INTRODUCTION

Iodine is one of the most important elements that are necessary for the normal functioning of the human body. Iodine is present in the thyroid gland at 80% of the body's total content. The thyroid gland is the main storehouse for iodine, and it synthesizes the most important regulatory hormone, thyroxine (T4), and triiodothyronine (T3). These hormones affect the growth and development of the body, including the thyroid gland itself. The production of these hormones depends on the availability of iodine, particularly in the diet. A deficiency in iodine can lead to serious health problems, including the development of goiter and an increased risk of hypothyroidism.

Materials and methods: The determination of iodine was carried out using the Ecotest-120 pH meter. "Ekomi-I" was used as an ion-selective electrode. Silver chloride electrode "ESR 10101" was used as a reference electrode.

Results and Discussion: The developed procedure has a suitable level of linearity (correlation coefficient = 0.9995%), correctness (variation coefficient = 1.58%), repeatability (variation coefficient = 6.67%), and analytical area (0.03–209.4 μg/mL analyte in the test solution). The procedure allows us to determine iodine in the form of iodides with an accuracy comparable to the accuracy of neutron activation analysis and can be recommended as an alternative to titrimetric methods existing in the world-leading pharmacopoeias.

Keywords: Iodine in the iodides form, kelp thallus, pharmacopoeial analysis, validation.
implementation of this treatment type. The content of this element can be up to 5%, calculated on the dry weight of the crude herbal drug (CHD). For example, land plants absorb only 0.2–0.5 µg in terms of one gram plant mass. For this reason, kelp thallus has long been included in the State Pharmacopoeia of the Russian Federation, as well as in the British and European Pharmacopoeias as a biological source of iodine.

When developing procedures for the quantitative determination of iodine, it is desirable to know in which form it occurred in a natural object. Available literature data show that the share of iodides in kelp accounts for approximately 90% of the total iodine content; the remaining 10% are iodinated amino acids, the most are iotyrosine.

Different chemical and physicochemical methods are used to determine the iodine in the kelp thallus. So, the laboratory titrimetric method (iodometry) is used to determine the gross iodine content according to the State Pharmacopoeia of Russian Federation XIV edition (SPRF XIV), European Pharmacopoeia (EP), and British Pharmacopoeia (BP) that is insufficiently selective, especially in the case of natural objects.

It includes several stages: preliminary burning of the analyzed sample in an oxygen atmosphere, followed by absorption of the reaction products with a mixture of water and sodium hydroxide, as in the case of or by calcination in an open flame of the burner as in the case of. In some works, iodine content was determined by a gas chromatograph equipped with an electron-capture detector. The analysis can be performed by high-performance liquid chromatography after extraction of iodine compounds with ionic liquids containing a pyridinium cation.

Some attempts have been made to use atomic absorption spectrometry (AAS) to determine the total iodine content. However, the determination of this nonmetal AAS method can be determined by the AAS method only indirectly by determining the metals that form the iodine compounds with accurate stoichiometry. However, this nonmetal can be determined by the AAS method only indirectly, namely by determining metals that form compounds with iodine with exact stoichiometry. The most widely used methods in the iodine determination in natural objects are the mass spectrometry with inductively coupled plasma and the older labor-intensive Sandell–Koltgoff kinetic method. The Sandell–Koltgoff method is extremely sensitive. However, it is not highly selective. A more modern method is inductively coupled plasma mass spectrometry (ICP-MS), which has a large dynamic range (up to 5–7 orders of magnitude). Nevertheless, this method is not without significant drawbacks. It is expensive and requires the almost complete mineralization of the sample because the introduction of solutions containing a high level of organic substances into inductively coupled plasma is difficult and also addicted to significant matrix effects, which are associated with the transportation of the analyzed solution to the plasma and strong isobaric influences from the components of the matrix.

