International Journal of Toxicology, 26(Suppl. 3):31–77, 2007

ISSN: 1091-5818 print / 1092-874X online
DOI: 10.1080/10915810701663150

Final Report on the Safety Assessment of Ricinus Communis (Castor) Seed Oil, Hydrogenated Castor Oil, Glyceryl Ricinoleate, Glyceryl Ricinoleate SE, Ricinoleic Acid, Potassium Ricinoleate, Sodium Ricinoleate, Zinc Ricinoleate, Cetyl Ricinoleate, Ethyl Ricinoleate, Glycol Ricinoleate, Isopropyl Ricinoleate, Methyl Ricinoleate, and Octyldodecyl Ricinoleate[1]

The oil derived from the seed of the Ricinus communis plant and its primary constituent, Ricinoleic Acid, along with certain of its salts and esters function primarily as skin-conditioning agents, emulsion stabilizers, and surfactants in cosmetics, although other functions are described. Ricinus Communis (Castor) Seed Oil is the naming convention for castor oil used in cosmetics. It is produced by cold pressing the seeds and subsequent clarification of the oil by heat. Castor oil does not contain ricin because ricin does not partition into the oil. Castor oil and Glyceryl Ricinoleate absorb ultraviolet (UV) light, with a maximum absorbance at 270 nm. Castor oil and Hydrogenated Castor Oil reportedly were used in 769 and 202 cosmetic products, respectively, in 2002; fewer uses were reported for the other ingredients in this group. The highest reported use concentration (81%) for castor oil is associated with lipstick. Castor oil is classified by Food and Drug Administration (FDA) as generally recognized as safe and effective for use as a stimulant laxative. The Joint Food and Agriculture Organization (FAO)/World Health Organization (WHO) Expert Committee on Food Additives established an acceptable daily castor oil intake (for man) of 0 to 0.7 mg/kg body weight. Castor oil is hydrolyzed in the small intestine by pancreatic enzymes, leading to the release of glycerol and Ricinoleic Acid, although 3,6-epoxyoctanedioic acid, 3,6-epoxydecanedioic acid, and 3,6-epoxydodecanedioic acid also appear to be metabolites. Castor oil and Ricinoleic Acid can enhance the transdermal penetration of other chemicals. Although chemically similar to prostaglandin E_1, Ricinoleic Acid did not have the same physiological properties. These ingredients are not acute toxicants, and a National Toxicology Program (NTP) subchronic oral toxicity study using castor oil at concentrations up to 10% in the diet of rats was not toxic. Other subchronic studies of castor oil produced similar findings. Undiluted castor oil produced minimal ocular toxicity in one study, but none in another. Undiluted castor oil was severely irritating to rabbit skin in one study, only slightly irritating in another, mildly irritating to guinea pig and rat skin, but not irritating to miniature swine skin. Ricinoleic Acid was nonirritating in mice and in one rabbit study, but produced well-defined erythema at abraded and intact skin sites in another rabbit study. Zinc Ricinoleate was not a sensitizer in guinea pigs. Neither castor oil nor Sodium Ricinoleate was genotoxic in bacterial or mammalian test systems. Ricinoleic Acid produced no neoplasms or hyperplasia in one mouse study and was not a tumor promoter in another mouse study, but did produce epidermal hyperplasia. Castor oil extract had a strong suppressive effect on S_{180} body tumors and ARS ascites cancer in male Kunming mice. No dose-related reproductive toxicity was found in mice fed up to 10% castor oil for 13 weeks. Female rats injected intramuscularly with castor oil on the first day after estrus had suppressed ovarian folliculogenesis and anti-implantation and abortive effects. Castor oil used as a vehicle control in rats receiving subcutaneous injections had no effect on spermatogenesis. A methanol extract of *Ricinus communis* var. *minor* seeds (ether-soluble fraction) produced anti-implantation, anticonceptive, and estrogenic activity in rats and mice. Clinically, castor oil has been used to stimulate labor. Castor oil is not a significant skin irritant, sensitizer, or photosensitizer in human clinical tests, but patients with occupational dermatoses may have a positive reaction to castor oil or Ricinoleic Acid. The instillation of a castor oil solution into the eyes of nine patients resulted in mild and transient discomfort and minor epithelial changes. In another study involving 100 patients, the instillation of castor oil produced corneal epithelial cell death and continuity breaks in the epithelium. Because castor oil contains Ricinoleic Acid as the primary fatty acid group, the Cosmetic Ingredient Review (CIR) Expert Panel considered the safety test data on the oil broadly applicable to this entire group of cosmetic ingredients. The available data demonstrate few toxic effects. Although animal studies indicate no significant irritant or sensitization potential, positive reactions to Ricinoleic Acid in selected populations with identified dermatoses did suggest that sensitization reactions may be higher in that population. Overall, however, the clinical experience suggests that sensitization reactions are seen infrequently. In the absence of inhalation toxicity data on these ingredients, the Panel determined that these ingredients can be used safely in aerosolized cosmetic products because the particle sizes produced are not respirable. Overall, the CIR Expert Panel concluded that these cosmetic ingredients are safe in the practices of use and concentrations as described in this safety assessment.

Received 14 May 2007; accepted 24 August 2007.

Address correspondence to Wilbur Johnson, Jr., Senior Scientific Analyst and Writer, Cosmetic Ingredient Review, 1101 17th Street, NW, Suite 412, Washington, DC 20036, USA.

[1]Reviewed by the Cosmetic Ingredient Review Expert Panel.

INTRODUCTION

The safety of the following ingredients in cosmetics is reviewed in this report: Ricinus Communis (Castor) Seed Oil, Hydrogenated Castor Oil, Glyceryl Ricinoleate, Glyceryl Ricinoleate SE, Ricinoleic Acid, Potassium Ricinoleate, Sodium Ricinoleate, Zinc Ricinoleate, Cetyl Ricinoleate, Ethyl Ricinoleate, Glycol Ricinoleate, Isopropyl Ricinoleate, Methyl Ricinoleate, and Octyldodecyl Ricinoleate. For the sake of brevity, Ricinus Communis (Castor) Seed Oil will be referred to as castor oil.

Glyceryl Ricinoleate and Glyceryl Ricinoleate SE are esters of glycerin and Ricinoleic Acid (component of castor oil). A Cosmetic Ingredient Review (CIR) safety assessment (Elder 1988) of Glyceryl Ricinoleate in cosmetics was published with the conclusion that the available data are insufficient to support the safety of this ingredient as used in cosmetics. The following data were needed for completion of this safety assessment: (1) 28-day chronic dermal toxicity (guinea pigs) and (2) clinical sensitization and photosensitization studies (or an appropriate ultraviolet spectrum instead of the photosensitization data).

Glyceryl Ricinoleate and Glyceryl Ricinoleate SE are included in the present safety assessment in the expectation that the available data on castor oil, Ricinoleic Acid, salts of Ricinoleic Acid, and other esters of Ricinoleic Acid will contribute to a resolution of the CIR Expert Panel's data needs for Glyceryl Ricinoleate and Glyceryl Ricinoleate SE.

CHEMISTRY

Definition and Structure

Ricinus Communis (Castor) Seed Oil

According to the *International Cosmetic Ingredient Dictionary and Handbook* (Gottschalck and McEwen 2004), Ricinus Communis (Castor) Seed Oil (CAS no. 8001-79-4) is defined as the fixed oil that is obtained from the seeds of *Ricinus communis*. Other names for this oil include: Castor Oil, Castor Oil (Ricinus communis L.), Castor Seed Oil, Ricinus Communis, and Ricinus Communis Oil (Gottschalck and McEwen 2004), and Ricinus Oil (TNO BIBRA International Ltd. 1999). As given by TNO BIBRA International Ltd. (1999), the structural formula is:

where R represents a fatty acyl group [$CH_3(CH_2)_5CH(OH)CH_2CH{=}CH(CH_2)_7COOH$] that is typically derived from Ricinoleic Acid. Ricinoleic Acid accounts for 87% to 90% of the fatty acyl groups, and the following other fatty acids comprise the remaining fatty acyl groups: oleic acid (2% to 7%), linoleic acid (3% to 5%), palmitic acid (1% to 2%), stearic acid (1%), dihydrostearic acid (1%), and trace amounts of other fatty acyl groups (not specified) (TNO BIBRA International Ltd. 1999).

According to other sources, Castor Oil contains 2.4% lauric acid (Larsen et al. 2001), 2% to 5% linoleic acid (Maier et al. 1999), and globulin, cholesterol, lipase, vitamin E, and β-sitosterol (Scarpa and Guerci 1982).

Hydrogenated Castor Oil

According to the *International Cosmetic Ingredient Dictionary and Handbook* (Gottschalck and McEwen 2004), Hydrogenated Castor Oil (CAS no. 8001-78-3) is defined as the end product of controlled hydrogenation of Ricinus Communis (Castor) Seed Oil. Castor Oil, Hydrogenated is another technical name for this ingredient, and Castor Wax (trade name) and Montane 481 (trade name mixture) are other names under which Hydrogenated Castor Oil is marketed (Gottschalck and McEwen 2004).

Ricinoleic Acid, Its Salts, and Simple Esters

Table 1 includes the structures, definitions, and cosmetic ingredient functions of Glyceryl Ricinoleate, Glyceryl Ricinoleate SE, Ricinoleic Acid, Potassium Ricinoleate, Sodium Ricinoleate, Zinc Ricinoleate, Cetyl Ricinoleate, Ethyl Ricinoleate, Glycol Ricinoleate, Isopropyl Ricinoleate, Methyl Ricinoleate, and Octyldodecyl Ricinoleate.

Chemical and Physical Properties

Properties of castor oil and Hydrogenated Castor Oil are summarized in Table 2 and properties of the following ingredients are summarized in Table 3: Ricinoleic Acid, Ethyl Ricinoleate, Methyl Ricinoleate, Sodium Ricinoleate, Potassium Ricinoleate, and Zinc Ricinoleate.

Composition/Impurities

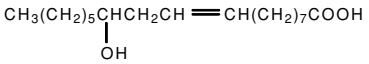
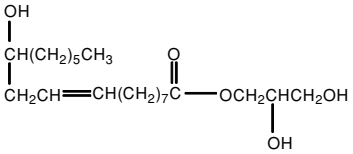
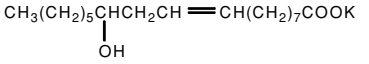
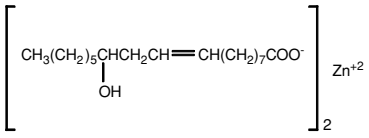
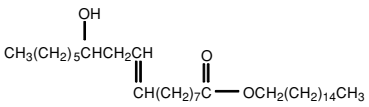
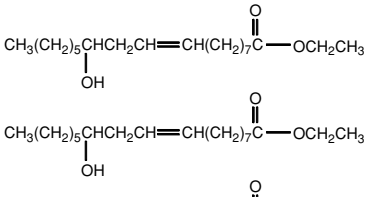
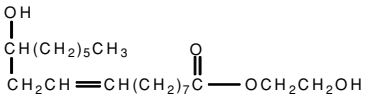
Castor Oil

The following three peaks were observed in a gas-liquid chromatogram on castor oil: palmitic acid (1.7%), C18 acids (stearic, oleic, and linoleic acids) (15.9%), and Ricinoleic Acid (82.4%) (Kato and Yamaura 1970).

According to the *Food Chemicals Codex* (National Academy of Sciences 1996), the requirements for castor oil are as follows: free fatty acids (passes test), heavy metals, as Pb (not more than 10 mg/kg), hydroxyl value (between 160 and 168), iodine value (between 83 and 88), saponification value (between 176 and 185), and specific gravity (between 0.952 and 0.966). The test for free fatty acids is described as follows: free fatty acids (10 g) are dissolved in 50 ml of a mixture of equal volumes of alcohol and ether (which has been neutralized to phenolphthalein with not more than 7 ml of 0.1 N sodium hydroxide). Phenolphthalein (1 ml) is added, followed by titration with 0.1 N sodium hydroxide until the solution remains pink (after shaking).

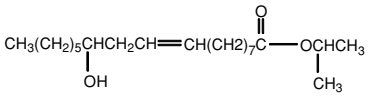
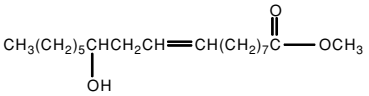
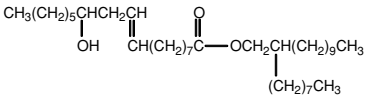
Similarly, the United States Pharmacopeia (Committee of Revision of the United States Pharmacopeial Convention 2004a)

TABLE 1
Structures, definitions, and functions of ricinoleic acid, its salts, and esters (Gottschalck and McEwen 2004)

Structure	Definition	Function
	**Ricinoleic Acid** (CAS nos. 141-22-0 and 7431-95-0) is an unsaturated fatty acid. Other names include: 12-Hydroxy-9-Octadecenoic Acid; 9-Octadecenoic Acid, 12-Hydroxy-; Ricinic Acid; and Ricinolic Acid	Surfactant–cleansing agent
	**Glyceryl Ricinoleate** (CAS nos.141-08-2, 1323-38-2, and 5086-52-2) is the monoester of glycerin and ricinoleic acid. Other names include: Glycerin Monoricinoleate; Glycerol Monoricinoleate; Glyceryl Monoricinoleate, 12-Hydroxy-9-Octadecenoic Acid, Monoester with 1,2,3-Propanediol; Ricinoleic Acid Monoglyceride; and Ricinolein, 1-Mono-	Skin-conditioning agent—emollient and surfactant–emulsifying agent
See above	**Glyceryl Ricinoleate SE** is a self-emulsifying grade of Glyceryl Ricinoleate containing sodium and/or potassium stearate	Skin-conditioning agent—emollient and surfactant–emulsifying agent
	**Potassium Ricinoleate** (CAS no. 7492-30-0) is the potassium salt of Ricinoleic Acid. Other names include: 12-Hydroxy-9-Octadecenoic Acid, Monopotassium Salt and 9-Octadecenoic Acid, 12-Hydroxy-, Monopotassium Salt	Surfactant–cleansing agent and surfactant–emulsifying agent
See above, except with Na in place of K	**Sodium Ricinoleate** (CAS no. 5323-95-5) is the sodium salt of Ricinoleic Acid. Other names include: 12-Hydroxy-9-Octadecenoic Acid, Sodium Salt; 9-Octadecenoic Acid, 12-Hydroxy, Sodium Salt; and Sodium Ricinate	Surfactant–cleansing agent and surfactant–emulsifying agent
	**Zinc Ricinoleate** (CAS no. 13040-19-2) is the zinc salt of Ricinoleic Acid. Other names include: 12-Hydroxy-9-Octadecenoic Acid, Zinc Salt and 9-Octadecenoic Acid, 12-Hydroxy-, Zinc Salt	Anticaking agent, deodorant agent, and opacifying agent
	**Cetyl Ricinoleate** (CAS no.10401-55-5) is the ester of cetyl alcohol and Ricinoleic Acid. Other names include: Hexadecyl 12-Hydroxy-9-Octadecenoate; 12-Hydroxy-9-Octadecenoic Acid, Hexadecyl Ester; 9-Octadecenoic Acid, 12-Hydroxy-, Hexadecyl Ester; and Ricinoleic Acid, Hexadecyl Ester	Skin-conditioning agent—occlusive
	**Ethyl Ricinoleate** (CAS no. 55066-53-0) is the ester of ethyl alcohol and Ricinoleic Acid. Other names include: Ethyl 12-Hydroxy-9-Octadecenoate; 12-Hydroxy-9-Octadecenoic Acid-, Ethyl Ester; 9-Octadecenoic Acid, 12-Hydroxy-, Ethyl Ester; and Ricinoleic Acid, Monoethyl Ester	Fragrance ingredient and skin-conditioning agent—emollient
	**Glycol Ricinoleate** (CAS nos. 106-17-2 and 40275-40-9) is the ester of ethylene glycol and Ricinoleic Acid. Other names include: 1,2-Ethanediol Monoricinoleate; Ethylene Glycol Monoricinoleate; Glycol Monoricinoleate; 2-Hydroxyethyl 12-Hydroxy-9-Octadecenoate; 9-Octadecenoic Acid, 12-Hydroxy-, 2-Hydroxyethyl Ester; and Ricinoleic Acid, 2-Hydroxyethyl Ester	Emulsion stabilizer and skin-conditioning agent—emollient

(Continued on next page)

TABLE 1
Structures, definitions, and functions of ricinoleic acid, its salts, and esters (Gottschalck and McEwen 2004) *(Continued)*

Structure	Definition	Function
	**Isopropyl Ricinoleate** (CAS no. 71685-99-9) is the ester of isopropyl alcohol and Ricinoleic Acid. Other names include: 12-Hydroxy-9-Octadecenoic Acid, 1-Methylethyl Ester; 1-Methylethyl, 12-Hydroxy-9-Octadecenoate; 9-Octadecenoic Acid, 12-Hydroxy-, 1-Methylethyl Ester; and Ricinoleic Acid, Isopropyl Ester	Skin-conditioning agent—emollient
	**Methyl Ricinoleate** (CAS nos. 141-24-2 and 23224-20-6) is the ester of methyl alcohol and ricinoleic acid. Other names include: Castor Oil Acid, Methyl Ester; 12-Hydroxy-9-Octadecenoic Acid, Methyl Ester; Methyl Castor Oil Fatty Acid; Methyl 12-Hydroxy-9-Octadecenoate; 9-Octadecenoic Acid, 12-Hydroxy-, Methyl Ester; and Ricinoleic Acid, Methyl Ester	Skin-conditioning agent—emollient
	**Octyldodecyl Ricinoleate** (CAS nos. 79490-62-3 and 125093-27-8) is the ester of Octyldodecanol and Ricinoleic Acid. Other names include: 12-Hydroxy-9-Octadecenoic Acid, 2-Octyldodecyl Ester; 9-Octadecenoic Acid, 12-Hydroxy-, 2-Octyldodecyl Ester; 2-Octyldodecyl 12-Hydroxy-9-Octadecenoate; and 2-Octyldodecyl Ricinoleate	Skin-conditioning agent—occlusive

has published the following requirements for castor oil: free fatty acids ("the free fatty acids in 10 g require for neutralization not more than 3.5 ml of 0.10 N sodium hydroxide") heavy metals (0.001%), hydroxyl value (between 160 and 168), iodine value (between 83 and 88), saponification value (between 176 and 182), and specific gravity (between 0.957 and 0.961).

Hydrogenated Castor Oil

According to Nikitakis and McEwen (1990), Hydrogenated Castor Oil consists primarily of glyceryl-tri-hydroxystearate. An earlier source (Binder et al. 1970) indicates that 12-hydroxystearic acid is the principal constituent of Hydrogenated Castor Oil.

The National Formulary (Committee of Revision of the United States Pharmacopeial Convention 2004b) has published the following requirements for Hydrogenated Castor Oil: free fatty acids (free fatty acids in 20 g require for neutralization not more than 11.0 ml of 0.1 N sodium hydroxide), heavy metals (0.001%), hydroxyl value (between 154 and 162), iodine value (not more than 5), and saponification value (between 176 and 182).

Methods of Production

Castor Oil

According to Kathren et al. (1959), castor oil is extracted from the bean of the tropical plant, *Ricinus communis*, by either of the following two methods: (1) use of a solvent [not stated] or (2) mechanical crushing, grinding, and pressing. The former method is more efficient, leaving a more dessicated residue. This residue, known as pomace, contains ricin and a separate allergen (Ratner and Gruehl 1929). Following extraction from the castor bean, neither of these two fractions is present in castor oil (Ordman 1955).

According to a more recent reference (Cornell University 2001), ricin does not partition into the oil because of its water solubility. Therefore, castor oil does not contain ricin, provided that cross-contamination does not occur during its production.

Other sources indicate that castor oil is produced via the cold pressing of the seeds of *Ricinus communis* (Hui 1996) and by cold expression and subsequent clarification of the oil by heat (Gennaro 1990).

Hydrogenated Castor Oil

According to Nikitakis and McEwen (1990), Hydrogenated Castor Oil is obtained by the controlled hydrogenation of pure Castor Oil. Another source (Campbell & Co. 2002) indicates that Hydrogenated Castor Oil (hard, brittle wax) results from the addition of hydrogen to Castor Oil in the presence of a nickel catalyst.

Ricinoleic Acid

One of the simplest methods for obtaining Ricinoleic Acid is the hydrolysis of castor oil. Ricinoleic acid may then be separated from hydrolyzed castor Oil by successive additions of urea (i.e., urea complexing) (Chakravarty and Bose 1963). According to a more recent source, Ricinoleic Acid results from the saponification of castor oil (Lewis 1997).

TABLE 2
Chemical and physical properties of Castor Oil and Hydrogenated Castor Oil

Property	Value	Reference
	Castor Oil	
Color/Form	Colorless to pale-yellow viscous liquid	Lewis 2000
Taste	Slightly acrid	National Toxicology Program (NTP) 2003
Specific gravity	0.945 to 0.965 at 25°/25°C; 0.961 to 0.963 at 15.5°/15.5°C	NTP 2003
Density	0.953 to 0.965 g/ml at 20°C	NTP 2003
Viscosity	6 to 8 poises at 25°C	NTP 2003
	283 cP at 37°C	Fredholt et al. 2000
Solubility	In water (<1 mg/ml at 20°C); in DMSO (≥100 mg/ml at 20°C); In 95% ethanol (≥ 100 mg/ml at 20°C); in methanol (miscible); In acetone (≥100 mg/ml at 20°C)	NTP 2003
	Miscible in absolute alcohol, glacial acetic acid, chloroform, and ether	Lewis 2000
Surface tension	39.0 dynes/cm at 20°C; 35.2 dynes/cm at 80°C	NTP 2003
Refractive index	1.4784 at 20°C; 1.473 to 1.477 at 25°C; 1.466 to 1.473 at 40°C	NTP 2003
Optical rotation	Not less than +3.5°	NTP 2003
Flash point	229°C (445°F)	NTP 2003
Autoignition temperature	448°C (840°F)	NTP 2003
Melting point	−12°C	NTP 2003
Boiling point	313°C	NTP 2003
Freezing point	−10°C	NTP 2003
Saponification value	178	NTP 2003
Iodine value	85	NTP 2003
Acid value	<4	NTP 2003
Reichert-Meissl value	<0.5	NTP 2003
Polenske value	<0.5	NTP 2003
Acetyl value	144 to 150	NTP 2003
Hydroxyl value	161 to 169	NTP 2003
	Hydrogenated Castor Oil	
Color/Form	White to cream waxy solid	Nikitakis and McEwen 1990
Odor	As specified by the buyer	Nikitakis and McEwen 1990
	Slight characteristic odor	Allegri et al. 1981
Taste	Tasteless	Allegri et al. 1981
Identification	Positive: Close match to a standard IR spectrum with no indication of foreign materials	Nikitakis and McEwen 1990
Specific gravity	0.98 to 1.04 at 25°/25°C	Nikitakis and McEwen 1990
Melting range	78° to 90°C	Nikitakis and McEwen 1990
Acid value	5.0 maximum	Nikitakis and McEwen 1990
Saponification value	175 to 185	Nikitakis and McEwen 1990
Iodine value	As specified by the buyer	Nikitakis and McEwen 1990
	<5.0	Budavari 1989
Hydroxyl value	155 max	KIC Chemicals, Inc. 2004

(Continued on next page)

TABLE 2
Chemical and physical properties of Castor Oil and Hydrogenated Castor Oil *(Continued)*

Property	Value	Reference
Density	0.98 g/cm^3	Physical & Theoretical Chemistry Lab 2004
Solubility	Insoluble in water and organic solvents.	Lewis 1997
	Soluble in chloroform; very slightly soluble in benzene; virtually insoluble in alcohol, ethyl ether, carbon disulfide, and water	Allegri et al. 1981
Drying loss	When dried under vacuum in presence of phosphoric acid anhydride, should lose no more than 0.1% of weight	Allegri et al. 1981
Sulfuric ash	Not more than 0.01%	Allegri et al. 1981

Methyl Ricinoleate

According to Masri et al. (1962), Methyl Ricinoleate has been obtained by alcoholysis of castor oil, followed by fractional distillation of the crude esters and further purification by low temperature crystallization. According to a more recent source, Methyl Ricinoleate is a product of either the esterification of Ricinoleic Acid or the alcoholysis of castor oil; the product is purified by vacuum distillation (Lewis 1997).

Ultraviolet Absorption

Castor Oil

A ultraviolet (UV) absorption spectrum on castor oil indicates maximum absorbance at 270 nm; absorption peaks at 280 and 260 nm were also observed (Sasol North America, Inc. no date).

Glyceryl Ricinoleate

A UV absorption spectrum on Glyceryl Ricinoleate indicates maximum absorbance at 270 nm and an absorption peak at 280 nm (Sasol North America, Inc. no date).

Analytical Methods

Castor Oil

Castor oil has been analyzed using the following methods: mass spectroscopy (Ayorinde et al. 2000), gas-liquid chromatography (Kato and Yamaura 1970; Ramsey et al. 1980), thin-layer chromatography (Srinivasulu and Mahapatra 1973), and nonaqueous reverse-phase high-performance liquid chromatography–mass spectrometry (Stübiger et al. 2003).

Methyl Ricinoleate

Methyl Ricinoleate has been analyzed by nuclear magnetic resonance (NMR) spectroscopy (Wineburg and Swern 1973) and gas-liquid chromatography (Paulus and Champion 1972).

Reactivity

Castor Oil

Castor oil is combustible when exposed to heat, and spontaneous heating may occur (Lewis 2000).

According to Gunstone (1989), Castor Oil is a precursor of octadeca-9,11-dienoic acid (dehydration reaction); 12-hydroxystearic acid (hydrogenation); heptanol and undec-10-enoic acid (pyrolysis); and sebacic acid, 10-hydroxydecanoic acid, octan-2-ol, and octan-2-one (alkaline fusion).

According to Achaya (1971), the hydrogenation of Castor oil produces 12-hydroxystearate, stearate, 12-ketostearate, and ricinoleyl alcohol. Dehydrated castor oil is a dehydration product of castor oil, and contains a fair proportion of conjugated dienes.

Hydrogenated Castor Oil

Hydrogenated Castor Oil is combustible and is incompatible with strong oxidizing agents (Physical and Theoretical Chemistry Lab 2004).

USE

Purpose in Cosmetics

Ricinus Communis (Castor) Seed Oil functions as a fragrance ingredient and a skin-conditioning agent—occlusive in cosmetic products, and Hydrogenated Castor Oil functions as a skin-conditioning agent—occlusive and a viscosity increasing agent—nonaqueous (Gottschalck and McEwen 2004). The cosmetic ingredient functions of Ricinoleic Acid and some of its salts and esters are included in Table 1. Except for the anticaking, deodorant, and opacifying agent functions of Zinc Ricinoleate and the surfactant function of Ricinoleic Acid, Potassium Ricinoleate, Sodium Ricinoleate, Glyceryl Ricinoleate, and Glyceryl Ricinoleate SE, these ingredients function as skin-conditioning agents in cosmetic products. In addition to functioning as a skin-conditioning agent, Ethyl Ricinoleate also functions as a fragrance ingredient. Glycol Ricinoleate also functions as an emulsion stabilizer, and Glyceryl Ricinoleate and Glyceryl Ricinoleate SE also function as surfactants in cosmetics.

