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Safety Assessment of Glycerin as Used in Cosmetics

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Abstract

This is a safety assessment of glycerin as used in cosmetics. Glycerin functions as a denaturant, fragrance ingredient, hair conditioning agent, humectant, oral care agent, oral health-care drug, skin protectant, skin conditioning agent—humectant, and viscosity-decreasing agent. The Cosmetic Ingredient Review Expert Panel (Panel) reviewed relevant animal and human data. The Panel concluded that glycerin is safe as a cosmetic ingredient in the practices of use and concentration described in this safety assessment.

Keywords

cosmetics, safety, glycerin

Introduction

This is a review of the available scientific literature and submitted unpublished data relevant to assessing the safety of glycerin as used in cosmetics. Glycerin is reported to function in cosmetics as a denaturant, fragrance ingredient, hair conditioning agent, humectant, oral care agent, oral health-care drug, skin protectant, skin conditioning agent—humectant, and viscosity-decreasing agent.[1]

Much of the information in this safety assessment was obtained from summary information made available by the Organization for Economic Cooperation and Development Screening Information Data Set, the European Commission-European Chemicals Bureau, and the European Chemicals Agency.[2-4]

Chemistry

Definition and Structure

Glycerin (CAS no. 56-81-5) is the polyhydric alcohol that conforms generally to the structure in Figure 1.[1] The molecular formula is $C_3H_8O_3$. Glycerin (also referred to as glycerol in the literature) is a simple triol, that is, it has 3 hydroxyl groups.

Glycerin is naturally occurring in all animals and plant matter in combined form as glycerides in fats and oils or in intracellular spaces as lipids.[3] While the chemicals are identical, there is naturally occurring glycerin derived from plants and animals and synthetic glycerin obtained from nontriglyceride sources.[5]

Physical and Chemical Properties

Glycerin is a clear, syrupy liquid (Table 1).[2,6] It can be in a crystallized state, but seldom is because of its tendency to supercool and the pronounced effect of small amounts of water in depressing the freezing point.[3]

Glycerin has solvent properties similar to those of water and simple aliphatic alcohols.[5] The chemical is completely miscible with water, methanol, ethanol, and the isomers of propanol, butanol, and pentanol. Glycerin is also fully miscible with phenol, glycol, propanediols, amines, and heterocyclic compounds containing a nitrogen atom in the ring (eg, pyridine, quinoline). Glycerin is less soluble in acetone, diethyl ether, and dioxane and is almost insoluble in hydrocarbons, long-chain aliphatic alcohols, fatty oils, and halogenated solvents such as chloroform.

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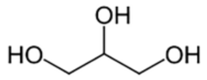

Figure 1. Glycerin.

Table 1. Chemical and Physical Properties of Glycerin.

Property	Value	Reference
Physical form	Liquid, syrupy	2,6
Color	Clear	6
Odor	Odorless, mild	5
Molecular weight (g/mol)	92.09	56
Density/specific gravity at 20°C	1.26	2
Viscosity (kg/s·m)	1.41	2
Vapor pressure (mm Hg) at 50°C	0.0025	6
Vapor density (mm Hg)	3.17	6
Melting point (°C)	18	2
	17.9	6
Boiling point (°C)	290	2
Water solubility	Miscible	2
Log K_{ow}	−1.76	2
Disassociation constants (pK_a)	0.07×10^{-13}	2

Method of Manufacture

Natural glycerin is obtained as a byproduct in the hydrolysis of fats and oils.[5] Multiple methods exist for the synthetic manufacture of glycerin. The starting materials for synthetic production of glycerin include allyl chloride, acrolein, propylene oxide, sugars, certain polyalcohols, fats, or epichlorohydrin.[5]

In one method, allyl chloride is oxidized with hypochlorite to produce dichlorohydrin, which is then converted, without isolation, to epichlorohydrin by ring closure with calcium hydroxide or sodium hydroxide.[5] Epichlorohydrin is hydrolyzed to yield glycerin by heating to 80°C to 200°C with a 10% to 15% aqueous solution of sodium hydroxide or sodium carbonate at atmospheric pressure or overpressure. The yield of glycerin, calculated from allyl chloride, is 98%, obtained as a dilute (10%-25%) solution containing 5% to 10% sodium chloride and <2% other impurities. The aqueous glycerin solution is concentrated in a multistage evaporation plant under vacuum to produce a glycerin concentration of >75%, after separating precipitated sodium chloride. The glycerin solution is then distilled under high vacuum and the co-distilled water is separated by fractional condensation. The glycerin is treated further to remove color impurities and odorous material; this can be accomplished, for example, using activated carbon.

A second method involves the oxygenation of propene to acrolein, which is then reduced under Meerwein-Ponndorf-Verley conditions to yield allyl alcohol.[5] The allyl alcohol is then epoxidated with hydrogen peroxide and the resulting glycidol is hydrolyzed to produce glycerin.

Impurities

The US Pharmacopeia-National Formulary standards state that the amount of any individual impurity in glycerin cannot exceed 0.1% and that the total for all impurities, including diethylene glycol and ethylene glycol, must not exceed 1%.[7]

The US Food and Drug Administration (FDA) notes that glycerin is a byproduct of biodiesel fuel produced from the *Jatropha* species of plant.[8] There is a possibility that toxic impurities, including phorbol esters, may be present in glycerin produced this way. Conventional impurity tests may not detect these toxins, and glycerin from this source should therefore not be used in human and animal food, medical products, cosmetics, and other FDA-regulated products. The FDA advises industry to be aware of the potential for substitution or use of oils, glycerin, and proteins derived from the *Jatropha* plant.

