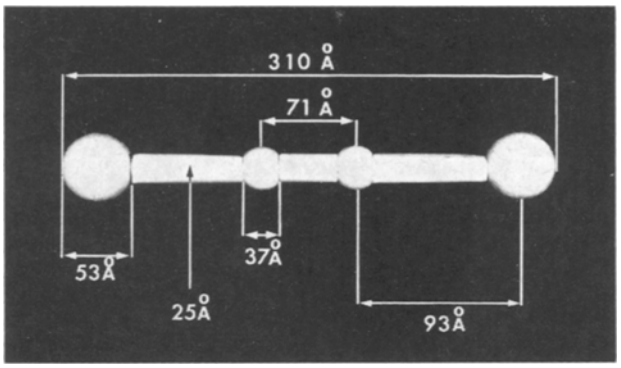

Fig. 2. Proposed model for the structural protein sub-unit of *B. undatum* periostracum.

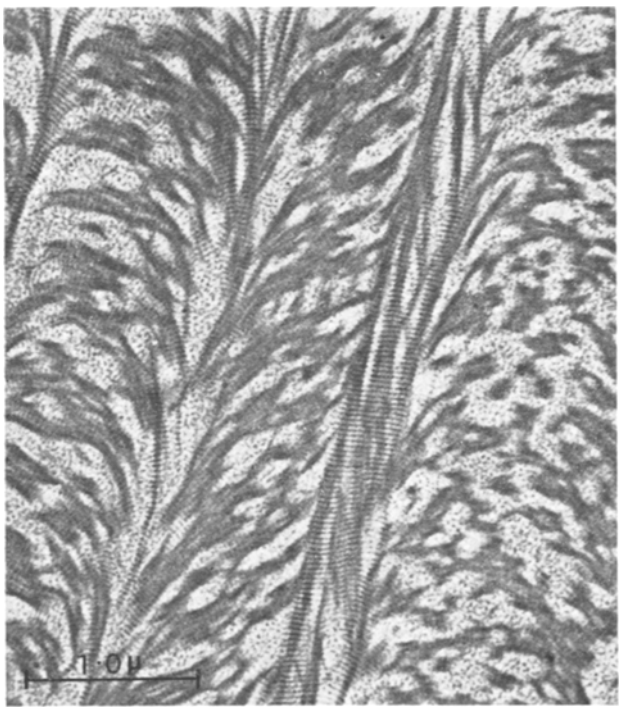

Fig. 3. Electron micrograph of a section of *B. undatum* periostracum made in a plane perpendicular to the surface of the periostracum (horizontal in this picture). Fixed in 5% glutaraldehyde pH 7.0 cacodylate buffer, 0.2 *M* sucrose. Post-fixed in osmic acid and embedded in Epon. Stained with uranyl acetate and Reynold's lead stain. × 35,000.

structure built up from ribbons in which features of the striated pattern already described can be clearly seen.

This type of parabolic lamellar structure, with the appearance of the flowering of systems of bundles or sheets of fibres, has been noted elsewhere; principally in arthropod cuticle[6,7] but also in the tunic of tunicates[8]. A similar phenomena has been noted for certain liquid crystal systems[9] and in the globules of oothecal structure protein secreted into the lumen of the left collateral gland of the praying mantis[10].

An explanation of this type of parabolic lamellar structure has been proposed[6] on the basis of Moiré patterns formed from fibril systems seen in oblique section. These systems consist of superimposed sheets of fibres, each sheet having all of the individual fibre axes parallel, and with every sheet rotated through a small angle relative to that above it. A regular progression of rotation, always in the same sense, down the stack of sheets is proposed. Oblique sectioning through such a stack gives rise to the artefact of parabolic lamellae as has been demonstrated by model building[10].

On the basis of the available evidence therefore we suggest that the periostracum of this gastropod is organized from stacked sheets of ribbons or fibres, composed from small asymmetric sub-units as described above, in which the mean fibre axis of each sheet is specifically rotated relative to that of the sheets immediately above and below it and in which the plane of the sheets is parallel to the surface of the periostracum[11].

