presence of biological compounds with phosphate bonds of higher energy than glycerophosphate (*2*).

It has since been established that this acceleration is achieved in the absence of nucleotides by a direct phosphate transfer, that is, without passing through the intermediary stage of inorganic phosphate. When a mixture of P^{32}-labeled phosphocreatine, synthesized enzymatically (*3*), unlabeled inorganic phosphate, and glycerol was incubated with intestinal phosphatase at 38° C for 15 and 30 min, the glycerophosphate in excess of that synthesized in the absence of phosphocreatine (representing two-thirds to three-fourths of the total glycerophosphate) had about the same specific activity as the phosphocreatine, while the specific activity of the inorganic phosphate was quite low. If the phosphate transfer had passed through the intermediary stage of inorganic phosphate, then the specific activity of the glycerophosphate synthesized could not be higher than that of the inorganic phosphate. Similar results were obtained with radioactive phosphocreatine, fructose, and inorganic phosphate: at the end of 15 min of incubation more than half of the total fructose-phosphate formed derived its phosphorus directly from the phosphocreatine. Other observations indicate that phosphopyruvate glucose-1-phosphate (and similar compounds with relatively high phosphate bond energy) can also participate as phosphate donors in such a direct phosphate transfer.

TABLE 1

SPECIFIC ACTIVITIES OF THE PHOSPHATE SPECIES IN THE PHOSPHORYLATION OF GLYCEROL AND FRUCTOSE AT 38° C WITH PHOSPHOCREATINE LABELED WITH P^{32}

	Initial molar concentration	% Acceleration of synthesis	Specific activity cpm/γ P	
A			0 min	30 min.
Phosphocreatine	0.0252		1090	895
Inorganic P	0.448		0	9.3
Glycerol	1.63		–	–
Glycerophosphate	0.0	290	0	606*
B			0 min	15 min
Phosphocreatine	0.030		1828	1750
Inorganic P	0.444		0	13.3
Fructose	2.34		–	–
Fructose-phosphate	0.0	375	0	730*

* Corrected for the nonlabile P impurities present in the phosphocreatine preparation.

In Table 1 the specific activities (cpm/γ P) of the different phosphate species from two such experiments are given. The general composition of the enzymatic incubation mixture was the same as previously reported (*2*).

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Hatching Eggs of Floodwater Mosquitoes in Media that Promote Plant Growth[1]

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In a natural environment, eggs of floodwater mosquitoes are deposited on soil, in debris, or among plants in places subject to transient submergence. In such sites they remain in a viable state for a year or longer, and usually hatch when they are flooded with water at a proper temperature. However, submergence in this manner is not a dependable provocation for hatching eggs of these mosquitoes in the laboratory, according to numerous observers. Water in natural habitats must contain stimulants to hatching that are not found in tap water. Several observers have noted that durable eggs of the genus *Aedes* may be stimulated to hatch by infusions made from plants that grow in the natural oviposition sites and by cultures of bacteria and yeasts. Abdel-Malek (*1*) found that eggs of *Aedes trivittatus* Coq. hatched erratically in dilute solutions of chemicals that regulate plant growth such as 1-naphthaleneacetic, 3-indoleacetic, and 3-indolebutyric acids. No one seems to have devised a way to get eggs to hatch in consistently high percentages in a few hours, as occurs in nature.

Larvae of floodwater mosquitoes hatch after two phases of growth have been completed in the egg. The first is an increase in number of cells, which continues until the fully formed embryo occupies all of the interior of the egg. The second is an increase in size of the embryo, until the shell of the egg is ruptured. Between the two periods, eggs may be dormant for months. Temperature regulates the rate of increase of cells, and the nature of the solution surrounding the eggs has much to do with initiating escape of the larvae. The final act of hatching involves an increase in size of the embryo in a manner similar to the elongation phase of growth of plant tissue. Media that stimulate one might affect the other similarly.

Eggs of the floodwater species, *Psorophora discolor* (Coq.) may hatch at any time after the embryos are fully formed. Maturation of the embryo requires about 4 days at a temperature between 22 and 26° C. If kept on a moist surface at a temperature within this range, larvae may hatch whenever the eggs are submerged in a suitable medium. Eggs kept on a moist substratum at a temperature of 15–20° C in the laboratory have survived and yielded vigorous larvae after at least 9 months. However, prolonged exposure of eggs to temperatures as low as 15–20° C will prevent any medium from causing hatching until the eggs are conditioned for several days at a temperature favorable for hatching.

