ploratory walking to the quick searching behaviour, but produced no song within 5 min.

The numbers above the bars in Figs 1, 2 and 3 are the numbers of males tested with each stimulus; the height of the bars shows the proportion of males that sang within 5 min. For *A. fasciatus*, *A. allardi*, *A. tinnulus* and *P. ambitiosus*, the proportion of males that sang in response to paper conditioned by conspecific females was significantly greater than the proportion that sang on the control paper ($P < 0.05$ in 2×2 G tests of independence[1]; Figs 1 and 2). In the absence of visual, acoustic, or tactile cues, males of the four species sang in response to some factor, presumably chemical stimuli left on the substrate by females. None of the *Eunemobius carolinus* and *A. sparsalsus* males gave a positive response in the presence of conspecific female paper.

A. fasciatus, *A. allardi*, and *A. tinnulus* males showed a significantly greater response to conspecific female paper than to paper conditioned by conspecific males (Fig. 3). Furthermore, *A. fasciatus* males showed a greater response to paper conditioned by adult females than to paper conditioned by *A. fasciatus* juveniles (Fig. 3). These results suggest a sexual role for chemical communication in these species. Ground crickets of these genera produce quiet, high pitched songs relative to those of field crickets (subfamily Gryllinae), or even other nemobiine genera (for example, *Eunemobius*)[2]. The softer, higher pitched songs are not likely to carry far to attract females at a distance. Perhaps the song-releasing stimuli left on the substrate by females are pheromones which indicate areas where singing males are most likely to attract females. The stimuli may be important in species isolation; I have experimental evidence of specificity at the species level[2].

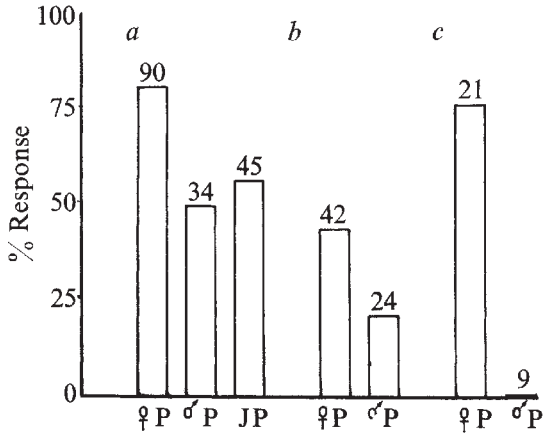

Fig. 3 Proportions of *A. fasciatus* (*a*), *A. allardi* (*b*) and *A. tinnulus* males (*c*) that sang in tests with female-conditioned paper (♀P) male-conditioned paper (♂P); and juvenile-conditioned paper (JP) for *A. fasciatus*. For each species a significantly greater proportion sang on female-conditioned paper than on male-conditioned paper. A greater proportion of *A. fasciatus* males sang on female-conditioned paper than on juvenile-conditioned paper. Numbers above bars refer to number of males tested.

Previous studies have described pheromone-like communication in crickets[3,4] and chemically-induced stridulation has been reported for the Douglas fir beetle[5]. The data reported here, however, demonstrate song-releasing chemical stimuli in insects well known for their acoustic communication. Further studies may reveal something about the evolutionary origins of cricket song. In some cockroach species (for example, *Blattella germanica*) males raise their wings and expose gustatorial glands (which females feed on) in response to antennal contact with a female pheromone[6,7]. This is of interest here since crickets are believed to have a cockroach-like ancestor[8].

At present, the origin and nature of the song-releasing stimuli are unknown. Initial attempts to isolate stimuli from the faecal pellets of females have resulted in ambiguous bioassays.

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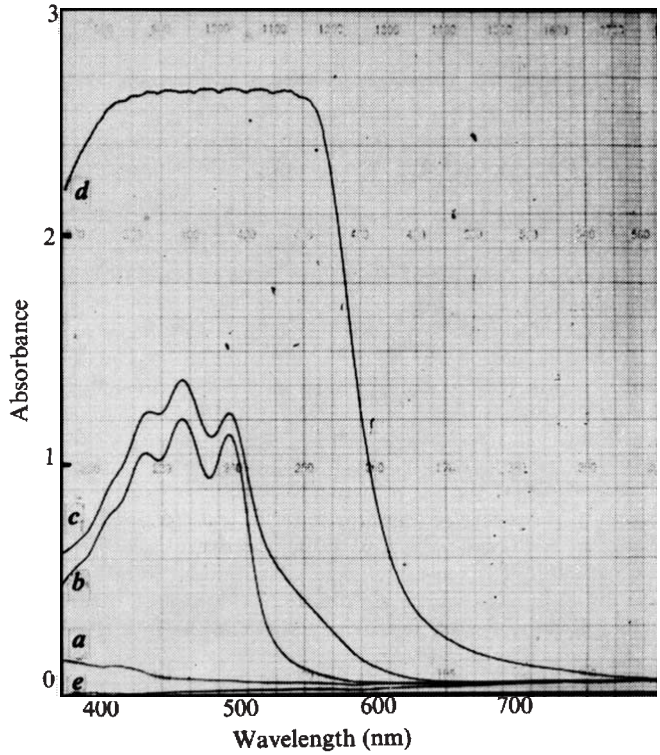
Changeable coloration of cornea in the fish *Hexagrammos octogrammus*

