COMPOSITION AND MICROSTRUCTURE INVESTIGATION FOR THE MODELING AND CLASSIFICATION OF DIETARY FIBER DERIVED FROM PLANTS

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Abstract: Investigation of the composition and microstructure of dietary fiber derived from plants showed that the stabilizers investigated differ with regard to size and shape of the particles and the density of particle distribution. The composition and microstructure of dietary fiber derived from plants have been studied using electron microscopy. Spectrometric profiles of chemical composition have been obtained, and the content of the predominant chemical elements in food microstructure stabilizers has been determined. Some similarity concerning the content of certain chemical elements and the ratio of the contents of different elements has been detected upon the analysis of food structure stabilizers of the same type (carboxymethylcellulose, gum, and sodium pyrophosphate). Mathematical processing of photomicrographs of structure stabilizer samples has been performed, and masks for the assessment of the content of microcavities in the particles of the structure stabilizers investigated have been created.

Key words: dietary fiber, stabilizer, microstructure, electron microscopy, histogram, carboxymethylcellulose, sodium alginate, sodium pyrophosphate, xanthan gum

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INTRODUCTION

Dairy products are among the most important components of human food. They account for 20% of protein supply and 30% of fat supply in the human diet. Creation of products with predefined properties and rational use of raw materials are the priority directions for the development of dairy products manufacturing technologies [1, 5, 6].

Adherence to scientifically based formulations and compliance of the final product composition with regulatory requirements concerning the composition of raw materials are among the most important issues to be controlled during the assessment of quality of dairy products. Hundreds of dairy products available on the market are in constant demand and often actively marketed; therefore sellers and manufacturers of dairy products alike are always tempted to adulterate these products. Therefore, reliable methods for the identification of raw materials found in dairy products are necessary to prevent faulty and adulterated foods from being sold [2, 7, 8, 9].

The question of reliable determination of the type of components found in dairy products is currently especially acute due to the widespread adulteration of foods with texture stabilizers. The use of these components implies adding them to foods to induce gelling of liquid systems. Structure stabilizers currently in use comprise anionic polysaccharides, both natural (pectin, agar, agaroid, and pyrophosphate) and artificial (oxidized starch). Alginates, cellulose derivatives, and carboxymethyl cellulose (CMC), as well as various gums, are widely used abroad [3, 10, 11, 12]. Classification of stabilizers can be based on one of the following criteria: description of all compounds as polysaccharide materials, assignment of names referring to botanical species, origin (plant, animal, or artificial), or chemical properties. Classification taking the origin of the stabilizers into account is currently preferred; according to this classification, all stabilizers are assigned to groups of modified natural or semi-synthetic stabilizers, chemically modified natural stabilizers or compounds similar to them, or synthetic gums obtained by chemical synthesis.

Agar is one of the classic structure stabilizers widely used in confectionery industry. However, the increasing shortage of agar sources necessitates the replacement of agar with other structure stabilizers. Various types of pectins are an example of promising structure stabilizers. They are currently used in food and pharmaceutical industry. Pectins are capable of forming gel systems characterized by a specific set of physical and chemical properties. Furthermore, pectin was shown to exert beneficial effects on the human organism, and the resources for pectin production are virtually unlimited [4, 13, 14, 15].

The aim of the present work was to compare the microstructure and composition of various structure stabilizers of plant origin for the subsequent development of procedures for the detection of adulterated products.

OBJECTS AND METHODS OF THE STUDY

Texture stabilizers of five different types, namely, carboxymethylcellulose CMC Akutsel 3265, CMC 4500-6000, sodium alginate NO4-600, sodium pyrophosphate SAPP 40, and xanthan gum, were the objects of the present study.

Scanning electron microscopy (SEM) analysis of stabilizer microstructure was conducted using a scanning electron microscope JSM-7500 FA. Photographs at magnification ranging from 100 to 500× were produced for each sample.

