Available online at www.sciencedirect.com
SCIENCE DIRECT®
Food Chemistry 87 (2004) 313–319
Food Chemistry
www.elsevier.com/locate/foodchem

# Hygienic quality evaluation of the egg product used as ingredient in fresh egg pasta

C. Alamprese *, M. Rossi, E. Casiraghi, A. Hidalgo, F. Rauzzino

*Dipartimento di Scienze e Tecnologie Alimentari e Microbiologiche, University of Milan, Via Celoria 2, 20133 Milano, Italy*

Received 1 September 2003; received in revised form 18 December 2003; accepted 18 December 2003

**Abstract**

The aim of this study was to verify the applicability of egg product legal chemical indexes (3OH-butyric acid, succinic acid, lactic acid) and of uracil to fresh pasta, in order to evaluate the hygienic-sanitary quality of the egg ingredient. The analytical characterisation of 20 commercial fresh egg pasta samples and of several ingredients demonstrated that 3OH-butyric acid and uracil are able to highlight the use of egg products of poor hygienic-sanitary quality. For instance, the use of incubator reject eggs in the production of fresh egg pasta (at 20% egg) can be detected by a value of 3OH-butyric acid above 2.83 mg/kg d.m., combined with the presence of uracil. Uracil, which is absent in sound eggs and is not detectable in wheat products, represents a more reliable index for the evaluation of the hygienic quality of eggs in pasta, instead of succinic acid already present in the wheat ingredients.


*Keywords:* Egg product; Fresh egg pasta; Hygienic quality; Organic acid; Uracil

## 1. Introduction

According to Italian legislation (D.P.R. 187/01), fresh egg pasta must be produced using wheat flour and/or semolina and at least four shelled hen eggs (no less than 200 g) per kilogram of wheat product. The eggs may be substituted by an equal quantity of liquid egg product. The possibility to use egg products is a novelty compared to the previous legislation (L. 580/67), which compelled the sole use of fresh eggs.

European Economic Community (EEC) Directive No. 437/89 (EEC, 1989) establishes hygienic-sanitary criteria to be conformed to during the production and marketing of egg products, intended for both retail consumers and food industry. Microbiological legal specifications (Table 1) are accomplished through a thermal treatment equivalent at least to pasteurisation. Furthermore, EEC Directive limits the content of some organic acids, as shown in Table 1.

A 3OH-butyric acid content exceeding the legal limit is an index of the fraudulent addition of incubator reject eggs to egg products (Littman, Schulte, & Acker, 1982; Robinson, Barnes, & Taylor, 1975; Rossi, Hidalgo, Pompei, & Giuffrida, 1999; Salwin, Staruszkiewicz, & Bond, 1972); succinic acid is an index of microbial spoilage (Stijve & Diserens, 1987); lactic acid develops with both microbial and embryonic growth (Stijve & Diserens, 1987).

Besides the criteria provided by European legislation, several authors (Hidalgo, Rossi, Pompei, & Casiraghi, 2003; Littman et al., 1982; Morris, Hoerninf, & St. Angelo, 1989; Rossi et al., 1999) suggested other indexes related to hygienic quality of eggs and egg products. Particularly relevant is uracil whose precursor is uridine, a nucleoside naturally present in eggs. The presence of detectable quantities of uracil in egg products is an evidence of both egg microbial deterioration (Hidalgo et al., 2003; Morris et al., 1989) and addition of incubator reject eggs, the latter if supported by the presence of 3OH-butyric acid (Rossi et al., 1999).

Since eggs are the characterizing ingredient for fresh egg pasta, the aim of this study was to verify the applicability of egg product legal chemical indexes (3OH-butyric acid, succinic acid, lactic acid) and of uracil to fresh pasta, in order to evaluate the hygienic quality of

* Corresponding author. Tel.: +39-0250316625; fax +39-0250316632.
*E-mail address:* cristina.alamprese@unimi.it (C. Alamprese).

