Quality and Safety of Meat Products In Russia: Results of Monitoring Samples from Manufacturers and Evaluation of Analytical Methods

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Abstract
Violations in manufacturing and products that do not meet the declared markings are currently an acute problem in ensuring the safety and quality of food. Meat products are often falsified due to the high added value and multicomponent composition. Despite the efforts of regulatory organizations, the covert replacement of various types of meat with cheaper or low-grade compounds is widespread. Consumer societies, individuals, food quality control state institutions are interested in the results of food quality, safety and conformity monitoring. This article presents information on results of food products’ conformity and possible analytical methods used to control meat products’ composition, the results of meat product monitoring as conducted by the V. M. Gorbatov Federal National Center for Food Systems (Moscow, Russia), data on the prevalence of various types of falsification, and proposals to improve the quality and safety control of meat products in Russia. According to the national regulatory framework, which includes national and international safety standards and regulations, has a strict control over the content of a large number of components of a diverse chemical nature is needed. That leads to the development of analytical methods and devices that can reliably evaluate components in food products added even in micro amounts. A direct relationship between the introduction of a new, more accurate method for identifying a product’s components and the reduction in cases of the corresponding fraud has been detected.

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Introduction

In modern society, the quantity and quality of consumed products has a significant impact on the health of the population. In this regard, reliable information on the sources of raw materials for the production of meat products is of fundamental importance, because many technologies allow the replacement of raw material (for example, muscle tissue) with protein-containing additives of various animal or vegetable origins. Moreover, cases of falsification of meat products using animal species not encompassed by the technology are becoming widespread.\(^1\)

The counterfeiting of products is divided into several categories:

- qualitative falsification by the introduction of unlabeled additives or changes in the species composition;
- quantitative falsification by the violation of the ratio of the components of in the formulation;
- assortment falsification by the substitution of popular classic meat products with other products derived from cheaper raw materials; and
- information falsification by inaccurate or distorted information about the product.

The falsification of raw materials by changing the species composition not only changes the properties of the end products but also poses a danger to consumers' health. The greatest risks are associated with substitutions of raw materials with animal meat, the use of which is prohibited or limited in connection with the possibility of its infection by prions or viruses. In some cases, the use of undeclared components (e.g., meat and milk proteins, soy, mustard) can cause allergic reactions, a risk of which the consumer remains uninformed. In addition, falsification of raw materials can violate the moral code of consumers whose national or religious views do not allow the consumption of meat from certain species of livestock and poultry.

To ensure safe and healthy food, joint efforts are required. Therefore, cooperation is intensifying among sectorial agricultural institutes to create a unified monitoring system. Obviously, for the smooth functioning of all units of an integrated trophological chain, coordinated actions by specialists in various fields are necessary. In this regard, the tasks of the food and processing industries are intertwined with the tasks of health care.\(^2\)

At the V. M. Gorbatov Federal National Center for Food Systems (Moscow, Russia), a concept to guarantee the comprehensive quality and safety of meat products has been developed, which includes the use of barrier technologies, predictive microbiology, critical control points, production management principles, monitoring of the safety and quality of the product throughout the chain of production, transportation, and sale.\(^3\)

The aims of the study are: (1) monitor food products' conformity; (2) identify the most falsified foods; (3) present a unified monitoring system for food nonconformity assessment.

Variety of Analytical Methods

At all stages of production, it is crucial to control the composition of meat products (i.e., raw materials, intermediate products, and finished products). To ensure the effectiveness of such control, reliable and productive analytical methods are needed that will allow researchers to detect individual ingredients and the molecular identifiers reflecting the content of different types of raw materials in the finished product.\(^4\) Multistage technological treatments (i.e., fine grinding, salting, heat treatment, formulation) may make it difficult to identify the structure of muscle tissue, as recommended by traditional methods. But methods have been developed in scientific research to solve such problems.

To determine the composition of food products, methods of electrophoresis and immunodiffusion have been tested, but their long duration seriously limits widespread application.\(^5\) Chromatographic methods with high sensitivity, accuracy, and speed are highly promising;\(^6\) however, they require expensive equipment and are currently not well adapted for use with complex multicomponent substances such as meat products.

