

displacement activities are also at a minimum. However, it appeared that several *U. inversa* individuals, confined to a terrarium too small for their number, became unnaturally aggressive towards one another. Of these, one female was seen to be performing displacement feeding—the one and only case of a displacement activity observed in this particular species. The female concerned appeared to be trapped in a corner, away from her burrow, and completely surrounded by four males. She ran about nervously, trying unsuccessfully to make her escape, and even attempted to climb the vertical sides of the terrarium, falling down again and again. Finally, she began to feed, in a highly 'nervous' fashion, and, since her chelipeds appeared to be scooping up no mud whatsoever, it appears that this must be yet another case of displacement feeding.

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<sup>1</sup> Tinbergen, N., *Quart. Rev. Biol.*, **27**, 1 (1952).

<sup>2</sup> Bastock, M., Morris, D., and Moynihan, M., *Behaviour*, **6**, 66 (1953).

<sup>3</sup> Morris, D., *Behaviour*, **6**, 271 (1954).

### Cytochemical Research on Coelenterate Nematocysts

A NOTE by T. M. Yanagita and T. Wada,<sup>1</sup> suggesting, after C. H. Brown<sup>2</sup>, the possible keratin nature of coelenterate nematocysts has directed my attention to the subject. In 1952 I had observed the pronounced metachromatic colour produced by dilute thionin on large nematocysts from a siphonophore, *Halistemma* sp. It is known that the metachromatic colour of a dye, as taken up by a substance, is characteristic of an acidic mucopolysaccharide content, associated or not, with proteins to form mucoproteins<sup>3</sup>. The fact interested me, since I had studied such substances in the envelopes of pagurid and cephalopod spermatophores<sup>4</sup>.

Another aspect of the nematocyst's contents has been studied by J. Boisseau<sup>5</sup>—the presumably toxic substance enclosed in the enidocyst. He concludes that an albumin content is associated with a phenolic compound; the latter differs according to the type of enidocyst and the species investigated.

In view of these different facts, I undertook to investigate the matter more closely, with the few cytochemical procedures available to me. Two species were chosen: a siphonophore, *Abylopsis tetragona* Ott. (*Halistemma* are not as frequent off the coast of Algiers), and an actinian, *Anemonia sulcata* Penn.

(1) *Abylopsis* has four types of nematocysts localized in the daetylozooid; I shall use Weill's<sup>6</sup> nomenclature to designate them eventually. I ascertained the protein constituent by means of a few colour reactions which are weakly positive (ninhydrin, Sakagushi, Millon, Adamkiewicz). The cystine content was revealed by the lead sulphide reaction. It is positive on every part of all enidocysts, thereby confirming my predecessors' findings. The phenolic group was studied with the azo and chromaffine reactions and others special to *orthodiphenols*, according to procedures given by Lison<sup>7</sup>. I found that all nematocysts contain a small amount of *orthodiphenol*. This may differ according to the parts of the cyst.

The mucopolysaccharide content was analysed by means of techniques summarized by Lison<sup>7</sup>. The different kinds of cysts react positively to the Bauer test. Staining with toluidine blue gives different results: the invaginated (or discharged) threads and fluid content of the enidocysts take a strongly metachromatic hue, implying the presence of a sulpho-mucopolysaccharide compound; the mastigophore capsules alone are to a lesser degree metachromatic too: this would indicate a simple mucopolysaccharide content (as defined by K. Meyer<sup>3</sup>). The other nematocyst capsules are coloured orthochromatically. They can, however, be made to colour metachromatically by treatment with 10 per cent chromic acid. These capsules would thus contain a neutral mucoprotein.

(2) *Anemonia sulcata* has several kinds of nematocysts, localized in different parts of the body. There are, besides, spirocysts in the tentacles, not considered to be true enidocysts (Weill<sup>6</sup>). The protein content includes arginine, specially abundant in spirocysts, on which it confers basic properties; tryptophan is present in small quantity; cystine can be revealed by the lead sulphide and by the sodium nitroprussate reactions—the latter after reducing the S—S linkage in the amino-acid. The result is more strongly positive on the spirocysts than on the basitrichia or other kinds. The Chevrement and Fredericq test could not be of any use in this case, since it is given by other reducing agents, such as diphenols. It is likely the S—S bridges in the protein account for the sensitivity of certain parts to alkaline sodium sulphide, such as was found by Brown<sup>2</sup> and which I confirmed on the basitrichia capsules. The spirocysts are not wholly soluble.

The presence of an *orthodiphenol* has been identified in each type of nematocyst; the amount is very slight, as shown by the weak chromaffine reaction. The spirocysts contain an amino-*orthodiphenol* instead (giving a positive azo-reaction in acidic medium, as well as in alkaline solution).

The polysaccharide content gives a positive Bauer reaction, but as a rule very faint. Staining by toluidine blue is not strongly metachromatic on basitrichia and the contents of atrichia, since colour is due to the carboxylic groups. These nematocysts seem to be made of simple mucoproteins. But the mastigophores and atrichia capsules are not coloured; neither are the spirocysts, which, being acidophilic, do not take basic dyes. The two former can, however, be made to stain metachromatically after chromic oxidation. This would indicate that they are made up of neutral mucoproteins. The spirocysts remain uncoloured in these conditions; since they are slightly Bauer-positive, I think this indicates a low polysaccharide content linked to a basic protein. I would suggest it is a glycoprotein, as defined by Meyer<sup>3</sup>, on account of its low carbohydrate content.

A more detailed account will be published elsewhere.

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<sup>1</sup> Yanagita, T. M., and Wada, T., *Nature*, **173**, 171 (1954).

<sup>2</sup> Brown, C. H., *Nature*, **166**, 439 (1950).

<sup>3</sup> Meyer, K., "Advances in Protein Chemistry", **2**, 249 (1945).

<sup>4</sup> Hamon, M., "Recherches sur les spermatophores" (Thèse, Alger, 1942).

<sup>5</sup> Boisseau, J. P., *Bull. Soc. Zool. Fr.*, **77**, 151 (1952).

<sup>6</sup> Weill, R., *Trav. Sta. Zool. Wimereux*, X and XI, 1-701 (Thèse, Paris, 1934).

<sup>7</sup> Lison, L., "Histochimie et Cytochimie animales" (Paris, 1953).