0308-8146/$ - see front matter © 2004 Elsevier Ltd. All rights reserved.
doi:10.1016/j.foodchem.2003.12.030

Table 1
Analytical specifications for egg products (EEC, 1989)

| |
|---|
| Microbiological criteria |
| Salmonellae: absence in 25 g or ml of egg product |
| Mesophilic aerobic bacteria: $M = 10^5$ in 1 g or 1 ml |
| Enterobacteriaceae: $M = 10^2$ in 1 g or 1 ml |
| Staphylococci: absence in 1 g of egg product |
| Other criteria |
| 3OH-butyric acid: <10 mg/kg in the dry matter |
| Lactic acid: <1000 mg/kg in the dry matter |
| Succinic acid: <25 mg/kg in the dry matter |

*M*, Maximum value for the number of bacteria

the egg ingredient. To reach this aim, 20 samples of commercial fresh egg pasta of different brands were analysed, together with several samples of wheat flour, semolina, and whole egg products of different hygienic quality.

## 2. Materials and methods

### 2.1. Egg products

Six whole egg samples of different hygienic qualities were analysed: an industrial pasteurised whole egg product (WE1); an industrial pasteurised yolk-enriched (in which the yolk to albumen ratio is higher than the natural value) whole egg product (WE2); two raw whole egg mixes obtained by shelling and homogenising either fresh grade A eggs (EEC, 1991) (WE3) or shell eggs stored at 37 °C for 18 days (WE4); two raw whole egg mixes produced by breaking grade A eggs, leaving them in contact with shells at room temperature (about 25 °C) either for 1 day (WE5) or for 3 days (WE6), removing shells, and homogenising. Samples WE5 and WE6 were prepared in order to simulate the effect of illegal practices (use of broken eggs and/or centrifugation) during egg products manufacture, which lead to high microbial contamination (Hidalgo et al., 2003).

### 2.2. Wheat products

Commercial samples of wheat flour (F1, F2, F3, F4), type "00" (L. 580/67), and semolina (S1, S2, S3, S4) were analysed.

### 2.3. Commercial fresh egg pasta

Twenty different commercial brands of *lasagna* style fresh egg pasta were purchased on the market. At least five packages per brand were used for analysis. According to the labels, eight samples were made with semolina, while the others contained semolina and flour in different ratios. The egg ingredient ranged between 17% and 30%, with an average of 21.8%; three brands did not declare the amount of egg.

Sampling for analysis was performed by blending the content of five packages, belonging to the same lot. Samples are identified by alpha-numeric codes, where the letter indicates the production factory and the number distinguishes the different brands produced in the same factory. Two samples, labelled as organic pasta, are identified by the abbreviation "org".

### 2.4. Dry matter

Dry matter of pasta and wheat products was determined by a gravimetric method according to the Italian Metodi Ufficiali di Analisi dei Cereali (D.M. 21.9.67). Dry matter of whole egg products was determined according to the AOAC 925.30 method (1995). Results are expressed as g/100 g.

### 2.5. Free acidity

Pasta free acidity was determined according to the analytical method of the Italian Istituto Superiore di Sanità, as modified by Acquistucci, Fantauzzi, D'Egidio, Iori, Onori, and Grossi (2000). Results are expressed as acidity degrees corresponding to millilitres of 1 N NaOH needed to titrate 100 g of pasta on dry matter basis.

### 2.6. 3OH-butyric acid

3OH-butyric acid was evaluated using the Boehringer Mannheim enzymatic kit (R-Biopharm GmbH, Darmstadt, Germany). Pasta, semolina and flour samples were prepared for analysis according to the kit method for protein-containing samples, modified as follows: 5 g sample, 15 mL 1 M perchloric acid (Merck, Darmstadt, Germany) and 10 mL HPLC-grade water (BDH Laboratory Supplies, Poole, UK) were blended for 5 min at 8000 $min^{-1}$ using an Ultra Turrax T25 homogeniser (Janke & Kunkel IKA Labortechnik, Staufen, Germany). The mix was then centrifuged at 12,000*g* for 11 min at 15 °C. The supernatant was recovered and brought to pH 7–8 with the addition of solid $KHCO_3$ (Merck) and placed first at −28 °C for 10 min and then at 4 °C for other 10 min. Cooling caused the separation of a precipitate that was discarded. The supernatant was kept at 37 °C for 60 min, centrifuged at 25,000*g* for 11 min at 15 °C, and then filtered through a 0.45 μm HA Millipore membrane (Millipore Corp., Bedford, MA).