To establish the species of native (not processed and not subjected to technological impact) raw materials, enzyme methods can also be used that establish differences in the enzyme spectra among
various animal species. However, these methods are unsuitable for products that have undergone heat treatment and are unable to determine the quantitative characteristics of the composition of meat products.

A variety of analytical reactions (for example, for glycogen), such as spectral control of certain compounds cannot be considered acceptable due to the large number of false positive and false negative results.

Molecular genetic methods of analysis, despite their significant progress, can still only be considered as confirmatory methods implemented in specialized laboratories for samples selected according to the results of preliminary screening.

Enzyme-linked immunosorbent assay (ELISA), which is highly sensitive, is also used for these purposes. This method is most convenient for establishing the species of meat and determining the presence and amount of additives of vegetable proteins such as soy. An important issue that determines the applicability of immunodetection in food control is the stability of detected biomarkers after enzymatic and heat treatment of manufactured foodstuffs. We have shown the advantages of troponins – thermostable biomarkers of muscle tissues – for meat authentication. An ELISA of troponin I was developed and characterized. It was found that this ELISA allowed researchers to distinguish between mammalian (beef, pork, lamb, horse) and bird (chicken, turkey, duck) meat sources.

The multiplex monitoring of specific peptides by mass spectrometry was considered for the confirmatory level of meat products’ control, based on multiple responses monitoring (MRM-MS). Proteins from the meat matrix were extracted and digested with trypsin, and the peptide mixtures were analyzed by high-performance liquid chromatography with QQQ mass spectrometric detection. Candidate proteins were characterized by using modeling in the Skyline program. To select biomarkers, peptides were considered whose length exceeded six amino acids. The choice of species-specific protein and its marker amino acids was based on the following factors: high content in muscle tissues, good signal-to-noise ratio in meat samples, high specificity, the absence of missing cleavages, and cleavage sites specific to trypsin at both ends. The main proteins selected to determine the authenticity of meat products were myoglobin and lactate dehydrogenase.

Priorities for Monitoring
Nutrition is the most important factor ensuring the maintenance of the population’s health, working capacity, and creative potential. The relative cheapness of vegetable protein components compared to raw materials of animal origin has led to the active introduction of vegetable matter, primarily soy protein, in the formulation of meat products. This trend is not in itself unfavorable, but consumers have the right to know about the presence of additives.

Because the cost of soy protein is almost half that of animal protein, the introduction of soy in meat products has recently become extremely common. According to the Test Center of the V. M. Gorbatov Federal National Center for Food Systems, more than two thirds of the products received for certification from manufacturers contain unmarked soybean inclusions. In addition to the economic reasons for the falsification of meat products, it is necessary to take into account the fact that almost all soy is a transgenic (genetically modified [GM]) product. Until recently, Russia allowed the use of only three varieties of GM soybean and 15 GM varieties (corn, potatoes, sugar beets, rice, microorganisms) in the finished products. The usual addition of soy protein with meat products technology is 2–10%.

In connection with the increasing requirements for the quality and safety of meat products, the government actively identifies the raw materials and finished products to determine freshness, leaneness and grade of beef and pork trimming; density and melting point of bacon; the content of bone residue (mainly in mechanically deboned poultry meat); the mass fraction of moisture, fat, protein, salt, nitrite in the finished product; the content of plant proteins and other components.

Results of Meat Products Monitoring
Monitoring often reveals violations of the qualitative composition of products (Fig. 1). Over 20% of the total number of monitored Russian meat processing plants generates products that contain a large number of unacceptable additives of one or several
items at the same time. Of all these enterprises, 30% produce cooked sausage products, which include two types of unacceptable vegetable additives; 32% use one type of additives; and only 15% do not use unauthorized additives at all.\textsuperscript{13, 14}

In 2018, the V. M. Gorbatov Federal National Center for Food Systems Test Center of the checked over 10,000 samples of cooked products and smoked meats, as well as 12,500 samples for 8 months of 2019. Consumer requirement to assess food conformity with histological method has increased greatly due to reliable and comparable results.

