sacrificed after the study was terminated were submitted for microscopic examination.

Results:

The results from this study were complicated by an outbreak of mucoid enteritis in the rabbit colony. This study was originally scheduled to be a 90 day dermal administration toxicity test in rabbits for the test article. However, numerous animals in all groups exhibited signs of mucoid enteritis. These signs included depression, diarrhea, abdominal bloating, dehydration and death. Twenty deaths occurred before the study was terminated on April 27, 1995 (37 days after the start of the study). The performing contract lab states that 3/8 deaths in the high dose group occurred early in the study; one each on Day 5, 8 and 9. They concluded that these deaths may have been related to the test article, but the sponsor says that these deaths were not related to the test article. It is not possible to determine if these deaths were or were not related to the test article due to the outbreak of mucoid enteritis in the rabbit colony.

The only other measurement that was obtained for this study was dermal irritation. Dermal irritation (moderate to severe) was observed for all dose groups. The extent of dermal irritation did not appear to be dose-dependent. The dermal irritation observed in all three dose groups appeared to be greater than that observed in the placebo group.

Key Study Findings:

The major finding noted in this study after twice daily treatment for 37 days was moderate to severe dermal irritation in all test article treated animals. The extent of dermal irritation did not appear to be dose dependent. In addition, the dermal irritation observed in all three dose groups appeared to be greater than that observed in the placebo group.

A possible test article mortality in high dose animals was noted in this study. However, it is not possible to determine this with certainty due to the mortality induced by an outbreak of mucoid enteritis in the rabbit colony in this study. Therefore, the study needed to be repeated in healthy rabbits to get accurate data for this toxicity test. How the sponsor chose to address this will be described in the following reviewed nonclinical studies.

Repeat Dose Toxicology Study #5:

7-Day dermal pilot study in rabbits, 0.5% 5-FU cream

Note: At the time of conduct of this study, the sponsor had determined that clinically the 0.5% 5-fluorouracil cream was equally as efficacious as the —— 5-fluorouracil cream and produced less dermal irritation in humans. Therefore, the sponsor determined to pursue development of the drug product as a 0.5% 5-fluorouracil cream. The sponsor requested if all additional nonclinical studies could be conducted with a dose response with the 0.5% 5-fluorouracil cream due to the level of irritation seen in nonclinical studies after application of —— 5-fluorouracil cream. The sponsor was informed that this would be acceptable. The purpose of this study was

to determine tolerable doses of the 0.5% 5-fluorouracil cream to use in the repeat of the 90 day repeat dose rabbit toxicology study.

Study Title: 7-Day dermal pilot study in rabbits, 0.5% 5-FU cream
Study No: DL-PC-6025-9701
Amendment #, Vol #: 000, 10
Conducting laboratory: [redacted]
Date of study initiation: 4/21/97
GLP compliance: Yes
QA- Report: Yes (X) No ()
Methods:

Hair from the entire dorsal trunk of each animal was removed with electric clippers. A 12 x 12 cm dorsal area was divided into four 6 x 6 cm test sites. The sites were designated left front, right front, left back and right back. Elizabethan collars were placed on each animal prior to dosing and worn throughout the study.

Half the daily dose of the test material was applied to the appropriate 6 x 6 cm test site and spread evenly over the entire test site. The site remained uncovered during the treatment period. The second half of the dose was administered to the test site following six hours of exposure. Residual material was wiped from the exposure site with paper towel moistened with water prior to each dose application. For Groups 1, 2 and 4, the same site was dosed daily. For group 3, the test site was moved to a new quadrant daily.

The frequency of dosing was twice daily for seven days for groups 1, 2 and 3 and twice daily for three days for group 4. The duration of the study for groups 1 – 3 was 16 days with test material administration for the first 7 days. The duration of the study for group 4 was for 14 days with test article administration during the first 3 days.

Dosing:

- *species/strain:* New Zealand White rabbits
- *#/sex/group or time point:* 2/sex/group
- *age:* ~10 weeks
- *weight:* 2.1-2.5 kg males; 2.0-2.3 kg females
- *satellite groups used for toxicokinetics or recovery:* N/A
- *dosage groups in administered units:* refer to dosing table below
- *route, formulation, volume, and infusion rate:* route = topical, 0.7 ml/kg/interval (1.4 ml/kg/day) of vehicle or test article, 1.4 g/kg/day total dose of 0.5% 5-fluorouracil cream

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Dosing Table

Group	Treatment	Dose (mg/kg/day)	Dose site rotation schedule
1	5-fluorouracil vehicle cream	0	Left front dosed for 7 days
2	0.5% 5-fluorouracil cream	7	Left front dosed for 7 days
3	0.5% 5-fluorouracil cream	7	Four sites rotated daily for 7 days as follows: left front, right front, left back, right back
4	0.5% 5-fluorouracil cream	7	Left front dose for 3 consecutive days*

* - Group 4 was initiated after the review of the data from groups 1 – 3. Group 4 was designed to determine the recovery period after three consecutive days daily dosing at the same site.

Drug, lot#, radiolabel, and % purity: 0.5% 5-fluorouracil cream – lot# 970057
5-fluorouracil vehicle cream – lot # 970051

Formulation/vehicle: Same as clinical formulation except with the addition of [redacted] as a [redacted]

Observations and times:

- *Clinical signs:* twice daily
- *Local dermal signs:* Degree of dermal irritation was determined daily on days 1-8 and on days 12 and 16 for groups 1–3. Degree of dermal irritation was determined daily on days 0-4 and on days 7, 9, 11 and 14 group 4. Skin reactions were scored according to the Draize method.
- *Body weights:* animals were weighed on days 8 and 16 in groups 1–3 and on days 4 and 14 in group 4

Results:

- **Clinical signs** – No treatment related deaths or clinical signs were noted in this study.
- **Local dermal signs** – No dermal irritation was noted in group 1 animals (7 day single site application of 5-fluorouracil vehicle cream).

  Most of the animals in group 2 (7 day single site application of 0.5% 5-fluorouracil cream) exhibited slight erythema by day 3. All animals in this group had moderate erythema with very slight to slight edema at the end of the dosing period (Day 7). All animals had severe erythema by day 12. Other dermal effects seen in this group at the end of the treatment phase included: slight desquamation, slight fissuring, eschar and white or blanched tissue. Most animals

continued to exhibit severe erythema (3/4) and blanched skin (2/4) at the end of the study (9 days after the last dose).

The animals in group 3 (administration site rotated daily) exhibited very slight or slight erythema on the day after the initial dosing at each respective site. After a three day recovery, most animals continued to exhibit very slight or slight erythema. On the day following a second dose, all animals had slight to moderate erythema and/or very slight to slight edema at the first three sites (the fourth site was only dosed once). During the 9 to 11 day recovery period most animals exhibited moderate to severe erythema, very slight to slight edema, slight to moderate atonia, slight to moderate desquamation and/or slight fissuring. A few animals had eschar, exfoliation, and/or white or blanched tissue. These changes were still evident in most animals at study termination that had the administration sites treated twice. For the administration site that was treated once, all animals were free of irritation after a 12 day recovery period.

Group 4 (dosed at the same site for three consecutive days followed by a 12 day recovery period) was added on after the results from the previous two test groups were obtained. The sponsor decided that this dosing regimen may be less irritating to the animals and may provide adequate time for recovery prior to the next dose application (there would be 9 days between dosing at each site if 4 dosing sites were used). All of the animals in group 4 had slight erythema on the day after the initial dose. Most (3/4) of the animals had moderate erythema on the day after the third dose. After a 6 day recovery period, two of the four animals did not exhibit any signs of dermal irritation and the other two had slight erythema. After a 9 day recovery period, all animals did not exhibit any signs of dermal irritation.

- **Body weights** No treatment related effects on body weight were noted in this study.

Key Study Findings:

Severe dermal irritation was noted in the animals treated for seven days on the same treatment site with 0.5% 5-fluorouracil cream, which did not recover completely by the end of the study (9 days after end of dosing). Moderate dermal irritation was noted in animals treated for seven days on alternating treatment sites (4 total), which did not fully recover by the end of the study (9 days after end of dosing). Moderate dermal irritation was noted in animals treated for three days on the same site with a full recovery by the end of the study (12 days after end of dosing).

Based on the results from this experiment, the sponsor recommended the dosing regimen for the 90 day rabbit dermal toxicity study should consist of using four sites per animal with

rotating the site after three consecutive twice daily doses and clip the appropriate dose site on the day prior to rotation. The sponsor believed that this would be the best protocol design to allow complete healing of the site prior to another application of the test material and would therefore allow for a full 90 day study to be completed with the 0.5% 5-fluorouracil formulation.

My recommendation based on the results of this study was to consider another non-rodent species for conduct of the long term toxicology study. I recommended an 8 week repeat dose study in micropigs with a 2 week dose range study to support the dose selection for the pivotal study. The sponsor accepted this recommendation and the next two studies describe the results from the studies conducted in micropigs.

Repeat Dose Toxicology Study #6:

14-Day dermal irritation study in Yucatan micropigs

Study Title:	14-Day dermal irritation study in Yucatan micropigs
Study No:	DL-PC-6025-9703
Amendment #, Vol #:	000, 10
Conducting laboratory:	[redacted]
Date of study initiation:	10/14/97
GLP compliance:	Yes
QA- Report:	Yes (X) No ()
Methods:	

The hair was clipped from the administration site prior to treatment and was re-clipped on an as needed basis. Animals were given a daily dermal application of test article for 14 days at either 160 or 40 mg/kg of the cream formulation to 16 or 4 cm^2/kg, respectively. Test article was applied to the test site with a syringe and applied evenly with a glass rod. The treatment area was unabraided and unoccluded. Prior to the application of the next dose, the test article was gently wiped off with gauze moistened with water and blotted dry to remove any residual material.

