NTP
National Toxicology Program
U.S. Department of Health and Human Services

NTP Technical Report on the Photococarcinogenesis Study of Aloe Vera [CAS No. 481-72-1 (Aloe-emodin)] in SKH-1 Mice (Simulated Solar Light and Topical Application Study)

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NTP TECHNICAL REPORT

ON THE

PHOTOCOCARCINOGENESIS

STUDY OF ALOE VERA

[CAS NO. 481-72-1 (Aloe-emodin)]

IN SKH-1 MICE

(SIMULATED SOLAR LIGHT AND TOPICAL APPLICATION STUDY)

NATIONAL TOXICOLOGY PROGRAM
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FOREWORD

The National Toxicology Program (NTP) is an interagency program within the Public Health Service (PHS) of the Department of Health and Human Services (HHS) and is headquartered at the National Institute of Environmental Health Sciences of the National Institutes of Health (NIEHS/NIH). Three agencies contribute resources to the program: NIEHS/NIH, the National Institute for Occupational Safety and Health of the Centers for Disease Control and Prevention (NIOSH/CDC), and the National Center for Toxicological Research of the Food and Drug Administration (NCTR/FDA). Established in 1978, the NTP is charged with coordinating toxicological testing activities, strengthening the science base in toxicology, developing and validating improved testing methods, and providing information about potentially toxic substances to health regulatory and research agencies, scientific and medical communities, and the public.

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The photococarcinogenesis study of Aloe vera was conducted at the Food and Drug Administration's (FDA) National Center for Toxicological Research (NCTR) under an interagency agreement between the FDA and the National Institute of Environmental Health Sciences (NIEHS). The studies were monitored by a Toxicology Study Selection and Review Committee composed of representatives from the NCTR and other FDA centers, NIEHS, and other *ad hoc* members from other governmental agencies and academia. The interagency agreement was designed to use the staff and facilities of the NCTR in the testing of FDA priority chemicals and to provide FDA scientists and regulatory policymakers with information for hazard identification and risk assessment.

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SUMMARY

Background
Extracts from the Aloe vera plant are widely used in skin care products. We studied the effects of synthetic solar light on the skin of hairless mice that had been treated with creams containing various Aloe vera extracts.

Methods
We applied creams containing Aloe vera plant extracts (aloe gel, whole leaf, or decolorized whole leaf) or aloe-emodin to groups of 36 male and female hairless mice in the morning; other groups received creams containing no aloe. In the afternoon groups of animals were exposed to synthetic solar light for four hours. Other groups were not exposed to light and were control groups. The treatments and exposures were performed five days per week for 40 weeks, during which the animals were monitored for development of skin cancers.

Results
Mice exposed to synthetic solar light developed significant increases in squamous cell neoplasms and squamous cell nonneoplastic lesions of the skin whether or not they received treatment with cream. Female mice, but not male mice, treated with aloe gel or aloe-emodin and exposed to simulated solar light had increased numbers of squamous cell neoplasms when compared with mice treated with the carrier cream without the aloe gel or aloe-emodin and exposed to the same intensity of light. For both male and female mice, inclusion of aloe whole leaf extract or decolorized leaf extract in the cream increased the number of squamous cell neoplasms when the animals were exposed to simulated solar light.

Conclusions
We conclude that aloe gel or aloe-emodin had a weak enhancing effect on the photocarcinogenic activity of simulated solar light in female but not male hairless mice. We conclude that aloe whole leaf extract and decolorized leaf extract had a weak enhancing effect on the photocarcinogenic activity of simulated solar light in both male and female hairless mice.

ABSTRACT

ALOE VERA

CAS No. 481-72-1 (Aloe-emodin)

Chemical Formula: $C_{15}H_{10}O_5$ (Aloe-emodin) Molecular Weight: 270.24 (Aloe-emodin)

Synonyms: *Aloe barbadensis* Miller; *Aloe vera* Tournefort ex Linné; *Aloe vulgaris* Lamark; Barbados aloe; Curaçao aloe

The popular recognition of the *Aloe barbadensis* Miller (Aloe vera) plant as a therapeutic dermatologic agent has led to the widespread incorporation of Aloe vera leaf extracts in skincare products. Studies have suggested that Aloe vera in skincare preparations may enhance the induction of skin cancer by ultraviolet radiation. A 1-year study was conducted in mice to determine whether the topical application of creams containing Aloe vera plant extracts (aloe gel, whole leaf, or decolorized whole leaf) or creams containing aloe-emodin would enhance the photocarcinogenicity of simulated solar light (SSL).

1-Year Study

Groups of 36 male and 36 female Crl:SKH-1 (hr^-/hr^-) hairless mice received topical applications of control cream or creams containing 3% or 6% (w/w) aloe gel, whole leaf, or decolorized whole leaf or 7.46 or 74.6 µg/g aloe-emodin to the dorsal skin region each weekday morning. The mice were irradiated with SSL emitted from filtered 6 kW xenon arc lamps each weekday afternoon. The topical applications of creams and irradiance exposures were conducted 5 days per week for a period of 40 weeks. A 12-week recovery/observation period followed the 40-week treatment/exposure period. Additional groups of 36 male and 36 female mice received no cream and were exposed to 0.00, 6.85, 13.70, or 20.55 mJ • CIE/cm^2 SSL per day.

Mice that received no cream treatment and were exposed to increasing levels of SSL showed significant SSL exposure-dependent decreases in survival and significant increases in the in-life observations of skin lesion onset, incidence, and multiplicity, and significant SSL exposure-dependent increases in the incidences and multiplicities of histopathology-determined squamous cell nonneoplastic skin lesions (squamous hyperplasia and focal atypical hyperplasia) and squamous cell neoplasms (papilloma, carcinoma *in situ*, and/or carcinoma). Squamous cell neoplasms were not detected in mice that received no SSL exposure.

The topical treatment with the control cream of mice that were exposed to SSL did not impart a measurable effect when compared with comparable measurements in mice that received no cream treatment and were exposed to the same level of SSL, suggesting that the control cream used in these studies did not alter the efficiency of the SSL delivered to mice or the tolerability of mice to SSL.

The application of aloe gel creams to mice had no effect on body weights, survival, or the in-life observations of skin lesion onset, incidence, or multiplicity. The administration of aloe gel creams to male mice had no effect on the incidences or multiplicities of histopathology-determined squamous cell nonneoplastic skin lesions or neoplasms. Female mice treated with aloe gel creams (3% and 6%) had significantly increased multiplicities of squamous cell neoplasms.

There were no treatment-related effects on body weights, survival, or the in-life observations of skin lesion onset, incidence, or multiplicity in mice treated with the whole leaf creams. In male mice exposed to SSL and treated with the 6% whole leaf cream, a significant increase was observed in the multiplicity of squamous cell neoplasms.

Female mice exposed to SSL and treated with the 3% whole leaf creams had significantly decreased multiplicity of squamous cell nonneoplastic lesions and significantly increased multiplicity of squamous cell neoplasms. Female mice exposed to SSL and treated with the 6% whole leaf cream had significantly decreased multiplicity of squamous cell nonneoplastic lesions.

The application of decolorized whole leaf creams to mice had no effect on body weights, survival, or the in-life observations of skin lesion onset, incidence, or multiplicity. Male mice administered the 3% decolorized whole leaf cream had significantly increased multiplicity of squamous cell neoplasms. Female mice administered the 3% decolorized whole leaf cream had significantly decreased multiplicity of squamous cell nonneoplastic skin lesions and significantly increased multiplicity of squamous cell neoplasms. In female mice that received the 6% decolorized whole leaf cream, there was a significant increase in the multiplicity of squamous cell neoplasms.

As with the Aloe vera plant extracts, the application of aloe-emodin creams to mice had no measurable effect on body weights, survival, or the in-life observations of skin lesion onset, incidence, or multiplicity. The administration of aloe-emodin creams to male mice had no effect on the incidence or multiplicity of histopathology-determined nonneoplastic skin lesions or squamous cell neoplasms. Female mice treated with the 74.6 µg/g aloe-emodin cream had significantly decreased multiplicity of histopathology-determined squamous cell nonneoplastic skin lesions and significantly increased multiplicity of squamous cell neoplasms.

CONCLUSIONS

These experiments investigated the potential of topical application of creams containing extracts of *Aloe barbadensis* Miller plant (aloe gel, whole leaf, or decolorized whole leaf) or aloe-emodin to alter the photocarcinogenic activity of filtered xenon arc simulated solar light (SSL) in male and female SKH-1 hairless mice. Data on skin lesions were collected both on digital images during the in-life phase and by histopathologic evaluation at necropsy. No effects of creams upon SSL-induced skin lesions were identified from data collected during the in-life phase.

Aloe Gel or Aloe-emodin

Under the conditions of these studies, there was a weak enhancing effect of aloe gel or aloe-emodin on the photocarcinogenic activity of SSL in female but not in male SKH-1 mice based on an increase in the multiplicity of histopathologically determined squamous cell neoplasms.

Aloe Whole Leaf or Decolorized Whole Leaf

Under the conditions of these studies, there was a weak enhancing effect of aloe whole leaf or decolorized whole leaf on the photocarcinogenic activity of SSL in both male and female SKH-1 mice based on an increase in the multiplicity of histopathologically determined squamous cell neoplasms.

A summary of the Technical Reports Review Subcommittee comments and the public discussion on this Technical Report appears on page 12.

Summary of the 1-Year Photococarcinogenesis Study of Aloe vera and 13.70 mJ • CIE/cm^2 SSL in SKH-1 Mice

	Aloe Gel: Male	Aloe Gel: Female	Whole Leaf: Male	Whole Leaf: Female
Concentrations in cream	0%, 3%, or 6%	0%, 3%, or 6%	0%, 3%, or 6%	0%, 3%, or 6%
Body weights	No effect	No effect	No effect	No effect
Survival rates	No effect	No effect	No effect	No effect
In-life observed skin lesion onset	No effect	No effect	No effect	No effect
In-life observed skin lesion incidence	No effect	No effect	No effect	No effect
In-life observed skin lesion multiplicity[a]	No effect	No effect	No effect	No effect
Incidence of histopathology determined focal atypical squamous hyperplasia	No effect	Decreased (35/36, 27/36, 29/36)	No effect	No effect
Multiplicity of histopathology determined focal atypical squamous hyperplasia[a]	No effect	Decreased (4.4, 2.4, 3.1)	No effect	Decreased (4.4, 3.5, 3.2)
Incidence of histopathology determined squamous neoplasms (papilloma, carcinoma *in situ*, and/or carcinoma)	No effect	No effect	No effect	No effect
Multiplicity of histopathology determined squamous neoplasms (papilloma, carcinoma *in situ*, and/or carcinoma)[a]	No effect	Increased (6.4, 9.2, 8.1)	Increased (5.8, 6.4, 8.4)	Increased (6.4, 8.7, 7.7)

[a] The number of skin lesions per animal at risk in the group

Summary of the 1-Year Photococarcinogenesis Study of Aloe vera and 13.70 mJ • CIE/cm^2 SSL in SKH-1 Mice

	Decolorized Whole Leaf: Male	Decolorized Whole Leaf: Female	Aloe-emodin: Male	Aloe-emodin: Female
Concentrations in cream	0%, 3%, or 6%	0%, 3%, or 6%	0, 7.46, or 74.6 µg/g	0, 7.46, or 74.6 µg/g
Body weights	No effect	No effect	No effect	No effect
Survival rates	No effect	No effect	No effect	No effect
In-life observed skin lesion onset	No effect	No effect	No effect	No effect
In-life observed skin lesion incidence	No effect	No effect	No effect	No effect
In-life observed skin lesion multiplicity[a]	No effect	No effect	No effect	No effect
Incidence of histopathology determined focal atypical squamous hyperplasia	No effect	Decreased (35/36, 32/36, 27/36)	No effect	No effect
Multiplicity of histopathology determined focal atypical squamous hyperplasia[a]	No effect	Decreased (4.3, 2.5, 3.5)	No effect	Decreased (4.4, 3.5, 3.1)
Incidence of histopathology determined squamous neoplasms (papilloma, carcinoma *in situ*, and/or carcinoma)	No effect	No effect	No effect	No effect
Multiplicity of histopathology determined squamous neoplasms (papilloma, carcinoma *in situ*, and/or carcinoma)[a]	Increased (5.8, 8.0, 6.4)	Increased (6.4, 10.0, 9.3)	No effect	Increased (6.4, 7.9, 8.9)

[a] The number of skin lesions per animal at risk in the group

NATIONAL TOXICOLOGY PROGRAM BOARD OF SCIENTIFIC COUNSELORS TECHNICAL REPORTS REVIEW SUBCOMMITTEE

The members of the Technical Reports Review Subcommittee who evaluated the draft NTP Technical Report on Aloe vera on February 27, 2008, are listed below. Subcommittee members serve as independent scientists, not as representatives of any institution, company, or governmental agency. In this capacity, subcommittee members have five major responsibilities in reviewing the NTP studies:

- to ascertain that all relevant literature data have been adequately cited and interpreted,
- to determine if the design and conditions of the NTP studies were appropriate,
- to ensure that the Technical Report presents the experimental results and conclusions fully and clearly,
- to judge the significance of the experimental results by scientific criteria, and
- to assess the evaluation of the evidence of carcinogenic activity and other observed toxic responses.

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SUMMARY OF TECHNICAL REPORTS REVIEW SUBCOMMITTEE COMMENTS

On February 27, 2008, the draft Technical Report on the photococarcinogenesis study of Aloe vera received public review by the National Toxicology Program's Board of Scientific Counselors' Technical Reports Review Subcommittee. The review meeting was held at the National Institute of Environmental Health Sciences, Research Triangle Park, NC.

Dr. M.D. Boudreau, National Center for Toxicological Research (NCTR), introduced the study of photococarcinogenicity of Aloe vera by describing the use of aloe in cosmetic and therapeutic preparations, the various plant extracts used in the studies, the design and methodology of the photococarcinogenesis study, and the survival, body weights, and skin lesions of the animals in the study. In addition, Dr. Boudreau explained the distinction between normal in-life observations that note the presence of lesions and the subsequent histopathology diagnoses that identify the type of each lesion. The proposed conclusions for the 1-year study were:

In male and female SKH-1 hairless mice treated topically with aloe gel creams and exposed to simulated solar light (SSL), the onset, incidence, and multiplicity of in-life observed skin lesions and the incidence of squamous cell neoplasms (papilloma, carcinoma *in situ*, and carcinoma) did not differ from comparable measurements made in mice treated with control cream and exposed to SSL. The multiplicity of squamous cell neoplasms was significantly increased in female, but not male, mice treated topically with aloe gel creams and exposed to SSL when compared to mice treated with control cream and exposed to the same dose of SSL.

In male and female SKH-1 hairless mice treated topically with aloe whole leaf creams and exposed to SSL, the onset, incidence, and multiplicity of in-life observed skin lesions and the incidence of squamous cell neoplasms did not differ from the comparable measurements made in mice treated with control cream and exposed to SSL. The multiplicity of squamous cell neoplasms was significantly increased in mice treated topically with aloe whole leaf creams and exposed to SSL when compared to mice treated with control cream and exposed to the same dose of SSL.

In male and female SKH-1 hairless mice treated topically with decolorized aloe whole leaf creams and exposed to SSL, the onset, incidence, and multiplicity of in-life observed skin lesions and the incidence of squamous cell neoplasms did not differ from the comparable measurements made in mice treated with control cream and exposed to SSL. The multiplicity of squamous cell neoplasms was significantly increased in mice treated topically with decolorized aloe whole leaf creams and exposed to SSL when compared to mice treated with control cream and exposed to the same dose of SSL.

In male and female SKH-1 hairless mice treated topically with aloe-emodin and exposed to SSL, the onset, incidence, and multiplicity of in-life observed skin lesions and the incidence of squamous cell neoplasms did not differ from the comparable measurements made in mice treated with control cream and exposed to SSL. The multiplicity of squamous cell neoplasms was significantly increased in female mice, but not male mice, treated topically with aloe-emodin creams and exposed to SSL when compared to mice treated with control cream and exposed to the same dose of SSL.

Dr. Crump, the first principal reviewer, noted the complexity of this type of study. He asked for more detail of the source of the statistical analyses used and the definitions of the parameters and tests. He felt he was in general agreement with the conclusions but found them difficult to follow.

Dr. Cattley, the second principal reviewer, asked for clarification about the distinction between the presumptive positive control material and the other test articles and about the light used in the present studies and UVB radiation. He asked for more clarification of the study rationale and of the neoplasm types that led to the conclusion statements.

Dr. Bunton, the third principal reviewer, felt the conclusions were more restatements of findings than interpretive conclusion statements. She asked for clarification of notes in the appendixes regarding which tissue sites received histopathologic analysis and which did not. Dr. Bunton also inquired about the 12-week period following exposure to irradiation before animal sacrifice.

Dr. Boudreau agreed to expand the description of statistical tests used and the characterization of the aloe-emodin material. She explained that the final 12-week period was an observation period to follow the progression of lesions following exposure.

Dr. P.C. Howard, NCTR, added that this study followed the standard exposure procedures for photocarcinogenesis experiments, with the only difference being an enhanced histopathologic examination of the animals. He noted that the light used in these studies simulated solar light in its distribution of wavelengths and was different from pure UVB radiation in its tumorigenic efficiency.

In discussion of the proposed conclusions, Dr. Bunton inquired if the language could more than restate the findings, and Dr. Crump inquired if the standard levels of evidence were inappropriate for this type of study. Dr. J.R. Bucher, NIEHS, replied that this was the second photococarcinogenesis study reviewed so far, and the program was still trying to develop language for studies that may show either cocarcinogenic or protective effects. Dr. Howard noted that in the previous study the benchmark for conclusions was based on the ability of the test compound to alter the carcinogenic effects of the simulated solar light.

At this point, an alternative conclusion drafted by the NTP was displayed:

These experiments investigated the potential of topical application of creams containing extracts of *Aloe barbadensis* Miller plant (aloe gel, whole leaf, decolorized whole leaf, or aloe-emodin) to alter the photocarcinogenic activity of filtered xenon arc simulated solar light (SSL) in male and female SKH-1 hairless mice.

Aloe Gel or Aloe-emodin
Under the conditions of these studies, there was no effect of aloe gel or aloe-emodin on the photocarcinogenic activity of SSL in male SKH-1 mice. There was a weak enhancing effect of aloe gel or aloe-emodin on the photocarcinogenic activity of SSL in female SKH-1 mice based on an increase in the multiplicity of histopathologically determined squamous cell neoplasms. Neither aloe gel nor aloe-emodin affected the SSL-induced onset, incidence, or multiplicity of in-life observed skin lesions or the SSL-induced incidence of histopathologically determined squamous cell neoplasms.

Aloe Whole Leaf or Decolorized Whole Leaf
Under the conditions of these studies, there was a weak enhancing effect of aloe whole leaf or decolorized whole leaf on the photocarcinogenic activity of SSL in both male and female SKH-1 mice based on an increase in the multiplicity of histopathologically determined squamous cell neoplasms. Neither aloe whole leaf nor decolorized whole leaf affected the SSL-induced onset, incidence, or multiplicity of in-life observed skin lesions or the SSL-induced incidence of histopathologically determined squamous cell neoplasms.

Dr. Mirsalis said this set of conclusions was clearer. He felt that use of the traditional categories of levels of evidence might not be appropriate for this type of study.

Dr. Crump suggested that stating incidences were not increased was less meaningful for studies where overall incidences approached 100% in all groups. Dr. Howard noted that in this type of study time of onset was often more significant than the overall incidence of animals with skin lesions, particularly at the higher light intensities.

Drs. Crump and Mirsalis suggested adding one sentence noting that no differences in lesions were detected in the in-life observations. Dr. R.L. Melnick, NIEHS, suggested that the modifier "weak" was undefined by any formal criteria and may be difficult for subsequent readers to interpret.

Following a lunch recess, the panel reconvened to consider a revised version of the conclusions:

These experiments investigated the potential of topical application of creams containing extracts of *Aloe barbadensis* Miller plant (aloe gel, whole leaf, or decolorized whole leaf) or aloe-emodin to alter the photocarcinogenic activity of filtered xenon arc simulated solar light (SSL) in male and female SKH-1 hairless mice. Data on skin lesions were collected both on digital images during the in-life phase and by histopathologic evaluation at necropsy. No effects of creams upon SSL-induced skin lesions were identified from data collected during the in-life phase.

Aloe Gel or Aloe-emodin
Under the conditions of these studies, there was a weak enhancing effect of aloe gel or aloe-emodin on the photocarcinogenic activity of SSL in female but not in male SKH-1 mice based on an increase in the multiplicity of histopathologically determined squamous cell neoplasms.

Aloe Whole Leaf or Decolorized Whole Leaf
Under the conditions of these studies, there was a weak enhancing effect of aloe whole leaf or decolorized whole

leaf on the photocarcinogenic activity of SSL in both male and female SKH-1 mice based on an increase in the multiplicity of histopathologically determined squamous cell neoplasms.

Following discussion, Dr. Mirsalis moved, and Dr. Novak seconded, that the conclusions be accepted as rewritten. The motion was approved unanimously with eight votes.

INTRODUCTION

ALOE VERA

CAS No. 481-72-1 (Aloe-emodin)

Chemical Formula: $C_{15}H_{10}O_5$ (Aloe-emodin) Molecular Weight: 270.24 (Aloe-emodin)

Synonyms: *Aloe barbadensis* Miller; *Aloe vera* Tournefort ex Linné; *Aloe vulgaris* Lamark; Barbados aloe; Curaçao aloe

Chemical and Physical Properties

The *Aloe barbadensis* Miller (Aloe vera Linné) plant, commonly referred to as Aloe vera, has green fleshy leaves covered by a thick cuticle or rind and a clear pulp (Figure 1). The main feature of the Aloe vera plant is its high water content, ranging from 99% to 99.5% (Atherton, 1998). The remaining 0.5% to 1.0% solid material contains over 75 different, potentially active compounds, including water- and fat-soluble vitamins, minerals, enzymes, simple and complex polysaccharides, phenolic compounds, and organic acids. Compositional studies on the structural components of the Aloe vera plant leaf portions have found that the rind composes 20% to 30% and the pulp 70% to 80% of the whole leaf weight. On a dry weight basis, the percentages of the rind and pulp represented as lipids (2.7% and 4.2%) and proteins (6.3% and 7.3%) only account for a minor fraction (Femenia *et al.*, 1999). The percentages of soluble sugars (11.2% and 16.5%), primarily glucose, and ash (13.5% and 15.4%), in particular calcium, are relatively high in the rind and pulp, respectively. However, the nonstarch polysaccharides and lignin represent the bulk of each leaf fraction, accounting for 62.3% and 57.6% of the dry weight of the rind and pulp, respectively. Two commercially important products are obtained from the leaf pulp of the Aloe vera plant: aloe gel and aloe latex.

Aloe Gel

Aloe gel is the clear, mucilaginous, aqueous extract of the inner leaf pulp (Figure 1) and serves as the water and energy storage component of the plant (Yaron, 1993; Paez *et al.*, 2000). Mechanical extrusion of the inner leaf pulp produces a 70% yield of aloe gel (g gel/100 g pulp) with a water content of 99% to 99.5% (Femenia *et al.*, 1999). The aloe gel of field-grown Aloe vera has a pH of 4.4 to 4.7 and a total soluble solids content of 0.56% to 0.66%; however, seasonal fluctuations and fluctuations due to water availability have been noted (Genet and van Schooten, 1992; Wang and Strong, 1995; Waller *et al.*, 2004). The high acidity of the aloe gel may be due to the accumulation of organic acids, such as malic acid, via crassulacean acid metabolism (CAM). Chemical analysis of aloe gel indicates that, as with the rind and pulp, lipids and proteins are minor fractions of the dry weight, representing 5.1% and 8.9%, respectively; however, the amount of soluble sugars (27.8%) detected in the gel is substantially higher than that in the rind or pulp (Wang and Strong, 1995). The ash content is relatively high in all fractions of the plant but in particular in the aloe gel, where it accounts for 23.6% of the dry matter. Sodium, potassium, calcium, and magnesium are the predominant minerals detected in all leaf fractions; however, calcium is the main mineral present in the rind and pulp fractions, while sodium and potassium are higher in the aloe gel. Nonstarch polysaccharides and lignin represent 35% of the dry mass of the aloe gel (Femenia *et al.*, 1999). Aloe gel polysaccharides consist of linear chains of glucose and mannose molecules, and because there is considerably more mannose present than glucose, the molecules are referred to as polymannans. These linear chains range in size from a few to several thousand subunits. The major polysaccharide, aceman-

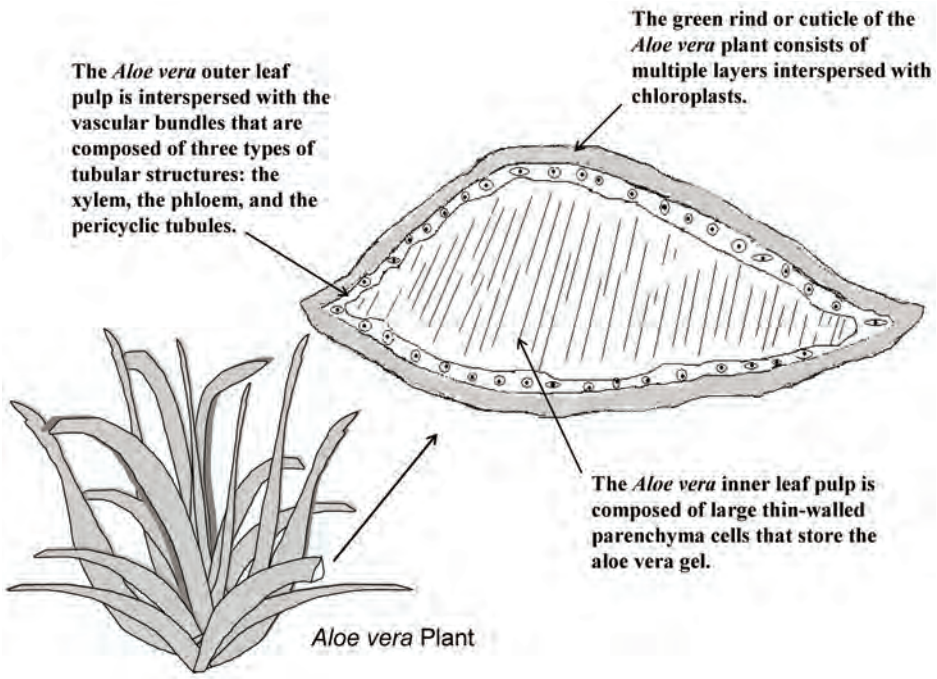


Figure 1
Schematic Representation of the Aloe vera Plant and a Cross-section through an Aloe vera Leaf

nan, is composed of one or more polymers of various chain lengths with molecular weights ranging from 30 to 40 kDa or greater and consisting of repeating units of glucose and mannose in a 1:3 ratio (Gowda *et al.*, 1979; Mandal and Das, 1980a; Yaron, 1993; Femenia *et al.*, 1999; Chow *et al.*, 2005). The polysaccharide sugar moieties of acemannan are linked by β glycosidic bonds to form linear chains with random *O*-acetyl groups and a low degree of galactose side chain branching. The β-1→4 glycosidic bond configuration of acemannan is an important consideration when examining the reported therapeutic effects of aloe gel, since humans lack the capacity to digest these bonds enzymatically. The size and structure of the polysaccharide polymers result in the formation of a colloidal system within the leaf pulp tissue that increases the viscosity and opacity of the mostly aqueous solution (Danhof, 1998). The chemical bonds within the carbohydrate polymers contribute to these qualities but are susceptible to degradation by endogenous and exogenous bacteria (Gorloff, 1983; Yaron, 1993; Waller *et al.*, 2004). Chemically preserved fresh aloe gel that is stored at room temperature or is incubated at 40° C for 48 hours exhibits degradation in its rheological properties, a decrease in the content and composition of polysaccharides, and a substantial increase in the mannose:glucose ratio, from 2.9:1 in the fresh aloe gel to 13.4:1 in the incubated aloe gel (Yaron, 1993). Others have found significant variation in commercial products' content when compared with plant-derived aloe gel (Mandal and Das, 1980a,b; Ross *et al.*, 1997; Turner *et al.*, 2004).

Whole Leaf Extract

Whole leaf extract, also referred to as whole leaf Aloe vera juice, is the aqueous extract of the whole Aloe vera leaf with lignified fibers removed. The whole leaf extract contains both the aloe gel and aloe latex. Aloe latex is a bitter, yellow plant exudate that is stored and transported along the margins of Aloe vera leaves via pericyclic tubules, which are located within the leaf pulp just beneath and adjacent to the leaf rind (Figure 1). The restricted distribution of the latex within the margins of the Aloe vera plant leaf suggests that aloe latex is a source of secondary metabolites, compounds that do not function directly in plant growth and development and often serve as a plant defense strategy (Chauser-Volfson and Gutterman, 1996; Wink, 2003). A wide variety of secondary compounds has been isolated from the aloe latex (Reynolds, 1985a). The isolated compounds are largely phenolic in nature, and many are anthraquinone *C*-glycosides, anthrones, and free anthraquinones (Park *et al.*, 1998). The levels of anthraquinone *C*-glycosides in aloe latex are quite variable; however, they may constitute up to 30% of the dry weight of aloe latex (Groom and Reynolds, 1987). Aloe latex contains four major *C*-glycosyl constituents: aloin A, aloin B, Aloesin, and Aloeresin A (Figure 2) (Saccù *et al.*, 2001). Aloin A, a *C*-glycoside anthrone, also referred to as barbaloin, is the major component of the latex (Birch and Donovan, 1955; Hay and Haynes, 1956; Reynolds, 1985b). Aloin A and its epimer, Aloin B, also referred to as isobarbaloin (Figure 2), have a 9-anthrone skeleton and a β-D-glucopyranosyl substituent (Mannitto *et al.*, 1990). Aloesin, also knownas Aloeresin B, is a 5-methyl chromone with an 8-β-D-glucopyranosyl substituent (Haynes *et al.*, 1970),and Aloeresin A is a 5-methyl chromone withan 8-β-D-glucopyranosyl-2-*O*-trans-*p*-coumarol substituent (Gramatica *et al.*, 1982). Several other *O*- and *C*-glycosides of anthrones and anthraquinones, as well as free anthrones, dianthrones, and a small amount of free anthraquinones, have been isolated from aloe latex, including aloe-emodin, an anthraquinone derived from barbaloin and isobarbaloin (Zonta *et al.*, 1995; Okamura *et al.*, 1996, 1997; Saleem *et al.*, 1997; Park *et al.*, 1998). The occurrence of endogenous free anthraquinones and anthrones in aloe latex results from oxidative processes rather than from metabolic synthesis (Figure 2) (Franz and Grün, 1983; Hattori *et al.*, 1988; Saleem *et al.*, 1997). In addition, aloe latex contains a number of aromatic aldehydes and ketones (Saccù *et al.*, 2001). On a dry weight basis, aloe latex is reported to also contain an acid insoluble resin (16% to 63%), significant ash content (24.5%), and a small quantity of essential oils that are responsible for the odor of the latex (Mapp and McCarthy, 1970). The sugar moiety in aloins is D-glucose, and studies indicate that carbon atom 1 of the D-glucose moiety is linked directly to carbon atom 10 of the anthracene ring in a β-configuration. The carbon–carbon bond is quite resistant to acid and alkaline conditions, and cleavage by oxidation, rather than hydrolysis, is achieved only under the drastic conditions of acid in combination with an oxidant (Hay and Haynes, 1956). In addition, the β-(1→10) C–C bond is resistant to β–glucosidases of plants and most plant bacteria (Vyth and Kamp, 1979; Joshi, 1998). Intestinal microflora of humans and animals are able to cleave the β-*C*-glucosyl bond, although considerable variation in response among animal species is noted (Mapp and McCarthy, 1970; Hattori *et al.*, 1988; Che *et al.*, 1991).

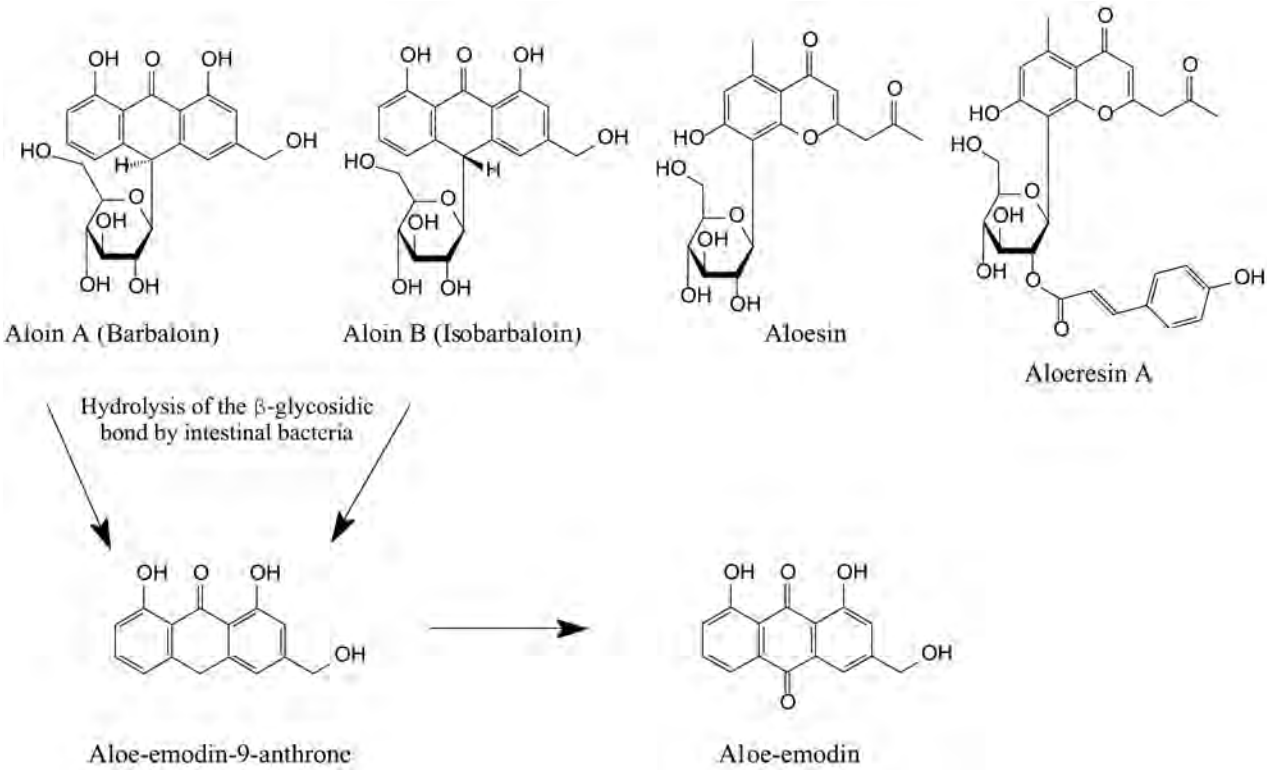


FIGURE 2
Structures of Aloe vera Latex-derived Anthraquinone *C*-glycosides, Anthrone, and Anthraquinone

Decolorized Whole Leaf Extract

Whole leaf extract that undergoes activated carbon adsorption to remove the phenolic components of aloe latex is termed decolorized whole leaf extract and is also referred to as whole leaf Aloe vera gel. Although aloe gel and the decolorized whole leaf extract are similar in that each contain little or no aloe latex, charcoal adsorption changes the physical and chemical properties of the whole leaf extract. Decolorized whole leaf extract differs from aloe gel in that it exhibits a degradation in rheological properties and a loss of approximately 19% to 23% of the complex polysaccharide content (Pelley *et al.*, 1998).

Aloe-emodin

Aloe-emodin is a plant-derived hydroxy-anthraquinone that is present in low levels in the leaves and roots of a number of plants such as Aloe vera. Aloe-emodin occurs as orange crystalline needles, with a melting point of 223° to 224° C when recrystallized with toluene. It is soluble in hot alcohol and in ether, benzene (yielding a yellow color), ammonia water, and sulfuric acid (yielding a crimson color) (*Merck*, 1989).

Production, Use, and Human Exposure

Aloe barbadensis Miller, commonly referred to as Aloe vera, is one of approximately 420 species of Aloe belonging to the lily family (family *Liliaceae*, tribe *Aloineae*) that originated in South Africa but is now indigenous to dry subtropical and tropical climates, including the southern United States (Grindlay and Reynolds, 1986; Viljoen and Van Wyk, 2000). Aloes are perennial succulents or xerophytes, and as such, they are adaptable to habitats with low or erratic water availability, are characterized by the capacity to store large volumes of water in their tissue, and are able to utilize CAM, which is an adaptation to the photosynthetic pathway in hot climates that involves the formation of malic acid (Ni *et al.*, 2004; Takahashi *et al.*, 2005). Only a few species of Aloe are of commercial importance; Aloe vera is considered to be the most potent and, therefore, is the most popular (Eshun and He, 2004; Ni *et al.*, 2004).

The most common Aloe vera extracts for topically applied cosmetics and skin care products are aloe gel and decolorized whole leaf extracts; whole leaf extracts, which contain both aloe gel and aloe latex, also appear in suntan lotions and in sunburn skin treatments (Grindlay and Reynolds, 1986). While some retail products state the source of the principle Aloe vera ingredients, many do not. Both classes of Aloe vera leaf products, aloe gel and aloe latex, are reported to possess a wide range of pharmacologic activities; however, these claims are not supported by well-controlled studies.

Considerable variation in the quality of Aloe vera plant products exists due to differences in growing, harvesting, processing, and storage techniques. Harvesting of Aloe vera plant leaves is generally performed by hand (Grindlay and Reynolds, 1986). At the processing step, the leaves are cleaned with water and a mild chlorine solution. Aloe gel is obtained from the fillet of the leaf pulp either by manual removal of the outer layers of the leaf with a knife or by machine. Either method is flawed and has the potential to contaminate the aloe gel with aloe latex (Grindlay and Reynolds, 1986). The whole leaf extract is obtained by grinding the whole, fresh leaves without removal of the rind. Extraneous material and lignified fibers are then removed by homogenizing and filtering the whole leaf extract (Yaron, 1993). Activated carbon adsorption to produce decolorized whole leaf extract is the first processing step where an extract is intentionally subjected to chemical alteration. Decolorized whole leaf extract has a lower content of complex polysaccharides than either aloe gel or the whole leaf extracts (Pelley *et al.*, 1998). The processed extracts are difficult to keep stable, a problem that may cause differences in product potency; therefore, the aloe gel or whole leaf extracts may undergo a "stabilization" process before being bottled. This process involves pasteurization, adulteration with chemical preservatives and additives, concentration, and/or drying (Gorloff, 1983; Grindlay and Reynolds, 1986; Yaron, 1993; Simal *et al.*, 2000).

Aloe vera plant extracts are currently found as ingredients in a myriad of health care and cosmetic products (Table I1) (CIR, 2004). Aloe vera leaf extracts are used as external analgesics, humectants, oral care agents, and skin conditioning agents in cosmetics (CTFA, 2004). The leaf polysaccharides of the Aloe vera plant are used in cosmetics as film formers, humectants, and skin-conditioners, and the Aloe vera leaf water is used as a fragrance ingredient (CTFA, 2004). Aloe vera is available in a large range of skin moisturizers, face and hand creams, cleansers, soaps, suntan lotions, shampoos and hair tonics, shaving preparations, bath aids, makeup and

fragrance preparations, and baby lotions and wipes (Gallagher and Gray, 2003).

Absorption, Distribution, Metabolism, and Excretion

Aloe Gel

There are no reports of studies to determine the absorption, distribution, metabolism, or excretion of topically applied aloe gel in animal or human skin. Metabolic disposition studies on aloe gel after oral or intravenous administration have been conducted. These studies will be discussed in a subsequent Technical Report that examines the effects of oral administration of Aloe vera in rats and mice.

Whole Leaf Extract

The ability of free anthraquinones, anthranoid glycosides, or aloe latex to be absorbed, distributed, metabolized, and excreted has been studied following oral ingestion; however, studies have not examined these effects following topical administration of aloe latex or whole leaf aloe extracts in experimental animals or humans.

Decolorized Whole Leaf Extract

There are no reports of studies to determine the absorption, distribution, metabolism, or excretion of topically applied decolorized whole leaf extract in experimental animals or humans.

Aloe-emodin

Pharmacokinetic studies after oral administration of aloe-emodin have been conducted in experimental animals and humans; however, there are no reports of studies that examined the absorption, distribution, metabolism, or excretion of topically applied aloe-emodin in experimental animals or humans.

Biological Effects

Aloe Gel

Cell Proliferation

In vitro: There are several reports about the stimulatory effects of Aloe vera components on cell proliferation (Danhof and McAnalley, 1983; Davis *et al.*, 1987); however, the identities of the substances responsible for influencing cell proliferation are currently not known. Because no single definitive active ingredient has been identified, some authors suggest that there may be synergism between the polysaccharides and other components in the aloe gel.

The occurrence of lectin-like substances in Aloe vera was first described by Winters *et al.* (1981), who reported that fractions prepared by differential centrifugation from fresh leaf and commercial Aloe vera extracts contained high levels of lectin-like substances. Fresh leaf fractions of Aloe vera promoted the attachment and growth of normal human cells but not tumor cells, while the commercial Aloe vera fractions were found equally cytotoxic to normal human cells and tumor cells. Substances in fractions of Aloe vera whole leaf and gel extracts were found to induce proliferation in fibroblast and neuron-like cells (Bouthet *et al.*, 1995). Aloe vera was more potent in stimulating the growth of cells when treated prior to attachment than in the treatment of adherent cell cultures. Since the adherence of cells to a matrix is an essential factor for growth, the results suggested that Aloe vera may improve cell attachment. The number of human fibroblasts treated with fresh Aloe vera was shown to increase in a dose-dependent fashion, while cytotoxicity was observed in cells treated with aloe latex (Danhof and McAnalley, 1983). In contrast to the effects observed with treatment with plant-derived Aloe vera, a commercial gel was found to have the opposite effect, suggesting that substances were added during the processing that altered the lectin-like activities and resulted in the disruption of cell attachment and growth. However, when cytotoxicity assays were conducted with an *in vitro* system that mimicked human skin, the effective concentration to kill 50% of cells (EC_{50}) could not be determined, since the Aloe vera at a 100% concentration was found essentially nontoxic and actually stimulated cellular activity (Bowles, 1994).

Experimental animals: Burn or wound healing is a response to tissue injury resulting in the restoration of tissue integrity. The growth of endothelial, epithelial, and fibroblast cells plays a critical role in wound healing processes (Mantle *et al.*, 2001). Animal studies and clinical trials on the proliferative effects of Aloe vera have focused primarily on the duration to reepithelialization of wounds, with few, if any, studies examining the effects of Aloe vera on healthy skin. The results from published studies are conflicting and likely reflect the use of different, poorly characterized, complex commercial products,

rather than plant-derived components, making comparisons difficult.

The wound healing effects of plant-derived Aloe vera were compared with that of a 1% silver sulfadiazine cream on second-degree burns in guinea pigs (Kaufman *et al.*, 1988). The postburn reepithelialization, wound contraction, and hair follicle count were significantly lower in animals treated with Aloe vera compared with silver sulfadiazine-treated animals; however, the thickness of tissue granulation was significantly higher, suggesting that topical application of Aloe vera hindered the wound healing process. Kaufman *et al.* (1988) reported similar results in guinea pigs when Aloe vera was used to treat burn wounds. When topical agents, including scarlet red ointment, benzoyl peroxide lotion, bacitracin ointment, silver sulfadiazine cream, plant-derived Aloe vera, tretinoin cream, capsaicin cream, and mupirocin ointment, were compared for their wound healing effects on full-thickness excision wounds in guinea pigs, Aloe vera was found to have no effect on the rate of reepithelialization or wound contraction (Watcher and Wheeland, 1989).

Clinical trials: The temporal and histological effects of Aloe vera were compared with Vaseline™ gauze treatment in 27 male and female patients with partial thickness burn wounds (Visuthikosol *et al.*, 1995). The Aloe vera used in this study was from Thailand, and the plant species was not specified. Aloe vera treatment enhanced angiogenesis and collagen formation and shortened healing times. In contrast, a significant delay in wound healing was associated with the use of a commercial preparation of Aloe vera compared with a standard wound management procedure in 21 women with wound complications from gynecologic surgery (Schmidt and Greenspoon, 1991).

Clinical trials have also examined the effectiveness of Aloe vera in the treatment of radiation-induced dermatitis and psoriasis. The ability of Aloe vera to heal ulcers, rashes, or poorly healed scars associated with radiation therapy was evaluated in two phase III trials in women who had undergone radiation therapy to the breast or chest wall (Williams *et al.*, 1996). The plant-derived Aloe vera was incorporated into an inert gel. The first double-blind, placebo-controlled study examined the effect of Aloe vera versus a placebo gel on 194 patients undergoing radiotherapy for breast cancer. There was no difference between the treatment and placebo groups. It was speculated that the inert carrier gel might have some beneficial effect, so a second trial study was conducted with 108 patients to compare Aloe vera with no treatment. The trial was not blinded; however, the results were identical on both treatment arms, with Aloe vera offering no benefit. Similar effects were observed in a phase III study involving 225 patients who received either topical Aloe vera or aqueous cream applications three times a day throughout the radiation treatment and for two additional weeks after completion of radiation treatment (Heggie *et al.*, 2002). The aqueous cream was significantly better than Aloe vera in reducing treatment-related side effects.

The efficacy of a plant-derived and preserved, but otherwise untreated, Aloe vera test agent in the treatment of psoriasis was examined in a double-blind, placebo-controlled, randomized, intraindividual right/left comparison study. The Aloe vera or a placebo gel was applied twice daily to symmetrical test lesions for a period of 4 weeks. The sum score of erythema, infiltration, and desquamation was measured and was found to be statistically significant in favor of the placebo treatment (Paulsen *et al.*, 2005).

Immunomodulatory Effects

The ability of Aloe vera and polysaccharides to reduce the severity of acute inflammatory responses has been evaluated in several animal models. Unfortunately, many of these studies were conducted using either complex commercial products or poorly characterized plant components. A commercial topical Aloe vera solution was used to evaluate leukocyte adhesion and cytokine production in a second-degree burn wound rat model (Duansak *et al.*, 2003). Burn wounds were untreated or treated with either saline or Aloe vera. The daily treatment with Aloe vera significantly reduced leukocyte adhesion and serum levels of TNF-α and IL-6. Saline treatment also decreased levels of these parameters, but significance was only observed for serum levels of IL-6.

The ability of Aloe vera extract to influence lymphocyte function and prevent the suppression of delayed-type hypersensitivity and contact hypersensitivity by ultraviolet (UV) irradiation has been examined by several investigators. Strickland *et al.* (1994) investigated the ability of Aloe vera to ameliorate the UV irradiation-induced immune suppression in mice. UVB irradiation and fluorescein isothiocynate sensitization was used to induce local immune suppression. A plant source of Aloe vera was incorporated into a cream base consisting of petrolatum, mineral oil, and lanolin. The topical application

of the Aloe vera cream to irradiated skin ameliorated UVB suppression of contact hypersensitivity and delayed-type hypersensitivity responses. Aloe vera did not prevent the formation of UV-induced DNA pyrimidine dimers nor did Aloe vera accelerate the repair of these lesions in UV-irradiated mouse skin; however, the Aloe vera partially protected the number and morphology of accessory cells such as Langerhan and dendritic epidermal cells. In an effort to identify and characterize the Aloe vera components that offered protection against UVB-impairment of accessory cell function, Lee *et al.* (1999) conducted *in vitro* studies and isolated at least two small-molecular-weight (less than 1 kDa) immuno - modulators that were capable of restoring UVB-suppressed activity *in vivo*. The components offered 50% and 81% recovery of accessory cells at low UVB doses (less than 180 J/m^2) but were ineffective at higher UVB doses.

Byeon *et al.* (1998) investigated whether protection from UV-induced immunosuppression was afforded by single or multiple agents and whether protection decreased upon storage of the gel. For these studies, mice were administered UV irradiation in combination with topical application of a crude aloe gel extract or a highly purified oligosaccharide fraction. The results indicated that maximum protection of contact hypersensitivity was afforded by the gel extract. However, the activity of the gel was shown to decay with time, despite storage of the material as lyophilized powder. Within 3 to 9 months of storage, none of the gels prevented UV-induced suppression of contact hypersensitivity, and the commercial preparations of gel were uniformly inactive, even when tested within a month after being prepared. In contrast, time and storage of the gel extracts had little effect on the protection afforded for delayed-type hypersensitivity, even after 12 months of storage, suggesting that protection from UV-induced radiation is mediated by at least two separate factors in crude aloe gel extract. In addition, a purified oligosaccharide fraction offered *in vivo* protection against UV-induced suppression of delayed-type hypersensitivity. Furthermore, when mice were injected with spent culture media of keratinocytes that were exposed to the oligosaccharide and UVB irradiation, the media reduced IL-10 levels and blocked the immunosuppression of UV irradiation, suggesting that oligosaccharides may prevent UV-induced suppression of delayed-type hypersensitivity by suppressing keratinocyte-derived cytokines.

The effects of Aloe vera on UVB-induced erythema and increased blood flow was examined in 12 male and female volunteers (Crowell *et al.*, 1989). The men and women received UVB irradiation to two sites on each forearm and hourly applications of plant-derived Aloe vera. Doppler blood flow measurements and clinical assessment of erythema at 6 and 24 hours indicated there were no significant alterations in blood or qualitative differences in erythema in aloe-treated areas compared with untreated control sites.

Whole Leaf Extract

Antioxidant/Oxidative Effects

The antioxidant activities of anthraquinone and anthrones of Aloe vera have been evaluated using different model systems (Hutter *et al.*, 1996; Lee *et al.*, 2000; Yen *et al.*, 2000). An aloesin derivative from Aloe vera was found to exhibit potent antioxidant activity and inhibit cyclooxygenase-2 and thromboxane A_2 synthase. In the presence of a lipid, methyl linoleate, lipid peroxidation was induced by the UVA irradiation of two Aloe vera whole leaf extracts that differed in their anthraquinone content; however, the amounts of lipid peroxides formed were higher in the leaf extract that contained lower amounts of anthraquinones (Xia *et al.*, 2007).

Decolorized Whole Leaf Extract

Biological effects induced by decolorized whole leaf Aloe vera extracts *per se* were not found; however, in the previous discussion of results from published studies, the test substances described in these studies reflect the use of different, poorly characterized crude extracts from plants or complex commercial products. Though not stated as such, many of these test substances are likely to contain the decolorized whole leaf extract.

Aloe-emodin

Antioxidant/Oxidative Effects

In vitro studies on the photobiological and photochemical properties of aloe-emodin were conducted in human skin fibroblasts (Wamer *et al.*, 2003). Cells were incubated with aloe-emodin and exposed to UV or visible light. Cells pretreated with aloe-emodin showed increased sensitivity to both UVA and visible light. Significant photooxidative damage to both RNA and DNA was associated with the phototoxicity induced by aloe-emodin. Oxidative damage was observed even at

low levels of phototoxicity, which suggested that photooxidative damage may cause, rather than result from, cellular death induced by aloe-emodin. The phototoxicity mechanism for aloe-emodin appears to involve the generation of reactive oxygen species and the subsequent formation of stable photoproducts with cellular components (Vargas *et al.*, 2002). Aloe-emodin was found to generate singlet oxygen efficiently when irradiated with UV light, and the survival of human skin fibroblast in the presence of aloe-emodin was found to decrease when irradiated (Vath *et al.*, 2002).

Reproductive and Developmental Toxicity

Reviews of literature revealed no reproductive, embryolethal, teratogenic, or developmental toxicity associated with the topical application of Aloe vera (Mengs, 1986; Parry and Matambo, 1992).

Carcinogenicity

No studies were found that examined the effects of chronic topical application of Aloe vera plant extracts and exposure to ultraviolet radiation in experimental animals or humans.

Dermal Toxicity

Aloe Gel

Despite reports of the wound-healing and emollient properties of aloe gel, the topical application of aloe gel has been reported to cause contact dermatitis, erythema, and photodermatitis (Hunter and Frumkin, 1991; Domínguez-Soto, 1992; Ernst, 2000).

Whole Leaf Extract

Reviews of the literature revealed no studies that examined dermal toxicity associated with the topical application of whole leaf extracts of Aloe vera.

Aloe-emodin

Reviews of the literature revealed no studies that examined dermal toxicity associated with the topical application of aloe-emodin.

Mutagenicity/Genotoxicity

Aloe Gel

Reviews of the literature revealed no studies that examined mutagenicity or genotoxicity associated with aloe gel.

Whole Leaf Extract

Tumor-promoting, as well as antimutagenic activities, have been ascribed to aloe latex. SW480 colorectal tumor cells, VACO235 adenoma cells, and normal colonic epithelial cells were exposed to dihydroxyanthraquinones (0.2 to 5 mg/mL) present in laxatives to determine if these compounds stimulated growth and the secretion of urokinase (Schörkhuber *et al.*, 1998). Concentrations of 5 mg/mL caused between 50% to 70% cell loss in colorectal carcinoma SW480 cells; however, DNA synthesis was not similarly reduced. Dihydroxyanthraquinone treatment caused an approximate doubling in the number of premalignant VACO235 cells, whereas the growth of normal rat colonic epithelial cells was not affected. Urokinase secretion was increased by all dihydroxyanthraquinones in a dose-dependent manner, and this was the predominant effect of the dihydroxyanthraquinones in the SW480 carcinoma cells. Urokinase facilitates metastasis by matrix degradation and digestion of normal cells, and it was suggested that the release of urokinase caused the loss of cells observed in the SW480 carcinoma line.

Aloe-emodin

Aloe-emodin, a component of aloe latex, and other dihydroxyanthraquinones were examined for activities associated with tumor promotion, such as stimulation of cell proliferation and enhancement of malignant transformation (Wölfle *et al.*, 1990). The *in vivo* treatment of primary rat hepatocytes with danthron, aloe-emodin, crysophanol, and rhein resulted in a two- to threefold increase of DNA synthesis, whereas emodin was essentially inactive. This marked stimulation of DNA synthesis was in the range with other known tumor promoters, such as phenobarbital and hexachlorocyclohexane. The results suggested that anthraquinones possessing hydroxyl groups in two positions may have tumor promoting activities.

Strickland *et al.* (2000) found that painting aloe-emodin in an ethyl alcohol vehicle on the skin of mice in conjunction with exposure to UVB irradiation resulted in the

development of melanin-containing skin tumors. In addition, the application of ethanol and aloe-emodin combined with the exposure of mice to UVB irradiation for 33 weeks was shown to cause mutations in the p53 gene, whereas in the absence of UV irradiation, mice failed to develop tumors or p53 gene mutations (Badgwell *et al.*, 2004).

Mutagenic and genotoxic activities in bacteria and eukaryotic cells have been shown for some, but not all, anthraquinones found in aloe latex. Westendorf *et al.* (1990) investigated naturally occurring hydroxyanthraquinones for mutagenicity and cell-transforming activity. Aloe-emodin exhibited dose-related effects in mutation assays, in rat hepatocyte DNA-repair induction assays, and in assays to determine malignant transformation of C3H/M mouse fibroblasts. Müller *et al.* (1996) investigated the genotoxicities of several anthraquinone derivatives found as natural constituents in plants and showed that some of the 1,8-dihydroxyanthraquinone derivatives, including aloe-emodin, were intercalating agents that inhibited the interaction between topoisomerase II and DNA. The compounds induced a moderate increase in *Tk*-mutations and a dose-dependent induction of micronuclei. A micronucleus test indicated that danthron was more potent than aloe-emodin, which was more potent than emodin. Kodama *et al.* (1987) observed DNA strand breaks and the generation of free radical and hydrogen peroxide by some anthraquinone derivatives from plant sources. Subsequently, Mueller *et al.* (1998a,b) and Mueller and Stopper (1999) showed that some anthraquinone derivatives are biotransformed by cytochrome P450 1A2 *in vitro* and that this may be relevant for the disposition of anthraquinone derivatives *in vivo*.

The dihydroxyanthraquinones emodin, danthron, and aloe-emodin were investigated for genotoxicity in a number of *in vitro* assays, including mutation and micronucleus assays in mouse L5178Y cells, kinetochore analysis, topoisomerase II assay, and comet assays (Müller *et al.*, 1996; Mueller and Stopper 1999). Emodin, danthron, and aloe-emodin reduced the amount of monomer DNA generated by topoisomerase II, indicating that all three compounds were capable of inhibiting the topoisomerase II-mediated decatenation. Furthermore, a modified comet assay showed that pretreatment of the cells with the test compounds reduced the effects of etoposide, an inhibitor of topoisomerase II. Danthron and aloe-emodin, but not emodin, increased the fraction of DNA moving into comet tails at concentrations of 50 µM in single-cell gel-electrophoresis assays. Results of these assays indicated that danthron and aloe-emodin are genotoxic.

Study Rationale

The phototoxicity of topically applied health and cosmetic ingredients is of concern because of the large body surface area that is exposed to sunlight. Studies have shown that a number of secondary metabolites of plant origin, such as the anthraquinones in Aloe vera, are photosensitizers and may be transformed to reactive intermediates when they absorb UV radiation and/or may yield active oxygen species (Hirose *et al.*, 1990; Downum, 1992; Kersten *et al.*, 1999). *In vitro* studies that examined the photobiological and photochemical properties of anthraquinones and anthrones from Aloe vera demonstrated that cells pretreated with anthraquinones and exposed to ultraviolet light A (UVA) generated significant photooxidative damage to both cellular RNA and DNA (Wamer *et al.*, 2003). The mechanism of phototoxicity was shown to involve the generation of reactive oxygen species, the induction of lipid peroxides, and the formation of stable phototoproducts with cellular components (Vargas *et al.*, 2002; Vath *et al.*, 2002; Xia *et al.*, 2007). Recent studies also raise questions about the ability of aloe products to cause cell proliferation and promote cellular growth, processes associated with carcinogenicity. While people who use cosmetic products that contain Aloe vera are unavoidably exposed to sunlight, it is not known whether the use of Aloe vera-containing cosmetics with concomitant exposure to sunlight results in any deleterious effects (Boudreau and Beland, 2006). The purpose of this 1-year photococarcinogenesis study was to assess whether or not the topical application of Aloe vera substances alters the process of photocarcinogenesis in SKH-1 hairless mice exposed to simulated solar light.

MATERIALS AND METHODS

PROCUREMENT AND CHARACTERIZATION

Aloe vera Plant Extracts

Three lyophilized *Aloe barbadensis* Miller (Aloe vera) test articles were obtained from Pangea Phytoceuticals, Inc. (Harlingen, TX), as aloe gel, whole leaf, and decolorized whole leaf extracts (Appendix D). The aloe gel extract (lot numbers 010510AG, 030109AG, and 032903AG) consisted of the inner leaf gel of hand-filleted Aloe vera leaves with the pulp removed. No further treatments were performed on this material. The whole leaf extract (lot numbers 013008ND and 030109ND) was produced by grinding whole Aloe vera leaves and treating the slurry with cellulase (23 mg/L) to remove the rind components and to maximize yields. This product contained the inner leaf gel and the aloe latex. The decolorized whole leaf extract (lot numbers 013008AC and 030221D) was prepared in an identical manner as the whole leaf extract previously described, with the exception that the slurry was further treated with activated carbon (1.0%, w/w) to remove the latex anthraquinone components from the extract. The different lots of *Aloe barbadensis* test articles were combined and used in the 1-year study. Analyses to determine the homogeneity/content of malic acid, aloin A, and aloe-emodin and the average molecular weight of each of the Aloe vera plant extracts were performed by the Chemistry Support Unit at the National Center for Toxicological Research (NCTR, Jefferson, AR) (Appendix D). Glycosyl linkage analyses of alcohol-insoluble fractions of each of the Aloe vera plant extracts were performed by the Complex Carbohydrate Research Center at the University of Georgia (Athens, GA). Reports on analyses performed in support of the study on the effect of Aloe vera on the photocarcinogenicity of simulated solar light are on file at the National Center for Toxicological Research.

High-performance liquid chromatography (HPLC) was used to determine the homogeneity and concentrations of malic acid, aloin A, and aloe-emodin in the aloe gel, whole leaf, and decolorized whole leaf test articles. The average molecular weight of the polysaccharides in each Aloe vera plant extract was determined using size exclusion HPLC. Glycosyl linkage analysis was performed using gas chromatography-mass spectrometry.

To ensure stability, the bulk aloe gel, whole leaf, and decolorized whole leaf test articles were stored in color-coded, high-density polyethylene pails with snap lids at approximately –20° C. At the end of the study, HPLC analyses of the three bulk chemicals were performed by the Chemistry Support Unit at NCTR; no degradation was detected.

Aloe-emodin

Aloe-emodin [1,8-dihydroxy-3-(hydroxymethyl) anthracene-9,10-dione; CAS No. 481-72-1] was purchased from Sigma-Aldrich (St. Louis, MO) and was received in a single lot (013K1075), with a stated purity of ≥95%. The compound was stored in amber vials at approximately 4° C. The purity of a 52 µg/mL solution of aloe-emodin was determined by the Chemistry Support Unit at NCTR using HPLC; the average purity for three replicate samples was 99.8%.

Control Cream

The base cream used as the control cream was obtained from Cosmetech Laboratories, Inc. (Fairfield, NJ) in four lots (CLI 120604-G, -H, -I, and -J). The control cream was formulated according to the recipe described in Table 1. The absence of malic acid, aloin A, and aloe-emodin from the control cream was confirmed for each lot using HPLC. All lots had a specific gravity of 0.962 that remained constant, with pH values of approximately 6.0 and a mean viscosity of 11,400 centipoise. The base cream was formulated with a 10% "hole" for the incorporation of the test articles and was stored in high-density polyethylene bottles with screw-cap lids at approximately 4° C.

Table 1
Formulation of Control Cream Used in the 1-Year Simulated Solar Light Study of Aloe vera

Item No.	Phase[a]	Ingredient	Supplier/Lot No.	% (w/w)	Batch Size(g)
1	A	Deionized Water		63.57	12,713.46
2	A	Glycerin 96%	Compton/381039	4.00	800.00
3	A	Carbopol 981 (2% Soln.)	B.F. Goodrich/ ECIN2Cd484	10.00	2,000.00
4	B	Ceteryl Alcohol	Croda CS5-5130	1.00	200.00
5	B	Mineral Oil 65/75	Penreco/K3008D	3.00	600.00
6	B	Dimethicone DC200-100	Dow Corning/ DC0000530183	1.00	200.00
7	B	DC 345 Cyclomethicone	Dow Corning/ DC00014292997	3.00	600.00
8	B	Lipomulse 165	Lipo/P-196K2	2.50	500.00
9	B	MYRJ 52S	Lipo/P205E2	0.75	150.00
10	C	NaOH (25% Aq. Soln.) to pH 6.0	PCI/000358A	0.18	36.54
11	D	Germaben II	ISP/0420080405	1.00	200.00
		Total		90.00	18,000.00

[a] Manufacturing instructions: heat phase A to 70° C. Heat phase B to 70° C. Add phase B to phase A. Cool to 40° C and add phases C and D. Homogenize mixture for 3 minutes and package.

Preparation and Analysis of Dose Formulations

The dose formulations were prepared twice weekly by dissolving the appropriate amount of each of the four test articles in distilled, deionized water and then thoroughly mixing these solutions with the control cream to provide 10% of the final weight of the dosed creams (Table D2). The dose formulations were dispensed into glass scintillation vials sealed with Teflon®-lined screw caps and stored at approximately 4° C for up to 3 days. Homogeneity studies (a 6% dose formulation of aloe gel and a 78 µg/g dose formulation of aloe-emodin) and stability studies (3% dose formulations for each of the three Aloe vera plant extracts and 7.46 and 74.6 µg/g aloe-emodin dose formulations) were performed by the Chemistry Support Unit using HPLC. Homogeneity was confirmed and stability was confirmed for up to 3 days for samples stored in glass vials sealed with Teflon®-lined screw caps at 4° C.

Weekly dose certifications of the dosed cream formulations of the Aloe vera plant extracts and aloe-emodin were conducted by the Chemistry Support Unit using HPLC. Quantitation of malic acid and aloin A was used for certification of the dose formulation concentrations of the three Aloe vera plant extract test articles (Tables D3 through D5). Dose formulations of aloe-emodin were directly measured for concentrations of the test chemical (Table D6).

Light Source and Irradiance Dosimetry

This study was conducted in a similar manner as that of other photococarcinogenesis studies conducted in the NTP/FDA Center for Phototoxicology at NCTR (NTP, 2007). The sources of irradiance were glass-filtered (WG320/1 mm; SCHOTT North America, Inc., Elmsford, NY) 6.5 kW xenon arc lamps (Atlas Electric, Inc., Spokane, WA), and exposures were monitored with solar light PMA-1101 broad-band dosimeters (Solar

Light Company, Inc., Glenside, PA) attached to the center-front of each animal rack. The source of irradiance is commonly referred to as simulated solar light (SSL). In the present study, the mice were exposed to filtered SSL at 0, 0.3, 0.6, or 0.9 minimal erythema doses (MED) of light (NTP, 2007). MED is defined as the minimal amount of radiation that causes slight erythema within 24 hours after irradiation.

The actual measured exposure to light in this study was based on the convention of the Commission Internationale de l'Eclairage (CIE, 1987, 1999). The light exposures were determined by measuring the irradiance from the SSL source in mW/cm^2 and multiplying the irradiance by the human erythema action spectrum to obtain a weighted irradiance in mW • CIE/cm^2. Since exposure to 1 mW • CIE/cm^2 is equivalent to 1 mJ • CIE/cm^2, the weighted irradiances from the SSL lamp source are multiplied by the length of exposure to calculate the daily exposures received by the mice. Based on this convention, 0.0, 0.3, 0.6, and 0.9 MED doses of SSL were equivalent to 0.00, 6.85, 13.70, and 20.55 mJ • CIE/cm^2, respectively. Daily exposures of light in this Technical Report are given as mJ • CIE/cm^2. Refer to Appendixes E and F for additional information on the spectral irradiance and the dosimetry of the lamp source.

