### **Parasitic Bronchitis of Livestock in Japan**

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From the viewpoint of livestock population and economic damage, it can be said that the major lungworm diseases of livestock in Japan are substantially swine metastrongylosis and bovine dictyocaulosis.

In Japan, lungworms of several species, which will be described below, cause parasitic bronchitis of animals, except swine and cattle.

The combined infection of *Metastrongylus* pudendotectus (Wostokov, 1905) and *M. apri* (Gmelin, 1790) is very common among wild boars.

Though several reports have been made on the outbreak of dictyocaulosis caused by *Dictyocaulus filaria* (Rudolphi, 1809) among sheep, the sheep involved were exclusively those imported from the United States or Australia into Japan.

The parasitic infestation with the small lungworm, *Muellerius capillaris* (Müller, 1889), has rarely been recognized among sheep and goats.

Parasitic bronchitis in *Capricornis crispus* (Temminck), which is a natural monument, has recently been investigated owing to its increased death rate among animals of this species in which *Protostrongylus* sp., which had not been recorded in Japan, was detected from all the lungs examined.

In general, there is a direct or indirect relationship between feeding and management of livestock and the occurrence of lungworm diseases.

The atomspheric temperature in Japan, where such temperature is quite variable by seasons, has no great influence on the occurrence of swine lungworm disease. In this country, however, the frequency of occurrence of this disease is rather high among pigs fed garbage in the dirty piggery on small farms. This might be due to the unclean environment produced by the utilization of garbage because such environment is favorable for an increase in earthworms acting as intermediate hosts for swine lungworms.

On the other hand, the occurrence of dictyocaulosis in calves is closely related to climatic conditions, especially temperature, precipitation and humidity, and to the amount of grass which is much influenced by these conditions. Besides, the population density of cattle has a great influence upon the severity and spread of this disease in a herd.

### Swine lungworm disease

This is one of the most important diseases of the helminthic infestations of piglets in Japan. Two species, *M. apri* and *M. pudendotectus*, have been known to be causal parasites of this disease, but *M. salmi* (Gedoelst, 1923) has not been recorded. *M. apri* is distributed all over Japan but *M. pudendotectus* is a less important species which is distributed locally. These two species induce a combined infection. Neither of them causes a single infection independently from each other.

### Incidence

Statistics from slaughterhouses show that pigs were infected with this parasite at the rate of 60 to 70 per cent, and that the economic damage caused by this disease was very serious up to about seven or eight years ago. Recently, this disease has been declining gradually in severity owing to the technical improvement in pig enterprises, especially efficient management and the application of the very effective anthelmintic, tetramisole. The rate of infection of this disease is estimated to be about 20 to 30 percent at present.

### Life cycle

One species of earthworm, Eisenia foetida, is only one known intermediate host of *Metastrongylus* spp. in Japan. It is a common inhabitant in the porcine manure piled up around the piggery. When engorged by earthworms of this species, Metastrongylus eggs hatch in the body of the earthworms. After the 1st and 2nd larval stages, 3rd-stage larvae (infective larvae) appear on the eighth to tenth day after infection and are then harbored by the earthworms.

When a pig is experimentally fed earthworms harboring infective-stage larvae, it is easily infected with lungworm disease. The author has proved, however, that no pig is, at least positively, fond of capturing earthworms, though there may be a few chances for it to capture them.

As a result of their experimental study, Ueno et al. (1965)<sup>14)</sup> revealed an interesting biological phenomenon that infective larvae harbored in the alimentary tract of earthworms finally came out in the lumen of the tract, from piercing through the wall, and were mixed with feces to be discharged from the body little by little. Encysted larvae show a strong resistance to environmental sensitization. They may be transmitted to pigs directly or by some mechanical intermediary.



Fig. 1. Mode of Metastrongylus infection in pigs

### Pathogenicity

The pathogenicity of the genus Metastrongy-

lus should be considered not only with the direct pathogenicity of this parasite itself but also with the symptoms of the combined infection with this parasite and virus pneumonia (VP) or swine epizootic pneumonia (SEP). and with the importance of transmission of swine influenza virus (SIV).