Dosing:

- *species/strain:* male Yucatan micropigs
- *#/sex/group or time point:* 2 males/group
- *age:* not stated
- *weight:* not stated
- *satellite groups used for toxicokinetics or recovery:* Refer to dosing table below
- *dosage groups in administered units:* Refer to dosing table below
- *route, form, volume, and infusion rate:* route = topical, for additional information refer to table below

Dosing Table

Treatment	Cream dose (mg/kg/day)	Active dose (mg/kg/day)
5-fluorouracil vehicle cream	160	0
——— 5-fluorouracil cream	160	—
0.5% 5-fluorouracil cream	40	0.2
0.5% 5-fluorouracil cream	160	0.8

Drug, lot#, radiolabel, and % purity: 0.5% 5-fluorouracil cream – lot# 970080
——— 5-fluorouracil cream – lot# EXP97K075
5-fluorouracil vehicle cream – lot # 970051

Formulation/vehicle: Same as clinical formulation except with the addition of ——— as a ———

Observations and times:

- *Clinical signs:* daily
- *Local dermal signs:* daily; graded accorded to Draize scale
- *Body weights:* prior to treatment and on days 1, 7 and 14
- *Food consumption:* daily
- *Hematology:* prior to treatment and at terminal sacrifice (day 14)
- *Clinical chemistry:* prior to treatment and at terminal sacrifice (day 14)
- *Toxicokinetics:* days 1 and 14 at pretreatment and 0.5, 1, 2, 4, 8 and 24 hours postdose
- *Gross pathology:* at sacrifice
- *Histopathology:* The following organs were preserved from each animal in 10% buffered formalin: adrenals, aorta, brain, epididymides, esophagus, eyes, femur with marrow, heart, large intestines (cecum, colon, rectum), small intestines (duodenum, jejunum, ileum), kidneys, larynx/pharynx, liver, lungs, lymph nodes (mandibular and mesenteric), gross lesions, mammary glands, pancreas, peripheral nerve, pituitary, prostate and seminal vesicles, salivary glands, skeletal muscle, skin (treated and untreated), spinal cord, spleen, stomach, testes, thymus, thyroid and parathyroids, tongue, trachea and urinary bladder.

Histological examination of the untreated skin and the administration site for all dose groups was performed in this study. Other preserved tissues did not undergo microscopic evaluation.

Results:

- **Clinical signs** No treatment related deaths or clinical signs were noted in this study.

- **Local dermal signs** Dermal observations on test article treated animals were generally first noted on or after Day 10 and included darkened areas (<1-3 cm), erythema (Grade 1) and small ulcerations and papules. No dermal observations were noted in vehicle treated animals.
- **Body weights** No treatment related effects on body weight were noted in this study.
- **Food Consumption** No treatment related effects on food consumption were noted in this study.
- **Hematology** No treatment related effects on hematology parameters were noted in this study.
- **Clinical chemistry** No treatment related effects on clinical chemistry parameters were noted in this study.
- **Toxicokinetics** Concentrations of 5-fluorouracil in all toxicokinetic samples were below the limit of quantitation of — ng/ml. All toxicokinetic samples were analyzed using a validated — method (—, —) with a quantitation range of — ng/ml for 5-fluorouracil.
- **Gross pathology** No treatment related effects on gross pathology were noted in this study.
- **Histopathology** Microscopic changes in test article treated skin included a minimal to mild increase in melanin pigment and epidermal hyperplasia. More prominent changes included increased perivascular inflammatory cell infiltrates, intraepidermal pustule formation and ulceration. The epidermis adjacent to pustules/ulcers occasionally showed focal parakeratotic hyperkeratosis. All changes were limited to small foci less than 2 mm in diameter. The microscopic changes exhibited a dose response relationship.

Key Study Findings:

It appeared that the 5-fluorouracil cream was reasonably tolerated in micropigs at the highest dose tested in this study. Toxicokinetic analysis indicated that no systemic absorption of the 5-fluorouracil occurred in micropigs from the 5-fluorouracil cream under the conditions of this study.

It should be noted that the sponsor did not submit the protocol for the dose ranging to the agency prior to initiation for review. Even though the sponsor did accept the recommendation to select another species for the proposed nonclinical study besides rabbit, the sponsor did not carry out a comparison of 1X/day vs 2X/day application regimen in micropigs. However, it did appear that the 1X/day application does start to cause dermal effects prior to completion of the 14 day

dosing period. Therefore, for this test article, which has as part of it's therapeutic benefit the potential to cause severe irritation to the skin, it is probably appropriate to apply it 1X/day to reduce the irritation potential to allow for a full 8 weeks of treatment. The results of this study verify that the micropig is a better animal model for evaluating the dermal toxicity of the 5-fluorouracil cream than the rabbit. The rabbit showed severe dermal irritation after only 3 days of repeat application.

Repeat Dose Toxicology Study #7:

8-week dermal toxicity study in Yucatan micropigs

Study Title:	8-week dermal toxicity study in Yucatan micropigs
Study No:	DL-PC-6025-9704
Amendment #, Vol #:	000, 11-12
Conducting laboratory:	[redacted]
Date of study initiation:	3/6/98
GLP compliance:	Yes
QA- Report:	Yes (X) No ()
Methods:	

The hair was clipped from the administration site prior to treatment and was re-clipped on an as needed basis. Animals were given a daily dermal application of test article for 8 weeks. Test article was applied to the test site with a syringe and applied evenly with a glass rod. The treatment area was unabraided and unoccluded. Prior to the application of the next dose, the test article was gently wiped off with gauze moistened with water and blotted dry to remove any residual material.

Note: Due to the level of dermal irritation noted in males in the mid and high dose groups after 3 weeks of treatment, it was decided to dose all treatment groups on one site for 4 weeks and then rotate treatment for the last 4 weeks on another site.

Dosing:

- *species/strain:* Yucatan micropigs
- *#/sex/group or time point:* Refer to dosing table below
- *age:* 1-5 months
- *weight:* 6.3-11.6 kg males; 8.5-11.0 kg females
- *satellite groups used for toxicokinetics or recovery:* Refer to dosing table below
- *dosage groups in administered units:* Refer to dosing table below
- *route, form, volume, and infusion rate:* route = topical, for additional information refer to table below

Dosing Table

Treatment	Cream dose (mg/kg/day)	Dose Area (cm^2/kg)	Active dose a(mg/kg/day)	Number of Animals	
				Males	Females
5-fluorouracil vehicle cream	320	32	0	6	6
0.5% 5-fluorouracil cream	40	4	0.2	4	4
0.5% 5-fluorouracil cream	160	16	0.8	4	4
0.5% 5-fluorouracil cream	320	32	1.6	4	4

Drug, lot#, radiolabel, and % purity: 0.5% 5-fluorouracil cream – lot# 970080
5-fluorouracil vehicle cream – lot # 970051

Formulation/vehicle: Same as clinical formulation except with the addition of ——— as a ———

Observations and times:

- *Clinical signs:* daily
- *Local dermal signs:* weekly; graded accorded to Draize scale
- *Body weights:* weekly
- *Food consumption:* daily
- *Ophthalmology:* prior to treatment and prior to terminal sacrifice (week 8)
- *Hematology:* prior to treatment and at weeks 4 and 8
- *Clinical chemistry:* prior to treatment and weeks 4 and 8
- *Urinalysis:* prior to treatment and prior to terminal sacrifice (week 8)
- *Toxicokinetics:* week 4 and last day of dosing (week 8) at pretreatment and 0.5, 1, 2, 4, 8 and 24 hours postdose
- *Gross pathology:* at sacrifice
- *Organs weighed:* adrenals, brain, heart, kidneys, liver, lungs, pituitary gland, prostate, salivary glands, spleen, thymus, thyroid, uterus, ovaries and testes
- *Histopathology:* The following organs were preserved from each animal in 10% buffered formalin: adrenals, aorta, brain, cervix, epididymides, esophagus, eyes, femur with marrow, gall bladder, heart, large intestines (cecum, colon, rectum), small intestines (duodenum, jejunum, ileum), kidneys, lacrimal gland, larynx/pharynx, liver, lungs, lymph nodes (mandibular and mesenteric), gross lesions, mammary glands, ovaries, oviducts, pancreas, peripheral nerve, pituitary, prostate and seminal vesicles, salivary glands, skeletal muscle, skin (treated and untreated), spinal cord, spleen, sternum with marrow, stomach, testes, thymus, thyroid and parathyroids, tongue, trachea, urinary bladder, uterus and vagina.

Histological examination of all preserved tissues was performed for all dose groups in this study.

Results:

- **Clinical signs** No treatment related deaths or clinical signs were noted in this study.
- **Local dermal signs** Several dermal observations were noted in low, mid and high dose groups. The skin changes noted included fissuring, ulceration, pustule formation, darkening of areas, lightening of areas, coalescence of darkened areas and blistering. Dermal changes noted in the mid and high dose animals were of the same severity and were more severe than those noted in low dose animals. Papules were noted in on vehicle treated as well as low, mid and high dose groups. This effect is probably related to vehicle cream and not 5-fluorouracil.
- **Body weights** No treatment related effects on body weight were noted in this study.
- **Food Consumption** No treatment related effects on food consumption were noted in this study.
- **Ophthalmology** No treatment related ophthalmologic effects were noted in this study.
- **Hematology** No treatment related effects on hematology parameters were noted in this study.
- **Clinical chemistry** No treatment related effects on clinical chemistry parameters were noted in this study.
- **Urinalysis** No treatment related effects on urinalysis parameters were noted in this study.
- **Toxicokinetics** Toxicokinetic data indicated that the plasma concentration of 5-fluorouracil increased with increasing dose level. At week 4 and week 8, peak concentration occurred mainly 0.5-1 hours post-dose in both males and females. No detectable plasma levels were noted at the 4 hr timepoint and later. No obvious differences in the systemic exposure of 5-fluorouracil between males and females were noted in this study. Only plasma levels were provided in the study report. No AUC data was provided in this study report. This may be due to not all animals in each group registered measurable plasma levels.

