Furgiuele et al (1964) have found that lesion of the amygdala decreases depression of the locomotor activity in rats, induced by single dose of imipramine or desipramine.

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A new method for measuring variations of rat paw volume

S. H. FERREIRA, *Department Pharmacology, Faculty of Medicine Ribeirão Preto, SP. 14.100, Brazil*

A useful parameter for assessing the anti-inflammatory activity of new compounds is their effect on the increase of rat paw volume induced by phlogogenic stimuli. In this communication a new method is described for measuring the volume of the rat paw by determining the time required for a constant delivery pump to replace the volume occupied by the paw. The volume is read after the paw has been removed from the chamber, and is thus less sensitive to the operator bias. An advantage of the method is that it does not involve the use of mercury.

A schematic diagram of the apparatus is shown in Fig. 1 (left panel). A constant volume peristaltic pump delivers saline to the side entry tube of the translucid polycarbonate chamber from the reservoir beneath the chamber. The saline contains lauryl sulphate, 2 mg ml^{-1}, and ethanol, 50 ml $litre^{-1}$, to reduce surface tension. The saline overflow leaves the chamber from a V-shaped notch in the top edge, flows down the side and forms into drops on the thin piece of polyethylene attached to its bottom. The volume of the drops can be control by adjusting the size of this piece. The flow (2·4 $\mu l\ min^{-1}$) was adjusted to provide 20 drops per 10 s (20 μl/drop).

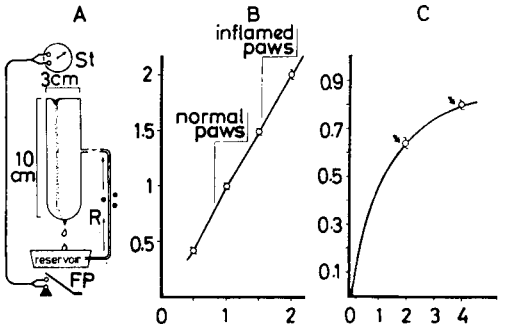

FIG. 1. The replacement volume technique. (A) gives a graphic representation of the method. A roller pump (R) maintains a constant flow dripping from the bottom of the cuvette. The paw is removed after 20 s of immersion and the fluid displaced is measured by the time necessary to refill the cuvette, indicated by the fall of the second drop. The stopwatch is activated by a foot switch (FS) simultaneously with the removal of the paw. (B) is a calibration curve. The piston of a syringe was immersed into the cuvette to displace a pre-defined volume (abscissa: ml). The volume calculated by the method is given on the ordinate (ml). The values are the mean ±s.e.m. of 5 measurements. (C) shows the mean value ±s.e.m. of five measurements of a single paw which received 100 μg of carrageenan. The increase in volume (ordinates: ml) was calculated by subtracting the value of the contralateral paw, injected with saline (0·1 ml). The arrows indicate that a sixth measurement fell within the s.e.m. Ordinate: time (h).

For measurements the rat paw is immersed in the chamber up to the tibiotarsic articulation for 15–20 s until the overflow drop frequency is constant and equal to that before paw introduction. The paw is removed and simultaneously a foot-switch activates a stopwatch (Digital Gauges, England). The time required to refill the chamber (indicated by the second drop to fall from the bottom of the chamber) is recorded. The volume displaced by the paw is directly related to refil time because pump delivery is constant. A calibration curve obtained by immersing a syringe piston in the chamber is shown in Fig. 1. There was linearity when the volume of the piston was varied from the values equivalent to a normal paw to these observed with inflamed paws. The right panel (C) shows the volume change of a rat paw after treatment with 100 μg of carrageenan (Marine Coloids, USA). The increment of the rat paw volume can be calculated either by subtracting the value measured at any time from the value obtained at zero time or by subtracting the volume of contralateral paw (control) injected with saline. Routinely two readings of the same paw were made. If there was a difference greater than 1 s between readings, a third reading was made. A trained experimenter takes about 2 min to measure both paws. The replacement volume technique was successfully used for measuring the variations of mouse paw volume. In this instance the size of the chamber was reduced (1·5 cm diameter/7·5 cm high).

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