Identification of total aromas of plant protein sources

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Received June 15, 2017; Accepted in revised form September 30, 2017; Published August 25, 2020

Abstract:
Introduction. Due to the deficit and high cost of complete animal protein, the search and analysis of alternative sources is an actual scientific trend. Lentils is a good alternative to animal protein, but the pronounced bean smell and taste limit its full or partial use in food production. The aim of the work was to determine the total aromas of lentils when germinated to eliminate the bean taste and smell.

Study objects and methods. The object of the study was brown lentil beans germinated under laboratory conditions. Samples of the equilibrium gas phase formed over samples of wet and sprouted beans were investigated. The analysis of total aromas was carried out on a laboratory odor analyzer MAG-8 (“electronic nose”) by the method of piezoelectric quartz micro-weighing with an array of sensors.

Results and discussion. The study results showed qualitative and quantitative differences in the equilibrium gas phase over samples of wet and germinated grain. The quantitative analysis showed that the content of volatile compounds over sprouted grain is 12% less than over wet. The qualitative composition of the samples of wet and sprouted grain differed by 60%, which confirmed the influence of germination on the composition of the equilibrium gas phase and the possibility of eliminating bean odor. Testing showed that the use of pre-processed lentil grains allows to replace up to 50% of raw meat in minced products (minced food, chopped food) without changing the smell of the products.

Conclusion. According to the results obtained, preliminary processing of lentils by germination will allow using this bean culture as an alternative source of animal protein to expand the range, and improve the quality of meat and dairy products.

Keywords: Lentils, germination, amino acid composition, biological value, total flavors, total analytic signal, the equilibrium gas phase

Funding: The authors are grateful to the administration of Voronezh State University of Engineering Technologies (VSUET) for the opportunity to carry out the study in specialized laboratories.

Please cite this article in press as: Antipova LV, Kuchmenko TA, Osmachkina AA, Osipova NA. Identification of total aromas of plant protein sources. Foods and Raw Materials. 2020;8(2):362–368. DOI: http://doi.org/10.21603/2308-4057-2020-2-362-368.
proved the benefits of legume proteins traditionally grown in Russia (Saratov, Samara, Orenburg and Penza regions and Altai Krai), the potential of which can be significantly increased by pre-germination [1].

Among alternative sources of protein, legumes occupy priority positions in terms of protein quantity and quality. That is why they are preferred when creating, for example, meat food systems [5, 6]. Among them, lentils, known for its healing properties, should be distinguished. By the quantity and quality of amino acids, the lentil protein is closest to the first grade beef protein. Compared to other legumes, lentils have a more balanced amino acid composition, containing an increased amount of valuable vitamins, macro-, microelements (including iodine), and less oligosaccharides that cause intestinal flatulence.

Lentils is one of few cultures containing only one digestive enzyme inhibitor that acts on trypsin (Table 1).

Lentils have good botanical properties, develop and fruit well in Central Russia (Chernozemye, Volga region).

The choice of lentils as an object of the study is justified by the results of chemical analysis obtained by Howell, according to which this culture has a number of obvious advantages compared to other legumes [9]. However, the test results achieved in the conditions of the “Kalacheevsky” meat factory showed the limited use of lentils even in small quantities (5–8%) due to its bean taste and smell. In this regard, germination is of obvious advantages compared to other legumes [9].

Our previous results proved that germinating significantly improved the biological value of raw materials, namely the content of proteins, vitamins, and minerals. The amount of amino acids increased by a factor of 1.5–2 (Fig. 1).

The amino acid composition of the protein becomes more balanced, with the score close to the score of the reference protein (Fig. 2). A significant increase in lysine and tryptophan, the most valuable amino acids, can be mentioned. Lysine is a deficient amino acid, which, combined with vitamins, strengthens the immune system, promotes calcium absorption from the intestine, as well as contributes to cellular protein and bone tissue formation. Tryptophan is involved in the serotonin (the hormone of happiness) formation; mood, sleep quality, level of pain threshold and susceptibility to various irritants and inflammations depend on its concentration.

An increase in the content of minerals and vitamins, as well as a decrease in the number of oligosaccharides, extremely insufficient, and ungeneralized [13–16]. Lentil products (flour, concentrate, isolate) combine well and replace food systems of animal origin (meat, dairy).