  In low dose animals, no detectable plasma levels were noted after 4 weeks. Plasma levels ranged from ———— ng/ml in males and from ———— ng/ml in females after 8 weeks of treatment.

In mid dose animals, plasma levels ranged from ——— in males and from ——— in females after 4 weeks of treatment. Plasma levels ranged from ——— ng/ml in males and from ——— ng/ml in females after 8 weeks of treatment.

In high dose animals, plasma levels ranged from ——— ng/ml for males and from ———ng/ml for females after 4 weeks of treatment. Plasma levels ranged from ——— ng/ml for males and from ——— ng/ml in females after 8 weeks of treatment.

All toxicokinetic samples were analyzed using a validated ——— method ——— with a quantitation range of ——— ng/ml for 5-fluorouracil.

- **Gross pathology** — No treatment related effects on gross pathology were noted in this study.

- **Histopathology** — Moderate to severe microscopic changes were observed in the skin at the administration site of low, mid and high dose animals. The administration site changes were characterized by multifocal to confluent areas of acanthosis, orthokeratoic hyperkeratosis, increased melanin pigment within the epidermis and intraepidermal pustules, which frequently ruptured to form superficial ulcers with adherent serocellular crusts. Minimal dermal inflammation was noted at treated sites. Macroscopically these changes appeared as raised crusted areas, which were more darkly pigmented than adjacent untreated skin.

  Skin changes at vehicle treated administration sites in control animals were characterized by minimal acanthosis, hyperkeratosis and perivascular dermal inflammation.

Key Study Findings:

The major toxicity noted in this study was skin changes that ranged from moderate to severe at the treatment site noted in all dose groups. Macroscopically the skin changes included fissuring, ulceration, pustule formation, darkening of areas, lightening of areas, coalescence of darkened areas and blistering. Corresponding microscopic changes included multifocal to confluent areas of acanthosis, orthokeratoic hyperkeratosis, increased melanin pigment within the epidermis and intraepidermal pustules, which frequently ruptured to form superficial ulcers with adherent serocellular crusts. This level of treatment site effects occurred even when the dose site was rotated after 4 weeks. No systemic toxicity was noted in this study. Toxicokinetic data indicate that minimal systemic absorption occurred after topical application of 0.5% 5-fluorouracil cream to micropigs.

Due to the dermal effects noted at the treatment site in all dose groups, a dermal (local) NOAEL could not be established in this study. The NOAEL for systemic effects was 1.6 mg/kg/day (43.2 mg/m^2/day). The NOAEL for the systemic effects in this study is ~6 times the maximum human dose (43.2 mg/m^2/day ÷ 7.4 mg/m^2/day).

Due to the inherent nature of the 5-fluorouracil cream (anticipated dermal irritation) it was acceptable to rotate the treatment site after 4 weeks of treatment and to have a 1X/day treatment schedule instead of 2X/day. In addition, 8 weeks of treatment was acceptable for this drug product since moderate to severe dermal effects were noted by the end of this treatment period. Extending the nonclinical study to 12 weeks (90 days) would not have been humane treatment of the animals and would not have provided any additional data for safety purposes.

Reproductive Toxicology Studies (5-fluorouracil):

Note: No reproductive toxicity studies were conducted with the drug product. The sponsor will rely on literature data for 5-fluorouracil included in the NDA submission.

There are a large number of articles in the literature on the reproductive toxicity of 5-fluorouracil. Many of these articles show that 5-fluorouracil effects development in animal models. A summary of the reproductive and teratogenic effects of 5-fluorouracil demonstrated with *in vivo* animal models based on the literature submitted will be provided below.

In the Physician's Desk Reference (in the Efudex label)[46], it is reported for 5-fluorouracil that "Doses of 125 to 250 mg/kg, administered intraperitoneally, have been shown to induce chromosomal aberrations and changes in chromosome organization of spermatogonia in rats. Spermatogonial differentiation was also inhibited by fluorouracil, resulting in transient infertility. However, in studies with a strain of mouse which is sensitive to the induction of sperm head abnormalities after exposure to a range of chemical mutagens and carcinogens, fluorouracil was inactive at oral doses of 5 to 80 mg/kg/day. In female rats, fluorouracil administered intraperitoneally at doses of 25 and 50 mg/kg during the preovulatory phase of oogenesis significantly reduced the incidence of fertile matings, delayed the development of preimplantation and postimplantation embryos, increased the incidence of preimplantation lethality and induced chromosomal anomalies in these embryos. Single dose intravenous and intraperitoneal injections of 5-fluorouracil have been reported to kill differentiated spermatogonia and spermatocytes (at 500 mg/kg) and to produce abnormalities in spermatids (at 50 mg/kg) in mice."

5-Fluorouracil was shown to be teratogenic in a number of studies in laboratory rodents (mice, rats and hamsters) after parental administration. Teratogenicity was observed in inbred mice (strains 129/Rr and BALB/cRr) that were given 5-fluorouracil intraperitonally at doses ranging from 10 to 40 mg/kg on days 9-13 of gestation.[47] Tail, skull and limb defects were identified in both strains at all doses. Maximum teratogenicity was observed when 5-fluorouracil

[46] Physician's Desk Reference (1999) Efudex. Medical Economics Co., Inc. Newy Jersey, pp. 1364-1365.

[47] Dagg CP (1960) Sensitive stages for the production of developmental abnormalities in mice with 5-fluorouracil. *Am. J. Anat.* 106: 89-96.

was administered on gestational days 10-12. Teratogenicity was noted in mice at doses ≥10 mg/kg/day (30 mg/m^2/day). The lowest teratogenic dose in mice is ~4 times the maximum human dose (30 mg/m^2/day ÷ 7.4 mg/m^2/day).

Pregnant Sprague Dawley rats were injected intraperitoneally with 10-30 mg/kg of 5-fluorouracil on day 9 of gestation[48]. Fetuses were evaluated on gestation day 20. The number of externally malformed fetuses increased in a dose related manner at doses ≥15 mg/kg. The most common defect was micro-anophthalmos (presence of vestigial eyes). Growth retardation was also noted in the 5-fluorouracil treated groups. Teratogenicity was noted in rats at doses ≥15 mg/kg/day (90 mg/m^2/day). The lowest teratogenic dose in rats is ~12 times the maximum human dose (90 mg/m^2/day ÷ 7.4 mg/m^2/day).

Two studies conducted in Wistar rats showed that when 5-fluorouracil was administered intraperitoneally on gestation day 9, a dose of 10 mg/kg was not teratogenic, while teratogenicity was observed at doses of 15 or 20 mg/kg[49,50]. In another study, intraperitoneal doses of 12 to 37 mg/kg of 5-fluorouracil given on day 11 or 12 of gestation were teratogenic[51].

Malformed fetuses were observed when Syrian golden hamsters were given intramuscular injections of 5-fluorouracil at doses of 3-9 mg/animal (~33-100 mg/kg) on days 8-11 of pregnancy[52]. Fetuses were evaluated on gestational day 15. Malformations included defects of the tail and limbs and cleft palate. The malformation rate was related to the dose and time of drug administration. As organogenesis advanced, higher doses of 5-fluorouracil were needed to produce malformed embryos. A dose of 4.5 mg/animal (50 mg/kg) given on day 9 of pregnancy was lethal to all embryos, while a dose of 8 mg/animal (90 mg/kg) was required to obtain the same effect when given on day 10. Teratogenicity was noted in hamsters at doses ≥33 mg/kg/day (155 mg/m^2/day). The lowest teratogenic dose in hamsters is ~20 times the maximum human dose (155 mg/m^2/day ÷ 7.4 mg/m^2/day).

No embryotoxic effects were observed in the fetuses of two rhesus monkeys treated parenterally with 20 and 40 mg/kg of 5-fluorouracil on day 20 of pregnancy when evaluated on gestation day 100[53]. Two monkeys treated with 20 mg/kg 5-fluorouracil in this study on consecutive days between gestational days 20 to 24 did demonstrate some effects. One monkey produced a very small fetus while absorption occurred in the other. However, when eight rhesus monkeys in this study were treated with doses exceeding 40 mg/kg 5-fluorouracil on various gestational days between 17 and 27, all eight monkeys aborted 5-30 days after treatment. The

[48] Kuwagata M, Takashima H and Nagao T (1998) A comparison of the *in vivo* and *in vitro* response of rat embryos to 5-fluorouracil. *J. Vet. Med. Sci.* 60: 93-99.
[49] Wilson JG, Jordan RL and Schumacher H (1969) Potentiation of the teratogenic effects of 5-fluorouracil by natural pyridines: I. Biological aspects. *Teratology* 2: 91-98.
[50] Wilson JG (1964) Teratogenic interaction of chemical agents in the rat. *J. Pharmacol. Expl. Ther.* 144: 429-436.
[51] Murphy (1962) Teratogenic effects in rats of growth inhibiting chemicals, including studies on thalidomide. *Clin. Proc. Child. Hosp.* 18: 307-322.
[52] Shah RM and MacKay RA (1978) Teratological evaluation of 5-fluorouracil and 5-bromo-2-deoxyuridine on hamster fetuses. *J. Embryol. Exp. Morph.* 43: 47-54.
[53] Wilson JG (1971) Use of rhesus monkeys in teratological studies. *Federation Proc.* 30: 104-109.

embryolethal dose in monkeys was >40 mg/kg/day (480 mg/m^2/day). The embryolethal dose in monkeys is ~65 times the maximum human dose (480 mg/m^2/day ÷ 7.4 mg/m^2/day).

