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TETRACYCLINE LEVELS IN SKIN SURFACE FILM AFTER ORAL ADMINISTRATION OF TETRACYCLINE TO NORMAL ADULTS AND TO PATIENTS WITH ACNE VULGARIS*

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Tetracycline is widely used in the treatment of acne vulgaris, and several studies have demonstrated significant clinical improvement in patients on such therapy (1, 2). Yet, why this drug is helpful in acne is not completely understood. Small doses do result in a reduction in the number of bacteria on the skin surface, (3) as well as in the free fatty acid content of surface lipids (4). The beneficial effect of tetracycline may be due to its antibiotic action against *Corynebacterium acnes*. However, the fact that other antibacterial agents, notably penicillin, which are equally effective against *C. acnes in vitro* (5) do not produce equal clinical improvement in acne or reduce the free fatty acid content of skin surface lipids, leads to consideration of other possible mechanisms of action of tetracycline in this condition.

Tetracycline has been demonstrated in the skin of animals (7, 8) and it has been demonstrated by fluorescence in human hair, nails, and epitheliomas (9–11). Gould and Ritchie (12) using microbiological assay methods, reported whole skin levels of tetracycline approaching 80% of those of the serum. Cullen and Crounse (13) on the other hand, were not able to demonstrate, by fluorometry, tetracycline in full thickness biopsies of normal skin from the buttock, although it was present in the inflamed skin of patients with dermatitis or psoriasis.

The present study was undertaken to determine whether tetracycline appears on the skin surface in significant quantities after oral administration to normal adults and to patients with acne.

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Tetracycline hydrochloride used in this study was furnished by Lederle Laboratories and Squibb Laboratories.

MATERIALS AND METHODS

Experimental subjects were divided into three groups.

Group I consisted of 5 normal adult volunteers who took full therapeutic doses (1–1.5 gm, or 14–19 mgm per kilogram body weight) each day for 14 days.

Group II consisted of 3 normal adults who took gradually increasing doses of tetracycline up to full doses, followed by a period of decreasing doses.

Group III consisted of 24 patients with acne who were taking .250 grams of tetracycline daily for periods of several weeks or months.

Collection of surface film material (SFM). Experimental subjects were requested to avoid applying cosmetics and rubbing or cleansing their forehead skin throughout periods of observation. At approximately mid-afternoon, the surface film material (SFM) was collected by lightly rubbing the skin of the forehead area, between the eyebrows and scalp hairline, with a cotton pledget moistened with absolute alcohol. This cotton pledget was then thoroughly rinsed with several changes of absolute ethyl alcohol and pressed as dry as possible. The washing alcohol was then collected in a previously tared 10 ml glass beaker and allowed to evaporate to dryness at room temperature, a process which required 24–36 hours. The quantity of residue SFM was determined by weighing. The contents of the beaker were then redissolved in 5 cc of 0.1 N HCl solution and analyzed for tetracycline content by the spectrofluorometric method of Kohn.

Tetracycline levels in freshly drawn aliquots of serum from the above patients were also determined by the fluorometric method of Kohn.

Measurements of fluorescence were made in a Turner model 110 fluorometer, with filters providing excitation and emission measurements of 405 and 520 mμ respectively. Readings in unknowns were compared to those given by freshly prepared standard solutions of tetracycline and SFM from individuals not taking the drug. Background fluorescence corresponding to 0.05 to 0.15 μg tetracycline per 100 mg surface film was recorded in surface film from patients not taking tetracycline.

RESULTS

Group I

Table I shows the levels of tetracycline in surface film of five adults who took full doses of

TABLE I
Group I
Levels of tetracycline in skin surface film from patients on full therapeutic doses

Patient	Daily dose of tetracycline (mg./kg.)	μg. Tetracycline per ml. serum	μg Tetracycline per 100 mg. surface film
1	17.3	1.2	0.58 (0.25–0.89)
2	18.9	2.5	0.76 (0.12–1.7)
3	13.4	1.5	1.26 (0.53–1.9)
4	17.6	1.5	0.91 (0.52–1.4)
5	19.1	1.4	0.93 (0.4–1.9)

