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# WAYS OF INFECTION OF VERTEBRATE ANIMALS WITH SARCOSPORIDIA AND MORPHOLOGY OF THEIR CAUSATIVE AGENTS

**Abstract.** Spores are protozoan single - celled organisms that parasitize the body cavity, intestines, and blood cells of annelids, molluscs, arthropods, vertebrates and humans. Due to parasitic activity, digestive and contractile vacuoles are absent: they absorb nutrients through the entire body; they do not have motor organoids, only the sex cells and microgametes contain flagellum.

Spores reproduce by asexual, sexual, and sporogony processes. The process of asexual reproduction occurs through the division of the nucleus into many nuclei, i.e., as a result of the process of schizogony, young individuals called merozoites emerge. Sexual reproduction is the emergence of gametes in the process of gametogony. The zygote formed as a result of copulation is covered with a thick coat that turns into an oocyst. After this, the process of sporogony begins. As a result of the process of sporogony within the oocysts, first sporoblasts are formed, from which small sporozoites appear and they are located inside the spores.

Among the sporous organisms, coccidiomorpha are the protozoan which cannot exist independently and only parasitically live inside the cell. The body forms are oval, roundish, or arched. They are found in the intestinal cells, myocytes, hepatocytes, and in kidneys and blood cells of humans, vertebrates (mammals, birds, reptiles). The life cycle of coccidia is very complex.

Oocysts via water and grass enter the intestines of the intermediate host; the cell wall of the oocysts under the influence of intestinal juices is dissolved and eight sporozoids released. For further development of sporozoids, it is necessary to penetrate the intestinal epithelial cell; when they penetrate the cell, the body rounds up and turns into schizont. The nucleus of schizont divides several times. The division of these nuclei underlies the process of schizogony. This is the process of asexual reproduction. Around each nucleus, formed as a result of schizogony, cytoplasm is accumulated, which turns into small merozoites. The process of schizogony is repeated several times, so the number of parasites increases. After schizogony, the gametogony process begins. This process occurs in the final host, resulting in gametes. The final hosts of parasites expell sporocysts (oocysts) to the external environment with feces. Accordingly, this process is repeated.

**Key words:** sarcocyst, sarcosporidiosis, fauna, life cycle, micromorphology, merozoite, definitive host, schizogony, gametogony, sporogony.

**Introduction.** Sarcosporidiosis-zooanthroponosic disease of animals and humans, characterized by lesion of the muscle caused by single-celled protozoa. Sarcocystosis is often recorded in a chronic form and is externally unnoticeable invasion. Pathogens of sarcocystosis occur in the form of trophozite (endozite, cytozoit) and cyst (micro-and macrocysts) in the body of intermediate hosts (pets) [1].

Sarcocysts are parasites that are common in vertebrates, including sheep (Sarcocystis ovicanis, Sarcocystis ovifelis), goats (Sarcocystis capracanis), cattle (Sarcocystis bovicanis, Sarcocystis bovifelis, Sarcocystis bovihominis), camels (Sarcocystis camelicanis), pigs (Sarcocystis) suicanis, Sarcocystis suihominis), and other animals. The external structure of sarcocyst species is very similar to each other. They reproduce through two hosts, through definitive (final) and intermediate hosts. The final host happen to be a human, dog, cat, and other predatory meat-eating vertebrates. On the other hand, ungulates, rodents, seals, birds (except for predatory birds), reptiles, fish nurseries happen to be intermeiate hosts.

Definitive hosts of the parasite expell sporocysts (oocysts) to the external environment with feces. The initial phase of development is the trophozoite. It has a shape of sickle, banana, spindle, or pea; the body size is 6-15 microns, and the width is 2-7 microns. Parasites consist of a nucleus, cytoplasm, and cell wall.

The body size of cysts visible to naked eye reaches 10-20 mm. The shape of this species resemble spindles, ellipses, or cylinders, with two-layer walls. The cystic cavity is filled with trophozoites and is divided into many chambers. The typical form of sporocysts is ellipsoid, the body size is 14.3x10.2 microns. There are no polar bodies. The wall happens to be shiny, colorless, very thin, and tender

In the cavity of the Sporocyst, there are four sporozoits with a body size of 9-11x2-3 microns and round residual body. Sarcocystoses are found in all regions of Kazakhstan [2].