The inexpensive ionometry method is a real alternative to the Sandell–Koltgoff method and ICP-MS, as iodine is presented mainly in the iodides form in the kelp. This method provides an effective combination of the determination step with the fast, simple, and safe step of sample preparation. Unfortunately, in recent decades, the number of works devoted to the use of direct ionometry for the determination of iodine in pharmaceutical objects is very small. Perhaps it is worth mentioning the work, in which the authors developed a solid-state ion-selective electrode containing 1,3-dihexadecylimidazolium ionic liquid. According to the developers, depending on the composition of the anionic part of the ionic liquid, the electrode can be selective both with respect to thiocyanates and iodides. However, this electrode is not easily accessible and standardized, as it is not produced by modern industry. Solid-state electrodes (including chemically and mechanically stable crystalline membranes, for example, made of Ag2S) doped with AgI) are the most widely used in the analysis practice. They are rather convenient, industrially fabricated and, accordingly, standardized. Nevertheless, in pharmacognostic practice, the ionometry method is currently not widely used, despite the availability and relative cheapness of such iodide-selective electrodes and equipment in general. Until now, iodine is determined in factory laboratories using the laborious, insufficiently selective titrimetric method used in pharmacopoeias.

The purpose of this study was to develop and validate a simple, effective procedure for the quantitative determination of iodine in the form of iodide by ionometry in the kelp thallus. The developed technique can be recommended as an alternative to titrimetric methods existing in the world-leading pharmacopoeias.

**Materials and Methods**

**Devices and materials**

The determination of iodides was carried out by using the “Ecotest-120” pH meter. “Ekom-I” was used as an ion-selective electrode. Silver chloride electrode “ESR 10101” was used as a reference electrode. The background electrolyte was a 1M KNO3 solution.
Standard samples, reagents, and standard solutions

High-quality laboratory grade water (conductivity <4.3 μS·cm⁻¹ at 20°C), potassium nitrate (high-grade purity, Himmed, Russia, Moscow), potassium iodide (ISO, European Pharmacopoeia Reagent, ≥99.5%, Sigma-Aldrich, Germany, Steinheim) were used in analysis.

The background electrolyte solution was prepared as follows: 101 g (accurately weighed) of potassium nitrate was quantitatively transferred to a 1000-mL volumetric flask, 600 mL of water was added, stirred until the sample was completely dissolved, the solution volume was adjusted to the mark with the same solvent, and stirred. The initial standard solution was prepared by dissolving an exact portion of potassium iodide (16.6 g), previously dried at a temperature of 105°C–110°C to constant weight, in a 1000-mL volumetric flask; the solution volume was adjusted to the mark with water, and stirred (concentration 1–10⁻¹ mol/L [209.4 μg/mL]). The following standard solutions were prepared by sequential dilution and iodide concentrations were obtained: 1 × 10⁻⁴ mol/L (0.30 μg/mL), 1 × 10⁻³ mol/L (2.70 μg/mL), 1 × 10⁻² mol/L (23.9 μg/mL); 1 × 10⁻¹ mol/L (209.4 μg/mL), 1 × 10⁻² mol/L (309.4 μg/mL), and 1 × 10⁻¹ mol/L (0.30 μg/mL). For this purpose, 10 mL of a solution with a higher concentration was placed in a 100-mL volumetric flask; the solution volume was adjusted to the mark with water. High-quality laboratory grade water was used as a compensation solution.

Sample

The kelp thallus sample was purchased in the Moscow pharmacy network. The sample is corresponded the requirements of SPRF XIV edition.[14]

Samples’ preparation

Accurately weighed 5 g of the CHD, crushed to a 3-mm particle size, is placed in a 500-mL conical flask. 200 mL of water with a temperature of 90°C is added. The sample is incubated for 1 h at room temperature (25°C). The obtained extract is filtered through five layers of gauze into a 500-mL measuring cylinder, the CHDs are squeezed out, and the volume of the obtained extract is measured. The coefficient φ is calculated as follows: \( \phi = \frac{5 \cdot V/200 \cdot m}{V/40 \cdot m} \), where \( V \) is the extraction volume, mL and \( m \) is the kelp thallus sample weight, g.

Drawing a calibration curve

For drawing a calibration curve, standard solutions with concentrations of 1 × 10⁻⁴, 1 × 10⁻³, 1 × 10⁻², 1 × 10⁻¹, and 1 × 10⁻⁴ mol/L were used. Its preparation is described in “Standard samples, reagents, and standard solutions” section. The instrument (“Ecotest-120” pH meter) automatically recalculates the molarity (mol/L) in μg/mL when the analyte concentration is determined by the calibration curve.

Procedure

5 mL of a background electrolyte solution is added to 45 mL of standard and test solutions, as well as a comparison solution. Then the concentration of iodides in the test solution is measured according to the calibration graph.