1-Year Study

Study Design

Groups of 36 male and 36 female SKH-1 mice were treated in the morning, 5 days per week, with 75 µL (approximately 2 mg/cm^2) of control cream, 3% or 6% aloe gel cream, 3% or 6% whole leaf cream, 3% or 6% decolorized whole leaf cream, or 7.46 or 74.6 µg/g aloe-emodin cream for 40 weeks; additional groups of 36 male and 36 female mice received no cream (Table 2).

Table 2
Level of Exposure to Simulated Solar Light in Mice in the 1-Year Study of Aloe vera

Cream Application	No Light (0.00 mJ • CIE/cm^2/day)	Low Exposure (6.85 mJ • CIE/cm^2/day)	Medium Exposure (13.70 mJ • CIE/cm^2/day)	High Exposure (20.55 mJ • CIE/cm^2/day)
None	36 males 36 females	36 males 36 females	36 males 36 females	36 males 36 females
Control cream	36 males 36 females		36 males 36 females	
3% Aloe gel			36 males 36 females	
6% Aloe gel	36 males 36 females		36 males 36 females	
3% Whole leaf			36 males 36 females	
6% Whole leaf	36 males 36 females		36 males 36 females	
3% Decolorized whole leaf			36 males 36 females	
6% Decolorized whole leaf	36 males 36 females		36 males 36 females	
Aloe-emodin 7.46 µg/g			36 males 36 females	
Aloe-emodin 74.6 µg/g	36 males 36 females		36 males 36 females	

Mice were irradiated with SSL in the afternoon. Mice that received no cream treatment were exposed to 0.00, 6.85, 13.70, or 20.55 mJ • CIE/cm^2 SSL per day; mice dosed with control cream, 6% aloe gel, 6% whole leaf, 6% decolorized whole leaf, or 74.6 µg/g aloe-emodin cream were exposed to 0.00 or 13.70 mJ • CIE/cm^2 SSL per day; mice dosed with 3% aloe gel, 3% whole leaf, 3% decolorized whole leaf, or 7.46 µg/g aloe-emodin cream were exposed to 13.70 mJ • CIE/cm^2 SSL per day. A 12-week recovery/observation period followed the 40-week treatment/exposure period.

Creams were dispensed with a positive-displacement repeater pipette (Eppendorf Repeater® Plus, Fisher Scientific, Houston, TX) equipped with a 2.5 mL reservoir and were applied with a gloved finger to the dorsal skin region of the animal for 30 seconds. A single-channel, battery operated electronic timer (Fisher Scientific) was used to maintain consistency in the duration of the cream applications. The site of application extended from the nape of the neck to the base of the tail and midway along both sides of the animal. Untreated animals were not handled.

The mice were housed in stainless steel racks that allowed horizontal exposure to the SSL light source. The racks were placed at preset positions around the light source with the front of the animal cage approximately 2 meters from the light source. The duration of exposure was based on the dose of SSL. Typical durations for SSL exposure levels of 6.85, 13.70, and 20.55 mJ • CIE/cm^2 were 30, 70, and 90 minutes, respectively.

Source and Specification of Animals

Male and female Crl:SKH-1 (hr^-/hr^-) BR hairless mice were obtained from Charles River Laboratories (Wilmington, MA). Mice were approximately 4 weeks old upon receipt, quarantined for 2 weeks, and acclimated for 1 week to the animal room environment prior to the start of the study. Mice were approximately 8 weeks old at the beginning of the study. Twelve mice of each sex were examined during quarantine for serological pathogens, parasites, and bacterial pathogens. Additional screenings of sentinel pairs of each sex were conducted at 20, 26, and 39 weeks according to the protocols of the NCTR Sentinel Animal Program (Appendix H).

Animal Maintenance

Mice were housed individually in a compartment, with six compartments per cage, six cages per column, and two columns per rack. Feed and water were available *ad libitum*, except during periods of SSL exposure. Due to the design of the racks, neither feed consumption nor water consumption was measured during the course of the study. Cages were rotated daily, and racks were changed weekly. Further details of animal maintenance are given in Table 3. Information on feed composition and contaminants is provided in Appendix G.

TABLE 3
Experimental Design and Materials and Methods in the 1-Year Simulated Solar Light Study of Aloe vera

Study Laboratory	National Center for Toxicological Research (Jefferson, AR)
Strain and Species	Crl:SKH-1 (hr^-/hr^-) hairless mice
Animal Source	Charles River Laboratories (Wilmington, MA)
Time Held Before Studies	2 weeks quarantine plus 1 week acclimation
Average Age When Studies Began	8 weeks
Date of First Dose and Exposure	May 12, 2003
Duration of Dosing and Exposure	40 weeks cream application and exposure to light; 12 weeks recovery/observation
Date of Last Dose	March 19, 2004

TABLE 3
Experimental Design and Materials and Methods in the 1-Year Simulated Solar Light Study of Aloe vera

Scheduled Necropsy Dates
May 11-June 16, 2004

Average Age at Scheduled Necropsy
61 weeks

Size of Study Groups
36 males and 36 females

Method of Distribution
Animals were distributed randomly into groups of approximately equal initial mean body weights.

Animals per Cage
1 per compartment, six compartments per cage

Method of Animal Identification
Tail tattoo

Diet
Autoclaved NIH-31 open formula meal/pelleted diet (Purina Mills, Richmond, IN), available *ad libitum*, except during light exposure

Water
Millipore-filtered tap water (Jefferson, AR, municipal supply) via automatic watering system (Edstrom Industries, Waterford, WI), available *ad libitum*, except during light exposure

Cages
Lenderking model EXP355-72 stainless steel cage/racks (Lenderking Caging Corp., Millersville, MD)

Animal Room Environment
Temperature: 25° ± 3° C
Relative humidity: 50% ± 20%
Room fluorescent light: 12 hours/day
Room air changes: 10/hour

Dose and Exposure Concentrations
No-cream groups exposed to 0.00, 6.85, 13.70, or 20.55 mJ • CIE/cm^2 SSL per day;
Control cream, 6% aloe gel cream, 6% whole leaf cream, 6% decolorized whole leaf cream, or aloe-emodin 74.6 µg/g cream groups exposed to 0.00 or 13.70 mJ • CIE/cm^2 SSL per day;
3% aloe gel cream, 3% whole leaf cream, 3% decolorized whole leaf cream, or aloe-emodin 7.46 µg/g cream groups exposed to 13.70 mJ • CIE/cm^2 SSL per day

Type and Frequency of Observation
Observed twice daily; body weights were recorded initially, weekly, and when an animal was removed from the study; clinical findings were recorded weekly and when an animal was removed from the study. Skin lesions were digitally photographed weekly.

Method of Sacrifice
Carbon dioxide asphyxiation

Necropsy
Necropsies were performed on all animals, except where noted. Skin lesions were digitally photographed, mapped, and labeled for correlation with microscopic findings.

Histopathology
Histopathology was limited to the examination of gross lesions and tissue masses, skin at the site of application, and control skin samples and was performed on all animals including those that died during the study or were removed early.

Clinical Examinations and Pathology

All animals were observed twice daily. Body weights were recorded initially, weekly, and when an animal was removed from the study; clinical findings were recorded weekly and when an animal was removed from the study. Mice were examined weekly for the presence of skin lesions that were consistent with the development of ultraviolet (UV) light-induced skin tumors. The size of each lesion was measured with a digital vernier caliper (Digimatic 500, Mitutoyo America Corporation, Aurora, IL), and the size and location of the individual skin lesions were recorded in the NCTR Multi-Generation Support System database. Digital images of mice with noted skin lesions were captured each week and were used to determine the in-life skin lesion latency, incidence, and multiplicity. Mice were removed from the study when the size of a skin lesion was greater than or equal to 5 mm. Animals also were removed from the study when individual skin lesions could no longer be discerned due to the merging of the lesions or when the health or welfare of an animal was inconsistent with continuance on the study.

Mice removed from the study due to morbidity, skin lesion size, significant skin lesion merging, or at study termination were euthanized by carbon dioxide inhalation. A complete necropsy was performed on all animals. Digital photographs of mice were taken after euthanasia but prior to necropsy. Gross skin tumor lesions were mapped by the study pathologists and numerically labeled on the printed photographs to serve as a guide for the identification and trimming of trace gross lesions (TGLs) reported on the individual animal necropsy record (IANR) and to correlate the numbers of the TGLs with the microscopic findings of the study pathologist. At necropsy, all major organs and tissues were examined grossly, removed, and placed in 10% neutral buffered formalin fixative. Descriptions of gross lesions were recorded on the IANR. All gross skin lesions were trimmed, processed, mounted in paraffin-plastic polymer blocks, sectioned at 5 µm, and stained with hematoxylin and eosin. Additional samples of normal skin from the right and left front and rear dorsal skin and from the abdominal ventral skin were removed, fixed, and similarly processed. At the discretion of the study pathologist, samples of skin tumor lesions from sacrificed and moribund animals were frozen in liquid nitrogen. The photographically documented gross skin lesions were correlated with the IANR entries, and all sections of skin and skin tumor lesions were examined microscopically. When applicable, nonneoplastic skin lesions were graded for severity.

Tissues other than skin were not evaluated by histopathology. Microscopic evaluations were completed by the study pathologist, and the micropathology data were entered into the NCTR Laboratory Data Acquisition System. A microscopic finding was recorded with the corresponding gross observation for skin when possible. Microscopic findings were tabulated by the individual animal record in the pathology report and classified as either nonneoplastic or neoplastic findings. Neoplasia was further defined upon microscopic evaluation by type (squamous cell papilloma, squamous cell carcinoma *in situ*, and squamous cell carcinoma) and was summarized by counts of each neoplasm type by treatment. As part of the pathological evaluation of skin lesions, the incidences (percentage of animals per treatment with neoplasms or nonneoplastic lesions) by lesion type and the multiplicity (average number of lesions per animal) were determined. After completion of all microscopic evaluations, the tissue slides, paraffin blocks, and residual wet tissues were stored in the NCTR pathology archives.

An internal review of the histopathology data was conducted by a quality control pathologist to assure the accuracy, standardization, and completeness of the histopathology examination and reporting process. The quality control pathologist reviewed the skin and skin lesions from 10% of the animals of each dose group. The quality control pathologist evaluated the gross IANR, the gross-to-microscopic correlation, and the histopathology of each case, and concurrence or nonconcurrence was documented. In the case of nonconcurrence, the quality control pathologist consulted with the study pathologist and consensus was achieved.

An independent quality assessment laboratory conducted an external pathology data review and quality assessment. The pathology quality assessment review consisted of a reexamination by quality assessment pathologists of all slides and diagnoses from 20% of the male and female mice from each of the groups that were not exposed to SSL and from the no cream, control cream, and high dose groups exposed to 13.70 mJ • CIE/cm^2 SSL. No animals were included in the quality assessment review from the male and female no cream treatment groups exposed to 6.85 or 20.55 mJ • CIE/cm^2 SSL. Any differences between the quality assessment pathologists and the study pathologist were discussed,

and differences that were not resolved were forwarded to the Pathology Working Group (PWG) for resolution.

The quality assessment report, pathology tables, and the report of the study pathologist were reviewed by the PWG coordinator, who reviewed selected skin slides and addressed any inconsistencies in the diagnoses made by the study pathologist and quality assessment pathologists. Representative examples of treatment-related skin lesions and examples of inconsistent diagnoses between the study pathologist and the quality assessment pathologists were presented by the coordinator to the PWG for review. The PWG consisted of the quality assessment pathologist, the study pathologist, and other pathologists experienced in laboratory animal pathology. Each participant examined all slides and discussed his/her observations with the group. The final diagnoses for reviewed lesions represent a consensus between the study pathologist, quality assessment pathologists, and the PWG.

Statistical Methods

Animal Removal/Survival Analyses

A Cox proportional hazards regression model was used to analyze the removal/survival data for this study (Cox, 1972). The hazard function is the slope of the removal/survival curve and indicates the instantaneous probability of removal at a given time. Removal rates are expected to change over the course of a study, but at any particular time, the removal hazard functions of any two groups are assumed to be proportional to each other. The ratio of the hazards is essentially a relative risk. The Cox model analyses were performed by treating the study as a large, one-way design with 18 treatment groups for each sex. Hazard ratios were computed relative to the no cream/no SSL group or the control cream only group, as relevant. Mice removed prior to dosing or because of being missexed were eliminated from the removal/survival analyses; otherwise, animals found dead, moribund, or harvested for tumor-size threshold or for excessive tumor burden were considered uncensored, while animals accidentally killed or terminally sacrificed were censored at that point. Because most of the uncensored events fell into the protocol harvest category, this is a removal time analysis rather than a survival analysis.

Differences were considered significant if the single-sided P value was less than 5%. Cox regression model estimates of hazard functions were used to compute the relative hazard ratios for the treated groups as a percentage of the relevant control group. Kaplan-Meier curves (Kaplan and Meier, 1958) along with confidence regions were generated for the contrasts of interest.

Body Weight Analyses

Body weight data were preprocessed using LOESS regression analysis within each animal to minimize issues associated with outliers and to "rasterize" the data into equally spaced intervals (i.e., display weight gain in discrete increments). LOESS regression analysis is a nonparametric data smoothing technique that fits a curve to the data using moving data windows. For the body weight data in this study, the windows were the time points from 0 to 52 weeks in 4-week intervals, with zero corresponding to the start of the study.

A mixed effects linear model with Aloe vera dose level as the predictor and time as a repeated measures predictor was fit to the smoothed, rasterized data using an autoregressive covariance structure to accommodate dependence among repeated measurements of body weights on the same animals. The data were treated as 11 one-way designs for each sex, representing each of the four Aloe vera test articles, the control cream, and the no cream control groups for each of 0.00 and 13.70 mJ • CIE/cm^2 SSL, plus one SSL-only analysis for the no cream mice. Another analysis was run for each sex on the no cream animals using SSL as the predictor. Contrasts were used to compare Aloe vera and SSL treatment groups and compute linear trends by weeks on study. For the treatment groups that received no cream, the exposure levels of SSL were evenly spaced (0.00, 6.85, 13.70, and 20.55 mJ • CIE/cm^2), so linear trends were tested. The reported P values were two sided, and values less than or equal to 0.05 were considered statistically significant.

Analysis of Skin Lesion Onset, Incidence, and Multiplicity

In-Life Assessment of Skin Lesion Onset and Incidence

Digital images of mice with noted skin lesions were captured each week of the study and were used to determine in-life skin lesion onset, incidence, and multiplicity. A Cox regression model was used to compare first lesion onset times among dose groups. The data were treated as 11 one-way designs for each sex, representing each of the four Aloe vera test articles, the control cream, and the no cream control groups for each of 0.00 and 13.70 mJ • CIE/cm^2 SSL, plus one SSL-only analysis for the no

cream mice. For each sex, an additional analysis was run for the SSL-only groups using SSL level as the predictor. The time of appearance of skin lesions was considered an uncensored event; animals that developed no skin lesions were considered to be censored at their removal times. Kaplan-Meier curves also were generated, along with the mean and median time to lesiononset for each treatment group. P values were determined from the Cox model using contrasts with the no cream/no SSL group or with the within-SSL control cream group, where appropriate. These P values are two sided.

In a second approach, the in-life skin lesion incidence was compared among Aloe vera dose groups using the continuity-corrected Poly-3 test (Bailer and Portier, 1988a,b; Portier and Bailer, 1989) with Bieler and Williams' (1993) modification for each sex, SSL level, and Aloe vera test article. The P values presented for the in-life skin lesion incidence are two sided. Two-sided tests were used since Aloe vera was being evaluated as a cocarcinogen and since it was possible for the cream to exhibit a beneficial effect. Because of the sparsity of skin lesions in mice that were not exposed to SSL, no analyses were conducted on these data. It should be noted that this test was run for each study week following a convention established in previous phototoxicity studies (NTP, 2007).

Histopathology Tumor Incidence

Calculation of Tumor Incidence

The incidences of neoplasms or nonneoplastic skin lesions are presented in Tables A1, A3, B1, and B3 as the number of animals bearing such lesions at a specific anatomic site and the number of animals with that site examined microscopically. For calculation of statistical significance, the incidences of most neoplasms (Tables A2 and B2) and all nonneoplastic lesions are given as the numbers of animals affected at each site examined microscopically. Tables A2 and B2 also give the survival-adjusted neoplasm rate for each group and each site-specific neoplasm. This age-adjusted rate accounts for differential mortality by assigning a reduced risk of neoplasm, proportional to the third power of the fraction of time on study, only to site-specific, lesion-free animals that do not reach terminal sacrifice.

Analysis of Histopathology Tumor Incidence

A statistical analysis was conducted for any neoplastic or nonneoplastic skin lesion having an incidence of three or more in any group. The histopathology data were treated as 11 one-way designs for each sex, representing each of the four Aloe vera test articles, the control cream, and the no cream control groups for each of 0.00 and 13.70 mJ • CIE/cm^2 SSL, plus one SSL-only analysis for the no cream mice. The incidences of neoplastic and nonneoplastic skin lesions were analyzed using the Poly-3 test with Bieler and Williams' (1993) modification. Equally spaced test article doses were used in all tests, although the dose concentrations for aloe-emodin (0, 7.46, and 74.6 µg/g) were not evenly spaced. Tests of significance included pairwise comparisons of each treated group with controls and a test for an overall dose-related trend. Continuity-corrected Poly-3 tests were used in the analysis of lesion incidence, and reported values are one sided. The significance of lower incidences or decreasing trends in lesions is represented as 1– P with the letter N added (e.g., P = 0.99 is presented as P = 0.01N).

Analysis Of Skin Lesion Multiplicity

In-life Assessment of Skin Lesion Multiplicity

Two models were used to analyze the in-life tumor multiplicity results, a Poisson regression model and a modified Wei-Lin-Weisfeld (WLW) model. The two methods produced similar findings, and the results reported are from the more common Poisson regression model. In all models, the data were treated as 11 one-way designs for each sex, representing each of the four Aloe vera test articles, the control cream, and the no cream control groups for each of 0.00 and 13.70 mJ • CIE/cm^2 SSL, plus one SSL-only analysis for the no cream mice.

The endpoint for the skin lesion multiplicity was an ever growing count of lesions over time. Of course, once an animal is removed from the study, its count no longer grows. Given the large number of early removals, the analysis should account for this. Prior analyses have compared the dose groups at each week using a Kruskal-Wallis analysis (NTP, 2007). The difficulty with this method is that the time chosen to focus upon is arbitrary and that it has no inherent method to account for removals that occur before the week being analyzed.

For this study, the Kruskal-Wallis analysis was dropped in favor of a Poisson regression using the logarithm of removal week as an offset. The inferences are, therefore, on the rate of lesion formation rather than the count. Overdispersion was examined and found not to be a

problem with this endpoint. Since the rate of lesion formation may vary over time, the analysis was conducted at each week as in previous studies. Analyzing by week requires shifting the offset time suitably for each week. The final week results are regarded as definitive, although the early results are of potential interest. Suitable contrasts were used to form pairwise comparisons. As in the rest of the analyses, aloe-emodin was treated as evenly spaced for the purposes of linear trend tests even though it was not.

The SSL-only analysis contained weeks with no lesions in the no SSL and low SSL groups of animals. The analysis above cannot handle zero-incidence groups. For this analysis, conducted by week, fractional lesions were added to these groups. The results are conservative since adding fractional lesions to these groups will lessen the significance of the SSL effect.

The tumor burden plots of the Poisson analysis above suggested that the lesion formation rate might be changing in a way related to Aloe vera dose, but by the final week, the rates had evened out. A modified WLW analysis was used in the hope of detecting this transient difference in treatments. It is really just a multivariate Cox model that handles within-animal correlation using a sandwich estimator. This analysis appeared to perform well in a previous study (NTP, 2007), but it did not detect a significant transient difference in this study and did not appear to perform as well.

The reported P values are two sided, and values less than 5% are considered statistically significant.

Analysis of Histopathology Tumor Multiplicity

The histopathology tumor multiplicities are numbers of tumors of specific morphology type present at necropsy for each animal. Since early removal is a common event in this study, an adjustment for age is an important consideration. The response is a count of tumors for each animal, which suggested a Poisson model. The natural extension to age-adjust a Poisson model is to use age at removal as an offset time to create a Poisson rate. Since overdispersion did appear to be present, a negative binomial model was used rather than the simpler pure Poisson model. For each morphology type and pool considered, the data were treated as 11 one-way designs for each sex, representing each of the four Aloe vera test articles, the control cream, and the no cream control groups for each of 0.00 and 13.70 mJ • CIE/cm^2 SSL, plus one SSL-only analysis for the no cream mice. Contrasts were used to compare doses to control and to form tests of linear trend.

As a confirmation of the Poisson results, a relative effects model (Brunner *et al.*, 2001) was run on the animal-specific removal-corrected lesion rates (lesion count divided by weeks on study for each animal). The results of the relative effects model analysis were similar and were not reported since the Poisson model was considered a more powerful statistical model.

The reported P values are one sided, and values less than 5% are considered statistically significant. P values corresponding to beneficial trends have an N appended.

Quality Assurance Methods

This study was conducted in compliance with Food and Drug Administration Good Laboratory Practice Regulations (21CFR, Part 58). The Quality Assurance Unit of the NCTR performed audits and inspections of protocols, procedures, data, and reports throughout the course of these studies. Independent and separate audits were conducted for the completeness and accuracy of the pathology data, pathology tables, pathology specimens, and the pathology report.

RESULTS

REMOVAL/SURVIVAL

The dispositions of mice on the 1-year photococarcinogenesis study are shown in Table 4. Thirty-six male and 36 female mice were initially allocated to each of the 18 different treatment groups. With the exception of three missexed mice, one mouse that died prior to dosing, and the autolysis of one animal, all mice were microscopically examined by the study pathologist.

No Cream

The effects of SSL exposure on the Cox survival hazard ratios for male and female mice that did not receive cream treatment are summarized in Table 5 and graphically represented with Kaplan-Meier survival curves in Figure 3. The control group in this analysis was the group of mice that was not exposed to simulated solar light (SSL). When compared to the survival of mice not exposed to SSL, the survival of male and female mice exposed to 6.85, 13.70, or 20.55 mJ • CIE/cm^2 SSL showed a significant exposure-related increase in the hazard ratios and a decrease in survival. In male mice, exposure to SSL (6.85, 13.70, or 20.55 mJ • CIE/cm^2) significantly decreased the likelihood of survival and increased the relative hazards ratios by 17-, 138-, and 1,208-fold, respectively. Female mice exposed to 13.70 or 20.55 mJ • CIE/cm^2 SSL had significantly higher Cox relative hazard ratios (20- and 235-fold, respectively) when compared with the control group.

Control Cream

The effect of SSL exposure on the survival of male and female mice administered control cream is shown in Table 5 and is graphically represented in Figure 4. The survival of mice that were exposed to 13.70 mJ • CIE/cm^2 SSL was significantly decreased by 29- and 18-fold in males and females, respectively, when compared with control cream mice not exposed to SSL. At the same SSL exposure level, the Cox hazard ratios for male and female control cream mice were not significantly different from those of the no cream mice (Table 6).

Aloe Gel, Whole Leaf, Decolorized Whole Leaf, and Aloe-emodin Creams

The effects of SSL exposure on the survival of mice administered aloe gel, whole leaf, decolorized whole leaf, or aloe-emodin creams are compared in Table 5. In pairwise comparison tests with animals that were not exposed to SSL but received the same cream treatment, mice exposed to SSL had significantly decreased survival and increased hazard ratios, regardless of the test article treatment. Kaplan-Meier survival curves are shown for mice administered aloe gel (Figure 5), whole leaf (Figure 6), decolorized whole leaf (Figure 7), or aloe-emodin (Figure 8) creams, and a statistical summary of the Cox survival hazard ratios by SSL level relative to the control cream groups is presented in Table 6. In the absence of SSL, the survival curves of male and female mice treated with aloe gel, whole leaf, or decolorized whole leaf creams and the survival curve of female mice treated with aloe-emodin were shifted to the right of the survival curves for the control cream mice, suggesting improved survival with the application of the dosed cream treatments. The Cox hazard ratios of these same treatment groups were also lower than those of the control cream mice; however, with the exception of female mice treated with aloe-emodin, differences in the Cox hazard ratios were not significant (Table 6). In male and female mice exposed to SSL, the survival curves of mice that received aloe gel, whole leaf, or decolorized whole leaf creams were similar to the control cream groups, and there were no significant differences in the Cox hazard ratios (Table 6). Only female mice treated with 7.46 µg/g aloe-emodin cream had a significantly higher Cox hazard ratio and a leftward shift in the survival curve, suggesting decreased survival relative to control cream mice.

TABLE 4
Disposition of Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Test Article	SSL Exposure Level (mJ • CIE/cm^2)			
	0.00	6.85	13.70	20.55
Male				
No Cream				
Mice initially in study	36	36	36	36
Natural death	1	2	0	0
Accident	0	1	0	0
Moribund	0	6	0	3
Harvest (skin lesion ≥5 mm)	0	5	36	33
Terminal sacrifice	35	22	0	0
Discard	0	0	0	0
Examined microscopically	36	36	36	36
Control Cream				
Mice initially in study	36		35	
Natural death	0		1	
Moribund	5		3	
Harvest (skin lesion ≥5 mm)	0		31	
Terminal sacrifice	30		0	
Discard	1[a]		0	
Examined microscopically	35		35[b]	
3% Aloe Gel				
Mice initially in study	0		36	
Natural death			0	
Moribund			5	
Harvest (skin lesion ≥5 mm)			31	
Terminal sacrifice			0	
Discard			0	
Examined microscopically			36	
6% Aloe Gel				
Mice initially in study	36		36	
Natural death	0		0	
Moribund	1		1	
Accidental death	1		0	
Harvest (skin lesion ≥5 mm)	0		35	
Terminal sacrifice	34		0	
Discard	0		0	
Examined microscopically	36		36	
3% Whole Leaf				
Mice initially in study	0		36	
Natural death			2	
Moribund			1	
Harvest (skin lesion ≥5 mm)			32	
Terminal sacrifice			1	
Discard				
Examined microscopically			36	

TABLE 4
Disposition of Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Test Article	SSL Exposure Level (mJ • CIE/cm^2) 0.00	6.85	13.70	20.55
Male (continued)				
6% Whole Leaf				
Mice initially in study	36		36	
Natural death	2		1	
Moribund	4		2	
Harvest (skin lesion ≥5 mm)	0		32	
Terminal sacrifice	30		1	
Discard				
Examined microscopically	35[c]		36	
3% Decolorized Whole Leaf				
Mice initially in study	0		36	
Natural death			1	
Moribund			2	
Harvest (skin lesion ≥5 mm)			33	
Terminal sacrifice			0	
Discard			0	
Examined microscopically			36	
6% Decolorized Whole Leaf				
Mice initially in study	36		36	
Natural death	2		0	
Moribund	3		4	
Harvest (skin lesion ≥5 mm)	1		32	
Terminal sacrifice	30		0	
Discard				
Examined microscopically	36		36	
7.46 µg/gram Aloe-emodin				
Mice initially in study	0		36	
Natural death			2	
Moribund			3	
Harvest (skin lesion ≥5 mm)			30	
Terminal sacrifice			0	
Discard			1[a]	
Examined microscopically			35	
74.6 µg/gram Aloe-emodin				
Mice initially in study	36		36	
Natural death	0		1	
Moribund	7		0	
Harvest (skin lesion ≥5 mm)	1		34	
Terminal sacrifice	27		1	
Discard	1[a]		0	
Examined microscopically	35		36	

TABLE 4
Disposition of Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Test Article	SSL Exposure Level (mJ • CIE/cm^2)			
	0.00	6.85	13.70	20.55
Female				
No Cream				
Mice initially in study	36	36	36	36
Natural death	1	1	0	0
Moribund	5	4	1	1
Harvest (skin lesion ≥5 mm)	0	7	34	35
Terminal sacrifice	30	24	1	0
Examined microscopically	36	36	36	36
Control Cream				
Mice initially in study	36		36	
Natural death	0		0	
Moribund	7		1	
Harvest (skin lesion ≥5 mm)	1		33	
Terminal sacrifice	28		2	
Examined microscopically	36		36	
3% Aloe Gel				
Mice initially in study			36	
Natural death			0	
Moribund			4	
Harvest (skin lesion ≥5 mm)			32	
Terminal sacrifice			0	
Examined microscopically			36	
6% Aloe Gel				
Mice initially in study	36		36	
Natural death	2		1	
Moribund	4		4	
Harvest (skin lesion ≥5 mm)	0		31	
Terminal sacrifice	30		0	
Examined microscopically	36		36	
3% Whole Leaf				
Mice initially in study			36	
Natural death			1	
Moribund			3	
Harvest (skin lesion ≥5 mm)			31	
Terminal sacrifice			1	
Examined microscopically			36	
6% Whole Leaf				
Mice initially in study	36		36	
Natural death	0		3	
Moribund	2		1	
Harvest (skin lesion ≥5 mm)	1		32	
Terminal sacrifice	33		0	
Examined microscopically	36		36	

TABLE 4
Disposition of Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Test Article	SSL Exposure Level (mJ • CIE/cm^2)			
	0.00	6.85	13.70	20.55
Female (continued)				
3% Decolorized Whole Leaf				
Mice initially in study			36	
Natural death			2	
Moribund			0	
Harvest (skin lesion ≥5 mm)			34	
Terminal sacrifice			0	
Examined microscopically			36	
6% Decolorized Whole Leaf				
Mice initially in study	36		36	
Natural death	0		1	
Moribund	4		2	
Harvest (skin lesion ≥5 mm)	0		33	
Terminal sacrifice	32		0	
Examined microscopically	36		36	
7.46 µg/gram Aloe-emodin				
Mice initially in study			36	
Natural death			0	
Moribund			4	
Harvest (skin lesion ≥5 mm)			32	
Terminal sacrifice			0	
Examined microscopically			36	
74.6 µg/gram Aloe-emodin				
Mice initially in study	36		36	
Natural death	1		2	
Moribund	2		1	
Harvest (skin lesion ≥5 mm)	0		33	
Terminal sacrifice	33		0	
Examined microscopically	36		36	

[a] One animal missexed
[b] One animal died prior to dosing and was discarded.
[c] One animal autolyzed, and no tissue was taken.

TABLE 5
Effect of SSL on Removal Time (Hazard Ratio Relative to No Light Control) by Test article

Treatment	Linear Trend[a]	SSL Exposure Level (mJ • CIE/cm^2)[b]		
		6.85	13.70	20.55
Male				
No Cream	10.39*	16.69*	138.48*	1,208.16*
Control Cream	—[c]	—	28.93*	—
Aloe Gel Cream	—	—	146.17*	—
Whole Leaf Cream	—	—	22.24*	—
Decolorized Whole Leaf Cream	—	—	28.67*	—
Aloe-emodin Cream	—	—	20.49*	—
Female				
No Cream	6.49*	1.99	20.21*	234.92*
Control Cream	—	—	18.16*	—
Aloe Gel Cream	—	—	31.43*	—
Whole Leaf Cream	—	—	50.50*	—
Decolorized Whole Leaf Cream	—	—	43.38*	—
Aloe-emodin Cream	—	—	58.19*	—

* Significant at 5%
[a] Linear trend hazard ratio represents the increase in hazard with a single experimental SSL level increment. This is 6.85 mJ • CIE/cm^2.
[b] Pairwise comparisons are relative to the no light group for the test article which was assigned a value of 1.0.
[c] Value of statistic cannot be computed (Linear Trend), or article was not tested at 6.85 or 20.55 mJ • CIE/cm^2 SSL.

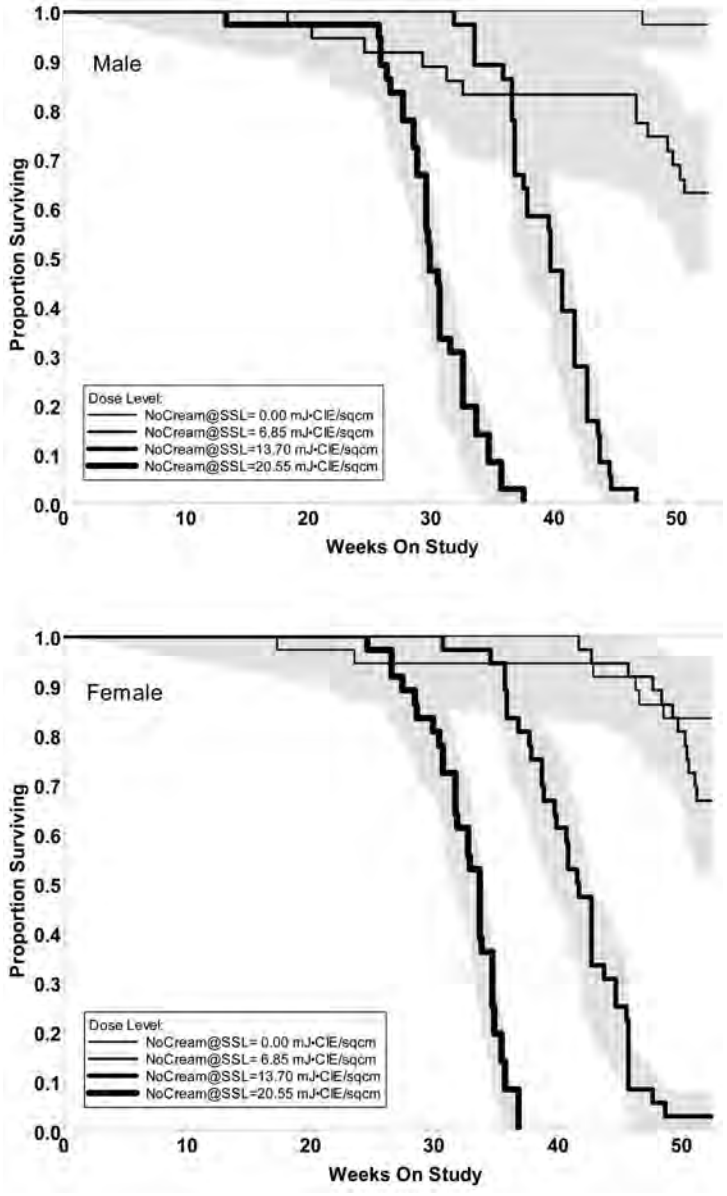


FIGURE 3
Kaplan-Meier Survival Curves for Mice Administered No Cream and Exposed to 0.00, 6.85, 13.70, or 20.55 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

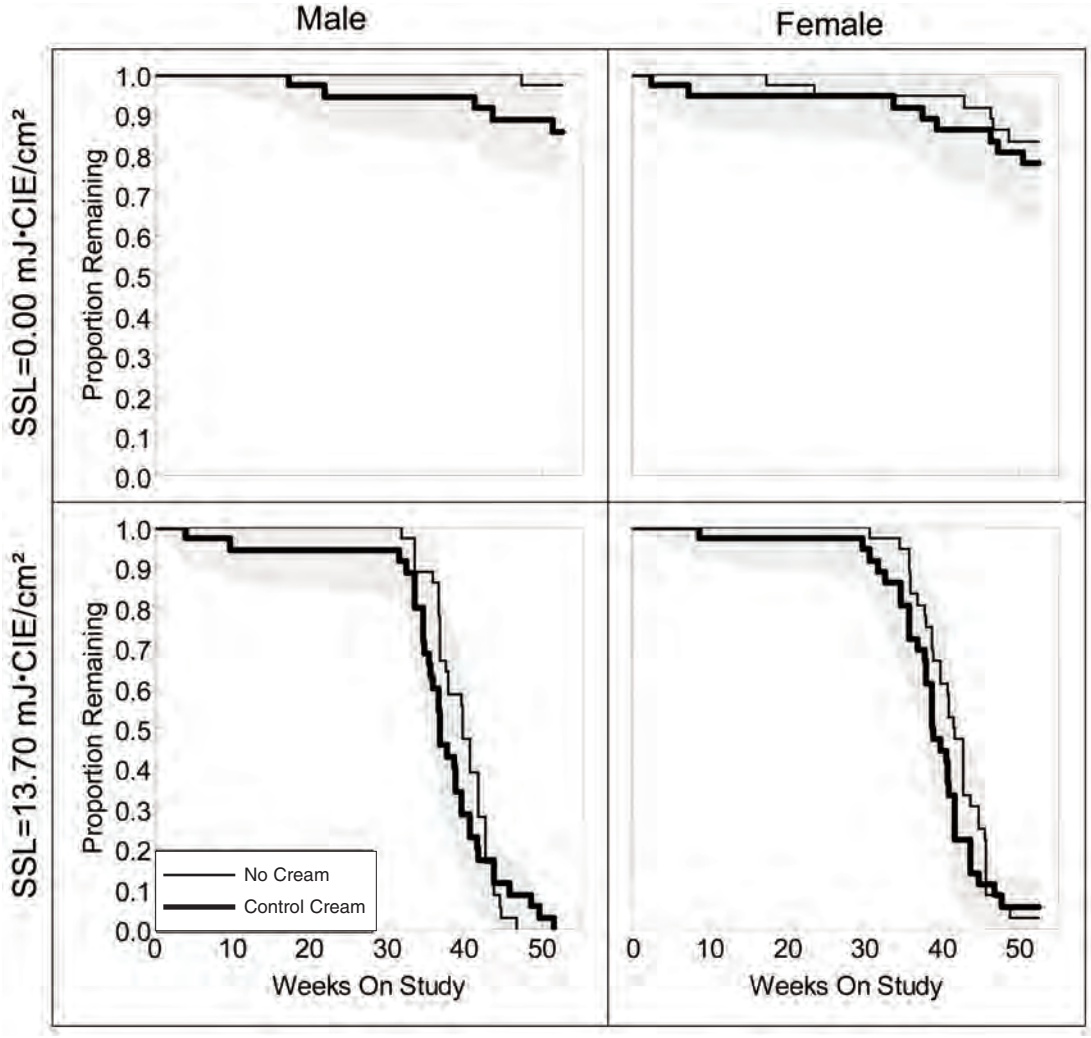


FIGURE 4
Kaplan-Meier Survival Curves for Mice Administered Control Cream or No Cream and Exposed to 0.00 or 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

TABLE 6
Effect of Test Article on Removal Time (Hazard Ratio Relative to Control Cream Group) by SSL

Treatment	Linear Trend[a]	Test Article Dose[b]	
		Low	High
Male			
0.00 mJ • CIE/cm^2 SSL			
Control Cream[c]	—[d]	—	5.62
Aloe Gel Cream	—	—	0.19
Whole Leaf Cream	—	—	1.15
Decolorized Whole Leaf Cream	—	—	1.19
Aloe-emodin Cream	—	—	1.67
13.70 mJ • CIE/cm^2 SSL			
Control Cream	—	—	1.17
Aloe Gel Cream	0.97	1.08	0.94
Whole Leaf Cream	0.94	0.80	0.88
Decolorized Whole Leaf Cream	1.08	1.17	1.18
Aloe-emodin Cream	1.09	0.98	1.18
Female			
0.00 mJ • CIE/cm^2 SSL			
Control Cream[b]	—	—	1.42
Aloe Gel Cream	—	—	0.68
Whole Leaf Cream	—	—	0.33
Decolorized Whole Leaf Cream	—	—	0.44
Aloe-emodin Cream	—	—	0.32*
13.70 mJ • CIE/cm^2 SSL			
Control Cream	—	—	1.27
Aloe Gel Cream	1.09	1.02	1.18
Whole Leaf Cream	0.96	0.70	0.93
Decolorized Whole Leaf Cream	1.03	1.39	1.06
Aloe-emodin Cream	1.01	1.50*	1.01

* Significant at 5%
[a] Linear trend hazard ratio represents the increase in hazard with a single experimental dose increment. For aloe-emodin, this is an approximately log(dose) step; for the other test articles, this is 3%.
[b] Low dose and high dose for all test articles except aloe-emodin = 3% and 6%, respectively; low dose and high dose for aloe-emodin = 7.46 and 74.6 µg/g, respectively.
[c] Control Cream is presented as the relative hazard to the No Cream control group at the same SSL exposure level.
[d] Value of statistic cannot be computed (Linear Trend), or article was not tested at 0.00 mJ • CIE/cm^2 SSL (Low Dose).

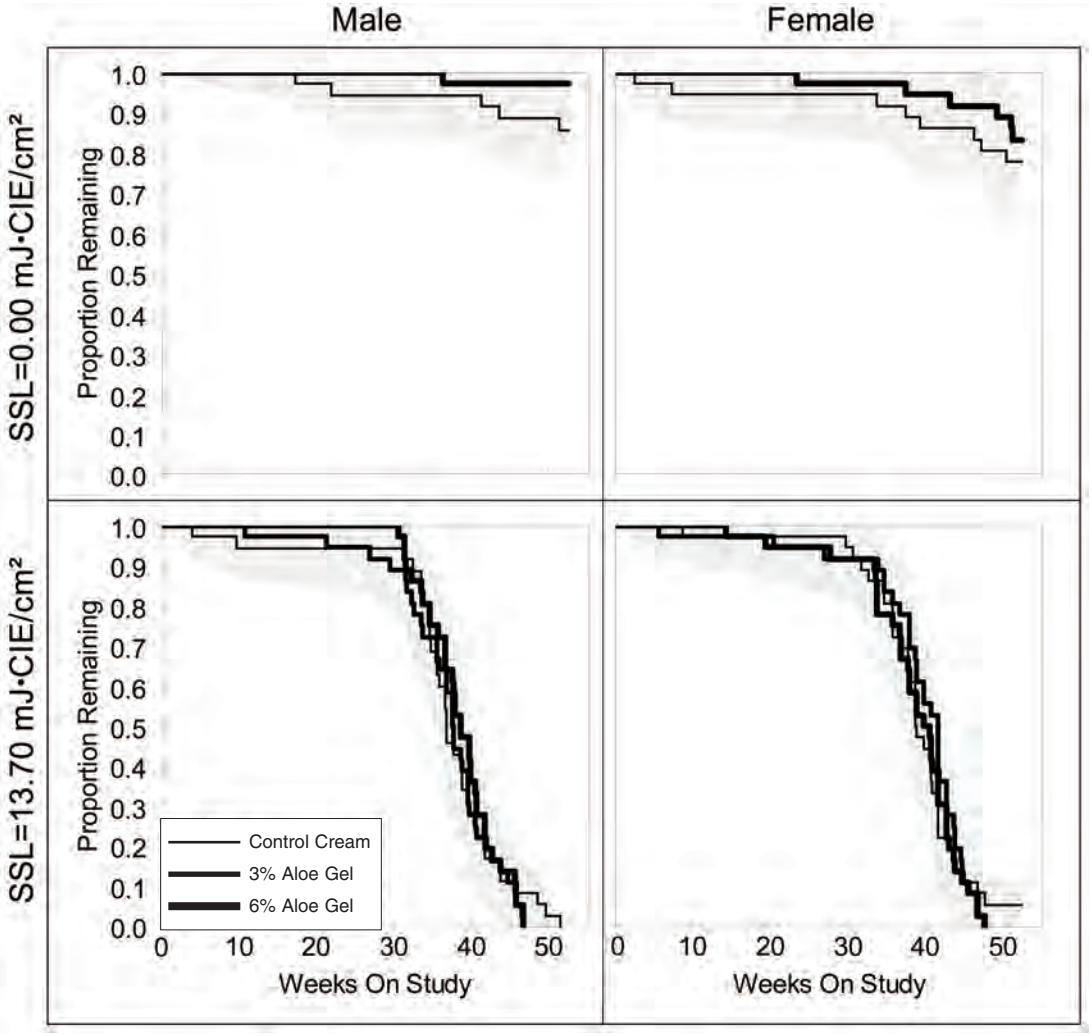


FIGURE 5
Kaplan-Meier Survival Curves for Mice Administered Aloe Gel Creams or Control Cream and Exposed to 0.00 or 13.70 mJ C CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

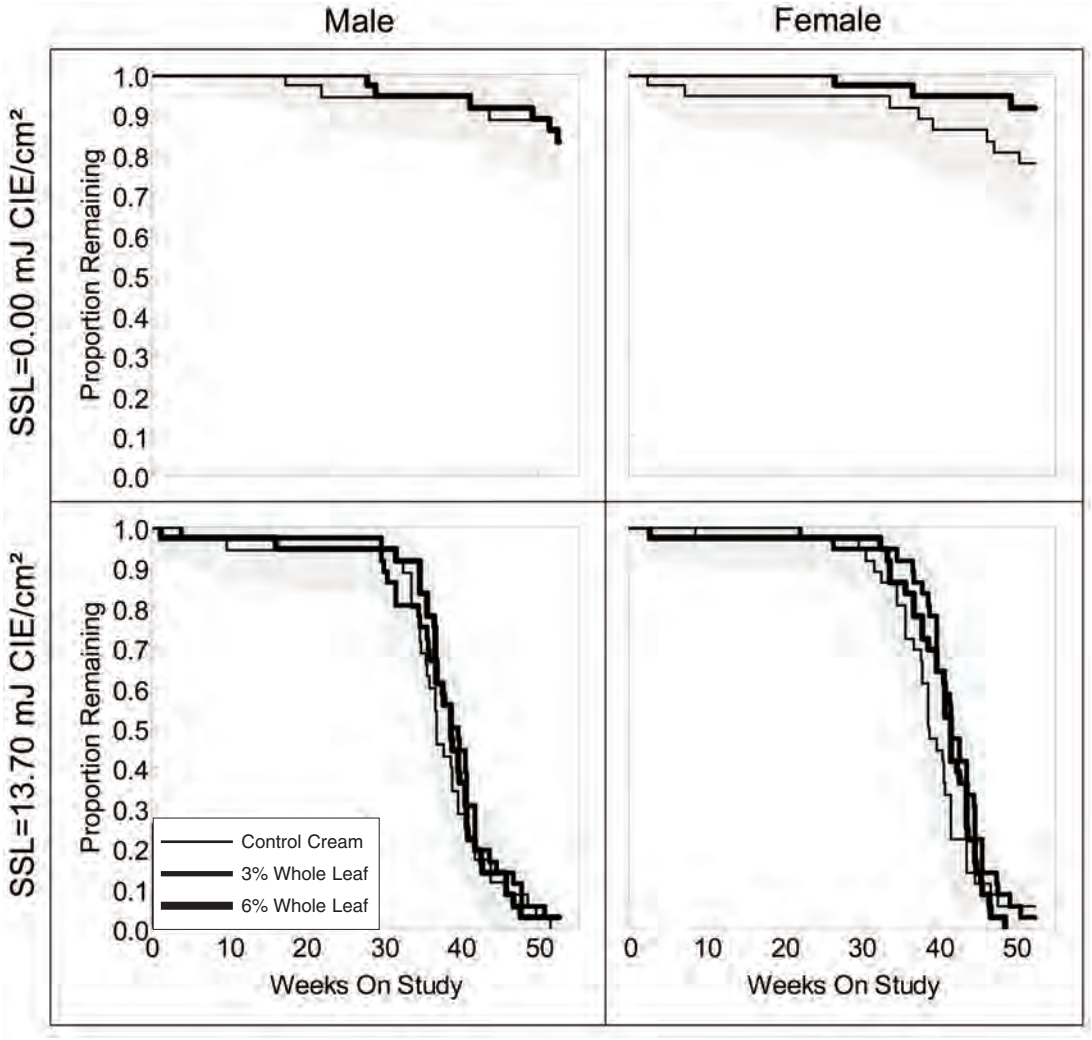


FIGURE 6
Kaplan-Meier Survival Curves for Mice Administered Whole Leaf Creams or Control Cream and Exposed to 0.00 or 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

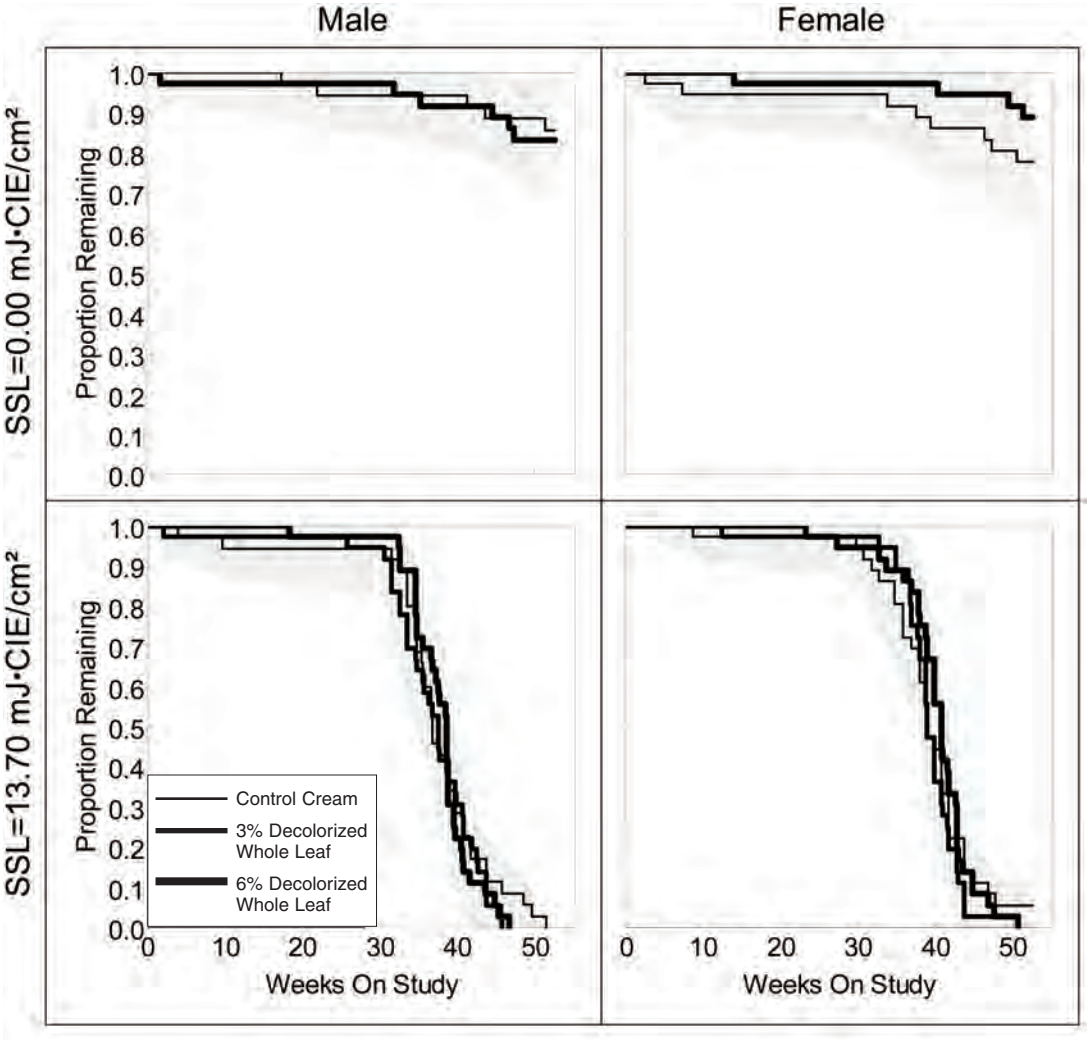


FIGURE 7
Kaplan-Meier Survival Curves for Mice Administered Decolorized Whole Leaf Creams or Control Cream and Exposed to 0.00 or 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

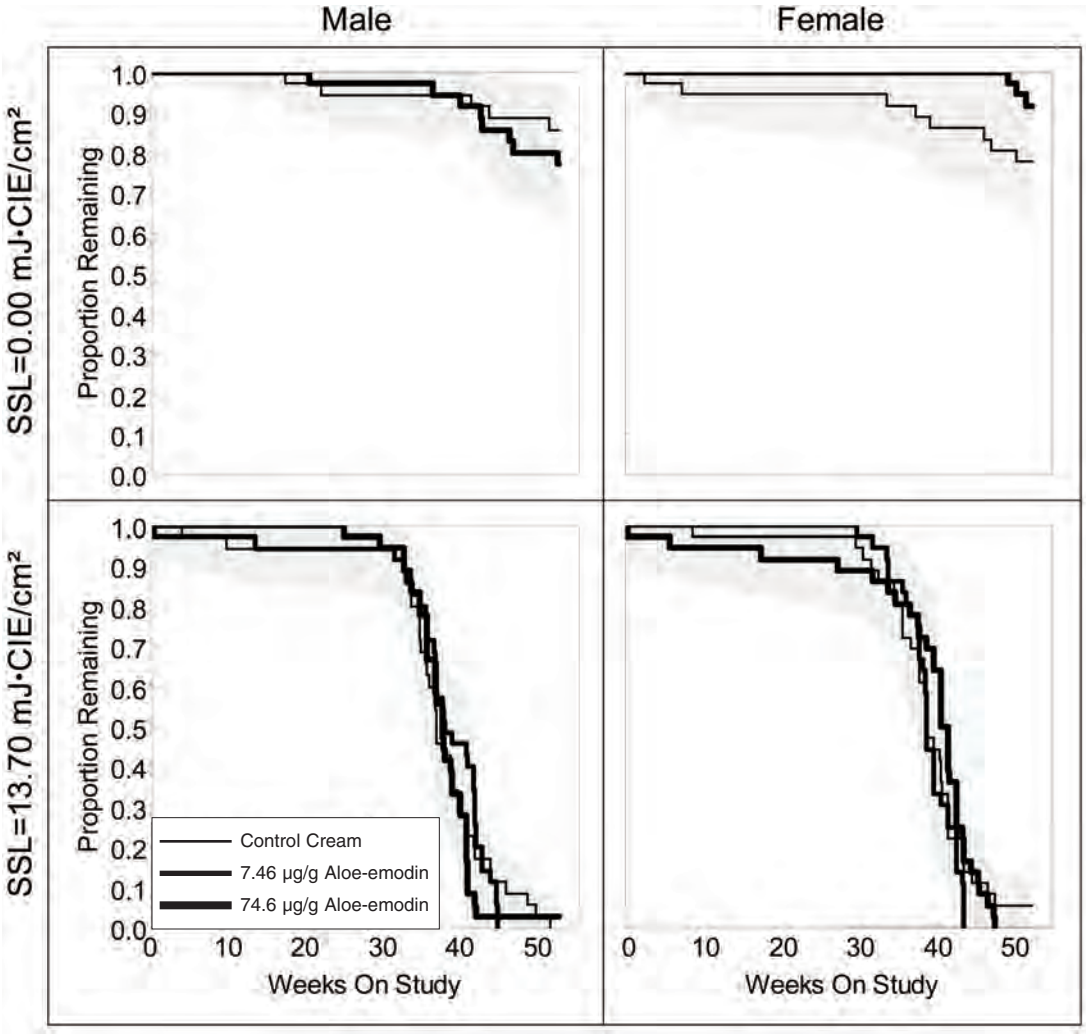


FIGURE 8
Kaplan-Meier Survival Curves for Mice Administered Aloe-emodin Creams or Control Cream and Exposed to 0.00 or 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

Body Weights

Body weight tables are presented in Appendix C.

No Cream

Tables C1 and C2 show mean body weights, percentage of mean body weights relative to controls (mice not exposed to SSL), survival, and statistical tests of dose trends and pairwise comparisons to controls for mice exposed to SSL in the absence of any cream treatment. Exposure to SSL at 6.85, 13.70, or 20.55 mJ • CIE/cm^2 had no significant dose-trend effects on the mean body weights of male or female mice that did not receive cream treatment during the 1-year study. In comparison tests with mice that were not exposed to SSL, significant differences in body weight means were observed in female mice only and only at the end of the study.

Control Cream

The results of the body weight analysis of mice that did not receive cream treatment compared with mice administered control cream are shown for 0.00 and 13.70 mJ • CIE/cm^2 SSL in Tables C3 and C4. In the absence of SSL, no significant differences were noted between the mean body weights of mice that did not receive cream treatment and the body weights of mice administered control cream during the 1-year study, with the exception of male mice at week 52. No significant differences were noted between the no cream and control cream males exposed to 13.70 mJ • CIE/cm^2 SSL at any time point.

Aloe Gel, Whole Leaf, Decolorized Whole Leaf, and Aloe-emodin Creams

With few exceptions, the administration of aloe gel (Tables C5 and C6), whole leaf (Tables C7 and C8), decolorized whole leaf (Tables C9 and C10), or aloe-emodin (Tables C11 and C12) creams to male and female mice had no significant effect on mean body weights when compared with the body weights of mice at the same time point that were administered the control cream and exposed to the same level of SSL.

In-Life Skin Lesion Onset

The time to skin lesion onset was computed for each animal and used to generate Kaplan-Meier curves. The median time for skin lesion onset (the time point in the study when 50% of the animals in a treatment group had at least one measurable skin lesion) was calculated, and probability values were determined based on the median time to skin lesion onset values for each treatment group. The reference cell for contrasts, other than the no cream group, was the control cream group. The probability values presented are two sided.

No Cream

The Kaplan-Meier in-life curves and Cox contrasts for skin lesion onset in male and female mice that did not receive cream treatment but were exposed to SSL are shown in Figure 9. Significant SSL exposure-related trends in the onset of skin lesions were observed in male and female mice. In pairwise comparison tests with control mice that were not exposed to SSL, the median weeks of skin lesion onset significantly decreased as SSL exposure levels increased.

Control Cream

The Kaplan-Meier in-life curves and Cox contrasts for skin lesion onset in male and female mice that were exposed to 13.70 mJ • CIE/cm^2 SSL and received either no cream or the control cream are shown in Figure 10. No significant differences were observed in the median weeks of skin lesion onset in pairwise comparison tests between the control cream and no cream groups.

Aloe Gel, Whole Leaf, Decolorized Whole Leaf, and Aloe-emodin Creams

The Kaplan-Meier in-life curves and Cox contrasts for skin lesion onset in male and female mice administered control cream or aloe gel, whole leaf, decolorized whole leaf, or aloe-emodin creams and exposed to 13.70 mJ • CIE/cm^2 SSL are shown in Figures 11 through 14. Linear trend tests, represented by P values for the 6% aloe gel, 6% whole leaf, 6% decolorized whole leaf, or 74.6 µg/g aloe-emodin groups, showed no significant dose-related differences in the median weeks of skin lesion onset in male or female mice. In pairwise comparison tests with the control cream groups, the median weeks of skin lesion onset were similar among male and female mice administered 3% or 6% aloe gel,whole leaf, or decolorized whole leaf creams or 7.4 or 74.6 µg/g aloe-emodin.

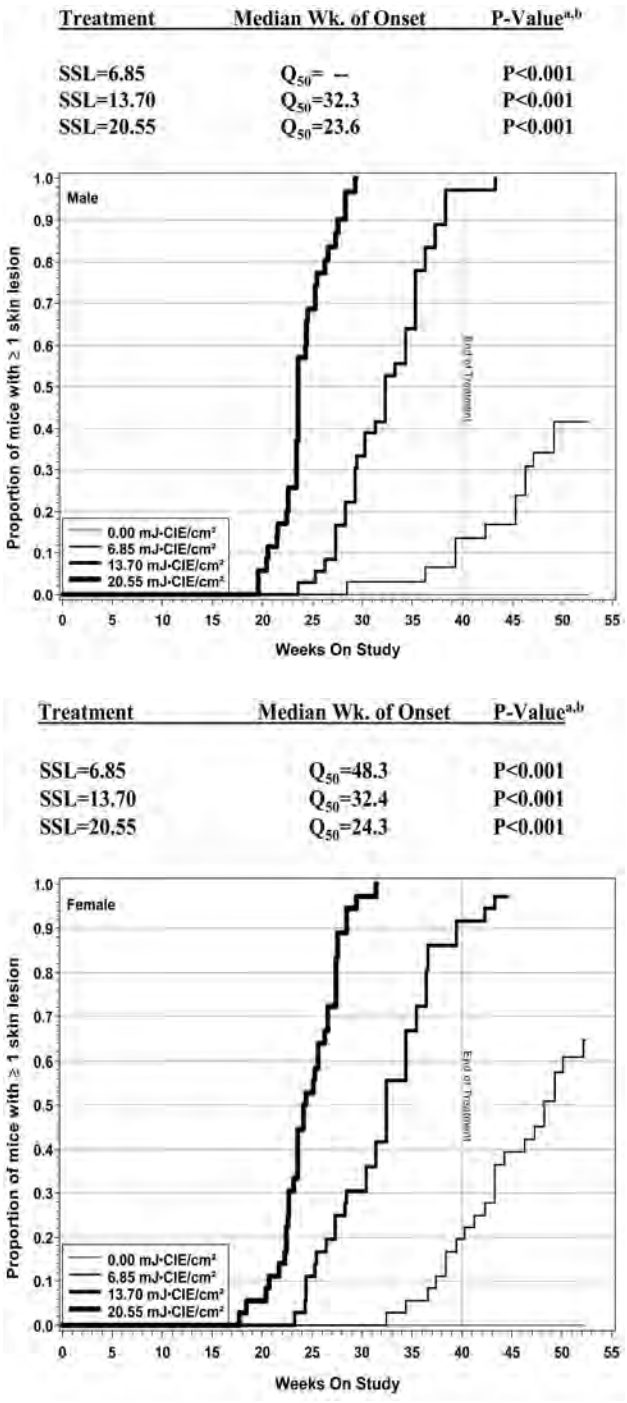


FIGURE 9
Kaplan-Meier Curves and Cox Contrasts for In-life Skin Lesion Onset in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

[a] P values for 6.85 and 13.70 mJ • CIE/cm^2 SSL exposures represent pairwise comparisons to the 0.00 mJ • CIE/cm^2 SSL control.

[b] The P value for the 20.55 mJ • CIE/cm^2 SSL exposure represents pairwise comparison to the 0.00 mJ • CIE/cm^2 SSL control and the results of a linear trend test.

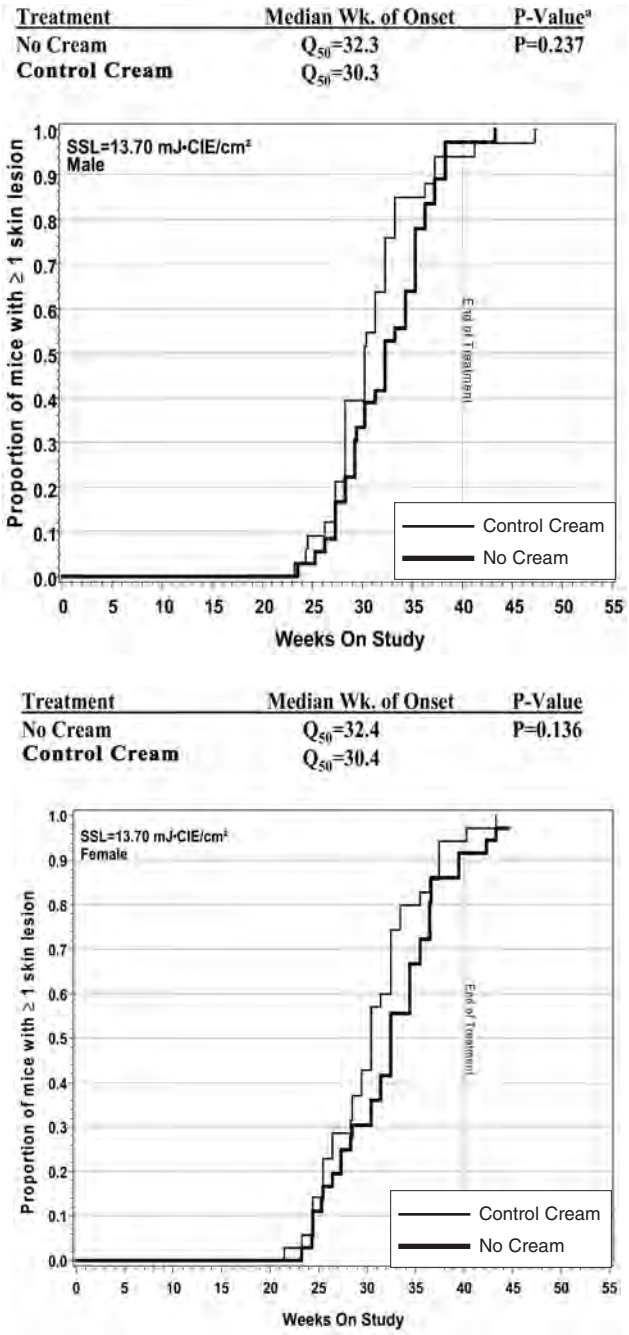
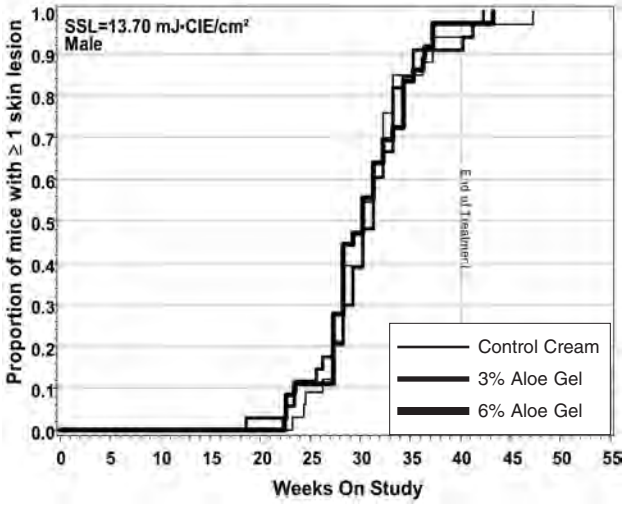


FIGURE 10
Kaplan-Meier Curves and Cox Contrasts for In-life Skin Lesion Onset in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and No Cream

[a] P values represent pairwise comparisons to the control cream groups.

