PRODUCT MONOGRAPH

Tri-halogenated

PrULTRAVATE®
Halobetasol Propionate Topical Cream
0.05 % w/w

Halobetasol Propionate Topical Ointment
0.05 % w/w

Topical Corticosteroid

Bausch Health, Canada Inc.
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Canada

Date of Revision:
October 15, 2019

Submission Control No: 230873

NAME OF DRUG

PrULTRAVATE®
Halobetasol Propionate Topical Cream
0.05 % w/w

Halobetasol Propionate Topical Ointment
0.05 % w/w

THERAPEUTIC CLASSIFICATION

Topical Corticosteroid

INDICATIONS AND CLINICAL UES

ULTRAVATE® (halobetasol propionate) Cream and Ointment are high to super-high potency topical corticosteroids indicated for the relief of inflammatory manifestations of resistant or severe psoriasis and corticosteroid-responsive dermatoses. These products are not recommended for use in children.

CONTRAINDICATIONS

ULTRAVATE® (halobetasol propionate) Cream and Ointment are contraindicated in patients who are hypersensitive to halobetasol propionate, to other corticosteroids, or to any of the ingredients in these products.

ULTRAVATE® (halobetasol propionate) Cream and Ointment are contraindicated in viral diseases of the skin including herpes simplex, vaccinia and varicella. They are also contraindicated in untreated bacterial, tubercular and fungal infections involving the skin.

WARNINGS

Use in Pregnancy
There are no clinical trials of ULTRAVATE® in pregnant women. Therefore, this product should be used during pregnancy only if the potential benefit justifies the potential risk to the fetus.

Corticosteroids have been shown to be teratogenic and embryotoxic in laboratory animals at low doses when administered systemically. Some corticosteroids have been shown to be teratogenic after topical application. Halobetasol propionate has been shown to be teratogenic in rats and rabbits at low doses. The human topical dose of ULTRAVATE ® was embryotoxic in rabbits.

Nursing Mothers
Systemically administered corticosteroids appear in human milk and can suppress growth, interfere with endogenous corticosteroid production, or cause other adverse effects. It is not known whether topical administration of corticosteroids could result in sufficient systemic absorption to produce detectable quantities in human milk. Because many drugs are excreted in human milk, caution should be exercised when administering ULTRAVATE ® to a nursing woman.

ULTRAVATE® (halobetasol propionate) Cream or Ointment are not to be used with occlusive dressing. These products are not formulated for ophthalmic use and should not be used in or near the eyes.

ULTRAVATE® (halobetasol propionate) Cream or Ointment are for Dermatological use only.

PRECAUTIONS

General

In the presence of bacterial infections of the skin, an appropriate antibacterial agent should be used as primary therapy. If it is considered necessary, the topical corticosteroid may be used as an adjunct to control inflammation, erythema, and itching. If a favourable response does not occur within a few days to a week, the steroid should be discontinued until the infection has been adequately controlled.

Significant systemic absorption may occur when steroids are applied over large areas of the body. To minimize this possibility, when long term therapy is anticipated, interrupt treatment periodically or treat one area of the body at a time.

ULTRAVATE ® Ointment produced HPA axis suppression when used at recommended doses of 7 grams per day for one week in patients with psoriasis. These effects were reversible upon discontinuation of treatment.

Laboratory Tests
Patients receiving a large dose of a high potency topical steroid applied to a large surface area should be evaluated periodically for evidence of HPA axis suppression. This may be done by using the ACTH stimulation, A.M. plasma cortisol and urinary free-cortisol tests. Patients receiving super-potent corticosteroids should not be treated for more than 2 weeks at a time and it is recommended that only small areas be treated at any one time due to the increased risk of HPA suppression.

Prolonged use of topical corticosteroid products may produce atrophy of the skin and subcutaneous tissues. If this occurs, treatment should be discontinued.

Topical corticosteroids should be used with caution in patients with stasis dermatitis and other skin diseases associated with impaired circulation, hypersensitive patients and patients with

glaucoma.

Patients should be advised to inform subsequent physician of the prior use of corticosteroids.

Carcinogenesis, Mutagenesis
Long-term animal studies have not been performed to evaluate the carcinogenic potential of halobetasol propionate. Positive mutagenicity studies were observed in two genotoxicity assays. Halobetasol was positive in a Chinese hamster micronucleus test *in vivo* and in a mouse lymphoma gene mutation assay *in vitro.* In other genotoxicity tests including Ames/Salmonella assay, sister chromatid exchange test, chromosome aberration studies of germinal and somatic cells of rodents and in a mammalian spot test for point mutations, halobetasol propionate was not found to be genotoxic.

Use in Children

ULTRAVATE® (halobetasol propionate) Cream or Ointment should not be used in children. Because of the higher ratio of skin surface area to body mass, children are at greater risk for HPA axis suppression, glucocorticoid insufficiency after withdrawal of treatment and Cushing's syndrome while on treatment.

INFORMATION FOR PATIENTS

Patients using **ULTRAVATE®** (halobetasol propionate) Cream or Ointment should receive the following information:

1. This medication is to be used as directed by the physician and should not be used longer than the prescribed time period. It is for external use only. Avoid contact with eyes.

2. The medication should not be used for any disorder other than for which it was prescribed.

3. The treated skin area should not be bandaged or otherwise covered or wrapped so as to be occlusive.

4. Any signs of local adverse reactions should be reported to your physician.

ADVERSE REACTIONS:

A total of 1018 patient have been studied in **ULTRAVATE®** (halobetasol propionate) clinical trials, 596 received the ointment formulation, 341 received the cream formulation and 81 received both formulations. The incidence of adverse reactions with **ULTRAVATE®** cream and ointment were those commonly observed with topical corticosteroids.

The most frequently reported adverse reaction across all clinical trials with **ULTRAVATE®** Ointment was stinging (2%). Other adverse reactions related and probably related that were

reported at less than 1% were: burning, erythema, acne, skin atrophy, pruritus, leukoderma, telangiectasia, pustulation, dry skin, bruise, rash, lichenified dermatitis, paraesthesia, urticaria, and fungal infection.
The most frequently reported adverse reaction across all clinical trials with **ULTRAVATE®** Cream was also stinging (3%). Other adverse reactions related and probably related that were reported at less than 1% were: pruritus, burning skin, dry skin, leukoderma, erythema, skin atrophy, sore joint, and eye pressure.

The following adverse skin reactions have been reported with the use of topical corticosteroids and may occur more frequently with high potency corticosteroids such as **ULTRAVATE®** Cream and Ointment. These reactions are listed in an approximately decreasing order of occurrence: burning, itching, irritation, dryness, folliculitis, hypertrichosis, acneiform eruptions, hypopigmentation, perioral dermatitis, allergic contact dermatitis, maceration of the skin, secondary infection, skin atrophy, striae and miliaria. Systemic absorption of topical corticosteroids has produced reversible HPA axis suppression, manifestations of Cushing's syndrome, hyperglycemia, and glucosuria in some patients. In rare instances, treatment (or withdrawal of treatment) of psoriasis with corticosteroids is thought to have provoked the pustular form of the disease.

SYMPTOMS AND TREATMENT OF OVERDOSE

Topically applied **ULTRAVATE®** (halobetasol propionate) Cream or Ointment can be absorbed in sufficient amounts to produce systemic effects including reversible HPA axis suppression with the potential for glucocorticosteroid insufficiency after withdrawal of treatment. If HPA axis suppression is noted, withdraw the drug gradually by reducing the amount and frequency of application. Recovery of HPA axis function is generally prompt and complete upon discontinuation of topical corticosteroids. Infrequently, signs and symptoms of glucocorticosteroid insufficiency may occur requiring supplemental systemic corticosteroids.

