The liver fluke Opisthorchis felineus: biology, epidemiology and carcinogenic potential

Mariya Y. Pakharukova and Viatcheslav A. Mordvinov

The liver fluke Opisthorchis felineus is a member of the triad of epidemiologically important liver fluke species belonging to the family Opisthorchiidae and the major agent causing opisthorchiasis over a vast territory, covering Russia, Kazakhstan and several European countries. The similarity between the diseases caused by O. felineus and other liver flukes, O. viverrini and Clonorchis sinensis, in clinical manifestations and course suggests that the scenarios of their development and, possibly, complications have much in common. The International Agency for Research on Cancer classified O. viverrini and C. sinensis as group 1 agents and the major factors inducing cholangiocarcinoma in endemic regions. However, a carcinogenic potential of O. felineus is poorly studied. This review characterizes O. felineus, briefs the epidemiological situation in Western Siberia, the world’s largest opisthorchiasis focus, and assesses the carcinogenic potential of this liver fluke. The review is based on a comprehensive analysis of the published medical data on opisthorchiasis and its complications in Western Siberia. Results of performed analysis reflect the actual epidemiological situation in opisthorchiasis focus and suggest an association of this disease with bile duct cancer.

Keywords: Life cycle stages, Liver cancer, Liver fluke, Opisthorchis felineus, Russia

Introduction

The liver flukes Opisthorchis felineus (Rivolta, 1884), O. viverrini (Poirier, 1886), and Clonorchis sinensis (Loas, 1907) are the three epidemiologically significant species of the family Opisthorchiidae (class Trematoda, phylum Platyhelminthes), affecting the human and animal liver and bile ducts. The International Agency for Research on Cancer classified the liver flukes O. viverrini and C. sinensis as group 1 agents, i.e., the agents carcinogenic to humans, and as the major factors inducing cholangiocarcinoma in endemic regions.

These members of the family Opisthorchiidae have been described in numerous research papers, comprehensively summarized in several recent reviews. This review focuses on the biology, epidemiology, and carcinogenic potential of O. felineus, the parasite presenting a serious public health threat in several regions of Eurasia.

Opisthorchiasis caused by O. felineus is spread over the territory of the former Soviet Union and Eastern Europe. The foci of this disease are confined to river plains. The West Siberian Lowland, one of the largest lowland plains of the globe, is most illustrative in this case, since it houses the largest world focus of opisthorchiasis caused by O. felineus in the Ob–Irtysh basin.

Although opisthorchiasis caused by O. felineus is endemic in Eastern Europe and Western Siberia, focal outbreaks causing significant human morbidity can also occur in Western Europe. O. felineus has been found in the Iberian and Balkan Peninsulas, Germany and Italy. According to preliminary estimates, at least 1.6 million people are infected with O. felineus of the total of 17 millions infested with the opisthorchid flukes.

From the standpoint of the official medical statistics, cholangiocarcinoma is not among the major cancer types in Russia. Unfortunately, the consolidated data of official statistics are inappropriate to clarify whether there is a correlation between cholangiocarcinoma and opisthorchiasis caused by O. felineus.

For this review, we have analyzed the published Russian medical data on the epidemiology of opisthorchiasis and its complications in Western Siberia. In our opinion, the results of this analysis reflect the current epidemiological situation in the Ob–Irtysh opisthorchiasis focus and suggest a direct association between this disease and bile duct cancer.

Methods

For this review, we analyzed the relevant original research papers as well as statistical reports of clinics and morgues published in leading reviewed Russian medical research and applied journals.
The main sources for the literature search were Pubmed, Elibrary (http://elibrary.ru/defaultx.asp) and Google Scholar (http://scholar.google.com/) from 1970–2015. Keywords used (in Russian and in English) were: opisthorchiasis, Opisthorchis felineus, liver flukes, Russia, Western Siberia, Tobolsk, Tyumen, Novosibirsk, Tomsk, liver cancer, bile duct cancer, cholangiocarcinoma, cholangitis, autopsy. Only full text papers were selected. All volumes of the Annals of Surgical Hepatology (Moscow) and Medical Parasitology (Moscow) from 1970 to 2015 were analyzed. Additionally, some papers were advised by our competent colleagues. All cited papers included information about the place of study, description of materials and methods, including statistics and analysis of clinical records, and/or autopsy data. The papers on the O. felineus carcinogenic potential have pathomorphological descriptions of the liver; in addition, most papers contain histopathological images.