Substances that promote growth of plant tissues vary in effectiveness. Thimann (*3*) states that short sections of etiolated oat coleoptiles will elongate slightly in water, more in water containing purified growth-promoting sub-

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stances, and vastly more when sugar is added to the medium. He further states that a suitable medium for the purpose of growing plant tissue contains sucrose 1%, potassium chloride M/100, and indoleacetic acid 1–5 mg/l. Caplan and Steward (*2*) have shown that dilutions of milk from ripe coconuts, and corn lyophilized while in the milk stage contain factors that stimulate growth of carrot explants many times more than purified indoleacetic acid alone. The purpose of this paper is to show the order of increasing influence of various types of media on the hatching of eggs of a common species of floodwater mosquito, *Psorophora discolor*.

TABLE 1

INFLUENCE OF VARIOUS MEDIA ON THE HATCHING OF LARVAE FROM EGGS OF *Psorophora discolor* IN THE LABORATORY

Medium	Age of medium	Eggs treated	Larvae hatched					
			0–4 hr		5–24 hr		Total	
	days	no.	no.	%	no.	%	no.	%
Extract of canned corn	0	120	93	77	5	4	98	82
Coconut milk (5%)	0	330	243	74	21	7	264	80
Glucose, KCl, and indoleacetic acid	2–7	180	137	76	2	−1	139	77
Glucose, KCl, and acetic acid	3–5	240	141	59	21	9	162	68
Glucose, and acetic acid	2–3	180	107	60	11	6	118	66
Glucose, and ethyl alcohol	3	260	49	19	102	39	151	58
Sucrose, KCl, and indoleacetic acid	0	20	0	0	98	81	98	81
Glucose, KCl, and acetic acid	0	180	0	0	127	71	127	71
Glucose, KCl, and indoleacetic acid	0	180	0	0	116	64	116	64
Glucose, and acetic acid	0	90	0	0	52	58	52	58
Glucose, and KCl	0	90	0	0	53	59	53	59
Glucose	0	180	0	0	102	57	102	57
Indoleacetic acid (5–10 ppm)	0	60	0	0	26	43	26	43
Indolepropionic acid (2–10 ppm)	0	70	2	3	24	34	26	37
Indolebutyric acid (2–10 ppm)	0	70	0	0	25	36	0	36
Yeast suspension	0	40	0	0	5	12	5	12
Indoleacetic acid (0.1–2.0 ppm)	0	250	1	−1	13	5	14	6
Acetic acid (20–100 ppm)	0	300	6	2	4	1	10	3
Indoleacetic acid (20 ppm)	0	30	3	10	0	0	3	10
Tap water	0	340	4	1	2	−1	6	2
Potassium chloride	0	30	0	0	0	0	0	0

Preparations for observing the effects of various media on the rate of hatching of eggs were carried out in the following manner. Solutions were prepared by dissolving the ingredients in tap water without sterilization. Some were used as soon as they were prepared, and others were aged at 20° C for periods of time shown in Table 1. All solutions of glucose and sucrose contained 10,000 ppm of the sugar. Potassium chloride was used at the concentration of 7460 ppm. Indoleacetic acid, in all solutions containing sugar, was used at a concentration of 5 ppm; otherwise dilutions were as shown in the table. Acetic acid and ethyl alcohol were used in concentrations of 100 ppm. The extract of canned corn (used in lieu of lyophilized corn) was composed of the turbid filtrate from a mixture of 1 part of cream-style corn in 4 parts of water. Coconut milk was diluted to 5% of normal. The yeast suspension consisted of four pellets of living dried yeast dropped into a tube of tap water containing the eggs. Eggs were submerged in the media, and records were made of the number of larvae hatched after 4 and 24 hr. Some media caused hatching for 48 and 72 hr, but no data are included for intervals longer than 24 hr. All eggs were stored on moist cellucotton for 4–5 months at 15–18° C and conditioned for 10 days at 26–27° C before submerging them.

Hatching of conditioned, durable eggs of *Psorophora discolor* was stimulated by substances that effect plant growth, as is seen in Table 1. Tap water alone had hardly any effect on the process when eggs had been conditioned only 10 days. Purified substances, when used in concentrations of 2–10 ppm, stimulated 36–43% of the eggs to hatch within an interval of 24 hr, but they were ineffective during the first 4 hr. Similarly, solutions containing glucose, when freshly prepared, caused no hatching within 4 hr, but within 24 hr 57–71% of the eggs were hatched. Diluted fresh coconut milk and a freshly prepared extract of commercially canned cream-style corn caused hatching of 80–82% of the eggs. The last two media caused most hatching within 4 hr.

Aging of all solutions containing sugar advanced the time of hatching so that nearly all eggs hatched within 4 hr after submergence. However, the total number of eggs hatched was less in all aged media (with one exception) than was the case with relatively sterile coconut milk and extracts of canned corn. The exception was the aged medium containing glucose and indoleacetic acid, which caused as high a percentage to hatch as rapidly as those containing coconut milk and corn extract. Aged media containing ethyl alcohol were inferior to others in ability to stimulate hatching. Only 19% of eggs treated with media containing alcohol hatched within 4 hr, and 58% hatched within 24 hr.

