been proven to reduce the risk of coronary heart disease and inflammatory conditions (e.g. rheumatoid arthritis). Plant oils and animal fats contain little or no omega-3 fatty acids (with a couple of notable exceptions: rapeseed and linseed), so the primary source of the omega-3 polyunsaturated lipids in the diet is marine fish, such as salmon. Therefore, correct marketing of salmon and salmon products promotes the consumption of 'healthy fats'. In other words, carotenoids render salmon more attractive to the consumer, and this in turn yields human health benefits. Indeed, providing animals with carotenoids is a means of achieving what nature really has intended, naturally. <-