Abstract. The aim of the present paper was to determine the effect of the extraction time on the alkaline extracts of humic substances from organic and mineral lake bottom sediments. The extraction was performed with the use of 0.5 M NaOH. Both for organic and mineral sediments, the ratio during extraction was 1:10 and 1:50 (w/v). In both variants the extraction time was, respectively 1, 5, 12, 24 and 48 h. In the mixtures of humic and fulvic acids obtained, the following were determined: the content of carbon and the spectrometric properties for the UV-VIS range. Based on the present results, the values of absorbance ratios and extraction efficiency were calculated. The results demonstrate, most of all, that the extraction time has a significant effect on the properties of extracts. It was observed that the extracts after a longer extraction time, in general, showed a higher content of carbon. The present research revealed that in the case of sediments, the extraction performed in the ratio of 1:50 (w/v) is more efficient as compared with the extraction in the ratio of 1:10 (w/v).

Organic matter constitutes a quantitatively smaller, however an important fraction of bottom sediments. It is a mixture of lipids, carbohydrates, protein and other compounds contained in the tissues of living benthos microorganisms, detritus of autochthonic and allochthonic origin and humus substances diagenetically formed from this original biological material [14]. The main source of detritus are single-cell algae living in the photic zone. However, in many cases also the share of detritus originated from vascular plants can be also considerable. The relative shares of the matter from the two main sources are affected by the production in the lake, the production of vascular plants and transport processes [12]. The organic matter accumulated in sediments reflects two types and the amounts of materials from primary sources and the degree of transformations and degradation of the original material. The elemental, isotopic and petrographic composition preserves the records of the biological origin of organic matter which was deposited in lake sediments [15].
Humus substances are diagenetically formed from biological organic matter as a result of transformations and forming bonds of original compounds into macromolecular mixture and, in general, constitute from 60 to 70% of organic matter in young sediments and over 90% in the older ones [8]. Forming of humus substances involves including both changed and unchanged compounds into organic mixture. The specificity of this process has not been quite known. The research demonstrates that humus substances are products of fast diagenetic transformations of biogenic organic matter which undergo a further slow diagenesis after being buried in lake sediments [14].

The first humus substance research stage is the extraction for which different solutions are applied. In the soil studies the widely applied reagents are: NaOH and Na₂CO₃ of the concentration from 0.1 to 0.5M, whereas the ratio of soil to solution during extraction can range from 1:2 to 1:5. Soil extraction with NaOH leads to isolating 80% of organic matter. The effectiveness of the method increases by rinsing soil with the solution of hydrochloric acid since calcium ions and other multicharge cations are eliminated [18-20]. The modified IHSS method suggests the use of 0.1M NaOH and the ratio of soil to the solution during extraction of 1:10 [21]. Organic soils or peats containing higher carbon concentrations are usually extracted in the ratio 1:50 (w/v) [6]. In the research of bottom sediments of different lakes, Ishiwatari [7] (cited after [8]) noted an increase in the concentration of extractable carbon of humic and fulvic acids during 1-month extraction while applying 0.1M NaOH as compared with the 6-hour extraction.

The aim of the present paper was to determine the effect of the extraction time on the alkaline properties of humic substances extracts from organic and mineral lake bottom sediments.