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| Inhibitors          | Their action                                                                 | Inhibitors          | Their action                                                                 |
|---------------------|------------------------------------------------------------------------------|---------------------|------------------------------------------------------------------------------|
| 1. Protease inhibitors | Binding trypsin and chymotrypsin, which are secreted by the animal’s pancreas, thereby reducing the efficiency of digestion of feed. | 1. Trypsin inhibitor | Inhibiting the activity of trypsin, which is synthesized by the pancreas in the form of zymogen, activated in the duodenum 12, and catalyzes the hydrolysis of proteins and peptides. Trypsin is involved in the activation of many digestive enzymes through a limited proteolysis mechanism. |
| 2. Urease Inhibitors | Suppressing urease activity, thereby reducing urea hydrolysis with ammonia formation, and neutralizing its toxic effect on the body. |                     |                                                                              |
| 3. Lipoxigenase inhibitors | Inhibiting the activity of lipoxigenase involved in the enzymatic rancidity of fats during storage with the formation of aldehydes and ketones, which give an unpleasant smell and taste. |                     |                                                                              |

**Table 1 Digestive enzyme inhibitors in legumes**

![Amino acid composition of lentil grains, beef and poultry meat](image)
The study was to assess the total aromas of lentils during germination, to eliminate extraneous odor by partial or complete replacement of animal proteins in food systems of animal origin.

STUDY OBJECTS AND METHODS

The object of study was brown lentil beans (State Standard 7066-77) germinated in laboratory conditions at Voronezh State University of Engineering Technologies.

Grains of untreated lentils were germinated at 21–23°C for 3–4 days, preventing their complete drying out.

The general chemical composition of sprouted lentils is presented in Table 2, and the amino acid composition in Fig. 1. It can be seen from the data that germinated lentil grains have significant advantages in the content of the most important nutrients.

The study of total aromas was carried out on a MAG-8 laboratory (experimental) odor analyzer (Fig. 3) with the electronic nose methodology, by the method of piezoelectric quartz weighing with an array of sensors [20–22]. We analyzed three samples: wet grain, germinated grain and water.

The sensor array consisted of eight sensors based on BAW-type piezoelectric quartz resonators with an oscillation frequency of 10.0 MHz and with diverse film sorbents on electrodes.

Coatings are selected in accordance with the test objective (possible emission from samples of various organic compounds):

Sensor 1 – Multilayer Carbon Nanotubes, MCNT;
Sensor 2 – Polyethylene glycol succinate, PEGS;
Sensor 3 – Polyethylene glycolsebacinate, PEGSb;
Sensor 4 – Polyethylene Glycol Adipate, PEGA;
Sensor 5 – Polyethylene glycol-2000, PEG-2000;
Sensor 6 – Dicyclohexane-18-Crown-6, DCH18C6;
Sensor 7 – Twin-40, Tween;
Sensor 8 – Polyethylene Glycolphthalate, PEGP.

Grain samples were placed in glass tubes (10 g in each), tightly closed, kept at room temperature (20 ± 1°C) for at least 20 min to saturate the equilibrium gas phase over the samples. Then, we determined moisture content, which amounted to 51.2%. 3 cm³ of the equilibrium gas phase was taken through a membrane with individual syringes and introduced into the detection cell.

Background of the array of sensors was from 15 to 30 Hz·s. The measurement time was 60 s, the mode for recording sensor responses was uniform with a step of 1 s, the optimal algorithm for presenting

Table 2 Effect of germination on chemical composition of lentil seeds

| Indicators   | Content in 100 g of product Before germination | After germination |
|--------------|-----------------------------------------------|------------------|
| Proteins, g  | 26.15                                         | 29.56            |
| Fat, g       | 1.2                                           | 1.1              |
| Carbohydrates, g | 53.7                                         | 41.06            |
| including glucose | 8.45                                         | 13.64            |
| Oligosaccharides: |                                              |                  |
| raffinose    | 0.9                                           | 0.5              |
| stachyose    | 2.7                                           | 2.1              |
| verbascose   | 1.4                                           | 0.8              |
| Starch       | 33.8                                          | 24.12            |
| Cellulose    | 3.65                                          | 3.04             |
| Ash          | 3.65                                          | 3.31             |
| Moisture     | 12.33                                         | 18.1             |
| Minerals, mg |                                              |                  |
| calcium      | 84.23                                         | 84.62            |
| phosphorus   | 401.16                                        | 400.3            |
| magnesium    | 78.9                                          | 76.3             |
| iron         | 12.06                                         | 12.32            |
| sodium       | 56.12                                         | 55.91            |
| potassium    | 659.18                                        | 659.51           |
| Vitamins, mg |                                              |                  |
| B1           | 0.5                                           | 0.78             |
| B2           | 0.21                                          | 0.48             |
| PP           | 1.8                                           | 2.21             |
| C            | –                                              | 0.04             |
| β-carotene   | 0.03                                          | 0.08             |

1 State Standard 7066-77. Lentil for human consumption (plate-shaped). Requirements for state purchases and deliveries. Moscow: Izdatel'stvo standartov; 2003. 8 p.
responses was based on the maximum responses of individual sensors. The measurement error was 10%.