The data on incidence were extracted from the Rospotrebnadzor official report.18 We excluded the articles that duplicated data from original articles or lacked details about the methods as well as review articles, conference reports and theses.

We tried to cover all the papers in the central Russian journals related to the epidemiological situation with regard to opisthorchiasis and recording of liver cancer cases in endemic regions and their potential association with O. felineus infection over the last 40 years. However, we cannot exclude that some papers containing valuable information could have been overlooked.

We did not directly analyze the reports from morgues and did not participate in autopsies.

**Life cycle**

Opisthorchis felineus as a typical representative of the family Opisthorchiidae has a complex life cycle with alternation of two intermediate hosts and the definitive host. In the 1930s, Vogel was the first to describe the life cycle of these liver flukes and experimentally demonstrated that mollusks are obligatory intermediate hosts in the life cycle of these parasites.9 A general characteristic of all Opisthorchiidae is their high specificity to the first intermediate host. Typically, one or several closely related snail species act as the first intermediate host. Typically, the rediae produce numerous free-swimming cercariae, which leave the snail for their second intermediate host (Figure 1, Table 1).

Fish, the second intermediate host of O. felineus, belong to the family Cyprinidae (Table 1). The next 3 weeks to 2 months, the O. felineus cercariae develop into metacercariae in the fish body. As has been shown, metacercariae in the Ob River tributaries become invasive (able to infect humans and animals) 30–40 days after they enter the fish body.19 The duration of this period depends on the fish species and water temperature. Metacercariae are localized to the subcutaneous fat and muscle tissues at a depth of 1–2 mm. However, they may sometimes gather in quite unusual sites, for example, pectoral fins, gills or gut walls.9

The definitive host is infected when eating raw fish containing O. felineus metacercariae. The total list of definitive host species for this parasite comprises 33 species and subspecies of mammals, first and foremost, belonging to the order Carnivora: domestic cats, dogs, wolves, foxes, bears and badgers.9 In the gastrointestinal tract of the definitive host, the metacercaria emerges from its cyst and the excysted fluke enters the bile ducts of the liver. The liver flukes reach sexual maturity (maritae) after 25–30 days.9 The adult maritae produce numerous eggs, which may enter freshwater aquatic bodies with feces and sediment there to the bottom in shallow water. The larvae they contain, miracidia, are able to retain viability in freshwater aquatic bodies for 84–104 days if they entered these bodies in the early spring. Moreover, they are able to survive the period of water freezing–thawing during the overall winter and retain their viability to the July of the next summer if they entered these water bodies in the late fall.20 Infected fish is the only source of O. felineus infection for humans and animals. The O. felineus metacercariae are tolerant to drying, high salt concentration, and low temperatures and can be killed only by a high temperature.21 The typical sources of O. felineus infection in Russia are sun-dried, light-salted, and light-pickled fish. In addition, aboriginal populations in Western Siberia traditionally eat cold-served slices of frozen fish.

**Epidemiology**

Cases of opisthorchiasis caused by O. felineus are regularly recorded in several regions of the former Soviet Union, in particular, in the Volga–Kama basin (Central Russia),22 Ukraine9 and Kazakhstan.9 However, the highest O. felineus prevalence is observed in Western Siberia.9

In 1986–1996, snails in the Novosibirsk oblast were screened for the prevalence of liver fluke infection. The Bithyniidae snails sampled in 10 sites displayed an infection rate of only 2%.23 Further screening in 1995–2010 involving 1422 individuals demonstrated that the prevalence of liver flukes in the snails B. troscheli in the Ob River basin near Novosibirsk did not exceed 1%. In the Khanty-Mansiisk Autonomous Okrug (lower reaches of the Ob and Irtysh Rivers), this family was represented by B. troscheli; 1.08% (of 185 assayed individuals) of these snails were infected in 2006 in the Ob River and 0.76% (132 individuals) in the Irtysh River.15 Thus, the snail infection with liver fluke at a level of 1% is sufficient to maintain a 98% infection rate of O. felineus in fish.

