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The Limited Role of Corticosteroids in Ameliorating Experimental Doxorubicin Skin Toxicity in the Mouse

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Summary. *Local skin necrosis is a serious complication of doxorubicin (Adriamycin) infiltration in man. Intradermal (ID) doxorubicin injection was used in the mouse to create skin lesions, after wich a number of local corticosteroid interventions were studied. The most effective local antidote was low-dose (2.5 mg) hydrocortisone (HC), but only against the low-dose doxorubicin challenge (0.05 mg). Other andidotes with lesser effectiveness included dexamethasone-sodium bicarbonate and an intermediate dose of HC (6.25 mg). Larger doses of ID HC did not prevent local doxorubicin toxicity and were inherently toxic to the skin. Systemic corticosteroids were similarly ineffective. The superiority of the low ID HC dose, the ineffectiveness of additions (sodium bicarbonate, topical HC cream), and the resistance of the high-dose doxorubicin ID challenge (0.5 mg) suggests a limited role for corticosteroids in the management of experimental doxorubicin skin toxicity.*

Introduction

In a previous report we have described an experimental murine model that allows quantitation of doxorubicin (Adriamycin) skin toxicity [8]. A number of antidotes have been reported to reduce local skin damage in man following inadvertent venous extravasation of doxorubicin [2, 3, 10, 11, 15]. Problems exist, however, in evaluating these anecdotal case reports: (1) Different and often unknown amounts of drug were infiltrated; (2) infiltration occurred into different anatomical areas; and (3) nonstandard approaches to local management were employed. A variety of pharmacologic approaches have been used, most involving local corticosteroids.

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If left untreated, there is great potential for severe and prolonged local skin toxicity following doxorubicin extravasation in man [3, 12]. Lesions on the dorsum of the hand are particularly serious since important deep tissues are readily involved, leading to neural and vascular necrosis and often functional impairment of the affected limb. Often reconstructive surgery is required since extensive lesions may not heal appropriately and can slowly expand in size and depth over a period of months.

In this report we describe the limited effectiveness of several corticosteroid antidotes to doxorubicin skin infiltration in a mouse model. An attempt was made to simulate the clinical reports involving corticosteroids; thus, antidotes tested include hydrocortisone (HC) [12], dexamethasone-bicarbonate [15], and hydrocortisone injection plus topical cream applications [2]

Materials and Methods

Adult female BALB/c mice (Jackson Laboratories, Bar Harbor, Maine) weighing an average of 25 g undergo the removal of dorsal hair from a 3 × 3 cm area 24 h prior to doxorubicin injection. One to two applications of the topical depilatory agent Neet lotion (containing thioglycolic acid, mineral oil, calcium hydroxide, sorbitol, and steryl and cetyl alcohols) is used for this purpose. The experimental model [8] uses ID injections of 0.5 and 0.05 mg doxorubicin (Adriamycin, Adria Laboratories, Wilmington, Delaware), each diluted into 0.05 ml 0.9% sodium chloride USP (without preservatives).

Local antidotes included hydrocortisone sodium succinate (Solu-Cortef, 100 mg/2 ml Redi-Vial, Upjohn Laboratories, Kalamazoo, Michigan USA), dexamethasone sodium phosphate (Decadron, 4 mg/ml multidose vial, Merck, Sharpe and Dohme Laboratories, West Point, PA, USA), and sodium bicarbonate injection USP (8.4% or 50 mEq/50 ml, Abbott Laboratories, Chicago, ILL, USA).

Animals were divided into treatment groups of four each and caged together for the remainder of the experiment. Control injections were performed for each antidote tested. Each antidote

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was injected ID at 0.05 ml volume given proximal to and immediately after the ID doxorubicin. The perpendicular widths of skin lesions were measured by micrometer daily. Three parameters were assessed: induration, erythema, and ulceration. In our previous report, these toxicity parameters were shown to increase in a doxorubicin dose-dependent fashion with respect to: (1) the total area of the lesion [the area under a toxicity-time curve (cm^2/day)]; (2) the peak toxicity level (cm^2) and the duration of each toxicity (days).

