AN IMPROVED MODEL FOR BIOASSAY OF TOPICAL CORTICOSTEROIDS

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Pretreatment of skin with topical corticosteroids followed by pricking the skin with histamine solution, results in relative suppression of histamine response. The reduction in the diameter of the wheal has been used to assess the relative efficacy of topical corticosteroids. The wheal being three dimensional, we have improved the above model by measuring the volume rather than the diameter of the wheal, which was found to be more accurate and reproducible.

Key words : Topical corticosteroid, Improved method for assay.

Various methods of testing the efficacy of topical corticosteroids have been developed. Reddy and Singh[1] developed a very simple technique using the ability of local corticosteroids to suppress the histamine wheal induction. In this technique they measured the diameter of the wheal produced in response to pricking with histamine solution. We have now improved their technique by measuring the volume of the wheal instead of its diameter.

Materials and Methods

Ten healthy volunteers aged 15 to 30 years were selected for the study. None of them had used systemic or topical corticosteroids or antihistaminics for atleast 8 weeks prior to the study.

Flexural aspects of both forearms were cleaned with 70% alcohol and two sites 5 cm apart were selected on each forearm avoiding any prominent vein. Initial reading by the spherometer was taken for each site by keeping the forearm flat on the table-top. The exact sites for all the three prongs of the spherometer were marked so that the second reading could be taken at the same site. A circular glass cup, open at both ends and with an even surface was placed on the selected site and 0.5 ml of 0.15% histamine acid phosphate solution was poured to a depth of 2 mm and a prick was made by an automatic pricking needle. The solution was kept at the site for 30 seconds and then the glass cup was removed and the solution was wiped off with cotton wool. The maximum and minimum diameters of the wheal were measured after 12 minutes. The spherometer was kept on the previously marked sites and the second reading was taken. The difference between these two readings on the spherometer was the height of the wheal.

Volume of the wheal was calculated by using formula $\pi r^2 h$, where r denotes radius (half of the diameter) and h denotes the height of the wheal (The difference between the two readings on the spherometer).

0.5 ml of (0.1%) fluocinolone acetonide cream on two sites on one forearm and a bland cream on the other forearm as control was applied with tuberculin syringes. Fresh occlusive dressings with corticosteroids and the bland cream were applied on the respective sites daily for three days after cleaning with 70% alcohol. During these days the subjects were instructed to avoid wetting the dressings. Histamine test was repeated after 72 hours of the start of the experiment.

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Results

On comparing the mean volume of the wheals at the experimental sites i.e. corticosteroid site and the control site, there was no difference ($P>0.9$) before occlusion with corticosteroid cream. We have, however, observed statistically highly significant ($P<0.001$) reduction in the mean volume of the wheals after 72 hours of occlusion with corticosteroid cream (Table I).

Comments

A large number of methods are described in the literature for assessing the potency of topical corticosteroids on animals and on human subjects. Animal models cannot predict the response in human beings because of obvious reasons. McKenzie and Stoughton[2] technique is still the most popular method till date but it has certain limitations. It assesses the capacity of the corticosteroid to induce vasoconstriction of healthy blood vessels from normal state. This is, however, not the situation in clinical practice. Kaidbey and Kligman[3] found that potency ratings achieved in the vasoconstrictor test do not always parallel the therapeutic effect on dermatitic skin. These authors prefer to measure the degree of suppression of experimentally induced contact allergic dermatitis. Burrows and Stoughton[4] evaluated the potency of topical corticosteroids by studying their relative capacity in different concentrations to prevent the induction of contact sensitization with the application of DNCB on corticosteroid treated skin. These techniques are, however, not practicable for routine practice as these induce sensitization.

Reddy and Singh's technique[1] is an improvement because it assesses the relative ability of topical corticosteroid preparations to suppress histamine effect on blood vessels. It appears to be closer to the clinical situations than other techniques. Ive and Comaish[5] have, however, described that Reddy and Singh's technique appeared to match more sophisticated methods of measurement. In this technique, the diameter of the wheal is measured and the reduction in size is corre-

Table I. Effect of the corticosteroid in reducing the size of the histamine wheal.

	Volume of the wheal in mm³			
	Before occlusion		72 hours after occlusion	
	Test site	Control site	Test site	Control site
1	113.14	111.53	3.78	113.62
2	120.40	117.42	13.71	120.48
3	44.19	42.54	4.99	41.97
4	86.87	86.15	11.58	86.00
5	86.88	86.74	20.41	86.82
6	131.25	133.51	8.49	132.74
7	51.55	48.05	2.67	49.01
8	63.25	62.87	8.38	62.69
9	51.04	49.28	5.56	50.04
10	75.43	75.68	3.90	73.67
Mean	82.40	81.38	8.35	81.70
±S. D.	±30.95	±31.63	±5.54	±32.07
P values	>0.9		<0.001*	

* Statistically highly significant.

lated with the relative potency of the corticosteroids. Wheal is, however, three dimensional, and we have observed that even in the same subject the size of the wheal does vary at different sites. This is probably due to the fact that the wheal becomes larger in diameter where there is loose areolar tissue. We could feel that in these cases the height of the wheal was less. We, therefore, felt that the volume of the wheal may be a more sensitive indicator.

A simple technique was developed using a very cheap and easily available spherometer by which we could measure volume with a fairly good accuracy. The difference in the volumes of the wheals between various control sites was insignificant.

Volumes of histamine-induced wheals 72 hours after application and occlusion with fluocinolone acetonide cream (0.1%) at two different sites in the same individuals compared with the volumes of wheals at the control sites are shoum in table I. There was statistically highly significant reduction in the volume of wheals in comparison to control sites after 72 hours of corticosteroid application but there was no difference between the volumes of the wheals produced at the two control sites.

References

1. Reddy BSN and Singh G : A new model for human bioassay of topical corticosteroids, Brit J Dermatol, 1976; 94 : 191-193.
2. McKenzie AW and Stoughton RB : Method for comparing percutaneous absorption of steroids, Arch Dermatol, 1962; 86 : 608-610.
3. Kaidbey KH and Kligman AM : Assay of topical corticosteroids by suppression of experimental inflammation in humans, J Invest Dermatol, 1974; 63 : 292-294.
4. Burrows WM and Stoughton RB : Inhibition of induction of human contact sensitization by topical gluco-corticoids, Arch Dermatol, 1976; 112 : 175-178.
5. Ive A and Comaish S : Topical therapy, in : Recent Advances in Dermatology-5, Editors, Rook A and Savin J, Churchill Livingstone, Edinburgh, 1980; p 292.