WHITEPAPER

Intra Calferol - Sunny side up

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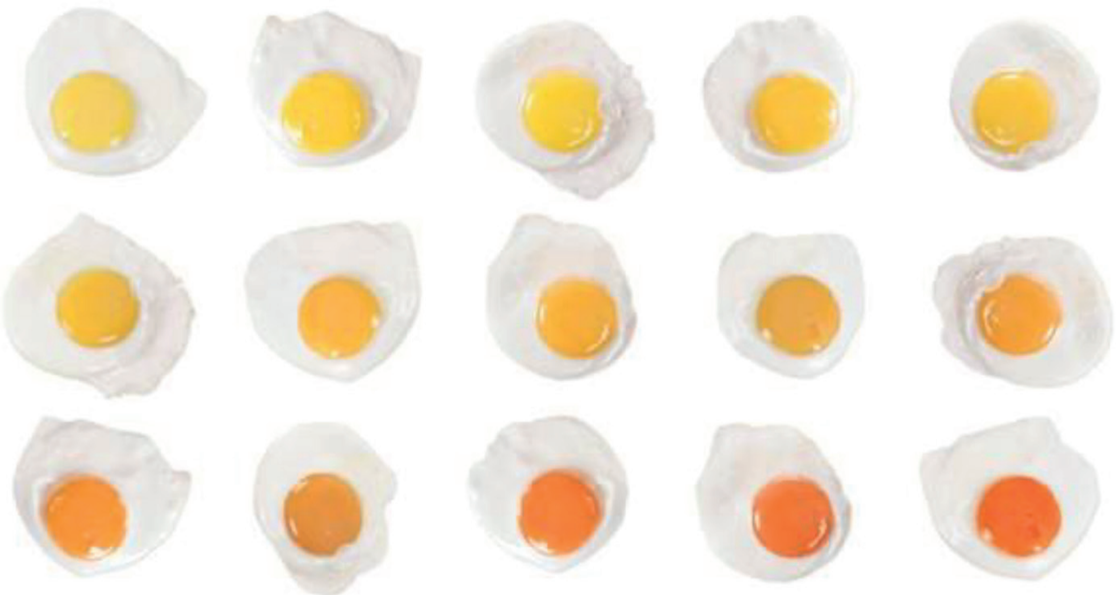
'People eat first with their eyes' is an expression that has long influenced the presentation of food, but also consumer attitudes and shopping tendencies towards food. Consumers make choices on food based on their assessment of quality, which includes the visual appearance of the food itself. Parameters such as uniformity, colour intensity, and smoothness are just some of the examples that consumers use to inspect and judge the quality of a food. When it comes to eggs, one of the important sensory factors used to evaluate quality is the colour of the yolk.



Although yolk colour can be influenced by many factors, one of the determining factors for yolk colour is the health of the flock. Any disruption to the immune status, gastrointestinal or reproductive health can influence the pigment absorption from feed or deposition of pigment into the yolk, causing changes in yolk colour. Vitamin D3, a nutritional substance that has seen extensive use in laying flocks to improve skeletal and eggshell quality, also has the ability to influence yolk colour indirectly. Studies have shown that Vitamin D3 not only plays an important role in the regulation of calcium metabolism in the hen, but also has significant roles in the modulation of the mammalian immune system, with trials reporting a robust immunomodulatory property response when fed to chickens.[1]

The application of precision supplemental nutrition via the drinking water has become increasingly popular over the years to support and maintain the production of high quality eggs in older laying flocks. As feed intake is often compromised during periods of environmental and disease stress, supplemental nutrition via the drinking water can ensure continuous intake of important and sensitive nutritional factors that supports flock health and immunity.

The influence of the mentioned supplemental nutrition on egg quality has been evaluated in an Italian field study with 22.000 Lohmann Brown hens reared in an aviary system. The aviary system consisted of 4 separate boxes. Box 2 was selected as the control, whereas Box 4 was selected as the Intra Calferol trial group. Intra Calferol, a concentrated Vitamin D3 supplement with additional chelated calcium, magnesium and phosphorus releaser was supplemented via the drinking water according to the general protocol and recommended dosage of 1L product in 1000L of water. The trial started when the birds were 69 weeks of age and continued for a duration of 12 weeks. Egg collection, lab analysis and data statistical analysis have been conducted by the Dept of Animal Nutrition of the University of Bologna.

The trial set-up

Daily egg collection generally started at 7.30am starting from box 1 to box 4 and lasted a total of about 1.5 hours. From the collected eggs, a total of 400 eggs were randomly sampled at the start of the trial (T0), after 6 weeks (T1) and after 12 weeks, at the end of the trial (T2). The colour of the yolks were evaluated based on the L*a*b* colour system (CIELAB colour space, International Commission on Illumination). This system classifies colours in terms of their lightness (L), redness (a) and yellowness (b).

Significant difference on Yellowness (b)

The Intra Calferol supplementation was clearly observed in the yellowness of the egg yolk. Although at the start of the trial (week 69), the control group showed a significantly 'yellower' yolk, a shift was seen by the end of the trial period at week 81 and the egg yolks of the Intra Calferol supplemented group had significantly yellower yolks **(figure 1)**.