Statistical significance was assessed at the 0.05 level according to Student's *t*-test for analysis, and the results of antidote treatments were compared with those recorded in 0.05 ml saline-treated control animals receiving ID injections of 0.5 and 0.05 mg doxorubicin. The specific doses of the antidotes (each diluted to 0.05 ml total volume) were as follows: 2.5 mg HC (low dose): 6.25 mg HC (intermediate dose), 12.5 mg HC (high dose): 0.1 mg/0.025 mEq dexamethasone-sodium bicarbonate; and 2.5 mg ID HC followed by a daily application of 0.5% HC cream USP (E. Fougers & Co., Hicksville, NY USA) for days.

Results

Seven different ID antidotes were evaluated for their ability to prevent ID doxorubicin-induced skin lesions. Doxorubicin dose-dependent lesions reached maximal toxic areas 5–10 days after injection. Lesions then slowly healed over 25–45 days. Typical skin lesions were ovoid in nature with concentric areas of induration, erythema, and ulceration (in descending order of size).

The results of the seven pharmacologic interventions in the prevention of ulceration are shown in Table 1. In this series, the lowest ID HC dose tested (2.5 mg) proved more effective in the prevention of skin toxicity than either of the higher doses, the highest (12.5 mg) being locally toxic. This dose was also given by IM injection (into the thigh of a hind leg) to test whether a systemic corticosteroid effect was involved. Neither the total toxicity areas nor the peak lesion size were affected by the IM HC injection following either doxorubicin challenge. However, more rapid healing was noted in mice given 0.05 mg doxorubicin.

HC had little effect in preventing skin toxicity following the highest doxorubicin dose, with the exception that the intermediate HC dose did significantly reduce the duration of ulceration following 0.5 mg doxorubicin ($P < 0.05$, Table 1).

The combination of dexamethasone and sodium bicarbonate was effective in reducing the skin toxicity caused by 0.05 mg ID doxorubicin. There was also a suggestion, though not statistically significant, that this drug combination can reduce the extent of ulceration following the larger doxorubicin dose. Sodium bicarbonate alone, tested at a single dose of 0.05 ml or 0.05 mEq, did not reduce the skin toxicity caused by either doxorubicin dose. Rather it signif-

Table 1. Comparison of skin toxicity for seven corticosteroid-containing antidotes against two ID doxorubicin challenge doses

Antidote	Total toxicity AUC ($cm^2 \cdot$ day)			Peak level (cm^2)			Damage duration (days)		
	Doxorubicin (mg) ID			Doxorubicin (mg) ID			Doxorubicin (mg) ID		
	None	0.05	0.5	None	0.05	0.5	None	0.05	0.05
Normal saline ID (control)	0	0.528	8.235	0	0.069	0.660	0	17.75	29.50
Low ID dose hydrocortisone	0	0.005[a]	10.333	0	0.003[a]	0.735	0	0.75[a]	31.50
Intermediate ID dose hydrocortisone	0	0.139[a]	3.618	0	0.030[a]	0.305	0	7.50[a]	22.50[a]
High ID dose hydrocortisone	0.964	0.178[b]	10.51	0.165	0.038[a]	0.753	5.00	6.75[a]	28.00
High IM dose hydrocortisone	0	0.230	8.774	0	0.063	0.727	0	4.25[a]	26.67
Dexamethasone–sodium bicarbonate ID	0	0.034[a]	4.778	0	0.0075[a]	0.455	0	1.75[a]	27.00
Sodium bicarbonate ID	0	0.671	7.346	0	0.125[c]	0.680	0	13.50	31.25
Hydrocortisone ID and topical cream	0	0.379	6.358	0	0.054	0.477	0	8.75	35.00[c]

[a] Denotes $P < 0.05$ difference favoring treatment over control
[b] Marginal significance ($P < 0.07$)
[c] Denotes $P < 0.05$ difference favoring control over treatment