Treatment	Median Wk. of Onset	P-Value[a,b]
Control Cream	Q_{50}=30.3	
3% Aloe Gel	Q_{50}=31.3	P=0.993
6% Aloe Gel	Q_{50}=30.5	P=0.835



Treatment	Median Wk. of Onset	P-Value[a,b]
Control Cream	Q_{50}=30.4	
3% Aloe Gel	Q_{50}=33.4	P=0.239
6% Aloe Gel	Q_{50}=30.9	P=0.412

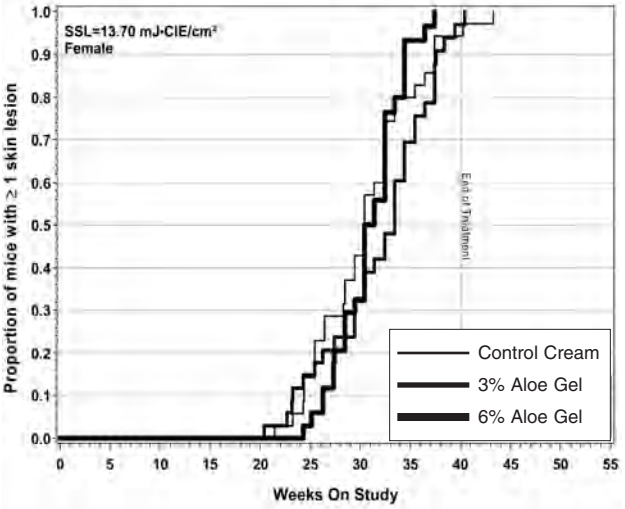


FIGURE 11
Kaplan-Meier Curves and Cox Contrasts for In-life Skin Lesion Onset in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Aloe Gel Creams and Control Cream

[a] P values for the 3% aloe gel cream groups represent pairwise comparisons to the control cream groups.

[b] P values for the 6% aloe gel cream groups represent pairwise comparisons to the control cream groups and the results of linear trend tests.

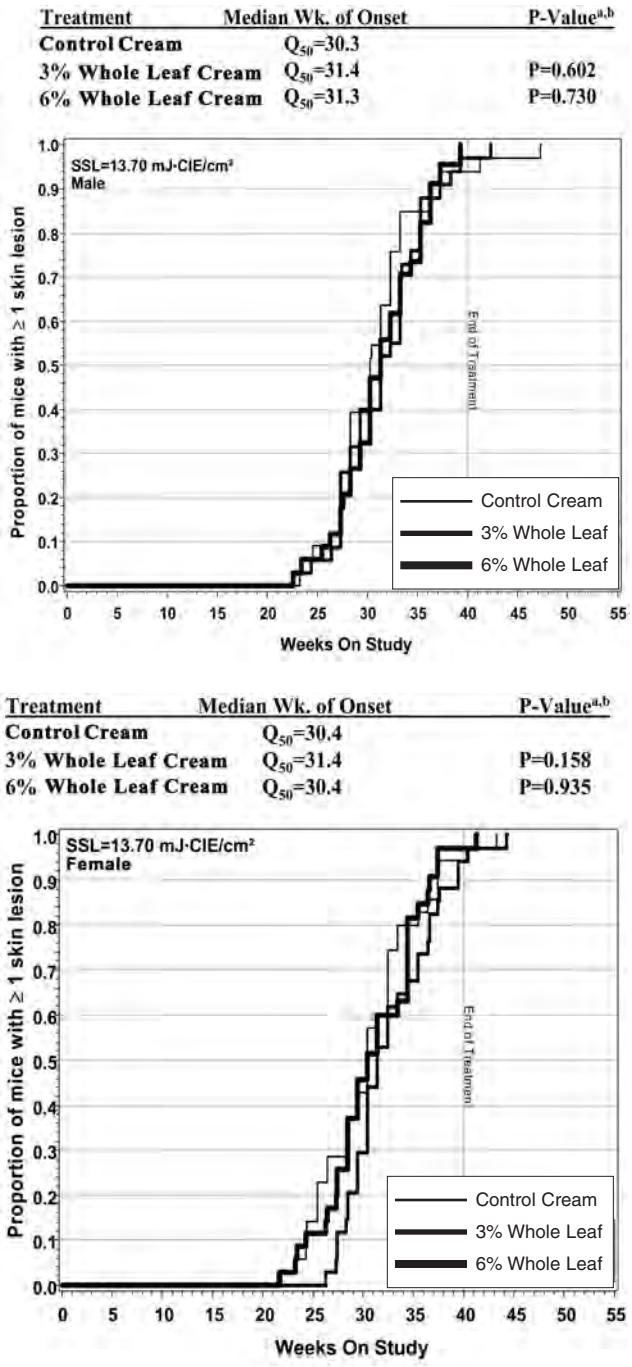


FIGURE 12
Kaplan-Meier Curves and Cox Contrasts for In-life Skin Lesion Onset in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Whole Leaf Creams and Control Cream

[a] P values for the 3% whole leaf cream groups represent pairwise comparisons to the control cream groups.

[b] P values for the 6% whole leaf cream groups represent pairwise comparisons to the control cream groups and the results of linear trend tests.

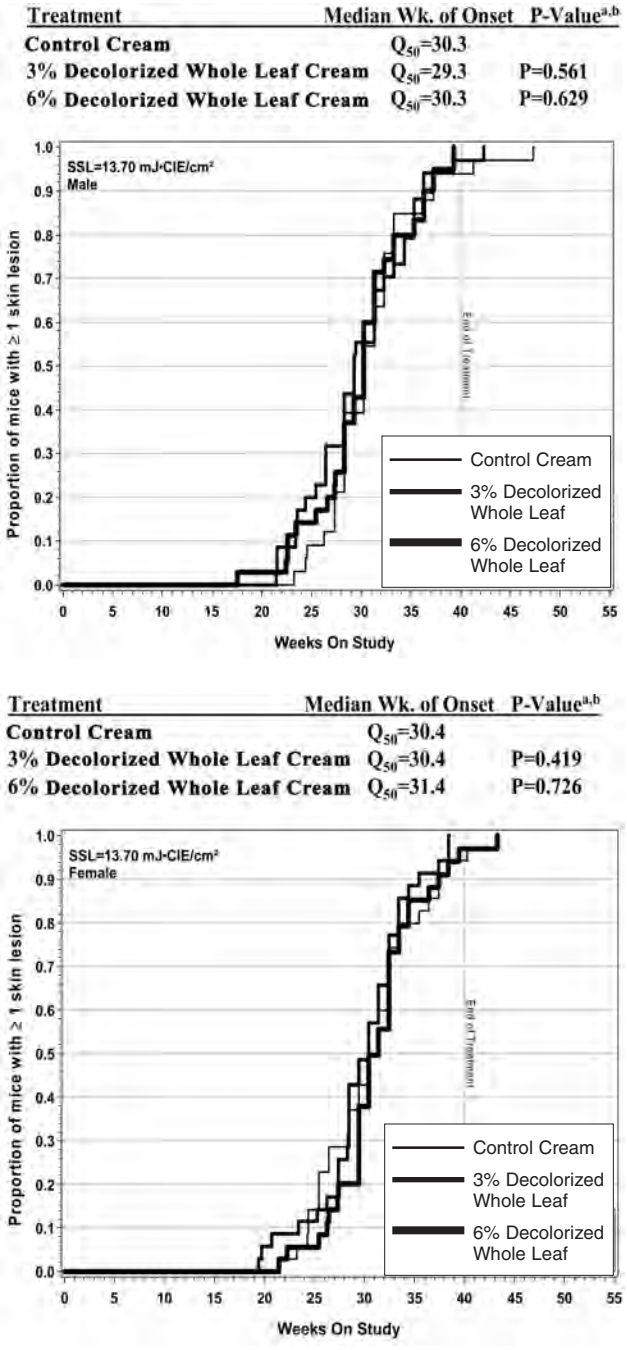


FIGURE 13
Kaplan-Meier Curves and Cox Contrasts for In-life Skin Lesion Onset in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Decolorized Whole Leaf Creams and Control Cream

[a] P values for the 3% decolorized whole leaf cream groups represent pairwise comparisons to the control cream groups.

[b] P values for the 6% decolorized whole leaf cream groups represent pairwise comparisons to the control cream groups and the results of linear trend tests.

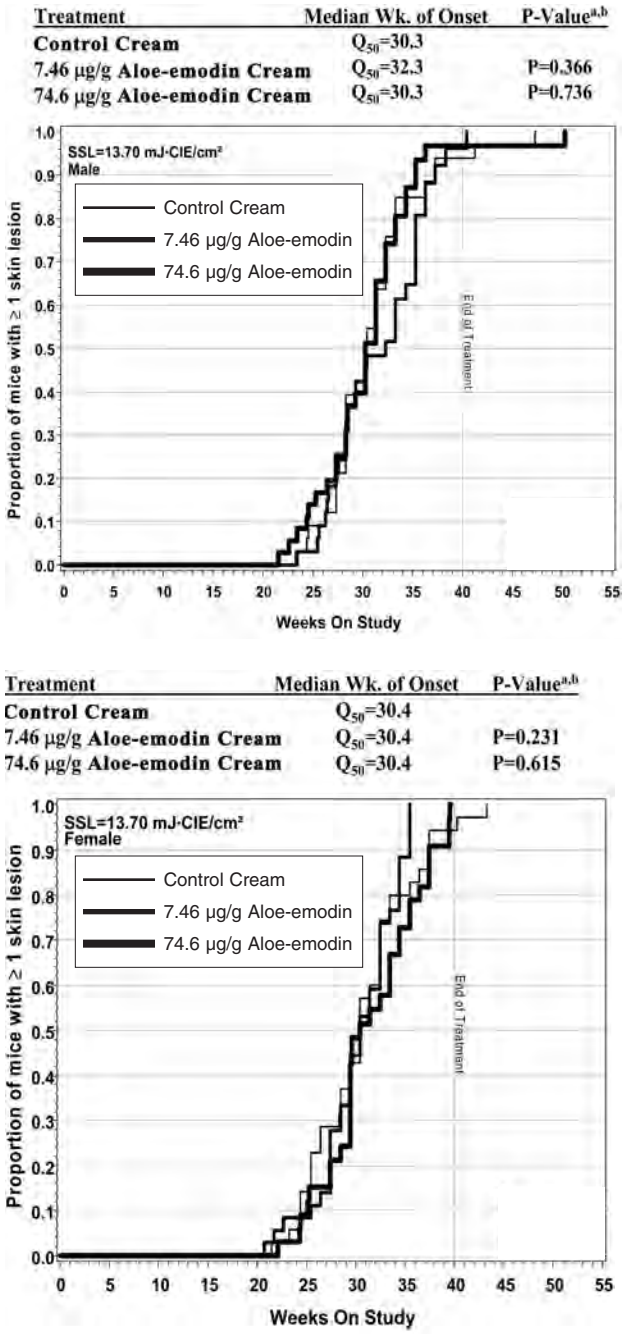


FIGURE 14
Kaplan-Meier Curves and Cox Contrasts for In-life Skin Lesion Onset in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Aloe-emodin Creams and Control Cream

[a] P values for the 7.46 µg/g aloe-emodin cream groups represent pairwise comparisons to the control cream groups.

[b] P values for the 74.6 µg/g aloe-emodin cream groups represent pairwise comparisons to the control cream groups and the results of linear trend tests.

In-Life Skin Lesion Incidence and Multiplicity

No Cream

The Poly-3 probability values for the age-adjusted incidence rates and the Poisson probability values for multiplicities of skin lesions determined in-life in mice that did not receive cream treatment but were exposed to SSL are shown in Table 7. The control group for analysis was the group of mice that was not exposed to SSL. The effects of increasing exposure levels of SSL on mouse skin lesion incidence and multiplicity were quite remarkable, and significant linear dose trends for the SSL effects were observed in both male and female mice. In pairwise comparison tests that examined the multiplicities of skin lesions in control mice that were not exposed to SSL, skin lesion incidence rates were significantly increased in all exposed groups of male and female mice. Similarly, significant Poisson P values were observed in pairwise comparison tests of the multiplicities of skin lesions in control mice (0.00 mJ • CIE/cm^2 SSL) with those in male mice exposed to 13.70 or 20.55 mJ • CIE/cm^2 of SSL and those in female mice exposed to 6.85, 13.70, or 20.55 mJ • CIE/cm^2 SSL.

Control Cream

The comparisons of the effects of no cream treatment with the control cream on the incidence and multiplicities of in-life determined skin lesions in mice are shown in Table 8. The age-adjusted incidence rates of male and female mice administered either no cream or control cream and exposed to 13.70 mJ • CIE/cm^2 SSL approached 100% among both treatments and sex groups, and the P values of 1.00 for the Poly-3 tests reflected that the incidence rates did not differ between the treatment groups. Similarly, there were no significant differences in the Poisson tests of skin lesion multiplicities in control cream or no cream male or female mice exposed to 13.70 mJ • CIE/cm^2 SSL.

Table 7
Effects of SSL Exposure on the Incidences and Multiplicities of Skin Lesions Detected In-life at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream[a]

	0.00 mJ • CIE/cm^2 SSL	6.85 mJ • CIE/cm^2 SSL	13.70 mJ • CIE/cm^2 SSL	20.55 mJ • CIE/cm^2 SSL
Male				
Adjusted incidence rate	0/35.7 (0.0%)	12/29.3 (40.9%)	36/36.0 (100.0%)	34/34.1 (99.6%)
First incidence (weeks)	—[b]	28	23	19
Poly-3 test (incidence)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE	0.0 ± 0.0	1.0 ± 0.2	6.4 ± 0.5	8.8 ± 0.7
Relative treatment effects regression	P = 0.006*	P = 0.062	P = 0.014*	P = 0.011*
Female				
Adjusted incidence rate	0/32.8 (0.0%)	22/34.9 (63.0%)	35/35.6 (98.3%)	36/36.0 (100.0%)
First incidence (weeks)	—	32	23	17
Poly-3 test (incidence)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE	0.0 ± 0.0	1.7 ± 0.2	5.6 ± 0.4	9.1 ± 0.6
Relative treatment effects regression	P = 0.007*	P = 0.044*	P = 0.016*	P = 0.011*

* Significant at P≤0.05

[a] P values for the 0.00 mJ • CIE/cm^2 SSL groups represent linear trend test results; otherwise, P values represent pairwise comparisons to the control group (0.00 mJ • CIE/cm^2). The additive standard error of the mean is an approximation; the true error structure is multiplicative, rather than additive. All P values are two sided. SE = standard error.

[b] Not applicable; no lesions in the animal group

TABLE 8
Effects of No Cream and Control Cream on the Incidences and Multiplicities of Skin Lesions Detected In-life at the Site of Application in Mice Exposed to 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera[a]

	No Cream	Control Cream
Male		
Adjusted incidence rate	36/36.0 (100.0%)	33/33.0 (100.0%)
First incidence (weeks)	23	23
Poly-3 test (incidence)		P = 1.00
Overall mean lesion multiplicity ± SE	6.4 ± 0.5	6.5 ± 0.5
Poisson regression (multiplicity)		P = 0.789
Female		
Adjusted incidence rate	35/35.6 (98.3%)	35/35.0 (100.0%)
First incidence (weeks)	23	21
Poly-3 test (incidence)		P = 1.00
Overall mean lesion multiplicity ± SE	5.6 ± 0.4	6.1 ± 0.5
Poisson regression (multiplicity)		P = 0.454

[a] The additive standard error of the mean is an approximation; the true error structure is multiplicative, rather than additive. All P values are two sided. SE = standard error.

Aloe Gel, Whole Leaf, Decolorized Whole Leaf, and Aloe-emodin Creams

The effects of aloe gel, whole leaf, decolorized whole leaf, and aloe-emodin creams on the incidences and multiplicities of in-life determined skin lesions in mice are shown in Tables 9 through 12. In male and female mice exposed to 13.70 mJ • CIE/cm^2 SSL, there were no significant dose-related trend differences in skin lesion incidences or multiplicities due to any of the treatments. Additionally, in pairwise comparison tests of skin lesion incidences or multiplicities, there were no significant differences between mice administered control cream and mice administered aloe gel, whole leaf, decolorized whole leaf, or aloe-emodin creams.

Nonneoplastic Skin Lesions

This section describes the biologically noteworthy histopathology changes in nonneoplastic lesions that occurred in the study. The skin at the site of application of the creams (coincident with the site of SSL exposure) was the target tissue of the study. Summaries of the incidences of nonneoplastic lesions are presented in Appendix A for male mice and Appendix B for female mice.

The skin of the SKH-1 hairless mouse examined in this study had a number of unique characteristics. The epidermis in the untreated SKH-1 mouse usually consisted of one or two layers of squamous cells (Plate 1A). Skin hair shafts and adnexal structures, such as sebaceous glands, were either absent or were atypical in location and development. There were often prominent cystic structures in the dermis that appeared to be remnants of hair follicles. The cysts were usually lined by squamous or low cuboidal epithelium and were either empty or contained small amounts of keratinized debris and occasionally, fragmented hair shafts.

The exposure of SKH-1 mice to SSL and/or aloe creams was associated with diffuse thickening of the epidermis. This change was documented using the term "squamous hyperplasia" (Plate 1B). The severity of squamous hyperplasia was graded and scored according to the number of epithelial cell layers in the epidermis: two to four cell layers was graded as minimal hyperplasia and received a score of 1; four to six cell layers was graded as mild hyperplasia and received a score of 2; six to nine cell layers was graded as moderate and received a score of 3; and more than nine cell layers was graded as marked and received a score of 4.

The exposure of SKH-1 mice to SSL and/or aloe creams was also associated with focal nodular thickening of the epidermis; the term "focal atypical squamous hyperplasia" was used to describe this condition (Plate 1C). Focal atypical squamous hyperplasia was differentiated from squamous hyperplasia by its nodular characteristic, which was usually detected by gross examination. The squamous cells that composed the nodule of focal atypical squamous hyperplasia resembled normal epidermis; however, dysplastic changes, including the lack of cohesion or orientation, pleomorphism, nuclear atypia, and basal disorganization, occurred. In addition, increased numbers of normal and abnormal mitotic figures, inflammatory changes, and increased amounts of keratin were often present over the superficial layers of the hyperplastic epidermis. The incidence and multiplicity of focal atypical squamous hyperplasia was documented according to the number of nodules detected in the epidermis.

No Cream

The incidences, severity scores, and multiplicities of nonneoplastic skin lesions at the site of application in male and female mice that were not administered cream and were exposed to SSL are given in Table 13. Significant SSL exposure-related trends in the incidences of squamous hyperplasia were observed in both male and female mice that were not administered cream, and the average severity scores for squamous hyperplasia increased as the exposure levels of SSL increased. In pairwise comparison tests with control mice not exposed to SSL, the incidences of squamous hyperplasia were significantly increased in all exposed groups.

Significant SSL exposure-related trends occurred in the incidences of focal atypical squamous hyperplasia in male and female mice. In pairwise comparison tests with mice that were not administered cream and not exposed to SSL, significantly increased incidences of focal atypical squamous hyperplasia were observed at each SSL exposure level. No foci of atypical squamous hyperplasia were detected in male or female mice not exposed to SSL. The multiplicities of focal atypical squamous hyperplasia showed significant SSL exposure-related trends in male and female mice. At each SSL exposure

TABLE 9
Effects of Control Cream and Aloe Gel Creams on the Incidences and Multiplicities of Skin Lesions Detected In-life at the Site of Application in Mice Exposed to 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera[a]

	Control Cream	3% Aloe Gel	6% Aloe Gel
Male			
Adjusted incidence rate	33/33.0 (100.0%)	33/33.2 (99.4%)	36/36.0 (100.0%)
First incidence (weeks)	24	20	23
Poly-3 test (incidence)	P = 1.00	P = 1.00	P = 1.00
Overall mean lesion multiplicity ± SE	6.5 ± 0.5	5.6 ± 0.5	6.1 ± 0.5
Poisson regression (multiplicity)	P = 0.493	P = 0.191	P = 0.493
Female			
Adjusted incidence rate	35/35.0 (100.0%)	33/33.2 (99.4%)	33/33.3 (99.0%)
First incidence (weeks)	21	20	24
Poly-3 test (incidence)	P = 1.00	P = 1.00	P = 1.00
Overall mean lesion multiplicity ± SE	6.1 ± 0.5	5.3 ± 0.4	5.9 ± 0.5
Poisson regression (multiplicity)	P = 0.768	P = 0.227	P = 0.768

[a] P values for the control cream groups represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to the control. All P values are two sided. The additive standard error of the mean is an approximation; the true error structure is multiplicative, rather than additive. SE = standard error.

TABLE 10
Effects of Control Cream and Whole Leaf Creams on the Incidences and Multiplicities of Skin Lesions Detected In-life at the Site of Application in Mice Exposed to 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera[a]

	Control Cream	3% Whole Leaf	6% Whole Leaf
Male			
Adjusted incidence rate	33/33.0 (100.0%)	34/34.2 (99.5%)	33/33.4 (98.9%)
First incidence (weeks)	23	22	22
Poly-3 test (incidence)	P = 1.00	P = 1.00	P = 1.00
Overall mean lesion multiplicity ± SE	6.5 ± 0.5	5.6 ± 0.5	7.7 ± 0.5
Poisson regression (multiplicity)	P = 0.127	P = 0.157	P = 0.127
Female			
Adjusted incidence rate	35/35.0 (100.0%)	34/34.2 (99.4%)	34/34.3 (99.2%)
First incidence (weeks)	21	26	21
Poly-3 test (incidence)	P = 1.00	P = 1.00	P = 1.00
Overall mean lesion multiplicity ± SE	6.1 ± 0.5	6.7 ± 0.5	6.8 ± 0.5
Poisson regression (multiplicity)	P = 0.324	P = 0.377	P = 0.324

[a] P values for the control cream groups represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to the control. All P values are two sided. The additive standard error of the mean is an approximation; the true error structure is multiplicative, rather than additive. SE = standard error.

TABLE 11
Effects of Control Cream and Decolorized Whole Leaf Creams on the Incidences and Multiplicities of Skin Lesions Detected In-life at the Site of Application in Mice Exposed to 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera[a]

	Control Cream	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf
Male			
Adjusted incidence rate	33/33.0 (100.0%)	34/34.1 (99.7%)	33/33.7 (98.0%)
First incidence (weeks)	23	21	17
Poly-3 test (incidence)	P = 0.663	P = 1.00	P = 1.00
Overall mean lesion multiplicity ± SE	6.5 ± 0.5	7.6 ± 0.6	6.4 ± 0.5
Poisson regression (multiplicity)	P = 0.799	P = 0.168	P = 0.799
Female			
Adjusted incidence rate	35/35.0 (100.0%)	35/35.0 (100.0%)	34/34.2 (99.3%)
First incidence (weeks)	21	19	21
Poly-3 test (incidence)	P = 1.00	P = 1.00	P = 1.00
Overall mean lesion multiplicity ± SE	6.1 ± 0.5	6.8 ± 0.5	6.7 ± 0.5
Poisson regression (multiplicity)	P = 0.447	P = 0.314	P = 0.447

[a] P values for the control cream groups represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to the control. All P values are two sided. The additive standard error of the mean is an approximation; the true error structure is multiplicative,

TABLE 12
Effects of Control Cream and Aloe-emodin Creams on the Incidences and Multiplicities of Skin Lesions Detected In-life at the Site of Application in Mice Exposed to 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera[a]

	Control Cream	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Male			
Adjusted incidence rate	33/33.0 (100.0%)	31/31.6 (98.3%)	34/34.3 (99.0%)
First incidence (weeks)	24	24	22
Poly-3 test (incidence)	P = 1.00	P = 1.00	P = 1.00
Overall mean lesion multiplicity ± SE	6.5 ± 0.5	7.3 ± 0.5	7.4 ± 0.5
Poisson regression (multiplicity)	P = 0.246	P = 0.302	P = 0.246
Female			
Adjusted incidence rate	35/35.0 (100.0%)	35/35.2 (99.5%)	33/33.0 (99.9%)
First incidence (weeks)	21	20	22
Poly-3 test (incidence)	P = 1.00	P = 1.00	P = 1.00
Overall mean lesion multiplicity ± SE	6.1 ± 0.5	6.4 ± 0.5	6.6 ± 0.5
Poisson regression (multiplicity)	P = 0.526	P = 0.700	P = 0.526

[a] P values for the control cream groups represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to the control. All P values are two sided. The additive standard error of the mean is an approximation; the true error structure is multiplicative, rather than additive. SE = standard error.

TABLE 13
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

	0.00 mJ • CIE/cm^2 SSL	6.85 mJ • CIE/cm^2 SSL	13.70 mJ • CIE/cm^2 SSL	20.55 mJ • CIE/cm^2 SSL
Male				
Squamous Hyperplasia				
Overall rate[a]	1/36 (2.8%)	20/36 (55.6%)	35/36 (97.2%)	28/36 (77.8%)
Poly-3 test (incidence)[b]	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Average severity[c]	1.0	1.3	1.7	1.8
Focal Atypical Squamous Hyperplasia				
Overall rate	0/36 (0.0%)	12/36 (33.3%)	33/36 (91.7%)	32/36 (88.9%)
Poly-3 test (incidence)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.0 ± 0.0 (0.0-0.0)	0.7 ± 0.2 (0.3-1.0)	3.3 ± 0.4 (2.6-3.9)	2.9 ± 0.4 (2.2-3.6)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.0 ± 0.0 (0.0-0.0)	0.7 ± 0.2 (0.3-1.0)	4.2 ± 0.5 (3.4-5.1)	4.9 ± 0.7 (3.8-6.1)
Relative treatment effect regression (multiplicity)[b]	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Female				
Squamous Hyperplasia				
Overall rate	1/36 (2.8%)	32/36 (88.9%)	36/36 (100.0%)	34/36 (94.4%)
Poly-3 test (incidence)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Average severity	1.0	1.3	1.8	1.9
Focal Atypical Squamous Hyperplasia				
Overall rate	0/36 (0.0%)	15/36 (41.7%)	32/36 (88.9%)	34/36 (94.4%)
Poly-3 test (incidence)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.0 ± 0.0 (0.0-0.0)	0.8 ± 0.2 (0.4-1.2)	2.8 ± 0.4 (2.2-3.5)	3.8 ± 0.4 (3.0-4.5)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.0 ± 0.0 (0.0-0.0)	0.8 ± 0.3 (0.4-1.3)	3.4 ± 0.4 (2.6-4.1)	6.2 ± 0.8 (4.8-7.6)
Relative treatment effect regression (multiplicity)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*

* Significant at P≤0.05
SE = standard error; C.I. = confidence interval
[a] Number of animals with specified nonneoplastic skin lesion/number of animals with skin examined microscopically
[b] P values for control group (0.00 mJ • CIE/cm^2) represent linear trend tests; otherwise, P values represent pairwise comparisons to control group.
[c] Average severity grade of lesions in affected animals: 1 = minimal, 2 = mild, 3 = moderate, 4 = marked

level, the multiplicity of foci of atypical squamous hyperplasia was significantly greater than that of the control groups.

Control Cream

The incidences, severity scores, and multiplicities of nonneoplastic skin lesions at the site of application in mice administered control cream and exposed to SSL at 0.00 or 13.70 mJ • CIE/cm^2 SSL are compared in Table 14 with same-sex animals at the same SSL exposure level that did not receive cream. In the absence of SSL exposure, there was a mild, albeit significant, increase in the incidence of squamous hyperplasia in female mice administered control cream when compared with mice that did not receive cream. At 13.70 mJ • CIE/cm^2 SSL, there were no significant differences in the incidences or severity scores of squamous hyperplasia in no cream or control cream male or female mice.

There were no foci of atypical squamous hyperplasia detected in male or female mice not exposed to SSL, and the incidences and multiplicities of focal atypical squamous hyperplasia did not differ between male or female mice that received either no cream treatment or the control cream and exposure to 13.70 mJ • CIE/cm^2 SSL.

Aloe Gel Creams

The effects of aloe gel creams on the incidences, severity scores, and multiplicities of nonneoplastic skin lesions at the site of application in male and female mice exposed to 0.00 or 13.70 mJ • CIE/cm^2 SSL are shown in Table 15. In comparison tests with the control cream groups in the absence or presence of SSL exposure, the severity scores of squamous hyperplasia were not affected by the application of the aloe gel creams in male or female mice, and there were no significant dose-related trend effects in the Poly-3 analyses of the incidences of squamous cell hyperplasia.

Focal atypical squamous hyperplasia was not observed in male or female mice treated with control cream or 6% aloe gel cream in the absence of SSL. At 13.70 mJ • CIE/cm^2 SSL, no dose-related trend effects were found in the incidences of focal atypical squamous hyperplasia, although pairwise comparison tests with the control cream group indicated a significantly decreased incidence of focal atypical squamous hyperplasia in female mice administered 3% aloe gel cream. A significantly negative Poisson dose-related trend and significantly decreased multiplicities of focal atypical squamous hyperplasia were detected in both dosed groups of female mice exposed to 13.70 mJ • CIE/cm^2 SSL. No significant dose-related trends or differences in the multiplicities of focal atypical squamous hyperplasia were observed in male mice.

Whole Leaf Creams

The incidences, severity scores, and multiplicities of nonneoplastic skin lesions at the site of application in mice administered whole leaf creams are shown in Table 16. The incidence of squamous hyperplasia was unaffected by treatment in the absence of SSL in male mice. The incidence of squamous hyperplasia in female mice was significantly decreased in a pairwise comparison test with the control cream group in the absence of SSL, and the average severity score of this lesion was also decreased. There were no significant dose-related effects of the whole leaf creams on the incidences and severity scores of squamous hyperplasia in male or female mice exposed to 13.70 mJ • CIE/cm^2 SSL, and pairwise comparison tests with the control cream groups were similar in all groups.

Focal atypical squamous hyperplasia did not occur in male or female mice administered control cream or 6% whole leaf cream in the absence of SSL (Table 16). There were no significant dose-related effects in focal atypical squamous hyperplasia incidences or multiplicities in male mice. At 13.70 mJ • CIE/cm^2 SSL, a significant negative dose-related trend was observed in the incidences of focal atypical squamous hyperplasia in female mice; however, pairwise comparisons showed no significant differences in the incidences of this lesion in female mice administered 3% or 6% whole leaf creams and the control cream groups. A significant negative dose-related trend was observed in the multiplicities of focal atypical squamous hyperplasia in female mice, and pairwise comparison tests with the control cream group indicated significantly decreased multiplicities in females administered 3% and 6% whole leaf creams.

Decolorized Whole Leaf Creams

Similar effects that were observed in mice administered whole leaf creams were observed in mice administered decolorized whole leaf creams (Table 17). The incidence of squamous hyperplasia was unaffected by treatment in the absence of SSL in male mice. In female mice administered 6% decolorized whole leaf cream and not exposed to SSL, the incidence of squamous hyperplasia was significantly decreased in a pairwise comparison test

TABLE 14
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream and Control Cream

	No Cream	Control Cream
Male		
0.00 mJ • CIE/cm^2 SSL		
Squamous Hyperplasia		
Overall rate[a]	1/36 (2.8%)	1/35 (2.9%)
Poly-3 test (incidence)[b]	P = 0.736	
Average severity[c]	1.0	2.0
Focal Atypical Squamous Hyperplasia		
Overall rate	0/36 (0.0%)	0/35 (0.0%)
Poly-3 test (incidence)	—[d]	
Overall mean lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)	0.0 (0.0-0.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)	0.0 (0.0-0.0)
Poisson regression (multiplicity)[b]	—	
13.70 mJ • CIE/cm^2 SSL		
Squamous Hyperplasia		
Overall rate	35/36 (97.2%)	31/35 (88.6%)
Poly-3 test (incidence)	P = 0.373N	
Average severity	1.7	1.7
Focal Atypical Squamous Hyperplasia		
Overall rate	33/36 (91.7%)	31/35 (88.6%)
Poly-3 test (incidence)	P = 0.760	
Overall mean lesion multiplicity (95% C.I.)	3.3 (2.6-3.9)	3.1 (2.4-3.7)
Age-adjusted lesion multiplicity (95% C.I.)	4.3 (3.6-5.1)	4.3 (3.6-5.2)
Poisson regression (multiplicity)	P = 0.470	

TABLE 14
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream and Control Cream

	No Cream	Control Cream
Female		
0.00 mJ • CIE/cm^2 SSL		
Squamous Hyperplasia		
Overall rate	1/36 (2.8%)	10/36 (27.8%)
Poly-3 test (incidence)	P = 0.002*	
Average severity	1.0	1.2
Focal Atypical Squamous Hyperplasia		
Overall rate	0/36 (0.0%)	0/36 (0.0%)
Poly-3 test (incidence)	—	
Overall mean lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)	0.0 (0.0-0.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)	0.0 (0.0-0.0)
Poisson regression (multiplicity)	—	
13.70 mJ • CIE/cm^2 SSL		
Squamous Hyperplasia		
Overall rate	36/36 (100.0%)	35/36 (97.2%)
Poly-3 test (incidence)	P = 1.000N	
Average severity	1.8	2.1
Focal Atypical Squamous Hyperplasia		
Overall rate	32/36 (88.9%)	35/36 (97.2%)
Poly-3 test (incidence)	P = 0.385	
Overall mean lesion multiplicity (95% C.I.)	2.8 (2.2-3.5)	3.3 (2.7-3.8)
Age-adjusted lesion multiplicity (95% C.I.)	3.5 (2.9-4.2)	4.4 (3.7-5.1)
Poisson regression (multiplicity)	P = 0.065	

* Significant at P≤0.05
C.I. = confidence interval
[a] Number of animals with specified nonneoplastic skin lesion/number of animals with skin examined microscopically
[b] P values represent pairwise comparison with the control cream group. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. A lower incidence in the control cream group is indicated by N.
[c] Average severity grade of lesions in affected animals: 1 = minimal, 2 = mild, 3 = moderate, 4 = marked
[d] Value of statistic cannot be computed.

TABLE 15
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe Gel Creams

	Control Cream	3% Aloe Gel[a]	6% Aloe Gel
Male			
0.00 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate[b]	1/35 (2.9%)		1/36 (2.8%)
Poly-3 test (incidence)[c]			P = 0.745
Average severity[d]	2.0		1.0
Focal Atypical Squamous Hyperplasia			
Overall rate	0/35 (0.0%)		0/36 (0.0%)
Poly-3 test (incidence)			—[e]
Overall mean lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Poisson regression (multiplicity)[c]			—
13.70 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	31/35 (88.6%)	33/36 (91.7%)	36/36 (100.0%)
Poly-3 test (incidence)	P = 0.141	P = 0.622	P = 0.282
Average severity	1.7	1.6	1.8
Focal Atypical Squamous Hyperplasia			
Overall rate	31/35 (88.6%)	28/36 (77.8%)	34/36 (94.4%)
Poly-3 test (incidence)	P = 0.531	P = 0.640N	P = 0.760
Overall mean lesion multiplicity (95% C.I.)	3.1 (2.4-3.7)	2.6 (2.0-3.2)	3.3 (2.7-3.8)
Age-adjusted lesion multiplicity (95% C.I.)	4.3 (3.6-5.2)	3.6 (3.0-4.4)	4.4 (3.7-5.2)
Poisson regression (multiplicity)	P = 0.488	P = 0.131N	P = 0.488

TABLE 15
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe Gel Creams

	Control Cream	3% Aloe Gel[a]	6% Aloe Gel
Female			
0.00 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	10/36 (27.8%)		13/36 (36.1%)
Poly-3 test (incidence)			P = 0.372
Average severity	1.2		1.0
Focal Atypical Squamous Hyperplasia			
Overall rate	0/36 (0.0%)		0/36 (0.0%)
Poly-3 test (incidence)			—
Overall mean lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Poisson regression (multiplicity)			—
13.70 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	35/36 (97.2%)	34/36 (94.4%)	33/36 (91.7%)
Poly-3 test (incidence)	P = 0.798N	P = 1.000N	P = 0.994N
Average severity	2.1	2.0	2.1
Focal Atypical Squamous Hyperplasia			
Overall rate	35/36 (97.2%)	27/36 (75.0%)	29/36 (80.6%)
Poly-3 test (incidence)	P = 0.087N	P = 0.031N*	P = 0.157N
Overall mean lesion multiplicity (95% C.I.)	3.3 (2.7-3.8)	1.8 (1.3-2.3)	2.3 (1.7-2.9)
Age-adjusted lesion multiplicity (95% C.I.)	4.4 (3.7-5.2)	2.4 (1.9-2.9)	3.1 (2.5-3.7)
Poisson regression (multiplicity)	P = 0.013N*	P≤0.001N*	P = 0.013N*

* Significant at P≤0.05
C.I. = confidence interval
[a] Article not tested @ 0.00 mJ • CIE/cm^2 SSL
[b] Number of animals with specified nonneoplastic skin lesion/number of animals with skin examined microscopically
[c] P values for control cream represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to control. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. Negative trends and P values are appended with an N.
[d] Average severity grade of lesions in affected animals: 1 = minimal, 2 = mild, 3 = moderate, 4 = marked
[e] Value of statistic cannot be computed.

TABLE 16
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Whole Leaf Creams

	Control Cream	3% Whole Leaf[a]	6% Whole Leaf
Male			
0.00 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate[b]	1/35 (2.9%)		0/35 (0.0%)
Poly-3 test (incidence)[c]			P = 0.496N
Average severity[d]	2.0		
Focal Atypical Squamous Hyperplasia			
Overall rate	0/35 (0.0%)		0/35 (0.0%)
Poly-3 test (incidence)			—[e]
Overall mean lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Poisson regression (multiplicity)[c]			—
13.70 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	31/35 (88.6%)	34/36 (94.4%)	34/36 (94.4%)
Poly-3 test (incidence)	P = 0.272	P = 0.666	P = 0.440
Average severity	1.7	1.5	1.7
Focal Atypical Squamous Hyperplasia			
Overall rate	31/35 (88.6%)	29/36 (80.6%)	34/36 (94.4%)
Poly-3 test (incidence)	P = 0.311	P = 0.362N	P = 0.515
Overall mean lesion multiplicity (95% C.I.)	3.1 (2.4-3.7)	3.0 (2.4-3.6)	3.6 (2.9-4.2)
Age-adjusted lesion multiplicity (95% C.I.)	4.3 (3.6-5.2)	4.1 (3.4-4.9)	4.9 (4.1-5.8)
Poisson regression (multiplicity)	P = 0.235	P = 0.360N	P = 0.235

TABLE 16
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Whole Leaf Creams

	Control Cream	3% Whole Leaf	6% Whole Leaf
Female			
0.00 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	10/36 (27.8%)		2/36 (5.6%)
Poly-3 test (incidence)			P = 0.006N*
Average severity	1.2		1.0
Focal Atypical Squamous Hyperplasia			
Overall rate	0/36 (0.0%)		0/36 (0.0%)
Poly-3 test (incidence)			—
Overall mean lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Poisson regression (multiplicity)			—
13.70 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	35/36 (97.2%)	35/36 (97.2%)	32/36 (88.9%)
Poly-3 test (incidence)	P = 0.082N	P = 1.000N	P = 0.276N
Average severity	2.1	1.8	2.0
Focal Atypical Squamous Hyperplasia			
Overall rate	35/36 (97.2%)	33/36 (91.7%)	29/36 (80.6%)
Poly-3 test (incidence)	P = 0.017N*	P = 0.783N	P = 0.071N
Overall mean lesion multiplicity (95% C.I.)	3.3 (2.7-3.8)	2.8 (2.3-3.3)	2.5 (1.9-3.0)
Age-adjusted lesion multiplicity (95% C.I.)	4.4 (3.8-5.1)	3.5 (3.0-4.1)	3.2 (2.7-3.8)
Poisson regression (multiplicity)	P = 0.010N*	P = 0.034N*	P = 0.010N*

* Significant at P≤0.05
C.I. = confidence interval
[a] Article not tested @ 0.00 mJ • CIE/cm^2 SSL
[b] Number of animals with specified nonneoplastic skin lesion/number of animals with skin examined microscopically
[c] P values for control cream represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to control. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. Negative trends and P values are appended with an N.
[d] Average severity grade of lesions in affected animals: 1 = minimal, 2 = mild, 3 = moderate, 4 = marked
[e] Value of statistic cannot be computed.

with the control cream group, and the lesion had a lower average severity score. The incidences and severity scores of squamous hyperplasia in male and female mice administered decolorized whole leaf creams and exposed to 13.70 mJ • CIE/cm^2 SSL showed no significant dose-related trends.

Focal atypical squamous hyperplasia did not occur in male or female mice administered control cream or 6% decolorized whole leaf cream in the absence of SSL (Table 17). No treatment-related differences in the incidences of focal atypical squamous hyperplasia were observed in male mice exposed to 13.70 mJ • CIE/cm^2 SSL. In female mice exposed to 13.70 mJ • CIE/cm^2 SSL, a significant negative dose-related trend occurred in the incidences of focal atypical squamous hyperplasia, and the incidence of this lesion was significantly decreased in females administered 6% decolorized whole leaf cream. No significant dose-related trends were observed in the multiplicities of focal atypical squamous hyperplasia in male or female mice administered decolorized whole leaf creams, and the only significant difference in pairwise comparison tests with the control cream groups was a decreased multiplicity in females exposed to 13.70 mJ • CIE/cm^2 SSL and administered 3% decolorized whole leaf cream.

Aloe-emodin Creams

The incidences, severity scores, and multiplicities of nonneoplastic skin lesions at the site of application in male and female mice administered aloe-emodin creams and exposed to 0 or 13.70 mJ • CIE/cm^2 SSL are presented in Table 18. In a pairwise comparison test with the control cream group in the absence of SSL exposure, 74.6 µg/g aloe-emodin cream significantly decreased the incidence of squamous hyperplasia in female mice. Average severity scores of squamous hyperplasia in male and female mice administered aloe-emodin creams and exposed to 13.70 mJ • CIE/cm^2 SSL were similar to those of the control cream groups, and the incidence rates showed no significant dose-related trends or differences in pairwise comparisons with the control cream groups.

A single incidence of focal atypical squamous hyperplasia occurred in a male mouse administered 74.6 µg/g aloe-emodin cream and not exposed to SSL (Table 18). In female mice exposed to 13.70 mJ • CIE/cm^2 SSL, a significant negative dose-related trend was observed in the incidences of focal atypical squamous hyperplasia. Similar trend effects were not observed in male mice, and there were no differences in pairwise comparison tests with the male or female control cream groups. A significant negative dose-related trend in the multiplicities of focal atypical squamous hyperplasia was observed in female mice exposed to 13.70 mJ • CIE/cm^2 SSL, and the multiplicity was significantly decreased in 74.6 µg/g aloe-emodin females compared to the control cream group. Similar effects were not observed in male mice.

Skin Neoplasms

This section describes the statistically significant changes in incidences of neoplasms that occurred in the study. The skin at the site of application of the creams (coincident with the site of SSL exposure) was the target tissue of the study. Summaries of the incidences of neoplasms and statistical analyses of primary neoplasms are presented in Appendix A for male mice and Appendix B for female mice.

The exposure of mice to SSL resulted in a time- and exposure-dependent formation of proliferative neoplasms in the epidermis. The proliferative skin lesions consisted of a continuum of morphologic alterations that began with focal atypical squamous hyperplasia and progressed with increased proliferation and dysplastic changes to three morphologically distinct types of squamous cell neoplasms in the epidermis: squamous cell papilloma, squamous cell carcinoma *in situ*, and squamous cell carcinoma. Squamous cell papilloma was a solitary focus, usually sessile, with arborized projections elevated above the surface of the skin and consisted of a core of fibrovascular tissue contiguous with the dermis and covered by an orderly arrangement of multiple layers of well-differentiated, often hyperkeratotic, stratified squamous epithelia (Plate 2). Squamous cell carcinoma *in situ* was characterized as a small nodule that at times rose above the surface of the skin and ranged in size from 1 to several millimeters in diameter (Plate 3A). Squamous cell carcinoma *in situ* was a discrete nodule that lacked the orderly arrangement of squamous epithelia, consisted of masses of sheets of cells that lacked cohesion and orientation, had borders that were sharply demarcated from and often compressed the underlying dermis, and was frequently observed in multiples and with other tumor types. Squamous cell carcinoma was characterized by microscopy as downward-projecting sheets, nests, and anastomosing cords of neoplastic squamous cells that extended into the dermis and, in some instances, penetrated the skeletal muscle and invaded the subcutaneous tissue (Plate 3B). Grossly, squamous cell

TABLE 17
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Decolorized Whole Leaf Creams

	Control Cream	3% Decolorized Whole Leaf[a]	6% Decolorized Whole Leaf
Male			
0.00 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate[b]	1/35 (2.9%)		1/36 (2.8%)
Poly-3 test (incidence)[c]			P = 0.754N
Average severity[d]	2.0		2.0
Focal Atypical Squamous Hyperplasia			
Overall rate	0/35 (0.0%)		0/36 (0.0%)
Poly-3 test (incidence)			—[e]
Overall mean lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Poisson regression (multiplicity)[c]			—
13.70 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	31/35 (88.6%)	35/36 (97.2%)	35/36 (97.2%)
Poly-3 test (incidence)	P = 0.116	P = 0.379	P = 0.304
Average severity	1.7	1.5	1.5
Focal Atypical Squamous Hyperplasia			
Overall rate	31/35 (88.6%)	34/36 (94.4%)	30/36 (83.3%)
Poly-3 test (incidence)	P = 0.254N	P = 0.564	P = 0.448N
Overall mean lesion multiplicity (95% C.I.)	3.1 (2.4-3.7)	2.9 (2.3-3.5)	3.3 (2.6-4.0)
Age-adjusted lesion multiplicity (95% C.I.)	4.3 (3.6-5.3)	4.2 (3.4-5.1)	4.5 (3.8-5.4)
Poisson regression (multiplicity)	P = 0.401	P = 0.408N	P = 0.401

TABLE 17
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Decolorized Whole Leaf Creams

	Control Cream	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf
Female			
0.00 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	10/36 (27.8%)		1/36 (2.8%)
Poly-3 test (incidence)			P = 0.001N*
Average severity	1.2		1.0
Focal Atypical Squamous Hyperplasia			
Overall rate	0/36 (0.0%)		0/36 (0.0%)
Poly-3 test (incidence)			—
Overall mean lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Poisson regression (multiplicity)			—
13.70 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	35/36 (97.2%)	34/36 (94.4%)	35/36 (97.2%)
Poly-3 test (incidence)	P = 0.925N	P = 0.969N	P = 1.000N
Average severity	2.1	2.0	1.9
Focal Atypical Squamous Hyperplasia			
Overall rate	35/36 (97.2%)	32/36 (88.9%)	27/36 (75.0%)
Poly-3 test (incidence)	P = 0.003N*	P = 0.422N	P = 0.015N*
Overall mean lesion multiplicity (95% C.I.)	3.3 (2.7-3.8)	1.9 (1.5-2.2)	2.7 (1.9-3.5)
Age-adjusted lesion multiplicity (95% C.I.)	4.3 (3.6-5.2)	2.5 (2.0-3.1)	3.5 (2.9-4.2)
Poisson regression (multiplicity)	P = 0.089N	P≤0.001N*	P = 0.089N

* Significant at P≤0.05
C.I. = confidence interval
[a] Article not tested @ 0.00 mJ • CIE/cm2 SSL
[b] Number of animals with specified nonneoplastic skin lesion/number of animals with skin examined microscopically
[c] P values for control cream represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to control. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. Negative trends and P values are appended with an N.
[d] Average severity grade of lesions in affected animals: 1 = minimal, 2 = mild, 3 = moderate, 4 = marked
[e] Value of statistic cannot be computed.

TABLE 18
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe-emodin Creams

	Control Cream	7.46 µg/g Aloe-emodin[a]	74.6 µg/g Aloe-emodin
Male			
0.00 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate[b]	1/35 (2.9%)		1/35 (2.9%)
Poly-3 test (incidence)[c]			P = 0.759
Average severity[d]	2.0		1.0
Focal Atypical Squamous Hyperplasia			
Overall rate	0/35 (0.0%)		1/35 (2.9%)
Poly-3 test (incidence)			P = 0.499
Overall mean lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Poisson regression (multiplicity)[c]			—[e]
13.70 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	31/35 (88.6%)	32/35 (91.4%)	32/36 (88.9%)
Poly-3 test (incidence)	P = 0.558	P = 0.669	P = 0.714
Average severity	1.7	1.6	1.7
Focal Atypical Squamous Hyperplasia			
Overall rate	31/35 (88.6%)	30/35 (85.7%)	33/36 (91.7%)
Poly-3 test (incidence)	P = 0.552	P = 0.652N	P = 0.785
Overall mean lesion multiplicity (95% C.I.)	3.1 (2.4-3.7)	2.9 (2.3-3.5)	3.5 (2.7-4.2)
Age-adjusted lesion multiplicity (95% C.I.)	4.3 (3.6-5.3)	4.1 (3.3-5.0)	4.8 (4.0-5.8)
Poisson regression (multiplicity)	P = 0.263	P = 0.355N	P = 0.263

TABLE 18
Incidences, Severities, and Multiplicities of Nonneoplastic Skin Lesions at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe-emodin Creams

	Control Cream	7.46 µg/g Aloe-emodin[a]	74.6 µg/g Aloe-emodin
Female			
0.00 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	10/36 (27.8%)		2/36 (5.6%)
Poly-3 test (incidence)			P = 0.005N*
Average severity	1.2		1.5
Focal Atypical Squamous Hyperplasia			
Overall rate	0/36 (0.0%)		0/36 (0.0%)
Poly-3 test (incidence)			—
Overall mean lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.0 (0.0-0.0)		0.0 (0.0-0.0)
Poisson regression (multiplicity)			—
13.70 mJ • CIE/cm^2 SSL			
Squamous Hyperplasia			
Overall rate	35/36 (97.2%)	34/36 (94.4%)	34/36 (94.4%)
Poly-3 test (incidence)	P = 0.787N	P = 0.516N	P = 1.000N
Average severity	2.1	1.9	1.8
Focal Atypical Squamous Hyperplasia			
Overall rate	35/36 (97.2%)	32/36 (88.9%)	27/36 (75.0%)
Poly-3 test (incidence)	P = 0.027N*	P = 0.350N	P = 0.077N
Overall mean lesion multiplicity (95% C.I.)	3.3 (2.7-3.8)	2.6 (2.1-3.1)	2.3 (1.8-2.9)
Age-adjusted lesion multiplicity (95% C.I.)	4.4 (3.8-5.1)	3.5 (2.9-4.1)	3.1 (2.6-3.8)
Poisson regression (multiplicity)	P = 0.011N*	P = 0.051N	P = 0.011N*

* Significant at P≤0.05
C.I. = confidence interval
[a] Article not tested @ 0.00 mJ • CIE/cm^2 SSL
[b] Number of animals with specified nonneoplastic skin lesion/number of animals with skin examined microscopically
[c] P values for control cream represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to control. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. Negative trends and P values are appended with an N.
[d] Average severity grade of lesions in affected animals: 1 = minimal, 2 = mild, 3 = moderate, 4 = marked
[e] Value of statistic cannot be computed.

carcinoma often appeared as a nodular mass with an irregular surface that was often ulcerated and crater-formed with large masses of keratin that occupied the central crater-formed depression of the mass.

In addition to the three morphologically distinct types of squamous cell neoplasms in the epidermis, combinations of these neoplasms were analyzed. These included the combination of squamous cell carcinoma *in situ* and/or squamous cell carcinoma and the combination of squamous cell papilloma, squamous cell carcinoma *in situ*, and/or squamous cell carcinoma.

No Cream

The incidences of skin neoplasms at the site of application in male and female mice that did not receive cream and were exposed to SSL are summarized in Table 19. The control group for this analysis was the group of mice not exposed to SSL, and the site of application for this analysis was the area of dorsal skin that was exposed to SSL and extended from the nape of the neck to the base of the tail and midway along both sides of the animal.

There were no skin neoplasms detected in male or female control mice. The results of the Poly-3 incidence analysis showed that the incidences of squamous cell papilloma, squamous cell carcinoma *in situ*, and squamous cell carcinoma, when considered as separate lesion types or in combination, were highly dependent on exposure to SSL, and significant SSL exposure-response trends were observed in male and female mice.

In pairwise comparison tests with the control mice, the exposure of mice to SSL significantly enhanced the incidences of squamous cell papilloma and squamous cell carcinoma *in situ* at the site of application, when the neoplasm types were considered separately or in combination with squamous cell carcinoma. Mice exposed to 13.70 or 20.55 mJ • CIE/cm^2 SSL had significantly increased incidences of squamous cell carcinoma.

The results of the Relative Treatment Effect linear trend analyses for neoplasm multiplicities in male and female mice that did not receive cream treatment are also shown in Table 19 and Figure 15. Significant positive exposure-related trends were observed in the multiplicities of squamous cell neoplasms in the no cream male and female mice exposed to SSL for each morphology type and combination of neoplasms that were evaluated by histopathology. In male and female mice, SSL exposure significantly enhanced the multiplicities of squamous cell papilloma, squamous cell carcinoma *in situ*, and the combinations of the squamous cell carcinoma morphologies and all squamous cell neoplasms. The multiplicities of squamous cell carcinoma were significantly enhanced in mice exposed to 13.70 or 20.55 mJ • CIE/cm^2 SSL but not in mice exposed to 6.85 mJ • CIE/cm^2 SSL.

Control Cream

No skin neoplasms of any morphology type or combination occurred in no cream and control cream male and female mice not exposed to SSL. Increased incidences of skin neoplasms occurred in no cream and control cream mice exposed to 13.70 mJ • CIE/cm^2 SSL; however, no significant differences were detected for the incidences of neoplasms or combinations of neoplasms between treatments in male or female mice.

The results of the Poisson tests of neoplasm multiplicities by neoplasm type are given in Figure 16. In pairwise comparison tests, there were no significant differences in the multiplicities of skin neoplasms, either by morphology type or combination of morphology types, between no cream or control cream male or female mice.

Aloe vera Creams at 0.00 mJ • CIE/cm2 SSL

With the exception of a single squamous cell papilloma in a female mouse administered 6% whole leaf cream (Table B1), there were no skin neoplasms in Aloe vera cream-treated mice not exposed to SSL.

TABLE 19
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

	0.00 mJ • CIE/cm^2 SSL	6.85 mJ • CIE/cm^2 SSL	13.70 mJ • CIE/cm^2 SSL	20.55 mJ • CIE/cm^2 SSL
Male				
Squamous Cell Papilloma				
Overall rate[a]	0/36 (0.0%)	5/36 (13.9%)	22/36 (61.1%)	10/36 (27.8%)
Adjusted rate[b]	0/35.7 (0.0%)	5/29.1 (17.2%)	22/27.8 (79.2%)	10/14.6 (68.5%)
Terminal rate[c]	0/35 (0.0%)	3/22 (13.6%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	—[e]	327	235	200
Poly-3 test (incidence)[d]	P≤0.001*	P = 0.015*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.3 ± 0.1 (0.0-0.5)	1.1 ± 0.2 (0.7-1.4)	0.5 ± 0.1 (0.2-0.7)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.3 ± 0.1 (0.0-0.5)	1.4 ± 0.3 (0.9-1.9)	0.8 ± 0.2 (0.4-1.1)
Relative treatment effect regression (multiplicity)[d]	P≤0.001*	P = 0.027*	P≤0.001*	P≤0.001*
Squamous Cell Carcinoma *in situ*				
Overall rate	0/36 (0.0%)	7/36 (19.4%)	31/36 (86.1%)	27/36 (75.0%)
Adjusted rate	0/35.7 (0.0%)	7/29.3 (23.9%)	31/33.0 (93.8%)	27/28.6 (94.5%)
Terminal rate	0/35 (0.0%)	3/22 (13.6%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	—	327	235	181
Poly-3 test (incidence)	P≤0.001*	P = 0.002*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.4 ± 0.2 (0.0-0.8)	2.5 ± 0.4 (1.9-3.2)	2.8 ± 0.4 (2.1-3.4)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.4 ± 0.3 (0.0-0.9)	3.4 ± 0.5 (2.5-4.3)	4.8 ± 0.7 (3.7-5.9)
Relative treatment effect regression (multiplicity)	P≤0.001*	P = 0.006*	P≤0.001*	P≤0.001*
Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	0/36 (0.0%)	19/36 (52.8%)	26/36 (72.2%)
Adjusted rate	0/35.7 (0.0%)	0/28.5 (0.0%)	19/26.5 (71.7%)	26/27.9 (93.3%)
Terminal rate	0/35 (0.0%)	0/22 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	—	—	235	181
Poly-3 test (incidence)	P≤0.001*	—[f]	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.0 ± 0.0 (0.0-0.0)	0.7 ± 0.1 (0.5-1.0)	1.4 ± 0.2 (1.1-1.8)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.0 ± 0.0 (0.0-0.0)	0.9 ± 0.2 (0.7-1.2)	2.4 ± 0.4 (1.8-3.0)
Relative treatment effect regression (multiplicity)	P≤0.001*	P = 1.000	P≤0.001*	P≤0.001*

TABLE 19
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

	0.00 mJ • CIE/cm^2 SSL	6.85 mJ • CIE/cm^2 SSL	13.70 mJ • CIE/cm^2 SSL	20.55 mJ • CIE/cm^2 SSL
Male (continued)				
Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	7/36 (19.4%)	35/36 (97.2%)	33/36 (91.7%)
Adjusted rate	0/35.7 (0.0%)	7/29.3 (23.9%)	35/35.2 (99.4%)	33/33.3 (99.2%)
Terminal rate	0/35 (0.0%)	3/22 (13.6%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	—	327	235	181
Poly-3 test (incidence)	P≤0.001*	P = 0.002*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.4 ± 0.2 (0.0-0.8)	3.3 ± 0.4 (2.6-3.9)	4.2 ± 0.4 (3.5-4.9)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.4 ± 0.3 (0.0-0.9)	4.3 ± 0.5 (3.5-5.2)	7.2 ± 0.8 (5.9-8.5)
Relative treatment effect regression (multiplicity)	P≤0.001*	P = 0.006*	P≤0.001*	P≤0.001*
Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	9/36 (25.0%)	35/36 (97.2%)	33/36 (91.7%)
Adjusted rate	0/35.7 (0.0%)	9/29.3 (30.7%)	35/35.2 (99.4%)	33/33.3 (99.2%)
Terminal rate	0/35 (0.0%)	5/22 (22.7%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	—	327	235	181
Poly-3 test (incidence)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.7 ± 0.4 (0.1-1.3)	4.3 ± 0.4 (3.6-5.1)	4.7 ± 0.5 (3.8-5.5)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.7 ± 0.4 (0.1-1.4)	5.7 ± 0.6 (4.7-6.7)	8.0 ± 0.8 (6.5-9.4)
Relative treatment effect regression (multiplicity)	P≤0.001*	P = 0.001*	P≤0.001*	P≤0.001*

TABLE 19
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

	0.00 mJ • CIE/cm^2 SSL	6.85 mJ • CIE/cm^2 SSL	13.70 mJ • CIE/cm^2 SSL	20.55 mJ • CIE/cm^2 SSL
Female				
Squamous Cell Papilloma				
Overall rate	0/36 (0.0%)	16/36 (44.4%)	20/36 (55.6%)	16/36 (44.4%)
Adjusted rate	0/32.8 (0.0%)	16/34.4 (46.5%)	20/28.1 (71.1%)	16/20.7 (77.1%)
Terminal rate	0/30 (0.0%)	11/24 (45.8%)	0/1 (0.0%)	0/0 (0.0%)
First incidence (days)	—	320	242	186
Poly-3 test (incidence)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.9 ± 0.2 (0.6-1.2)	1.5 ± 0.3 (1.0-2.0)	1.1 ± 0.3 (0.6-1.7)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.9 ± 0.2 (0.6-1.2)	1.9 ± 0.4 (1.2-2.5)	1.8 ± 0.5 (0.9-2.7)
Relative treatment effect regression (multiplicity)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Squamous Cell Carcinoma *in situ*				
Overall rate	0/36 (0.0%)	9/36 (25.0%)	28/36 (77.8%)	23/36 (63.9%)
Adjusted rate	0/32.8 (0.0%)	9/34.5 (26.1%)	28/31.7 (88.3%)	23/26.1 (88.0%)
Terminal rate	0/30 (0.0%)	5/24 (20.8%)	1/1 (100.0%)	0/0 (0.0%)
First incidence (days)	—	299	215	172
Poly-3 test (incidence)	P≤0.001*	P = 0.002*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.5 ± 0.2 (0.2-0.8)	2.2 ± 0.4 (1.5-2.9)	1.6 ± 0.3 (1.2-2.1)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.6 ± 0.2 (0.2-0.9)	2.7 ± 0.5 (1.9-3.6)	2.6 ± 0.4 (1.9-3.4)
Relative treatment effect regression (multiplicity)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	2/36 (5.6%)	23/36 (63.9%)	17/36 (47.2%)
Adjusted rate	0/32.8 (0.0%)	2/33.4 (6.0%)	23/29.2 (78.8%)	17/21.6 (78.8%)
Terminal rate	0/30 (0.0%)	2/24 (8.3%)	0/1 (0.0%)	0/0 (0.0%)
First incidence (days)	—	366 (T)	250	199
Poly-3 test (incidence)	P≤0.001*	P = 0.240	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.1 ± 0.0 (0.0-0.1)	0.9 ± 0.2 (0.7-1.2)	0.8 ± 0.2 (0.5-1.1)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.1 ± 0.0 (0.0-0.1)	1.1 ± 0.2 (0.8-1.5)	1.3 ± 0.3 (0.8-1.7)
Relative treatment effect regression (multiplicity)	P≤0.001*	P = 0.246	P≤0.001*	P≤0.001*

TABLE 19
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

	0.00 mJ • CIE/cm^2 SSL	6.85 mJ • CIE/cm^2 SSL	13.70 mJ • CIE/cm^2 SSL	20.55 mJ • CIE/cm^2 SSL
Female (continued)				
Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	10/36 (27.8%)	34/36 (94.4%)	28/36 (77.8%)
Adjusted rate	0/32.8 (0.0%)	10/34.5 (29.0%)	34/34.7 (97.9%)	28/30.0 (93.4%)
Terminal rate	0/30 (0.0%)	6/24 (25.0%)	1/1 (100.0%)	0/0 (0.0%)
First incidence (days)	—	299	215	172
Poly-3 test (incidence)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.6 ± 0.2 (0.3-0.9)	3.1 ± 0.5 (2.3-3.9)	2.4 ± 0.4 (1.8-3.1)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	0.6 ± 0.2 (0.3-1.0)	3.9 ± 0.6 (3.0-4.8)	3.9 ± 0.6 (2.9-4.9)
Relative treatment effect regression (multiplicity)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	19/36 (52.8%)	35/36 (97.2%)	32/36 (88.9%)
Adjusted rate	0/32.8 (0.0%)	19/35.0 (54.4%)	35/35.4 (98.8%)	32/33.0 (97.0%)
Terminal rate	0/30 (0.0%)	12/24 (50.0%)	1/1 (100.0%)	0/0 (0.0%)
First incidence (days)	—	299	215	172
Poly-3 test (incidence)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*
Overall mean lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	1.5 ± 0.3 (1.0-2.0)	4.6 ± 0.8 (3.4-5.9)	3.6 ± 0.5 (2.7-4.4)
Age-adjusted lesion multiplicity ± SE (95% C.I.)	0.00 ± 0.0 (0.0-0.0)	1.5 ± 0.3 (1.0-2.1)	5.8 ± 0.9 (4.2-7.3)	5.7 ± 0.8 (4.4-7.0)
Relative treatment effect regression (multiplicity)	P≤0.001*	P≤0.001*	P≤0.001*	P≤0.001*

* Significant at P≤0.05
C.I. = confidence interval; SE = standard error
[a] Number of neoplasm-bearing animals/number of animals with skin examined microscopically
[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] P values for control group represent linear trend tests; otherwise, P values represent pairwise comparisons to control group. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice.
[e] Not applicable; no neoplasms in animal group
[f] Value of statistic cannot be computed.