DOSAGE AND ADMINISTRATION

Apply a thin layer of **ULTRAVATE®** (halobetasol propionate) Cream or Ointment to the affected skin and rub in gently and completely. Apply twice daily, or as directed by your physician. Treatment is to be discontinued when the dermatologic disorder is controlled.

Treatment with **ULTRAVATE®** (halobetasol propionate) Cream or Ointment should be limited to 50g per week. The duration of therapy should not exceed two weeks without patient re-evaluation. **ULTRAVATE®** (halobetasol propionate) Cream and Ointment are not to be used with occlusive dressing.

PHARMACEUTICAL INFORMATION

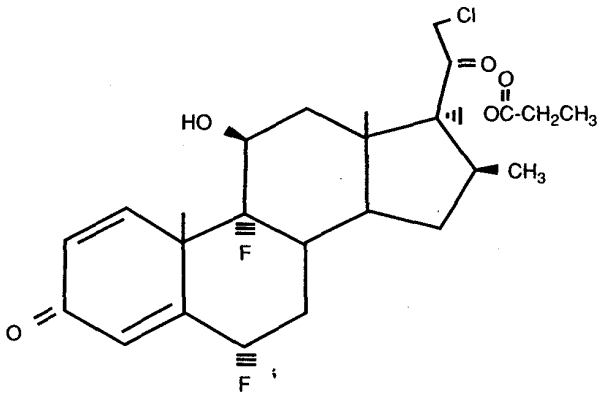
Drug Substance

USAN Name: Halobetasol Propionate

Trade Name: ULTRAVATE®

Chemical Name: 21-Chloro-6α,9-difluoro-11β-hydroxy-16β-methyl-pregna-1,4-diene-3-20-dione,17-propionate

Structural Formula:



Molecular Formula: $C_{25}H_{31}ClF_2O_5$

Molecular Weight: 484.973 g/mol

Physicochemical Properties

Description: Halobetasol propionate is a white crystalline powder or solid

Solubility: Insoluble in water, soluble in ethanol (37%), dimethyl sulfoxide (>50%), soluble in diesters (e.g. dibutyl adipate), slightly soluble in long-chain monoesters (isopropyl myristate).

Melting Point: The melting range is between 200°C and 216°C with decomposition.

COMPOSITION

Ointment

Halobetasol Propionate 0.05% w/w
Petrolatum
Propylene Glycol
Dehymuls E
Beeswax

Cream

Halobetasol Propionate 0.05% w/w
Water
Cetyl Alcohol
Steareth-21
Isopropyl Isostearate
Isopropyl Palmitate
Glycerin
Diazolidinyl Urea (Germall II)
Methylchloroisothiazolinone (and)
Methylisothiazolinone (Kathon CG)

STABILITY AND STORAGE RECOMMENDATIONS

ULTRAVATE® (halobetasol propionate) Cream and Ointment should be stored at controlled room temperature between 15°C and 25°C.

AVAILABILITY OF DOSAGE FORMS

ULTRAVATE® (halobetasol propionate) OINTMENT: **ULTRAVATE®** Ointment is packaged in 50g aluminum tubes providing 0.5 mg of halobetasol propionate per gram.

ULTRAVATE® (halobetasol propionate) CREAM: **ULTRAVATE®** Cream is packaged in 50g aluminum tubes providing 0.5 mg of halobetasol propionate per gram.

INFORMATION FOR THE PATIENT:

1. This medication is to be used as directed by the physician and should not be used longer than the prescribed time period. It is for external use only. Avoid contact with eyes.

2. This medication should not be used for any disorder than that for which it is prescribed.
3. The treated skin should not be bandaged or otherwise covered or wrapped so as to be

occlusive.

4. Any signs of local adverse reaction should be reported to your physician.

PHARMACOLOGY

Animals *in vivo*

Halobetasol propionate at concentrations ranging from 0.001% to 1%, was evaluated *in vivo* for anti-inflammatory activity following single topical application, using several animal models, including ultraviolet light (UVB) induced inflammation in guinea pigs, croton oil-induced inflammation in mice, rats and rabbits, and oxazolone-induced inflammation in mice and rats. The anti-inflammatory activity was demonstrated by reduction in erythema, edema and/or local temperature. Anti-proliferative activity of halobetasol propionate was examined by repeated topical and single local applications in hexadecane-stimulated epidermal hyperplasia in guinea pigs and cotton pellet granuloma in rats, respectively. Halobetasol propionate was evaluated at 1% concentration in the hexadecane model and at doses ranging from 0.2 mcg/pellet to 250 mcg/pellet in the cotton pellet granuloma model. Antiproliferative activity was demonstrated by reduction in epidermal hyperplasia or granuloma weight.

Atrophogenicity as demonstrated by hypotrophy of the epidermal and subepidermal tissue was examined following repeated topical application of 1%-2% halobetasol propionate to guinea pig skin.

In most of these tests, halobetasol propionate was compared to clobetasol propionate at similar concentrations or doses. In most studies, the test materials were prepared in solutions consisting of dimethylacetamide/acetone/ethanol or pyridine/water/diethylether.

In general, the results in these models indicate that the anti-inflammatory and anti-proliferative activity exhibited by halobetasol propionate at the concentrations tested is better than the low potency corticosteroid hydrocortisone and equivalent to or exceeding that of CBP. Halobetasol propionate exhibited atrophogenic effects that exceeded the effects of HC but were comparable to or exceeded that of the CBP, the ultra potent steroid.

Percutaneous absorption and excretion of halobetasol propionate was examined in rats, dogs and guinea pigs. Single, topical application of tritium labelled halobetasol propionate at 0.02% and 0.05% in cream, and 0.05% and 0.2% in ointment was made to the intact, occluded skin of rats and dogs for 6 hours. Halobetasol propionate 0.05% ointment was tested similarly in guinea pigs. These treatments resulted in absorption ranging from 2.6%-3.4% of the applied radioactivity in rats, 0.8%-3.6% in dogs, and 5.5%-6% in guinea pigs, as determined by recovery of radioactivity in the excreta. The percutaneous absorption of halobetasol propionate 0.05% ointment in guinea pigs is somewhat higher than that observed in rats and dogs.

In a separate study the influence of ethyl lactate or propylene glycol on percutaneous absorption of 0.05% halobetasol propionate from an ointment was examined using tritium labelled

halobetasol propionate. Test material was applied to the intact, occluded skin of guinea pigs for 6 hours and absorption was determined by recovery of radioactivity in the excreta. The results showed lower absorption than that noted in the above studies, ranging between 0.46%-1.45% of the applied radioactivity. The lower absorption probably can be related to the lower total dose and area of application compared to previously mentioned studies.

In rats, dogs and guinea pigs following topical application, more than 80% of the test material that was detected in the excreta was found in the feces.

Absorption, distribution, metabolism and excretion of halobetasol propionate was also examined following oral and parenteral administration of radiolabelled halobetasol propionate in rats, dogs and guinea pigs.

The excretory data obtained in rats, dogs and guinea pigs following oral and intravenous administration of 0.02 mg/kg of tritium labelled halobetasol propionate show that independently of the route of administration similar proportions of the radioactivity administered were eliminated in urine and feces, indicating complete absorption of the oral dose. In rats and guinea pigs, elimination was relatively fast and 90% or greater proportion of the administered radioactivity was detected in the excreta 168 hours after dosing. On the other hand, in the dog elimination was slower and less than 90% of the administered radioactivity was detected in the excreta during the same period.

In all 3 species, elimination was predominately via the feces as was apparent following percutaneous application. The absorption and elimination of doses higher than 0.02 mg/kg were also examined in rats and dogs. In rats the oral administration of a higher dose (1.0 mg/kg) was also completely absorbed and eliminated. In the dog, only 68% of the administered radioactivity was eliminated after the oral administration of a higher dose (0.1 mg/kg) suggesting lower absorption at the higher dose.