Egg products were prepared for analysis following the same procedure, but weighing 15 g sample and homogenising them without water addition.

The detection limits for the various products analysed (Tables 2–4) were calculated on the basis of the absorbance difference ($\Delta A = 0.020$) indicated in the kit instructions, and considering an average dry matter for

Table 2
3OH-butyric acid, succinic acid, and uracil contents (average ± SD) in egg products samples of different hygienic-sanitary quality

| Sample | 3OH-butyric acid (mg/kg d.m.) | Succinic acid (mg/kg d.m.) | Uracil (mg/kg d.m.) |
|---|---|---|---|
| WE1 | 3.3 ± 0.2 | 13.7 ± 1.9 | n.d. |
| WE2 | n.d. | n.d. | n.d. |
| WE3 | n.d. | 8.24 ± 0.02 | n.d. |
| WE4 | 3.86 ± 0.36 | 14.93 ± 0.49 | n.d. |
| WE5 | 5.66 ± 2.17 | 18.24 ± 4.73 | 19.45 ± 1.10 |
| WE6 | 7.57 ± 5.15 | 562.73 ± 2.80 | 29.81 ± 1.54 |
| WE7[a] | 127.26 ± 83.94 | 12.00 ± 0.35 | 13.30 ± 13.9 |

n.d., not detectable; <2.70 mg/kg d.m. for 3OH-butyric acid; <9.70 mg/kg d.m. for succinic acid; <5.20 mg/kg d.m. for uracil (considering a dry matter of 23.7 g/100g).

[a] Average value of four incubator reject egg lots candled at the eighth and 18th day (Giuffrida, 1998).

Table 3
3OH-butyric acid, succinic acid and uracil contents (average ± SD) in wheat products analysed

| Sample | 3OH-butyric acid (mg/kg d.m.) | Succinic acid (mg/kg d.m.) | Uracil (mg/kg d.m.) |
|---|---|---|---|
| S1 | 1.91 ± 0.14 | 81.94 ± 0.71 | n.d. |
| S2 | n.d. | 60.63 ± 0.02 | n.d. |
| S3 | n.d. | 63.86 ± 4.73 | n.d. |
| S4 | n.d. | 82.73 ± 2.80 | n.d. |
| *Average* | | 72.29 ± 11.68 | |
| F1 | n.d. | 45.63 ± 1.98 | n.d. |
| F2 | n.d. | 42.45 ± 1.46 | n.d. |
| F3 | n.d. | 46.41 ± 0.06 | n.d. |
| F4 | n.d. | 49.84 ± 0.31 | n.d. |
| *Average* | | 46.08 ± 3.03 | |

n.d., not detectable; <1.86 mg/kg d.m. for 3OH- butyric acid; <1.07 mg/kg d.m. for uracil (considering a dry matter of 85 g/100g).

each type of product. Results are expressed as mg/kg of dry matter.

### 2.7. Succinic acid

The analysis of succinic acid was carried out using the Boehringer Mannheim (R-Biopharm GmbH) enzymatic kit. The preparation of the samples was performed as for the analysis of 3OH-butyric acid. The detection limit of succinic acid for the various products analysed were calculated as for 3OH-butyric acid (Tables 2–4). Results are expressed as mg/kg of dry matter.

### 2.8. Uracil

The uracil content was determined following the HPLC method described for eggs by Morris (1987), as modified by Rossi and Pompei (1995).

