AN ABSTRACT OF THE THESIS OF

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Title FASCIOLOIDIASIS OF CATTLE, DEER AND ELK IN OREGON AND SOUTHERN WASHINGTON

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The biology, history and synonyms of Fascioloides magna were reviewed and the known distribution given.

Studies have shown that Cervidae are the natural hosts and domestic ruminants the accidental hosts. The parasite is reportedly pathogenic to goats and sheep but causes severe tissue reactions in cattle. The trematode is unable to reproduce in cattle but may occasionally in sheep and goats.

During this study 107 Oregon and 19 Washington cattle were traced from abattoirs to the source of infection. The main source of cattle infections for Oregon and southern Washington was the Columbia River area. Other enzootic areas were Wheeler, Klamath, Douglas and Coos Counties in Oregon and Yakima County in Washington.

Survey data showed that F. magna was either rare or did not occur in deer and elk in the Coastal Mountain Range or in Benton

County of the Willamette Valley in Oregon. There was a high incidence of infected elk in the Cascade Range of Lane County. Reports indicated the incidence was also high in deer of the Columbia River area from Portland to Astoria.

On the major study area, Tenasillahae Island in the Columbia River, 94 percent of the deer and 77 percent of cattle were infected. Two proven snail hosts, Stagnicola palustris and Pseudosuccinea columella were the only Lymnaeidae snails found on the island and it was concluded that they were intermediate hosts for F. magna in this area.

Stagnicola palustris, P. columella, Lymnaea auricularia and Stagnicola bulimoides were found in Oregon and S. palustris in Washington. No naturally infected snails were found nor were any experimental infections successful.

Pigmentation of the liver and lymph nodes, omentum and diaphragm was found in each case of infected deer and elk even when no closed cysts were present. A massive infection in one Tenasillahae Island deer resulted in adhesions, hyperplasia, necrosis of the liver and extensive pigmentation.

The ova from cattle were darker colored, thicker shelled and shorter than those from deer and elk.

Fluke ova incubated in an "egg bath" hatched by the 12th day.

A theory of the reabsorption of F. magna cysts by the omentum

of deer as a means of eliminating the cysts from the liver was presented.

It was concluded that F. magna was of minor importance as a state-wide problem in Oregon. The Columbia River area was considered a major local problem and would become increasingly important as the use of islands and diked areas are more extensively used for raising livestock.

FASCIOLOIDIASIS OF CATTLE, DEER AND ELK IN OREGON AND SOUTHERN WASHINGTON

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FASCIOLOIDIASIS OF CATTLE, DEER AND ELK IN OREGON AND SOUTHERN WASHINGTON

INTRODUCTION

Fascioloidiasis is a disease which occurs in domestic and wild ruminants. It is caused by the large American liver fluke, Fasciloides magna (Bassi, 1875) Ward 1917. The life cycle of this parasite and the interrelationship between natural and accidental hosts is shown in Figure 1.

Deer, elk and moose have been reported as the natural hosts for F. magna because; 1) viable ova are freely passed in the feces of infected animals, thereby permitting continuation of the life cycle and 2) light infections are relatively non-pathogenic. In infected cattle viable ova are not commonly passed with the feces and in sheep the parasite is highly pathogenic. Cattle do not ordinarily show signs of infection and the parasite appears to have a short life span. The bovine liver builds scar tissue and calcium cysts which encapsulate the parasite. As a result of encystation the ova of the liver fluke cannot escape; thus, the parasite is unable to complete its life cycle (60, p. 211).

In sheep the disease condition is frequently acute even in light infections. Encystation occurs only rarely in this host. If it does occur the ova escape and reproduction may take place (60, p. 214;

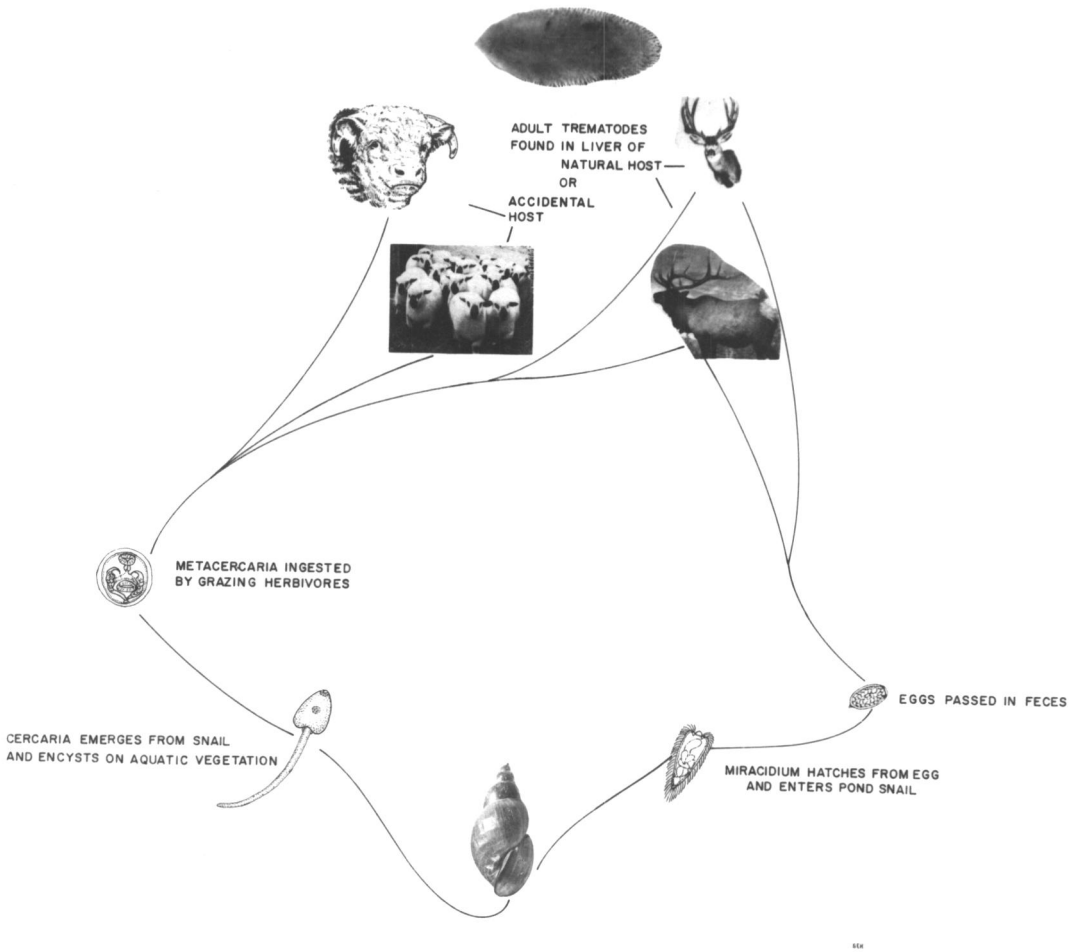


Figure 1. LIFE CYCLE OF FASCIOLOIDES MAGNA (BASSI, 1874)

28, p. 345).

There is no known effective method of treating infected animals (28, p. 345). In domestic animals fascioloidiasis may be controlled by erradicating snails and, in some instances, eliminating infected deer and elk.

The importance of this disease has not previously been recognized in the northwestern states, although the parasite has been observed occasionally from specific areas of Oregon and Washington. This study was initiated in March 1963 to determine: a) the distribution and incidence of F. magna in Bovidae and Cervidae, b) the importance of this disease problem c) the snail intermediate hosts, d) snail host bionomics and e) the incubation period of F. magna ova.

REVIEW OF LITERATURE

History and Nomenclature

The Fascioloides magna literature has been extensively reviewed by Stiles (58, p. 175-177) in 1894 and 1895, Hall (30, p. 432-436) in 1912, Swales (60, p. 178-181) in 1935 and Price (46, p. 122-124) in 1953. A brief summary of Stiles' review follows.

The large American liver fluke was discovered by Bassi, 1875, during an epizootic among deer in Royal Park near Turin, Italy. He believed the parasite had been introduced to the park in elk from America. Bassi named the parasite Distomum magnum but apparently gave an inadequate description of it. Between 1887 and 1889 American authors, not realizing they were working with the same parasite or because they did not accept Bassi's description, proposed new names. In 1887 Curtice mistakenly identified F. magna in three Kansas cattle as Fasciola hepatica (Distoma hepaticum). Two years later Dinwiddie found the parasite in Texas and mistakenly identified it as Fasciola hepatica. Later both authors changed their identifications to Fasciola magna. Sonsino in 1890 called this trematode Fasciola gigantia. Hassall in 1891 called it Fasciola carnosa and then changed the name to Fasciola americana. That same year Francis named it Distomum texanicum and Leidy gave it the name Distoma crassum. Leidy declared this parasite identical to one found in a

young male Chinese [According to Swales (60, p. 179) however, this fluke was a different species]. Perroncito in 1882 titled his description "Distoma grande. Distoma magno (Distoma magnum, Bassi)" which agreed with Bassi's identification. Stiles supposed the term "grande" not a new Latin name for this species, but included Distoma grande in the list of synonyms because of the confusion in Perroncito's title. Stossich, 1892, rejected the names Fasciola gigantia and F. magna and renamed the parasite Cladocoelium giganteum.

The confusion was eliminated in 1892 when Leuckart and Stiles compared specimens of both European and American authors and discovered they were the same species (58, p. 178). After establishing this fact, Stiles (58, p. 225-243) proposed the parasite be named Fasciola magna (Bassi, 1875) Stiles 1894.

Stiles (58, p. 225-243) also published the first complete, accurate description of F. magna. He demonstrated the differences between this trematode and F. hepatica, describing the external and internal anatomy, the egg, and miracidium.

In 1917 Ward (62) stated that Fasciola magna differed from F. hepatica as it lacked an anterior cone and the vitellaria were ventral to the intestinal branches instead of intermingling. He contended these differences constituted a new genus and proposed Fascioloides magna (Bassi, 1875) Ward 1917. The synonyms are listed in Table I.

TABLE I. LIST OF SYNONYMS FOR FASCIOLOIDES MAGNA.

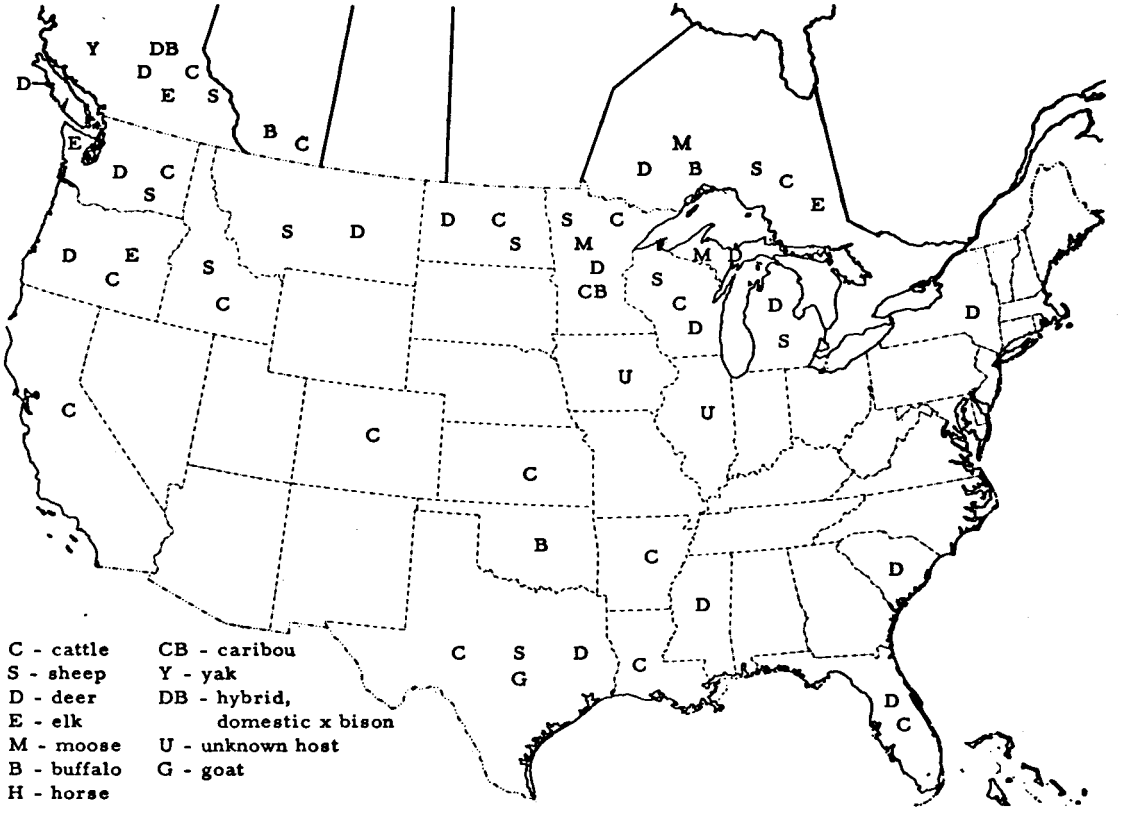
Parasite	Author	Year
Distomum magnum	Bassi	1875
Distoma grande	Perroncito	1882
Distoma hepaticum	Curtice	1887
Fasciola hepatica	Dinwiddie	1889
Fasciola gigantia	Sonsino	1890
Fasciola carnosa	Hassall	1891
Fasciola americana	Hassall	1891
Distomum texanicum	Francis	1891
Distoma crassum	Leidy	1891
Cladocoelium giganteum	Stossich	1892
Fasciola magna (Bassi, 1875)	Stiles	1894
Fascioloides magna (Bassi, 1875)	Ward	1917

Slusarski (56), 1955, adopted the earlier combination, Fasciola magna, (Bassi, 1875) Stiles 1894, since he did not consider Ward's Fascioloides justifiable. Erhardová (15), 1961, also did not recognize Ward (62) and used the name Fasciola magna.

Distribution and Incidence

United States and Canada

Most reports of F. magna have originated from the United States. Figure 2 shows the distribution of known enzootic areas.



[1] Location of horse unknown.

Figure 2. Known distribution of F. magna in the United States and Canada.