Media containing sugar were suitable for development of certain bacteria and yeasts, and these may have added

some stimulus to hatching. Yeast alone in tap water provides no stimulant for hatching that is very effective within 24 hr. The suspension used caused no hatching within 4 hr and very little within 24 hr. Therefore yeast alone is not likely to have added a stimulant to the aged media containing sugar that brought about hatching within 4 hr.

Durable eggs of *Psorophora discolor*, when submerged in media that promote growth in plants after a proper period of conditioning, hatch in a manner analogous to the elongation reaction of plant tissue. Water alone permits little or no hatching; purified stimulants cause less than half of the eggs to hatch; sugar in the medium may favor microorganisms that shorten the interval between submergence and hatching and increase percentage of hatch; freshly prepared extract of commercially canned cream-style corn and dilutions of coconut milk hasten hatching and increase the number of larvae hatched.

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A New Apparatus for Recording of Ecologic and Climatic Factors, Especially Temperature

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For many purposes it is of interest to know for what length of time the temperature has been above a certain point, e.g., 0° C. A recorder that can supply this information may be constructed using the following principle.

If a ray of light of constant intensity illuminates a photographic plate, and the plate after a certain period is removed and developed, the degree of darkness will be a function of the time during which the plate has been illuminated. If the illuminated spot moves as a function of temperature, and such an apparatus is placed under varying temperatures, one may, after a certain period, remove and develop the plate. By photometry of the picture thus obtained, one may tell something about which temperatures occurred during the time of exposure, and the frequency of occurrence of each temperature.

A paper impregnated with a salt of a radioactive element of sufficiently high stability, e.g., a salt of radium, supplies a constant source of radiation that affects a photographic plate.

On this principle, the apparatus shown in Fig. 1 has been constructed.

A pointer (4) is connected with a bimetal strip (2) which bends with temperature. At the tip of the pointer, a metal plate is placed, and through this a slit (5) is cut. The slit is 1 cm long and consists of two parts, an outer one 0.5 cm long and 0.4 mm wide, and an inner one 0.5 cm long and 0.2 mm wide.

The system of bimetal strip and pointer is fastened in a metal case (1). The pointer moves parallel to the lid of the case (6).

The lid seen from below is pictured in Fig. 1 c. There is a frame (7) with two springs (8). In this frame a

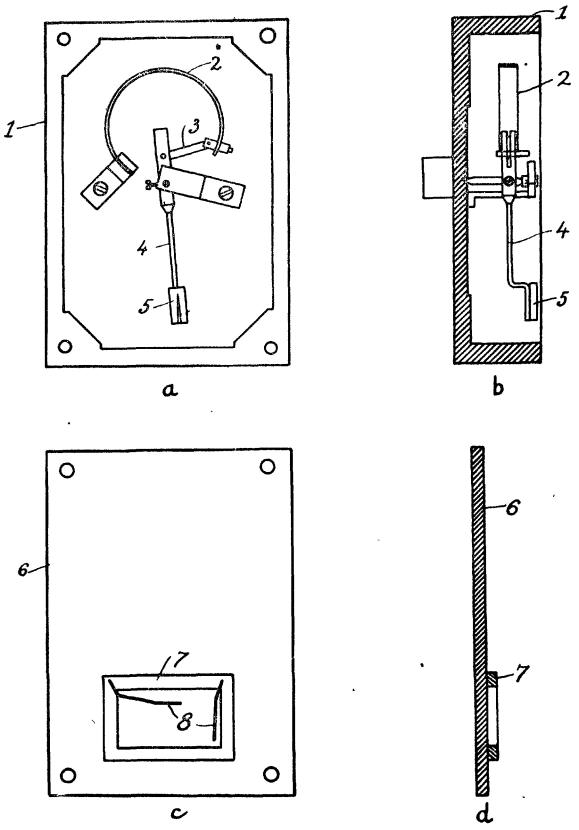

FIG. 1.

photographic plate is fastened so that two sides of the plate are pressed towards two sides of the frame. Thus the photographic plate is placed in a known position.

On the under side of the slit, a piece of paper impregnated with radium sulfate is fastened. Only the α- and β-particles going through the slit can reach the photographic plate. Some of the γ-rays going through the metal plate will reach the photographic plate, but if a fine-grained type of plate is employed, the photographic effect of the γ-rays is negligible in comparison to the effect of the α-particles.

When a photographic plate is placed in the frame, the lid fastened to the case, and the apparatus left under varying temperatures, different parts of the plate will be exposed to radiation at different temperatures. Each time a certain temperature occurs, one definite sector of the plate will be exposed, and in the course of time the exposures will be superimposed and added.