THE USE OF THE INDICATOR PARASITE *ANISAKIS SIMPLEX* IN POPULATION SURVEYS OF PACIFIC HERRING *CLUPEA PALLASII* (CLUPEIFORMES: CLUPEIDAE) IN SAKHALIN WATERS

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**Abstract**

Quantitative indices of herring infestation in Sakhalin waters reveal a high infection rate of fish in the basin of the Sea of Okhotsk, off the east coast of the island. Moreover, in certain years almost 100% of herring were infested. The so-called local populations as well as widely migrating Sakhalin Hokkaido herring are confined to the area of this waterbody. In the basin of the Sea of Japan, off the west coast of Sakhalin, the indicator values of herring infestation are much lower. The local population as well as widely migrating herring of Sakhalin Hokkaido population are confined to this marine area. The causality analysis of herring infestation in these areas allows to confirm our earlier conclusions about population structure of herring inhabiting the waters around Sakhalin Island.

**Key words:** herring, nematodes, anisakis, extensiveness of invasion, intensity of invasion, index of abundance

**INTRODUCTION**

Parasitological surveys of fish were conducted in the northwestern part of the Pacific Ocean in different years. As a result, a great number of disparate faunistic and systematic works have been published, including materials on fish nematodes in the studied region (Oshima 1972, Kurochkin 1974, Solovieva 1994, Vyalova 2003, Vyalova and Vinogradov 2003, Gaevskaya 2005, Butorina 2006, Rybnikova et al. 2009, Rybnikova and Pushnikova 2010, Pushnikova and Rybnikova 2012, Aseeva et al. 2013). According to Solovieva (1994), mass parasites registered in all the Far Eastern seas are larvae of the nematode *Anisakis simplex* found in 42 species of fish, including the Pacific herring (56.6% infestation rate).
Analyzing the role of cod in the life cycle of *A. simplex* in the Baltic Sea, the researchers note that infestation of these predatory fish with the third-stage larvae occurs through the herring which is the main food source for cod. The latter, in its turn, receives anisakis larvae at the same third stage of development when feeding on crustaceans, mainly euphausiids. Actually, in Baltic coastal waters herring is the main source of infection with anisakid nematodes not only to cod, but also to pike perch, since it is a significant part of the pike perch diet (Gaevskaya 2005).

The use of anisakids as biological indicators of the population structure and the biological characteristics of their hosts confirms the earlier revealed differentiation pattern. This method is particularly important in studying the population structure of animals, their stock separation and the organization of rational fishing.

Quantitative indices of infestation with the larvae of *A. simplex* were used to distinguish populations of pink salmon in Sakhalin waters (Vyalova 2003). Indicator parasites allowed to solve the problem of differentiation of local stocks of sockeye salmon (Konovalov 1971). Differences in the intensity of invasion of Alaska pollock, surveyed in the waters around Hokkaido Island, with the larvae of *A. simplex* and *Contracaecum osculatum* allowed to recommend the given nematodes as bioindicators for stock separation of this mass and commercially important fish (Konishi and Sakurai 2002).

Five species of helminths, including *A. simplex*, *Contracaecum sp.*. and *Hysterothy- lacium sp.*, were used as biological tags for stock separation in Pacific herring in California waters (Moser and Hsieh 1992).

Due to the significant differences in the species composition of helminths, including anisakids, and the frequency of their occurrence in flying squid (*Ommastrephes bar-trami*), it was concluded that in the eastern and western North Pacific Ocean it formed two large groupings which did not mix during the spawning period (Bower and Margolis 1991).

An excursion into the history of the use of anisakids as biological indicators in population surveys of fish and invertebrates shows how useful these parasites can be in such cases. A parasitological method is especially effective when used in conjunction with other methods, for example, ecogeographical, morphometric or genetic methods.

Information about infestation of Pacific herring with the larvae of this parasite is vital and of great practical interest. As a mass type of parasite, sufficiently large and easily recognizable larvae of *A. simplex* can be used as indicator parasites of the commercially exploited Pacific herring populations.

**MATERIAL AND METHODS**

The paper is based on samples of spawning Pacific herring caught in the Sea of Japan and the Sea of Okhotsk (Fig. 1). In performing biological analysis of newly caught herring, nematode larvae were retrieved from the body cavity of fish; the number of larvae was counted in each individual. The data for the study were collected in 1977-2008. Over 5,000 fish samples were analyzed.
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**RESULTS AND DISCUSSION**

Quantitative indices of herring infestation in Sakhalin waters reveal a high infection rate of fish in the basin of the Sea of Okhotsk, off the east coast of the island. Moreover, in certain years almost 100% of herring were infested (Pushnikova and Rybnikova 2012). The so-called local populations as well as widely migrating Sakhalin Hokkaido herring are confined to the area of this waterbody. In the basin of the Sea of Japan, off the west coast of Sakhalin, the indicator values of herring infestation are much lower. The local population (De Kastri population) as well as widely migrating herring of Sakhalin Hokkaido population are confined to this marine area.

