endocarp is present but not sclerified (produced with p-chlorophenoxyacetic acid); and (c) syconia containing achenes in which the endocarp is present and completely sclerified (produced with benzothiazol-2-oxyacetic acid). Achenes in each of these types do not contain an embryo.

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Protective Effect of Glycine on Sperm Exposed to Light

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Spermatozoa of the purple sea urchin, *Strongylocentrotus purpuratus*,[2] were found to be about a million times as sensitive as eggs to ultraviolet light (*1*) at wavelengths absorbed by nucleoproteins. Spermatozoa of a number of other sea urchins, *S. franciscanus, Arbacia punctulata*, the sand dollar *Dendraster eccentricus*, and the starfish *Pateria miniata* were also found to be highly sensitive to these radiations (*2*). The injurious effect of ultraviolet light on *S. purpuratus* is partially reversed by small dosages of monochromatic visible light, especially at 4350 A (*3*). However, visible light also has an injurious effect on spermatozoa, immotilizing them and rendering them incapable of fertilizing the eggs. Dosages that do not immotilize the spermatozoa cause a slight delay in cleavage (*3*). The recent discovery by Tyler (*4–6*) that glycine protects spermatozoa against deterioration after dilution of the spawn suggested the possibility of protecting the spermatozoa against radiations by this means.

Accordingly, in all the following experiments on this species, the spawn issuing from a testis of *S. purpuratus* was diluted 1:400 in sea water solution containing 0.05*M* glycine (this being the concentration found optimal by Tyler), a similar suspension in sea water alone serving as control. A drop (about 0.01 ml) was withdrawn from each vessel at various periods and tested by adding to a dish containing about 300 eggs in 0.3 ml sea water, which was stirred by gently blowing with a pipette. Although there is variation in the longevity of sperm from different animals, untreated but diluted spermatozoa kept at 13° C in the dark lose their viability in 1–3 hr. In diffuse daylight (100–200 ft-c) and at a higher temperature (21° C), they remain virile less than an hour. In the presence of glycine, however, they remained virile for more than 20 hr (the longest time tested) in the dark at 13° C. Spermatozoa of the purple sea urchin are therefore protected by glycine in the same manner as those of the other species tested by Tyler and coworkers.

Glycine, however, offers no protection against injury by short ultraviolet radiations. Spermatozoa without as well as with glycine, irradiated with 50 ergs/mm^2 of λ 2654A from a quartz monochromator, were used to fertilize normal eggs. The resulting zygotes in both cases showed an 80-min delay in the first cleavage over unirradiated controls (for methods see [*3*]). Nor did the presence of glycine enhance photoreactivation by visible light of spermatozoa injured by ultraviolet radiations, the cleavage delay being the same in experimental and control.

On the other hand, glycine had a strong protective effect on spermatozoa treated with visible light. In most of these experiments the source of radiations was a GE CH-4 spotlamp (at 30 cm), and the light was filtered throug 8 in. of water and a #3060 Corning glass filter, which cuts off most of the long ultraviolet (λ 3660A and possibly somewhat shorter) but transmits most of the visible (30% at 4000A). The intensity, measured by a GE exposure meter, was about 3900 ft-c. Spermatozoa in sea water exposed to this light lost their viability in 5–10 min. In glycine solution they fail to fertilize eggs only after exposures of about an hour or more. Addition of glycine to spermatozoa that have been immotilized with white light does not revive them, whether the suspension had been treated in sea water or in glycine sea water. Treated with sunlight (1800 ft-c) through water cells to remove heat and the Corning #3060 filter to remove the ultraviolet radiations, the spermatozoa were killed in 10 min in sea water, but those in glycine sea water were still fully viable after 33 min.

Of the wavelengths tested, the visible light that is most effective in immotilizing spermatozoa of the purple sea urchin is the blue, λ 4350A; a dosage of 20,000–40,000 ergs/mm^2 affects almost 100% of the spermatozoa, as judged both by immotilization and by the failure to induce fertilization membranes or cleavage (the latter check being necessary since occasionally one finds membranes so close to the egg that fertilization might be questioned, yet cleavage occurs). Violet light, λ 4050A, is less effective than blue, immotilizing only after dosages of 60,000–70,000 ergs/mm^2. Wavelength 3660A in the ultraviolet was not effective even at dosages of 80,000 ergs/mm^2, the highest tried. Monochromatic yellow (5780A) was also ineffective even at a dosage of 80,000 ergs/mm^2. The compound responsible for the injury therefore seems to be something which absorbs strongly in the blue-violet portion of the visible.