Materials and research methods. In the complex educational and research laboratory at the Institute of Natural Science and Geography and the laboratory of Invertebrate Zoology at the Department of Biology in the period from 2018 to 2019, study was conducted. The target area of research was the southeastern region of Kazakhstan.

A comparative study of the morphology of sarcocysts, cystozoids, and sporocysts from samples of various vertebrate species under a light microscope showed the presence of different shapes and sizes of cysts, merozoits, and the structure of the cyst wall. The volume and shape of cysts usually depend on their age and level of maturation. A constant diagnostic sign of mature sarcocysts of various animal species under a light microscope is the structure of the cystic wall and the size of increasing, complete stages of sarcocyst development-merozoits.

When determining the type of sarcosporidia, both morphological and biological properties of the parasite, as well as the intermediate and definitive host and the species it belongs play an important role. Sarcosporidian are related to specific parasitic group according to the intermediate owners. From samples taken from various vertebrate species, when studying the development cycle and morphology of sarcocysts, it was found that these animals have sarcocysts that differ significantly in the size of merozoites. The dependence of the merozoite size on the taxonomic belonging of the definitive host was studied [3].

Results and Discussions. The morphological feature of various sarcosporidia species usually depends on the taxonomic belonging of the definitive host. Definitive hosts morphological comparative data of various sarcosporidia are shown (table 1). The study mainly identified the size of merozoites and sarcocysts and the structure of the wall.

Species	Inter- mediate host	Definitive host	Average size of merozoites (microns)	Structure of the cyst wall	Thickness of the cyst wall
Sarcocystis citellivulpes	Yellow ground squirrel	Fox, corsac fox	4.1x12.1	Horizontal wavy	1.4-3.3
Sarcocystis citellibuteonis		Buzzard	1.6x7.0	Smooth	0.6-1.0
Sarcocystis muris	House mouse	Cat	4.1x13.8	Smooth	1.1-2.0
Sarcocystis dispersa		Owl	2.0x7.0	Smooth	0.4-1.0
Sarcocystis cerne	Vole	Kestrel	2.1x8.4	Smooth	0.4-1.0

Table 1 – Size of merozoites and sarcocysts and structure of their wall

Sarcosporidia merozoites with definitive hosts as a predatory mammals (fox, corsak, dog, cat) are large, with thick, horizontal wavy cyst wall(under a light microscope). There is only one distinction: walls of sarcocysts of house mice are smooth. Merozoites sarcosporidia of predatory birds as a definitive host (buzzard, owl, kestrel) are small, the walls of the cysts are not horizontal horizontal wavy but are smooth and thin (table 2) [4].

Species	Intermediate host	Definitive host	Average size of merozoites (microns)
Sarcocystis bovicanis	Cattle	Dog, wolf, fox	3.7-10.5
Sarcocystis ovifelis	Sheep	Cat	3.4-13.3
Sarcocystis ovicanis	Sheep	Dog, buzzard, shibor	4.7-14.2
Sarcocystis moulei	Goat	Dog, wolf, corsac	5.0-16.0
Sarcocystis suicanis	Pig	Dog, cat	2.5-14.1
Sarcocystis eduicanis	Horse	Dog	4.3-11.1
Sarcocystis gruneri	Maral	Dog, wolf	1.0-12.2
Sarcocystis orientalis	Siberian mountain goat	Dog, wolf, fox	3.4-18.4

Table 2 - Comparative data of sarcocysts identified from predatory mammals as a definitive host

To determine the life cycle of sarcocysts, it is very difficult to find definitive owners, that is predatory animals, especially the wild ones. In the selection of definitive hosts of the sarcocysts under reasearch, it can significantly help to determine the definitive host dependence on the above-mentioned morphological features. This allows you to predict definitive hosts among predatory mammals or birds.

The results of a study of sarcocysts in which rodents (zorman, house mice) are intermediate hosts are presented in the following section. With brief information about the distribution of animal data and the detection of sarcocysts of wild rodents given, collected material is analyzed.