ALTHOUGH fish are considered 'lower' vertebrates, the visual apparatus of some species has a high degree of adaptability unparallelled in other vertebrates. For example, members of the Hexagrammidae can change their cornea rapidly from colourless in the dark, to deep red in bright light[1,2]. We have studied this ability in *Hexagrammos octogrammus* Pallas, a common shallow-water fish of the Japan Sea, and found that it is caused by the effects of illumination on the distribution of coloured cytoplasm in the corneal chromatophores.

Specimens of *H. octogrammus* caught by night or kept in the dark have a cornea as transparent and colourless as that of most fish. But specimens collected in their natural environment by day, or kept in an illuminated aquarium, have a bright orange or deep red cornea. If a light-adapted fish is placed in the dark, its cornea gradually pales, becom-

Fig. 1 Absorption spectra measured in pupil zone of corneas from *H. octogrammus* in different adaptation states: *a*, in full darkness; *c*, intermediate (yellow) coloration observed after about 30 min in sunlight after full dark adaptation; *d*, full colortion after 2 h in sunlight; *b*, yellow cornea of *Pleurogrammus monopterigius* in full colour; *e*, control.



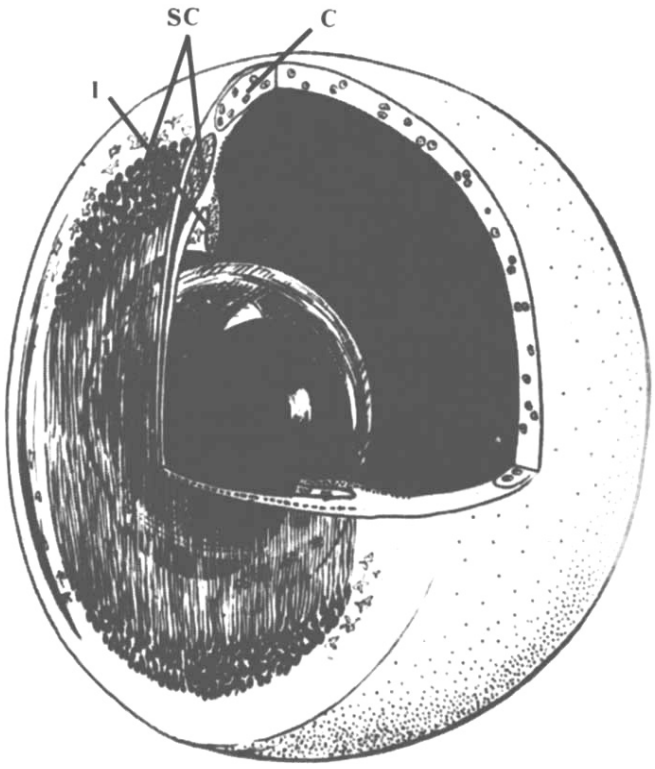


Fig. 2 Reconstruction of anterior part of the eye in *H. octogrammus*. SC, Chromatophores; C, chondreal sclera; I, iris.

ing yellow and then colourless. Similarly, in bright illumination, a colourless cornea becomes yellow and then red, even at night. The speed of colour change depends on the intensity of illumination, and when a fish is transferred from direct sunlight to darkness, complete decoloration usually takes 1.5–2 h. If the fish is kept continuously in light intensities of about 10 lx, its cornea becomes yellow without reddening.

The cornea of *H. octogrammus* has surprising absorptive properties as a colour filter. Its transmittance in the pupil zone in the light-adapted state does not exceed 0.3% for wavelengths of 400–500 nm. It absorbs more than 10% of the light up to a wavelength of 580 nm (Fig. 1). The most saturated coloration reported so far for a fish cornea has been that of the South American *Astronotus* (Cichlidae)[3]—yet in this species the minimum transmittance is not less than 4%, absorption above 10% being observed only for wavelengths shorter than 510 nm.

