Research into quality of pectins from pea seed coats

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Abstract: For the first time, the complete set of quality indicators and sorption properties was studied in pectins extracted from the seed coats of peas, using standard and modified methods. It is established that organoleptic and physico-chemical indicators of pectins from pea seed coats meet the requirements of GOST 29186-91; moreover, safety and microbiological indicators conform to both GOST 29186-91 and SanPiN 2.3.2.1078-01. According to the main analytical characteristics, pectins from pea seed coats are highly esterified and meet the requirements for industrial pectins. The sorption properties of the studied pectins are the most pronounced when exposed to lead, cadmium and copper ions, and to a lesser extent, zinc ions. The obtained data allows us to recommend pectins from pea seed coats as structure-forming agents and sorbents of heavy metal ions in the production of functional foods.

Keywords: pea seed coats, pectin, analysis of quality indicators, degree of esterification, sorption properties

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INTRODUCTION

According to GOST R 51806-2001, pectin is a dry, powdery product, ranging from light beige to light grey in colour. It consists mainly of polygalacturonic acid, which is partially esterified with methanol, and its salts, having a mass fraction of galacturonic acid in a dry, deashed substance of at least 65%. It is typically extracted from the tissues of higher plants, and has a stabilising, thickening, gel-forming capability, as well as functional properties. Pectin is intended to be consumed simultaneously with food or to be used as a food supplement or physiologically functional food ingredient.

Due to their ability to swell, pectins ensure the integrity and stability of plant tissues, and regulate water exchange [1]. With metal salts, pectins form insoluble, stable compounds, which are not adsorbed in the human intestine. The complex forming properties of pectins are the result of the presence of the carboxyl and hydroxyl groups of galacturonic acid in their molecules. Pectins are capable of binding to heavy metal ions entering the digestive system, as well as preventing their secondary resorption when they enter the gastrointestinal tract with bile or as part of other digestive secretions. According to the data presented in the study [2], pectins with a degree of esterification below 20.0% are the most effective sorbents of strontium ions. It has also been established that pectin-containing drugs are able to remove antimony, radioactive strontium, mercury and lead from the human body, and can increase the liver’s detoxication function [3]. In the article [4], the authors showed that pectin can be used to stabilise liposomal drug delivery systems. Pectin extracts are the source of a number of macro- and micro-nutrients, organic acids and amino acids. Their increased anti-radiological properties are also noted. Moreover, these properties are especially pronounced in native pectins that are soluble in liquids and not subjected to dehydration [5]. Pectins also exhibit antimicrobial activity against non-spore-forming causative agents of food poisoning such as staphylococcus, dysentery bacteria and other microorganisms, which, according to E.B. Lazareva et al., can be attributed to their antioxidant properties [6].

Therefore, the demand for pectin, which acts as a natural functional ingredient, is increasing in the areas of food production, pharmaceutical enterprises and the dietary supplement industry. However, in the Russian Federation, the development of the secondary market is hindered by a lack of domestic pectin production. This results in high prices for pectin and consumer dependency on imports. Thus, the search for sources of relatively inexpensive pectin-containing raw materials for domestic production of affordable and high-quality pectin is very relevant.

Commercially, pectins are currently obtained from apple pomace, beet pulp, sunflower capitulum and citrus peels (lime, lemon, orange, grapefruit) [1, 7]. In addition, the possibility of extracting pectins from pumpkin [8], daikon [9], kiwi [10] and other crops has recently been studied. A number of researchers suggest the use of secondary raw assets as a source of pectins. They propose cotton cell walls [8], seed coats [11–13] and nuts [14], the bark of coniferous trees [15] and berry pomace [16].

During pea processing, waste is produced in the form of coats, which comprises up to 14% of the total seed mass. These coats contain valuable components such as fibre, hemicellulose, pectin substances, antioxidant proteins, essential amino acids, starch, B vitamins, etc. [17, 18]. D. Brassar et al. found that pectin is localised in the cell walls of the parenchymal tissue of pea coats [19]. It was shown that pectin substances in pea seed coats are composed of two parts: water-soluble and water-insoluble, with insoluble proteopectin predominating [20].

Thus, along with other non-traditional sources, seed coats as a by-product of pea processing can be considered as a raw material for obtaining high-quality pectin and reducing the dependency of domestic producers on imports.

The purpose of this study was to conduct a comprehensive assessment of the quality of pectins extracted from the seed coats of peas, as well to determine the possible future direction of their use in the food industry.