Female	RTE Trend	Male	RTE Trend
Papilloma	**P<0.001**	Papilloma	**P<0.001**
Carcinoma *in situ*	**P<0.001**	Carcinoma *in situ*	**P<0.001**
Carcinoma	**P<0.001**	Carcinoma	**P<0.001**
All Neoplasms	**P<0.001**	All Neoplasms	**P<0.001**

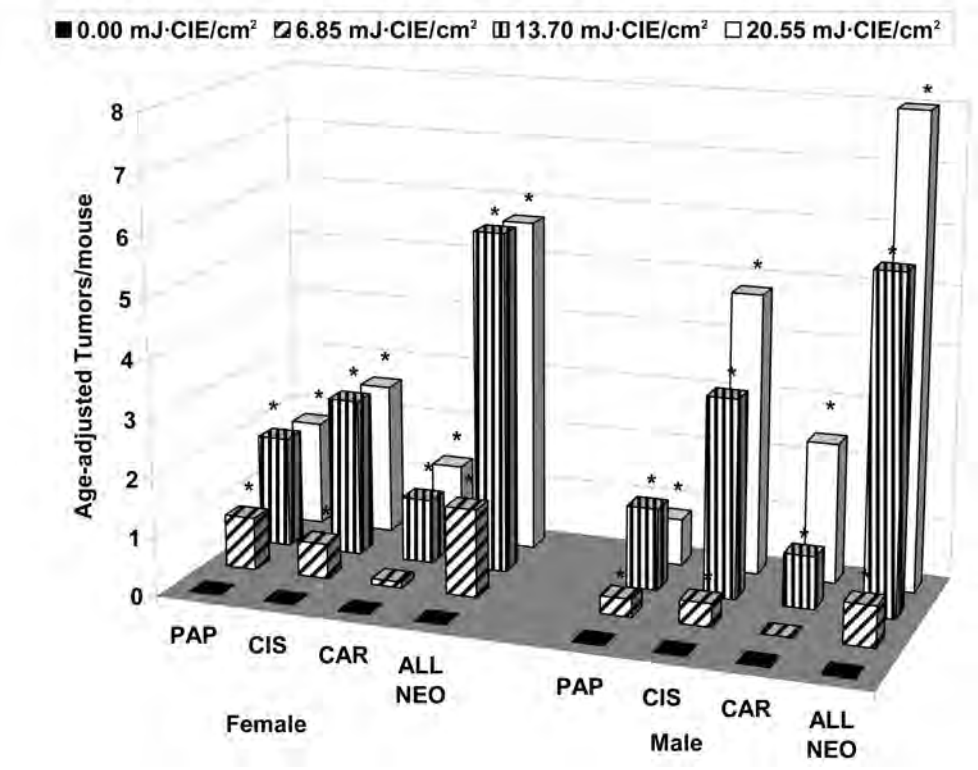


FIGURE 15
Comparisons of the Effects of SSL Exposure on Multiplicity of Squamous Cell Skin Neoplasms at the Site of Application in No Cream Mice in the 1-Year Simulated Solar Light Study of Aloe vera

* Asterisks above chart bars denote significant pairwise comparisons with the 0.00 mJ • CIE/cm^2 SSL controls ($P \leq 0.05$). Significant P values for exposure-related trends are bolded.
RTE = relative treatment effect; PAP = papilloma; CIS = carcinoma *in situ*; CAR = carcinoma; ALL NEO = PAP + CIS + CAR

Female	Poisson Tests
Papilloma	P=0.305
Carcinoma *in situ*	P=0.350
Carcinoma	P=0.492
All Neoplasms	P=0.065

Male	Poisson Tests
Papilloma	P=0.256
Carcinoma *in situ*	P=0.404N
Carcinoma	P=0.472N
All Neoplasms	P=0.439

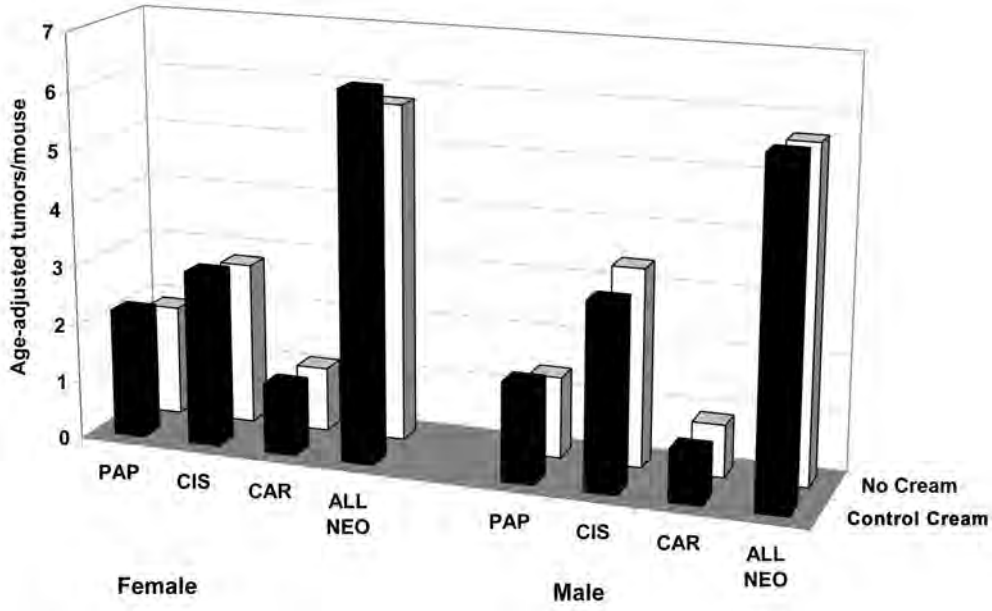


FIGURE 16
Comparisons of the Effects of Control Cream and No Cream on Multiplicity of Squamous Cell Skin Neoplasms at the Site of Application in Mice Exposed to 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

PAP = papilloma; CIS = carcinoma *in situ*; CAR = carcinoma; ALL NEO = PAP + CIS + CAR

Aloe Gel Creams

The incidences of skin neoplasms at the site of application in male and female mice administered aloe gel creams and exposed to 13.70 mJ • CIE/cm^2 SSL are presented in Table 20. There were no treatment-related effects on the incidences of skin neoplasms, either by morphology or combination, in male mice. The induction of squamous cell papilloma showed a positive dose-related trend in female mice, and the incidence of squamous cell papilloma in the 6% aloe gel group was significantly increased. In pairwise comparison tests with the control cream group, the aloe gel creams did not significantly alter the incidences of squamous cell carcinoma *in situ* or squamous cell carcinoma, either when considered separately or in combinations with each other or with squamous cell papilloma.

The exposure of male mice to 13.70 mJ • CIE/cm^2 SSL and the topical application of aloe gel creams did not result in any treatment-related differences in the multiplicities of squamous cell neoplasms, either by morphology type or combination (Table 20; Figure 17). The topical application of aloe gel creams resulted in significant positive trends in the multiplicities of squamous cell papilloma and the combination of all squamous cell neoplasms in female mice exposed to 13.70 mJ • CIE/cm^2 SSL. In pairwise comparison tests with mice that received the control cream, female mice that received the 3% or 6% aloe gel cream had significantly increased multiplicities of squamous cell papilloma and the combination of all squamous cell neoplasms; the multiplicities of squamous cell carcinoma *in situ* and the combination of the squamous cell carcinoma morphologies were significantly increased in females dosed with 3% aloe gel cream. There were no differences in the multiplicities of squamous cell carcinoma between female mice administered control cream and those administered 3% or 6% aloe gel creams.

Whole Leaf Creams

The incidences of skin neoplasms at the site of application in male and female mice administered whole leaf creams and exposed to 13.70 mJ • CIE/cm^2 SSL are presented in Table 21. The incidences of squamous cell papilloma in male and female mice showed significant positive trends, and significantly increased incidences of squamous cell papilloma occurred in male and female mice administered 6% whole leaf cream. The incidences of squamous cell carcinoma *in situ*, squamous cell carcinoma, or combinations of neoplasm types were not altered in male or female mice by the application of whole leaf creams, and no significant dose-related trends were observed for these individual neoplasms or combinations.

As shown in Table 21 and Figure 18, the topical application of creams containing the whole leaf extract of Aloe vera and the concomitant exposure to 13.70 mJ • CIE/cm^2 SSL resulted in significant dose-related trend effects in the multiplicities of squamous cell papilloma in male and female mice and the multiplicities of the combination of all squamous cell neoplasms in male mice. In pairwise comparison tests with the control cream group, the multiplicities of squamous cell carcinoma *in situ*, the combination of squamous cell carcinoma morphologies, and the combination of all squamous cell neoplasms were significantly increased in female mice administered 3% whole leaf cream. There were no differences in the multiplicities of squamous cell neoplasms by morphology type or combination between males administered control cream or 3% whole leaf cream. The multiplicities of squamous cell papilloma in males and females administered 6% whole leaf cream and the multiplicity of all squamous cell neoplasms combined in males administered 6% whole leaf cream were significantly increased.

Decolorized Whole Leaf Creams

The incidences of skin neoplasms at the site of application in male and female mice administered decolorized whole leaf creams and exposed to 13.70 mJ • CIE/cm^2 SSL are presented in Table 22. The incidences of squamous cell neoplasms by morphology type or combination of types were not affected by the decolorized whole leaf creams in male mice, and significant dose-related trends were not observed. As observed with the aloe gel and the whole leaf creams, the induction of squamous cell papilloma showed a significant positive trend in female mice administered decolorized whole leaf creams. In contrast, the incidences of squamous cell carcinoma showed a significant negative trend in females. In pairwise comparisons with the control cream group, females administered 6% decolorized whole leaf cream had a significantly increased incidence of squamous cell papilloma; the incidence of squamous cell carcinoma was significantly decreased in females administered 3% decolorized whole leaf cream.

TABLE 20
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe Gel Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	3% Aloe Gel	6% Aloe Gel
Male			
Squamous Cell Papilloma			
Overall rate[a]	16/35 (45.7%)	19/36 (52.8%)	25/36 (69.4%)
Adjusted rate[b]	16/22.5 (71.0%)	19/24.1 (78.8%)	25/28.9 (86.4%)
Terminal rate[c]	0/0 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	221	228	214
Poly-3 test (incidence)[d]	P = 0.056	P = 0.357	P = 0.090
Overall mean lesion multiplicity (95% C.I.)	1.3 (0.6-2.0)	1.8 (1.0-2.5)	1.6 (1.1-2.0)
Age-adjusted lesion multiplicity (95% C.I.)	1.7 (1.1-2.5)	2.4 (1.6-3.4)	2.1 (1.5-3.1)
Poisson regression (multiplicity)[d]	P = 0.255	P = 0.160	P = 0.255
Squamous Cell Carcinoma *in situ*			
Overall rate	26/35 (74.3%)	28/36 (77.8%)	29/36 (80.6%)
Adjusted rate	26/29.0 (89.6%)	28/29.8 (93.9%)	29/32.3 (89.8%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	235	221	214
Poly-3 test (incidence)	P = 0.619N	P = 0.431	P = 0.697
Overall mean lesion multiplicity (95% C.I.)	2.3 (1.6-2.9)	2.5 (1.9-3.1)	2.9 (2.2-3.6)
Age-adjusted lesion multiplicity (95% C.I.)	3.2 (2.4-4.1)	3.4 (2.7-4.4)	3.9 (3.1-5.0)
Poisson regression (multiplicity)	P = 0.159	P = 0.357	P = 0.159
Squamous Cell Carcinoma			
Overall rate	15/35 (42.9%)	18/36 (50.0%)	20/36 (55.6%)
Adjusted rate	15/22.7 (66.1%)	18/24.2 (74.4%)	20/26.5 (75.4%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	221	222	214
Poly-3 test (incidence)	P = 0.254	P = 0.355	P = 0.312
Overall mean lesion multiplicity (95% C.I.)	0.7 (0.4-0.9)	0.8 (0.5-1.0)	0.8 (0.5-1.0)
Age-adjusted lesion multiplicity (95% C.I.)	0.9 (0.7-1.3)	1.1 (0.8-1.5)	1.0 (0.7-1.4)
Poisson regression (multiplicity)	P = 0.390	P = 0.333	P = 0.390
Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma			
Overall rate	30/35 (85.7%)	31/36 (86.1%)	34/36 (94.4%)
Adjusted rate	30/31.4 (95.6%)	31/31.6 (98.1%)	34/35.1 (96.8%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	221	221	214
Poly-3 test (incidence)	P = 0.558	P = 0.585	P = 0.684
Overall mean lesion multiplicity (95% C.I.)	2.9 (2.2-3.7)	3.2 (2.6-3.9)	3.6 (2.9-4.4)
Age-adjusted lesion multiplicity (95% C.I.)	4.1 (3.3-5.1)	4.5 (3.7-5.5)	4.9 (4.1-6.0)
Poisson regression (multiplicity)	P = 0.151	P = 0.313	P = 0.151

TABLE 20
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe Gel Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	3% Aloe Gel	6% Aloe Gel
Male (continued)			
Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma			
Overall rate	31/35 (88.6%)	31/36 (86.1%)	36/36 (100.0%)
Adjusted rate	31/32.1 (96.6%)	31/31.6 (98.1%)	36/36.0 (100.0%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	221	221	214
Poly-3 test (incidence)	P = 0.202	P = 0.716	P = 0.432
Overall mean lesion multiplicity (95% C.I.)	4.2 (3.0-5.3)	5.0 (3.9-6.0)	5.2 (4.2-6.2)
Age-adjusted lesion multiplicity (95% C.I.)	5.8 (4.7-7.2)	6.8 (5.6-8.4)	7.1 (5.8-8.7)
Poisson regression (multiplicity)	P = 0.135	P = 0.186	P = 0.135
Female			
Squamous Cell Papilloma			
Overall rate	21/36 (58.3%)	25/36 (69.4%)	29/36 (80.6%)
Adjusted rate	21/26.5 (79.2%)	25/28.7 (87.0%)	29/30.5 (95.0%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	237	235
Poly-3 test (incidence)	P = 0.012*	P = 0.283	P = 0.018*
Overall mean lesion multiplicity (95% C.I.)	1.7 (1.1-2.2)	2.9 (2.0-3.8)	2.8 (2.1-3.4)
Age-adjusted lesion multiplicity (95% C.I.)	2.2 (1.6-3.0)	3.8 (2.9-5.0)	3.6 (2.7-4.8)
Poisson regression (multiplicity)	P = 0.025*	P = 0.015*	P = 0.025*
Squamous Cell Carcinoma *in situ*			
Overall rate	30/36 (83.3%)	32/36 (88.9%)	28/36 (77.8%)
Adjusted rate	30/31.9 (93.9%)	32/32.7 (97.9%)	28/30.3 (92.4%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	237	235
Poly-3 test (incidence)	P = 0.532N	P = 0.398	P = 0.650N
Overall mean lesion multiplicity (95% C.I.)	2.3 (1.7-2.8)	3.2 (2.5-3.9)	2.6 (1.9-3.2)
Age-adjusted lesion multiplicity (95% C.I.)	3.0 (2.4-3.8)	4.2 (3.5-5.2)	3.4 (2.8-4.2)
Poisson regression (multiplicity)	P = 0.252	P = 0.028*	P = 0.252
Squamous Cell Carcinoma			
Overall rate	25/36 (69.4%)	22/36 (61.1%)	23/36 (63.9%)
Adjusted rate	25/29.8 (83.8%)	22/27.8 (79.1%)	23/28.8 (79.9%)
Terminal rate	1/2 (50.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	243	194
Poly-3 test (incidence)	P = 0.397N	P = 0.444N	P = 0.480N
Overall mean lesion multiplicity (95% C.I.)	0.9 (0.7-1.1)	0.9 (0.6-1.1)	0.8 (0.6-1.0)
Age-adjusted lesion multiplicity (95% C.I.)	1.2 (0.9-1.5)	1.2 (0.9-1.5)	1.1 (0.9-1.4)
Poisson regression (multiplicity)	P = 0.407N	P = 0.496N	P = 0.407N

TABLE 20
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe Gel Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	3% Aloe Gel	6% Aloe Gel
Female (continued)			
Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma			
Overall rate	35/36 (97.2%)	33/36 (91.7%)	32/36 (88.9%)
Adjusted rate	35/35.0 (100.0%)	33/33.2 (99.4%)	32/32.9 (97.3%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	237	194
Poly-3 test (incidence)	P = 0.233N	P = 1.000N	P = 0.567N
Overall mean lesion multiplicity (95% C.I.)	3.1 (2.6-3.7)	4.1 (3.3-4.8)	3.4 (2.7-4.0)
Age-adjusted lesion multiplicity (95% C.I.)	4.2 (3.5-5.0)	5.4 (4.6-6.3)	4.5 (3.8-5.4)
Poisson regression (multiplicity)	P = 0.288	P = 0.035*	P = 0.288
Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma			
Overall rate	35/36 (97.2%)	33/36 (91.7%)	33/36 (91.7%)
Adjusted rate	35/35.0 (100.0%)	33/33.2 (99.4%)	33/33.3 (99.0%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	237	194
Poly-3 test (incidence)	P = 0.766N	P = 1.000N	P = 0.994N
Overall mean lesion multiplicity (95% C.I.)	4.8 (3.9-5.7)	7.0 (5.7-8.3)	6.1 (5.0-7.2)
Age-adjusted lesion multiplicity (95% C.I.)	6.4 (5.3-7.6)	9.2 (7.8-10.8)	8.1 (6.9-9.6)
Poisson regression (multiplicity)	P = 0.049*	P = 0.006*	P = 0.049*

* Significant at P≤0.05
C.I. = confidence interval
[a] Number of neoplasm-bearing animals/number of animals with skin examined microscopically
[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] P values for control cream represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to control. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. Negative trends and P values are appended with an N.

Female	Poisson Trend
Papilloma	**P=0.025**
Carcinoma *in situ*	P=0.252
Carcinoma	P=0.407N
All Neoplasms	**P=0.049**

Male	Poisson Trend
Papilloma	P=0.255
Carcinoma *in situ*	P=0.159
Carcinoma	P=0.390
All Neoplasms	P=0.135

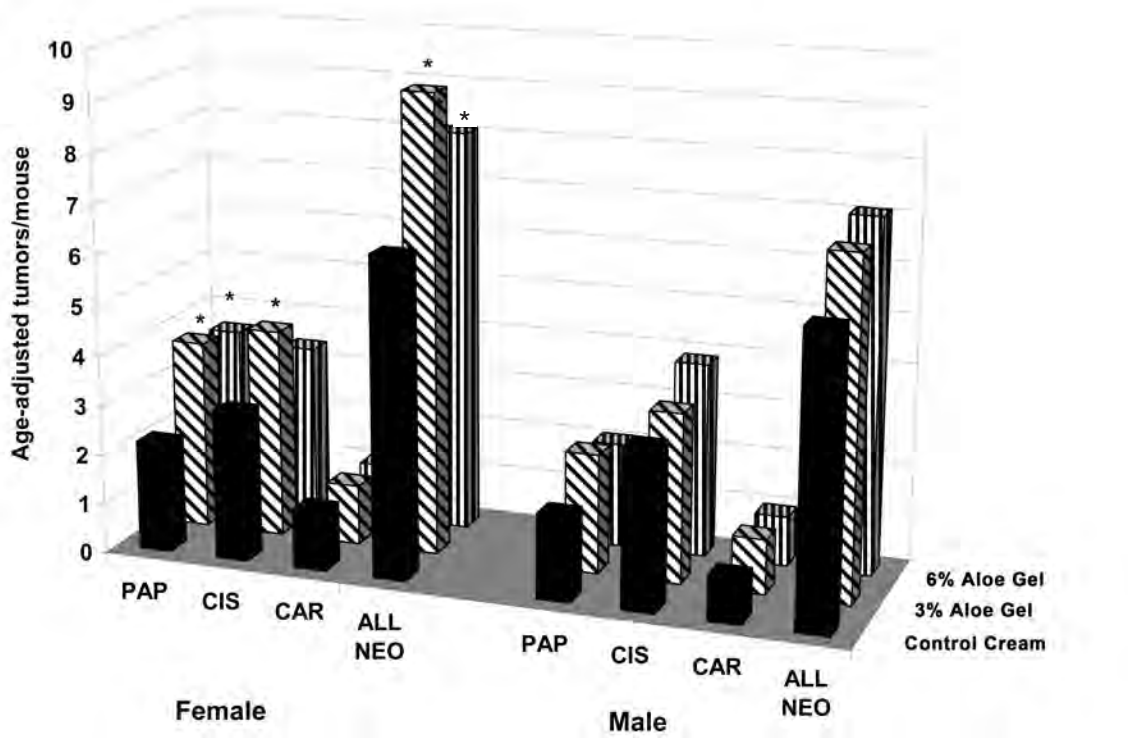


FIGURE 17
Comparisons of the Effects of Control Cream and Aloe Gel Creams on Multiplicity of Squamous Cell Skin Neoplasms at the Site of Application in Mice Exposed to 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

* Asterisks above chart bars denote significant pairwise comparisons with the control cream group ($P \leq 0.05$).
Significant P values for dose-related trends are bolded.
PAP = papilloma; CIS = carcinoma in situ; CAR = carcinoma; ALL NEO = PAP + CIS + CAR

TABLE 21
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Whole Leaf Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	3% Whole Leaf	6% Whole Leaf
Male			
Squamous Cell Papilloma			
Overall rate[a]	16/35 (45.7%)	20/36 (55.6%)	27/36 (75.0%)
Adjusted rate[b]	16/22.5 (71.0%)	20/25.3 (79.2%)	27/30.0 (89.9%)
Terminal rate[c]	0/0 (0.0%)	1/1 (100.0%)	1/1 (100.0%)
First incidence (days)	221	242	221
Poly-3 test (incidence)[d]	P = 0.018*	P = 0.341	P = 0.030*
Overall mean lesion multiplicity (95% C.I.)	1.3 (0.6-2.0)	1.6 (1.0-2.2)	2.3 (1.7-2.9)
Age-adjusted lesion multiplicity (95% C.I.)	1.7 (1.2-2.5)	2.1 (1.5-3.0)	3.1 (2.2-4.3)
Poisson regression (multiplicity)[d]	P = 0.023*	P = 0.248	P = 0.023*
Squamous Cell Carcinoma *in situ*			
Overall rate	26/35 (74.3%)	29/36 (80.6%)	28/36 (77.8%)
Adjusted rate	26/29.0 (89.6%)	29/31.2 (93.1%)	28/30.5 (91.9%)
Terminal rate	0/0 (0.0%)	1/1 (100.0%)	1/1 (100.0%)
First incidence (days)	235	208	221
Poly-3 test (incidence)	P = 0.462	P = 0.492	P = 0.578
Overall mean lesion multiplicity (95% C.I.)	2.3 (1.6-2.9)	2.6 (1.8-3.4)	3.3 (2.4-4.1)
Age-adjusted lesion multiplicity (95% C.I.)	3.2 (2.4-4.2)	3.5 (2.7-4.6)	4.5 (3.5-5.8)
Poisson regression (multiplicity)	P = 0.070	P = 0.340	P = 0.070
Squamous Cell Carcinoma			
Overall rate	15/35 (42.9%)	17/36 (47.2%)	15/36 (41.7%)
Adjusted rate	15/22.7 (66.1%)	17/24.6 (69.2%)	15/23.4 (64.1%)
Terminal rate	0/0 (0.0%)	1/1 (100.0%)	0/1 (0.0%)
First incidence (days)	221	208	242
Poly-3 test (incidence)	P = 0.502N	P = 0.542	P = 0.577N
Overall mean lesion multiplicity (95% C.I.)	0.7 (0.4-0.9)	0.6 (0.4-0.8)	0.6 (0.4-0.8)
Age-adjusted lesion multiplicity (95% C.I.)	0.9 (0.7-1.3)	0.8 (0.6-1.2)	0.8 (0.6-1.2)
Poisson regression (multiplicity)	P = 0.370N	P = 0.361N	P = 0.370N
Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma			
Overall rate	30/35 (85.7%)	32/36 (88.9%)	32/36 (88.9%)
Adjusted rate	30/31.4 (95.6%)	32/32.7 (97.9%)	32/32.7 (97.9%)
Terminal rate	0/0 (0.0%)	1/1 (100.0%)	1/1 (100.0%)
First incidence (days)	221	208	221
Poly-3 test (incidence)	P = 0.403	P = 0.600	P = 0.598
Overall mean lesion multiplicity (95% C.I.)	2.9 (2.2-3.7)	3.2 (2.4-4.0)	3.9 (3.1-4.7)
Age-adjusted lesion multiplicity (95% C.I.)	4.1 (3.3-5.1)	4.3 (3.5-5.4)	5.3 (4.3-6.5)
Poisson regression (multiplicity)	P = 0.082	P = 0.390	P = 0.082

TABLE 21
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Whole Leaf Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	3% Whole Leaf	6% Whole Leaf
Male (continued)			
Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma			
Overall rate	31/35 (88.6%)	32/36 (88.9%)	32/36 (88.9%)
Adjusted rate	31/32.1 (96.6%)	32/32.7 (97.9%)	32/32.7 (97.9%)
Terminal rate	0/0 (0.0%)	1/1 (100.0%)	1/1 (100.0%)
First incidence (days)	221	208	221
Poly-3 test (incidence)	P = 0.543	P = 0.727	P = 0.722
Overall mean lesion multiplicity (95% C.I.)	4.2 (3.0-5.3)	4.8 (3.6-6.0)	6.2 (5.0-7.4)
Age-adjusted lesion multiplicity (95% C.I.)	5.8 (4.7-7.2)	6.4 (5.2-7.9)	8.4 (6.8-10.3)
Poisson regression (multiplicity)	P = 0.022*	P = 0.293	P = 0.022*
Female			
Squamous Cell Papilloma			
Overall rate	21/36 (58.3%)	26/36 (72.2%)	29/36 (80.6%)
Adjusted rate	21/26.5 (79.2%)	26/30.7 (84.8%)	29/31.2 (93.1%)
Terminal rate	2/2 (100.0%)	0/1 (0.0%)	0/0 (0.0%)
First incidence (days)	208	243	236
Poly-3 test (incidence)	P = 0.039*	P = 0.396	P = 0.048*
Overall mean lesion multiplicity (95% C.I.)	1.7 (1.1-2.2)	2.6 (1.8-3.5)	3.1 (2.3-3.9)
Age-adjusted lesion multiplicity (95% C.I.)	2.2 (1.6-3.0)	3.2 (2.4-4.2)	4.0 (3.0-5.2)
Poisson regression (multiplicity)	P = 0.010*	P = 0.068	P = 0.010*
Squamous Cell Carcinoma *in situ*			
Overall rate	30/36 (83.3%)	31/36 (86.1%)	28/36 (77.8%)
Adjusted rate	30/31.9 (93.9%)	31/33.2 (93.4%)	28/31.5 (88.9%)
Terminal rate	2/2 (100.0%)	1/1 (100.0%)	0/0 (0.0%)
First incidence (days)	208	243	236
Poly-3 test (incidence)	P = 0.260N	P = 0.710N	P = 0.363N
Overall mean lesion multiplicity (95% C.I.)	2.3 (1.7-2.8)	3.7 (2.9-4.4)	2.3 (1.6-2.9)
Age-adjusted lesion multiplicity (95% C.I.)	3.0 (2.4-3.8)	4.6 (3.7-5.7)	2.9 (2.3-3.7)
Poisson regression (multiplicity)	P = 0.452N	P = 0.014*	P = 0.452N

TABLE 21
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Whole Leaf Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	3% Whole Leaf	6% Whole Leaf
Female (continued)			
Squamous Cell Carcinoma			
Overall rate	25/36 (69.4%)	20/36 (55.6%)	18/36 (50.0%)
Adjusted rate	25/29.8 (83.8%)	20/27.9 (71.8%)	18/26.4 (68.2%)
Terminal rate	1/2 (50.0%)	1/1 (100.0%)	0/0 (0.0%)
First incidence (days)	208	243	228
Poly-3 test (incidence)	P = 0.070N	P = 0.173N	P = 0.095N
Overall mean lesion multiplicity (95% C.I.)	0.9 (0.7-1.1)	0.7 (0.5-1.0)	0.6 (0.4-0.8)
Age-adjusted lesion multiplicity (95% C.I.)	1.2 (0.9-1.5)	0.9 (0.7-1.2)	0.8 (0.6-1.1)
Poisson regression (multiplicity)	P = 0.067N	P = 0.141N	P = 0.067N
Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma			
Overall rate	35/36 (97.2%)	33/36 (91.7%)	32/36 (88.9%)
Adjusted rate	35/35.0 (100.0%)	33/34.0 (97.0%)	32/33.3 (96.2%)
Terminal rate	2/2 (100.0%)	1/1 (100.0%)	0/0 (0.0%)
First incidence (days)	208	243	228
Poly-3 test (incidence)	P = 0.201N	P = 0.483N	P = 0.357N
Overall mean lesion multiplicity (95% C.I.)	3.1 (2.6-3.7)	4.4 (3.6-5.2)	2.9 (2.2-3.5)
Age-adjusted lesion multiplicity (95% C.I.)	4.2 (3.4-5.0)	5.5 (4.6-6.5)	3.7 (3.1-4.5)
Poisson regression (multiplicity)	P = 0.255N	P = 0.039*	P = 0.255N
Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma			
Overall rate	35/36 (97.2%)	34/36 (94.4%)	34/36 (94.4%)
Adjusted rate	35/35.0 (100.0%)	34/34.2 (99.4%)	34/34.3 (99.2%)
Terminal rate	2/2 (100.0%)	1/1 (100.0%)	0/0 (0.0%)
First incidence (days)	208	243	228
Poly-3 test (incidence)	P = 0.844N	P = 1.000N	P = 0.998N
Overall mean lesion multiplicity (95% C.I.)	4.8 (3.9-5.7)	7.0 (5.7-8.3)	6.0 (4.8-7.1)
Age-adjusted lesion multiplicity (95% C.I.)	6.4 (5.3-7.6)	8.7 (7.4-10.3)	7.7 (6.5-9.1)
Poisson regression (multiplicity)	P = 0.104	P = 0.018*	P = 0.104

* Significant at P≤0.05
C.I. = confidence interval
[a] Number of neoplasm-bearing animals/number of animals with skin examined microscopically
[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] P values for control cream represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to control. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. Negative trends and P values are appended with an N.

Female	Poisson Trend
Papilloma	**P=0.010**
Carcinoma *in situ*	P=0.452N
Carcinoma	P=0.067N
All Neoplasms	P=0.104

Male	Poisson Trend
Papilloma	**P=0.023**
Carcinoma *in situ*	P=0.070
Carcinoma	P=0.370N
All Neoplasms	**P=0.022**

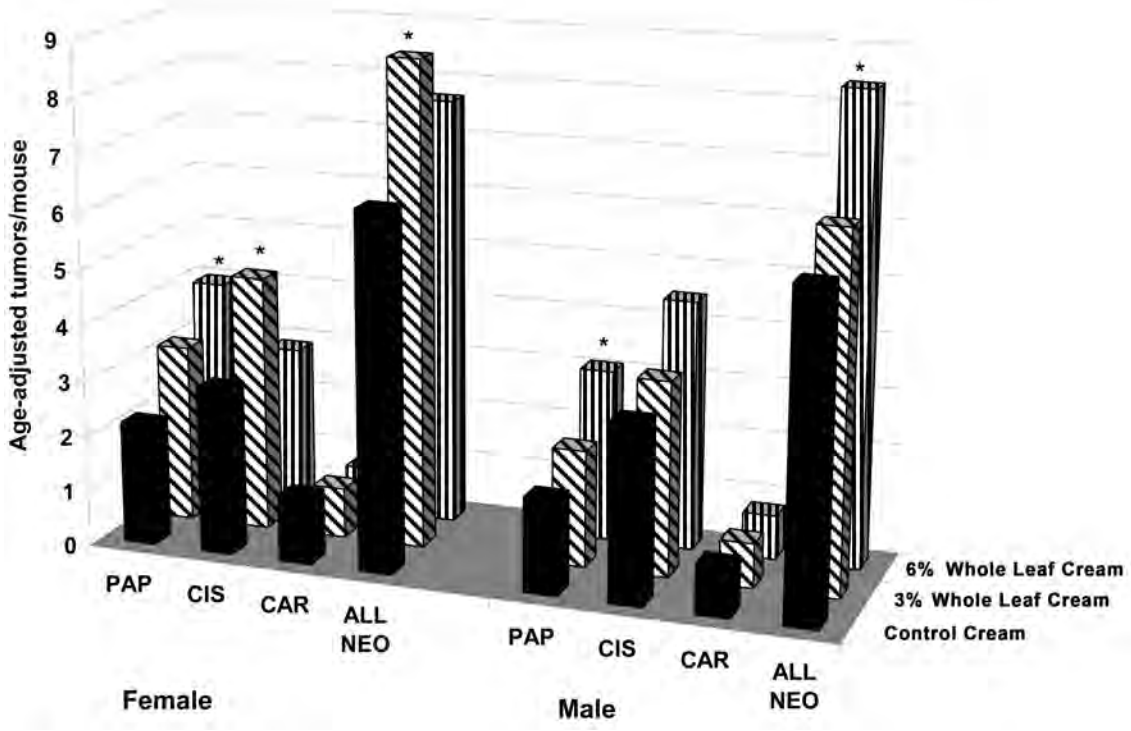


FIGURE 18
Comparisons of the Effects of Control Cream and Whole Leaf Creams on Multiplicity of Squamous Cell Skin Neoplasms at the Site of Application in Mice Exposed to 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

* Asterisks above chart bars denote significant pairwise comparisons with the control cream group ($P \leq 0.05$).
Significant P values for dose-related trends are bolded.
PAP = papilloma; CIS = carcinoma *in situ;* CAR = carcinoma; ALL NEO = PAP + CIS + CAR

A significant positive trend occurred in the multiplicities of squamous cell papilloma and the combination of all squamous cell neoplasms in female mice administered decolorized whole leaf creams (Table 22; Figure 19). In pairwise comparisons with the control cream group, significant increases occurred in the multiplicities of squamous cell papilloma and all squamous cell neoplasms combined in female mice administered 3% or 6% decolorized whole leaf creams. Male and female mice administered 3% decolorized whole leaf cream had significantly increased multiplicities of carcinoma *in situ*, the combination of squamous cell carcinoma morphologies, and the combination of all squamous cell neoplasms.

Aloe-emodin Creams

The incidences of skin neoplasms at the site of application in male and female mice administered aloe-emodin creams and exposed to 13.70 mJ • CIE/cm^2 SSL are given in Table 23. The incidences of squamous cell papilloma in female mice administered aloe-emodin creams showed a significant positive trend that was of similar magnitude to those found for the incidences of squamous cell papilloma in the other aloe cream-treated groups. Compared with the control cream groups, 7.46 µg/g males and 74.6 µg/g females had significantly increased incidences of squamous cell papilloma.

There were no significant dose-related trends or pairwise comparisons with the control cream group in the multiplicities of skin neoplasms in male mice (Table 23; Figure 20). As observed with the other Aloe vera creams, female mice administered aloe-emodin creams apparently were more sensitive to the induction of squamous cell neoplasms than males. The multiplicities of squamous cell papilloma, squamous cell carcinoma *in situ*, the combination of squamous cell carcinoma morphologies, and the combination of all squamous cell neoplasms showed significant positive trends in female mice. In comparison tests with the control cream group, the multiplicities of squamous cell papilloma were significantly increased in 7.46 and 74.6 µg/g females, and the multiplicities of squamous cell carcinoma *in situ*, the combination of squamous cell carcinoma morphologies, and the combination of all squamous cell neoplasms were significantly increased in 74.6 µg/g females.

TABLE 22
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Decolorized Whole Leaf Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf
Male			
Squamous Cell Papilloma			
Overall rate[a]	16/35 (45.7%)	19/36 (52.8%)	20/36 (55.6%)
Adjusted rate[b]	16/22.5 (71.0%)	19/24.4 (77.7%)	20/25.5 (78.4%)
Terminal rate[c]	0/0 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	221	221	242
Poly-3 test (incidence)[d]	P = 0.296	P = 0.401	P = 0.367
Overall mean lesion multiplicity (95% C.I.)	1.3 (0.6-2.0)	1.6 (1.0-2.3)	1.4 (1.0-1.9)
Age-adjusted lesion multiplicity (95% C.I.)	1.7 (1.1-2.6)	2.3 (1.5-3.3)	2.0 (1.3-2.9)
Poisson regression (multiplicity)[d]	P = 0.340	P = 0.204	P = 0.340
Squamous Cell Carcinoma *in situ*			
Overall rate	26/35 (74.3%)	29/36 (80.6%)	26/36 (72.2%)
Adjusted rate	26/29.0 (89.6%)	29/30.6 (94.7%)	26/29.7 (87.5%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	235	214	227
Poly-3 test (incidence)	P = 0.468N	P = 0.361	P = 0.583N
Overall mean lesion multiplicity (95% C.I.)	2.3 (1.6-2.9)	3.5 (2.6-4.4)	2.6 (1.9-3.4)
Age-adjusted lesion multiplicity (95% C.I.)	3.2 (2.4-4.2)	5.0 (3.9-6.4)	3.6 (2.8-4.7)
Poisson regression (multiplicity)	P = 0.295	P = 0.026*	P = 0.295
Squamous Cell Carcinoma			
Overall rate	15/35 (42.9%)	14/36 (38.9%)	20/36 (55.6%)
Adjusted rate	15/22.7 (66.1%)	14/22.1 (63.4%)	20/25.8 (77.5%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	221	214	227
Poly-3 test (incidence)	P = 0.171	P = 0.559N	P = 0.238
Overall mean lesion multiplicity (95% C.I.)	0.7 (0.4-0.9)	0.6 (0.3-0.8)	0.6 (0.4-0.8)
Age-adjusted lesion multiplicity (95% C.I.)	0.9 (0.7-1.3)	0.8 (0.5-1.1)	0.9 (0.6-1.2)
Poisson regression (multiplicity)	P = 0.430N	P = 0.303N	P = 0.430N
Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma			
Overall rate	30/35 (85.7%)	31/36 (86.1%)	30/36 (83.3%)
Adjusted rate	30/31.4 (95.6%)	31/32.1 (96.5%)	30/31.8 (94.4%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	221	214	227
Poly-3 test (incidence)	P = 0.543N	P = 0.735	P = 0.690N
Overall mean lesion multiplicity (95% C.I.)	2.9 (2.2-3.7)	4.1 (3.1-5.0)	3.3 (2.5-4.0)
Age-adjusted lesion multiplicity (95% C.I.)	4.1 (3.2-5.2)	5.8 (4.5-7.2)	4.5 (3.6-5.6)
Poisson regression (multiplicity)	P = 0.335	P = 0.042*	P = 0.335

TABLE 22
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Decolorized Whole Leaf Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf
Male (continued)			
Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma			
Overall rate	31/35 (88.6%)	33/36 (91.7%)	32/36 (88.9%)
Adjusted rate	31/32.1 (96.6%)	33/33.6 (98.4%)	32/33.1 (96.8%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	221	214	227
Poly-3 test (incidence)	P = 0.679	P = 0.666	P = 0.809
Overall mean lesion multiplicity (95% C.I.)	4.2 (3.0-5.3)	5.7 (4.4-7.0)	4.7 (3.7-5.7)
Age-adjusted lesion multiplicity (95% C.I.)	5.8 (4.6-7.3)	8.0 (6.5-9.9)	6.4 (5.2-8.0)
Poisson regression (multiplicity)	P = 0.297	P = 0.045*	P = 0.297
Female			
Squamous Cell Papilloma			
Overall rate	21/36 (58.3%)	30/36 (83.3%)	31/36 (86.1%)
Adjusted rate	21/26.5 (79.2%)	30/32.6 (92.1%)	31/32.9 (94.3%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	229	235
Poly-3 test (incidence)	P = 0.020*	P = 0.077	P = 0.029*
Overall mean lesion multiplicity (95% C.I.)	1.7 (1.1-2.2)	3.0 (2.1-3.9)	3.5 (2.6-4.5)
Age-adjusted lesion multiplicity (95% C.I.)	2.2 (1.6-3.0)	4.0 (3.1-5.3)	4.5 (3.5-5.9)
Poisson regression (multiplicity)	P = 0.002*	P = 0.007*	P = 0.002*
Squamous Cell Carcinoma *in situ*			
Overall rate	30/36 (83.3%)	32/36 (88.9%)	29/36 (80.6%)
Adjusted rate	30/31.9 (93.9%)	32/33.3 (96.1%)	29/31.5 (92.0%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	229	229
Poly-3 test (incidence)	P = 0.469N	P = 0.601	P = 0.604N
Overall mean lesion multiplicity (95% C.I.)	2.3 (1.7-2.8)	3.9 (3.2-4.7)	3.0 (2.3-3.8)
Age-adjusted lesion multiplicity (95% C.I.)	3.0 (2.4-3.8)	5.3 (4.3-6.4)	3.9 (3.2-4.8)
Poisson regression (multiplicity)	P = 0.086	P = 0.001*	P = 0.086

TABLE 22
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Decolorized Whole Leaf Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf
Female (continued)			
Squamous Cell Carcinoma			
Overall rate	25/36 (69.4%)	15/36 (41.7%)	17/36 (47.2%)
Adjusted rate	25/29.8 (83.8%)	15/23.7 (63.2%)	17/25.5 (66.6%)
Terminal rate	1/2 (50.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	244	235
Poly-3 test (incidence)	P = 0.043N*	P = 0.039N*	P = 0.074N
Overall mean lesion multiplicity (95% C.I.)	0.9 (0.7-1.1)	0.6 (0.4-0.8)	0.7 (0.5-1.0)
Age-adjusted lesion multiplicity (95% C.I.)	1.2 (0.9-1.5)	0.8 (0.6-1.1)	0.9 (0.7-1.3)
Poisson regression (multiplicity)	P = 0.194N	P = 0.066N	P = 0.194N
Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma			
Overall rate	35/36 (97.2%)	33/36 (91.7%)	32/36 (88.9%)
Adjusted rate	35/35.0 (100.0%)	33/33.9 (97.4%)	32/33.3 (96.2%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	229	229
Poly-3 test (incidence)	P = 0.170N	P = 0.573N	P = 0.357N
Overall mean lesion multiplicity (95% C.I.)	3.1 (2.6-3.7)	4.5 (3.8-5.3)	3.8 (2.9-4.6)
Age-adjusted lesion multiplicity (95% C.I.)	4.2 (3.4-5.0)	6.0 (5.1-7.2)	4.8 (4.1-5.8)
Poisson regression (multiplicity)	P = 0.169	P = 0.008*	P = 0.169
Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma			
Overall rate	35/36 (97.2%)	34/36 (94.4%)	34/36 (94.4%)
Adjusted rate	35/35.0 (100.0%)	34/34.4 (98.8%)	34/34.2 (99.3%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	229	229
Poly-3 test (incidence)	P = 0.780N	P = 0.928N	P = 1.000N
Overall mean lesion multiplicity (95% C.I.)	4.8 (3.9-5.7)	7.5 (6.2-8.9)	7.3 (5.7-8.8)
Age-adjusted lesion multiplicity (95% C.I.)	6.4 (5.2-7.7)	10.0 (8.4-12.0)	9.3 (7.8-11.1)
Poisson regression (multiplicity)	P = 0.007*	P = 0.002*	P = 0.007*

* Significant at P≤0.05
C.I. = confidence interval
[a] Number of neoplasm-bearing animals/number of animals with skin examined microscopically
[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] P values for control cream represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to control. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. Negative trends and P values are appended with an N.

Female	Poisson Trend
Papilloma	**P=0.002**
Carcinoma *in situ*	P=0.086
Carcinoma	P=0.194N
All Neoplasms	**P=0.007**

Male	Poisson Trend
Papilloma	P=0.340
Carcinoma *in situ*	P=0.295
Carcinoma	P=0.430N
All Neoplasms	P=0.297

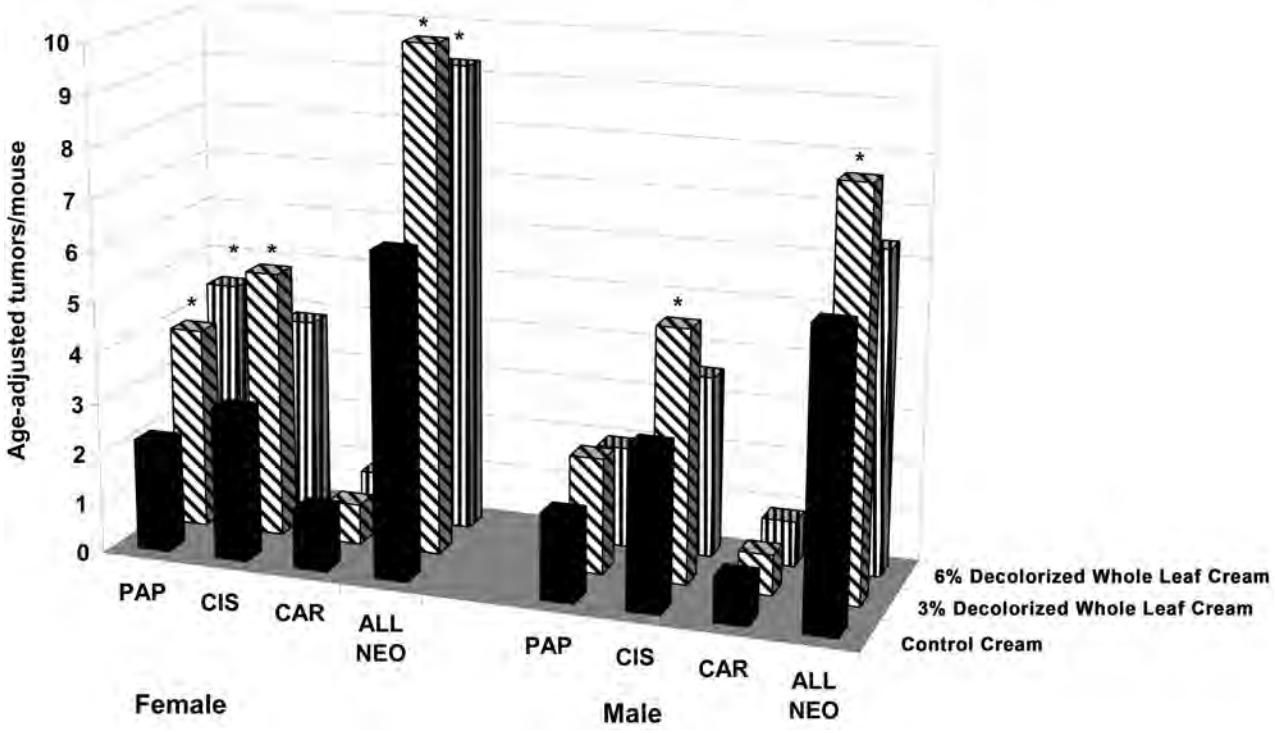


FIGURE 19
Comparisons of the Effects of Control Cream and Decolorized Whole Leaf Creams on Multiplicity of Squamous Cell Skin Neoplasms at the Site of Application in Mice Exposed to 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

* Asterisks above chart bars denote significant pairwise comparisons with the control cream group ($P \leq 0.05$).
Significant P values for dose-related trends are bolded.
PAP = papilloma; CIS = carcinoma *in situ;* CAR = carcinoma; ALL NEO = PAP + CIS + CAR

TABLE 23
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe-emodin Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Male			
Squamous Cell Papilloma			
Overall rate[a]	16/35 (45.7%)	27/35 (77.1%)	19/36 (52.8%)
Adjusted rate[b]	16/22.5 (71.0%)	27/29.4 (91.7%)	19/24.9 (76.2%)
Terminal rate[c]	0/0 (0.0%)	0/0 (0.0%)	1/1 (100.0%)
First incidence (days)	221	228	207
Poly-3 test (incidence)[d]	P = 0.425	P = 0.014*	P = 0.460
Overall mean lesion multiplicity (95% C.I.)	1.3 (0.6-2.0)	1.7 (1.2-2.2)	1.6 (1.0-2.3)
Age-adjusted lesion multiplicity (95% C.I.)	1.7 (1.2-2.5)	2.4 (1.7-3.5)	2.3 (1.6-3.2)
Poisson regression (multiplicity)[d]	P = 0.187	P = 0.130	P = 0.187
Squamous Cell Carcinoma *in situ*			
Overall rate	26/35 (74.3%)	24/35 (68.6%)	30/36 (83.3%)
Adjusted rate	26/29.0 (89.6%)	24/27.8 (86.3%)	30/32.2 (93.1%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/1 (0.0%)
First incidence (days)	235	228	207
Poly-3 test (incidence)	P = 0.361	P = 0.511N	P = 0.486
Overall mean lesion multiplicity (95% C.I.)	2.3 (1.6-2.9)	2.0 (1.5-2.5)	2.6 (2.0-3.1)
Age-adjusted lesion multiplicity (95% C.I.)	3.2 (2.5-4.1)	2.8 (2.1-3.6)	3.6 (2.8-4.6)
Poisson regression (multiplicity)	P = 0.288	P = 0.265N	P = 0.288
Squamous Cell Carcinoma			
Overall rate	15/35 (42.9%)	21/35 (60.0%)	23/36 (63.9%)
Adjusted rate	15/22.7 (66.1%)	21/26.3 (79.7%)	23/28.1 (82.0%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/1 (0.0%)
First incidence (days)	221	230	207
Poly-3 test (incidence)	P = 0.088	P = 0.175	P = 0.113
Overall mean lesion multiplicity (95% C.I.)	0.7 (0.4-0.9)	0.8 (0.6-1.0)	0.9 (0.7-1.1)
Age-adjusted lesion multiplicity (95% C.I.)	0.9 (0.7-1.3)	1.1 (0.8-1.4)	1.2 (0.9-1.6)
Poisson regression (multiplicity)	P = 0.128	P = 0.278	P = 0.128
Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma			
Overall rate	30/35 (85.7%)	28/35 (80.0%)	32/36 (88.9%)
Adjusted rate	30/31.4 (95.6%)	28/30.1 (93.2%)	32/33.7 (95.1%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	0/1 (0.0%)
First incidence (days)	221	228	207
Poly-3 test (incidence)	P = 0.605N	P = 0.578N	P = 0.723N
Overall mean lesion multiplicity (95% C.I.)	2.9 (2.2-3.7)	2.7 (2.2-3.3)	3.4 (2.8-4.1)
Age-adjusted lesion multiplicity (95% C.I.)	4.1 (3.3-5.1)	3.8 (3.1-4.8)	4.8 (4.0-5.9)
Poisson regression (multiplicity)	P = 0.185	P = 0.351N	P = 0.185

TABLE 23
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe-emodin Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Male (continued)			
Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma			
Overall rate	31/35 (88.6%)	30/35 (85.7%)	33/36 (91.7%)
Adjusted rate	31/32.1 (96.6%)	30/31.1 (96.3%)	33/33.7 (98.1%)
Terminal rate	0/0 (0.0%)	0/0 (0.0%)	1/1 (100.0%)
First incidence (days)	221	228	207
Poly-3 test (incidence)	P = 0.509	P = 0.787N	P = 0.704
Overall mean lesion multiplicity (95% C.I.)	4.2 (3.0-5.3)	4.5 (3.6-5.4)	5.1 (4.1-6.1)
Age-adjusted lesion multiplicity (95% C.I.)	5.8 (4.7-7.2)	6.3 (5.1-7.8)	7.1 (5.8-8.7)
Poisson regression (multiplicity)	P = 0.133	P = 0.335	P = 0.133
Female			
Squamous Cell Papilloma			
Overall rate	21/36 (58.3%)	27/36 (75.0%)	29/36 (80.6%)
Adjusted rate	21/26.5 (79.2%)	27/30.3 (89.1%)	29/31.1 (93.3%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	223	192
Poly-3 test (incidence)	P = 0.028*	P = 0.176	P = 0.049*
Overall mean lesion multiplicity (95% C.I.)	1.7 (1.1-2.2)	2.8 (2.1-3.4)	2.6 (1.8-3.3)
Age-adjusted lesion multiplicity (95% C.I.)	2.2 (1.6-2.9)	3.6 (2.8-4.7)	3.5 (2.7-4.5)
Poisson regression (multiplicity)	P = 0.029*	P = 0.018*	P = 0.029*
Squamous Cell Carcinoma *in situ*			
Overall rate	30/36 (83.3%)	27/36 (75.0%)	31/36 (86.1%)
Adjusted rate	30/31.9 (93.9%)	27/30.3 (89.1%)	31/31.5 (98.3%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	223	223
Poly-3 test (incidence)	P = 0.246	P = 0.369N	P = 0.356
Overall mean lesion multiplicity (95% C.I.)	2.3 (1.7-2.8)	2.4 (1.7-3.1)	3.1 (2.4-3.8)
Age-adjusted lesion multiplicity (95% C.I.)	3.0 (2.4-3.8)	3.2 (2.5-4.1)	4.2 (3.4-5.3)
Poisson regression (multiplicity)	P = 0.046*	P = 0.369	P = 0.046*

TABLE 23
Incidences and Multiplicities of Skin Neoplasms at the Site of Application in Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe-emodin Creams at 13.70 mJ • CIE/cm^2 SSL

	Control Cream	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Female (continued)			
Squamous Cell Carcinoma			
Overall rate	25/36 (69.4%)	22/36 (61.1%)	19/36 (52.8%)
Adjusted rate	25/29.8 (83.8%)	22/27.8 (79.0%)	19/25.6 (74.2%)
Terminal rate	1/2 (50.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	223	223
Poly-3 test (incidence)	P = 0.191N	P = 0.438N	P = 0.252N
Overall mean lesion multiplicity (95% C.I.)	0.9 (0.7-1.1)	0.8 (0.5-1.0)	0.9 (0.6-1.2)
Age-adjusted lesion multiplicity (95% C.I.)	1.2 (0.9-1.5)	1.0 (0.7-1.3)	1.2 (1.0-1.6)
Poisson regression (multiplicity)	P = 0.368	P = 0.280N	P = 0.368
Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma			
Overall rate	35/36 (97.2%)	32/36 (88.9%)	32/36 (88.9%)
Adjusted rate	35/35.0 (100.0%)	32/33.4 (95.8%)	32/32.2 (99.4%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	223	223
Poly-3 test (incidence)	P = 0.669N	P = 0.275N	P = 1.000N
Overall mean lesion multiplicity (95% C.I.)	3.1 (2.6-3.7)	3.2 (2.4-4.0)	4.0 (3.3-4.8)
Age-adjusted lesion multiplicity (95% C.I.)	4.2 (3.4-5.1)	4.2 (3.5-5.1)	5.5 (4.5-6.6)
Poisson regression (multiplicity)	P = 0.049*	P = 0.462	P = 0.049*
Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma			
Overall rate	35/36 (97.2%)	34/36 (94.4%)	33/36 (91.7%)
Adjusted rate	35/35.0 (100.0%)	34/34.6 (98.4%)	33/33.0 (99.9%)
Terminal rate	2/2 (100.0%)	0/0 (0.0%)	0/0 (0.0%)
First incidence (days)	208	223	192
Poly-3 test (incidence)	P = 0.912N	P = 0.860N	P = 1.000N
Overall mean lesion multiplicity (95% C.I.)	4.8 (3.9-5.7)	5.9 (4.7-7.1)	6.6 (5.3-7.9)
Age-adjusted lesion multiplicity (95% C.I.)	6.4 (5.3-7.7)	7.9 (6.6-9.4)	8.9 (7.5-10.6)
Poisson regression (multiplicity)	P = 0.015*	P = 0.087	P = 0.015*

* Significant at P≤0.05
C.I. = confidence interval
[a] Number of neoplasm-bearing animals/number of animals with skin examined microscopically
[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] P values for control cream represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to control. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. Negative trends and P values are appended with an N.

Female	Poisson Trend
Papilloma	**P=0.029**
Carcinoma *in situ*	**P=0.046**
Carcinoma	P=0.368
All Neoplasms	**P=0.015**

Male	Poisson Trend
Papilloma	P=0.187
Carcinoma *in situ*	P=0.288
Carcinoma	P=0.128
All Neoplasms	P=0.133

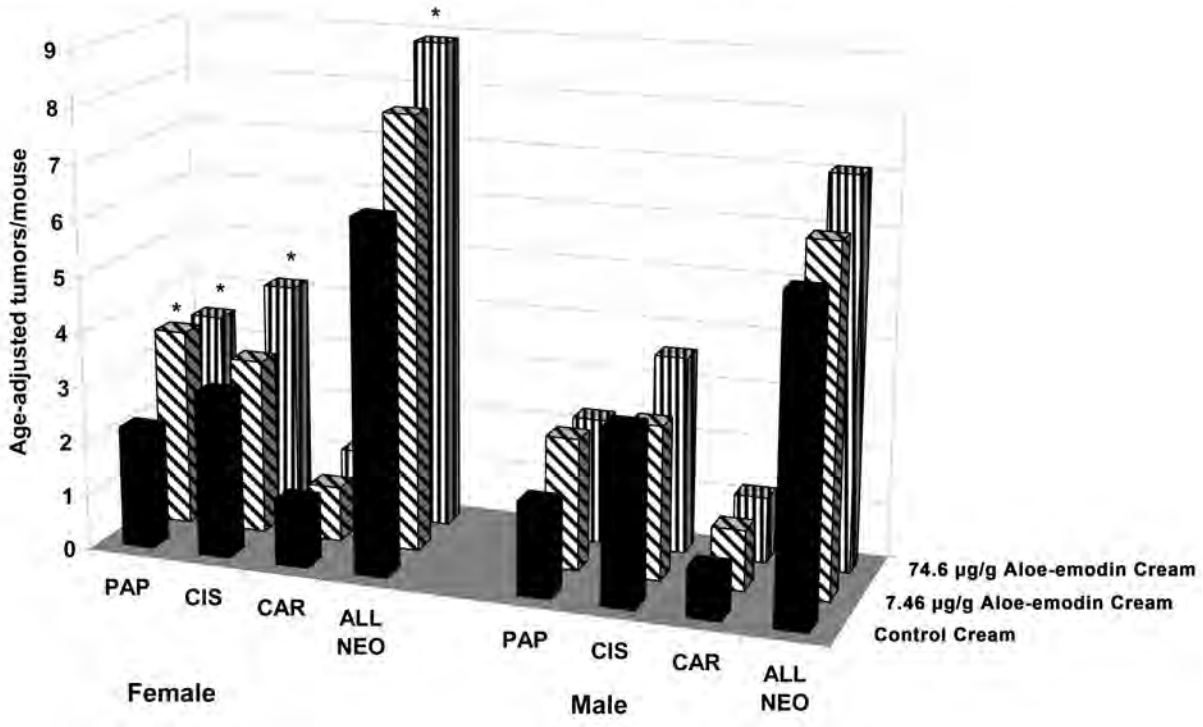


FIGURE 20
Comparisons of the Effects of Control Cream and Aloe-emodin Creams on Multiplicity of Squamous Cell Skin Neoplasms at the Site of Application in Mice Exposed to 13.70 mJ • CIE/cm^2 SSL in the 1-Year Simulated Solar Light Study of Aloe vera

* Asterisks above chart bars denote significant pairwise comparisons with the control cream group ($P \leq 0.05$). Significant P values for dose-related trends are bolded.
PAP = papilloma; CIS = carcinoma *in situ;* CAR = carcinoma; ALL NEO = PAP + CIS + CAR

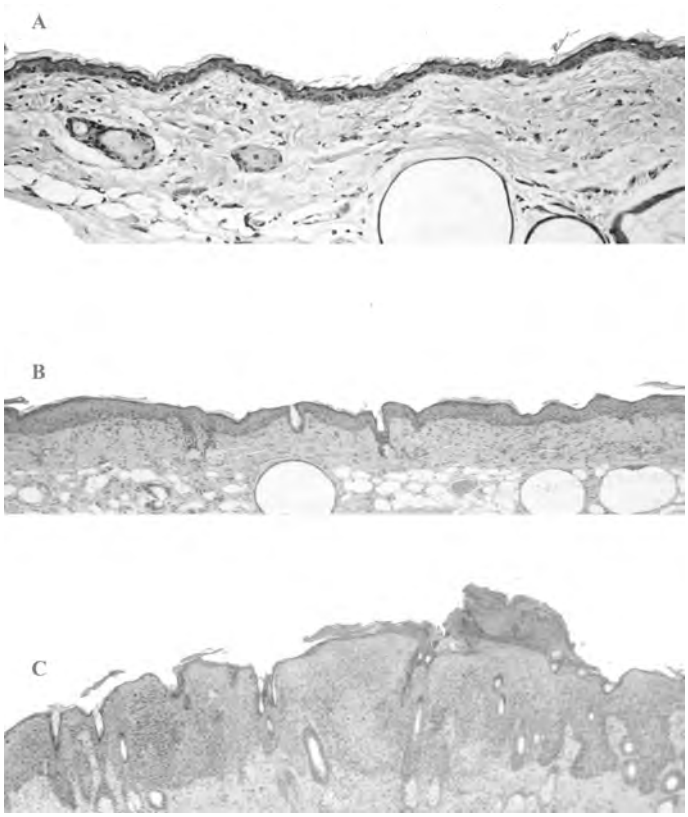


Plate 1
Panel A: normal mouse squamous cell epithelium. H&E
Panel B: mildly diffuse squamous cell hyperplasia of the epidermis. H&E
Panel C: marked focal atypical hyperplasia of the epidermis. H&E

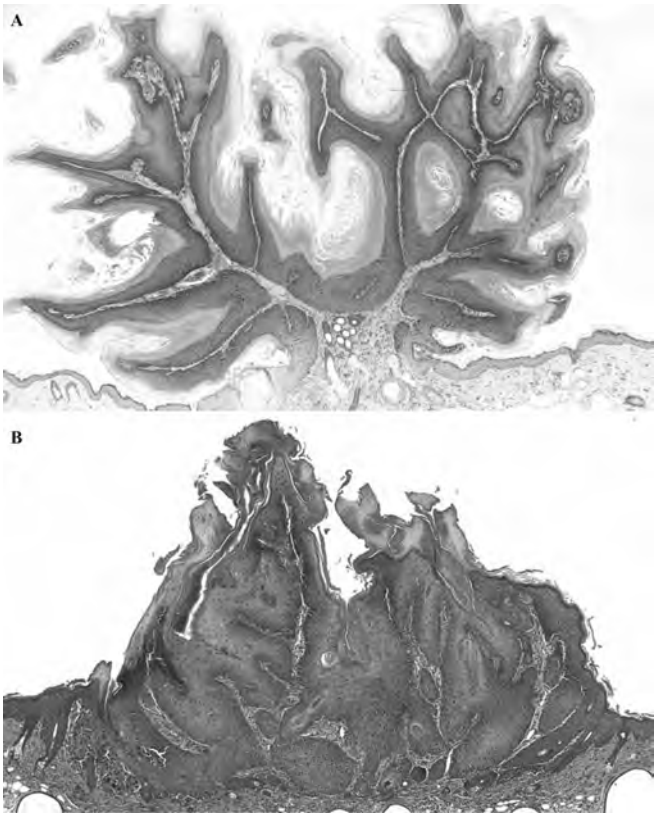


Plate 2
Panel A: pedunculated-based squamous cell papilloma of the epidermis. H&E
Panel B: sessile-based squamous cell papilloma of the epidermis. H&E

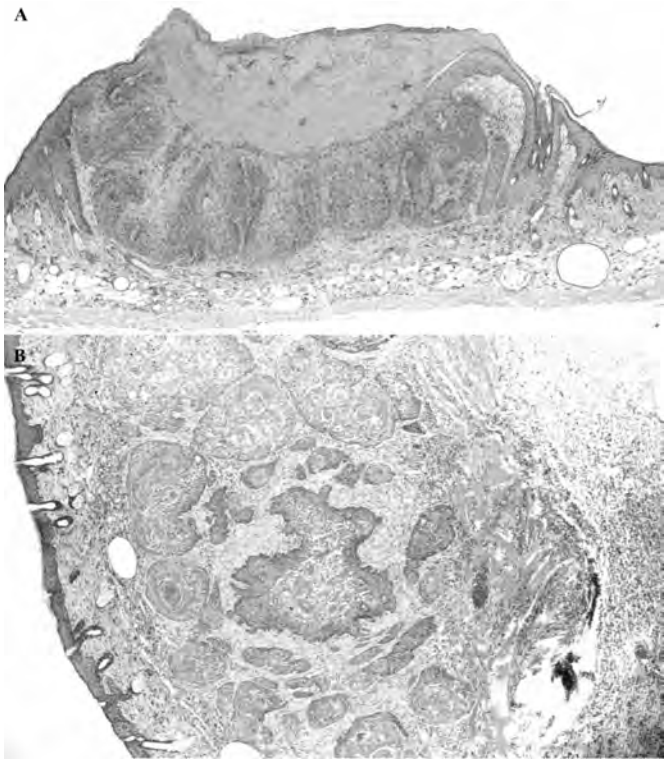


Plate 3
Panel A: squamous cell carcinoma *in situ* of the epidermis. H&E
Panel B: squamous cell carcinoma of the epidermis. H&E

DISCUSSION AND CONCLUSIONS

Squamous cell neoplasia is one of the most common human malignancies, representing one third of all new cancers diagnosed in the United States (Jemal *et al.*, 2007). The number of new cases diagnosed per year in the United States alone approaches one million and continues to rise. The primary pathogenic factor for the development of squamous cell neoplasia in the general population is overexposure to sunlight-derived ultraviolet (UV) radiation (Mukhtar and Elmets, 1996; Armstrong and Kricker, 2001). UV radiation may act as both an initiator and a promoter of skin cancer; therefore, UV light is considered a complete carcinogen.

The 1-year photococarcinogenesis study on Aloe vera was designed to test the hypothesis that the topical application of creams that contain extracts or components (in the case of aloe-emodin) of the Aloe vera plant would alter the process of simulated solar light (SSL)-induced photocarcinogenesis in SKH-1 hairless mice. In order to test this hypothesis, male and female SKH-1 mice were treated in the mornings, 5 days/week, with either control cream or a cream that contained one of the Aloe vera plant extracts (aloe gel, whole leaf, or decolorized whole leaf) or aloe-emodin. The mice were then exposed to SSL (0.00 or 13.70 mJ • CIE/cm^2) in the afternoons on the same days. This treatment regime continued for a period of 40 weeks and was followed by a 12-week observation/recovery period.

The Aloe vera extracts selected for study were obtained from the fresh whole leaves of the *Aloe barbadensis* Miller plant, commonly referred to as Aloe vera; however, each extract represented either a different portion of the plant leaf or a different extraction method. For example, the aloe gel was obtained from the inner parenchyma tissue of hand-filleted Aloe vera leaves. The whole leaf and the decolorized whole leaf extracts, which are two commonly used products obtained from commercial processing methods for Aloe vera, were obtained from fresh, whole Aloe vera leaves. The whole leaf extract represented the natural whole leaf juice and contained the leaf gel and leaf latex, while the decolorized whole leaf extract represented a cost-effective means of producing leaf gel without the labor-intensive process of hand-filleting the leaves. The decolorized whole leaf extract differed from both the whole leaf extract and aloe gel in that the activated charcoal removed the aloe latex found in the whole leaf extracts and also some of the native, high molecular weight polysaccharides found in aloe gel.