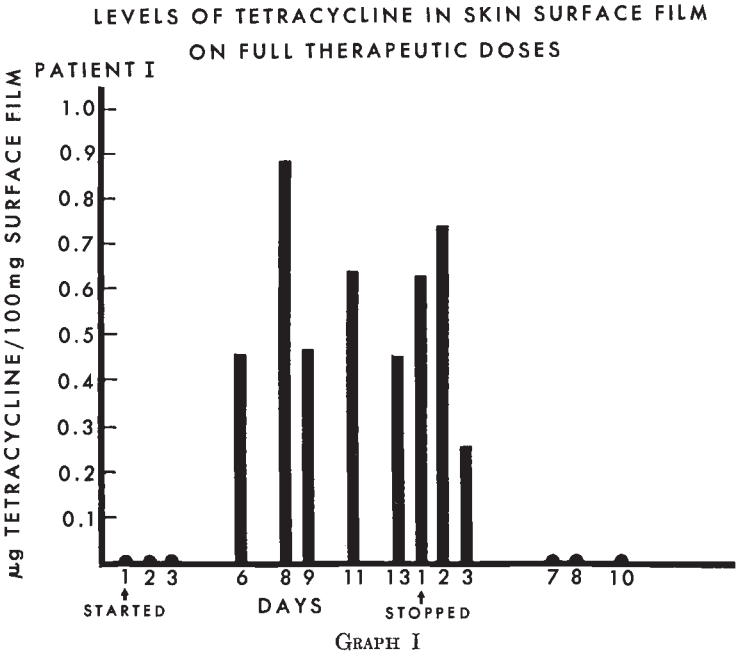
tetracycline for 14 days. Tetracycline appeared in surface film 4 to 8 days after starting tetracycline and measured from 0.4 μg to 1.9 μg per 100.0 mg. of surface film material. Corresponding serum levels of tetracycline ranged from 1.4 μg to 2.5 μg ml. of serum. Tetracycline was measured in surface film from 4 to 6 days after discontinuing tetracycline therapy. Though levels of tetracycline varied from day to day, it is seen that the levels reached bacteriostatic ranges comparable to those found in serum. Results from representative patients are recorded in Graphs I and II.

Group II

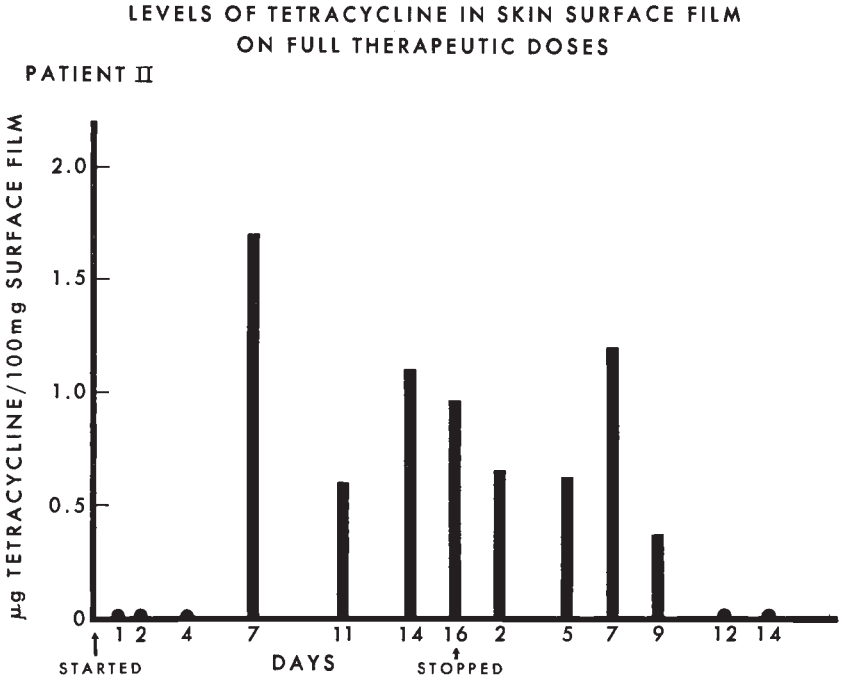
Group II patients who took gradually increasing and decreasing amounts of tetracycline over a sixteen day period demonstrated skin levels of tetracycline 5 to 7 days after initiating treatment measuring from 0.2 to 1.5 μg per 100 mg. of surface film material. Serum levels ranged from 0.4 to 4.5 μg per ml. of serum. Results from a representative patient are recorded in Graph III.

Group III

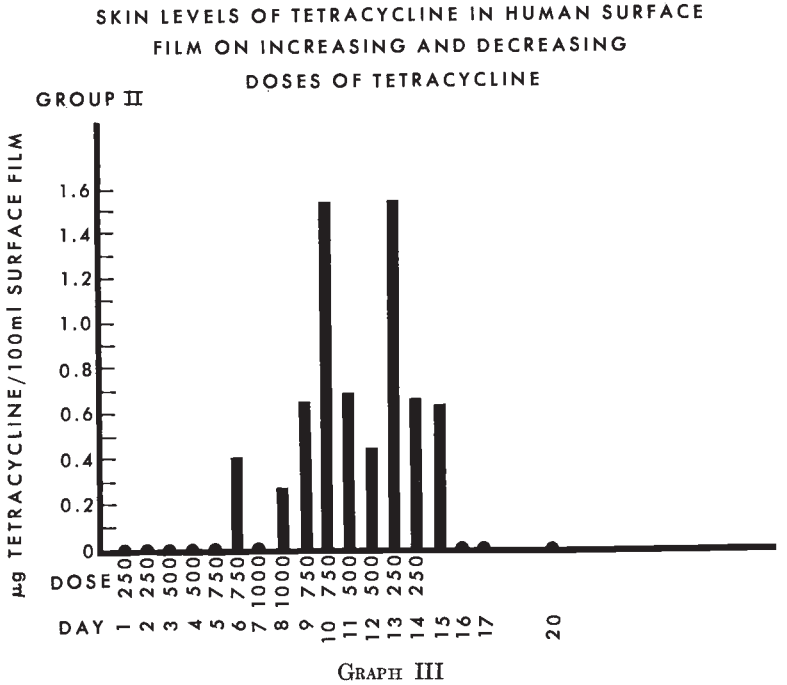
Group III consisted of patients who had been taking small doses of tetracycline over a long period. Skin surface film levels were demonstrated in 13 out of 23 patients ranging from .4 to 1.5 μg per 100 mg. surface film collected. Serum levels in these patients ranged from .5 to 2.5 μg per ml. Eight patients with demonstrable tetracycline in their sera failed to