EXPERIMENTAL PART

Pectins extracted from the pea seed coats of Temp (smooth seeds) and Amior (wrinkled seeds) varieties of the FSBSI “Federal scientific centre of grain legumes and cereal cultures” (Orel, Russia) selection were used as the object of research.

Organoleptic and physico-chemical indicators of the quality of pectins were assessed according to GOST 29186-91. Pectin was placed on a sheet of white paper and visually examined to deduce the shape of the particles, and their colour, smell and taste. Extraneous matter visible to the naked eye was evaluated visually. The mass fraction of moisture was determined by drying a portion of pectin at a temperature of 103 °C until it reached constant weight.

The degree of esterification of pectins was assessed by the titrimetric method, determining
free and – following saponification – esterified carboxyl groups of polygalacturonic acid in a portion of pectin, purified from soluble ballast impurities and cations.

Nitrates were determined by their reduction to nitrites with subsequent photocolorimetric measurement of the azo dye colour intensity, which is formed by the interaction of nitrite with sulphanilic acid and \( \alpha \)-naphthylamine in acetic acid by the Griess reaction.

The mass proportion of the fibre fraction was determined by sifting the pectin through a 0.5 mm soil sieve having a woven wire mesh and weighing the residue on the sieve, matching to the percentage of fibre fraction.

The pH of a 1% pectin solution was determined by dissolving pectin in distilled water, then heating the solution for 10–15 minutes at a temperature of 50–60 °C, decanting from the insoluble residue and determining pH on an I-500 ionometric transducer.

The mass proportion of ballast substances was determined by pouring pectin with acidified 70% ethanol, followed by washing with pure 75% and then with 96% ethyl alcohol and drying to constant weight at a temperature ranging from 80 to 85 °C.

The mass proportion of acetyl groups in pectin was determined by alkaline hydrolysis followed by distillation in the presence of magnesium sulphate solution and titration with a 0.1 N alkali solution in the presence of phenolphthalein.

The content of free carboxyl groups in pectin was determined by dissolving pectin in distilled water, followed by titration with a 0.1 N alkali solution in the presence of Hinton's indicator. The number of methoxylated carboxyl groups was determined in the same solution. After determining the content of free carboxyl groups, a 0.5 N alkali solution was added to the neutralised sample, to hydrolyse the methoxylated carboxyl groups. Then to the solution was added a 0.5 N hydrochloric acid solution, the excess of which was titrated with a 0.1 N alkali solution. To calculate the amount of methoxylated carboxyl groups, corrections were made for acetyl groups, which are also susceptible to hydrolysis under these conditions. The content of pure pectin was calculated from the titrimetric data after the determination of free and methoxylated carboxyl groups, taking the content of ballast substances into account.

The ash content was determined by burning a portion of pectin in a pre-calcined and weighed crucible on a hot plate until the emission of combustion products ceased, followed by calcination in a muffle furnace until completely reduced to ash.

Evaluation of the gelatinous ability of pectin was carried out by determining the maximum tensile strength of the jelly using a Tarr-Baker tester in a series of tests with different contents of tartaric acid.

The lead content in pectins was determined according to GOST 30178-96; arsenic – according to GOST R 51766-2001; zinc – according to GOST 26927-8 and copper – according to GOST 30178-96. The content of cesium-137 and strontium-90 was evaluated using a scintillation gamma spectrometer along with the "Progress" software. Microbiological indicators were also determined: NMAFAM – according to GOST 10444.15-94; CB – according to GOST R 52816-2007; pathogenic microorganisms, including salmonella – according to GOST R 52814-2007 and mould fungii – according to GOST 10444.12-88.

Evaluation of the adsorption capacity of pectins was carried out according to the method developed in the Kazan State Technical University [21]. The authors of this article modified this technique as follows: a 1.0% aqueous solution of pectin was prepared, which was then infused for 3 hours, mixed with a solution of a heavy metal with a concentration of 100.0 mg/dm\(^3\), rested for 3 h and then the resulting dispersed phase was filtered.

For the preparation of solutions of heavy metals, 100.0 mg/dm\(^3\) state standard samples (SSS) of the solution's composition were used. The samples used were lead ions (SSS 7252-96), cadmium (SSS 6070-91), copper (SSS 6074-91) and zinc (SSS 7256-96). Sample preparation was carried out in a programmable two-chamber furnace PDP-Lab in order to determine the residual quantities of heavy metal ions in model solutions. The residual amount of heavy metal ions was determined by the voltammetric method according to MU 31-04 / 04 (FR 1.31.2004.00986) on a TA-Lab analyser.