Table 1: The share of identified discrepancies in food products of the total number of tests for the period 2018–2019 in comparison with 2015 (data of the Test Center of the V. M. Gorbatov Federal National Center for Food Systems)

| Percentage of inconsistencies (to the total number of samples studied) | 2015 | 2018  | 2019  |
|---------------------------|------|-------|-------|
| According to microbiological indicators |      |       |       |
| Meat and meat products    | 16%  | 28%   | 27%   |
| Fish, fish products, and other seafood | 2%   | 5%    | 6%    |
| Milk and dairy products   | 9.5% | 15%   | 13%   |
| Bakery and confectionery products | -    | 17.2% | 12%   |
| Ready products of public food service | 9%   | 10%   | 11%   |
| According to physicochemical indicators |      |       |       |
| Meat and meat products    | 25%  | 23.3% | 24%   |
| Fish, fish products, and other seafood | -    | -     | -     |
| Frozen fish, fish products, and other seafood (glaze) | 2.5% | 1.0%  | 3.0%  |
| Milk and dairy products   | 5%   | 9%    | 7%    |
| Bakery and confectionery products | 2.5% | 1.0%  | -     |
Due to the fact that meat products are multi-component matrices, it was interesting to study the presence of undeclared components in products other than meat-containing ones.

As can be seen from Table 1, the most effective were results of histological studies. The percentage of identified nonconformities increased from 45% to 49% for 2018–19. Because the samples often come in anonymized form, we can assume many counterfeit products. The following unlabeled components were revealed more often than others collagen and animal proteins in sausages, carrageenans, starches, gums, and other additives that are not approved for use. Many samples with signs that meat had been treated with various solutions (injection) were revealed. Extensive studies of canned products were also carried out, during which the most found component not declared in the regulatory documentation was starch. Furthermore, many deviations were noted in the species identification of the raw material composition: the use of poultry meat, soybean, and so on. According to microbiological testing, the most frequently detected indicators were the following: a number of mesophilic, aerobic, and facultative anaerobic microorganisms, coliform bacteria, *Listeria monocytogenes*, and *Salmonella*. Most often, an excess of the total microbial number was detected in cooked sausage products, chilled and frozen meat, and chicken semifinished products. In the study of microbiological safety indicators of canned food products (industrial sterility) for 2018, 26 cases of noncompliance with the declared requirements were identified. Inconsistencies in physicochemical parameters were most often expressed in the following deviations: an underestimated amount of protein (both in cooked sausages produced in accordance with GOST and in canned products), an increased content of salt, and an increased fat content. According to the fatty acid composition, 19.6% of the tested dairy products did not correspond to official regulations, and the presence of phytosterols was established in 6.2%. The dynamics of identifying falsifications are interesting. In 2015, there were 3 times more inconsistencies in fatty acids in milk and dairy

| Component Composition (Histological Method) |
|---------------------------------------------|
| Meat and meat products                      |
|                                            |
| Fish                                        |

| According to fatty acid composition          |
|---------------------------------------------|
| Dairy products                              |
|                                            |
|                                            |

| According to the content of phytosterols     |
|---------------------------------------------|
| Dairy products                              |
|                                            |
|                                            |

| According to toxicological indicators (toxic elements, antibiotics, pesticides) |
|--------------------------------------------------------------------------------|
| Meat and meat products | 0.12% | 0.6% | - |
| Non-fish fishing objects | 0.2% | 0.1% | - |

| According to the presence of GM organisms   |
|---------------------------------------------|
| Plant protein isolates, flour, including soybean |
|                                            |
|                                            |

| According to the component composition (PCR) |
|---------------------------------------------|
| Meat and meat products | 18% | 11.4% | 10% |
| Fish                        | 13% | 14% | 22% |
products than in 2019. This is explained by the introduction of a method for detecting phytosterols, which reduced the number of false positive results, as well as the active work of regulatory and public organizations and the media reporting on falsifications of milk fat.

