Control of the stability of the results of studies of cadmium content using the method of additions in cow's milk samples

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Abstract. The relevance of the problem under consideration is due to the need to use hardware to cover a wide range in the determination of toxic elements (heavy metals) in food. The primary and important task is to ensure control of the quantitative content of cadmium, the introduction and application of fast and reliable methods of their research. The analysis of the data obtained shows that all the results on the study of milk for the content of cadmium, obtained by stripping voltammetry and atomic absorption spectrometry by assessing the precision and operational control of the error using the method of additions, are satisfactory. The implementation of the methods of stripping voltammetry and atomic absorption spectrometry achieves the best precision of research results in the testing laboratory, both under repeatability conditions and under conditions of intermediate precision. The results obtained indicate that in the range of less than 0.01 mg of cadmium per 1 kg of cow's milk, prevail (42.2\%) over other ranges (during monitoring). The least registered sample results are (1.4\%) with the range of obtained data - 0.01-0.05 mg/kg.

1. Introduction
In the foreign and domestic literature, the issue of the influence of nutrition on human health has been widely studied [1-5], information on contaminants, their types and effects on the body as a whole, routes of intake of toxicants and preventive measures to reduce the level of food contamination with toxic substances is presented in detail, as well as technological methods for reducing the residual amounts of contaminants in food products [6-14]. Ensuring testing of food products and food raw materials at the highest level of compliance with modern international quality standards is a component that determines the competitiveness of laboratories. The processes of updating the laboratory base in modern conditions of the development of scientific and technological progress are objectively necessary [14-20].

Analytical laboratories that monitor safety indicators for the purposes of state supervision and certification are equipped with equipment for photometric, atomic absorption, chromatographic...
The development of effective algorithms for optimizing the laboratory base of equipment, as well as the search for a solution that will allow laboratories to provide reliable test results at minimal cost in the shortest possible time is an important and urgent task [21-28]. The relevance of the problem under consideration is due to the need for hardware to cover a wide range in the determination of toxic elements. The primary and important task is to ensure control of the quantitative content of toxic elements, the introduction and application of fast and reliable methods of their analysis [29–35]. Implementation of quality management systems is of great importance [36, 37].

This paper considers such a toxic element as cadmium, which is mandatory for control in food products, in accordance with the requirements of the Technical Regulations of the Customs Union 021/2011 (TR CU 021) "On food safety", as amended on August 8, 2019 (figure 1).

![Figure 1. Permissible levels of cadmium in milk and dairy products according to the requirements of technical regulations, mg/kg, not more.](image)

Cadmium is classified as a toxicant of the highest hazard group; it is a highly toxic cumulative poison with a broad spectrum of action.

### 2. Material and methods
The development and approbation in the laboratory of modified test methods of the conformity confirmation method is an important and urgent task.

Conducting comparative tests with these methods in order to ensure control of cadmium in food products, analyzing the accuracy, precision, repeatability of the results obtained makes it possible to judge the effectiveness of the application of these methods, and also allows a comparative analysis of the results obtained. An analysis was carried out in order to optimize the methods used and to develop...
the most economical and effective option for optimizing the equipment used in the laboratory while maintaining the metrological characteristics of the accuracy of the results obtained.

The methodological basis is the standards for research methods, test methods, operating manuals and instructions for the use of equipment.

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Quality control of the analysis results during the implementation of the method in the laboratory provides for the control of the stability of the analysis results. The object of research is cow's milk samples.

Equipment for research of cadmium content in food raw materials and food products is shown in figure 2.

![Research Instrument Base](image)

**Figure 2.** Research Instrument Base.

In the course of the research, the standard deviation (RSD) of the intralaboratory repeatability of $S_r$ and the intermediate precision $S_{IR}$ of atomic absorption analysis and stripping voltammetry were evaluated.

For research purposes, standard samples of the composition of a solution of cadmium ions were used (figure 3).