All studies were performed in three replications with the subsequent processing of the obtained results using the Excel MS Office statistical package and determining the error of the mean and standard deviation at a significance level of 95%.

The studies were conducted in the Federal State Budget Educational Institution of Higher Education "Orel State University of Economics and Trade" (Orel), as well as in the "Orel Reference Centre" of the Federal service for veterinary and phytosanitary surveillance (Orel).

**RESULTS AND DISCUSSION**

The pectins extracted from the seed coats of the Tempa and Amior pea varieties according to their organoleptic characteristics met the requirements of GOST 29186-91 for food pectin. They presented as a fine, odourless powder of light-yellow colour having a sour taste peculiar to pectins. The colouring of pectins from pea seed coats correlates strongly with their purification from related substances, including colouring materials (Amior variety coats), during the extraction process.

Humidity of pectins from pea seed coats was 12.29–12.46% (Table 1), which was caused by their increased hygroscopicity.

Mass proportion of nitrite did not exceed the normalised GOST 29186-91 values. Impurities in the pectin were absent. The content of the fibrous
Analytical characteristics of pectins from pea seed coats

| Indicator                                      | Pectin from seed coats of pea varieties | Temp          | Amior         |
|------------------------------------------------|----------------------------------------|---------------|---------------|
| Mass proportion of pure pectin,%               |                                        | 77.37 ± 0.10  | 78.00 ± 0.12  |
| Mass proportion of ballast substances,%       |                                        | 16.04 ± 0.04  | 15.31 ± 0.02  |
| pH of 1% solution                              |                                        | 3.40 ± 0.01   | 3.50 ± 0.01   |
| Degree of esterification,%                    |                                        | 61.81 ± 0.21  | 63.03 ± 0.18  |
| Mass proportion of free carboxyl groups,%     |                                        | 7.20 ± 0.01   | 7.02 ± 0.01   |
| Mass proportion of esterified groups,%        |                                        | 11.66 ± 0.02  | 11.97 ± 0.04  |
| Mass proportion of methoxylated carboxyl groups,% |                                | 11.28 ± 0.02  | 11.56 ± 0.01  |
| Mass proportion of free carboxyl groups,%     |                                        | 18.48 ± 0.02  | 18.58 ± 0.04  |
| Gel-forming ability, °TB                      |                                        | 234.50 ± 2.20 | 236.00 ± 2.50 |

Table 1
Physico-chemical properties of pectin of pea seed coats

Таблица 1
Физико-химические показатели пектинов из семенных оболочек гороха

| Indicator                                      | Norm (according to GOST 29186-91) | Pectin from seed coats of pea varieties |
|------------------------------------------------|-----------------------------------|----------------------------------------|
| Moisture content, %                            | not more than 10.00               | 12.29 ± 0.04                           |
| Mass fraction of nitrates in the calculation on ion NO₃,% | not more than 0.18               | 0.06 ± 0.00                           |
| Impurities visible to the naked eye            | not permitted                     | absent                                 |
| Mass proportion of fibrous fractions larger than 0.5 mm,% | not more than 20.00               | 8.70 ± 0.05                           |

Table 2
Safety and microbiological indicators of pectin from pea seed coats

Таблица 2
Показатели безопасности и microbiологические показатели пектинов
из семенных оболочек гороха

| Indicator                                      | Acceptable level, not more than | Pectin from seed coats of pea varieties |
|------------------------------------------------|---------------------------------|----------------------------------------|
| Toxic elements, mg/kg:                         |                                 |                                        |
| Lead                                           | 10.0                            | 0.11                                   |
| Arsenic                                        | 3.0                             | not found                             |
| Copper                                         | 50                              | 0.10                                   |
| Zinc                                           | 25                              | 0.08                                   |
| Radionuclides, Bq/kg:                          |                                 |                                        |
| Cesium-137                                     | 160                             | not found                             |
| Strontium-90                                   | 90                              | not found                             |
| Microbiological indicators, CFU:               |                                 |                                        |
| NMAFAM in 1 g                                  | 5.00 · 10⁰¹²                     | 1.20 · 10³                          |
| GB (coliforms) in 0.1 g                        | not permitted                   | not found                             |
| Pathogens, including salmonella, in 25 g       | not permitted                   | not found                             |
| Mould fungi in 1 g                             | 5.00·10¹                        | not found                             |