In Kazakhstan, there is a vast habitat area of wellow ground squirrel, which covers the southern (Kyzylorda, Shymkent, Zhambyl) and South-Eastern (Almaty region). It is also very common in the western part of Kazakhstan (Atyrau, West Kazakhstan, and Aktobe regions), in Central Kazakhstan (in the Western regions of Karaganda and Akmola regions). Yellow ground squirrels are vertebrate animals that live in groups. They live mainly in clay-desert steppes and semi-desert lands, also, in hardened sands. In S.M. Puck's work, seven species of coccidia were identified in this animals. Individuals that belong to the genus Eimeria-5 species, genus Isospora-2 species, taxoplasm and sarcocysts [5].

379 yellow ground squirrels of different ages and genders were selected for the study. Microscopy of native preparations revealed 28 (7.4) infected rodents. In 2018, the overall infection rate of yellow ground squirrels was significantly higher (7.6%). This indicator is also associated with the study of adult animals. Among young population fewer infected ones were encountered, and compared to males, female infection (11.4%) is dominant, but in 2019-vice versa. Therefore, in males and females, there is no difference. Only 5.6% of the total number of examined yellow ground squirrels were infected with Sarcocystis citellivulpes, 0.5% - Sarcocystis cittellibuteonis, and in two species simultaneously-0.4%. In the habitats of yellow ground squirrels, the definitive host of sarcocysts, i.e. representatives of the order of predators fox and corsacs are widely common [6].

Sarcocystis cittellibuteonis cysts according to ultrastructural characteristics are thin cysts that are widespread in mammals, birds, and reptiles. The thin wall of cysts differs from the cysts of Sarcocystis citellivulpes by the morphology of the cystic walls and a smaller size of cystozoids at the optical level. The cystic wall of sarcocystis citellivulpes is equipped with thick, that is, long glands consisting of fibrils.

The attention-grabbing diversity of the structure of the cystic walls of sarcocysts species and the formation of a specific cystic wall in each cystic phase of parasite development can be explained by hereditary features. Questions about the charactristics of the cystic walls of cattle sarcocysts were discussed in the works of M.D. Novak, V. M. Fedoseenko, V.A. Orazalinova, sheep sarcocysts - A.O. Heydorn, Mehihorn [7].

Experimental work carried out in farm animals has shown that only asexual reproduction – schizogony – occurs in the intermediate host, while in the definitive host – the process of sexual reproduction: gametogony and sporogony. At the first infection, an acute symptom of sarcocystosis is observed in young animals, when inside the body is undergoing a process of mass asexual reproduction. The disease can quickly proceed and become fatal. After the formation of the sarcosporidium development period, a chronic phase of the disease occurs in the muscles, which is usually observed when examining muscle cysts.

Several methods are used during the study of sarcospores. Fecal examination using the fulleborn method--this method is simple and widely used in practice. To do this, after about 10-20 grams of feces taken, it is put it in a glass container of 100-200 ml volume, in which a pre-prepared saturated solution of table salt is added(to prepare a solution of table salt, mix 350 grams of table salt in 1 liter of water and boil

for half an hour). After that, it is completely crushed up with a stick. The stool is diluted with a solution of salt in the concentrate and mixed again with a stick. Feces are re-filtered with a filter and poured into a clean glass container for 30-40 minutes to settle. During the 'settlement', sarcocysts float to the surface of the liquid. They are removed from the surface of the liquid, placed in a slide, covered with glass, and examined under the microscope.

The Darling method uses precipitation and flotation methods together. First, 3-5 grams of feces are mixed with a small amount of water, the resulting mixture is filtered with a filter and poured into a centrifuge tube. In the opposite tube of centrifuge fecal filtrate or water is added, and their weight is balanced. Then the test tubes are turned over by a centrifuge device for 5 minutes at a speed of 3000 rpm (at that time sarcocysts precipitate). Then drain the liquid on the surface of the precipitation and pour into the latter the Darling liquid prepared with the addition of glycerol solutions with a density of 1.205 kg/m and the equal amount of saturated table salt. The precipitation is well mixed with the Darling liquid. It is shaken so that there is no spill outside and rotated for 3 minutes at a speed of 1000 rpm. This way, Sarcocysts in the fecal liquids float on the liquid surface. They are removed from the surface of the liquid and examined under a microscope on a slide with a covering glass.