![Characteristics of a standard sample composition of the solution of cadmium ions](image)

**Figure 3.** Characteristics of a standard sample composition of the solution of cadmium ions.

**Table 1.** Analytical lines of cadmium for atomic absorption spectrometry (determination of lead).

| Analytical line wavelengths, nm | Relative ratio weakening of sensitivity |
|--------------------------------|----------------------------------------|
| 228.80                        | 1.0                                    |
| 326.11                        | 410                                    |

Table 2 shows the distinctive features of voltammetric methods of analysis using the analyzer "TA-4".
Table 2. The range of determined cadmium content on the analyzer "TA-4".

| Determined content range, mg/kg | Indicator electrode               |
|--------------------------------|-----------------------------------|
| from 0.0015 to 1.0 inclusive   | Mercury film on a silver backing  |
| from 0.0015 to 1.0 inclusive   | Silver modified                   |

The repeatability and reproducibility of the results of measurements of cadmium in milk are presented in Table 3.

Table 3. Convergence and reproducibility of cadmium measurements.

| Mass fraction of cadmium in the milk of milk, m⁻¹ | Convergence r, m⁻¹ | Relative standard deviation of convergence 100S/m | Reproducibility R, m⁻¹ | Relative standard deviation of reproducibility 100S/m |
|-------------------------------------------------|-------------------|-----------------------------------------------|----------------------|-----------------------------------------------|
| 0.01                                            | 0.0034            | 12                                            | 0.0034               | 40                                            |
| 0.1                                             | 0.017             | 6                                             | 0.017                | 20                                            |
| 0.5                                             | 0.055             | 4                                             | 0.055                | 12                                            |
| 1.0                                             | 0.090             | 3                                             | 0.090                | 9                                             |

The main document of the research procedure is the test facility quality manual.

The control by the method of additions during the implementation of various methods in this work was carried out according to the approved research scheme.

During the operational control of the analysis procedure using the control procedure to control the error using the additive method, the control means were working samples of a stable composition and the same samples with a known addition of cadmium.

Under the conditions of intra-laboratory precision, the analysis of samples with and without added cadmium was carried out.

3. Results and discussion

Atomic absorption spectrometry is widely used in the determination of heavy metals (toxic elements) in various branches of science and industry.

Voltammetric analyzers, as a rule, with their own electrodes, software and methodological support. The voltammetric analyzer can have one electrochemical cell or several. Examples of voltammetric analyzers: with one electrochemical cell:

- "ABS-1.1",
- "IVA-5",
- "Ecotest-VA",
- "AVA 3",
- "AKV-07MK",
- "Pan-arsenic",
- "PU-1";

With three electrochemical cells:

- "TA-4",
- "TA-07"
- others.

The maximum levels of cadmium according to the national standard of the People's Republic of China GB 2762-2012 meet the requirements of the Codex Alimentarius standards, unless there is no Codex standard. The maximum levels of cadmium in accordance with the requirements of TR CU 021 comply with, and for some types of products are more stringent, the requirements of the Codex Alimentarius standards, except in cases where there is no Codex standard. In comparison with the maximum permissible concentration (MPC) of cadmium in food products regulated by the requirements
of TR CU 021, the maximum levels of cadmium in accordance with the national standard of China GB 2762-2012 meet or below the requirements of CU TR 021, i.e. MPCs for cadmium for some types of products were stricter in TR CU 021.

It should be noted changes in the national standard of the People’s Republic of China GB 2762-2012 in the direction of expanding the range of product groups for which standard cadmium indicators have been established, compared to GB 2762-2005. In the national standard GB 2762-2005, only maximum levels of cadmium were presented for the following types of products: cereals (rice, soybeans, peanuts, flour, corn, millet, sorghum), potatoes; animal meat, liver and kidneys of animals; fruit; stem vegetables (other than celery); leafy vegetables, celery, edible mushrooms; other vegetables; a fish; fresh egg.