Several parameters (onset, incidence, and multiplicity) were evaluated during the in-life phase of the study and following the histopathology of mouse tissues to determine the effects of Aloe vera extracts or aloe-emodin on SSL-induced photocarcinogenesis. Skin lesion onset (the rate at which specified doses of UV radiation cause skin cancer under laboratory conditions) is the convention used by industry to determine whether a test substance alters SSL-induced photocarcinogenesis, with histologic confirmation not routinely performed for skin lesions observed during the in-life phase of photococarcinogenesis studies (Forbes *et al.*, 2003). Incidence is the percentage of animals within a particular treatment that have at least one detectable in-life skin lesion or histopathology-detected squamous cell nonneoplastic lesion or neoplasm. Multiplicity, on the other hand, is the total number of events (in-life skin lesion or the histopathology-detected squamous cell nonneoplastic lesion or neoplasm) per animal within a treatment group.

Skin lesion onset and multiplicity determined during in-life observations of mice and incidence, whether based on in-life observations or histopathology data, were not significantly different in Aloe vera or aloe-emodin treated groups when compared with the control cream groups. In contrast, the multiplicities of the combination of all squamous cell neoplasms (papilloma, carcinoma *in situ*, and carcinoma) were significantly increased in Aloe vera and aloe-emodin treated mice in pairwise comparisons with control cream animals, suggesting a photococarcinogenic-enhancing effect induced by each of the test articles.

The mice that did not receive cream and were exposed to SSL served as the experimental positive controls to confirm that the basic conditions of the experiment were able to produce a positive result (squamous cell neoplasms), even in the event that none of the mice that received the test substances produced a positive result. Because of

the design of the animal rack/cage system used in this study and the direction of the incidence of light from the SSL lamp source, the primary site of mouse skin exposure to SSL extended from the nape of the neck to the base of the tail and midway along both sides of the animal and was coincident with the site of application for the control, Aloe vera, and aloe-emodin creams.

The exposure of untreated (no-cream) male and female mice to increasing amounts of SSL resulted in exposure-dependent decreases in survival and increases in the Cox relative hazard ratios. Because body weight and morbidity did not differ significantly in mice exposed to SSL, the decreased survival was the result of removal of mice that had reached a protocol-defined limit on skin lesion size or skin lesion merging.

Concomitant with a dose-dependent decrease in survival, there was a significant SSL exposure-dependent decrease in the time to in-life detection of skin lesion onset and a significant increase in the incidences and multiplicities of in-life detected skin lesions. A previous photococarcinogenesis study of glycolic acid and salicyclic acid that was conducted in a similar manner and at the same test facility as the current study showed similar group median times to skin lesion onset for mice that received no cream treatment and were exposed to equivalent doses of SSL (NTP, 2007). In the previous study, untreated mice exposed to 6.85, 13.70, or 20.55 mJ • CIE/cm^2 had median times to skin lesion onset of >53.0, 35.5, or 24.0 weeks for males and >53.0, 33.0, or 24.0 weeks for females, respectively. In the current study, the median times to skin lesion onset for the untreated groups exposed to 6.85, 13.70, or 20.55 mJ • CIE/cm^2 of SSL were >53.0, 32.3, or 23.6 weeks for male mice and 48.3, 32.4, or 24.3 weeks for female mice, respectively. Other comparisons of in-life skin lesion data between these studies could not be conducted because in the previous study, in-life skin lesion incidence and multiplicity data were not reported for the untreated groups.

In general, incidence and multiplicity of squamous cell neoplasms would be expected to increase with increased exposure levels of SSL (Gallagher and Lee, 2006; Gloster and Neal, 2006; Faurschou *et al.*, 2007). Significant SSL exposure-related trends in the incidences and multiplicities of squamous cell neoplasms were observed in male and female mice that received no cream. In addition, with the exception of squamous cell carcinoma in mice exposed to 6.85 mJ • CIE/cm^2 SSL, pairwise comparisons indicated that the incidences of squamous neoplasms (squamous cell papilloma, squamous cell carcinoma *in situ*, and squamous cell carcinoma) were significantly increased at each of the SSL exposure levels. The previous photococarcinogenesis study with glycolic acid and salicyclic acid showed similar trends in the induction of squamous cell neoplasms in mice that received no cream. The major differences between the two studies was that in the current study, the incidences of squamous cell carcinoma tended to be lower, while the incidences of squamous cell papilloma and squamous cell carcinoma *in situ* tended to be higher. Cutaneous squamous cell carcinoma is a larger, more invasive lesion than squamous cell papilloma or squamous cell carcinoma *in situ* (Utikal *et al.*, 2007; Veness, 2007). In the current study, mice were removed when the size of lesions reached 5 mm to reduce the problem of merging of large lesions, whereas in the glycolic and salicyclic acid study, lesions were allowed to reach 10 mm. It is likely that the lower incidences of squamous cell carcinoma in the current study were due to this earlier removal of animals from the study.

Epidemiologic and experimental studies have demonstrated that increased incidences of skin cancer are the result of increased exposure to solar UV radiation (Bush *et al.*, 1999). Furthermore, since light sources that contain UVB are carcinogenic (IARC, 1992), significant positive exposure-related trends in incidences and multiplicities of squamous cell neoplasms were anticipated for mice that received SSL exposure and no cream. Nevertheless, confirmation of these results was necessary to establish an exposure trend with SSL as a comparison tool for the study design – to test the effects of topical application of Aloe vera plant extracts on the photocarcinogenesis of SSL.

The efficiency, tolerability, and applicability of topical agents are directly related to employed vehicles. Common vehicles are complex mixtures consisting of diverse ingredients that belong to six major groups: hydrophilic and lipophilic bases, emulsifiers, gel-forming agents, preservatives, and antioxidants (Daniels and Knie, 2007). Ideally, topical control creams should demonstrate the baseline results obtained when a test substance does not produce a measurable positive result. Data collected from the hairless mouse model have indicated that the properties of a control cream may, at times, shorten the time to tumor formation and/or lower the minimum dose of radiation required to induce erythema, also known as the MED (Jacobs *et al.*, 2004). In the current study, the topical application of the oil-in-water

emulsion-type control cream had no effect on the survival of male or female mice either in the absence or presence of SSL when compared with the respective untreated control animals. Additionally, neither body weight nor the in-life detection of skin lesion onset, incidence, or multiplicity were significantly affected by the topical application of the control cream in comparison tests with the untreated control animals at the same level of SSL. With the exception of a significantly increased incidence of squamous cell hyperplasia (a diffuse thickening of the epidermis), the application of the control cream also had no significant effect on the histopathology-detected incidences of skin neoplasms or nonneoplastic skin lesions at the site of application in male or female mice, either in the absence of SSL or in the presence of 13.70 mJ • CIE/cm^2 of SSL, when compared with their respective untreated controls. Similarly, the multiplicities of skin neoplasms or nonneoplastic skin lesions were not affected by the application of control cream in male or female mice when compared with their respective untreated controls. *In toto*, these results suggest that the control cream used in this study did not impart a measurable effect when compared with the group that received no cream treatment and the same dose of SSL.

Aloe vera gel is most popularly recognized as a topical agent in the treatment of burns and insect bites; however, the topical application of Aloe vera gel has been reported to cause contact dermatitis, erythema, and photodermatitis (Hunter and Frumkin, 1991; Ernst, 2000; Mascolo *et al.*, 2004). In the photococarcinogenesis study described here, the inclusion of the aloe gel in topical creams at amounts up to 6% (w/w) had no deleterious effect on the survival and body weights of SKH-1 mice, and the in-life skin lesion onset was similar to that of the control cream mice. However, the histopathology enumeration and statistical analysis of the multiplicity of squamous cell neoplasms and nonneoplastic lesions revealed significant dose-related increases in the numbers of squamous cell papilloma and the total number of squamous cell neoplasms and a significant negative dose trend in the multiplicities of focal atypical squamous hyperplasia in female mice treated with the aloe gel creams. Furthermore, the multiplicities of squamous cell papilloma, squamous cell carcinoma *in situ*, the combination of all carcinomas, and all squamous cell neoplasms were significantly increased in female mice treated with the 3% aloe gel cream when compared to control cream mice. Only the multiplicity of squamous cell carcinoma was unaffected by the treatment of female mice with the 3% aloe gel cream. As in the case of mice that received no cream treatment and were exposed to SSL, the lower numbers of squamous cell carcinomas may have resulted from removal of animals from the study before greater numbers of squamous cell carcinomas could develop. On the other hand, the lower multiplicities of focal atypical hyperplasia may represent a progression of this nonneoplastic lesion type to a neoplasm. Nevertheless, the topical application of aloe gel induced a significantly increased multiplicity of squamous cell neoplasms when compared with the control cream. Squamous cell neoplasms or nonneoplastic lesions in aloe gel treated mice that were not exposed to SSL were either not detected or did not differ from the comparable measurements made in control cream animals.

As with the aloe gel, the survival, body weights, and in-life skin lesion onset of SKH-1 mice were not affected by the incorporation of the whole leaf extract into the topical creams used in these studies. The statistical results on the multiplicities of squamous cell neoplasms and nonneoplastic lesions showed significantly increased numbers and significant dose-related trends in the multiplicities of pathology-detected squamous cell papilloma in male and female mice treated with the whole leaf creams, while a significant negative trend and lower numbers of focal atypical hyperplasia were observed in female mice. When all neoplasm types were considered together, significantly increased numbers and significant dose-related trends were observed in male mice. In female mice treated with 3% whole leaf creams, significantly greater than control levels were shown in the multiplicities of squamous cell carcinoma *in situ* and in the combinations of all squamous cell carcinomas and all squamous cell neoplasms. As the multiplicities of squamous cell carcinoma were not different from control cream levels in male or female mice, the significant differences in the multiplicities of all squamous cell carcinomas in female mice treated with the 3% whole leaf extract cream and of the total of all neoplasms in male mice treated with the 6% whole leaf extract cream were likely a reflection of the significantly increased numbers of squamous cell carcinoma *in situ* in the female mice and the significantly increased numbers of squamous cell papilloma in male mice.

The aloe gel and whole leaf extracts used in these studies differed primarily in the content of leaf latex, which

is the sole source of the Aloe vera plant-derived anthraquinones. The leaf latex resides along the margins of the Aloe vera leaf, and the aloe gel was obtained from the inner parenchyma of the leaf pulp, which is devoid of the leaf latex. On the other hand, the whole leaf extract was produced from the whole leaf and contained both the leaf pulp and the leaf latex. The 3% and 6% aloe gel creams contained approximately 81 and 162 mg/kg of aloin, respectively; whereas, the 3% and 6% whole leaf creams contained more than a sixfold higher concentration of aloin. The similarity in the findings of this study between mice treated with the aloe gel and those treated with the whole leaf extract suggested that the latex, and especially the anthraquinone components, did not have a major impact on the induction of squamous cell neoplasms by SSL in the SKH-1 mouse.

As with the aloe gel and whole leaf extracts of the Aloe vera plant, creams that contained the decolorized whole leaf extract had no apparent effect on the survival, body weights, or in-life skin lesion onset of male or female mice. The histopathology enumeration and statistical analysis of the multiplicity of squamous cell neoplasms similarly revealed significant dose-related increases in the numbers of squamous cell papilloma and in the total number of squamous cell neoplasms in female mice treated with the decolorized whole leaf creams. In addition, the numbers of squamous cell papilloma, squamous cell carcinoma *in situ*, the total number of all carcinomas, and the total number of squamous cell neoplasms were significantly increased, and the number of focal atypical hyperplasia was decreased in female mice treated with the 3% decolorized whole leaf cream in comparison tests with control cream mice. These results were very consistent with the results detected in mice treated with the aloe gel and the whole leaf creams. As previously observed, only the multiplicity of squamous cell carcinoma was unaffected by the treatment of female mice with the 3% decolorized whole leaf cream, likely reflecting the larger size of this neoplasm type and the removal of animals from the study before greater numbers of the larger squamous cell carcinomas could develop.

Unlike male mice treated with the aloe gel and whole leaf extracts, male mice treated with the decolorized whole leaf cream displayed a similar profile as female mice in the multiplicities of squamous cell neoplasms. Male mice treated with the 3% decolorized whole leaf extract showed significantly increased multiplicity of squamous cell carcinoma *in situ* and significantly increased numbers of all squamous carcinomas and total squamous cell neoplasms when compared with control cream mice. The higher numbers of all squamous cell carcinomas and all squamous cell neoplasms apparently mirrored the multiplicity of squamous cell carcinoma *in situ*, since the numbers of squamous cell carcinoma and squamous cell papilloma in male mice were not significantly different from the control cream mice.

Similar to the whole leaf extract, the decolorized whole leaf extract was produced from the whole leaf of the Aloe vera plant. However, because the decolorized whole leaf extract was treated with activated charcoal, the leaf latex components were removed, and the resulting product was more similar to the aloe gel extract than it was to the whole leaf extract. The results of the study from the topical application of the decolorized whole leaf extract reflected similarly the results from the aloe gel and whole leaf treatments, suggesting that components other than those in the plant latex contributed to the significantly increased multiplicities of squamous cell neoplasms compared to those of the controls.

Aloe-emodin is an aglycone anthraquinone naturally present in the leaves of Aloe vera, and its presence in the Aloe vera plant is thought to result from the oxidation of aloin, the principal anthraquinone glycoside in the Aloe vera latex, rather than via direct endogenous synthesis. Some studies have shown that aloe-emodin is phototoxic and, potentially, photocarcinogenic (CIR, 2004). *In vitro* studies on the photobiological and photochemical properties of aloin and aloe-emodin found that significant photooxidative damage to both RNA and DNA was associated with the phototoxicity induced by aloe-emodin (Wamer *et al.*, 2003). Vargas *et al.* (2002) exposed human red blood cells to aloe-emodin and irradiated the cells with UVA. Aloe-emodin caused cell lysis (>50%) within 70 minutes after irradiation; no hemolysis occurred in the dark controls. The free radical scavengers reduced glutathione and superoxide dismutase had little protective effect on the photohemolytic process. Photoperoxidation and significant amounts of hydroperoxides were formed in irradiated aloe-emodin/linoleic acid in a phosphate-buffered solution (Vargas *et al.*, 2002). Aloe-emodin was found to generate singlet oxygen efficiently when irradiated with UV light (Vath *et al.*, 2002). Furthermore, the application of ethanol and aloe-emodin combined with the exposure of mice to UVB irradiation for 33 weeks was shown to cause mutations in the p53 gene; whereas in the absence of UV irradiation, mice failed to develop tumors

or p53 gene mutations (Badgwell *et al.*, 2004). Although aloe-emodin is not technically an extract of the Aloe vera plant, aloe-emodin was included as a component of these studies, since based on the literature, as noted above, it was predicted that aloe-emodin may have photococarcinogenic properties *in vivo*.

As with the Aloe vera plant extracts, aloe-emodin had no significant effect on the survival, body weights, or skin lesion onset of mice when compared with mice treated with the control cream. Similar to the other Aloe vera treatments, the histopathology enumeration and Poisson regression statistical analysis of the multiplicity of squamous cell neoplasms detected significantly greater than control and dose-related increases in the numbers of squamous cell papilloma, squamous cell carcinoma *in situ*, all squamous cell carcinomas, and the total number of squamous cell neoplasms in female mice treated with the 74.6 µg/g aloe-emodin cream and significantly greater than control numbers of squamous cell papilloma in female mice treated with the 7.4 µg/g aloe-emodin cream.

Aloe-emodin was incorporated into the creams in this study as a single compound rather than as a complex mixture as with the Aloe vera extracts. *In vitro* studies on the photobiological and photochemical properties of anthraquinones from the Aloe vera plant showed that aloe-emodin, the aglycone of aloin, is cytotoxic and generates reactive oxygen species and stable photoproducts when irradiated with UV light (Vargas *et al.*, 2002; Vath *et al.*, 2002; Wamer *et al.*, 2003). Still, results were similar between the aloe-emodin treatment and the Aloe vera extract treatments. These results are perplexing, since the Aloe vera extracts that contained the aloe latex, in particular the whole leaf extract, and those that lacked or had only trace amounts of latex, in particular the aloe gel and decolorized whole leaf extracts, induced similar effects as the aloe-emodin component alone.

Aloe vera is widely regarded as a therapeutic dermatologic agent, especially in the treatment of burns and wounds (Heggers *et al.*, 1996). The popular recognition of Aloe vera as a dermatologic agent has led to the widespread incorporation of Aloe vera extracts in healthcare and cosmetic products (Strickland and Pelley, 2004). Scientific evidence for the efficacy of Aloe vera is limited, and studies that use commercial preparations have been difficult to reproduce. One factor that contributes to the lack of scientific evidence for the efficacy of Aloe vera is undoubtedly the lack of standardized procedures for the harvesting, handling, and processing of Aloe vera. As stated previously, the Aloe vera extracts used in this study were handled and processed under very strict guidelines. Each of the plant extracts and aloe-emodin produced consistent, yet quite similar, photococarcinogenesis-enhancing effects, especially in female SKH-1 mice. These effects included increased multiplicity of pathology-defined squamous cell neoplasms.

CONCLUSIONS

These experiments investigated the potential of topical application of creams containing extracts of *Aloe barbadensis* Miller plant (aloe gel, whole leaf, or decolorized whole leaf) or aloe-emodin to alter the photocarcinogenic activity of filtered xenon arc simulated solar light (SSL) in male and female SKH-1 hairless mice. Data on skin lesions were collected both on digital images during the in-life phase and by histopathologic evaluation at necropsy. No effects of creams upon SSL-induced skin lesions were identified from data collected during the in-life phase.

Aloe Gel or Aloe-emodin

Under the conditions of these studies, there was a weak enhancing effect of aloe gel or aloe-emodin on the photocarcinogenic activity of SSL in female but not in male SKH-1 mice based on an increase in the multiplicity of histopathologically determined squamous cell neoplasms.

Aloe Whole Leaf or Decolorized Whole Leaf

Under the conditions of these studies, there was a weak enhancing effect of aloe whole leaf or decolorized whole leaf on the photocarcinogenic activity of SSL in both male and female SKH-1 mice based on an increase in the multiplicity of histopathologically determined squamous cell neoplasms.

A summary of the Technical Reports Review Subcommittee comments and the public discussion on this Technical Report appears on page 12.

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APPENDIX A
SUMMARY OF LESIONS IN MALE MICE IN THE 1-YEAR SIMULATED SOLAR LIGHT STUDY OF ALOE VERA

TABLE A1a
Summary of the Incidence of Neoplasms in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 0.00 mJ • CIE/cm^2 SSL[a]

	No Cream	Control Cream	6% Aloe Gel	6% Whole Leaf	6% Decolorized Whole Leaf	74.6 µg/g Aloe-emodin
Disposition Summary						
Animals initially in study	36	36	36	36	36	36
Early deaths						
Accidental death			1			
Moribund		5	1	4	3	7
Natural deaths	1			1	2	
Skin lesion greater than 5 mm					1	1
Survivors						
Died last week of study				1		
Terminal sacrifice	35	30	34	30	30	27
Missexed		1				1
Animals examined microscopically	36	35	36	35[b]	36	35
Integumentary System						
Skin, control	(36)	(35)	(36)	(35)	(36)	(35)
Skin, site of application	(36)	(35)	(36)	(35)	(36)	(35)
Sebaceous gland, adenoma, one			1 (3%)			
Systems Examined with No Neoplasms Observed						
Alimentary System						
Cardiovascular System						
Endocrine System						
General Body System						
Genital System						
Hematopoietic System						
Musculoskeletal System						
Nervous System						
Respiratory System						
Special Senses System						
Urinary System						
Neoplasm Summary						
Total animals with primary neoplasms[c]			1			
Total primary neoplasms			1			
Total animals with benign neoplasms			1			
Total benign neoplasms			1			

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm. The number given with the neoplasm name indicates the number of that neoplasm per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.
[b] One animal autolyzed, and no tissue was taken.
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE A1b
Summary of the Incidence of Neoplasms in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 6.85 mJ • CIE/cm^2 SSL[a]

	No Cream	
Disposition Summary		
Animals initially in study	36	
Early deaths		
Accidental death	1	
Moribund	5	
Natural deaths	2	
Skin lesion greater than 5 mm	5	
Survivors		
Died last week of study	1	
Terminal sacrifice	22	
Animals examined microscopically	36	
Integumentary System		
Skin, control	(36)	
Squamous cell papilloma, one	1	(3%)
Skin, site of application	(36)	
Lymphoma malignant	1	(3%)
Squamous cell carcinoma *in situ*, one	5	(14%)
Squamous cell carcinoma *in situ*, two	1	(3%)
Squamous cell carcinoma *in situ*, greater than five	1	(3%)
Squamous cell papilloma, one	2	(6%)
Squamous cell papilloma, two	2	(6%)
Squamous cell papilloma, four	1	(3%)
Sebaceous gland, adenoma, one	1	(3%)
Systemic Lesions		
Multiple organs[b]		(36)
Lymphoma malignant	1	(3%)
Systems Examined with No Neoplasms Observed		
Alimentary System		
Cardiovascular System		
Endocrine System		
General Body System		
Genital System		
Hematopoietic System		
Musculoskeletal System		
Nervous System		
Respiratory System		
Special Senses System		
Urinary System		
Neoplasm Summary		
Total animals with primary neoplasms[c]	10	
Total primary neoplasms	15	
Total animals with benign neoplasms	5	
Total benign neoplasms	7	
Total animals with malignant neoplasms	8	
Total malignant neoplasms	8	

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm. The number given with the neoplasm name indicates the number of that neoplasm per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.
[b] Number of animals with any tissue examined microscopically
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE A1c
Summary of the Incidence of Neoplasms in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL[a]

	No Cream	Control Cream	3% Aloe Gel	6% Aloe Gel	3% Whole Leaf
Disposition Summary					
Animals initially in study	36	36	36	36	36
Early deaths					
Moribund		3	5	1	1
Natural deaths				1	2
Skin lesion greater than 5 mm	36	31	31	35	32
Survivors					
Terminal sacrifice					1
Missexed					
Animals examined microscopically	36	35[b]	36	36	36
Integumentary System					
Skin, control	(36)	(35)	(36)	(36)	(36)
Squamous cell carcinoma, one					
Squamous cell papilloma, one					
Skin, site of application	(36)	(35)	(36)	(36)	(36)
Basal cell carcinoma, one					
Fibrosarcoma	1 (3%)				
Lymphoma malignant					
Squamous cell carcinoma, one	13 (36%)	9 (26%)	11 (31%)	15 (42%)	12 (33%)
Squamous cell carcinoma, two	5 (14%)	4 (11%)	5 (14%)	4 (11%)	5 (14%)
Squamous cell carcinoma, three	1 (3%)	2 (6%)	2 (6%)		
Squamous cell carcinoma, four				1 (3%)	
Squamous cell carcinoma in situ, one	11 (31%)	8 (23%)	6 (17%)	3 (8%)	10 (28%)
Squamous cell carcinoma in situ, two	7 (19%)	6 (17%)	7 (19%)	7 (19%)	7 (19%)
Squamous cell carcinoma in situ, three	3 (8%)	5 (14%)	3 (8%)	8 (22%)	4 (11%
Squamous cell carcinoma in situ, four	2 (6%)	1 (3%)	5 (14%)	4 (11%)	2 (6%)
Squamous cell carcinoma in situ, five	3 (8%)	1 (3%)	4 (11%)	3 (8%)	1 (3%)
Squamous cell carcinoma in situ, greater than five	5 (14%)	5 (14%)	3 (8%)	4 (11%)	5 (14%)
Squamous cell papilloma, one	14 (39%)	7 (20%)	7 (19%)	10 (28%)	6 (17%)
Squamous cell papilloma, two	5 (14%)	3 (9%)	4 (11%)	5 (14%)	7 (19%)
Squamous cell papilloma, three	1 (3%)	3 (9%)	1 (3%)	5 (14%)	1 (3%)
Squamous cell papilloma, four	1 (3%)	1 (3%)	1 (3%)	4 (11%)	1 (3%)
Squamous cell papilloma, five		1 (3%)	2 (6%)		2 (6%)
Squamous cell papilloma, greater than five	1 (3%)	1 (3%)	4 (11%)	1 (3%)	3 (8%)
Systemic Lesions					
Multiple organs[c]	(36)	(35)	(36)	(36)	(36)
Lymphoma malignant					

TABLE A1c
Summary of the Incidence of Neoplasms in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	6% Whole Leaf	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Disposition Summary					
Animals initially in study	36	36	36	36	36
Early deaths					
Moribund	2	2	4	3	
Natural deaths	1	1		2	1
Skin lesion greater than 5 mm	32	33	32	30	34
Survivors					
Terminal sacrifice	1				1
Missexed				1	
Animals examined microscopically	36	36	36	35	36
Integumentary System					
Skin, control	(36)	(36)	(36)	(34)	(36)
Squamous cell carcinoma, one		1 (3%)			
Squamous cell papilloma, one			1 (3%)		1 (3%)
Skin, site of application	(36)	(36)	(36)	(35)	(36)
Basal cell carcinoma, one					1 (3%)
Fibrosarcoma					
Lymphoma malignant	1 (3%)		1 (3%)	1 (3%)	
Squamous cell carcinoma, one	9 (25%)	9 (25%)	18 (50%)	16 (46%)	14 (39%)
Squamous cell carcinoma, two	5 (14%)	4 (11%)	1 (3%)	4 (11%)	9 (25%)
Squamous cell carcinoma, three	1 (3%)	1 (3%)	1 (3%)	1 (3%)	
Squamous cell carcinoma, four					
Squamous cell carcinoma in situ, one	2 (6%)	5 (14%)	4 (11%)	4 (11%)	5 (14%)
Squamous cell carcinoma in situ, two	7 (19%)	3 (8%)	8 (22%)	7 (20%)	8 (22%)
Squamous cell carcinoma in situ, three	2 (6%)	4 (11%)	1 (3%)	5 (14%)	6 (17%)
Squamous cell carcinoma in situ, four	7 (19%)	4 (11%)	6 (17%	5 (14%)	7 (19%)
Squamous cell carcinoma in situ, five	4 (11%)	5 (14%)	2 (6%)	2 (6%)	2 (6%)
Squamous cell carcinoma in situ, greater than five	6 (17%)	8 (22%)	5 (14%)	1 (3%)	2 (6%)
Squamous cell papilloma, one	6 (17%)	5 (14%)	4 (11%)	13 (37%)	5 (14%)
Squamous cell papilloma, two	10 (28%)	7 (19%)	7 (19%)	5 (14%)	4 (11%)
Squamous cell papilloma, three	2 (6%)	2 (6%)	5 (14%)	4 (11%)	4 (11%)
Squamous cell papilloma, four	3 (8%)		2 (6%)	2 (6%)	3 (8%)
Squamous cell papilloma, five	1 (3%)	1 (3%)	1 (3%)	1 (3%)	
Squamous cell papilloma, greater than five	5 (14%)	4 (11%)	1 (3%)	2 (6%)	3 (8%)
Systemic Lesions					
Multiple organs	(36)	(36)	(36)	(35)	(36)
Lymphoma malignant	1 (3%)		1 (3%)	1 (3%)	

TABLE A1c
Summary of the Incidence of Neoplasms in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	No Cream	Control Cream	3% Aloe Gel	6% Aloe Gel	3% Whole Leaf
Systems Examined with No Neoplasms Observed					
Alimentary System					
Cardiovascular System					
Endocrine System					
General Body System					
Genital System					
Hematopoietic System					
Musculoskeletal System					
Nervous System					
Respiratory System					
Special Senses System					
Urinary System					
Neoplasm Summary					
Total animals with primary neoplasms[d]	36	31	31	36	32
Total primary neoplasms	73	57	65	74	66
Total animals with benign neoplasms	22	16	19	25	20
Total benign neoplasms	22	16	19	25	20
Total animals with malignant neoplasms	36	30	31	34	32
Total malignant neoplasms	51	41	46	49	46

TABLE A1c
Summary of the Incidence of Neoplasms in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	6% Whole Leaf	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Systems Examined with No Neoplasms Observed					
Alimentary System					
Cardiovascular System					
Endocrine System					
General Body System					
Genital System					
Hematopoietic System					
Musculoskeletal System					
Nervous System					
Respiratory System					
Special Senses System					
Urinary System					
Neoplasm Summary					
Total animals with primary neoplasms	33	33	32	31	33
Total primary neoplasms	71	63	68	73	74
Total animals with benign neoplasms	27	19	20	27	19
Total benign neoplasms	27	19	21	27	20
Total animals with malignant neoplasms	33	31	30	29	32
Total malignant neoplasms	44	44	47	46	54

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm. The number given with the neoplasm name indicates the number of that neoplasm per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.
[b] One animal died prior to dosing and was discarded.
[c] Number of animals with any tissue examined microscopically
[d] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE A1d
Summary of the Incidence of Neoplasms in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 20.55 mJ • CIE/cm^2 SSL[a]

	No Cream
Disposition Summary	
Animals initially in study	36
Early deaths	
Moribund	3
Skin lesion greater than 5 mm	33
Animals examined microscopically	36
Integumentary System	
Skin, control	(36)
Squamous cell papilloma, one	1 (3%)
Subcutaneous tissue, mast cell tumor malignant	1 (3%)
Skin, site of application	(36)
Squamous cell carcinoma, one	10 (28%)
Squamous cell carcinoma, two	10 (28%)
Squamous cell carcinoma, three	5 (14%)
Squamous cell carcinoma, greater than five	1 (3%)
Squamous cell carcinoma *in situ,* one	2 (6%)
Squamous cell carcinoma *in situ,* two	8 (22%)
Squamous cell carcinoma *in situ,* three	2 (6%)
Squamous cell carcinoma *in situ,* four	7 (19%)
Squamous cell carcinoma *in situ,* five	3 (8%)
Squamous cell carcinoma *in situ,* greater than five	5 (14%)
Squamous cell papilloma, one	5 (14%)
Squamous cell papilloma, two	3 (8%)
Squamous cell papilloma, three	2 (6%)
Systems Examined with No Neoplasms Observed	
Alimentary System	
Cardiovascular System	
Endocrine System	
General Body System	
Genital System	
Hematopoietic System	
Musculoskeletal System	
Nervous System	
Respiratory System	
Special Senses System	
Urinary System	
Neoplasm Summary	
Total animals with primary neoplasms[b]	33
Total primary neoplasms	65
Total animals with benign neoplasms	11
Total benign neoplasms	11
Total animals with malignant neoplasms	33
Total malignant neoplasms	54

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm. The number given with the neoplasm name indicates the number of that neoplasm per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.
[b] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE A2a
Statistical Analysis of Primary Neoplasms in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

	0.00 mJ • CIE/cm^2	6.85 mJ • CIE/cm^2	13.70 mJ • CIE/cm^2	20.55 mJ • CIE/cm^2
Skin (Site of Application): Squamous Cell Papilloma				
Overall rate[a]	0/36 (0.0%)	5/36 (13.9%)	22/36 (61.1%)	10/36 (27.8%)
Adjusted rate[b]	0/35.7 (0.0%)	5/29.1 (17.2%)	22/27.8 (79.2%)	10/14.6 (68.5%)
Terminal rate[c]	0/35 (0.0%)	3/22 (13.6%)	0/0	0/0
First incidence (days)	—[e]	327	235	200
Poly-3 test[d]	P≤0.001	P = 0.015	P≤0.001	P≤0.001
Skin (Site of Application): Squamous Cell Carcinoma *in situ*				
Overall rate	0/36 (0.0%)	7/36 (19.4%)	31/36 (86.1%)	27/36 (75.0%)
Adjusted rate	0/35.7 (0.0%)	7/29.3 (23.9%)	31/33.0 (93.8%)	27/28.6 (94.5%)
Terminal rate	0/35 (0.0%)	3/22 (13.6%)	0/0	0/0
First incidence (days)	—	327	235	181
Poly-3 test	P≤0.001	P = 0.002	P≤0.001	P≤0.001
Skin (Site of Application): Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	0/36 (0.0%)	19/36 (52.8%)	26/36 (72.2%)
Adjusted rate	0/35.7 (0.0%)	0/28.5 (0.0%)	19/26.5 (71.7%)	26/27.9 (93.3%)
Terminal rate	0/35 (0.0%)	0/22 (0.0%)	0/0	0/0
First incidence (days)	—	—	235	181
Poly-3 test	P≤0.001	—[f]	P≤0.001	P≤0.001
Skin (Site of Application): Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	7/36 (19.4%)	35/36 (97.2%)	33/36 (91.7%)
Adjusted rate	0/35.7 (0.0%)	7/29.3 (23.9%)	35/35.2 (99.4%)	33/33.3 (99.2%)
Terminal rate	0/35 (0.0%)	3/22 (13.6%)	0/0	0/0
First incidence (days)	—	327	235	181
Poly-3 test	P≤0.001	P = 0.002	P≤0.001	P≤0.001
Skin (Site of Application): Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	9/36 (25.0%)	35/36 (97.2%)	33/36 (91.7%)
Adjusted rate	0/35.7 (0.0%)	9/29.3 (30.7%)	35/35.2 (99.4%)	33/33.3 (99.2%)
Terminal rate	0/35 (0.0%)	5/22 (22.7%)	0/0	0/0
First incidence (days)	—	327	235	181
Poly-3 test	P≤0.001	P≤0.001	P≤0.001	P≤0.001

[a] Number of neoplasm-bearing animals/number of animals with skin examined microscopically
[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] P values for the control group (0.00 mJ • CIE/cm^2) represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to the control group. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice.
[e] Not applicable; no neoplasms in animal group
[f] Value of statistic cannot be computed.

TABLE A2b
Statistical Analysis of Primary Neoplasms in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	No Cream	Control Cream	3% Aloe Gel	6% Aloe Gel	3% Whole Leaf
Skin (Site of Application): Squamous Cell Papilloma					
Overall rate[a]	22/36 (61.1%)	16/35 (45.7%)	19/36 (52.8%)	25/36 (69.4%)	20/36 (55.6%)
Adjusted rate[b]	22/27.8 (79.2%)	16/22.5 (71.0%)	19/24.1 (78.8%)	25/28.9 (86.4%)	20/25.3 (79.2%)
Terminal rate[c]	0/0	0/0	0/0	0/0	1/1 (100.0%)
First incidence (days)	235	221	228	214	242
Poly-3 trend test[d]			P = 0.056		P = 0.018
Poly-3 vehicle pairwise comparison[e]	P = 0.331N		P = 0.357	P = 0.090	P = 0.341
Skin (Site of Application): Squamous Cell Carcinoma *in situ*					
Overall rate	31/36 (86.1%)	26/35 (74.3%)	28/36 (77.8%)	29/36 (80.6%)	29/36 (80.6%)
Adjusted rate	31/33.0 (93.8%)	26/29.0 (89.6%)	28/29.8 (93.9%)	29/32.3 (89.8%)	29/31.2 (93.1%)
Terminal rate	0/0	0/0	0/0	0/0	1/1 (100.0%)
First incidence (days)	235	235	221	214	208
Poly-3 trend test			P = 0.619N		P = 0.462
Poly-3 vehicle pairwise comparison	P = 0.425N		P = 0.431	P = 0.697	P = 0.492
Skin (Site of Application): Squamous Cell Carcinoma					
Overall rate	19/36 (52.8%)	15/35 (42.9%)	18/36 (50.0%)	20/36 (55.6%)	17/36 (47.2%)
Adjusted rate	19/26.5 (71.7%)	15/22.7 (66.1%)	18/24.2 (74.4%)	20/26.5 (75.4%)	17/24.6 (69.2%)
Terminal rate	0/0	0/0	0/0	0/0	1/1 (100.0%)
First incidence (days)	235	221	222	214	208
Poly-3 trend test			P = 0.254		P = 0.502N
Poly-3 vehicle pairwise comparison	P = 0.446N		P = 0.355	P = 0.312	P = 0.542
Skin (Site of Application): Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma					
Overall rate	35/36 (97.2%)	30/35 (85.7%)	31/36 (86.1%)	34/36 (94.4%)	32/36 (88.9%)
Adjusted rate	35/35.2 (99.4%)	30/31.4 (95.6%)	31/31.6 (98.1%)	34/35.1 (96.8%)	32/32.7 (97.9%)
Terminal rate	0/0	0/0	0/0	0/0	1/1 (100.0%)
First incidence (days)	235	221	221	214	208
Poly-3 trend test			P = 0.558		P = 0.403
Poly-3 vehicle pairwise comparison	P = 0.391N		P = 0.585	P = 0.684	P = 0.600
Skin (Site of Application): Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma					
Overall rate	35/36 (97.2%)	31/35 (88.6%)	31/36 (86.1%)	36/36 (100.0%)	32/36 (88.9%)
Adjusted rate	35/35.2 (99.4%)	31/32.1 (96.6%)	31/31.6 (98.1%)	36/36.0 (100.0%)	32/32.7 (97.9%)
Terminal rate	0/0	0/0	0/0	0/0	1/1 (100.0%)
First incidence (days)	235	221	221	214	208
Poly-3 trend test			P = 0.202		P = 0.543
Poly-3 vehicle pairwise comparison	P = 0.533N		P = 0.716	P = 0.432	P = 0.727

TABLE A2b
Statistical Analysis of Primary Neoplasms in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	6% Whole Leaf	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Skin (Site of Application): Squamous Cell Papilloma					
Overall rate	27/36 (75.0%)	19/36 (52.8%)	20/36 (55.6%)	27/35 (77.1%)	19/36 (52.8%)
Adjusted rate	27/30.0 (89.9%)	19/24.4 (77.7%)	20/25.5 (78.4%)	27/29.4 (91.7%)	19/24.9 (76.2%)
Terminal rate	1/1 (100.0%)	0/0	0/0	0/0	1/1 (100.0%)
First incidence (days)	221	221	242	228	207
Poly-3 trend test		P = 0.296		P = 0.425	
Poly-3 vehicle pairwise comparison	P = 0.030	P = 0.401	P = 0.367	P = 0.014	P = 0.460
Skin (Site of Application): Squamous Cell Carcinoma *in situ*					
Overall rate	28/36 (77.8%)	29/36 (80.6%)	26/36 (72.2%)	24/35 (68.6%)	30/36 (83.3%)
Adjusted rate	28/30.5 (91.9%)	29/30.6 (94.7%)	26/29.7 (87.5%)	24/27.8 (86.3%)	30/32.2 (93.1%)
Terminal rate	1/1 (100.0%)	0/0	0/0	0/0	0/1 (0.0%)
First incidence (days)	221	214	227	228	207
Poly-3 trend test		P = 0.468N		P = 0.361	
Poly-3 vehicle pairwise comparison	P = 0.578	P = 0.361	P = 0.583N	P = 0.511N	P = 0.486
Skin (Site of Application): Squamous Cell Carcinoma					
Overall rate	15/36 (41.7%)	14/36 (38.9%)	20/36 (55.6%)	21/35 (60.0%)	23/36 (63.9%)
Adjusted rate	15/23.4 (64.1%)	14/22.1 (63.4%)	20/25.8 (77.5%)	21/26.3 (79.7%)	23/28.1 (82.0%)
Terminal rate	0/1 (0.0%)	0/0	0/0	0/0	0/1 (0.0%)
First incidence (days)	242	214	227	230	207
Poly-3 trend test		P = 0.171		P = 0.088	
Poly-3 vehicle pairwise comparison	P = 0.577N	P = 0.559N	P = 0.238	P = 0.175	P = 0.113
Skin (Site of Application): Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma					
Overall rate	32/36 (88.9%)	31/36 (86.1%)	30/36 (83.3%)	28/35 (80.0%)	32/36 (88.9%)
Adjusted rate	32/32.7 (97.9%)	31/32.1 (96.5%)	30/31.8 (94.4%)	28/30.1 (93.2%)	32/33.7 (95.1%)
Terminal rate	1/1 (100.0%)	0/0	0/0	0/0	0/1 (0.0%)
First incidence (days)	221	214	227	228	207
Poly-3 trend test		P = 0.543N		P = 0.605N	
Poly-3 vehicle pairwise comparison	P = 0.598	P = 0.735	P = 0.690N	P = 0.578N	P = 0.723N
Skin (Site of Application): Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma					
Overall rate	32/36 (88.9%)	33/36 (91.7%)	32/36 (88.9%)	30/35 (85.7%)	33/36 (91.7%)
Adjusted rate	32/32.7 (97.9%)	33/33.6 (98.4%)	32/33.1 (96.8%)	30/31.1 (96.3%)	33/33.7 (98.1%)
Terminal rate	1/1 (100.0%)	0/0	0/0	0/0	1/1 (100.0%)
First incidence (days)	221	214	227	228	207
Poly-3 trend test		P = 0.679		P = 0.509	
Poly-3 vehicle pairwise comparison	P = 0.722	P = 0.666	P = 0.809	P = 0.787N	P = 0.704

[a] Number of neoplasm-bearing animals/number of animals with skin examined microscopically
[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] P values represent the results of linear trend tests. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. A negative trend is indicated by N.
[e] Poly-3 pairwise comparison to the control cream group. A lower incidence than that in the control cream group is indicated by N.

TABLE A3a
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 0.00 mJ • CIE/cm^2 SSL[a]

	No Cream	Control Cream	6% Aloe Gel	6% Whole Leaf	6% Decolorized Whole Leaf	74.6 µg/g Aloe-emodin
Disposition Summary						
Animals initially in study	36	36	36	36	36	36
Early deaths						
Accidental death			1			
Moribund		5	1	4	3	7
Natural deaths	1			1	2	
Skin lesion greater than 5 mm					1	1
Survivors						
Died last week of study				1		
Terminal sacrifice	35	30	34	30	30	27
Missexed		1				1
Animals examined microscopically	36	35	36	35[b]	36	35
Integumentary System						
Skin, control	(36)	(35)	(36)	(35)	(36)	(35)
Cyst epithelial inclusion	2 (6%)	3 (9%)	1 (3%)	3 (9%)	4 (11%)	3 (9%)
Hyperplasia, squamous	2 (6%)	2 (6%)	2 (6%)	1 (3%)	1 (3%)	
Inflammation, pyogranulomatous	3 (8%)		3 (8%)	3 (9%)	3 (8%)	
Dermis, inflammation, chronic active	34 (94%)	31 (89%)	34 (94%)	32 (91%)	36 (100%)	32 (91%)
Epidermis, necrosis	2 (6%)		1 (3%)	1 (3%)	1 (3%)	
Skin, site of application	(36)	(35)	(36)	(35)	(36)	(35)
Cyst epithelial inclusion	1 (3%)	2 (6%)	4 (11%)	3 (9%)	5 (14%)	9 (26%)
Granuloma				1 (3%)		
Hyperplasia, squamous atypical, one, focal						1 (3%)
Hyperplasia, squamous	1 (3%)	1 (3%)	1 (3%)		1 (3%)	1 (3%)
Inflammation, pyogranulomatous					1 (3%)	
Dermis, inflammation, chronic active	36 (100%)	33 (94%)	36 (100%)	34 (97%)	36 (100%)	33 (94%)
Epidermis, necrosis		1 (3%)			1 (3%)	

Systems Examined with No Lesions Observed
Alimentary System
Cardiovascular System
Endocrine System
General Body System
Genital System
Hematopoietic System
Musculoskeletal System
Nervous System
Respiratory System
Special Senses System
Urinary System

[a] Number of animals examined microscopically at the site and the number of animals with lesion. The number given with the lesion name indicates the number of that lesion per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.
[b] One animal autolyzed, and no tissue was taken.

TABLE A3b
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 6.85 mJ • CIE/cm^2 SSL[a]

	No Cream
Disposition Summary	
Animals initially in study	36
Early deaths	
Accidental death	1
Moribund	5
Natural deaths	2
Skin lesion greater than 5 mm	5
Survivors	
Died last week of study	1
Terminal sacrifice	22
Animals examined microscopically	36
Integumentary System	
Skin, control	(36)
Abscess	2 (6%)
Cyst epithelial inclusion	4 (11%)
Dermis, inflammation, chronic active	30 (83%)
Subcutaneous tissue, edema	1 (3%)
Skin, site of application	(36)
Abscess	1 (3%)
Cyst epithelial inclusion	12 (33%)
Hyperplasia, squamous atypical, one, focal	6 (17%)
Hyperplasia, squamous atypical, two, focal	3 (8%)
Hyperplasia, squamous atypical, three, focal	1 (3%)
Hyperplasia, squamous atypical, four, focal	1 (3%)
Hyperplasia, squamous atypical, five, focal	1 (3%)
Hyperplasia, squamous	20 (56%)
Inflammation, pyogranulomatous	1 (3%)
Dermis, inflammation, chronic active	33 (92%)

Systems Examined with No Lesions Observed
Alimentary System
Cardiovascular System
Endocrine System
General Body System
Genital System
Hematopoietic System
Musculoskeletal System
Nervous System
Respiratory System
Special Senses System
Urinary System

[a] Number of animals examined microscopically at the site and the number of animals with lesion. The number given with the lesion name indicates the number of that lesion per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.

TABLE A3c
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL[a]

	No Cream	Control Cream	3% Aloe Gel	6% Aloe Gel	3% Whole Leaf
Disposition Summary					
Animals initially in study	36	36	36	36	36
Early deaths					
Moribund		3	5	1	1
Natural deaths		1			2
Skin lesion greater than 5 mm	36	31	31	35	32
Survivors					
Terminal sacrifice			1		
Missexed					
Animals examined microscopically	36	35[b]	36	36	36
Integumentary System					
Skin, control	(36)	(35)	(36)	(36)	(36)
Abscess	1 (3%)		1 (3%)		1 (3%)
Cyst epithelial inclusion	4 (11%)	4 (11%)	3 (8%)	3 (8%)	
Developmental malformation			1 (3%)		
Granuloma				2 (6%)	
Hyperplasia, squamous atypical, one, focal				1 (3%)	
Hyperplasia, squamous	2 (6%)	2 (6%)		2 (6%)	3 (8%)
Inflammation, pyogranulomatous	4 (11%)		2 (6%)		3 (8%)
Dermis, inflammation, chronic active	22 (61%)	30 (86%)	26 (72%)	30 (83%)	31 (86%)
Epidermis, necrosis	1 (3%)	1 (3%)	3 (8%)	1 (3%)	
Skin, site of application	(36)	(35)	(36)	(36)	(36)
Abscess					
Cyst epithelial inclusion	5 (14%)	3 (9%)	7 (19%)	4 (11%)	6 (17%)
Granuloma					
Hyperplasia, squamous atypical, one, focal	5 (14%)	5 (14%)	2 (6%)	4 (11%)	4 (11%)
Hyperplasia, squamous atypical, two, focal	9 (25%)	6 (17%)	10 (28%)	10 (28%)	5 (14%)
Hyperplasia, squamous atypical, three, focal	6 (17%)	7 (20%)	5 (14%)	6 (17%)	4 (11%)
Hyperplasia, squamous atypical, four, focal	3 (8%)	4 (11%)	4 (11%)	4 (11%)	7 (19%)
Hyperplasia, squamous atypical, five, focal	2 (6%)	5 (14%)	3 (8%)	5 (14%)	4 (11%)
Hyperplasia, squamous atypical, greater than five, focal	8 (22%)	4 (11%)	4 (11%)	5 (14%)	5 (14%)
Hyperplasia, squamous	35 (97%)	31 (89%)	33 (92%)	36 (100%)	34 (94%)
Inflammation, pyogranulomatous	2 (6%)			1 (3%)	1 (3%)
Dermis, inflammation, chronic active	34 (94%)	32 (91%)	32 (89%)	34 (94%)	32 (89%)
Epidermis, necrosis			2 (6%)	3 (8%)	2 (6%)
Subcutaneous tissue, inflammation, pyogranulomatous	1 (3%)				

TABLE A3c
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	6% Whole Leaf	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Disposition Summary					
Animals initially in study	36	36	36	36	36
Early deaths					
Moribund	2	2	4	3	
Natural deaths	1	1		2	1
Skin lesion greater than 5 mm	32	33	32	30	34
Survivors					
Terminal sacrifice	1				1
Missexed				1	
Animals examined microscopically	36	36	36	35	36
Integumentary System					
Skin, control	(36)	(36)	(36)	(34)	(36)
Abscess			1 (3%)		1 (3%)
Cyst epithelial inclusion	3 (8%)	2 (6%)	4 (11%)	2 (6%)	6 (17%)
Developmental malformation					
Granuloma			1 (3%)		
Hyperplasia, squamous atypical, one, focal					
Hyperplasia, squamous		1 (3%)	1 (3%)	1 (3%)	1 (3%)
Inflammation, pyogranulomatous	3 (8%)		1 (3%)	4 (12%)	2 (6%)
Dermis, inflammation, chronic active	30 (83%)	27 (75%)	33 (92%)	28 (82%)	34 (94%)
Epidermis, necrosis	4 (11%)	2 (6%)		5 (15%)	1 (3%)
Skin, site of application	(36)	(36)	(36)	(35)	(36)
Abscess	1 (3%)				
Cyst epithelial inclusion	2 (6%)	5 (14%)	4 (11%)	3 (9%)	2 (6%)
Granuloma	1 (3%)				
Hyperplasia, squamous atypical, one, focal	4 (11%)	4 (11%)	5 (14%)	5 (14%)	8 (22%)
Hyperplasia, squamous atypical, two, focal	5 (14%)	14 (39%)	2 (6%)	5 (14%)	5 (14%)
Hyperplasia, squamous atypical, three, focal	12 (33%)	9 (25%)	4 (11%)	5 (14%)	4 (11%)
Hyperplasia, squamous atypical, four, focal	4 (11%)		10 (28%)	8 (23%)	4 (11%)
Hyperplasia, squamous atypical, five, focal	1 (3%)	3 (8%)	4 (11%)	3 (9%)	5 (14%)
Hyperplasia, squamous atypical, greater than five, focal	8 (22%)	4 (11%)	5 (14%)	4 (11%)	7 (19%)
Hyperplasia, squamous	34 (94%)	35 (97%)	35 (97%)	32 (91%)	32 (89%)
Inflammation, pyogranulomatous				2 (6%)	
Dermis, inflammation, chronic active	33 (92%)	34 (94%)	34 (94%)	35 (100%)	32 (89%)
Epidermis, necrosis	2 (6%)	6 (17%)	5 (14%)	7 (20%)	1 (3%)
Subcutaneous tissue, inflammation, pyogranulomatous					

TABLE A3c
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL[a]

	No Cream	Control Cream	3% Aloe Gel	6% Aloe Gel	3% Whole Leaf
Systems Examined with No Lesions Observed					
Alimentary System					
Cardiovascular System					
Endocrine System					
General Body System					
Genital System					
Hematopoietic System					
Musculoskeletal System					
Nervous System					
Respiratory System					
Special Senses System					
Urinary System					

TABLE A3c
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	6% Whole Leaf	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Systems Examined with No Lesions Observed					
Alimentary System					
Cardiovascular System					
Endocrine System					
General Body System					
Genital System					
Hematopoietic System					
Musculoskeletal System					
Nervous System					
Respiratory System					
Special Senses System					
Urinary System					

[a] Number of animals examined microscopically at the site and the number of animals with lesion. The number given with the lesion name indicates the number of that lesion per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.
[b] One animal died prior to dosing and was discarded.

TABLE A3d
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 20.55 mJ • CIE/cm^2 SSL[a]

	No Cream
Disposition Summary	
Animals initially in study	36
Early deaths	
Moribund	3
Skin lesion greater than 5 mm	33
Animals examined microscopically	36
Integumentary System	
Skin, control	(36)
Abscess	1 (3%)
Cyst epithelial inclusion	1 (3%)
Inflammation, pyogranulomatous	1 (3%)
Dermis, inflammation, chronic active	20 (56%)
Epidermis, necrosis	1 (3%)
Skin, site of application	(36)
Abscess	1 (3%)
Cyst epithelial inclusion	1 (3%)
Hyperplasia, squamous atypical, one, focal	7 (19%)
Hyperplasia, squamous atypical, two, focal	9 (25%)
Hyperplasia, squamous atypical, three, focal	3 (8%)
Hyperplasia, squamous atypical, four, focal	6 (17%)
Hyperplasia, squamous atypical, five, focal	2 (6%)
Hyperplasia, squamous atypical, greater than five, focal	5 (14%)
Hyperplasia, squamous	28 (78%)
Dermis, inflammation, chronic active	26 (72%)
Epidermis, necrosis	5 (14%)

Systems Examined with No Lesions Observed
Alimentary System
Cardiovascular System
Endocrine System
General Body System
Genital System
Hematopoietic System
Musculoskeletal System
Nervous System
Respiratory System
Special Senses System
Urinary System

[a] Number of animals examined microscopically at the site and the number of animals with lesion. The number given with the lesion name indicates the number of that lesion per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.

APPENDIX B
SUMMARY OF LESIONS IN FEMALE MICE IN THE 1-YEAR SIMULATED SOLAR LIGHT STUDY OF ALOE VERA

TABLE B1a
Summary of the Incidence of Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 0.00 mJ • CIE/cm^2 SSL[a]

	No Cream	Control Cream	6% Aloe Gel	6% Whole Leaf	6% Decolorized Whole Leaf	74.6 µg/g Aloe-emodin
Disposition Summary						
Animals initially in study	36	36	36	36	36	36
Early deaths						
Moribund	5	7	2	2	3	
Natural deaths	1		2			1
Skin lesion greater than 5 mm		1		1		
Survivors						
Died last week of study			2		1	2
Terminal sacrifice	30	28	30	33	32	33
Animals examined microscopically	36	36	36	36	36	36
Integumentary System						
Skin, control	(36)	(36)	(36)	(36)	(36)	(36)
Lymphoma malignant					1 (3%)	
Subcutaneous tissue, lipoma		1 (3%)				
Subcutaneous tissue, lymphoma malignant		1 (3%)				
Skin, site of application	(36)	(36)	(36)	(36)	(36)	(36)
Lymphoma malignant		1 (3%)			1 (3%)	
Squamous cell papilloma, one				1 (3%)		
Systemic Lesions						
Multiple organs[b]	(36)	(36)	(36)	(36)	(36)	(36)
Lymphoma malignant		1 (3%)			1 (3%)	
Systems Examined with No Neoplasms Observed						
Alimentary System						
Cardiovascular System						
Endocrine System						
General Body System						
Genital System						
Hematopoietic System						
Musculoskeletal System						
Nervous System						
Respiratory System						
Special Senses System						
Urinary System						
Neoplasm Summary						
Total animals with primary neoplasms[c]		2		1	1	
Total primary neoplasms		2		1	1	
Total animals with benign neoplasms		1		1		
Total benign neoplasms		1		1		
Total animals with malignant neoplasms		1			1	
Total malignant neoplasms		1			1	

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm. The number given with the neoplasm name indicates the number of that neoplasm per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.
[b] Number of animals with any tissue examined microscopically
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE B1b
Summary of the Incidence of Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 6.85 mJ • CIE/cm^2 SSL[a]

	No Cream
Disposition Summary	
Animals initially in study	36
Early deaths	
Moribund	1
Natural death	1
Skin lesion greater than 5 mm	7
Survivors	
Died last week of study	3
Terminal sacrifice	24
Animals examined microscopically	36
Integumentary System	
Skin, control	(36)
Squamous cell carcinoma, one	2 (6%)
Squamous cell papilloma, one	6 (17%)
Skin, site of application	(36)
Lymphoma malignant	1 (3%)
Squamous cell carcinoma, one	2 (6%)
Squamous cell carcinoma *in situ*, one	4 (11%)
Squamous cell carcinoma *in situ*, two	2 (6%)
Squamous cell carcinoma *in situ*, three	2 (6%)
Squamous cell carcinoma *in situ*, five	1 (3%)
Squamous cell papilloma, one	6 (17%)
Squamous cell papilloma, two	4 (11%)
Squamous cell papilloma, three	6 (17%)
Systemic Lesions	
Multiple organs[b]	(36)
Lymphoma malignant	1 (3%)
Systems Examined with No Neoplasms Observed	
Alimentary System	
Cardiovascular System	
Endocrine System	
General Body System	
Genital System	
Hematopoietic System	
Musculoskeletal System	
Nervous System	
Respiratory System	
Special Senses System	
Urinary System	
Neoplasm Summary	
Total animals with primary neoplasms[c]	23
Total primary neoplasms	36
Total animals with benign neoplasms	18
Total benign neoplasms	22
Total animals with malignant neoplasms	13
Total malignant neoplasms	14

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm. The number given with the neoplasm name indicates the number of that neoplasm per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.
[b] Number of animals with any tissue examined microscopically
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE B1c
Summary of the Incidence of Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL[a]

	No Cream	Control Cream	3% Aloe Gel	6% Aloe Gel	3% Whole Leaf
Disposition Summary					
Animals initially in study	36	36	36	36	36
Early deaths					
Moribund	1	1	4	4	2
Natural deaths		1	1		
Skin lesion greater than 5 mm	34	33	32	31	31
Survivors					
Died last week of study					1
Terminal sacrifice	1	2			1
Animals examined microscopically	36	36	36	36	36
Integumentary System					
Skin, control	(36)	(36)	(36)	(36)	(36)
Lymphoma malignant					
Squamous cell carcinoma, one			2 (6%)		
Squamous cell carcinoma in situ, one	1 (3%)				1 (3%)
Squamous cell papilloma, one	1 (3%)			1 (3%)	2 (6%)
Sebaceous gland, adenoma					
Skin, site of application	(36)	(36)	(36)	(36)	(36)
Lymphoma malignant					1 (3%)
Squamous cell carcinoma, one	16 (44%)	20 (56%)	15 (42%)	17 (47%)	16 (44%)
Squamous cell carcinoma, two	5 (14%)	4 (11%)	5 (14%)	6 (17%)	2 (6%)
Squamous cell carcinoma, three	1 (3%)	1 (3%)	2 (6%)		2 (6%)
Squamous cell carcinoma, four	1 (3%)				
Squamous cell carcinoma in situ, one	10 (28%)	9 (25%)	5 (14%)	7 (19%)	4 (11%)
Squamous cell carcinoma in situ, two	5 (14%)	7 (19%)	6 (17%)	6 (17%)	6 (17%)
Squamous cell carcinoma in situ, three	7 (19%)	6 (17%)	5 (14%)	3 (8%)	4 (11%)
Squamous cell carcinoma in situ, four	2 (6%)	3 (8%)	8 (22%)	2 (6%)	2 (6%)
Squamous cell carcinoma in situ, five	1 (3%)	3 (8%)	4 (11%)	5 (14%)	3 (8%)
Squamous cell carcinoma in situ, greater than five	3 (8%)	2 (6%)	4 (11%)	5 (14%)	12 (33%)
Squamous cell papilloma, one	7 (19%)	5 (14%)	4 (11%)	7 (19%)	5 (14%)
Squamous cell papilloma, two	5 (14%)	6 (17%)	5 (14%)	4 (11%)	8 (22%)
Squamous cell papilloma, three	3 (8%)	3 (8%)	5 (14%)	4 (11%)	5 (14%)
Squamous cell papilloma, four		5 (14%)	2 (6%)	8 (22%)	2 (6%)
Squamous cell papilloma, five	3 (8%)	1 (3%)	3 (8%)	1 (3%)	
Squamous cell papilloma, greater than five	2 (6%)	1 (3%)	6 (17%)	5 (14%)	6 (17%)
Systemic Lesions					
Multiple organs[b]	(36)	(36)	(36)	(36)	(36)
Lymphoma malignant					1 (3%)

TABLE B1c
Summary of the Incidence of Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	6% Whole Leaf	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Disposition Summary					
Animals initially in study	36	36	36	36	36
Early deaths					
Moribund	1		2	4	1
Natural deaths	3	2	1		2
Skin lesion greater than 5 mm	32	34	33	32	33
Survivors					
Died last week of study					
Terminal sacrifice					
Animals examined microscopically	36	36	36	36	36
Integumentary System					
Skin, control	(36)	(36)	(36)	(36)	(36)
Lymphoma malignant			1 (3%)		
Squamous cell carcinoma, one	1 (3%)				
Squamous cell carcinoma *in situ*, one	1 (3%)				
Squamous cell papilloma, one	1 (3%)	2 (6%)	2 (6%)	1 (3%)	1 (3%)
Sebaceous gland, adenoma				1 (3%)	
Skin, site of application	(36)	(36)	(36)	(36)	(36)
Lymphoma malignant			1 (3%)		
Squamous cell carcinoma, one	14 (39%)	10 (28%)	9 (25%)	19 (53%)	11 (31%)
Squamous cell carcinoma, two	4 (11%)	4 (11%)	7 (19%)	1 (3%)	3 (8%)
Squamous cell carcinoma, three		1 (3%)	1 (3%)	2 (6%)	4 (11%)
Squamous cell carcinoma, four					1 (3%)
Squamous cell carcinoma *in situ*, one	6 (17%)	3 (8%)	7 (19%)	9 (25%)	5 (14%)
Squamous cell carcinoma *in situ*, two	10 (28%)	3 (8%)	4 (11%)	4 (11%)	6 (17%)
Squamous cell carcinoma *in situ*, three	4 (11%)	6 (17%)	2 (6%)	3 (8%)	6 (17%)
Squamous cell carcinoma *in situ*, four	4 (11%)	4 (11%)	6 (17%)	5 (14%)	5 (14%)
Squamous cell carcinoma *in situ*, five	1 (3%)	7 (19%)	5 (14%)	2 (6%)	3 (8%)
Squamous cell carcinoma *in situ*, greater than five	3 (8%)	9 (25%)	5 (14%)	4 (11%)	6 (17%)
Squamous cell papilloma, one	4 (11%)	7 (19%)	6 (17%)	4 (11%)	7 (19%)
Squamous cell papilloma, two	6 (17%)	7 (19%)	7 (19%)	4 (11%)	9 (25%)
Squamous cell papilloma, three	6 (17%)	6 (17%)	5 (14%)	6 (17%)	4 (11%)
Squamous cell papilloma, four	4 (11%)	2 (6%)	3 (8%)	3 (8%)	2 (6%)
Squamous cell papilloma, five	3 (8%)	3 (8%)		6 (17%)	2 (6%)
Squamous cell papilloma, greater than five	6 (17%)	5 (14%)	10 (28%)	4 (11%)	5 (14%)
Systemic Lesions					
Multiple organs	(36)	(36)	(36)	(36)	(36)
Lymphoma malignant			1 (3%)		

TABLE B1c
Summary of the Incidence of Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	No Cream	Control Cream	3% Aloe Gel	6% Aloe Gel	3% Whole Leaf
Systems Examined with No Neoplasms Observed					
Alimentary System					
Cardiovascular System					
Endocrine System					
General Body System					
Genital System					
Hematopoietic System					
Musculoskeletal System					
Nervous System					
Respiratory System					
Special Senses System					
Urinary System					
Neoplasm Summary					
Total animals with primary neoplasms[c]	35	35	33	33	34
Total primary neoplasms	73	76	81	81	81
Total animals with benign neoplasms	20	21	25	29	26
Total benign neoplasms	21	21	25	30	28
Total animals with malignant neoplasms	34	35	33	32	33
Total malignant neoplasms	52	55	56	51	53

TABLE B1c
Summary of the Incidence of Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	6% Whole Leaf	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Systems Examined with No Neoplasms Observed					
Alimentary System					
Cardiovascular System					
Endocrine System					
General Body System					
Genital System					
Hematopoietic System					
Musculoskeletal System					
Nervous System					
Respiratory System					
Special Senses System					
Urinary System					
Neoplasm Summary					
Total animals with primary neoplasms	34	34	35	34	33
Total primary neoplasms	78	79	80	78	80
Total animals with benign neoplasms	29	30	31	27	29
Total benign neoplasms	30	32	33	29	30
Total animals with malignant neoplasms	32	33	33	32	32
Total malignant neoplasms	48	47	47	49	50

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm. The number given with the neoplasm name indicates the number of that neoplasm per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.
[b] Number of animals with any tissue examined microscopically
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE B1d
Summary of the Incidence of Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 20.55 mJ • CIE/cm^2 SSL[a]

	No Cream
Disposition Summary	
Animals initially in study	36
Early deaths	
Moribund	1
Skin lesion greater than 5 mm	35
Animals examined microscopically	36
Integumentary System	
Skin, control	(36)
Skin, site of application	(36)
Squamous cell carcinoma, one	8 (22%)
Squamous cell carcinoma, two	7 (19%)
Squamous cell carcinoma, three	1 (3%)
Squamous cell carcinoma, four	1 (3%)
Squamous cell carcinoma *in situ*, one	7 (19%)
Squamous cell carcinoma *in situ*, two	4 (11%)
Squamous cell carcinoma *in situ*, three	7 (19%)
Squamous cell carcinoma *in situ*, four	3 (8%)
Squamous cell carcinoma *in situ*, five	1 (3%)
Squamous cell carcinoma *in situ*, greater than five	1 (3%)
Squamous cell papilloma, one	8 (22%)
Squamous cell papilloma, two	3 (8%)
Squamous cell papilloma, three	2 (6%)
Squamous cell papilloma, five	1 (3%)
Squamous cell papilloma, greater than five	2 (6%)

TABLE B1d
Summary of the Incidence of Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 20.55 mJ • CIE/cm^2 SSL

	No Cream
Systems Examined with No Neoplasms Observed	
Alimentary System	
Cardiovascular System	
Endocrine System	
General Body System	
Genital System	
Hematopoietic System	
Musculoskeletal System	
Nervous System	
Respiratory System	
Special Senses System	
Urinary System	
Neoplasm Summary	
Total animals with primary neoplasms[b]	32
Total primary neoplasms	56
Total animals with benign neoplasms	16
Total benign neoplasms	16
Total animals with malignant neoplasms	28
Total malignant neoplasms	40

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm. The number given with the neoplasm name indicates the number of that neoplasm per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.
[b] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE B2a
Statistical Analysis of Primary Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

	0.00 mJ • CIE/cm^2	6.85 mJ • CIE/cm^2	13.70 mJ • CIE/cm^2	20.55 mJ • CIE/cm^2
Skin (Control Site): Squamous Cell Papilloma				
Overall rate[a]	0/36 (0.0%)	6/36 (16.7%)	1/36 (2.8%)	0/36 (0.0%)
Adjusted rate[b]	0/32.8 (0.0%)	6/33.6 (17.8%)	1/19.0 (5.3%)	0/8.8 (0.0%)
Terminal rate[c]	0/30 (0.0%)	4/24 (16.7%)	0/1 (0.0%)	0/0
First incidence (days)	—[e]	352	250	—
Poly-3 test[d]	P = 0.158	P = 0.014	P = 0.397	—[f]
Skin (Control Site): Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	2/36 (5.6%)	0/36 (0.0%)	0/36 (0.0%)
Adjusted rate	0/32.8 (0.0%)	2/34.1 (5.9%)	0/18.3 (0.0%)	0/8.8 (0.0%)
Terminal rate	0/30 (0.0%)	0/24 (0.0%)	0/1 (0.0%)	0/0
First incidence (days)	—	292	—	—
Poly-3 test	P = 0.520	P = 0.245	—	—
Skin (Control Site): Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	2/36 (5.6%)	1/36 (2.8%)	0/36 (0.0%)
Adjusted rate	0/32.8 (0.0%)	2/34.1 (5.9%)	1/18.7 (5.3%)	0/8.8 (0.0%)
Terminal rate	0/30 (0.0%)	0/24 (0.0%)	0/1 (0.0%)	0/0
First incidence (days)	—	292	299	—
Poly-3 test	P = 0.271	P = 0.245	P = 0.395	—
Skin (Control Site): Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	8/36 (22.2%)	2/36 (5.6%)	0/36 (0.0%)
Adjusted rate	0/32.8 (0.0%)	8/34.3 (23.3%)	2/19.4 (10.3%)	0/8.8 (0.0%)
Terminal rate	0/30 (0.0%)	4/24 (16.7%)	0/1 (0.0%)	0/0
First incidence (days)	—	292	250	—
Poly-3 test	P = 0.070	P = 0.003	P = 0.142	—
Skin (Site of Application): Squamous Cell Papilloma				
Overall rate	0/36 (0.0%)	16/36 (44.4%)	20/36 (55.6%)	16/36 (44.4%)
Adjusted rate	0/32.8 (0.0%)	16/34.4 (46.5%)	20/28.1 (71.1%)	16/20.7 (77.1%)
Terminal rate	0/30 (0.0%)	11/24 (45.8%)	0/1 (0.0%)	0/0
First incidence (days)	—	320	242	186
Poly-3 test	P≤0.001	P≤0.001	P≤0.001	P≤0.001

TABLE B2a
Statistical Analysis of Primary Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

	0.00 mJ • CIE/cm^2	6.85 mJ • CIE/cm^2	13.70 mJ • CIE/cm^2	20.55 mJ • CIE/cm^2
Skin (Site of Application): Squamous Cell Carcinoma *in situ*				
Overall rate	0/36 (0.0%)	9/36 (25.0%)	28/36 (77.8%)	23/36 (63.9%)
Adjusted rate	0/32.8 (0.0%)	9/34.5 (26.1%)	28/31.7 (88.3%)	23/26.1 (88.0%)
Terminal rate	0/30 (0.0%)	5/24 (20.8%)	1/1 (100.0%)	0/0
First incidence (days)	—	299	215	172
Poly-3 test	P≤0.001	P = 0.002	P≤0.001	P≤0.001
Skin (Site of Application): Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	2/36 (5.6%)	23/36 (63.9%)	17/36 (47.2%)
Adjusted rate	0/32.8 (0.0%)	2/33.4 (6.0%)	23/29.2 (78.8%)	17/21.6 (78.8%)
Terminal rate	0/30 (0.0%)	2/24 (8.3%)	0/1 (0.0%)	0/0
First incidence (days)	—	366 (T)	250	199
Poly-3 test	P≤0.001	P = 0.240	P≤0.001	P≤0.001
Skin (Site of Application): Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	10/36 (27.8%)	34/36 (94.4%)	28/36 (77.8%)
Adjusted rate	0/32.8 (0.0%)	10/34.5 (29.0%)	34/34.7 (97.9%)	28/30.0 (93.4%)
Terminal rate	0/30 (0.0%)	6/24 (25.0%)	1/1 (100.0%)	0/0
First incidence (days)	—	299	215	172
Poly-3 test	P≤0.001	P≤0.001	P≤0.001	P≤0.001
Skin (Site of Application): Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma				
Overall rate	0/36 (0.0%)	19/36 (52.8%)	35/36 (97.2%)	32/36 (88.9%)
Adjusted rate	0/32.8 (0.0%)	19/35.0 (54.4%)	35/35.4 (98.8%)	32/33.0 (97.0%)
Terminal rate	0/30 (0.0%)	12/24 (50.0%)	1/1 (100.0%)	0/0
First incidence (days)	—	299	215	172
Poly-3 test	P≤0.001	P≤0.001	P≤0.001	P≤0.001

(T) Terminal sacrifice
[a] Number of neoplasm-bearing animals/number of animals with skin examined microscopically
[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] P values for the control group (0.00 mJ • CIE/cm^2) represent the results of linear trend tests; otherwise, P values represent pairwise comparisons to the control group. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice.
[e] Not applicable; no neoplasms in animal group
[f] Value of statistic cannot be computed.