Halobetasol propionate was extensively metabolised by all three species. According to inverse isotope dilution analysis in the rat, dog and guinea pig, only 0.02% to 0.3% of the radioactivity detected in the urine was determined as intact halobetasol propionate. Tissue distribution of orally and intravenously (IV) administered halobetasol propionate to the rat showed that peak levels in blood, tissues and organs were detected at 4 hours. Very little retention (< 0.06% of dose) of radioactivity was noted in organs and tissues 168 hours after oral and IV dosing in rats.

Pharmacodynamics

Four trials comparing the vasoconstrictor activity of halobetasol propionate cream and ointment formulations or ethanolic solutions to various marketed topical corticosteroids or investigational formulations were undertaken. Two of the studies were done using the traditional McKenzie-Stoughton method or a modification of that method. In these studies, the test substance is allowed to remain on the skin for 16 hours and the test site graded at least at 18 hours following application. In the modified method, other time periods are also graded.

The other two studies used an Area Under the Curve (AUC) method, in which the test substance

remains on the skin for a much shorter time period, 6-8 hours. The degree of blanching was then evaluated at multiple times following the removal of the test substance.

Both studies using the McKenzie-Stoughton method showed halobetasol propionate solutions, ointment and cream, to exert an extremely strong vasoconstrictive effect, equal or superior to other corticosteroids classified in the ultra-potent category such as TEMOVATE™ or DIPROLENE™ cream or ointment formulations. In the two AUC studies, results were more variable, with high potency and mid-potency corticosteroids showing statistically equivalent rankings with the ultra-potent corticosteroids. See Table 1.

Table 1
Mean Score/Rank at 18 Hours

	McKenzie-Stoughton Methodology		Area Under Curve Methodology	
Drugs Studied	CG82/82*	46A50-0001	46R87-0001	DE118-006†
Halobetasol Propionate 0.05% Ointment	--	2.73[A]	2.6[A]	--
Halobetasol Propionate 0.05% Cream	--	2.23[A,B]	1.8[B,C]	6.71[B,C]
TEMOVATE 0.05% Ointment	--	2.43[A,B]	--	--
TEMOVATE 0.05% Cream	--	2.43[A,B]	--	9.54[A]
DIPROLENE 0.05% Ointment	--	2.20[B]	2.1[A,B,C]	
DIPROLENE 0.05% Cream	--	2.23[A,B]	--	
LIDEX 0.05% Cream	--	--	2.4[A,B]	
WESTCORT Ointment	--	1.10[C]	0.8[D]	
Halobetasol Propionate 0.02% Solution	--	--	1.4[C,D]	
Halobetasol Propionate 0.05% Solution	--	--	1.6[C]	
ELECON Cream	--	--	--	6.46[C]
MAXIVATE Cream	--	--	--	4.66[D]
KENALOG Cream	--	--	--	4.26[D,E]
ARISTOCORT Cream	--	--	--	3.00[E,F]
HYTONE Cream	--	--	--	2.71[F]
Hydrocortisone Ethanolic Solutions	11[B]	--		
Halobetasol Propionate Ethanolic Solutions	58[A]	--		
Clobetasol Ethanolic Solutions	56[A]	--		

*Overall reaction intensity at all periods measured
†Over the entire time period measured
Scores/ranks with the same letter in each column are not statistically significantly different from other (P □0 05)

Pharmacokinetics

Percutaneous systemic effects of 0.02% and 0.05% halobetasol propionate ointments and DERMOVATE™ ointment, containing 0.05% clobetasol 17-propionate as the active ingredient, were evaluated in six healthy male volunteers by assessing serum cortisol levels. The subjects ranged in age from 32 to 47 years (mean = 39.1 years). The ointments were applied, without an occlusive dressing, in dosages of 12 g once daily at 4 p.m. on two successive days (Days 2 and 3) to 2400 cm^2 skin surface of the trunk (5 mg ointment/cm^2) of six volunteers in a randomized cross-over comparative study. Serum cortisol levels at 8 a.m. were determined before (Days 1 and 2), during (Days 3 and 4) and after (Days 8 and 9) application of the above-mentioned

ointments, by radioimmunoassay, using the COAT-A-COUNT™ kit (Diagnostics Products Corp., Los Angeles). The normal range of serum cortisol at 8 a.m. for this assay is 7-32 mcg/dl.

All three ointments tested produced a reversible lowering of the serum cortisol levels. Clobetasol propionate 0.05% ointment and halobetasol propionate 0.05% ointment produced closely similar and statistically significant suppression of morning serum cortisol levels. However, no individual level was below the lower limit of normal for this assay. During the post-treatment phase, mean serum cortisol levels reached 93% and 98% of the baseline values for clobetasol 0.05% and halobetasol propionate 0.05% ointments, respectively. Halobetasol propionate 0.02% ointment showed a trend toward lowered cortisol levels, but the change in values from baseline was not statistically significant. See Table 2.

Table 2
Mean Natural Logarithms of Serum Cortisol Levels
(Mean Values mcg/dl)
Ointment
Normal Range, 7-32 mcg/dl

Period	**Halobetasol Propionate 0.05%**		**Clobetasol 0.05%**		**Halobetasol Propionate 0.02%**	
	Mean	(Mean)	Mean	(Mean)	Mean	(Mean)
	LN	(□g/dl)	LN	(□g/dl)	LN	(□g/dl)
Baseline	2.7111	(15.04)	2.7394	(15.48)	2.7584	(15.77)
During Treatment	2.3921	(10.94)	2.4619	(11.72)	2.5897	(13.33)
Post Treatment	2.6907	(14.74)	2.6672	(14.40)	2.7044	(14.95)

In an open-label study, halobetasol propionate was administered topically to five men and five women with localized psoriasis to determine systemic effects. The age range was 28 to 76 years (mean = 38 years). On the two pre-treatment days and on Days 11-15 post-treatment, salicylic acid 5% ointment was applied twice daily to remove scales. Halobetasol propionate 0.05% ointment, 2.5 g was applied twice a day for 10 days to psoriatic lesions. One 8 a.m. morning plasma cortisol level was taken before treatment; three during treatment; and one was obtained post-treatment, 5 days after the end of therapy.

A global assessment of therapeutic effect was made at the end of 10 days' treatment with halobetasol propionate 0.05% ointment using a 5-point scale where 1 = healed, 2 = marked improvement, 3 = moderate improvement, 4 = poor improvement, and 5 = no improvement.

Plasma cortisol levels were measured by a radioimmunoassay (RIA) where the normal range was 5.8 to 36.4 mcg/dl when obtained at 8 a.m. One patient had blood drawn between 9:15 a.m. and 10:15 a.m. The other subjects had blood drawn at the correct time.

Results in 9 patients did not show any statistically significant differences between the 8 a.m. plasma cortisol values obtained before or after treatment with those recorded during treatment with halobetasol propionate 0.05% ointment. See Table 3.

Table 3
Mean Plasma Cortisol Levels

(*mc*g/dl)
Normal Range, 5.8-36.4 *mc*g□/dl

Period	Subjects	Mean	S.D.	Min	Max
Pre-Treatment	9	25.3	8.6	11	38
During Treatment	9	21.5	5.9	11	31
Post Treatment	9	23.8	7.1	13	35

Following ten days' treatment with halobetasol propionate 0.05% ointment, 8 of 10 patients were healed and 2 showed marked improvement in their psoriatic lesions. Folliculitis was reported at the site of application in one patient.

In an open-label evaluation of the effects on the hypothalamic-pituitary-adrenal (HPA) axis, 7 grams per day of halobetasol propionate 0.05% ointment was applied to psoriatic plaques of six men and one woman whose ages ranged from 20-65 years (mean = 47 years).

Halobetasol propionate 0.05% ointment was applied to lesions covering up to 30% of their body surface area twice daily for seven days. Three baseline cortisol plasma levels, two during treatment and two post-treatment were determined by radioimmunoassay, using the COAT-A-COUNT KIT (Diagnostics Products Corp., Los Angeles). Two consecutive 24-hour urines were collected pre-treatment and two during treatment to determine 17-hydroxycorticoid excretion.