Extent of Use in Cosmetics

Frequency of use data based on industry reports to the Food and Drug Administration (FDA) in 2002 indicate that Ricinus

TABLE 3
Chemical and physical properties of Ricinoleic Acid, its salts, and esters

Property	Value	Reference
	Ricinoleic Acid	
Molecular weight	298.47	Lide and Frederikse 1993
Color/Form	Yellow fatty acid	Grant 1972
	Yellow, viscous liquid	Nikitakis and McEwen 1990
	Colorless to yellow, viscous liquid	Lewis 1997
Odor	As specified by the buyer	Nikitakis and McEwen 1990
Identification	Positive: Close match to a standard IR spectrum with no indication of foreign materials	Nikitakis and McEwen 1990
Specific gravity	0.940 at 25°/25°C	Nikitakis and McEwen 1990
Density	0.9450	Lide and Frederikse 1993
	0.940 at 27.4°/4°C	Lewis 1997
Solubility	Insoluble in water	Grant 1972
	Soluble in alcohol, acetone, ether, and chloroform	Budavari 1989
	Soluble in ethyl alcohol and diethyl ether	Lide and Frederikse 1993
	Soluble in most organic solvents; insoluble in water	Lewis 1997
Refractive index	1.4716 at 20°C	Nikitakis and McEwen 1990
	1.4716	Lide and Frederikse 1993
	1.4697 at 20°C	Lewis 1997
Optical rotation	$[\alpha]^{22/D}$ of +6.67°C	Budavari 1989
	+6.67° at 22°C	Nikitakis and McEwen 1990
	$[\alpha]^{12/D}$of +5.05	Lide and Frederikse 1993
	Dextrorotatory	Lewis 1997
Melting point	5.5°C	Nikitakis and McEwen 1990
	α : 7.7°C; β: 16°C; γ: 5.5°C	Lide and Frederikse 1993
	5.5°C	Lewis 1997
Boiling point	226.8°C	Lide and Frederikse 1993
	226°C at 10 mm Hg	Lewis 1997
Combustibility	Combustible	Lewis 1997
Neutralization value	187.98	Budavari 1989
	188	Nikitakis and McEwen 1990
Iodine value	85.05	Budavari 1989
	85	Nikitakis and McEwen 1990
	Potassium Ricinoleate	
Color/Form	White paste	Lewis 1997
Solubility	Soluble in water	Lewis 1997
Combustibility	Combustible	Lewis 1997
	Sodium Ricinoleate	
Color/Form	White or slightly yellow powder	Lewis 1997
Odor	Nearly odorless	Lewis 1997
Solubility	Soluble in water or alcohol	Lewis 1997
Combustibility	Combustible	Lewis 1997
	Zinc Ricinoleate	
Color/Form	Fine white powder	Lewis 1997
Odor	Faint fatty acid odor	Lewis 1997
Density	1.10 at 25°/25°C	Lewis 1997

(Continued on next page)

TABLE 3
Chemical and physical properties of Ricinoleic Acid, its salts, and esters *(Continued)*

Property	Value	Reference
Melting point	92°C to 95°C	Lewis 1997
Combustibility	Combustible	Lewis 1997
	Methyl Ricinoleate	
Color/Form	Colorless liquid	Lewis 1997
Solubility	Insoluble in water; soluble in alcohol and ether	Lewis 1997
Density	0.9236 at 22°/4°C	Lewis 1997
Refractive index	1.4628	Lewis 1997
Boiling point	245°C at 10 mm Hg	Lewis 1997
Flash point	−4.5°C	Lewis 1997
Combustibility	Combustible	Lewis 1997
	Ethyl Ricinoleate	
Molecular weight	326.52	Lide and Frederikse 1993
Density	0.9045	Lide and Frederikse 1993
Refractive index	1.4618	Lide and Frederikse 1993
Optical rotation	$[\alpha]^{22/D}$ of +5.3	Lide and Frederikse 1993
Boiling point	258°C	Lide and Frederikse 1993

Communis (Castor) Seed Oil and Hydrogenated Castor Oil are being used in a total of 769 and 202 cosmetic products, respectively. Glyceryl Ricinoleate (16 cosmetic products), Ricinoleic Acid (6 cosmetic products), Sodium Ricinoleate (12 cosmetic products), Zinc Ricinoleate (3 cosmetic products), and Cetyl Ricinoleate (55 cosmetic products) were also reported. These data are given in Table 4 (FDA 2002).

Use concentration data obtained from an industry survey by the Cosmetic, Toiletry, and Fragrance Association (CTFA 2004) indicate that Ricinus Communis (Castor) Seed Oil and Hydrogenated Castor Oil are being used in cosmetics at concentrations up to 81% and 39%, respectively (see Table 4).

Other maximum ingredient use concentrations reported in Table 4 are as follows: Glyceryl Ricinoleate (up to 12%), Zinc Ricinoleate (up to 2%), Cetyl Ricinoleate (up to 10%), and Octyldodecyl Ricinoleate (up to 5%). Of the product categories listed in Table 4, the highest reported use concentration for Ricinus Communis (Castor) Seed Oil is associated with lipsticks. Two other sources indicate that lipstick contains 44% *w*/*w* castor oil (Hui 1996) and 10% to 67% castor oil (Smolinske 1992).

Certain ingredients in this group are reportedly used in a given product category, but the concentration of use is not available. For other ingredients in this group, information regarding use concentration for specific product categories is provided, but the number of such products is not known. In still other cases, an ingredient is not in current use, but may be used in the future. For example, Potassium Ricinoleate, Ethyl Ricinoleate, Glycol Ricinoleate, Isopropyl Ricinoleate, and Methyl Ricinoleate are not currently reported to FDA as in use, nor are there industry survey data indicating a current use concentration.

Cosmetic products containing Ricinus Communis (Castor) Seed Oil, Hydrogenated Castor Oil, and Ricinoleic Acid and its salts and esters are applied to most areas of the body, and could come in contact with the oral, ocular, or nasal mucosae. These products may be used on a daily basis, and could be applied frequently over a period of several years.

Product categories for Ricinus Communis (Castor) Seed Oil, Hydrogenated Castor Oil, and salts and esters of Ricinoleic Acid include potential aerosol applications. Bower (1999) characterized the typical diameter of anhydrous hair spray particles in the 60- to 80-μm range (typically, $<1\%$ are below 10 μm). Pump hair sprays, in contrast, have typical particle diameters of ≥ 80 μm. Johnsen (2004) reported that the mean particle diameter is around 38 μm in a typical aerosol spray. In practice, he stated that aerosols should have at least 99% of particle diameters in the 10- to 110-μm range. For comparison, Jensen and O'Brien (1993) reported a mean aerodynamic diameter of 4.25 ± 1.5 μm for respirable particles, which is smaller than any of the cosmetic product particle sizes given above.

Noncosmetic Use

Castor Oil, Hydrogenated Castor Oil, and Ricinoleic Acid Salts and Esters

According to Aplin and Eiseo (1997), castor oil is used as an industrial lubricant and as a medicinal purgative; the authors noted that medicinal Castor Oil does not contain ricin. McKeon et al. (2000) confirmed lubricant and anti-fungal uses and additional uses in paints, coatings, and plastics.

The following uses have been reported for Hydrogenated Castor Oil (Budavari 1989): in water-repellent coatings, candles,

TABLE 4
Current uses and concentrations of Ricinus Communis (Castor) Seed Oil and Ricinoleic Acid and its salts and esters in cosmetics

Product category (total no. of formulations)	Ingredient uses in each product category (FDA 2002)	Use concentrations (CTFA 2004) (%)
	Ricinus Communis (Castor) Seed Oil	
Baby products		
Shampoos (29)	—	—
Lotions, oils, powders, and creams (60)	1	—
Other (34)	2	—
Bath products		
Oils, tablets, and salts (143)	1	22
Soaps and detergents (421)	—	0.008
Bubble baths (215)	—	—
Capsules (2)	—	—
Other (196)	10	—
Eye makeup		
Eyebrow pencils (102)	10	3
Eyeliners (548)	11	4–8
Eye shadow (576)	9	2–5
Eye lotions (25)	—	—
Eye makeup remover (100)	1	—
Mascara (195)	16	0.4–3
Other (152)	11	13
Fragrance products		
Colognes and toilet waters (684)	—	0.1
Perfumes (235)	—	0.3–1
Powders (273)	—	—
Sachets (28)	—	—
Other (173)	—	—
Noncoloring hair care products		
Conditioners (651)	20	0.0002–5
Sprays/aerosol fixatives (275)	1	—
Straighteners (63)	—	—
Permanent waves (207)	—	—
Rinses (42)	5	19
Shampoos (884)	—	0.005
Tonics, dressings, etc. (598)	34	0.005–52
Wave sets (53)	—	16
Other (277)	6	—
Hair—coloring products		
Dyes and colors (1690)	27	2
Tints (49)	—	—
Rinses (20)	—	0.0005
Shampoos (32)	—	—
Color sprays (5)	—	—
Lighteners with color (5)	—	—
Bleaches (120)	1	0.04
Other (55)	—	—
Makeup		
Blushers (245)	10	0.08–45
Face powders (305)	1	1

(Continued on next page)

TABLE 4
Current uses and concentrations of Ricinus Communis (Castor) Seed Oil and Ricinoleic Acid and its salts and esters in cosmetics
(Continued)

Product category (total no. of formulations)	Ingredient uses in each product category (FDA 2002)	Use concentrations (CTFA 2004) (%)
Foundations (324)	5	0.05–6
Leg and body paints (4)	—	—
Lipsticks (962)	492	15–81
Makeup bases (141)	4	0.002–20
Rouges (28)	1	38–48
Makeup fixatives (20)	—	—
Other (201)	25	40–49
Nail care products		
Basecoats and undercoats (44)	—	—
Cuticle softeners (19)	1	—
Creams and lotions (15)	1	—
Nail extenders (1)	—	—
Nail polishes and enamels (123)	—	69
Nail polish and enamel removers (36)	8	—
Other (55)	3	—
Personal hygiene products		
Bath soaps and detergents (421)	18	$\leq$ 0.008
Underarm deodorants (247)	—	0.01–3
Douches (5)	—	—
Feminine deodorants (4)	—	—
Other (308)	1	0.003
Shaving products		
Aftershave lotions (231)	2	0.004–0.03
Beard softeners (0)	—	—
Mens talcum (7)	—	—
Preshave lotions (14)	—	—
Shaving creams (134)	—	—
Shaving soaps (2)	—	—
Other (63)	1	0.03
Skin care products		
Skin cleansing creams, lotions, liquids, and pads (775)	4	0.004
Depilatories (34)	2	—
Face and neck creams, lotions, powders, and sprays (310)	1	1–7
Body and hand creams, lotions, powders, and sprays (840)	3	0.02–55
Body and hand sprays	—	0.004
Foot powders and sprays (35)	—	—
Moisturizers (905)	8	7
Night creams, lotions, powders, and sprays (200)	—	0.00004
Paste masks/mud packs (271)	2	—
Skin fresheners (184)	—	—
Other (725)	8	—
Suntan products		
Suntan gels, creams, liquids, and sprays (131)	1	6
Indoor tanning preparations (71)	—	—
Other (38)	1	10
Total uses/ranges for Ricinus Communis (Castor) Seed Oil	769	0.00004–81

TABLE 4
Current uses and concentrations of Ricinus Communis (Castor) Seed Oil and Ricinoleic Acid and its salts and esters in cosmetics
(Continued)

Product category (total no. of formulations)	Ingredient uses in each product category (FDA 2002)	Use concentrations (CTFA 2004) (%)
	Hydrogenated Castor Oil	
Baby products		
Other (34)	—	5
Bath products		
Oils, tablets, and salts (143)	1	19
Soaps and detergents (421)	1	0.0003–0.7
Bubble baths (215)	—	4
Other (196)	—	3
Eye makeup		
Eyebrow pencils (102)	11	4–26
Eyeliners (548)	25	3–39
Eye shadow (576)	6	2–19
Eye lotions (25)	—	0.5
Eye makeup		
Eye makeup remover (100)	1	7
Mascara (195)	—	5
Other (152)	14	22–39
Fragrance products		
Colognes and toilet waters (684)	—	0.0003
Perfumes (235)	—	5
Other (173)	—	7
Noncoloring hair care products		
Shampoos (884)	—	0.02
Tonics, dressings, etc. (598)	—	3
Other (277)	1	—
Makeup		
Blushers (245)	2	3
Foundations (324)	4	0.9–5
Leg and body paints (4)	—	1
Lipsticks (962)	20	0.5–33
Makeup fixatives (20)	—	3–7
Other (201)	24	0.04–13
Nail care products		
Creams and lotions (15)	1	—
Oral hygiene products		
Other (6)	—	1–5
Personal hygiene products		
Underarm deodorants (247)	2	2–8
Feminine deodorants (4)	—	2
Other (308)	65	—
Shaving products		
Aftershave lotions (231)	2	—
Skin care products		
Skin cleansing creams, lotions, liquids, and pads (775)	3	1
Face and neck creams, lotions, powders, and sprays (310)	—	1–2
Body and hand creams, lotions, powders, and sprays (840)	3	0.0003–1
Foot powders and sprays (35)	—	8

(Continued on next page)

TABLE 4
Current uses and concentrations of Ricinus Communis (Castor) Seed Oil and Ricinoleic Acid and its salts and esters in cosmetics *(Continued)*

Product category (total no. of formulations)	Ingredient uses in each product category (FDA 2002)	Use concentrations (CTFA 2004) (%)
Moisturizers (905)	3	1–5
Night creams, lotions, powders, and sprays (200)	1	1–3
Other (725)	9	0.1–1
Suntan products		
Suntan gels, creams, liquids, and sprays (131)	1	0.8–3
Other (38)	2	—
Total uses/ranges for Hydrogenated Castor Oil	202	0.0003–39
Glyceryl Ricinoleate		
Eyebrow pencil (102)	—	11
Eyeliner (548)	1	2
Eyeshadow (576)	5	—
Other eye makeup products (152)	1	2–12
Lipsticks (962)	5	—
Other shaving products (63)	1	—
Body and hand creams, lotions, powders, and sprays (840)	2	—
Night creams, lotions, powders, and sprays (200)	1	—
Total uses/ranges for Glyceryl Ricinoleate	16	12
Ricinoleic Acid		
Bath preparations		
Oils, tablets and salts (143)	1	—
Fragrance preparations		
Colognes and toilet waters (684)	1	—
Other fragrance preparations (173)	4	—
Total uses/ranges for Ricinoleic Acid	6	—
Sodium Ricinoleate		
Baby products		
Other baby products (34)	1	—
Bath Preparations		
Soaps and detergents (421)	10	—
Noncoloring Hair Preparations		
Shampoos (884)	1	—
Total uses/ranges for Sodium Ricinoleate	12	—
Zinc Ricinoleate		
Noncoloring hair preparations		
Hair sprays/aerosol fixatives (275)	—	1
Personal hygiene products		
Underarm deodorants (247)	2	2
Skin care preparations		
Skin cleansing creams, lotions, liquids, and pads (775)	1	—
Body and hand skin care preparations (sprays) (840)	—	1
Total uses/ranges for Zinc Ricinoleate	3	1–2
Cetyl Ricinoleate		
Makeup preparations		
Blushers (245)	—	3

TABLE 4
Current uses and concentrations of Ricinus Communis (Castor) Seed Oil and Ricinoleic Acid and its salts and esters in cosmetics *(Continued)*

Product category (total no. of formulations)	Ingredient uses in each product category (FDA 2002)	Use concentrations (CTFA 2004) (%)
Foundations (324)	3	2–3
Lipsticks (962)	26	0.5–10
Other makeup preparations (201)	1	—
Skin care preparations		
Skin cleansing creams, lotions, liquids, and pads (775)	2	0.1–0.5
Body and hand skin care preparations (840)	1	—
Moisturizers (905)	15	0.1–2
Paste masks (mud packs) (271)	3	—
Other skin care preparations (725)	3	4
Suntan preparations		
Other suntan preparations (38)	1	—
Total uses/ranges for Cetyl Ricinoleate	55	0.1–10
Octyldodecyl Ricinoleate		
Fragrance preparations		
Other fragrance preparations (173)	—	3
Makeup preparations		
Lipsticks (962)	—	3–5
Total uses/ranges for Octyldodecyl Ricinoleate	—	3–5

shoe polish, carbon paper, and ointments; for impregnating paper, wood, and cloth; for electrical condenser impregnation; as a solid lubricant; and as a pressure mold release agent in the manufacture of formed plastics and rubber goods. The use of Hydrogenated Castor Oil in waterproofing fabrics has also been reported (Lewis 1997).

According to FDA's *OTC (Over-the-Counter) Drug Review Ingredient Status Report* (FDA 2003a), castor oil is classified as generally recognized as safe and effective for use as a stimulant laxative, but not generally recognized as safe and effective for use as a wart remover. For use as a stimulant laxative, the single daily dose reported for one product ranges from 15 to 60 ml for adults and children $\geq$12 years (Drugstore.com, Inc. 2004).

FDA had issued a proposed rule in 1982 stating that Castor oil is generally recognized as safe and effective and not misbranded when used as an active ingredient (laxative) in OTC laxative drug products (FDA 1985) and a final rule is pending (FDA 2003a). FDA has concluded that castor oil does not meet monograph conditions and is not generally recognized as safe and effective for use as a wart remover in OTC wart remover drug products (FDA 1990).

Castor oil is included in the list of inactive ingredients (excipients) present in approved oral, intramuscular, and topical drug products or conditionally approved drug products that are currently marketed for human use (FDA 2003b).

FDA-approved direct/indirect food additive uses of Castor oil and the following Ricinoleic Acid salts and esters are summarized in Table 5: Potassium Ricinoleate, Sodium Ricinoleate, Zinc Ricinoleate, Glycol Ricinoleate, Isopropyl Ricinoleate, and Methyl Ricinoleate.

The Joint Food and Agriculture Organization (FAO)/World Health Organization (WHO) Expert Committee on Food Additives (1980) established an acceptable daily intake (for man) of 0 to 0.7 mg/kg body weight for castor oil. The determination of range was based on consideration of the lack of adequate long-term studies.

Ricinoleic Acid

Noncosmetic uses of Ricinoleic Acid are as follows: turkey red oil, textile finishing, source of sebacic acid and heptanol, ricinoleate salts, and 12-hydroxystearic acid (Lewis 1997).

Zinc Ricinoleate

The following noncosmetic uses of Zinc Ricinoleate have been reported: fungicide, emulsifier, greases, lubricants, waterproofing, lubricating-oil additive, and stabilizer in vinyl compounds (Lewis 1997).

Methyl Ricinoleate

Noncosmetic uses of Methyl Ricinoleate include: plasticizer, lubricant, cutting oil additive, and wetting agent (Lewis 1997).

TABLE 5
Direct/indirect food additive uses of Castor Oil, Hydrogenated Castor Oil, and Ricinoleic acid salts and esters

Use	Specification/listing	Code of Federal Regulations (CFR)
Diluent	Not more than 500 ppm Castor Oil in the finished food may be used safely as a diluent in color additive mixtures for food use that are exempt from certification.	21 CFR 73.1
Release/antisticking agent	Provided that Castor Oil meets the specifications of the United States Pharmacopeia, it is permitted for use as a release agent and antisticking agent, not to exceed 500 ppm in hard candy.	21 CFR 172.876
Natural flavoring substance and adjuvant	Castor Oil is included in the list of natural flavoring substances and natural adjuvants that are permitted for direct addition to food for human consumption.	21 CFR 172.510
Plasticizer	Castor Oil is approved for use as a plasticizer (total not to exceed 30% by weight of rubber product, unless otherwise specified) in rubber articles that have been declared safe for use in producing, manufacturing, packing, processing, preparing, treating, packaging, transporting, or holding food.	21 CFR 177.2600
Component of food-contact surface	Hydrogenated Castor Oil may be used safely as a component of the uncoated or coated food-contact surface of paper and paperboard intended for use in producing, manufacturing, packaging, processing, preparing, treating, packing, transporting, or holding aqueous and fatty foods.	21CFR176.170
Component of food-contact surface	Hydrogenated Castor Oil may be used safely as a component of the uncoated or coated food-contact surface of paper and paperboard intended for use in producing, manufacturing, packaging, processing, preparing, treating, packing, transporting, or holding dry food.	21CFR176.180
Component of defoaming agents	Castor Oil, Hydrogenated Castor Oil, Glycol Ricinoleate, Isopropyl Ricinoleate, and Methyl Ricinoleate are among the substances that are permitted for use in the formulation of defoaming agents that have been declared safe for use in the manufacture of paper and paperboard intended for use in packaging, transporting, or holding food.	21 CFR 176.210
Component of surface lubricants	Castor Oil is included in the list of substances permitted for use in surface lubricants that have been declared safe for use in the manufacture of metallic articles that contact food. May be used in surface lubricants employed to facilitate the drawing, stamping, or forming of metallic articles from rolled foil or sheet stock by further processing, provided that the total residual lubricant remaining in the metallic article in the form in which it contacts food does not exceed 0.2 mg per square inch of food-contact surface.	21 CFR 178.3910
Component of cellophane	Hydrogenated Castor Oil is included in the list of components of cellophane that may be safely used for packaging food.	21CFR 177.1200

TABLE 5
Direct/indirect food additive uses of Castor Oil, Hydrogenated Castor Oil, and Ricinoleic acid salts and esters *(Continued)*

Use	Specification/listing	Code of Federal Regulations (CFR)
Component of closure-sealing gaskets	Hydrogenated Castor Oil may be employed in the manufacture of closure-sealing gaskets that may be safely used on containers intended for use in producing, manufacturing, packing, processing, preparing, treating, packaging, transporting, or holding food.	21CFR177.1210
Component of cross-linked polyester resins	Hydrogenated Castor Oil may be used in the production of or may be added to cross-linked polyester resins that may be safely used as articles or components of articles that are intended for repeated use in contact with food.	21CFR 177.2420
Textiles and textile fiber components derived from Castor Oil	Fats, oils, fatty acids, and fatty alcohols derived from Castor Oil are included in the list of substances that are permitted for use in textiles and textile fibers that have been declared safe for use as articles or components of articles that are intended for use in producing, manufacturing, packing, processing, preparing, treating, packaging, transporting, or holding food.	21 CFR 177.2800
Component of adhesives	Polymeric esters of polyhydric alcohols and polycarboxylic acids prepared from glycerin and phthalic anhydride and modified with Castor Oil are permitted for use in adhesives that have been declared safe for use as components of articles intended for use in packaging, transporting, or holding food. Methyl Ricinoleate is also permitted for use in these adhesives.	21 CFR 175.105
Component of resinous and polymeric coatings	Castor Oil, Hydrogenated Castor Oil, Potassium Ricinoleate, Sodium Ricinoleate, and Zinc Ricinoleate are included in the list of substances employed in the production of resinous and polymeric coatings that have been declared safe for use as the food-contact surface of articles intended for use in producing, manufacturing, packing, processing, preparing, treating, packaging, transporting, or holding food.	21 CFR 175.300

BIOLOGICAL PROPERTIES

Absorption, Distribution, Metabolism, and Excretion

Castor Oil

In a study by Paul and McKay (1942), two rabbits (weight = 3 kg) were fed 6% castor oil in the diet for 18 days; fecal collection occurred during the last ten days. The utilization (uncorrected for metabolic fat) of castor oil was 92.1%, which the authors considered to be efficient utilization. For both rabbits, the percentage of fat in the feces was 2.2%.

In a study by Stewart and Sinclair (1945), adult rats (number, weights, and strain not stated) received a diet containing 48.4% castor oil for 4 to 6 weeks. Control rats received stock ration only. Feces were collected from three rats on the castor oil diet. At the end of the feeding period, excised organs/tissues were ground thoroughly and samples of phospholipid fatty acids were obtained from the liver, small intestine, and muscle; glyceride fatty acids were obtained from the liver and fat depots. There was no evidence of catharsis in any of the animals.

Average percentages of Ricinoleic Acid in the phospholipid fatty acids were as follows: liver (test: 1.3 ± 0.6% [9 analyses]; controls: 1.7 ± 1.1% [7 analyses]), small intestine (test: 4.9 ± 1.7% [8 analyses]; controls: 6.0 ± 4.4% [4 analyses]), and skeletal muscle (test: 3.6 ± 2.9% [8 analyses]; controls: 4.0 ± 1.7% [7 analyses]). The following values are average percentages of Ricinoleic Acid in glycerides and cholesterol esters: fat depots (test: 6.8 ± 4.2% [11 analyses]; controls: 0.5 ± 0.5% [7 analyses]) and liver (test: 7.2 ± 2.4% [8 analyses]; controls: 5.6 ± 4.1% [5 analyses]).

The authors concluded that the feeding of castor oil did not lead to the appearance of significant amounts of Ricinoleic Acid in phospholipids of the small intestine, liver, and skeletal muscle,

nor in glycerides of the liver. Additionally, they concluded that ricinoleic acid is a component acid of the glycerides in the fat depots, comprising 7% of the total fatty acids. The fatty acids excreted by each of three rats amounted to 2.1%, 2.2%, and 3.6% of those ingested. Total body fat in these three animals was also determined, and it was calculated that 1% to 2% of absorbed Ricinoleic Acid was deposited in the fat depots. The authors concluded that Ricinoleic Acid was rapidly metabolized (Stewart and Sinclair 1945).

Watson and Gordon (1962) studied the digestion, absorption, and metabolism of castor oil (medicinal grade) in eight male Sprague-Dawley rats (weights = 100 to 200 g). The composition of the castor oil was as follows: ricinoleic acid (90.0%), linoleic acid (4.7%), oleic acid (3.2%), stearic acid (1.0%), palmitic acid (1.0%), and palmitoleic acid (0.1%). In the first experiment, four rats were fed rat chow ad libitum, and the remaining four were fasted overnight. On the following morning, castor oil (1.0 ml) was dosed via stomach tube and chyle was collected over a 24-h period. The mean values for percent recovery of Ricinoleic Acid in fasted and fed rats were 6.8% and 24.2%, respectively ($p < .01$).

In the second experiment, seven weanling rats were fed a diet consisting of rat chow that had been mixed with castor oil (20% by weight). The control group was fed an olive oil-supplemented diet. After 4 and 8 weeks of feeding, an epididymal fat pad was removed from each rat in both groups, and fatty acid composition was determined using gas-liquid chromatography. Mean values for Ricinoleic Acid in the fat pad after 4 and 8 weeks of feeding were 9.1 ± 1.7 and $9.7 \pm 1.0\%$, respectively. Ricinoleic Acid was undetectable in the fat pads of control rats fed olive oil. For rats fed castor oil, random analyses of feces indicated a considerable fraction of hydroxystearic acid. However, hydroxystearic acid was undetectable in the feces of rats fed a normal diet. The authors suggested that hydrogenation of Ricinoleic Acid in the gut lumen by intestinal bacteria would be a likely explanation for this finding.

The relationship between dose and absorption of castor oil was studied in the final experiment. Polyethylene cannulae were inserted into the thoracic duct and duodenum of each of two rats, and the animals received 0.5 N saline overnight. On the following morning, one rat received 0.2 ml and one rat received 0.6 ml of castor oil, and the rats were killed 45 min post dosing. Only the high dose induced diarrhea. Intestines (small and large) were excised, homogenized, and tissue lipids were extracted. Chyle was also collected and extracted. Total lipid estimation and gas-liquid chromatography (GLC) analysis of fatty acids were performed on the extracts. For the rat dosed with 0.2 ml castor oil, the percentage of Ricinoleic Acid in the chyle was 18.1% and the percentage detected in the small bowel lipids was 6.2%. For the rat dosed with 0.6 ml Castor Oil, the percentage of Ricinoleic Acid in the chyle was 3.0% and the percentage in the small bowel was 3.1%. Ricinoleic Acid was undetectable in large bowel lipids from the rat with diarrhea. The result indicates a close correlation between the dose administered and the percentage of Ricinoleic Acid in the feces, i.e., greater absorption at the lower dose (Watson and Gordon 1962).

Castor oil is metabolized to Ricinoleic Acid by pancreatic lipase in hamsters (Gaginella and Bass 1978). Thompson (1980) reported that Castor Oil is a triglyceride that is hydrolyzed in the small intestine in humans by pancreatic enzymes, leading to the release of glycerol and Ricinoleic Acid.

In a study by Hagenfeldt et al. (1986), castor oil was administered intragastrically to germ-free and conventional rats (number not stated). Urine was collected at intervals over a 24-h period. The following epoxydicarboxylic acids were detected in the urine of both germ-free and conventional rats: 3,6-epoxyoctanedioic acid; 3,6-epoxydecanedioic acid; and 3,6-epoxydodecanedioic acid. These acids were not detected in urine collected from the rats prior to dosing with castor oil, and they also were not detected in steam-sterilized castor oil. The authors stated that results for the germ-free rat indicate that the cyclization of Ricinoleic Acid (hydroxy fatty acid in castor oil) to form an epoxy compound occurs endogenously and does not require the presence of intestinal bacteria.

In a study by Ihara-Watanabe et al. (1999), two groups of five male Wistar rats (3 weeks old) received 10% castor oil in the diet (cholesterol-enriched and cholesterol-free, respectively) for 20 days. In both dietary groups, a very small quantity of Ricinoleic Acid was present in perirenal adipose tissue, but not in the serum or hepatic tissue. It was also noted that the perirenal fatty acid profiles did not reflect those of the dietary fats, either in the absence or presence of dietary cholesterol. The fecal recovery of Ricinoleic Acid was approximately 0.5% of the total ingested. It was concluded that castor oil was readily absorbed and metabolized.

Hydrogenated Castor Oil

In a study by Binder et al. (1970), groups of 15 male albino rats (Slonaker substrain of Wistar strain, weights = 43 to 83 g) received diets containing the following oils: group 1 (fed 1% Hydrogenated Castor Oil (HCO) + 19% corn oil for 16 weeks), group 2 (1% HCO + 19% corn oil for 8 weeks, then 20% corn oil for 8 weeks), group 3 (10% HCO + 10% corn oil for 16 weeks), and group 4 (10% HCO + 10% corn oil for 8 weeks, then 20% corn oil for 8 weeks). The control group received 20% corn oil in the diet for 16 weeks. The sample of HCO that was incorporated into the diet contained 86.5% 12-hydroxystearic acid, 10.3% nonoxygenated acids, and 3.2% 12-ketostearic acid. Thus, the effective dietary concentrations of Hydrogenated Castor Oil in the 1% and 10% diets were 0.865% and 8.65%, respectively. Results relating to the toxicity of HCO are summarized in the section Subchronic Oral Toxicity later in the report.