No adequate and well-controlled studies have been conducted in pregnant women with either the topical or parenteral forms of 5-fluorouracil. Multiple congenital anomalies (hypoplasia of the aorta, lung, thymus, and GI tract and urinary tract abnormalities) were reported in the fetus of a 41 year old Caucasian woman who received 600 mg doses of 5-fluorouracil intravenously five times weekly for one month beginning at 11-12 fetal weeks[54]. It was concluded in the literature report that the case most likely involved a basic genetic or chromosomal abnormality due to the advanced age of the mother. However, it could not be ruled out that 5-fluorouracil may have affected the ongoing development of some structures. In another case, 500 mg of 5-fluorouracil was administered daily for 15 doses over an 11 week period during the second and third trimesters of pregnancy[55]. No evidence of teratogenicity was observed in this neonate.

In the Physician's Desk Reference (in the Efudex label)[56],it is reported for 5-fluorouracil that "One birth defect (cleft lip and palate) has been reported in the newborn of a patient using Efudex as recommended. One birth defect (ventricular septal defect) and cases of miscarriage have been reported when Efudex was applied to mucous membrane areas." However, a cause and effect relationship to these birth defects has not been established for Efudex. The sponsor notes that teratogenicity was not reported in other cases of women treated topically with 5-fluorouracil. Five pregnant women were treated for human papillomaviurs infection with topical 5-fluorouracil (5%) on the lower genital tract at various times during the first 16 weeks of gestation[57]. Intravaginal doses ranged from 1 to 2.5 grams. One patient underwent amniocentesis that detected a 47,XXX complement of chromosomes. All of the patients delivered healthy infants at term. In another report, three women treated vaginally with Efudex (5% 5-fluorouracil cream) during the first trimester of pregnancy gave birth to normal infants[58]. Two other cases were reported of women treated vaginally with Efudex (total dose of 185-260 mg 5-fluorouracil) who became pregnant during the period of conception. Both women gave birth to normal infants[59].

In summary, 5-fluorouracil administered parenterally was shown to impair fertility in rats, was teratogenic in rodents (mice, rats and hamsters) and was embryolethal in monkeys. Teratogenicity was noted in mice at doses ≥10 mg/kg/day (30 mg/m^2/day). The lowest teratogenic dose in mice is ~4 times the maximum human dose (30 mg/m^2/day ÷ 7.4 mg/m^2/day). Teratogenicity was noted in rats at doses ≥15 mg/kg/day (90 mg/m^2/day). The lowest teratogenic

[54] Stephens JD, Golbus MS, Miller TR, Wilber RR and Epstein CJ (1980) Multiple congenital anomalies in a fetus exposed to 5-fluorouracil during the first trimester. *Am. J. Obstet. Gynecol.* 137: 747-749.
[55] Stadler HE and Knowles J (1971) Fluorouracil in pregnancy: Effect on the neonate. *J.A.M.A.* 217: 214-215.
[56] Physician's Desk Reference (1999) Efudex. Medical Economics Co., Inc. Newy Jersey, pp. 1364-1365.
[57] Van Le L, Pizzuti KJ, Greenberg M and Reid R (1991) Accidental use of low-dose 5-fluorouracil in pregnancy. *J. Reprod. Med.* 36: 872-874.
[58] Kopelman JN and Miyazawa K (1990) Inadvertent 5-fluorouracil treatment in early pregnancy: A report of three cases. *Reprod. Toxicol.* 4: 233-235.
[59] Odom LDJ, Plouffe L and Butler WJ (1990) 5-Fluorouracil exposure during the period of conception: Report on two cases. *Am. J. Obstet. Gynecol.* 163: 76-77.

dose in rats is ~12 times the maximum human dose (90 mg/m^2/day ÷ 7.4 mg/m^2/day). Teratogenicity was noted in hamsters at doses ≥33 mg/kg/day (155 mg/m^2/day). The lowest teratogenic dose in hamsters is ~20 times the maximum human dose (155 mg/m^2/day ÷ 7.4 mg/m^2/day). The embryolethal dose in monkeys was >40 mg/kg/day (480 mg/m^2/day). The embryolethal dose in this study is ~65 times the maximum human dose (480 mg/m^2/day ÷ 7.4 mg/m^2/day). It should be noted that the amount of systemic 5-fluorouracil after topical administration of the 0.5% 5-fluorouracil cream formulation to human patients will probably be much lower than the doses used parenterally in these animal studies due to the relatively low level of systemic absorption from the 0.5% 5-fluorouracil cream.

Two cases of birth defects have been reported following topical administration of 5.0% 5-fluorouracil cream (dermal or mucous membrane areas). However, a definite cause and effect relationship for these birth defects has not been established for topical 5-fluorouracil. No evidence of teratogenicity was observed in infants from ten other pregnant women who were treated vaginally with 5.0% 5-fluorouracil cream.

It is recommended that Pregnancy Category X is the appropriate pregnancy category for [redacted] (0.5% 5-fluorouracil cream). This recommendation is based on the results of the literature teratogenicity studies conducted in mice, rats and hamsters that demonstrated a strong teratogenic signal in rodents.

Genotoxicity Studies (5-fluorouracil):

Note: No genotoxicity studies were conducted with the drug product. The sponsor will rely on literature data for 5-fluorouracil included in the NDA submission.

Numerous articles exist in the literature describing the mutagenicity of 5-fluorouracil. Many of these articles indicate that 5-fluorouracil has genotoxic effects in both mammalian and non-mammalian systems. A summary of the genotoxic effects of 5-fluorouracil in various model systems described in the submitted literature will be provided below.

In vitro non-mammalian cell systems

The 1998 PDR reports (under the fluorouracil injection label) that 5-fluorouracil was mutagenic to *Salmonella typhimurium* strains TA1535, TA1537 and TA1538[60]. Negative results were observed in a mutagenicity study using strains TA100, TA98 or TA92 with or without S-9 mix at doses of 1-50 µg/plate[61]. In this study, 5-fluorouracil had a strong lethal effect on cells at a concentration of 5 µg/plate without S-9 mix. In a study sponsored by the National Toxicology Program, 5-fluorouracil was negative in the TA100, TA1535, TA1537 and TA98 strains at doses of 0 - 3.3 µg/plate with and without S-9 mix[62].

[60] Physician's Desk Reference (1998) Fluorouracil injection. Medical Economics Co. Inc. New Jersey, pp. 2463-2464.

[61] Seino Y, Nagao M, Yahagi T, Hoshi A, Kawachi T and Sugimura T (1978) Mutagenicity of several calses of antitumor agents to *Salmonella typhimurium* TA98, TA100 and TA92. *Cancer Res.* 38: 2148-2156.

[62] Zeiger E, Anderson B, Haworth S, Lawlor T, Mortelmans K and Speck W (1987) *Salmonella* mutagenicity tests: III. Results from the testing of 255 chemicals. *Environ. Mutagen.* 9: 1-110.

5-Fluorouracil was mutagenic to *Bactillus subtilis*[63] strain 168 and in the survival count rec-assay with *Bactillus subtilis*[64]. 5-Fluorouracil was reported to produce petite mutations in *Saccharomyces cerevisiae*[65].

In vitro mammalian cell systems

Oncogenic transformation of C3H/10T½ clone 8 mouse embryo cells has been induced *in vitro* by exposure to 5-fluorouracil at concentrations of 10^{-4} and 10^{-5} M, but not at concentrations of 10^{-6} M or lower[66]. In the same paper, oncogenic transformation was produced in the same cell line with a metabolite of 5-fluorouracil (5-fluoro-2'-deoxyuridine), and the transformed cells produced malignant tumors when injected into immunosuppressed syngeneic mice.

5-Fluorouracil was positive with and without S9 mix in microwell mouse lymphoma assays using L5178 $tk^{+/-}$ cell clone at concentration of 2-8 µg/ml[67].

5-Fluorouracil was clastogenic (caused breaks but no gaps or rearrangements) in Chinese hamster fibroblasts at concentrations of 100 µg/ml, but not at concentrations between 0.1 and 10 µg/ml[68].

In vivo mammalian cell systems

5-Fluorouracil was positive in the mouse bone marrow micronulceus test at intraperitoneal doses of 100 or 500 mg/kg administered twice at an interval of 24 hours[69]. Increased rates of micronuclei formation were found after 54 hours at lower doses of 5-fluorouracil (15 or 50 mg/kg). In another literature study, increased micronuclei were observed in inbred and outbred strains of Swiss CD-1 mice at an intraperitoneal dose of 50 mg/kg administered twice at an interval of 24 hours[70].

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[63] Gause GF, Kochetkova GV, Dudnik YV, Saruchanova LE and Laiko AV (1967) Isolation of mutants with altered DNA base composition in *Bacillus subtilis* 168. *Nature* 214: 714-715.
[64] SakamotoY, Fujikawa K, Nagayabu O, Yamamoto KS, Kikuchi Y and Kondo S (1985) Studies on combined test system for detection of mutagenicity of antineoplastics. *Mutat. Res.* [Abstract] 147: 272.
[65] IRAC (9181) Monographs on the evaluation of carcinogenic risks of chemicals to humans: 5-fluorouracil. Lyon, France, World Health Organization, International Agency for Research on Cancer. 26: 217-235.
[66] Jones PA, Benedict WF, Baker MS, Mondal S, Rapp U and Heidelberger C (1976) Oncogenic transformation of C3H/10T½ clone 8 mouse embryo cells by halogenated pyrimidine nucleosides. *Cancer Res.* 36: 101-107.
[67] Sofuni T, Honma M, Hayashi M, Shimada H, Tanaka N, Wakuri S, Awogi T, Yamamoto KI, Nishi Y and Nakadate M (1996) Detection of in vitro clastogens and spindle cell poisons by the mouse lymphoma assay using the microwell method: Interim report of an international collaborative study. *Mutagenesis* 11: 349-355.
[68] Maier P and Schmid W (1976) Ten model mutagens evaluated by the micronucleus test. *Mutat. Res.* 40: 325-328.
[69] Maier P and Schmid W (1976) Ten model mutagens evaluated by the micronucleus test. *Mutat. Res.* 40: 325-328.
[70] Aeschbacher HU, Gottwick D, Meier H and Poot AW (1979) Mutagen-sensitive strain of mice. *Mutat. Res.* 59: 301-304.