Physical examination and clinical laboratory tests were done pre- and post-treatment.

The normal range of plasma cortisol values was 5-25 mcg/dl. The mean baseline plasma cortisol level was 18.9 +/- (S.D.) mcg/dl, the mean during treatment cortisol level was 15.4 +/- 5.0 mcg/dl, and the mean post-treatment cortisol level was 19.6 +/- 7.8 mcg/dl. None of the mean plasma cortisol levels were suppressed below 9.0 mcg/dl (lower limit of normal, 5 mcg/dl) and the lowest individual value was 5 mcg/dl.

The normal range of urinary 17-hydroxycorticoids is 4-14 mg/24 hr for males and 2-10 mg/24 hr for females. The mean baseline excretion for the males was 6.6 +/- 1.4 (S.D.) mg/24 hr and the mean baseline excretion for the female patient was 3.5 mg/24 hr. The mean 17-hydroxycorticoid excretion during treatment was 5.1 +/- 1.4 (S.D.) mg/24 hr for the males and 3.0 mg/24 hr for the female. None of the mean or individual urinary 17-hydroxycorticoid values for males or for the female were suppressed below the lower limit of the normal range. See Table 4.

Table 4

Mean Period Cortisol and 17-OH Corticoid Levels											
	Plasma Cortisol (□g/dl) (Normal Range, 5-25 □g/dl)					**17-OH Corticoids (mg/24hr)** (Normal Range 4-14 mg/24hr 2-10 □g/dl)					
						Males				Female	
Period	N	Mean	S.D.	Min	Max	N	Mean	Min	Max	N	Mean
Baseline	7	18.9	4.1	13	24	6	6.6	4	8	1	3.5
During Treatment	7	15.4	5.0	9	21.5	6	5.1	4	7.5	1	3.0
Post Treatment	7	19.6	7.8	11	35.5						

Results from the battery of laboratory tests conducted both pre- and post-therapy were considered to be within normal limits for psoriatic patients.

One patient developed urticaria on Study Day 13 (Post Treatment Day 3), which subsided within a few hours. The subject was patch tested with the treatment medication, ointment vehicle and petrolatum on Study Day 14 without further reaction. Her post-treatment plasma cortisol levels were 31 and 40 □g/dl on Study Days 14 and 15, respectively, indicating a normal response to a stressful situation. Two additional patients reported mild irritation or itching after the first one or two applications on excoriated areas.

It was concluded that halobetasol propionate 0.05% ointment at a level of 7 g/day results in a slight suppression of the plasma cortisol which returns to or exceeds the baseline value within 5 days after the end of treatment. All values were within normal limits for morning plasma cortisol levels and, therefore, treatment with 7 g/day of halobetasol propionate 0.05% ointment is considered not to cause significant adrenal suppression.

A randomized two-way cross-over study was performed in six healthy male volunteers age 30-46 years (mean = 38.1) to determine systemic absorption of halobetasol propionate 0.05% cream and ointment formulations. Subjects received halobetasol propionate, 0.05 mg, 0.1 mg and 0.25 mg orally in ethanolic solution in the first phase of the study. In the second phase, the same subjects received ten grams of ointment or cream, equivalent to 5 mg halobetasol propionate, on two different occasions separated by a two-week wash-out period applied to 2,000 cm^2 of normal skin on the trunk. This medication was left on unoccluded for 12 hours. Urine was collected over 96 hours and analyzed for apparent halobetasol propionate by radioimmunoassay. Excretion of apparent halobetasol propionate into the urine was relatively slow with both cream and ointment formulations. The major portion of apparent halobetasol propionate appeared in the urine within 48-72 hours of application. Over the entire 96-hour collection period a mean of 725 +/- 420 ng (S.D.) for cream and 951 +/- 310 ng (S.D.) for the ointment had been excreted into the urine. In the oral study a mean of 0.73% (range, 0.55% - 0.90%) was found in the urine as apparent halobetasol propionate. This represents about 2.0% and 2.6% of the applied 5 mg of active halobetasol propionate in cream and ointment, respectively.

No adverse reactions were reported.

It was concluded that halobetasol propionate 0.05% is absorbed to a similar extent from the cream and ointment formulations. The extent of percutaneous absorption lies within the range of that reported for other topical corticosteroids such as triamcinolone acetonide (0.6% - 2.3%), diflorasone diacetate (1.1%) and halometasone (1.3%) in cream formulations and halomethasone (6.5%) in ointment formulation.

Controlled Clinical Trials

Halobetasol propionate cream and ointment formulations were evaluated in thirteen (13) well-controlled clinical trials (9 using the ointment and 4 with the cream), seven active control, four paired comparison vehicle control and two parallel group vehicle control in patients with plaque psoriasis, chronic eczema and atopic dermatitis. A total of 937 patients received halobetasol propionate. In the active control studies, halobetasol propionate was as effective as Dermovate. In the paired comparison and parallel group studies, halobetasol propionate was statistically and clinically superior to the vehicles.

TOXICOLOGY

Animals

Acute Toxicity Studies on Halobetasol Propionate (as pure compound)

Rat
The acute oral (LD_{50}) toxicity of halobetasol propionate was determined in rats. Halobetasol propionate was administered as a suspension in 0.05% CMC at oral dose levels of 1000, 3000, and 6000 mg/kg study groups of 5/sex/group. There was reduction of spontaneous motility in the high dose group lasting more than 6 hours, with no effects after 24 hours. The acute oral LD_{50} determined was greater than 6000 mg/kg (practically nontoxic).

Hamster
The acute oral (LD_{50}) toxicity of halobetasol propionate was determined in Chinese hamsters. Halobetasol propionate was administered orally as a suspension in 0.5% CMC at a dose level of 6000 mg/kg to a study group of 5/sex. Three (3) deaths (1 male/2 female) occurred during the study with hyperreflexia and dyspnea observed after dosing. No symptoms were observed at 24 hours. The acute oral (LD_{50}) was greater than 6000 mg/kg.

Dog
The acute oral (LD_{50}) toxicity of halobetasol propionate was determined in beagle dogs. Halobetasol propionate was administered via gelatin capsules at an oral dose level of 3000 mg/kg to a study group of 1/sex. Emesis occurred in the male after dosing. General toxicity signs were an initial marked but a reversible decrease in eosinophils, with decreased cortisol values observed during the 2-week study. There were no deaths. The acute oral (LD_{50}) of halobetasol propionate was greater than 3000 mg/kg.

Acute Toxicity Studies on 0.05% Halobetasol Propionate Cream

Rabbit
The ocular irritation potential of 0.05% halobetasol propionate cream or cream vehicle was determined in rabbits. 0.1 g of the test material or vehicle was instilled into the conjunctival eye sac in the eyes of 6 rabbits. After 1 minute, the eyes of 3 rabbits were washed out and all animals graded for irritation at 1, 24, 48 and 72 hours, and at 6 and 7 days. No irritation was observed. 0.05% halobetasol propionate cream was not classified as an eye irritant.

Rabbit
The primary skin irritation potential of 0.05% halobetasol propionate cream was determined in rabbits. Topical applications of 0.5 ml of the test material was applied to the intact or abraded skin sites of rabbits under 24-hour patch occlusion. A primary skin irritation index of 1.21 (mild irritation) was obtained and 0.05% halobetasol propionate cream was not classified as a primary skin irritant.

Rabbit
The dermal irritation potential of 0.05% halobetasol propionate cream or cream vehicle after repeated application was determined in rabbits. The test materials were topically applied at a dose level of 3.0 g to the depilated backs of rabbits (5/sex/group) under 24-hour occlusion daily for 5 days. The fifth dressing was removed after 8 hours. Minimal irritation was observed during the first 2 days and none thereafter. There was a slight body weight increase. There were no mortalities observed during the study.