3.1. Control of intermediate precision

In terms of repeatability and intermediate precision, five average measurement results were obtained for bovine milk samples.

The acceptability of the determination results was assessed in accordance with GOST R ISO 5725-6 "Accuracy (correctness and precision) of methods and measurement results". The range between the maximum and minimum values of all five analysis results \((X_{\text{max}} - X_{\text{min}})\) was compared with the absolute value of the critical range for five analysis results \(C_{R0.95}(5)\).

The critical range factor \(f(n)\) for the five results is 3.9.

The analyzer is controlled by pressing the control buttons displayed on the display of the test equipment.

The permissible discrepancy between two parallel results obtained in the same laboratory in one series of measurements (the convergence \(r\)) depends on the mass fraction of cadmium in milk and, at a confidence level of \(P=0.95\), does not exceed the established values of the regulatory documents.

The analyzer is controlled by the software of the test process at all stages of measurements. The widespread use of computer software significantly expands the capabilities of the devices, allows more reliable separation of the analytical signal, reduce the measurement error to 2-5%, automate statistical processing and calculation of analysis results.

For the Kvant-2AT spectrometer, when determining cadmium = 46.8%.

For the analyzer "TA-4" when determining cadmium = 50.7%.

The analysis of the obtained research results presented in table 4 showed that the condition \((X_{\text{max}} - X_{\text{min}}) \leq C_{R0.95}(5)\) is fulfilled for all measurement results.

Table 4. Results of studies of milk for cadmium content (standardized level of permissible concentration 0.03 mg/kg).

| №  | TA-4 | Kvant-2AT | TA-4 | Kvant-2AT | TA-4 | Kvant-2AT |
|----|------|----------|------|----------|------|----------|
|    | \(C_D = 0.05\) sample preparation stage | \(C_D = 0.05\) measurement stage | \(C_D = 0.01\) stage sample preparation | \(C_D = 0.01\) measurement stage |
| 1  | 0.0091 | 0.0087 | 0.0168 | 0.0161 | 0.0169 | 0.0169 |
| 2  | 0.0077 | 0.0085 | 0.0164 | 0.0166 | 0.0174 | 0.0174 |
| 3  | 0.0084 | 0.0075 | 0.0163 | 0.0169 | 0.0171 | 0.0173 |
| 4  | 0.0086 | 0.0076 | 0.0171 | 0.0163 | 0.0176 | 0.0177 |
| 5  | 0.0088 | 0.0092 | 0.0161 | 0.0171 | 0.0177 | 0.0181 |
| X_{\text{avr}} | 0.00852 | 0.0083 | 0.01654 | 0.0166 | 0.01734 | 0.01748 |

Assessing the precision of results \((X_{\text{max}} - X_{\text{min}}) \leq C_{R0.95}(5)\)

| 0.0014<0.0004 | 0.0016<0.0004 | 0.001<0.0008 | 0.001<0.0008 | 0.002<0.0008 | 0.002<0.0008 |

3.2. Monitoring the stability of analysis results using the addition method

Operational control of the analysis procedure was carried out by the performer by comparing the result of a separate control procedure \(K_c\) with the calculated control standard \(K\) (figure 4).
Figure 4. Operational control of the analysis procedure.

In accordance with the analysis methods, the results of control measurements of the cadmium concentration in the averaged working sample of milk - $X_{(n)}$ and in the averaged working sample with a known cadmium addition – $X_{(n)+d}$.

The results of the operational control of the analysis procedure using the control procedure to control the error using the addition method are presented in table 5.