For wheat products and pasta analysis, sample preparation was changed per sample size and sample to reagent ratio, in order to obtain an adequately concentrated sample solution. An aliquot of minced pasta (10.5 g) or wheat product (7 g) was homogenised with 7 mL HPLC-grade water (BDH), 11.2 mL of 6% v/v perchloric acid (Merck) and 1.7 mL HPLC-grade acetonitrile (BDH) using an Ultra Turrax T25 (Janke & Kunkel) at 8000 $min^{-1}$ for 3 min. The homogenised solution was centrifuged at 12,000*g* for 11 min at 15 °C and then filtered through a 0.45 μm HA Millipore membrane. Injection volume was 100 μL. A calibration curve was prepared using eight different uracil (Merck) solutions from 0.2 to 10 mg/L. Based on the calibration curve, the detection limit was calculated as the value of

Table 4
Free acidity, 3OH-butyric acid, succinic acid and uracil contents (average ± SD) in commercial fresh egg pasta samples analysed

| Sample | Free acidity (°) | 3OH-butyric acid (mg/kg d.m.) | Succinic acid (mg/kg d.m.) | Uracil (mg/kg d.m.) |
|---|---|---|---|---|
| A1org | 2.44 ± 0.07 | n.d. | 61.23 ± 0.88 | n.d. |
| B1 | 1.50 ± 0.01 | 2.41 ± 0.08 | 45.73 ± 1.77 | n.d. |
| C1 | 2.05 ± 0.07 | 2.91 ± 0.31 | 78.45 ± 1.88 | n.d. |
| D1 | 1.95 ± 0.01 | n.d. | 58.96 ± 1.39 | 1.25 ± 0.01 |
| E1 | 2.18 ± 0.10 | n.d. | 69.12 ± 0.74 | n.d. |
| E2org | 1.87 ± 0.06 | n.d. | 81.67 ± 0.69 | n.d. |
| F1 | 1.59 ± 0.01 | n.d. | 64.63 ± 1.14 | n.d. |
| G1 | 1.30 ± 0.11 | n.d. | 54.09 ± 0.57 | n.d. |
| G2 | 2.04 ± 0.16 | n.d. | 58.31 ± 2.89 | n.d. |
| G3 | 1.67 ± 0.01 | n.d. | 53.12 ± 1.34 | 2.42 ± 0.03 |
| G4 | 1.64 ± 0.05 | n.d. | 50.20 ± 0.63 | n.d. |
| G5 | 1.58 ± 0.11 | n.d. | 53.98 ± 0.99 | n.d. |
| H1 | 2.05 ± 0.01 | n.d. | 56.12 ± 1.12 | 1.23 ± 0.12 |
| H2 | 2.31 ± 0.11 | n.d. | 140.97 ± 6.73 | 2.01 ± 0.04 |
| H3 | 2.04 ± 0.11 | n.d. | 55.46 ± 1.76 | 1.01 ± 0.01 |
| H4 | 2.18 ± 0.10 | 3.76 ± 0.32 | 100.80 ± 4.16 | 1.05 ± 0.04 |
| H5 | 2.06 ± 0.05 | n.d. | 126.53 ± 8.49 | 0.90 ± 0.01 |
| I1 | 1.92 ± 0.03 | n.d. | 63.28 ± 2.41 | 0.94 ± 0.01 |
| I2 | 1.77 ± 0.10 | n.d. | 64.66 ± 4.70 | 0.90 ± 0.01 |
| L1 | 1.98 ± 0.06 | n.d. | 64.84 ± 1.60 | 1.96 ± 0.02 |

n.d., not detectable; <2.28 mg/kg d.m. for 3OH-butyric acid; <0.87 mg/kg d.m. for uracil (considering a dry matter of 70 g/100g).

the intercept of the regression line plus three times the standard error of estimate (Miller & Miller, 1988).

Results are expressed as mg/kg of dry matter.