TABLE B2b
Statistical Analysis of Primary Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	No Cream	Control Cream	3% Aloe Gel	6% Aloe Gel	3% Whole Leaf
Skin (Control Site): Squamous Cell Papilloma					
Overall rate[a]	1/36 (2.8%)	0/36 (0.0%)	0/36 (0.0%)	1/36 (2.8%)	2/36 (5.6%)
Adjusted rate[b]	1/19.0 (5.3%)	0/15.9 (0.0%)	0/16.3 (0.0%)	1/15.9 (6.3%)	2/19.5 (10.2%)
Terminal rate[c]	0/1 (0.0%)	0/2 (0.0%)	0/0	0/0	0/1 (0.0%)
First incidence (days)	250	—[f]	—	265	313
Poly-3 trend test[d]			P = 0.268		P = 0.410
Poly-3 vehicle pairwise comparison[e]	P = 0.535N		—[g]	P = 0.500	P = 0.280
Skin (Control Site): Squamous Cell Carcinoma					
Overall rate	0/36 (0.0%)	0/36 (0.0%)	2/36 (5.6%)	0/36 (0.0%)	0/36 (0.0%)
Adjusted rate	0/18.3 (0.0%)	0/15.9 (0.0%)	2/17.4 (11.5%)	0/15.3 (0.0%)	0/18.9 (0.0%)
Terminal rate	0/1 (0.0%)	0/2 (0.0%)	0/0	0/0	0/1 (0.0%)
First incidence (days)	—	—	279	—	—
Poly-3 trend test			P = 0.660		P = 0.287
Poly-3 vehicle pairwise comparison	—		P = 0.251	—	—
Skin (Control Site): Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma					
Overall rate	1/36 (2.8%)	0/36 (0.0%)	2/36 (5.6%)	0/36 (0.0%)	1/36 (2.8%)
Adjusted rate	1/18.7 (5.3%)	0/15.9 (0.0%)	2/17.4 (11.5%)	0/15.3 (0.0%)	1/19.3 (5.2%)
Terminal rate	0/1 (0.0%)	0/2 (0.0%)	0/0	0/0	0/1 (0.0%)
First incidence (days)	299	—	279	—	313
Poly-3 trend test			P = 0.660		P = 0.166
Poly-3 vehicle pairwise comparison	P = 0.533N		P = 0.251	—	P = 0.538
Skin (Control Site): Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma					
Overall rate	2/36 (5.6%)	0/36 (0.0%)	2/36 (5.6%)	1/36 (2.8%)	2/36 (5.6%)
Adjusted rate	2/19.4 (10.3%)	0/15.9 (0.0%)	2/17.4 (11.5%)	1/15.9 (6.3%)	2/19.5 (10.2%)
Terminal rate	0/1 (0.0%)	0/2 (0.0%)	0/0	0/0	0/1 (0.0%)
First incidence (days)	250	—	279	265	313
Poly-3 trend test			P = 0.357		P = 0.104
Poly-3 vehicle pairwise comparison	P = 0.279N		P = 0.251	P = 0.500	P = 0.280
Skin (Site of Application): Squamous Cell Papilloma					
Overall rate	20/36 (55.6%)	21/36 (58.3%)	25/36 (69.4%)	29/36 (80.6%)	26/36 (72.2%)
Adjusted rate	20/28.1 (71.1%)	21/26.5 (79.2%)	25/28.7 (87.0%)	29/30.5 (95.0%)	26/30.7 (84.8%)
Terminal rate	0/1 (0.0%)	2/2 (100.0%)	0/0	0/0	0/1 (0.0%)
First incidence (days)	242	208	237	235	243
Poly-3 trend test			P = 0.012		P = 0.039
Poly-3 vehicle pairwise comparison	P = 0.322		P = 0.283	P = 0.018	P = 0.396
Skin (Site of Application): Squamous Cell Carcinoma *in situ*					
Overall rate	28/36 (77.8%)	30/36 (83.3%)	32/36 (88.9%)	28/36 (77.8%)	31/36 (86.1%)
Adjusted rate	28/31.7 (88.3%)	30/31.9 (93.9%)	32/32.7 (97.9%)	28/30.3 (92.4%)	31/33.2 (93.4%)
Terminal rate	1/1 (100.0%)	2/2 (100.0%)	0/0	0/0	1/1 (100.0%)
First incidence (days)	215	208	237	235	243
Poly-3 trend test			P = 0.532N		P = 0.260N
Poly-3 vehicle pairwise comparison	P = 0.315		P = 0.398	P = 0.650N	P = 0.710N

TABLE B2b
Statistical Analysis of Primary Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	6% Whole Leaf	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Skin (Control Site): Squamous Cell Papilloma					
Overall rate	1/36 (2.8%)	2/36 (5.6%)	2/36 (5.6%)	1/36 (2.8%)	1/36 (2.8%)
Adjusted rate	1/17.7 (5.7%)	2/16.2 (12.3%)	2/17.8 (11.2%)	1/15.7 (6.4%)	1/16.6 (6.0%)
Terminal rate	0/0	0/0	0/0	0/0	0/0
First incidence (days)	292	299	255	271	292
Poly-3 trend test		P = 0.203		P = 0.344	
Poly-3 vehicle pairwise comparison	P = 0.522	P = 0.234	P = 0.256	P = 0.497	P = 0.509
Skin (Control Site): Squamous Cell Carcinoma					
Overall rate	1/36 (2.8%)	0/36 (0.0%)	0/36 (0.0%)	0/36 (0.0%)	0/36 (0.0%)
Adjusted rate	1/17.7 (5.6%)	0/15.3 (0.0%)	0/16.6 (0.0%)	0/15.1 (0.0%)	0/16.1 (0.0%)
Terminal rate	0/0	0/0	0/0	0/0	0/0
First incidence (days)	286	—	—	—	—
Poly-3 trend test		—[g]		—	
Poly-3 vehicle pairwise comparison	P = 0.522	—	—	—	—
Skin (Control Site): Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma					
Overall rate	2/36 (5.6%)	0/36 (0.0%)	0/36 (0.0%)	0/36 (0.0%)	0/36 (0.0%)
Adjusted rate	2/18.4 (10.9%)	0/15.3 (0.0%)	0/16.6 (0.0%)	0/15.1 (0.0%)	0/16.1 (0.0%)
Terminal rate	0/0	0/0	0/0	0/0	0/0
First incidence (days)	265	—	—	—	—
Poly-3 trend test		—		—	
Poly-3 vehicle pairwise comparison	P = 0.264	—	—	—	—
Skin (Control Site): Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma					
Overall rate	3/36 (8.3%)	2/36 (5.6%)	2/36 (5.6%)	1/36 (2.8%)	1/36 (2.8%)
Adjusted rate	3/18.9 (15.9%)	2/16.2 (12.3%)	2/17.8 (11.2%)	1/15.7 (6.4%)	1/16.6 (6.0%)
Terminal rate	0/0	0/0	0/0	0/0	0/0
First incidence (days)	265	299	255	271	292
Poly-3 trend test		P = 0.203		P = 0.344	
Poly-3 vehicle pairwise comparison	P = 0.140	P = 0.234	P = 0.256	P = 0.497	P = 0.509
Skin (Site of Application): Squamous Cell Papilloma					
Overall rate	29/36 (80.6%)	30/36 (83.3%)	31/36 (86.1%)	27/36 (75.0%)	29/36 (80.6%)
Adjusted rate	29/31.2 (93.1%)	30/32.6 (92.1%)	31/32.9 (94.3%)	27/30.3 (89.1%)	29/31.1 (93.3%)
Terminal rate	0/0	0/0	0/0	0/0	0/0
First incidence (days)	236	229	235	223	192
Poly-3 trend test		P = 0.020		P = 0.028	
Poly-3 vehicle pairwise comparison	P = 0.048	P = 0.077	P = 0.029	P = 0.176	P = 0.049
Skin (Site of Application): Squamous Cell Carcinoma *in situ*					
Overall rate	28/36 (77.8%)	32/36 (88.9%)	29/36 (80.6%)	27/36 (75.0%)	31/36 (86.1%)
Adjusted rate	28/31.5 (88.9%)	32/33.3 (96.1%)	29/31.5 (92.0%)	27/30.3 (89.1%)	31/31.5 (98.3%)
Terminal rate	0/0	0/0	0/0	0/0	0/0
First incidence (days)	236	229	229	223	223
Poly-3 trend test		P = 0.469N		P = 0.246	
Poly-3 vehicle pairwise comparison	P = 0.363N	P = 0.601	P = 0.604N	P = 0.369N	P = 0.356

TABLE B2b
Statistical Analysis of Primary Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	No Cream	Control Cream	3% Aloe Gel	6% Aloe Gel	3% Whole Leaf
Skin (Site of Application): Squamous Cell Carcinoma					
Overall rate	23/36 (63.9%)	25/36 (69.4%)	22/36 (61.1%)	23/36 (63.9%)	20/36 (55.6%)
Adjusted rate	23/29.2 (78.8%)	25/29.8 (83.8%)	22/27.8 (79.1%)	23/28.8 (79.9%)	20/27.9 (71.8%)
Terminal rate	0/1 (0.0%)	1/2 (50.0%)	0/0	0/0	1/1 (100.0%)
First incidence (days)	250	208	243	194	243
Poly-3 trend test			P = 0.397N		P = 0.070N
Poly-3 vehicle pairwise comparison	P = 0.424		P = 0.444N	P = 0.480N	P = 0.173N
Skin (Site of Application): Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma					
Overall rate	34/36 (94.4%)	35/36 (97.2%)	33/36 (91.7%)	32/36 (88.9%)	33/36 (91.7%)
Adjusted rate	34/34.7 (97.9%)	35/35.0 (100.0%)	33/33.2 (99.4%)	32/32.9 (97.3%)	33/34.0 (97.0%)
Terminal rate	1/1 (100.0%)	2/2 (100.0%)	0/0	0/0	1/1 (100.0%)
First incidence (days)	215	208	237	194	243
Poly-3 trend test			P = 0.233N		P = 0.201N
Poly-3 vehicle pairwise comparison	P = 0.709		P = 1.000N	P = 0.567N	P = 0.483N
Skin (Site of Application): Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma					
Overall rate	35/36 (97.2%)	35/36 (97.2%)	33/36 (91.7%)	33/36 (91.7%)	34/36 (94.4%)
Adjusted rate	35/35.4 (98.8%)	35/35.0 (100.0%)	33/33.2 (99.4%)	33/33.3 (99.0%)	34/34.2 (99.4%)
Terminal rate	1/1 (100.0%)	2/2 (100.0%)	0/0	0/0	1/1 (100.0%)
First incidence (days)	215	208	237	194	243
Poly-3 trend test			P = 0.766N		P = 0.844N
Poly-3 vehicle pairwise comparison	P = 0.907		P = 1.000N	P = 0.994N	P = 1.000N

TABLE B2b
Statistical Analysis of Primary Neoplasms in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	6% Whole Leaf	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Skin (Site of Application): Squamous Cell Carcinoma					
Overall rate	18/36 (50.0%)	15/36 (41.7%)	17/36 (47.2%)	22/36 (61.1%)	19/36 (52.8%)
Adjusted rate	18/26.4 (68.2%)	15/23.7 (63.2%)	17/25.5 (66.6%)	22/27.8 (79.0%)	19/25.6 (74.2%)
Terminal rate	0/0	0/0	0/0	0/0	0/0
First incidence (days)	228	244	235	223	223
Poly-3 trend test		P = 0.043N		P = 0.191N	
Poly-3 vehicle pairwise comparison	P = 0.095N	P = 0.039N	P = 0.074N	P = 0.438N	P = 0.252N
Skin (Site of Application): Squamous Cell Carcinoma *in situ* and/or Squamous Cell Carcinoma					
Overall rate	32/36 (88.9%)	33/36 (91.7%)	32/36 (88.9%)	32/36 (88.9%)	32/36 (88.9%)
Adjusted rate	32/33.3 (96.2%)	33/33.9 (97.4%)	32/33.3 (96.2%)	32/33.4 (95.8%)	32/32.2 (99.4%)
Terminal rate	0/0	0/0	0/0	0/0	0/0
First incidence (days)	228	229	229	223	223
Poly-3 trend test		P = 0.170N		P = 0.669N	
Poly-3 vehicle pairwise comparison	P = 0.357N	P = 0.573N	P = 0.357N	P = 0.275N	P = 1.000N
Skin (Site of Application): Squamous Cell Papilloma, Squamous Cell Carcinoma *in situ*, and/or Squamous Cell Carcinoma					
Overall rate	34/36 (94.4%)	34/36 (94.4%)	34/36 (94.4%)	34/36 (94.4%)	33/36 (91.7%)
Adjusted rate	34/34.3 (99.2%)	34/34.4 (98.8%)	34/34.2 (99.3%)	34/34.6 (98.4%)	33/33.0 (99.9%)
Terminal rate	0/0	0/0	0/0	0/0	0/0
First incidence (days)	228	229	229	223	192
Poly-3 trend test		P = 0.780N		P = 0.912N	
Poly-3 vehicle pairwise comparison	P = 0.998N	P = 0.928N	P = 1.000N	P = 0.860N	P = 1.000N

[a] Number of neoplasm-bearing animals/number of animals with skin examined microscopically
[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] P values represent the results of linear trend tests. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. A negative trend is indicated by N.
[e] Poly-3 pairwise comparison to the control cream group. A lower incidence than that in the control cream group is indicated by N.
[f] Not applicable; no neoplasms in animal group
[g] Value of statistic cannot be computed.

TABLE B3a
Summary of the Incidence of Nonneoplastic Lesions in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 0.00 mJ • CIE/cm^2 SSL[a]

	No Cream	Control Cream	6% Aloe Gel	6% Whole Leaf	6% Decolorized Whole Leaf	74.6 µg/g Aloe-emodin
Disposition Summary						
Animals initially in study	36	36	36	36	36	36
Early deaths						
Moribund	5	7	2	2	3	
Natural deaths	1		2			1
Skin lesion greater than 5 mm		1		1		
Survivors						
Died last week of study			2		1	2
Terminal sacrifice	30	28	30	33	32	33
Animals examined microscopically	36	36	36	36	36	36
Integumentary System						
Skin, control	(36)	(36)	(36)	(36)	(36)	(36)
Abscess		2 (6%)	1 (3%)			1 (3%)
Cyst epithelial inclusion	3 (8%)	6 (17%)	3 (8%)	3 (8%)	4 (11%)	1 (3%)
Edema		1 (3%)			1 (3%)	
Hemorrhage		1 (3%)				
Hyperplasia, squamous	1 (3%)	5 (14%)	1 (3%)	4 (11%)		
Inflammation, pyogranulomatous		1 (3%)		1 (3%)		2 (6%)
Dermis, inflammation, chronic active	32 (89%)	32 (89%)	35 (97%)	36 (100%)	34 (94%)	36 (100%)
Subcutaneous tissue, abscess						1 (3%)
Skin, site of application	(36)	(36)	(36)	(36)	(36)	(36)
Abscess		1 (3%)				
Cyst epithelial inclusion	3 (8%)		2 (6%)	2 (6%)	5 (14%)	6 (17%)
Edema					1 (3%)	
Granuloma		1 (3%)				
Hyperplasia, squamous	1 (3%)	10 (28%)	13 (36%)	2 (6%)	1 (3%)	2 (6%)
Dermis, inflammation, chronic active	33 (92%)	36 (100%)	36 (100%)	36 (100%)	36 (100%)	36 (100%)

Systems Examined with No Lesions Observed
Alimentary System
Cardiovascular System
Endocrine System
General Body System
Genital System
Hematopoietic System
Musculoskeletal System
Nervous System
Respiratory System
Special Senses System
Urinary System

[a] Number of animals examined microscopically at the site and the number of animals with lesion. The number given with the lesion name indicates the number of that lesion per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.

TABLE B3b
Summary of the Incidence of Nonneoplastic Lesions in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 6.85 mJ • CIE/cm^2 SSL[a]

	No Cream
Disposition Summary	
Animals initially in study	36
Early deaths	
Moribund	1
Natural death	1
Skin lesion greater than 5 mm	7
Survivors	
Died last week of study	3
Terminal sacrifice	24
Animals examined microscopically	36
Integumentary System	
Skin, control	(36)
Cyst epithelial inclusion	7 (19%)
Hyperplasia, squamous	1 (3%)
Dermis, inflammation, chronic active	35 (97%)
Skin, site of application	(36)
Cyst epithelial inclusion	6 (17%)
Hyperplasia, squamous atypical, one, focal	10 (28%)
Hyperplasia, squamous atypical, two, focal	2 (6%)
Hyperplasia, squamous atypical, three, focal	1 (3%)
Hyperplasia, squamous atypical, five, focal	1 (3%)
Hyperplasia, squamous atypical, greater than five, focal	1 (3%)
Hyperplasia, squamous	32 (89%)
Dermis, inflammation, chronic active	36 (100%)

Systems Examined with No Lesions Observed
Alimentary System
Cardiovascular System
Endocrine System
General Body System
Genital System
Hematopoietic System
Musculoskeletal System
Nervous System
Respiratory System
Special Senses System
Urinary System

[a] Number of animals examined microscopically at the site and the number of animals with lesion. The number given with the lesion name indicates the number of that lesion per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.

TABLE B3c
Summary of the Incidence of Nonneoplastic Lesions in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL[a]

	No Cream	Control Cream	3% Aloe Gel	6% Aloe Gel	3% Whole Leaf
Disposition Summary					
Animals initially in study	36	36	36	36	36
Early deaths					
Moribund	1	1	4	4	2
Natural deaths				1	1
Skin lesion greater than 5 mm	34	33	32	31	31
Survivors					
Died last week of study					1
Terminal sacrifice	1	2			1
Animals examined microscopically	36	36	36	36	36
Integumentary System					
Skin, control	(36)	(36)	(36)	(36)	(36)
Abscess				3 (8%)	
Cyst epithelial inclusion	3 (8%)	3 (8%)	5 (14%)	5 (14%)	4 (11%)
Hemorrhage	1 (3%)				
Hyperplasia, squamous atypical, one, focal		1 (3%)	1 (3%)	1 (3%)	1 (3%)
Hyperplasia, squamous	12 (33%)	3 (8%)			
Inflammation, pyogranulomatous	1 (3%)		3 (8%)		
Dermis, inflammation, granulomatous					
Dermis, inflammation, chronic active	29 (81%)	29 (81%)	30 (83%)	35 (97%)	32 (89%)
Epidermis, necrosis	1 (3%)		2 (6%)		
Subcutaneous tissue, abscess		1 (3%)			
Subcutaneous tissue, inflammation, chronic	1 (3%)				
Skin, site of application	(36)	(36)	(36)	(36)	(36)
Cyst		1 (3%)			
Cyst epithelial inclusion	5 (14%)	5 (14%)	10 (28%)	5 (14%)	8 (22%)
Granuloma		1 (3%)		1 (3%)	
Hyperplasia, squamous atypical, one, focal	7 (19%)	8 (22%)	10 (28%)	10 (28%)	6 (17%)
Hyperplasia, squamous atypical, two, focal	8 (22%)	4 (11%)	8 (22%)	5 (14%)	10 (28%)
Hyperplasia, squamous atypical, three, focal	8 (22%)	7 (19%)	3 (8%)	5 (14%)	5 (14%)
Hyperplasia, squamous atypical, four, focal	3 (8%)	7 (19%)	3 (8%)	3 (8%)	4 (11%)
Hyperplasia, squamous atypical, five, focal	1 (3%)	4 (11%)	1 (3%)	4 (11%)	6 (17%)
Hyperplasia, squamous atypical, greater than five, focal	5 (14%)	5 (14%)	2 (6%)	2 (6%)	2 (6%)
Hyperplasia, squamous	36 (100%)	35 (97%)	34 (94%)	33 (92%)	35 (97%)
Inflammation, pyogranulomatous	2 (6%)	1 (3%)			2 (6%)
Dermis, inflammation, chronic active	36 (100%)	36 (100%)	36 (100%)	36 (100%)	35 (97%)
Epidermis, necrosis		1 (3%)	3 (8%)		1 (3%)

Systems Examined with No Lesions Observed
Alimentary System
Cardiovascular System
Endocrine System
General Body System
Genital System
Hematopoietic System
Musculoskeletal System
Nervous System
Respiratory System
Special Senses System
Urinary System

TABLE B3c
Summary of the Incidence of Nonneoplastic Lesions in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 13.70 mJ • CIE/cm^2 SSL

	6% Whole Leaf	3% Decolorized Whole Leaf	6% Decolorized Whole Leaf	7.46 µg/g Aloe-emodin	74.6 µg/g Aloe-emodin
Disposition Summary					
Animals initially in study	36	36	36	36	36
Early deaths					
Moribund	1		2	4	1
Natural deaths	3	2	1		2
Skin lesion greater than 5 mm	32	34	33	32	33
Survivors					
Died last week of study					
Terminal sacrifice					
Animals examined microscopically	36	36	36	36	36
Integumentary System					
Skin, control	(36)	(36)	(36)	(36)	(36)
Abscess				1 (3%)	
Cyst epithelial inclusion	3 (8%)	5 (14%)	2 (6%)	3 (8%)	1 (3%)
Hemorrhage	1 (3%)				
Hyperplasia, squamous atypical, one, focal			1 (3%)	1 (3%)	1 (3%)
Hyperplasia, squamous	1 (3%)		4 (11%)		
Inflammation, pyogranulomatous		1 (3%)	1 (3%)		1 (3%)
Dermis, inflammation, granulomatous	1 (3%)				
Dermis, inflammation, chronic active	30 (83%)	33 (92%)	29 (81%)	31 (86%)	30 (83%)
Epidermis, necrosis		1 (3%)			
Subcutaneous tissue, abscess					
Subcutaneous tissue, inflammation, chronic					
Skin, site of application	(36)	(36)	(36)	(36)	(36)
Cyst					
Cyst epithelial inclusion	1 (3%)	7 (19%)	5 (14%)	5 (14%)	
Granuloma	2 (6%)				
Hyperplasia, squamous atypical, one, focal	5 (14%)	12 (33%)	6 (17%)	8 (22%)	6 (17%)
Hyperplasia, squamous atypical, two, focal	7 (19%)	9 (25%)	6 (17%)	5 (14%)	5 (14%)
Hyperplasia, squamous atypical, three, focal	6 (17%)	9 (25%)	3 (8%)	8 (22%)	5 (14%)
Hyperplasia, squamous atypical, four, focal	6 (17%)	1 (3%)	4 (11%)	6 (17%)	5 (14%)
Hyperplasia, squamous atypical, five, focal	3 (8%)		1 (3%)	3 (8%)	4 (11%)
Hyperplasia, squamous atypical, greater than five, focal	2 (6%)	1 (3%)	7 (19%)	2 (6%)	2 (6%)
Hyperplasia, squamous	32 (89%)	34 (94%)	35 (97%)	34 (94%)	34 (94%)
Inflammation, pyogranulomatous			2 (6%)	1 (3%)	
Dermis, inflammation, chronic active	33 (92%)	35 (97%)	33 (92%)	35 (97%)	35 (97%)
Epidermis, necrosis		4 (11%)	3 (8%)	1 (3%)	1 (3%)

Systems Examined with No Lesions Observed
Alimentary System
Cardiovascular System
Endocrine System
General Body System
Genital System
Hematopoietic System
Musculoskeletal System
Nervous System
Respiratory System
Special Senses System
Urinary System

[a] Number of animals examined microscopically at the site and the number of animals with lesion. The number given with the lesion name indicates the number of that lesion per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.

TABLE B3d
Summary of the Incidence of Nonneoplastic Lesions in Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: 20.55 mJ • CIE/cm^2 SSL[a]

	No Cream
Disposition Summary	
Animals initially in study	36
Early deaths	
Moribund	1
Skin lesion greater than 5 mm	35
Animals examined microscopically	36
Integumentary System	
Skin, control	(36)
Cyst epithelial inclusion	1 (3%)
Dermis, inflammation, chronic active	19 (53%)
Skin, site of application	(36)
Cyst epithelial inclusion	2 (6%)
Hyperplasia, squamous atypical, one, focal	5 (14%)
Hyperplasia, squamous atypical, two, focal	6 (17%)
Hyperplasia, squamous atypical, three, focal	5 (14%)
Hyperplasia, squamous atypical, four, focal	7 (19%)
Hyperplasia, squamous atypical, five, focal	4 (11%)
Hyperplasia, squamous atypical, greater than five, focal	7 (19%)
Hyperplasia, squamous	34 (94%)
Dermis, inflammation, chronic active	32 (89%)
Epidermis, necrosis	3 (8%)

Systems Examined with No Lesions Observed
Alimentary System
Cardiovascular System
Endocrine System
General Body System
Genital System
Hematopoietic System
Musculoskeletal System
Nervous System
Respiratory System
Special Senses System
Urinary System

[a] Number of animals examined microscopically at the site and the number of animals with lesion. The number given with the lesion name indicates the number of that lesion per animal. On March 2, 2005, pathological examinations of tissues or organs other than skin were suspended.

APPENDIX C
BODY WEIGHT DATA

TABLE C1
Mean Body Weights and Survival of Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

Weeks on Study	0.00 mJ • CIE/cm^2 SSL			6.85 mJ • CIE/cm^2 SSL			
	No. of Survivors	Av. Wt.[a] (g)	P Value[b]	No. of Survivors	Av. Wt. (g)	P Value[c]	Wt. (% of Controls)
0	36	29.9 ± 0.4	0.563	36	29.7 ± 0.4	0.630	99.1
4	36	33.6 ± 0.4	0.794	36	33.4 ± 0.4	0.805	99.6
8	36	35.4 ± 0.4	0.511	36	35.1 ± 0.4	0.574	99.1
12	36	36.0 ± 0.4	0.947	36	35.7 ± 0.5	0.631	99.2
16	36	36.4 ± 0.4	0.644	36	36.3 ± 0.5	0.839	99.7
20	36	36.7 ± 0.4	0.818	33	36.7 ± 0.5	0.791	100.0
24	36	36.8 ± 0.4	0.323	33	36.9 ± 0.5	0.708	100.2
28	36	37.3 ± 0.4	0.843	32	37.5 ± 0.6	0.632	100.6
32	36	37.3 ± 0.4	0.524	30	37.5 ± 0.5	0.309	100.6
36	36	37.2 ± 0.4	0.976	29	37.5 ± 0.5	0.201	100.9
40	36	37.1 ± 0.4		29	37.1 ± 0.5	0.456	100.1
44	36	37.1 ± 0.5		29	36.5 ± 0.5	0.651	98.2
48	35	37.3 ± 0.5		26	37.0 ± 0.6	0.844	99.0
52	35	37.8 ± 0.5		22	37.1 ± 0.6	0.536	98.2
Mean for weeks							
0-52		36.1 ± 0.4			36.0 ± 0.5		99.6

Weeks on Study	13.70 mJ • CIE/cm^2 SSL				20.55 mJ • CIE/cm^2 SSL			
	No. of Survivors	Av. Wt. (g)	P Value[c]	Wt. (% of Controls)	No. of Survivors	Av. Wt. (g)	P Value[c]	Wt. (% of Controls)
0	36	29.5 ± 0.4	0.452	98.52	36	29.6 ± 0.4	0.604	99.0
4	36	33.1 ± 0.3	0.414	98.57	36	33.8 ± 0.4	0.642	100.8
8	36	34.4 ± 0.3	0.105	97.29	36	35.2 ± 0.4	0.736	99.4
12	36	35.1 ± 0.3	0.150	97.64	36	36.1 ± 0.4	0.801	100.4
16	36	35.5 ± 0.3	0.125	97.51	35	36.3 ± 0.4	0.967	99.7
20	36	36.0 ± 0.3	0.290	98.30	35	37.0 ± 0.4	0.494	100.9
24	36	36.8 ± 0.3	0.961	99.92	35	37.5 ± 0.4	0.237	101.7
28	36	37.2 ± 0.3	0.843	99.69	28	37.1 ± 0.4	0.987	99.5
32	35	37.3 ± 0.4	0.939	100.12	11	38.2 ± 0.8	0.333	102.5
36	31	37.5 ± 0.4	0.667	100.71	1	33.4	0.840	89.9
40	17	37.8 ± 0.6	0.502	102.02	0			
44	3	38.3 ± 1.2	0.217	103.15	0			
48	0				0			
52	0				0			
Mean for weeks								
0-52		35.7 ± 0.4		99.5		35.4 ± 0.4		99.4

[a] Mean ± standard error
[b] P value represents linear trend in body weight with increasing simulated solar light (SSL) level.
[c] P value represents pairwise comparison of body weight to 0.00 mJ • CIE/cm^2 SSL group.

TABLE C2
Mean Body Weights and Survival of Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: No Cream

Weeks on Study	0.00 mJ • CIE/cm^2 SSL			6.85 mJ • CIE/cm^2 SSL			
	No. of Survivors	Av. Wt.[a] (g)	P Value[b]	No. of Survivors	Av. Wt. (g)	P Value[c]	Wt. (% of Controls)
0	36	23.6 ± 0.4	0.720	36	23.9 ± 0.2	0.608	101.2
4	36	25.9 ± 0.4	0.324	36	26.1 ± 0.3	0.647	101.0
8	36	27.3 ± 0.4	0.647	36	27.8 ± 0.3	0.308	102.1
12	36	28.2 ± 0.4	0.675	36	29.0 ± 0.3	0.156	102.8
16	36	28.9 ± 0.4	0.821	36	29.7 ± 0.3	0.152	102.7
20	35	29.6 ± 0.4	0.541	36	30.4 ± 0.3	0.166	102.6
24	34	30.2 ± 0.5	0.428	36	31.1 ± 0.4	0.125	103.0
28	34	30.6 ± 0.4	0.439	36	31.7 ± 0.4	0.078	103.4
32	34	30.7 ± 0.4	0.115	36	31.9 ± 0.4	0.053	103.7
36	34	30.9 ± 0.5	0.052	36	31.9 ± 0.4	0.064	103.5
40	34	31.2 ± 0.5		36	32.0 ± 0.4	0.215	102.4
44	33	31.8 ± 0.5		34	32.3 ± 0.5	0.203	101.6
48	31	31.7 ± 0.5		32	33.0 ± 0.6	0.013*	104.0
52	30	31.9 ± 0.6		24	33.3 ± 0.7	0.003*	104.5
Mean for weeks							
0-52		29.5 ± 0.4			30.3 ± 0.4		102.7

Weeks on Study	13.70 mJ • CIE/cm^2 SSL				20.55 mJ • CIE/cm^2 SSL			
	No. of Survivors	Av. Wt. (g)	P Value[c]	Wt. (% of Controls)	No. of Survivors	Av. Wt. (g)	P Value[c]	Wt. (% of Controls)
0	36	23.7 ± 0.3	0.974	100.08	36	23.5 ± 0.2	0.828	99.5
4	36	26.3 ± 0.3	0.432	101.67	36	26.4 ± 0.3	0.352	102.0
8	36	27.7 ± 0.3	0.443	101.55	36	27.6 ± 0.3	0.570	101.2
12	36	28.5 ± 0.3	0.547	101.18	36	28.6 ± 0.3	0.475	101.4
16	36	29.1 ± 0.3	0.672	100.81	36	29.2 ± 0.3	0.565	101.1
20	36	29.9 ± 0.3	0.636	100.93	36	30.1 ± 0.4	0.343	101.8
24	36	30.6 ± 0.3	0.524	101.35	36	30.9 ± 0.4	0.257	102.3
28	36	31.0 ± 0.3	0.548	101.25	32	31.2 ± 0.5	0.229	101.9
32	35	31.4 ± 0.4	0.219	102.27	22	31.6 ± 0.5	0.059	102.7
36	30	31.5 ± 0.4	0.186	101.98	3	30.4 ± 1.2	0.030*	98.4
40	22	31.7 ± 0.4	0.297	101.59	0			
44	11	32.2 ± 0.7	0.318	101.29	0			
48	2	36.7 ± 1.7	0.001*	115.71	0			
52	1	35.8	0.001*	112.13	0			
Mean for weeks								
0-52		30.4 ± 0.5		103.127		28.9 ± 0.5		101.2

* Significant at P≤0.05
[a] Mean ± standard error
[b] P value represents linear trend in body weight with increasing simulated solar light (SSL) level.
[c] P value represents pairwise comparison of body weight to 0.00 mJ • CIE/cm^2 SSL group.

TABLE C3
Mean Body Weights and Survival of Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and No Cream

Weeks on Study	Control Cream No. of Survivors	Control Cream Av. Wt.[a] (g)	No Cream No. of Survivors	No Cream Av. Wt. (g)	No Cream P Value[b]	No Cream Wt. (% of Controls)
0.00 mJ • CIE/cm^2 SSL						
0	36	29.7 ± 0.4	36	29.9 ± 0.4	0.704	100.9
4	36	33.2 ± 0.4	36	33.6 ± 0.4	0.639	101.0
8	36	35.0 ± 0.4	36	35.4 ± 0.4	0.605	101.0
12	36	35.9 ± 0.4	36	36.0 ± 0.4	0.868	100.3
16	35	36.6 ± 0.4	36	36.4 ± 0.4	0.965	99.7
20	34	36.9 ± 0.4	36	36.7 ± 0.4	0.879	99.4
24	33	37.1 ± 0.4	36	36.8 ± 0.4	0.917	99.2
28	33	37.5 ± 0.5	36	37.3 ± 0.4	0.971	99.4
32	33	37.6 ± 0.5	36	37.3 ± 0.4	0.885	99.2
36	33	37.5 ± 0.5	36	37.2 ± 0.4	0.902	99.3
40	33	37.2 ± 0.5	36	37.1 ± 0.4	0.863	99.8
44	31	36.9 ± 0.5	36	37.1 ± 0.5	0.357	100.7
48	31	36.5 ± 0.6	35	37.3 ± 0.5	0.078	102.2
52	30	36.8 ± 0.7	35	37.8 ± 0.5	0.032*	102.6
Mean for weeks						
0-52		36.0 ± 0.5		36.1 ± 0.4		100.3
13.70 mJ • CIE/cm^2 SSL						
0	34	29.6 ± 0.4	36	29.5 ± 0.4	0.842	99.6
4	34	32.1 ± 0.4	36	33.1 ± 0.3	0.093	103.1
8	34	33.7 ± 0.4	36	34.4 ± 0.3	0.227	102.1
12	33	34.6 ± 0.5	36	35.1 ± 0.3	0.419	101.4
16	33	35.4 ± 0.4	36	35.5 ± 0.3	0.810	100.5
20	33	36.0 ± 0.4	36	36.0 ± 0.3	0.925	100.2
24	33	36.9 ± 0.4	36	36.8 ± 0.3	0.851	99.8
28	33	37.3 ± 0.5	36	37.2 ± 0.3	0.834	99.7
32	32	37.3 ± 0.5	35	37.3 ± 0.4	0.859	100.0
36	21	37.7 ± 0.7	31	37.5 ± 0.4	0.740	99.4
40	10	37.5 ± 0.8	17	37.8 ± 0.6	0.688	101.0
44	4	35.8 ± 1.0	3	38.3 ± 1.2	0.634	106.9
48	3	36.6 ± 0.9	0			
52	0		0			
Mean for weeks						
0-52		28.7		35.7 ± 0.4		101.1

* Significant at P≤0.05
[a] Mean ± standard error
[b] P value represents pairwise comparison to control cream at the same exposure to SSL.

TABLE C4
Mean Body Weights and Survival of Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and No Cream

Weeks on Study	Control Cream No. of Survivors	Control Cream Av. Wt.[a] (g)	No Cream No. of Survivors	No Cream Av. Wt. (g)	P Value[b]	Wt. (% of Controls)
0.00 mJ • CIE/cm^2 SSL						
0	34	23.7 ± 0.4	36	23.6 ± 0.4	0.901	99.7
4	34	25.7 ± 0.4	36	25.9 ± 0.4	0.791	100.7
8	34	27.0 ± 0.4	36	27.3 ± 0.4	0.693	101.0
12	34	28.1 ± 0.4	36	28.2 ± 0.4	0.919	100.2
16	34	28.9 ± 0.4	36	28.9 ± 0.4	0.932	100.2
20	34	29.5 ± 0.4	35	29.6 ± 0.4	0.879	100.3
24	34	30.1 ± 0.4	34	30.2 ± 0.5	0.840	100.3
28	34	30.6 ± 0.5	34	30.6 ± 0.4	0.849	100.2
32	34	30.8 ± 0.5	34	30.7 ± 0.4	0.983	99.9
36	33	30.9 ± 0.5	34	30.9 ± 0.5	0.940	99.9
40	31	31.2 ± 0.6	34	31.2 ± 0.5	0.702	100.1
44	31	31.8 ± 0.6	33	31.8 ± 0.5	0.917	99.9
48	29	31.8 ± 0.6	31	31.7 ± 0.5	0.655	99.9
52	28	32.0 ± 0.5	30	31.9 ± 0.6	0.272	99.6
Mean for weeks						
0-52		29.4 ± 0.5		29.5 ± 0.4		100.1
13.70 mJ • CIE/cm^2 SSL						
0	35	23.7 ± 0.3	36	23.7 ± 0.3	0.921	99.8
4	35	25.7 ± 0.3	36	26.3 ± 0.3	0.158	102.5
8	35	26.6 ± 0.3	36	27.7 ± 0.3	0.019*	104.0
12	35	27.5 ± 0.3	36	28.5 ± 0.3	0.031*	103.6
16	35	28.1 ± 0.3	36	29.1 ± 0.3	0.019*	103.8
20	35	28.6 ± 0.3	36	29.9 ± 0.3	0.007*	104.3
24	35	29.5 ± 0.3	36	30.6 ± 0.3	0.017*	103.7
28	35	30.4 ± 0.3	36	31.0 ± 0.3	0.143	102.2
32	32	30.5 ± 0.3	35	31.4 ± 0.4	0.049*	103.0
36	26	31.0 ± 0.4	30	31.5 ± 0.4	0.166	101.4
40	16	31.1 ± 0.5	22	31.7 ± 0.4	0.310	102.1
44	5	30.1 ± 0.9	11	32.2 ± 0.7	0.255	107.0
48	2	29.0 ± 2.0	2	36.7 ± 1.7	0.001*	126.7
52	2	29.8 ± 2.1	1	35.8	0.005*	120.1
Mean for weeks						
0-52		28.7		30.4 ± 0.5		106.0

* Significant at $P \leq 0.05$
[a] Mean ± standard error
[b] P value represents pairwise comparison to control cream at the same exposure to SSL.

TABLE C5
Mean Body Weights and Survival of Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe Gel Creams

Weeks on Study	Control Cream			3% Aloe Gel Cream[a]				6% Aloe Gel Cream			
	No. of Survivors	Av. Wt.[b] (g)	P Value[c]	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)
0.00 mJ • CIE/cm^2 SSL											
0	36	29.7 ± 0.4						36	29.5 ± 0.4	0.746	99.3
4	36	33.2 ± 0.4						36	32.3 ± 0.4	0.142	97.1
8	36	35.0 ± 0.4						36	33.8 ± 0.4	0.066	96.5
12	36	35.9 ± 0.4						36	34.6 ± 0.4	0.065	96.6
16	35	36.6 ± 0.4						36	35.2 ± 0.3	0.054	96.2
20	34	36.9 ± 0.4						35	35.8 ± 0.4	0.124	96.9
24	33	37.1 ± 0.4						35	36.4 ± 0.4	0.463	98.1
28	33	37.5 ± 0.5						35	36.8 ± 0.4	0.402	98.0
32	33	37.6 ± 0.5						35	37.1 ± 0.4	0.656	98.7
36	33	37.5 ± 0.5						34	36.8 ± 0.3	0.641	98.2
40	33	37.2 ± 0.5						34	36.4 ± 0.4	0.584	98.1
44	31	36.9 ± 0.5						34	36.3 ± 0.4	0.960	98.5
48	31	36.5 ± 0.6						34	36.6 ± 0.4	0.366	100.3
52	30	36.8 ± 0.7						34	37.1 ± 0.5	0.187	100.8
Mean for weeks											
0-52		36.0 ± 0.5							35.3 ± 0.4		98.1
13.70 m • CIE/cm^2 SSL											
0	34	29.6 ± 0.4	0.492	36	29.6 ± 0.3	0.983	99.95	36	29.2 ± 0.4	0.492	98.5
4	34	32.1 ± 0.4	0.267	36	32.9 ± 0.4	0.226	102.44	36	32.8 ± 0.4	0.267	102.2
8	34	33.7 ± 0.4	0.275	36	34.4 ± 0.4	0.273	102.10	36	34.4 ± 0.4	0.275	102.1
12	33	34.6 ± 0.5	0.270	35	35.1 ± 0.4	0.530	101.35	36	35.4 ± 0.4	0.270	102.1
16	33	35.4 ± 0.4	0.321	35	35.8 ± 0.4	0.624	101.06	36	36.0 ± 0.4	0.321	101.9
20	33	36.0 ± 0.4	0.320	35	36.1 ± 0.5	0.863	100.47	36	36.6 ± 0.4	0.320	101.8
24	33	36.9 ± 0.4	0.803	34	36.9 ± 0.4	0.668	100.11	36	37.1 ± 0.4	0.803	100.5
28	33	37.3 ± 0.5	0.726	33	37.2 ± 0.4	0.651	99.95	36	37.5 ± 0.4	0.726	100.7
32	32	37.3 ± 0.5	0.889	30	37.0 ± 0.5	0.347	99.07	32	37.6 ± 0.4	0.889	100.6
36	21	37.7 ± 0.7	0.997	23	36.5 ± 0.7	0.095	96.91	26	37.7 ± 0.5	0.997	99.9
40	10	37.5 ± 0.8	0.298	10	35.7 ± 1.6	0.013*	95.20	13	36.3 ± 0.6	0.298	97.0
44	4	35.8 ± 1.0	0.089	5	32.3 ± 3.5	0.001*	90.06	5	34.8 ± 0.6	0.089	97.2
48	3	36.6 ± 0.9		0				0			
52	0			0				0			
Mean for weeks											
0-52		35.4			35.0 ± 0.8		99.06		35.5 ± 0.4		100.4

* Significant at $P \leq 0.05$
[a] Article not tested at 0.00 mJ • CIE/cm^2 SSL
[b] Mean ± standard error
[c] P value represents linear trend test.
[d] P value represents pairwise comparison to control cream at the same exposure to SSL.

TABLE C6
Mean Body Weights and Survival of Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe Gel Creams

Weeks on Study	Control Cream			3% Aloe Gel Cream[a]				6% Aloe Gel Cream			
	No. of Survivors	Av. Wt.[b] (g)	P Value[c]	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)
0.00 mJ • CIE/cm^2 SSL											
0	34	23.7 ± 0.4						36	23.8 ± 0.4	0.953	100.2
4	34	25.7 ± 0.4						36	25.0 ± 0.4	0.325	97.4
8	34	27.0 ± 0.4						36	25.8 ± 0.4	0.076	95.5
12	34	28.1 ± 0.4						36	26.7 ± 0.4	0.039*	95.0
16	34	28.9 ± 0.4						36	27.4 ± 0.4	0.038*	95.1
20	34	29.5 ± 0.4						36	28.2 ± 0.4	0.059	95.6
24	34	30.1 ± 0.4						35	29.0 ± 0.5	0.095	96.3
28	34	30.6 ± 0.5						35	29.5 ± 0.5	0.118	96.6
32	34	30.8 ± 0.5						35	29.8 ± 0.4	0.161	97.0
36	33	30.9 ± 0.5						35	30.0 ± 0.4	0.171	97.0
40	31	31.2 ± 0.6						34	30.1 ± 0.5	0.177	96.4
44	31	31.8 ± 0.6						33	30.5 ± 0.5	0.112	95.8
48	29	31.8 ± 0.6						33	31.1 ± 0.5	0.184	97.9
52	28	32.0 ± 0.5						30	31.3 ± 0.6	0.164	97.9
Mean for weeks											
0-52		29.4 ± 0.5							28.4 ± 0.5		96.7
13.70 mJ • CIE/cm^2 SSL											
0	35	23.7	0.691	35	23.4 ± 0.3	0.531	98.58	36	23.9 ± 0.3	0.691	100.9
4	35	25.7 ± 0.3	0.482	35	25.6 ± 0.3	0.986	99.96	36	26.0 ± 0.4	0.482	101.5
8	35	26.6 ± 0.3	0.129	35	27.1 ± 0.3	0.371	101.81	36	27.4 ± 0.4	0.129	103.1
12	35	27.5 ± 0.3	0.240	35	28.1 ± 0.3	0.278	102.12	36	28.1 ± 0.4	0.240	102.3
16	35	28.1 ± 0.3	0.209	35	28.7 ± 0.4	0.233	102.28	35	28.8 ± 0.5	0.209	102.5
20	35	28.6 ± 0.3	0.089	35	29.6 ± 0.5	0.079	103.30	34	29.6 ± 0.5	0.089	103.5
24	35	29.5 ± 0.3	0.146	34	30.0 ± 0.4	0.170	101.57	34	30.4 ± 0.5	0.146	102.9
28	35	30.4 ± 0.3	0.411	33	30.4 ± 0.4	0.692	99.97	33	30.8 ± 0.5	0.411	101.5
32	32	30.5 ± 0.3	0.721	33	30.5 ± 0.4	0.832	99.83	33	30.8 ± 0.5	0.721	100.9
36	26	31.0 ± 0.3	0.704	29	30.9 ± 0.5	0.755	99.48	27	30.9 ± 0.5	0.704	99.6
40	16	31.1 ± 0.4	0.761	20	30.5 ± 0.5	0.745	98.19	18	30.6 ± 0.6	0.761	98.4
44	5	30.1 ± 0.5	0.914	7	30.7 ± 0.8	0.588	102.17	5	31.2 ± 0.5	0.914	103.7
48	2	29.0 ± 0.9		0				0			
52	2	29.8 ± 2.0		0				0			
Mean for weeks											
0-52		28.7 ± 2.1			28.8 ± 0.4		100.77		29.1 ± 0.5		101.7

* Significant at $P \leq 0.05$
[a] Article not tested at 0.00 mJ • CIE/cm^2 SSL
[b] Mean ± standard error
[c] P value represents linear trend test.
[d] P value represents pairwise comparison to control cream at the same exposure to SSL.

TABLE C7
Mean Body Weights and Survival of Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Whole Leaf Creams

Weeks on Study	Control Cream No. of Survivors	Av. Wt.[b]	P Value[c]	3% Whole Leaf Cream[a] No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)	6% Whole Leaf Cream No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)
0.00 mJ • CIE/cm^2 SSL											
0	36	29.7 ± 0.4						36	29.2 ± 0.4	0.457	98.3
4	36	33.2 ± 0.4						36	31.9 ± 0.4	0.044*	95.9
8	36	35.0 ± 0.4						36	33.4 ± 0.4	0.013*	95.2
12	36	35.9 ± 0.4						36	34.1 ± 0.4	0.010*	95.2
16	35	36.6 ± 0.4						36	34.5 ± 0.4	0.004*	94.4
20	34	36.9 ± 0.4						36	35.3 ± 0.4	0.034*	95.8
24	33	37.1 ± 0.4						36	36.1 ± 0.4	0.246	97.3
28	33	37.5 ± 0.5						35	36.2 ± 0.4	0.098	96.5
32	33	37.6 ± 0.5						34	36.6 ± 0.4	0.198	97.5
36	33	37.5 ± 0.5						34	36.7 ± 0.4	0.300	97.9
40	33	37.2 ± 0.5						34	36.6 ± 0.4	0.523	98.6
44	31	36.9 ± 0.5						33	36.7 ± 0.4	0.902	99.4
48	31	36.5 ± 0.6						33	36.8 ± 0.5	0.516	100.8
52	30	36.8 ± 0.7						31	37.6 ± 0.5	0.174	102.1
Mean for weeks											
0-52		36.0 ± 0.5							35.1 ± 0.4		97.5
13.70 mJ • CIE/cm^2 SSL											
0	34	29.6 ± 0.4	0.701	35	30.2 ± 0.3	0.361	101.88	35	29.9 ± 0.5	0.701	100.8
4	34	32.1 ± 0.4	0.475	35	33.2 ± 0.4	0.059	103.61	35	32.5 ± 0.4	0.475	101.4
8	34	33.7 ± 0.4	0.387	35	34.5 ± 0.4	0.196	102.35	35	34.3 ± 0.4	0.387	101.6
12	33	34.6 ± 0.5	0.337	35	35.3 ± 0.4	0.273	101.99	35	35.2 ± 0.4	0.337	101.8
16	33	35.4 ± 0.4	0.765	35	35.9 ± 0.4	0.448	101.36	34	35.6 ± 0.4	0.765	100.7
20	33	36.0 ± 0.4	0.799	35	36.4 ± 0.4	0.479	101.25	34	36.2 ± 0.4	0.799	100.6
24	33	36.9 ± 0.4	0.776	35	36.9 ± 0.4	0.976	100.10	34	36.8 ± 0.5	0.776	99.7
28	33	37.3 ± 0.5	0.803	35	37.6 ± 0.4	0.613	100.87	34	37.2 ± 0.4	0.803	99.7
32	32	37.3 ± 0.5	0.650	29	37.9 ± 0.3	0.714	101.59	33	37.2 ± 0.4	0.650	99.5
36	21	37.7 ± 0.7	0.564	24	37.6 ± 0.4	0.574	99.74	28	37.4 ± 0.5	0.564	99.3
40	10	37.5 ± 0.8	0.661	16	37.3 ± 0.7	0.348	99.60	13	38.2 ± 0.8	0.661	101.9
44	4	35.8 ± 1.0	0.573	6	35.2 ± 1.4	0.018*	98.33	5	37.6 ± 0.7	0.573	104.9
48	3	36.6 ± 0.9	0.921	2	29.3 ± 2.4	0.001*	80.07	1	40.6	0.921	111.0
52	0			1	32.2			1	41.1		
Mean for weeks											
0-52		35.4 ± 0.6			35.0 ± 0.6		99.44		36.4 ± 0.5		101.8

* Significant at $P \leq 0.05$
[a] Article not tested at 0.00 mJ • CIE/cm^2 SSL
[b] Mean ± standard error
[c] P value represents linear trend test.
[d] P value represents pairwise comparison to control cream at the same exposure to SSL.

TABLE C8
Mean Body Weights and Survival of Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Whole Leaf Creams

Weeks on Study	Control Cream			3% Whole Leaf Cream[a]				6% Whole Leaf Cream			
	No. of Survivors	Av. Wt.[b] (g)	P Value[c]	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)
0.00 mJ • CIE/cm^2 SSL											
0	34	23.7 ± 0.4						36	23.7 ± 0.3	0.987	100.1
4	34	25.7 ± 0.4						36	25.2 ± 0.3	0.445	98.0
8	34	27.0 ± 0.4						36	26.2 ± 0.3	0.242	97.1
12	34	28.1 ± 0.4						36	27.1 ± 0.3	0.136	96.4
16	34	28.9 ± 0.4						36	27.8 ± 0.4	0.126	96.4
20	34	29.5 ± 0.4						36	28.6 ± 0.4	0.187	97.0
24	34	30.1 ± 0.4						36	29.4 ± 0.4	0.270	97.5
28	34	30.6 ± 0.5						35	29.9 ± 0.5	0.330	97.9
32	34	30.8 ± 0.5						35	30.2 ± 0.5	0.428	98.3
36	33	30.9 ± 0.5						35	30.3 ± 0.5	0.421	98.2
40	31	31.2 ± 0.6						34	30.1 ± 0.4	0.361	96.5
44	31	31.8 ± 0.6						34	30.3 ± 0.4	0.126	95.3
48	29	31.8 ± 0.6						34	30.8 ± 0.5	0.127	96.8
52	28	32.0 ± 0.5						33	31.3 ± 0.5	0.121	97.6
Mean for weeks											
0-52		29.4 ± 0.5							28.6 ± 0.4		97.4
13.70 mJ • CIE/cm^2 SSL											
0	35	23.7 ± 0.3	0.862	36	23.6 ± 0.3	0.805	99.51	35	23.8 ± 0.2	0.862	100.3
4	35	25.7 ± 0.3	0.418	36	25.8 ± 0.3	0.803	100.45	35	26.0 ± 0.2	0.418	101.5
8	35	26.6 ± 0.3	0.285	36	26.9 ± 0.3	0.587	100.95	35	27.1 ± 0.3	0.285	101.9
12	35	27.5 ± 0.3	0.411	36	27.6 ± 0.3	0.861	100.30	35	27.9 ± 0.3	0.411	101.4
16	35	28.1 ± 0.3	0.516	36	28.2 ± 0.3	0.794	100.43	35	28.4 ± 0.3	0.516	101.1
20	35	28.6 ± 0.3	0.170	36	29.2 ± 0.3	0.221	101.99	35	29.3 ± 0.3	0.170	102.3
24	35	29.5 ± 0.3	0.212	35	29.9 ± 0.4	0.345	101.29	35	30.1 ± 0.3	0.212	102.0
28	35	30.4 ± 0.3	0.764	34	30.4 ± 0.4	0.832	100.09	35	30.2 ± 0.3	0.764	99.5
32	32	30.5 ± 0.3	0.322	34	30.6 ± 0.4	0.773	100.37	35	30.1 ± 0.4	0.322	98.6
36	26	31.0 ± 0.4	0.276	33	31.1 ± 0.4	0.777	100.18	30	30.6 ± 0.4	0.276	98.5
40	16	31.1 ± 0.5	0.217	23	31.2 ± 0.5	0.722	100.52	23	31.1 ± 0.4	0.217	100.2
44	5	30.1 ± 0.9	0.994	12	31.8 ± 0.6	0.410	105.80	8	32.2 ± 0.8	0.994	107.2
48	2	29.0 ± 2.0	0.754	3	33.2 ± 2.3	0.008*	114.59	1	31.6	0.754	109.1
52	2	29.8 ± 2.1		1	31.1	0.102	104.52	0			
Mean for weeks											
0-52		28.7 ± 0.6			29.3 ± 0.5		102.21		29.1 ± 0.3		101.8

* Significant at $P \leq 0.05$
[a] Article not tested at 0.00 mJ • CIE/cm^2 SSL
[b] Mean ± standard error
[c] P value represents linear trend test.
[d] P value represents pairwise comparison to control cream at the same exposure to SSL.

TABLE C9
Mean Body Weights and Survival of Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Decolorized Whole Leaf Creams

Weeks on Study	Control Cream			3% Decolorized Whole Leaf Cream[a]				6% Decolorized Whole Leaf Cream			
	No. of Survivors	Av. Wt.[b] (g)	P Value[c]	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)
0.00 mJ • CIE/cm^2 SSL											
0	36	29.7 ± 0.4						35	29.0 ± 0.4	0.310	97.7
4	36	33.2 ± 0.4						35	32.2 ± 0.4	0.135	96.9
8	36	35.0 ± 0.4						35	34.1 ± 0.4	0.168	97.3
12	36	35.9 ± 0.4						35	34.9 ± 0.4	0.171	97.4
16	35	36.6 ± 0.4						35	35.3 ± 0.4	0.096	96.6
20	34	36.9 ± 0.4						35	35.5 ± 0.4	0.056	96.1
24	33	37.1 ± 0.4						35	35.7 ± 0.4	0.077	96.2
28	33	37.5 ± 0.5						35	36.1 ± 0.4	0.075	96.2
32	33	37.6 ± 0.5						34	36.0 ± 0.4	0.044*	95.8
36	33	37.5 ± 0.5						33	36.0 ± 0.4	0.044*	96.2
40	33	37.2 ± 0.5						33	35.6 ± 0.4	0.029*	95.8
44	31	36.9 ± 0.5						33	35.3 ± 0.5	0.044*	95.6
48	31	36.5 ± 0.6						30	35.7 ± 0.5	0.365	97.9
52	30	36.8 ± 0.7						30	36.2 ± 0.6	0.601	98.3
Mean for weeks											
0-52		36.0 ± 0.5							34.8 ± 0.4		96.7
13.70 mJ • CIE/cm^2 SSL											
0	34	29.6 ± 0.4	0.648	35	29.5 ± 0.3	0.800	99.52	36	29.4 ± 0.3	0.648	99.1
4	34	32.1 ± 0.4	0.052	35	33.1 ± 0.4	0.074	103.16	36	33.2 ± 0.3	0.052	103.4
8	34	33.7 ± 0.4	0.111	35	34.7 ± 0.4	0.092	102.83	36	34.6 ± 0.3	0.111	102.7
12	33	34.6 ± 0.5	0.218	35	35.3 ± 0.4	0.233	102.01	36	35.3 ± 0.3	0.218	102.1
16	33	35.4 ± 0.4	0.349	35	36.4 ± 0.4	0.092	102.76	36	35.9 ± 0.3	0.349	101.5
20	33	36.0 ± 0.4	0.491	35	36.4 ± 0.4	0.427	101.30	35	36.4 ± 0.3	0.491	101.3
24	33	36.9 ± 0.4	0.873	35	36.7 ± 0.4	0.748	99.55	35	36.9 ± 0.3	0.873	99.9
28	33	37.3 ± 0.5	0.943	34	37.5 ± 0.4	0.671	100.63	35	37.3 ± 0.3	0.943	100.1
32	32	37.3 ± 0.5	0.930	30	37.2 ± 0.4	0.935	99.75	35	37.5 ± 0.3	0.930	100.6
36	21	37.7 ± 0.7	0.689	21	37.6 ± 0.7	0.958	99.64	25	37.3 ± 0.5	0.689	99.0
40	10	37.5 ± 0.8	0.601	11	37.6 ± 0.5	0.588	100.23	8	36.7 ± 1.0	0.601	98.0
44	4	35.8 ± 1.0	0.893	2	36.7 ± 0.3	0.222	102.31	3	37.9 ± 0.2	0.893	105.6
48	3	36.6 ± 0.9		0				0			
52	0			0				0			
Mean for weeks											
0-52		35.4 ± 0.6			35.7 ± 0.4		101.1		35.7 ± 0.4		101.1

* Significant at $P \leq 0.05$
[a] Article not tested at 0.00 mJ • CIE/cm^2 SSL
[b] Mean ± standard error
[c] P value represents linear trend test.
[d] P value represents pairwise comparison to control cream at the same exposure to SSL.

TABLE C10
Mean Body Weights and Survival of Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Decolorized Whole Leaf Creams

Weeks on Study	Control Cream			3% Decolorized Whole Leaf Cream[a]				6% Decolorized Whole Leaf Cream			
	No. of Survivors	Av. Wt.[b]	P Value[c]	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)
0.00 mJ • CIE/cm^2 SSL											
0	34	23.7 ± 0.4						36	23.9 ± 0.3	0.751	100.8
4	34	25.7 ± 0.4						36	25.9 ± 0.2	0.733	100.8
8	34	27.0 ± 0.4						36	27.3 ± 0.3	0.645	101.0
12	34	28.1 ± 0.4						36	28.2 ± 0.3	0.845	100.4
16	34	28.9 ± 0.4						35	28.6 ± 0.3	0.799	99.1
20	34	29.5 ± 0.4						35	28.9 ± 0.3	0.422	98.0
24	34	30.1 ± 0.4						35	29.3 ± 0.3	0.216	97.2
28	34	30.6 ± 0.5						35	29.9 ± 0.3	0.315	97.7
32	34	30.8 ± 0.5						35	30.1 ± 0.3	0.334	97.8
36	33	30.9 ± 0.5						35	30.1 ± 0.3	0.265	97.4
40	31	31.2 ± 0.6						35	30.1 ± 0.4	0.181	96.5
44	31	31.8 ± 0.6						34	30.6 ± 0.4	0.073	96.2
48	29	31.8 ± 0.6						34	30.9 ± 0.4	0.039*	97.2
52	28	32.0 ± 0.5						32	31.0 ± 0.4	0.008*	96.6
Mean for weeks											
0-52		29.4 ± 0.5							28.9 ± 0.3		98.4
13.70 mJ • CIE/cm^2 SSL											
0	35	23.7 ± 0.3	0.710	36	24.2 ± 0.3	0.240	102.19	36	23.9 ± 0.2	0.710	100.7
4	35	25.7 ± 0.3	0.101	36	26.5 ± 0.3	0.046*	103.45	36	26.4 ± 0.2	0.101	102.8
8	35	26.6 ± 0.3	0.046*	36	27.9 ± 0.3	0.006*	104.62	36	27.5 ± 0.2	0.046*	103.3
12	35	27.5 ± 0.3	0.123	36	28.6 ± 0.3	0.013*	104.02	36	28.2 ± 0.2	0.123	102.5
16	35	28.1 ± 0.3	0.126	35	29.0 ± 0.3	0.033*	103.36	36	28.8 ± 0.2	0.126	102.4
20	35	28.6 ± 0.3	0.158	35	29.5 ± 0.3	0.044*	103.11	36	29.3 ± 0.3	0.158	102.2
24	35	29.5 ± 0.3	0.628	35	30.0 ± 0.3	0.289	101.58	35	29.6 ± 0.3	0.628	100.2
28	35	30.4 ± 0.3	0.998	35	30.5 ± 0.3	0.837	100.29	34	30.1 ± 0.3	0.998	99.0
32	32	30.5 ± 0.3	0.335	35	30.9 ± 0.4	0.467	101.20	34	30.7 ± 0.3	0.335	100.6
36	26	31.0 ± 0.4	0.298	31	31.5 ± 0.4	0.307	101.43	32	31.2 ± 0.3	0.298	100.5
40	16	31.1 ± 0.5	0.775	13	31.4 ± 0.8	0.583	100.87	20	31.3 ± 0.5	0.775	100.6
44	5	30.1 ± 0.9	0.671	1	25.7	0.586	85.55	5	31.8 ± 0.8	0.671	105.7
48	2	29.0 ± 2.0	0.156	1	26.4	0.661	91.15	1	33.6	0.156	115.9
52	2	29.8 ± 2.1		0				0			
Mean for weeks											
0-52		28.7 ± 0.6			28.6 ± 0.4		100.22		29.4 ± 0.3		102.8

* Significant at $P \leq 0.05$
[a] Article not tested at 0.00 mJ • CIE/cm^2 SSL
[b] Mean ± standard error
[c] P value represents linear trend test.
[d] P value represents pairwise comparison to control cream at the same exposure to SSL.

