Ciliate Protozoa from the Rumen of the Japanese Serow, 
*Capricornis crispus* (Temminck)

Soichi IMAI  
Department of Parasitology, Nippon Veterinary and Zootechnical College,  
Musashino-shi, Tokyo 180

Masafumi ABE  
Biological Institute, Faculty of Science, Tohoku University,  
Sendai-shi, Miyagi 980

Keiji OGIMOTO  
Department of Animal Science, Faculty of Agriculture, Tohoku University,  
Sendai-shi, Miyagi 980

(Received for publication June 11, 1980)

Abstract. The composition of rumen ciliate protozoa from the Japanese serow was investigated. The average number of total ciliates per milliliter of rumen contents was $1.4 \times 10^6$. Eleven species, one of trichostomatid and ten of entodiniomorphid ciliates, were recognized. Of them, ten species had already been described in the domestic ruminants in Japan, but one was detected specifically from the Japanese serow. This species was described as the new form, *Epidinium ecaudatum* forma *capricornis* forma nov. This form had bifurcated and trifurcated caudal spines. Species or formae with trifurcated spines had been described not in the genus *Epidinium* but in the genus *Ophryoscolex*. The fact may be one of the characters showing the phylogenetical relationship between these two genera.

The descriptions of ciliates living in the rumen of wild ruminants have been published by investigators on antelopes [13, 15, 28, 39, 40], ibex [6, 7, 35], musk-ox [10, 30], chamois [5], and various deers [10, 27, 34, 36, 37, 41]. Little is known, however, about the ciliate fauna of wild ruminants in Japan, although a report has been made on that of domestic ruminants in the country [20].

The present paper describes the ciliates in the Japanese serow, *Capricornis crispus* (Temminck), which is found only in Japan, with one new form of epidinian species, *Epidinium ecaudatum* forma *capricornis*.

Materials and Methods  
Samples of rumen contents were obtained from 30 adult and 1 young Japanese serows. Four of these animals had been captured around Sendai City, Miyagi Prefecture, and kept at the Yagiya zoological park, Sendai City, for various seasons over a period from 1970 to 1974 (the Miyagi group). They were constantly fed apples, sweet potatoes, dried small sardines and hay or fresh grass. None of them had come in contact with any other ruminant during their life in the park. Samples were collected from them by the aid of a rumen catheter. The other 27 animals had been captured in Gifu Prefecture in December, 1979 and January, 1980 (the Gifu group).

The rumen contents of the Miyagi group were filtrated through a double-folded sheet of gauze into flasks. For the identification of ciliates by optical microscopy, they were treated with 50% (W/V) sucrose solution and washed with 0.9% (W/V) NaCl.
solution in such manner as reported previously [21].

The resulting samples were fixed and stained in MFS solution, which was 10% formalin solution containing 0.1% methylgreen and 0.8% NaCl.

Specimens from the Gifu group were at first fixed in 70% (V/V) ethanol. They were refixed and stored in MFS solution. Some of them were stained with Mayer's hematoxylin for whole mounts. The others were treated in the following manner for scanning electron microscopy. They were washed 5 times overnight with distilled water and refixed in Párducz's fixative [31] at room temperature for 15 minutes. Then they were washed 5 times with distilled water and treated in the same manner as described previously [22].

Measurement of the ciliates was made by an ocular micrometer. Mean length and width of the body were obtained from 20 individuals selected at random.

Samples for the protozoal count were not pre-treated with 30% sucrose solution in order to prevent loss of protozoa. Protozoa contained in a 1:5 dilution of sample in MFS solution were counted with a plankton counter deck glass.

The genera and species of these ciliates were identified mainly on the basis of the descriptions published by previous workers [13, 25, 26].

The terminology of the ophryoscolecid ciliates for the orientation conforms to the system proposed by Lubinsky [29] and by Dehoryt [11]. For the genus of the subfamily Entodiniinae, the side closest to the macronucleus was named the right. For the genera of the subfamilies Diplodiniinae and Ophryoscolecinae, it was reversed and named the left.

**Results**

Table 1 shows the average numbers of the total ciliates and the composition of the 4 genera which appeared. The ciliates of the genus *Entodinium* appeared at a high frequency, or about 80% of the total ciliates. The large entodiniomorphid group, consisting of *Elytroplastron* and *Epidinium*, was usually found at the rate of less than $10^4$/ml.