Résumé. Le périostracum du gastéropode *Buccinum undatum* L. se compose de rubans de protéine fibreuse disposés en lames formant des stries répétées. Ces rubans sont accolés et joints bout à bout. Leurs éléments constitutifs (subunités) sont en forme de haltères ressemblant à la molécule de fibrinogène.

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[6] Y. Bouligand, C. r. hebd. Séanc. Acad. Sci. Paris *261*, 3665 (1965).
[7] M. Locke, J. Biophys. Biochem. Cytol. *10*, 589 (1961).
[8] A. C. Neville, Adv. Insect Physiol. *4*, 213 (1967).
[9] C. Robinson, Molec. Crystals *1*, 467 (1966).
[10] W. Kenchington and N. E. Flower, J. Microsc. *89*, 263 (1969).
[11] We thank the Department of Physics at the University of Lancaster for providing facilities of their electron microscope.

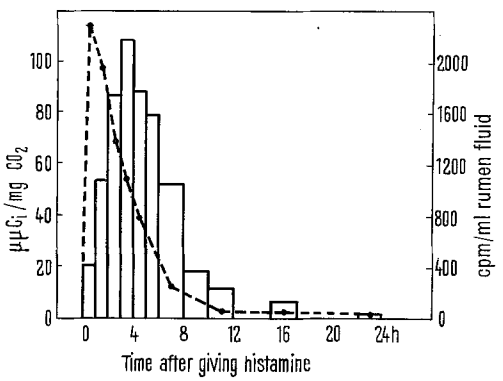
In vivo Rupture of the Imidazole Ring of Histamine

Histamine is catabolized through various pathways in mammalian tissues but the imidazole ring always remains intact. When the substance is injected into animals it is largely excreted in the urine in the form of various metabolites[1]. Schayer[2] injected two rats and a guinea-pig with about 0.1 μg of ring-$^{14}C_2$-histamine per g and detected no radioactivity in the expired air. In a forth animal, injected with about 10 times this quantity of $^{14}C_2$-histamine he detected minute quantities of $^{14}CO_2$ which he thought might be due to an impurity in the histamine used.

We have recently done some experiments which suggest that in ruminant animals the imidazole ring of dietary histamine is ruptured. In such animals the food is thoroughly fermented by micro-organisms in the rumen before it passes to the intestines. Previous work has shown that when $^{14}C_2$-histamine and carrier histamine were given by mouth the biological activity disappeared

rapidly from the rumen of sheep[3]. The radioactivity disappeared more slowly, indicating that histamine inactivation takes place in the rumen. Less than 30% of the administered radioactivity was recovered in urine and faeces[4]. Since this greatly differed from observations on injected histamine we examined the possibility that the imidazole ring of histamine is split by ruminal microorganisms leading to the exhalation of $^{14}CO_2$.

In the present experiments Scottish Blackface ewes were given 800 g of pelleted dried grass daily. Exhaled air was collected by placing the animals head in a loosely-fitting hood. Air was pumped out of the hood so as to maintain a slightly negative pressure and a small, known fraction of this air flow was passed through a series of absorption towers containing NaOH solution. At the end of each collection period the quantity of expired CO_2 was determined by titration of an aliquot of the contents of each of the absorption towers with acid and the remainder was precipitated with $BaCl_2$. The precipitated $BaCO_3$ was washed, dried and weighed and duplicate samples were suspended in POPOP-PPO-toluene scintilation fluid using Cab-O-Sil as gelling agent. The radioactivity was measured in a Panax scintillation counter. About 10,000 counts were recorded and the amount of $^{14}CO_2$ exhaled was calculated with due allowance for atmospheric temperature and pressure, atmospheric CO_2, background radioactivity and efficiency for counting. Radioactivity of centrifuged rumen liquor was measured by plating 100 µl and counting in a Beckman flow-counter (Lowbeta II).



