Indian Journal of Pharmaceutical Education and Research | Association of Pharmaceutical Teachers of India

Buzzer-Assisted Plethysmometer for the Measurement of Rat Paw Volume

Kulkarni A.R*, Ganguly K, Kiran Chaturvedi, Karale S and Singh R.M

Department of Pharmacology, S.E.T's College of Pharmacy, Dharwad - 580 002, Karnataka

ABSTRACT

Submitted: 1-10-2010 *Revised: 28-12-2010* *Accepted: 19-4-2011*

A sensitive and reproducible method of measuring rat paw volume was developed by connecting electrical buzzer to conventional plethysmometer.This methodology combined ease of operation and precision as the end point is detected by buzzer system. Standard error of mean (SEM) were calculated for the volume measured by both conventional as well as buzzer-assisted plethysmometer and results indicated that calculated values of SEM were 0.02 and 0.037 for buzzer-assisted and conventional plethysmometer respectively. Presently developed buzzer-assisted plethysmometer may be a useful tool for the students carrying out research on inflammation.

Keywords: Plethysmometer, Edema, Inflammation

INTRODUCTION

Carrageenan induced rat paw edema is a convenient method for assessing an agent for acute anti-inflammatory activity by examining its ability to reduce or prevent the development of paw swelling[1]. This model was first time developed by Winter et al [2] and has long been used to assess the anti-inflammatory properties of agents such as nonsteroidal anti-inflammatory drugs (NSAIDs) which inhibit prostaglandin production. Over the period of time, various modifications have been suggested for improvement of this model to increase accuracy of the volume measurement so as to minimize the errors. For instance, sensitive and reproducible method of measuring mouse paw volume was developed by interfacing a Mettler Delta Range top-loading balance with a microcomputer[3]. This method had high precision with the advantages of computer-controlled data processing and archivable storage of data. Recently, digital plethysmometers are available for measurement of change in small volume[4]. However, the buzzer-assisted plethysmometer developed here is a modified version of conventional plethysmometer with some modification and can be prepared by students. This paper gives details of the equipment and design with an emphasis to its reproducibility.

MATERIALS AND METHODS

Design of the plethysmometer

Buzzer connections and modifications to conventional plethysmometer were made as per the figure 1. An electrical buzzer which works on 6V DC battery was adopted. Two electrical leads of the buzzer were introduced into plethysmometer such that when mercury touches certain level, leads get connected with each other with the help of mercury so that buzzer starts blowing. Therefore, when mercury reaches certain level, buzzer should start blowing and should be off below this particular level.

Measurement of standard volume

Wooden cylinders of known volume (0.5, 0.75 and 1.0 ml) were completely immersed into mercury of the plethysmometer. As a result the mercury level in the plethysmometer increases in accordance to their volume. The raised mercury levels were adjusted to original level with and without help of buzzer. All the readings were taken as average of five measurements and standard error of mean (SEM) was calculated.

Measurements of rat paw volume

Wistar albino rats were purchased from Venkateshwara enterprises, Bangalore, India. Animals were maintained under conventional laboratory conditions, at temperature 25±2°C, and a 12 h natural light period. Commercial pellet diet (Lipton India) and drinking water were provided *ad libitum*. Animal experiments were carried out after obtaining the clearance from the institutional animal ethical committee. Wistar albino rat of 158 g was selected for measurement of paw volume. The paw was completely immersed into mercury up to the tibiotarsal articulation. As a result the mercury level in the plethysmometer increased and the raised mercury level was adjusted to original level with and without help of buzzer. All the readings were taken average of five measurements and standard error of mean (SEM) was calculated.

RESULTS AND DISCUSSION

An electrical buzzer which works on low voltage battery (6V DC) was adopted. Electrical connections were made as shown in figure 1. The buzzer alerts when mercury touches certain level in plethysmometer due to closing of the circuit. In case of conventional plethysmometer, adjustment of level

***Address for Correspondence:**

Kulkarni A.R, Department of Pharmacology, S.E.T's College of Pharmacy, Dharwad-580 002, Karnataka

E-mail : kulkarni73@yahoo.com

of mercury is done by close observation. Before, measurement of the actual rat paw volume, plethysmometer was calibrated with standard wooden cylinders of known volume. Measurement of volume of standard wooden cylinders is also done to confirm the reproducibility of the measurement.