After 4 weeks of feeding, some of the animals on the 1% HCO (three rats) and 10% HCO (three rats) diets were necropsied and excised abdominal adipose tissue from each group was pooled. At weeks 4 and 8, sets of three rats on the corn oil diet were used. At the end of the 16-week feeding period, adipose tissue from rats in the various dietary groups was analyzed. Adipose tissue

was not pooled. After 8, 12, and 16 weeks, lipids were extracted from three rats on each diet. Methyl esters were recovered from the extracted lipids and chromatographed.

The deposition of hydroxystearic acid in abdominal fat and other body lipids was reported. In all cases, it was accompanied by hydroxypalmitic acid, hydroxymyristic acid, and hydroxylauric acid (hydroxy-stearic acid metabolites). The percent composition of these HCO-derived hydroxy fatty acids in rat lipids was as follows: 12-hydroxystearic acid (≈81%), 10-hydroxypalmitic acid (≈17%), 8-hydroxymyristic acid (≈1.6%), and 6-hydroxylauric acid (≈0.4%). After 4 weeks on the 1% HCO diet, HCO-derived fatty acids accounted for 0.90% (by weight) of the abdominal fat fatty acids. This proportion decreased to ≈0.35% over the 8- to 16-week feeding period. Compared to the abdominal fatty acids, the acids obtained from the carcass lipids contained a smaller proportion of HCO-derived hydroxyacids (≈0.28% over the 8- to 16-week feeding period). The period encompassing 4 weeks of feeding on the 10% HCO diet yielded the greatest content of HCO-derived hydroxy acids in the lipids (i.e., 4.4% hydroxy acids in abdominal fat). At 16 weeks, this proportion had decreased to <2% (about same as in the carcass lipids).

A rapid decrease in the amount of HCO-derived hydroxy fatty acids in the tissues was noted when the HCO diet was changed to the control diet (Binder et al. 1970).

Ricinoleic Acid and Methyl Ricinoleate

Uchiyama et al. (1963) studied the accumulation of hydroxy acids in depot fat after rats were fed with Ricinoleic Acid in two experiments. In the first experiment, adult male rats (number, strain, and weights not stated) were fed Ricinoleic Acid (5% emulsion, 20 ml) for 7 days. In the second experiment, the animals were fed for 27 days. Lipid extraction from the fat tissue was followed by hydrolysis to yield a fatty acid mixture. A gas-liquid chromatogram indicated appreciable amounts of the following hydroxy fatty acids with shorter chain lengths than Ricinoleic Acid: 10-hydroxyhexadecenoic acid (experiment 1: 0.60% of total fatty acids; experiment 2: 0.33% of total fatty acids), 8-hydroxytetradecenoic acid (experiment 1: 0.03% of total fatty acids; experiment 2: 0.08% of total fatty acids), and 6-hydroxydodecenoic acid (experiment 2: 0.03% of total fatty acids). Ricinoleic Acid comprised 0.51% of total fatty acids in experiment 1 and 3.85% of total fatty acids in experiment 2.

In a study by Okui et al. (1964), the metabolism of hydroxy fatty acids was evaluated using male albino rats (weight = 120 ± 10 g). The rats received 1.5 g Ricinoleic Acid or an emulsion containing 5% (*w*/*v*) Ricinoleic Acid by stomach tube three times per day for a desired period. Feces were collected every 24 h until the animals were killed. The animals were killed 20 h after the last dose, and subcutaneous adipose tissue was removed for analyses. Oral dosing with Ricinoleic Acid resulted in a fecal excretion rate (for Ricinoleic Acid) ranging from 1% to 4%. Following oral administration for up to 30 days, the accumulation of hydroxy acids by 5% of fatty acids in fat tissue was noted. Analyses of adipose tissue indicated an occurrence of shorter chain hydroxy acids other than Ricinoleic Acid.

Ricinoleic Acid was also injected intraperitoneally to determine whether another route of administration would lead to the formation of shorter chain hydroxy acids. After Ricinoleic Acid was injected intraperitoneally daily for 7 days, Ricinoleic Acid accumulated in the adipose tissue; no shorter chain hydroxy acids were reported (Okui et al. 1964).

In a study by Rao et al. (1969), Ricinoleic Acid or Methyl Ricinoleate was administered via gastric intubation to male rats (minimum weight of 400 g) with a cannulated thoracic duct. Lymph was collected for 48 h, and the lipids were then extracted and separated into various lipid classes. Results indicated that Ricinoleic Acid was present in the triglyceride, diglyceride, monoglyceride, and free fatty acid fractions. Peak absorption of Ricinoleic Acid occurrred within 30 min post administration. Ricinoleic Acid was not present in the phospholipid or cholesterol ester fractions of the lymph lipids.

Percutaneous Absorption

Ricinoleic Acid

Butcher (1952) evaluated the skin penetration of Ricinoleic Acid in vivo using rats that were 20 to 30 days old. In order to increase the fluorescence of Ricinoleic Acid, either one part of methyl anthranilate or methylcholanthrene was added to 99 parts Ricinoleic Acid. The test substance was either gently rubbed on to skin that had been clipped free of hair. Biopsies were taken at various intervals post application. The preparations were observed using a Spencer microscope with a quartz condenser. Ricinoleic Acid was retained mainly in the outer strata of the epidermis. There was little evidence of deeper penetration in biopsies that were taken at 2 h post application.

In a study by Baynes and Riviere (2004), the percutaneous absorption of a radiolabeled [^{3}H]Ricinoleic Acid (specific activity = 20.0 mCi/mmol) mixture was evaluated using porcine skin membranes or silastic (polydimethylsioloxane) membranes in the Bronaugh flow-through diffusion cell system. [^{3}H]Ricinoleic Acid (5%) mixtures were prepared in water containing either 5% mineral oil or 5% PEG 200. Other [^{3}H]Ricinoleic Acid mixtures were formulated with the following three commonly used cutting fluid additives: triazine, linear alkylbenzene sulfonate, and triethanolamine. At 8 h after topical exposure, Ricinoleic Acid absorption (based on amount recovered in receptor fluid) ranged from 1% to 13% in silastic membranes and 0.1% to 0.3% in porcine skin membranes. For most mixtures, peak absorption of Ricinoleic Acid occurred within 3 h. The greatest Ricinoleic Acid peak concentrations were associated with the control mixtures containing PEG in both membranes.

At the conclusion of the 8-h perfusion experiments, as much as 5% of the dose of Ricinoleic Acid was detected in the dosed skin, and as much as 16% of the dose was detected in the stratum

corneum with mineral oil formulations. With polyethylene glycol (PEG) formulations, the amount of Ricinoleic Acid remaining in the skin tissues was considerably less. The cutting fluid additives significantly decreased Ricinoleic Acid partitioning from the formulation into the stratum corneum in PEG-based mixtures. Additionally, the individual additives or combinations of these additives significantly reduced Ricinoleic Acid permeability in silastic membranes and porcine skin (Baynes and Riviere 2004).

Skin Penetration Enhancement

Ricinoleic Acid

In a study by Song et al. (2001), the effects of *cis*-9-octadecenoic acid (oleic acid) and a group of chemically related *cis*- (Ricinoleic Acid) and *trans*- (ricinelaidic acid) 12-monohydroxylated derivatives and their ethyl and methyl esters on the skin penetration of model hydrophobic (hydrocortisone) and hydrophilic (5-fluorouracil) drugs were evaluated in vitro using hairless mouse skin. Test solutions were prepared by placing either test compound (5-fluorouracil, 5.4 mg; hydrocortisone, 15 mg) with or without an unsaturated fatty acid (50 mg each) or fatty acid ester (50 mg each) in a screw-capped glass vial, and adding propylene glycol (final volume = 1 ml). The mixture was sonicated to effect complete dissolution. Permeation studies were conducted using Vlia-Chien diffusion cells and full thickness abdominal skin from female hairless HRS/J mice (6 weeks old). Ricinoleic Acid enhanced the in vitro transdermal permeation of 5-fluorouracil.

The transdermal penetration rate of hydroquinone from propylene glycol was very slow. However, in the presence of oleic acid, the penetration rate accelerated and was enhanced approximately 1800-fold. In the presence of Ricinoleic Acid, the enhancement of hydroquinone skin penetration was less than 1.5-fold. Regarding the penetration of 5-flurouracil, fluxes were much higher in the presence of oleic acid (333-fold increase; $p < .001$) than in the presence of Ricinoleic Acid (<5-fold increase) (Song et al. 2001).

Transepidermal Water Loss

Castor Oil

Lieb et al. (1988) evaluated transepidermal water loss (TEWL) using a modification of a flow-through diffusion cell that was originally designed by Bronaugh and Stewart (1985). The membrane was described as full-thickness, cartilage-stripped skin obtained from the ventral ear of the male Syrian golden hamster. Castor oil (10 μl) was placed on the skin (0.32-cm^2 area), and a TEWL rate–versus-time study was conducted. Tritiated water (HTO) permeated through the membrane into the diffusion cell's receptor compartment. Flux (TEWL, μg/cm^2/h) was determined (using a scintillation counter) by measuring the HTO that adsorbed to anhydrous calcium chloride in the receptor cell. Castor oil caused a marked decrease in the TEWL rate. For untreated skin, the mean TEWL rate was approximately 450 μg/cm^2/h at 1 h. Mean values for the TEWL rate were approximately 400 μg/cm^2/h at 1 h and 250 μg/cm^2/h at 24 h. Additionally, castor oil was the only substance that exhibited a burst effect, i.e., initially, the membrane became fully hydrated and swollen and was more permeable, providing an "increased push" effect to pass water through the membrane toward the anhydrous desiccant.

Effect on Smooth Muscle

Castor Oil and Ricinoleic Acid

Mathias et al. (1978) evaluated the effect of Castor Oil and Ricinoleic Acid on smooth muscle of the rabbit small intestine. Anesthetized male New Zealand white rabbits (weights between 1.5 and 3.0 kg) were used in ileal loop studies. Castor oil (dose = 0.85 ml/kg) was administered into the orad end of the ileal loop, and myoelectric recording techniques were used. Ricinoleic Acid was administered by intraluminal infusion (2 μg/kg/min [6 mM] into the orad end). The term migrating action potential complex (MAPC) was used to describe the myoelectric activity that has been observed in certain abnormal states. MAPC has been defined as action potential discharge activity occurring for longer than 2.5 s, on at least two consecutive electrode sites at 2.5-cm intervals, and having a propagation velocity of $\cong$1.0 cm/s. The myoelectric activity of castor oil and Ricinoleic Acid was similar to the MAPC that was observed in the cholera enterotoxin-infected loops. Castor oil induced an alteration in myoelectric activity.

In experiments that were designed to study the motor response in segments of the duodenum, mid-jejunum, terminal ileum, and sigmoid colon (four experiments, respectively), Ricinoleic Acid was diluted with 0.9% sodium chloride solution to a volume that would deliver 1.14 ml/h. No MAPC activity was observed in control experiments in which 0.9% sodium chloride solution was perfused into the duodenum at a rate of 1.14 ml/h. When compared to control values, there was no statistically significant difference in the slow wave frequency or slow wave propagation velocity in loops exposed to castor oil or Ricinoleic Acid (Mathias et al. 1978).

Lodge (1994) evaluated the potential for Ricinoleic Acid to induce vasoconstriction using tissues from male New Zealand white rabbits. After removal of the thoracic aorta and left and right external jugular veins, the vessels were cleaned and cut into rings. Following removal of the endothelium, the rings were suspended between two wire holders in tissue baths. Ricinoleic Acid was diluted with ethanol to produce final bath concentrations ranging from 0.1 to 30 μg/ml (molar concentration range of 0.335 to 100.50 μM). Ricinoleic Acid induced vasoconstriction ($EC_{50} = 0.24 \pm 0.04$ μg/ml) that was concentration-dependent and sensitive to the inhibitory effect of ifetroban, but not indomethacin. Ricinoleic acid also evoked concentration-dependent force development (constriction) in the aorta ($EC_{50} = 4.7 \pm 0.7$ μg/ml), and this response was antagonized by ifetroban.

Sodium Ricinoleate, Castor Oil, and Ricinoleic Acid

Stewart and Bass (1976a) evaluated the effects Sodium Ricinoleate, castor oil, and Ricinoleic Acid on the digestive contractility of the canine small bowel using four male, mixed breed dogs (weights between 10 and 15 g). After feeding, each animal received a single bolus infusion of Sodium Ricinoleate (500 mg in 30 ml of saline) into the duodenum. Saline (30-ml volume) served as the control. A duodenal cannula was implanted in each animal, and circular muscle activity was recorded for 1 h. Each animal was dosed with Sodium Ricinoleate in two experiments, and with isotonic saline (30 ml) in two control experiments. In additional experiments, alterations in digestive contractile patterns were monitored after administration (gastric tube) of castor oil or Ricinoleic Acid (volume = 10 ml) one h after feeding. Each treatment was repeated twice in each of two animals.

Intraduodenal administration of Sodium Ricinoleate produced a biphasic response in the proximal and mid-jejunum consisting of brief initial stimulation, followed by inhibition of digestive contractility. Compared to the control experiments, these stimulatory and inhibitory effects on intestinal contractility were described as significant ($p < .05$) changes in the average force per contraction. The changes in mid-jejunal digestive contractility were similar to those observed in the proximal jejunum.

The oral administration of Ricinoleic Acid (10 ml) induced alterations in digestive patterns that were qualitatively similar to those induced by the intraduodenal administration of Sodium Ricinoleate. Brief initial stimulation, followed by an apparent depression of intestinal contractility, was observed after oral dosing with Castor Oil (10 ml) (Stewart and Bass 1976a).

Sodium Ricinoleate and Methyl Ricinoleate

In a study by Stewart et al. (1975), Sodium Ricinoleate depressed the smooth muscle twitch response of the electrically driven guinea-pig ileum preparation at concentrations ranging from 1.25×10^{-5} to 4×10^{-4} M. Methyl Ricinoleate failed to depress the smooth muscle twitch response over this range of test concentrations. Similar results were reported for spontaneously contracting rabbit jejunum.

Antinociceptive Activity

Ricinoleic Acid and Potassium Ricinoleate

Vieira et al. (2000) evaluated the antinociceptive activity of Ricinoleic Acid (in peanut oil), dissolved in a vehicle containing 10% ethanol, 10% Tween 80, and 80% saline, using groups of 5 male albino Dunkin-Hartley guinea pigs (weights = 250 to 350 g) and groups of 8 to 10 male Swiss mice (weights = 20 to 25 g). A series of experiments was performed. In the first experiment, a single topical application of Ricinoleic Acid (900 μg/mouse) was made to the ventral surface of the right paw of each mouse. Test substance application was followed by intraplantar injection with carrageenan (to induce edema) 30 min later. Ricinoleic acid significantly reduced the reaction time to a heat stimulus, compared to treatment with the vehicle. When intraplantar injection with carageenan was preceded by repeated (8 days) topical applications of Ricinoleic Acid (900 μg/mouse), again, a marked prolongation of paw withdrawal latency to heat, compared to the vehicle control group, was noted.

Another experiment involved the repeated local application (8 days) of Ricinoleic Acid (900 μg/mouse) or the repeated (4 days) intradermal injection of Potassium Ricinoleate (30 μg/mouse) on the ventral surface of the right paw. Freund's adjuvant had been injected into the hindpaw on the first day. The paw withdrawal latency to a painful stimulus (heat) after topical application was increased over at least 2 weeks. The intradermal injection of Potassium Ricinoleate induced a significant antinociceptive effect that lasted for at least 3 weeks (Vieira et al. 2000).

Effect on Enzyme Activity

Ricinoleic Acid

In a study by Gaginella et al. (1978), isolated villus cells from the small intestine of the hamster were used to determine whether Ricinoleic Acid stimulates intestinal adenylate cyclase. In the adenylate cyclase assay, activity of the enzyme was determined using a method that is based upon the conversion of [$\alpha-^{32}$P]ATP to [^{32}P]cyclic AMP. The cell homogenates (30 to 50 μg of protein per tube) were incubated for 20 min. Ricinoleic Acid concentrations of 10^{-9} to 5×10^{-3} M did not cause dose-dependent stimulation of adenylate cyclase. Significant ($p < .05$), but slight, stimulation of adenylate cyclase was noted at a concentration of 10^{-5} M, and higher concentrations inhibited basal activity. Prostaglandin E_1, which is chemically similar to Ricinoleic Acid, was a potent stimulant of adenylate cyclase activity, causing dose-related stimulation ($p < .001$) of adenylate cyclase at concentrations of 10^{-9} to 10^{-4} M.

Simon and Kather (1980) studied the modulation of adenylate cyclase and cAMP-phosphodiesterase, key enzymes of cAMP metabolism, by Ricinoleic Acid in human colonic mucosa. The experiments were performed using histologically normal colonic mucosa obtained from eight patients (five males, three females) who were undergoing hemicolectomy for carcinoma or diverticulosis. Adenylate cyclase activity was determined according to the method of Salomon et al. (1974), and cyclic AMP phosphodiesterase activity was determined according to the method of Pöch (1971). Ricinoleic Acid was tested at concentrations ranging from 1×10^{-8} to 5×10^{-4} mol/l. Basal adenylate cyclase activity in homogenates of human colonic mucosa averaged 250 pmol cAMP/mg protein/15 min. Mean basal cAMP phosphodiesterase activity was 150 ± 15 pmoles per mg protein per minute.

Over the range of concentrations tested, dose-related stimulation of the large bowel adenylate cyclase was not observed. Basal activity of adenylate cyclase was inhibited at concentrations above 1×10^{-5} mol/l. Ricinoleic Acid did not influence soluble cAMP phosphodiesterase activity over the range of concentrations tested. Ricinoleic Acid was an ineffective stimulus of human colonic adenylate cyclase, and also was not a competitive

inhibitor of soluble cAMP phosphodiesterase activity (Simon and Kather 1980).

In a study by Vanderhoek et al. (1980), the effect of Ricinoleic Acid on the oxygenation of [1-^{14}C]arachidonic acid by platelet cyclooxygenase and lipoxygenase enzymes was monitored using an oxygen electrode and by analysis of the radioactive products formed. The enzymes were obtained from human platelet concentrates (3 to 4 days old). Ricinoleic Acid did not appear to have an effect on either enzyme. The concentration of Ricinoleic Acid that was required for half-maximal inhibition of lipoxygenase or cyclooxygenase activity was >300 μM.

Beubler and Schrirgi-Degen (1992) studied the stimulation of enterocyte protein kinase C by laxatives in vitro using intestinal epithelial cell (from female Sprague-Dawley rats) preparations. A protein kinase C assay system was used to determine the activity of protein kinase C. This assay system is based on protein kinase C–catalyzed transfer of the γ-phosphate group of adenosine-5′-triphosphate to a peptide that is specific for protein kinase C. Ricinoleic Acid (dissolved in DMSO) stimulated protein kinase C activity in a concentration-dependent manner within the concentration range of 2 to 200 μg/ml.

Effect on Uridine Uptake

At a concentration of 50 μM, Ricinoleic Acid suppressed the uptake of uridine into human diploid fibroblasts in culture (Polgar and Taylor 1977).

Effect on Lipid Metabolism

Castor Oil

Ihara-Watanabe et al. (1999) evaluated the effect of the following diets on lipid metabolism using groups of five male Wistar rats (3 weeks old): 10% castor oil, 10% high-oleic safflower oil, and 10% coconut oil. The diets were prepared in the absence or presence of dietary cholesterol, and were fed ad libitum for 20 days. On day 20, blood samples were obtained and liver and perirenal adipose tissues were excised. Serum and hepatic lipids were analyzed for total cholesterol, high-density lipoprotein, triacylglycerols, and phospholipids.

Body weight gain and food intake were lowest for rats fed the castor oil diet enriched with cholesterol, but not for rats fed cholesterol-free diets. Body weight gain to consumption ratios were similar among all of the groups. Compared to rats fed the high-oleic safflower oil diet (enriched with cholesterol), rats fed the castor oil diet enriched with cholesterol had decreased liver weights (g/100 g of body weight). Data on liver weights were not presented in the study.

Values for total cholesterol and phospholipids in the serum were significantly decreased in rats fed castor oil, compared to rats fed high-oleic safflower oil in the absence or presence of cholesterol. For cholesterol-enriched diets, the lowest value for serum high-density lipoproteins was reported for the castor oil dietary group. No significant differences in serum triacylglycerol occurred between either of the dietary groups (with or without cholesterol in the diet).

Compared to the high-oleic safflower oil dietary group (with or without cholesterol), significantly decreased ($p < .05$) hepatic triacylglycerol concentrations were reported for the castor oil dietary group. For cholesterol-enriched diets, the castor oil and coconut oil dietary groups had significantly increased ($p < .05$) hepatic cholesterol when compared to the high-oleic safflower oil dietary group. It was concluded that castor oil had a hypocholesterolemic effect in rats in this study (Ihara-Watanabe et al. 1999).

Effect on Leukocyte Infiltration and Prostaglandin Synthesis

Castor Oil

Ohia et al. (1992) used castor oil as a vehicle control in a study evaluating the effects of steroids and immunosuppressive drugs on endotoxin-induced uveitis in rabbits. The vehicle control group consisted of four male New Zealand rabbits (weights = 1.5 to 2.0 kg). The animals were injected intramuscularly with castor oil (dose = 2 mg/kg). At 1 h post injection, both eyes of each rabbit were cleansed and anesthetized. This procedure was followed by the intravitreal injection of *Escherichia coli* endotoxin (100 ng). The animals were killed 24 h after injection of the endotoxin. Aqueous humor was obtained and leukocytes were counted. The eyes were then enucleated for microscopic examination. Compared to saline-treated controls, castor oil significantly reduced the infiltration of leukocytes and the prostaglandin E_2 (inflammatory mediator) content of the aqueous humor and iris-ciliary body. The authors concluded that Castor Oil had an anti-inflammatory effect in this study.

Effect on Cytokine-Induced Endothelial Cell Activation

Sodium Ricinoleate

De Caterina and Bernini (1998) evaluated the relationship between structural differences in fatty acids and the inhibition of endothelial activation, taking into consideration that dietary long-chain fatty acids may influence processes that involve endothelial activation (e.g., inflammation and atherosclerosis). To test for a potential stimulatory effect of fatty acids on adhesion molecule expression, sodium salts of saturated, monounsaturated (Sodium Ricinoleate included), and n-6 and n-3 polyunsaturated fatty acids were each incubated alone (test concentration = 25 μM) with human saphenous vein endothelial cells (HSVECs) for 72 h. The 72-h incubation period was followed by 16 h of stimulation with the cytokine interleukin (IL)-1α in the continuous presence of fatty acid salt. At the end of the incubation period, vascular cell adhesion molecule-1 (VCAM-1) expression was assessed using a cell surface enzyme immunoassay (EIA).

Sodium Ricinoleate significantly inhibited (considered modest; $p < .05$) cytokine-induced endothelial adhesion molecule

expression. DHA inhibited VCAM-1 expression ($p < .01$) most potently (De Caterina and Bernini 1998).

De Caterina and Bernini (1998) also conducted an experiment to determine whether the fatty acid sodium salts may induce endothelial activation in the absence of cytokines. The salts were incubated with endothelial cell monolayers for 24 h. Incubation periods prior to detection of adhesion molecule surface expression varied from 0 to 72 h. Neither Sodium Ricinoleate (25 μM) nor any other fatty acid sodium salt induced adhesion molecule expression, as assessed by EIA.

Ricinoleic Acid

In a study by De Caterina et al. (1995), the effect of Ricinoleic Acid on VCAM-1 expression was evaluated using rabbit coronary artery endothelium in vitro. VCAM-1 (marker for endothelial cell activation) is a surface protein that is not expressed by endothelial cells in vitro, but is inducible by exposure to inflammatory stimuli such as bacterial endotoxin, IL-1, or tumor necrosis factor α (TNFα). Ricinoleic Acid significantly reduced IL-4–induced VACM-1 expression (50% inhibitory concentration between 10 and 100 μM). Neither cell number nor viability was increased in this concentration range. Ricinoleic Acid did not inhibit TNFα-induced VACM-1 expression to any significant extent.

Effect on Prostaglandin Synthesis

Castor Oil

Gao et al. (1999) evaluated the effect of castor oil in the diet on the synthesis of prostaglandin E_2 (PGE_2) and the induction of labor using two groups of eight pregnant Wistar rats (at gestation day 18; test and control groups, respectively). Two milliliters of castor oil–containing diet were administered by gavage daily for a total of four feedings. The diet (labor-inducing diet) consisted of castor oil (30 ml) + one chicken egg, blended and heated to a thick consistency. At 4 h after the fourth feeding, the pregnant females were killed. Compared to the control group, a significant increase in concentrations of PGE_2 in tissues of the intestinal mucosa, placenta, amnion, and amniotic cells was noted in test animals. The authors stated that the increased synthesis of PGE_2 "is a key of" the initiation of labor that is induced by a castor oil diet.

Cytotoxicity

Sodium Ricinoleate

Gaginella et al. (1977b) evaluated the cytotoxicity of Ricinoleic Acid using epithelial cells that were isolated from the small intestines of male Syrian hamsters. Cytotoxicity was assessed by exclusion of trypan blue, the release of intracellular (prelabeled) ^{51}Cr, and inhibition of the cellular uptake of 3-O-methylglucose. The results produced by all three methods indicated that Sodium Ricinoleate produced a dose-dependent cytotoxicity.

For the trypan blue exclusion method, Sodium Ricinoleate caused dose-related increases in cytotoxicity at concentrations of 0.1 to 5.0 mM. Death of all cells was noted at a concentration of 2.0 mM. Results for control incubation in the ^{51}Cr release assay indicated that 26.8 $\pm$ 1.7% of ^{51}Cr, initially within or bound to the cells, was released into the medium. Sodium Ricinoleate induced concentration-dependent (0.1 to 5 mM Sodium Ricinoleate) increases in ^{51}Cr release, from 4.2% to 54.8% above the control value. In the 3-O-methylglucose uptake assay, Sodium Ricinoleate inhibited the uptake of 3-O-methylglucose over a concentration range of 0.1 to 2.0 mM (Gaginella et al. 1977b).

In a study by De Caterina and Bernini (1998), the cytotoxicity of Sodium Ricinoleate and other fatty acid sodium salts in human saphenous vein endothelial cell cultures was evaluated. Cell count, trypan blue exclusion, and [^{3}H]leucine incorporation into total cell-associated and released proteins were monitored. For the latter of the three, trichloroacetic acid precipitation of total cell extracts and cell supernates was performed. At concentrations $\leq$25 μM, Sodium Ricinoleate and other fatty acid sodium salts did not induce significant toxicity (based on cell count and trypan blue exclusion) for periods up to 72 h prior to cytokine stimulation.

Antimicrobial Activity

Castor Oil

Morris et al. (1979) evaluated the antimicrobial activity of castor oil on *Staphylococcus aureus*, *Escherichia coli*, and *Candida albicans* using the Petri plate–paper disc procedure. Paper discs were soaked with 20 μl of a 10% Castor Oil solution, and the discs were immediately applied to solidified, seeded agar (1 disc per plate). The plates were incubated for 10 to 24 h. Discs containing 95% ethanol (20 μl) served as negative controls. Castor Oil did not have antimicrobial activity on any of the strains tested.

Sodium Ricinoleate

Whiteside-Carlson et al. (1955) studied the effect of Sodium Ricinoleate on cell division using a laboratory strain of *E. coli*. Sodium Ricinoleate was tested over a concentration range of 1 to 5 mg/ml of medium. The cultivation of *E. coli* on nutrient agar that was saturated with Sodium Ricinoleate caused the development of filamentous forms during the first h of logarithmic growth. By the end of the log phase, organisms of normal morphology were observed. When agar containing Sodium Ricinoleate was adjusted to pH values of 5 to 8, filament formation was more marked and persisted for longer periods at the more alkaline pH value. The utilization of liquid proteose-peptone medium (with periodic readjustment of the pH to a range of 7.6 to 7.8) resulted in maintenance of the cells in filamentous form throughout the entire growth cycle. The tendency toward filament formation was reduced by lowering the incubation temperature to below 37°C. The results of this study indicate

that Sodium Ricinoleate interfered with cell division in *E. coli*, causing the organisms to develop as filaments.

Mordenti et al. (1982) studied the activity of Sodium Ricinoleate against *Streptococcus mutans* strain 6715-13 (plaque) in vitro. The minimum inhibitory concentration of Sodium Ricinoleate against *S. mutans* was 1% (or 3.12×10^{-2} M). During testing for the minimum inhibitory concentration, the following four distinct concentration-dependent cell growth and acid production phenomena were identified: (1) no cell growth, pH > 6.8; (2) cell growth, medium pH > 6.8; (3) cell growth, medium pH of 5.2 to 6.8; and (4) cell growth, medium pH < 5.2. It was concluded that Sodium Ricinoleate had bactericidal activity in this assay. In subsequent experiments, wire-adherent plaque specimens were treated with various concentrations of Sodium Ricinoleate. The intact plaque samples survived in each of these tests. The authors stated that the ability for Sodium Ricinoleate to kill bacteria in suspension, but not in intact plaque, clearly demonstrates the absence of a correlation between antibacterial activity against cells in suspension and antiplaque activity.