Hall in 1912(30, p. 433-435) summarized the known distribution. The parasite was reported from an infected white-tailed deer (Odocoileus virginianus) from Mississippi and infected cattle (Bos taurus) from Colorado, Arkansas, Kansas, Texas and Louisana. No host was listed for an infection in Oklahoma. He also listed a report by Curtice of infected dairy cattle in Marin County, California. There were several unconfirmed reports from other states. Included was a report of infected sheep (Ovis aries) which may have come from Washington. A second case of F. magna infected sheep from Ovando, Montana was reported by Hall (31) in 1914.

In 1916 Hadwen (29, p. 511) discovered F. magna in a black-tailed deer (Odocoileus hemionus columbianus) on Texada Island in the Straits of Georgia, British Columbia. Kermode (33), 1916, also reported an epizootic among deer on the same Island.

Of 400 buffalo (Bison bison) slaughtered in 1923 at the National Buffalo Park, Wainwright, Alberta, Canada, Cameron (2, p. 334; 3, p. 417) found there were several infected.

Sinitsin (54, p. 273, 274; 55, p. 186), 1930 and 1933, searched for an enzootic area throughout the southern states but observed cattle infections only in Houston and Fort Worth, Texas slaughter houses.

Swales (59, p. 472, 473, 476) included F. magna in a check list of important helminths in Canada, 1933. In 1935 Swales' list

(60, p. 180) of previous reports for the United States included cattle, wapiti (Cervus canadensis), moose (Alces alces americana), white-tailed deer, sheep a "goat (Capra hircus)" and a horse (equus caballus). For Canada he listed buffalo and black-tailed deer. He reported that the coast deer (O. h. columbianus) of British Columbia and Vancouver Island were severely infected. During his study he found natural infections in yak (Bos grunniens), hybrid domestic cattle X bison (Bos taurus X Bison bison), cattle, white-tailed deer, mule deer (Odocoileus hemionus) and wapiti. He was able to experimentally infect the following animals: sheep, domestic rabbits (Lepus cuniculus var. domesticus) and guinea pigs (Cavia porcellus).

In addition to the states already discussed Krull (37, p. 509), 1933, reported that this trematode occurred in Illinois and Iowa; however, no hosts were given.

The first report in moose was provided by Fenstermacher (18, p. 10) in Minnesota, 1934. Infected moose were again reported from the same area in 1942 by Fenstermacher and Olsen (19, p. 241; 44, p. 404, 406). A year later they found 19 of 92 Minnesota deer infected (45, p. 5). Olsen (43, p. 26, 27), 1949, discovered 36 of 52 deer infected in Refugio County, Texas and reported that goats, sheep, deer and cattle from the coastal prairie of Texas were infected.

In Florida, Schwart (48), 1938, found F. magna in a large

number of deer and in one cow.

Whitlock (63, p. 245-246), 1939, reported that 20 percent of the white-tailed deer in the upper peninsula of Michigan were infected but only six cases had been reported from the lower peninsula.

Dinaburg (14, p. 102-103), 1939, found 23 diseased deer in Florida and recorded one parasitized white-tailed deer from South Carolina.

Cowan (11, p. K42-K43) in 1941 noted an infected British Columbia deer. He again reported the disease near Courteney, the Alberni Valley and Englishman's River Valley, British Columbia in 1946 (12, p. 85).

Erickson and Highby (17) in 1942 reported a Woodland caribou (Rangifer caribou) of Red Lake, Minnesota had once been infected with F. magna, but at the time Dr. Reuel Fenstermacher examined it, only a cyst and eggs were observed.

Dikmans (13, p. 219), 1945, reported F. magna occurring in cattle, sheep and goats in Minnesota, Idaho, Texas, Louisana, Arkansas and the Canadian provinces of British Columbia, Alberta and Ontario. This information is not included in Figure 2 since Dikmans stated that it was partially presumptive.

Shaw (50, p. 15) in 1947 reported finding the trematode in an Oregon elk. Also, he (51) found the parasite in a white-tailed deer collected near Clatskanie, Oregon.

Kingscote (34, p. 33, 35), 1949, reported this parasite in cattle and sheep in the District of Sudbury, Ontario, Canada and in elk, deer, buffalo, cattle and sheep in the Burwash District, Ontario. He also reported a heavy infection in an elk herd. No hosts were given for the parasites which occurred in the Rainy River area, Kenora Districts and Frontenac County of Ontario. In addition to these, Renfrew, Hastings, Peterborough, Bruce, Carleton, Algoma and Nipissing counties or districts were enzootic for F. magna. Of 203 deer from widely separated areas of Ontario, 13.2 percent were infected. Fecal examinations of 112 deer revealed 11.6 percent infected. Post mortem examinations of elk disclosed 78 infections; by fecal examination, 36 of 77 animals checked were positive. His incidence data were exclusive of known infected elk herds. Examination of ten moose droppings showed four infected. The one liver examined was negative. Kingcote's records show 1200 head of cattle and sheep in Ontario have suffered or died from this fluke. Nine of 29 flocks of sheep were positive and 44 beef livers in one abattoir were condemned. Throughout this extensive survey sixteen counties or districts with a total of 42 townships or localities were checked.

Kingscote (35, p. 203) also reported the occurrence of F. magna in deer and moose in the upper Michigan peninsula.

Cheatum (10, p. 217), 1951, reported that studies conducted in the New York Adirondack Mountains revealed that deer frequently

suffered from fascioloidiasis.

Price (46, p. 122-124) in 1953 reviewed the literature describing the major contributions and giving the known distribution of F. magna at the time.

Shumard and Eveleth (52, p. 61) reported the parasite in a steer from North Dakota in 1954.

Campbell and Todd (7) in 1954 recorded three infections in northern Wisconsin sheep. Campbell (4, p. 36, 41, 43), 1957, added 208 infected deer, two cattle and 25 sheep from Wisconsin.

Griffiths (26), 1955, gave another report of F. magna in deer, cattle and sheep in Minnesota but did not cite the number of infections. His check list of 1962 (28, p. 343) included infected cattle from Idaho, Texas, Minnesota, Arkansas, Louisiana and three Canadian provinces, British Columbia, Alberta and Ontario; sheep from Washington, Idaho, North Dakota, Minnesota, Wisconsin, Michigan and Ontario; and deer from Oregon, North Dakota, Texas, Minnesota, Wisconsin, Michigan, New York, South Carolina, Florida and British Columbia. He also gave a complete account of the occurrences in Minnesota. Of 15,608 Minnesota cattle slaughtered between 1952 and 1960, 30.8 percent of the livers were condemned.

Senger (49, p. 5, 9) in 1963 reported F. magna in a large number of Montana deer but did not state the species of deer involved.

Knapp and Shaw (36), 1963, reported finding the infection in four

cattle and two black-tailed deer. All were from Douglas County, Oregon.

Europe

The original discovery of F. magna by Bassi, according to Stiles (58, p. 175), took place in Italy in 1875. Swales (60, p. 180, 207), 1935, listed sheep, wapiti, blue bull (Boselaphus tragocamelus), fallow deer, (Dama dama), red deer (Cervus elaphus) and sambur (Cervus unicolor) as having been reported from Italy and red deer from Gorlitz, Germany.

In 1956 Erhardová and Kotrly (16) recorded the presence of F. magna in Czechoslovakian wild ruminants. The specific animals were not made known. Erhardová (15), 1961, reported this trematode in Italy, Poland, Germany and Czechoslovakia.

Slusarski (56, p. 52-55) briefly reviewed the European literature on the distribution of Fasciola magna (Fascioloides magna) and described its occurrence in a red deer from Lower Silesia. Again in 1956 Slusarski (57) reported the occurrence of Fasciola magna in red deer and cattle from Upper and Lower Silesia, Poland.

Life History

Hall (30, p. 432) in 1912 stated: "From the close relation of this fluke and the common liver fluke of sheep, and from the fact that

the infected range of the two forms is very nearly the same, cattle frequently being infected with both flukes at the same time, it is probable that the life history of the two flukes will be found very similar and the intermediate hosts perhaps the same."

Sinitsin (54, p. 274), 1930, was first to record an intermediate host for F. magna. Naturally infected Stagnicola bulimoides techella (Galba bulimoides techella) were collected in Southern Texas.

Krull (38) in 1933 confirmed Sinitsin's work by experimentally infecting S. b. techella and recovered cercariae. Also he showed that Fossaria modicella rustica (Lea) and Pseudosuccinea columella (Say) could be experimentally infected. The intermolluscan phase took 40 days in F. m. rustica and 49 days in P. columella. Again in 1933 he (37) demonstrated that Fossaria modicella could serve as an intermediate host.

Swales (60, p. 181) found naturally infected Fossaria parva (Lea) and Stagnicola palustris nuttalliana (Lea) and described their bionomics. He also was able to artificially infect both species. The intermolluscan stage took from 49 to 58 days.

Working in Canada, Swales (60) conducted the first complete life history study. He found the egg had an average length of 148 μ and a width of 94 μ. Campbell (6, p. 309) described the eggs as being 114 μ long by 96 μ wide. Stiles (58, p. 242) found measurements of 109 μ to 168 μ in length and 75 μ to 96 μ in width.

Swales (60, p. 187) claimed the duration of incubation was 35 days. Eggs were rapidly destroyed when fecal humidity dropped below 50 percent. At a mean temperature of 24° C (maximum 27° C, mimimum 21° C) the eggs reached the morula stage in ten days and hatched as early as 29 days.

In a detailed study of the development of the egg and miracidia, Campbell (6, p. 308-313) reported an incubation period of 21 days. The earliest hatching occurred on the 17th day. Water containing the eggs was aerated and held at 25±1° C. Abnormal embryonation occurred and was always lethal when the ova were incubated at 34° C.

The morphology and behavior of the miracidia have been described by Swales (60, p. 191-192), Campbell (6, p. 314-315) and Campbell and Todd (9, p. 343). The latter (9, p. 343) pointed out in 1955 that the diffusion of possible attractants under experimental conditions was insignificant in guiding the miracidium to the snail. Apparently he (6, p. 318) corrected himself in 1961 by stating that there was a chemical stimulus provided by the snail. In the same paper he also demonstrated positive phototaxis in the miracidia.

Swales (60, p. 192-193) and Campbell and Todd (8, p. 226, 228) described the morphology and metamorphosis of the miracidium to sporocyst and redial stages. Swales (60, p. 196, 201) gave an excellent account of the description of the daughter rediae and the cercarial development and explained that the emergence of the

cercariae took place most frequently during dark hours. Cercaria did not encyst on the surface of the water but usually attached to objects within three centimeters of the water surface.

In 1953 Wu and Kingscote (64) reported having artificially infected Lymnaea stagnalis (L.). They (65, p. 90) were able to induce hatching of F. magna eggs on the 19th through the 26th days in cultures having water changed every two days. Cercariae were collected from mass exposed snails. Rediae developed in the repiratory chamber, mantle, the margin of the foot and external surface of the gut.

Griffiths (26), 1955, reported Stagnicola palustris (Müller) as an experimental intermediate host in Minnesota. In 1959 he (27) was able to collect a naturally infected Stagnicola (Hinkleyia) caperata (Say). In 1962 he (28, p. 344) experimentally infected both S. palustris and S. caperata.

Erhardová, (15) in Czechoslovakia experimentally infected 80 percent of Galba truncatula. He also was able to experimentally infect Lymnaea peregra peregra and Lymnaea peregra ovata. The intermolluscan cycle was completed in 40 to 44 days.

Knapp and Shaw (36), 1963, reported S. palustris (Müller) and S. bulimoides as being associated with enzootic areas of F. magna. As was mentioned earlier S. palustris is a vector of this parasite but the role of S. bulimoides is not known. A closely related snail

S. b. techella has been proven to be a host (54, p. 274; 38).

Campbell and Todd (8, p. 225) reported Stagnicola reflexa as a possible intermediate host. Natural infections for this snail have not been reported. As a result of abnormal shedding of metacercariae, Campbell considered this snail an unsatisfactory host.

An extensive study of the ecology and life history of Lymnaea humilus (Say) (Fossaria parva (Lea)) was conducted by McGraw (40, p. 118; 41, p. 16-27). The characteristic mud flat habitat was described and plant growth listed. Lynmaea humilus was able to tolerate dry periods in natural conditions. This snail was considered to be an incipient land species.

Olsen (42, p. 401-402) studied the bionomics of S. b. techella, a snail host for F. magna. The most favorable habitat was the Texas Gulf Coast region where the clay soils are uniformly flat, low and poorly drained and the climate mild and moist. His study showed that the snail lived in both permanent and temporary pools. This snail had a cycle of two generations annually with the young estivating in the soil until fall rains.

Friedl (21, p. 75) in 1961 published a study on in vitro survival of the rediae of F. magna in various media. Survival was checked in relation to the presence or absence of various compounds in an effort to better understand the metabolic requirements of this form. That same year he (22, p. 247) developed methods of obtaining

bacteria-free rediae of F. magna and tested their survival in various media under axenic conditions. In 1960 and 1961 Friedl (20; 23, p. 772) described a method of hatching embryonated ova of F. magna by subjecting them to a partial vacuum and by exposing them to a nitrogen atmosphere. Large numbers of miracidia were easily obtained. Again in 1961 he (24, p. 776) studied the effect of free amino acids in hemolymph of Lymnaea stagnalis jugularis (Say) on the survival of F. magna rediae by chromatographic methods. In 1964 he (25, p. 7) reported a method of securing bacteria-free L. s. jugularis for use in studying and evaluating selectively introduced parasites.

The known intermediate hosts of F. magna are listed in Table II.

TABLE II. KNOWN INTERMEDIATE HOSTS OF F. MAGNA

Species	Natural Infection	Experimental Infection	Locality of Natural Infections	Author
Stagnicola bulimoides techella	x		Texas	Sinitsin (54, p. 274)
		x		Krull (37)
Fossaria modicella rustica		x		Krull (37)
Pseudosuccinea columella		x		Krull (37)
Fossaria modicella		x		Krull (37)
Fossaria parva	x	x	British Columbia	Swales (59, p. 181)
Stagnicola palustris nuttalliana	x	x	British Columbia	Swales (59, p. 181)
Lymnaea stagnalis		x		Wu and Kingscote (64)
Stagnicola palustris		x		Griffiths (26)
Stagnicola caperata	x		Minnesota	Griffiths (27)
		x		Griffiths (27, p. 344)
Galba truncatula		x		Erhardová (15)
*Lymnaea peregra peregra		x		Erhardová (15)
*Lymnaea peregra ovata		x		Erhardová (15)
Stagnicola reflexa		x		Campbell and Todd (7, p. 225)

*Unconfirmed

Pathogenicity

The pathogenicity of F. magna was recognized at the time of the helminth's discovery in Turin, Italy. At that time the disease condition was described as typical fascioliasis (60, p. 178). This description most likely referred to as acites, submandibular edema and emaciation.