The iodide content (X,%) is calculated by the following formula:

\[
X = \frac{(C - C_0) \times 100 \times 100}{1000000 \times 0 \times (100 - W')}
\]

\[
\phi = \frac{2 \times (C - C_0)}{0 \times (100 - W')} \times 100 \%
\]

where \( C \) is the iodide concentration in the analyzed solution, μg/mL; \( C_0 \) is the iodide concentration in the compensation solution; \( \phi \) is the CHD water absorption coefficient; \( a \) is the weight of sample, g; and \( W \) is the CHD humidity, %.

RESULTS AND DISCUSSION

Feasibility demonstration

The selectivity of industrially manufactured crystalline solid-state and selective electrodes is achieved by the vacancy charge transfer mechanism, in which vacancies are filled only with a certain mobile ion (Ag⁺), as the shape, size, and charge distribution of the vacancy correspond only to a certain mobile ion. As a result, extraneous ions (NO₃⁻, ClO₄⁻, SO₄²⁻, and many others) that do not precipitate with the Ag⁺ ion practically do not affect the potential of such an electrode; ions of other halides also do not significantly affect the determined component analytical signal due to the significant difference in the solubility products between AgI and AgBr, AgCl.

Table 1 illustrates well the foregoing. Table 1 presents approximate selectivity coefficients. They are the ratio of the solubility product of the silver salt
of the element being determined—iodine—to the solubility product of the silver salt of the interfering component X.\(^{38}\) The presented data show that the selectivity of the iodine-selective electrode with respect to the analyte in the presence of interfering ions is extremely high.

So, iodide in the form of iodide is determined 1,000,000 times more sensitive than chloride. Chloride is a matrix ion, the most widely distributed in nature, including in CHDs. Strict differentiation of cells by tissues, as in higher land plants, is absent in thallus of brown algae, although their thallus are multicellular. The result of this is a relatively uniform distribution of iodine in the thallus cells, which is confirmed by the available literature data\(^{11}\) and shown in this work using the example of studying the effect of the degree of kelp thallus crushing on I\(^–\) extraction. The data are shown in Table 2.

Table 1 shows that the crushing degree influence on the iodide extraction effectiveness in the aqueous phase is insignificant in the studied range. The results can be explained by the extreme hydrophilicity of the inorganic form I\(^–\) compared to organic biologically active compounds contained in higher plants. This makes the analyte easy to recover. In the kelp thallus chemical analysis, it is difficult to study the effect on iodide extraction of indicators such as CHD, extractant ratio, and extraction ratio due to the peculiarities of the physicochemical determination method used. With a decrease in the extractant volume, the extraction viscosity increases, which makes it difficult to establish an equilibrium membrane potential. It also influences the correct analyte determination. With extractant increasing volume, as well as dilution, the metrological characteristics of the technique significantly deteriorate. The determination error increases significantly and can exceed 10%.

Brown algae are marine phyto-organisms. When water is added to the dried CHD, the latter is intensively absorbed by the plant cells, which affects the final extraction volume. This effect must be taken into account in the iodine quantitative determination in the form of iodide in the kelp thallus using the introduction of a special coefficient. Table 3 shows the data reflecting the coefficient determination taking into of CHD water absorption coefficient (\(\varphi\)). So, the obtained data analysis shows that the coefficient \(\varphi\) is 0.798—the volume of the final extraction differs by 20% from the initial volume of the added extractant.

### Linearity

The electrode potential of standard solutions was sequentially measured. The solution preparation is described in the section “Standard samples, reagents, and standard solutions.” Table 4 shows the standard electrode potential values. According to Table 4, a graph was plotted: the potential of the indicator electrode—on the \(y\)-axis; \(p\)-function for iodide ions (negative decimal logarithm from the equilibrium concentration of iodide)—on the \(x\)-axis. The graph is shown in Figure 1. The presented data show that the analytical signal is directly proportional to the analyte concentration in the entire working range of the ion-selective electrode. The correlation coefficient (\(R\)) obtained from the calibration line analysis was more
than 0.9995%. The result confirms the quantitation method linearity. The relative standard deviation (RSD) calculated at the found iodide concentration in the analyzed solution (105.6 µg/mL) was less than 5% ($n = 3, P = 0.95$).