Other Biological Effects

Castor Oil

Capasso et al. (1986) evaluated the effect of castor oil on the formation of histamine, 5-hydroxytryptamine (5-HT), and prostaglandin-like material (PG-LM) in the rat intestine using male, fasted Wistar-Nossan rats (number and weights not stated). Castor oil (dose = 2 ml/rat) was administered by gavage. Following the onset of diarrhea, the animals were killed, and sections of colon removed. Untreated rats served as controls. Castor oil induced an approximately fourfold increase in PG-LM ($p < .01$) and a threefold increase in histamine and 5-HT ($p < .01$). A decrease in the Castor oil–induced colonic formation of PG-LM was noted following pretreatment of the animals with indomethacin (inhibitor of prostaglandin biosynthesis) or hydrocortisone. It was concluded that these data support the idea that the laxative effect of castor oil is the result of increased intestinal production of PG-LM, histamine, and 5-HT.

A study was conducted by Pinto et al. (1989) to determine whether oral administration of castor oil leads to changes in the levels of platelet-activating factor (PAF) formed by intestinal tissue, and whether these changes are related to intestinal damage. Fasted, male Wistar-Nossan rats (number not stated; weights = 130 to 140 g) were used. Castor oil (dose = 2 ml/rat) was administered orally, and the animals were killed.

At 3 h post dosing, the formation of PAF by segments of rat intestine was significantly increased (compared to control rats treated with olive oil) in the following intestinal segments: duodenum ($p < .001$), jejunum ($p < .01$), ileum ($p < .2$), and colon ($p < .1$). At macroscopic examination, extensive regions of hyperemia were noted in the entire intestinal mucosa. The duodenum and jejunum regions were the most severely affected. No increase in PAF was noted in control rats dosed with olive oil.

Compared to control rats dosed with olive oil, castor oil also increased the intraluminal release of acid phosphatase (AP) in the duodenum and jejunum ($p < .01$), and a similar trend was reported for the ileum and colon ($p < 0.2$ to .1). A correlation between increased release of AP and intestinal hyperemia was established. The authors noted that the results of this study suggest that PAF is involved in the mechanism underlying castor oil–induced intestinal damage (Pinto et al. 1989).

In a study by Blair et al. (2000), the affinity of castor oil and numerous other chemicals for the estrogen receptor (from uteri of ovariectomized Sprague-Dawley rats) was evaluated using a validated estrogen receptor competitive binding assay. In this assay, the ability for a chemical to compete with [^{3}H]estradiol ([^{3}H]E_2) for the estrogen receptor was determined, based on IC_{50} (concentration resulting in 50% inhibition of [^{3}H]estradiol binding) values. All assays were replicated a minimum of two times. The mean IC_{50} for Castor oil was $>1.0 \times 10^{-4}$ M. Mean IC_{50} values for 4-heptyloxyphenol and phenophthalein (both classified as moderate estrogen receptor binders) were $6.75 \times 10^{-5} \pm 0.75 \times 10^{-5}$ and $6.73 \times 10^{-6} \pm 1.79 \times 10^{-6}$, respectively. The binding affinity of castor oil for the estrogen receptor was not classified.

Ricinoleic Acid

In a study by Grainger et al. (1982), the effect of Ricinoleic Acid on lymph flow in outerperfused segments of cat ileum was evaluated. The cats were anesthetized and a segment of ileum with intact innervation and lymphatic drainage was isolated and perfused by the intact mesenteric artery. A cannula was inserted into a large lymphatic vessel emerging from the mesenteric pedicle, and lymph flow was determined by observing lymph movement in a calibrated pipette that was connected to the lymphatic cannula. The mean control (± *SE*) value for intestinal lymph flow in the experiments was 0.048 ± 0.005 ml/min × 100 g. Net fluid secretion was stimulated by intraluminal instillation of 5 mM Ricinoleic Acid. The peak increase in intestinal lymph flow produced by Ricinoleic Acid in all experiments was 6.3 ± 1.2 ($n = 5$) times the control. The maximal fluid excretion rate observed with Ricinoleic Acid was 0.43 ± 0.23 ml/min × 100 g.

Yagaloff et al. (1995) evaluated the inhibitory activity of a series of fatty acids and fatty acid derivatives on [^{3}H]leukotriene B_4 binding (LTB_4) to pig neutrophil membranes using the LTB_4 receptor binding assay. The most potent chemicals were 15-hydroxy-DGLA (20:3; $K_i = 1$ μM), eicosadienoic acid (20:2; $K_i = 3$ μM), and Ricinelaidic Acid (18:1; $K_i = 2$ μM). Ricinoleic Acid (18:1) had a K_i of 9 μM.

ANIMAL TOXICOLOGY

Acute Oral Toxicity

Castor Oil

In a study by Capasso et al. (1994), castor oil (2 ml) was administered orally to ten male Wistar rats (weights = 160 to

180 g). The animals were killed and two segments from standardized regions of the duodenum and jejunum were visibly evaluated for macroscopic damage. Mucosal injury was graded according to the following scale: 0 (normal), 1 (hyperemia), 2 (hyperemia with evidence of hemorrhage into the lumen), and 3 (severe hemorrhage into the lumen).

Copious diarrhea was reported for all animals on days 3, 5, and 7 post dosing. Macroscopic damage, characterized mainly by vasocongestion, was observed throughout the duodenum and jejunum. The injury observed ranged from mild (at 1 h) to severe (at 5 h), and was less severe at 7 h. Injury was not observed at 0.5 or 9 h after dosing. Castor oil–induced mucosal damage was associated with statistically significant intraluminal release of acid phosphatase.

In additional experiments (groups of 10 rats), mucosal damage induced by castor oil was exacerbated in the presence of a nitric oxide synthase inhibitor (N^{ω}-nitro-l-arginine methyl ester), suggesting that nitric oxide provides protection against castor oil-induced mucosal damage (Capasso et al. 1994).

In a study involving male Crl:CD BR rats, the findings suggested that castor oil–induced diarrhea is the result of activation of NK_1 and NK_2 receptors by endogenous tachykinins (Croci et al. 1997).

In a study by Johnson et al. (1993), castor oil was administered orally to 12 ponies (17 months to 20 years old; weights = 160 to 250 kg). The ponies were randomly assigned to two treatment groups and one control group (four ponies per group). Each animal received a single 2.5 ml/kg body weight dose of castor oil via nasogastric tube. Diarrhea occurred within 24 h of dosing. The animals were killed and necropsied at 24, 48, and 72 h post dosing. Intestinal tissues were removed and subjected to gross and microscopic examination.

In ponies killed at 24 or 48 h, gross lesions were most severe in the cecum and ventral colon. At 24 h, the mucosa of the distal jejunum, ileum, cecum, and ventral colon was diffusely red. Most of the ponies dosed with castor oil had generalized hyperemia of the intestinal mucosa at 24, 48, and 72 h post dosing, and mesenteric, cecal, and colonic lymph nodes were edematous. Mucosal and submucosal edema of the ileum, cecum, and ventral colon was noted in all ponies dosed with castor oil. At 72 h post dosing, focal acute ulceration and intense mucosal hyperemia of the glandular stomach was observed in three of four ponies dosed with castor oil.

Microscopic changes in the superficial enterocytes of the cecum and ventral colon that were characterized as follows: loss of microvilli; distortion of the cytoplasmic terminal web; expansion of the cytoplasmic matrix, with formation of precipitates; and widening of intracellular spaces between junctional complexes. Castor oil induced acute superficial enterocolitis in ponies. Dosing was associated with transient diarrhea, neutrophilic inflammation, and mucosal erosion in the cecum and ventral colon (Johnson et al. 1993).

Hydrogenated Castor Oil

The acute oral toxicity of a 25% solution of Hydrogenated castor oil in olive oil was evaluated in a study involving ten male Wistar rats (average body weight = 155 g). The test solution (dose = 10 g/kg) was administered via a gastric probe. Toxic signs were not observed in any of the animals, indicating that the test substance was well tolerated. The LD_{50} was >10 g/kg (Allegri et al. 1981).

Ricinoleic Acid

In a study by Okui et al. (1964), male albino rats (number not stated; weight = 120 ± 10 g) were injected intraperitoneally (i.p.) with 100 mg Ricinoleic Acid. All of the animals were dead within 24 h. The i.p. injection of a 5% Ricinoleic Acid emulsion (1 ml) was tolerable, but caused an increase in the following: amount of ascites, conglutination of peritoneal organs, and swelling of the liver. However, histological changes were not observed in liver specimens.

Cetyl Ricinoleate

In an acute oral toxicity test performed by Laboratoire de Recherche et d'Experimentation (1994), Cetyl Ricinoleate was evaluated using five female NMRI EOPS mice (weights = 19 to 22 g). Oral dosing (single dose 2000 mg/kg) was followed by a 6-day observation period. None of the animals died, and clinical and behavioral evaluations were normal. Weight gain was considered satisfactory. The authors stated that Cetyl Ricinoleate was non-toxic in this study.

Ethyl Ricinoleate

The acute oral toxicity of Ethyl Ricinoleate was evaluated using 10 rats (weights and strain not stated). The acute oral LD_{50} exceeded 5.0 g/kg; one animal receiving this dose died. The following clinical signs were observed during the study: isolated instances of diarrhea, chromorhinorrhea, ptosis, and chromodacryorrhea. At necropsy, gross observations were normal for eight animals. Necropsy of the remaining two animals revealed the following changes in either one or both animals: red and/or yellow areas in the intestines, red areas in the stomach, mottled liver, mottled spleen, mottled kidneys, dark lungs, red exudate in the anogenital area, and a large amount of blood in the bladder (Research Institute for Fragrance Materials [RIFM] 2000).

Acute Dermal Toxicity

Ethyl Ricinoleate

The acute dermal toxicity of Ethyl Ricinoleate was evaluated using six rabbits (weights and strain not stated). The acute dermal LD_{50} exceeded 5.0 g/kg; one animal receiving this dose died. The following clinical signs were observed during the study: isolated instances of lethargy, diarrhea, ptosis, and yellow nasal discharge, and moderate erythema (six rabbits), slight edema (one rabbit), and moderate edema (five rabbits). At necropsy,

gross observations were normal for three animals. Necropsy of the remaining animals revealed the following changes: red areas in the intestines, mottled liver, white nodules in the liver, dark areas in the lungs, mottled kidneys, pale kidneys, dark spleen, and dark areas in the stomach (RIFM 2000).

Acute Intravenous Toxicity

Castor Oil

In a study by Lorenz et al. (1982), castor oil was administered intravenously (single bolus dose = 0.1 ml/kg) to each of eight anesthetized adult mongrel dogs (four males, four females; mean weight = 20.5 kg). Blood for histamine assays was obtained at 2, 5, 10, 15, and 20 min post dosing. The incidence of the following signs was determined: erythema, hives (papules and wheals), edema, defecation, tachypnea, hypotension, and death (circulatory arrest). None of the animals died, and erythema (one animal; score not stated) was the only clinical sign that was reported. An increase in blood histamine occurred in one of the eight animals. At 2 min post dosing, the increase was 0.6 ng/ml of whole blood (maximum response) in this animal. The authors concluded that castor oil was ineffective in terms of its ability to cause histamine release.

Short-Term Oral Toxicity

Castor Oil

Masri et al. (1962) conducted an experiment in which ten male weanling rats (4 weeks old; mean weight = 42.2 ± 0.6 g) were fed (ad libitum) 10% castor oil for approximately 5 weeks. A similar control group (mean weight = 42.2 ± 0.5 g) received corn oil in the diet. The animals were observed for growth and catharsis throughout the duration of the study. Castor oil in the diet did not induce catharsis in any of the 10 rats, and growth of these animals was comparable to that observed in the control group. At the end of the experiment, mean weights for test and control rats were 155.6 ± 4.6 and 160.5 ± 4.1 g, respectively. Grossly, there were no abnormalities. Additionally, no abnormalities of the perirenal fat were noted.

Short-Term Subcutaneous Toxicity

Castor Oil

In a study by Migally (1979), the morphology of the adrenal cortex was examined after injection of a commonly used vehicle consisting of castor oil and benzyl benzoate. Twenty-five adult C57 BL/6J mice (weights not stated) were divided into the following groups: group I (five intact control mice); group II (five sham-injected mice); group III (five mice each injected with 0.1 ml castor oil); group IV (five mice each injected with 0.1 ml benzyl benzoate; and group V (five mice each injected with 0.1 ml castor oil and benzyl benzoate [4:1]). The mice were injected subcutaneously daily for four weeks and then killed. Necropsy was then performed and tissues prepared for light and electron microscopic examination.

Using electron microscopy, parenchymal cells containing electron-dense lipid inclusions were observed in the zona fasciculata of adrenal glands from mice injected with castor oil. By light microscopy, there was no difference between adrenal glands from control mice and mice injected with the vehicle consisting of castor oil and benzyl benzoate; however, by electron microscopy, the following variations were observed: profound disruption of parenchymal mitochondria in the zona fasciculata and a noticeable increase in size for some of the mitochondria. The vesicular orientation of the inner mitochondrial membrane was lost (i.e., random orientation), and diminution of the mitochondrial matrix was also observed.

Unlike the controls, the macrophages of exposed animals had large vacuoles filled with globular material. In the zona reticularis and inner zona fasciculata, parenchymal mitochondria with circular orientation of the inner membrane (normally few) were noticeably increased in number. The authors noted the appearance of similar mitochondria following adrenocorticotrophic hormone (ACTH) stimulation, and that the alteration of ACTH release has been shown to result from stress. The author concluded that subcutaneous injection of the castor oil–benzyl benzoate vehicle caused morphological alterations that are suggestive of stress-induced changes (Migally 1979).

Subchronic Oral Toxicity

Castor Oil

In a study by the National Toxicology Program (NTP) (1992), the subchronic oral toxicity of castor oil was evaluated using groups of 10 male and 10 female F344/N rats (mean body weights = 126 to 132 g [males]; 107 to 110 g [females]) and groups of 10 male and 10 female $B6C3F_1$ mice (mean body weights = 22.6 to 23.0 [males]; 17.2 to 17.7 [females]). In the core study, five groups of test animals (mice or rats) received diets containing 0.62%, 1.25%, 2.5%, 5.0%, or 10% castor oil continuously for 13 weeks. Two control groups of 20 rats and 20 mice received diets that did not contain castor oil.

Ten additional rats/sex were included at each dose level for evaluation of hematological and clinical chemistry parameters. On days 5 and 21, these animals were anesthetized with CO_2 and blood samples were obtained. These animals were killed following blood collection on day 21. Blood samples for hematology and clinical chemistry were also collected from core-study rats at the end of the study. Also, at the end of the 13-week study, all core-study animals were killed and complete necropsies were performed, including complete microscopic examination on tissues, on all test and control animals.

No effect on survival or significant differences in average food consumption were noted in any of the five groups of male and female rats fed castor oil. There were no significant differences in mean body weights between test and control groups.

The following hematological effects were reported for male rats: a slight decrease in mean corpuscular hemoglobin

concentration (MCHC) (10% castor oil diet), a statistically significant decrease ($p < .05$) in mean corpuscular volume (MCV) (10% castor oil diet), a decrease in mean corpuscular hemoglobin (MCH) (5% and 10% castor oil diets), and an increase in platelets (1.25%, 5%, and 10% castor oil diets). The only hematological abnormality reported for female rats was a statistically significant ($p < .05$) decrease in reticulocyte counts (0.62% or 10% castor oil diets). The authors did not consider the hematological effects to be biologically significant. A dose-related increase (treatment-related) in the activity of serum alkaline phosphatase was noted in male and female rats at days 5 and 21, and at the end of the study.

Increased heart-to-body weight ratios were reported for male rats that received 0.62%, 2.5%, and 10.0% castor oil diets; however, no increase in absolute heart weight was noted. The observed differences in organ-to-body weight ratios were not considered treatment related. Microscopically, no morphologic changes were associated with the slight differences in organ weights between the dietary groups.

Compared to controls, liver weights were increased in male and female mice receiving diets containing 5% or 10% castor oil. Increased kidney weights were reported for female mice that received 5% or 10% castor oil in the diet. Microscopically, there was no evidence of morphologic changes that could be been associated with the slight differences in organ weights between the groups tested. Additionally, there was no evidence of treatment-related lesions in any of the tissues or organs that were examined (all dietary groups).

The authors concluded that diets containing up to 10% castor oil to B6C3F$_1$ mice and F344/N rats was not associated with toxicity to any specific organ, organ system, or tissue in this study (NTP 1992).

Hydrogenated Castor Oil

In a preliminary feeding trial (preparatory to the 16-week feeding study reported below) by Binder et al. (1970), the subchronic oral toxicity of 5%, 10%, and 20% Hydrogenated Castor Oil [HCO] (effective concentrations of 4.33%, 8.65%, and 17.3%, respectively) in the diet was evaluated using groups of three weanling female rats (Slonaker substrain of Wistar strain). The sample of HCO that was incorporated into the diet contained 86.5% 12-hydroxystearic acid, 10.3% nonoxygenated acids, and 3.2% 12-ketostearic acid. The three groups were fed for 90 days, and the control group received diet only. At necropsy, organ weights were recorded and numerous tissues were prepared for microscopic examination. Prior to necropsy, blood samples were obtained for hematological evaluation.

A reduced growth rate in rats fed 8.65% and 17.3% Hydrogenated Castor Oil, respectively, was the only abnormality that was noted. The authors stated that it is likely that Hydrogenated Castor Oil was poorly digested because of its high melting point. Therefore, the poor body weight gains may have been due to the lower caloric density of the diets containing Hydrogenated Castor Oil at concentrations $\geq$8.65%.

To confirm the authors' explanation of the results, the experimental trial was repeated with Hydrogenated Castor Oil at concentrations of 1%, 5%, and 10% in corn oil (effective concentrations of 0.865%, 4.33%, and 8.65%, respectively). The amount of corn oil added to the individual diets was 9%, 15%, and 10%. Corn oil (20%) was present in the control diet. Necropsy was performed at the end of the 90-day feeding period. The growth rate for rats fed 8.65% Hydrogenated Castor Oil in the diet was equivalent to that of the other dietary groups. Based on organ weight determinations and the results of hematological and microscopic evaluations, no adverse effects were observed (Binder et al. 1970).

In the 16-week feeding study (Binder et al. 1970), groups of 15 male albino rats (Slonaker substrain of Wistar strain, weights = 43 to 83 g) received diets containing the following oils: group 1 (fed 0.865% HCO + 19% corn oil for 16 weeks), group 2 (0.865% HCO + 19% corn oil for 8 weeks, then 20% corn oil for 8 weeks), group 3 (8.65% HCO + 10% corn oil for 16 weeks), and group 4 (8.65% HCO + 10% corn oil for 8 weeks, then 20% corn oil for 8 weeks).

The effective dietary concentrations of Hydrogenated Castor Oil in the 0.865% and 8.65% diets were 0.865% and 8.65%, respectively. The control group received 20% corn oil in the diet for 16 weeks.

At 4, 8, 12, and 16 weeks, the number of animals on the HCO diets that were alive was 33, 30, 12, and 6 rats, respectively. The corresponding number of rats remaining on the control diet was 15, 15, 12, and 6.

Weight differences due to the different diets at 4 and 8 weeks were not statistically significant at the 95% level. However, at 12 weeks, rats on the 8.65% HCO diet weighed significantly less than rats on the control diet or 0.865% HCO diet. At 16 weeks, the weight gain of rats on the control diet was significantly greater than that of rats on the 8.65% HCO diet for 16 weeks or on the 8.65% HCO diet for 8 weeks + control diet for the remaining 8 weeks. The weight gain of rats on the 0.865% HCO diet was not significantly different from that of rats on the control diet. Except for the reduced growth rate in rats receiving the 8.65% HCO diet, no adverse effects were observed (Binder et al. 1970).

Ocular Irritation/Toxicity

Castor Oil

Carpenter and Smyth (1946) evaluated the ocular irritation potential of castor oil using albino rabbits. The eyelids were retracted, and undiluted castor oil (0.5 ml) was applied to the center of the cornea. At approximately 1 min post instillation, the eyelids were released. Reactions were scored 18 to 24 h later. To determine the score for ocular injury, the individual numerical scores for each eye were added together and then divided by the number of eyes (usually 5). An ocular injury score of 5.0 represents severe injury, and corresponds to necrosis (covering approximately three-fourths of the corneal surface) that is visible only after staining or a more severe necrosis covering a smaller

area. An injury grade of 1 (maximum score possible = 20) was reported for castor oil.

In a study by Guillot et al. (1979), the effects of various oils (used in cosmetics) on the rabbit eye were assessed according to the official French methods. Following the instillation of undiluted castor oil (no ocular rinsing), no corneal involvement was found. However, slight congestion of the iris and conjunctiva was observed.

Castor oil served as one of the vehicle controls in a study by Behrens-Baumann et al. (1986) investigating the effect of topical cyclosporin A on the abraded and intact rabbit cornea. Two castor oil vehicle control groups of outbred male rabbits were used. In both groups (10 eyes/group), 10 drops of castor oil were instilled daily for 3 weeks, and all eyes were examined using a slit-lamp. At the end of the 3-week period, the animals were killed and their corneas were excised and prepared for scanning and transmission electron microscopy. For eyes (no corneal abrasion) treated with Castor oil and examined by scanning electron microscopy, there were increased numbers of medium dark and dark epithelial cells (compared to the normal epithelium). The cell borders were intact and the number of holes was reduced. By transmission electron microscopy, corneal endothelial cells were intact. By scanning electron microscopy, an intact cell pattern, with more cell microstructures than normal, for the corneal endothelium was reported. Ultrathin sections appeared normal. The authors concluded that castor oil did not damage the rabbit corneal epithelium or endothelium (Behrens-Baumann et al. 1986).

Skin Irritation

Castor Oil

Butcher (1951) evaluated the effect of castor oil and other substances on rat epidermis using, primarily, 22-day-old albino or Long-Evans rats or rats of either strain in which the hair follicles were in the resting stage; older animals were also used. A total of 216 rats were used in the experiments. Using dry cotton, castor oil was gently swabbed onto a large area of the dorsum (clipped free of hair) daily for 5 days to 2 weeks. At the end of the application period, small areas of skin were obtained from the backs of treated and control (one rat/litter) rats and examined microscopically. Castor oil was described as having a mild effect on the epidermis. The alteration was mainly confined to the stratum granulosum, where the cells were more numerous and the granules were more evident, compared to the litter-mate controls.

In a study by Meyer et al. (1976), castor oil (undiluted, 4 drops) was rubbed onto either side of the shaved left flank of each of six albino guinea pigs (weights = 300 to 400 g) and six young pigs (weights = 10 to 12 kg) within 30 s on 10 successive days. The opposite side served as the control field, and was massaged only. The animals were killed on day 11. A tissue section (1 × 1 cm) was excised from the center of the test fields and prepared for microscopic examination. Five to eight sections of 7 μm thickness per test area were used for evaluation. A micrometer eyepiece was inserted into the microscope, and epidermal width measurements (25 per individual field; 400× magnification) were made. Castor oil induced slight reddening at the application site.

Microscopically (pigs and guinea pigs), hyperplasia, with an increase in the cell and nuclear density of the basal cell layers, was observed. Additionally, widening of the granular layer and thick keratohyalin granules were observed. Average epidermal widths (25 measurements) were as follows at the end of the 10-day application period: guinea pigs (control, 27.9 μm; test, 94.3 μm) and pigs (control, 26.5 μm; test, 32.3 m) (Meyer et al. 1976).

Rantuccio et al. (1981) evaluated the effect of undiluted castor oil on the skin of five female albino rabbits (6 months old; mean weight = 2500 g). The test substance (0.5 ml) was gently massaged into depilated skin of the right flank of each rabbit daily for 30 days. Following each daily application, the test site was covered with a simple protective dressing. Two small skin punch biopsies were obtained from the right and left (control) flank on days 10, 20, and 30. Macroscopic skin changes included slight erythema and slight edema. Microscopic examination of punch biopsies obtained at day 10 revealed "clear-cut" acanthosis with vacuolization and disorganization of the basal layer. These changes were associated with some infiltration of the dermis by mononuclear cells and fibrocytes. After days 20 and 30, the epithelial changes became progressively more evident. However, the appearance of the infiltrate remained unchanged.

In a study by Guillot et al. (1979), the effect of various oils that are used in cosmetics on rabbit skin was assessed according to the official French methods. An open skin irritation test and repeated application test (8 weeks) were performed. The authors stated that the two samples of undiluted castor oil, as well as 10% aqueous castor oil, were well tolerated.

In a study by Motoyoshi et al. (1979), the skin irritation potential of castor oil (undiluted) was evaluated using six albino angora rabbits (average weight = 2.6 kg). The test substance (0.1 g of liquid or semisolid) was applied for 24 h to two test areas (clipped free of hair) on the dorsal surface of each animal. A plastic collar was wrapped around the neck of each animal and remained throughout the application period, after which reactions were scored according to the following scale: − (no reddening) to + + + (severe reddening, beet redness). At 30 min, after reactions were scored, the test areas were clipped free of hair and castor oil was reapplied according to the same procedure. A third application of Castor Oil occurred 48 h later, and reactions were scored. Reactions were scored again at 72 h. Castor oil was severely irritating to the skin of rabbits (score = 3).

The skin irritation potential of Castor oil was also evaluated using guinea pigs, rats, and miniature swine in this study.

Undiluted castor oil (0.1 g) was applied daily to dorsal skin (clipped free of hair) in the mid-lumbar region of six male Hartley guinea pigs (weights = 350 to 500 g) and six male Wistar rats (weights = 250 to 350 g). The period of testing, the frequency of application, and the method of evaluation of skin reactions were

the same as those in the preceding test involving albino angora rabbits. Castor oil was mildly irritating to the skin of guinea pigs and rats.

The skin irritation potential of undiluted castor oil was evaluated using six miniature swine of the Pitman-Moore strain (1 month old). The test substance (0.05 g) was introduced under a 15-mm-diameter patch that was secured with adhesive tape. The entire trunk was wrapped with rubberized cloth during the 48-h exposure period. Reactions were scored after patch removal. The grading scale was the same as that used in the experiment involving albino angora rabbits that is summarized above. Castor oil was not irritating to the skin of miniature swine (Motoyoshi et al. 1979).

Ricinoleic Acid

In a study by Vieira et al. (2000), 8 to 10 male Swiss mice received a single topical application of Ricinoleic Acid (in peanut oil, 10 mg/mouse) on the ventral surface of the paw. Neither erythema nor edema of the paw was observed.

Vieira et al. (2001) presented results on the skin irritation potential of Ricinoleic Acid, using groups of six male albino Dunkin-Hartley guinea pigs, in a study evaluating pro- and anti-inflammatory effects of Ricinoleic Acid (concentration not stated). Ricinoleic Acid (0.1 ml, in peanut oil) was administered topically to the entire eyelid surface. The thickness of the eyelid was measured in mm using ophthalmic microcallipers. Topical treatment with Ricinoleic Acid (10, 30, or 100 mg/guinea pig) caused eyelid reddening and edema. A moderate, dose-dependent eyelid edema was reported as follows: 0.12 ± 0.05 mm (10 mg Ricinoleic Acid), 0.18 ± 0.02 mg (30 mg), and 0.23 ± 0.1 mm (100 mg). Maximal edema was achieved at 2 h post-application. The application of Ricinoleic acid vehicle did not cause any appreciable edema.

Cetyl Ricinoleate

In a skin irritation study conducted by the Laboratoire de Recherche et d'Experimentation (1994), Cetyl Ricinoleate (test concentration not stated) was evaluated using three male New Zealand albino rabbits. Erythema and edema were observed in three and two rabbits, respectively, up to 72 h post application. At 72 h, one rabbit had an erythema score of 2 and the remaining two had an erythema score of 1. Edema (score = 1) was observed in two rabbits at 72 h. All reactions were totally reversible at 7 days post application. Cetyl Ricinoleate was a nonirritant in this study.

Zinc Ricinoleate

In a study by International Bio-Research, Inc. (1977a), the skin irritation potential of 10% Griillocin HY 77 (now TEGODEO HY 77) in soybean oil was evaluated using six New Zealand albino rabbits (weight range = 2.4 to 2.6 kg). TEGODEO HY 77 (trade name mixture for Zinc Ricinoleate) has the following composition: Zinc Ricinoleate (more than 50%), triethanolamine (10% to 25%), dipropylene glycol (1% to 5%), and lactic acid (1% to 5%). The diluted test substance (concentration after dilution not stated) was applied to clipped skin on two areas of the back of each animal; one of the areas was abraded. The test sites were covered with occlusive patches (secured with adhesive tape) and the entire trunk was wrapped in a rubber sleeve. The patches remained in place for 24 h and reactions were scored at 24 and 72 h post application as follows according the Draize scale: 0 (no erythema) to 4 (severe erythema [beet redness] to slight eschar formation [injuries in depth] and 0 (no edema) to 4 (severe edema, raised more than 1 mm and extending beyond area of exposure).

