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Qualimetric assessment as a tool of the quality and safety management system of meat products with undeclared components

Elizaveta Kryuchenko[a], Yulia Kuzlyakina[a] and Valentina Zamula[a]*

[a] *V.M. Gorbatov Federal Research Center for Food Systems, Quality Management and Conformity Assessment Department, 109316, Moscow, Russia*

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ABSTRACT

The range of food components used in food production is constantly growing. The presence of food components is determined by the recipe, but however, contamination or unscrupulous use of them is possible. The presence of undeclared food components is fraught with, in addition to falsification, the risk of harm to the health of the consumer. It is important to determine the exact content of food components in a food product. This is especially true for substances that can cause food intolerance. It is possible to detect trace amounts of food components using various methods. Regulatory authorities in many countries, realizing the seriousness of the problem, are adopting laws, regulations and standards requiring food labelling to indicate the possible presence of food components that can lead to a state of hypersensitivity, thereby ensuring their exclusion. In the Russian Federation, such requirements are contained in TR CU 022/2011. The control of food components, including allergens, is required by all voluntary standards that set requirements for food safety management systems such as BRC, IFS, ISO 22000, etc. However, the scope of control measures is much wider than just food labelling and these measures are impossible without modern methodology and analytical methods. To control the presence of trace amounts of food components in food products, various methods are used — both qualitative and quantitative. The concentration of an additive (e.g., allergen, food component) that can cause a serious health hazard can be in the micro- and nanogram levels, and approaches are constantly being developed to increase the sensitivity of allergen detection methods in food products. These are immunoanalytical, mass spectrometric, chromatographic, histological methods, methods based on nucleic acid amplification, proteomic analysis, methods using biosensors. This article discusses such methods, and their advantages and disadvantages, and presents a qualimetric assessment of these methods in order to determine the most effective one, which will provide consumers with high-quality and safe products.

1. Introduction

Food hypersensitivity is a serious problem that affects approximately 3 to 10% of adults and 8% of children worldwide (*Osborne*, 2011). Reducing the risk of adverse food hypersensitivity reactions in food consumers requires the elimination of certain substances from the diet. However, it will not be effective without clear information about the presence of undeclared food components, in particular allergens, in products. This is especially true for those components that are not included in the recipe,

***Corresponding author:** Elizaveta Kryuchenko, *l.kryuchenko@fncps.ru*

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but may potentially appear in the product in small quantities. EU labelling rules require clear labelling of 14 substances or products causing allergies or intolerances, while the US requires eight.

In the Russian Federation, the requirements for the labelling of food products containing the most common components, the use of which may cause allergic reactions or is contraindicated in certain types of diseases, are contained in Technical regulation of the Eurasian Customs Union № 022 "Food products in terms of their labelling" (TR CU 022/2011). The list of these food components is harmonized with the EU.

An analysis of publication activity for the period from 1990 to 2023 in the Scopus, PubMed and E-library databases revealed a sharp increase, approximately since 2005, in the number of scientific publications dealing with this topic. Interestingly, the e-library found 89 publications in 2023 for the keyword "food allergen" and 189 for "food intolerance". At the same time, 18840 and 15532 publications were issued by Scopus and PubMed, respectively. This indicates the lag of domestic science in this area, although since 2015 there has been a significant increase in interest in this topic, which is reflected in the growth of publication activity.

At present, worldwide there are no uniform established values for the minimum concentration of a particular food component that can cause an undesirable effect on the health of the consumer, the threshold dose. On the other hand, fast and sensitive methods for detecting food components are needed, especially when it comes to substances that are not included in the formulation of the product and contained in trace amounts. A mini-review of modern methods and approaches for the detection of food components in meat products, as well as their qualimetric assessment is presented.

2. Materials and methods

Qualimetric assessment of methods for determining food components in meat products includes several successive stages (*Bazrova*, 2015).

