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Why should I use CAPSANTAL on laying hen or broiler diets?

Reliance on natural feed ingredients (corn, gluten meal, alfalfa meal...) to provide a sufficient amount of oxycarotenoids for adequate pigmentation of broilers and egg yolks is not always possible. Major problems in the use of natural ingredients for pigmentation include:

- a) Variation in oxycarotenoid content,
- b) Bioavailability of the oxycarotenoids,
- c) Instability of the oxycarotenoids in the feed ingredient and finished feed,
- d) Poor correlation between chemical assay of oxycarotenoid content and active biological performance.

The instability of the oxycarotenoids in feed ingredients is relative and depends on the source, storage conditions, length of storage, addition of fat and/or an antioxidant, and probably other less well defined factors.

During the years, as stability data have accumulated and improvements have been made in handling and storing the products, better retention of either carotenes or xanthophylls or both has been achieved. The carotenoid content of corn varies with the time of the year it is received, storage time and conditions, the year of the crop... Also, the rate of loss of xanthophylls and carotenes in dehydrated alfalfa meal is accelerated by heat and light.

That's why the addition of CAPSANTAL natural pigmenters to the feed provides a safer supply of xanthophylls to the diet and assures that the final product (either eggs -egg yolk- or broiler -skin and shank-) really reaches the desired pigmentation.

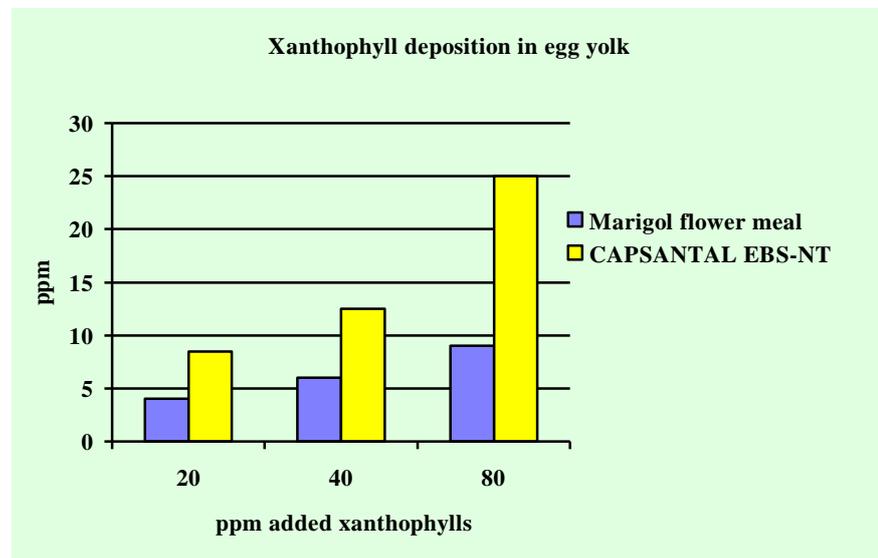
The natural pigments most widely used are lutein, obtained from marigold flowers, and capsanthin, obtained from Capsicum peppers. These



pigments present several characteristics that may affect their absorbability and deposition rate on poultry production.

Capsantal products are not just marigold flower or red pepper meal; those are just the raw materials that undergo a complex process to reach a final product with a constant and certified xanthophyll content and pigmenting ability.

Moreover, ITPSA, as an antioxidant producer, is also aware of the possible instability of xanthophylls from natural sources. As a result the CAPSANTAL products are extremely stable (not



just in the original package, but also after having been mixed in the diet) and protected with the action of the ethoxyquin as antioxidant.

Why the CAPSANTAL products are better absorbed by the animal than marigold flower or pepper meal?