GRAPH I



GRAPH II



GRAPH III

TABLE II

Group III

Levels of tetracycline in skin surface film from patients on prolonged low dosage therapy

Case no.	Duration of therapy	μg Tetracycline per 100 mg surface film
1	2 weeks	0.35
2	5 months	1.85
3	4 months	—
4	3 months	0.47
5	2 months	1.73
6	3 months	1.68
7	2 months	1.07
8	2 months	1.43
9	2 months	—
10	2 months	—
11	2 weeks	—
12	1 month	0.48
13	1 month	0.26
14	1 month	—
15	1 month	—
16	3 months	0.91
17	4 months	0.63
18	9 months	—
19	2 weeks	—
20	8 months	—
21	2 weeks	—
22	4 months	1.85
23	4 months	1.18

show the drug in their SFM. Two patients had no demonstrable tetracycline in their sera at the time the sample was drawn. Results are shown in Table II.

COMMENT

We have very carefully avoided use of the term "sebum" in referring to the material removed from the surface of the forehead by wiping it with alcohol. The surface film material probably is composed of a variety of materials, including sebum, dried sweat, desquamating keratin and variable amounts of extraneous material including Colorado dust and Denver mud. Thus any measurements of tetracycline levels in this heterogeneous and undefined material must not be interpreted as precise measurements of anything, and it is not surprising that the measurements indicated wide variation in tetracycline levels from day to day. We can only conclude that significant and probably bacteriostatically effective quantities of tetracycline do reach the skin surface film even in patients on small quantities of this drug.

There is reason to believe that the concentrations of tetracycline on the skin surface are higher than we recorded. There are several possible reasons why this could be so: 1) the presence of extraneous material in the weighed amounts of surface film 2) inadvertent wiping or washing of the forehead by the patient from one collection period to another 3) decomposition of tetracycline on the skin surface and during the evaporation time required for our analytical procedure. A better method of collection of tetracycline is in order for more rapid analysis of the drug. However, when known amounts of tetracycline were added to cotton sponges, 80% was recovered by our analytical methods. We experimented with other methods of collection, including the cup method, cigarette paper method, and wiping the forehead with ether, but found our method of collection to be superior.

Do the data permit any conclusions about the route by which tetracycline reaches the skin surface film? Because of the observed time lag of 4–8 days between the institution of therapy and the appearance of measurable quantities of tetracycline in the skin surface film, it is tempting to thick that the probable route is via the sebaceous glands, which are known in the rat to have a turnover time of about this duration (15). The trans-sebaceous route also seems attractive because of the known lipid solubility of tetracycline base in which form the drug should exist at the slightly alkaline pH of serum, which would favor its solubility in the lipid laden sebaceous cells.

On the other hand, the delay of several days in the appearance of measurable quantities of tetracycline in the skin surface film material does not completely rule out transepidermal penetration, or, even more likely, its delivery by sweat. In these, the reason for the lag period of several days might be that it takes such a period for measurable quantities of the drug to accumulate in the surface film. Actually, it is quite possible for all three routes, sebaceous, transepidermal, or through the sweat glands, to be operative to greater or lesser degree.

Obviously, a study of this sort raises more questions than it answers about the pharmacology of tetracycline in the skin. In par-

ticular, we have not gone as far as we had hoped in measuring levels of tetracycline attained within the pilosebaceous lumens, in which acne centers. We have only shown that tetracycline does attain significant levels on the skin surface, even after small amounts have been taken orally, and that there is a lag period of several days between administration of the drug and its appearance on the skin surface.

SUMMARY

Tetracycline was demonstrated in significant quantities in surface film material collected from the forehead after oral administration of tetracycline to normal adults and to patients with acne. A delay of four to eight days occurred before tetracycline appeared in the surface film material and remained in surface film material for three to eight days after the drug was discontinued.

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