Table 1. List of potentially effective research methods for determining the presence of food components, including allergens, in meat products

№	Method name	The essence of the method
1.	Enzyme immunoassay (ELISA)	The method allows determination of the presence of hidden proteins in meat products that can lead to a hypersensitivity reaction.
2.	Lateral Flow Device (LFD)	The LFD is a test strip (rapid test). The method is based on an immunochromatographic approach in which proteins in a sample interact with antibodies and are conjugated simultaneously and held for a single short period.
3.	Polymerase chain reaction (PCR)	The method is characterized by amplification of DNA segments. PCR analysis can be viewed as a series of sequential steps: sampling, sample preparation, DNA extraction, DNA quantification, titration, PCR setup, equipment operation, software analysis, manual analysis.
4.	Mass spectrometry	The method allows protein identification, determines the primary amino acid sequences of the protein, identifies post-translational modifications and quantifies proteins in meat products.
5.	Histological method	The method is based on the identification of plant components of protein origin in various types of meat raw materials and finished meat products in accordance with their microstructural features using histological preparations.
6.	Liquid chromatography high-resolution mass spectrometry (LC-HRMS)	The method directly analyses the cleaved peptide fragments of the desired proteins by their differences in molecular weights.
7.	Surface plasmon resonance (SPR) biosensors	The method allows real-time detection of compounds interacting with an immobilized target molecule: an antibody against the desired protein or a single-stranded DNA molecule capable of hybridizing with a specific DNA fragment of food components.

At the first stage, a literature search of methods was carried out and for further assessment, those for which there was not found enough data on their effective use were excluded. As a result, a list of potentially effective research methods was formed that effectively and accurately determines the presence of undeclared protein-containing food components in meat products (Table 1).

Next, a qualimetric assessment was carried out to determine the current and expected (required) parameters in order to determine and predict changes in the compared characteristics (*Yankovskaya*, 2019; *Novak*, 2020).

The values of the weighting coefficients were calculated by the direct assessment method, which involves assigning points (C_{ij}) to each of the assessed indicators using scales. The average score C_i was calculated for each indicator using the formula:

$$C_i = \frac{\sum_{j=1}^{N} C_{ij}}{N}, \quad (1)$$

where C_{ij} is the score attributed to the i-th indicator by each expert;

N is the number of experts.

The weight coefficient of the i-th indicator was determined by the formula:

$$M_i = \frac{C_i}{\sum_{i=1}^{n} C_i}, \quad (2)$$

where C_i is the average score attributed to the i-th indicator; n is the number of evaluated indicators.

2. Results and discussion

Taking into account the principle of decomposition, a property tree diagram of quality indicators of the most appropriate method for determining food components in meat products was developed (Figure 1). The developed property tree made it possible to establish a range of indicators that determine the quality of methods for determining food components in meat products, build a hierarchical structure of the concept of quality of methods for determining food components in meat products, establish weight coefficients and conduct a comparative assessment (*Dunchenko*, 2019).

The histological method is defined as the most established, standardized method applicable to the study of food components, including allergens. Comprehensive studies of the structural features of vegetable protein components used in the production of meat products have been carried out in the V. M. Gorbatov Federal Research Center for Food Systems for fifteen years. The features of changes in vegetable protein components' microstructures during technological processing have been studied, and histological methods for their identification in any type of meat raw materials, semi-finished products and finished products have been developed by center experts. Based on the results of the research, the center developed the methodology GOST 31474-2012 "Meat and meat products. Histological method of identification of plant protein additives".

However, histological studies of vegetable protein preparations used in the meat industry have shown that similar technological variants of different proteins have similar microstructural identification indicators, which makes it difficult to analyse and interpret the results. So, for example, the structure and tinctorial properties of soy and wheat texturates are the same. Particles of some versions of the albumin preparation have microstructural characteristics similar to soy isolated proteins. Thus, the use of traditional histological methods for the detection of such proteins cannot always provide reliable information about their content in the composition of meat products.