The most severe pathogenesis occurred in sheep. This was originally recognized by Hall (30, p. 432) in 1912. He stated that a large number of sheep, presumably from Washington, had died as a result of this parasite. Again in 1914 Hall (31) said that a large number of Montana sheep had died of fascioloidiasis and described a characteristic black pigmentation of the hepatic lymph nodes and certain areas of the liver. He did not believe that the parasite caused severe pathogenesis in cattle. In sheep, however, many deaths resulted.

Swales (60, p. 214; 61, p. 92-93) described the effect of F. magna on sheep (Ovis aries) as acutely pathogenic owing to their inability to confine the fluke. However, Campbell (4) was able to show that mechanical damage was not the principle cause of death in sheep but rather that the fluke produced an "antimetabolite" which disseminated throughout the body during the burrowing stages. Since sheep were unable to confine the fluke, dissemination of the toxin was prolonged. Rarely was the fluke found encysted by fibrous tissue. If this

happened the fluke sometimes matured and the eggs along with the toxic products of the fluke escaped through the biliary system as in the natural definitive hosts, Cervidae (39, p. 26). Evidence by Campbell (7), Krull (39, p. 27), Price (47, p. 150) and others support the hypothesis that F. magna can occasionally reproduce in goats and sheep.

Swales (60, p. 208-210; 61, p. 83-85) showed that F. magna provoked severe tissue reactions in cattle which resulted in encystation of the parasite in a thick fibrous cyst. Fibrous tissue which formed the cyst blocked afferent and efferent bile ducts. Bile duct blockage resulted in the accumulation of ova and vomitus from the fluke. A black pigment associated with fascioloidiasis was found to discolor liver tissue and lymph nodes. Pigmentation occurred primarily during the migratory and closed cyst stages.

According to Campbell (5, p. 774) the black pigmentation had for years been referred to as melanosis, melanoid pigment (46, p. 124) and other "non-committal terms" but was commonly understood to be melanin. Sinclair (53, p. 69) in 1949 was first to describe the pigment as hematin but did not say how he arrived at this conclusion. Campbell's tests described the pigment as a hem compound, possibly hematin (5, p. 773-774).

Swales (61, p. 83), 1936, compared tissue reactions of Bos taurus, Bison bison, B. taurus X B. bison, Cervus canadensis,

Odocoileus virginianus and Ovis aries in relation to fascioloidiasis. Host defense reactions in the liver were recorded for each animal. The data had been obtained to support his hypothesis that Cervidae were the natural hosts and that cattle were accidental hosts.

Evidently Hall (32) in 1930 was first to consider the Cervidae as natural hosts. Descriptions by Hadwen (29, p. 511) of the cysts and dilated bile ducts containing flukes indicated that deer were natural hosts. The ducts were quite large, containing two or three flukes and sometimes "inky" bile.

Swales (60, p. 208-211) described the open cavities of Cervidae livers and the lack of pigmentation that occurred. He also demonstrated the relatively low pathogenicity and reproductive capacity of F. magna in these hosts. On the basis of this information he judged that Cervidae were the natural hosts. Since this time, Olsen (43, p. 27), Cowan (12, p. 85), Shumard and Eveleth (51, p. 61), Griffiths (28, p. 345) and others have supported the hypothesis.

Whitlock (63, p. 245-246) stated that deer are tolerant of moderate infections. Only heavy infestations caused emaciation or death. He gave an account of an apparently healthy doe harboring 22 flukes.

Cowan (11, p. K42-K43) reported a deer in good condition infected with 23 flukes and nine heavily infected deer which died of hepatic portal hemorrhaging due to lesions caused by the flukes. The hemorrhaging apparently occurred as the deer were being chased

by domestic dogs. He also reported an instance where F. magna rendered deer more vulnerable to attacks by predators.

Kingscote (34, p. 33, 35), 1949, recorded heavily infected elk in Ontario, Canada. Older animals suffered extensive damage while younger ones were less severely affected. Up to 176 flukes were found in one liver. He did not state whether the flukes were mature or immature.

Control

Swales (60, p. 212) stated the control of fascioloidiasis can be accomplished by two methods: (1) erradicating snail hosts with various applications of copper sulfate and (2) eliminating infected Cervidae from areas grazed by cattle and sheep.

Griffiths (28, p. 345) and Swales (60, p. 211) have agreed that anthelmintic treatment of cattle probably would be ineffective because of the parasite's location in closed cysts of the liver.

Kingscote (35, p. 207) reported that heavily infected elk herds were eliminated for the protection of domestic ruminants in Ontario, Canada. He also reported on combined control methods involving the administration of hexachloroethane to cattle and sheep, erradication of snails with copper sulfate and selection of safe hay pastures.

MATERIALS AND METHODS

Enzootic Area Locations

Bovidae

Reports of infected cattle were obtained through federal and state inspected abattoirs, particularly those located in Portland, Oregon. Most inspectors were contacted and the characteristics of the disease were explained to them. They were asked to report any infections and include in the report the back tag and brand or ear tag identification to aid in tracing the animals. If the back tag was given, the cattle were traceable through the United States Meat Inspection records. State records were used to trace the cattle on which brand and ear tags were received. Frequently no identification was received and the animals were traced through the commission companies and shippers. If a large number of animals were involved the work was simplified; whereas, tracing one or two infected cattle often failed.

After locating the origin of infection the livestock producers were questioned to confirm the report and to locate the grazing areas where the animals may have become infected.

Slaughter houses in Portland, Roseburg, Bandon, Tillamook and some plants in the Willamette Valley were asked to report information on infected cattle.

Cervidae

Reports of infected animals were obtained through examination of hunter killed deer and elk or by special kill permits issued by the Oregon State Game Commission for examination and recovery of fluke eggs. A description of the infection was given to the Oregon State Game Commission field men who aided in searching for infected animals during the deer and elk hunting seasons.

Checking deer and elk livers in Millacoma Forest near Coos Bay and MacDonald Forest near Corvallis, Oregon (Figure 5) was undertaken because of the large number of animals killed and because hunters were required to check in and out at the same station. As they checked into the area they were requested to bring in livers for examination.

Deer and elk kills were also examined in Clatsop and Lane Counties, Oregon. No livers were examined from animals killed near Coos or Klamath Counties, Oregon. Neither were any examined in southern Washington although ranchers in Cowlitz County where cattle infections had been reported were questioned on the occurrence of F. magna in deer.

Tenasillahae Island Study

Tenasillahae Island is 1700 acres of low land located near Clifton, Oregon in the Columbia River (Figure 3). The island was diked in

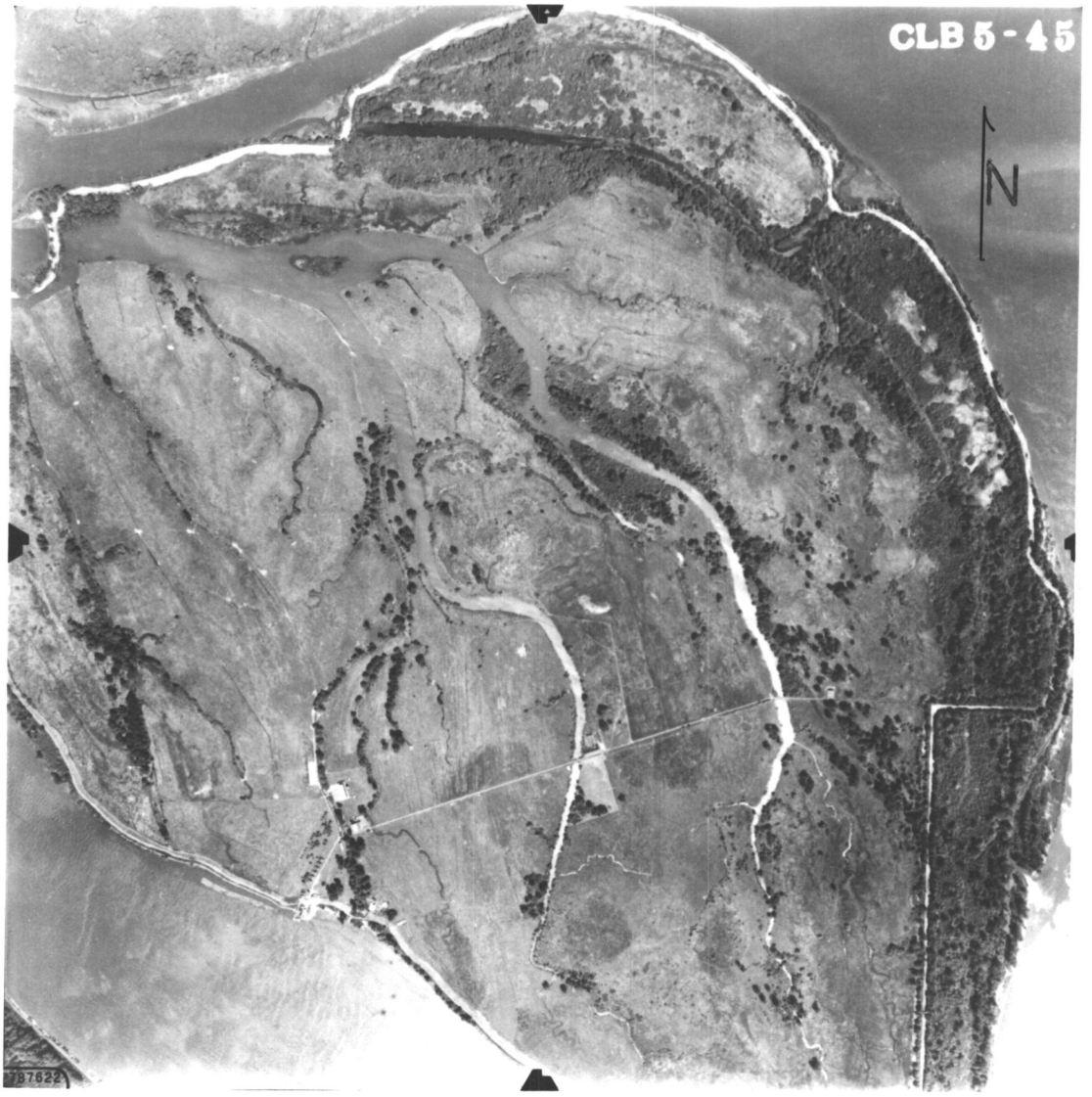


Figure 3. Aerial photograph of Tenasillahae Island showing the numerous water channels and ponds. The dike is the white line surrounding the island.

1907 and has approximately 250 acres of standing water in channels, ponds and drainage ditches, 450 acres of brush and 1000 acres of grazing land. The water table is within a few inches of ground level; therefore, most of the grazing land is soft throughout the winter and spring. The high point on the island is a 13 foot dike. As the tide recedes, water gates open and the island water flows out. The incoming tides of approximately coastal height close the gates, keeping the water level fairly constant throughout the year.

Large numbers of confirmed reports of F. magna infected beef encouraged the use of this island as a research area. Other reasons were ease of surveying the snail and deer populations and ease of killing and examining infected deer.

The island was systematically searched for snails and the dates of collection, identification and bionomics recorded.

An estimate of the number of deer inhabiting the island was obtained by day and spotlight counts. The incidence of infection in deer was indicated by fecal and liver examinations.

Liver Diagnosis

Cattle livers collected in the abattoirs were cooled (7° C) until transported to the laboratory for examination. Liver pigmentation, fluke burrowings, number and description of closed cysts, number and maturity of flukes present and presence of ova in the gall bladder

were recorded.

Detection of infected deer and elk livers was made by examining 1) lymph nodes and liver tissue for pigmentation, 2) the bile for F. magna ova and 3) the liver capsule for scar tissue. Infected livers were returned to the laboratory where flukes were counted, ova recovered from the cavities and ducts and descriptions of the lesions recorded.

Intermediate Host Investigations

When an enzootic area was located a survey was made to determine what species of snails were present.

Snails collected from enzootic areas were examined for cercariae or rediae by crushing and observing under a binocular microscope. Snails from each area were also held in aquaria to determine if they would shed cercariae.

Examinations of large numbers of snails were made. If not infected, these presumably disease-free snails from each area were individually or mass exposed to F. magna miracidia. Each exposure experiment varied in the number of miracidia per individual snail. Other controls on the infection experiments were water temperature at the time of exposure and during incubation, age of miracidia, age of the snails, snail species and type of aquaria (Tables IV-VII). If exposed snails survived laboratory conditions, they were observed

for at least 90 days for possible shedding of cercariae. If none were shed by the end of the culture period, the snails were examined by crushing as described previously.

Most experiments were conducted in aquaria provided with water filters and aeration. Distilled water was generally used. Snail food consisted of fresh lettuce.

Since Physaidae, Succineidae and Planorbidae have not been involved as snail hosts they were not exposed during this study.

Snail ecology was recorded at the time of collection. Data noted were pH of the water, type of pond or drainage ditch, depth at which the snails appeared to congregate, bottom description, relation of snails to grass surrounding the body of water, water movement, approximate number of snails and presence or absence of snails at various seasons.

F. magna Ova Incubation and Hatching

Two methods of egg incubation were used. In the first, eggs from bovine liver cysts and uteri of the flukes were contained separately in finger bowls and incubated in shallow aerated water at room temperature (maximum 26.5° C, minimum 15.5° C, mean 20.2° C).