Rabbit
The dermal irritation potential of 0.05% halobetasol propionate cream, after repeated application for 5 days and a follow-up period of 3 days, was determined in rabbits. The test material was topically applied at 0.5 g under 24-hour patch occlusion to the backs of rabbits (3/sex) for days 1-4, with the fifth dressing removed after 8 hours. Moderate irritation was observed during the study which appeared to clear after 3 days of non-treatment. There was a slight body weight reduction. No mortalities were observed during the study.

Acute Toxicity Studies on 0.05% Halobetasol Propionate Ointment

Rat
The acute oral (LD_{50}) toxicity of 0.05% halobetasol propionate ointment was determined in rats. 0.05%, halobetasol propionate ointment was orally administered undiluted at a dose level of 7.5 mg/kg to a study group of 5/sex. No mortalities or other signs of toxicity were observed. The acute oral (LD_{50}) of 0.05% halobetasol propionate ointment was greater than 7.5 mg/kg.

Rabbit
The ocular irritation potential of 0.05% halobetasol propionate ointment or vehicle was determined in rabbits. 0.1 g of the test material or vehicle was instilled into the conjunctival eye sac in the eyes of 6 rabbits. After 1 minute, the eyes of 3 rabbits were washed out and all animals graded for irritation at 1, 6, 24, 48, and 72 hours, and at 6 and 8 days. No irritation was observed. 0.05% halobetasol propionate ointment was not classified as an eye irritant.

Rabbit
The ocular irritation potential of 0.05% halobetasol propionate ointment was determined in rabbits. 0.1 ml of the test material or vehicle was instilled into the conjunctival eye sac in the eyes of 8 rabbits. After 2 seconds, the eyes of 2 rabbits were washed out, and all animals were graded for irritation at 24, 48, and 72 hours and at 7 days. Slight irritation was observed. 0.05% halobetasol propionate ointment was not classified as an eye irritant.

Rabbit
The primary skin irritation potential of 0.05% halobetasol propionate ointment was determined in rabbits. Topical applications of 0.5 ml of the test material was applied to the intact or abraded skin sites of rabbits under 24-hour patch occlusion. A primary skin irritation index of 0.42 (minimal irritation) was obtained and 0.05% halobetasol propionate ointment was not classified as a primary skin irritant.

Rabbit
The dermal irritation potential of 0.05% halobetasol propionate ointment or ointment vehicle after repeated application was determined in rabbits. The test materials were topically applied at a dose level of 5.0 g to the depilated backs of rabbits (3/sex/group) under 24-hour occlusion daily for 5 days. The fifth dressing was removed after 8 hours. There was a slight body weight decrease. There were no mortalities or irritation observed during the study.

Acute Toxicity Studies on 0.05% Halobetasol Propionate Solution

Rabbit
The ocular irritation potential of 0.05% halobetasol propionate solution or solution vehicle was determined in rabbits. 0.01 ml of the test material was instilled into the conjunctival eye sac in the eyes of 6 rabbits. After 1 minute, the eyes of 3 rabbits were washed out and all animals graded for irritation at 6, 24, 48, and 72 hours and at 6 and 7 days. Minimal irritation was observed. 0.05% halobetasol propionate solution would not be classified as an eye irritant.

Rabbit
The dermal irritation potential of 0.05% halobetasol propionate solution or vehicle after repeated application for 5 days and a follow-up period of 3 days was determined in rabbits. The test material was topically applied at 0.5 ml under 24 hr. patch occlusion to the backs of rabbits (3/sex) for days 1-4, with the fifth dressing removed after 8 hrs. No irritation and no mortalities were observed during the study.

Sensitization Studies on 0.05% Halobetasol Propionate Cream, Ointment, and Solution

Guinea Pig
The sensitization potential of 0.05% halobetasol propionate cream or vehicle was determined in guinea pigs. To groups of 10/sex/group, topical applications of the test materials were made under occlusion on sites injected with adjuvant. At the second induction during week 2, the test materials were reapplied under occlusion on the sites pretreated with a 10% sodium lauryl sulfate. Challenge applications to test and control study groups were made 14 days later. Under the conditions of the modified maximization test in guinea pigs, 0.05% halobetasol propionate

cream was non-sensitizing.

Guinea Pig
The sensitization potential of 0.05% halobetasol propionate ointment or vehicle was determined in guinea pigs. To groups of 10/sex/group, topical applications of the test materials were made under occlusion on sites injected with adjuvant. At the second induction during week 2, the test materials were reapplied under occlusion on the sites pretreated with 10% sodium lauryl sulfate. Challenge applications to test and control study groups were made 14 days later. Under the conditions of the modified maximization test in guinea pigs, 0.05% halobetasol propionate ointment was non-sensitizing.

Guinea Pig
The sensitization potential of 0.05% halobetasol propionate solution or solution vehicle was determined in guinea pigs. To test or vehicle control groups of 10/sex/group, the first induction phase consisted of separate 0.1 ml i.d. injections of the adjuvant, the 0.05% halobetasol propionate solution or vehicle, and the 0.05% halobetasol propionate solution or vehicle in combination with adjuvant. The second induction was followed one week later with a 0.2 ml topical application made of the test material or vehicle under occlusion to application sites pretreated the day before with 10% sodium lauryl sulfate. Challenge applications of the test solution or vehicle solution were made after 2 weeks, and a second challenge 2 weeks later.

Under the conditions of this modified maximization study, 0.05% halobetasol propionate solution was non-sensitizing in guinea pigs. Mild to moderate dermal sensitizing responses were, however, associated with a vehicle solution excipient.

Guinea Pig
The sensitization potential of 0.05% halobetasol propionate solution or solution vehicle was determined in guinea pigs. To test or vehicle control groups of 10/sex/group, the first induction phase consisted of separate 0.1 ml i.d. injections of the adjuvant, the 0.05% halobetasol propionate solution or vehicle, and the 0.05% halobetasol propionate solution or vehicle in combination with adjuvant. The second induction was followed one week later with a 0.4 ml topical application made of the test material or vehicle under occlusion to application sites pretreated the day before with 10% sodium lauryl sulfate. Challenge applications of the test solution or vehicle solution were made after 2 weeks.

Under the conditions of this Magnusson and Kligman maximization study, 0.05% halobetasol propionate solution was non-sensitizing in guinea pigs.

Subacute Toxicity Studies on Halobetasol Propionate (as pure compound and ointment)

Oral – Rat
The subacute oral toxicity potential of halobetasol propionate was determined in rats. Halobetasol propionate was administered orally by gavage at dose levels of 0.01, 0.1 and 1.0 mg/kg/day (in 0.5% CMC) at a dose volume of 10 ml/kg to study groups of 10 or 15/sex/group for 3 months. A similar test group of animals received the vehicle, 0.5% CMC, and served as a control. At termination of treatment, a one-month recovery period was conducted

involving 5/sex/group from the 0.1 and 1.0 mg/kg dose groups and of the vehicle control group.

During the study, 1 male rat of the high dose group was found dead (Day 40). Test material related changes included piloerection, reduction of spontaneous activity, muscle hypotonia, and loss of hair in all dose groups. These symptoms except for hair loss were, in general, reversible. Body weights were markedly reduced in all dose levels, with some improvement in weight gain during recovery. Food consumption values were similarly reduced during the study. Clinical chemistry effects included increased but reversible ALAT (SGPT) values in the mid and high dose groups. Hematological changes were reduced packed cell volume and dose related lymphopenia and neutrophilia which were reversible during recovery. Urinalysis revealed blood and protein in the urine which were present after the recovery. Organ weights of the liver, heart and kidney were increased, with decreased adrenal, spleen and thymus weights. The organ weight changes in the mid and high dose groups were generally correlated with microscopic evaluations.