TABLE C11
Mean Body Weights and Survival of Male Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe-emodin Creams

Weeks on Study	Control Cream No. of Survivors	Av. Wt.[b] (g)	P Value[c]	7.46 µg/g Aloe-emodin Cream[a] No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)	74.6 µg/g Aloe-emodin Cream No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)
0.00 mJ • CIE/cm^2 SSL											
0	36	29.7 ± 0.4						36	29.0 ± 0.4	0.327	97.6
4	36	33.2 ± 0.4						36	32.1 ± 0.5	0.130	96.6
8	36	35.0 ± 0.4						36	34.0 ± 0.5	0.170	97.1
12	36	35.9 ± 0.4						35	35.1 ± 0.5	0.219	98.0
16	35	36.6 ± 0.4						35	35.7 ± 0.5	0.185	97.5
20	34	36.9 ± 0.4						34	35.9 ± 0.5	0.176	97.3
24	33	37.1 ± 0.4						34	36.3 ± 0.4	0.357	97.9
28	33	37.5 ± 0.5						34	36.8 ± 0.4	0.381	98.0
32	33	37.6 ± 0.5						34	36.9 ± 0.4	0.396	98.1
36	33	37.5 ± 0.5						34	36.3 ± 0.5	0.160	97.0
40	33	37.2 ± 0.5						32	36.2 ± 0.6	0.114	97.4
44	31	36.9 ± 0.5						30	37.0 ± 0.5	0.398	100.2
48	31	36.5 ± 0.6						28	36.7 ± 0.5	0.826	100.6
52	30	36.8 ± 0.7						28	36.8 ± 0.5	0.769	100.0
Mean for weeks											
0-52		36.0 ± 0.5							35.3 ± 0.5		98.1
13.70 mJ • CIE/cm^2 SSL											
0	34	29.6 ± 0.4	0.548	35	29.3 ± 0.4	0.604	98.95	36	29.3 ± 0.5	0.548	98.8
4	34	32.1 ± 0.4	0.928	35	32.6 ± 0.3	0.407	101.55	36	32.0 ± 0.4	0.928	99.8
8	34	33.7 ± 0.4	0.822	35	33.8 ± 0.3	0.856	100.32	36	33.9 ± 0.3	0.822	100.4
12	33	34.6 ± 0.5	0.641	35	34.8 ± 0.3	0.834	100.42	36	34.9 ± 0.3	0.641	100.9
16	33	35.4 ± 0.4	0.823	33	35.5 ± 0.3	0.834	100.28	36	35.5 ± 0.3	0.823	100.4
20	33	36.0 ± 0.4	0.806	33	36.0 ± 0.3	0.746	100.04	36	36.1 ± 0.3	0.806	100.5
24	33	36.9 ± 0.4	0.833	33	36.6 ± 0.3	0.435	99.25	36	36.8 ± 0.4	0.833	99.7
28	33	37.3 ± 0.5	0.855	33	37.2 ± 0.3	0.653	99.76	35	37.3 ± 0.4	0.855	100.2
32	32	37.3 ± 0.5	0.716	32	37.5 ± 0.3	0.901	100.58	34	37.8 ± 0.5	0.716	101.3
36	21	37.7 ± 0.7	0.653	24	37.7 ± 0.5	0.898	100.05	24	38.3 ± 0.5	0.653	101.6
40	10	37.5 ± 0.8	0.845	16	36.6 ± 0.8	0.234	97.61	10	37.8 ± 0.6	0.845	101.0
44	4	35.8 ± 1.0	0.106	4	37.0 ± 1.1	0.388	103.17	1	36.1	0.106	100.6
48	3	36.6 ± 0.9	0.070	0				1	35.4	0.070	96.7
52	0			0				1	33.9		
Mean for weeks											
0-52		35.4 ± 0.6			35.4 ± 0.4		100.2		35.4 ± 0.4		100.1

[a] Article not tested at 0.00 mJ • CIE/cm^2 SSL
[b] Mean ± standard error
[c] P value represents linear trend test.
[d] P value represents pairwise comparison to control cream at the same exposure to SSL.

TABLE C12
Mean Body Weights and Survival of Female Mice in the 1-Year Simulated Solar Light Study of Aloe vera: Control Cream and Aloe-emodin Creams

Weeks on Study	Control Cream			7.46 µg/g Aloe-emodin Cream[a]				74.6 µg/g Aloe-emodin Cream			
	No. of Survivors	Av. Wt.[b]	P Value[c]	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)	No. of Survivors	Av. Wt. (g)	P Value[d]	Wt. (% of Controls)
0.00 mJ • CIE/cm^2 SSL											
0	34	23.7 ± 0.4	0.819					36	23.9 ± 0.3	0.819	100.6
4	34	25.7 ± 0.4	0.943					36	25.7 ± 0.3	0.943	100.2
8	34	27.0 ± 0.4	0.971					36	27.0 ± 0.3	0.971	100.1
12	34	28.1 ± 0.4	0.933					36	28.0 ± 0.3	0.933	99.8
16	34	28.9 ± 0.4	0.721					36	28.6 ± 0.3	0.721	99.2
20	34	29.5 ± 0.4	0.449					36	29.0 ± 0.3	0.449	98.4
24	34	30.1 ± 0.4	0.209					36	29.3 ± 0.3	0.209	97.4
28	34	30.6 ± 0.5	0.363					36	30.0 ± 0.3	0.363	98.1
32	34	30.8 ± 0.5	0.478					36	30.3 ± 0.3	0.478	98.6
36	33	30.9 ± 0.5	0.481					36	30.4 ± 0.3	0.481	98.5
40	31	31.2 ± 0.6	0.458					36	30.6 ± 0.3	0.458	97.9
44	31	31.8 ± 0.6	0.272					36	30.9 ± 0.4	0.272	97.3
48	29	31.8 ± 0.6	0.133					36	31.1 ± 0.4	0.133	98.0
52	28	32.0 ± 0.5	0.025*					33	31.2 ± 0.4	0.025*	97.4
Mean for weeks											
0-52		29.4 ± 0.5							29.0 ± 0.3		98.7
13.70 mJ • CIE/cm^2 SSL											
0	35	23.7 ± 0.3	0.726	36	23.5 ± 0.2	0.596	99.00	34	23.9 ± 0.2	0.726	100.7
4	35	25.7 ± 0.3	0.800	36	25.2 ± 0.3	0.354	98.39	34	25.8 ± 0.3	0.800	100.5
8	35	26.6 ± 0.3	0.978	36	26.2 ± 0.3	0.384	98.54	34	26.6 ± 0.3	0.978	100.0
12	35	27.5 ± 0.3	0.877	36	27.1 ± 0.3	0.370	98.55	34	27.6 ± 0.3	0.877	100.3
16	35	28.1 ± 0.3	0.307	36	28.1 ± 0.3	0.962	99.92	34	28.5 ± 0.3	0.307	101.7
20	35	28.6 ± 0.3	0.163	36	28.9 ± 0.4	0.613	100.79	33	29.2 ± 0.3	0.163	102.1
24	35	29.5 ± 0.3	0.757	36	29.4 ± 0.4	0.775	99.57	33	29.6 ± 0.3	0.757	100.4
28	35	30.4 ± 0.3	0.950	36	30.0 ± 0.4	0.359	98.66	32	30.3 ± 0.3	0.950	99.7
32	32	30.5 ± 0.3	0.354	34	30.8 ± 0.4	0.780	100.90	31	30.9 ± 0.3	0.354	101.3
36	26	31.0 ± 0.4	0.281	29	31.3 ± 0.5	0.599	100.76	29	31.5 ± 0.4	0.281	101.4
40	16	31.1 ± 0.5	0.549	12	32.2 ± 0.9	0.360	103.45	23	31.6 ± 0.4	0.549	101.5
44	5	30.1 ± 0.9	0.676	0				6	32.0 ± 0.7	0.676	106.3
48	2	29.0 ± 2.0		0				0			
52	2	29.8 ± 2.1		0				0			
Mean for weeks											
0-52		28.7 ± 0.6			28.4 ± 0.4		99.87		29.0 ± 0.3		101.3

* Significant at P≤0.05
[a] Article not tested at 0.00 mJ • CIE/cm^2 SSL
[b] Mean ± standard error
[c] P value represents linear trend test.
[d] P value represents pairwise comparison to control cream at the same exposure to SSL.

APPENDIX D
CHEMICAL CHARACTERIZATION AND DOSE FORMULATION STUDIES

CHEMICAL CHARACTERIZATION AND DOSE FORMULATION STUDIES

Procurement and Characterization

Aloe vera Plant Extracts

Three lyophilized *Aloe barbadensis* Miller (Aloe vera) test articles were obtained from Pangea Phytoceuticals, Inc. (Harlingen, TX), as aloe gel, whole leaf, and decolorized whole leaf extracts. The aloe gel extract (lot numbers 010510AG, 030109AG, and 032903AG) consisted of the inner leaf gel of hand-filleted Aloe vera leaves with the pulp removed. No further treatments were performed on this material. The whole leaf extract (lot numbers 013008ND and 030109ND) was produced by grinding whole Aloe vera leaves and treating the slurry with cellulase (23 mg/L) to remove the rind components and to maximize yields. This product contained the inner leaf gel and the aloe latex. The decolorized whole leaf extract (lot numbers 013008AC and 030221D) was prepared in an identical manner as the whole leaf extract previously described, with the exception that the slurry was further treated with activated carbon (1.0%, w/w) to remove the latex anthraquinone components from the extract. The different lots of *Aloe barbadensis* test articles were combined and used in the 1-year study. Analyses to determine the homogeneity, content of malic acid, aloin A, and aloe-emodin, and the average molecular weight of each of the Aloe vera plant extracts were performed by the Chemistry Support Unit at the National Center for Toxicological Research (NCTR, Jefferson, AR). Glycosyl linkage analyses of alcohol-insoluble fractions of each of the Aloe vera plant extracts were performed by the Complex Carbohydrate Research Center at the University of Georgia (Athens, GA). Reports on analyses performed in support of the study on the effect of Aloe vera on the photocarcinogenicity of simulated solar light are on file at the National Center for Toxicological Research.

High-performance liquid chromatography (HPLC) was used to determine the homogeneity and concentrations of malic acid (system A; Table D1), aloin A (system B), and aloe-emodin (system D) in the aloe gel, whole leaf, and decolorized whole leaf test articles. The average molecular weight of the polysaccharides in each Aloe vera plant extract was determined using size exclusion HPLC by system C with photodiode array, refractive index, and Rayleigh laser light-scattering detection; instrument calibration was verified using certified dextran polysaccharide standard reference materials obtained from Polymer Standard Service, Gmbh (Mainz, Germany). For glycosyl linkage analysis, samples of the Aloe vera plant extracts were permethylated, depolymerized, reduced, and acetylated; the resultant partially methylated alditol acetates were analyzed using gas chromatography-mass spectrometry on a Hewlett-Packard system (Palo Alto, CA) with a Supelco 2330 30 m capillary column (Supelco, Bellefonte, PA) as described by York *et al*. (1986).

Homogeneity of each of the three Aloe vera plant extracts was confirmed. Average concentrations of malic acid and aloin A were determined from nine samples of each test article. The average concentrations of aloe-emodin were determined from three replicate samples of each test article:

	Average Concentration (µg/g)		
Bulk Aloe vera Plant Extract	***Malic Acid***	***Aloin A***	***Aloe-emodin***
Aloe gel	233,000	2,710	Not detected
Whole leaf	219,000	16,500	17
Decolorized whole leaf	246,000	82	Not detected

Average molecular weights of 2,290,000, 51,500, and 66,600 Kdaltons were found for the polysaccharides of the aloe gel, whole leaf, and decolorized whole leaf extracts, respectively. 4-Mannose was determined to be the major glycosyl residue in the aloe gel test article; 4-glucose was also found. 4-Mannose was also the predominant

glycosyl residue in the whole leaf test article; in addition, terminal glucose was a major component, and terminal mannose and 4-glucose were also found. 4-Mannose and terminal glucose were the primary residues in the decolorized whole leaf test article, and terminal fructose was also seen.

To ensure stability, the bulk aloe gel, whole leaf, and decolorized whole leaf test articles were stored in color-coded, high-density polyethylene pails with snap lids at approximately –20° C. At the end of the study, HPLC analyses of the three bulk chemicals using systems A (quantitating malic acid) and B (quantitating aloin A) were performed by the Chemistry Support Unit at NCTR; no degradation was detected.

Aloe-emodin

Aloe-emodin was obtained from Sigma-Aldrich (St. Louis, MO) in one lot (013K1075). The purity of a 52 µg/mL solution of aloe-emodin in acetonitrile:0.5 M sulfuric acid (75:25) was determined using HPLC by system D; the average purity for three replicate samples was 99.8%. Aloe-emodin was stored in amber vials at approximately 4° C.

Control Cream

The base cream used as the control cream was obtained from Cosmetech Laboratories, Inc. (Fairfield, NJ), in four lots (CLI 120604-G, -H, -I, and -J) that were not combined and were used in the 1-year study. The control cream was formulated according to the recipe described in Table 1 and the lots did not vary in the supplier/lot number of the ingredients. The absence of malic acid, aloin A, and aloe-emodin from the control cream was confirmed for each lot using HPLC by systems A, B, and C, respectively. All lots had a specific gravity of 0.962 that remained constant, with pH values of approximately 6.0 and a mean viscosity of 11,400 centipoise. The base cream was formulated with a 10% "hole" for the incorporation of the test articles, and it was stored in high-density polyethylene bottles with screw-cap lids at approximately 4° C.

Preparation and Analysis of Dose Formulations

The dose formulations were prepared twice each week by dissolving the appropriate amount of each of the four test articles in distilled, deionized water and then thoroughly mixing these solutions with the control cream to provide 10% of the final weight of the dosed creams (Table D2). The dose formulations were dispensed into glass scintillation vials, sealed with Teflon®-lined screw caps, and stored at approximately 4° C for up to 3 days.

Homogeneity studies (a 6% dose formulation of aloe gel and a 78 µg/g dose formulation of aloe-emodin) and stability studies (3% dose formulations for each of the three Aloe vera plant extracts and 7.46 and 74.6 µg/g aloe-emodin dose formulations) were performed by the Chemistry Support Unit at NCTR using HPLC. HPLC systems A (quantitating malic acid) and B (quantitating aloin A) were used for analysis of the dose formulations of the Aloe vera plant extracts, and system D (quantitating aloe-emodin) was used for the analysis of the dose formulations of aloe-emodin. Homogeneity was confirmed, and stability was confirmed for up to 3 days for samples stored in glass vials sealed with Teflon®-lined screw caps at 4° C.

Weekly dose certifications of the dosed cream formulations of the Aloe vera plant extracts and aloe-emodin were conducted by the Chemistry Support Unit at NCTR using HPLC by the same systems used for the dose formulation homogeneity and stability studies. Quantitation of malic acid and aloin A was used for certification of the dose formulation concentrations of the three Aloe vera plant extract test articles; target concentrations of malic acid and aloin A in these dose formulations are indicated in Tables D3, D4, and D5. Dose formulations of aloe-emodin were directly measured for concentrations of the test chemical; target concentrations were equivalent to the dose concentrations of aloe-emodin (Table D6). While an acceptability range of ± 10% of target concentration was desirable, the goal of the dose certification was to enable calculation of the dose being administered to an animal at a specific time point. In general, the dosed cream formulations were within the desirable acceptability range.

TABLE D1
High-Performance Liquid Chromatography Systems Used in the 1-Year Simulated Solar Light Study of Aloe vera[a]

Detection System	Column	Solvent System
System A Ultraviolet photodiode array with monitoring at 210 nm	Dionex Ion Pac®ICE-AS6, 250 mm × 9 mm, 8 µm particle size (Dionex Corporation, Sunnyvale, CA)	0.2 mM sulfuric acid, pH 2.2, isocratic; flow rate 0.8 mL/minute
System B Ultraviolet photodiode array with monitoring at 360 nm	Phenomenex Prodigy™ ODS3, 250 mm × 4.6 mm, 5 µm particle size (Phenomenex, Torrance, CA)	Acetonitrile:0.01 M sodium phosphate monobasic (25:75), pH 4.4, isocratic; flow rate 1.0 mL/minute
System C Ultraviolet photodiode array with monitoring at 360 nm	YMC-Pack Diol-300, 300 mm × 8 mm, 5 µm particle size, 30 nm pore size (YMC Co., Ltd., Kyoto, Japan)	50 mM NaH_2PO_4:0.05% NaN_3 (25:75); flow rate 1.0 mL/minute
System D Ultraviolet/visible photodiode array scanning from 200 to 800 nm or with monitoring at 428 nm	Phenomenex Prodigy™ ODS3, 250 mm × 4.6 mm, 5 µm particle size (Phenomenex)	Acetonitrile:0.01 M sodium phosphate monobasic (80:20), pH 4.4, isocratic; flow rate 1.0 mL/minute

[a] High-performance liquid chromatographs were manufactured by Waters Corporation (Milford, MA).

TABLE D2
Preparation and Storage of Dose Formulations in the 1-Year Simulated Solar Light Study of Aloe vera

Preparation
Premix solutions of the four test articles dissolved in distilled deionized water were prepared, and these solutions were then mixed with the control cream to provide 10% of the final weight of the dosed cream formulations. The dose formulations were prepared twice each week.

Chemical Lot Numbers

Aloe Gel
010510AG
030109AG
032903AG

Whole Leaf
013008ND
030109ND

Decolorized Whole Leaf
013008AC
030221D

Aloe-emodin
013K1075

Control Cream
CLI 120604-G
CLI 120604-H
CLI 120604-I
CLI 120604-J

Maximum Storage Time
3 days

Storage Conditions
Stored in glass scintillation vials with Teflon®-lined screw caps at approximately 4° C

Study Laboratory
National Center for Toxicological Research (Jefferson, AR)

TABLE D3
Malic Acid and Aloin A Concentrations in the Aloe Gel Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Dose Formulation Concentration (%)	Malic Acid			Aloin A		
		Target Concentration (µg/g)	Determined Concentration[a] (µg/g)	Difference from Target (%)	Target Concentration (µg/g)	Determined Concentration[a] (µg/g)	Difference from Target (%)
May 12, 2003	3	6,990	6,930 ± 40	–1	81.3	92 ± 2	+13
May 14, 2003	6	13,980	14,170 ± 310	+1	162.6	159 ± 2	–2
May 21, 2003	3	6,990	8,860 ± 80	+27	81.3	97 ± 0	+19
May 19, 2003	6	13,980	15,140 ± 0	+8	162.6	176 ± 1	+8
May 26, 2003	3	6,990	6,470 ± 10	–7	81.3	83 ± 0	+2
May 28, 2003	6	13,980	14,410 ± 180	+3	162.6	166 ± 2	+2
June 2, 2003	3	6,990	6,660 ± 120	–5	81.3	79 ± 4	–3
June 4, 2003	6	13,980	13,940 ± 230	0	162.6	167 ± 2	+3
June 11, 2003	3	6,990	6,500 ± 50	–7	81.3	85 ± 0	+5
June 9, 2003	6	13,980	14,230 ± 80	+2	162.6	155 ± 4	–5
June 18, 2003	3	6,990	6,550 ± 50	–6	81.3	84 ± 1	+3
June 16, 2003	6	13,980	15,370 ± 40	+10	162.6	159 ± 2	–2
June 25, 2003	3	6,990	6,390 ± 170	–9	81.3	86.5 ± 1.6	+6
June 23, 2003	6	13,980	14,160 ± 60	+1	162.6	160 ± 4	–2
June 30, 2003	3	6,990	6,400 ± 80	–8	81.3	79 ± 0	–3
July 2, 2003	6	13,980	13,160 ± 10	–6	162.6	147 ± 0	–10
July 7, 2003	3	6,990	6,390 ± 10	–9	81.3	81.4 ± 0.4	0
July 9, 2003	6	13,980	13,310 ± 30	–5	162.6	164 ± 2	+1
July 16, 2003	3	6,990	6,730 ± 30	–4	81.3	77.1 ± 1.6	–5
July 14, 2003	6	13,980	13,790 ± 170	–1	162.6	165 ± 1	+1
July 21, 2003	3	6,990	6,350 ± 50	–9	81.3	75.9 ± 0.9	–7
July 23, 2003	6	13,980	13,720 ± 40	–2	162.6	149 ± 2	–8
July 30, 2003	3	6,990	6,870 ± 90	–2	81.3	87 ± 1	+7
July 28, 2003	6	13,980	13,520 ± 10	–3	162.6	145 ± 1	–11
August 4, 2003	3	6,990	6,930 ± 70	–1	81.3	95.9 ± 0.2	+18
August 6, 2003	6	13,980	13,630 ± 110	–3	162.6	140 ± 1	–14
August 13, 2003	3	6,990	6,480 ± 10	–7	81.3	82 ± 0	+1
August 11, 2003	6	13,980	14,040 ± 0	0	162.6	141 ± 1	–13
August 20, 2003	3	6,990	6,660 ± 70	–5	81.3	77.8 ± 0.8	–4
August 18, 2003	6	13,980	14,190 ± 160	+2	162.6	164 ± 1	+1
August 25, 2003	3	6,990	6,420 ± 140	–8	81.3	75.6 ± 1.8	–7
August 27, 2003	6	13,980	13,450 ± 100	–4	162.6	152 ± 1	–7
September 1, 2003	3	6,990	6,630 ± 160	–5	81.3	80.1 ± 0.3	–1
September 3, 2003	6	13,980	14,160 ± 320	+1	162.6	157 ± 4	–3
September 10, 2003	3	6,990	6,460 ± 40	–8	81.3	73.8 ± 2.8	–9
September 8, 2003	6	13,980	14,420 ± 290	+3	162.6	155 ± 5	–5

TABLE D3
Malic Acid and Aloin A Concentrations in the Aloe Gel Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Dose Formulation Concentration (%)	Malic Acid			Aloin A		
		Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)	Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)
September 17, 2003	3	6,990	6,590 ± 20	–6	81.3	79.0 ± 1.1	–3
September 15, 2003	6	13,980	6,910 ± 150	–51	162.6	83.7 ± 3.2	–49
September 24, 2003	3	6,990	3,120 ± 10	–55	81.3	39.8 ± 0.3	–51
September 22, 2003	6	13,980	13,560 ± 200	–3	162.6	149 ± 2	–8
September 29, 2003	3	6,990	6,310 ± 170	–10	81.3	79.5 ± 1.0	–2
October 1, 2003	6	13,980	14,400 ± 260	+3	162.6	147 ± 3	–10
October 6, 2003	3	6,990	6,800 ± 110	–3	81.3	91.2 ± 0.8	+12
October 8, 2003	6	13,980	13,020 ± 490	–7	162.6	141 ± 3	–13
October 13, 2003	3	6,990	6,920 ± 140	–1	81.3	97.0 ± 1.1	+19
October 15, 2003	6	13,980	13,900 ± 20	–1	162.6	145 ± 1	–11
October 22, 2003	3	6,990	6,930 ± 290	–1	81.3	71.5 ± 0.9	–12
October 20, 2003	6	13,980	15,090 ± 1,710	+8	162.6	175 ± 27	+8
October 27, 2003	3	6,990	5,790 ± 250	–17	81.3	75.3 ± 0.8	–7
October 29, 2003	6	13,980	14,710 ± 190	+5	162.6	143 ± 2	–12
November 3, 2003	3	6,990	6,260 ± 140	–10	81.3	64.9 ± 0.7	–20
November 5, 2003	6	13,980	13,470 ± 30	–4	162.6	156 ± 1	–4
November 10, 2003	3	6,990	5,880 ± 130	–16	81.3	74.9 ± 0.1	–8
November 12, 2003	6	13,980	13,450 ± 290	–4	162.6	141 ± 5	–13
November 17, 2003	3	6,990	5,890 ± 190	–16	81.3	74.1 ± 0.1	–9
November 19, 2003	6	13,980	13,030 ± 200	–7	162.6	148 ± 2	–9
November 26, 2003	3	6,990	6,600 ± 160	–6	81.3	81.9 ± 0.7	+1
November 24, 2003	6	13,980	13,500 ± 230	–3	162.6	142 ± 0	–13
December 3, 2003	3	6,990	6,660 ± 100	–5	81.3	83.2 ± 0.8	–1
December 1, 2003	6	13,980	13,620 ± 50	–3	162.6	151 ± 0	–7
December 8, 2003	3	6,990	6,090 ± 110	–13	81.3	83.7 ± 0.1	+3
December 10, 2003	6	13,980	14,200 ± 30	+2	162.6	148 ± 1	–9
December 15, 2003	3	6,990	6,740 ± 30	–4	81.3	83.8 ± 0.1	+3
December 17, 2003	6	13,980	14,170 ± 20	+1	162.6	153 ± 1	–6
December 24, 2003	3	6,990	6,500 ± 60	–7	81.3	78.8 ± 1.2	–3
December 22, 2003	6	13,980	14,660 ± 240	+5	162.6	154 ± 5	–5
December 29, 2003	3	6,990	6,310 ± 60	–10	81.3	73.3 ± 1.3	–10
December 31, 2003	6	13,980	13,570 ± 440	–3	162.6	163 ± 0	0
January 5, 2004	3	6,990	6,870 ± 1,190	–2	81.3	83.5 ± 14.2	+3
January 7, 2004	6	13,980	16,920 ± 150	+21	162.6	195 ± 2	+20

TABLE D3
Malic Acid and Aloin A Concentrations in the Aloe Gel Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Dose Formulation Concentration (%)	Malic Acid			Aloin A		
		Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)	Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)
January 12, 2004	3	6,990	6,530 ± 40	–7	81.3	80.8 ± 0.7	–1
January 14, 2004	6	13,980	11,850 ± 20	–15	162.6	115 ± 3	–29
January 19, 2004	3	6,990	5,880 ± 80	–16	81.3	75.6 ± 0.4	–7
January 21, 2004	6	13,980	11,230 ± 10	–20	162.6	130 ± 1	–20
January 26, 2004	3	6,990	6,130 ± 70	–12	81.3	83.7 ± 1.0	+3
January 28, 2004	6	13,980	13,730 ± 150	–2	162.6	150 ± 1	–8
February 2, 2004	3	6,990	6,000 ± 70	–14	81.3	84.7 ± 2.1	+4
February 4, 2004	6	13,980	12,780 ± 170	–9	162.6	160 ± 4	–2
February 11, 2004	3	6,990	6,390 ± 80	–9	81.3	80.6 ± 1.6	–1
February 9, 2004	6	13,980	13,320 ± 160	–5	162.6	166 ± 1	+2
February 18, 2004	3	6,990	6,070 ± 60	–13	81.3	73.4 ± 0.4	–10
February 16, 2004	6	13,980	13,180 ± 500	–6	162.6	154 ± 5	–5
February 23, 2004	3	6,990	6,170 ± 120	–12	81.3	71.3 ± 2.2	–12
February 25, 2004	6	13,980	12,470 ± 190	–11	162.6	128 ± 3	–21
March 1, 2004	3	6,990	5,890 ± 60	–16	81.3	77.5 ± 1.2	–5
March 3, 2004	6	13,980	12,880 ± 320	–8	162.6	150 ± 3	–8
March 8, 2004	3	6,990	7,000 ± 770	0	81.3	84.4 ± 10.7	+4
March 10, 2004	6	13,980	14,960 ± 420	+7	162.6	194 ± 6	+19
March 15, 2004	3	6,990	6,310 ± 0	–10	81.3	76.8 ± 0.4	–6
March 17, 2004	6	13,980	17,970 ± 2,450	+29	162.6	213 ± 33	+31

[a] Results of duplicate analyses

TABLE D4
Malic Acid and Aloin A Concentrations in the Whole Leaf Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Dose Formulation Concentration (%)	Malic Acid			Aloin A		
		Target Concentration (µg/g)	Determined Concentration[a] (µg/g)	Difference from Target (%)	Target Concentration (µg/g)	Determined Concentration[a] (µg/g)	Difference from Target (%)
May 12, 2003	3	6,570	6,240 ± 20	–5	495	420 ± 310	–15
May 14, 2003	6	13,140	12,750 ± 60	–3	990	986 ± 1	0
May 21, 2003	3	6,570	6,650 ± 20	+1	495	500 ± 3	+1
May 19, 2003	6	13,140	13,380 ± 0	+2	990	1,040 ± 0	+5
May 26, 2003	3	6,570	6,110 ± 80	–7	495	494 ± 1	0
May 28, 2003	6	13,140	12,950 ± 80	–1	990	888 ± 23	–10
June 2, 2003	3	6,570	6,560 ± 170	0	495	524 ± 31	+6
June 4, 2003	6	13,140	12,800 ± 30	–3	990	1,040 ± 6	+5
June 11, 003	3	6,570	6,100 ± 90	–7	495	497 ± 5	0
June 9, 2003	6	13,140	13,240 ± 60	+1	990	993 ± 0	0
June 18, 2003	3	6,570	6,610 ± 30	+1	495	483 ± 4	–2
June 16, 2003	6	13,140	13,990 ± 50	+6	990	1,000 ± 7	+1
June 25, 2003	3	6,570	12,320 ± 50	+88	495	1,030 ± 4	+108
June 23, 2003	6	13,140	13,260 ± 20	+1	990	1,040 ± 6	+5
June 30, 2003	3	6,570	6,050 ± 50	–8	495	513 ± 3	+4
July 2, 2003	6	13,140	12,390 ± 0	–6	990	990 ± 8	0
July 7, 2003	3	6,570	6,250 ± 30	–5	495	498 ± 2	+1
July 9, 2003	6	13,140	12,480 ± 10	–5	990	1,040 ± 0	+5
July 16, 2003	3	6,570	6,220 ± 10	–5	495	456 ± 0	–8
July 14, 2003	6	13,140	12,710 ± 10	–3	990	1,028 ± 14	+4
July 21, 2003	3	6,570	6,040 ± 90	–8	495	456 ± 7	–8
July 23, 2003	6	13,140	12,700 ± 90	–3	990	952 ± 6	–4
July 30, 2003	3	6,570	6,370 ± 20	–3	495	494 ± 9	0
July 28, 2003	6	13,140	12,660 ± 70	–4	990	932 ± 1	–6
August 4, 2003	3	6,570	6,370 ± 0	–3	495	533 ± 3	+8
August 6, 2003	6	13,140	12,520 ± 150	–5	990	925 ± 21	–7
August 13, 2003	3	6,570	6,380 ± 20	–3	495	509 ± 0	+3
August 11, 2003	6	13,140	12,970 ± 20	–1	990	1,000 ± 10	+1
August 20, 2003	3	6,570	6,380 ± 30	–3	495	482 ± 7	–3
August 18, 2003	6	13,140	13,870 ± 30	+6	990	998 ± 19	+1
August 25, 2003	3	6,570	6,340 ± 0	–4	495	491 ± 3	–1
August 27, 2003	6	13,140	12,480 ± 40	–5	990	966 ± 3	–2
September 1, 2003	3	6,570	5,920 ± 10	–10	495	448 ± 8	–9
September 3, 2003	6	13,140	12,910 ± 90	–2	990	910 ± 1	–8

TABLE D4
Malic Acid and Aloin A Concentrations in the Whole Leaf Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Dose Formulation Concentration (%)	Malic Acid			Aloin A		
		Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)	Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)
September 10, 2003	3	6,570	6,260 ± 30	–5	495	460 ± 6	–7
September 8, 2003	6	13,140	12,830 ± 100	–2	990	955 ± 7	–4
September 17, 2003	3	6,570	6,100 ± 50	–7	495	490 ± 4	–1
September 15, 2003	6	13,140	12,250 ± 90	–7	990	938 ± 3	–5
September 24, 2003	3	6,570	6,450 ± 30	–2	495	452 ± 3	–9
September 22, 2003	6	13,140	12,400 ± 120	–6	990	941 ± 11	–5
September 29, 2003	3	6,570	5,940 ± 80	–10	495	469 ± 0	–5
October 1, 2003	6	13,140	12,460 ± 40	–5	990	857 ± 16	–13
October 6, 2003	3	6,570	6,480 ± 0	–1	495	575 ± 2	+16
October 8, 2003	6	13,140	12,460 ± 80	–5	990	921 ± 0	–7
October 13, 2003	3	6,570	6,580 ± 60	0	495	456 ± 3	–8
October 15, 2003	6	13,140	11,410 ± 130	–13	990	826 ± 10	–17
October 22, 2003	3	6,570	6,720 ± 60	+2	495	451 ± 3	–9
October 20, 2003	6	13,140	12,500 ± 80	–5	990	1,010 ± 10	+2
October 27, 2003	3	6,570	6,000 ± 210	–9	495	439 ± 9	–11
October 29, 2003	6	13,140	13,120 ± 240	0	990	900 ± 7	–9
November 3, 2003	3	6,570	2,690 ± 190	–59	495	211 ± 6	–57
November 5, 2003	6	13,140	12,420 ± 90	–5	990	948 ± 7	–4
November 10, 2003	3	6,570	5,680 ± 170	–14	495	485 ± 6	–2
November 12, 2003	6	13,140	12,450 ± 230	–5	990	914 ± 1	–8
November 17, 2003	3	6,570	5,760 ± 0.0	–12	495	467 ± 2	–6
November 19, 2003	6	13,140	12,310 ± 310	–6	990	908 ± 3	–8
November 26, 2003	3	6,570	6,070 ± 220	–8	495	491 ± 3	–1
November 24, 2003	6	13,140	13,060 ± 730	–1	990	943 ± 4	–5
December 3, 2003	3	6,570	6,100 ± 190	–7	495	492 ± 5	+2
December 1, 2003	6	13,140	12,950 ± 610	–1	990	938 ± 8	–5
December 8, 2003	3	6,570	5,730 ± 20	–13	495	499 ± 3	+1
December 10, 2003	6	13,140	13,640 ± 180	+4	990	961 ± 3	–3
December 15, 2003	3	6,570	6,590 ± 490	0	495	508 ± 2	+3
December 17, 2003	6	13,140	12,190 ± 50	–7	990	975 ± 5	–2
December 24, 2003	3	6,570	5,730 ± 90	–13	495	482 ± 3	–3
December 22, 2003	6	13,140	12,530 ± 80	–5	990	934 ± 4	–6
December 29, 2003	3	6,570	6,200 ± 40	–6	495	480 ± 0	–3
December 31, 2003	6	13,140	12,450 ± 110	–5	990	1,030 ± 0	+4

TABLE D4
Malic Acid and Aloin A Concentrations in the Whole Leaf Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Dose Formulation Concentration (%)	Malic Acid			Aloin A		
		Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)	Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)
January 5, 2004	3	6,570	5,910 ± 50	–10	495	521 ± 1	+5
January 7, 2004	6	13,140	12,210 ± 0	–7	990	1,050 ± 10	+6
January 12, 2004	3	6,570	6,410 ± 240	–2	495	521 ± 6	+5
January 14, 2004	6	13,140	14,850 ± 10	+13	990	949 ± 3	–4
January 19, 2004	3	6,570	5,610 ± 60	–15	495	490 ± 1	–1
January 21, 2004	6	13,140	12,580 ± 0	–4	990	975 ± 3	–2
January 26, 2004	3	6,570	6,100 ± 90	–7	495	509 ± 10	+3
January 28, 2004	6	13,140	12,660 ± 50	–4	990	994 ± 1	0
February 2, 2004	3	6,570	5,680 ± 60	–14	495	543 ± 3	+10
February 4, 2004	6	13,140	12,050 ± 50	–8	990	1,000 ± 0	+1
February 11, 2004	3	6,570	6,150 ± 20	–6	495	521 ± 5	+5
February 9, 2004	6	13,140	12,520 ± 100	–5	990	1,030 ± 20	+4
February 18, 2004	3	6,570	5,760 ± 20	–12	495	471 ± 3	–5
February 16, 2004	6	13,140	12,110 ± 160	–8	990	990 ± 14	0
February 23, 2004	3	6,570	6,220 ± 80	–5	495	454 ± 3	–8
February 25, 2004	6	13,140	12,610 ± 60	–4	990	719 ± 159	–27
March 1, 2004	3	6,570	5,890 ± 10	–10	495	430 ± 2	–13
March 3, 2004	6	13,140	11,970 ± 30	–9	990	936 ± 4	–5
March 8, 2004	3	6,570	6,050 ± 30	–8	495	477 ± 11	–4
March 10, 2004	6	13,140	11,990 ± 10	–9	990	1,030 ± 10	+4
March 15, 2004	3	6,570	5,970 ± 40	–9	495	498 ± 1	+1
March 17, 2004	6	13,140	12,680 ± 30	–4	990	987 ± 10	0

[a] Results of duplicate analyses

TABLE D5
Malic Acid and Aloin A Concentrations in the Decolorized Whole Leaf Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Dose Formulation Concentration (%)	Malic Acid			Aloin A		
		Target Concentration (µg/g)	Determined Concentration[a] (µg/g)	Difference from Target (%)	Target Concentration (µg/g)	Determined Concentration[a] (µg/g)	Difference from Target (%)
May 14, 2003	3	7,380	7,970 ± 420	+8	2.46	2.9 ± 0.1	+18
May 12, 2003	6	14,760	15,000 ± 300	+2	4.92	5.1 ± 0.2	+4
May 21, 2003	3	7,380	8,020 ± 140	+9	2.46	3.2 ± 0.2	+30
May 19, 2003	6	14,760	14,530 ± 20	–2	4.92	7.5 ± 0.4	+52
May 28, 2003	3	7,380	7,570 ± 30	+3	2.46	2.4 ± 0.1	–2
May 26, 2003	6	14,760	14,370 ± 210	–3	4.92	4.8 ± 0.1	–2
June 2, 2003	3	7,380	7,540 ± 0	+2	2.46	2.3 ± 0.1	–7
June 4, 2003	6	14,760	15,150 ± 150	+3	4.92	4.6 ± 0.2	–7
June 9, 2003	3	7,380	7,360 ± 50	0	2.46	2.4 ± 0.0	–2
June 11, 003	6	14,760	14,580 ± 90	–1	4.92	4.9 ± 0.0	0
June 18, 2003	3	7,380	7,600 ± 60	+3	2.46	2.1 ± 0.1	–15
June 16, 2003	6	14,760	16,050 ± 0	+9	4.92	4.0 ± 0.1	–19
June 23, 2003	3	7,380	7,780 ± 50	+5	2.46	3.3 ± 0.2	+34
June 25, 2003	6	14,760	14,180 ± 30	–4	4.92	5.0 ± 0.2	+2
June 30, 2003	3	7,380	7,660 ± 270	+4	2.46	2.7 ± 0.4	+10
July 2, 2003	6	14,760	14,960 ± 380	+1	4.92	5.1 ± 0.2	+4
July 9, 2003	3	7,380	7,200 ± 10	–2	2.46	2.8 ± 0.3	+14
July 7, 2003	6	14,760	13,330 ± 90	–10	4.92	4.6 ± 0.1	–7
July 16, 2003	3	7,380	7,210 ± 50	–2	2.46	2.3 ± 0.0	–7
July 14, 2003	6	14,760	14,470 ± 50	–2	4.92	5.0 ± 0.0	+2
July 23, 2003	3	7,380	7370 ± 10	0	2.46	2.3 ± 0.1	–7
July 21, 2003	6	14,760	13,740 ± 100	–7	4.92	4.6 ± 0.0	–7
July 30, 2003	3	7,380	7,510 ± 80	+2	2.46	2.4 ± 0.0	–2
July 28, 2003	6	14,760	14,810 ± 130	0	4.92	4.5 ± 0.2	–9
August 6, 2003	3	7,380	7,450 ± 80	+1	2.46	2.3 ± 0.0	–7
August 4, 2003	6	14,760	14,750 ± 60	0	4.92	5.4 ± 0.0	+10
August 13, 2003	3	7,380	7,430 ± 50	+1	2.46	2.7 ± 0.7	+10
August 11, 2003	6	14,760	14,750 ± 20	0	4.92	4.9 ± 0.2	0
August 18, 2003	3	7,380	7,310 ± 160	–1	2.46	2.6 ± 0.3	+6
August 20, 2003	6	14,760	14,750 ± 10	0	4.92	4.7 ± 0.0	–4
August 25, 2003	3	7,380	7,370 ± 90	0	2.46	2.4 ± 0.1	–2
August 27, 2003	6	14,760	14,510 ± 20	–2	4.92	4.6 ± 0.1	–7
September 3, 2003	3	7,380	7,380 ± 40	0	2.46	2.3 ± 0.0	–7
September 1, 2003	6	14,760	14,230 ± 40	–4	4.92	4.4 ± 0.0	–11
September 10, 2003	3	7,380	7,590 ± 110	+3	2.46	2.2 ± 0.0	–11
September 8, 2003	6	14,760	15,280 ± 370	+4	4.92	4.7 ± 0.0	–4

TABLE D5
Malic Acid and Aloin A Concentrations in the Decolorized Whole Leaf Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Dose Formulation Concentration (%)	Malic Acid			Aloin A		
		Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)	Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)
September 15, 2003	3	7,380	6,990 ± 510	–5	2.46	2.3 ± 0.1	–7
September 17, 2003	6	14,760	14,600 ± 50	–1	4.92	4.7 ± 0.1	–4
September 24, 2003	3	7,380	6,510 ± 10	–12	2.46	1.9 ± 0.0	–23
September 22, 2003	6	14,760	14,280 ± 30	–3	4.92	4.6 ± 0.1	–7
October 1, 2003	3	7,380	7,270 ± 30	–1	2.46	2.0 ± 0.0	–19
September 29, 2003	6	14,760	14,730 ± 20	0	4.92	4.8 ± 0.1	–2
October 6, 2003	3	7,380	7,340 ± 60	–1	2.46	2.8 ± 0.0	+14
October 8, 2003	6	14,760	15,620 ± 120	+6	4.92	4.5 ± 0.0	–9
October 15, 2003	3	7,380	7,340 ± 90	–1	2.46	2.0 ± 0.0	–19
October 13, 2003	6	14,760	14,920 ± 480	+1	4.92	4.3 ± 0.0	–13
October 22, 2003	3	7,380	8,110 ± 50	+10	2.46	2.4 ± 0.0	–2
October 20, 2003	6	14,760	14,600 ± 90	–1	4.92	4.9 ± 0.0	0
October 29, 2003	3	7,380	7,920 ± 40	+7	2.46	2.2 ± 0.0	–11
October 27, 2003	6	14,760	14,240 ± 190	–4	4.92	4.6 ± 0.1	–7
November 3, 2003	3	7,380	7,390 ± 50	0	2.46	2.0 ± 0.0	–19
November 5, 2003	6	14,760	14,750 ± 190	0	4.92	4.8 ± 0.0	–2
November 12, 2003	3	7,380	7,470 ± 70	+1	2.46	2.4 ± 0.0	–2
November 10, 2003	6	14,760	14,320 ± 430	–3	4.92	5.5 ± 0.1	+12
November 17, 2003	3	7,380	6,940 ± 190	–6	2.46	2.3 ± 0.1	–7
November 19, 2003	6	14,760	14,670 ± 80	–1	4.92	4.2 ± 0.0	–15
November 24, 2003	3	7,380	8,170 ± 350	+11	2.46	2.7 ± 0.6	+10
November 26, 2003	6	14,760	14,870 ± 240	+1	4.92	4.7 ± 0.1	–4
December 3, 2003	3	7,380	7,660 ± 90	+4	2.46	2.5 ± 0.1	+2
December 1, 2003	6	14,760	15,390 ± 740	+4	4.92	4.4 ± 0.1	–11
December 10, 2003	3	7,380	7,950 ± 60	+8	2.46	2.3 ± 0.0	–7
December 8, 2003	6	14,760	15,370 ± 380	+4	4.92	5.0 ± 0.1	+2
December 15, 2003	3	7,380	7,840 ± 350	+6	2.46	2.4 ± 0.0	–2
December 17, 2003	6	14,760	15,370 ± 160	+4	4.92	4.7 ± 0.0	–4
December 22, 2003	3	7,380	7,320 ± 40	–1	2.46	2.3 ± 0.0	–7
December 24, 2003	6	14,760	14,510 ± 420	–2	4.92	4.8 ± 0.0	–2
December 29, 2003	3	7,380	7,320 ± 350	–1	2.46	2.5 ± 0.0	+2
December 31, 2003	6	14,760	15,480 ± 10	+5	4.92	5.0 ± 0.0	+2
January 7, 2004	3	7,380	7,790 ± 500	+6	2.46	2.6 ± 0.0	+6
January 5, 2004	6	14,760	15,150 ± 840	+3	4.92	4.7 ± 0.1	–4
January 12, 2004	3	7,380	7,430 ± 110	+1	2.46	2.6 ± 0.0	+6
January 14, 2004	6	14,760	17,750 ± 180	+20	4.92	4.6 ± 0.1	–7

TABLE D5
Malic Acid and Aloin A Concentrations in the Decolorized Whole Leaf Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Dose Formulation Concentration (%)	Malic Acid			Aloin A		
		Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)	Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)
January 21, 2004	3	7,380	7,000 ± 40	–5	2.46	2.5 ± 0.0	+2
January 19, 2004	6	14,760	15,400 ± 30	+4	4.92	4.7 ± 0.0	–4
January 26, 2004	3	7,380	7,040 ± 50	–5	2.46	3.2 ± 0.1	+30
January 28, 2004	6	14,760	14,520 ± 110	–2	4.92	4.9 ± 0.0	0
February 4, 2004	3	7,380	7,040 ± 130	–5	2.46	2.5 ± 0.0	+2
February 2, 2004	6	14,760	16,510 ± 110	+12	4.92	6.3 ± 1.5	+28
February 11, 2004	3	7,380	7,150 ± 60	–3	2.46	2.6 ± 0.1	+6
February 9, 2004	6	14,760	14,030 ± 120	–5	4.92	4.9 ± 0.0	0
February 16, 2004	3	7,380	7,010 ± 120	–5	2.46	2.6 ± 0.0	+6
February 18, 2004	6	14,760	13,700 ± 60	–7	4.92	5.3 ± 0.1	+8
February 23, 2004	3	7,380	7,250 ± 90	–2	2.46	2.2 ± 0.0	–11
February 25, 2004	6	14,760	14,630 ± 60	–1	4.92	4.5 ± 0.1	–9
March 3, 2004	3	7,380	7,080 ± 60	–4	2.46	2.4 ± 0.0	–2
March 1, 2004	6	14,760	13,850 ± 20	–6	4.92	4.9 ± 0.0	0
March 8, 2004	3	7,380	7,050 ± 10	–4	2.46	2.3 ± 0.1	–7
March 10, 2004	6	14,760	14,110 ± 30	–4	4.92	5.0 ± 0.0	+2
March 17, 2004	3	7,380	7,390 ± 30	0	2.46	2.0 ± 0.0	–19
March 15, 2004	6	14,760	14,070 ± 90	–5	4.92	4.9 ± 0.1	0

[a] Results of duplicate analyses

TABLE D6
Aloe-emodin Concentrations in the Aloe-emodin Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Target Concentration (µg/g)	Determined Concentration[a] (µg/g)	Difference from Target (%)
May 12, 2003	74.6	73.4 ± 0.0	–2
May 14, 2003	7.46	7.1 ± 0.0	–5
May 19, 2003	7.46	7.2 ± 0.1	–3
May 21, 2003	74.6	72.6 ± 3.0	–3
May 26, 2003	74.6	68.6 ± 0.3	–8
May 28, 2003	7.46	6.9 ± 0.0	–8
June 2, 2003	74.6	77.8 ± 0.6	+4
June 4, 2003	7.46	6.9 ± 0.2	–8
June 9, 2003	7.46	7.0 ± 0.1	–6
June 11, 2003	74.6	73.1 ± 2.5	–2
June 16, 2003	74.6	80.7 ± 0.8	+8
June 18, 2003	7.46	6.2 ± 0.1	–17
June 23, 2003	74.6	71.9 ± 1.0	–4
June 25, 2003	7.46	9.6 ± 0.4	+29
June 30, 2003	74.6	63.9 ± 2.0	–14
July 2, 2003	7.46	6.8 ± 0.1	–9
July 7, 2003	74.6	65.5 ± 1.6	–12
July 9, 2003	7.46	9.6 ± 0.0	+29
July 14, 2003	74.6	73.7 ± 0.5	–1
July 16, 2003	7.46	6.4 ± 0.3	–14
July 21, 2003	74.6	63.0 ± 0.8	–16
July 23, 2003	7.46	5.7 ± 0.0	–24
July 28, 2003	7.46	6.2 ± 0.3	–17
July 30, 2003	74.6	63.6 ± 1.4	–15
August 4, 2003	7.46	6.3 ± 0.6	–16
August 6, 2003	74.6	71.3 ± 0.0	–4
August 11, 2003	74.6	74.6 ± 1.0	0
August 13, 2003	7.46	4.6 ± 0.2	–38
August 18, 2003	74.6	71.9 ± 0.4	–4
August 20, 2003	7.46	6.6 ± 0.1	–12
August 25, 2003	74.6	84.7 ± 0.3	+14
August 27, 2003	7.46	6.9 ± 0.0	–8
September 1, 2003	7.46	6.9 ± 0.0	–8
September 3, 2003	74.6	59.9 ± 0.1	–20
September 8, 2003	74.6	75.4 ± 0.0	+1
September 10, 2003	7.46	7.2 ± 0.1	–3
September 15, 2003	7.46	6.7 ± 0.1	–10
September 17, 2003	74.6	77.4[b]	+4
September 22, 2003	7.46	7.2 ± 0.2	–3
September 24, 2003	74.6	64.5 ± 0.2	–14
September 29, 2003	7.46	6.9 ± 0.2	–8
October 1, 2003	74.6	75.5 ± 0.6	+1
October 6, 2003	74.6	73.2 ± 0.7	–2
October 8, 2003	7.46	7.2 ± 0.1	–3
October 13, 2003	74.6	66.6 ± 2.1	–11
October 15, 2003	7.46	7.1 ± 0.1	–5
October 20, 2003	7.46	7.2 ± 0.3	–3
October 22, 2003	74.6	67.5 ± 0.1	–10
October 27, 2003	7.46	6.7 ± 0.4	–10
October 29, 2003	74.6	94.3 ± 1.1	+26

TABLE D6
Aloe-emodin Concentrations in the Aloe-emodin Dose Formulations Administered Dermally to SKH-1 Mice in the 1-Year Simulated Solar Light Study of Aloe vera

Date Sampled	Target Concentration (µg/g)	Determined Concentration (µg/g)	Difference from Target (%)
November 3, 2003	74.6	73.0 ± 3.2	–2
November 5, 2003	7.46	5.8 ± 0.0	–22
November 10, 2003	74.6	87.1 ± 3.0	+17
November 12, 2003	7.46	9.1 ± 0.2	+22
November 17, 2003	74.6	65.2 ± 0.4	–13
November 19, 2003	7.46	8.4 ± 0.6	+13
November 24, 2003	74.6	63.5 ± 0.4	–15
November 26, 2003	7.46	4.9 ± 0.4	–34
December 1, 2003	74.6	49.7 ± 1.5	–33
December 3, 2003	7.46	7.7 ± 0.1	+3
December 8, 2003	7.46	9.0 ± 0.7	+21
December 10, 2003	74.6	53.2 ± 1.1	–29
December 15, 2003	74.6	71.6 ± 11.0	–4
December 17, 2003	7.46	8.6 ± 0.0	+15
December 22, 2003	74.6	82.2 ± 0.1	+10
December 24, 2003	7.46	7.5 ± 0.2	+1
December 29, 2003	74.6	81.4 ± 0.4	+9
December 31, 2003	7.46	8.3 ± 0.1	+11
January 5, 2004	7.46	8.3 ± 0.2	+11
January 7, 2004	74.6	77.1 ± 1.0	+3
January 12, 2004	74.6	62.0 ± 0.2	–17
January 14, 2004	7.46	7.4 ± 0.5	–1
January 19, 2004	7.46	8.0 ± 0.1	+7
January 21, 2004	74.6	68.7 ± 0.1	–8
January 26, 2004	74.6	69.2 ± 0.4	–7
January 28, 2004	7.46	8.6 ± 0.1	+15
February 2, 2004	7.46	14.7 ± 0.1	+97
February 4, 2004	74.6	70.1 ± 0.7	–6
February 9, 2004	7.46	10.4 ± 0.0	+39
February 11, 2004	74.6	70.9 ± 0.8	–5
February 16, 2004	74.6	75.1 ± 1.7	+1
February 18, 2004	7.46	7.8 ± 0.0	+5
February 23, 2004	74.6	76.6 ± 0.6	+3
February 25, 2004	7.46	9.7 ± 0.1	+30
March 1, 2004	74.6	64.8 ± 1.1	–13
March 3, 2004	7.46	8.4 ± 0.0	+13
March 8, 2004	7.46	5.9 ± 0.0	–21
March 10, 2004	74.6	68.4 ± 0.3	–8
March 15, 2004	74.6	105 ± 1	+41
March 17, 2004	7.46	6.2 ± 0.1	–17

[a] Results of duplicate analyses
[b] n = 1

APPENDIX E
SPECTRAL IRRADIANCE OF THE SIMULATED SOLAR LIGHT

SPECTRAL IRRADIANCE OF THE SIMULATED SOLAR LIGHT

METHODS

Simulated solar light (SSL) was created by filtering the output from 6.5 kilowatt (kW) long-arc xenon arc lamps (Atlas Electric, Inc., Spokane, WA) through 1 mm thick SCHOTT WG-320 glass filters (SCHOTT North America, Inc., Elmsford, NY); two xenon arc lamps (SSL-1 and SSL-2) were used during the 1-year study. At generally weekly intervals during the 1-year study, irradiance of the filtered light was measured 2 meters from the light source using a calibrated Optronics OL-754 spectroradiometer (Optronic Laboratories, Inc., Orlando, FL). Excel® was used to calculate the mean and standard error of the 46 measurements of the irradiance (expressed in W/cm^2 per nm units) at each wavelength from 250 to 450 nm. The relative standard error for each wavelength was determined by dividing each standard error by the corresponding mean and multiplying by 100.

The average weighted irradiance and weighted standard error at each wavelength (expressed in $W \bullet CIE/cm^2$ per nm units) was determined by multiplying the average irradiance and standard error by the appropriate weighting value (S_{er}) published by the Commission Internationale de l'Éclairage (CIE) that reflects the intrinsic effectiveness of the wavelength to induce erythema (CIE, 1999). Light between 250 and 298 nm is the most effective at inducing erythema and is accordingly assigned a weighting value of 1. For the spectral range of 250 to 450 nm, the values of S_{er} derived from the human erythema action spectrum are defined as:

S_{er} Value	Wavelength (nm)
1	250 to 298
$10^{0.094\ (298\text{-wavelength})}$	299 to 328
$10^{0.015\ (140\text{-wavelength})}$	329 to 400
0	401 to 450

RESULTS

Average irradiance and average weighted irradiance values for SSL-1 are presented in Table E1, and they are graphically represented in Figures E1A (semilogarithmic scale) and E1B (linear scale), respectively. Similar data for SSL-2 are presented in Table E2 and Figures E2A and E2B. The spectral outputs from the two glass-filtered 6.5 kW xenon arc lights were quite consistent over the course of the study. As shown in Tables E1 and E2, the relative standard errors of the irradiance values over the 45 weeks of study were highest at the low end of the spectra between 250 and 260 nm and lowest at the upper wavelengths between 400 and 450 nm. The largest contribution of the spectra of the light sources to the weighted irradiance is from light emitted between 295 and 320 nm (Figures E1B and E2B).