In the second method, eggs from fluke uteri and from the livers of both Bovidae and Cervidae were incubated in an "egg bath". The "egg bath" (Figure 4) was a glass aquarium measuring six and

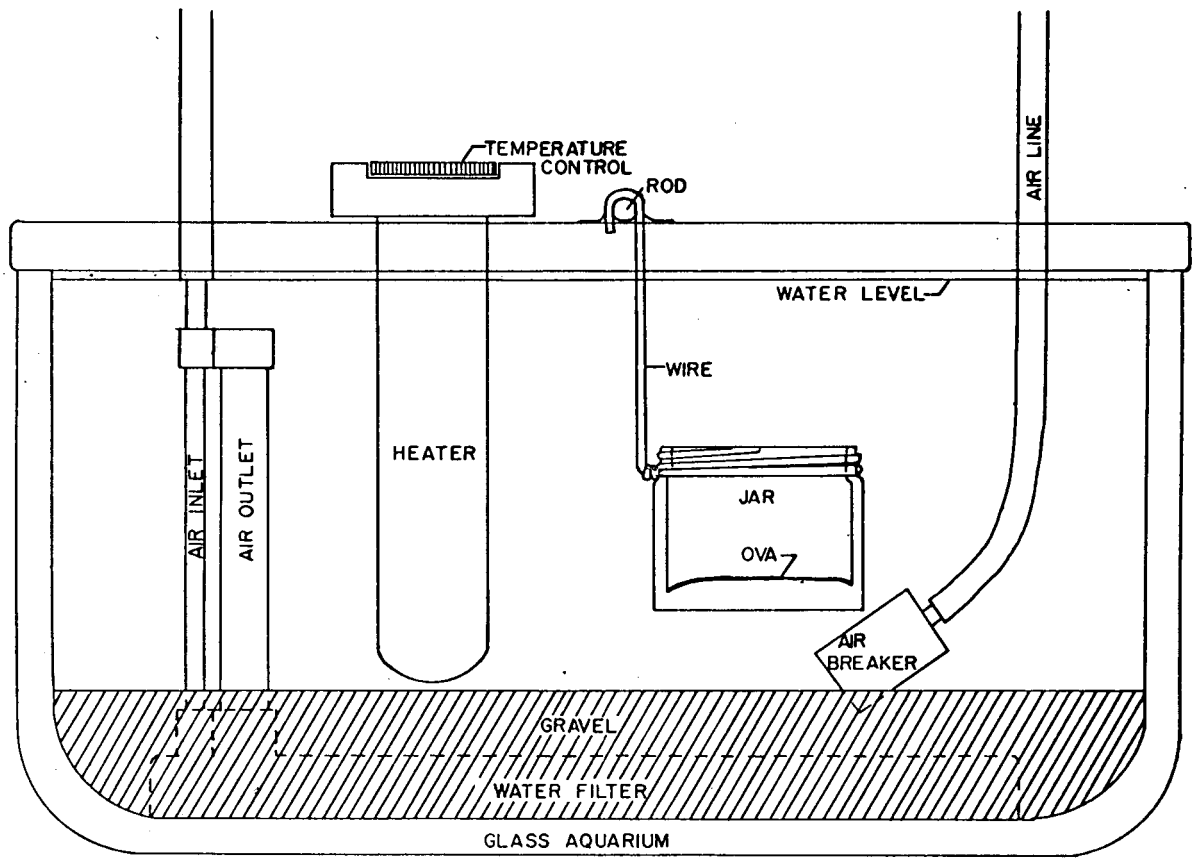


Figure 4. Egg bath.

one-half inches in width, 11 inches in length and six inches in height with a volume of one gallon. An air operated gravel filter placed in the bottom of the aquarium filtered and aerated the water as it bubbled through the outlet tube. An adjustible heater kept the water at the desired temperatures of 25-28° C. Ova were allowed to settle to the bottom of the small wide-mouthed jar completely filled with distilled water. Stone air breakers were placed on the bottom of the aquarium and the jars slowly lowered into the water and suspended from glass rods. As the jars were lowered only a slow exchange of water occurred and the eggs were not disturbed. A small stream of bubbles provided adequate circulation of aerated water over the layer of eggs. Water was added to the bath when needed but no other care was necessary. Thus clean, aerated water of the desired temperature flowed over the eggs throughout their incubation.

Eggs were checked for development by drawing some from the bottom of the jar into a small pipette and examining them microscopically. The air breaker was turned off when the eggs were removed or replaced and until they had settled.

Photographs of the best developed eggs were made daily after development began. When the eggs appeared to be totally developed they were washed in a 325 mesh-per-square-inch sieve with room temperature distilled water, returned to the same jar and allowed to sit in daylight, but not direct sunlight, for one hour. If no hatching

occurred, this process was repeated each day until the first miracidia were observed.

F. magna Ova Morphology

The morphology of eggs taken from cattle and deer and elk livers were compared. A percentage of eggs having a small, tail-like projection described by Swales (60, p. 187) was obtained. Size comparisons of eggs in Oregon elk feces were compared with measurements reported by Swales (60, p. 187).

RESULTS

Definitive Host Distribution

Bovidae

One-hundred and seven reports of Fascioloides magna in Oregon cattle were obtained through packing establishments. These were traced to the following areas: 91 to Tenasillahae Island (Clatsop County), five to Antelope (Wheeler County), two to Klamath Falls (Klamath County), one to Roseburg (Douglas County), three to Glide (Douglas County), one to Langlois (Coos County), two to Portland (Multnomah County) and two untraceable (Figure 5).

As indicated by the infection reports and interviews with meat inspectors, Tenasillahae Island was a major source of infections in Oregon. This study area will be discussed in a separate section.

Inspectors of five Portland slaughter houses stated they had seen infected cattle from many areas of the Columbia River. One inspector[1] stated that infected beef from Deer Island had been seen previously but no reports from that Island were obtained during the study.

The incidence of this disease is not known in Wheeler and Klamath Counties. Several inspectors from Portland, Langlois and Roseburg

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[1]Dr. Albert J. Jenish, Kenton Station, United States Meat Inspection, Portland, Oregon.

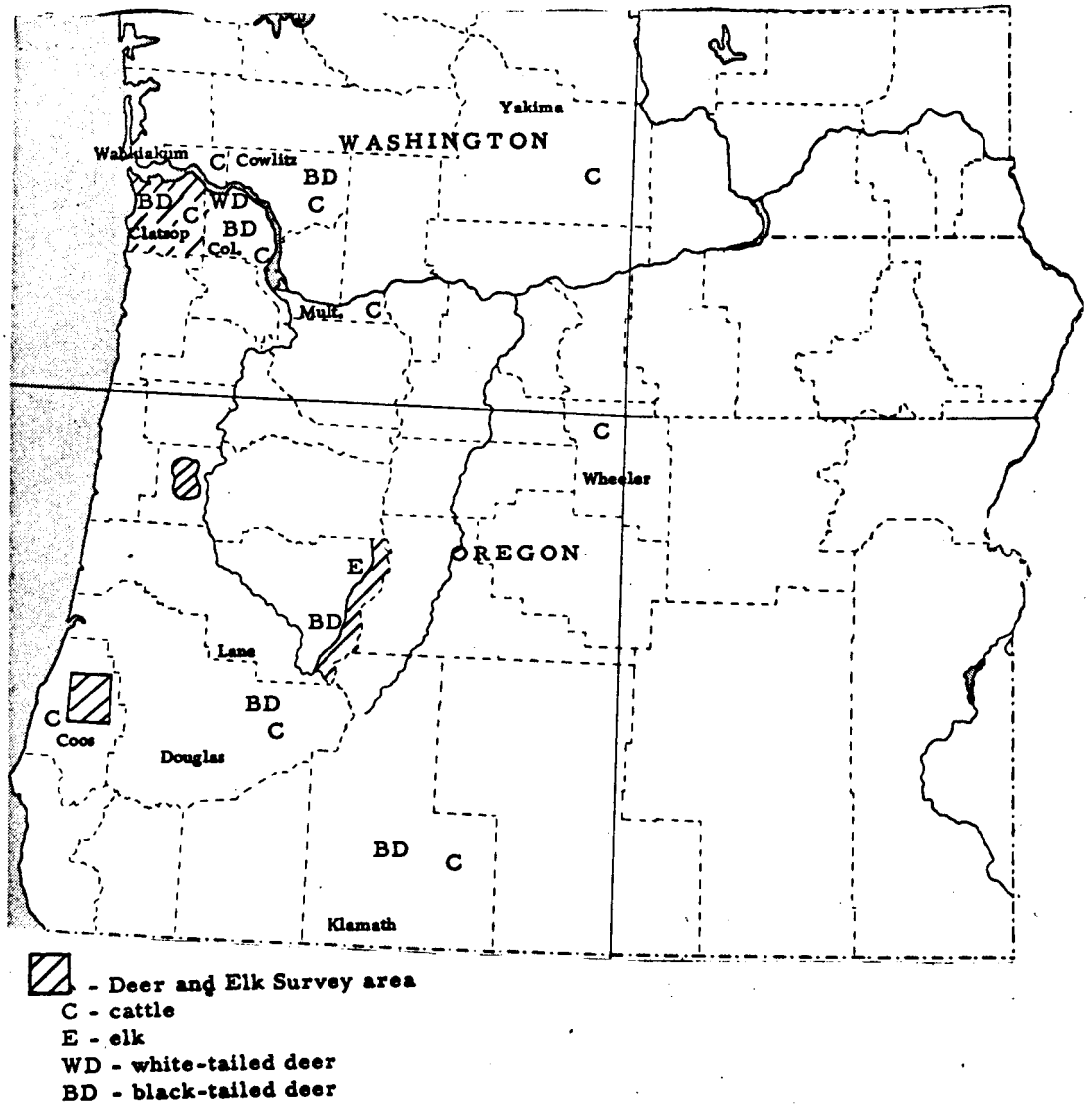

Figure 5. Fascioloides magna enzootic areas and survey locations in Oregon.

were questioned about the occurrence of F. magna in Klamath County; each agreed it commonly occurred in that area. The five beef from Antelope reportedly became infected while grazing near the John Day River.

Cattle were grazed in the Oakridge area where the parasite was enzootic in elk. However, a Eugene meat inspector who has examined cattle livers from this area stated that F. magna had not been observed.

Infected Washington cattle were discovered by Portland meat inspectors. Six diseased animals were traced to Woodland (Cowlitz County), 11 to Cathlamet (Wahkiakum County) and two to Topenish (Yakima County) (Figure 5). Several unconfirmed reports came from Sunnyside near Topenish. Woodland and Cathlamet are on the banks of the Columbia River, Woodland across the river from Deer Island and Cathlamet near Tenasillahae Island. Woodland ranchers described the characteristics of the disease and stated it commonly occurred in cattle throughout the low, flat diked area. No contact was made with livestock producers in Topenish, Cathlamet or Sunnyside.

Cervidae

A total of 150 black-tailed deer and approximately 108 elk were examined during the 1963 Oregon hunting season. In Millacoma Forest 35 deer and approximately 50 Roosevelt elk livers were

inspected. No infections were found. One hundred and fifteen deer examined from MacDonald Forest were negative. Approximately 55 Roosevelt elk were examined in Clatsop County but all were negative. Two infected Rocky Mountain elk were found in the Oakridge area of Lane County where Shaw (51) had previously reported one F. magna infection in the same host.

Six fecal examinations for fluke ova were made from elk near the Columbia River. All were negative. Fecal examinations were made of 21 elk in the Cascade range of Lane County of which 19 or 90.5 percent were positive for F. magna ova. Fecal examinations of deer on Tenasillahae Island are reported in a separate section.

Deer and elk infection reports were also obtained through interviews with hunters and ranchers. Hunters gave adequate descriptions of F. magna infections in elk from the Cascade Mountain area in Lane County. One rancher near Oakridge described six infected elk which he had killed and one infected black-tailed deer, the only deer recorded for that region. Hunters near Clifton, Oregon (Tillamook County) gave good descriptions of several fluke infected deer from small islands in the Columbia River and one infected deer near Knappa, ten miles west of Clifton. One Woodland, Washington rancher described F. magna infections in several deer killed on the diked lowland. Also, a specimen of F. magna from an elk on the Olympic peninsula of Washington has been placed in the parasite

collection at Portland State College.[2]

The Veterinary Medicine Department's parasite specimen collection contains a fluke collected in 1947 from a black-tailed deer from Klamath County. Two infected deer also have been reported from Douglas County by Knapp and Shaw (36). These are the only Cervidae infections recorded for those areas.

No Cervidae infections have been reported in Multnomah, Wheeler and Coos Counties in Oregon and Wahkiamkum and Yakima Counties in Washington.

Tenasillahae Island Study

Bovidae

During this study 91 F. magna infected cattle were traced to this island. Reports of infected animals submitted by meat inspectors usually did not indicate the total number of animals examined from a given area. For this reason only 79 animals could be used to obtain an estimate of incidence. Sixty-one or 77 percent of the 79 were found to be infected. Some cattle were infected with both F. magna and F. hepatica. Of 24 cattle examined in July 1964, 17 were infected with F. hepatica, 14 with F. magna, and ten with both trematodes. Three were not parasitized.

[2]R. W. Macy, Professor of Biology, Portland State College, Portland, Oregon.

Cervidae

Four parasitized deer were killed during the study and 13 fecal examinations were made. Providing that no duplications occurred regarding the fecal samples, 16 of 17 or 94 percent of the deer were infected.

An estimate of the total number of deer inhabiting the island was made. Thirteen deer were counted during a spotlight survey. It was estimated that not over 25 percent of the population could have been obtained, thus indicating the total deer population was not less than 52[3]. A second estimate of the island deer population indicated that there were between 35 and 75 and possibly as high as 100 animals present.[4] Approximations were based on observations of deer and their indices (tracks and fecal deposits).

Intermediate Hosts

Frequent attempts were made to collect snails between August 1963 and June 1964. Stagnicola palustris and P. columella were collected during the spring and summer. Attempts to collect Lymnaeid snails during other times of the year were unsuccessful; however, Physa spp. and Planorbis spp. were found at all times. None of the

[3]Wesley Batterson, Game Agent, Northwest Region, Oregon State Game Commission.

[4]Ira D. Luman, Chief of Big Game, Oregon State Game Commission.

collected snails were infected with F. magna.

During August 1963, P. columella were found in a small pond on the northwest side of the island but no other site was found to contain Lymnaeid snails that season. Both P. columella and S. palustris were found in drainage ditches the following spring and summer. Pseudosuccinea columella were concentrated on the west and north portions of the island. Stagnicola palustris were more widely distributed but concentrated on the south and east sections of the island (Figure 3).

Large adult P. columella and S. palustris were collected in permanent stagnant ponds along the east dike during July 1964. Only a few snails were found and in all cases these were exceptionally large. No young specimens were collected in this habitat.

Frequent attempts to collect snails in the same areas during other times of the year failed.

The drainage ditches, where most of the snails were located, contained shallow, slow-running water, mud bottoms and were banked with heavy grass. Most frequently the snails were seen crawling about on the bottom. During heavy rains when water levels rose above the confines of the ditches, Physa spp. were frequently seen crawling about in the surrounding grass; however, the Lymnaeid snails were not observed beyond the grass banks.