In the compression method of study, 25 grams of samples of heart, sub-cutaneous, tongue, and abdominal meat are taken from deceased or slaughtered animals. From the selected meat samples, a preparation of 1.5x0.3 centimeters is made, placed in a slide; 1-2 drops of isotonic solution are instilled; the surface is covered with a covering glass, clamped between the glasses and examined under a microscope. In addition, for a clear detection of parasitic trophozoites with an average magnifier microscope, they should be dyed. 2-3 drops of 0.5% aqueous solution of methylene blue and frozen alkaline acid are instilled; after 3 minutes, 2-3 drops of 25% smelling salts are poured on the particles; then, the particles are compressed and examined under a microscope. Sarcocysts can also be detected using a fluorescent microscope [8,9].

Conclusion. According to the taxonomical characteristics of the definitive hosts, it was found that some morphological features of sarcocysts differ abruptly. If the definitive host of sarcocysts happens to be predatory mammals, cystozoites in the cysts are large, its wall is thich and with glands, and if the definitive host of sarcocysts happens to be a predatory birds: merozoites are small, walls of sarcocystis are thin and smooth.

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#### ОМЫРТҚАЛЫ ЖАНУАРЛАРДЫҢ САРКОСПОРИДИЛЕРМЕН ЗАЛАЛДАНУ ЖОЛДАРЫ ЖӘНЕ ОЛАРДЫҢ ҚОЗДЫРГЫШТАРЫНЫҢ МОРФОЛОГИЯСЫ

**Аннотация.** Саркоспоридилер - Sarcocystis туысына Sporozoa класына жататын қарапайымдылардың паразиттік түрі болып табылады, табигатта кең таралган. Жершарының әртүрлі елдерінен омыртқалы жануарлардың көптеген түрлерінен табылған. Әсіресе үй жануарларының саркоспоридилермен залалдануы жабайы жануарларға қарағанда басымырақ болып келеді.

Саркоцистоз - кең таралган және аз зерттелген иесінің бұлшық ет ұлпасын зақымдайтын жануарлар мен адамның протозойлық ауруы. Олардың қоздыргыштары саркоспоридилер немесе 170 жылдан астам зерттеу тарихы бар ет споралары болып табылады.

Саркоцисталарды жұқтыру әсіресе топталып тіршілік ететін жануарларда басымырақ болады, сонымен қатар жабайы жануарлардағы саркоспоридилер үй жануарларымен салыстырғанда нашар зерттелген.

Саркоспоридилер жасушаішілік циста түзуші кокцидиялар, екі иесінің ағзасында өтетін облигатты гетероксенді тіршілік циклі тән. Жыныстық (гаметогония) мен спорогония процесі соңгы иесі - жыртқыш жануарлардың агзасында өтеді, соның нәтижесінде ооцисталар нәжіспен бірге сыртқы ортаға бөлінеді. Олар аралық иесінің ағзасына түскенде жыныссыз көбею процесі жүреді, соның нәтижесінде негізінен мерозоит – гамонтпен толтырылған бұлшық ет цисталары қалыптасады.

Морфологиялық құрылысы сопақ, ұштары дөңгелектелген ұршық тәрізді немесе 12-16х3-9 мкм өлшемдерімен дога тәрізді споралармен толтырылған ұзындығы 0,17-1,6 миллиметр болып келетін цилиндрлік цисталар түзеді. Ядросы ортасында кариосомасы бар сопақ немесе дөңгелек пішінді болып келеді. Цисталары тағаммен бірге адамның ас қорыту жолына түседі (инвазирленген жануарлардың етінен). Асқазанда және аш ішекте олар ферменттердің әсерінен сыртқы қабығы ериді, паразиттер қан тамырларына

өтіп және қанның ағысымен бүкіл денеге таралады. Содан кейін аяқ-қол, көмей, тіл, жүрек бұлшық еттерінде олар жаңа цисталарды түзеді.

Саркоспоридилердің клиникасы аз зерттелген. Тіпті қарқынды зақымдалғанның өзінде де саркоцистоз клиникалық белгісіз өтуі мүмкін. Цисталарда саркоцистин деп аталатын улы заттар болады. Гистологиялық зерттеу кезінде жануарлардың бұлшық етінде - гиалинді қайта туу, ал адамда - ұсақ қан құйылу байқалады. Биопсия арқылы алынған нативті және тұрақты боялған препараттарда немесе бұлшық еттердің гистологиялық кесінділерінен цисталар анықталады. Цисталар рентгенологиялық зерттеу кезінде сирек анықталуы мүмкін.