Histopathological examination revealed expected dose related corticosteroid related changes including ballooned hepatocytes, with marked adrenal, thymic and splenic atrophy, and respiratory tract infections. Most changes were reduced or reversible after recovery. On the basis of numerous observed effects including body weight reduction, minimal liver effects, reduced lymphatic elements in axillary lymph nodes in the 0.01 mg/kg dose group, the no effect level is less than 0.01 mg/kg/day.

In a study performed in parallel to the above, the reference control steroid, clobetasol propionate, was similarly evaluated for subacute oral toxicity potential at a dose level of 0.1 mg/kg/day to a study group of 15/sex for 3 months, followed by a one-month recovery period. Test material effects were similar in type and comparable to the steroid related effects reported on halobetasol propionate (at 0.1 mg/kg). The effects produced were reversible.

Oral – Dog

The subacute oral toxicity potential of halobetasol propionate was determined in beagle dogs. Halobetasol propionate was administered orally via gelatin capsules to dogs at dose levels of 0.01, 0.03, and 0.1 mg/kg/day to study groups of 3/sex/group (low and mid dose) and 6/sex/group (high dose) for 3 months. The test material was administered as a premix with lactose. A control group of 3/sex was administered 10 mg/kg/day of lactose in gelatin capsules.

At termination of treatment, a 1-month recovery period was conducted involving 3/sex/group of the high dose group dogs.

No mortalities related to test material administration were observed during the study. Changes associated with administration of halobetasol propionate included general signs of soft, bloody feces in the high dose group, diarrhea, decreased body weight in the high dose, no changes in food consumption, increased but reversible ALAT levels in mid and high dose levels, increased alkaline phosphatase values at high dose, marked with reversible dose dependent increase of an alpha 3 globulins with a slight decrease in other globulins, decreased but slightly reversible dose related cortisol levels, increased but reversible triglycerides in the high dose group, hematological changes included decreased hemoglobin, rbc, and hematocrit in the high dose

group, dose related lymphopenia and neutrophilia, with eosinopenia, and bacteria in urine (high dose). Organ weight changes included reduced adrenals, non or minimally reversible, increased liver, increased spleen and kidneys (high dose). Histopathological examination revealed marked atrophy of the adrenal and lymphatic tissues, ballooning and/or vacuolization of the hepatocytes, and inflammation of the urogenital tract. Most changes were dose dependent or occurring at the high dose and were reduced or reversible following recovery other than the adrenals. There was no ocular, auditory, or neurological changes observed during the study.

In the high dose group (0.1 mg/kg), an increased incidence of slight deviations in repolarization (diphasic or notched T-wave, reversion of polarity) was detected in 11 of 12 animals. Alternations continued through the one-month recovery period in 3 of 6 animals.

Clobetasol propionate, the reference steroid, was similarly evaluated in a 3-month subacute oral toxicity study at a dose level of 0.03 mg/kg/day (as a lactose premix) in gelatin capsules to a study group of 3 males and female dogs. 2/3 male dogs showed variations of the T-wave polarity.

The dogs receiving oral doses of both steroids reacted to the treatment by developing stress along with a suppression of the lymphatic system and apparently increased susceptibility to infectious diseases in a dose related manner. The deviation in the form of the T-Waves was therefore not considered to indicate drug related cardiomyopathy.

In the absence of cardiac lesions found at necropsy, the absence of electrocardiographic changes other than non-specific T-wave changes, presence of toxic effects that can produce non-specific T-wave effects in dogs, the electrocardiographic findings are not considered indicative of cardiotoxicity.

Other results of this study revealed comparable steroid related effects of clobetasol propionate to that of halobetasol propionate, other than a decrease in body weight present in the clobetasol propionate group and in liver changes which were more pronounced in the dogs treated with halobetasol propionate.

Dermal – Rat

The subacute dermal toxicity potential of halobetasol propionate ointment was determined in rats for 3 months. The test material was topically administered to study groups of 6/sex/group for 3 months at daily dose levels of 0.05%, 0.1% and 0.2% halobetasol propionate ointment at a dose volume of 400 mg/kg of ointment (equivalent to 0.2, 0.4, and 0.8 mg/kg/day of halobetasol propionate). Control groups were similarly treated with the vehicle ointment and 0.05% clobetasol propionate ointment served as a reference control. Separate study groups of 4/sex/group of rats were also treated with test or control ointments for 3 months and were maintained on study for 1 month without treatment for recovery.

During the study there were no treatment related mortalities observed in the rats. Test material related changes observed during the study included skin changes (red/blue colouration), decreased body weights in the males, decreased food consumption, increased ALAT (SGPT) levels, hematological changes consisting of lymphopenia and neutrophilia, slight anemia,

hematuria, and hypo gamma globulinemia, the organ weight of the adrenals and thymus were reduced. At necropsy, the condition of the animals was considered cachectic. Histopathological examination revealed expected steroid related changes including epidermal thinning, distended macrophages in the lungs, vacuolated and distended hepatocytes of the liver, moderate adrenocortical atrophy and atrophy of the lymphoid organs, and hyperplasia of the islets of Langerhans in the pancreas.

The changes observed were, in general, dose related and reversible or reduced by the end of the recovery study. Similar changes, which were less severe, were observed in the 0.05% clobetasol propionate ointment test groups.

Dermal – Dog
The subacute dermal toxicity potential of halobetasol propionate ointment was determined in dogs following topical administrations for 3 months. The test material was topically administered to study groups of 6/sex/group for 3 months at daily dose levels of 0.05%, 0.1%, and 0.2% halobetasol propionate ointment at a dose volume of 400 mg/kg/day of ointment (equivalent to 0.2, 0.4, and 0.8 mg/kg/day of halobetasol propionate). All application sites were occluded daily for 6 hours. Control groups were similarly treated with the vehicle ointment, and 0.05% clobetasol propionate ointment served as a reference control. After 3 months, study groups of 2/sex/group of dogs were maintained on study for 1 month without treatment for recovery.

During the study, 1 dog in the mid and high dose was killed due to severely infected skin wounds. Another dog was killed in the low dose due to self-inflicted injury. Test material related changes observed during the study included skin lesions, erythema, hair loss, papules/scabs and abscesses, no remarkable body weight or food consumption changes, increased ALAT, alkaline phosphatase, and alpha 2 globulins, decreased cortisol, with hypogammaglobulinemia, dose related anemia in the halobetasol propionate treated animals, hematological changes included increased ESR, lymphopenia and neutrophilia, eosinopenia, hematuria and hemoglobinuria, with no other changes observed during ophthalmic, auditory or electrocardiac examinations. Organ weight changes revealed decreased adrenals and increased liver weights. Histopathological examination revealed expected steroid related changes consisting of adrenocortical and marked thymic atrophy, cytoplasmic vacuolation of the hepatocytes, dermal changes were thinning of the stratum corneum and focal panniculus myopathy of the skin, and an increased incidence of granulomatous reactions associated with helminth larvae in the lungs, liver and lymph nodes.

Most changes observed were dose related and reversible or reduced following a 1-month recovery period. Comparable changes were observed in the 0.05% clobetasol propionate ointment dose group.

Reproductive Studies on Halobetasol Propionate

Fertility and Reproduction

Rat
Halobetasol propionate was studied in rats for potential effects on fertility, general reproductive pregnancy and prenatal development. Halobetasol propionate was orally administered at daily dose levels of 0.008, 0.020, and 0.050 mg/kg/day (as a lactose premix suspended in CMC) to study groups of 20 male and 20 female rats. Similarly, groups were treated with the vehicle, and clobetasol propionate was dosed at 0.05 mg/kg/day and served as a reference control. The males were treated for 60 days and the females for 14 days prior to mating until termination of a 12-day mating period. Treatment was continued until Day 15 of pregnancy.