Table E1
Irradiance and Weighted Irradiance for Light Source SSL-1 in the 1-Year Simulated Solar Light Study of Aloe vera[a]

Wavelength (nm)	Irradiance ($W/cm^2/nm$)	Relative Standard Error for Irradiance[b] (%)	S_{er}	Weighted Irradiance ($W \cdot CIE/cm^2/nm$)
250	6.55E-09 ± 2.42E-10	3.699168624	1	6.55E-09 ± 2.42E-10
251	6.97E-09 ± 2.54E-10	3.649718914	1	6.97E-09 ± 2.54E-10
252	7.36E-09 ± 2.63E-10	3.577519384	1	7.36E-09 ± 2.63E-10
253	7.70E-09 ± 2.69E-10	3.49654579	1	7.70E-09 ± 2.69E-10
254	8.11E-09 ± 2.80E-10	3.449496691	1	8.11E-09 ± 2.80E-10
255	8.47E-09 ± 2.86E-10	3.376091945	1	8.47E-09 ± 2.86E-10
256	8.84E-09 ± 2.94E-10	3.327272727	1	8.84E-09 ± 2.94E-10
257	9.19E-09 ± 3.00E-10	3.260125355	1	9.19E-09 ± 3.00E-10
258	9.50E-09 ± 3.05E-10	3.207928459	1	9.50E-09 ± 3.05E-10
259	9.81E-09 ± 3.04E-10	3.096842698	1	9.81E-09 ± 3.04E-10
260	1.02E-08 ± 3.12E-10	3.066554034	1	1.02E-08 ± 3.12E-10
261	1.06E-08 ± 3.19E-10	3.020847906	1	1.06E-08 ± 3.19E-10
262	1.08E-08 ± 3.19E-10	2.944830388	1	1.08E-08 ± 3.19E-10
263	1.11E-08 ± 3.20E-10	2.886323859	1	1.11E-08 ± 3.20E-10
264	1.14E-08 ± 3.23E-10	2.826419276	1	1.14E-08 ± 3.23E-10
265	1.17E-08 ± 3.33E-10	2.833405382	1	1.17E-08 ± 3.33E-10
266	1.20E-08 ± 3.27E-10	2.720254309	1	1.20E-08 ± 3.27E-10
267	1.24E-08 ± 3.33E-10	2.692195963	1	1.24E-08 ± 3.33E-10
268	1.26E-08 ± 3.37E-10	2.661491432	1	1.26E-08 ± 3.37E-10
269	1.29E-08 ± 3.43E-10	2.647310718	1	1.29E-08 ± 3.43E-10
270	1.33E-08 ± 3.43E-10	2.586233102	1	1.33E-08 ± 3.43E-10
271	1.37E-08 ± 3.52E-10	2.563097939	1	1.37E-08 ± 3.52E-10
272	1.40E-08 ± 3.55E-10	2.528927591	1	1.40E-08 ± 3.55E-10
273	1.42E-08 ± 3.57E-10	2.511388238	1	1.42E-08 ± 3.57E-10
274	1.45E-08 ± 3.56E-10	2.449695603	1	1.45E-08 ± 3.56E-10
275	1.48E-08 ± 3.57E-10	2.415252965	1	1.48E-08 ± 3.57E-10
276	1.51E-08 ± 3.65E-10	2.407196138	1	1.51E-08 ± 3.65E-10
277	1.55E-08 ± 3.68E-10	2.377437191	1	1.55E-08 ± 3.68E-10
278	1.59E-08 ± 3.78E-10	2.380731347	1	1.59E-08 ± 3.78E-10
279	1.63E-08 ± 3.81E-10	2.342480626	1	1.63E-08 ± 3.81E-10
280	1.66E-08 ± 3.86E-10	2.320694487	1	1.66E-08 ± 3.86E-10
281	1.69E-08 ± 3.86E-10	2.283586276	1	1.69E-08 ± 3.86E-10
282	1.72E-08 ± 3.88E-10	2.251702801	1	1.72E-08 ± 3.88E-10
283	1.76E-08 ± 3.91E-10	2.218286542	1	1.76E-08 ± 3.91E-10
284	1.80E-08 ± 4.02E-10	2.240711198	1	1.80E-08 ± 4.02E-10
285	1.83E-08 ± 4.03E-10	2.202592652	1	1.83E-08 ± 4.03E-10
286	1.88E-08 ± 4.13E-10	2.203681149	1	1.88E-08 ± 4.13E-10
287	1.92E-08 ± 4.22E-10	2.200037252	1	1.92E-08 ± 4.22E-10
288	1.95E-08 ± 4.28E-10	2.190292332	1	1.95E-08 ± 4.28E-10
289	1.99E-08 ± 4.29E-10	2.156989905	1	1.99E-08 ± 4.29E-10
290	2.10E-08 ± 5.61E-10	2.669885367	1	2.10E-08 ± 5.61E-10
291	2.25E-08 ± 5.34E-10	2.369411699	1	2.25E-08 ± 5.34E-10
292	2.37E-08 ± 5.50E-10	2.322038763	1	2.37E-08 ± 5.50E-10
293	2.68E-08 ± 6.41E-10	2.395640001	1	2.68E-08 ± 6.41E-10
294	3.21E-08 ± 7.64E-10	2.375282347	1	3.21E-08 ± 7.64E-10
295	4.10E-08 ± 1.06E-09	2.58986821	1	4.10E-08 ± 1.06E-09
296	5.58E-08 ± 1.53E-09	2.741466226	1	5.58E-08 ± 1.53E-09
297	7.71E-08 ± 2.10E-09	2.72490607	1	7.71E-08 ± 2.10E-09
298	1.08E-07 ± 2.98E-09	2.754940256	1	1.08E-07 ± 2.98E-09
299	1.50E-07 ± 4.16E-09	2.77425756	0.8053784	1.21E-07 ± 3.35E-09
300	2.02E-07 ± 5.41E-09	2.678929387	0.6486344	1.31E-07 ± 3.51E-09
301	2.68E-07 ± 6.93E-09	2.586651773	0.5223962	1.40E-07 ± 3.62E-09

Table E1
Irradiance and Weighted Irradiance for Light Source SSL-1 in the 1-Year Simulated Solar Light Study of Aloe vera

Wavelength (nm)	Irradiance ($W/cm^2/nm$)	Relative Standard Error for Irradiance (%)	S_{er}	Weighted Irradiance ($W \cdot CIE/cm^2/nm$)
302	3.43E-07 ± 8.61E-09	2.511607137	0.4207266	1.44E-07 ± 3.62E-09
303	4.34E-07 ± 1.05E-08	2.4178564	0.3388442	1.47E-07 ± 3.56E-09
304	5.32E-07 ± 1.20E-08	2.25385867	0.2728978	1.45E-07 ± 3.27E-09
305	6.35E-07 ± 1.36E-08	2.14761056	0.219786	1.40E-07 ± 3.00E-09
306	6.35E-07 ± 1.36E-08	2.14761056	0.1770109	1.12E-07 ± 2.41E-09
307	7.42E-07 ± 1.50E-08	2.023799714	0.1425608	1.06E-07 ± 2.14E-09
308	8.60E-07 ± 1.90E-08	2.210580983	0.1148154	9.88E-08 ± 2.18E-09
309	9.73E-07 ± 2.19E-08	2.249872283	0.0924698	9.00E-08 ± 2.03E-09
310	1.10E-06 ± 2.26E-08	2.043182679	0.0744732	8.23E-08 ± 1.68E-09
311	1.22E-06 ± 2.52E-08	2.05614724	0.0599791	7.34E-08 ± 1.51E-09
312	1.36E-06 ± 2.64E-08	1.943219645	0.0483059	6.57E-08 ± 1.28E-09
313	1.50E-06 ± 2.75E-08	1.833961495	0.0389045	5.83E-08 ± 1.07E-09
314	1.61E-06 ± 2.92E-08	1.809175007	0.0313329	5.05E-08 ± 9.14E-10
315	1.71E-06 ± 3.03E-08	1.778795978	0.0252348	4.30E-08 ± 7.66E-10
316	1.80E-06 ± 3.18E-08	1.764048435	0.0203236	3.66E-08 ± 6.46E-10
317	1.90E-06 ± 3.26E-08	1.716433835	0.0163682	3.11E-08 ± 5.34E-10
318	1.99E-06 ± 3.35E-08	1.683519475	0.0131826	2.62E-08 ± 4.42E-10
319	2.08E-06 ± 3.42E-08	1.648568807	0.010617	2.20E-08 ± 3.63E-10
320	2.16E-06 ± 3.55E-08	1.642763033	0.0085507	1.85E-08 ± 3.04E-10
321	2.25E-06 ± 3.63E-08	1.616035709	0.0068865	1.55E-08 ± 2.50E-10
322	2.33E-06 ± 3.71E-08	1.593711079	0.0055463	1.29E-08 ± 2.06E-10
323	2.43E-06 ± 3.82E-08	1.57057088	0.0044668	1.09E-08 ± 1.71E-10
324	2.50E-06 ± 3.98E-08	1.591459236	0.0035975	9.00E-09 ± 1.43E-10
325	2.54E-06 ± 3.99E-08	1.573479344	0.0028973	7.35E-09 ± 1.16E-10
326	2.58E-06 ± 4.03E-08	1.561471327	0.0023335	6.03E-09 ± 9.41E-11
327	2.64E-06 ± 4.11E-08	1.558047878	0.0018793	4.95E-09 ± 7.72E-11
328	2.70E-06 ± 4.10E-08	1.519113684	0.0015136	4.08E-09 ± 6.20E-11
329	2.74E-06 ± 4.22E-08	1.537728759	0.0014622	4.01E-09 ± 6.17E-11
330	2.80E-06 ± 4.25E-08	1.517587704	0.0014125	3.95E-09 ± 6.00E-11
331	2.85E-06 ± 4.31E-08	1.512460014	0.0013646	3.89E-09 ± 5.88E-11
332	2.89E-06 ± 4.33E-08	1.496332462	0.0013183	3.82E-09 ± 5.71E-11
333	2.94E-06 ± 4.32E-08	1.470268916	0.0012735	3.74E-09 ± 5.51E-11
334	2.99E-06 ± 4.39E-08	1.469257432	0.0012303	3.68E-09 ± 5.40E-11
335	3.03E-06 ± 4.40E-08	1.452054341	0.0011885	3.60E-09 ± 5.23E-11
336	3.07E-06 ± 4.42E-08	1.440479892	0.0011482	3.53E-09 ± 5.08E-11
337	3.11E-06 ± 4.44E-08	1.428236485	0.0011092	3.45E-09 ± 4.93E-11
338	3.16E-06 ± 4.52E-08	1.43142667	0.0010715	3.38E-09 ± 4.84E-11
339	3.20E-06 ± 4.49E-08	1.405707071	0.0010351	3.31E-09 ± 4.65E-11
340	3.23E-06 ± 4.54E-08	1.405055516	0.001	3.23E-09 ± 4.54E-11
341	3.27E-06 ± 4.55E-08	1.389856913	0.0009661	3.16E-09 ± 4.39E-11
342	3.31E-06 ± 4.59E-08	1.386678689	0.0009333	3.09E-09 ± 4.29E-11
343	3.35E-06 ± 4.57E-08	1.365504207	0.0009016	3.02E-09 ± 4.12E-11
344	3.38E-06 ± 4.60E-08	1.360181427	0.000871	2.95E-09 ± 4.01E-11
345	3.42E-06 ± 4.57E-08	1.336639554	0.0008414	2.88E-09 ± 3.85E-11
346	3.53E-06 ± 2.12E-08	0.60023992	0.0008128	2.87E-09 ± 1.72E-11
347	3.56E-06 ± 2.12E-08	0.593578964	0.0007852	2.80E-09 ± 1.66E-11
348	3.60E-06 ± 2.07E-08	0.576754983	0.0007586	2.73E-09 ± 1.57E-11
349	3.63E-06 ± 2.04E-08	0.562563363	0.0007328	2.66E-09 ± 1.50E-11
350	3.66E-06 ± 2.03E-08	0.553787975	0.0007079	2.59E-09 ± 1.43E-11
351	3.69E-06 ± 2.03E-08	0.549066497	0.0006839	2.53E-09 ± 1.39E-11
352	3.72E-06 ± 1.99E-08	0.534881765	0.0006607	2.46E-09 ± 1.32E-11
353	3.75E-06 ± 2.00E-08	0.533730928	0.0006383	2.40E-09 ± 1.28E-11
354	3.78E-06 ± 2.02E-08	0.533993946	0.0006166	2.33E-09 ± 1.25E-11

Table E1
Irradiance and Weighted Irradiance for Light Source SSL-1 in the 1-Year Simulated Solar Light Study of Aloe vera

Wavelength (nm)	Irradiance ($W/cm^2/nm$)	Relative Standard Error for Irradiance (%)	S_{er}	Weighted Irradiance ($W \cdot CIE/cm^2/nm$)
355	3.81E-06 ± 2.01E-08	0.528624837	0.0005957	2.27E-09 ± 1.20E-11
356	3.86E-06 ± 2.00E-08	0.517580731	0.0005754	2.22E-09 ± 1.15E-11
357	3.88E-06 ± 2.00E-08	0.514398033	0.0005559	2.16E-09 ± 1.11E-11
358	3.90E-06 ± 2.02E-08	0.51813472	0.000537	2.10E-09 ± 1.09E-11
359	3.92E-06 ± 1.99E-08	0.50629271	0.0005188	2.04E-09 ± 1.03E-11
360	3.96E-06 ± 1.98E-08	0.501411492	0.0005012	1.98E-09 ± 9.95E-12
361	4.00E-06 ± 1.95E-08	0.489223287	0.0004842	1.93E-09 ± 9.47E-12
362	4.02E-06 ± 2.00E-08	0.496943324	0.0004677	1.88E-09 ± 9.33E-12
363	4.03E-06 ± 1.97E-08	0.490172841	0.0004519	1.82E-09 ± 8.92E-12
364	4.06E-06 ± 1.94E-08	0.478829026	0.0004365	1.77E-09 ± 8.48E-12
365	4.09E-06 ± 1.95E-08	0.477140437	0.0004217	1.73E-09 ± 8.23E-12
366	4.13E-06 ± 1.96E-08	0.475212596	0.0004074	1.68E-09 ± 8.00E-12
367	4.19E-06 ± 1.96E-08	0.46943229	0.0003936	1.65E-09 ± 7.73E-12
368	4.24E-06 ± 1.98E-08	0.467378263	0.0003802	1.61E-09 ± 7.53E-12
369	4.29E-06 ± 2.00E-08	0.465882691	0.0003673	1.58E-09 ± 7.35E-12
370	4.32E-06 ± 1.95E-08	0.452076526	0.0003548	1.53E-09 ± 6.92E-12
371	4.28E-06 ± 1.92E-08	0.449566286	0.0003428	1.47E-09 ± 6.59E-12
372	4.24E-06 ± 1.91E-08	0.450173505	0.0003311	1.40E-09 ± 6.32E-12
373	4.23E-06 ± 1.88E-08	0.445016567	0.0003199	1.35E-09 ± 6.02E-12
374	4.24E-06 ± 1.88E-08	0.443654574	0.000309	1.31E-09 ± 5.82E-12
375	4.27E-06 ± 1.92E-08	0.449723899	0.0002985	1.27E-09 ± 5.73E-12
376	4.30E-06 ± 1.94E-08	0.450375485	0.0002884	1.24E-09 ± 5.59E-12
377	4.34E-06 ± 1.95E-08	0.449213714	0.0002786	1.21E-09 ± 5.43E-12
378	4.40E-06 ± 1.99E-08	0.452579497	0.0002692	1.18E-09 ± 5.36E-12
379	4.47E-06 ± 2.01E-08	0.449047482	0.00026	1.16E-09 ± 5.22E-12
380	4.57E-06 ± 2.01E-08	0.440045153	0.0002512	1.15E-09 ± 5.05E-12
381	4.58E-06 ± 2.00E-08	0.437839478	0.0002427	1.11E-09 ± 4.87E-12
382	4.54E-06 ± 2.00E-08	0.441137134	0.0002344	1.06E-09 ± 4.70E-12
383	4.50E-06 ± 1.97E-08	0.437228364	0.0002265	1.02E-09 ± 4.46E-12
384	4.48E-06 ± 1.94E-08	0.432487974	0.0002188	9.80E-10 ± 4.24E-12
385	4.47E-06 ± 1.94E-08	0.433351756	0.0002113	9.44E-10 ± 4.09E-12
386	4.47E-06 ± 1.93E-08	0.430961974	0.0002042	9.13E-10 ± 3.94E-12
387	4.48E-06 ± 1.96E-08	0.437957397	0.0001972	8.83E-10 ± 3.87E-12
388	4.53E-06 ± 1.95E-08	0.431257133	0.0001905	8.63E-10 ± 3.72E-12
389	4.64E-06 ± 2.03E-08	0.437875311	0.0001841	8.55E-10 ± 3.74E-12
390	4.76E-06 ± 2.06E-08	0.433137491	0.0001778	8.46E-10 ± 3.66E-12
391	4.78E-06 ± 2.06E-08	0.430769302	0.0001718	8.20E-10 ± 3.53E-12
392	4.81E-06 ± 2.10E-08	0.437483988	0.000166	7.98E-10 ± 3.49E-12
393	4.91E-06 ± 2.08E-08	0.423742686	0.0001603	7.88E-10 ± 3.34E-12
394	5.09E-06 ± 2.17E-08	0.427375498	0.0001549	7.88E-10 ± 3.37E-12
395	5.58E-06 ± 2.46E-08	0.440275441	0.0001496	8.34E-10 ± 3.67E-12
396	5.79E-06 ± 2.50E-08	0.431093992	0.0001445	8.37E-10 ± 3.61E-12
397	5.87E-06 ± 2.59E-08	0.440405988	0.0001396	8.20E-10 ± 3.61E-12
398	5.61E-06 ± 2.56E-08	0.456707166	0.0001349	7.57E-10 ± 3.46E-12
399	5.18E-06 ± 2.28E-08	0.440116091	0.0001303	6.75E-10 ± 2.97E-12
400	4.98E-06 ± 2.17E-08	0.435781007	0.0001259	6.27E-10 ± 2.73E-12
401	4.93E-06 ± 2.12E-08	0.429730269	0	—[c]
402	4.92E-06 ± 2.16E-08	0.439018402	0	—
403	4.93E-06 ± 2.13E-08	0.431799385	0	—
404	4.96E-06 ± 2.16E-08	0.434297502	0	—
405	5.03E-06 ± 2.17E-08	0.432235284	0	—
406	5.05E-06 ± 2.21E-08	0.437931904	0	—
407	5.14E-06 ± 2.27E-08	0.442163966	0	—

TABLE E1
Irradiance and Weighted Irradiance for Light Source SSL-1 in the 1-Year Simulated Solar Light Study of Aloe vera

Wavelength (nm)	Irradiance ($W/cm^2/nm$)	Relative Standard Error for Irradiance (%)	S_{er}	Weighted Irradiance ($W \cdot CIE/cm^2/nm$)
408	5.42E-06 ± 2.40E-08	0.442346709	0	—
409	5.36E-06 ± 2.40E-08	0.448253701	0	—
410	5.25E-06 ± 2.32E-08	0.440882687	0	—
411	5.41E-06 ± 2.40E-08	0.444358419	0	—
412	5.64E-06 ± 2.48E-08	0.440008506	0	—
413	5.41E-06 ± 2.37E-08	0.438687017	0	—
414	5.25E-06 ± 2.32E-08	0.44255402	0	—
415	5.21E-06 ± 2.28E-08	0.438444388	0	—
416	5.21E-06 ± 2.30E-08	0.441637849	0	—
417	5.23E-06 ± 2.31E-08	0.442304974	0	—
418	5.30E-06 ± 2.33E-08	0.439785979	0	—
419	5.79E-06 ± 2.68E-08	0.462018959	0	—
420	5.84E-06 ± 2.63E-08	0.450016561	0	—
421	5.47E-06 ± 2.51E-08	0.458899108	0	—
422	5.34E-06 ± 2.43E-08	0.454799563	0	—
423	5.34E-06 ± 2.37E-08	0.443137238	0	—
424	5.38E-06 ± 2.36E-08	0.438345832	0	—
425	5.38E-06 ± 2.37E-08	0.440473191	0	—
426	5.36E-06 ± 2.39E-08	0.445491595	0	—
427	5.36E-06 ± 2.35E-08	0.437839465	0	—
428	5.36E-06 ± 2.41E-08	0.449119511	0	—
429	5.36E-06 ± 2.37E-08	0.442394129	0	—
430	5.36E-06 ± 2.39E-08	0.445535371	0	—
431	5.37E-06 ± 2.33E-08	0.434424178	0	—
432	5.39E-06 ± 2.37E-08	0.440200243	0	—
433	5.42E-06 ± 2.45E-08	0.451946024	0	—
434	5.45E-06 ± 2.44E-08	0.44753724	0	—
435	5.49E-06 ± 2.47E-08	0.450302817	0	—
436	5.57E-06 ± 2.50E-08	0.447914802	0	—
437	5.76E-06 ± 2.63E-08	0.457204046	0	—
438	6.05E-06 ± 2.75E-08	0.454310519	0	—
439	5.99E-06 ± 2.79E-08	0.465464176	0	—
440	5.63E-06 ± 2.56E-08	0.454526486	0	—
441	5.71E-06 ± 2.66E-08	0.466032953	0	—
442	5.80E-06 ± 2.69E-08	0.464169092	0	—
443	5.68E-06 ± 2.64E-08	0.464789271	0	—
444	5.64E-06 ± 2.65E-08	0.469121185	0	—
445	5.66E-06 ± 2.71E-08	0.478041563	0	—
446	5.61E-06 ± 2.65E-08	0.472207565	0	—
447	5.60E-06 ± 2.56E-08	0.456947269	0	—
448	5.65E-06 ± 2.62E-08	0.463977314	0	—
449	6.02E-06 ± 2.90E-08	0.482736107	0	—
450	8.71E-06 ± 4.65E-08	0.533183612	0	—

[a] Irradiance and weighted irradiance values are presented as mean ± standard error for 46 measurements at each wavelength; W = watts; S_{er} = CIE human erythema action spectrum weighting function (CIE, 1999); weighted irradiance = irradiance • S_{er}.
[b] Determined by dividing each irradiance standard error by the corresponding mean and multiplying by 100.
[c] Not applicable; $S_{er} = 0$

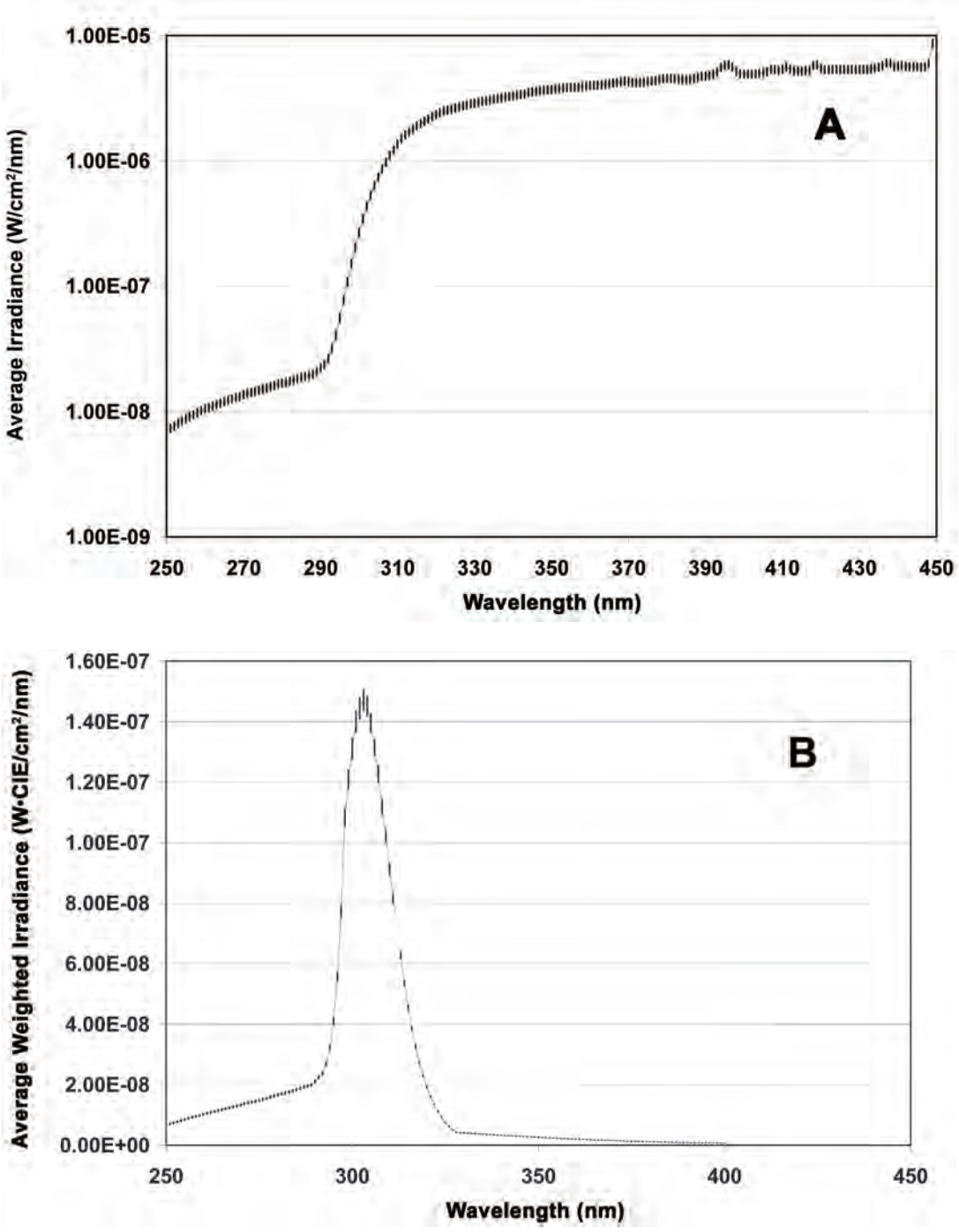

Figure E1
Average Irradiance (A) and Average Weighted Irradiance (B) for Light Source SSL-1 in the 1-Year Simulated Solar Light Study of Aloe vera

Average ± standard error (n = 46 at each wavelength); W = watts; irradiance was weighted by application of the CIE human erythema action spectrum weighting function (CIE, 1999).

TABLE E2
Irradiance and Weighted Irradiance for Light Source SSL-2 in the 1-Year Simulated Solar Light Study of Aloe vera[a]

Wavelength (nm)	Irradiance ($W/cm^2/nm$)	Relative Standard Error for Irradiance[b] (%)	S_{er}	Weighted Irradiance ($W \cdot CIE/cm^2/nm$)
250	6.28E-09 ± 1.68E-10	2.67101478	1	6.28E-09 ± 1.68E-10
251	6.62E-09 ± 1.82E-10	2.75176768	1	6.62E-09 ± 1.82E-10
252	6.96E-09 ± 1.85E-10	2.65562024	1	6.96E-09 ± 1.85E-10
253	7.30E-09 ± 1.87E-10	2.56874288	1	7.30E-09 ± 1.87E-10
254	7.61E-09 ± 1.95E-10	2.55669218	1	7.61E-09 ± 1.95E-10
255	7.95E-09 ± 1.95E-10	2.4483249	1	7.95E-09 ± 1.95E-10
256	8.25E-09 ± 1.99E-10	2.41148018	1	8.25E-09 ± 1.99E-10
257	8.57E-09 ± 1.98E-10	2.31023689	1	8.57E-09 ± 1.98E-10
258	8.85E-09 ± 2.08E-10	2.34443675	1	8.85E-09 ± 2.08E-10
259	9.14E-09 ± 2.09E-10	2.28722077	1	9.14E-09 ± 2.09E-10
260	9.50E-09 ± 2.18E-10	2.29238518	1	9.50E-09 ± 2.18E-10
261	9.83E-09 ± 2.18E-10	2.21598926	1	9.83E-09 ± 2.18E-10
262	1.01E-08 ± 2.24E-10	2.22283005	1	1.01E-08 ± 2.24E-10
263	1.03E-08 ± 2.27E-10	2.19268529	1	1.03E-08 ± 2.27E-10
264	1.06E-08 ± 2.24E-10	2.11496157	1	1.06E-08 ± 2.24E-10
265	1.08E-08 ± 2.31E-10	2.12980989	1	1.08E-08 ± 2.31E-10
266	1.12E-08 ± 2.28E-10	2.0394925	1	1.12E-08 ± 2.28E-10
267	1.14E-08 ± 2.28E-10	2.0032289	1	1.14E-08 ± 2.28E-10
268	1.16E-08 ± 2.32E-10	1.99878471	1	1.16E-08 ± 2.32E-10
269	1.19E-08 ± 2.40E-10	2.01136632	1	1.19E-08 ± 2.40E-10
270	1.23E-08 ± 2.42E-10	1.97394897	1	1.23E-08 ± 2.42E-10
271	1.27E-08 ± 2.44E-10	1.92547216	1	1.27E-08 ± 2.44E-10
272	1.30E-08 ± 2.46E-10	1.89627938	1	1.30E-08 ± 2.46E-10
273	1.32E-08 ± 2.45E-10	1.86062801	1	1.32E-08 ± 2.45E-10
274	1.34E-08 ± 2.55E-10	1.9025242	1	1.34E-08 ± 2.55E-10
275	1.36E-08 ± 2.51E-10	1.83701867	1	1.36E-08 ± 2.51E-10
276	1.39E-08 ± 2.47E-10	1.77149204	1	1.39E-08 ± 2.47E-10
277	1.43E-08 ± 2.52E-10	1.7628052	1	1.43E-08 ± 2.52E-10
278	1.46E-08 ± 2.58E-10	1.76261261	1	1.46E-08 ± 2.58E-10
279	1.50E-08 ± 2.53E-10	1.68763493	1	1.50E-08 ± 2.53E-10
280	1.54E-08 ± 2.66E-10	1.72614815	1	1.54E-08 ± 2.66E-10
281	1.57E-08 ± 2.69E-10	1.72002086	1	1.57E-08 ± 2.69E-10
282	1.59E-08 ± 2.62E-10	1.64553252	1	1.59E-08 ± 2.62E-10
283	1.63E-08 ± 2.75E-10	1.68973965	1	1.63E-08 ± 2.75E-10
284	1.66E-08 ± 2.75E-10	1.65832542	1	1.66E-08 ± 2.75E-10
285	1.70E-08 ± 2.77E-10	1.63086461	1	1.70E-08 ± 2.77E-10
286	1.73E-08 ± 2.78E-10	1.60120946	1	1.73E-08 ± 2.78E-10
287	1.78E-08 ± 2.80E-10	1.57705396	1	1.78E-08 ± 2.80E-10
288	1.81E-08 ± 2.83E-10	1.56161406	1	1.81E-08 ± 2.83E-10
289	1.84E-08 ± 2.91E-10	1.5793542	1	1.84E-08 ± 2.91E-10
290	2.02E-08 ± 3.44E-10	1.70215663	1	2.02E-08 ± 3.44E-10
291	2.16E-08 ± 3.37E-10	1.55974598	1	2.16E-08 ± 3.37E-10
292	2.26E-08 ± 3.49E-10	1.5457129	1	2.26E-08 ± 3.49E-10
293	2.53E-08 ± 3.65E-10	1.44454308	1	2.53E-08 ± 3.65E-10
294	3.02E-08 ± 3.65E-10	1.20951662	1	3.02E-08 ± 3.65E-10
295	3.85E-08 ± 4.33E-10	1.12444803	1	3.85E-08 ± 4.33E-10
296	5.22E-08 ± 5.33E-10	1.0206982	1	5.22E-08 ± 5.33E-10
297	7.26E-08 ± 7.68E-10	1.05909511	1	7.26E-08 ± 7.68E-10
298	1.02E-07 ± 1.04E-09	1.02043832	1	1.02E-07 ± 1.04E-09
299	1.41E-07 ± 1.30E-09	0.92081974	0.805378	1.13E-07 ± 1.04E-09
300	1.90E-07 ± 1.78E-09	0.93565585	0.648634	1.23E-07 ± 1.15E-09
301	2.52E-07 ± 2.29E-09	0.90719223	0.522396	1.32E-07 ± 1.20E-09
302	3.24E-07 ± 2.73E-09	0.84428088	0.420727	1.36E-07 ± 1.15E-09

Table E2
Irradiance and Weighted Irradiance for Light Source SSL-2 in the 1-Year Simulated Solar Light Study of Aloe vera

Wavelength (nm)	Irradiance ($W/cm^2/nm$)	Relative Standard Error for Irradiance (%)	S_{er}	Weighted Irradiance ($W \cdot CIE/cm^2/nm$)
303	4.10E-07 ± 3.21E-09	0.78494793	0.338844	1.39E-07 ± 1.09E-09
304	5.03E-07 ± 3.62E-09	0.71895305	0.272898	1.37E-07 ± 9.87E-10
305	6.04E-07 ± 4.27E-09	0.70605673	0.219786	1.33E-07 ± 9.37E-10
306	7.06E-07 ± 4.49E-09	0.6362962	0.177011	1.25E-07 ± 7.95E-10
307	8.07E-07 ± 4.52E-09	0.55995642	0.142561	1.15E-07 ± 6.44E-10
308	9.23E-07 ± 7.46E-09	0.80830162	0.114815	1.06E-07 ± 8.57E-10
309	1.05E-06 ± 8.06E-09	0.76643456	0.09247	9.73E-08 ± 7.46E-10
310	1.16E-06 ± 9.37E-09	0.80543846	0.074473	8.67E-08 ± 6.98E-10
311	1.29E-06 ± 9.97E-09	0.7727798	0.059979	7.74E-08 ± 5.98E-10
312	1.43E-06 ± 8.13E-09	0.5693118	0.048306	6.90E-08 ± 3.93E-10
313	1.54E-06 ± 8.51E-09	0.55405291	0.038905	5.98E-08 ± 3.31E-10
314	1.63E-06 ± 9.65E-09	0.59320772	0.031333	5.10E-08 ± 3.02E-10
315	1.72E-06 ± 1.00E-08	0.5836024	0.025235	4.33E-08 ± 2.53E-10
316	1.81E-06 ± 1.02E-08	0.56251439	0.020324	3.69E-08 ± 2.07E-10
317	1.90E-06 ± 1.05E-08	0.55136176	0.016368	3.11E-08 ± 1.72E-10
318	1.99E-06 ± 1.12E-08	0.56257875	0.013183	2.62E-08 ± 1.47E-10
319	2.07E-06 ± 1.08E-08	0.52369516	0.010617	2.20E-08 ± 1.15E-10
320	2.15E-06 ± 1.10E-08	0.511642	0.008551	1.84E-08 ± 9.41E-11
321	2.23E-06 ± 1.17E-08	0.52647813	0.006887	1.54E-08 ± 8.09E-11
322	2.33E-06 ± 1.15E-08	0.49361482	0.005546	1.29E-08 ± 6.38E-11
323	2.40E-06 ± 1.21E-08	0.50439799	0.004467	1.07E-08 ± 5.40E-11
324	2.43E-06 ± 1.21E-08	0.49632253	0.003598	8.75E-09 ± 4.34E-11
325	2.48E-06 ± 1.24E-08	0.5011491	0.002897	7.18E-09 ± 3.60E-11
326	2.53E-06 ± 1.27E-08	0.50205676	0.002334	5.90E-09 ± 2.96E-11
327	2.59E-06 ± 1.31E-08	0.50760722	0.001879	4.86E-09 ± 2.47E-11
328	2.64E-06 ± 1.32E-08	0.50087451	0.001514	4.00E-09 ± 2.00E-11
329	2.69E-06 ± 1.31E-08	0.48748645	0.001462	3.93E-09 ± 1.92E-11
330	2.74E-06 ± 1.27E-08	0.46383442	0.001413	3.86E-09 ± 1.79E-11
331	2.78E-06 ± 1.34E-08	0.48172068	0.001365	3.79E-09 ± 1.83E-11
332	2.83E-06 ± 1.37E-08	0.48288011	0.001318	3.73E-09 ± 1.80E-11
333	2.87E-06 ± 1.36E-08	0.47286579	0.001274	3.66E-09 ± 1.73E-11
334	2.91E-06 ± 1.36E-08	0.46607371	0.00123	3.58E-09 ± 1.67E-11
335	2.96E-06 ± 1.40E-08	0.47351542	0.001189	3.51E-09 ± 1.66E-11
336	3.00E-06 ± 1.33E-08	0.44461434	0.001148	3.44E-09 ± 1.53E-11
337	3.04E-06 ± 1.37E-08	0.45113421	0.001109	3.37E-09 ± 1.52E-11
338	3.08E-06 ± 1.44E-08	0.46619704	0.001072	3.30E-09 ± 1.54E-11
339	3.12E-06 ± 1.39E-08	0.4456068	0.001035	3.23E-09 ± 1.44E-11
340	3.16E-06 ± 1.43E-08	0.45354712	0.001	3.16E-09 ± 1.43E-11
341	3.20E-06 ± 1.45E-08	0.45462013	0.000966	3.09E-09 ± 1.40E-11
342	3.23E-06 ± 1.48E-08	0.45881033	0.000933	3.02E-09 ± 1.38E-11
343	3.27E-06 ± 1.49E-08	0.45597802	0.000902	2.95E-09 ± 1.34E-11
344	3.31E-06 ± 1.50E-08	0.45518523	0.000871	2.88E-09 ± 1.31E-11
345	3.39E-06 ± 1.79E-08	0.5277368	0.000841	2.85E-09 ± 1.51E-11
346	3.43E-06 ± 1.83E-08	0.53324323	0.000813	2.79E-09 ± 1.49E-11
347	3.46E-06 ± 1.81E-08	0.52443175	0.000785	2.72E-09 ± 1.42E-11
348	3.49E-06 ± 1.80E-08	0.51626556	0.000759	2.65E-09 ± 1.37E-11
349	3.52E-06 ± 1.81E-08	0.51469888	0.000733	2.58E-09 ± 1.33E-11
350	3.56E-06 ± 1.83E-08	0.51471273	0.000708	2.52E-09 ± 1.30E-11
351	3.59E-06 ± 1.85E-08	0.51598549	0.000684	2.45E-09 ± 1.27E-11
352	3.62E-06 ± 1.90E-08	0.52586567	0.000661	2.39E-09 ± 1.26E-11
353	3.65E-06 ± 1.89E-08	0.51841253	0.000638	2.33E-09 ± 1.21E-11
354	3.67E-06 ± 1.86E-08	0.5067904	0.000617	2.27E-09 ± 1.15E-11
355	3.70E-06 ± 1.87E-08	0.50518536	0.000596	2.21E-09 ± 1.11E-11

TABLE E2
Irradiance and Weighted Irradiance for Light Source SSL-2 in the 1-Year Simulated Solar Light Study of Aloe vera

Wavelength (nm)	Irradiance ($W/cm^2/nm$)	Relative Standard Error for Irradiance (%)	S_{er}	Weighted Irradiance ($W \cdot CIE/cm^2/nm$)
356	3.73E-06 ± 1.86E-08	0.49925256	0.000575	2.15E-09 ± 1.07E-11
357	3.75E-06 ± 1.90E-08	0.50698961	0.000556	2.08E-09 ± 1.06E-11
358	3.77E-06 ± 1.87E-08	0.49670802	0.000537	2.02E-09 ± 1.00E-11
359	3.79E-06 ± 1.93E-08	0.50824219	0.000519	1.97E-09 ± 9.99E-12
360	3.82E-06 ± 1.89E-08	0.49332643	0.000501	1.92E-09 ± 9.46E-12
361	3.86E-06 ± 1.89E-08	0.48870385	0.000484	1.87E-09 ± 9.14E-12
362	3.88E-06 ± 1.91E-08	0.49164594	0.000468	1.82E-09 ± 8.93E-12
363	3.89E-06 ± 1.89E-08	0.48488655	0.000452	1.76E-09 ± 8.53E-12
364	3.92E-06 ± 1.88E-08	0.47874252	0.000437	1.71E-09 ± 8.20E-12
365	3.96E-06 ± 1.89E-08	0.47767027	0.000422	1.67E-09 ± 7.97E-12
366	4.00E-06 ± 1.89E-08	0.47340564	0.000407	1.63E-09 ± 7.71E-12
367	4.05E-06 ± 1.91E-08	0.47151066	0.000394	1.59E-09 ± 7.52E-12
368	4.10E-06 ± 1.88E-08	0.45930435	0.00038	1.56E-09 ± 7.16E-12
369	4.16E-06 ± 1.87E-08	0.44939796	0.000367	1.53E-09 ± 6.86E-12
370	4.18E-06 ± 1.91E-08	0.45563481	0.000355	1.48E-09 ± 6.76E-12
371	4.14E-06 ± 1.86E-08	0.44856664	0.000343	1.42E-09 ± 6.37E-12
372	4.10E-06 ± 1.80E-08	0.43755842	0.000331	1.36E-09 ± 5.95E-12
373	4.09E-06 ± 1.76E-08	0.43133369	0.00032	1.31E-09 ± 5.64E-12
374	4.11E-06 ± 1.79E-08	0.43493161	0.000309	1.27E-09 ± 5.52E-12
375	4.13E-06 ± 1.78E-08	0.43088578	0.000299	1.23E-09 ± 5.32E-12
376	4.17E-06 ± 1.77E-08	0.42475185	0.000288	1.20E-09 ± 5.10E-12
377	4.20E-06 ± 1.79E-08	0.42488119	0.000279	1.17E-09 ± 4.98E-12
378	4.26E-06 ± 1.80E-08	0.42241662	0.000269	1.15E-09 ± 4.84E-12
379	4.34E-06 ± 1.80E-08	0.41583618	0.00026	1.13E-09 ± 4.69E-12
380	4.44E-06 ± 1.80E-08	0.4059829	0.000251	1.11E-09 ± 4.52E-12
381	4.45E-06 ± 1.81E-08	0.40664248	0.000243	1.08E-09 ± 4.39E-12
382	4.42E-06 ± 1.82E-08	0.41301328	0.000234	1.04E-09 ± 4.27E-12
383	4.38E-06 ± 1.80E-08	0.40991236	0.000227	9.92E-10 ± 4.07E-12
384	4.36E-06 ± 1.78E-08	0.40791703	0.000219	9.53E-10 ± 3.89E-12
385	4.34E-06 ± 1.77E-08	0.40838574	0.000211	9.18E-10 ± 3.75E-12
386	4.35E-06 ± 1.74E-08	0.40060962	0.000204	8.88E-10 ± 3.56E-12
387	4.36E-06 ± 1.71E-08	0.39315633	0.000197	8.59E-10 ± 3.38E-12
388	4.41E-06 ± 1.75E-08	0.39730972	0.000191	8.40E-10 ± 3.34E-12
389	4.52E-06 ± 1.77E-08	0.39167905	0.000184	8.32E-10 ± 3.26E-12
390	4.64E-06 ± 1.84E-08	0.39698831	0.000178	8.24E-10 ± 3.27E-12
391	4.66E-06 ± 1.85E-08	0.39829543	0.000172	8.00E-10 ± 3.19E-12
392	4.68E-06 ± 1.81E-08	0.38560143	0.000166	7.77E-10 ± 3.00E-12
393	4.79E-06 ± 1.84E-08	0.383328	0.00016	7.68E-10 ± 2.95E-12
394	4.97E-06 ± 1.88E-08	0.37836317	0.000155	7.69E-10 ± 2.91E-12
395	5.45E-06 ± 2.13E-08	0.38984336	0.00015	8.16E-10 ± 3.18E-12
396	5.69E-06 ± 2.16E-08	0.37900999	0.000145	8.22E-10 ± 3.12E-12
397	5.76E-06 ± 2.12E-08	0.36719485	0.00014	8.05E-10 ± 2.95E-12
398	5.52E-06 ± 2.08E-08	0.37728698	0.000135	7.45E-10 ± 2.81E-12
399	5.08E-06 ± 1.90E-08	0.37429835	0.00013	6.62E-10 ± 2.48E-12
400	4.88E-06 ± 1.77E-08	0.36219461	0.000126	6.14E-10 ± 2.23E-12
401	4.83E-06 ± 1.72E-08	0.35654725	0	—[c]
402	4.82E-06 ± 1.66E-08	0.34457926	0	—
403	4.83E-06 ± 1.69E-08	0.35045199	0	—
404	4.87E-06 ± 1.65E-08	0.33839448	0	—
405	4.93E-06 ± 1.68E-08	0.34027637	0	—
406	4.96E-06 ± 1.71E-08	0.34560201	0	—
407	5.04E-06 ± 1.74E-08	0.345184	0	—
408	5.33E-06 ± 1.88E-08	0.35233137	0	—

TABLE E2
Irradiance and Weighted Irradiance for Light Source SSL-2 in the 1-Year Simulated Solar Light Study of Aloe vera

Wavelength (nm)	Irradiance ($W/cm^2/nm$)	Relative Standard Error for Irradiance (%)	S_{er}	Weighted Irradiance ($W \cdot CIE/cm^2/nm$)
409	5.28E-06 ± 1.84E-08	0.34929286	0	—
410	5.16E-06 ± 1.78E-08	0.34571618	0	—
411	5.32E-06 ± 1.84E-08	0.34643472	0	—
412	5.55E-06 ± 1.86E-08	0.33516899	0	—
413	5.33E-06 ± 1.82E-08	0.34165625	0	—
414	5.17E-06 ± 1.68E-08	0.32599767	0	—
415	5.12E-06 ± 1.65E-08	0.32118325	0	—
416	5.12E-06 ± 1.64E-08	0.32057402	0	—
417	5.14E-06 ± 1.64E-08	0.31948389	0	—
418	5.21E-06 ± 1.66E-08	0.31813018	0	—
419	5.69E-06 ± 2.00E-08	0.35244761	0	—
420	5.77E-06 ± 1.93E-08	0.33429219	0	—
421	5.39E-06 ± 1.69E-08	0.31351402	0	—
422	5.26E-06 ± 1.64E-08	0.31251455	0	—
423	5.26E-06 ± 1.63E-08	0.31040037	0	—
424	5.30E-06 ± 1.64E-08	0.30959529	0	—
425	5.30E-06 ± 1.58E-08	0.29842676	0	—
426	5.29E-06 ± 1.59E-08	0.29994357	0	—
427	5.28E-06 ± 1.61E-08	0.30413424	0	—
428	5.29E-06 ± 1.60E-08	0.30314489	0	—
429	5.28E-06 ± 1.58E-08	0.29892696	0	—
430	5.29E-06 ± 1.54E-08	0.29117883	0	—
431	5.30E-06 ± 1.54E-08	0.29112794	0	—
432	5.32E-06 ± 1.57E-08	0.29433917	0	—
433	5.35E-06 ± 1.54E-08	0.28870386	0	—
434	5.37E-06 ± 1.60E-08	0.29773056	0	—
435	5.42E-06 ± 1.59E-08	0.29305918	0	—
436	5.50E-06 ± 1.60E-08	0.29018818	0	—
437	5.68E-06 ± 1.68E-08	0.295051	0	—
438	5.97E-06 ± 1.77E-08	0.29605739	0	—
439	5.94E-06 ± 1.82E-08	0.30678652	0	—
440	5.56E-06 ± 1.58E-08	0.284848	0	—
441	5.64E-06 ± 1.63E-08	0.28873543	0	—
442	5.73E-06 ± 1.61E-08	0.28045504	0	—
443	5.62E-06 ± 1.59E-08	0.28355699	0	—
444	5.57E-06 ± 1.54E-08	0.27695956	0	—
445	5.60E-06 ± 1.57E-08	0.28080431	0	—
446	5.55E-06 ± 1.55E-08	0.27966912	0	—
447	5.54E-06 ± 1.58E-08	0.28482643	0	—
448	5.59E-06 ± 1.57E-08	0.28175936	0	—
449	5.94E-06 ± 1.91E-08	0.3222755	0	—
450	8.60E-06 ± 3.63E-08	0.42215745	0	—

[a] Irradiance and weighted irradiance values are presented as mean ± standard error for 46 measurements at each wavelength; W = watts; S_{er} = CIE human erythema action spectrum weighting function (CIE, 1999); weighted irradiance = irradiance • S_{er}.
[b] Determined by dividing each irradiance standard error by the corresponding mean and multiplying by 100.
[c] Not applicable; $S_{er} = 0$

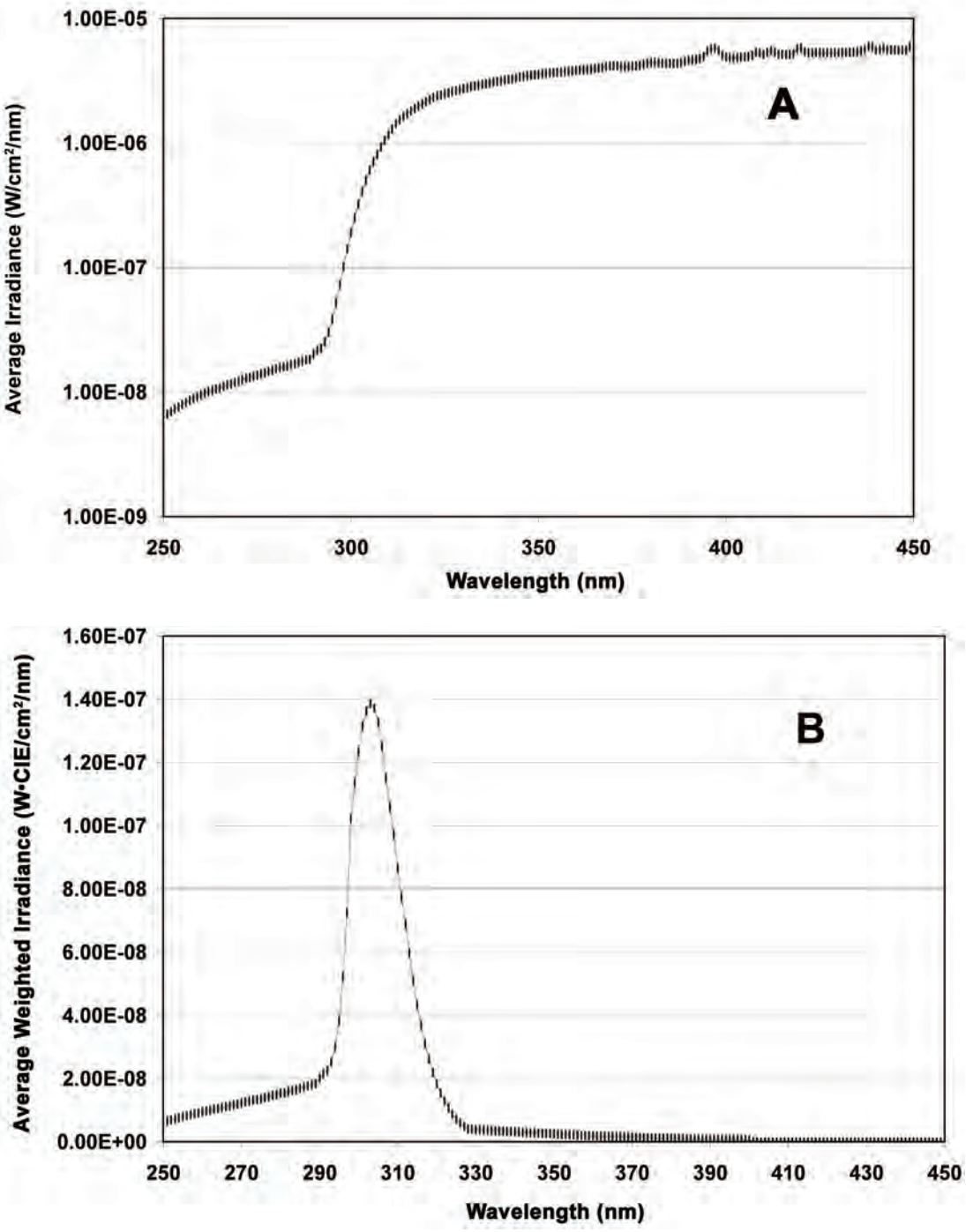

Figure E2
Average Irradiance (A) and Average Weighted Irradiance (B) for Light Source SSL-2 in the 1-Year Simulated Solar Light Study of Aloe vera

Average ± standard error (n = 46 at each wavelength); W = watts; irradiance was weighted by application of the CIE human erythema action spectrum weighting function (CIE, 1999).

APPENDIX F
DOSIMETRY OF THE SIMULATED SOLAR LIGHT

DOSIMETRY OF THE SIMULATED SOLAR LIGHT

Methods and Results

SKH-1 mice were housed in Lenderking EXP355-72 (Lenderking Caging Products, Millersville, MD) animal racks. Each animal rack had 72 individual animal compartments, and animals housed in a single rack received the same dose of simulated solar light (SSL). Mice were administered SSL each weekday afternoon for 40 weeks. An individual animal rack may have housed mice from more than one treatment definition group; however, the mice on a given animal rack all received the same dose of SSL and were all of the same sex.

As described in Appendix E, the spectral irradiance of the SSL from a filtered 6.5 kWatt (kW) xenon arc light source was measured using a spectroradiometer and recorded in units of W/cm^2 per nm. Measured irradiance was multiplied by human erythema action spectrum weighting factors defined by the Commission Internationale de l'Éclairage (CIE) to generate full-spectrum weighted irradiance with units of $W \bullet CIE/cm^2$ (Appendix E; CIE, 1999). Because 1 W/second equals 1 Joule (J), weighted irradiances can be converted to units of $mJ \bullet CIE/cm^2$ following timed exposures to SSL.

The target dose of irradiation administered to an animal rack was based on a weekly accumulation of doses dispensed in equivalent increments Monday through Friday of the same week. The target doses of irradiance for the Aloe vera study were based on historical data from published studies conducted at the Argus Research Laboratories (Sambuco *et al.*, 2003) and were defined as 0 SSL, 0.3 $MED^{instrumental}$ SSL, 0.6 $MED^{instrumental}$ SSL, and 0.9 $MED^{instrumental}$ SSL. These values were determined using a Solar Light PMA 2101 (Solar Light Company, Inc., Glenside, PA) erythemally weighted dosimeter and were determined to be equivalent to the following daily doses:

$$0.3\ MED^{instrumental}\ SSL \approx 6.85\ mJ \bullet CIE/cm^2$$

$$0.6\ MED^{instrumental}\ SSL \approx 13.70\ mJ \bullet CIE/cm^2$$

$$0.9\ MED^{instrumental}\ SSL \approx 22.55\ mJ \bullet CIE/cm^2$$

A summary of the dose of SSL delivered to each treatment group throughout the study is presented in Table F1. The results indicated that male mice were exposed to 99.68% to 100.10% of the target doses, with a relative standard error of approximately 0.3%. Female mice were exposed to 99.50% to 100.13% of the target doses, while relative standard error values ranged from 0.16% to 0.57%. Overall, the results indicated that mice were exposed to 99.50% to 100.13% of the target doses, with a relative standard error less than 0.6% in all dose groups.

TABLE F1
Doses of Light in the 1-Year Simulated Solar Light Study of Aloe vera[a]

Group	Animal Rack Number	Daily Light Dose[b]	Targeted Daily Weighted Irradiance (mJ • CIE/cm^2)	Determined Daily Weighted Irradiance (mJ • CIE/cm^2)[c]	Relative Standard Error (%)	Percent of Target
Male						
No cream	18	0.3 MED	6.85	6.8283 ± 0.0326	0.48	99.68
No cream	19	0.6 MED	13.70	13.7140 ± 0.0420	0.31	100.10
Control cream	19	0.6 MED	13.70	13.7140 ± 0.0420	0.31	100.10
Aloe gel 3%	20	0.6 MED	13.70	13.7115 ± 0.0427	0.31	100.08
Aloe gel 6%	20	0.6 MED	13.70	13.7115 ± 0.0427	0.31	100.08
Whole leaf 3%	21	0.6 MED	13.70	13.6976 ± 0.0419	0.31	99.98
Whole leaf 6%	21	0.6 MED	13.70	13.6976 ± 0.0419	0.31	99.98
Decolorized whole leaf 3%	22	0.6 MED	13.70	13.6994 ± 0.0443	0.32	100.00
Decolorized whole leaf 6%	22	0.6 MED	13.70	13.6994 ± 0.0443	0.32	100.00
Aloe-emodin 7.46 µg/g	23	0.6 MED	13.70	13.7013 ± 0.0440	0.32	100.01
Aloe-emodin 74.6 µg/g	23	0.6 MED	13.70	13.7013 ± 0.0440	0.32	100.01
No cream	24	0.9 MED	20.55	20.5481 ± 0.0543	0.26	99.99
Female						
No cream	4	0.3 MED	6.85	6.8589 ± 0.0109	0.16	100.13
No cream	5	0.6 MED	13.70	13.6531 ± 0.0684	0.50	99.66
Control cream	5	0.6 MED	13.70	13.6531 ± 0.0684	0.50	99.66
Aloe gel 3%	6	0.6 MED	13.70	13.6485 ± 0.0675	0.49	99.62
Aloe gel 6%	6	0.6 MED	13.70	13.6485 ± 0.0675	0.49	99.62
Whole leaf 3%	7	0.6 MED	13.70	13.6316 ± 0.0677	0.50	99.50
Whole leaf 6%	7	0.6 MED	13.70	13.6316 ± 0.0677	0.50	99.50
Decolorized whole leaf 3%	8	0.6 MED	13.70	13.6347 ± 0.0669	0.49	99.52
Decolorized whole leaf 6%	8	0.6 MED	13.70	13.6347 ± 0.0669	0.49	99.52
Aloe-emodin 7.46 µg/g	9	0.6 MED	13.70	13.6366 ± 0.0682	0.50	99.54
Aloe-emodin 74.6 µg/g	9	0.6 MED	13.70	13.6366 ± 0.0682	0.50	99.54
No cream	10	0.9 MED	20.55	20.4488 ± 0.1172	0.57	99.51

[a] Groups of male (racks 15, 16, and 17) and female (racks 1, 2, and 3) mice exposed to 0.00 mJ • CIE/cm^2 of simulated solar light are not presented in this table.
[b] MED = minimal erythema dose
[c] Mean daily rack dose ± standard error

APPENDIX G
INGREDIENTS, NUTRIENT COMPOSITION, AND CONTAMINANT LEVELS IN NIH-31 RAT AND MOUSE RATION

TABLE G1
Ingredients of NIH-31 Rat and Mouse Ration

Ingredients[a]	Percent by Weight
Ground whole hard wheat	35.5
Ground #2 yellow shelled corn	21.0
Ground whole oats	10.0
Wheat middlings	10.0
Fish meal (60% protein)	9.0
Soybean meal (48.5% protein)	5.0
Alfalfa meal (17% protein)	2.0
Corn gluten meal (60%)	2.0
Dicalcium phosphate[b]	1.5
Soy oil	1.5
Brewer's dried yeast	1.0
Ground limestone[b]	0.5
Premixes	0.5
Salt	0.5

[a] Ingredients are ground to pass through a U.S. Standard Screen No. 16 before mixing.
[b] The specific ingredient requirement is for cadmium content not to exceed 1 mg/kg.

TABLE G2
Vitamins and Minerals in NIH-31 Rat and Mouse Ration[a]

	Amount	Source
Vitamins		
A	22,000,000 IU	Vitamin A palmitate or acetate
D_3	3,800,000 IU	D-activated animal sterol
K_3	20 g	Menadione activity
Choline	700 g	Choline chloride
dl-α-tocopheryl acetate	15 g	
Folic acid	1 g	
Niacin	20 g	
d-Pantothenic acid	25 g	*d*-Calcium pantothenate
Riboflavin	5 g	
Thiamine	65 g	Thiamine mononitrate
B_{12}	14 g	
Pyridoxine	2 g	Pyridoxine hydrochloride
Biotin	0.12 g	*d*-Biotin
Minerals		
Magnesium	400 g	Magnesium oxide
Manganese	100 g	Manganese oxide
Iron	60 g	Iron sulfate
Zinc	10 g	Zinc oxide
Copper	4 g	Copper sulfate
Iodine	1.5 g	Calcium iodate
Cobalt	0.4 g	Cobalt carbonate

[a] Per ton (2,000 pounds) of finished product

TABLE G3
Nutrient Composition of NIH-31 Rat and Mouse Ration[a]

Nutrient	Mean ± SD	Number of Samples
Crude protein (% by weight)	21.0 ± 1.0	11
Crude fat (% by weight)	5.82 ± 0.84	11
Volatiles (% by weight)	6.56 ± 0.57	11
Vitamins		
A (µg/g)	10.80 ± 1.82	11
E (µg/g)	55.9 ± 5.0	11
B_1 (mg/g)	0.092 ± 0.004	11
Minerals		
Selenium (µg/g)	0.37 ± 0.10	11

[a] Analyses for nutrient content of NIH-31 diet were performed by standard operating procedures developed and/or validated by the NCTR Division of Chemistry.

TABLE G4
Contaminant Levels in NIH-31 Rat and Mouse Ration[a]

	Mean ± SD	Number of Lots (Number Positive)
Arsenic (µg/g)	0.12 ± 0.05	11 (9)
Cadmium (µg/g)	<MDL	11 (0)
Lead (µg/g)	0.54 ± 0.11	11 (8)
Aflatoxin B_1 (ppb)	<MDL	11 (0)
Aflatoxin B_2(ppb)	<MDL	11 (0)
Aflatoxin G_1 (ppb)	<MDL	11 (0)
Aflatoxin G_2 (ppb)	<MDL	11 (0)
Total Fumonisin (ppb)	483 ± 149	11 (11)
Pesticides (ppb)		
Heptachlor	<MDL	5 (0)
Total DDT	<MDL	5 (0)
Dieldrin	<MDL	5 (0)
PCB	<MDL	5 (0)
Malathion	<MDL	5 (0)
Lindane	<MDL	5 (0)

[a] Analyses for nutrient and contaminant content of NIH-31 diet were performed by standard operating procedures developed and/or validated by the NCTR Division of Chemistry. MDL = minimum detectable level.

APPENDIX H
SENTINEL ANIMAL PROGRAM

SENTINEL ANIMAL PROGRAM

METHODS

Rodents used in the Carcinogenesis Program of the National Toxicology Program are produced in optimally clean facilities to eliminate potential pathogens that may affect study results. The Sentinel Animal Program is part of the periodic monitoring of animal health that occurs during the toxicologic evaluation of chemical compounds. Under this program, the disease state of the rodents is monitored via serology on sera from extra (sentinel) animals in the study rooms. These animals and the study animals are subject to identical environmental conditions. The sentinel animals come from the same production source and weanling groups as the animals used for the studies of chemical compounds.

Serum samples were collected from two pairs of sentinel mice at each time point during the 1-year study. Blood from each animal was collected and allowed to clot, and the serum was separated. The serum was analyzed by enzyme-linked immunosorbent assay (ELISA) for the presence of specific antibodies by the Research Animal Diagnostic Laboratory (University of Missouri, Columbia, MO). The laboratory serology methods and viral/mycoplasma agents for which testing was performed are tabulated below; the times at which blood was collected during the studies are also listed.

Method and Test	Time of Analysis
Mice	
1-Year Study	
ELISA	
Ectromelia virus	20, 26, and 39 weeks
EDIM (epizootic diarrhea of infant mice)	20, 26, and 39 weeks
GDVII (mouse encephalomyelitis virus)	20, 26, and 39 weeks
LCM (lymphocytic choriomeningitis virus)	20, 26, and 39 weeks
MHV (mouse hepatitis virus)	20, 26, and 39 weeks
Mycoplasma pulmonis	20, 26, and 39 weeks
Parvo NS-l	20, 26, and 39 weeks
Parvovirus	20, 26, and 39 weeks
PVM (pneumonia virus of mice)	20, 26, and 39 weeks
Polyoma virus	20, 26, and 39 weeks
Reovirus 3	20, 26, and 39 weeks
Sendai	20, 26, and 39 weeks

RESULTS

All test results were negative.

APPENDIX I
INGREDIENT USE DATA FOR *ALOE BARBADENSIS* LEAF, *ALOE BARBADENSIS* LEAF EXTRACT, AND *ALOE BARBADENSIS* LEAF JUICE

Table I1
Ingredient Use Data for *Aloe Barbadensis* Leaf, *Aloe Barbadensis* Leaf Extract, and *Aloe Barbadensis* Leaf Juice[a]

Ingredient	Product Category	Concentration of Raw Material in Product	Concentration of Aloe in Raw Material (% solids)
Leaf	Personal cleanliness products	0.05%	200× (45% solids)
Leaf	Body and hand creams, lotions, and powders (excluding shaving preparations)	0.00%	200× (powder)
Leaf	Body and hand sprays	0.01%	0.25%
Leaf	Skin care preparations	0.00%	200× (powder)
Leaf	Suntan preparations	0.00%	200× (powder)
Leaf extract	Baby lotions, oils, powders, and creams	0.009%-0.1% 0.10% 1% 0.10%	Not available 5% 1× 40×
Leaf extract	Baby products	0.01%	40×
Leaf extract	Bubble baths	0.20%	4%
Leaf extract	Bath preparations	0.1%-0.5%	0.6%-4%
Leaf extract	Eyeliner	0.10%	5%
Leaf extract	Eye shadow	0.10% 0.05%-2% 0.20%	Not available 4%-5% 1× (2% solids)
Leaf extract	Eye lotion	0.10%	5%
Leaf extract	Mascara	0.10%	5%
Leaf extract	Eye makeup preparations	0.1%-1% 0.001%-0.2%	Not available 4%-5%
Leaf extract	Colognes and toilet water	3% 0.30%	0.01% 4%
Leaf extract	Powders (dusting and talcum)	0.009%-0.01% 0.01%	Not available 100% (powder)
Leaf extract	Fragrance preparations	0.50%	4%
Leaf extract	Hair conditioners	6% 0.2%-0.5% 0.05% 0.01%	0.01% 0.3%-4% 50% (0.03% solids) 1× (0.05%-1% solids)
Leaf extract	Hair sprays (aerosol fixatives)	6% 0.20% 0.05% 0.00%	0.01% 4% 50% (0.03% solids) 1×
Leaf extract	Hair straighteners	2% 0.05%	0.50% 50% (0.03% solids)
Leaf extract	Rinses (noncoloring)	0.10%	0.01%
Leaf extract	Shampoos (noncoloring)	0.10% 0.01%-0.5% 0.05% 0.0001%-0.01%	0.60% 3%-4% 50% (0.03% solids) 1× (0.05%-1% solids)
Leaf extract	Tonic, dressings, and other hair grooming aids	0.001%-0.2%	0.6%-5%
Leaf extract	Hair preparations (noncoloring)	0.00% 0.01%	0.50% 50% (0.005% solids)
Leaf extract	Blushers (all types)	0.05% 0.10%	Not available 5%
Leaf extract	Face powders	0.10% 0.20%	5% 1× (2% solids)

TABLE I1
Ingredient Use Data for *Aloe Barbadensis* Leaf, *Aloe Barbadensis* Leaf Extract, and *Aloe Barbadensis* Leaf Juice

Ingredient	Product Category	Concentration of Raw Material in Product	Concentration of Aloe in Raw Material (% solids)
Leaf extract	Foundations	0.07	Not available
		0.01%-0.5%	0.2%-0.6%
		0.02%-0.2%	4%-5%
		0.20%	1× (2% solids)
Leaf extract	Lipstick	0.8%-6%	Not available
		0.1%-5%	0.6%-5%
		1%-6%	25%
		0.30%	1× (2% solids)
Leaf extract	Makeup bases	0.20%	Not available
		0.50%	4%
		0.20%	1× (2% solids)
Leaf extract	Rouges	1%	Not available
Leaf extract	Makeup preparations	0.00%	4%
Leaf extract	Cuticle softeners	0.01%	4%
Leaf extract	Bath soaps and detergents	0.10%	0.01%
		0.05%	1% (0.0005% solids)
		0.01%-0.5%	4%-5%
		0.10%	1× (0.05%-1% solids)
Leaf extract	Deodorants (underarm)	<0.01%-0.5%	2%-4%
		0.10%	40× (16% solids)
Leaf extract	Personal cleanliness products	0.10%	0.01%
		0.01%	100% (powder)
Leaf extract	Aftershave lotions	0.50%	0.60%
		0.30%	4%
Leaf extract	Preshave lotions (all types)	0.10%	0.60%
Leaf extract	Shaving cream (aerosol, brushless, and lather)	0.003%-1%	0.6%-4%
Leaf extract	Shaving preparations	0.20%	0.60%
Leaf extract	Skin cleansing (cold creams, cleansing lotions, liquids, and pads)	0.002%-0.5%	0.6%-4%
		0.05%	1% (0.0005% solids)
		0.00%	1×
		0.01%	40×
Leaf extract	Depilatories	0.10%	0.60%
Leaf extract	Face and neck creams, lotions, and powders (excluding shaving preparations)	0.00009%-1%	0.6%-4%
Leaf extract	Body and hand creams, lotions, and powders (excluding shaving preparations)	0.1%-3%	0.1%-4%
		0.10%	1% (0.001% solids)
Leaf extract	Body and hand sprays	0.06%	0.60%
Leaf extract	Moisturizing creams, lotions, and powders	0.1%-0.5%	0.6%-4%
		0.40%	40%
		0.01%	100% (powder)
Leaf extract	Night creams, lotions, and powders	0.1%-0.5%	0.6%-4%
Leaf extract	Paste masks (mud packs)	0.50%	4%
		1%	50% (0.5% solids)
Leaf extract	Skin fresheners	0.0006%-0.5%	0.3%-4%
Leaf extract	Skin care preparations	0.10%	0.60%
		0.01%	7%
		0.01%	100% (powder)
		0.05%	10×
Leaf extract	Suntan gels, creams, and liquids	0.10%	1% (0.7%-1.2% solids)
		0.05%	25%
Leaf extract	Suntan preparations	2%	5%
		0.30%	25%

TABLE I1
Ingredient Use Data for *Aloe Barbadensis* Leaf, *Aloe Barbadensis* Leaf Extract, and *Aloe Barbadensis* Leaf Juice

Ingredient	Product Category	Concentration of Raw Material in Product	Concentration of Aloe in Raw Material (% solids)
Leaf juice	Baby shampoos	1%	1× (0.5% solids)
Leaf juice	Baby lotions, oils, powders, and creams	1%-2%	1× (0.5%-2%)
Leaf juice	Bath oils, tablets, and salts	0.10%	100% (powder)
		0.10%	1× (0.5% solids)
Leaf juice	Bubble baths	0.10%	1× (0.5% solids)
Leaf juice	Eye liner	1%	1× (0.5% solids)
Leaf juice	Eye makeup remover	1%	1× (1% solids)
Leaf juice	Eye lotion	0.05%	100% (powder)
		2%	1× (0.5% solids)
Leaf juice	Eye makeup remover	1%-2%	1× (0.5%-1% solids)
Leaf juice	Eye makeup preparations	0.01%	100% (powder)
		0.1%-5%	1× (0.5% solids)
Leaf juice	Powders (dusting and talcum)	0.00%	200× (92% solids)
Leaf juice	Hair conditioners	2%	0.90%
		0.01%-1%	100%
Leaf juice	Hair sprays (aerosol fixatives)	0.00%	0.90%
		0.00%	1×
Leaf juice	Hair straighteners	0.00%	0.50%
Leaf juice	Shampoos (noncoloring)	0.00%	Not available
		2%	0.90%
		0.01%	100%
Leaf juice	Tonics, dressings, and other hair grooming aids	0.10%	0.90%
Leaf juice	Hair dyes and colors (all types requiring caution statement)	0.10%	5%
Leaf juice	Hair preparations	0.00%	0.90%
Leaf juice	Blushers (all types)	0.10%	100% (powder)
Leaf juice	Face powders	0.10%	Not available
		0.05%	100% (powder)
		0.10%	1× (0.5 solids)
Leaf juice	Foundations	0.01%	100% (powder)
		4%	1× (0.5% solids)
Leaf juice	Lipstick	0.50%	(92% solids)
Leaf juice	Rouges	0.05%	1× (0.5% solids)
Leaf juice	Makeup preparations	0.20%	1× (0.5% solids)
Leaf juice	Nail creams and lotions	2%	1× (2% solids)
Leaf juice	Bath soaps and detergents	0.50%	(5% solids)
		0.00%	0.10%
		0.05%-1%	1× (0.5%-1% solids)
		0.01%	200× (~90% solids)
Leaf juice	Douches	1%	1× (0.5% solids)
Leaf juice	Feminine hygiene deodorants	0.10%	Not available
Leaf juice	Personal cleanliness products	1%	1× (0.5% solids)
Leaf juice	Aftershave lotions	0.50%	(5% solids)
		0.10%	(92% solids)
		1%-5%	1× (0.5%-2% solids)
Leaf juice	Shaving cream (aerosol, brushless, and lather)	0.10%	100%
Leaf juice	Skin cleansing, (cold creams, cleansing lotions, liquids, and pads)	0.50%	(5% solids)
		0.01%-0.5%	0.1%-0.5%
		2%-7%	1× (0.5%-2% solids)
Leaf juice	Face and neck creams, lotions, and powders (excluding shaving preparations)	0.50%	(5% solids)
		0.1%-0.5%	0.1%-1%
		1%-2%	1× (0.5%-1% solids)

TABLE I1
Ingredient Use Data for *Aloe Barbadensis* Leaf, *Aloe Barbadensis* Leaf Extract, and *Aloe Barbadensis* Leaf Juice

Ingredient	Product Category	Concentration of Raw Material in Product	Concentration of Aloe in Raw Material (% solids)
Leaf juice	Body and hand creams, lotions, and powders (excluding shaving preparations)	0.50%	(5% solids)
		0.20%	(92% solids)
		3%	0.90%
		1%-5%	1× (0.5%-1% solids)
		0.01%	200× (~90% solids)
Leaf juice	Body and hand sprays	0.001%	0.10%
Leaf juice	Moisturizing creams, lotions, and powders	0.50%	(5% solids)
		0.20%	100% (powder)
		3%	1× (0.5%)
Leaf juice	Night creams, lotions, and powders	0.50%	(92% solids)
		1%-5%	1× (0.5%-1% solids)
Leaf juice	Paste masks (mud packs)	5%	1× (0.5% solids)
Leaf juice	Skin fresheners	2%	1× (0.5%-1%)
		0.03%	10× (5% solids)
Leaf juice	Skin care preparations	0.01%	(92% solids)
		0.10%	100% (powder)
		0.1%-3%	1× (0.5%-2% solids)
		0.10%	40× (21% solids)
Leaf juice	Suntan gels, creams, and liquids	3%-4%	1× (0.5% solids)
		0.10%	40× (21% solids)
Leaf juice	Indoor tanning preparations	0.02%	(92% solids)
Leaf juice	Suntan preparations	5%	1× (0.5% solids)

[a] Data are the results of a survey of the cosmetic industry that was conducted by the Cosmetic, Toiletry, and Fragrance Association on use levels of the aloe raw materials in final products in 2002. The table was prepared in September 2002 and was provided by Linda Loretz, Ph.D., Director, Safety and Regulatory Toxicology, Cosmetic, Toiletry, and Fragrance Association.

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