Analysis Station JEOL JED-2300 was used to investigate the composition of the structure stabilizers. X-ray microanalysis performed with this device yielded spectrometric profiles from which the chemical composition of the structure stabilizers was inferred.

Computational processing of photomicrographs revealing the stabilizer structure involved assessment of the content of microcavities using Corel Photo Paint X3 software; masks were created by extracting elements according to color, transformation of the photograph into a binary image and determination of the content of the elements of interest using a histogram.

RESULTS AND DISCUSSION

Investigation of the microstructure was performed for five types of structure stabilizers with different bulk densities (Table 1).

Table 1. Bulk density of the structure stabilizers investigated

Structure stabilizer	Bulk density, g/dm^3
CMC Akutsel 3265	450
CMC 4500-6000	490
Sodium alginate NO4-600	600
Sodium pyrophosphate SAPP 40	710
Xanthan gum	830

Microphotographs of CMC Akutsel 3265 at magnifications of 100, 200, and 500× are shown in Fig. 1. These photomicrographs show that the structure of CMC Akutsel 3265 comprises particulate elements shaped as elongated fibers with a rough surface; particle diameter ranges from 20 to 30 μm. The bulk density of CMC Akutsel 3265 equaled 450 g/dm^3 and was the lowest among those of all the structure stabilizers studied.

CMC (carboxymethyl cellulose) is a salt of a weak carboxylic acid produced in a reaction of sodium monochloroacetate with alkaline cellulose according to the following scheme:

$$\zeta\text{-OH} + \text{NaOH} \rightarrow \zeta\text{-ONa} + \text{H}_2\text{O}$$
$$\zeta\text{-ONa} + \text{Cl-CH}_2\text{-COONa} \rightarrow \zeta\text{-O-CH}_2\text{-COONa} + \text{+NaCl}$$

Each anhydropyranose unit of the carboxymethylcellulose molecule contains three -OH groups capable of reacting with sodium monochloroacetate. The substitution of all three OH groups is theoretically possible; however, the degree of substitution in real CMC samples ranges from 0.4 to 1.2. The pK values of the carboxyl groups are 4.0 and 4.4 at degrees of substitution equal to 0.5 and 0.8, respectively. Approximately 90% and 10% of the carboxyl groups are ionized at pH 7.0 and 5.0, respectively. Carboxymethyl cellulose is an ionogenic cellulose ester, and therefore its stabilizing effect depends on salt concentration and other properties of the medium.

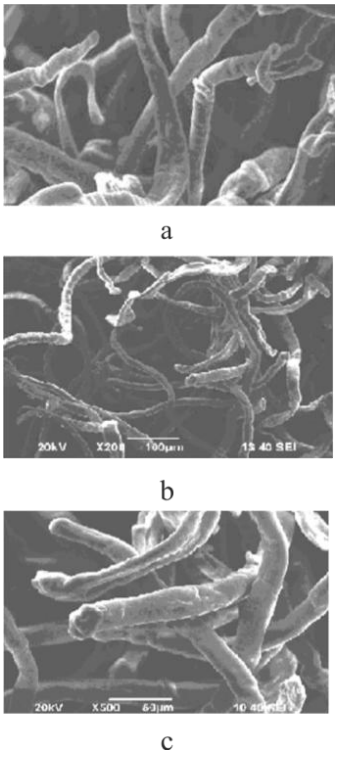

Fig. 1. Microstructure of CMC Akutsel 3265 at magnifications of 100 (a), 200 (b), and 500× (c).

The composition of CMC Akutsel 3265 is shown in Table 2.

The results demonstrate the predomination of oxygen (42.78 %) in CMC Akutsel 3265. Chlorine content is the lowest (0.07 %).