A comparative analysis of herring infestation with anisakis larvae in different studied areas revealed the spots with the highest and lowest rates of infestation (Table 1). A high degree of invasion with the larvae of *A. simplex* was noted in spawning her-
ring in the southern part of Peter the Great Bay (Solovyeva 2002), in Lake Tunaicha, in Sovetskaya Gavan Bay and off the northeast coast of Sakhalin Island. Low rates of infestation were found in surveyed herring caught in waters off southwestern Sakhalin and in Sakhalin Gulf (see Table 1). In the early 1990s very high infestation rates in herring with anisakids were observed in the areas of the Sea of Okhotsk basin. Thus, the extensiveness of invasion during this period in herring in the Bay of Patience was 95%, in Lake Tunaicha 80%, in the bays of the northeastern coast of Sakhalin it ranged from 50% to 83%, in Sakhalin Gulf was 59.3%. For the waterbody of the northern part of the Sea of Japan (the Tatar Strait), these indices were much lower: 23% off southwestern Sakhalin, 14% off northwestern Sakhalin.

Table 1
Multiyear data on the infestation of herring with the larvae of *Anisakis simplex* in different areas of the Seas of Japan and Okhotsk

<table>
<thead>
<tr>
<th>Material</th>
<th>Fish length, cm</th>
<th>Number of fish</th>
<th>Extensiveness of invasion, %</th>
<th>Intensity of invasion</th>
<th>Index of abundance</th>
</tr>
</thead>
<tbody>
<tr>
<td>area</td>
<td>years</td>
<td>fluctuations</td>
<td>mean</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1. Peter the Great Bay, Posyet Bay, Narva Bay, Ussuri Bay, Amur Bay, Razdolnaya River</td>
<td>1996-2001</td>
<td>22.0-39.0</td>
<td>29.05</td>
<td>108</td>
<td>82.3</td>
</tr>
<tr>
<td>1992</td>
<td>26.0-32.0</td>
<td>29.29</td>
<td>100</td>
<td>23.0</td>
<td>1-12</td>
</tr>
<tr>
<td>1994-1995</td>
<td>20.0-37.5</td>
<td>28.04</td>
<td>296</td>
<td>32.3</td>
<td>1-15</td>
</tr>
<tr>
<td>3. Sovetskaya Gavan Bay</td>
<td>2000</td>
<td>22.0-37.0</td>
<td>34.08</td>
<td>100</td>
<td>95.0</td>
</tr>
<tr>
<td>4. Northwestern Sakhalin</td>
<td>1977-1979</td>
<td>20.5-28.0</td>
<td>25.20</td>
<td>231</td>
<td>14.5</td>
</tr>
<tr>
<td>1985-1986</td>
<td>19.0-31.5</td>
<td>26.80</td>
<td>342</td>
<td>51.4</td>
<td>1-9</td>
</tr>
<tr>
<td>1990-1991</td>
<td>21.5-31.5</td>
<td>26.34</td>
<td>150</td>
<td>14.0</td>
<td>1-10</td>
</tr>
<tr>
<td>5. Aniva Bay</td>
<td>1977-1979</td>
<td>17.5-27.0</td>
<td>19.60</td>
<td>100</td>
<td>20.0</td>
</tr>
<tr>
<td>6. Lake Tunaicha, southeastern Sakhalin</td>
<td>1977-1979</td>
<td>12.2-25.5</td>
<td>19.30</td>
<td>371</td>
<td>30.7</td>
</tr>
<tr>
<td>1992</td>
<td>15.0-29.0</td>
<td>20.37</td>
<td>200</td>
<td>80.0</td>
<td>1-155</td>
</tr>
<tr>
<td>7. Gulf of Patience, Lake Nevskoye</td>
<td>1977-1979</td>
<td>17.3-27.0</td>
<td>22.60</td>
<td>100</td>
<td>47.0</td>
</tr>
<tr>
<td>1982-1983</td>
<td>17.5-32.0</td>
<td>24.00</td>
<td>1,345</td>
<td>37.85</td>
<td>1-45</td>
</tr>
<tr>
<td>1993</td>
<td>19.5-31.5</td>
<td>24.04</td>
<td>100</td>
<td>95.0</td>
<td>2-33</td>
</tr>
<tr>
<td>1981</td>
<td>21.0-26.5</td>
<td>23.44</td>
<td>99</td>
<td>61.6</td>
<td>1-15</td>
</tr>
<tr>
<td>8. Northeastern Sakhalin</td>
<td>1992,1998</td>
<td>15.5-31.2</td>
<td>25.56</td>
<td>400</td>
<td>72.5</td>
</tr>
<tr>
<td>2003</td>
<td>25.4-32.0</td>
<td>26.0</td>
<td>50</td>
<td>80.0</td>
<td>1-14</td>
</tr>
<tr>
<td>10. North Sakhalin</td>
<td>2003</td>
<td>25.4-32.0</td>
<td>28.80</td>
<td>50</td>
<td>80.0</td>
</tr>
<tr>
<td>11. Iony-Kashevarov area</td>
<td>2006</td>
<td>26.0-32.0</td>
<td>28.74</td>
<td>57</td>
<td>50.0</td>
</tr>
</tbody>
</table>
On the whole, it was noted that the lowest degree of infestation was characteristic of herring which inhabited waters off the western Sakhalin coast. And, despite the revealed tendency toward the growth of invasion rates, herring inhabiting waters off the southwestern Sakhalin (the Sakhalin-Hokkaido population) were least infected. In addition, we recorded increased infestation in fish in local groupings, including lake herrings (Lake Tunaicha, Lake Nevskoye) (Rybnikova et al. 2009, Rybnikova and Pushnikova 2015).

