

into shreds and shaken with calcium augmented serum did not take up any calcium or phosphate whatsoever.

Another reason for believing that it is a colloidal form of calcium and phosphate which is impregnated on the membrane and not calcium and phosphate ion as such is given by the experimental findings with magnesium. When the magnesium in the blood is augmented by injection there is no such loss on the filtration membrane as is found with calcium. A typical experiment is given in Table III. The agreement between the serum magnesium and the

TABLE III.  
Effect of MgSO<sub>4</sub> Injection on Magnesium Diffusibility.  
Dog Fl. Wt. 14 kilos. Injected subcutaneously with 10 cc. of 20% MgSO<sub>4</sub>.

Time, hr.	Total Mg.	Diffusible Mg.	Total Mg. calculated
Control	2.15	1.90	2.15
0.25	5.10	4.10	4.50
1.3	5.25	4.05	5.20
4.5	2.35	2.05	2.20

values calculated from the ultrafiltrate and concentrate magnesium here is gratifying. Because of the close chemical relationship of calcium and magnesium, if adsorption of the cations were involved, both elements would be expected to show a similar behavior.

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Effect of Infra-Red Light on Subsequent Fertilization of the Eggs of Certain Marine Invertebrates.\*

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A study has been made of the effect of near infra-red radiation on the subsequent fertilization of the eggs of some marine invertebrates. The species studied were 2 sea urchins (*Strongylocentrotus purpuratus* and *S. franciscanus*) and a worm (*Urechis caupo*). Light from a Mazda standard projection lamp (gas filled, 30 volts, 30 amperes, with the tungsten filament radiating at about 3200°K) was passed through a slit 1x18 mm., 10 cm. from the filament, and

\* This work was founded upon and includes a portion of the work done by one of us (S.C.B.) with the assistance of M. H. Simmers under grants from the Committee on the Effects of Radiation on Living Organisms (National Research Council).

through a monochromator consisting of 2 glass collimating lenses having a maximum thickness of 6 mm. each, and two 60° quartz prisms. An adjustable outlet slit was so arranged as to pass that portion of the spectrum lying within the approximate limits 0.8-1.2 $\mu$  as determined by calculation from the known dispersion. The wave length of maximum energy was determined by the use of a small vacuum thermocouple and found to lie at about 0.8-0.9 $\mu$ , as would be expected from the nature of the source. Rough calculation suggests that the energy flux in this region was probably of the order of magnitude of 0.1 cal. cm.<sup>-2</sup> min.<sup>-1</sup>

Unfertilized eggs in a single layer were irradiated for periods of 15 to 45 minutes in special thin glass cells, controls in similar cells being placed 2 cm. away on the top of the monochromator. The experiments were done in darkness, the source being in a separate room, so that the controls received only that small amount of infra-red and scattered visible light which passed through the monochromator and was reflected by the walls of the dark room. After irradiation the eggs were fertilized and further development watched under ordinary artificial illumination. For purposes of this article the term fertilization is used to designate the formation of fertilization membranes. The eggs of *Urechis* showed quantitatively identical effects upon this process and upon the disappearance of the germinal vesicle and subsequent cleavage.

In all cases the proportion of eggs fertilized decreased. The table shows the results of the experiments under the following column heads in order: Species; experiment number; duration of irradiation;  $n_2$  and  $n_1$ , the total numbers of eggs in the control and irra-

TABLE I.  
The effects of infra-red light on subsequent fertilization of eggs.  
See text for details.

Species	Exp. No.	Irradiation min.	$n_2$	$n_1$	$p_2$	$p_1$	$p_2 - p_1$	$\epsilon_{p_2 - p_1}$	$\frac{p_2 - p_1}{p_2} \cdot 100$	$\frac{p_2 - p_1}{\epsilon_{p_2 - p_1}}$
<i>Strongylocentrotus purpuratus</i>	I	30	1341	1263	.541	.321	-.221	$\pm .019$	-40.7	11.6
"	II	30	1439	1414	.051	.011	-.040	$\pm .020$	-78.4	2.0
"	III	30	951	923	.358	.046	-.312	$\pm .053$	-87.1	5.9
"	IV	45	1123	1292	.300	.089	-.211	$\pm .016$	-70.3	13.2
"	V	45	1265	928	.596	.186	-.410	$\pm .019$	-68.7	21.6
<i>S. franciscanus</i>	VI	30	461	570	.269	.219	-.050	$\pm .027$	-18.5	1.8
<i>Urechis caupo</i>	VII	15	509	628	.203	.081	-.122	$\pm .015$	-60.0	8.1
"	VIII	30	1376	1211	.307	.190	-.117	$\pm .016$	-36.3	7.3
"	IX	45	279	298	.570	.416	-.154	$\pm .041$	-27.0	3.8

diated samples;  $p_2$  and  $p_1$ , the corresponding fractions successfully fertilized;  $p_2 - p_1$ , the decrease in the fraction fertilized;  $\epsilon_{p_2 - p_1}$ , the standard error of this difference for simple samples;  $\frac{(p_2 - p_1)}{p_2} \cdot 100$ , the per cent decrease in successful fertilization, and  $\frac{p_2 - p_1}{\epsilon_{p_2 - p_1}}$ , the ratio of the decrease to its standard error. The last two columns, therefore, show the magnitude of the effect and its statistical significance. Since all the experiments agree in showing a decrease in fertilization, they may all be considered significant; either experiments II or VI alone would not be significant. In the first case unavoidable ageing of sperm and eggs before use greatly lowered the percentage fertilized and probably accounts for the poor result. Exp. VI represents our only experiment with *S. franciscanus*, and in it relatively few eggs were used. The relatively inconclusive result in this case may be due to either chance error of small samples, or more resistant eggs. The former seems more probable.

The decrease in fertilization is probably not due to heating; thermocouples showed at no time during irradiation a rise of more than  $0.04^\circ\text{C}$ . in the temperature of the sea water in which the eggs were lying. Previous studies by Simmers, using a Pt-Fe microthermocouple as described by Whitaker,<sup>1</sup> have shown that the temperature within these eggs does not during irradiation rise above that of the surrounding medium enough to affect a measuring system sensitive to  $0.02^\circ\text{C}$ . The total temperature difference between control and irradiated eggs was certainly not as much as  $0.06^\circ\text{C}$ .

Even this slight rise in temperature might be suspected to coincide with some evaporation of sea water from the irradiated sample. But since repeated renewal of this sea water during irradiation did not affect the outcome, evaporation was not a factor.

There appears to remain only one possibility: that infra-red light of wave length  $0.8\text{-}1.2\mu$  affects photochemically some constituent of these eggs and thus inhibits subsequent fertilization. We know of no previous experiments in which biological effects attributed to infra-red light may not have been produced indirectly as a result of heating. Unless some factor which we have not considered is operative, our results are, therefore, the first demonstration of a photochemical effect of infra-red light on living cells.

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<sup>1</sup> Whitaker, D. M., *Science*, 1929, 70, 263.