Liver Pathology

Bovidae

Bovine livers infected with F. magna always showed pigmentation, even with light infections. The hepatic lymph nodes were severely blackened by iron porphyrin deposits similar to those on the deer liver shown in Figure 8. Pigmentation of the liver tissue may also be seen in Figures 6 and 7. In heavy infections the iron porphyrin was deposited throughout the body cavity, especially in the diaphragm, omentum, lungs and dorsal peritoneum. In one case the entire carcass of a heavily infected bull was condemned by a Portland meat inspector[5] because of severe pigmentation.

According to Swales (60, p. 210) F. magna fluke ova are not discharged from the infected bovine liver. However, during this study one severely infected liver, containing many tracks and recently developed cysts, held nearly 800 eggs in the gall bladder. The eggs were examined and appeared to be infertile. They were incubated in aerated finger bowls but no embryonation occurred.

The cattle flukes were found in two developmental stages; the migratory or burrowing stage and the cyst stage. The most extensive damage occurred during the migratory stage. Many livers

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[5]Dr. D. L. Welch, Inspector-in-charge, Swift and Company, Portland, Oregon.

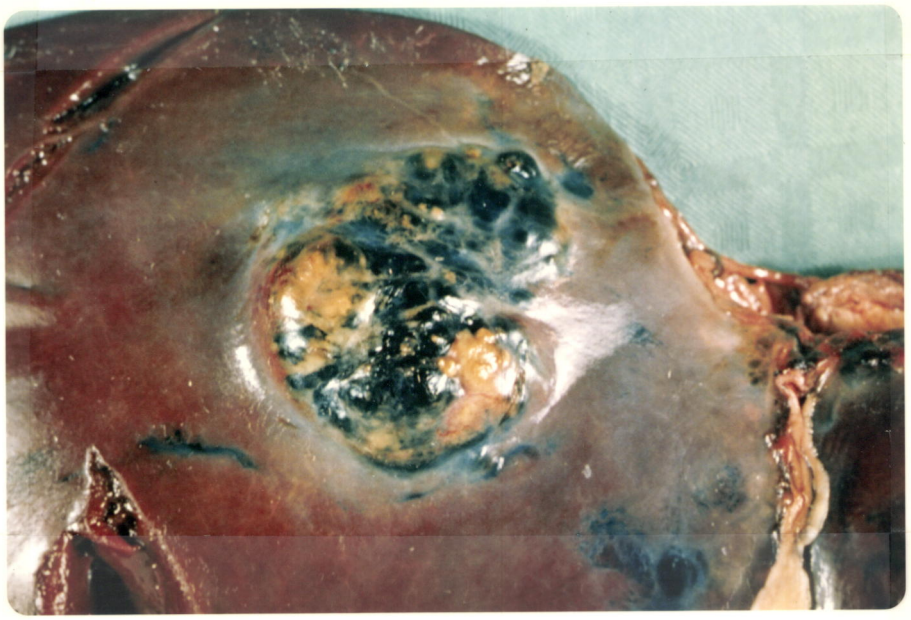


Figure 6. Cyst stage of F. magna in a bovine liver. Note pigmentation and scar tissue of the cyst.

6 cm

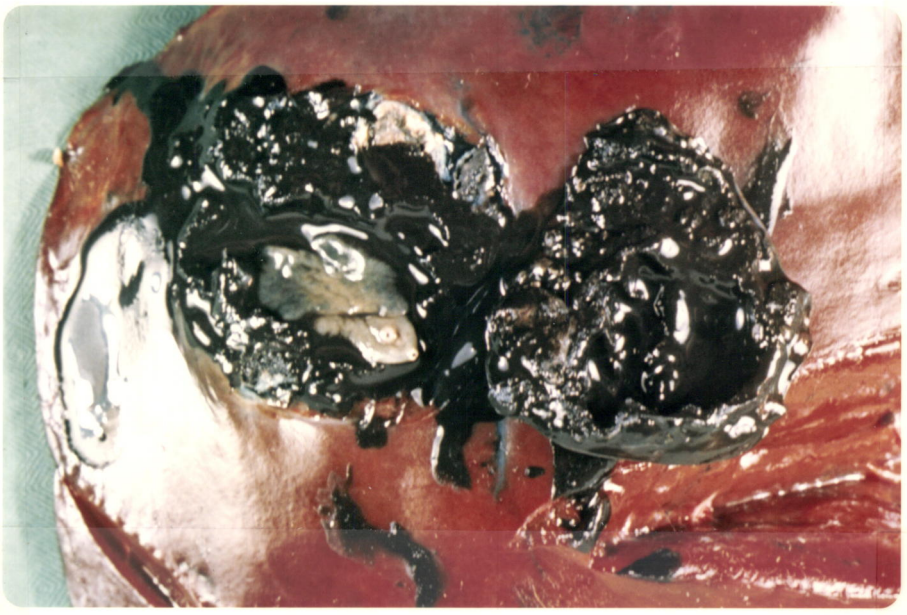


Figure 7. Opened cyst shows the black fluid and a dead fluke. Note coloration of the fluke and calcification of cyst.

showed migration tracks of the immature flukes. The fluke tracks were from ten to 15 centimeters in length and from 1.5 to 2.0 centimeters in diameter. The tracks often led inward from a small black scar on the capsule, although frequently the area near the base of the caudate lobe and the thickest portion of the liver contained most of the tracks. The tracks filled with bloody fluid which eventually healed leaving red fibrous tissue. The flukes became confined by scar tissue and calcareous-like deposits.

The movements of the fluke produced a hollow area which developed a cyst. Bile ducts intercepted by the cavity probably continued to flow for a short time, but because of rapid tissue fibrosis and deposition of a calcareous substance the bile ducts soon were blocked. After the bile ducts were blocked there was no inlet or outlet; thus a true cyst was formed. The cysts became packed with a black fluid consisting of fluke ova, pigment and debris (Figure 7). Figure 6 illustrates the characteristic pigmentation, scar tissue and bone-like material of the cyst. Many livers which were examined contained late cyst stages which were being reabsorbed. Flukes were whitish colored and appeared to be necrotic (Figure 7).

Cervidae

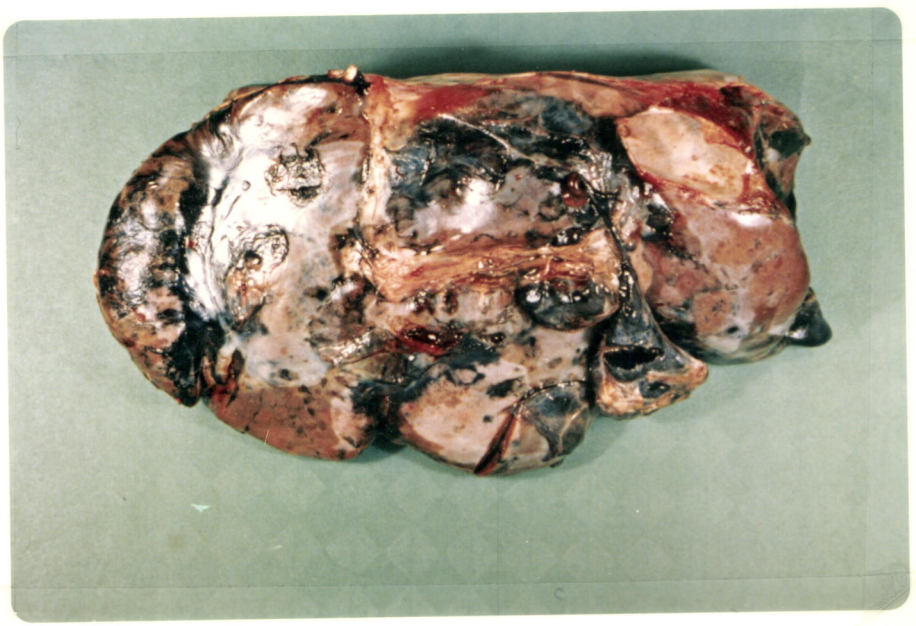
The deer and elk livers which were examined did not show the severe tissue reaction observed in cattle although pigmentation of the

lymph nodes and streaking of liver tissue occurred in each case. The livers in lightly infected deer were not swollen but lymph nodes were greatly enlarged. The heavily infected liver in Figure 8 was nearly three times the normal sized liver shown in Figure 9.

Flukes were found in two developmental stages in elk and deer although a third or migratory stage is known to occur (60, p. 206). Infected livers were examined from two elk from Lane County and four deer from Tenasillahae Island. No migratory stages were found; however, thin reddish scar tissue linings indicated early cavity stages. The mature cavity stages found in deer and elk livers were white, thin-walled and contained large afferent and efferent bile ducts.

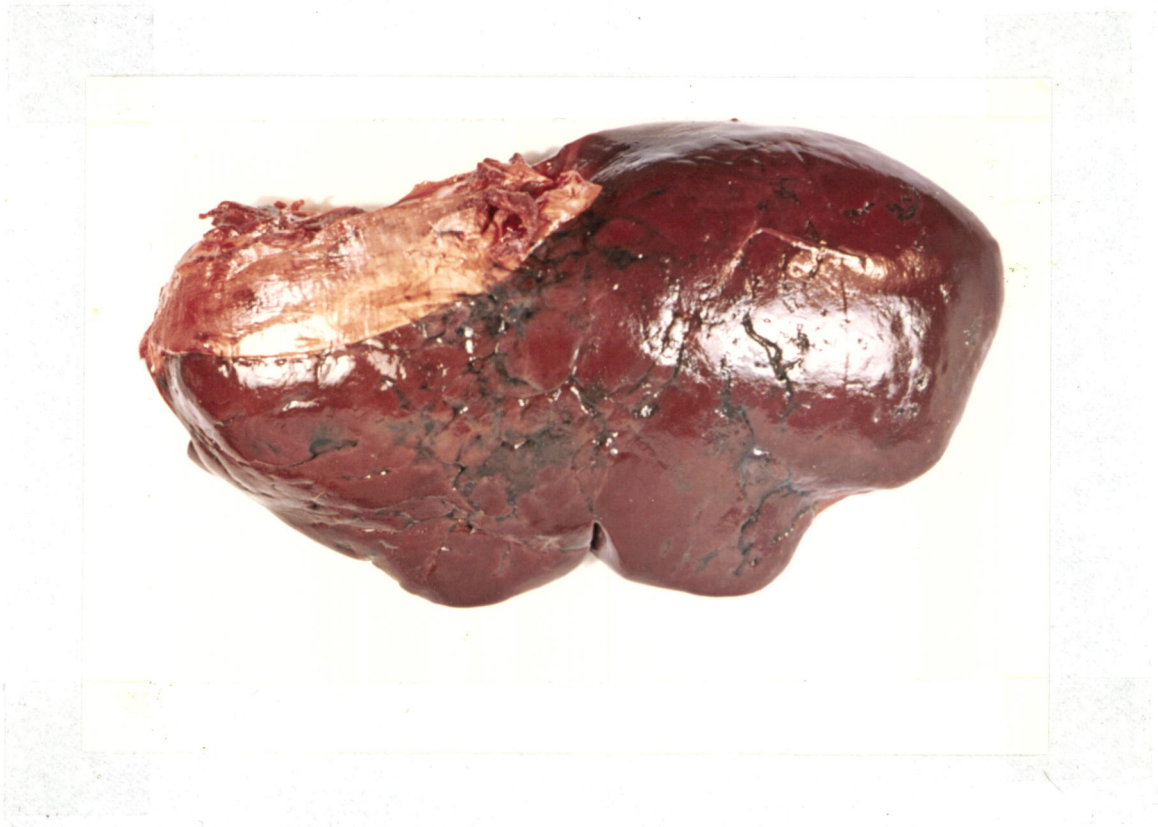
Usually two flukes were present in each cavity whereas in cattle the number occurring was more variable. The cyst stage was less common in deer and elk than in cattle; however, two deer livers from Tenasillahae Island and two elk livers from Lane County contained cysts. The liver (Figure 8) which contained 61 adult flukes also contained several closed cysts. The cysts found in deer and elk, however, were filled with eggs and did not show pigment. Frequently remains of the fluke were found within the cyst.

One cyst of a Tenasillahae Island deer appeared to have been forced outside the liver capsule and was suspended by a thin peduncle of fibrous tissue. Several other cysts, completely separated from the livers of three deer, were in the omentum.



10 cm

Figure 8. Deer liver from Tenasillahae Island containing 61 adult flukes. Note the extensive pigmentation and large size of the liver. Enlarged black lymph nodes can be seen hanging from the main bile duct which is the flesh-colored band of tissue near the center of the liver.



10 cm

Figure 9. Deer liver from Tenasillahae Island containing four flukes. Note the pigmentation of the liver capsule. Compare this liver with the liver in Figure 8 which contained 61 flukes.

The deer infected with 61 flukes was emaciated and the liver adhered to the diaphragm, omentum, parietal peritoneum and right kidney. Pigmentation occurred throughout the body cavity, especially in the hepatic lymph nodes, liver capsule (Figure 8), diaphragm, lungs and omentum. Moderate pigmentation occurred in the parietal and visceral peritoneum and the liver was about three times normal weight. The hepatic lymph nodes were greatly enlarged.