At present, nothing is known about wormtoxin secreted by this parasite.

The pathogenicity of this parasite varies with the number of parasites present. It is presented mainly as the functional insufficiency of the pulmonary lobule and emphysema. The former is caused by the mechanical blockage in small and medium bronchi which control the function of lobules distributed in the marginal parts of the lung. Piglets harboring numerous parasites of this species are affected with dyspnea and show retarded growth.

MacKenzie (1963)<sup>8)</sup> reported that the combined infection of VP and Metastrongylus caused severe and more extensive pneumonitis than the single infection of either of the two agents. Shope (1941)<sup>12)</sup> and Kammer & Hanson (1962)<sup>7)</sup> suggested the possibility of this parasite to be the carrier of SIV masking this virus.

It is very interesting to study *Toxoplasma* gondii in future since this pathogen may be transmitted by Metastrongylus parasites themselves and their eggs and 3rd-stage larvae.

The author has some experience in this problem. When piglets were infected with 3rdstage larvae of Metastrongylus released from pepsinated earthworms collected from a piggery contaminated with Metastrongylus and Toxoplasma, most of them showed a typical symptom of toxoplasmosis unexpectedly. The sera of these infected piglets showed an extraordinarily high antibody titer when examined by the dye test.

### **Clinical symptoms and diagnosis**

When infected, adult pigs manifested no specific clinical symptoms, except coughing, but piglets show frequent coughing, increment in respiration, and dyspnea after subjected to a sudden forced movement. They gradually become thin, suffering from anemia.

The exact diagnosis of this disease can be made when eggs of Metastrongylus are detected from feces. Generally speaking, an animal harbors much more parasites of this species than it is expected to do from the number of eggs detected from it. It may be considered that when a fecal sample contains 100 epg (eggs per gram), it is estimated to keep 60 to 100 parasites.

Though Metastrongylus eggs can be floated on a solution saturated with sodium chloride or sucrose, Bello (1961)" found a saturated MgSO<sub>4</sub> solution to be most suitable for this purpose.

An ecological diagnosis is made to know the extent of infestation with swine lungworms on a given hog farm. It is made by detecting developing-stage larvae of lungworms from the body of intermediate hosts collected around the piggery. In this method, such part of the earthworm body as called "the heart" is exposed anatomically in the 10th to 12th segment of the earthworm. It is solely disposed between two pieces of slide glass to be pressed and examined for the existence of larvae under the microscope at a low-power magnification.

#### Immunity

It has been proved by the hemoagglutination test and CF test that antibody exists in the sera of pigs infected with swine lungworms. Some pigs were exposed to reinfection after they had been freed from infection with swine lungworms by some anthelmintic treatment and allowed to stand for about three weeks. Others were inoculated subcutaneously or intramuscularly with the physiological saline emulsion of adult lungworms. No immunity, however, was produced in any pig of either group to prevent it from reinfection with this parasite.

On the other hand, Onishi (1968<sup>9)</sup>, 1973<sup>10)</sup>) reported that when inoculated perorally or parenterally with 3rd-stage larvae of Metastrongylus partially inactivated by irradiation with 50,000 r, guinea pigs were conferred a strong immunity against reinfection with the parasite.

### Treatment and prophylaxis

Tetramisole was developed by Thienpont et al. (1966)<sup>13)</sup> as a wide-spectrum anthelmintic against nematode parasites. It was reported to be a very effective agent against swine lungworms. After that, its efficacy was confirmed by Ueno et al. (1967)<sup>15)</sup> and other workers.

When a dose of 7.5 to 10 mg/kg of this compound is administered to infected piglets, it is so effective as to remove nearly completely lungworms of all the stages covering from seven-day-old larvae to adult worms.

It is the most effective mode of administration of this agent to mix the agent with feed. There is no significant difference in efficiency between subcutaneous and intramuscular injection.