At 24 h post application, well-defined erythema was observed at the abraded test sites of all six rabbits and slight erythema was observed at the intact sites of four rabbits. Well-defined erythema was observed at the intact sites of two rabbits during the 24-h reading. At 72 h post application, five rabbits had mild erythema at the abraded site and two rabbits had the same reaction at the intact site. Using the data for abraded and intact test sites, a total primary irritation index of 1.1 was calculated (International Bio-Research, Inc. 1977a).

Effect on Intestinal Membranes

Ricinoleic Acid

In a study by Morehouse et al. (1986), a single 0.1 ml dose of Ricinoleic Acid (100 mg/ml) was administered intragastrically to fasted, specific pathogen-free mice (strain CD-1, number not stated). This dosage of Ricinoleic Acid was determined, on a weight basis, to be approximately the dosage that is used therapeutically in humans. Groups of mice were killed at various intervals, and light microscopy and transmission and scanning electron microscopy were used to identify structural alterations. At 2 h post dosing, the duodenal villi were markedly shortened when compared to control duodenal villi. This erosion of the villi throughout the duodenum caused massive exfoliation of columnar and goblet cells, filling the lumen with cellular debris and mucus. Disruption of the mucosal barrier resulted in continuity between the intestinal lumen and lamina priopria of the villi, with the loss of formed blood elements and lamina propria constituents into the intestinal lumen. The mucosal damage was much more localized at 4 h post dosing, and the erosion of the villi had been largely repaired. Repair was complete at 6 h post dosing.

Sodium Ricinoleate

Gaginella et al. (1977a) studied the morphology of the colon after perfusion of the organ with Sodium Ricinoleate solution, using 25 male New Zealand white rabbits. A Foley catheter was inserted and the colon was perfused with control solution (1.25 ml per minute) for 20 min. The colon was then flushed and perfused with the test solutions. Ten rabbits were perfused with 10 mM Sodium Ricinoleate and the remaining three groups of five were perfused with 2.5, 5.0, and 7.5 mM, respectively. The animals were killed and full thickness samples of colon

were obtained and prepared for light or scanning electron microscopy. In additional experiments, tissue was obtained from animals that were not perfused and those perfused with control buffer (contained PEG 400) for 60 or 160 min.

Using scanning electron microscopy, mucous membranes of colons perfused with control buffer for 60 min and unperfused colons appeared normal. After 1 h of perfusion with 10 mM Sodium Ricinoleate, patchy loss of epithelial cells was apparent. The loss of epithelial cells was confirmed using light microscopy. A dose response-relationship for mucosal damage was noted. The most severe damage resulted from perfusion with 10 mM Sodium Ricinoleate. Lesser degrees of change and lesser areas of mucosal surface affected were associated with lower concentrations. Small amounts of DNA (due to loss of DNA into the lumen) accumulated in the control buffer, and perfusion with 2.5 mM Sodium Ricinoleate did not cause a significant increase in these amounts. However, DNA accumulation increased significantly after perfusion with 5.0, 7.5, and 10 mM Sodium Ricinolete, and a plateau was reached with 7.5 mM solutions.

The control perfusate that was used contained low (PEG 400) and high-molecular-weight (PEG 4000) polyethylene glycols. The absorption of PEG 400 increased progressively with increases in the concentration of Sodium Ricinoleate (above 2.5 mM) perfused. Sodium Ricinoleate (2.5 mM) did not increase absorption of the individual polymers that comprise PEG 400 (molecular weight 242 to 594). The absorption of lower molecular weight polymers (242 to 462) increased significantly ($p < .005$) in the presence of 5, 7.5, and 10 mM Sodium Ricinoleate. For higher molecular weight polymers (506 to 594), increased absorption in the presence of 5 mM Sodium Ricinoleate was not observed. However, significantly increased absorption ($p < .05$) of the higher molecular weight polymers was observed during perfusion with 7.5 and 10 mM Sodium Ricinoleate (Gaginella et al. 1977a).

Comedogenicity

Castor Oil

Fulton et al. (1976) evaluated the comedogenicity of cosmetics and cosmetic ingredients. Castor oil was applied to the base of the internal right ear of each of three white albino rabbits on Monday through Friday for 2 weeks. The left ear served as the control. Comedogenicity was clinically evaluated on the following Monday using the scale: 0 (no increase in visible hyperkeratosis) to 5 (severe lesions, such as those seen following the application of coal tar). Six-millimeter punch biopsies (right ear) were obtained for microscopic examination, and comedogenicity was evaluated according to the same scale. A score of 1 (increase in visible hyperkeratosis extending to the possible presence of comedones) was reported for castor oil, which was considered nonsignificant because repeated testing in different rabbits often failed to produce identical effects.

In a study by Morris and Kwan (1983), the comedogenicity of castor oil was evaluated using three New Zealand rabbits. Castor oil was applied to the external ear five times weekly (one application per day). The actual amount applied ranged from 5 to 10 mg/cm^2. The animals were killed after the 14th application and several full-thickness sections of the test site and contralateral untreated ear were prepared for microscopic examination. Comedogenic activity was based on the degree of follicular hyperkeratosis and other morphologic changes in the majority of pilosebaceous units, when compared to control sections. Comedogenicity was graded according to the following scale: 0 (negative: pilosebaceous unit normal in appearance when compared to control [untreated] ear section) to 5 (severe: widely dilated follicles, filled with packed keratin; follicular epithelial hyperplasia causing partial or total involution of sebaceous glands and ducts; possible inflammatory changes). Comedogenicity scores for castor oil were 0 to 1, corresponding to a slight increase in keratin content within the follicle, with essentially no change in follicular epithelium.

Fulton (1989) evaluated the comedogenicity of castor oil (mixed in propylene glycol at 9:1 dilution; test concentration = 10%) using three New Zealand albino rabbits. The test substance (1 ml) was applied onto the entire inner surface of one ear 5 days per week for 2 weeks. Untreated ears served as controls. Follicular keratosis was evaluated both macroscopically (visually) and microscopically, using a micrometer to measure the width of the follicular keratosis. Follicular keratosis was evaluated according to the following scale: 0 or 1 (no significant increase in follicular keratosis) to 4 or 5 (an extensive increase in follicular keratosis). The following scale was used to evaluate skin irritation that resulted from repeated application of the test substance: 0 (no irritation) to 5 (epidermal necrosis and slough). A follicular keratosis score of 1 and an irritation score of 0 was reported for castor oil.

Skin Sensitization

Zinc Ricinoleate

International Bio-Research, Inc (1977b) conducted a skin sensitization study on Grillocin HY 77 (now TEGODEO HY 77) using 30 white guinea pigs (average weight = 300 g). TEGODEO HY 77 (trade name mixture for Zinc Ricinoleate) has the following composition: Zinc Ricinoleate (more than 50%), triethanolamine (10% to 25%), dipropylene glycol (1% to 5%), and lactic acid (1% to 5%) (Degussa 2001). Test and control groups consisted of 20 and 10 guinea pigs, respectively. A closed patch containing the trade mixture (0.5 ml, undiluted) was placed on the left shoulder, clipped free of hair, of each animal and remained in place for 6 h. Treatments were repeated once weekly for 3 weeks.

At two weeks after the last exposure, test and control guinea pigs were challenged (same procedure, right side) with duplicate closed patches. At 24 h post removal of the patches, the test site on each animal was treated with a depilatory. Sites were then rinsed and reactions were graded 2, 24, and 48 h later according to the method of Draize. Clinical observations were

normal throughout the study. Erythema and eschar formation were graded according to the following scale: 0 (no erythema) to 4 (severe erythema [beet redness] to slight eschar formation [injuries in depth]). The following scale for edema formation was used: 0 (no edema) to 4 (severe edema [raised more than 1 mm and extending beyond area of exposure]). A score of 0 (erythema and edema) was reported for each test and control animal during each of the three grading periods. Grillocyn HY 77 did not produce a positive reaction in any of the test animals (International Bio-Research, Inc. 1977b).

Allergenicity

Hydrogenated Castor Oil

Lehrer et al. (1980) investigated the presence of castor bean allergens in Castor Wax (Hydrogenated Castor Oil) using both in vitro and in vivo analyses. The experiments in this study involving human subjects are summarized under the subsection Allergenicity in the Clinical Assessment of Safety section of this report. The following solvents were used to prepare aqueous extracts of Castor Wax: phosphate buffered saline (PBS), urea-NaCl, Triton X-100, or sodium lauryl sulfate (SLS). The aqueous extracts prepared were analyzed for total organic matter and protein (expressed as percent of total organic matter or dry weight). Results from assays for protein in the aqueous extracts of Castor Wax were as follows: 100% protein (urea-NaCl extract), 17.0% (SLS extract), 12.9% (PBS extract), and 0.34% (Triton X-100 extract) protein.

In a qualitative precipitation analysis, Castor Wax aqueous extracts were analyzed for castor bean antigens using an immunodiffusion procedure involving rabbit antiserum (50 μl of antigen or antiserum in each well). The antiserum was produced in rabbits that had been immunized with extracts of castor bean in Freund's complete adjuvant. Of the aqueous extracts tested, only the SLS Castor Wax extract reacted with the rabbit antisera. No reactivity with the antiserum was observed when the SLS solution alone was tested. Because the reaction was unusually strong, suggesting a false-positive reaction, the extract was tested with normal rabbit serum. However, the same precipitin line was observed. It was concluded that the reaction of the SLS Castor Wax extract with rabbit serum proteins was probably a nonspecific reaction.

The aqueous extracts of Castor Wax were also analyzed for allergens by determining their reactivity (48-h passive cutaneous anaphylaxis [PCA] reaction) with mouse anticastor reaginic (immunoglobulin E [IgE]) antibody. CFW female mice were sensitized with reaginic antibody that was produced in BDF mice that had been immunized with heated castor bean extracts and *Bordetella pertussis* adjuvant. The mice were challenged 48 h later by intravenous injection of extracts of Castor Wax in 0.5% Evans blue solution and killed at 30 min post injection. Nonsensitized mice challenged with Castor Wax extracts served as controls. The reactions in control mice were always classified as negative. The antiserum did not react with PBS, urea-NaCl, or Triton X-100 extracts of Castor Wax, but a positive response (positive PCA titer of 40, mean of three determinations) resulted from the reaction of the antiserum with the SLS extract of Castor Wax.

The radioallergosorbent test (RAST) inhibition assay was used for further analysis of allergens in the extracts of Castor Wax. This assay was capable of detecting up to 7.5 μg castor allergen. Patient serum (100 μl) and Castor Wax extract (100 μl) were incubated with antigen-coated discs. Incubation was followed by the addition of radioiodinated anti-IgE and another incubation period. Normal serum incubated with antigen coated discs and test serum incubated with discs coated with a non–cross-reacting antigen served as controls. Assay results were expressed as percent inhibition of the patients' RAS test. Neither the PBS nor Triton X-100 extract of Castor Wax had an inhibitory effect on the RAS test, whereas the urea-NaCl extract induced a small degree (13.4%) of inhibition. However, the SLS Castor Wax extract induced significant inhibition (40.1%) of the RAS reaction.

Based on the nonspecific reaction of the SLS Castor Wax extract with rabbit serum proteins in the first experiment of this study, the authors examined the possibility that this extract may inhibit the RAST nonspecifically using a heterologous RAST inhibition assay. An SLS or Triton X-100 extract of Castor Wax was incubated with reaginic antisera from a patient who was sensitive to peanuts, and then assayed in the peanut RAST. The SLS extract of Castor Wax inhibited the peanut RAST by 37%, suggesting that the results for the RAST inhibition assay in the preceding paragraph may be due entirely or, in part, to a nonspecific inhibition of the RAST reaction, rather than indicating that the SLS extract of Castor Wax contains specific castor allergens.

The results of these experiments (both in vitro and in vivo) indicate that allergens are present at low concentrations in Castor Wax, because the SLS Castor Wax extract induced positive PCA reactions in mice and a positive direct RAS test for reaginic castor allergens (Lehrer et al. 1980).

NEUROTOXICITY

Sodium Ricinoleate

Fox et al. (1983) applied Sodium Ricinoleate (0.01% to 1.0%) to the serosal surface of the rat (anesthetized male Sprague-Dawley rats, weights = 200 to 225 g) jejunum every 5 min for 0.5 h. At 30 days post application, treated and untreated jejunal segments were removed and examined microscopically. At a concentration of 0.1%, Sodium Ricinoleate significantly reduced ($p < .02$) the number of ganglion cells in the myenteric plexus. The highest test concentration (1% Sodium Ricinoleate) significantly reduced the number of cells in the myenteric plexus ($p < .01$) and the submucosal plexus ($p < .02$). The lowest test concentration (0.01%) did not significantly reduce the number of ganglion cells in the myenteric or submucosal plexus.

Renal Toxicity

Castor Oil

Langer et al. (1968) evaluated effects of castor oil on the rat kidney, following intraluminal injection, using five male white albino rats (weights = 180 to 220 g). The kidney was exposed and proximal tubuli were punctured (using Leitz stereomicroscope) and filled with castor oil. At 15 to 60 min, the proximal tubular segments (filled with oil) were fixed in situ. The kidney was excised and prepared for both light and electron microscopic examination. The injection of castor oil resulted in dilatation of the proximal convoluted tubular lumen and compression of the brush border of proximal tubular cells, although a cessation of the normal pinocytosis was induced in the presence of tubular fluid. A toxic effect of castor oil on the tubular epithelium was not observed.

In a study by Racusen et al. (1986) evaluating the nephrotoxicity of cyclosporine, castor oil served as the vehicle control. Two control groups of eight adult male Munich-Wistar rats (weights = 220 to 270 g) were used. Group 1 vehicle controls (vehicle-ischemia group) underwent bilateral renal artery clamping, and were then dosed intraperitoneally (i.p.) daily (4-day period) with a volume of castor oil vehicle that was equivalent to the i.p. dose of cyclosporine (dose = 60 mg/kg). Group 2 vehicle controls (vehicle-sham group) underwent sham surgery. The incisions were closed prior to daily i.p. dosing with castor oil. On postoperative day 3, feed was withdrawn and urine was collected over a period of 12 h for clearance studies. On postoperative day 4, the animals were killed and tissues prepared for light and electron microscopy (confined to cortical proximal convoluted tubules).

In separate studies, renal blood flow and the glomerular filtration rate were determined. Two groups of eight saline control groups (saline-ischemia and saline-sham groups, respectively) were treated according to the same procedure.

Tubular vacuolization was a prominent feature in vehicle-ischemia control rats, and was observed to a minor degree in the vehicle-sham control rats. The clear vacuoles observed using light microscopy probably corresponded to two ultrastructural changes, dilated endoplasmic reticulum and lipid droplets. With the exception of the saline-sham group, dilated endoplasmic reticulum was observed in all control animals. However, lipid droplets were not observed in any of the control animals. The presence of eosinophilic cytoplasmic inclusions was reported for two of eight vehicle-ischemia rats and four of eight vehicle-sham rats, but not for saline-control rats. Tubular necrosis (confined entirely to outer medullary pars recta) was observed in half of the vehicle-ischemia rats and in all saline-ischemia rats, but was not observed in saline-sham or vehicle-sham control groups. Tubular regeneration was noted in vehicle-ischemia and saline-ischemia control rats.

The renal clearance study involved two control groups that were dosed with castor oil (five rats) or mineral oil (six rats). Two 20-min inulin clearance determinations were made, and the vehicle was then administered i.p. (at dose volume comparable to 60 mg/kg cyclosporine dose; administered to test group of seven) in a region of the upper right abdomen. Dosing was followed by five 20-min clearance periods. Compared to the mineral oil control group (six rats), a significant decrease (25% decrease; $p < .05$) in renal blood flow was noted in rats (group of five) dosed i.p. with castor oil. Renal vascular resistance was also significantly higher ($p < .05$). No significant changes in inulin clearance were reported. A significant decrease in blood pressure was noted in all groups; however, blood pressure remained within the physiologic and autoregulatory range (Racusen et al. 1986).

GENOTOXICITY

Castor Oil

Zeiger et al. (1988) evaluated the mutagenicity of castor oil (United States Pharmacopeia purity grade; in dimethylsulfoxide [DMSO]) in the preincubation assay using *Salmonella typhimurium* strains TA97, TA98, TA100, TA1535, and TA1537. The assay was conducted, with and without metabolic activation, according to a modification of the procedure by Haworth et al. (1983). Castor oil was tested at doses up to 10,000 μg/plate and classified as nonmutagenic (all strains, with and without metabolic activation) over the range of doses tested in this assay.

Hachiya (1987) evaluated the mutagenicity of castor oil using *S. typhimurium* strains TA97, TA98, TA100, TA102, TA1537 and strain WP2/pKM102 of *Escherichia coli*. In each strain, castor oil was tested at doses ranging from 100 to 5000 μg/plate with and without metabolic activation. Results were negative for all strains tested, both with and without metabolic activation. Cytotoxicity (at doses of 100 to 5000 μg/plate) was noted only in strain WP2/pKM102, with and without metabolic activation.

NTP (1992) reported a study in which groups of male and female B6C3F$_1$ mice received diets containing 0.62%, 1.25%, 2.5%, 5.0%, and 10% castor oil (in DMSO) for 13 weeks. Blood samples were obtained at the end of the 13-week study. There was no evidence of induction of micronuclei in peripheral erythrocytes.

NTP (1992) also evaluated the induction of sister chromatid changes by castor oil (in DMSO) in Chinese hamster ovary cells. The cells were incubated with castor oil at concentrations up to 5000 μg/ml with and without metabolic activation. Mitomycin-C and cyclophosphamide served as positive controls without and with metabolic activation, respectively. DMSO served as the solvent control. Castor oil did not induce sister chromatid exchanges either with or without metabolic activation.

Sodium Ricinoleate

In a study by Haworth et al. (1983), the mutagenicity of Sodium Ricinoleate was evaluated according to the preincubation procedure of the Salmonella assay (Ames et al. 1975) as described by Yahagi et al. (1975). Sodium Ricinoleate (in distilled water) was tested at concentrations up to 33 μg/plate

at one testing laboratory, and up to 333.3 μg/plate at another laboratory. The following *S. typhimurium* strains were used with and without metabolic activation: TA98, TA100, TA1535, and TA1537. Distilled water served as the vehicle control. The positive controls were as follows: 2-aminoanthracene, 4-nitro-*o*-phenylenediamine, sodium azide, and 9-aminoacridine. Results for Sodium Ricinoleate were negative with and without metabolic activation.

DNA Binding of Reaction Product

Methyl Ricinoleate

Frankel et al. (1987) evaluated the fluorescence formed by the interaction of DNA with lipid oxidation products. The keto ester, methyl ketoricinoleate, was prepared from Methyl Ricinoleate by allylic oxidation with a CrO_3-pyridine complex in methylene chloride. Results from this assay indicated that methyl ketoricinoleate (relative concentration = 1 mM/3 ml) produced very little fluorescence (mean = 29 ± 2 units) with DNA in the presence of ferric chloride–ascorbic acid. The mean value for methyl linolenate hydroperoxides (used as reference) was 362 ± 9 units. Mean values were given as the fluorescence intensity of DNA after 72 h (excitation: 315 nm; emission: 420 nm).

CARCINOGENICITY

Ricinoleic Acid

In a study by Boyland et al. (1966), 2% Ricinoleic Acid, in gum tragacanth, was injected intravaginally into each of 20 BALB/c female mice (5 to 6 weeks old). Injections were made twice per week, and each animal received a total of 100 injections. According to the same dosing schedule, each of 20 positive control and 30 vehicle control mice received 62 injections of 0.3% dimethylbenz(*a*)anthracene and 100 injections of gum tragacanth, respectively. An additional 30 mice served as the untreated control group.

None of the 20 mice injected with Ricinoleic Acid had neoplasms or hyperplastic lesions of the corpus uteri, cervix uteri, vagina, or perineal skin. However, benign lung adenomas were observed in ten of the 13 mice dosed with Ricinoleic Acid and in 6 of the 24 vehicle control mice that were killed after the 14th month of dosing. Benign lung adenomas were also observed in 9 of the 20 untreated control mice that were killed after the 14th month. The difference in benign adenoma incidence between Ricinoleic Acid–treated and untreated control mice was not statistically significant ($p > .1$). Additionally, compared to controls, there was no tendency for Ricinoleic Acid–treated mice with adenomas to have a higher average number of tumor nodules per mouse. Malignant tumors of the vagina and perineal skin were observed in 15 of the 20 (75%) positive control mice. The authors stated that the results of this study do not provide any definite evidence of the carcinogenicity of Ricinoleic Acid (Boyland et al. 1966).

Tumor Promotion Activity

Castor Oil, Ricinoleic Acid, and Glyceryl Ricinoleate

Shubik (1950) evaluated the tumor promotion activity of castor oil, Ricinoleic Acid, Glyceryl Ricinoleate and the following other chemicals using groups of mice (strain and weights not stated): 20% turpentine in liquid paraffin, 0.3% acridine in liquid paraffin, 0.5% fluorene in liquid paraffin, 1% phenanthrene in liquid paraffin, silver nitrate (10% aqueous), and oleic acid. Castor oil, Ricinoleic Acid, Glyceryl Ricinoleate, and oleic acid were not diluted prior to testing.

Initially, the nine substances were tested using groups of three mice (one group per substance). The test substances were applied to a small area of skin in the interscapular region (clipped free of hair). Four applications per mouse, spaced over a period of 2 weeks, were made. At the end of the application period, the animals were killed and tissues were obtained for microscopic examination. The following substances induced slight epidermal hyperplasia: castor oil, Ricinoleic Acid, Glyceryl Ricinoleate, oleic acid, fluorene, and phenanthrene. Turpentine induced minimal histologic change, and acridine induced marked epidermal hyperplasia. Silver nitrate induced hyperplasia most marked in the hair follicles.

In the tumor promotion experiment (groups of 10 mice; strain and weights not stated), each mouse received a single application of 9,10-dimethyl-1,2-benzanthracene in liquid paraffin. The nine test substances (one per group) were then applied semiweekly for 20 weeks. For all groups of mice tested, no tumors were recorded. Castor oil, Ricinoleic Acid, and Glyceryl Ricinoleate, as well as the other test substances that were evaluated, were not tumor promoters in this study (Shubik 1950).

Ricinoleic Acid

Bull et al. (1988) investigated the effect of Ricinoleic Acid (unoxidized) on DNA synthesis and the induction of ornithine decarboxylase activity. The authors noted that unsaturated fatty acids are readily oxidized to a variety of biologically active derivatives, and have suggested that autoxidation products of polyunsaturated fatty acids may also play a role in the enhancement of tumorigenesis. They noted that this suggestion is based on published results (Bull et al. 1984) indicating that hydroperoxy and hydroxy derivatives of arachidonic acid and linoleic acids, the primary autoxidation products of major dietary unsaturated fatty acids, induce ornithine decarboxylase (ODC) and stimulate DNA synthesis (two components of mitogenesis) in colonic mucosa in vivo.

Male Sprague-Dawley rats (weights = 100 to 125 g) fed the following semisynthetic diet (weight percentages indicated) were starved for 24 h prior to intrarectal (i.r.) instillation of Ricinoleic Acid (vehicle: 5% ethanol in 50 mM $NaHCO_3$, pH 8.0): casein (20%), DL-methionine (0.2%), salt mix (3.6%), vitamin mix (2%), beef fat (5%), dextrose (45.2%), cellulose (5%), and cornstarch (19%). Two experiments were performed using Ricinoleic Acid test concentrations of 5 and 10 mM. Test groups

consisted of three to five rats per dose level. The vehicle control group (three rats per experiment) was dosed with 5% ethanol in 50 mM $NaHCO_3$, and positive control animals received 6 mM deoxycholate. Data were presented as an average of the two experiments. The measurement of DNA synthesis in the colon was based on the incorporation of [^{3}H]dThd 12 h after i.r. instillation of Ricinoleic Acid. ODC activity was determined in colonic mucosa that was harvested 3 h after i.r. instillation of Ricinoleic Acid.

Ricinoleic Acid was inactive as a stimulator of DNA synthesis and inducer of ODC activity in the rat colon. Concentrations up to 10 mM resulted in insignificant increases in [^{3}H]dThd incorporation and did not increase the level of ODC activity above control values (Bull et al. 1988).

Anticancer Activity

Extract of Castor Oil

Xie et al. (1992) evaluated the anticancer/antitumor activity of an extract of castor oil using male Kunming mice (weights = 18 to 22 g) and mice bearing S_{180} ARS ascitic fluid tumor cells. Castor oil was saponified and acidified to obtain a coarse, fatty acid mixture that was recrystallized and refined to produce the active anticancer component. Using Tween-80 (Polysorbate 80), an emulsion containing this component was produced. A polyphase liposome from castor oil extract was also produced. The inhibitory action of the anticancer component on S_{180} tumor cells was evaluated. Ascites was obtained from mice that had been inoculated with S_{180} tumor cells (with well-grown ascites) that had been washed with physiological brine in order to collect the cancer cells. The physiological brine was diluted and injected (2 ml) subcutaneously into the right front axilla of each mouse. On the day after injection of the tumor cells, the mice were randomly divided into groups.

The three dilution control groups (8 to 10 mice per group) were injected with 200, 300, or 400 mg/kg of a physiological brine solution containing an equal load of Tween-80. The four polyphase liposome control groups (eight mice per group) were injected with blank liposomes (doses of 300, 300, 400, and 400 mg/kg, respectively). For dilution control and polyphase liposome control groups, injections were made into the abdominal cavity daily for 7 days, after which the mice were killed. Subcutaneous tumors were removed and weighed in order to calculate the tumor suppression rate (%).

Tumor suppression rates for dilution controls were as follows: 30.6% (200 mg/kg dose group), 37% (300 mg/kg dose group), and 53.1% (400 mg/kg dose group). The following tumor suppression rates were reported for the polyphase liposome control groups: 40.2% (300 mg/kg dose group), 36.4% (the other 300 mg/kg dose group), 55.7% (400 mg/kg dose group), and 58% (the other 400 mg/kg dose group).

In another experiment, the inhibitory action of the anticancer component extracted from castor oil on ARS ascites cancer was evaluated according to a similar procedure. Ascites was obtained from mice inoculated with ARS cancer cells (with well-grown ascites) that had been washed with physiological brine in order to collect the cancer cells. The physiological brine was diluted and injected (0.2 ml) into the abdominal cavity of each mouse. On the day after injection of the cancer cells, each of nine mice was injected with a polyphase liposome that was produced from castor oil extract (dose = 200 mg/kg) daily for 7 days. At the end of the treatment period, the animals were monitored (for survival) for an additional 35 days.

Study results indicate that the castor oil extract had a strong suppressive effect on S_{180}body tumors in mice. Its suppressive effect on ARS ascites cancer was also very strong, with 64% of the ascites tumors in mice being completely cured. Compared to controls, the life extension rate was more than 136% ($p < .0012$) (Xie et al. 1992).

REPRODUCTIVE AND DEVELOPMENTAL TOXICITY

Castor Oil

NTP (1992) (see earlier Subchronic Oral Toxicity section) fed groups of rats and mice diets containing 0.62%, 1.25%, 2.5%, 5.0%, and 10% castor oil, respectively, continuously for 13 weeks. A slight decrease in epididymal weight (6% to 7%) was observed in mid- and high-dose groups of male rats; however, this finding was not dose related. No effects on any other male reproductive end point (testes weight and epididymal sperm motility, density, or testicular spermatid head count) or female reproductive endpoint (estrous cycle length, or time spent in each phase of the cycle) were noted. Microscopically, there were no treatment-related lesions in any organ or tissue. These results indicate that there was little or no evidence of any reproductive toxicity in rats that was associated with castor oil in the diet. For male and female mice, castor oil in the diet had no adverse effects on any male or female reproductive parameter.

Litvinova and Fedorchenko (1994) evaluated the influence of single doses of different plant oils on the estrous cycle and fertility using female Wistar rats (number and weights not stated). Castor oil (0.2 ml, single dose) was injected intramuscularly on the first day after estrus. Study of the estrous cycle was based on the following: the cytologic picture that was obtained from vaginal smears, the phase structure (proestrus, estrus, metaestrus, and diestrus) of the cycle, and the frequency of estrus over a 15-day observation period prior to injection of castor oil (control) and a similar length of time after injection (test). Fertility was studied using intact animals (control) and animals that were treated with castor oil. On day 2, after castor oil injection (i.e., during diestrus), the females were mated with male rats. Control females were also mated with male rats on day 2. The first day of pregnancy was defined by the presence of spermatozoa in vaginal smears. Pregnant females were killed on day 20. The estrous cycle phases were recorded 3 to 4 times during the 15-day observation period.