The total analytical signal is generated by using the integrated signal processing algorithm of eight sensors in the form of a “visual imprint”. To determine the total composition of sample smell, we used the full “visual imprints” of the peaks (the largest responses of eight sensors), constructed from the maximum responses of the sensors in the equilibrium gas phase of the samples during the measurement time (no more than 1 min). The similarity and difference in the composition of the volatile odor fraction over the analyzed samples was established [20]. Slight differences in the composition of the gas mixture were established by comparing the kinetic “visual imprints” constructed from the responses of all the sensors recorded at different times over the entire measurement interval. The nature of the components mixture is more apparent in such analytical signals. Both types of signals, as well as the area of the figures are calculated automatically in the instrument software.

The following criteria for assessing differences in the smell of the analyzed samples are selected:

- A qualitative characteristic – the form of a “visual imprint” with characteristic distributions along the response axes, was determined by the set of compounds in the equilibrium gas phase.

Quantitative characteristics:

1) The total area of the full “visual imprint” ($S_\Sigma$, Hz·s) was used to estimate the total intensity of the aroma proportional to the concentration of volatile substances, including water. This parameter was constructed from all signals of all sensors for the full measurement time;

2) The maximum signal of sensors with the most active or specific sorbent films ($\Delta F_{\text{max}}$, Hz) was applied to assess the content of individual classes of organic compounds in the EGP by the normalization method [21, 22];

3) The identification parameter ($A_{ij}$) was used to identify individual classes of compounds in a mixture. This parameter was calculated from the signals of the sensors in the analyzed samples and for standard compounds.

Sensor responses were recorded, processed and compared in the software of the MAG Soft analyzer.

**RESULTS AND DISCUSSION**

In the course of experimental studies (Table 3), the total content of volatile compounds in the EGP was found to correlate with the total analytical signal of the “electronic nose” – the area of the “visual imprint” of the response peaks.

Insignificant differences were found in the total odor intensity over samples of wet and germinated grains, however, the contribution to the total sorption response of different classes of compounds is not equal. To establish differences in the composition (qualitative and quantitative) of the volatile odor fraction, we analyzed the total content of readily volatile components in the equilibrium gas phase over the samples (Fig. 4).

The shape of the “visual imprint” of the sensor responses in the array showed insignificant differences in the chemical composition of the equilibrium gas phase over samples of wet and germinated grain. The content of volatile compounds in the equilibrium gas phase over germinated grain was less by 12% than over wet grain.

Additionally, we noted the change in the quantitative composition of the odor above the samples according to the relative content of the main classes of volatile compounds, evaluated by the normalization method (Table 4).

**Table 3** “Visual imprint” area of the sensor signals (S1-S8) in the equilibrium gas medium above the samples

| Samples        | S1-MCNT | S2-PEGS | S3-PEGSb | S4-PEGa | S5-PEG-2000 | S6-DCH18C6 | S7-Tween | S8-PEGP | $S_{max}$ Hz·s |
|----------------|---------|---------|----------|---------|-------------|------------|----------|---------|----------------|
| Wet grain      | 9       | 13      | 9        | 14      | 14          | 9          | 11       | 11      | 353            |
| Germinated     | 8       | 13      | 8        | 12      | 13          | 9          | 11       | 11      | 312            |
| grain          |         |         |          |         |             |            |          |         |                |
| Water          | 18      | 21      | 15       | 24      | 23          | 17         | 21       | 22      | 1135           |

MCNTs – multilayer carbon nanotubes, PEGS – polyethylene glycol succinate, PEGSb – polyethylene glycol sebacinate, PEGA – polyethylene glycol adipate, PEG-2000 – polyethylene glycol, DCH18C6 – dicyclohexane-18-Crown-6, Tween – Twin 40, PEGP – polyethylene glycolphthalate
After germination, a decrease in the intensity of aroma was noted in the universal indicator, as well as in the “O-containing”, “alcohols, ketones, amines” and “alcohols, ketones, water” indicators.

