Authentication of organic eggs by LC fingerprinting and isotope ratio analysis

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Introduction

People's growing awareness of health and environment has led to an increased public interest in the quality of foods and food production systems. Organic production benefits from fair competition between producers and sustained consumer confidence, which requires regular confirmation assessments of the identity of organic produce apart from administrative (traceability) controls.

Aim

The aim of the present study was to develop and modify fingerprint methodology for the verification of Dutch organic eggs versus conventional (barn/free range) eggs.

Materials and Methods

Eggs from Dutch farms with laying hens (24 organic, 12 barn, 12 free range) were collected in 2009. Three samples of three pooled yolks or whites were used for analysis. LC fingerprinting and isotope ratio mass spectrometry (IRMS) was applied. For LC fingerprinting carotenoids were extracted from the yolks, and subsequently analysed by HPLC with a reversed phase C18 column and diode array detection at 445 nm. Averages of the triplicate measurements were subjected to multivariate data analysis (Principal Component Analysis (PCA) and k-nearest neighbour (kNN); Pirouette 4.01, Infometrix). Models to predict the production system of the eggs (organic or conventional) from the LC fingerprints were estimated and optimised.

Carbon ($\delta^{13}C$) and nitrogen ($\delta\ ^{15}N$) isotope values were determined using EA-IRMS of dehydrated egg whites. Pooled samples were ground and homogenised before being weighed into tin capsules and analysed in duplicate simultaneously for isotopic abundance as well as total organic carbon and nitrogen. International and working reference standards, and blanks were included during each run for calibration. Isotopic ratios ($^{13}C/^{12}C$ and $^{15}N/^{14}N$) are expressed as isotopic deviations δ defined as:

$$\delta(‰) = \frac{R_s - R_{Ref}}{R_{Ref}} \times 1000$$

where R_s is the isotopic ratio measured for the sample and R_{Ref} that of the international standards.

Results and Discussion

LC fingerprinting

All samples were analysed for their carotenoid profiles by HPLC. PCA on the data showed clustering of organic samples (Fig. 1). The egg samples were kNN classified by their production system: success rates are presented in Table 1. All organic samples, as well as 92% of the conventional egg samples, were classified correctly.

Table 1. Predictions of egg samples into classes applying an optimized kNN model (organic vs. conventional).

Predicted / Actual Class	Correct class (#)	Incorrect class (#)	Success rate
Organic	24	0	100%
Conventional	24	2	92%

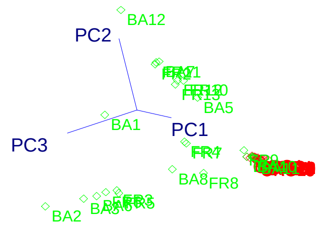

Figure 1. Plot of the first three dimensions of PCA on the carotenoid (LC) profiles of organic (red) and conventional (green) eggs.

Isotope ratio analysis

The results of IRMS are presented in Fig. 2. The organic eggs were generally differentiated from free range and barn eggs by their $\delta^{15}N$ values. Organic eggs are laid by hens which consume only organic feed which has higher $\delta^{15}N$ values than conventional feed. Plants fertilised with organic manures and composts have higher $\delta^{15}N$ values than synthetically fertilised plants. This $\delta^{15}N$ value of feed is transferred via diet into the egg. Free range eggs fall in an intermediate zone between organic and barn eggs as hens can forage outside. Furthermore, these hens fertilise the soil with their manure, also raising the $\delta^{15}N$ values of the vegetation. In this study we found some free range eggs have higher $\delta^{15}N$ values suggesting they have good outdoor access, supplementing their commercial feed. However animal (e.g. milk products) and fish products in feed can also contribute to a higher $\delta^{15}N$ value of the eggs and are allowed w.r.t. EU regulations. Carbon isotopes show that these eggs are consistent with a plant diet, whereby more positive $\delta^{13}C$ values are typical of C4 plants (ie. maize, sorghum and millet), and more negative $\delta^{13}C$ values are typical of C3 plants (ie. wheat, barley and soybeans).

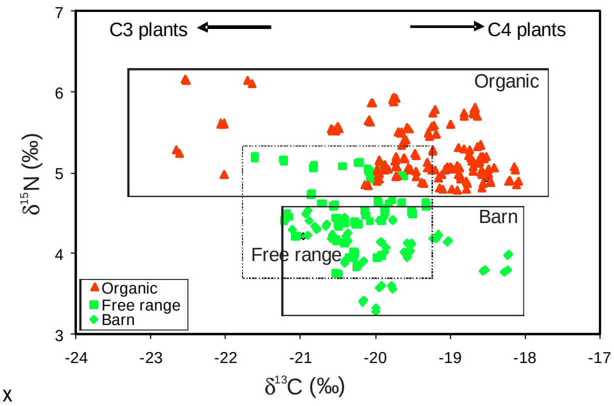

Figure 2. Stable carbon and nitrogen isotopes of organic (red), conventional (green) eggs.

Conclusion

The present study showed that the production system of eggs (organic vs. conventional) can be verified with analytical tools, which is a valuable addition to administrative controls.

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Acknowledgement
This research was (partly) financed by the Dutch Ministry of Agriculture, Nature and Food Quality.