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THE CELLULASE CONTENT OF VARIOUS SPECIES OF COCKROACHES

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Abstract—Cellulase was demonstrated in many groups of cockroaches. The enzyme is secreted by the salivary glands and in the lumen of the intestines. Wide variation was shown in the content of enzyme per unit weight of insect and in its origin and distribution between the intestines and the salivary glands. As exemplified by the blattids, which produce the largest amount of cellulase per unit weight, there is no loss of cellulase-producing activity by the omnivorous species to correspond with their lessened dependence on wood, as contrasted with the wood-eating *Cryptocercus* and *Panesthia*. Although some species were alike, there was no consistent familial relationship in the overall content of cellulase and its production by the salivary glands. There was no consistent pattern of evolutionary development.

CELLULASE has been shown to be produced by the salivary glands and by the microflora or microfauna of other parts of the alimentary tract in many species of cockroaches (WHARTON *et al.*, 1965). These two sources of the enzyme appear to be independent of one another, so that it might be expected that in some species of cockroach only one of these sources would function. Yet even in *Cryptocercus punctulatus* which depends for survival on the cellulase generated by the xylophagous protozoa of the hindgut (CLEVELAND *et al.*, 1934), the salivary glands produce some cellulase. On the other hand, omnivorous forms, like *Periplaneta americana*, which do not appear to depend on cellulase activity since they may subsist on a diet of egg alone (WHARTON *et al.*, 1965), may produce considerable amounts of the enzyme in both the salivary glands and the midgut. Active salivary glands have not been observed in the absence of intestinal cellulase, but this may be explained either by the accumulation of cellulase from the saliva by the food or by its production by micro-organisms in the midgut, or both. If cellulose is a part of the diet of the cockroach, we might further expect to find characteristic enzyme patterns which reflect the ecological experience of the different groups.

MATERIALS AND METHODS

The salivary glands and intestines were excised and treated according to the methods previously described (WHARTON *et al.*, 1965). 0·5 ml of an extract was added to 0·5 ml of a 1·0% solution of carboxymethyl cellulose (D.S.O.52) and incubated at pH 4·5 and 50°C for 1 hr. Under these conditions, one cellulase unit produces 0·50 mg of reducing sugar (as glucose) (REESE and MANDELS, 1963).

RESULTS AND DISCUSSION

Examination of many taxa fed on a diet of Purina Laboratory Chow—except for *Cryptocercus punctulatus* which was fed on wood—shows that considerable variations occur in the origin, distribution, and content of cellulase (Table 1). *Panchlora nivea* and *Blattella germanica* appear to lack cellulase. *Supella supellectilium* generates a very small amount in the salivary glands and less in the intestines; *Panesthia javanica* contains it as a bacterial product in its foregut (CLEVELAND *et al.*, 1934). *C. punctulatus* contains it in the protozoa of the hindgut (CLEVELAND *et al.*, 1934), and has minute to large amounts in the salivary glands, showing much greater variance in this respect than the other species examined. *P. americana* has moderate to large amounts in the salivary glands and most of the remainder in the midgut and caeca; *Gromphadorhina brunneri* produces large amounts and is also distinguished by an exceptionally active salivary gland system. Variations of these types occur in other species.

It is evident (Table 1) that the blattids produce by far the largest amount of cellulase per unit weight of insect. This ratio does not vary with the size of the insect, as shown by *B. orientalis* and *E. floridana* among the blattids, and *N. cinerea* and *L. maderae* among the blaberids. There is also no consistent familial relationship between overall content of cellulase and its production by the salivary glands. For example, in the family Blattidae, *Eurycotis floridana* of the subfamily Polyzosteriinae, and *Blatta orientalis* of the subfamily Blattinae, differ not only in their total content of cellulase which might be attributed to differences in size, but most especially in the relative activities of their salivary glands. Although the total content of cellulase in *B. orientalis* compares favourably with that of *P. americana*, which is in the same subfamily, the activities of the salivary glands are quite different. A similar difference is seen in *Periplaneta*. Among the Blaberidae, *Capucina patula* and *Panchlora nivea*, of the subfamily Panchlorinae, are characterized as having little or no cellulase, respectively, but *Leucophaea maderae* and *Gromphadorhina brunneri* of the subfamily Oxyhaloinae contain respectively moderate and large amounts of cellulase and differ in the relative activity of the salivary glands to the whole, which in *G. brunneri* reaches the exceptionally high ratio of 50 per cent. Even between species of the same genus great differences may exist as shown by the very weak activity of the salivary glands of *P. australasiae* compared with *P. americana* and *P. fuliginosa*. In *Blaberus*, however, there is a greater interspecific consistency in total cellulase content and the activity of the salivary glands.

The Panchloroid complex (MCKITTRICK, 1964) consists of subfamilies that differ greatly from one another not only in size and appearance but also in the content and distribution of cellulase, as shown by the extremes of *Panchlora* which has little or no cellulase and *Gromphadorhina* which is rich in this enzyme. Differences in size, which might seem to account for the quantitative differences in total cellulase in this group, are evidently not responsible for the relative activity of the salivary glands to the whole, as illustrated by *Capucina* 1 : 9, *Nauphoeta* 1 : 19, and *Gromphadorhina* 1 : 2.