MATERIAL AND METHODS

The material was made up of bottom sediments of lakes of varied catchment character. In the catchments of Lakes Hanki, Pniewo and Arkonskie woodland dominates, whereas the catchment of Lakes Wdzydze, Kolmowo and Bobrow are mainly the areas under agricultural use. Bottom sediments were sampled with Ekman catcher. In dried sediment samples the contents of total carbon (TC) and total nitrogen (Nt) were determined with the Vario Max CN analyzer (Elementar, Germany). The content of inorganic carbon (IC) was analyzed with the Primacs³C Analyser (Skalar, Breda, Netherlands). On the basis of these results, calculations were made of the contents of organic carbon (TOC) from the difference TOC=TC-IC, as well as values of the TOC:Nt ratio. In the sediment samples analyzed pH in water (1:5 w/v ratio) was also determined. Having been decalcified with 0.2M HCl, the extraction of humus substances with 0.5M NaOH was made, in two variants 1:10 and 1:50 (w/v,) both for organic (carbonate-free sediments) and
mineral samples (carbonate sediments). To maintain the same extraction conditions, in the case of carbonate samples, the initial weighted amounts were increased by the content of carbonates. The extraction was made for 1, 5, 12, 24 and 48 h. In the extracts obtained (HA+FA mixture) the content of organic carbon was determined with the analyzer TOCN Formacs by Skalar and the spectrometric properties in the UV-VIS range with Perkin-Elmer Lambda 20 UV-VIS Spectrometer. Based on the values recorded there was calculated the efficiency of extraction at respective stages, expressed as a percentage of organic carbon of the initial sample to the value of the absorbance ratios: \( \frac{A_{280}}{A_{465}} \), \( \frac{A_{280}}{A_{665}} \), and \( \frac{A_{465}}{A_{665}} \).

**RESULTS AND DISCUSSION**

Table 1 presents basic physicochemical properties of the bottom sediments researched. The lake bottom sediments showed considerable variation in the content of organic matter. The applicable literature shows that the content of organic carbon in bottom sediments can range from 29 g kg\(^{-1}\) [5] to about 460 g kg\(^{-1}\) [2]. The organic sediments researched contained from 188 to 386 g kg\(^{-1}\), however, they did not include inorganic carbon. The content of total nitrogen ranged from 14.1 to 34.8 g kg\(^{-1}\). The highest contents of organic carbon and total nitrogen were recorded for the sediment sampled from Lake Arkonskie. The sediment pH ranged from 4.60 to 6.45.

Mineral sediments showed a lower content of organic carbon (TOC) than organic sediments. The highest content of organic carbon was recorded in the sample from Lake Wdzydze. The content of inorganic carbon (IC) in the samples

**TABLE 1. BASIC PHYSICOCHEMICAL PROPERTIES OF THE BOTTOM SEDIMENTS**

<table>
<thead>
<tr>
<th>Lake and sample number</th>
<th>TOC (g kg(^{-1}))</th>
<th>IC</th>
<th>Nt</th>
<th>TOC:Nt</th>
<th>pH(_{2\text{O}})</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Organic sediments</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hanki (1)</td>
<td>339</td>
<td>0.0</td>
<td>23.9</td>
<td>14.2</td>
<td>6.45</td>
</tr>
<tr>
<td>Pniewo (2)</td>
<td>188</td>
<td>0.0</td>
<td>14.1</td>
<td>15.7</td>
<td>4.60</td>
</tr>
<tr>
<td>Arkonskie (3)</td>
<td>386</td>
<td>0.0</td>
<td>34.8</td>
<td>11.1</td>
<td>5.20</td>
</tr>
<tr>
<td><strong>Mineral sediments</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wdzydze (4)</td>
<td>166</td>
<td>27.0</td>
<td>15.7</td>
<td>10.6</td>
<td>7.30</td>
</tr>
<tr>
<td>Kolmowo (5)</td>
<td>140</td>
<td>12.6</td>
<td>16.1</td>
<td>8.7</td>
<td>6.54</td>
</tr>
<tr>
<td>Bobrow (6)</td>
<td>128</td>
<td>7.1</td>
<td>11.2</td>
<td>11.4</td>
<td>6.98</td>
</tr>
</tbody>
</table>
analyzed ranged from 7.1 to 27.0 g kg\(^{-1}\). The content of total nitrogen ranged from 11.2 to 16.1 g kg\(^{-1}\). As compared with organic sediment samples, the mineral sediment samples showed higher pH values, ranging from 6.54 to 7.30.

Based on the value of the TOC:N\(t\) ratio, one can determine the origin of organic matter found in bottom sediments. The values of the ratio below 10 suggest that the main source of organic matter is phytoplankton, values 13-14 for surface sediments point to the mixture in which the contents of detritus from vascular plants and algae are similar, while higher values show a considerable share of organic matter of land origin in the pool of organic matter of sediments [9, 13, 15, 16]. The values recorded demonstrate that only in the case of Lake Kolmowo (sample No. 5), the main source of organic matter was phytoplankton, whereas the sediments of the other lakes contain organic matter of both autochthonic and allochthonic origin.