Table 2. Component composition of CMC Akutsel 3265

Element	Relative content, mass. %
Carbon	31.15±0.93
Nitrogen	21.90±0.65
Oxygen	42.78±1.28
Sodium	4.01±0.12
Chlorine	0.07±0.002

The microphotograph shown in Fig. 1-a was used to determine the content of microcavities in CMC Akutsel 3265. Filtering of background elements was necessary in this case. It was accomplished by increasing the contrast and correcting the mask manually. The results of microcavity detection in CMC Akutsel 3265 are shown in Fig. 2.

The content of microcavities in CMC Akutsel 3265 was 51.24±2.2% according to the histogram.

Thus, a low bulk density of 450 g/dm^3 is characteristic of CMC Akutsel 3265; the structure elements of this material have the form of elongated fibers with a diameter of 20–30 μm. Chemical elements present in CMC Akutsel 3265 include carbon, nitrogen, oxygen, sodium, and chlorine. The content of microcavities in this structure stabilizer amounted to 51.24±2.2%.

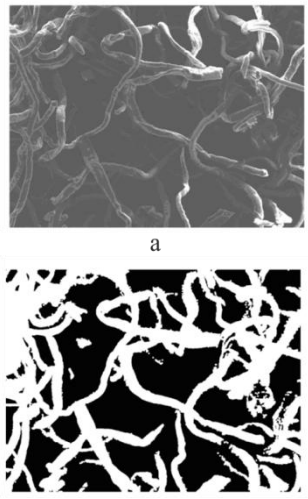

Fig. 2. The results of assessment of the content of microcavities in CMC Akutsel 3265:
a – photomicrograph at a magnification of 100×;
b – mask of the photomicrograph shown in Fig. 2-a.

Photomicrographs of the structure of CMC 4500-6000 at a magnification of 100, 200, and 500× are shown in Fig. 3.

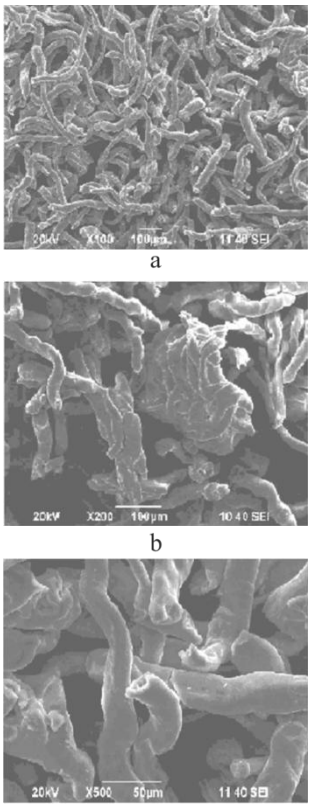

Fig. 3. Microstructure of CMC 4500-6000 at magnifications of 100 (a), 200 (b) and 500× (c).

These photomicrographs reveal the similarity of the microstructure of the structure stabilizer CMC 4500-6000 to that of CMC Akutsel 3265, with a denser arrangement of the elements in the former due to a higher bulk density (490 g/dm^3). As evident from Fig. 3-b and 3-c, some fibers in CMC 4500-6000 interweave, forming large clusters of particulate elements of more than 200 μm in size. The diameter of the fibers is 20–35 μm.

Results of the analysis of the chemical composition of CMC 4500-6000 are shown in Table 3. The content of oxygen, sodium, and chlorine in this stabilizer is higher than in CMC Akutsel 3265, while the content of carbon and nitrogen is lower.

Table 3. Component composition of CMC 4500–6000

Element	Content, mass %
Carbon	28.84±0.86
Nitrogen	17.29±0.52
Oxygen	48.01±1.44
Sodium	5.63±0.17
Chlorine	0.24±0.01

The results of microcavity detection in CMC 4500-6000 are illustrated by Fig. 4. The photomicrograph shown in Fig. 3a was used for the analysis.