In the area of the Ob–Irtysh basin (Western Siberia), fish screening for assessment of the O. felineus metacercaria infection is conducted on a regular basis. In 2002–2009, 997 cyprinid fish belonging to 11 species were assayed in the Tyumen oblast near the city of Tobolsk (five sites in the lower reaches of the Irtysh River as well as 10 sites in lakes and Irtysh tributaries). These data demonstrate that the O. felineus infection rate depends on individual species. In particular, the highest rate was observed in the ide Leuciscus idus (96.3%) and common dace L. leuciscus (98%) versus 36.2 and 31.7% in the common bream Abramis brama and roach Rutilus rutilus, respectively. Several species, such as the common carp Cyprinus carpio, tench
Tinca tinca, goldfish Carassius auratus gibelio, and crucian carp Carassius carassius, were free of any O. felineus infection.24 Similar data on the prevalence of O. felineus in different fish species were obtained by Karpenko et al.23 in the Novosibirsk oblast (middle reaches of the Ob River and its tributaries). The highest infection rate was observed in the ide L. idus (40.4%), common dace L. leuciscus, and sunbleak Leucaspius delineatus (43%). According to our own data of 2012–2015 (27 catches, 523 ide individuals from the Ob River near Novosibirsk), the number of O. felineus metacercariae per individual was 362 ± 177 (unpublished data). Note that the rate of infected fish increases with their age. In particular, the infection rate of dace yearlings and 2-year-old individuals is 12% versus 96.7% among the 5-year-old fish.24 Thus, the fish can be infected during its entire life.

These data demonstrate that the ide L. idus is the major source of O. felineus infection in Western Siberia. This is an abundant species of commercial value. In addition, the roach R. rutilus and dace L. leuciscus are also an important infection source; people eat these fishes and feed animals with them (Figure 2).

In 2013, Rospotrebnadzor presented the data on the structure of helminth infestation. Opisthorchiasis in Russia accounts for 79.0% of all cases; diphyllobothriasis 18.2%; dirofilariasis 5%; cystic echinococcosis 1.3%; alveolar echinococcosis 0.1%; taeniasis 0.1%; beef tapeworm infection 0.3%; clonorchiasis 0.5%; and trichiniasis for 0.1%.18 The official statistics for average opisthorchiasis infection rate in Russia is 20 cases per 100 000 population; however, this rate for the Khanty-Mansiisk Autonomous Okrug (the central part of the Ob–Irtysh opisthorchiasis focus) (Figure 3) is approximately 1000 per 100 000 population. The regions with the infection rate exceeding 100 per 100 000 population are regarded as endemic and are also associated with the Ob–Irtysh basin (Ob, Irtysh, Tura, and Tom' Rivers).18,25 The highest prevalence of opisthorchiasis is recorded in the Khanty-Mansiisk Autonomous Okrug, Yamalo-Nenets Autonomous Okrug (Yamal), as well as Tyumen, Tomsk, Omsk and Novosibirsk oblasts (Figure 3).

The actual level of O. felineus infection among the population may be deduced from short-term screenings in some villages of endemic regions (Kato-Katz and ELISA tests in fecal samples).26

![Figure 1. Life cycle of Opisthorchis felineus. The eggs are shed in the biliary tree, enter the intestine, and are passed with feces. They need to be ingested by freshwater gastropod snails, the first intermediate host, to develop into sporocysts, rediae, and free-swimming cercariae, the stage infective for the second intermediate host, cyprinid fish. Humans and other fish-eating mammals may serve as a definitive host by ingesting raw, slightly salted, or frozen fish. Entering the host body, metacercariae infect the biliary tract of mammals, where they mature into adult worms, the sexual stage, over approximately 1 month. The lifespan of an adult liver fluke in the human body can reach over 20 years.9,54 This figure is available in black and white in print and in color at Transactions online.](https://academic.oup.com/trstmh/article-abstract/110/1/28/2461623)
Table 1. Second intermediate hosts of Opisthorchis felineus in Western Siberia