### 2.9. Lactic acid

To determine lactic acid, two methods were used: the HPLC method described above for uracil analysis, but using a refractive index detector (Hidalgo et al., 2003) and an enzymatic kit method (R-Biopharm GmbH). The preparation of the samples for the enzymatic analysis was carried out following the method described for 3OH-butyric acid.

All the analyses were performed in duplicate.

## 3. Results and discussion

### 3.1. Analytical methods

The sample preparation suggested for eggs in the enzymatic kit instructions demonstrated to be not feasible for the analysis of pasta and wheat products, as this preparation method requires a heating step at 100 °C for 15 min. Due to this treatment, the gelatinisation of starch occurs, which impairs clarification of sample solution. Therefore, the enzymatic kit sample preparation suggested for protein-containing samples was adopted, modifying the ratios between sample size and reagents, in order to avoid excessive viscosity of homogenised sample mix. A heating phase at 37 °C was also introduced, in order to produce the formation of floccules, subsequently removed through centrifugation and filtration, prior to spectrophotometric determination. The same preparation method was also applied to the egg product in order to obtain data comparable to those pertaining to pasta and wheat products.

While developing the enzymatic kit method for lactic acid in pasta, an unsatisfactory extraction yield (about 50%) was observed. Extraction yield was evaluated analysing a sample of home-made pasta prepared with eggs and wheat products of pre-determined lactic acid contents. As an alternative, HPLC analysis of lactic acid in pasta was attempted, however, the peak ascribed to lactic acid, which is well separated in egg products, resulted hidden by other peaks in pasta chromatogram. Consequently, lactic acid was not determined, since the analytical methods commonly used are not feasible for pasta products.

### 3.2. Egg products

Table 2 shows the level of 3OH-butyric acid, succinic acid and uracil in the analysed egg samples of different hygienic-sanitary quality. For comparison, average literature values relating to samples of incubator reject eggs (sample WE7) are also reported (Giuffrida, 1998). All experimental samples present levels of 3OH-butyric acid below the legal limits (10 mg/kg d.m.). On the other hand, the 3OH-butyric acid literature value for sample WE7 is abundantly above this limit. 3OH-butyric acid is present only in fertilised eggs and its concentration is not related to any microbial development but only to embryo metabolism. Besides, neither the hen breed nor the incubator procedures, nor thermal treatment affect the level of 3OH-butyric acid in incubator reject eggs (Jones & Ellingworth, 1979; Robinson et al., 1975; Salwin et al., 1972).

Succinic acid which is, on the contrary, an index linked to bacterial and not embryo metabolism (Stijve & Diserens, 1987) results abundant only in eggs left to deteriorate for 3 days (sample WE6). The same amount of succinic acid was found in liquid raw eggs having a microbial count of about $10^9$ cfu/g (Hidalgo et al., 2003). In all other egg samples, this acid is below the legal limit (25 mg/kg d.m.).

The results concerning uracil show that this compound is absent in whole egg samples made either with fresh eggs (samples WE1, WE2 and WE3) and stored eggs (sample WE4), yet it is present in the incubator reject eggs (WE7) and in the eggs deteriorated for 1 day (WE5) or 3 days (WE6), being higher in WE6.

It is noteworthy how uracil turns out to be an hygienic quality index more sensitive than the legal limit for succinic acid. In fact uracil was even found in sample WE5, presenting a level of succinic acid below the legal limit. According to Hidalgo et al. (2003), the presence of detectable quantities of uracil in egg products is enough to indicate an unacceptable microbial development.

### 3.3. Wheat products

Table 3 shows the results of the analyses carried out on flour and semolina samples. They do not contain detectable quantities of 3OH-butyric acid, except for sample S1 presenting a value equivalent to the detection limit in wheat products. Thus in theory, the possible presence of detectable quantities of 3OH-butyric acid in fresh pasta should comes from the contribution of eggs. This aspect will be thoroughly treated further on.