In a few minutes after the administration of this compound, the pig being to suffer from transitory coughing and its respiration increases in rate for a while. There is a trend that the larger the number of swine lungworms harbored by a pig, the more frequently these symptoms appear in the pig.

It is rather difficult to control swine lungworm infection. Establishment and practice of a control program of swine lungworm infection are dependent on the rearing environment of swine.

The economic loss caused by this infection will be blocked at a minimum level if sufficient attention is paid to the following points: (1) An effective anthelmintic must be applied periodically not only to piglets but also to full-grown pigs in a piggery contaminated with the parasite. (2) No manure heap is made around the piggery. (3) Ditches around the piggery must be drained well and kept always clean to prevent feces from being scattered.

# Bovine parasitic bronchitis (Bovine dictyocaulosis)

This disease is caused by the infection of *Dictyocaulus viviparus*. It is commonly called "husk" or "hoose" and is a serious parasitic disease which is usually found among calves in many European countries.

It is well known that when infected once with this parasite spontaneously or experimentally in the field, cattle can acquire a strong resistance to the reinfection. On the basis of this fact, a unique vaccine has been developed in the domain of parasitic disease.

### Incidence

Although bovine parasitic bronchitis has occurred in several pastures in Japan, the substantial damage of cattle is rather small because major yearling calves are kept in cowsheds.

Even if yearling calves are placed in the pasture, the pasturing period is limited to a few months. Recently, however, the conventional manner of rearing has been transformed into pasturing. The establishment of a largescale pasture and the expansion of the existing pasture have been encouraged.

Consequently, several pastures have already been contaminated by the etiological parasite spread by yearling calves infected subclinically with the parasite (carriers) and introduced from a pasture contaminated with the parasite into a newly established pasture.

It is possible to recognize the pattern of outbreak of this disease in the field; that is, clinical symptoms usually appear over a period from late summer to early autumn and usually disappear within two or three months.

In numerous pastures where no effective countermeasures have been taken immediately after the occurrence of this diseases, about 30 to 47 per cent of calves have died suffering from husk, and the growth of surviving calves has been retarded distinctly to cause great economic damage.

### Life cycle

D. viviparus is parasitic to the trachea and bronchi of cattle and deposits embryonated eggs in these organs. These eggs come up by coughing and are swallowed. They pass through the alimentary tract of cattle and hatch out to be first-stage larvae, which are discharged with the excretion of feces on the pasture ground. These larvae grow into infective (3rd-stage) larvae in four or five days, and infect cattle when ingested with grass.

Infective larvae are so resistant to environmental conditions, especially cold weather, that they can live over winter even in the northern part of the Japanese mainland and in Hokkaido. First-stage larvae, however, are so sensitive to environmental conditions that only ten percent of them can reach the 3rd stage, or become infective larvae.

The migratory course of this parasite inside the body of the host animal was reported in detail by Jarrett et al. (1957)<sup>30</sup>, Poynter et al. (1960)<sup>10</sup>, and Douvres & Lucker (1958)<sup>20</sup> in cattle or guinea pigs.

When ingested by cattle with grass, infective larvae take off their sheath, pass through the wall of the small intestine and the mesenteric lymph node to the lymph vessel, and attain the lung at the end of 24 hours after infection.

The prepatent period of this parasite is about 25 to 27 days. The largest number of 1st-stage larvae appears about 35 days after the infection. Then the larvae in feces begin to decrease gradually in number, reaching a minimum around the 45th to 50th day after the infection.

Most of the parasites in the lung disappear within two months, when no reinfection occurs successively.

### Pathogenicity

Since *D. viviparus* is more strongly pathogenic than any other helminth, it causes an infection in a herd of cattle which may result in an epizootic, bring about a high fatality, and affect weight gain seriously.

In a newly infected calf, the tissue reaction of the lung to this parasite begins to appear about ten days after the infection. As one of the specific pathological changes of this reaction in its initial stage, alveloar collapse mostly occurs in the diaphragmatic lobe. Infiltration with eosinophils, macrophages and giant cells can be recognized as a histopathological change.