Table 5. The results of the operational control of the analysis procedure using the method of additions (examination of samples for the content of cadmium).

| №  | Stage addition | Sample preparation | measurements |
|----|----------------|--------------------|--------------|
|    |                | TA-4               | Kvant-2AT    | TA-4           | Kvant-2AT    |
|    |                | $K_a$ K            | $K_a$ K      | $K_a$ K        | $K_a$ K      |
| 1  |                | -0.0092 0.01111    | -0.0064 0.01051 | -0.006 0.01142 | -0.005 0.010631 |
| 2  |                | -0.007 0.00747    | -0.007 0.00715 | 0.0046 0.008676 | 0.0036 0.008162 |
| 3  |                | -0.002 0.00203    | -0.0017 0.00187 | -0.00118 0.00211 | -0.00082 0.001951 |
| 4  |                | -0.0082 0.00878   | -0.0062 0.00837 | -0.007 0.008904 | -0.0036 0.008615 |

Analyzing the obtained research results presented in table 5, we came to the conclusion that the condition $|K_a| \leq K$ is fulfilled, the analysis procedure is recognized as satisfactory.

In order to control the stability of the analysis results obtained by different methods, operational control of the error was carried out using the method of additions.

Operational control of the analysis procedure was carried out by comparing the result of a separate control procedure $K_a$ with the calculated control standard $K$.

The results of the operational control of the milk test procedure using the control procedure to control the error using the addition method are presented in table 6.

Table 6. Results of studies of milk for cadmium content (standardized level of permissible concentration 0.05 mg/kg).

|      | Kvant-2AT | TA-4 | Kvant-2AT | TA-4 | Kvant-2AT | TA-4 |
|------|-----------|------|-----------|------|-----------|------|
|      | addition at the stage of sample preparation $c = 0.5$ | additive at the stage of measurement with $= 0.5$ |
|      | additive at the stage of sample preparation $c = 0.03$ | additive at the stage of measurement with $= 0.03$ |

| $X_{(5)}$ ave | 0.00852 | 0.0083 | 0.01654 | 0.0166 | 0.01734 | 0.01748 |
|--------------|--------|-------|--------|-------|--------|--------|
| $X_{ave}$    | 0.00841 | 0.01657 | 0.0166 | 0.01734 | 0.01741 |

**Evaluation of the precision of results obtained by different methods**

- $1.3 \% < 17 \%$
- $0.18 \% < 17 \%$
- $0.4 \% < 17 \%$

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|      | Kvant-2AT | TA-4 | Kvant-2AT | TA-4 | Kvant-2AT | TA-4 |
|------|-----------|------|-----------|------|-----------|------|
|      | addition at the stage of sample preparation $c = 0.5$ | additive at the stage of measurement with $= 0.5$ |
|      | additive at the stage of sample preparation $c = 0.03$ | additive at the stage of measurement with $= 0.03$ |

| $X_{(5)}$ ave | 0.00852 | 0.0083 | 0.01654 | 0.0166 | 0.01734 | 0.01748 |
|--------------|--------|-------|--------|-------|--------|--------|
| $X_{ave}$    | 0.00841 | 0.01657 | 0.0166 | 0.01734 | 0.01741 |

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- $1.3 \% < 17 \%$
- $0.18 \% < 17 \%$
- $0.4 \% < 17 \%$
The research results (table 6) are considered satisfactory.

3.3. Research results monitoring
We monitored the results of studies on the content of cadmium in cow's milk from January 2019 to October 2020. We evaluated the distribution of research results depending on the concentration of cadmium in cow's milk (figure 5).

![Figure 5. Distribution of test results depending on the concentration (mg/kg) of cadmium in cow's milk samples.]

The obtained monitoring results indicate that in the range of less than 0.01 mg of cadmium per 1 kg of cow's milk, prevail (42.2%) over other ranges. The least registered sample results (1.4%) with the range of obtained data - 0.01-0.05 mg/kg.

4. Conclusion
The analysis of the data obtained shows that all the results on the study of the cadmium content in the cooled yolk by the assessment of precision and on-line control of the error using the method of additions are satisfactory.

By implementing the methods of stripping voltammetry and atomic absorption spectrometry, the best precision of the results of studies of cadmium in the testing laboratory is achieved, both under conditions of intermediate precision and under conditions of repeatability.

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