The number of species having appeared in one sero was 4.9 on the average, ranging from 2 to 7.

Eleven species, composed of 1 trichostomatid and 10 entodiniomorphid species, were revealed in the present studies. All of them had already been described in the domestic ruminants in Japan [20], except one which was detected specifically from the Japanese serow and proposed as a new form. All the 11 species of ciliates are mentioned below.

1. *Dasytricha ruminantium* (Schuberg, 1888)

This trichostomatid species was relatively small with rows of cilia arranged in spiral. The macronucleus was elliptical and the situation definite. The vestibulum was located at the anterior end of the body. The range of body length was 70–100 μm and that of body width 40–50 μm.

Occurrence: Two individuals (6.6%) of the hosts examined.

2. *Entodinium exiguum* (Dogiel, 1925)

The body was rounded and flattened bilaterally. The macronucleus was short and thick. The range of body length was 20–27 μm and that of body width 15–18 μm.

Occurrence: Fourteen individuals (45.2%) of the hosts examined.

3. *Entodinium nanulum* (Dogiel, 1922)

This species resembled the preceding one, although the body was longer and the posterior part of the body more slender in the former. The length of the macronucleus was about one-third of that of the body.

<table>
<thead>
<tr>
<th>Table 1. Average densities of total ciliates and the composition of genera</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Total count of ciliates (log10/ml)</strong></td>
</tr>
<tr>
<td><strong>Dasytricha</strong></td>
</tr>
<tr>
<td><strong>Entodinium</strong></td>
</tr>
<tr>
<td><strong>Elytroplastron</strong></td>
</tr>
<tr>
<td><strong>Epidinium</strong></td>
</tr>
</tbody>
</table>

<sup>1)</sup> Average of 31 samples.  
<sup>2)</sup> Standard deviation.  
<sup>3)</sup> Range.
The range of body length was 32–45 μm and that of body width 18–25 μm.

Occurrence: Twenty-six individuals (83.9%) of the hosts examined. This was one of the most commonly and abundantly found species in the Japanese serow.

4. Entodinium minimum (Schuberg, 1888)

The body was asymmetrical and somewhat triangular in shape. The right surface was strongly convex, and the left one flattened or concave. The macronucleus was rod-shaped and long. The range of body length was 30–50 μm and that of body width 20–32 μm.

Occurrence: Five individuals (16.1%) of the hosts examined.

5. Entodinium simplex (Dogiel, 1925)

The body was ovoid and flattened bilaterally. The macronucleus was thick in the anterior part and thin in the posterior part. The length of the macronucleus was about one-third of that of the body. The range of body length was 35–50 μm and that of body width 25–40 μm.

Occurrence: Twenty-nine individuals (93.5%) of the hosts examined. Like E. nanelium, this was one of the most commonly found species in the Japanese serow.

6. Entodinium dubardii (Buisson, 1923)

This species closely resembled E. simplex, except that its macronucleus was of approximately the same thickness in the anterior as in the posterior part, as described by Wertheim [38]. The range of body length was 40–55 μm and that of body width 30–40 μm.

Occurrence: Eleven individuals (35.5%) of the hosts examined.

7. Entodinium longinucleatum

(Dogiel, 1925)

The body was ovoid with a very long macronucleus both ends of which were smoothly rounded and which usually occupied the entire right side of the body. The range of body length was 50–80 μm and that of body width 40–60 μm.

Occurrence: Two individuals (6.5%) of the hosts examined.

8. Entodinium lobospinosum

(Dogiel, 1925)

This species had 2 different posterior processes. The right process was sharp and long, while the left one was blunt. The range of body length was 35–45 μm and that of body width 20–28 μm. The processes varied widely in length.

Occurrence: Three individuals (9.7%) of the hosts examined.

9. Entodinium ekendrae (Das-Gupta, 1935)

This species had 3 different posterior processes. The right process was sharp and long. Of the 2 left processes, one was short and sharp, and the other short and blunt. The micronucleus was situated at the posterior end of the macronucleus. The range of body length was 37–45 μm and that of body width 25–35 μm.