<table>
<thead>
<tr>
<th>Common name</th>
<th>Latin name</th>
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<tbody>
<tr>
<td>Ide</td>
<td>Leuciscus idus</td>
</tr>
<tr>
<td>Dace</td>
<td>Leuciscus leuciscus</td>
</tr>
<tr>
<td>Roach</td>
<td>Rutilus rutilus</td>
</tr>
<tr>
<td>Crucian carp</td>
<td>Carassius carassius</td>
</tr>
<tr>
<td>Silver crucian carp</td>
<td>Carassius auratus gibello</td>
</tr>
<tr>
<td>Common bream</td>
<td>Abramis brahma</td>
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<tr>
<td>Nase</td>
<td>Chondrostoma nasus</td>
</tr>
<tr>
<td>Common rudd</td>
<td>Scardinius erythrophthalmus</td>
</tr>
<tr>
<td>Common or European carp</td>
<td>Cyprinus carpio</td>
</tr>
<tr>
<td>Tench</td>
<td>Tinca tinca</td>
</tr>
<tr>
<td>Gudgeon</td>
<td>Gario gario</td>
</tr>
<tr>
<td>Minnow</td>
<td>Phoxinus phoxinus</td>
</tr>
<tr>
<td>Sunbleak</td>
<td>Leucaspius delineatus</td>
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In particular, it has been shown that the infection rate among the rural population of the Tomsk oblast is 18–20% (291 individuals were examined); Tyumen oblast, 37% (134 individuals); and Chelyabinsk oblast, 18.6% (129 individuals).26

The results of short-term screening studies comply with the data of long-term observations in the regions with a high rate of liver fluke infection. Of special value are the autopsy data, since they most precisely reflect the infection rate. In particular, almost half cases (45.0%, 1014) of 2253 autopsies in the city morgue of Tobolsk during 1950–1987 showed signs of liver fluke invasion (retrospective analysis of autopsy records).27 According to the autopsy data, the prevalence of opisthorchiasis in Tomsk during 1983–2002 varied in the range of 10–18.3% (343 autopsies, prospective autopsy studies).28,29 The age related incidence of infection (1983–1987; 641 autopsies): 10–19 years old, 33.3% male (M), 50% female (F); 20–29, 26% M, 20% F; 30–39, 17% M, 27% F; 40–49, 26% M, 33% F; 50–59, 43% M, 54% F; and 60–69, 28.5% M, 37% F.27

Summing up, we should emphasize that the data published by individual teams of Russian scientists suggest a high rate of O. felineus infection among the population of endemic regions, which varies from 10 to 45%, which considerably exceeds the data of official statistics.

Carcinogenic potential of Opisthorchis felineus

Although the mechanism underlying development of the cholangiocarcinoma during liver fluke infection has been studied only fragmentarily, the accumulated data, mainly with O. viverrini, allow for a preliminary scheme of how this type of tumor can be induced. O. viverrini can affect the bile duct epithelium by mechanical injury; the impact of reactive oxygen intermediates (ROI) generated by the host cells in the focus of inflammation; and a direct action of certain proteins secreted by liver flukes, which induce proliferation and inhibit cell apoptosis.30–32 All these processes cause the injuries of genetic material, mutations, fixed during DNA replication. Eventually, accumulation of mutations can lead to malignant transformation of cholangiocytes and development of cholangiocarcinoma.30,33,34

According to recent data, the carcinogenic liver flukes O. viverrini and S. haematobium synthesize specific oxysterols and catechol estrogens able to act as genotoxic agents.36,35 It is assumed that these compounds damage the host DNA and in combination with the above mentioned factors enhance development of malignant tumors in the tissues injured by parasites.