5-Fluorouracil was administered intraperitoneally to male CD-1 mice at doses of 12.5, 25, 50 or 100 mg/kg in a micronucleus assay that isolated peripheral blood reticulocytes[71]. Assays were performed at 24 hour intervals from 0 to 120 hours. Increased incidences of micronuclei were noted at all doses tested in this study. Peak responses at the lower doses (12.5 and 25 mg/kg/) occurred at 48-72 hours, while peak responses at the higher doses (50 and 100 mg/kg) were delayed to 96 hours.

5-Fluorouracil was tested in a mouse fetal liver micronucleus assay.[72] 5-Fluorouracil was administered to female (C3H X SWF)F_1 mice intraperitoneally on gestational day 13 at doses of 0.01 to 0.3 mmol/kg. Mice were euthanized after 27 hours. Increased micronuclei in fetal liver were observed at doses of 0.05 mmol/kg or greater. In the same paper, mice treated with 0.2 mmol/kg of 5-fluorouracil were euthanized at various time points between 0 and 48 hours after treatment. The peak frequency of micronuclei occurred at 27 hours.

5-fluorouracil was negative in a dominant lethal mutation assay performed in CFLP mice[73]. Male mice (15/group) were administered 5-fluorouracil by oral gavage for 5 consecutive days at doses of 6.25 and 25 mg/kg/day. After completion of the 5-day dosing period, each male was caged sequentially with two undosed females each week for 8 consecutive weeks. No effect on pre-implantation loss was noted in this study. The sporadic post-implantation deaths at occasional time points were not considered evidence of a dominant lethal effect.

Micronucleus and metaphase analyses were performed on Wistar-derived rat bone-marrow 12 and 24 hours following a single intraperitoneal injection of 250 mg/kg of 5-fluorouracil[74]. Increased micronuclei and chromosome aberrations were detected at both time points with the greatest response noted at 24 hours. Chromosomal aberrations were detected in Sprague Dawley rat peripheral blood lymphocytes following intraperitoneal administration of 5-fluorouracil at a dose of 50 mg/kg[75]. Blood samples were taken after 3, 12, 24 or 48 hours or weekly up to 20 weeks after 5-fluorouracil administration. A 13 fold increased in chromosome damage was observed at the first time point. Within 48 hours the effect was drastically reduced and the levels returned to spontaneous values after one week.

Sister chromatid exchanges (SCE) and chromosomal breakage were evaluated in two human patients treated with a single intravenous dose of 5000 mg of 5-fluorouracil[76]. Assays were performed using short-term phytohemagglutitin stimulated lymphocyte cultures by means

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[71] Ohuchida A, Furukawa A, Yoshida J, Watanabe M, Aruga F, Miwa Y, Shinkawa K and Kinae N (1992) Micronucleus assays on 5-fluorouracil and 6-mercaptopurine with mouse peripheral blood reticulocytes. *Mutat. Res.* 278: 139-143.

[72] Nakamura M, Fort FL and Kikuchi Y (1993) Fetal liver micronucleus assay in mice of 5-fluorouracil and related compounds. *Mutat. Res.* 291: 29-34.

[73] James DA and Smith DM (1982) Analysis of results from a collaborative study of the dominant lethal assay. *Mutat. Res.* 97: 303-314.

[74] Albanese R (1987) The cytonucleus test in the rat: A combined metaphase and micronuclues assay. *Mutat. Res.* 182: 309-321.

[75] Rosselli F, Zaccaro L, Venturi M and Rossi AM (1990) Persistence of drug-induced chromosome aberrations in peripheral blood lymphocytes of the rat. *Mutat. Res.* 232: 107-114.

[76] Musilova J, Michalova K and Urban J (1979) Sister-chromatid exchanges and chromosomal breakage in patients treated with dytostatics. *Mutat. Res.* 67: 289-294.

of bromodeoxyuridine substitution and fluorescence plus Giemsa staining technique. No rise in the SCE frequency or the number of chromosomal breaks were noted during the study period (5-60 days).

A slight increase in numerical and structural chromosome aberrations in peripheral blood lymphocytes were noted in 3 out of 4 patients with solid tumors treated parenterally with 5-fluorouracil[77]. The cumulative dose of 5-fluorouracil for the one negative patient was 0.24 g while the total dose for one of the positive patients was 1.0 g. The doses for the other 2 patients were not reported in the paper. Chromosomal abnormalities in peripheral blood lymphocytes were also observed in a study of four cancer patients who had been treated with 5-fluorouracil[78]. However, the dose of 5-fluorouracil administered to these patients were not reported in the paper.

In vivo non-mammalian cell systems

5-Fluorouracil was tested in a Drosophila wing spot test for somatic mutation that makes use of two wing cell markers (*mwh* and *flr*) that can result in three categories of spots: 1) *mwh* single spots, 2) *flr* single spots, and 3) twin spots with adjacent *mwh* and *flr* area.[79] An acute feeding study using 5 mM of 5-fluorouracil for six hours resulted in all three types of spots. In contrast, chronic feeding for 48 hours with 0.1 mM gave small single spots only.

In summary, 5-fluorouracil was mutagenic and caused chromosomal damage in several *in vivo* and *in vitro* mammalian and non-mammalian systems. Negative results were observed in a few studies in the literature articles submitted with the NDA. However, the number of positive results far outweigh the number of negative results. In is not unexpected that compounds such as 5-fluorouracil that have an effect on DNA, RNA and protein synthesis would have positive effects on mutagenicity and chromosomal damage.

Genotoxicity Studies (microsponge):

Genetic Toxicology Study #1:

Salmonella/mammalian-microsome plate incorporation mutagenicity assay – Ames test

Study Title: Salmonella/mammalian-microsome plate incorporation mutagenicity assay – Ames test
Study No: Study B0165S
Study Type: Ames test
Study Endpoint: *In vitro* bacterial mutagenicity
Amendment #, Vol #: 000, 13
Conducting Laboratory: [redacted]

[77] Bridge MF and Melamed MR (1972) Leukocyte chromosome abnormalities in advanced nonhematopoietic cancer. *Cancer Res.* 32: 2212-2220.
[78] Amato RS, Mitra J, Kabakow B and Blinick G (1966) Effects of thio-TEPA and 5-fluorouracil on chromosomes of human lymphocytes in vivo. *Federation Proc.* [Abstract] 25: 561.
[79] Graf U, Frei H, Kagi A, Katz AJ and Wurgler FE (1989) Thirty compounds tested in the Drosophilia wing spot test. *Mutat. Res.* 222: 359-373.

Study Dates: 10/3/91 – 11/5/91
GLP Compliance: Yes
QA- Reports: Yes (X) No ( )

Methods:

Indicator Strains: *Salmonella typhimurium* strains: TA98, TA100, TA1535, TA1537 and TA1538
Test Article: Acrylates ———
Drug Lot Number: 915484
Vehicle Control: DMSO
Concentrations: 0, 100, 333, 1000, 3333 and 5000 µg/plate (three replicate cultures/concentration; ± metabolic activation)
Cytotoxic Effects: No cytotoxicity noted at 5000 µg/plate in the dose ranging study. Slight precipitate was noted at all doses in pivotal study.
Criteria Evaluated: Reversion frequency, viability, integrity of background lawn
Metabolic Activation System: Aroclor-1254-induced rat liver S9

Positive Controls, Negative Controls, and Concentrations Tested

Strain	Mutagen (Positive Control)	Concentration (µg/plate)
With Activation		
TA98	2-Aminoanthracene	1.0
TA100	2-Aminoanthracene	1.0
TA1535	2-Aminoanthracene	1.0
TA1537	2-Aminoanthracene	1.0
TA1538	2-Aminoanthracene	1.0
Without Activation		
TA98	2-Nitrofluorene	1.0
TA100	Sodium Azide	1.0
TA1535	Sodium Azide	1.0
TA1537	9-Aminoacridine	75
TA1538	2-Nitrofluorene	1.0

Results:

No evidence of cytotoxicity was noted at the highest concentration tested in this assay. The histidine+ and tryptohan+ revertant values were not significantly higher in Acrylates ——— treated cultures than in negative controls. Positive controls yielded an appropriate increase in revertants in each tester strain.

Summary:

Acrylates — was negative in the Ames test under the conditions of this assay.