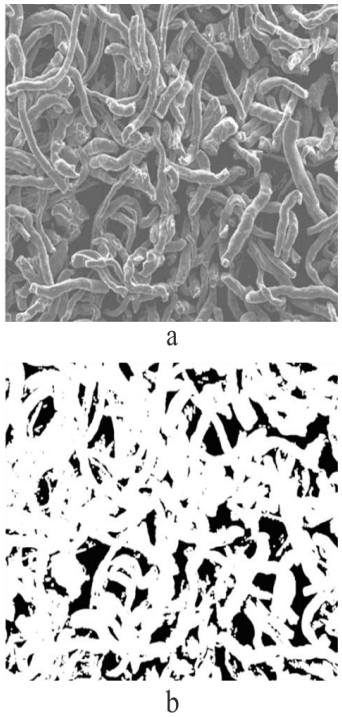

Fig. 4. The results of evaluation of the content of microcavities in CMC 4500-6000:
a – photomicrograph at a magnification of 100×;
b – mask of the photomicrograph shown in Fig. 4-a.

Since the elements of CMC 4500-6000 could not be reliably separated from the background using the color parameter, the selection borders were reduced by two pixels using contour selection after color selection.

The content of microcavities in CMC 4500-6000 was 20.28±1.2% according to the histogram.

The results show that the microstructure of CMC 4500-6000 is constituted by irregularly shaped fibers of 20–35 μm in diameter, the chemical components of this stabilizer include carbon, nitrogen, oxygen, sodium, and chlorine, and the content of microcavities equals 20.28±1.2%.

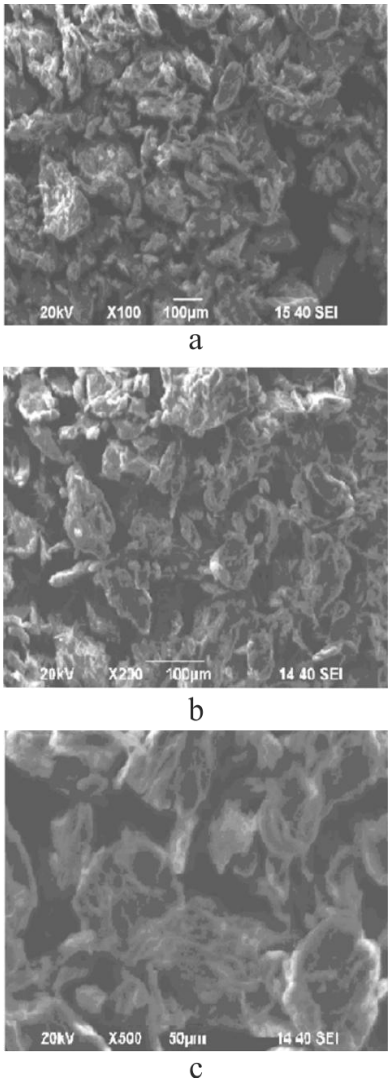

Fig. 5. Microstructure of sodium alginate NO4-600 at magnifications of 100 (a), 200 (b) and 500× (c).

Photomicrographs of sodium alginate NO4-600 at a magnification of 100, 200, and 500× are shown in Fig. 5. As evident from Fig. 5, structural elements of sodium alginate NO4-600 are particles of irregular shape ranging from 20 to 250 μm in size. Crystalline formations are present on the surface of the particles. Both rounded and elongated particles were detected. The bulk density of sodium alginate NO4-600 is 600 g/dm^3.

The composition of sodium alginate NO4-600 is illustrated by Table 4; as evident from the data obtained, the presence of calcium and the absence of nitrogen distinguish sodium alginate NO4-600 from the structure stabilizers described above. Oxygen is the predominant component of sodium alginate NO4-600 (52.91 %), and the content of chlorine (0.19 %) is the lowest.

Table 4. Component composition of sodium alginate NO4-600

Element	Content, mass. %
Carbon	37.37±1.12
Oxygen	52.91±1.58
Sodium	9.26±0.28
Chlorine	0.19±0.006
Calcium	0.27±0.01

Photographs of the microcavities in the structure of sodium alginate NO4-600 are shown in Fig. 6; these images were obtained by processing the photomicrograph in Fig. 5-a.