**Correctness**
The procedure correctness was proved by the method of additives by adding the exact volume of the solution with the iodide concentration of 2100 µg/mL. Three parallel determinations were made for each additive, adding the initial solution to the analyzed solution, so that the final analyte concentration was in a linear concentration range [Figure 1]. The correctness index was evaluated by calculating the recovery, that is, as the ratio between the total content of the introduced I– and the initially available I– in the test solution, expressed as a percentage. The method correctness determination results are presented in Table 5. The data presented in Table 5 show that the correctness of the iodine quantitative determination in the form of iodides for the average value of each of the three determinations is in the range from 95.0% to 105.0%. Statistical characteristics calculated on the basis of recovery [Table 5] are presented in Table 6. The obtained data indicate that the analyte quantitative determination variation coefficient ($n = 9$) does not exceed 4.0% and amounts to 1.58%.

**Repeatability**
To determine the repeatability, the coefficient of variation was calculated according to the results of the iodine quantitative determination in the form of iodines ($n = 6$) in the test solution. Table 7 presents the

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**Table 5: Correctness of iodine determination in the form of iodides in kelp thallus by ionometry**

| The iodide content in the extract, µg/mL | Introduced, µg/mL | The calculated iodide content, µg/mL | Was found, µg/mL | Recovery, % |
|----------------------------------------|-------------------|--------------------------------------|-----------------|------------|
| 105.2                                  | 12.7              | 117.4                                | 118.6           | 101.0      |
|                                        | 12.4              | 117.6                                | 118.3           | 100.6      |
|                                        | 12.2              | 117.4                                | 118.3           | 100.8      |
|                                        | 38.8              | 144.0                                | 147.6           | 102.5      |
|                                        | 41.4              | 146.6                                | 146.5           | 99.9       |
|                                        | 43.1              | 148.3                                | 147.7           | 99.6       |
|                                        | 116.1             | 221.3                                | 215.9           | 97.5       |
|                                        | 119.2             | 224.4                                | 221.1           | 98.5       |
|                                        | 116.6             | 221.8                                | 225.9           | 101.9      |

**Table 6: Openability statistical characteristics**

| Statistical characteristics | Results |
|-----------------------------|---------|
| Lowest value, %             | 97.5    |
| Highest value, %            | 102.5   |
| Average value, %            | 100.0   |
| Standard deviation          | 1.58    |
| The standard deviation of the average result | 0.53 |
| The variation coefficient (VC), % | 1.58 |
| Confidence interval ($P = 0.95$) | 1.22 |

**Figure 1:** Linear relationship between the $p$-function on the iodide’s equilibrium concentration and the electrode potential.
metrological results. Analysis of the data presented in Table 7 shows that the result variation coefficient of the iodine quantitative determination in the form of iodide in the kelp thallus \((n = 6)\) is 6.67%.

**Analytical area of the procedure**

The quantitative determination of the iodine procedure is applicable in the range of contents from 0.03 to 209.4 \(\mu\)g/mL analyte in the test solution. Correctness, linearity, and repeatability are established for interval values and values within the interval: linearity (0.03%–209.4%), correctness (0.03%–209.4%), and repeatability (approximately 0.42%). So, the given validation characteristics allow us to conclude that the developed technique can be used to determine iodine (in the form of iodides) in the kelp thallus. The data obtained using the developed methodology were in agreement with the data given in Küpper et al. \[11\]. Also, the obtained data were compared with the data obtained by the method from SPRF XIV edition. \[14\] The results are presented in Table 8. The data presented in Table 8 indicate that the developed procedure allows us to determine iodine in the form of iodides with an accuracy comparable to the accuracy of neutron activation analysis. At the same time, the result obtained using the methodology \[14\] cannot be considered satisfactory, as it turns out to be almost 2.5 times overstated due to the nonselectivity of the titrimetric method.

**Conclusion**

An effective procedure has been developed for the quantitative determination of iodine in the form of iodide in the kelp thallus by the ionometry method and its validation has been carried out. Linearity, correctness, and repeatability were estimated. It was shown that the procedure provides satisfactory results when the content of the analyte in the test solution is 0.03–209.4 \(\mu\)g/mL. Data comparison obtained using the developed procedure and the official method given in the State Pharmacopoeia of the Russian Federation reveals high selectivity and satisfactory accuracy (variation coefficient does not exceed 7%) of the ionometric approach. The procedure is promising for controlling the iodine content in kelp thallus in laboratory factories engaged in the production of herbal remedies as it does not require the use of expensive equipment and reagents.

The procedure can be used as an alternative procedure to existing ones. Also it can be recommended for inclusion in leading pharmacopoeias.

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**Conflicts of interest**

There are no conflicts of interest.

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