pipettes are made from pieces of capillary glass tubing of about 0.5 mm bore. The feeding end is flamed in a micro-burner until the opening is of the proper size, as determined in the first instance by actual trial on the insect, and later by comparing under a dissecting binocular with a tested pipette. The entrance canal should be short enough to permit the stylets to penetrate into the lumen of the pipette itself where they are bathed by a relatively large quantity of liquid, an important feature in connection with blood or serum suspensions, the surface of which dries quickly.

The procedure is as follows: The sandfly is etherized slightly and picked up by a leg with fine forceps. Using the cork vise with its spring as forceps, the wings are gently pressed together over the back, with the body close to and parallel with the surface of the cork. The vise with the immobilized insect, which quickly recovers from the anesthetic, is thrust into the glass tube, where it is securely held without further adjustment. Under a dissecting binocular the tip of the previously sterilized pipette is slipped over the stylets of the mouth parts, the labium folding at its base as the pipette pushes it back. This position simulates that assumed by the mouth-parts when the stylets pierce the skin in feeding naturally. The pipette is then manipulated so that the tip, with the stylets enclosed, carries the proboscis anteriorly, resulting in a bend at its base, thus accentuating the slight angle the proboscis normally makes with the head. This latter position was found to be essential for rapid feeding.

The free end of the pipette is then sterilized in the flame and a small drop of the suspension is allowed to run in. By means of another pipette fitted with rubber tube and mouth-piece this liquid is blown to the farther end, where it immediately surrounds the insect's stylets. Feeding usually begins at once and is completed within several minutes, at times within thirty seconds. It is necessary to use a fresh pipette for each insect as the minute opening is sealed by the drying of the suspension. Used pipettes may be washed by drawing water through them by means of a water suction pump.

The suspension commonly used was made as follows: Freshly defibrinated rabbit blood was centrifuged and the serum removed. Spleen tissue from a hamster heavily infected with Leishmania was ground up in this serum. A volume of spleen-serum suspension equal to that of the serum originally removed was then added to the rabbit corpuscles. Saline suspensions may also be used.

During the summer of 1926 over six hundred sandflies were fed by this method. After the considerable practice acquired in developing the technique the proportion of successful feeds was very high. The sandflies are not injured, and their subsequent behavior is quite comparable with that of naturally fed sandflies. Development of the flagellates of Leishmania was demonstrated in most of the sandflies fed with blood-spleen suspensions.

A similar method with certain modifications was successful in feeding mosquitoes (Culex). The apparatus is necessarily larger. The cork vise is cut so that the wings may be seized and held in approximately their normal position folded flat one over the other. The sandfly pipette is unsuitable on account of the length and flexibility of the mosquito's proboscis. Two pipettes are used, one fitting with the other with as little play as possible. They are made by drawing out capillary tubing to the proper diameter. The outer pipette should be broken off slantwise. Tips of both are smoothed in the flame. The outer one is slipped over the entire proboscis. The latter is thus held straight and the stylets enclosed within the labium lie approximately in the central axis of the pipette. The opening of the inner pipette is just large enough to admit the stylets but not the labium. As the inner pipette is slipped over the stylets the outer one is withdrawn slightly to allow for the bending of the labium which occurs as its tip is pushed back along the stylets. The suspension to be fed is then run in and further procedure is similar to that in the case of sandflies. Feeding experiments with mosquitoes were much less extensive than with sandflies, but served to demonstrate the feasibility of the technique.

This method could doubtless be adapted for feeding a variety of sucking insects in cases where other methods of artificial feeding, such as by means of membranes, fresh animal skin, etc., are uncertain or impracticable.⁴

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SPECIAL ARTICLES

EFFICIENCY OF PINHOLE PROBES1

By far the most efficient probe thus far has been the conical quill glass cone, yielding as much as s = 600 (s roughly in 10^{-6} atom) acoustic pressure

⁴ Waterston, J. A. "A Contribution to the Knowledge of the Bionomics of Sand-flies," Ann. Trop. Med. and Parasit., Vol. 16, p. 69. 1922.

¹ Advance Note from a Report to the Carnegie Institution of Washington, D. C.

with the given telephone exciter, or sound intensity; or over four times as much as a good plate pinhole (caet. par.). The glass probe is essentially a hollow truncated cone, so that the oscillating air current strikes a sharp circular edge. Pressure is observed in the interior of the cone, and dilation (relatively) on the outside. If we imagined an air current toward the inside were converted into pressure, whereas an outward jetlike current were not, at least in the same degree, we should simulate the action of the probe. Being an embouchure the need of an optimum diameter of pinhole is implied; but it will presently appear that the need of a sharp edge is even more imperative, and it seems natural that there should be more vorticity on one side of the pinhole than on the other.