Succinic acid instead was present in all wheat products, at higher levels in semolina: the amount of this acid was even higher than the legal limit for egg products.

No detectable quantities of uracil were found in wheat products. Therefore, the possible presence of uracil in fresh egg pasta could be ascribed to the use of deteriorated eggs or incubator reject eggs, the latter if 3OH-butyric acid is also present.

### 3.4. Commercial fresh egg pasta

The expiration dates reported on labels indicated a rather variable shelf life of the analysed fresh egg

pasta, related to packaging techniques (aseptic and non) and to conditions and number of thermal treatments applied during the production process. Shelf life normally varied between 15 and 55 days for those products submitted to only one pasteurisation, yet it varied between 60 and 90 days for products subjected to double pasteurisation, before and after packaging. All the samples purchased were analysed between one week and one month after manufacture, anyhow before the expiration date.

In order to obtain an indication about analytical indexes stability during a 30-day storage period, a sample of commercial pasta, withdrawn right after its packaging at the production plant and stored at 4 °C up to 30 days, was analysed. Besides the indexes related to egg products, the free acidity of pasta was evaluated, being an Italian legal index linked to pasta hygienic quality (D.P.R. 187/01).

For all considered parameters, no evolution was noticed during storage of this sample. Acidity, which must not exceed 7° by law, presented an average value of 2.3° during storage, indicating a substantial microbial stability. This was also confirmed by the average concentration of succinic acid (54.20 mg/kg d.m.), equivalent to the average levels observed in flour and semolina (Table 3), and by the absence of detectable quantities of uracil. The level of 3OH-butyric acid was below the detection limit.

Table 4 shows the analytical values obtained for the 20 samples of commercial fresh egg pasta. Free acidity of the analysed samples ranged between 1.30° and 2.44°, rather below the limit prescribed by law.

Only three samples showed detectable levels of 3OH-butyric acid. It should be considered that the absorbance difference, measured during spectrophotometric determination of 3OH-butyric acid in pasta, is due to the contributions of both wheat products, which give signals below the detection limit, and eggs. To assess the levels of 3OH-butyric acid in pasta, above which the possible use of incubator reject eggs becomes detectable, theoretical calculations were carried out (Table 5). A hypothetical pasta formulation containing 37% semolina, 37% flour, 20% eggs and 6% water (dry matter about 68%) was considered. Based on this formulation, three pasta samples were hypothesised containing three kinds of eggs: 100% incubator reject eggs (P1); a whole egg product at 20% incubator reject eggs (P2); a whole egg product having 3OH-butyric acid level equivalent to the legal limit (P3). For the incubator reject eggs, an average value of 3OH-butyric acid of 127.26 mg/kg d.m. (Table 2) was considered, corresponding to 30.16 mg/kg on an as is basis (egg dry matter 23.7 g/100g). The average contents of 3OH-butyric acid in flour and semolina were calculated according to the signals recorded for the samples reported in Table 3, although they were below the detection limit.

Using egg products at 100% (P1) or 20% (P2) incubator reject eggs, theoretical figures of 3OH-butyric acid above the detection limit (2.35 mg/kg d.m. of pasta) are obtained. On the other hand, using an egg product at a 3OH-butyric acid level equivalent to the legal limit (P3), a theoretical figure of 2.12 mg/kg d.m. is obtained. Adding to this value, which is below the detection limit, three times the average standard deviation of analysis

Table 5
3OH-butyric acid calculated concentration in three hypothetical pasta formulations containing egg products of different hygienic-sanitary quality