The injection of castor oil resulted in a reduction in the frequency of estrus by a factor of 1.4 and lengthening of the

interestrous phase by 39.6%. The authors concluded that the latter finding may be indicative of prolongation of luteolytic processes and an inhibition of folliculogenesis.

In female fertility experiments, mating occurred over a period of 3.22 ± 0.12 days. The index of fertility was calculated from the ratio of the number of mated females to the total number of females in the test. The fertility index for intact animals (control) was 100%, and pregnancy occurred in 100% of the cases. A decrease in the pregnancy index was reported for female rats injected intramuscularly with castor oil; however, no reliable difference between test and control values was detected.

Compared to controls, castor oil also caused a reduction in the number of corpora lutea in the ovaries, and this observation was said to have been a consequence of the suppression of folliculogenesis and ovulation. Additionally, the number of implantation sites in uteri from female rats injected with castor oil was decreased when compared to control rats. A decrease in the number of live fetuses in uteri and an increase in pre- and postimplantation deaths was also observed in rats injected with castor oil. The authors concluded that castor oil, injected intramuscularly, suppressed ovarian folliculogenesis and also had antiimplantation and abortive effects (Litvinova and Fedorchenko 1994).

Castor oil served as the vehicle control in a study evaluating the effect of long-term treatment with ICI 182,780 (an antiestrogen) on the rat testis (Oliveira et al. 2001). Two groups of four male, 30-day-old Sprague-Dawley rats served as vehicle controls. In one control group, castor oil (0.2 ml) was injected subcutaneously once per week, and the animals were killed 100 days after the first injection. The second group was dosed according to the same procedure; animals were killed 150 days after the first injection. The experiment was extended to 150 days (group 2 rats), based on results for group 1 ICI-treated and control animals on day 100.

Specifically, Group 1 ICI-treated rat testes were significantly heavier (35% heavier, $p < .05$) than those of the control group, and a significant increase in diameter and luminal areas of the semineferous tubules was observed. Additionally, in group 1 ICI-treated and control rats, spermatogenesis appeared normal and there was no evidence of degeneration or sloughing of germ cells.

On day 150, a 33% decrease (compared to day 100 results for ICI-treated rats; $p = .065$) in mean testicular weight was noted in ICI-treated rats. Seventy-five percent of the testes weighed less than the mean control value of 1.71 g. Atrophy of the semineferous tubules was observed in all but one of the testes of ICI-treated rats. Additionally, the rete testis and efferent ductule lumens in all ICI-treated rats were greatly dilated, compared to those of controls.

During the treatment period, mating studies were performed to compare the fertility of ICI-treated and control rats on days 45, 73, 100, 125, and 150. Males were housed individually with two adult females for a 15-day period. Through 100 days of treatment, there was no difference in male reproductive performance between ICI-treated and control rats. However, after day 100, 100% fertility was maintained in control males, but, by day 150, fertility had decreased to 25% in ICI-treated males (Oliveira et al. 2001).

Extract of Ricinus communis Seeds

The following study is included because castor oil may be extracted from the bean of the tropical plant, *Ricinus communis* by a method that involves the use of a solvent.

Okwuasaba et al. (1991) evaluated anticonceptive and estrogenic effects of a methanol extract of *Ricinus communis* var.*minor* seeds (ether-soluble fraction) using the following animals (groups of 10): adult albino Wistar rats (weights = 160 to 210 g), young albino Wistar rats (weights = 40 to 60 g), immature Swiss albino female mice (21 to 23 days old; weights = 10 to 13 g), and adult female New Zealand white rabbits (2.5 to 3.0 kg).

In the antifertility experiment, adult female rats and rabbits with regular estrus cycles were used. The females were mated overnight (3:1 female/male ratio), and the day of mating was considered day 0. Castor bean extract was injected subcutaneously (s.c. dose of 0.6 or 1.2 g/kg) into female rats once daily for two days, and mating occurred on the third day. Female rabbits were injected intramuscularly with Castor bean extract (dose = 200 mg/kg). Control animals were injected s.c. with corn oil for two consecutive days. A laparotomy was performed to confirm that implantation had occurred.

Castor bean extract (0.6 or 1.2 g/kg dose) prevented nidation in rats, and none of the treated rats delivered pups at term. The results for rabbits (200 mg/kg dose) paralleled those for rats. Nidation was prevented, and the extract protected the rabbits against pregnancy for over three gestation periods (i.e., 110 to 120 days). The normal gestation period for rabbits is 28 to 30 days. Effects induced by Castor bean extract were reversible in rats and rabbits. No fetal abnormalities were observed.

In the study to determine estrogenic activity, the following endpoints were used to estimate the estrogenicity of the Castor bean extract: uterine weight ratio, degree of vaginal cornification, and quantal vaginal opening. In one experiment, groups of 10 Swiss albino female mice (21 to 23 days old; weights = 10 to 13 g) were injected s.c. with 1.0 or 4.0 mg/kg Castor bean extract (in corn oil, 0.2 ml) daily for 4 days. In a similar experiment, bilateral ovariectomy was performed on young, immature rats (groups of five), and the wound was sutured. After a 15-day recovery period, the groups of rats were injected s.c. with 0.6 or 1.2 g/kg Castor bean extract (in corn oil, 0.2 ml) daily for 4 days. In both experiments, suitable controls (injected s.c. with corn oil only) were maintained. Additionally, estradiol-17β (reference hormone; dose = 10 μg/kg) was administered to rats (group of 10) and mice (group of 10). Mice and rats were necropsied 24 h after the last injection. Body and uterine wet weights were recorded.

Results for bilaterally ovariectomized rats indicated that doses of 0.6 and 1.2 g/kg induced increases in relative uterine weight of 116.7 ± 6.5 ($p < .01$) and 211.6 ± 8.2 mg/100 g body weight ($p < .001$), respectively, compared to a control value of 83.3 mg/100 g body weight. Compared to rats, Castor bean extract (1.0 and 4.0 mg/kg) induced a qualitatively similar effect in immature mice. In rats and mice, the effect on relative uterine weight appeared to have been dose-dependent. Estradiol-17β also increased uterine weight in rats and mice. The relative uterine weights (mg/100 g body weight) for rats and mice dosed with estradiol-17β were reported as follows: 309.2 ± 11.4 (rats) and 517.5 ± 12.3 (mice).

Cornification of the vaginal epithelium and quantal opening of the vagina were noted in immature mice and bilaterally ovariectomized rats (both dose groups for each). A dose-related decrease in the number of leukocytes and increases in cornified and epithelial cell counts were also reported for both species. No significant changes in body weight were observed.

The authors concluded that the ether-soluble portion of the methanol extract of *Ricinus communis* var. *minor* seeds possesses anti-implantation, anticonceptive, and estrogenic activity in rats and mice when administered subcutaneously (Okwuasaba et al. 1991).

CLINICAL ASSESSMENT OF SAFETY

Metabolism and Excretion

Castor Oil

Watson et al. (1963) studied the absorption and excretion of castor oil in five subjects (ages and weights not stated). The composition of the castor oil was as follows: palmitic acid (1%), palmitoleic acid (0.1%), stearic acid (1%), oleic acid (3.2%), linoleic acid (4.7%), and Ricinoleic Acid (90%). The castor oil (of medicinal purity) contained a trace amount of oil labelled with ^{131}I, and the doses administered ranged from 4 to 60 g (approximately 6 μCi of radioactivity per dose). Castor oil was administered to the five hypertensive patients on the day before a scheduled intravenous pyelogram; thereafter, only a light meal of coffee and toast was allowed. Stool collections were made during the first 24 h after dosing and during the subsequent 72 h. Urine was collected in 24-h samples. Small doses of oil were administered to the three normal volunteers, and free diet was allowed. Stools were collected at 2 and 3 days after dosing.

Fecal recovery of ^{131}I (%) ranged from 11.4% (for 10 g dose of castor oil) to 86.0% (for 44.4 g dose of castor oil). The authors concluded that castor oil can be absorbed by human subjects, that absorption is inversely related to the administered dose, and, at small doses (4 g), that absorption is virtually complete (Watson et al. 1963).

Hagenfeldt et al. (1986) administered castor oil (10 to 15 ml) orally to three healthy subjects. Urine was collected between 2 and 8 h post dosing. The following three epoxydicarboxylic acids were excreted in the urine: 3,6-epoxyoctanedioic acid; 3,6-epoxydecanedioic acid; and 3,6-epoxydodecanedioic acid. These three Ricinoleic Acid metabolites were also detected in the urine of rats as described earlier.

Induction of Labor

Castor Oil

Davis (1984) investigated the use of castor oil to stimulate labor using 196 patients with premature rupture of membranes (PROM), at least 4 h in duration, who were between 37 and 42 weeks of gestation. Patients were identified by reviewing charts of all patients who were admitted at an out-of-hospital birthing center from 1976 through 1981. Of the 196 patients, 107 (mean age = 28.6 years) were dosed orally with castor oil (2 oz) and 89 (mean age = 27.6 years) were not. Castor oil was administered only to PROM patients who had a latency period of at least 4 h. All patients were observed for labor onset for 24 h after the onset of PROM. If labor did not occur at this time, the patients were transferred to a hospital for oxytocin stimulation. The longest interval between rupture of the membranes and delivery was 48 h.

Of the 107 patients dosed with castor oil, 80 (75%) had labor onset. Spontaneous labor occurred in 52 (58%) of the 89 control patients. This difference between patients dosed with castor oil and controls was statistically significant ($p < .05$). The interval between castor oil administration and the onset of labor ranged from 1 to 13 h (mean = 4 h).

Labor outcomes were also evaluated for type of delivery, incidence of oxytocin stimulation, and infant well-being. The need for cesarean sections was nearly three times greater in the control group (15.7% incidence) than in patients dosed with castor oil (5.6% incidence). This difference was found to be statistically significant ($p < .01$). Additionally, the group dosed with castor oil did not require oxytocin stimulation as frequently (36% incidence) when compared to the control group (43% incidence), and the difference was not statistically significant.

Infant outcomes in the two groups were studied by observing Apgar scores at 1 and 5 min, and looking for any evidence of meconium staining. Two infants with Apgar scores of <7 were reported for both groups. At the time of membrane rupture, the presence of meconium was very low in both groups. Meconium staining was not observed after dosing with castor oil. It is also important to note that there were no maternal deaths and no significant maternal morbidity. The authors concluded that castor oil can be used safely and effectively to stimulate labor (Davis 1984).

In a study by Mitri et al. (1987), the amniotic fluid was examined at the time of rupture of membranes in 478 of the 498 women in labor. The amniotic fluid was meconium-stained in 174 subjects (mean age = 28.5 ± 0.65 years) and clear in the remaining 304 subjects (mean age = 25.9 ± 0.37 years). Cesarean section, low Apgar score, and the recent ingestion of castor oil were significantly more common in subjects with meconium-stained amniotic fluid. The authors concluded that

the passage of fetal meconium was predictive of poor labor outcome in terms of Apgar scores and the need for cesarean section. It was recommended that women who take castor oil in late pregnancy should be identified as a high-risk group for the passage of fetal meconium.

In a case report by Steingrub et al. (1988), a 33-year-old pregnant female (at week 40 of gestation) ingested castor oil to induce labor. Within 60 min of ingestion, cardiopulmonary arrest occurred and was reportedly due to amniotic fluid embolism.

Garry et al. (2000) evaluated the use of castor oil to induce labor in 52 pregnant women (mean age = 24.8 ± 6.7 years) at Saint Mary's Hospital in Brooklyn, New York. The untreated control group consisted of 48 pregnant women (mean age = 24.4 ± 4.9 years). The two groups of women did not differ in maternal age, parity, or gestational age. Castor oil was administered as a 60-ml dose in orange or apple juice, and its use was deemed successful only if active labor began within 24 h. Labor was defined as one or more contractions every 5 min, with cervical dilatation of 4 cm or more. Active labor was induced in 30 (57.7%) of the 52 women dosed with castor oil, compared to 2 of the 48 women in the control group ($p < .001$). The cesarean section rate for women dosed with castor oil was 19.2% (10 of 52 women), compared to 8.3% (4 of 48 controls) in the untreated control group. No relationship between dosing with castor oil, birth weight, and mode of delivery ($p = .66$) was found. Additionally, no adverse outcomes for the mother or fetus were identified. The authors concluded that women dosed with castor oil had an increased likelihood of initiation of labor within 24 h, compared to women who were not dosed with castor oil.

Ocular Irritation

Castor Oil

In a study by Secchi et al. (1990), a double-masked clinical trial was performed using a group of nine patients (ages not stated) with vernal keratoconjunctivitis. One eye was treated with 2% cyclosporine in castor oil four times daily for 15 days. The other eye was treated with castor oil solution only according to the same procedure. Mild and transient discomfort and minor epithelial changes were observed in both eyes.

In another clinical study (Gluud et al. 1981), vital staining was used to investigate the eyes of 100 patients (ages not stated) following the instillation of castor oil and the eyes of 50 patients following the instillation of 0.9% saline. One drop (0.01 ml) of sterile fluorescein–rose bengal (RB) dye mixture was instilled into the lower conjunctival sac and the ocular mucosa was examined using a slit lamp. White light was used to examine RB-stained spots and cobalt blue light was used to examine fluorescein-stained spots. The ocular mucosa was divided into five regions, and the number of stained spots in each region was counted/estimated and graded. The authors concluded that castor oil affected the cornea and conjunctiva, i.e, degeneration and death of epithelial cells (stained with RB) and continuity breaks in the epithelium (stained with fluorescein).

Skin Irritation/Dermal Toxicity

Castor Oil

Motoyoshi et al. (1979) evaluated the skin irritation potential of undiluted castor oil using 50 adult male volunteers (ages not stated). Individuals with known allergic reactions were excluded. Patches containing 0.05 g of the test material were applied to skin of the back and remained for 48 h. At the end of the application period, test sites were swabbed with dry gauze to remove any residual test material and reactions were scored 30 minutes later according to the following scale: − (negative reading) to + + ++ (bullous reaction). Castor oil was mildly irritating to the skin of human subjects.

In a study by Meyer et al. (1976), undiluted castor oil was applied to test fields delineated on the right thigh of each of three male subjects (22 to 31 years old). The control field was located on the opposite side. According to the test procedure, castor oil was rubbed onto the skin gently (<30 s) on 10 sucessive days. The control field was massaged only. A tissue section (1 × 1 cm) was excised from the center of each test field and five to eight sections (7 μm in thickness) per test area were subjected to microscopic examination. Macroscopic skin changes were not observed in either of the three subjects.

At microscopic examination, clear hyperchromasia and an increase in the number of cells were observed in the basal cell layer. Slight widening of the granular cell layer was also noted (Meyer et al. 1976).

Hydrogenated Castor Oil

In a study by de Groot (1994), members of the Dutch Contact Dermatitis Group and nonmember dermatologists patch tested 1 to 20 patients (ages not stated) either suffering from or suspected of suffering from contact allergy to cosmetic products. Details regarding the patch test procedure were not included. Patch test results indicated no reactions to 30% Hydrogenated Castor Oil in petrolatum.

Ethyl Ricinoleate

The skin irritation potential of 20% Ethyl Ricinoleate in petrolatum was evaluated using 32 healthy male volunteers (ages not stated) in a 48-h closed patch test. Skin irritation was not observed (RIFM 2000).

Allergenicity

Predictive Tests

Ethyl Ricinoleate

The skin sensitization potential of 20% Ethyl Ricinoleate in petrolatum was evaluated using 32 healthy male volunteers in the maximization test. The test substance was applied (under occlusion) to the same site on the forearms of each subject for five alternate-day 48-h periods. Each test site was pretreated with 5% aqueous sodium lauryl sulfate (under occlusion) for 24 h. At the end of a 10- to 14-day nontreatment period, challenge

patches were applied (under occlusion) to new test sites for 48 h. Challenge applications were preceded by 30-min applications of 2% aqueous sodium lauryl sulfate (under occlusion) on the left side. On the right side, the challenge application was not preceeded by treatment with sodium lauryl sulfate. A fifth site (challenge with petrolatum) served as the control. Reactions classified as either significantly irritating or allergic were not observed (RIFM 2000).

Provocative Tests

Castor Oil

Fujimoto et al. (1997) conducted a study involving 332 patients (25 males, 307 females; ages ≈20 to 70) suspected of having cosmetic contact dermatitis. The subjects were patch-tested with numerous cosmetic products and cosmetic ingredients. Patch tests (Finn chambers) were applied to the back for 48 h, and the test sites were examined at 30 minutes after patch removal, 1 day later, and 4 to 5 days after patch removal. None of the 49 patients patch-tested with castor oil had a positive reaction.

In a study by Hino et al. (2000), 346 patients (31 males, 315 females; ages ≈ 20 to 70 years) suspected of having cosmetic dermatitis were patch tested with various cosmetic products and cosmetic ingredients. Patch tests (Finn chambers) were applied to the back for 48 h. Test sites were examined according to the schedule in the preceding study. Of the 76 patients patch-tested with castor oil, one had a positive reaction. This reaction was observed only during the second reading (i.e., the day after the 30-min reading).

Hydrogenated Castor Oil

Lehrer et al. (1980) evaluated the sensitivity of three human subjects (with occupational hypersensitivity to castor allergens) to castor bean extract and four Castor Wax extracts (solvent = SLS, PBS, urea-NaCl, or Triton X-100). Seven normal laboratory personnel served as controls. The test substances were applied to the skin using both patch and prick tests. Only the castor bean extract and the SLS extract of Castor Wax were evaluated in the prick test, and the four Castor Wax extracts were evaluated in the patch test. In the prick test (to detect IgE-mediated hypersensitivity), a drop of allergen extract was applied to the skin. Following application to the skin, the underlying superficial layer of the skin was pricked with a 25-gauge needle. Reactions were scored 15 min after application of the allergen. A positive prick test reaction was defined as a wheal and flare reaction with edema >5 mm. Castor bean extract was tested at concentrations up to 1000 μg/ml and, SLS Castor Wax extract, at concentrations up to 22,000 μg/ml. Results from the prick test indicated no immediate skin reactivity to castor bean extract or the SLS Castor Wax extract.

In patch tests (to detect cell-mediated hypersensitivity), a semiocclusive patch containing either of the four Castor Wax extracts (concentration not stated) or paraffin (control) was applied to the skin for 24 and 48 h. Reactions were scored at 24 and 48 h post removal.

The three patients were highly sensitive to castor allergen in the prick test (positive skin reaction to 1.0 or 0.1 μg/ml castor bean extract). The patients also had a positive skin reaction to the SLS Castor Wax extract, where the level of sensitivity was 100- to 100,000-fold less than that induced by castor bean extract. None of the seven normal subjects had a delayed reaction to any of the four Castor Wax extracts that were tested. Prick test results (normal subjects) indicated no immediate skin reactivity to castor bean extract or the SLS Castor Wax extract.

Because immediate hypersensitivity to Castor Wax was observed in the three patients who were sensitive to castor bean, additional tests (prick tests) were performed to determine whether Castor Wax solubilized in different organic solvents or an underarm deodorant stick containing Castor Wax would induce irritation and elicit a positive immediate hypersensitivity reaction in these three patients. Positive skin reactions to Castor Wax (in different solvents or in a deodorant stick) were not detected.

The four Castor Wax extracts and paraffin (control) were evaluated in patch tests for cell-mediated hypersensitivity. None of the three patients had a delayed reaction to any of the four Castor Wax extracts (test concentration not stated).

The positive skin reactions (immediate skin reactivity) in sensitized individuals, together with the positive PCA reactions in mice and positive direct RAST for reaginic castor allergens (summarized earlier in this report) that were induced by SLS Castor Wax extract, indicate that allergens are present in Castor Wax at low concentrations.

A direct RAS study was conducted to determine whether the SLS extract of Castor Wax contains specific antigens or nonspecifically inhibits the RAS assay. Discs were coated with castor bean extract or the SLS Castor Wax extract and a direct RAS assay was performed using serum from one patient who was sensitive to castor allergens, a patient who was sensitive to peanuts, or pooled human serum (control). The patient sensitive to castor allergens had a significant RAS test ratio of 18.87 to castor bean allergen and a significant RAS test ratio of 8.04 to SLS Castor Wax extract. The patient sensitive to peanuts did not have a significant RAS test to either castor bean extract or the Castor Wax extract. The results from this direct RAS study indicate that SLS Castor Wax extract does contain Castor bean allergens, although the SLS Castor Wax extract can nonspecifically inhibit the RAS assay (Lehrer et al. 1980).

Ricinoleic Acid

In a retrospective study by Lim and Goh (2000), 202 consecutive patients (182 females, 20 males) with eczematous cheilitis who attended a contact and occupational dermatoses clinic in Singapore between January of 1996 and December of 1999 were patch-tested. Mean ages for female and male patients were 31.1 and 28.8 years, respectively. Patch tests were conducted according to International Contact Dermatitis Research Group (ICDRG) recommendations. Twenty-nine patients (2 males,

27 females) had positive reactions to Ricinoleic Acid. Of the 29 reactions, 22 were considered relevant positives.

Allergenicity/Photoallergenicity

Castor Oil

In a study by Hashimoto et al. (1990), 103 patients (ages not stated) suspected of having cosmetic-related contact dermatitis were patch tested with castor oil (as is). Both patch tests and photopatch tests were conducted according to ICDRG basic standards. The patch test units consisted of a combined use of both Finn chambers and Scanpor tape. One patient had a positive patch test reaction to castor oil. No positive photopatch test reactions were reported.

Case Reports

Case reports involving Castor Oil and Ricinoleic Acid esters are summarized in Table 6.

Occupational Exposure

Göhte et al. (1983) conducted a study involving 28 persons (14 men, 14 women; age range = 17 to 65 years) who were employed by a company involved with importing, preparing, and distributing plant products that are used in spices and as ingredients of health foods. The period of employment varied from two months to 20 years, and 25 of the 28 were smokers. Thirteen of the 28 developed the following work-related symptoms: rhinitis, conjunctivitis, asthma, itch, or urticaria. Blood samples from 25 subjects were subjected to IgE analysis using Phadebas IgE PRIST (paper radioimmunosorbent test). Phadebas RAST (radioallergosorbent test) was performed on 25 persons against antigens from an extract of castor oil bean. Fifteen subjects were prick-tested and 14 were patch-tested with an extract of castor oil bean. Of the three tests, only the RAST test yielded positive results; 1 of 25 subjects had an allergic reaction to an extract of castor oil bean. None of the subjects without work-related symptoms had positive reactions.

SUMMARY

Ricinus Communis (Castor) Seed Oil, Ricinoleic Acid, Glyceryl Ricinoleate, Glyceryl Ricinoleate SE, Potassium Ricinoleate, Sodium Ricinoleate, Zinc Ricinoleate, Cetyl Ricinoleate, Ethyl Ricinoleate, Glycol Ricinoleate, Isopropyl Ricinoleate, Methyl Ricinoleate, and Octyldodecyl Ricinoleate function, in cosmetics, as skin-conditioning agents, emulsion stabilizers, and surfactants. Ricinus Communis (Castor) Seed Oil also functions as a fragrance and Hydrogenated Castor Oil also functions as a viscosity-increasing agent—nonaqueous. In the case of Zinc Ricinoleate, functions include deodorant agent, opacifying agent, and anticaking agent.

Ricinoleic Acid accounts for 87% to 90% of the fatty acyl groups of Ricinus Communis (Castor) Seed Oil and the following other fatty acids comprise the remaining fatty acyl groups: oleic acid (2% to 7%), linoleic acid (3% to 5%), palmitic acid (1% to 2%), stearic acid (1%), dihydrostearic acid (1%), and trace amounts of other fatty acyl groups.

Hydrogenated Castor Oil consists primarily of glyceryltrihydroxystearate. The National Formulary has the following requirements for Hydrogenated Castor Oil: free fatty acids (free fatty acids in 20 g require for neutralization not more than 11.0 ml of 0.1 N sodium hydroxide), heavy metals (0.001%), hydroxyl value (between 154 and 162), iodine value (not more than 5), and saponification value (between 176 and 182).

Ricinus Communis (Castor) Seed Oil is produced by cold pressing of the seeds of *Ricinus communis* and subsequent clarification of the oil by heat. Castor Oil does not contain ricin because ricin does not partition into the oil, due to its water-solubility. Hydrogenated Castor Oil is the end product of controlled hydrogenation of Ricinus Communis (Castor) Seed Oil. One of the simplest methods for obtaining Ricinoleic Acid is the hydrolysis of castor oil.

Ricinus Communis (Castor) Seed Oil and Glyceryl Ricinoleate absorb UV light, with a maximum absorbance at 270 nm.

Reportedly, Ricinus Communis (Castor) Seed Oil and Hydrogenated Castor Oil were used in 769 and 202 cosmetic products, respectively, in 2002. The following use frequencies were reported for Ricinoleic Acid and its salts and esters: Glyceryl Ricinoleate (16), Ricinoleic Acid (6), Sodium Ricinoleate (12), Zinc Ricinoleate (3), and Cetyl Ricinoleate (55).

The highest reported use concentration for Ricinus Communis (Castor) Seed Oil is associated with lipsticks (81%). Other maximum ingredient use concentrations are: Hydrogenated Castor Oil (up to 39%), Glyceryl Ricinoleate (up to 12%), Zinc Ricinoleate (up to 2%), Cetyl Ricinoleate (up to 10%), and Octyldodecyl Ricinoleate (up to 5%).

Potassium Ricinoleate, Ethyl Ricinoleate, Glycol Ricinoleate, Isopropyl Ricinoleate, and Methyl Ricinoleate are not currently reported to FDA as in use, nor are there industry survey data indicating a current use concentration.

Castor oil is classified by FDA as generally recognized as safe and effective for use as a stimulant laxative. An acceptable daily intake (for man) of 0 to 0.7 mg/kg body weight has been established by FAO/WHO for castor oil.