Intermediate Host Investigations

Distribution and Bionomics

Stagnicola palustris, Stagnicola bulimoides, Lymnaea auricularia and Pseudosuccinea columella were the possible snail intermediate hosts found in Oregon (Figure 10). All enzootic areas of F. magna in Oregon were surveyed for possible snail hosts with the exception of Klamath County. According to Shaw (51) previous specimens of S. palustris were collected near Klamath Falls. No information was available on other Lymnaeid snails in that area. One collection trip was made to Coos County during the spring of 1963 and only shells of S. bulimoides were found. Three field trips were made to Lane County; however, no snails were found. One attempt was made to collect snails in Wheeler County but none were found. The only F. magna enzootic area surveyed in Washington was Cowlitz County

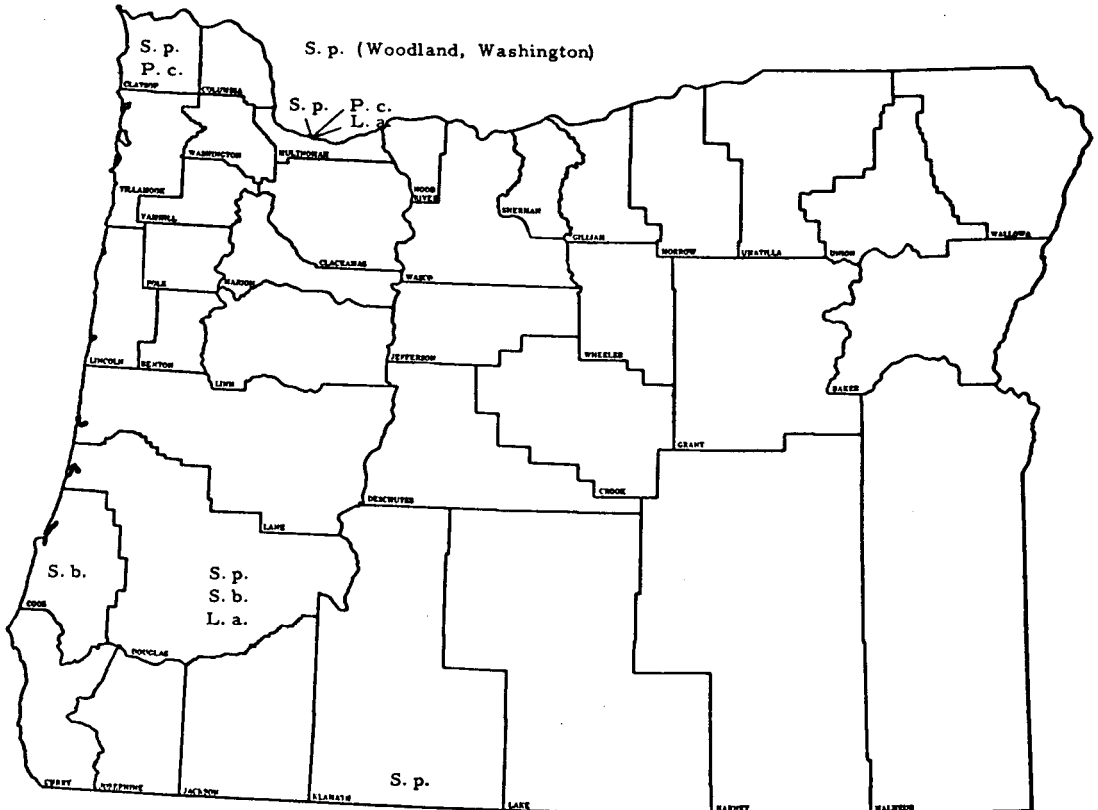


Figure 10. Distribution of Lymnaeidae snails in F. magna enzootic areas of Oregon.

where S. palustris was found.

PH readings were recorded during snail collections made in each enzootic area (Table III). There did not appear to be any correlation between pH values and snails species.

TABLE III. PH VALUES OF SNAIL HABITATS IN ENZOOTIC AREAS.

Area	Average pH	Snail Species
*Glide (Douglas Co.)	6.5	S. palustris S. bulimoides
Roseburg (Douglas Co.)	6.9	S. bulimoides
Langlois (Coos Co.)	7.1	S. bulimoides
Portland (Multnomah Co.)	6.9	S. palustris L. auricularia P. columella
Tenasillahae Island (Clatsop Co.)	6.9	S. palustris P. columella
Cascade Range (Lane Co.)	6.9	No snails found

*pH readings of 7.5 and 8.0 were obtained in this location previously.

Of the enzootic areas S. palustris was collected in Douglas, Multnomah, Klamath and Clatsop Counties in Oregon as well as in Cowlitz County, Washington. In each location this snail was associated with permanent, shallow, slow-moving water in grassy, mud-bottomed drainage ditches. Snails were most frequently seen crawling on mud bottoms but occasionally were observed on the grass at

the water's edge. According to Shaw (51) S. palustris is commonly found in ponds but during this study only small numbers were found in that habitat. This snail was seldom found in water over three or four feet deep and was most commonly observed in water two feet or less in depth.

Stagnicola bulimoides was found in the enzootic areas of Douglas and Coos Counties and was widely distributed throughout the Willamette Valley. It was most frequently found in grassy drainage ditches with partly mud bottoms and temporary, slow-running water. This species was commonly seen crawling on grass, mud banks or other exposed objects. During the summer months when drainage ditches dried, estivating snails were recovered from damp mud a few inches from the surface.

Lymnaea auricularia was found in many ponds in the Willamette Valley as well as the F. magna enzootic areas in Multnomah and Douglas Counties. This species was collected only in permanent ponds. Generally the snails were congregated in shallow water crawling on mud or clinging to vegetation. Occasionally a few snails were found in water three to four feet deep. This species was found at all times of the year.

Pseudosuccinea columella was found only in the Columbia River enzootic area in Oregon. Numerous snails of this species were collected from drainage ditches and a few large adults from permanent

ponds on Tenasillahae Island (Clatsop County). Collections were also made along the Columbia River in both Columbia and Multnomah Counties. With the exception of Tenasillahae Island, this species was found only in permanent stagnant ponds.

Physa spp. and Planorbidae snails were always found in the same habitat as described for the Lymnaeid snails. Succinea sp. was found only in one drainage ditch on Tenasillahae Island.

Experimental Exposures

All attempts to artificially infect the four species of Lymnaeid snails failed (Tables IV-VII). No attempt was made to expose Physa spp., Succinea sp. or Planorbis spp.

F. magna Ova Incubation and Hatching

F. magna ova taken from bovine liver cysts and from the uteri of flukes from the same source were incubated using the finger bowl technique. No eggs from the cysts hatched but ten to 15 percent of the ova removed from the uteri hatched after 38 days incubation. Since the finger bowl technique required constant care and because of poor success in hatching, the "egg bath" was devised.

The first experiment in which ova from cattle were incubated in the "egg bath" produced hatching on the 16th day. The ova were not observed until that time since hatching was not expected until the

TABLE IV. STAGNICOLA PALUSTRIS EXPOSURE EXPERIMENTS.

Aquaria	Miracidia		Temperature (° C)		Snails		Snail	Results
	No./Snail	Age	Exposure	Aquaria	Size	No.	Source	
5 gallon	2-3	8 hrs.	21	23.5	5-10mm	128	lab raised	died before 90 days
5 gallon	2-3	2-6 hrs.	27	27	3-5mm	116	lab raised	examined after 90 days
1 gallon	4-5	2-4 hrs.	27	27	3-6mm	50	Harrisburg	examined after 90 days
1 gallon	4-5	2-4 hrs.	29.5	29.5	3-6mm	50	Harrisburg	examined after 90 days
5 gallon	4/snail	1-2 hrs.	27	15.5-24	20-30mm	250	Woodland	died before 90 days
5 gallon	2-3	4-5 hrs.	24	24	4-9mm	300	North Portland	examined after 90 days
*30" diameter bowl, shallow, mud bottom	4	unknown	24	15.5-24	10-15mm	75	Tenasillahae Island	died before 90 days
1 gallon	5	unknown	15.5-24	15.5-24	20-30mm	51	Corvallis	died before 90 days

*Water used was from Tenasillahae Island.

TABLE V. LYMNAEA AURICULARIA EXPOSURE EXPERIMENTS.

Aquaria	Miracidia		Temperature (°C)		Snails		Snail	Results
	No./Snail	Age	Exposure	Aquaria	Size	No.	Source	
1 gallon	2-3	2-3 hrs.	24	24	6-7mm	70	North Portland	examined after 90 days
1 gallon	10/snail	1-2 hrs.	15.5-24	15.5-24	3-5mm	30	lab raised	examined after 90 days

TABLE VI. PSEUDOSUCCINEA COLUMELLA EXPOSURE EXPERIMENTS.

Aquaria	Miracidia No./Snail	Miracidia Age	Temperature (°C) Exposure	Temperature (°C) Aquaria	Snails Size	Snails No.	Snail Source	Results
1 gallon	4-5	2-4 hrs.	15.5-24	15.5-24	7-12mm	25	North Portland	died before 90 days
*30" diameter bowl, shallow, mud bottom	4	6 hrs.	24	15.5-24	4-8mm	100	Tenasillahae Island	died before 90 days

*Water used was from Tenasillahae Island.

TABLE VII. STAGNICOLA BULIMOIDES EXPOSURE EXPERIMENT.

Aquaria	Miracidia No./Snail	Miracidia Age	Temperature (°C) Exposure	Temperature (°C) Aquaria	Snails Size	Snails No.	Snail Source	Results
1 gallon	4-5	2-4 hrs.	15.5-24	15.5-24	4-7mm	100	Corvallis	died before 90 days

30th day (60, p. 187). Fluke eggs of deer or elk origin hatched in 12 days. Incubation temperature was held at 26±2° C. Incubation at 23° C using the same method was employed to observe effects on hatching. A large percentage of eggs were fully developed at this temperature and many had hatched by the beginning of the 16th day.

F. magna Ova Morphology

Twenty-eight F. magna ova washed from cavities of an infected elk liver from Oakridge, Oregon had an average length of 144.9 μ (range 137-155 μ) and a width of 90.0 μ (range 79-105 μ). Twenty-five ova in the feces of elk from Lane County averaged 151 μ in length and 94 μ in width. The length ranged from 136-166 μ and the width from 86-102 μ. Swales (60, p. 186) reported that F. magna ova in elk feces had an average length of 148 μ and a width of 94 μ.

Twenty-five ova from cattle liver cysts had an average length of 124 μ (range 107-171 μ) and a width of 91.4 μ (range 64-114 μ).

Fluke ova held in F. magna cysts of cattle were smaller, had thick shells and were black or dark brown in color (Figure 11). The normal ova from deer and elk were large with thin shells and were gold colored (Figure 12).

The small anti-opercular appendage described by Swales (60, p. 187) was present in 81 percent of 200 ova washed from liver cavities of deer from Tenasillahae Island. The appendage was lost if the ova were screened or washed vigorously.

DISCUSSION

Host Distribution

Bovidae

As stated by Griffiths (27, p. 344) F. magna appears to have little affect on the general health of cattle. Usually condemnation of the liver is the only loss involved. Meat inspectors look for the pathognomonic pigmentation (iron porphyrin) of the liver to identify F. magna infections. According to Campbell (4, p. 774) bile is possibly converted to iron porphyrin after ingestion by the fluke. The pigment accumulates in the lymph nodes and frequently appears in the diaphragm, lungs, omentum and dorsal peritoneum. In chronic infections the pigment spreads throughout the area of the body cavity. During this study the carcass of a heavily infected bull was condemned because of extensive pigmentation.

Tracing infected animals proved to be most time consuming. From this work came most of the information on the distribution of F. magna. The most important enzootic area discovered was bottom land near the Columbia River between Portland and Astoria, particularly Tenasillahae Island. Fascioloidiasis has undoubtedly increased in this area in the past 25 years. Damming of the Columbia River has increased the opportunity to use low lands for livestock

production. Heavier concentrations of livestock in areas occupied by infected wild ruminants will probably lead to an increase in the incidence of this disease.

The small number of bovine infection reports from other areas in Oregon indicates that the parasite is of minor importance as a disease problem. Klamath County may be an exception but there is not enough information to judge its importance. One report of four infected cattle in Wheeler County is the only incidence of the parasite in eastern Oregon. Since eastern Oregon is predominantly dry and has little area suitable for snail habitat, there is little chance of a wide-spread F. magna infection. Any infections occurring will probably be of minor and of local importance. In regions where water is available snail habitat may become improved by irrigation and in such cases this disease may become a problem.

Several infections, both confirmed and unconfirmed, have originated from Yakima County, Washington and one confirmed report was from the Olympic Peninsula in Washington. However, the importance of fascioloidiasis in Washington is not known.

From a statewide point of view F. magna as a disease problem in cattle of Oregon and southern Washington is of comparatively minor importance but is of major importance in certain areas of the Columbia River.

Cervidae

Swales (59, p. 207) stated that deer and elk having open cavities do not show pigmentation and that those having closed cysts do. He reasoned that the pigment produced in open cavities would be evacuated through the bile ducts. In closed cysts pigmentation would be unable to escape and would result in pigmentation of the lymph nodes and parenchymatous tissue. In contrast to Swales findings, Tenasillahae Island deer with light infections and having only open cavities showed pigmentation of the lymph nodes and liver capsules (Figure 9). This agrees with Campbell's (5, p. 772) statement that pigmentation may or may not occur in deer and elk livers containing only open cavities.

A comparison of a heavy and light infection occurring in Tenasillahae Island deer is illustrated in Figures 8 and 9. Although light or moderate infections generally are not pathogenic, heavy infections may cause acute fascioloidiasis, even in the normal host.

On the basis of the survey of "hunter killed" elk and deer in Millacoma Forest (Coos County), Tillamook County and MacDonald Forest (Benton County), the incidence of F. magna is either low or the parasite does not occur. No infected elk were found in Tillamook County; however, the survey did not include islands in or the areas adjacent to the Columbia River.

In contrast, the parasite is common in Rocky Mountain elk in the Cascade range of Lane County. Information on infected deer and elk was obtained by "hunter killed" animals and by interviewing hunters and ranchers. Interviews were generally non-productive but occasionally an observant hunter was able to describe the parasite and characteristic pigmentation. In such cases this information was accepted as a confirmed report. Hunters were difficult to contact because of the many roads entering and leaving the hunting areas. As a result only three kills were examined. Two were found to be infected.

Douglas, Klamath and Wheeler Counties and the Columbia River area were too difficult to survey and no attempt was made even though infected Cervidae are suspected as being responsible for bovine infections in those areas.

The Tenasillahae Island study and interviews with hunters of the Columbia River area indicated that the incidence of F. magna in deer in this region of Oregon and Washington was high. Consequently, fascioloidiasis could become a significant livestock disease problem in these areas. Although infected deer are known to exist in every location that infected bovine have been reported, the parasite is apparently of little significance in the rest of the state. Possible exceptions may be the Cascade range in Lane County and Klamath County where reports indicate that F. magna is common. No surveys

were made on other portions of the Cascade range.

In Rocky Mountain elk of Lane County the incidence of F. magna is high. The parasite appears to be uncommon or absent in Roosevelt elk and black-tailed deer of the Coastal Range in Oregon. Since no survey has been conducted in eastern Oregon the importance of this disease in that section of the state is not known. In Washington one report of an infected elk indicates that the parasite is present on the Olympic Peninsula but the incidence is not known.

Tenasillahae Island Study

Ninety-four percent of the island deer examined were found infected with F. magna. Deer were in close association with livestock and nearly every visit to the island provided an opportunity to observe this. A large number of infected deer in the presence of the proven intermediate hosts provided ample opportunity for the heavy infections found in cattle.