Genetic Toxicology Study #2:

Ames/Salmonella plate incorporation assay on microsponge with and without fluorescent light activation

Study Title:	Ames/Salmonella plate-incorporation assay on microsponge with and without fluorescent light activation
Study No:	— Study B0201S
Study Type:	Ames test
Study Endpoint:	*In vitro* bacterial mutagenicity
Amendment #, Vol #:	000, 13
Conducting Laboratory:	———
Study Dates:	11/13/92 – 12/4/92
GLP Compliance:	Yes
QA- Reports:	Yes (X) No ( )
Methods:	
Indicator Strains:	*Salmonella typhimurium* strains: TA98, TA100, TA102, TA1535, TA1537 and TA1538 *E. coli* strains: WP2 uvrA
Test Article:	Microsponge
Drug Lot Number:	— -05-11-L03
Vehicle Control:	DMSO
Concentrations:	0, 5, 16.7, 50, 167, 500, 1670, 3333 and 5000 µg/plate (three replicate cultures/concentration; ± metabolic activation)
Cytotoxic Effects:	No cytotoxicity noted at 5000 µg/plate in the dose ranging study. Slight precipitate was noted at doses ≥167 in pivotal study.
Criteria Evaluated:	Reversion frequency, viability, integrity of background lawn
Metabolic Activation System:	Aroclor 1254-induced rat liver S9
Light Activation System:	Cultures were exposed to four unshielded 40-watt cool white fluorescent bulbs for 30 minutes at a distance of 50 cm (delivers ~2-3 J/cm^2 total irradiant energy). A ——— film (———) was used to filter/reduce ambient room light.

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Positive Controls, Negative Controls, and Concentrations Tested

Strain	Mutagen (Positive Control)	Concentration (µg/plate)
With Light, Without Activation or With Activation*		
TA98	8-Methoxyposralen	50
TA100	8-Methoxyposralen	50
TA102	8-Methoxyposralen	500
TA1535	8-Methoxyposralen	50
TA1537	8-Methoxyposralen	50
TA1538	8-Methoxyposralen	50
WP2 uvrA	8-Methoxyposralen	500
Without Light, Without Activation		
TA98	2-Nitrofluorene	5.0
TA100	Sodium Azide	10.0
TA1535	Sodium Azide	10.0
TA1537	9-Aminoacridine	150
TA1538	2-Nitrofluorene	5.0
WP2 uvrA	ENNG	2.0
Without Light, With Activation		
TA98	2-Aminoanthracene	2.5
TA100	2-Aminoanthracene	2.5
TA1535	2-Aminoanthracene	2.5
TA1537	2-Aminoanthracene	2.5
TA1538	2-Aminoanthracene	2.5
WP2 uvrA	2-Aminoanthracene	2.5

* - Positive 8-Methoxyposralen (8-MOP) responses are expected only for strains TA1537, TA102, and WP2 uvrA. No 8-MOP increases are expected for strains TA1535, TA1538, TA98 or TA100.

Results:

No evidence of cytotoxicity was noted at the highest concentration tested in this assay. The histidine+ and tryptophan+ revertant values were not significantly higher in Microsponge treated cultures than in negative controls ± metabolic activation or ± light activation. Positive controls yielded an appropriate increase in revertants in each tester strain.

Summary:

Microsponge was negative in the Ames test ± metabolic activation or ± light activation under the conditions of this assay.

Genetic Toxicology Study #3:

Mutagenicity test on Acrylates ——— in the in vivo mouse micronucleus assay

Study Title:	Mutagenicity test on Acrylates ——— in the in vivo mouse micronucleus assay
Study No:	——— Report No. 18897-0-4550ECD
Study Type:	Mouse Micronucleus Assay
Study Endpoint:	*In vivo* bone marrow micronucleus formation
Amendment #, Vol #:	000, 10
Conducting Laboratory:	———
Study Dates:	12/26/97 – 11/6/97
GLP Compliance:	Yes
QA- Reports:	Yes (X) No ( )

Methods:

Species/Strain:	Crl:CD-1 (ICR) BD mice
#/Sex/Group:	6 males/group/timepoint (bone marrow harvested 24 hrs and 48 hrs after ip administration)
Test Article:	Acrylates ———
Drug Lot Number:	———13-L97030058B
Dose:	500, 1000 and 2000 mg/kg
Stock Concentration:	25, 50 and 100 mg/ml
Dose Volume:	20 ml/kg
Vehicle Control:	Corn Oil
Positive Control:	Cyclophosphamide (dose = 80 mg/kg; stock concentration = 8.0 mg/ml; dose volume = 10 ml/kg)
Route:	Intraperitoneal
Duration:	3 days
Criteria Evaluated:	Micronuclei in polychromatic erythrocytes

Results:

Treatment related clinical signs of toxicity included slight hypoactivity, squinted eyes, and rough hair coat in mid and high dose animals. Acrylates ——— was not cytotoxic to the bone marrow (defined as statistically significant decrease in the PCE:NCE ratio). No change in frequency of micronucleated polychromatic erythrocytes were noted in test article treated groups. Positive control animals yielded an appropriate increase in micronuclei in polychromatic erythrocytes.

Summary:

Acrylates ——— was negative in the mouse micronucleus assay under the conditions of this assay.

Genetic Toxicology Study #4:

Mutagenicity test with ——— in the Salmonella-Escherichia coli/mammalian-microsome reverse mutation assay with a confirmatory assay

Study Title:	Mutagenicity test with ——— in the Salmonella-Escherichia coli/mammalian-microsome reverse mutation assay with a confirmatory assay
Study No:	——— Study B0373S
Study Type:	Ames test
Study Endpoint:	*In vitro* bacterial mutagenicity
Amendment #, Vol #:	000, 13
Conducting Laboratory:	———
Study Dates:	5/13/98 – 6/2/98
GLP Compliance:	Yes
QA- Reports:	Yes (X) No ( )

Methods:

Indicator Strains:	*Salmonella typhimurium* strains: TA98, TA100, TA1535 and TA1537 *E. coli* strains: WP2 uvrA
Test Article:	——— glycol dimethacrylate ———
Drug Lot Number:	00357
Vehicle Control:	DMSO
Concentrations:	15.4, 51.2, 154, 512, 1540 and 5120 µg/ml with metabolic activation and 5.12, 15.4, 51.2, 154, 512 and 1540 µg/ml without metabolic activation (three replicate cultures/concentration)
Cytotoxic Effects:	Cytotoxicity was observed with tester strains TA100 and WP2 uvrA at ≥ 5,120 µg/ml with metabolic activation and at ≥ 1,030 µg/ml without metabolic activation.
Criteria Evaluated:	Reversion frequency, viability, integrity of background lawn
Metabolic Activation System:	Aroclor 1254-induced rat liver S9

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Positive Controls, Negative Controls, and Concentrations Tested

Strain	Mutagen (Positive Control)	Concentration (µg/plate)
	With Activation	
TA98	2-Aminoanthracene	2.5
TA100	2-Aminoanthracene	2.5
TA1535	2-Aminoanthracene	2.5
TA1537	2-Aminoanthracene	2.5
WP2 uvrA	2-Aminoanthracene	25
	Without Activation	
TA98	2-Nitrofluorene	1.0
TA100	Sodium Azide	2.0
TA1535	Sodium Azide	2.0
TA1537	ICR-191	2.0
WP2 uvrA	4-nitroquinoline-N-oxide	0.4

Results:

Cytotoxicity was noted in the dose range study and concentrations of test article were adjusted appropriately. The histidine+ and tryptohan+ revertant values were not significantly higher in —— treated cultures than in negative controls. Positive controls yielded an appropriate increase in revertants in each tester strain. An independent confirmatory assay yielded similar results.

Summary:

—— was negative in the Ames test under the conditions of this assay.

Genetic Toxicology Study #5:

Mutagenicity test on —— measuring chromosomal aberrations in Chinese hamster ovary (CHO) cells

Study Title: Mutagenicity test on —— measuring chromosomal aberrations in Chinese hamster ovary (CHO) cells
Study No: —— Study B0374S
Study Type: Chromosome Aberration Assay
Study Endpoint: *In vitro* mammalian cell chromosomal aberration
Amendment #, Vol #: 000, 13
Conducting Laboratory: ——
Study Dates: 5/13/98 – 7/22/98
GLP Compliance: Yes
QA- Reports: Yes (X) No ( )

Methods:

Indicator Strains:	Chinese Hamster Ovary Cells
Test Article:	——
Drug Lot Number:	003570
Concentrations:	10, 20, 39.9, 59.9, 118, 235 µg/ml (four replicate cultures/concentration; 17.8 hour exposure without metabolic activation; 3 hour exposure with metabolic activation)
Vehicle Control:	DMSO
Positive Controls:	Mitomycin C (0.75 µg/ml, non-activated system) Cyclophosphamide (5 µg/ml, activated system)
Cytotoxic Effects:	Cytotoxicity and reduced mitotic indices were noted in cultures treated with ≥ 336 µg/ml of test article ± metabolic activation
Criteria Evaluated:	Chromosome aberrations
Metabolic Activation System:	Aroclor 1254-induced rat liver S9

Results:

No increase in chromosomal aberrations as a result of exposure to —— was noted in this study. Positive controls yielded an appropriate increase in chromosomal aberrations.

Summary:

—— was negative in the Chinese hamster ovary cell chromosomal aberration assay under the conditions of this assay.

Genetic Toxicology Study #6:

Mutagenicity test on —— in the CHO/HGPRT forward mutation assay

Study Title:	Mutagenicity test on —— in the CHO/HGPRT forward mutation assay
Study No:	— Study B0375S
Study Type:	CHO/HGPRT forward mutation assay
Study Endpoint:	*In vitro* mammalian cell mutation assay
Amendment #, Vol #:	000, 13
Conducting Laboratory:	——
Study Dates:	5/13/98 – 6/18/98
GLP Compliance:	Yes
QA- Reports:	Yes (X) No ( )

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ON ORIGINAL

Methods:

Indicator Strains:	Chinese Hamster Ovary Cells
Test Article:	——
Drug Lot Number:	003570
Concentrations:	10, 50, 100 and 500 µg/ml without activation 10, 50, 100, 500, 1000, 1200, 1400, 1600 and 1800 µg/ml with activation (12 plates/concentration; 4 hour exposure; 7 day expression period)
Vehicle Control:	DMSO
Positive Controls:	5-Bromo-2'-deoxyuridine (50 µg/ml, non-activated system) 3-Methylcholanthrene (5 µg/ml, activated system)
Cytotoxic Effects:	Cytotoxicity was noted in cultures treated with ≥ 1000 µg/ml of test article without activation and at 2000 µg/ml with metabolic activation
Criteria Evaluated:	Reversion frequency
Metabolic Activation System:	Aroclor 1254-induced rat liver S9

Results:

Cytotoxicity was noted in the dose range study and concentrations of test article were adjusted appropriately. The 6-thioguanine+ revertant values were not significantly higher in —— treated cultures than in negative controls. Positive controls yielded an appropriate increase in revertants in Chinese hamster ovary cells.