Occurrence: Two individuals (6.5%) of the hosts examined.

10. Elytroplastra bubali (Dogiel, 1928)

This species resembled Polyplastra multivesiculatum (Dogiel et Fedorowa, 1925) which had been detected at a high frequency in the rumen of the domestic ruminants in Japan [20]. It was, however, distinguished easily from P. multivesiculatum by the following points. It has 3 big and 1 small skeletal plates and 4 contractile vacuoles located constantly on the left side of the macronucleus. P. multivesiculatum had 2 big and 3 small skeletal plates and more than 5 contractile vacuoles. The range of body length was 100–150 μm and
that of body width 75–90 μm. Optical microscopy revealed that the ciliates of *Ely. bubali* had ingested small ciliates, such as *E. longinucleatum* and *D. ruminantium*, and fragments of plants.

Occurrence: Twenty-nine individuals (93.5%) of the hosts examined.

11. *Epidinium ecaudatum* (Fiorentini, 1889) forma *capricornisi* forma nov. (Figs. 1–9)

Description: The body is elongate (length 1.5–2.6 times as much as width) and tapered toward the posterior end. The right surface is slightly concave, and the left surface convex. Five caudal spines arise from the posterior end of the body. They are approximately equal in length. Of them, one is on the right, another one on the left, two are on the upper and the other one is on the lower side. The extremities of the 2 spines on the upper side bifurcate. The one on the lower side trifurcates. Fine longitudinal striations extend over the cuticle. There are 3 skeletal plates on the
Rumen ciliates in Japanese serow

upper and the right sides. The macronucleus is situated on the upper left side adjacent to the edge of the skeletal plate. Its anterior end is blunt, while its posterior part gradually tapers to the posterior extremity. There is a notch in the middle region of the macronucleus. The ellipsoidal micronucleus is in the notch of the macronucleus. Two contractile vacuoles are on the left side. The narrow rectum and the cytopyge are situated at the dextroposterior end of the body.

Dimension: Measurements were made on 20 specimens selected at random. The results obtained are given below as the mean and the standard deviation, with the range shown in parentheses.

Length (L): 110.6±11.6 μm (90–130 μm)
Width (W): 57.7±4.8 μm (50–70 μm)
Length of spine: 12.6±2.3 μm (5–15 μm)
L/W: 1.93±0.25 (1.50–2.60)

Variation: The caudal spines of Epidinium ecaudatum forma capricornisi vary a little widely in length and morphology. In some individuals the trifurcated spine is obscure. Occurrence: Twenty-six individuals (83.9%) of the hosts examined.

REMARKS.
Epidinium ecaudatum forma capricornisi resembles Ep. ecaudatum forma parvicaudatum (Awerinzew et Mutafowa, 1914) and Ep. ecaudatum forma rusa (Benerjee, 1955) because of its possession of 5 caudal spines. Both formae, however, are lacking in bifurcated and trifurcated spines.

Discussion

No investigation has ever been conducted on the ciliate fauna in the rumen of the Japanese serow, Capricornis crispus (Témminck). The total number of ciliates in the rumen of the Japanese serow was 1.4×10^6/ml on the average. It was almost the same as that in cattle or sheep [20]. The fact suggests that as a nitrogen source for the host [19], the ciliate in the rumen may play almost the same role in the Japanese serow as in the domestic ruminants.

The present studies revealed 11 species, including 1 new form of the epidinian species, Epidinium ecaudatum forma capricornisi forma nov. All the species, except two, Ely. bubali and Ep. ecaudatum forma capricornisi, have been found at a relatively high frequency in cattle, sheep and goats in Japan [20]. Ely. bubali was first detected by Dogiel [14] from the water buffalo, and later reported from cattle in Japan [20]. It appeared very infrequently in cattle, but at a high frequency in the Japanese serow.

In the chamois inhabiting the southern part of Europe and closely related phylogenetically to the Japanese serow, 5 genera of ciliates, Entodinium, Diplodinium, Enoploplastron, Epidinium and Ophryoscolex, have been reported by Christl [5]. Of them, only 2 genera are common in the Japanese serow. On the other hand, all the 7 genera that appeared in these 2 wild animals have been detected from the domestic ruminants in U.S.S.R. [13] and Japan [20]. The facts may show the presence of a geographical feature in the distribution of ciliates. This feature appears to have been caused by the secondary infection of ciliates from the other species of ruminants inhabiting the same locality, in addition to the phylogenetic factor of the host related to the distribution.