| Pasta sample | Ingredients | Percent ingredient (%) | Ingredient 3OH-butyric acid average content (mg/kg) | 3OH-butyric acid weighed contribution | |
|---|---|---|---|---|---|
| | | | | mg/kg | mg/kg d.m. |
| P1 | Egg product at 100% I.R.E. | 20 | 30.16 | 6.03 | |
| | Semolina | 37 | 1.25 | 0.46 | |
| | Flour | 37 | 1.37 | 0.51 | |
| | *Sum* | | | 7.00 | 10.29 |
| P2 | Egg product at 20% I.R.E. | 20 | 6.54 | 1.31 | |
| | Semolina | 37 | 1.25 | 0.46 | |
| | Flour | 37 | 1.37 | 0.51 | |
| | *Sum* | | | 2.28 | 3.35 |
| P3 | Egg product at legal limit | 20 | 2.37 | 0.47 | |
| | Semolina | 37 | 1.25 | 0.46 | |
| | Flour | 37 | 1.37 | 0.51 | |
| | *Sum* | | | 1.44 | 2.12 |

I.R.E. = incubator reject eggs.

for pasta samples (Table 4), a theoretical figure is obtained equivalent to 2.83 mg/kg d.m., which is above the detection limit. Considering the samples B1, C1 and H4, showing detectable values of 3OH-butyric acid in Table 4, the calculated theoretical figure (2.83 mg/kg d.m.) excludes sample B1 from being suspected of containing incubator reject eggs. Also sample C1 should not be suspected of fraud, if considering the standard deviation associated to its level of 3OH-butyric acid. This hypothesis is also supported by the absence in both samples of uracil which, as proven by Rossi et al. (1999), accompanies 3OH-butyric acid in incubator reject eggs. Only sample H4 is seriously suspected of containing incubator reject eggs, presenting a level of 3OH-butyric acid above 2.83 mg/kg d.m. in combination with a detectable quantity of uracil.

Samples of commercial fresh egg pasta (Table 4) present an average succinic acid level of about 70 mg/kg d.m., above the legal limit for egg products. Particularly, samples H2, H4 and H5 present values much higher than average. The amount of succinic acid in a hypothetical fresh egg pasta containing 74% wheat product, 20% egg product at 25 mg/kg d.m. succinic acid (legal limit), and 6% water was calculated in order to define a possible minimum level of succinic acid in pasta which indicates the use of eggs having outlaw level of this acid. Due to a higher average value of succinic acid (Table 3), only semolina was considered as wheat ingredient in this formulation. The calculated content of succinic acid is 81.5 mg/kg d.m. Three times the average standard deviation of analysis for pasta samples (Table 4) was added to the calculated succinic acid concentration, obtaining a value of about 88 mg/kg d.m.

Comparing this value with those shown in Table 4 for commercial samples, it may be hypothesised that samples H2, H4 and H5 were produced using poor hygienic quality egg products. This suspect is confirmed by the presence of uracil.

Uracil, that must be absent in sound eggs (Hidalgo et al., 2003; Rossi et al., 1999) and is not detectable in wheat products, could represent a more reliable index for the evaluation of hygienic quality of eggs in pasta, instead of succinic acid.

Observing the analytical values shown in Table 4, it can be noticed that half of the examined pasta samples present detectable quantities of uracil. Five of them (H1–H5) were marketed under different brands, although produced in the same factory. Since these samples were acquired at different times, thus apparently belonging to different production lots, it can be assumed that their production factory purchases, as a rule, poor quality egg products. This kind of products can be found at competitive prices on the market of industrial egg products and allow the pasta production costs to drop. By chance, the only sample of pasta bought at a low-price discount store (H2) belongs to this group.

## 4. Conclusions

The analytical characterisation of raw materials and commercial fresh egg pasta demonstrated that analytical indexes taken into consideration are able to highlight the use of poor hygienic-sanitary quality egg products. For instance, the use of an egg product contaminated by incubator reject eggs in the production of fresh egg pasta (at 20% egg) can be detected by a value of 3OH-butyric acid above 2.83 mg/kg d.m., combined with the presence of uracil.

Uracil and succinic acid indicate the hygienic quality of eggs used in pasta production. However, while the presence of uracil in fresh egg pasta is enough to give the egg ingredient negative judgement, the level of succinic acid is more difficult to interpret, since this acid is naturally present in wheat products and seems to be thermolabile as well (Hidalgo et al., 2003).