Feed carotenoids are absorbed as free carotenoids. This means that hydroxycarotenoids have to be saponified first before they are absorbed. A

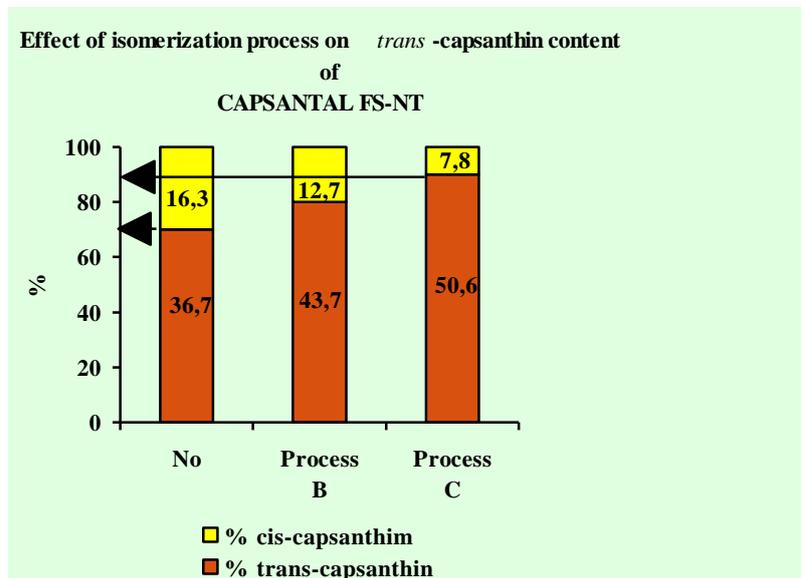


saponification process *in vitro* improves the digestibility of the originally esterified tagetes xanthophylls, compared with the *in vivo* saponification in the gut of the broiler by about 40 to 60% (in young chickens). Lutein in tagetes petals occurs mainly as diesters of palmitic and myristic acids.

Free lutein seems to be absorbed preferentially from the intestinal contents, and is transported in the serum to depot sites. With dietary lutein diester (untreated marigold flower meal), the tissue concentration obtained is generally only half that obtained with saponified dietary lutein (CAPSANTAL EBS NT). And the same can be stated for red pigments (CAPSANTAL FS NT).

ITPSA has been aware of these facts, and conducted its own research on these issues in the beginning of the 80's. As a result of this research on the

saponification process itself and on the increased deposition of saponified pigments of the diet, the natural pigments offered by our company (CAPSANTAL EBS NT and CAPSANTAL FS NT) are subjected to a saponification process that reaches almost 100% of saponification, increasing consequently their absorption.

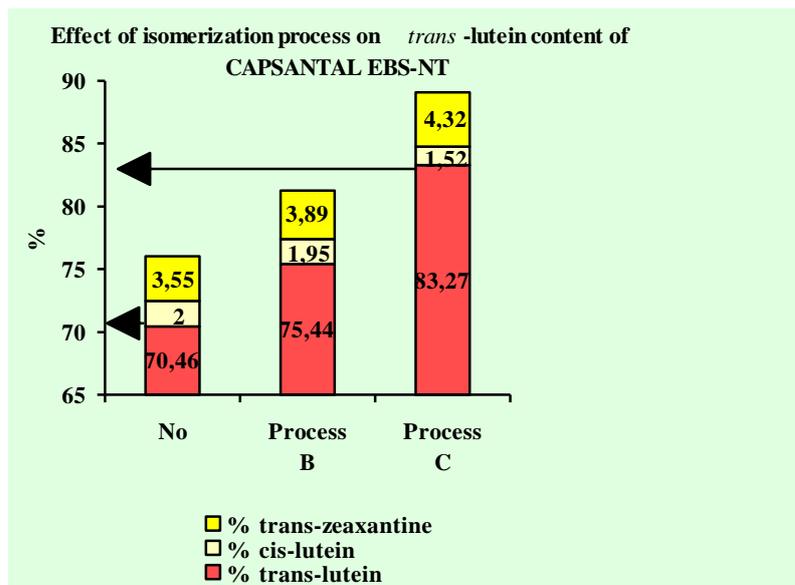




Is the saponification the only reason why the CAPSANTAL products are better absorbed?

