Studying the inhibitory effect of *Populus tremula* bark extract on the activity of enzymes

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Abstract. Opisthorchiasis is one of the most prevalent helminthisms in Russia. *Populus tremula* bark extract is used as anthelminthic remedy. It was shown that the extract contains salireposide, salicin, caffeic acid, salicyloyl salicin, tremulacin. The purpose of this paper was to study inhibitory effect of these substances on the enzyme activity. Yeast leucine aminopeptidase, yeast ATPase and human salivary amylase were tested. The study showed that the *P. tremula* bark extract inhibits all three enzymes, but although examined substances show the inhibitory effect on the enzymes none of them is as effective as the extract.

Opisthorchiasis is one of the most prevalent helminthisms in Russia. The Western Siberia region is known to be among the most dangerous locations of opisthorchiasis in the world. The infection rate among the local population in the area is considered to be 70-80 % [1]. Opisthorchiasis may cause development of cholangiocarcinoma [2].

*Populus tremula* bark drug is used as anthelminthic remedy [3]. Photocchemical analysis showed presence of salireposide (1), salicyloyl salicin (2), salicin (3), tremulacin (4), caffeic acid (5), ferulic acid (6), vanillic acid (7) in *P. tremula* bark [4].

Information regarding effect of particular substances contained in *P. tremula* bark on enzymes is lacking. Thus, as part of our ongoing in vitro characterization of this substances as Opisthorchis enzymes inhibitors, we...
have investigated inhibitory effect of substances 1-5 and *P.tremula* bark extract on enzymes, obtained from different available sources.

Three enzymes were studied: leucine aminopeptidase, ATPase, amylase. As a source of enzymes yeast extract was used for leucine aminopeptidase and ATPase and human saliva for amylase. Inhibitory effect analysis was carried out according to 5. The results are shown in table 1.

### Table 1. Effect of substances on enzymes

<table>
<thead>
<tr>
<th></th>
<th>leucine peptidase</th>
<th>ATPase</th>
<th>amylase</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>% inhibition</td>
<td>Conc., μM</td>
<td>% inhibition</td>
</tr>
<tr>
<td><em>P.tremula</em> bark extract</td>
<td>46,8</td>
<td>0,54 mg/ml</td>
<td>16,5</td>
</tr>
<tr>
<td>Tremulacin</td>
<td>49,67</td>
<td>1,01</td>
<td>19,07</td>
</tr>
<tr>
<td>Salireposide</td>
<td>55,95</td>
<td>1,31</td>
<td>0</td>
</tr>
<tr>
<td>Caffeic acid</td>
<td>77,78</td>
<td>2,96</td>
<td>15,5</td>
</tr>
<tr>
<td>Saliciroyl salicin</td>
<td>25,88</td>
<td>1,31</td>
<td>4,6</td>
</tr>
<tr>
<td>Salicin</td>
<td>0</td>
<td>1,86</td>
<td>20</td>
</tr>
</tbody>
</table>

Percent inhibition value was calculated using eq. 1 for leucine aminopeptidase and ATPase, and using eq. 2 for amylase.

\[
\text{%inhibition value} = \left( \frac{D_{\text{control}} - D_{\text{sample}}}{D_{\text{sample}}} \right) * 100\% 
\]

\[
\text{%inhibition value} = \left( 1 - \frac{D_{\text{control}} - D_{\text{sample}}}{D_{\text{control}} - D_{\text{enzyme}}} \right) * 100
\]

D – optical density.

The findings suggest that the *P.tremula* bark extract is capable of inhibiting all three enzymes, but none of examined substances is as effective as the extract. It may be explained by synergetic action of several extract components or by presence of more effective inhibitors with unspecified structure or undetected inhibitory activity in *P.tremula* bark extract.

### References