The results from the reproductive study on halobetasol propionate included males and females of the dose groups reacting to the treatment by a reduction in body-weight gain and, to a lesser extent, food consumption in a dose-related fashion. Fertility and reproductive performance remained unchanged. At 0.050 mg/kg, some tendency to an increased pre-implantation and, in particular, post-implantation rate of embryonic death was noted. In relation to maternal toxicity, the average weight of the fetuses examined near term was significantly diminished in the three experimental groups, in a dose-related fashion. Parallel to the reduction in fetal body weight, there were indications of delay of skeletal maturation at 0.020 mg/kg and, in particular, 0.050 mg/kg. Occasional anomalies and/or malformations were recorded for all groups. These included two fetuses from one litter of the 0.008 mg/kg dose group and one fetus of the 0.050 mg/kg dose group showing an omphalocele being consistent with a disturbance of ventral closure of the embryo. A "visceral anomaly" dilatation of the pelvic cavity of kidneys was also found in two fetuses from the litter the omphalocele was recorded for. Abnormal ossification of sternebrae was observed in all groups, including the vehicle control.

Based on the results of this study on halobetasol propionate, neither fertility nor general reproductive performance were impaired in the rat under the experimental conditions employed.

The treatment of rats with 0.050 mg/kg of the reference compound, clobetasol propionate, produced results similar to those recorded for halobetasol propionate at doses of 0.020 mg/kg (body weight of adults) or 0.050 mg/kg (fetal body weight). The live fetuses examined near term exhibited one instance each of cleft palate and dilatation of renal pelvic cavity as well as three instances of abnormal ossification of sternebrae.

Teratology

Oral – Rat
Halobetasol propionate was administered orally at dose levels of 0.008, 0.040, and 0.100 mg/kg/day from days 6 to 15 of pregnancy to study groups of 24 pregnant female rats each. Similar study groups were dosed with the vehicle (0.5% CMC), and a reference control group received clobetasol propionate at 0.100 mg/kg.

The results of this teratology study indicated a high degree of embryotoxicity associated with clobetasol propionate. Halobetasol propionate was, in contrast to the reference compound, devoid of an embryotoxic activity in the rat under the experimental conditions. At 0.040 and 0.100 mg/kg of halobetasol propionate, omphalocele and cleft palate were observed.

Cleft palate was observed to be slightly higher (2.1%) in the high dose group compared to the clobetasol propionate treated group. The omphalocele malformation was at a comparable rate in all drug treated groups. The marginal teratogenic action of halobetasol propionate as well as of clobetasol propionate was associated with maternal toxicity and fetotoxicity.

Oral – Rabbit
In a preliminary teratology study, halobetasol propionate was orally administered at dose levels of 0.01 and 0.04 mg/kg to study groups of 6 pregnant rabbits each from days 6 to 18 of pregnancy. In a similar study group of pregnant rabbits, clobetasol propionate was dosed at 0.02 mg/kg and served as the reference control.

The results from the screening teratology study in rabbits included dose related maternal and embryotoxicity. Halobetasol propionate was teratogenic in the rabbit at a dose producing maternal toxicity (0.01 mg/kg). The type of malformation observed was predominantly cleft palate. Omphalocele and malrotation of the fore-limbs were also observed. Clobetasol propionate, which was used as a reference compound, also produced cleft palate and malrotation of fore-limbs at a similar incidence. The dose administered (0.02 mg/kg) also induced maternal toxicity.

Failure of palatal closure is well known to occur in the rabbit fetus after treatment of the dams with a variety of corticosteroids. The mechanism of teratogenic action is considered to involve a disturbance of collagen synthesis in the embryonic tissues at a critical phase of development.

No instance of cleft palate was recorded to occur in a cumulative control population of the breed of rabbits used for this experiment.

The teratogenic action of halobetasol propionate as well as of the reference compound clobetasol propionate is attributed to the specific pharmacodynamic properties of these products. In the view of the clear-cut teratogenic effect that could be established in this preliminary study which allows also a qualitative and quantitative comparative assessment of teratogenic potency of the test substance and the reference compound, it was decided by the sponsor upon recommendation by the management of the testing facility not to conduct the planned main study.

Mutagenicity Studies

Ames/Salmonella
Halobetasol propionate was tested for mutagenic potential in a screening Ames/Salmonella microsome plate assay. An epoxide hydratase inhibitor and glutathione depleter, 1,1,1-trichloropropene 2,3-oxide was also included in the assay in order to increase the sensitivity of the assay for mutagenic epoxide potential. The results of this study showed halobetasol propionate to be nonmutagenic to bacterial cells with or without metabolic activation under the conditions of this assay.

Nuclear Anomaly Test in Somatic Interphase Nuclei of Chinese Hamster
Halobetasol propionate was tested to evaluate the potential mutagenic effects on somatic interphase cells in the bone marrow of Chinese hamsters. Halobetasol propionate was

administered at single oral dose levels of 750, 1500, 3000, and 6000 mg/kg daily for 2 consecutive days to study groups of 6 or 8/sex/group. The hamsters were sacrificed 24 hours after the second dose, and bone marrow smears were prepared. Control groups received the vehicle (0.5% CMC) and a positive control group was dosed with cyclophosphamide at 128 mg/kg. Nuclear anomalies were increased significantly in the bone marrow cells from animals dosed with the three lower doses of halobetasol propionate as compared to the number of nuclear anomalies in the controls. Under the experimental conditions, halobetasol propionate exerted a mutagenic action on hamster bone marrow somatic cells.

Another potent steroid, clobetasol propionate, in a similar study, was also found to produce nuclear anomalies in the bone marrow cells of Chinese hamsters at dose levels of 1250, 2500, and 5000 mg/kg.

Sister Chromatid Exchange
Halobetasol propionate was tested to evaluate the potential mutagenic effect on somatic cells (bone marrow) by the induction of sister chromatid exchange (SCE). Halobetasol propionate was administered as a single oral dose at levels of 1500, 3000, and 6000 mg/kg at a dose volume of 20 ml/kg in 0.5% CMC vehicle to Chinese hamster study groups of 4/sex/group. Two hours prior to dosing, the animals received a subcutaneous implant of a 45 mg tablet of 5-bromodeoxyuridine. After 24 hours, the hamsters were dosed i.p. with 10 mg/kg of colcemid and sacrificed, with bone marrow preparations made and stained for SCE evaluation. Control groups consisting of the vehicle and a positive control (100 mg/kg DMBA) were similarly evaluated. The results of this study showed no significant increase in the number of SCE found in comparison to the negative (vehicle) control.

Mouse Lymphoma
Halobetasol propionate was tested for mutagenic effects on L5178Y/TK +/- mouse lymphoma cells in vitro with and without microsomal activation. Results were expressed by the number of induced TK-/- mutants/$l0^6$ surviving cells. Initially in the assay tested with microsomal activation, the two low dose levels of 8.125 and 16.25 mg/ml did not produce increased mutant frequencies. In the 3 upper dose levels tested, 32.5-130 mg/ml, increased mutant frequencies were observed. Similar effects occurred when the dose levels were reassayed in the cells. Mutation frequency values were elevated at all 5 dose levels tested in the presence of metabolic activation. Again, precipitation of the test article was observed after a 4-hour treatment period. Under the experimental conditions, halobetasol propionate exerted mutagenic activity in the mouse lymphoma forward mutation system with and without metabolic activation. Clobetasol propionate was subsequently evaluated in a similar mouse lymphoma test and did not show evidence of mutagenic effects. When tested soluble concentrations did not produce a marked increase in the mutant frequency compared with the control. Concentrations in excess of 56 mg/ml without microsomal activation proved to be cytotoxic to the mouse lymphoma cells. Mutation frequencies were, therefore, not determined at this concentration.