The estimated number of deer present on the island was difficult to obtain because of the elusive nature of the animals and dense cover in many sections of the island (Figure 3). The results of deer elimination as a control measure may be determined by population trend counts. Population trend counts can be fairly reliable in determining increase or decrease of numbers of animals. Set counting routes and methods could be used throughout a period of several

years and the figures compared to indicate the trend.

Considerable conflict between the owner of the island and the Oregon State Game Commission arose from the fact that deer on the island were responsible for the existing infections in the cattle. Reports of deer and elk transmitting diseases to livestock are not common but this situation was the basis of a serious problem between the Oregon State Game Commission and a livestock producer.

The disease problem of Tenasillahae Island is an example of what is happening or may happen in similar Columbia River areas. The existing trend of crowding animals on irrigated pastures and the use of river islands may compound the problem. If such is the case, both the Washington and Oregon State Game Commissions may become involved in the responsibility of protecting livestock from the disease transmitted by deer and elk.

A precedent may have been set on responsibility of the Oregon Game Commission. It has for many years provided help to farmers and ranchers who are bothered by deer and elk. In some instances special kill permits are issued to the complainant, special hunts organized to eliminate animals causing the problems and partial payment for deer-proof fences made. It appears that this precedent of responsibility by the game commission should also apply to fascioloidiasis transmitted to cattle. In April 1964 the owner of the island was issued a kill permit upon the recommendation of the

Department of Veterinary Medicine of Oregon State University. It was issued on an experimental basis to observe the effect of eliminating island deer as a control measure for F. magna.

Recommendations were also made to the owner of the island[6] to treat drainage ditches with copper sulfate in order to erradicate snails. Early in the study snail control was thought to be impractical; however, when snails became available they were found most frequently in ditches. Since snails were confined to drainage ditches this treatment was thought to be practical. Further study on the distribution of the snail hosts will be necessary to confirm the findings. Any decrease in the incidence of F. magna will be a result of both snail and deer erradication.

Theory of Cyst Reabsorption in Deer

Several descriptions have been given of the cyst stage of F. magna infections (35, p. 204; 55, p. 186; 60, p. 208, 209). Bile cysts associated with the cavity become clogged and ultimately bile stops flowing. When this happens the cyst begins to fill with fluid, pigment, eggs and detritus. Presumably after the cyst has become packed the fluke dies and is then reabsorbed by the liver. Bovine livers examined during this study have contained all stages of the

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[6]Robert Fraser, Tenasillahae Island, Clifton, Oregon.

fluke including old cysts which were as small as four or five millimeters in diameter. It is possible that the decrease in size would result from reabsorption by the liver tissue. The length of time required to reabsorb a cyst is not known but the process may continue throughout the life span of the animal.

In three of the deer collected on Tenasillahae Island, cysts were found in the livers and also in the tissue of the omentum. In one instance a cyst had apparently worked to the surface of the liver and was suspended by a thin piece of tissue. The observation of a cyst nearly separated from the liver and of others enveloped in the omentum indicates that cysts may be eliminated in this manner. Since one of the functions of the omentum is to surround and isolate (1, p. 101), it is possible that this process is a defense mechanism of the host. The oval-shaped cysts found in the omentum were considerably smaller than cysts found in the liver; however, the reabsorption process would account for the smaller size.

Two points need further clarification. First, the origin of the capsule tissue of the cyst found in the omentum must be determined. If the tissue was from the liver then the cyst must have been formed by the liver. The instance in which a cyst was attached by a thin section of tissue then would be an intermediary stage since the cyst could not have formed in this position. In this case the capsule was definitely of hepatic origin. If cysts found in the omentum originated

there this means the eggs were released due to a rupture in the liver and that the omentum enveloped them. The problem with this explanation is the eggs would tend to be dispersed in the omentum rather than concentrated in well-formed cysts.

Second, more deer and elk need to be examined in order to establish whether or not this phenomenon generally occurs. Only animals which have been infected several times over a number of years could be used in checking this point since recent infections would not have progressed to this stage.

Intermediate Host Investigations

Stagnicola palustris and P. columella are proven intermediate hosts of F. magna (26; 38, p. 108). These two snails were the only Lymnaeidae found on Tenasillahae Island and it is assumed that they are the intermediate hosts for F. magna. Physidae, Planorbidae and Succineidae have not been incriminated as snail hosts for Fasciolidae and for that reason the Physa spp., Planorbis spp. and Succinea sp. found during the snail surveys were not experimentally exposed to the miracidia of F. magna.

This study has shown that S. palustris, L. auricularia and S. bulimoides inhabit the Douglas County enzootic area (Figure 10).

Lymnaea auricularia and S. bulimoides have not been proven to be intermediate hosts and it is not possible to say which of the three species are serving as intermediaries in that area. Since S. palustris is a proven host and was found in nearly every location surveyed it is possible to involve this species in each instance.

Pseudosuccinea columella was not found on the Washington side of the Columbia River but probably is present. Only one Lymnaeidae snail, S. palustris, was found in that area.

The snail host S. palustris was found in Klamath County, Oregon at an earlier date (51). Snails were not surveyed in the enzootic areas of Wahkiakum and Yakima Counties, Washington. Further investigations will be needed to determine which Lymnaeid snails are in the Cascade enzootic area.

The experiments listed in Tables 4-7 are a complete record of exposures made on the four species of Lymnaeidae snails (S. palustris, S. bulimoides, L. auricularia and P. columella).

Under laboratory conditions snails usually crawled out of the water and died from dehydration. Several methods were used in an attempt to solve the problem. A line of petroleum jelly was laid above the water level but this method was not satisfactory. Glass covers and screens were used but even greater numbers of deaths occurred. The crawling habits of S. bulimoides made it necessary to have wet surfaces. Cheese cloth absorbed water from the aquaria

and although it did not dry out it rotted in a short time. Even though the crawling problem was solved and mortality decreased many deaths still occurred. This snail survived for only one or two weeks in the laboratory.

Stagnicola palustris and L. auricularia were successfully raised in filtered aquaria although the latter adjusted better to laboratory conditions. Pseudosuccinea columella did not survive in filtered aquaria but did very well in large bowls containing mud and water from the natural habitat.

Fresh lettuce was eaten readily by all species of snails and good growth was obtained with most snails that survived.

Miracidia were more attracted to S. palustris and P. columella than S. bulimoides and L. auricularia. When all four species of snails were exposed in the same watch glass, the most miracidia could be seen swimming around P. columella and the least near L. auricularia. Miracidia were rarely attracted to S. bulimoides.

The principle reason for lack of success in recovering cercaria was death of the snails under laboratory conditions. No reason could be found for lack of infection in surviving snails.

F. magna Ova Incubation

Swales (60, p. 185) stated that he was able to hatch 0. 1 percent of the ova taken from cysts of a bovine liver. Sinitsin (55, p. 187)

hatched ova from bovine cysts and apparently incubated the eggs for 33 days at room temperature. During this current study ova from this source did not hatch except when they were taken from the uteri of live flukes. F. magna ova from bovine cysts were thick-walled, generally smaller and contained disintegrated remnants of the internal structures (Figure 11). The morphological differences of ova from Bovidae and Cervidae are illustrated in Figures 11 and 12.

All eggs which were incubated in aerated finger bowls in this study appeared nonviable. Eggs taken from the uteri of live flukes from bovine cysts were incubated at room temperature in aerated finger bowls and ten to 15 percent of the ova hatched on the 38th day.

Swales (60, p. 187, 189) established the ova incubation period at 29 to 35 days. Earliest hatching occurred on the 29th day but generally occurred on the 35th. Campbell in 1961 (6, p. 312) successfully hatched the ova in 17 days of incubation at 25° C and established a new incubation period. His method involved spreading the ova in a thin layer over the bottom of a petri dish half filled with water. Aeration was accomplished by diffusion through the shallow layer of water which was changed every two days.

Ova held in the "egg bath" used in this study hatched in 12 days. The early hatching time was credited to controlled constant temperature, filteration, aeration and circulation of the water contained in the egg bath. No special care was taken to spread the ova in a thin

Figure 11. Two ova taken from a F. magna cyst of a bovine. Note the thick walls.

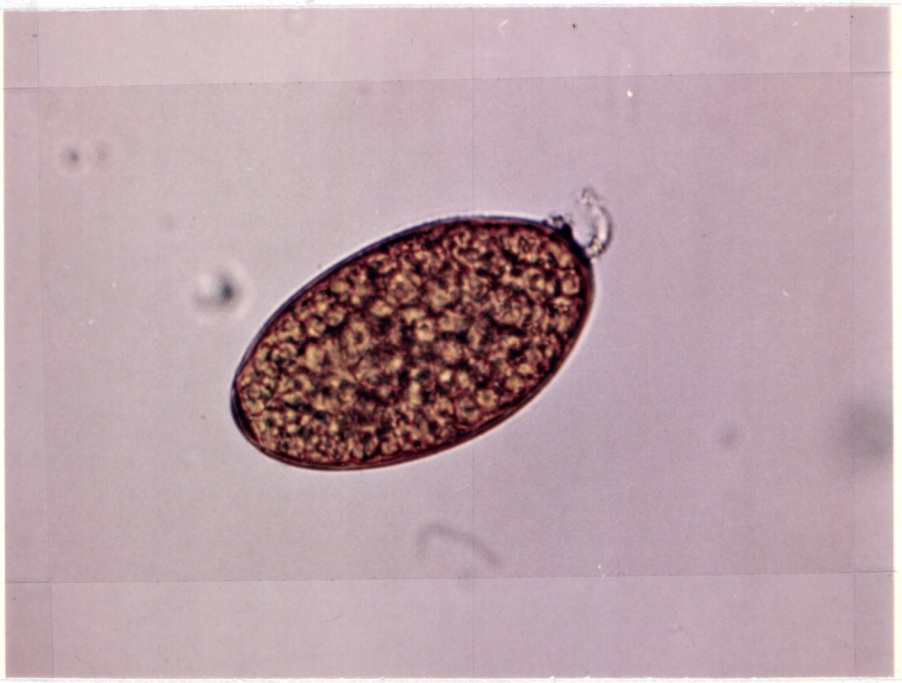

Figure 12. This viable ovum was photographed directly after removal from a bile duct of an infected deer. Note the thin walls and abopercular appendage.

layer since oxygenation was no problem.

Figure 12 is a photograph of an egg before incubation. The abopercular appendage described by Swales (60, p. 187) was seen in 20 percent of the ova found in elk and deer feces.

Figure 13 shows the anterior granular mass of the miracidia on the 10th day of incubation. Campbell (6, p. 310) illustrated the same stage miracidia on the 15th day of incubation. The anterior granular mass disappears after two days longer incubation. At this time hatching may be stimulated.

Ova were observed in various stages of development throughout their incubation. The egg shown on the right of Figure 13 is in the morula stage even though both eggs have been incubated for the same period. This trait is commonly seen regardless of the method employed in incubation (6, p. 308).

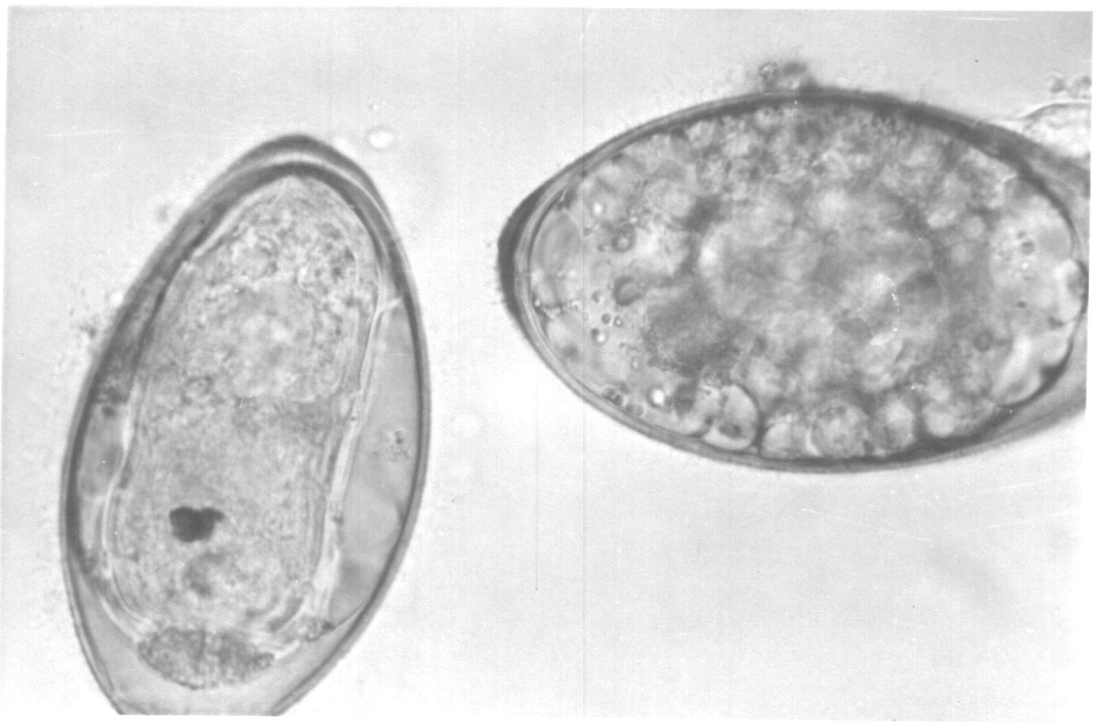

Figure 13. F. magna ova after 10 days incubation in the "egg bath". The egg on the left contains an active miracidia. The black eye spots are well developed. The miracidium should loose the anterior granular mass and hatch in two or three days. The egg on the right has not developed as rapidly. No explanation can be given for this phenomenon.

SUMMARY

Many important contributions to the biology of Fascioloides magna were summarized in the review of literature. The history and synonyms of F. magna were given as well as the known distribution throughout the world. Fascioloides magna was discovered in Italy but apparently was introduced into Europe through infected elk from America. This parasite has been reported from nearly every area of the United States and southern Canada (Figure 2). In Europe it was reported from Italy, Germany, Czechoslovakia and Poland.

Studies have shown that the natural hosts of F. magna were Cervidae and the common accidental hosts were cattle, sheep and goats. The parasite has been found to be most pathogenic to sheep and goats but also caused severe tissue reactions in cattle. Past work has shown that host-parasite incompatability prevents the parasite from reproducing in the accidental host.