Table 3
Analytical characteristics of pectins from pea seed coats

Таблица 3
Аналитические характеристики пектинов из семенных оболочек гороха

| Indicator                                      | Pectin from seed coats of pea varieties |
|------------------------------------------------|----------------------------------------|
| pH of 1% solution                              | 3.40 ± 0.01                           |
| Degree of esterification,%                    | 61.81 ± 0.21                          |
| Mass proportion of free carboxyl groups,%     | 7.20 ± 0.01                           |
| Mass proportion of esterified groups,%        | 11.66 ± 0.02                          |
| Mass proportion of methoxylated carboxyl groups,% | 11.28 ± 0.02                          |
| Mass proportion of free carboxyl groups,%     | 18.48 ± 0.02                          |

No radionuclides were found in the composition. Additionally, in the studied pectins there were no conditionally pathogenic microorganisms, pathogenic microorganisms or spoilage microorganisms.

In the study of safety indicators, it was found that the content of heavy metals in pectins from the coats of pea seeds is significantly lower than the allowable values (Table 2). No radionuclides were found in their composition. Additionally, in the studied pectins there were no conditionally pathogenic microorganisms, pathogenic microorganisms or spoilage microorganisms.

Evaluation of the analytical indicators of pectins obtained from pea seed coats revealed that the proportion of pure pectin in them was 77.37% (Temp variety) and 78.00% (Amior variety) (Table 3). The content of ballast substances (ash elements, protein substances, etc.) was 15.31% (Amior variety) and 16.04% (Temp variety), which meets international requirements for food pectin [8]. The acidity of the aqueous solutions of the studied pectins matched the pH values for pectins from other types of plant raw material.
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The degree of esterification (methoxylolation) determines such properties of pectic substances as solubility, gelation and the ability to react with metal ions. According to GOST R 51806-2001, highly esterified substances, in which the degree of esterification of carboxyl groups in the polygalacturonic acid molecule is 50.0% or more, are referred to as pectin. The permitted degree of esterification of the studied pectins, according to GOST 29186-91, allowed them to be classed as the highly esterified type B pectin (slow set) and testifies to their good gelation ability. The increased content of esterified groups also indicates the gelling properties of pectins from pea seed coats. The gelling power of the analysed pectins is comparable with indicators for industrial pectins according to GOST 29186-91. The presence of a significant amount of free carboxyl groups in the studied pectins confirms their complex formation capability [22].

Research of the ability of pectins from the seed coats of peas to bind heavy metal ions has shown that they display sorption properties towards lead, cadmium and copper to the greatest degree (Table 4).

Table 4

| Residual concentration in 1.0% pectin solution, mg/l | Pectin from seed coats of pea varieties |
|---------------------------------------------------|----------------------------------------|
|                                                   | Temp                                   |
|                                                   | Amior                                 |
| Lead                                              | 0.32 ± 0.01                           | 0.17 ± 0.01 |
| Cadmium                                           | 0.56 ± 0.02                           | 0.13 ± 0.01 |
| Copper                                            | 30.00 ± 0.02                          | 38.80 ± 0.02|

In 1.0% aqueous solutions of pectins containing lead ions in the amount of 100.0 mg/dm³, after infusion, heavy metal ions were not detected. It should also be noted that the pectin from the seed coats of the Amior pea variety bound copper and cadmium ions, respectively, 4.3 and 1.9 times more efficiently than the pectin from the coats of the Temp pea variety. The ability of pectins to bind zinc ions was less pronounced due to the low affinity of ions of this metal to the solutions of high esterified pectins from pea seed coats.

CONCLUSION

A comprehensive assessment of the quality of pectins from the pea seed coats showed that they meet the requirements for industrial pectins in terms of organoleptic, physico-chemical, microbiological and safety indicators. The studied pectins are classified as highly esterified, characterised by a high degree of purity and good gelation. An increased sorption capacity of pectins from pea seed coats towards lead, copper and cadmium ions was established. The obtained data provides an opportunity to recommend pectins from pea seed coats for use in the food industry as a gel-forming component in the composition of jelly food products of a functional designation.

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**Contribution**

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**Conflict of interests**

The authors declare no conflict of interests regarding the publication of this article.

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