## References


Acquistucci, R., Fantauzzi, P., D'Egidio, M. G., Iori, A., Onori, R., & Grossi, S. (2000). Ring test to revised the Italian method for the determination of free acidity in semolina and pasta. *Tecnica Molitoria, 51*, 113–121.

AOAC. (1995). *Official methods of analysis* (16th ed.). Gaithersburg, Maryland, USA: Association of Official Analytical Chemists.

D.M. 21.9.67. Metodi Ufficiali di Analisi dei Cereali. *Gazzetta Ufficiale, 285*.

D.P.R. 187/01. Regolamento per la revisione della normativa sulla produzione e commercializzazione di sfarinati e paste alimentari, a norma dell'articolo 50 della legge 22 febbraio 1994, no. 146. *Gazzetta Ufficiale, 117*, 6–12.

EEC. (1989). Directive No. 437/89 on the hygiene and health problems affecting the production and the placing on the market of egg products. *Official Journal of the European Community*, L212, 87–100.

EEC. (1991). Regulation No. 1274/91 for the application of the EEC Regulation No. 1907/90. *Official Journal of the European Community*, L121, 11–24.

Giuffrida, F. (1998). *Studio di indici analitici per l'individuazione delle uova di scarto di incubatoio nel misto d'uovo pastorizzato*. Thesis, University of Milan, Italy.

Hidalgo, A., Rossi, M., Pompei, C., & Casiraghi, E. (2003). Chemical indices for hygienic quality evaluation of pasteurized whole egg products. *Journal of Agricultural and Food Chemistry*, submitted.

Jones, J. M., & Ellingworth, C. E. (1979). Technical note. Detection of incubator rejects in whole eggs after normal commercial processing. *Journal of Food Technology, 14*, 199–203.

L. 580/67. Disciplina per la lavorazione e commercio dei cereali, degli sfarinati, del pane e delle paste alimentari. *Gazzetta Ufficiale, 189*.

Littman, S., Schulte, E., & Acker, L. (1982). Beurteilung des hygienischen Zustandes von Eiprodukten vor der Pasteurisierung über das Muster der organischen Sauren. *Zeitschrift für Lebensmittel-Untersuchung und-Forschung, 175*, 101–105.

Miller, J. C., & Miller, J. N. (1988). *Statistics for analytical chemistry* (2nd ed.). Chichester, West Sussex, England: Ellis Hortwood Limited Publishers.

Morris, C. E. (1987). Determination of uracil, uridine and formic acid in egg products by high-performance liquid chromatography. *Journal of Chromatography, 394*, 408–413.

Morris, C. E., Hoerninf, E. F., & St. Angelo, A. J. (1989). Uracil as a potentially useful indicator of spoilage in egg products. *Journal of Food Science, 54*, 581–583.

Robinson, D. S., Barnes, E. M., & Taylor, J. (1975). Occurrence of β-hydroxybutyric acid in incubator reject eggs. *Journal of the Science of Food and Agriculture, 26*, 91–98.

Rossi, M., & Pompei, C. (1995). Changes in some components and analytical values due to hen age. *Poultry Science, 74*(1), 152–160.

Rossi, M., Hidalgo, A., Pompei, C., & Giuffrida, F. (1999). A new approach to the identification of incubator reject eggs in pasteurized whole egg products obtained from fresh eggs. In *Proceeding of the VIII European Symposium on the Quality of Eggs and Egg Products* (pp. 451–456). Parma: Tipografia Benedettina S.c.r.l.

Salwin, H., Staruszkiewicz, W. F., & Bond, J. F. (1972). Formation of β-hydroxybutyric acid in eggs during incubation. *Journal of the Association of Official Analytical Chemists, 55*, 458–460.

Stijve, T., & Diserens, J. M. (1987). Simplified method for the routine determination of organic acids in eggs and egg products. *Deutsche Lebensmittel Rundschau, 83*(2), 44–47.