The content of microcavities inferred from the mask obtained (Fig. 6-b) was 33.79 ± 1.1%.

Thus, the basic structural elements of sodium alginate NO4-600 are dispersed particles of irregular shape with the size of 20–250 μm and crystalline formations on the surface. Carbon, oxygen, sodium, chlorine and calcium are among the components of sodium alginate NO4-600. Computational processing of the photomicrographs yielded a value of 33.79±1.1% for the content of microcavities in this structure stabilizer.

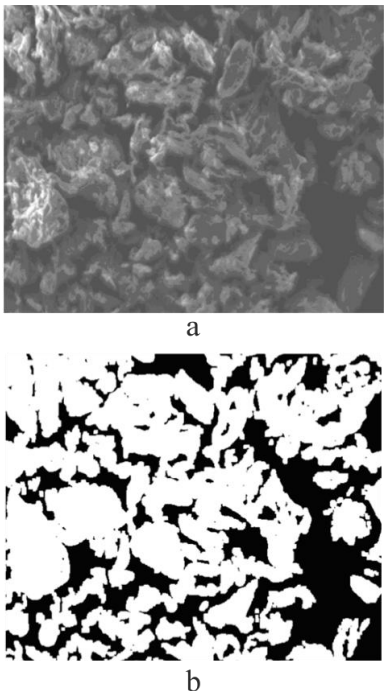

Fig. 6. Microcavities in sodium alginate NO4-600: a – photomicrograph at a magnification of 100×; b – mask of the photomicrograph shown in Fig. 6-a.

Photomicrographs of sodium pyrophosphate SAPP 40 at a magnification of 100, 200, and 500× are shown in Fig. 7.

Dense arrangement and high coverage are characteristic of the microstructure of sodium pyrophosphate SAPP 40. The particle size ranges from 5 to 90 μm. This structure stabilizer has a high bulk density of 710 g/dm^3.

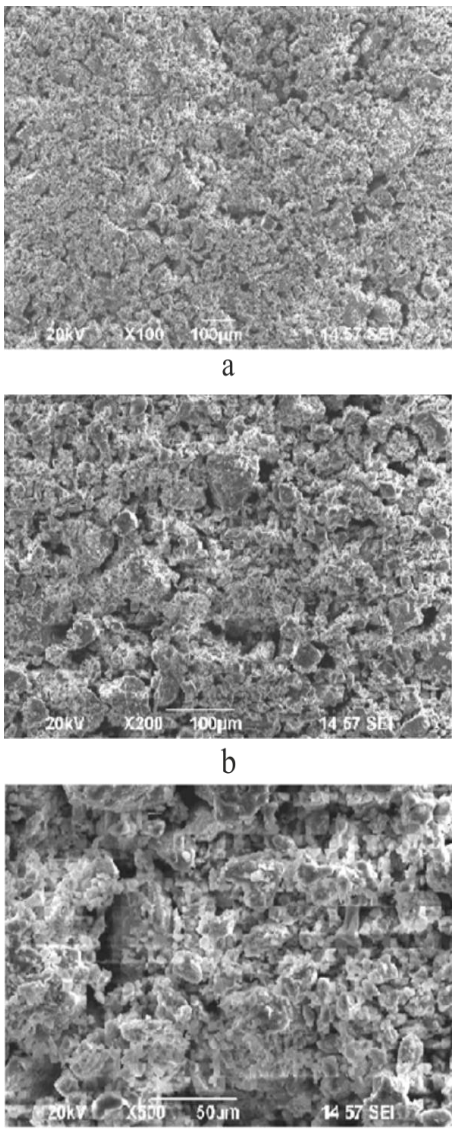

Fig. 7. Microstructure of sodium pyrophosphate SAPP 40 at magnifications of: a – 100; b – 200; c – 500×.