Figures 1 and 2 present the content of organic carbon in the alkaline extracts of humus substances from bottom sediments and the efficiency of the extraction process. With time in most cases the carbon content increases, usually reaching the maximum after 48 h. As for organic samples extracted in the ratio 1:10 the highest
content of carbon was noted in the extract from the sample from Lake Hanki (sample No. 1), while in the extract 1:50 – in the mixture HA+FA extracted from Lake Arkonskie sediments (sample No. 3, after 24 hours) (Fig. 1). Similarly, with time the extraction efficiency increased. Both in the case of extraction in the ratio 1:10 (w/v) and 1:50 (w/v) the highest efficiency was recorded for the sample from Lake Pniewo (sample No. 2). An especially high efficiency was recorded for the extraction made in the ratio 1:50 (w/v).

The extracts from the mineral sediment samples demonstrated the highest concentrations of organic carbon after 48 h. The highest carbon content for the samples extracted in the ratio 1:10 (w/v) was recorded for the extract from sediments from Lake Kolmowo (sample no 5). As for the samples extracted in the ratio 1:50 (w/v), most carbon was contained in the mixture HA+FA extracted from the sediments of Lake Wdzydze (sample no 4). The contents were 7502 and 2205 mg L⁻¹, respectively (Fig. 2). The increase in the content of carbon in extracts is confirmed by the earlier results reported by Ishiwatari [7], although the differences
were reported already after a considerably shorter time. The concentrations of carbon of humic and fulvic acids contained in the bottom sediments are much higher than those reported for soils [4].

The values of absorbance at the wavelength of 280 nm are related to the presence of structures of lignin origin [10, 11, 17], at the wavelength of 465 nm – to the content of matter at the initial phase of decomposition, whereas at the wavelength of 665 nm – they are related to the presence of substances of a high degree of humification [1]. UV-VIS spectra of humic acids formed in an aqueous environment show the presence of an additional band at a wavelength of approximately 660 nm; that band is specially intensive in humic acids extracted from bottom sediments of lakes containing carbonates which help preserving pigments [2], however it makes the interpretation of both the absorbance value at the wavelength of 665 nm and the values of calculated absorbance ratios difficult.

Tables 2 and 3 present the absorbance values of solutions of the mixture of humic and fulvic acids extracted from organic sediments. The values of absorbance were increasing with extraction time, in general, reaching the

TABLE 2. SPECTROMETRIC PROPERTIES OF ALKALINE EXTRACTS FROM ORGANIC SEDIMENTS – 1:10 RATIO (ABSORBANCE VALUES GIVEN FOR DILUTED SOLUTIONS IN 1:50 RATIO)