Chromosome Studies on the Male Germinal Epithelium/ Spermatocytes – Mouse
Halobetasol propionate was tested for mutagenic effect on the germinal epithelium, particularly on the potential formation of chromosomal aberrations in spermatocytes of mice. Halobetasol propionate was administered orally at daily dose levels of 333 and 1,000 mg/kg at a dose volume

of 20 ml/kg in 0.5% CMC. Dosing was conducted intermittently for 5 days (days 0, 2, 3, 5, and 9) to study groups of 15 male mice/group including vehicle control. Three days later, the groups were dosed with 10 mg/kg of colcemid and were killed. Drop preparations were made of the testicular parenchyma with 100 metaphases scored per animal. Results from this study indicated no evidence of mutagenic activity of halobetasol propionate in mouse spermatocytes. There were no dose-related increases in the frequency of chromosomal aberrations, however, there was an occurrence of a quadrivalent exchange figure in the low dose. Mortalities occurred at both the low and high-dose groups.

Clobetasol propionate was subsequently evaluated in a comparative chromosome study in mouse spermatocytes and did not produce any chromosomal aberrations.

Chromosome Studies on Male Germinal Epithelium/ Spermatogonia – Mouse
Halobetasol propionate was tested for mutagenic effect on the germinal epithelium particularly on the potential formation of chromosomal aberrations in spermatogonia of mice. Halobetasol propionate was administered orally for 5 consecutive days at daily dose levels of 1,667 and 5,000 mg/kg in a dose volume of 20 ml/kg in 0.5% CMC to study groups of 12 males/group including a vehicle control group. The mice were sacrificed one day after the last dose after receiving 10 mg/kg of colcemid. Drop-preparations were made of the testicular parenchyma, with 100 metaphases scored per animal. The results from this assay revealed no mutagenic activity of halobetasol propionate in mouse spermatogonia.

In a similar study clobetasol propionate was subsequently evaluated for chromosomal aberrations in the spermatogonia of mice and did not produce any chromosomal aberrations.

Chromosome Studies on Somatic Cells - Chinese Hamster
Halobetasol propionate was tested to evaluate the potential mutagenic effects on somatic cells (bone marrow) in Chinese hamsters. Halobetasol propionate was administered orally as a single dose for 2 consecutive days at dose levels of 1250, 2500, and 5000 mg/kg in 20 ml/kg of 0.5% CMC to study groups of 4/sex/group. Control groups of the vehicle and positive control (cyclophosphamide) were similarly tested. Colcemid (10 mg/kg) was dosed 2 hours after the second dose to the study groups, and all animals were killed 4 hours later. Chromosomal preparations were made from the bone marrow, with 100 metaphase plates examined from 2/sex/group. Frequencies of chromosomal aberrations and aberrant metaphases were similar in the treated and negative control groups. Therefore, the results of this study indicated no evidence of mutagenic effects of halobetasol propionate in the somatic cells of Chinese hamsters.

In a parallel study clobetasol propionate was similarly evaluated in the Chinese hamster and did not show evidence of mutagenic effects.

Mammalian Spot Test – Mouse
Halobetasol propionate was tested to evaluate the potential mutagenic effects on somatic cells *in vivo*. The test permits the detection of induced point mutations and other genetic events in the melanoblasts of embryos exposed *in utero* to the test material. Mutation induction is monitored postnatally for the presence on the fur of young mice for recessive spots (RS). Halobetasol propionate was administered as a single i.p. dose at 6 levels ranging from 18.75 to 600 mg/kg in

10 ml/kg of sesame oil on the 10th day of pregnancy to study groups of 71-73 pregnant female C57B1/6J mice each. A vehicle control and positive control (50 mg/kg - N-nitroso-n-ethyl urea (EMA)) were similarly evaluated.

Post-natal examinations of the fur were recorded at the age of 12-14 days and twice weekly for 3 weeks for the presence of RS as well as for cytotoxic effects on melanocytes by recording of white mid-ventral spots (WMVS). In the 3 highest dose levels, a high percentage of mortality and embryotoxicity was observed. The results of this study revealed no evidence of mutagenic effects observed in the surviving offspring. Dose-related cytotoxic effects on melanocytes, as well as embryotoxic effects were observed.

Similar results were observed with clobetasol propionate in a subsequent mammalian spot test study in mice.

Micronucleus Test in Mice
Halobetasol propionate was evaluated in this *in vivo* micronucleus test to determine its potential to damage chromosomes of bone marrow cells or damage the mitotic spindle apparatus in these cells. Halobetasol propionate was administered to groups of mice (5 males and 5 females per group) by intraperitoneal injection for two consecutive days. Nominal halobetasol propionate doses were 7.5, 40 and 75 mg/kg per day. The animals were sacrificed at 24 and 48 hours after the second injection. Animals administered the negative (vehicle) control, i.e. DMSO and corn oil, were also sacrificed at 24 and 48 hours after the second injection. Positive control mice (triethylenemelamine at 0.5 mg/kg) were sacrificed only at 24 hours after the second dose administration.

Slides were prepared from the bone marrow of the femurs of each animal. The slides were subsequently stained, blind coded, and microscopically evaluated for the incidence of micronucleated polychromatic erythrocytes (PCE). Also scored was the incidence of normochromatic erythrocytes in order to estimate PCE/NCE ratios, a measure of toxicity to the hemopoietic system. Halobetasol propionate did not induce any statistically significant increases in the number of micronucleated PCEs. The mean PCE/NCE ratio for all halobetasol propionate treated groups was significantly reduced when compared to vehicle controls at both the 24 and 48 hours time points. The lower PCE/NCE ratios are indicative of toxicity to the bone marrow hemopoietic cells.

In conclusion, halobetasol propionate is negative in this *in vivo* mouse bone marrow micronucleus assay, when tested up to toxic dose levels.

CHO/HGPRT Mammalian Cell Forward Gene Mutation Assay
Halobetasol propionate was evaluated for mutagenic potential in this forward gene mutation assay. This assay, conducted in vitro in Chinese hamster ovary fibroblasts, measures the ability of a test article to induce a deletion, frame shift, or base pair substitution.

Halobetasol propionate was evaluated for a five-hour dosing period at nominal concentrations of 25, 50, 75, and 125 *mc*g/ml of treatment medium. All dose levels were evaluated with and without Aroclor 1254 induced rat liver S-9 activation. Mutant frequencies among the halobetasol

propionate treated cultures were all at or below negative (vehicle, DMSO) control values for the three lower doses. The mutation frequency at 125 mcg /ml dose level was increased above vehicle and negative control values. However, this dose exceeded the limit of solubility in the treatment medium.

Clobetasol propionate was included in the study as a reference agent at nominal concentrations of 25, 50, 75, and 125 mcg/ml of treatment medium. Clobetasol propionate was evaluated only in the absence of S-9 metabolic activation. Mutant frequencies observed in the clobetasol propionate treated culture while slightly elevated over negative controls did not exhibit dose dependency and thus this compound was considered negative in this assay.

Humans

A standard dermatoxicity profile, consisting of four Human Dermal Safety Studies, was conducted in the United States in 284 normal volunteers of both sexes to determine the local tolerance of halobetasol propionate 0.05% ointment and cream and their respective vehicles. The tests consisted of the 21-Day Cumulative Irritation Test (30 subjects) with plasma cortisol levels determined at weekly intervals, Repeated Insult Patch Test (RIPT, Modified Draize Skin Sensitization Test) (215 subjects) and the Phototoxicity Test (10 subjects).

Results: Halobetasol propionate 0.05% ointment and cream are slightly to mildly irritating to volunteers when applied under occlusive patches. No sensitization was seen with either halobetasol propionate 0.05% ointment or cream. The products did not produce photocontact sensitization or phototoxicity. In addition, in the 24 subjects completing the trial in whom once weekly plasma cortisols were obtained, statistically significant reduction in plasma cortisol values were observed. However, none of the values declined below the normal lower limit of 5 mg/dl over a 3-week period.

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