ELISA remains the most widely used method for detecting food components and quantifying their presence. One reason for this is that the ELISA targets proteins in the food components that are the main causes of the hypersensitivity reaction. ELISA is relatively simple and can be performed by qualified personnel in a laboratory using relatively inexpensive equipment. The analysis can be completed within a few hours. ELISA test kits can detect the presence of gluten, lupine, soy and other protease inhibitors. There are currently no approved standards for the ELISA method for determining food components, including food allergens, in meat products (*Koeberl*, 2018).

Important advantages of ELISA include the presence of an express method (test strips). The method is well developed, has high sensitivity, which makes it possible to detect concentrations of up to 0.05 ng/ml of food components in meat products, the ability to use minimal volumes of the test material, the simplicity of the reaction, and the low

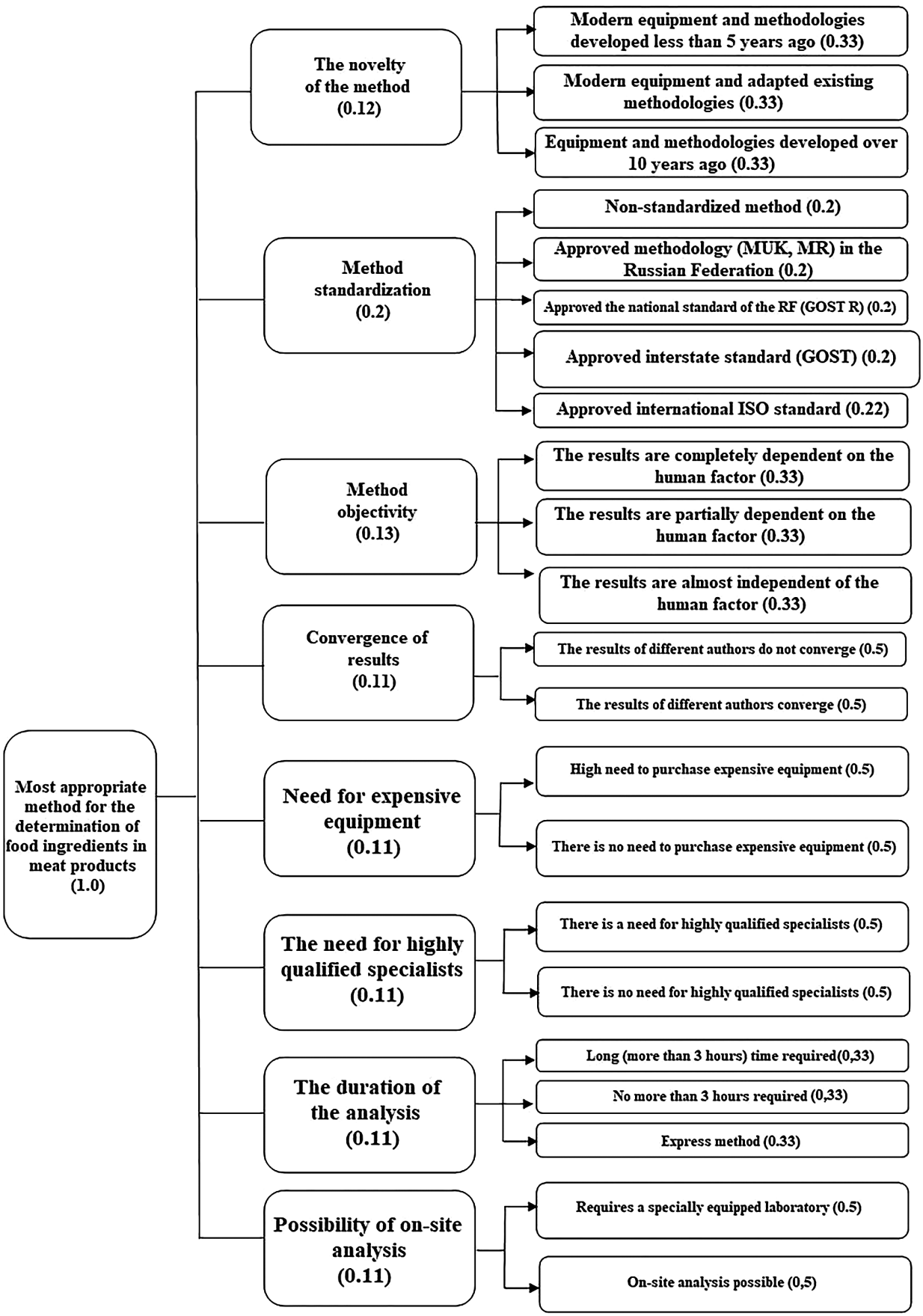

Figure 1. Property tree of quality indicators of the most appropriate method for determining food components in meat products

cost of diagnostic kits. ELISA test kits allow you to determine the target protein even in products that have undergone deep processing.

Immunohistochemical methods are currently being developed. They are highly specific and combine the advantages of traditional histological methods with the sensitivity of immunological ones.

Another method most commonly used is the polymerase chain reaction (PCR). The general principle of PCR is sequential and exponential amplification of the target DNA using a thermostable DNA polymerase. PCR can be applied to most food components containing DNA, and this method is required by law in many countries, for example, in the EU in accordance with Annex 3a of the European Directive 2003/89/EC. The specificity of the method allows, in most cases, unambiguous identification of the food component, which is especially important in the case of the presence of a protein in the product that can cause a hypersensitivity reaction. This makes the PCR method an important analytical tool. In addition, its high specificity makes it possible to check the results obtained by ELISA, which cannot be absolutely accurate due to the nature of the antibody-based ELISA method. Moreover, PCR is able to distinguish several protein targets that cannot be separated from other food components using ELISA. PCR requires a special laboratory room and trained personnel (*Cheng*, 2016).