Castor oil is a triglyceride that is hydrolyzed in the small intestine by pancreatic enzymes, leading to the release of glycerol and Ricinoleic Acid. Ricinoleic Acid was rapidly metabolized in a study of adult rats fed a diet containing 48.4% castor oil for 4 to 6 weeks. These animals did not have significant amounts of Ricinoleic Acid in phospholipids of the small intestine, liver, and skeletal muscle, nor in glycerides of the liver. A close correlation between the dose administered in the diet and the percentage of Ricinoleic Acid in the feces (greater absorption of the smaller dose) was noted in rats. Castor oil was readily absorbed and metabolized in two groups of male rats that were fed 10% castor oil in the diet (cholesterol-enriched and cholesterol-free, respectively) for 20 days. Fecal recovery of Ricinoleic Acid was

TABLE 6
Case reports with patch test results for Castor Oil and Ricinoleic Acid esters and salts

Subjects	Findings/patch test results	References
22-year-old female (with cheilitis)	Cheilitis worsened after application of various lipsticks and lip creams. Product concentrations of Castor Oil ranged from 10% to 67%. ++ patch test reactions to Castor Oil and each lipstick or lip cream that contained Castor Oil. Also, ++ patch test reactions to Ricinoleic Acid (as is), 30% Ricinoleic Acid in petrolatum, and purified Castor Oil (as is). No reactions to products that did not contain Castor Oil.	Sai 1983
23-year-old female (history of facial eczema) with acute dermatitis of the face	++ reactions to Castor Oil and make-up remover containing Castor Oil.	Brandle et al. 1983
29-year-old female (history of deodorant dermatitis)	Acute edematous dermatitis on the lips and perioral area, which spread to entire face and neck, after using a lipstick product containing Castor Oil. ++ patch test (Finn chambers, International Contact Dermatitis Research Group [ICDRG] recommendations) results for Castor Oil.	Andersen and Nielsen 1984
24-year-old female	Swelling of lips over 10-month period. Negative patch test results for Castor Oil and five brands of lipstick containing Castor Oil.	Yoshikawa et al. 1986
31-year-old female (no history of dermatitis)	Developed itchy rashes in the axillae, which spread over the entire body. Positive patch test reaction (++, at 38 and 96 h) to a deodorant that was used. Repeated open application test results for Castor Oil and a Zinc Ricinoleate mixture (Zinc Ricinoleate (88%), triethanolamine, zinc resinate, isostearic acid, dipropylene glycol, sodium lactate, and abietic acid) were positive.	Dooms-Goosens et al. 1987
45-year-old female (history of intolerance to cosmetics and adhesives)	Presented with an itchy dermatitis on both axillae, and red papules scattered over face and trunk. Patch test results indicated a red erythematous response to a deodorant that was used. Patch test results for Castor Oil were negative at 72 h and +/? at 96 h. Repeated open application test results for Castor Oil were negative after 7 days; ++ patch test results for 20% Glyceryl Ricinoleate (in petrolatum) at 72 and 96 h. Also, ++ reaction to Zinc Ricnoleate mixture (defined in preceding case report) at 96 h. This mixture also produced an erythematous test response in 1 of 10 control subjects.	Dooms-Goosens et al. 1987
40-year-old chemist (history of eczema due to chemical exposure on the job)	Developed eczema on the knee 10 days after joint surgery. Positive patch test results for surgical adhesive containing Castor Oil. Negative patch test results for Castor Oil.	Jost et al. 1989
20-year-old female	Allergic cheilitis (itching, swelling, and redness of the lips) after using a total of 16 lipstick products. Positive patch test results for Castor Oil and each product.	Fisher 1991
17-year-old female (with lip dermatitis)	Positive patch test reactions to Castor Oil and 3 lipsticks.	Fisher 1991

TABLE 6
Case reports with patch test results for Castor Oil and Ricinoleic Acid esters and salts *(Continued)*

Subjects	Findings/patch test results	References
17-year-old female (allergic to metals)	Developed itchy erythematovesicular reaction on right hand after using a liquid wart remover containing flexible collodion (contains 1.67% Castor Oil). + + + patch test reaction to wart remover. ++ patch test reactions to Castor Oil and 20% Castor Oil in petrolatum. Negative patch test reactions to Castor Oil and 20% Castor Oil in petrolatum in 15 control subjects.	Lodi et al. 1992
51-year-old female	Developed two roundish, erythematous vesicular plaques in left gluteal and right anterolateral neck regions (application sites) after 20 days of treatment with an anti-wart solution (flexible collodion, common component of wart removers, contains Castor Oil). ++ patch test reactions to Castor Oil 20% Castor Oil in petrolatum, and the wart remover.	Tabar et al. 1993
65-year-old male (history of rash following wood cutting in orchard)	Rash on face, neck, and hands. Same rash appeared 3 years earlier, following wood cutting. Patch test results for Castor Oil (100%) were negative.	Manzur et al. 1995
30-year-old female (history of mild atopic eczema)	Hand dermatitis after exposure to cream containing Castor Oil and zinc. + + + patch test reaction to Castor Oil. Negative results for 10 control subjects.	Wakelin et al. 1996
29-year-old female (nonatopic)	Sore, cracked lips, followed by vesicular facial edema, after use of the same lipstick product for several years. ++ patch test (Finn chambers, ICDRG criteria) results for Castor Oil.	Tan et al. 1997
24-year-old female (history of hay fever and dry lips)	Pruritic papules, swelling, and pigmentation on and around lips after applying lipstick. Negative patch test results for Castor Oil (as is).	Inoue et al. 1998
20-year-old army recruit	Rashes developed after applying a camouflage stick containing 47.1% Castor Oil. A mildly pruritic vesicular eruption appeared on the neck, chin, face, and hands; swelling of eyelid also observed. Symptoms lasted for 3 or 4 days.	Goon et al. 1999
43-year-old female (history of sinus aspergillosis and facial edema after oral dosing with tixocortol pivalate)	Recurrent facial dermatitis and cheilitis after use of a facial moisturizer containing 8% Castor Oil. +/+ patch test (Finn chambers) reactions to the moisturizer, Castor Oil, 15% Castor Oil in petrolatum, and 10% Castor Oil in petrolatum. + to ++ reactions to several lipstick and lip balm products containing Castor Oil.	Le Coz and Ball 2000
51-year-old female	Pruritic erythema in both axillae after use of a deodorant containing 76% Zinc Ricinoleate. One week later, developed acute contact dermatitis of the lips after using perfumed lipstick containing Glyceryl Ricinoleate. Lipstick was previously tolerated. + patch test reaction to deodorant and lipstick. + patch test reaction to 30% Glyceryl Ricinoleate in petrolatum. Re-patch testing yielded the following results: 76% Zinc Ricinoleate in petrolatum (?+ reaction on day 3) and no reactions to 30%, 15%, or 1% Zinc Ricinoleate in petrolatum 20% Glyceryl Ricinoleate in petrolatum (+ reaction on day 3), 30% Glyceryl Ricinoleate in petrolatum (+ reaction on day 3), and Castor Oil, as is (no reaction).	Magerl et al. 2001

(Continued on next page)

TABLE 6
Case reports with patch test results for Castor Oil and Ricinoleic Acid esters and salts *(Continued)*

Subjects	Findings/patch test results	References
58-year-old female (history of allergy to adhesive tapes)	After knee surgery, acute contact dermatitis around surgical incision rapidly became blistered and spread to right lower limb. Patch test results for Castor Oil (1% and 10% in petrolatum) were negative.	Reichert-Pénétrat et al. 2001
50-year-old female (history of allergy to adhesive tapes)	After hip surgery, acute dermatitis around surgical incision spread to lower right limb. Patch test results for Castor Oil (1% and 10% in petrolatum) were negative.	Reichert-Pénétrat et al. 2001
30-year-old female (with pigmented contact cheilitis)	Presented with scaly erythema with associated diffuse hyperpigmentation and itchy swelling for past one year after using various lipsticks. ++ patch test reactions to Ricinoleic Acid at day 3.	Leow et al. 2003

approximately 0.5% of the total ingested. 3,6-epoxyoctanedioic acid, 3,6-epoxydecanedioic acid, and 3,6-epoxydodecanedioic acid were excreted by rats given castor oil intragastrically. These acids were not detected in the urine prior to dosing with castor oil.

Following the oral administration of Hydrogenated Castor Oil to rats at dietary concentrations of 0.865% and 8.65%, the deposition of hydroxystearic acid in the abdominal fat and other body lipids was reported. In all cases, hydroxystearic acid was accompanied by hydroxypalmitic acid, hydroxymyristic acid, and hydroxylauric acid (hydroxystearic acid metabolites). Ricinoleic Acid or Methyl Ricinoleate administered by gastric intubation to male rats resulted in peak absorption of Ricinoleic Acid within 30 min post dosing in the following fractions: triglyceride, diglyceride, monoglyceride, and free fatty acid.

Castor oil (labeled with ^{131}I) was administered to five hypertensive patients on the day before a scheduled intravenous pyelogram, with results demonstrating that castor oil can be absorbed by human subjects, that absorption is inversely related to the administered dose, and, at small doses (4 g), that absorption is virtually complete.

Following oral administration of castor oil (10 to 15 ml) to three healthy subjects, the following three epoxydicarboxylic acids were excreted in the urine: 3,6-epoxyoctanedioic acid; 3,6-epoxydecanedioic acid; and 3,6-epoxydodecanedioic acid.

Castor oil caused a marked decrease in the transepidermal water loss rate from full-thickness, cartilage-stripped skin from the ventral ear of the male Syrian golden hamster. In another study, Ricinoleic Acid enhanced the in vitro transdermal permeation of 5-fluorouracil in hairless mouse skin.

Oral dosing with Sodium Ricinoleate, Ricinoleic Acid, or castor oil had both stimulatory and inhibitory effects on smooth muscle (canine small bowel) contraction in vivo. Sodium Ricinoleate, but not Methyl Ricinoleate, depressed the smooth muscle twitch response in a guinea pig ileum preparation at concentrations ranging from 1.25×10^{-5} to 4×10^{-4} M.

Unlike prostaglandin E_1 (chemically similar to Ricinoleic Acid), Ricinoleic Acid did not induce dose-dependent stimulation of adenylate cyclase in villus cell (from hamster small intestine) homogenates. Compared to saline-treated controls, castor oil significantly reduced the infiltration of leukocytes and the prostaglandin E_2 (inflammatory mediator) content of the rat aqueous humor and iris-ciliary body.

Sodium Ricinoleate significantly inhibited (considered modest; $p < .05$) cytokine-induced endothelial adhesion molecule expression when incubated with human saphenous vein endothelial cells. In a similar experiment, Ricinoleic Acid significantly decreased IL-4-induced VACM-1 expression (50% inhibitory concentration between 10 and 100 μM). Neither cell number nor viability was increased in this concentration range. Ricinoleic Acid did not inhibit TNFα-induced vascular cell adhesion molecule-1 expression to any significant extent. Castor oil in the diet induced a significant increase in prostaglandin E_2 concentrations in tissues of the intestinal mucosa, placenta, amnion, and amniotic cells in groups of pregnant Wistar rats.

Castor oil did not have antimicrobial activity in the *Staphylococcus aureus*, *Escherichia coli*, or *Candida albicans* bacterial strains. There was no correlation between Sodium Ricinoleate antibacterial activity against cells in suspension and antiplaque activity.

Following the oral administration of castor oil (2 ml), macroscopic damage, characterized mainly by vasocongestion, was observed throughout the duodenum and jejunum in rats. After male albino rats were dosed intraperitoneally with 100 mg Ricinoleic Acid, all animals were dead within 24 h. The acute oral LD_{50} for Ethyl Ricinoleate exceeded 5.0 g/kg in rats. A single oral dose (2.5 ml/kg) of castor oil caused generalized hyperemia of the intestinal mucosa up to 72 h post dosing in most of 12 ponies. An acute oral LD_{50} of >10 g/kg was reported for Hydrogenated Castor Oil in a study involving rats. The acute dermal LD_{50} for Ethyl Ricinoleate exceeded 5.0 g/kg in rabbits. One of the rabbits died. The skin reactions observed included

moderate erythema in six rabbits, moderate edema in five rabbits, and slight edema in one rabbit.

In an acute intravenous toxicity study in which eight mongrel dogs were dosed with castor oil (single bolus dose = 0.1 ml/kg), none of the animals died. Erythema was the only clinical sign that was reported. An increase in blood histamine occurred in one animal.

The short-term subcutaneous administration of a vehicle consisting of castor oil and benzyl benzoate to groups of five C_{57}Bl/6J mice caused morphological alterations suggestive of stress-induced changes. Using electron microscopy, the changes observed in the adrenal cortex included disruption of parenchymal mitochondria in the zona fasciculata, increase in the size of some of the mitochondria, loss of the vesicular orientation of the inner mitochondrial membrane, and diminution of the mitochondrial matrix. Mitochondrial changes were also observed in the zona reticularis and zona fasiculata.

Intragastric administration of Ricinoleic Acid (single 0.1 ml dose) to CD-1 specific pathogen–free mice resulted in the erosion of duodenal villi. In another study, perfusion of the rabbit colon with Sodium Ricinoleate solution (5.0, 7.5, and 10 mM) led to mucosal damage (i.e., the loss of epithelial cells). A dose-response relationship for mucosal damage was noted.

In an NTP subchronic oral toxicity study, the administration of diets containing up to 10% castor oil to groups of male and female F344/N rats continuously for 13 weeks was not associated with any toxicities.

The subchronic oral toxicity of Hydrogenated Castor Oil (dietary concentrations up to 17.3%) was evaluated in female rats in a preliminary feeding trial. A reduced growth rate in rats fed 8.65% and 17.3% Hydrogenated Castor Oil, respectively, was the only abnormality that was observed. Based on organ weight determinations and the results of hematological and microscopic examinations, no adverse effects were observed.

In a 16-week study, groups of 15 male albino rats were fed diets containing 0.865% or 8.65% Hydrogenated Castor Oil. Except for the reduced growth rate in rats fed 8.65% Hydrogenated Castor Oil, no adverse effects were observed.

The instillation of undiluted castor oil (0.5 ml) into the rabbit eye resulted in an ocular injury score of 1 (maximum score possible = 20). In contrast, undiluted castor oil induced slight congestion of the iris and conjunctiva in the rabbit eye in another study, but the instillation of castor oil (10 drops daily for 3 weeks) into the eyes of ten rabbits in a third study produced no damage to the corneal epithelium or endothelium.

Castor oil swabbed onto the dorsum of each of 216 albino or Long-Evans rats daily for five days to 2 weeks produced a mild effect on the epidermis. Undiluted castor oil was applied for 24 h to two areas on the dorsal surface of six albino angora rabbits. Severe skin irritation was observed. Undiluted castor oil applied to the dorsal skin of six male Hartley guinea pigs, six male Wistar rats, and six miniature swine produced mild skin irritation in guinea pigs and rats, but not in miniature swine. Repeated applications of undiluted castor oil to the flanks of five female albino rabbits daily for 30 days induced slight erythema and edema. Both open skin irritation and repeated application (8 weeks) tests for undiluted castor oil and 10% aqueous castor oil were well tolerated by rabbits in another study. Slight reddening of the application site (flank) was observed in a study in which six albino guinea pigs and six young pigs received applications of undiluted castor oil on 10 successive days.

In a predictive prick test in which seven normal subjects were tested with an SLS extract of Castor Wax (concentrations up to 22,000 μg/ml), no immediate skin reactivity was observed. None of the seven subjects patch tested with the SLS Castor Wax extract or the remaining three extracts of Castor Wax (concentrations not stated) had a delayed skin reaction. In a provocative prick test in the same study, three patients with occupational hypersensitivity to castor allergens were tested with the SLS extract of Castor Wax at concentrations up to 22,000 μg/ml. All three patients had a positive reaction to the SLS Castor Wax extract. None of the three patients patch-tested had a delayed reaction to the SLS Castor Wax extract or the remaining three extracts of Castor Wax (concentrations not stated).

The positive skin reactions (immediate skin reactivity) in sensitized individuals, together with the positive PCA reactions in mice and positive direct RAST for reaginic castor allergens that were induced by SLS Castor Wax extract, indicate that allergens are present in Castor Wax at low concentrations.

Neither erythema nor edema was observed following a single topical application of Ricinoleic Acid (in peanut oil) to the paws of 8 to 10 male Swiss mice. The application of Ricinoleic Acid (in peanut oil) to the entire eyelid surface of each of six male albino Dunkin-Hartley guinea pigs induced eyelid reddening and edema at doses of 10, 30, or 100 mg. Cetyl Ricinoleate was classified as a non-irritant following application to the skin of three male New Zealand albino rabbits. Diluted TEGODEO HY 77 (trade name mixture that generally contains >50% Zinc Ricinoleate) was applied, under occlusive patches and after patch removal. Well-defined erythema was observed at 48 and 72 h at the abraded sites of all six rabbits and at the intact sites of four rabbits. This mixture did not produce skin sensitization in a study involving 30 white guinea pigs.

Castor oil was not comedogenic when applied to the ears of rabbits for 2 weeks, but there was a slight increase in keratin content within the follicle, with essentially no change in the follicular epithelium.

Sodium Ricinoleate caused dose-related increases in cytotoxicity in cultures of epithelial cells that were isolated from the small intestines of male Syrian hamsters. In another study, Sodium Ricinoleate did not induce significant toxicity in human saphenous vein endothelial cell cultures at concentrations $\leq$25 μM. At concentrations of 0.1% and 1.0% Sodium Ricinoleate, applied to serosal surface or rat jejunum, a significant reduction in the number of ganglion cells in the myenteric or submucosal plexus was noted. This was not true for the lowest test concentration (0.01%). Castor oil did not have a toxic effect

on the tubular epithelium following intraluminal injection into the kidneys of five male white albino rats. In a study evaluating the nephrotoxicity of cyclosporine, castor oil served as the vehicle control.

Neither castor oil nor Sodium Ricinoleate was mutagenic to the following *Salmonella typhimurium* strains either with or without metabolic activation: TA98, TA100, TA102, TA1535, and TA1537. Results were also negative for castor oil in *Escherichia coli* strain WP2/pKM102. Groups of male and female mice that received diets containing 0.62%, 1.25%, 2.5%, 5.0%, and 10.0% castor oil (in DMSO) for 13 weeks had no evidence of induction of micronuclei in peripheral erythrocytes. Castor oil did not induce sister chromatid exchanges either with or without metabolic activation. Sodium Ricinoleate interfered with cell division in an *Escherichia coli* strain, causing the organisms to develop as filaments. Ricinoleic Acid was inactive as a stimulator of DNA synthesis and inducer of ornithine decarboxylase activity in the rat colon. Concentrations up to 10 mM resulted in insignificant increases in [^{3}H]dThd incorporation and did not increase the level of ornithine decarboxylase activity above control values.

None of the 20 mice injected intravaginally with 2% Ricinoleic Acid (in gum tragacanth) had neoplasms or hyperplastic lesions of the corpus uteri, cervix uteri, vagina, or perineal skin. However, benign lung adenomas were observed in 10 of the 13 mice dosed with Ricinoleic Acid and in 6 of the 24 vehicle control mice that were killed after the 14th month of dosing.

Neither undiluted castor oil, Ricinoleic Acid, nor Glyceryl Ricinoleate was a tumor promoter in a study involving groups of ten mice. However, the three test substances induced slight epidermal hyperplasia in groups of three mice following the application of each to a small area of skin in the interscapular region.

Castor oil extract had a strong suppressive effect on S_{180} body tumors in male Kunming mice. Its suppressive effect on ARS ascites cancer was also very strong, with 64% of the ascites tumors in mice being completely cured. Compared to controls, the life extension rate was more than 136% ($p < .0012$).

Groups of mice and rats fed diets containing 0.62%, 1.25%, 2.5%, 5.0%, and 10% Castor Oil continuously for 13 weeks had a slight decrease in epididymal weight (6% to 7%) in mid- and high-dose groups of male rats; however, this finding was not dose-related. No effects on any other male reproductive end point (testes weight and epididymal sperm motility, density, or testicular spermatid head count) or female reproductive endpoint (estrous cycle length, or time spent in each phase of the cycle) were noted.

Female Wistar rats injected intramuscularly with castor oil (0.2 ml, single dose) on the first day after estrus had suppressed ovarian folliculogenesis and anti-implantation and abortive effects. Anticonceptive and estrogenic effects of a methanol extract of *Ricinus communis* var.*minor* seeds (ether-soluble fraction) were studied in adult albino Wistar rats, young albino Wistar rats, immature Swiss albino female mice, and adult female New Zealand white rabbits. Subcutaneous administration of the extract produced anti-implantation, anticonceptive, and estrogenic activity in rats and mice.

Castor oil served as the vehicle control in a study evaluating the effect of long-term treatment with ICI 182,780 (an antiestrogen) on the rat testis. In the control group, four male Sprague-Dawley rats were injected subcutaneously with castor oil (0.2 ml) once per week and then killed 100 days after the first injection. Spermatogenesis appeared normal in each of the four control rats.

Castor oil (2 oz) was used to stimulate labor in 196 patients (between 37 and 42 weeks of gestation) with premature rupture of membranes. The need for cesarean sections was nearly three times greater in the control group (15.7% incidence) than in patients dosed with castor oil (5.6% incidence). No relationship between dosing with Castor Oil (administered as a 60-ml dose in orange or apple juice), birth weight, and mode of delivery was found in a study in which castor oil was used to induce labor in 52 pregnant women. The cesarean section rate for women dosed with castor oil was 19.2% (10 of 52 women), compared to 8.3% (4 of 48 controls) in the untreated control group. No adverse outcomes for the mother or fetus were identified.

Mild and transient discomfort and minor epithelial changes were observed in the eyes of nine patients after treatment (instillation) with a castor oil solution four times daily for 15 days. In another study involving 100 patients, the instillation of castor oil affected the cornea and conjunctiva. Specifically, the degeneration and death of epithelial cells and continuity breaks in the epithelium were observed.

Mild skin irritation was reported in 50 adult males patch tested with undiluted castor oil (0.05 g applied to back). None of the 1 to 20 patients, either suffering from or suspected of suffering from contact allergy to cosmetic products, patch tested with 30% Hydrogenated Castor Oil had irritant reactions. Undiluted castor oil rubbed gently onto the right thigh of each of three male subjects on 10 successive days produced, at microscopic examination of excised skin, clear hyperchromasia and an increase in the number of cells was observed in the basal cell layer. Slight widening of the granular cell layer was also noted. No skin irritation was seen with 20% Ethyl Ricinoleate in petrolatum using 32 healthy male volunteers in a 48-h closed patch test.

No skin sensitization or irritation was seen with 20% Ethyl Ricinoleate in a maximization test using 32 healthy male volunteers. In 49 patients with contact dermatitis patch-tested (48-h Finn chambers) with castor oil, none had a positive reaction. One of the 76 dermatitis patients patch-tested with Castor Oil had a positive reaction, observed only during the second reading (i.e., the day after the 30-min reading). Of 202 consecutive patients with eczematous cheilitis who attended a contact and occupational dermatoses clinic in Singapore, 29 patients had positive reactions to Ricinoleic Acid. Of the 29 positive reactions, 22 were considered relevant positives. In an allergenicity-photoallergenicity study, 103 patients suspected of having cosmetic-related dermatitis were patch tested

with castor oil. One patient had a positive patch test reaction. None of the patients had a positive photopatch test reaction.

In case reports that included patch test results for castor oil, Ricinoleic Acid, or esters of Ricinoleic Acid, both positive and negative reactions were reported. Of a group of 25 employees at a company involved with the preparation of plant products, 1 employee had an allergic reaction to an extract of the castor oil bean.

DISCUSSION

The CIR Expert Panel considered that the available data on Ricinus Communis (Castor) Seed Oil, Hydrogenated Castor Oil, Ricinoleic Acid, and salts and esters of Ricinoleic Acid in the current safety assessment are sufficient for evaluating the safety of Glyceryl Ricinoleate and Glyceryl Ricinoleate SE, as well. Because Ricinus Communis (Castor) Seed Oil contains Ricinoleic Acid as the primary fatty acid group, safety test data on the oil is considered broadly applicable to this entire group of cosmetic ingredients. Overall, the available data demonstrate few toxic effects in acute, subchronic, or chronic toxicity tests. Additionally, there were no genotoxic effects of castor oil in in vitro or in vivo tests. It was noted that the available data include a 50-week chronic study in which none of the 20 mice injected with Ricinoleic Acid twice per week had neoplasms or hyperplastic lesions of the corpus uteri, cervix uteri, vagina, or perineal skin. UV absorption spectra on Ricinus Communis (Castor) Seed Oil and Glyceryl Ricinoleate indicate maximum absorbance at 270 nm, suggesting that there would be no photosensitization potential of Glyceryl Ricinoleate or Ricinus Communis (Castor) Seed Oil in human subjects at relevant solar UV exposures.

Reactions classified as either significantly irritating or allergic were not observed in a maximization test on Ethyl Ricinoleate involving 32 healthy male subjects. Although there is a paucity of predictive patch test data on Ricinoleic Acid esters/salts, the Panel agreed that the negative guinea pig sensitization data on a trade name mixture containing more than 50% Zinc Ricinoleate, in conjunction with the human predictive patch test data, may be extended to support the safety of all of the cosmetic ingredients that are included in this safety assessment.

The Panel did consider the 22 positive reactions to Ricinoleic Acid in a retrospective study involving 202 consecutive patients with eczematous cheilitis. Recognizing that this study represents a case series on cheilitis that involves a select patient population, the Panel agreed that the incidence of sensitization reactions reported in the study may be expected in that select patient group, but not in the general population. Based on the Panel's experience and expertise in the area of patient patch testing, the incidence of positive reactions to Ricinoleic Acid is generally very low.

In the absence of inhalation toxicity data on Ricinus Communis (Castor) Seed Oil, Hydrogenated Castor Oil, Ricinoleic Acid, and salts and esters of Ricinoleic Acid in this safety assessment, the Panel determined that these ingredients can be used safely in aerosolized products because packaging and use ensure that particulates are not respirable (are greater than 10 μm). The Panel reasoned that the particle size of anhydrous hair sprays (60 to 80 μm) and pump hair sprays (>80 μm) is large compared to the median aerodynamic diameter of 4.25 $\pm$ 1.5 μm for a respirable particulate mass.

The CIR Expert panel recognizes that certain ingredients in this group are reportedly used in a given product category, but the concentration of use is not available. For other ingredients in this group, information regarding use concentration for specific product categories is provided, but the number of such products is not known. In still other cases, an ingredient is not in current use, but may be used in the future.

Although there are gaps in knowledge about product use, the overall information available on the types of products in which these ingredients are used and at what concentration indicate a pattern of use. Within this overall pattern of use, the Expert Panel considers all ingredients in this group to be safe.

CONCLUSION

The CIR Expert Panel concluded that Ricinus Communis (Castor) Seed Oil, Hydrogenated Castor Oil, Glyceryl Ricinoleate, Glyceryl Ricinoleate SE, Ricinoleic Acid, Potassium Ricinoleate, Sodium Ricinoleate, Zinc Ricinoleate, Cetyl Ricinoleate, Ethyl Ricinoleate, Glycol Ricinoleate, Isopropyl Ricinoleate, Methyl Ricinoleate, and Octyldodecyl Ricinoleate are safe as cosmetic ingredients in the practices of use and concentrations as described in this safety assessment.

REFERENCES

Achaya, K. T. 1971. Chemical derivatives of castor oil. *J. Am. Oil Chem. Soc.* 48:758–763.

Allegri, R., G. C. Deschel, V. Filippi, M. Gibellini, E. Portioli, and D. Veneri. 1981. Proposal for the pharmacopoeia: hydrogenated castor oil [translation]. *Boll. Chim. Farm.* 120:557–558.

Ames, B. N., J. McCann, and E. Yamasaki. 1975. Methods for detecting carcinogens and mutagens with the Salmonella/mammalian-microsome mutagenicity test. *Mutat. Res.* 31:347–364.

Andersen, K. E., and R. Nielsen. 1984. Lipstick dermatitis related to castor oil. *Contact Dermatitis* 11:253–254.

Aplin P.J., and Eiseo T. 1997. Ingestion of castor oil plant seeds. *Med. J. Aust.* 167:260–261.

Ayorinde, F. O., K. Garvin, and K. Saeed. 2000. Determination of the fatty acid composition of saponified vegetable oils using matrix-assisted laser desorption/ionization time-of-flight mass spectrometry. *Rapid. Commun. Mass Spectrom.* 14:608–615.

Baynes, R. E., and J. E. Riviere. 2004. Mixture additives inhibit the dermal permeation of the fatty acid, ricinoleic acid. *Toxicol. Lett.* 147:15–26.

Behrens-Baumann, W., S. Theuring, and H. Brewitt. 1986. The effect of topical cyclosporin A on the rabbit cornea. A clinical and electron microscopic study. *Graefes Arch. Clin. Exp. Ophthalmol.* 224:520–552.

Binder, R. G., A. N. Booth, D. J. Robbins, and G. Fuller. 1970. Hydroxystearic acid deposition and metabolism in rats fed hydrogenated castor oil. *Lipids* 5:832–837.

Blair, R. M., H. Fang, W. S. Branham et al. 2000. The estrogen receptor relative binding affinities of 188 natural and xenochemicals: Structural diversity of ligands. *Toxicol. Sci.* 54:138–153.

Bower, D. 1999. Unpublished information on hair spray particle sizes provided at the September 9, 1999 CIR Expert Panel meeting.[2]

Boyland, D., F. J. C. Roe, and B. C. V. Mitchley. 1966. Test of certain constituents of spermicide for carcinogenicity in genital tract of female mice. *Br. J. Cancer* 20:184–189.

Brandle, I., A. Boujnah-Khouadja, and J. Foussereau. 1983. Allergy to castor oil. *Contact Dermatitis* 9:424–425.

Bronaugh, R. L., and R. F. Stewart. 1985. Methods for in vitro percutaneous absorption studies. IV: The flow-through diffusion cell. *J. Pharm. Sci.* 74:64–66.

Beubler, E., and A. Schirgi-Degen. 1992. Stimulation of enterocyte protein kinase C by laxatives in vitro. *J Pharm Pharmacol* 45:59–62.

Budavari, S, ed. 1989. *The Merck Index. An Encyclopedia of Chemicals, Drugs, and Biologicals*, 11th ed., 290, 1307. Rahway, NJ: Merck & Co.

Bull, A. W., N. D. Nigro, W. A. Golembieski, J.D. Crissman, and L.J. Marnett. 1984. *In vivo* stimulation of DNA snthesis and induction of ornithine decarboxylase activity in rat colon by hydroperoxides, autoxidation products of unsaturated fatty acids. *Cancer Res* 44:4924–4928.

Bull, A. W., N. D. Nigro, and L. J. Marnett. 1988. Structural requirements for stimulation of colonic cell proliferation by oxidized fatty acids. *Cancer Res* 48:1771–1776.

Butcher, E. O. 1951. The effects of applications of various substances on the epidermis of the rat. *J Invest Dermatol* 16:85-90.

Butcher, E. O. 1952. The penetration of fat and fatty acid into the skin of the rat. *J Invest Dermatol* 21:43–48.

Campbell & Co. 2002. Castor oil & castor oil derivatives. Product data—castor waxes. http://www.campbelluv.com/castor.html [accessed September 2004].

Capasso, F., N. Mascolo, G. Autore, and V. Romano. 1986. Laxatives and the production of autacoids by rat colon. *J Pharm Pharmacol* 38:627–629.

Capasso, F., N. Mascolo, A. A. Izzo, and T. S. Gaginella. 1994. Dissociation of castor oil-induced diarrhea and intestinal mucosal injury in rat: Effect of N^G-nitro-l-arginine methyl ester. *Br J Pharmacol* 113:1127–1130.