Alcohols are the most commonly found compounds in natural essential oils. As part of essential oils, they do not only add a peculiar aroma, but also contribute to the manifestation of antiseptic activity against bacterial and viral infections, have analgesic, anesthetic and tonic effects, as well as regulate hormonal activity. The absence of toxicity is very important, therefore essential oils with a predominant alcohol content are relatively safe.

**Figure 4** “Visual prints” of the maximum sensor signals in the equilibrium gas phase above the samples. The rotary axis indicates the numbers of the sensors in the array (experimental part), and the vertical – the responses of the sensors at a particular point in time (ΔFmax, Hz)

Natural essential oils with a high content of ketones can cause side effects: neurotoxicity (negatively affect the functions of the nervous system), embryotrophic effect (dangerous during pregnancy), and hepatotropic effect (disrupt liver function).

Some amines are very toxic substances. Inhalation of their fumes and contact with skin are dangerous. Aliphatic amines affect the nervous system, as well as cause violations of the permeability of the walls of blood vessels and cell membranes, liver functions and the development of dystrophy.

We found that grain samples differed in quality composition. For a more visual presentation of the results, a spectrum of identification parameters was constructed (Fig. 5). The compositions of the equilibrium gas phase can be considered identical if the spectra coincide within the error (equal to or greater than 0.1).

Germinated grain contained fewer components in quality composition than wet grain. To establish such differences, the distribution by identification parameters of their absolute differences from water was traced (Fig. 6).

Differences by more than ± 0.1 units are significant and indicate a different composition of compounds in the equilibrium gas phase above the samples. In Fig. 5 the allowable difference interval is marked with black lines.

A significant change in the qualitative composition of the sprouted grain compared to wet grain was

**Table 4** Signal ratio of several sensors in the matrix for test samples

| Samples          | S1-MCNT | S2-PEGs | S3-PEGSb | S4-PEGA | S5-PEG-2000 | S6-DCH18C6 | S7-Tween | S8-PEGP |
|------------------|---------|---------|----------|---------|-------------|------------|----------|---------|
| Wet grain        | 10      | 14.4    | 10       | 15.6    | 15.6        | 10         | 12.2     | 12.2    |
| Germinated grain | 9.4     | 15.3    | 9.4      | 14.1    | 15.3        | 10.6       | 12.9     | 12.9    |
| Water            | 11.2    | 13      | 9.3      | 14.9    | 14.3        | 10.6       | 13       | 13.7    |

MCNT – multilayer carbon nanotubes, PEGS – polyethylene glycol succinate, PEGSb – polyethylene glycol sebacinate, PEGA – polyethylene glycol adipate, PEG-2000 – polyethylene glycol, DCH18C6 – dicyclohexane-18-Crown-6, Tween – Twin, PEGP – polyethylene glycol phthalate
established. Water was also very different from both grain samples, which suggests that not only water vapor, but also other organic volatile compounds were present in the equilibrium gas phase. The qualitative composition of the EGP above wet and germinated samples differed significantly (for selected points – by 60%).

In parallel, an analysis of the sensory characteristics of the grain was carried out. A significant decrease in the sharp smell of legumes after germination was found. This makes it possible to reduce the legume smell, which is a drawback when added to food products, or when traditional raw materials are completely replaced.

CONCLUSION

Testing in laboratory and pilot production conditions showed that the use of pre-processed lentil grains would allow replacing up to 50% of raw meat in minced products (ready-to-cook food, cupats) without changing the smell of the products. Smell is easily masked by spices and food additives. The products possess juiciness and attractive appearance.

The conducted studies opened up new prospects for the creation of meat and vegetable products enriched with biologically active substances, that have the possibility of wider use of domestic raw materials and the development of import-substituting technologies for healthy nutrition products.

Germinated lentils are supposed to be used both as part of meat systems and as an independent ingredient for salads, as well as when creating products that simulate meat for fasting, or when creating enriched extruded products for people who lose weight (bread, bran, etc.).

CONTRIBUTION

L.V. Antipova – 40%, T.A. Kuchmenko – 30%, A.A. Osmachkina – 20%, N.A. Osipova – 10 %.

CONFLICT OF INTERESTS

The authors declare no conflict of interest.

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