Олардың аралық иесі - ірі және ұсақ малдар, шошқа, жылқы, лама, түйе, қоян, тышқан болуы мүмкін. Түпкі иесі етқоректілер болып табылады. Соның салдарынан малда арықтау, анемия, өнімділіктің төмендеуі және тіпті өлімге экелу де байқалады.

Мақалада омыртқалы жануарлардағы Sarcocystis туысының өкілдерінің түрлері, омыртқалы жануарлардың залалдану жолдары және цисталардың көлемі мен мөлшері, морфологиясы туралы айтылған.

**Түйін сөздер:** саркоциста, саркоспоридиоз, фауна, даму циклы, микроморфология, мерозоит, дефинитивті иесі, шизогония, гаметогония, спорогония.

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### ПУТИ ЗАРАЖЕНИЯ ПОЗВОНОЧНЫХ ЖИВОТНЫХ САРКОСПОРИДАМИ И МОРФОЛОГИЯ ИХ ВОЗБУДИТЕЛЕЙ

**Аннотация.** Саркоспоридии – паразитный вид простейших, относящийся к роду Sarcocystis классу Sporozoa, широко распространенный в природе. На земле обнаружены многочисленные виды позвоночных животных в разных странах. Особенно преобладает зараженность саркоспоридами домашних животных, чем диких животных.

Саркоцистоз — это протозойная болезнь животных и человека, которая поражает мышечную ткань широко распространенный и мало исследованный. Их возбудителями являются саркопоридии или мясные споры, имеющие историю исследования более 170 лет.

Заражение саркоцистами особенно преобладает у животных, живущих группами, а также саркоспоридии у диких животных, которые исследованы хуже по сравнению с домашними животными.

Для саркоспоридии характерны внутриклеточные цистообразующие кокциды, облигатный гетероксенный жизненный цикл, протекающий в организме обоих хозяев. Половой процесс (гаметогонии) и спорогонии происходит в организме последнего хозяина – хищных животных, в результате ооцисты вместе с фекалиями попадают во внешнюю среду При попадании их в организм промежуточного хозяина происходит процесс бесполого размножения, в результате которого в основном формируются мышечные цисты, наполненные мерозоидом – гамонтом.

Морфологическое строение образует цилиндрические цисты длиной 0,17-1,6 миллиметров, имеющие овальное, с округленными концами бедрообразные или заполненные дугообразными спорами размером 12-16х3-9 мкм. Ядро представляет собой овальную или круглую форму с кариосомой в центре. Цисты (из мяса инвазированных животных) вместе с пищей поступают в пищеварительный тракт человека. В желудке и тонкой кишке они под воздействием ферментов, растворяют наружные оболочки и паразиты проникают в кровеносные сосуды и распределяются по всему телу с течением крови. Затем в мышцах конечностей, в гортани, языка, сердца они образуют новые цисты.

Клиника саркоспоридии мало изучена. Даже при интенсивном поражении саркоцистоз может пройти клинически незаметно. В цистах содержится ядовитое вещество, которое называется саркоцистин. При гистологическом исследовании наблюдается рецидив гиалина в мышцах животных, а у человека — мелкое кровоизлияние. В нативных и стойких окрашенных препаратах, полученных биопсией или из гистологических разрезах мышц, можно определить цисты. Цисты могут выявляться редко при рентгенологическом исследовании.

Их промежуточным хозяином может быть крупный и мелкий скот, свиньи, лошади, лама, верблюды, зайцы, и мыши. Конечным хозяином являются животные, которые являются плотоядными. В результате у животных наблюдается похудение, анемия, снижение продуктивности и даже смертность. В статье говорится о видах, морфологии и путях заражения позвоночных животных и количестве цист, представителей рода Sarcocystis у позвоночных животных.

**Ключевые слова:** саркоциста, саркоспоридиоз, фауна, цикл развития,, микроморфология, мерозоит, дефинитивный хозяин, шизогония, гаметогония, спорогония.

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