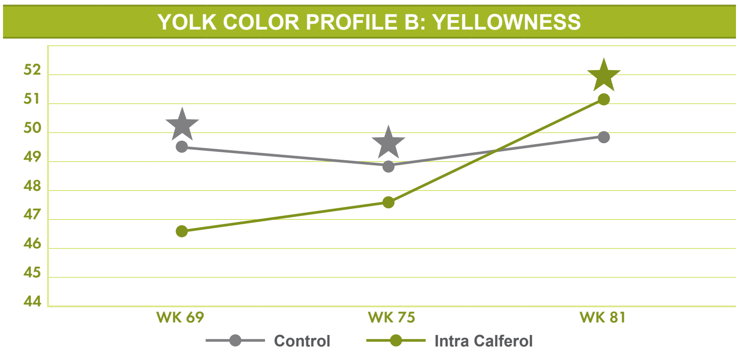


Figure 1: Yolk colour yellowness of control group compared to Intra Calferol supplemented group. Datapoints with an asterisk denotes significant differences.

Improved redness (a)

The results in **figure 2** demonstrated that when it came to the yolk redness, even though the treatment group yolks were significantly less red at the start of the trial (week 69), the supplementation of Intra Calferol resulted in an improved redness and thus was eventually similar to the control group in terms of redness. In short, supplementation of Intra Calferol helped the treatment group 'catch-up' to the control group.

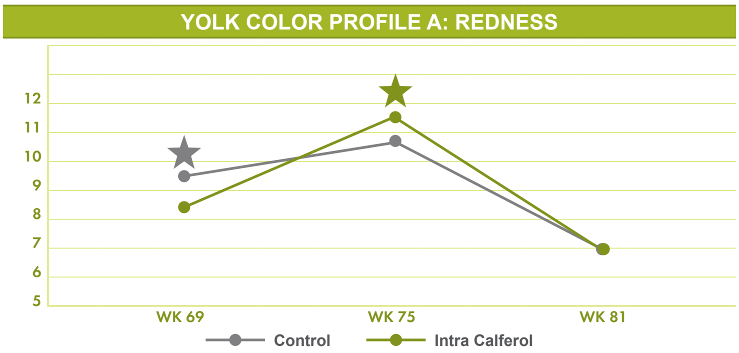


Figure 2: Yolk colour redness of control group compared to Intra Calferol supplemented group. Datapoints with an asterisk denotes significant differences.

Same Lightness (L)

The egg yolk lightness (L) for both groups showed a similar trend of increase over time with no statistical significance. However, since egg yolk redness (a) and yellowness (b) were significantly improved while lightness was similar between both groups, the improvement of the egg yolk visual quality was evaluated further based on the Delta-E gap.

Intra Calferol has closed the Delta E-gap

The Delta E quantifies the difference between two colours. Delta E is calculated on a scale from 0 to 100 (see below **table 1**). A low Delta E (< 1) indicates identical colouring, while a high Delta E level of 100 indicates exactly opposite colours.

DELTA E:	PERCEPTION DIFFERENCE:
<1	Colors not perceptible by the human eye
1 - 2	Hardly perceptible by close observation
2 - 10	Perceptible at a glance
11 - 49	Colors are more the opposite
100	Colors are exactly the opposite

table 1: L*a*b* colour system to calculate Delta E is based on the standard formulation: $\Delta E^*ab = ((\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2)0,5$

The Delta E value at T0 was 2,97 (perceptible at a glance). At T1 it already was closed to 1,82 and even further on T2 to 1,24, a difference hardly not perceptible. Intra Calferol group has clearly caught up, clearly closing the gap.

β-carotene concentrations confirm the better egg yolk colouring

Additionally to the Delta E values, the β-carotene concentration in the yolk was measured. Randomly 30 collected eggs per group were separately measured at week 81 (T2) and independently measured by the lab of a leading egg producer for β-carotene.

The improvement in egg yolk colour after supplementation with Intra Calferol could be related to the average increase of 13% in the β-carotene deposition. β-carotene is the organic pigment responsible for red-orange colouring and higher concentration of this pigment in the yolks of Intra Calferol supplemented eggs lead to a more intense, golden colour of the yolk which also reflected in the L*a*b* colour value.

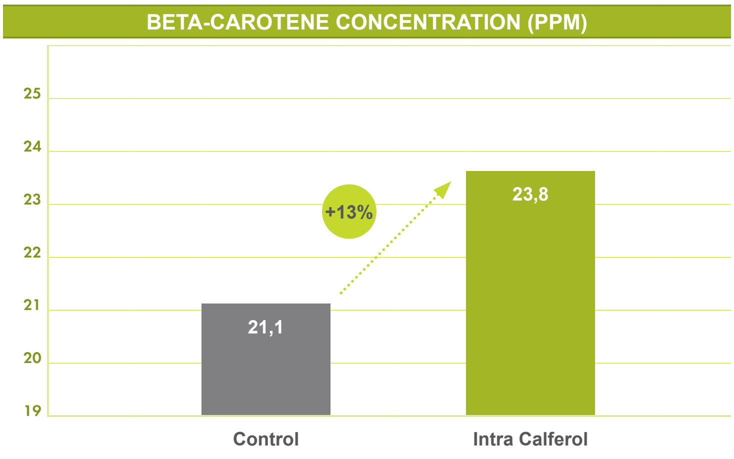


figure 3: Mean beta-carotene concentration of 30 randomly sampled egg yolks

In conclusion, older laying hens supplemented with Intra Calferol produced better quality eggs with more golden yolks with higher beta-carotene level. This was most likely attributed to the improved health status of the flock therefore facilitating more pigment absorption via the gastrointestinal tract and better pigment deposition in the yolk via the reproductive system.