Summary:

—— was negative in the CHO/HGPRT forward mutation assay under the conditions of this assay.

Overall Genetic Toxicology Summary:

The microsponge (Acrylic ——) was not genotoxic in the standard Ames test, the Ames test ± fluorescent light activation and an *in vivo* mouse micronucleus assay. Genotoxicity studies for —— were submitted in this NDA submission. —— is one of the individual crosslinked monomers contained in Acrylic —— formulation. These studies provide additional genotoxicity data for the microsponge system used in the 5-fluorouracil cream formulation in this NDA. —— was not genotoxic in the Ames test, Chinese hamster ovary cell chromosomal aberration assay or the CHO/HGPRT forward mutation assay.

Carcinogenicity Studies (5-fluorouracil):

Note: No carcinogenicity studies were conducted with the drug product. The sponsor will rely on literature data for 5-fluorouracil included in the NDA submission.

The carcinogenicity of 5-fluorouracil was examined in (C57BL/6 x C3H)F_1 mice[80]. Groups of 51 or 52 mice/sex were given 5-fluorouracil in their drinking water at doses of 30 or 60 ppm for 82 weeks and were observed for 4 weeks. Control mice were given tap water for 86 weeks. Mean total intakes of 5-fluorouracil per mouse for the entire study were 98 and 196 mg in males (4.25 and 8.5 mg/kg/day for a 40 g mouse) and 67 and 136 mg in females (2.9 and 5.9 mg/kg/day for a 40 g mouse), respectively. Survival in the 5-fluorouracil groups was comparable to controls and was greater than 80% in all groups at the end of the 86 week study period. Eight of 52 male mice (15%) treated at 60 ppm of 5-fluorouracil developed Harderian gland adenomas at a higher incidence than that of controls (4%). The analysis in the paper considered the difference to be marginal and noted that no dose response was found in this study for the Harderian gland adenomas. In addition, the paper stated that incidence was within the highly variable range for spontaneous tumors of this type. No significant increase in any other neoplastic lesions were noted in this study. The authors concluded that 5-fluorouracil was not carcinogenic when given continuously in the drinking water for 82 weeks to mice under the conditions of this study.

Reviewer's comment: The design of this carcinogencitiy study would not be acceptable under current agency criteria. The duration of treatment was not adequate (82 weeks instead of 104 weeks). The number of dose groups was note adequate (2 doses instead of 3 doses). The dose selection was not adequate (an MTD was not established in this study). Therefore, it can not be determined that 5-fluorouracil does not possess any potential carcinogenic risk based on the results of this study.

5-Fluorouracil was administered intraperitoneally to groups of 50 male and 50 female BALB/c mice at a dose of 30 mg/kg once a week for 50 weeks followed by an observation period for the remainder of their lives[81]. An equal number of control mice received sterile saline. The survival rate curves for both sexes showed no significant differences between treated and control mice. A significant increase in lung tumors was observed in both sexes (48% treated males, 24% for control males, 30% treated females, 18% control females). An increase in lymphoreticular tumors was noted in female mice only (42% treated females, 12% control females).

Reviewer's Comments: The utility of this study is limited because only tumors and pathologically altered organs visible at necropsy were examined microscopically. Therefore, additional tumors might have been noted if all tissues had been examined microscopically in this study. The design of this carcinogencitiy study would not be acceptable under current agency criteria. The duration of treatment was not adequate (50 weeks instead of 104 weeks). The number of dose groups was note adequate (1 dose instead of 3 doses). The dosing regimen was not acceptable (1X/week instead of daily). The dose selection was not adequate (an MTD was not established in this study). Therefore, the potential carcinogenic risk associated with 5-fluorouracil can not be adequately determined based on the results of this study. However, the increase in lung tumors in male and female mice and the increase in lymphoreticular tumors in female mice do provide a signal for carcinogenicity associated with 5-fluorouracil

[80] Iwagawa M, Nakamura M, Isumi K and Otsuka H (1991) Carcinogenicity ttesting of 5-fluorouracil in (C57BL/6 X C3H)F_1 mice. *J. Toxicol. Pathol.* 4: 129-135.

[81] Cavalier A, Alberti PF and Vitali R (1990) 5-Fluorouracil carcinogenesis in BALB/c mice. *Tumori* 76: 179-181.

administration.

A carcinogenicity study with 5-fluorouracil was performed in F344/DuCrj rats[82]. Groups of 50 rats were administered 5-fluorouracil at doses of 0, 62 or 125 ppm in their drinking water for 104 weeks. Assuming that rats weighted 400 grams and that water consumption was 12 ml/100 grams body weight/day, then these doses were equivalent to 0, 7.5 and 15 mg/kg/day. It is important to note that the survival rats were higher for the groups administered 5-fluorouracil compared to the controls for both male and female animals. It was stated in the abstract that no increase in the incidence of any neoplasia that was attributed to the administration of 5-fluorouracil was noted in this study.

Reviewer's comments: The carcinogenic potential of 5-fluorouracil can not be assessed from this study. Only an abstract was submitted for this study and without the full details of the study, it is difficult to assess the validity of this study. I would estimate that this study would not be acceptable by the agency standards due to an inadequate dose range tested (no MTD was established in the study).

The sponsor included additional references that describe studies conducted with 5-fluorouracil to assess potential carcinogenicity risk. The three studies described above were the ones most likely to have provided any insight for the potential carcinogenic risk of 5-fluorouracil. The remainder of the studies suffered from a number of deficiencies such as small group sizes, low doses, incomplete histopatholgy and short duration of exposure. None of the additional literature studies submitted the to the NDA would be acceptable to determine the potential carcinogenic risk associated with 5-fluorouracil.

The sponsor does note that the carcinogenic potential of 5-fluorouracil was reviewed by the International Agency for Research on Cancer in 1981[83] with an updated review in 1987[84]. It was concluded that there was inadequate evidence for carcinogenicity to humans. I would propose that there is inadequate data available to determine the potential carcinogenic risk associated with 5-fluorouracil. However, based on the strong genotoxicity signal noted in the literature, I would estimate that 5-fluorouracil would prove to be a strong carcinogenic agent in appropriately designed and conducted nonclinical carcinogenicity studies.

OVERALL SUMMARY AND EVALUATION:

Introduction:

5-Fluorouracil has been used for over 30 years as an antineoplastic agent administered by the parental (intravenous) route. It has also been used for almost 30 years in topical formulations of 1 to 5% for the treatment of actinic keratosis (i.e., Efudex® and Fluroplex®). The use of

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[82] Toyota K, Furukawa F, Sasada K, Yoshimura H and Takahashi M (1990) Carcinogenicity testin of the antineoplastic agent 5-fluorouracil in F344 rats. *Proc. Jpn. Cancer Assoc.* [Abstract] 49: 103.

[83] IRAC (1981) Monographs on the evaluation of carcinogenic risks of chemiicals to humans: 5-fluorouracil. Lyon, France, World Health Orgaaization, International Agency for Research on Cancer. 26: 217-235.

[84] IARC (1987) Monographs on the evaluation of carcinogenic risks to humans: 5-fluorouracil. Lyon, France, World Health Organization, International Agency for Research on Cancer, Supplement 7: 210-211.

topical 5-fluorouracil for actinic keratosis has been effective but is associated with severe skin irritation, which may limit its usefulness. The current product is a new formulation of 5-fluorouracil which —— the 5-fluorouracil in a microsponge delivery system. The microsponge polymer is composed of —— acrylate (Methyl Methacrylate/ —— Glycol Dimethacrylate) —— microspheres. The sponsor anticipates that less irritation will occur by incorporation of the drug into the microsponge polymer.

The nonclinical toxicology of systemically (oral, intravenous, etc) or topically administered 5-fluorouracil has been established previously in the literature. The focus of the nonclinical program for this topical formulation of 5-fluorouracil was to provide adequate nonclinical safety data for the new formulation of 5-fluorouracil. The sponsor has not conducted any studies to evaluate the carcinogencitiy, mutagenicity, or reproductive toxicity of 5-fluorouracil incorporated into the Microsponge® system. The sponsor will rely on relevant information for these categories as they pertain to the active ingredient, 5-fluorouracil, that have been derived from the literature and were included in the NDA submission. A brief summary of the significant toxicities and corresponding safety evaluation based on these studies is provided in the following section.

Safety Evaluation:

The nonclinical toxicology of systemically (oral, intravenous, etc) or topically administered 5-fluorouracil has been established previously in the literature. The main adverse effects noted with parenterally administered 5-fluorouracil are on the rapidly proliferating cells of the bone marrow and the gastrointestinal tract and include leukopenia, stomatitis, gastrointestinal ulceration and bleeding, and severe diarrhea. Central neurotoxicity, myocardial effects, and effects on the skin, have also occurred after parenteral administration in animals and humans. Following topical application, effects observed at the dermal administration site have included local inflammatory reactions, photosensitivity and hyperpigmentation.