The eggs of the purple sea urchin, although quite resistant to visible light filtered through the Corning #3060 filter, are also sensitive to the total radiations

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[2] Collected at Moss Beach, Calif., for the work at Stanford and off the rocks near the laboratory for the work at the Hopkins Marine Station.

of the CH-4 arc (intensity: 5000 ft-c) filtered only through the water cells to remove heat. After an exposure of 45 min fertilization by normal sperm was not observed. Eggs similarly exposed but in 0.05 glycine remained fertilizable, as indicated by fertilization membrane formation or cleavage for at least double this time. Some became sticky in the glycine solution and adhered to the slide.

To extend the observations to spawn of a related form, the sand dollar *D. eccentricus*,[3] and of the echiuroid worm *Urechis caupo*,[4] were tested in a similar manner. The spawn of the sand dollar, diluted 1:200, resisted prolonged exposure (over an hour) to visible light from the CH-4 lamp filtered through the water cells and the #3060 Corning filter; therefore it was tested without the latter filter. In the absence of glycine a deterioration of spermatozoa occurred only after 30 min and was not marked until 40 min, after which exposure very few normal eggs were fertilized by addition of such spermatozoa. In the presence of glycine, however, comparable injury did not occur until after about 60 min exposure. There was little evidence of any protective action when a 1:2000 suspension of sand dollar sperm was tried, suggesting also a mass effect, which was not further investigated. The eggs were also relatively insensitive to the radiations from this lamp and fertilized normally even after more than an hour of exposure. Only after 2 hr exposure was a slight delay in cleavage observed. Clearly, the sand dollar spermatozoa and eggs are much less sensitive to visible light (and long ultraviolet light of this light source) than those of the sea urchin, although the two animals are not too distantly related.

Spermatozoa of *U. caupo*, in a 1:400 suspension proved to be highly resistant to visible light (3900 ft-c) of the CH-4 lamp passed through the Corning #3060 filter, as they are to ultraviolet light (*7*). After 40 min exposure, 95% (equivalent to the control) of the unexposed eggs added to the exposed sperm suspension were fertilized. After 60 min exposure a decline in fertilizing power of the spermatozoa occurred, only about two thirds of the eggs added being fertilized. After 80 min exposure still fewer were fertilized, and after an exposure of 145 min only about a third were fertilized. In all cases, observations were made for cleavage and formation of free-swimming larvae, to make sure that those eggs which failed to show membranes did not later cleave. *Urechis* spermatozoa in glycine sea water showed no significantly greater resistance to visible light than those in plain sea water.

The spermatozoa of *Urechis* were next treated to the full radiations of the CH-4 arc passed only through the water filters to remove heat. They proved much less resistant to the total radiations, which include some long wavelength ultraviolet light, than to visible light alone. After 10 min all eggs added to exposed sperm were fertilized, but after 15 min exposure only about half of the eggs added to the exposed sperm were fertilized, and after 20–30 min none were fertilized or cleaved. Where fertilization occurred, trochophores appeared in a day (18° C).

Eggs of *Urechis* were also treated to the full radiations of the CH-4 arc (minus heat), and they proved quite resistant. After 60 min exposure to this lamp, they still cleaved normally when fertilized with normal sperm and gave rise to normal trochophores.

The experiments indicate that the spermatozoa of the purple sea urchin are much more sensitive to visible light injury than those of the sand dollar or the worm *Urechis*. The relatively greater effectiveness of the blue region of the spectrum in injuring sperm of the purple sea urchin suggests the presence in the sperm of some photosensitizer that has a maximal absorption in the blue. The mechanism of the sensitization of these spermatozoa to visible light should be susceptible to experimental analysis.

[3] Dredged in Monterey Bay between the Hopkins Marine Station and Fort Ord, Calif.

[4] Collected at Elkhorn Slough near Moss Landing, Calif.

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Enzymatic Synthesis of Higher Carbohydrates from Dextrose

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By means of alcoholic fermentation, Pigman (*1*) observed the production of unfermentable carbohydrates by action of mold enzymes on maltose and reported no such synthesis from dextrose. Pan and associates (*2*) reported on the synthesizing action of *Aspergillus niger* enzymes on maltose, but the enzyme system failed to synthesize from dextrose. Tsuchiya *et al.* (*3*) have demonstrated by chromatographic techniques that *A. niger* enzymes synthesize oligosaccharides from maltose, isomaltose, and cellobiose, but direct synthesis from dextrose was not demonstrated.

TABLE 1

Progress in Synthesis of Dextrose

Hours of incubation	Relative reducing value of mixture (%)
0	100
69½	93.4
142	89.4
167	88.5
191	88.3
214	87.7
238	87.4