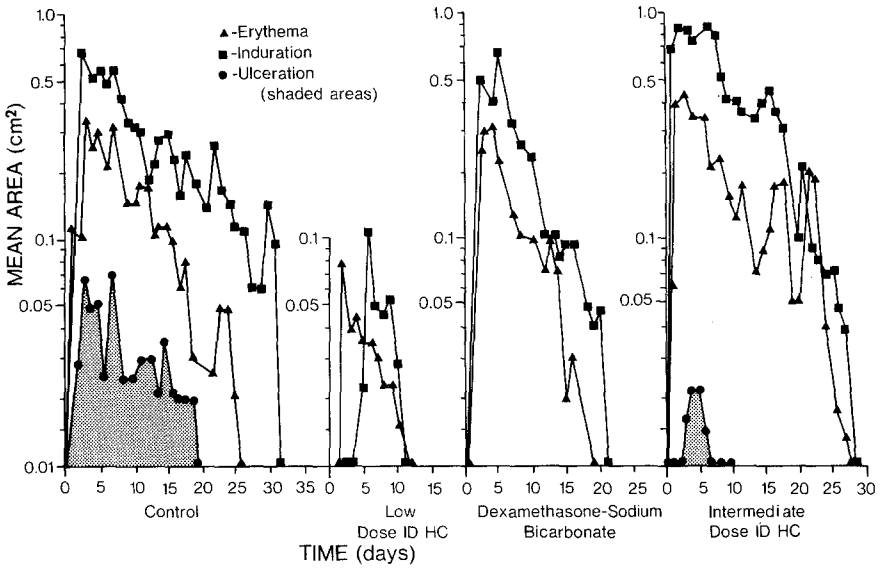

Fig. 1. Skin toxicity for three corticosteroid antidotes compared with control following 0.05-mg ID doxorubicin challenge

icantly increased peak toxicity associated with the 0.05 mg ID doxorubicin-treated animals ($P < 0.04$). Similarly, the addition of daily hydrocortisone cream applications to low-dose ID HC did not significantly reduce ulceration following doxorubicin.

The most effective local treatments for the 0.05-mg doses of doxorubicin were (in descending order) low-dose HC, dexamethasone-sodium bicarbonate, and intermediate-dose HC. These are graphically compared in Fig. 1.

Discussion

Several conclusions are possible from these experimental results: (1) Low ID doses of corticosteroids are more effective in preventing doxorubicin-induced skin toxicity than higher ID doses; (2) murine skin toxicity caused by high doses of concentrated doxorubicin is relatively resistant to local and systemic corticosteroid prevention; and (3) combinations of sodium bicarbonate or topical corticosteroid with low-dose ID HC do not improve results over those obtained with low-dose ID HC alone.

One limitation of these data is that the model depends upon ID injection of relatively concentrated drug solutions. This is not the same as clinical doxorubicin infiltrations, which typically involve larger volumes of more dilute drug delivered SC. Another overall limitation involves the usual caveat, which proscribes direct extrapolation of animal data to man.

It is interesting to note that both combination regimes (i.e., dexamethasone and sodium bicarbonate or hydrocortisone and topical cream) were slightly, but significantly, inferior to low-dose ID HC alone. Furthermore, sodium bicarbonate, used alone as an antidote, appeared to add significantly to doxorubicin-induced skin toxicity ($P < 0.05$). Thus, it seems that the positive clinical results of Zweig and Kabakow [15], who used dexamethasone plus sodium bicarbonate, could be explained by the effect of the corticosteroid alone.

Corticosteroids create diverse pharmacologic alterations, and the exact biochemical reason for the apparent effectiveness against ID doxorubicin noted in this study is unclear. That the higher doses of HC produced skin ulceration is not altogether surprising. It is well established that corticosteroids inhibit the biosynthesis of collagen [14] and have been associated with diverse overt skin toxicities in man [5]. In addition, even the 'low' ID HC dose of 2.5 mg (per 0.007 m^2 average mouse) found effective in this series represents an approximate equivalent of 350 mg/m^2 in man [9].