**Conclusion**

It should be noted that there is a direct relationship between the introduction of a new, more accurate method for identifying a product's components and the reduction in cases of the corresponding fraud. The replacement of animal protein with soy and the falsification of milk fat are good examples of this relationship. Food product certification is not obligatory any more in Russian Federation. Despite that food quality and safety aspects are of increasing interest in this country. The lack of careful control means that manufacturers, while maintaining acceptable levels of regulated substances, are able to use unnecessary plant components and overestimate their amount. It is not possible to confirm such a replacement in a routine testing and requires novel methods and tools. However, in practice, when determining the quality of a meat product, it is often necessary not only to establish the type and grade of a product but also to identify its composition. The purpose of such identification is to confirm the authenticity of a particular product as well as to comply with requirements or information about it indicated on the labeling and/or in the accompanying documents. According to the national regulatory framework, which includes national and international safety standards and regulations, a strict control over the content of a large number of components of a diverse chemical nature is needed. It leads to the development of analytical methods and devices that can reliably evaluate components in food products added even in micro amounts.

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**Conflict of Interest**

There are no conflicts to declare.

**References**

1. Ivankin A. N., Kulikovsky A. V., Vostrikova N. L., Chernukha I. M. Cis-, trans-conformational changes of bacterial fatty acids in comparison with analogues of animal and vegetable origin. *Applied Biochemistry and Microbiology*. 2014; 50(6): 604-610.
2. Vostrikova N. L., Ivankin A. N., Gorbunova N. A. Vegetable proteins-falsification of meat products and its identification. Ways of intensification of production and processing of agricultural products in modern conditions. *Materials of the International Scientific-Practical Conference*. Part 2. Volgograd; 2012: 23-24.
3. Kalinova Yu., Chernukha I., Ilyina T., Orlova O. Creation of a system to ensure the safety and quality of meat products in Russia. *Meat Technologies*. 2007; 3: 6-10.
4. Sarbacane S. T., Aubekerova L. S., Kurmanov B. K., Minaev Yu. M. Development of primers for identification of chicken DNA in meat products. In the collection: *Science, Education, Society: Problems and Prospects of Development*. 2014; 108-110.
5. Hopkins D. L. Chapter 12: The Eating Quality of Meat: II–Tenderness. *Lawrie’s Meat Science*. 2017; 357-381.
6. Wu Q., Yuan H., Zhang L., Zhang Y. Recent advances on multidimensional liquid chromatography mass-spectrometry for proteomics: from qualitative to quantitative analysis-a review. *Analytica Chimica Acta*. 2012; 731: 1–10.
7. Yurchenko S., Sats A., Tatar V., Kaart T., Jõudu I. Fatty acid profile of milk from Saanen and Swedish Landrace goats. *Food Chemistry*. 2018; 254: 326-332.
8. Astapova M. S. Comparative evaluation of
methods of identification of raw materials of animal and plant origin and food products. *Russian Journal of Veterinary Sanitation, Hygiene and Ecology*. 2019; 1: 90–99.

9. Galkin A. V., Trepalina E. Determination of the species of raw materials used in the meat processing industry. *Meat Series*. 2018; 1(71): 50-53.

10. Zvereva E. A., Kovalev L. I., Ivanov A. V., Kovaleva M. A., Zherdev A. V., Shishkin S. S., Lisitsyn A. B., Chernukha I. M., Dzantiev B. B. Enzyme immunoassay and proteomic characterization of troponin I as a marker of mammalian muscle compounds in raw meat and some meat products. *Meat Science*. 2015; 105: 46-52.

11. Vostrikova N. L., Chernukha I. M., Khvostov D. V. Methodological aspects of identification of tissue-specific proteins and peptides forming correcting properties of meat products. *Theory and Practice of Meat Processing*. 2018; 3(3): 36-55.

12. Vostrikova N. L., Ivankin A. N., Gorbunova N. A. Vegetable proteins-falsification of meat products and its detection. *Processing of Agricultural Raw Materials and Food Products (Moscow)*, 2012; 2: 23-25.

13. Pchelkina V. A. Histological methods of detection of plant proteins-allergens in meat products. *All about Meat (Moscow)*. 2016; 1: 50-53.

14. Pchelkina V. A. Development of immunohistochemical method for detection of soy proteins in meat products. *All about Meat (Moscow)*. 2016; 3: 20-23.