Proven snail intermediate hosts were listed. Control measures from past studies were given.

During this study 107 Oregon and 19 Washington cattle were traced from abattoirs to the source of infection. The main source of cattle infections for Oregon and southern Washington was the Columbia River area. Other enzootic areas were Wheeler, Klamath, Douglas and Coos Counties in Oregon and Yakima County in Washington (Figure 5).

Deer and elk hunter kills were examined in areas where bovine infections had been reported and in three hunting areas in Coos, Tillamook and Benton Counties. Results showed that F. magna is either rare or does not occur in deer and elk in the Coastal Mountain Range or in Benton County of the Willamette Valley in Oregon. There was a high incidence of infected elk in the Cascade Range of Lane County. Reports indicated the incidence was also high in deer of the Columbia River area from Portland to Astoria.

On the major study area, Tenasillahae Island in the Columbia River, (Figure 3) 94 percent of the deer and 77 percent of the cattle were infected. Two proven snail hosts, S. palustris and P. columella were the only Lymnaeidae snails found on the island and it was concluded that they were intermediate hosts for F. magna. Treating snails with copper sulfate and eliminating the deer from the island were recommendations made for experimental control measures.

Stagnicola palustris, P. columella, L. auricularia and S. bulimoides were the snails found in enzootic areas in Oregon. Only S. palustris was found in Washington although P. columella is probably also present. No naturally infected snails were found nor were any laboratory infections successful (Tables IV-VII).

Death of snails under laboratory conditions was considered to be the reason for lack of success in infecting and recovering F. magna cercariae. Of the four Lymnaeidae snails S. bulimoides was

the most difficult to raise under laboratory conditions.

Flukes were found in the burrowing and cyst stages in cattle and in the cavity and cyst stages in deer and elk. Pigmentation of the liver and lymph nodes, omentum and diaphragm was found in each of the infected deer and elk even when no closed cysts were present. A massive infection in one Tenasillahae Island deer resulted in adhesions, hyperplasia, necrosis of the liver and extensive pigmentation.

The ova from cattle were darker colored, thicker shelled and shorter than those from deer and elk (Figures 11 and 12).

Ova were incubated in an "egg bath" which allowed warmed, aerated, filtered water to flow over the layer of eggs throughout their incubation (Figure 4). The shortest hatching time obtained was 12 days.

A theory of the reabsorption of F. magna cysts by the omentum of deer as a means of eliminating the cyst from the liver was presented.

It was concluded that F. magna was of minor importance as a state-wide problem in Oregon and southern Washington. The Columbia River area was considered a major local problem and would become increasingly important as the use of islands and diked areas are more extensively used for raising livestock.

BIBLIOGRAPHY

1. Bone, Jesse F. Animal anatomy. Corvallis, O. S. C. Cooperative Association, 1958. 191 p.

2. Cameron, A. E. Notes on buffalo: anatomy, pathological conditions, and parasites. The Veterinary Journal 79:331-336. 1923.

3. Cameron, A. E. Some further notes on buffalo. The Veterinary Journal 80:413-417. 1924.

4. Campbell, William Cecil. Fascioloides magna (Trematoda) with special reference to the adult in relation to disease. Ph. D. thesis. Madison, University of Wisconsin, 1957. 119 numb. leaves. (Microfilm)

5. Campbell, William Cecil. Nature and possible significance of the pigment in Fascioloidiasis. Journal of Parasitology 46:769-775. 1960.

6. Campbell, William Cecil. Notes on the egg and miracidium of Fascioloides magna (Trematoda). Transactions of the American Microscopical Society 80:308-319. 1961.

7. Campbell, William Cecil and A. C. Todd. Natural infections of Fascioloides magna in Wisconsin sheep. The Journal of Parasitology 40:100. 1954.

8. Campbell, William Cecil and A. C. Todd. In vitro metamorphosis of the miracidium of Fascioloides magna (Bassi, 1875) Ward, 1917. Transactions of the American Microscopical Society 74:225-228. 1955.

9. Campbell, William Cecil and A. C. Todd. Behavior of the miracidium of Fascioloides magna (Bassi, 1875) Ward, 1917, in the presence of a snail host. Transactions of the American Microscopical Society 74:342-347. 1955.

10. Cheatum, E. L. Disease in relation to winter mortality of deer in New York. Journal of Wildlife Management 15:216-220. 1951.

11. Cowan, Ian McTaggart. Report upon some diseases and parasites of game birds and game and fur-bearing mammals in British Columbia. Report of Provincial Game Commission, 1941, p. K40-K45.

12. Cowan, Ian McTaggart. Parasites, diseases, injuries and anomalies of the Columbian black-tailed deer, Odocoileus hemionus columbianus (Richardson), in British Columbia. Canadian Journal of Research 24:71-103. 1946.

13. Dikmans, G. Check list of the internal and external animal parasites of domestic animals in North America. American Journal of Veterinary Research 6:211-241. 1945.

14. Dinaburg, A. G. Helminth parasites collected from deer, Odocoileus virginianus, in Florida. Proceedings of the Helminthological Society of Washington 6:102-104. 1939.

15. Erhardová, Božena. Vývojový cyklus motolice obrovské fasciola magna v podmínkách CSSR. (Developmental cycle of the trematode Fasciola magna in conditions of the Czechoslovakia.) (English summary p. 16) Zoologicke Listy. 24(1):9-16. 1961. (Abstracted in Biological Abstracts 36:46602. 1961)

16. Erhardová, Božena and A. Kotrly. Cizopasní cervi zažívaciho ústrojí našich volne žijících přežvýkavců. (The helminth parasite of the digestive tract of the Czechoslovakian wild ruminants.) Ceskoslovanská Parasitologie 2:41-68. 1955. (Abstracted in Biological Abstracts 30:20839. 1956)

17. Erickson, Arnold B. and P. R. Highby. Parasites of the woodland caribou. The Journal of Parasitology 28:423. 1942.

18. Fenstermacher, Reuel. Further studies of diseases affecting moose. Minneapolis, 1934. 26 p. (Minnesota. Agricultural Experiment Station. Bulletin 308)

19. Fenstermacher, Reuel and O. Wilford Olsen. Further studies of diseases affecting moose. III. The Cornell Veterinarian 32:241-254. 1942.

20. Friedl, Frank E. Induced hatching of operculate eggs. The Journal of Parasitology 46:454. 1960.

21. Friedl, Frank E. Studies on larval Fascioloides magna. I. Observations on the survival of rediae in vitro. The Journal of Parasitology 47:71-75. 1961.

22. Friedl, Frank E. Studies on larval Fascioloides magna. II. In vitro survival of axenic rediae in amino acids and sugars. The Journal of Parasitology 47:244-247. 1961.

23. Friedl, Frank E. Studies on larval Fascioloides magna. III. Mass hatching of miracidia by exposure to nitrogen. The Journal of Parasitology 47:770-772. 1961.

24. Friedl, Frank E. Studies on larval Fascioloides magna. IV. Chromatographic analyses of free amino acids in the hemolymph of a host snail. The Journal of Parasitology 47:773-776. 1961.

25. Friedl, Frank E. A method for securing the snail, Lymnaea stagnalis jugularis (Say), free from bacteria. Experimental Parasitology 15:7-13. 1964.

26. Griffiths, Henry J. Stagnicola palustris (Müller), an intermediate host for Fascioloides magna (Bassi, 1875), in Minnesota. The Journal of Parasitology 41:115. 1955.

27. Griffiths, Henry J. Stagnicola (Hinckleyia) caperata (Say), a natural intermediate host for Fascioloides magna (Bassi, 1875), in Minnesota. The Journal of Parasitology 45:146. 1959.

28. Griffiths, Henry J. Fascioloidiasis of cattle, sheep, and deer in northern Minnesota. Journal of the American Veterinary Medical Association 140:342-347. 1962.

29. Hadwen, Seymour. A new host for Fasciola magna, Bassi. Together with observations on the distribution of Fasciola hepatica., L. in Canada. Journal of the American Veterinary Medical Association 49:511-515. 1916.

30. Hall, Maurice Crowther. Our present knowledge of the distribution and importance of some parasitic diseases of sheep and cattle in the United States. 27th Report, Bureau of Animal Industry, United States Department of Agriculture 3:428-436. 1912.

31. Hall, Maurice Crowther. Society Proceedings, The Helminthological Society of Washington. The Journal of Parasitology 1:106. 1914.

32. Hall, Maurice Crowther. Parasites of elk and other wild ruminants. Proceedings: Biological Society, 741st meeting. Journal of the Washington Academy of Science 20:87-88. 1930.

33. Kermode, F. Investigation of reported disease in the blacktail or Columbian coast deer (Odocoileus columbianus). Annual Report British Columbia Provincial Museum, 1915. 12:12-13. 1916.

34. Kingscote, A. A. Report of the Ontario Veterinary College, Department of Parasitology, 1949, p. 27-36.

35. Kingscote, A. A. Liver rot (Fascioloidiasis) in ruminants. Canadian Journal of Comparative Medicine (14:203-208). 1950.

36. Knapp, Stuart E. and James N. Shaw. Occurrence of Fascioloides magna (Bassi) in Oregon cattle and deer. The Journal of Parasitology 49:339. 1963.

37. Krull, Wendell H. A new intermediate host for Fascioloides magna (Bassi, 1873) Ward, 1917. Science 78:508-509. 1933.

38. Krull, Wendell H. New snail hosts for Fasciola magna (Bassi, 1875) Stiles 1894. Proceedings of the Helminthological Society of Washington, 1933. The Journal of Parasitology 20:107-108. 1933.

39. Krull, Wendell H. Experiments with Fascioloides magna involving the final host. The North American Veterinarian 16: 26-28. 1935.

40. McCraw, Bruce M. The ecology of the snail Lymnaea humilis Say. Transactions of the American Microscopical Society 78: 101-121. 1959.

41. McCraw, Bruce M. Life history and growth of the snail Lymnaea humilis Say. Transactions of the American Microscopical Society 80:16-27. 1961.

42. Olsen, O. Wilford. Bionomics of the Lymnaeid snail, Stagnicola bulimoides techella, the intermediate host of the liver fluke in southern Texas. Journal of Agricultural Research 69: 389-403. 1944.

43. Olsen, O. Wilford. White-tailed deer as a reservoir host of the large American liver fluke. Veterinary Medicine:44:26-30. 1949.

44. Olsen, O. Wilford and Reuel Fenstermacher. Parasites of moose in northern Minnesota. American Journal of Veterinary Research 3:403-408. 1942.

45. Olsen, O. Wilford and Reuel Fenstermacher. The helminths of North American deer. Minneapolis, 1943. 20 p. (University of Minnesota. Agricultural Experiment Station. Technical Bulletin 159)

46. Price, Emmett W. The fluke situation in American ruminants. The Journal of Parasitology 39:119-134. 1953.

47. Price, Emmett W. Liver flukes of cattle and sheep. U. S. Dept. of Agriculture Yearbook of Agriculture 1956, p. 148-153.

48. Schwart, B. Fascioloides magna in cattle. Report of the Chief, Bureau of Animal Industry, Annual Reports of Department of Agriculture, 1938, p. 78.

49. Senger, Clyde M. Some parasites of Montana deer. Montana Wildlife, Montana Fish and Game Commission, Autumn 1963, p. 5-13.

50. Shaw, James N. Some parasites of Oregon wild life. Corvallis, 1947. 16 p. (Oregon. Agricultural Experiment Station. Station Technical Bulletin 11)

51. Shaw, James N., Professor Emeritus. Oregon State University. Personal communication. July 1964.

52. Shumard, R. F. and D. F. Eveleth. Liver flukes of cattle and sheep. North Dakota. Agricultural Experiment Station. Bimonthly Bulletin 17(2):59-65. 1954.

53. Sinclair, Luke R. Observations on liver flukes in cattle. Veterinary Medicine 44:68-69. 1949.

54. Sinitsin, D. F. A note on the life history of the large American fluke, Fasciola magna (Bassi). Science n. s. 73:273-274. 1930.

55. Sinitsin, D. F. Studien uber die Phylogenie der Trematoden. VI. The life histories of some American liver flukes. Zeitschrift fur Parasitenkunde 6:170-191. 1933.

56. Slusarski, Wieslaw. Studia nad europejskimi przedstawicielami przywry Fasciola magna (Bassi, 1875) Stiles, 1894. I. Ponowne wykrycie ogniska inwazji u jeleni na Slasku. Acta Parasitologica Polonica 3(1/5):1-59. 1955 (English Summary, p. 52-59)

57. Slusarski, Wieslaw. Dalsze studia nad europejskimi przedstawicielami Fasciola magna (Bassi, 1875) Stiles, 1894. II. Rozmieszczenie i biologia pasozyta w Polsce. Wiadomosci Parazytologiczne Warsaw 2(5):suppl. 165-166. 1956. (English summary, p. 166)

58. Stiles, Charles Wardell, The anatomy of the large American fluke (Fasciola magna), and a comparison with other species of the genus Fasciola, S. St. The Journal of Comparative Medicine and Veterinary Archives 15:161-178, 225-243, 299-313, 407-417, 457-462. 1894. 16:139-147, 213-222, 277-282. 1895.

59. Swales, W. E. A review of Canadian helminthology. Canadian Journal of Research 8:468-482. 1933.

60. Swales, W. E. The life cycle of Fascioloides magna (Bassi, 1874), the large liver fluke of ruminants, in Canada. Canadian Journal of Research 12:177-215. 1935.

61. Swales, W. E. Further studies on Fascioloides magna (Bassi, 1875) Ward, 1917, as a parasite of ruminants. Canadian Journal of Research 14:83-95. 1936.

62. Ward, H. B. On the structure and classification of North American parasitic worms. The Journal of Parasitology 4: 1-12. 1917.

63. Whitlock, S. C. The prevalence of disease and parasites in whitetail deer. Transactions of the Fourth North American Wildlife Conference 4:244-249. 1939.

64. Wu, Liang-yu and A. A. Kingscote. A note on Lymnea stagnalis (L.) as a snail host for Fascioloides magna (Bassi, 1875) (Trematoda). The Journal of Parasitology 39:568. 1953.

65. Wu, Liang-yu and A. A. Kingscote. Further study on Lymnaea stagnalis (L.) as a snail host for Fascioloides magna (Bassi, 1875) (Trematoda). The Journal of Parasitology 40:90-93. 1954.