Carpenter, C. P., and H. F. Smyth, Jr. 1946. Chemical burns of the rabbit cornea. *Am J Ophthal* 29:1363–1372.

Chakravarty, D., and A. Bose. 1963. A note on the preparation of ricinoleic acid by urea complexing. *J Pharm Pharmacol* 15:337–338.

Committee of Revision of the United States Pharmacopeial Convention. 2004a. The *United States Pharmacopoeia*, 27th ed., 348. Rockville, MD: United States Pharmacopeial Convention.

Committee of Revision of the United States Pharmacopeial Convention. 2004b. The National Formulary, 22nd ed., 2844. Rockville, MD: United States Pharmacopeial Convention.

Cornell University. 2001. Castor Oil. http://www.ansci.cornell.edu/plants/castorbean.html [accessed October 2003].

Cosmetic, Toiletry, and Fragrance Association (CTFA). 2004. Use concentration data from industry survey. Unpublished data submitted by CTFA, May 2004. 3 pages.[2]

Croci T., M. Landi, X. Emonds-Alt, G. Le Fur, J. P. Maffrand, and L. Manara. 1997. Role of tachykinins in castor oil diarrhea in rats. *Br J Pharmacol* 121:375–380.

Davis, L. 1984. The use of castor oil to stimulate labor in patients with premature rupture of membranes. *J Nurse Midwifery* 29:366–370.

De Caterina, R., and W. Bernini. 1998. Structural requirements for inhibition of cytokine-induced endothelial activation by unsaturated fatty acids. *J Lipid Res.* 39:1062–1070.

De Caterina, R., H. Tanaka, T. Nakagawa, P. J. Hauptman, and P. Libby. 1995. The direct effect of injectable cyclosporine and its vehicle, cremophor, on endothelial vascular cell adhesion molecule-1 expression. *Transplantation* 60:270–275.

[2]Available from the Director, Cosmetic Ingredient Review, 1101 17th Street, NW, Suite 412, Washington, DC 20036, USA.

De Groot, A. C., ed. 1994. *Patch testing. Test concentrations and vehicles for 3700 Chemicals*, 2nd ed., 11, 145. Amsterdam: Elsevier.

Degussa. 2001. Product data record. TEGODEO HY 77. Unpublished data submitted by CTFA, May 5, 2004. 2 pages.[2]

Dooms-Goossens, A. K. Dupré, A. Borghijs, C. Swinnen, M. Dooms, and H. Degreef. 1987. Zinc ricinoleate: Sensitizer in deodorants. *Contact Dermatitis* 16:292–294.

Drugstore.com, Inc. 2004. Rite Aid Castor oil, stimulant laxative U.S.P. Package details: Directions. http://www.drugstore.com [accessed December 2004].

Elder, R. L. ed. 1988. Final report on the safety assessment of glyceryl ricinoleate. *J. Am. Coll. Toxicol.* 7:721–739.

Fisher, A. A. 1991. Allergic cheilitis due to castor oil in lipsticks. *Cutis* 47:389–390.

Food and Drug Administration (FDA). 1985. Laxative drug products for over-the-counter human use; Tentative final monograph. *Federal Register* 50:2124–2158.

FDA. 1990. Wart remover drug products for over-the-counter human use. Final monograph. *Federal Register* 55:33246–33256.

FDA. 2002. Frequency of use of cosmetic ingredients.*FDA database*. Washington, DC: FDA.

FDA. 2003a. OTC Drug Review Ingredient Report. *FDA database*. Washington, DC: FDA.

FDA. 2003b. Inactive Ingredients Guide (Redacted), January 1996. http://www.fda.gov/cder/drug/iig/default.htm [accessed August 2003].

Fox, D. A., M. L. Epstein, and P. Bass. 1983. Surfactants selectively ablate enteric neurons of the rat jejunum. *J Pharmacol Exp Ther* 227:538–544.

Frankel, E. N., W. E. Neff, D. D. Brooks, and K. Fujimoto. 1987. Fluorescence formation from the interaction of DNA with lipid oxidation degradation products. *Biochim. Biophys. Acta* 919:239–244.

Fredholt, K., D. H. Larsen, and C. Larsen. 2000. Modification of in vitro drug release rate from oily parenteral depots using a formulation approach [translation]. *Eur J Pharm Sci* 11:231–237.

Fujimoto, M., N. Higashi, A. Kume, K. Ueda, and N. Hino. 1997. Patch test results in 332 patients suspected of cosmetic dermatitis. *Environ Dermatol* 4:268–276.

Fulton, J. E. Jr. 1989. Comedogenicity and irritancy of commonly used ingredients in skin care products. *J Soc Cosmet Chem* 40:321–333.

Fulton, J. E. Jr., S. Bradley, A. Aqundez, and T. Black. 1976. Non–comedogenic cosmetics. *Cutis* 17:344–351.

Gaginella, T. S., and P. Bass. 1978. Laxatives: An update on mechanism of action. *Life Sci.* 23:1001–1010.

Gaginella, T. S., V.S. Chadwick, J. C. Debongnie, J. C. Lewis, and S. F. Phillips. 1977a. Perfusion of rabbit colon with ricinoleic acid: Dose-related mucosal injury, fluid secretion, and increased permeability. *Gastroenterology* 73:95–101.

Gaginella, T. S., A. C. Haddad, V. L. W. Go, and S. F. Phillips. 1977b. Cytotoxicity of ricinoleic acid (castor oil) and other intestinal secretagogues on isolated intestinal epithelial cells. *J Pharmacol Exp Ther* 201:259–266.

Gaginella, T. S., S. F. Phillips, R. R. Dozois, and V. L. Go. 1978. Stimulation of adenylate cyclase in homogenates of isolated intestinal epithelial cells from hamsters. Effects of gastrointestinal hormones, prostaglandins, and deoxycholic and ricinoleic acids. *Gastroenterology* 74:11–15.

Gao, J., N.-.H. Sun, F.-Y. Wang, and N. Hao. 1999. Effects of castor oil-diet on the synthesis of prostaglandin E2 in pregnant rats [translation]. *Zhonghua Fu Chan Ke Za Zhi* 34:147–149.

Garry, D., R. Figueroa, J. Guillaume, and V. Cucco. 2000. Use of castor oil in pregnancies at term. *Altern Ther Health Med.* 6:77–79.

Gennaro, A. R., ed. 1990. *Remington's Pharmaceutical Sciences*, 18th ed., 785. Easton, PA: Mack Publishing.

Gluud, B. S., T. Boesen, and M. Norn. 1981. The effect of a fatty vehicle on the cornea and conjunctiva. In vivo staining following instilling of oil [translation]. *Ugeskr Laeger* 143:2345–2347.

Göhte, C. J., G. Wieslander, K. Ancker, and M. Forsbeck. 1983. Buckwheat allergy: Health food, an inhalation health risk. *Allergy* 38:155–159.

Goon, A. T.-J., P. P.-L. Ng, and S.-K. Ng. 1999. Allergic contact dermatitis from military camouflage. *Contact Dermatitis* 40:290–291.

Gottschalck, T. E., and G. N. McEwen, Jr., eds. 2004. *International Cosmetic Ingredient Dictionary and Handbook,* 10th ed. Washington, DC: CTFA.

Grainger, D. N., R. Cross, and J. A. Barrowman. 1982. Effects of various secretagogues and human carcinoid serum on lymph flow in the cat ileum. *Gastroenterology* 83:896–901.

Grant J, ed. 1972. *Hackh's Chemical Dictionary*, 4th ed., 584. New York: McGraw-Hill Book Co.

Guillot, J. P., J. Y. Giauffret, and M. C. Martini. 1979. Etude de tolerances oculaire et cutanee chez la lapin, de differentes matieres premieres utilisees en cosmetologie, et provenant de fabrications diverses (2eme partie). *Int. J. Cosmet. Sci.* 1:27–57.

Gunstone, F. D. 1989. Chemical reactions of fats and oils. *Biochem Soc Trans* 17:1141–1142.

Hachiya, N. 1987. Evaluation of chemical genotoxicity by a series of short-term tests. *Akita J Med* 14:269–292.

Hagenfeldt, L., L. Blomquist, and T. Midtvedt. 1986. Epoxydicarboxylic aciduria resulting from the ingestion of castor oil. *Clin Chim Acta* 161:157–163.

Hashimoto, Y., T. Sugai, A. Shoji, S. Asoh, K. Watanabe, and A. Inoue. 1990. Incidence of positive reactions in patch tests with ingredients of cosmetic products in 1989 and representative cases of cosmetic dermatitis (Translation). *Skin Res.* 32:115–124.

Haworth, S., T. Lawlor, K. Mortelmans, W. Speck, and E. Zeiger. 1983. *Salmonella* mutagenicity test results for 250 chemicals. *Environ Mutagen* 5:3–142.

Hino, N., M. Ikushima-Fujimoto, K. Ueda, A. Kume, and N. Higashi. 2000. Patch test results in 346 patients of suspected cosmetic dermatitis (1993–1996) [translation]. *Environ Dermatol* 7:144–153.

Hui, Y. H., ed. 1996. *Bailey's Industrial Oil & Fat Products. Industrial and Consumer Nonedible Products from Oils and Fats*, 5th ed., vol. 5., 368. New York: John Wiley & Sons.

Ihara-Watanabe, M., T. Shiroyama, G. Konda, H. Umekawa, T. Takahashi, T. Yamada and Y. Furuichi. 1999. Effects of castor oil on lipid metabolism in rats. *Biosci Biotechnol Biochem* 63:595–597.

Inoue, A, A. Shoji, and S. Aso. 1998. Allergic lipstick cheilitis due to ester gum and ricinoleic acid. *Contact Dermatitis* 39:39.

International Bio-Research, Inc. 1977a. Delayed contact sensitization (guinea pigs). Compound: "Grillocin HY 77" (undiluted) (Translation). Unpublished data submitted by CTFA. 18 pages.[2]

International Bio-Research, Inc. 1977b. Patch test for primary skin irritation and corrosivity of the compound: "Grillocin HY 77" (10% soy bean oil). Unpublished data submitted by CTFA. 7 pages.[2]

Jensen, P. A., and D. Obrien. 1993. Industrial hygiene. In: *Aerosol Measurement. Principles, Techniques and Applications*, ed. K. Willeke and P.A. Baron, 538–540. New York: John Wiley and Sons.

Johnsen, M. A. 2004. The influence of particle size. *Spray Technology and Marketing*. November:24–27.

Johnson, C. M., J. M. Cullen, and M. C. Roberts. 1993. Morphologic characterization of castor oil-induced colitis in ponies. *Vet Pathol* 30:248–255.

Jost, T., Y. Sell, and J. Foussereau. 1989. Contact allergy to Manilla resin. Nomenclature and physico-chemistry of Manilla, kauri, damar and copal resins. *Contact Dermatitis* 21:228–238.

Kathren, R. L., H. Price, J. C. Rogers. 1959. Air-borne castor-bean pomace allergy; a new solution to an old problem. *AMA Arch Ind Health* 19:487–489.

Kato, A., and Y. Yamaura.1970. A rapid gas chromatographic method for the determination of fatty acid compositions of soybean oil and castor oil. *Chem Ind* 39:1260.

KIC Chemicals, Inc. 2004. Hydrogenated castor oil, standard grade. Specifications. http://www.kicchem.com/hcostandard.htm [accessed September 2004].

Laboratoire De Recherche Et D'Experimentation. 1994. Acute oral toxicity and skin irritation and/or corrosion tests on cetyl ricinoleate. Unpublished data submitted by CTFA, April 13, 2004. 2 pages.[2]

Langer, K. H., W. Thoenes, and M. Wiederholt.1968. Light and electron microscopic investigations of proximal convoluted tubules of the rat kidney after intraluminal injection of oil [translation]. *Pflugers Arch* 302:149–165.

Larsen, S. W., E. Rinvar, O. Svendsen, J. Lykkesfeldt, G. J. Friis, and C. Larsen. 2001. Determination of the disappearance rate of iodine-125 labelled oils from the injection site after intramuscular and subcutaneous administration to pigs. *Int J Pharm* 230:67–75.

le Coz, C. J., and C. Ball. 2000. Recurrent allergic contact dermatitis and cheilitis due to castor oil. *Contact Dermatitis* 42:114–115.

Lehrer, S. B, R. M. Karr, D. J. G. Müller, and J.E. Salvaggio. 1980. Detection of castor allergens in castor wax. *Clin. Allergy* 10:33–41.

Leow, Y. H., S. H. Tan, and S. K. Ng. 2003. Pigmented contact cheilitis from ricinoleic acid in lipsticks. *Contact Dermatitis* 49:48–49.

Lewis, R. J. Sr. 1997. *Hawley's Condensed Chemical Dictionary*, 13th ed., 221, 749, 922, 972, 1027, 1200. New York: John Wiley & Sons.

Lewis, R. J., Sr., ed. 2000. *Sax's Dangerous Properties of Industrial Materials*,10th ed., 738. New York: John Wiley & Sons.

Lide, D. R., and H. P. R. Frederiske, eds. 1993. *CRC Handbook of Chemistry and Physics*, 74th ed., 3-453. Boca Raton, FL: CRC Press.

Lieb, L. M., R. A. Nash, J. R. Matias, and N. Orentreich. 1988. A new in vitro method for transepidermal water loss: A possible method for moisturizer evaluation. *J Soc Cosmet Chem* 39:107–119.

Lim, S. W., and C. L. Goh. 2000. Epidemiology of eczematous cheilitis at a tertiary dermatological referral centre in Singapore. *Contact Dermatitis* 43:322–326.

Litvinova, L. B., and T. V. Fedorchenko. 1994. The effect of plant oils on the female reproductive system [translation]. *Eksp Klin Farmakol* 57:49–51.

Lodge, N. J. 1994. Direct vasoconstrictor effects of sandimmune (cyclosporine A) are mediated by its vehicle Cremophor EL: Inhibition by the thromboxane A_2/prostaglandin endoperoxide receptor antagonist ifetroban. *J Pharmacol Exp Ther* 271:730–734.

Lodi, A., S. Leuchi, L. Mancini, G. Chiarelli, and C. Crosti. 1992. Allergy to castor oil and colophony in a wart remover. *Contact Dermatitis* 26:266–267.

Lorenz, W., A. Schmal, H. Schult et al. 1982. Histamine release and hypotensive reactions in dogs by solubilizing agents and fatty acids: Analysis of various components in cremophor El and development of a compound with reduced toxicity. *Agents Actions* 12:64–80.

Magerl, A., R. Heiss, and P. J. Frosch. 2001. Allergic contact dermatitis from zinc ricinoleate in a deodorant and glyceryl ricinoleate in a lipstick. *Contact Dermatitis* 44:119–121.

Maier, M., D. Staupendahl, H. R. Duerr, and H. J. Refior. 1999. Castor oil decreases pain during extracorporeal shock wave application. *Arch Orthop Trauma Surg* 119:423–427.

Manzur, F., F. El Sayed, and J. Bazex. 1995. Contact allergy to cinnamic aldehyde and cinnamic alcohol in Oléophytal. *Contact Dermatitis* 32:55.

Masri, M. S., L. A. Goldblatt, F. DeEds, and G. O. Kohler. 1962. Relation of cathartic activity to structural modifications of ricinoleic acid of castor oil. *J Pharm Sci* 51:999–1002.

Mathias, J. R., J. L. Martin, T. W. Burns, G. M. Carlson, and R. P. Shields. 1978. Ricinoleic acid effect on the electrical activity of the small intestine in rabbits. *J Clin Invest* 61:640–644.

McKeon, T. A., G. Q. Chen, and J. T. Lin. 2000. Biochemical aspects of castor oil biosynthesis. *Biochem Soc Trans* 28:972–974.

Meyer, F. U., C. Wollmann, N. Exner, and G. Exner. 1976. Comparative studies on the skin of guinea pigs, swine and man following the effect of various external agents [translation]. *Dermatol Monatsschr* 162:986–991.

Migally, N. 1979. Effect of castor oil and benzyl benzoate used as a vehicle for antiandrogens on the adrenal cortex. *Arch Androl* 2:365–369.

Mitri, F., G. J. Hofmeyr, and C. J. Van Geldren. 1987. Meconium during labor–self-medication and other associations. *SAMJ* 431–433.

Mordenti, J. J., R. E. Lindstrom, and J. M. Tanzer. 1982. Activity of sodium ricinoleate against in vitro plaque. *Pharm Sci* 71:1419–1421.

Morehouse, J. L., R. D. Specian, J. J. Stewart, and R. D. Berg. 1986. Translocation of indigenous bacteria from the gastrointestinal tract of mice after oral ricinoleic acid treatment. *Gastroenterology* 91:673–682.

Morris, J. A., A. Khettry, and E. W. Seitz. 1979. Antimicrobial activity of aroma chemicals and essential oils. *J. Am. Oil Chem. Soc.* 56:595–603.

Morris, W. E., and S. C. Kwan. 1983. Use of the rabbit ear model in evaluating the comedogenic potential of cosmetic ingredients. *J Soc Cosmet Chem* 34:215–225.

Motoyoshi, K., Y. Toyoshima, M. Sato, and M. Yoshimura. 1979. Comparative studies on the irritancy of oils and synthetic perfumes to the skin of rabbit, guinea pig, rat, miniature swine and man. *Cosmet Toilet* 94: 41.

Muldoon, D. F., E. A. Hassoun, and S. J. Stohs. 1992. Ricin-induced hepatic lipid peroxidation, glutathione depletion, and DNA single-strand breaks in mice. *Toxicon* 30:977–984.

National Academy of Sciences (NAS). 1996. *Food Chemicals Codex*, 4th ed., 94 Washington, DC: National Academy Press.

National Toxicology Program (NTP). 1992. NTP technical report on the toxicity studies of castor oil (CAS No. 8001-79-4) in F344/N rats and B6C3F1 mice (dosed feed studies). NTIS Report No. PB93151439.

NTP. 2003. *NTP Chemical Repository*. Castor Oil. Research Triangle Park, NC: NTP.

Nikitakis, J. M., and G. N. McEwen Jr., eds. 1990. *CTFA Compendium of Cosmetic Ingredient Composition—Descriptions I and II*. Washington, DC: CTFA.

Ohia, E. O., M. Mancino, and P. S. Kulkarni. 1992. Effects of steroids and immunosuppressive drugs on endotoxin-uveitis in rabbits. *J Ocul Pharmacol* 8:295–307.

Okui, S., M. Uchiyama, R. Sato, and M. Mizugaki. 1964. Metabolism of hydroxy fatty acids. III. Characterization of hydroxy acids in depot fat after feeding of ricinoleic acid. *J Biochem* 55:81–86.

Okwuasaba, F. K., U. A. Osunkwo, M. M. Ekwenchi, et al. 1991. Anticonceptive and estrogenic effects of a seed extract of Ricinus communis var. minor. *J Ethnopharmacol* 34:141–145.

Oliveira, C. A., K. Carnes, L. R. Franca, and R. A. Hess. 2001. Infertility and testicular atrophy in the antiestrogen-treated adult male rat. *Biol Reprod* 65:913–920.

Ordman, D. 1955. An outbreak of bronchial asthma in South Africa, affecting more than 200 persons, caused by castor bean dust from an oil-processing factory. *Int Arch Allergy* 7:10.

Paul, H., and C. M. McCay. 1942. The utilization of rats by herbivora. *Arch Biochem* 1:247–253.

Paulus, G. L., and M. H. Champion. 1972. Gas-liquid chromatographic determination of castor oil as methyl ricinoleate in lipstick. *J Soc Cosmet Chem* 23:313–319.

Physical & Theoretical Chemistry Lab. 2004. Safety (MSDS) data for hydrogenated castor oil. http://ptcl.chem.ox.ac.uk/MSDS/HY/hydrogenated_castor_oil.html [accessed September 2004].

Pinto, A., A. Calignano, N. Mascolo, G. Autore, and F. Capasso. 1989. Castor oil increases intestinal formation of platelet-activating factor and acid phosphatase release in the rat. *Br J Pharmacol* 96:872–874.

Pöch, G. 1971. Assay of phosphodiesterase with radioactively labeled cyclic 3′5′-AMP as substrate. *Naunyn-Schmiedeberg's Arch Pharmacol* 268:272–299.

Polgar, P., and L. Taylor. 1977. Effects of prostaglandin on substrate uptake and cell division in human diploid fibroblasts. *Biochem J*. 162:1–8.

Racusen, L. C., B. C. Kone, A. Whelton, and K. Solez.1986. Renal blood flow, glomerular filtration rate, and renal morphology in cyclosporine-induced acute renal failure in Munich-Wistar rats. *Am J Kidney Dis* 8:319–322.

Ramsey, J. D., T. D. Lee, M. D. Osselton, and A. C. Moffat. 1980. Gas liquid chromatographic retention indices of 296 nondrug substances on SE-30 or OV-1 likely to be encountered in toxicological analyses. *J Chromatogr* 184:185–206.

Rantuccio, F., D. Sinisi, A. Scardigno, and C. Coviello. 1981. Histological changes in rabbits after application of medicaments and cosmetic bases. II. *Contact Dermatitis* 7:94–97.

Rao, M. K., N. Risser, and E. G. Perkins. 1969. The incorporation of ricinoleic acid into rat lymph lipids. *Proc Soc Exp Biol Med* 131:1369–1372.

Ratner, B., and H. L. Gruehl. 1929. Respiratory anaphylaxis (asthma) and ricin poisoning induced with castor bean dust. *Am J Hyg* 10:236.

Reichert-Pénétrat, S., A. Barbaud, E. Reichert-Pénétrat, F. Granel, and J.-L. Schmutz. 2001. Allergic contact dermatitis from surgical paints. *Contact Dermatitis* 45:116–117.

Research Institute for Fragrance Materials (RIFM). 2000. Fragrance raw materials monographs. Ethyl ricinoleate. *FCT* 38:S91–S92.

Sai, S. 1983. Lipstick dermatitis caused by castor oil. *Contact Dermatitis* 9:75, 524.

Salomon, Y., C. Londos, and M. Rodbell. 1974. A highly sensitive adenylate cyclase assay. *Anal Biochem* 58:541–548.

Sasol North America, Inc. No date. UV spectrum for castor oil and glyceryl ricinoleate. Unpublished data submitted by CTFA, April 14, 2004. 1 page.[2]

Scarpa, A., and A. Guerci.1982. Various uses of the castor oil plant (Ricinus communis L.). A review. *J Ethnopharmacol* 5:117–137.

Secchi, A. G., M. S. Tognon, and A. Leonardi.1990. Topical use of cyclosporine in the treatment of vernal keratoconjunctivitis. *Am J Ophthalmol* 110:641–645.

Shubik, P. 1950. Studies on the promoting phase in the stages of carcinogenesis in mice, rats, rabbits, and guinea pigs. *Cancer Res* 10:13–17.

Simon, B., and H. Kather. 1980. Interaction of laxatives with enzymes of cyclic AMP metabolism from human colonic mucosa. *Eur J Clin Invest* 10:231–234.

Smolinske, S. C. 1992. *Handbook of Food, Drug, and Cosmetic Excipients*, 70. Boca Raton, FL: CRC Press.

Song, J. F., C. A. Lau-Cm, and K. H. Kim. 2001. Monohydroxylation and esterification as determinants of the effects of cis- and trans-9-octadecenoic acids on the permeation of hydrocortisone and 5-fluorouraci across hairless mouse skin in vitro. *Int J Pharmaceut* 212:153–160.

Srinivasulu, C., and S.N. Mahapatra. 1973. A rapid method for detecting groundnut oil in castor oil. *J Chromatogr* 86:261–262.

Steingrub, J. S., T. Lopez, D. Teres, and R. Steingart. 1988. Amniotic fluid embolism associated with castor oil ingestion. *Crit Care Med* 16:642–643.

Stewart, J. J., and P. Bass. 1976. Effects of ricinoleic acid and oleic acids on the digestive contractile activity of the canine small and large bowel. *Gastroenterology* 70:371–376.

Stewart, J. J, T. S. Gaginella, and P. Bass. 1975. Actions of ricinoleic acid and structurally related fatty acids on the gastrointestinal tract. I. Effects on smooth muscle contractility in vitro. *J Pharmacol Exp Ther* 195:347–354.

Stewart, W. C., and R. G. Sinclair. 1945. The absence of ricinoleic acid from phospholipids of rats fed castor oil. *Arch Biochem* 8:7.

Stübiger, G., E. Pittenauer, and G. Allmaier. 2003. Characterization of castor oil by on-line and of-line non-aqueous reverse-phase high-performance liquid chromatography-mass spectrometry (APCI and UV/MALDI). *Phytochem Anal* 14:337–346.

Tabar, A. I., M. D. Muro, S. Quirce, and J. M. Olaguibel. 1993. Contact dermatitis due to sensitization to lactic acid and castor oil in a wart remover solution. *Contact Dermatitis* 29:49–50.

Tan, B. B., A. L. Noble, M. E. Roberts, J. T. Lear, and J. S. English. 1997. Allergic contact dermatitis from oleyl alcohol in lipstick cross-reacting with ricinoleic acid in castor oil and lanolin. *Contact Dermatitis* 37:41–42.

Thompson, W. G. 1980. Laxatives: Clinical pharmacology and rational use. *Drugs* 19:49–58.

TNO BIBRA International Ltd. 1999. Toxicity profile. Castor oil. Surrey, United Kingdom: TNO BIBRA International Ltd. 9 pages.

Uchiyama, M., R. Sato, and M. Mizugaki. 1963. Characterization of hydroxyacids in depot fat after feeding of ricinoleic acid. *Biochem Biophys Acta* 70:344–346.

Vanderhoek, J. Y., R. W. Bryant, and J. M. Bailey. 1980. Communication. 15-hydroxy-5,8,11,13-eicosatetraenoic acid. A potent and selective inhibitor of platelet lipoxygenase. *J Biol Chem* 255:5996–5998.

Vieira, C, S. Evangelista, R. Cirillo, R. Terracciano, A. Lippi, C.A. Maggi, and S. Manzini. 2000. Antinociceptive activity of ricinoleic acid, a capsaicin-like compound devoid of pungent properties. *Eur J Pharmacol* 407:109–116.

Vieira, C., S. Tetzer, S. K. Sauer, et al. 2001. Pro- and anti-inflammatory actions of ricinoleic acid: Similarities and differences with capsaicin. *Naunyn-Schmiedeberg's Arch Pharmacol* 364:87–95.

Wakelin, S. H., A. J. Harris, and S. Shaw. 1996. Contact dermatitis from castor oil in zinc and castor oil cream. *Contact Dermatitis* 35:259.

Watson, W. C., and R. S. Gordon Jr. 1962. Studies on the digestion, absorption and metabolism of castor oil. *Biochem Pharmacol* 11:229–236.

Watson, W. C., R. S. Gordon Jr, A. Karmen, and A. Jover. 1963. The absorption and excretion of castor oil in man. *J Pharm Pharmacol* 15:183–188.

Whiteside-Carlson, V., B. F. Feaster, and W. W. Carlson. 1955. Filament induction in *Escherichia coli* by ricinoleic acid. *Proc Soc Exp Biol Med* 89:382–385.

Wineburg, J. P., and D. Swern. 1973. NMR chemical shift reagents in structural determination of lipid derivatives: III. Methyl ricinoleate and methyl 12-hydroxystearate. *J Am Oil Chem Soc* 50:142–146.

World Health Organization (WHO) .1980. Evaluation of certain food additives. Twenty-third report of the Joint FAO/WHO Expert Committee on Food Additives, 1–45. World Health Organization Technical Report Series 648. Geneva: WHO.

Xie, S. A., D. S. Liu, B. L. Li, Z. H. Wang, and J.Y. Tao. 1992. Antitumor effect of castor oil extract [translation]. *Zhongguo Zhong Yao Za Zh*i 17:560–561.

Yagaloff, K. A., L. Franco, F. Simko, and B. Burghardt. 1995. Essential fatty acids are antagonists of the leukotriene B4 receptor. *Prostaglandins Leukotriene Essent. Fatty Acids* 52:293–297.

Yahagi, T., M. Degawa, Y. Seino, T. Matsushima, M. Nagao, T. Sugimura, and Y. Hashimoto. 1975. Mutagenicity of carcinogenic azo dyes and their derivatives. *Cancer Lett* 1:91–96.

Yoshikawa, K., T. Kozuka, T. Doi, Y. Kataoka, K. Fujimoto, and S. Hashimoto. 1986. A case of allergic contact dermatitis to castor oil. 10th Anniversary Meeting of Japan Patch Test Research Group on Contact Dermatitis and Patch Test VII, Kobe, Japan, December 7–8, 1985. *Skin Res* 28(Suppl. 2):263–265.

Zeiger, E., B. Anderson, S. Haworth, T. Lawlor, and K. Mortelmans. 1988. Mutagenicity tests: IV. Results from the testing of 300 chemicals. *Environ Mol Mutagen* 11(Suppl. 12):1–158.