With the progress of the disease, a distinct contraction is seen between a normal and a consolidated pulmonary lobule. Such contracted area spreads to the anterior part of the lung. Dull plum-colored irregular areas of consolidation appear in the apical lobe most conspicuously and in the intermediate lobe to a lesser extent. A violent cellular response also occurs in them.

In the second-half stage of disease, or classical husk pneumonia, the features of edema and emphysema can be observed microscopically. These pathological lesions exist for a fairly long time, even after the removal of this parasite.

Although the cause of fog fever is yet unknown, the fever may be an allergic reaction induced under a singular immunological condition between this parasite and cattle.

There has been a cattle disease of unknown etiology called "kiri yoi" (fog inebriation) in the mountainous Chubu region of Japan contaminated with the infestation of D. viviparus. The symptoms of this disease agree well with those of fog fever clinically and immunologically.

### **Clinical symptoms and diagnosis**

A calf suffering from this disease breathes forcedly through the mouth, with the head extended. It is obliged to continue respiration with an extremely hard effort. Therefore, it cannot afford to any other activity, such as feeding. Moreover, it is almost impossible for the animal to eat, respiratory count often exceeding the pulse rate. Since these symptoms are manifested in the case of this disease, it is not always difficult to make a clinical diagnosis of the disease. The most correct diagnosis may be made when 1st-stage larvae are detected from feces. It is widely utilized for this purpose to isolate 1st-stage larvae by means of the Bermann's apparatus.

Ueno (1968 a & b)<sup>163,17)</sup> established the following technique to make the detection of 1st-stage larvae easy by using no special equipment in the field.

(1) A 3 g fecal sample is put on a piece of gauze  $(10 \times 10 \text{ cm})$ .

(2) The four corners of this piece of gauze are gathered to one point which is drawn up.

(3) With the gathered corners held upward, the sample wrapped with the piece of gauze is put into a pointed centrifuge tube about 50 ml in capacity.

(4) The gathered corners are fixed at the opening edge of the tube, into which faucet water is poured. All the tubes are allowed to stand overnight or for 24 hours to make larvae migrate into the water and settle at the bottom of the tube.

(5) The sediment is collected by the aid of a pipette, the tip of which has a relatively large oepning.

(6) The pipetted sediment is dropped on a slide glass for microscopic examination at a low-power magnification.

(7) The number of larvae per gram of feces (LPG) can be determined by dividing the number of detected larvae by three.

## Immunity and X-irradiated larval vaccine

It has been proved by many researchers that a moderate infection caused in calves by the significant attack of this parasite or by experimental inoculation with a controlled number of infective larvae results in an acquisition of solid immunity against reinfection.

Jarrett et al. (1958, 1959, 1960)<sup>4).5).6)</sup> of Glasgow University made an attempt to develop an X-irradiated larval vaccine on the basis of this fact. The vaccine is utilized practically at present. It contains larvae partially inactivated by irradiation with X-rays at 40 KR, and is given orally twice at a two-week interval before the pasturing of calves. Calves administered with this vaccine suffer from mild dictyocaulosis, which confers them immunity to the probable succeeding infection.

The disadvantages of this vaccine in field use are the early expiration date, necessity of double vaccination, and difficulty in the maintenance of its activity during transportation. Many problems may have to be solved, therefore, before the vaccine is applied practically in the field, except in fairly well organized regions.

### Treatment and prophylaxis

Naturally, the administration of this vaccine is very effective for the prophylaxis of this disease. The utilization of the vaccine, however, is rather limited because of the disadvantages mentioned above. As effective anthelmintics against swine lungworms, diethylcarbamazine, cyanacethydrazide, ascaridole-santonin aerosol and methylridine have been reported so far by many researchers. Practically, they do not always bring about satisfactory results.