Results of mean volume, standard deviation (SD) and standard error of mean (SEM) for standard wooden cylinder using different plethysmometers are shown in Table 1. Mean volume of 1.14 and 1.09 ml was obtained for 1 ml standard wooden cylinder using conventional plethysmometer and buzzer-assisted plethysmometer respectively. Results clearly indicated that deviations obtained for buzzer-assisted plethysmometer were comparatively less than that of conventional plethysmometer.

Results of standard error of mean obtained for standard wooden cylinders of different volume using different plethysmometers are shown in Table 2. It was observed that calculated values of SEM for the data measured by buzzer-assisted plethysmometer were comparatively less than that of conventional plethysmometer. Results also indicated that as volume of standard wooden cylinders increase, the SEM also increases however, quantum of increase is less in case of buzzer-assisted plethysmometer than the conventional one.

Results of mean rat paw volume measured by using different plethysmometers are shown in Table 3. A mean rat paw volume of 1.12 and 1.18 ml were obtained for conventional and buzzer-assisted plethysmometer respectively. It was observed that calculated values of SEM for the data measured by buzzer-assisted plethysmometer were comparatively less than that of conventional plethysmometer.

CONCLUSION

The newly developed electrical buzzer-assisted plethysmometer is a sensitive method of measuring rat paw volume and has got good reproducibility. The average volumes obtained for buzzer-assisted plethysmometer were very much close to the actual volume than the conventional one. The buzzer-assisted plethysmometer may be a useful tool for measurement of small volume associated with rat paw edema.

Table 1: Results of mean volume and SEM obtained for standard wooden cylinder of 1 ml using different plethysmometers

Instrument	Mean volume* (ml)	SD	SEM
Conventional plethysmometer	1.14	0.107497	0.0340
Buzzer-assisted plethysmometer	1.09	0.073786	0.0233

*n= Average of 5 determinations

Table 2: Comparison of SEM calculated for various fixed volumes

Instrument	SEM for various fixed volume* (ml)		
	0.5	0.75	1.0
Conventional plethysmometer	0.0150	0.0340	0.0416
Buzzer-assisted plethysmometer	0.0100	0.0100	0.0233

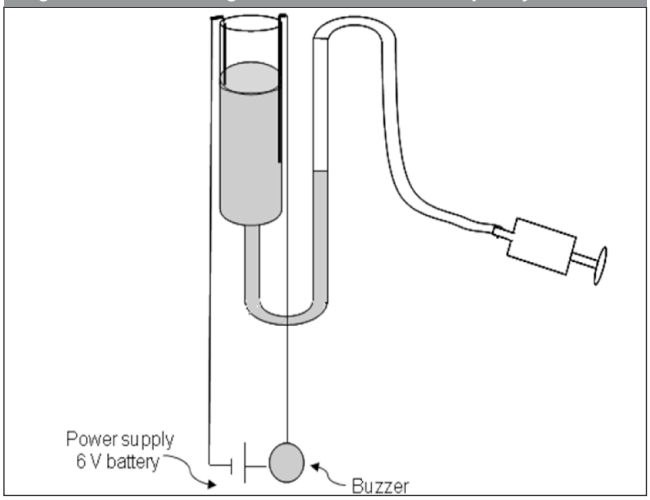
*n= Average of 5 determinations

Table 3. Results of mean rat paw volume and SEM measured using different plethysmometers

Instrument	Mean volume* (ml)	SEM
Conventional plethysmometer	1.12	0.037
Buzzer-assisted plethysmometer	1.18	0.020

*n= Average of 5 determinations

Fig. 1: Schematic diagram of Buzzer-assisted plethysmometer



ACKNOWLEDGEMENT

We thank Principal Dr. V. H. Kulkarni and Shri. H. V. Dambal, President, S.E.T's College of Pharmacy, Dharwad, India for providing necessary facilities.

REFERENCES

1. Turner RA. Screening methods in pharmacology. 5th ed. London: Academic Press Inc; 1965; 152.
2. Winter CA, Risley EA, Nuss GW. Carrageenan-induced oedema in the hind paw of rat as an assay for anti-inflammatory activity. ProcSoc Exp Biol Ther 1962; 111: 544-7.
3. Webb EF, Griswold DE. Microprocessor-assisted plethysmograph for the measurement of mouse paw volume. J Pharmacol Methods. 1984; 12(2):149-53.
4. Anonymous. Plethysmometer [online]. 2010 [cited 2010 Sep 23]. Available from: URL: http://www.ugobasile.com