Our laboratory is involved in studying the O. felineus biology,36–38 transcriptomics,39 and functional genomics,40 including the mechanisms of liver fluke metabolism associated with the synthesis of genotoxic compounds. We have recently succeeded in demonstrating that O. felineus possesses a functionally active cytochrome P450 form41 (unpublished data) as well as other enzymes involved in the metabolism of xenobiotics and able to generate oxysterols and catechol estrogens. Most likely, the specific genotoxic metabolites are a significant component of the liver fluke carcinogenic potential.

Since this country lacks systemic epidemiological and clinical programs for opisthorchiasis, the role of O. felineus in development of cholangiocarcinoma in humans is vague. Undoubtedly, the similarity in clinical manifestations and courses of the diseases caused by O. felineus and O. viverrini42–44 and the data on the pathogenesis of these two helminthiases acquired using experimental opisthorchiasis model45 suggest a rather high probability of O. felineus involvement in the induction of these tumors. Here we briefly the data published in the Russian medical literature on the complications associated with opisthorchiasis as well as the data from hospitals and morgues on autopsy results over a long time period.

As mentioned above, almost half the autopsies (1034, 45.0%) of the 2253 cases examined in Tobolsk in 1950–1987 detected the signs of liver fluke invasion (retrospective analysis of autopsy records).27,46 Of the patients infected with O. felineus, suppurative complications were observed in 46 (4.5%) cases and primary liver cancer, in 90 (8.7%).27 The average age of the patients with liver cancer (according to retrospective analysis of medical records) is 54.4–55.5 years.27

According to 343 autopsies of the cases with a long-term O. felineus invasion at the Tomsk city morgue, cholangiocarcinoma was observed in 5% patients (prospective autopsy studies).25,42–44 Of the 4756 opisthorchiasis patients treated at the Tomsk Hepatology Center, 141 were operated on for bile duct and pancreatic cancers (no data for individual cancer types).25,42–44 The consolidated data of specialized cancer centers in several Western Siberian cities over 10 years (1962–1971) demonstrate that 1225 primary liver cancer cases are recorded (retrospective analysis of medical records), which amounts to 9.4 cases per 100 000 population.48–50 Over the same period, this rate for the Yamalo-Nenets Autonomous Okrug was 8.2 per 100 000 population; for the Tobolsk region, 18.2; and for the Khanty-Mansiisk Autonomous Okrug, 20.7 per 100 000 population.

According to the data of the Tomsk regional cancer center (1990–2001), liver cancer is the seventh in the list of cancer diseases, accounting for 3.9%. In particular, the liver cancer morbidity among men during this period amounted to 11.2 cases per 100 000 population (retrospective analysis of medical records).51
Studies regarding additional risk factors for liver cancer were conducted (retrospective analysis of medical records). First, the association of primary liver cancer with viral hepatites in cities and localities of the Khanty-Mansiisk Autonomous Okrug, Yamalo-Nenets Autonomous Okrug, and Tyumen oblast was analyzed; the liver cancer incidence in these regions was not uniform. The localities and cities selected by random sampling were compared to the data on viral hepatitis morbidities there (retrospective analysis of medical records); however, no correlation was observed. Second, the correlation between liver cancer and cirrhosis was estimated. The retrospective analysis of medical records and autopsy records for 224 liver cancer cases detected a 21% rate of cirrhosis. Similar level of cirrhosis prevalence was found: 14.5% in the patients with liver cancer in the