Sheep Maud 31.1.67. Radioactivity of exhaled CO_2 (bars) and of rumen fluid (line) after administering 50 µCi of $^{14}C_2$-histamine-dihydrochloride and 200 mg histamine acid phosphate into the rumen by way of a permanent fistula. CO_2 was collected continuously for 12 h after dosing and then for 2 further 2 h periods at 15 and 22 h after dosing.

Specific activity of exhaled and rumen CO_2 after administering 50 µCi $^{14}C_2$-histamine dihydrochloride and 200 mg histamine acid phosphate into the rumen

Exhaled CO_2 Radioactivity (µµCi/mgCO_2)	Time after dosing (min)	Rumen CO_2 Radioactivity (µµCi/mgCO_2)	Time after dosing (min)
54	60–120	291	90
109	180–240	328	210
52	360–480	162	420

The results of a typical experiment are illustrated in the Figure. The sheep was given 50 µCi of $^{14}C_2$-histamine dihydrochloride (The Radiochemical Centre, Amersham) and 200 mg unlabelled histamine acid phosphate by way of a rumen fistula. Radioactivity in the rumen contents decreased rapidly to reach very low values after 10 h while the specific activity of the exhaled CO_2 increased initially, reaching a peak after 3–4 h, and then declined rapidly. In this experiment about 28% of the administered radioactivity was recovered in the exhaled air during the first 24 h after dosing. In 4 other experiments of this type 27–34% of the administered radioactivity was recovered as $^{14}CO_2$.

Further, the specific radioactivities of the exhaled air and the rumen contents were compared. Samples of rumen contents were aspirated at intervals after dosing and strained; CO_2 was expelled from the fluid with sulphuric acid and absorbed by diffusion in NaOH. The specific radioactivity was measured as for respiratory CO_2. The specific activity of ruminal CO_2 was from 3 to 5 times that of exhaled CO_2 (Table). It is thus clear that rupture of the imidazole ring is taking place in the fore-stomachs. But since the total quantities of CO_2 formed in the tissues greatly exceed those produced by the contents of the fore-stomachs, our data do not exclude the unlikely possibility that rupture of the imidazole ring might also take place in the tissues after absorption of histamine metabolites.

Rupture of the imidazole ring has previously been demonstrated in vitro using a 200-fold purified enzyme preparation from a strain of *Pseudomonas* which had been adapted to imidazole acetic acid[5]. In the presence of DPNH and oxygen this system metabolizes imidazole acetic acid to formylaspartic acid. Efforts to establish the pathways leading to the rupture of the imidazole ring by the contents of the fore-stomachs might meet with difficulties since histamine inactivation by rumen contents is not easily demonstrated in vitro[3].

In vivo rupture of the imidazole ring of histamine is probably not confined to the ruminant fore-stomach. It is, for example, not unlikely that some of the histamine formed in the large intestine of both ruminant and non-ruminant animals is degraded by intestinal bacteria through pathways leading to the rupture of the imidazole ring.

Zusammenfassung. Es wird gezeigt, dass Histamin auch durch das Aufreissen des Imidazolringes im Vormagen von Schafen katabolisiert werden kann. C^{14}-markiertes Histamin wurde in den Pansen des Schafes oral eingeführt und annähernd 30% davon in der Atmungsluft (CO_2) aufgefangen.

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1 R. W. SCHAYER, Physiol. Rev. *39*, 116 (1959).
2 R. W. SCHAYER, J. biol. Chem. *196*, 469 (1952).
3 Ø. V. SJAASTAD, Acta vet. scand. *8*, 157 (1967).
4 Ø. V. SJAASTAD, Acta vet. scand. *8*, 176 (1967).
5 O. HAYAISHI, H. TABOR and T. HAYAISHI, J. Am. chem. Soc. *76* 5570 (1954).
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