Results of the analysis of the composition of sodium pyrophosphate SAPP40 are shown in Table 5.

Table 5. Component composition of sodium pyrophosphate SAPP 40

Element	Content, mass. %
Carbon	8.21±0.25
Oxygen	51.94±1.55
Sodium	18.22±0.54
Phosphorus	21.63±0.65

The results of the quantitation of microcavities in sodium pyrophosphate SAPP 40 are shown in Fig. 8.

Analysis of the results presented in Fig. 8 showed that the content of microcavities in this stabilizer was 3.26±0.1%.

Thus, the structure of sodium pyrophosphate SAPP 40 is formed by closely spaced fine particles of irregular shape and a size of 5–90 µm. The content of microcavities is 3.26±0.1%.

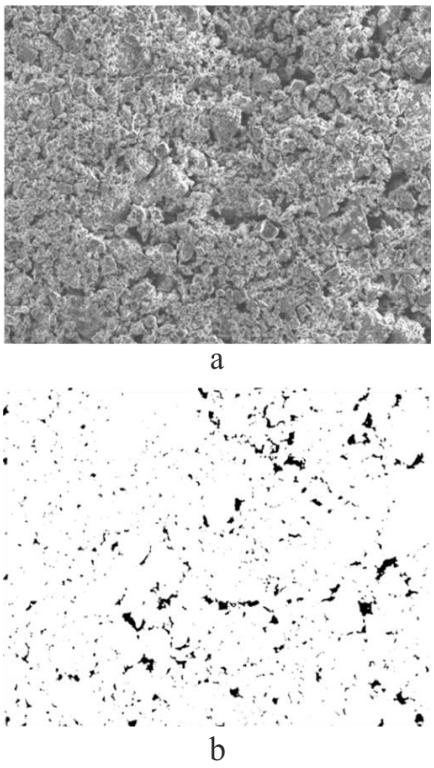

Fig. 8. Assessment of the content of microcavities in sodium pyrophosphate SAPP 40:
a – photomicrograph at a magnification of 100×;
b – mask of the photomicrograph shown in Fig. 8-a.

Photomicrographs of xanthan gum at a magnification of 100, 200, and 500× are shown in Fig. 9.

Xanthan gum is characterized by the highest bulk density among all structure stabilizers studied (830 g/dm^3) and has a dense fine structure. Most granules are elongated and range from 5 to 40 µm in size (Fig. 9-c). Such particle size allows for rapid formation of highly viscous solutions in both hot and cold food systems and results in the production of high-quality foods. Some of the structural elements of xanthan gum form conglomerates. Microcavities of irregular shape are also present in the structure (Fig. 9-a).

The composition of xanthan gum is shown in Table 6.

Xanthan gum contains carbon, nitrogen, oxygen, and potassium.

Quantitation of the content of microcavities in xanthan gum was performed using a microphotograph taken at a magnification of 500×, in order to make the quantitation error as low as possible. The results are presented in Fig. 10. The content of microcavities in the structure of xanthan gum was 6.51±0.3%.

Thus, the microstructure of xanthan gum is formed by closely spaced elongated granules of 5–40 µm in size. Carbon, nitrogen, oxygen, and potassium are found in this stabilizer. According to the results of computational processing, the content of microcavities in xanthan gum equals 6.51±0.3%.

