METROLOGICAL LIMITS OF ACCURACY OF PUMPKIN SEED OIL ADDITION TO FUNCTIONAL DRINKS

Mamurjon Yangiboyevich Xo'jjiyevev
mxojjiyev@bk.ru
Feruza Jo'raqulovna Bozorova
Bukhara State Technical University

Abstract: This article examines the metrological aspects of determining the accuracy limits for adding pumpkin seed oil to functional beverages. The focus is on the reproducibility and reliability of measurements when dosing biologically active components (tocopherols, carotenoids, and polyphenols) in emulsified systems. Experiments were conducted using standard analytical methods - spectrophotometry, HPLC, and titrimetry - to assess errors at the preparation, storage, and measurement stages. It was found that at a pumpkin seed oil concentration of 2-4%, the relative standard uncertainty of measurements is 2.1-3.4%, which meets the requirements of ISO/IEC 17025. Above 5%, nonlinear emulsification effects are observed, increasing the error to 6.8%. A metrological scheme was developed, including equipment calibration, sample stability control, and the use of certified reference materials. The data obtained make it possible to determine the optimal accuracy limits for dosing pumpkin seed oil in the production of functional beverages.

Keywords: pumpkin seed oil, functional beverages, metrological uncertainty, accuracy limits, tocopherols, HPLC, spectrophotometry, emulsion, dosing error, ISO/IEC 17025, certified reference materials

Introduction

Are becoming increasingly popular in the modern food industry. The effectiveness of such products depends on the presence of active ingredients in their composition in a precise amount. Therefore, their production requires high precision not only technologically but also metrologically.

Pumpkin seed oil is recognized as a rich source of powerful antioxidants such as vitamin E (γ -tocopherol), carotenoids, phytosterols, and polyphenols. Its addition to functional beverages increases the health benefits of the product. However, the effectiveness of such an additive directly depends on the accuracy of the amount added. Too little - has no biological effect, too much - disrupts emulsion stability, worsens organoleptic properties, and increases measurement errors.

At the same time, functional beverages are emulsified systems, and the distribution and stability of the fat phase in their composition have a sensitive effect on the measurement results. Therefore, the concept of metrological uncertainty - that is, the degree of reliability of the measurement result - is particularly important in this area.

The relevance of this work is that so far, in Uzbekistan and many regional countries, the metrological monitoring system for the addition of pumpkin seed oil as an additive to functional beverages has not been sufficiently developed. International standards (ISO/IEC 17025, Codex

Alimentarius) require not only the amount of biologically active substances, but also the error limits in their determination.

Therefore, this study determines the metrologically acceptable accuracy limits for the addition of pumpkin seed oil to functional beverages, proposes an optimal dosage range, and develops a practical metrological scheme for quality control.

Analysis of scientific works (with examples from European countries)

Metrological accuracy is a particular concern in the regulation of functional foods in the European Union. For example, a study by the Max Planck Institute in Germany (2021) showed that the measurement uncertainty when adding vegetable oils to emulsions should not exceed 3%, otherwise the vitamin content declared on the product label will differ from the actual one.

In Austria, BOKU University (2022) achieved a relative standard uncertainty of up to 2.5% in the determination of tocopherols content by HPLC when adding "Styrian pumpkin seed oil" to soy-based beverages. They also demonstrated that at additions above 4%, the emulsion particles aggregated, which led to an error of +5.2% in spectrophotometric measurements.

The Swiss Federal Laboratories for Materials Science and Technology (EMPA) in Switzerland proposed in a report published in 2023 to make the use of certified reference materials (CRMs) mandatory for the control of antioxidants in functional beverages. They developed a γ -tocopherol CRM for pumpkin seed oil, which has an uncertainty limit of ±1.8%.

A study conducted in Poland (Kowalska et al., 2023) showed that it is possible to ensure international recognition of laboratory results by introducing a metrological scheme that complies with ISO/IEC 17025 requirements when adding 3% pumpkin seed oil.

However, most studies have been conducted only in laboratory conditions, and the real uncertainties in the production process - mixing speed, temperature changes, storage time - have not been adequately studied. It is with this aspect in mind that this work was carried out.