Cohen [6] was unable to show a protective effect for local corticosteroid injections in a subcutaneous, doxorubicin-mouse model. In contrast to Cohen's work, our earlier results [7] and those of Rudolf et al. [13] showed that the subcutaneous mouse model produced inconsistent ulceration. This is probably because of the presence in the mouse of an attached, supportive muscle layer (the panniculus carnossus). This layer is directly adherent to mouse skin but is not found in human skin, which is nourished and supported by deep subcutaneous vessels and tissues. Other potential limitations of this study include the use of a single dose of bicarbonte and the necessity of injecting relatively small volumes of drug (0.05 ml) in the intradermal model used. The use of the small

volumes could produce local hypertonicity and irritation, which might augment the inherent toxicity of the drug. However, the separate injections of the antidotes alone should have controlled for any inherent (ID) antidote toxicities. Furthermore, all statistical comparisons were made against control animals receiving an equivalent Adriamycin challenge followed by 0.05 ml saline, thereby muting any local concentration effects upon Adriamycin toxicity. Concerning bicarbonate, a dose-ranging experiment will be necessary to firmly displace this antidote from clinical consideration. Nonetheless, we feel that our single-dose bicarbonate results were sufficiently negative to cast serious doubt on the future role of this antidote in the treatment of clinical Adriamycin extravasations.

In conclusion, we have been able to quantitate the limited effectiveness of several local corticosteroid therapies in the prevention of doxorubicin skin toxicity. High-dose doxorubicin infiltrations in this study remained relatively resistant to local corticosteroids. The antidotal role of corticosteroids appears to involve a local mechanism, since high-dose systemic therapy did not significantly alter toxicity. Furthermore, the consistent superiority of the lower corticosteroid doses and the nonadditive effects of the combination therapies confirm the limited role for corticosteroids in the management of local doxorubicin toxicity in the mouse. Thus different, more effective local antidotes must be sought. In this regard, several pharmacologic avenues appear promising: (1) Blockade of potential histaminergic and catecholamine mediators [4]; and (2) use of antioxidants [10] or complexing agents. These are currently being evaluated in our laboratory.

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References

1 Bachur NR (1975) Adriamycin (NSC-123127) Pharmacology. Cancer Chemother Rep [3] 6:153–158
2 Barlock AL, Howser DM, Hubbard SM (1979) Nursing management of adriamycin extravasation. Am J Nurs 79:94–96
3 Bowers DG, Lynch JB (1977) Adriamycin extravasation. Plast Reconstr Surg 61:86–92
4 Bristow MR, Billingham ME, Daniels JR (1979) (Abstract 477) Histamine and catecholamines mediate adriamycin cardiotoxicity. Proc Am Assoc Cancer Res/ASCO 20:118
5 Brunner MJ, Riddel JM Jr, Best WR (1951) Cutaneous side effects of ACTH, cortisone and pregnenolone therapy. J Invest Dermatol 16:205–210
6 Cohen MH (1979) Amelioration of adriamycin skin necrosis: An experimental study. Cancer Treat Rep 63:1003–1004
7 DiMarco A, Zunino F, Silvestrini R, et al (1971) Interaction of some daunomycin derivatives with deoxyribonucleic acid and their biological activity. Biochem Pharmacol 20:1323–1328
8 Dorr RT, Alberts DS, Chen HSG (1980) Experimental model of doxorubicin extravasation in the mouse. J Pharmacol Methods 4 (4):321–334
9 Freireich EJ, Gehan EA, Rall DP, Schhmidt LH, Skipper HE (1966). Qantitative comparison of toxicity of anticancer agents in mouse, rat, hamster, dog, monkey and man. Cancer Chemother Rep 50:219–244
10 Myers CE, McGuire W, Young R (1976) Adriamycin: Amelioration of toxicity by alpha-tocopherol. Cancer Treat Rep 60:961–962
11 Reilly JJ, Neifeld JP, Rosenberg SA (1977) Clincal course and management of accidental adriamycin extravasation. Cancer 40:2053–2056
12 Rudolph R, Stein RS, Patillo RA (1976) Skin ulcers due to adriamycin. Cancer 38:1087–1094
13 Rudolph R, Suzukim, and Luce JK (1979) Experimental skin necrosis produced by adriamycin. Cancer Treat Rep 63:529-537
14 Uitto J, Teir H, Mustakallio KK (1972) Corticosteroid-induced inhibition of the biosynthesis of human collagen. Biochem Pharmacol 21:2161–2167
15 Zweig JI, Kabakow B (1978) An apparently effective counter-measure for doxorubicin extravasation (Letter). JAMA 239:2116

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