Figure 2. The liver fluke Opisthorchis felineus in its natural habitat, neighborhood of Novosibirsk, Western Siberia. (A) El'tsovka River (a tributary of the Ob River), El'tsovka village. (B) A snail of the genus Bithynia from the El'tsovk River near the village of El'tsova. (C) An O. felineus metacercaria isolated from bile tissues. (D) Adult O. felineus flukes extracted from the bile ducts of a Syrian hamster (Mesocricetus auratus). (E) Sun-dried roach (Rutilus rutilus), a traditional beer snack in Russia. This figure is available in black and white in print and in color at Transactions online.
Khanty-Mansiisk Autonomous Okrug and 12% in Tobolsk. The prevalence of cirrhosis in the regions not endemic for opisthorchiasis (southern part of the Tyumen oblast, 26%) was approximately the same. Thus, any correlation was undetectable. The correlation between liver cancer and liver fluke invasion was searched for in individual localities of the Tyumen oblast, Khanty-Mansiisk Autonomous Okrug, and Yamalo-Nenets Autonomous Okrug with a high rate of liver fluke invasion versus southern part of the Tyumen oblast, displaying a low rate of population infection (retrospective study). This demonstrated a high direct correlation between the liver cancer incidence and opisthorchiasis infection in the northern Tyumen oblast, Khanty-Mansiisk Autonomous Okrug, and Yamalo-Nenets Autonomous Okrug (correlation coefficient, 0.66; mean error of correlation coefficient, 0.003). The liver fluke invasion is rare among the population of southern Tyumen oblast; any correlation

Figure 3. Prevalence of opisthorchiasis in Western Siberia according to the official statistics for 2012.24 This is the number of cases recorded when patients themselves sought medical help, thereby only approximately reflecting opisthorchiasis prevalence in the population. Nonetheless, these data allow the regions with highest prevalence to be distinguished: highly endemic >100 cases per 100 000 population; moderate 50–100 cases; and weakly endemic <50 cases per 100 000 population. This figure is available in black and white in print and in color at Transactions online.
between the liver cancer incidence and opisthorchiasis was undetectable.

Histological examination of 169 liver cancer patients in the Tyumen oblast detected cholangiocarcinoma at a rate of 77%; hepatocellular carcinoma, 18.9%; and other tumor types, 4.1%. The control group in the same study comprised 146 patients with liver cancer but without opisthorchiasis. They displayed cholangiocarcinoma at a rate of 34.2%; hepatocellular carcinoma, 55.5%; and other tumor types, 10.3% (retrospective prevalence case–control studies).

Another study in the same region gave similar statistics. Autopsies of 44 primary liver cancer cases detected O. felineus invasion in 42 cases and cholangiocarcinoma in 35 (79.5%) cases (prospective autopsy studies). Thus, the ratio of cholangiocarcinoma to hepatocellular tumors in the endemic regions is 4:1.

On the contrary, cholangiocarcinoma in the non endemic regions accounts for only 15% of all liver cancer cases. In particular, the rates of hepatocellular cancer and cholangiocarcinoma in Moscow, a non endemic region for liver fluke (according to the data of the Blokhin Russian Cancer Research Center; 1004 surgical interventions for primary liver cancer), were 85.1% and 14.9%, respectively (retrospective analysis of medical records). These levels match the published data on the cholangiocarcinoma epidemiology. This tumor is recorded at a rate of 19% in the United States, the region free of opisthorchiasis foci, vs Thailand, in particular, Khon Kaen Province, the region with the highest cholangiocarcinoma rate in the world, where this cancer type accounts for 71% of liver cancer cases.

Besides cholangiocarcinoma, opisthorchiasis may cause other complications, such as cholangites, cholecystites, cholangiobrosis and hepatic cysts, which can be regarded as a precancer. In addition, opisthorchiasis may induce development of hepatic abscesses and pancreatites. The structure of opisthorchiasis complications is described in the papers by researchers from the Tomsk Hepatology Center. In particular, observation of 4756 patients (prospective analysis) with opisthorchiasis demonstrated that 1170 (24.6%) required surgical intervention for bile duct strictures, 730 (62.4%), pancreatitis 188 (16.0%), hepatic cysts 37 (3.1%), sclerosing cholangitis 27 (2.3%), hepatic abscesses 31 (2.6%), and liver cirrhosis 43 (3.6%). Note that most of the patients subject to surgery had chronic opisthorchiasis with a O. felineus infection of over 10 years old (retrospective analysis of the clinical case records).