This changing corneal coloration can be compared with retinomotor and pupil reactions, which regulate the quantity of light reaching the receptor layer. The most likely function of the yellow and orange coloration is to improve the resolution of the eye by eliminating harmful loss of contrast of retinal image due to (1) a blue veil of light scattered on intraocular and extraocular media, and (2) chromatic aberration[4], which can be quite significant in the fish eye[5]. Thus shallow-water fishes like *H. octogrammus* searching for food on a well-lit substratum can benefit from a coloured cornea in spite of the loss of sensitivity because it is only at low light intensities that preretinal filters become a disadvantage. The exceptional saturation of the filters in *H. octogrammus* is tolerated because the fish can, so to speak, remove its sunglasses when it gets dark.

In most animals there is diffuse coloration of some part of the eye—in tree shrew, ground squirrels and diurnal snake the whole lens is yellow[4], in others, as in some squid[6].

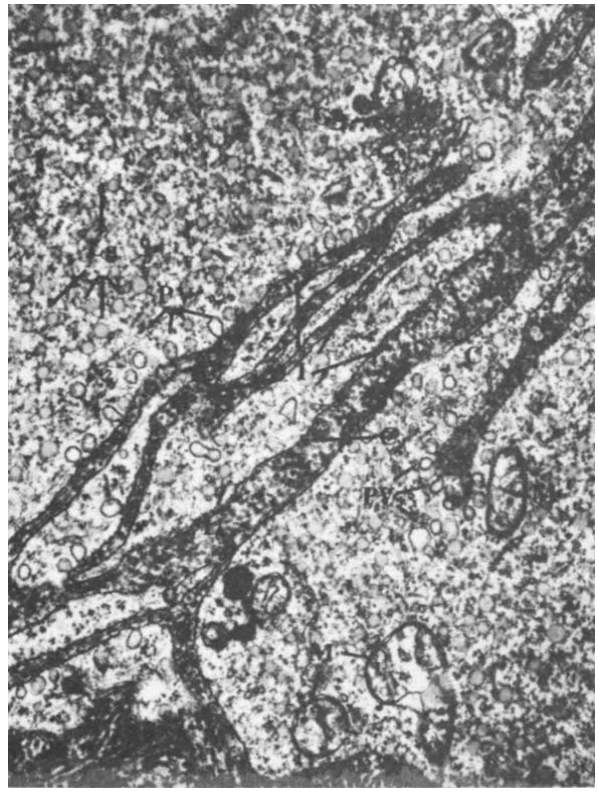
by specialised corneal chromatophores, which differ considerably in structure from ordinary dermal chromatophores.

Chromatophore cell bodies comprise two compact sickle-shaped masses near the upper and lower edges of the cornea, outside the pupil zone. In a vertical section of the cornea, cell masses are surrounded by fibrous tissue. Each chromatophore (50–70 μm in diameter) is pear shaped and gradually tapers into a single, flattened process. All processes are oriented vertically (in the eye of a living fish) and extend downwards or upwards, according to the position of the cells in the upper or lower cell mass, and overlap the pupil zone (Fig. 2).

Corneal coloration is altered by redistribution of coloured cytoplasm between cell bodies (Fig. 3) and their processes. Direct observation of the living fish under a dissecting microscope showed that in fish kept in bright light the processes are filled with cytoplasm, but in the dark they become empty and invisible.

There are two kinds of corneal chromatophore—deep red and yellow—resembling dermal erythrophores and xanthophores in their coloration. When a dark-adapted fish is exposed to bright light, the processes of the yellow chromatophores fill up first, giving a transient yellow colour to the cornea (Fig. 1*c*). The corneas of *Pleurogrammus monopterigius* and *H. stelleri* (both Hexagrammidae) have only yellow chromatophores, the colour of their corneas in the light-adapted state being similar to the intermediate coloration of that of *H. octogrammus* and the permanent colour of the cornea of pike and perch[7]. The absorption bands at 427, 455 and 485 nm reveal the carotenoid nature of the yellow pigmentation. The coloration of red cornea of *H. octogrammus* in light-adapted fish corresponds to absorption in the red corneal chromatophores, their absorption spectrum consisting of a single broad band (the half-band about 400–550 nm) with a single flattened maximum at

Fig. 3 Ultrastructure of the chromatophore from *H. octogrammus* cornea. Numerous lipid granules (LG), arborised invaginations of cell membrane (I), collagen fibres (C), pinocytosis vesicles (PV), mitochondria (M) and Golgi apparatus (GA) are visible. (× 29,750).



about 480 nm. The difference between the absorption spectra of the two types of corneal chromatophore, shown by microspectrophotometry, cannot be explained as resulting from differences in concentration of the same coloured substance.