Three forms with 5 caudal spines of Epidinium ecaudatum have been described. Ep. ecaudatum forma parvicaudatum has been detected all over the world, i.e., from cattle in U.S.S.R. [1, 13], U.S.A. [33], China [18] and New Zealand [8], from the gaur in India [24], from sheep in U.S.S.R. [13] and China [17], from goats in U.S.S.R. [13], and from the giraffe in South Africa [23]. Ep.
ecaudatum forma cattanei has been described from cattle in Italy [16], U.S.S.R. [1, 13], France [4], U.S.A. [3] and China [18], from zeus in India and Sri Lanka [26], from sheep in China [17], India [2] and Japan [20], and from goats in India [9]. Ep. ecaudatum forma rusa has been detected from the sambar in India [2]. All the 3 forms, however, have simple caudal spines not furcated. Species or formae with bifurcated or trifurcated spines have been found not in the genus Epidinium but in the genus Ophryoscolex. The fact may be one of the characters showing the phylogenetic relationship between these 2 genera.

Acknowledgments. The authors are greatly indebted to Dr. S. Nemoto, director, and Dr. S. Onodera, assistant director, and the staff of the Yagiya Zoological Park, Sendai, for arrangement in collecting samples, and to Dr. S. Takatsuki, of the Biological Institute, Faculty of Science, Tohoku University, Sendai, for supply of samples. They also wish to express their thanks to Mr. Y. Tazawa and Mr. K. Ike, of the Department of Parasitology, Nippon Veterinary and Zootecchnical College, Musashino, Tokyo, and to Mr. K. Suzuki, of the Biological Institute, Faculty of Science, Tohoku University, Sendai, for technical assistance.

This work was supported in part by Grant-in-Aid 476214 from the Scientific Research Fund of the Ministry of Education, Science and Culture, Japan.

References
RUMEN CILIATES IN JAPANESE SEROW


要約

ニホンカモシカ *Capricornis crispus* (Temminck) のルーメン内線毛虫相：今井壮一（日本獣医畜産大学寄生虫学教室），安倍正史（東北大学理学部生物学教室），肩元敬司（東北大学農学部畜衛生学教室）—ニホンカモシカ *Capricornis crispus* 51頭より得たルーメン内容に見出される線毛虫を検査した。その結果、ルーメン内容 1 ml あたりの平均線毛虫数は 1.4×10⁸ であり、反芻家畜のそれとはほぼ同様の数値が得られた。出現線毛虫種は11種が同定され、1 種は毛口類に属するもの、他10種はエントディニオモルファ類に属するものであった。これらのうち、10種は本邦の反芻家畜からも認められているものであったが、1 種はカモシカ特有のものであり、詳細な形態学的検討の結果、*Epidinium ecaudatum* の新型として、*Epidinium ecaudatum* forma *capricornisi* と命名した。本種は二叉、又は三叉に分岐した尾棘をもつが、現在まで三分岐した尾棘をもつルーメン内線毛虫は *Ophryoscolex* 属に限られており、*Epidinium* と *Ophryoscolex* との系統学的な関係を示唆する特徴の一つとして興味深いものと思われる。

Explanation of Figures

All the figures are photomicrographs of *Epidinium ecaudatum* forma *capricornisi* forma nov.

Figs. 2-4. Optical photomicrographs of an individual viewed from the lower side. Focus is adjusted on the lower (Fig. 2), the central (Fig. 3), and the upper (Fig. 4) side. A trifurcated spine is situated on the lower surface and bifurcated spines on the upper side. Each figure is at ×360.

Figs. 5-7. Optical photomicrographs of an individual viewed from the upper side. Focus is adjusted on the upper (Fig. 5), the central (Fig. 6), and the lower (Fig. 7) side. Each figure is at ×400.

Figs. 8 and 9. Scanning electron micrographs of the caudal spines. In Fig. 8 the trifurcated spine is evident. Fine longitudinal striations are shown on the body surface. ×1,400. In Fig. 9 the bifurcated spine is shown. ×1,970.