In many respects, however, the plate embouchure is more interesting, for here one can easily modify the bore and edge character of the pinhole, which in case of the glass quill tube cone has to be ground sharp to size. I have, therefore, given further attention to the simple plate device and the more important results are recorded in Figs. 2, 3, 4, the abscissas merely indicating the consecutive experiments. It was further found that the direction of the initial current in the telephone made considerable difference. Hence the latter is provided with a switch and its first position (I) is indicated by open circles, the opposite position (II) by black circles. It was thought that the difference was merely an expression of less efficiency of the telephone in the former case; but this is not true, as so many of the black circles are negative increments in s. In each case, the plate pinhole probe (plate on a quill tube 2 cm long, .35 cm in diameter) was tested both in the salient (s) and the reentrant (r) position in relation to the U-gauge (see Fig. 1). Diameters of the pinholes pricked by a fine cambric needle are also given. The very fine pinholes (diam. .02 cm) are singly too slow for convenient use. Finally the thickness, *v*, of the foil (plate) in which the pinhole is pricked from the outside of the tube is entered, making the record complete. Fig. 1 gives the adjustment of quill tube bc with pinhole at g to the pipe pactuated by the telephone at T, and the interferometer U-gauge, U. Pipe, spring break of the circuit and electric oscillation are in tune with the relation of bq(6.5 cm) to gc (10 cm) corresponding to a maximum fringe displacement s.

Pinholes Nos. 2, 3, Fig. 2, are similar but of varying size. The diameter effect within its range is not



definite, showing that some other factor determines its behavior. The salient pinhole in position I is, as a rule, strongly positive; the reentrant behavior in position II more strongly negative, but there are many exceptions.

Pinholes Nos. 1, 0, 4, 7, Fig. 3, were punctured in much thinner aluminum foil, and the favorable effect of this is at once apparent in the improved efficiency of the probes. In other respects the remarks already made apply. Nos. 4, 7 were constructed with greater skill.

These experiments at once indicate the nature of the missing factor; for heretofore the thickness of the foil has been ignored. It is clearly of greater importance than the diameter of pinhole.

Following this suggestion I next pricked pinholes in mica plate, split as thin as admissible and much below .01 mm. The results in Fig. 4 show the enormously increased efficiency obtained, ordinates being even five times as large as those in Fig. 2, referring to the original thick foil. No. 5 was only examined for diameter .02 cm. In No. 6 there is but little difference between the first two diameters. In puncturing the third, the hole was accidentally frayed to about twice the area wanted and beyond the admissible range. Hence the low efficiency.

There is, however, always difficulty in successfully enlarging the pinhole. For instance, in No. 8 the original efficiency (diam. .02) is very large, particularly in the negative. On enlarging the bore to .035 and .042 cm, its sensitivity is nearly lost. No. 9 is another peculiar case, in which the fine pinhole is negative in the salient and positive in the reentrant position, a rare inversion of the usual occurrence.

The final graph shows the corresponding behavior of an efficient glass pinhole, one of the best. The fine hole mica probe is thus of the same order of excellence.

If we take the highest of the s values corresponding to any thickness of foil ϑ , and plot s against ϑ , we get a graph of hyperbolic contour, giving a mean estimate of sensitivity based on the results obtained. The smallest manageable thickness of plate is thus essential; in other words, the pinhole should be a sharp circle and anything of the nature of a capillary tube, however short, is detrimental. The viscosity of air is here liable to ruin the experiment.

And yet the two sides of the pinhole behave quite differently to the current of air propelled through it by the alternating nodal pressure. Hence the production of vortices at the pinhole by the acoustic pressures seems alone to account for the observed results. The oscillating air columns in contact and in opposite phases at the pinhole are successively shooting vortices into each other and the pressure difference results because, owing to the structure of the pinhole in question, one of the air columns does this more efficiently than the other. As the pinhole dominates, the hydrodynamic forces in pulsating media (Bjerknes) have no relevancy.

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THE NUTRITION OF PLANARIAN WORMS

PLANARIAN worms exhibit differences in growth capacity according to their diet. They eat animal tissues with great readiness and show resulting variations in growth, depending upon the variety of tissue used and upon its condition as well. The worms thrive upon raw liver and have been kept indefinitely in our laboratory upon this as an exclusive diet. It was found,¹ however, that the power of liver to promote growth was diminished by heat and that the diminution depended upon the temperature to which the liver was subjected and upon the time of the exposure. Brain cortex is also an excellent food for the worms, and its power to promote growth is likewise diminished by heat.

In an attempt to determine whether the principle so important for the well-being of planarian worms could be separated from the intact tissue, the following procedure was employed. Liver from freshly killed guinea pigs was ground with sand and an equal weight of ether. The resulting paste was spread in a thin layer upon large glass plates and dried by an electric fan at room temperature for several hours. At the end of the drying the liver, which was in a highly friable condition, was ground to a fine powder in a mortar. It was then extracted five times with amounts of ether equivalent to the original weight of the liver, and the ether extract was evaporated with the electric fan. The product was a vaseline-like, brownish paste.

In all our experiments Planaria maculata was used. Each experimental group was kept in a finger bowl and consisted at the beginning of thirty worms. Since planarian worms multiply by fission neither average lengths nor number of worms alone could express the total growth, and it was decided to use as an index the total length of all the worms in a group. The length of the worms was estimated by placing them in a Petri dish over a piece of polar coordinated paper covered with a glass plate. As the worm glided along fully extended its anterior extremity was centered upon the center of the paper, and by following the movements of the worm with the Petri dish a really satisfactory estimate of the length of

¹Wulzen, R., Univ. of Calif. Pub. Physiology, 1926, VII, 1.