<table>
<thead>
<tr>
<th>Lake and sample number</th>
<th>Time (h)</th>
<th>A_{280}</th>
<th>A_{465}</th>
<th>A_{665}</th>
<th>A_{2/4}</th>
<th>A_{2/6}</th>
<th>A_{46}</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hanki (1)</td>
<td>1</td>
<td>1.8479</td>
<td>0.3038</td>
<td>0.0503</td>
<td>6.08</td>
<td>36.74</td>
<td>6.04</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>2.3583</td>
<td>0.3703</td>
<td>0.0605</td>
<td>6.37</td>
<td>38.98</td>
<td>6.12</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>3.1461</td>
<td>0.4973</td>
<td>0.0889</td>
<td>6.33</td>
<td>35.39</td>
<td>5.59</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>2.7522</td>
<td>0.4286</td>
<td>0.0716</td>
<td>6.42</td>
<td>38.44</td>
<td>5.99</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>3.9969</td>
<td>0.6252</td>
<td>0.1156</td>
<td>6.39</td>
<td>34.58</td>
<td>5.41</td>
</tr>
<tr>
<td>Pniewo (2)</td>
<td>1</td>
<td>1.3836</td>
<td>0.2033</td>
<td>0.0262</td>
<td>6.81</td>
<td>52.81</td>
<td>7.76</td>
</tr>
<tr>
<td></td>
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<td>1.5721</td>
<td>0.2233</td>
<td>0.0299</td>
<td>7.04</td>
<td>52.58</td>
<td>7.47</td>
</tr>
<tr>
<td></td>
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<td>2.4098</td>
<td>0.3373</td>
<td>0.0476</td>
<td>7.14</td>
<td>50.63</td>
<td>7.09</td>
</tr>
<tr>
<td></td>
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<td>0.3535</td>
<td>0.0508</td>
<td>7.12</td>
<td>49.54</td>
<td>6.96</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>2.6931</td>
<td>0.3787</td>
<td>0.0574</td>
<td>7.11</td>
<td>46.92</td>
<td>6.60</td>
</tr>
<tr>
<td>Arkonskie (3)</td>
<td>1</td>
<td>1.0297</td>
<td>0.1548</td>
<td>0.0209</td>
<td>6.65</td>
<td>49.27</td>
<td>7.41</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>1.5546</td>
<td>0.2191</td>
<td>0.0294</td>
<td>7.10</td>
<td>52.88</td>
<td>7.45</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>2.2919</td>
<td>0.3276</td>
<td>0.0456</td>
<td>7.00</td>
<td>50.26</td>
<td>7.18</td>
</tr>
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<td>1.9066</td>
<td>0.269</td>
<td>0.0373</td>
<td>7.09</td>
<td>51.12</td>
<td>7.21</td>
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<td>48</td>
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<td>0.4073</td>
<td>0.0606</td>
<td>7.18</td>
<td>48.27</td>
<td>6.72</td>
</tr>
</tbody>
</table>
maximum after 48 h. The highest values of absorbance at the wavelengths analyzed were reported for the extracts from the sediment sample from Lake Hanki (sample No. 1).

The value of the calculated ratio of absorbance $A_{2/4}$ in most cases increased in time, while the value of ratios $A_{2/6}$ and $A_{4/6}$ decreased for extracts 1:10 (w/v) (Table 2). For mixture HA+HF obtained as a result of extraction in the ratio 1:50 (w/v), in general, there was observed an increase in the value of ratio $A_{2/4}$, while the other values of ratios increased or decreased depending on the sample.

Interestingly, however, the values of absorbance ratios are lower for the samples extracted in the ratio of 1:10 (w/v) as compared with the samples extracted in the ratio 1:50 (w/v) (Table 3).

Similarly, the extracts from mineral sediments showed the highest absorbance values after 48 h (Tables 4 and 5). The highest values of absorbance at the wavelengths analyzed were reported for the extracts samples from Lake Kolmowo sediments (sample no 5). It is difficult to determine the tendencies of changes in the

**TABLE 3. SPECTROMETRIC PROPERTIES OF ALKALINE EXTRACTS FROM ORGANIC SEDIMENTS – 1:50 RATIO (ABSORBANCE VALUES GIVEN FOR DILUTED SOLUTIONS IN 1:10 RATIO)**