TABLE 1—CONTENT AND DISTRIBUTION OF CELLULASE IN THE SALIVARY GLANDS AND INTESTINAL TRACT OF DIFFERENT SPECIES OF COCKROACHES (*Blattaria**)

	Classification	Wt. in mg†	Total units per mg	Units per salivary gland	No. tested	Units per‡ whole intestine	No. tested	Ratio SG:WI
	Cryptocercidae							
	Cryptocercinae							
	Cryptocercus punctulatus Scudder	721	0·09	20·2	9	41	7	1:2
	Blattidae							
	Blattinae							
	Blatta orientalis (L.)	398	0·64	2·6	2	248	2	1:95
	Periplaneta americana (L.)	648	0·33	51·6	5	160	15	1:3
	P. australasiae (F.)	411	0·77	5·4	3	310	2	1:60
	P. fuliginosa (Serv.)	650	0·56	23·8	3	342	2	1:14
	Polyzosteriinae							
	Eurycotis floridana (Walk.)	1680	0·67	90·0	3	1040	2	1:12
	Blattellidae							
	Blattellinae							
	Supella supellectilium (Serv.)	52	0·02	0·6	2	0·3	2	1:1
	Blattella germanica (L.)	53	—	0	3	0	2	—
	Blaberidae							
Panchloroid complex	Pycnoscelinae							
	Pycnoscelus surinamensis (L.)	210	0·11	1·1	4	22	2	1:20
	Diplopterinae							
	Diploptera punctata (Esch.)	175	0·17	0·6	3	29	2	1:49
	Panchlorinae							
	Panchlora nivea (L.)	138	—	0	6	0·3	4	—
	Capucina patula (Walk.)	518	0·02	0·6	2	5·4	2	1:9
	Oxyhaloinae							
	Leucophaea maderae (F.)	2398	0·07	20·4	6	140	2	1:7
	Gromphadorhina brunneri Butler	7367	0·21	540·0	1	978	1	1:2
	Nauphoeta cinerea (Oliv.)	489	0·36	8·8	4	162	4	1:19
Blaberoid complex	Zetoborinae							
	Phortioeca phoraspoides (Walk.)	1029	0·16	7·5	2	162	1	1:22
	Blaberinae							
	Blaberus craniifer Burm.	3228	0·08	4·6	3	240	1	1:52
	B. discoidalis (Serv.)	2101	0·18	18·8	3	362	2	1:19
	B. giganteus (L.)	4002	0·05	5·0	4	214	3	1:43
	Byrsotria fumigata (Guér.)	1514	0·12	20·8	5	162	5	1:8

* McKittrick's classification (1964).
† Weights are the average of several individuals, but not of those actually tested.
‡ Unwashed. The washed out segments of the intestines are virtually devoid of cellulase; see also Wharton *et al.*, 1965.

In view of the great antiquity of the cockroach, and of the dependence of the primitive form, *Cryptocercus*, on its cellulolytic protozoan symbionts for digesting its diet of wood, broad relationships in the cellulase content of the taxonomic groups might have been established as new forms emerged. However, few such relationships are apparent and their general absence prompts an examination of the premise on which the expectation was founded. *Cryptocercus* depends for its existence on the cellulolytic activity of its symbiotic protozoa which in turn are intimately dependent on the physiology of the insect's metamorphosis for their continued existence (CLEVELAND *et al.*, 1934). Before this dependence developed, the symbionts must have had a casual association and, insofar as the digestion of wood is concerned, hydrolysis may have been accomplished outside the body by saprophytes or by means of the secretions of the insect's salivary glands. On the hypothesis that the salivary glands were the original source of cellulase, it would appear that *Cryptocercus* became increasingly dependent on protozoa as it depended more and more on wood for its diet. Thus, the present state of activity of the salivary glands in this insect is probably regressive as a consequence of symbiosis. This tendency may have continued in the more recent but closely related Blattidae, as seen in *B. orientalis*, whose salivary glands are responsible for only a small part of the considerable amount of cellulase produced. It is shown also in the same subfamily by *P. australasiae* but not so markedly by *P. fuliginosa* and less so by *P. americana*. On the other hand, in the further offshoot, the Polyzosteriinae, *Eurycotis floridana* contains much cellulase and has very active salivary glands.

Symbiosis between cockroaches and cellulolytic flagellates or other cellulolytic micro-organisms apparently becomes attenuated or lost as the cockroach becomes less dependent on wood and more omnivorous (GRASSÉ and NOIROT, 1959). A parallel loss of cellulase activity in the insect might be expected but the present data show no consistent pattern of cellulase diminution: *Cryptocercus*, for example, does not have a notably high level of cellulase. This may be due to the diversity of types of cellulolytic micro-organisms available to the insect as commensals or for establishing a symbiotic relationship, and the use which the particular species of cockroach makes of cellulose. Quite possibly, except for *Cryptocercus* which was kept on its natural diet of wood, the data do not represent the status of cellulase as it occurs in the different species in their natural habitats, for the cockroaches were all kept entirely on Laboratory Chow, which may change the normal intestinal microbia and affect cellulase production differently in the different species. Unfortunately, although much is known about the habitat of cockroaches, little is known about their actual diets (ROTH and WILLIS, 1960). Considering the dependence of the two extant xylophagous species of cockroach—on protozoa in the case of *Cryptocercus* and on bacteria in the case of *Panesthia*—one may surmise either that there was more than one origin of cellulolytic activity in the intestines of cockroaches or, if only one, that the symbiosis was tenuous and sensitive to ecological influences. No consistent pattern of evolutionary development was shown.

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