The causality analysis of herring infestation in these areas allowed to confirm our earlier conclusions about population structure of herring inhabiting the waters around Sakhalin island.

The allozyme study of Sakhalin herring made a great contribution to population and genetic surveys of Pacific herring. It was found that Pacific herring had a distinctive intraspecific population structure. In the Seas of Japan and Okhotsk there are several population totalities that can be interpreted as genetically isolated local stocks. Taking into account the significant interannual genetic variability within individual populations, as well as the peculiarities of population discrimination, we can presume that the local stock in Pacific herring is a subdivided or genetically heterogeneous population.

Genetically independent local stocks (subdivided populations) include: 1) local populations inhabiting Primorye waters and Sakhalin Gulf, 2) population totality of Sakhalin-Hokkaido herring, 3) totality of local populations inhabiting waters off northeastern Sakhalin and Lake Tunaicha (Fig. 2).

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Fig. 2. The two-dimensional distribution of the values of the roots of canonical variables (RCV) obtained by performing the discriminant analysis of five populations (1-5) of Pacific herring *Clupea pallasii*

1 – Primorye population, 2 – Sakhalin-Hokkaido population, 3 – Sakhalin Gulf population, 4 – Lake Tunaicha population, 5 – northeastern Sakhalin population; the dotted lines show different local stocks
It can be presumed that the local stock in Pacific herring is a subdivided or genetically heterogeneous population. The population consists of dynamic elements – subpopulations exchanging migrants among themselves. Differences between separate subdivided populations are supported comprehensively due to differentiating natural selection and gene drift (Rybnikova et al. 1998, Kartavtsev and Rybnikova 1999). It should be noted that the results of studying the allozyme variability of Pacific herring are consistent with the data obtained for the North-Okhotsk herring grouping as assessed by RAPD markers (Lapinsky et al. 2008), as well as with the data on the variability of the mitochondrial DNA control region in studying the intraspecies structure of Pacific herring found in the Sea of Japan and the southern Sea of Okhotsk (Gorbachev et al. 2011).

With the help of discriminant and factor analysis of characteristics or morphometric indices, variability and differentiation of multidimensional vectors-individuals into groups which have a clear biological meaning in representing local stocks of Pacific herring, were revealed. The obtained data of multivariate analysis of morphometric features confirm the previously published morphobiological and genetic information on the availability of at least three local stocks of Pacific herring in Sakhalin waters. The local stock of Sakhalin-Hokkaido herring, which was the main source of catches in the past in Sakhalin area, reveals a distinct morphological differentiation that distinguishes it from other herring groupings in the surveyed area (Fig. 3) (Kartavtsev et al. 2008).

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**Fig. 3.** The two-dimensional distribution of the values of the roots of canonical variables (RCV) obtained by performing the discriminant analysis of five populations (1-5) of Pacific herring *Clupea pallasii* and using allozyme loci and biological characteristics.
CONCLUSION

When analyzing the infestation of Pacific herring with the larvae of *A. simplex* in Sakhalin waters we found that indices of the extensiveness of invasion, the intensity of invasion and the abundance index in different areas differed due to the confinement of fish to different populations and to certain water bodies. The peculiarities of the regime of Sakhalin waters determine the distinctiveness of the seasonal dynamics of herring infestation. *A. simplex* larvae can be used as indicator parasites of Pacific herring populations commercially exploited by the fishery.

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WYKORZYSTANIE PASOŻYTA ANISAKIS SIMPLEX JAKO WSKAŹNIKA W BADANIACH POPULACYJNYCH ŚLEDZIA PACYFICZNEGO CLUPEA PALLASII (CLUPEIFORMES: CLUPEIDAE)

Streszczenie

Ilościowe wskaźniki skażenia śledzia pacyficznego w wodach Sachalinu wskazują na wysokie zanieczyszczenie ryb w dorzeczu Morza Ochockiego, u wschodniego wybrzeża wyspy. Co więcej, w niektórych latach śledź został zainfekowany prawie w 100%. Lokalne populacje śledzia, a także szeroko migrująca Sachalińsko-Hokkaidowska populacja śledzia są ograniczone do obszaru wodnego tego zbiornika. W basenie Morza Japońskiego, u zachodniego wybrzeża Sachalinu, wartości wskaźników zainfekowania śledzia są znacznie niższe. Podobnie lokalne i szeroko migrująca Sachalińsko-Hokkaidowska populacje śledzia są również ograniczone do tego obszaru wodnego. Analiza związków przyczynowo-skutkowych zainfekowania śledzi w tych obszarach pozwala potwierdzić uzyskane wcześniej wyniki dotyczące struktury populacji śledzi wód otaczających wyspę Sachalin.