Further studies at the Tomsk Hepatology Center confirm that the most frequent complications of opisthorchiasis are the bile duct strictures and cholecystites; the pancreatites caused by liver fluke invasion are second in the list of opisthorchiasis complications. Cholangiectasis underlies development of opisthorchiasis hepatic cysts; hepatic abscesses appear as a result of cyst empyema and as a complication of cholangiitis.

Similar data on the rate of surgical interventions in opisthorchiasis patients were obtained for the Khanty-Mansiisk Autonomous Okrug: 27.3% of the 812 opisthorchiasis patients at the Surgut City Hospital needed surgical intervention (prospective analysis).

Discussion

Opisthorchis felineus is a member of the triad of epidemiologically important liver fluke species belonging to the family Opisthorchiidae; however, its biology, epidemiology and carcinogenic potential are poorly studied as compared with O. viverrini and C. sinensis. This contradicts common sense, since the O. felineus distribution range covers vast areas in Europe and Asia and opisthorchiasis outbreaks caused by this helminth are expectable in many countries. Ever increasing migration and flows of tourists should be also taken into account. Correspondingly, patients with O. felineus invasion can be detected far beyond the endemic regions. Thus, the opisthorchiasis caused by O. felineus becomes a global-scale problem, out of the scope of biomedical problems in individual regions.

This review briefs the data on the rates of snail and fish infection by the liver fluke O. felineus in the world largest focus of opisthorchiasis as well as the prevalence of this pathogen in the population of Western Siberian regions within this focus. The consolidated data demonstrating that the rate of primary liver cancer in these regions is considerably higher as compared with the regions non endemic for opisthorchiasis and the average level for the overall country are reported for the first time.

It should be emphasized that when preparing this review, we had no access to the clinical records of the patients and the clinical material used for the corresponding original papers, which we reviewed. However, we tried to carefully select the papers for our review based on the criteria described in the Methods section. Nonetheless, we admit that not all the autopsies and examinations met the international standards. After so many years, we cannot verify these studies. We assume that the reviewed papers might contain not all necessary information about the quality of performed examinations; in such case, we report only the data extracted from the corresponding paper.

Unfortunately, the data on liver cancer prevalence have certain defects, in particular, the performed examinations and criteria for selection of control groups are insufficiently detailed. However, in the absence of other epidemiological data, the number of cases recorded in a particular hospital over a given time period is also valuable information, of course with indication that this is just recording of the number of cases rather than full-fledged epidemiological studies.

The data on incidence were extracted from the official reports of the Rospotrebnadzor, which are publicly available at the corresponding site. Thus, the official statistics reflect only the number of cases recorded when patients themselves sought medical help. Since the first stages in this disease are frequently asymptomatic, the official statistics as a rule record only chronic cases with pronounced symptoms. This prevents us from considering the corresponding data completely reliable, and we regard them rather as actual incidence. In addition, the Rospotrebnadzor official reports lack any data on autopsies. This prevents these data from being regarded as fully reflecting the actual incidence or actual prevalence.

Summing up the data on the carcinogenic potential of O. felineus, it should be emphasized that opisthorchiasis caused by this liver fluke and bile duct cancer are potentially connected. Analysis of the literature, briefed in this review, has allowed us to demonstrate that the liver cancer in the regions endemic for opisthorchiasis is considerably more prevalent as compared with the other regions of this country.

However, these data have been obtained by individual research teams and cannot provide a general picture of the association between opisthorchiasis and liver cancer. This requires large-scale
epidemiological studies based on experience in such type of research accumulated in other countries.³

Conclusions

Note, in conclusion that only the knowledge about biology of these parasites, including the structure and function of the genome, metabolic pathways, provision of life activities, detailed biology of developmental stages and host-parasite interactions will allow for satisfactory control of opisthorchiasis and the epidemiological situation.

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References

26 Ilyinskikh EN, Novitsky VV, Ilyinskikh NN, Lepyokhin AV. Opisthorchis felineus (Rivolta, 1884) and Metorchis bilis (Braun, 1890) infections in population of some regions of the Ob river basin [in Russian]. Parasitol (Mosk.) 2007;41:55–64.