The corneal chromatophores differ from other (dermal) chromatophores in having only one process, and having, in the cytoplasm of the cell body and process, homogeneous round granules of intermediate electron density (100–120 nm in diameter), which might be lipid droplets in which carotenoids are dissolved (Fig. 3). Both cell bodies and processes contain many microtubules (25 nm in diameter), lining the axis. No lipid granules have been found in the processes of dark-adapted fish, although microtubules and numerous pinocytosis vesicles are sometimes present. There are many invaginations of the cell membrane, up to 100 nm wide and several μm long, with pinocytosis vesicles attached to them. Unlike ordinary dermal chromatophores—erythrophores and xanthophores—the corneal chromatophores contain no pterinosomes[8].

Microtubules are directly concerned with dispersion and aggregation of pigment granules in melanophores[9,10], and antimitotic agents (colchicine and vinblastine) which destroy the normal function of microtubules may block the movement of pigment granules[11]. After injection of colchicine into the ocular bulb of *H. octogrammus* the response of cornea to a change of illumination is slower and weaker.

Changes of the corneal coloration caused by specific corneal chromatophores are not restricted to members of the Hexagrammidae. They have been observed, but to a lesser degree in several species of other families, such as the Blennidae, Cottidae and Tetraodontidae.

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Aversive behaviour of crown-of-thorns starfish to coral evoked by food-related chemicals

When *Acanthaster planci*, the coral-predating crown-of-thorns starfish, encounters living coral or when it is presented with coral extracts, it rears the arms near to the stimulus source aborally and retracts the tube feet of these arms into the ambulacral groove[1,2]. This aversive behaviour has generally been attributed to the effect of discharge of coral nematocysts[1–3], or to toxins released from nematocysts[4]. But arm rearing may be evoked before contact is made with corals, and both "withdrawal" responses are produced by non-coral food or food extracts which stimulate *A. planci* to feed (R.J.M., unpublished). This suggests that it is not nematocysts or their toxins which are responsible, but rather chemicals from coral tissue. The experiments described here show that this is the case: the function of the aversive response seems to be protection of the starfish against nematocyst discharge on contact with coral.

The role of nematocyst toxins was investigated by comparison of the effects on *A. planci* of extracts of whole coral tissue and homogenised isolated nematocysts from three coral species, including *Millepora dichotoma* ('fire coral'), which is only rarely attacked by *A. planci*. To validate comparisons between the extracts of each species of coral, and show up any differences in activity, it was necessary to equalise concentrations of non-stimulatory components of the extracts. Collins[5] found that only low molecular-weight components of coral extract, apparently amino acids, elicited arm withdrawal. It was assumed that soluble protein represented the bulk of non-stimulatory material in both nematocysts and tissue cells, and so the protein concentrations of the extracts were equalised—by dilution of the more concentrated extracts—after estimation by the method of Lowry *et al.*[6].

Table 1 summarises the effects of dripping extracts near the tips of arms of moving starfish. It shows that the homogenised nematocyst preparation of only one coral, *Millepora dichotoma*, evoked stronger responses than those produced by the corresponding whole coral extract, and even then the responses evoked were weaker than those produced by the whole tissue extract of *Acropora multicaulis*, in spite of the far greater protein concentration of the *Millepora* extract.

Moreover, the high efficiency of isolation of nematocysts of *Millepora dichotoma* (Fig. 1) made possible an estimate of the proportion of total tissue volume in intact *Millepora* occupied by nematocysts: it was approximately 2%. Thus the contribution made by nematocyst contents to the arm-rearing and tube-foot retraction activity of a whole coral extract seems to be negligible.

We even found that intact isolated nematocysts had no observable effect on the tube feet of *A. planci*—yet accidental splashes of nematocyst suspensions on to our skin produced typical stinging sensations.

These unexpected results suggested that nematocyst contents had leaked into the whole coral extracts during preparation, although most nematocysts remain undischarged. This objection was negated by testing extracts of mesenterial filaments, which contain greater concentrations of nematocysts than other coral tissues (Fig. 2) and may be removed intact from corals with large polyps. Even so, as Table 1 shows, the responses elicited by the mesenterial

Fig. 1 Photomicrograph of nematocysts isolated from 'fire coral', *Millepora dichotoma*, and used in the preparation of nematocyst extracts. The darker objects are fragments of coral skeleton. For further explanation, see text.