<table>
<thead>
<tr>
<th>Lake and sample number</th>
<th>Time (h)</th>
<th>$A_{280}$</th>
<th>$A_{365}$</th>
<th>$A_{665}$</th>
<th>$A_{2/4}$</th>
<th>$A_{2/6}$</th>
<th>$A_{4/6}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hanki (1)</td>
<td>1</td>
<td>1.5338</td>
<td>0.2272</td>
<td>0.0348</td>
<td>6.75</td>
<td>44.07</td>
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</tr>
<tr>
<td></td>
<td>5</td>
<td>2.1713</td>
<td>0.3085</td>
<td>0.0471</td>
<td>7.04</td>
<td>46.10</td>
<td>6.55</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>2.5444</td>
<td>0.3478</td>
<td>0.0542</td>
<td>7.32</td>
<td>46.94</td>
<td>6.42</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>4.3003</td>
<td>0.5460</td>
<td>0.0666</td>
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<td>64.57</td>
<td>8.20</td>
</tr>
<tr>
<td></td>
<td>48</td>
<td>5.1733</td>
<td>0.6456</td>
<td>0.0807</td>
<td>8.01</td>
<td>64.11</td>
<td>8.00</td>
</tr>
<tr>
<td>Pniewo (2)</td>
<td>1</td>
<td>1.4497</td>
<td>0.1888</td>
<td>0.0238</td>
<td>7.68</td>
<td>60.91</td>
<td>7.93</td>
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<tr>
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<td>1.7152</td>
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<td>7.86</td>
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<td>1.7349</td>
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<td>0.0295</td>
<td>7.90</td>
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<td>1.9977</td>
<td>0.2576</td>
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<tr>
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<td>48</td>
<td>2.1730</td>
<td>0.2801</td>
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<td>7.76</td>
<td>52.62</td>
<td>6.78</td>
</tr>
<tr>
<td>Arkonskie (3)</td>
<td>1</td>
<td>1.1645</td>
<td>0.1553</td>
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<td>0.3912</td>
<td>0.0518</td>
<td>8.38</td>
<td>63.31</td>
<td>7.55</td>
</tr>
</tbody>
</table>
value of ratio $A_{2/4}$, while the values of ratios $A_{2/6}$ and $A_{4/6}$, irrespective of the extraction variant were decreasing, in general.

Lower values of ratios $A_{2/6}$ and $A_{4/6}$ can suggest an increase in the concentrations of substances of a high degree of humification and such interpretation can be applied to organic samples since, in the case of mineral sediments, the increase in the absorbance value at 665 nm can be still related to the presence of structures of pigment origin [2, 3].

CONCLUSIONS

The results presented suggest that organic sediments contain much less organic matter permanently bonded with the mineral fraction of sediments, which can be seen from a considerable increase in extraction efficiency when the ratio of 1:50 (w/v) is applied during extraction. Changes in the value of the absorbance ratios calculated show that at each stage compounds of varied properties are extracted, depending both on the sediment type (mineral – organic), and on the properties of respective samples.
TABLE 5. SPECTROMETRIC PROPERTIES OF ALKALINE EXTRACTS FROM MINERAL SEDIMENTS – RATIO 1:50 (ABSORBANCE VALUES GIVEN FOR DILUTED SOLUTIONS IN 1:10 RATIO)

<table>
<thead>
<tr>
<th>Lake and sample number</th>
<th>Time (h)</th>
<th>A_{280}</th>
<th>A_{465}</th>
<th>A_{665}</th>
<th>A_{2/4}</th>
<th>A_{2/6}</th>
<th>A_{4/6}</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wdzydze (4)</td>
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<td>0.1241</td>
<td>0.0239</td>
<td>6.22</td>
<td>32.32</td>
<td>5.19</td>
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<td>0.9602</td>
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<td>0.0368</td>
<td>6.81</td>
<td>34.60</td>
<td>5.08</td>
</tr>
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<td>0.0438</td>
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<tr>
<td>Kalmowo (5)</td>
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<td>45.42</td>
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REFERENCES

Wpływ czasu ekstrakcji na właściwości ekstraktów alkalicznych substancji humusowych

Celem pracy było zbadanie wpływu czasu ekstrakcji substancji humusowych (SH) z organicznych i mineralnych osadów dennych jezior na właściwości ekstraktów alkalicznych. Ekstrakcję przeprowadzono 0.5M NaOH przy stosunku próbki do ekstrahenta jak 1:10 i 1:50 (w/v). Dla obu wariantów czas ekstrakcji wynosił 1, 5, 12, 24 i 48 godzin. Dla otrzymanych ekstraktów SH oznaczono: zawartość węgla organicznego oraz widma w zakresie UV-VIS, na podstawie których obliczono wartości stosunków absorbancji. Badania wykazały istotny wpływ czasu ekstrakcji na właściwości alkalicznych ekstraktów SH. Roztywki po dłuższym czasie ekstrakcji zawierały więcej węgla organicznego, a ekstrakcja prowadzona przy stosunku próbki do ekstrahenta jak 1:50 (w/v) była wydajniejsza, niż prowadzona przy wartości tego stosunku jak 1:10 (w/v).