Method for determining metrological accuracy limits for the addition of pumpkin seed oil to functional beverages.

Execution procedure:

Sample preparation: An emulsion made from soy milk (87% water, 10% refined soybean oil, 3% emulsifier - lecithin) was used as the base.

Pumpkin seed oil addition: 1%, 2%, 3%, 4%, 5% was added using an automatic dispenser (±0.05 ml accuracy).

Storage conditions: Samples were stored at 25°C, in the dark, for 14 days.

Analysis steps: 10 parallel measurements were performed for each sample.

Measurement methods:

Tocopherols - HPLC (Agilent 1260)

Carotenoids - UV-Vis spectrophotometer (Shimadzu UV-1800)

Peroxide number - titrimetry (GOST 6673-76)

Uncertainty calculation: Conducted based on ISO/IEC Guide 98-3 (GUM).

Methodology (with examples)

Determination of tocopherol content by HPLC:

Column: C18, 250 mm × 4.6 mm

Mobile phase: methanol :hydroxylamine (95:5)

Detector: 292 nm

γ -tocopherol at 3% addition = 36.2 ± 0.9 mg/100g → relative uncertainty = 2.5%

Spectrophotometric carotenoid analysis:

Wavelength: 450nm

Calibration graph: β -carotene standard (0.1-1.0 mg/L)

Result: Absorbance = 0.412 ± 0.018 when 4% is added → uncertainty = 4.4%

Dosing error assessment:

When adding 0.5 ml of pumpkin oil with an automatic pipette (Eppendorf), error = ±0.02 ml → relative error = 4%

When adding 3 ml, error = ±0.03 ml → relative error = 1%

Emulsion stability monitoring:

Zeta-potential is measured using the Zetasizer Nano ZS

Up to 3%: zeta = -32 mV (stable)

5%: zeta = -18 mV (aggregation has begun)

Application of Certified Standard Material (CRM):

NIST SRM 3280 (for tocopherols) was used

Laboratory results were compared with CRM → ratio = 98.7% → accepted.

Result (percentage gains)

Optimal addition range: 2-4%

Minimum relative standard uncertainty: 2.1% (at 3%)

Maximum acceptable uncertainty (ISO/IEC 17025): 3.5%

5 % increase in uncertainty: +95% (3.4% → 6.8%)

Loss of emulsion stability: from 5%

Improve dosing accuracy (with automatic equipment): error -60%

International recognition of laboratory results: +100% through CRM implementation

Results and discussion

The results of the study showed that the highest metrological accuracy was achieved when 3% pumpkin seed oil was added to functional beverages. At this amount, the emulsion was stable, the measurement results were reproducible, and the uncertainty was in accordance with the requirements of ISO/IEC 17025.

At concentrations below 2%, the amount of biologically active substances is close to the measurement limit, and the accuracy decreases (uncertainty up to 4-5%). In the range of 4-5%, emulsion particles aggregate, which causes systematic errors in spectrophotometric and chromatographic analyses.

In particular, errors in the dosing stage - manual addition, incorrect mixing - account for 40% of the total uncertainty. Therefore, it is necessary to use automatic dosing systems and calibrate each batch via CRM.

The storage period of the samples is also important: after 14 days, the peroxide number increases sharply, which distorts the measurement results. Therefore, metrological monitoring is designed for a period of only 14 days.

Conclusion: Metrologically acceptable accuracy limits for pumpkin seed oil addition to functional beverages should be in the range of 2-4%. 3% is recommended as optimal, as the relative standard uncertainty at this amount is around 2.1-2.5%, which is in line with international standards.

The developed metrological scheme includes:

- automatic dosing systems,
- certified standard materials,
- Multi-step control based on HPLC and spectrophotometry,
- emulsion stability through zeta potential.

This approach not only ensures product quality, but also confirms the authenticity of the ingredients indicated on the label, which increases consumer confidence and expands export opportunities. It is recommended to introduce such metrological systems in the Uzbek food industry in the future.

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