Tetramisole is also very effective against bovine Dictyocaulus, showing a marked efficacy in the extermination of swine lungworms. In Japan, the field application of this compound has been studied not only to eliminate the parasite in the lung but also to confer acquired immunity to calves exposed to natural infection.

Although no conclusion has yet been drawn from these studies, the following recommendation may be made under the actual pasturing conditions of yearling calves in Japan:

Calves should be administered with tetra misole twice at a 40-day interval after the initial administration, which is performed one week after the recognition of an outbreak of this infection. By this step the damage of bovine dictyocaulosis may be reduced to the minimum throughout the pasturing period.

### References

- Bello, T. R.: Comparison of the flotation of Metastrongylus and Ascaris eggs in 3 different leviation solution. Amer. J. Vet. Res. 22, 597-600 (1961).
- Douvres, F. W. & Lucker, J. T.: The morphogenesis of the parasitics stages of the cattle lungworm, *Dictyocaulus viviparus*, in experimentally infected guinea pigs. *J. Parasit.* 44, 28-29 (1958).
- Jarrett, W. F. H. et al.: The pathology of experimental bovine bronchitis. J. Pathol. Bacteriol. 73, 183-193 (1957).
- Jarrett, W. F. H. et al.: A field trial of parasitic bronchitis vaccine. Vet. Rec. 70, 451-454 (1958).
- Jarrett, W. F. H. et al.: Immunological studies on *Dictyocaulus viviparus* infection calves—double vaccination with irradiated larvae. *Amer. J. Vet. Res.* 20, 522-526 (1959).
- Jarrett, W. F. H. et al.: Immunological studies on *Dictyocaulus viviparus* infection, Immunity produced by the administration of irradiated larvae. *Immunology* 3, 145–151 (1960).
- 7) Kammer, H. & Hanson, R. P.: Studies on the transmission of swine influenza virus with Metastrongylus species in specific pathogens-free swine. J. Infect. Dis. 110, 99-102 (1962).
- MacKenzie, A.: Experimental observations on lungworm infection together with virus pneumonia in pigs. *Vet. Rec.* 75, 114–115 (1963).
- Onishi, T.: Immunological studies on Metastrongylus apri infection. I. Immuniz- ing activity produced by the administration of X-irradiated larvae to guinea pigs. Jap. J. Parasitol. 6, 516-524 (1968).
- Onishi, T.: Immunological studies on Metastrongylus apri infection. II. Immunizing activity of various oral dosages of larval vaccine between oral and subcutaneous inoculation. Jap. J. Vet. Sci. 35, 507-513 (1973).
- Poynter, R. et al.: Recent experiences with vaccination. Vet. Rec. 72, 1078-1090 (1960).
- 12) Shope, R. E.: The swine lungworm as a reservation and intermediate host for swine influenza virus. II. The transmission of swine influenza virus by the swine lungworm. J. Exp. Med. 74, 49-68 (1941).

- Thienpont, D. et al.: Tetramisole (R 8299). A new potent broad spectrum anthelmintic. Nature 209, 1084-1086 (1966).
- 14) Ueno, H. et al.: Spontaneous liberation of infective larvae from the earthworm, *Eisenia* .foetida, infected with Metastrongylus apri. Nat. Inst. Anim. Hlth Quart. 6, 89-100 (1966).
- 15) Ueno, H. et al.: Anthelmintic evaluation of tetramisole against the swine lungworm, Metastrongylus apri. Nat. Inst. Anim. Hlth

Quart. 7, 65-73 (1967).

- 16) Ueno, H.: A simple method for detection of first-stage larvae of *Dictyocaulus viviparus* from feces. I. Procedure and fundamental tests. J. Jap. Vet. Med. Ass. 21, 210-213 (1968<sup>a</sup>).
- 17) Ueno, H.: A simple method for detection of first-stage larvae of *Dictyocaulus viviparus* from feces. II. Comparison with other methods and field application. J. Jap. Vet. Med. Ass. 21, 255-259 (1968<sup>a</sup>).