A key advantage of the lateral flow device for detection is the rapidity and simplicity of the test, typically requiring little or no sample or reagent preparation. A huge advantage is that the method does not require expensive equipment. Also, the lateral flow device is an express method for determining the presence of food components in food products such as: lupine, soy, mustard, celery, gluten (*Dzantiev*, 2014).

The LC-HRMS/MS method is an analytical chemistry method that combines the physical separation capabilities of liquid chromatography with the mass analysis capabilities of mass spectrometry (MS). The advantage of the LC-HRMS/MS method is that it is able to detect the presence of several food components in meat products. However, the method has significant drawbacks: the need for expensive equipment, the complexity and duration of the analysis. These disadvantages complicate the method's widespread introduction into laboratory practice under production conditions at industry enterprises (*Stella*, 2020). Currently, there is an active standardization of methods based on LC-HRMS/MS.

The method of proteomic analysis, coupled with mass spectrometry, is a fairly new technology in relation to the detection of the presence of food components in products. The method requires expensive equipment and qualified personnel.

The use of SPR biosensors is a new tool for identifying food components. The use of biosensors makes it possible to identify food components in real time. Main disadvantages: expensive equipment, takes up a lot of space in the laboratory, it is difficult to simultaneously analyse several food components in one sample. At the same time, SPR biosensors have great potential for manufacturers and regulatory authorities (*Vasilescu*, 2020).

3. Conclusion

The conducted qualimetric evaluation of the main methods used to detect food components allows us to compare methods based on common characteristics.

The results of assessing the potential effectiveness of methods for determining the presence of food components in meat products allowed us to build the following sequence of effectiveness: histological method; ELISA; PCR; 2DE-MS; LFD; SPR.

The widespread use of many methods is complicated by the need to purchase expensive equipment, insufficient development of express method modifications, and the complexity of implementing most methods in production laboratories.

Considering the ever-growing range of food additives, the temptation for unscrupulous manufacturers to include unreasonable amounts of additives in formulations, the importance of in situ monitoring and control of food components, and the obligation of manufacturers to include food component control in production control programs, a qualimetric approach to choosing an appropriate method can serve as a reliable tool for the manufacturer.

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