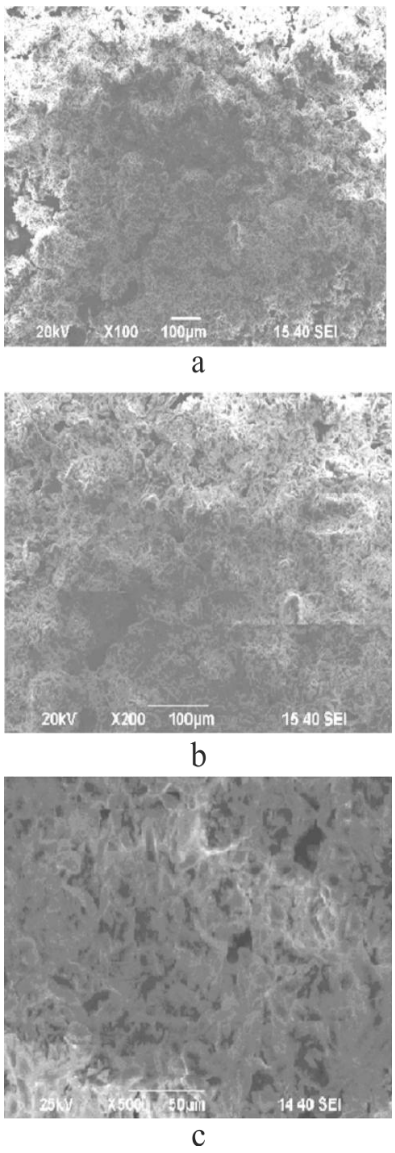

Fig. 9. Microstructure of xanthan gum at a magnification of 100 (a), 200 (b), and 500× (c).

Table 6. Composition of xanthan gum

Element	Content, mass %
Carbon	30.08±0.90
Nitrogen	23.42±0.70
Oxygen	46.38±1.39
Potassium	0.12±0.004

Analysis of the data shows that xanthan gum has the lowest specific surface area ($9.07 \cdot 10^{-6}$ cm^2) of all of the structure stabilizers investigated, and therefore its bulk density (830 g/dm^3) is the highest. Nitrogen was not detected in sodium alginate NO4-600. Sodium was not detected in xanthan gum. Chlorine was not detected in sodium pyrophosphate SAPP 40. In general, such elements as carbon, nitrogen, oxygen, sodium, and chlorine were detected in all structure stabilizers investigated, although the content of these elements varied.

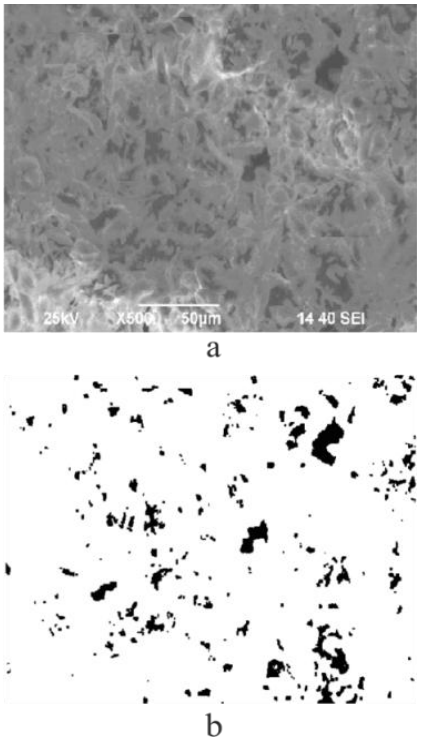

Fig. 10. Evaluation of the content of microcavities in xanthan gum:
a – photomicrograph at a magnification of 500×,
b – mask of the photomicrograph shown in Fig. 10-a.

Elongated fibers ranging from 15 to 60 µm in size were found among the structural elements of all types of carboxymethylcellulose investigated, with interweaving fibers forming large objects (above 200 µm in size) detected in CMC 4500-6000. The size of the elements of sodium alginate NO4-600 ranged from 20 to 250 µm. Sodium pyrophosphate SAPP 40 contained closely spaced fine particles of 5–90 µm in size. Structure elements of xanthan gum had a distinct elongated shape, their size was smaller (5–40 µm) than that of the other stabilizer particles, and the particles of this stabilizer were spaced more closely than in the others due to the high bulk density of the stabilizer (830 g/dm^3).

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