Acute oral toxicity studies were conducted with the —— 5-fluorouracil microsponge cream formulation and the microsponge —— Acrylates ——) alone. The estimated oral LD_{50} for rats was determined to be greater than 5 g/kg of —5-fluorouracil cream. The estimated oral LD_{50} for rats was determined to be greater than 5 g/kg of — Acrylates ——

Dermal and ocular irritation studies were conducted in rabbits with the —— 5-fluorouracil cream. In addition, a topical phototoxicity study was conducted in hairless mice with the 0.5% 5-fluorouracil cream. In the dermal irritation study, both Effudex® cream and the —— 5-fluorouracil cream were mild dermal irritants to rabbit skin. The —— 5-fluorouracil cream was slightly more irritating than the Effudex® cream in this study. The 5-fluorouracil vehicle cream was non-irritating to rabbit skin. The —— 5-fluorouracil cream was a mild ocular irritant in rabbits. The 0.5% 5-fluorouracil cream at a dose of 40 mg/kg (200 µl/25 cm^2) was not phototoxic in hairless mice.

Dermal and ocular irritation studies were conducted in rabbits with the microsponge polymer. The — Acrylates —— was not a dermal irritant to intact or abraded rabbit skin and was a very mild ocular irritant in rabbits.

A long term dermal toxicity study for the ——5-fluorouracil cream was conducted in rats. A second long term dermal toxicity study for the 0.5% 5-fluorouracil cream was conducted in micropigs. In the 90 day dermal toxicity study in rats, doses of 20, 40 and 120 mg/kg/day of the ——5-fluorouracil cream were applied as divided daily dermal doses, 7 days/week. The major site of toxicity for the treated animals was the skin. A dose-dependent increase in skin irritation after treatment with the test article was noted in this study. The degree of skin irritation ranged from moderate in the low dose group to severe in the high dose group. In all treated groups, the irritation observed after treatment was severe enough to warrant the moving of subsequent doses to an adjacent site. Similar effects have been documented for 5-fluorouracil cream (i.e., Effudex® cream) when used clinically. In addition, a statistically significant increase in segmented neutrophils was noted in both male and female rats in the high dose group compared to control animals. Similar effects have been documented for 5-fluorouracil when used clinically via iv administration.

Due to the skin irritation noted in all dose groups, a dermal (local) NOAEL could not be established in this study. The NOAEL for systemic effects (an increase in segmented neutrophils) was 40 mg/kg/day (240 mg/m^2/day). The NOAEL for the systemic effects in this study is ~32 times the maximum human dose (240 mg/m^2/day ÷ 7.4 mg/m^2/day).

In the 8 week dermal toxicity study in micropigs, doses of 0.8, 1.2 and 1.6 mg/kg/day of the 0.5% 5-fluorouracil cream were applied daily, 7 days/week. The major toxicity noted in this study was skin changes that ranged from moderate to severe at the treatment site noted in all dose groups. Macroscopically the skin changes included fissuring, ulceration, pustule formation, darkening of areas, lightening of areas, coalescence of darkened areas and blistering. Corresponding microscopic changes included multifocal to confluent areas of acanthosis, orthokeratoic hyperkeratosis, increased melanin pigment within the epidermis and intraepidermal pustules which frequently ruptured to form superficial ulcers with adherent serocellular crusts. This level of treatment site effects occurred even after the dose site was rotated after 4 weeks. No systemic toxicity was noted in this study. Toxicokinetic data indicate that minimal systemic absorption occurred after topical application of 0.5% 5-fluorouracil cream to micropigs.

Due to the dermal effects noted at the treatment site in all dose groups, a dermal (local) NOAEL could not be established in this study. The NOAEL for systemic effects was 1.6 mg/kg/day (43.2 mg/m^2/day). The NOAEL for the systemic effects in this study is ~6 times the maximum human dose (43.2 mg/m^2/day ÷ 7.4 mg/m^2/day).

Due to the inherent nature of the 5-fluorouracil cream (anticipated dermal irritation) it was acceptable to rotate the treatment site after 4 weeks of treatment and to have a 1X/day treatment schedule instead of 2X/day in the micropig study. In addition, 8 weeks of treatment was acceptable for this drug product since moderate to severe dermal effects were noted by the end of this treatment period. Extending the nonclinical study to 12 weeks (90 days) would not have been humane treatment of the animals and would not have provided any additional data for safety purposes.

Literature studies were submitted to address the teratogenicity of 5-fluorouracil after

parental administration. In summary, 5-fluorouracil administered parenterally was shown to impair fertility in rats, was teratogenic in rodents (mice, rats and hamsters) and was embryolethal in monkeys. Teratogenicity was noted in mice at doses ≥10 mg/kg/day (30 mg/m²/day). The lowest teratogenic dose in mice is ~4 times the maximum human dose (30 mg/m²/day ÷ 7.4 mg/m²/day). Teratogenicity was noted in rats at doses ≥15 mg/kg/day (90 mg/m²/day). The lowest teratogenic dose in rats is ~12 times the maximum human dose (90 mg/m²/day ÷ 7.4 mg/m²/day). Teratogenicity was noted in hamsters at doses ≥33 mg/kg/day (155 mg/m²/day). The lowest teratogenic dose in hamsters is ~20 times the maximum human dose (155 mg/m²/day ÷ 7.4 mg/m²/day). The embryolethal dose in monkeys was >40 mg/kg/day (480 mg/m²/day). The embryolethal dose in this study is ~65 times the maximum human dose (480 mg/m²/day ÷ 7.4 mg/m²/day). It should be noted that the amount of systemic 5-fluorouracil after topical administration of the 0.5% 5-fluorouracil cream formulation to human patients will probably be much lower than the doses used parenterally in these animal studies due to the relatively low level of systemic absorption from the 0.5% 5-fluorouracil cream.

It is recommended that Pregnancy category X is the appropriate pregnancy category for the 0.5% 5-fluorouracil cream. This recommendation is based on the results of the literature teratogenicity studies conducted in mice, rats and hamsters that demonstrated a strong teratogenic signal in rodents. Additional recommendations for the inclusion of potential reproductive toxicity information in the label will be described in more detail in the Labeling Review section below.

Literature studies were submitted to address the genotoxicity of 5-fluorouracil after parental administration. In summary, 5-fluorouracil was mutagenic and caused chromosomal damage in several *in vivo* or *in vitro* mammalian and non-mammalian systems. Negative results were observed in a few studies in the literature articles submitted with the NDA. However, the number of positive results far outweigh the number of negative results. In is not unexpected that compounds such as 5-fluorouracil that have an effect on DNA, RNA and protein synthesis would have positive effects on mutagenicity and chromosomal damage.

The sponsor included three genotoxicity study reports conducted with the microsponge polymer (~ Acrylic ———) used in the drug product. The microsponge polymer was not genotoxic in the standard Ames test, the Ames test ± fluorescent light activation and an *in vivo* mouse micronucleus assay. Three genotoxicity study reports for —— were submitted in this NDA submission. —— is one of the individual crosslinked monomers contained in the ~ Acrylic —— formulation. These studies provide additional genotoxicity data for the microsponge system used in the 5-fluorouracil cream formulation in this NDA. —— was not genotoxic in the Ames test, Chinese hamster ovary cell chromosomal aberration assay or the CHO/HGPRT forward mutation assay. Therefore, it is anticipated that the potential genotoxicity expressed for this drug product would be due to the active ingredient, 5-fluorouracil. It is not anticipated that the microsponge polymer used in the drug product would increase the genotoxic potential of 5-fluorouracil.

The literature articles submitted to the NDA to address the carcinogenic potential of 5-fluorouracil were inadequate. The studies suffered from a number of deficiencies such as small group sizes, low doses, incomplete histopatholgy and short duration of exposure. There is

inadequate data available to determine the potential carcinogenic risk associated with 5-fluorouracil. However, based on the strong genotoxicity signal noted in the literature, I would estimate that 5-fluorouracil would prove to be a strong carcinogenic agent in appropriately designed and conducted nonclincial carcinogenicity studies. However, due to the low level of systemic absorption noted for the 0.5% 5-fluorouracil cream, I would anticipate the risk for development of cancer from the use of the 0.5% 5-fluorouracil cream in the treatment of actinic keratosis would be low.

Clinical Relevance of Safety Issues:

The potential toxicity of 0.5% 5-fluorouracil cream has been adequately studied in nonclinical toxicology studies. No significant nonclinical toxicity findings were noted that would preclude the use of the 0.5% 5-fluorouracil cream in the treatment of actinic keratosis.

Conclusions:

Based on the nonclinical data available for the 0.5% 5-fluorouracil cream, my recommendation for NDA 20-985 is that it be approvable from a pharmacology/toxicology perspective provided that the recommended changes in the label discussed in the next section are incorporated into the label.

Labeling Review:

The entire ——— label is inserted below. Comments about the portions that relate to nonclinical pharmacology/toxicology will be inserted directly in the appropriate sections. Recommended sections to be deleted are marked by ~~strikeout~~. Recommended sections to be added are marked by highlight.

Note: The sponsor submitted only an annotated label to the NDA for review. It is recommended that all reference numbers be removed from the label for the final printed version. In addition, it has been determined by OPDRA to accept the tradename ——— without the ——. Therefore, it is recommended that all mention of —— be removed from the label for the final printed version.

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Draft

Labeling

APPEARS THIS WAY
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/S/

Barbara Hill, Ph.D.
Reviewing Pharmacologist

cc:
——: 20-985 (000)
HFD-340
HFD-540/DIV FILES
HFD-540/ TOX/JACOBS
HFD-540/PHARM/HILL
HFD-540/MO/VAUGHAN
HFD-540/CHEM/HATHAWAY
HFD-540/PM/LUTWAK
C:/MY DOCUMENTS/WORD/NDAS/NDA20985/20985000.DOC

Concurrence Only:
HFD-540/DivDir/JWILKIN /S/ 8/15/00
HFD-540/PharmTL/AJACOBS

DFS
7/13/00