Saponification is not the only reason. Feed carotenoids occur in natural compounds in both the *trans* and *cis* configurations. *Cis* carotenoids are formed from *trans* carotenoids mainly under the influence of excessive heat, light, and oxygen or as a result of manufacturing processes.

The ratio of *trans:cis* carotenoids in natural compounds may vary between 60 to 90% for *trans* carotenoids and 30 to 10% for *cis* carotenoids. The same principles apply to synthesized carotenoids (such as astaxanthin, canthaxanthin or zeaxanthin) which are also formed in both the *trans* and *cis* configurations. *Trans* carotenoids are considered to be more effective pigments: it might be concluded that individual feed carotenoids with a higher percentage of *trans* forms (our CAPSANTAL products) would have a better pigmenting efficiency than those with a lower percentage.



ITPSA started its own research on this topic several years ago. Currently we have a reliable isomerization process that increases the percentage of *trans* configurations of lutein and capsanthin. Consequently, our products offer a



higher proportion of *trans* lutein (CAPSANTAL EBS NT) or *trans* capsanthin (CAPSANTAL FS NT) than are present in the raw materials from which they derive.

What kind of control does ITPSA perform on CAPSANTAL products?

Pigments have been classically analyzed by spectrophotometry. This technique gives us a good idea of the pigment content. Some improvements have been made on the method, in order to have not just an absorption value for a fixed wavelength, but also the ratio between several maximal absorption wavelengths. These ratios give us more information on the composition of pigments. But the improvements on analytical techniques have not stopped there.

The analysis of carotenoids by HPLC has already had an impact on the control of pigmentation by the poultry industry. ITPSA has been working on this subject for a long time, and the analytical controls on pigment quality by HPLC are performed in our quality control laboratory.

The incorporation by ITPSA, several years ago, of the HPLC as a routine analytical technique to assure the quality control of pigments, has given us confidence in our products

Presently we have a faster availability of the analytical results and the control over the quality of our products is even greater than in the past. Using HPLC analysis to control our products, and consequently the diets, results in a more consistent pigmentation and benefit in the field

Generalities on the use of pigments.

The pigmentation of egg yolks and the pigmentation for broilers are often discussed together as one, overall pigmentation for poultry products. There are a number of similarities but there are also fundamental differences that should



be noted. The similarities are species, a nearly identical utilization of individual pigments, absorption, feed sources, biology, management, and colours.

The differences include the mechanisms of metabolic deposition, purpose of deposition, number of deposition sites, age of the bird, target tissue, market selection and economics, i.e., broiler industry needs relative to egg industry needs. Broilers are selected according to their appearance, as modified by pigments. Eggs are not selected according to yolk colour: that is evaluated later.

Colour is a more dominant factor for egg yolk appearance than for the appearance of broiler skin. In addition, the deposition of egg yolk pigment is relatively simple: the pigments that are absorbed are deposited almost directly and with little modification into the developing ova. There seem to be few other deposition sites in an actively laying hen; and once deposited in the yolk, the pigments are extremely stable and undergo almost no deterioration.

The deposition of pigment in broilers, though, is much more complex due to variations in the possible deposition sites and the unstable nature of the pigments in the epidermal locations. Although nearly the same pigments are utilized by both laying hens and by broilers, the relative efficiencies may vary dramatically. Thus, some pigments are more effective for egg yolk pigmentation than for broiler pigmentation. Egg yolk pigments are relatively easy to extract and quantify; and because the egg yolk is nearly homogeneous, colour is also determined easily. However, broiler skin pigments are not deposited uniformly throughout the skin, are not as easily extracted, and are not as easily quantified because of chemical reactions in the skin. The colour of broiler skin is also difficult to measure due to the lack of skin uniformity and of sample areas relative to the total surface area. In addition, broilers also deposit a portion of the absorbed dietary pigments into the fat, not just the skin. The ration of dietary pigments deposited in the skin to those deposited in the fat and the effect on subsequent appearance has not been explained.

It must be recognized that depth of colour as perceived by the eye in both egg yolk and broiler shanks or skin tissues is not directly proportional to the pigment deposited as determined by colourimetry at all concentration ranges. The deposition of pigment in broiler tissues from the feeding of one or more oxycarotenoids increases linearly as the level of oxycarotenoid intake increases



over a wide range of supplementation, whereas the visual observation of colour increase is logarithmically related to the colourimetric extract reading.

Other factors influencing pigmentation

Pigmentation involves more than having the right oxycarotenoids in the feed or ration, since numerous factors are involved. Thus, any level of oxycarotenoids supplied by the feed ingredients in the ration does not always produce the same pigmentation results. The quantity and type of dietary xanthophylls are not the only causes influencing the yolk colour. Some others factors are:

#Breed, strain and sex. Different breeds vary in their ability to deposit pigment in the yolk or in the skin. Fletcher et al (1970) compared the colour of yolks from 12 layer strains and in different housing conditions. Both factors affected the pigmentation of egg yolks (measured by reflectance colourimetry). Strain differences can cause 1/6 of the variation in density of yolk colour. Genetic factors also influence the ability of hens to absorb and deposit xanthophylls in the egg yolk, and there are differences even among individual hens within a single strain (Scott et al, 1968). Pigmentation intensity varies also among broiler strains, with males being more intensely pigmented than females.

#Housing conditions. Laying hens kept in cages are able to make better use of yolk pigments than hens kept on a litter floor. When broilers are raised in environmentally dark houses the intensity of pigmentation is stronger rather than in open-sided buildings.

#Diseases. Poultry diseases, such as coccidiosis, chronic respiratory disease (CRD), and Newcastle disease, depress shank colour and skin pigmentation. The inferior pigmentation is due to impaired intestinal absorption. Mycotic infections and mycotic contamination of feedstuffs also depress pigmentation. The pale-bird syndrome, which can be defined as the failure of chickens to realize the colour potential of their diets, is caused by mycotoxins, coccidia, reovirus and dietary unbalances. Theoretically, pale-bird syndrome could result from an impaired accumulation (absorption, transportation and deposition) of carotenoids or from an enhanced excretion of carotenoids.



#Market preparation practices. The pigmentation of broilers can be drastically affected during processing, the main factors being the temperature of scald water, scald time, the kind of plucker and the plucking time (Heath et al, 1975).

#Ration ingredients. Components or ingredients of the feed may have an effect on pigmentation intensity. There are reports on negative effects of soybean and fish meals, while vitamin E enhances pigmentation. The oxycarotenoids effective as pigmenters are structurally similar to the carotenes and vitamin A, and all are very susceptible to the oxidative destruction in the ration. Thus, the reason for including an antioxidant in the ration of broilers and layers is that it will reduce the rate of oxidative destruction of these unsaturated molecules including unsaturated fats during feed or ingredient storage. The effect of fat and oils on broiler and yolk pigmentation depends on the type and level of the fat.

#Fatty acids and fats with varying chain lengths and saturations gave markedly different results when incorporated on an equal weight basis into a diets containing a high level of lutein and fed to depleted birds for two days. Myristic, palmitic and stearic acids as well as tristearin (saturated, long chain fatty acids) supported the absorption of small amounts of lutein in serum. Short-chain, saturated fatty acids and long-chain unsaturated fatty acids improved markedly the absorption of lutein.

#The percentage of fat in isocaloric diets was important for good pigmentation. Adding about 6% cottonseed oil to a diet with white corn, soybean meal, and starch supplemented with a high level of lutein provided almost a threefold increase in the carotenoid found in the toe web over that found with no added fat in the diet. A similar dependency of egg yolk colour on the percentage of dietary fats was also noted by Hamilton and Parkhust.