A cosmetics raw material supplier reported that glycerin is 95% to 99.5% pure.[2] Impurities are water and trace levels of polyglycerol.

Use

Cosmetic

Glycerin is reported to function in cosmetics as a denaturant, fragrance ingredient, hair conditioning agent, humectant, oral care agent, oral health-care drug, skin protectant, skin conditioning agent—humectant, and viscosity-decreasing agent.[1]

The FDA collects information from manufacturers on the use of individual ingredients in cosmetics as a function of cosmetic product category in its Voluntary Cosmetic Registration Program (VCRP). In 2014, glycerin was the third most frequently reported ingredient in the VCRP database (after water and fragrance). Glycerin was reported to be used in 15,654 cosmetic products; 10,046 are leave-on products, 5,441 are rinse-off products, and 167 products are diluted for the bath (Table 2). These uses include 862 products for use near the eye, 160 lipsticks, 369 hair dyes and colors, 1,259 bath soaps and detergents, 7,756 skin care products, and 244 suntan preparations.[9] Glycerin is reported to be used in 125 baby products. Two uses for anhydrous glycerin (in an eye makeup preparation and a moisturizer) were reported in the VCRP; these uses were merged with the glycerin uses.

A survey was conducted by the Personal Care Products Council (Council) of the maximum use concentrations for this ingredient.[10] Glycerin is reported to be used at concentrations up to 79.2% in leave-on products, 99.4% in rinse-off products, and 47.9% in products diluted for the bath. It is used at up to 21% in baby products, 40.6% in eye lotions, 25% in perfumes, 47.3% in hair grooming aids, 68.6% in oral hygiene products, 99.4% in skin cleaning products, and 17.9% in suntan preparations.

Glycerin was reported to be used in aerosol/spray products that include hair sprays at concentrations at up to 30% (in propellant spray products at concentrations up to 10% and in pump spray products up to 30%), pump deodorants at up to 4%, face and neck products at up to 10%, body and hand products at

Table 2. Frequency and Concentration of Use According to Duration and Exposure of Glycerin.[a,9,10]

Use type	Uses	Maximum concentration (%)
Total/range	15,654	0.0001-99.4
Duration of use		
Leave-on	10,046	0.0001-79.2
Rinse-off	5,441	0.0007-99.4
Diluted for (bath) use	167	0.66-47.9
Exposure type[b]		
Eye area	862	0.025-40.6
Incidental ingestion	353	2-68.6
Incidental inhalation sprays	531	0.006-30c
	3,810[d]	0.075-47.3[d]
	2,643[e]	1.1-77.3[e]
Incidental inhalation powders	69	0.003-15
	53[f]	4.5-79.2[f]
	2,643[e]	1.1-77.3[e]
Dermal contact	12,710	0.003-99.4
Deodorant (underarm)	136[c]	0.1-10.4[g]
		0.019-4[h]
Hair noncoloring	1,911	0.015-47.3
Hair coloring	490	0.0007-20
Nail	57	0.0001-45
Mucous membrane	2,597	0.66-68.6
Baby	125	0.23-21

[a]Totals = rinse-off + leave-on + diluted for bath product uses.
[b]Because each ingredient may be used in cosmetics with multiple exposure types, the sum of all exposure type uses may not equal the sum total uses.
[c]Aerosol hair spray 0.11% to 10%; pump hair spray 0.11% to 30%; spray face and neck products 0.5% to 10%; spray body and hand product 0.006% to 5%; spray moisturizers 3.3%; aerosol suntan products 6%; pump spray suntan products 4.1% to 10%.
[d]It is possible these products *may* be sprays, but it is not specified whether the reported uses are sprays.
[e]Not specified whether a powder or a spray, so this information is captured for both categories of incidental inhalation.
[f]It is possible these products *may* be powders, but it is not specified whether the reported uses are powders.
[g]Not spray.
[h]Aerosol spray 0.019% to 0.05%; pump spray 2% to 4%.

up to 5%, moisturizing products at up to 3.3%, and suntan products at up to 10% (in propellant spray products at up to 6% and in pump spray products up to 10%). These propellant/pump spray products could possibly be inhaled. In practice, 95% to 99% of the droplets/particles released from cosmetic sprays have aerodynamic equivalent diameters >10 µm, with propellant sprays yielding a greater fraction of droplets/particles below 10 µm compared with pump sprays.[11-14] Therefore, most droplets/particles incidentally inhaled from cosmetic sprays would be deposited in the nasopharyngeal and bronchial regions and would not be respirable (ie, they would not enter the lungs) in any appreciable amount.[11,13] There is some evidence indicating that deodorant spray products can release substantially larger fractions of particulates having aerodynamic equivalent diameters in the range considered to be respirable.[11] However, the information is not sufficient to determine whether significantly greater lung exposures result from the use of deodorant sprays, compared to other cosmetic sprays.

Noncosmetic

Code of Federal Regulations (CFR) listings concerning glycerin are provided in Table 3. For example, glycerin is considered generally recognized as safe (GRAS) by the FDA for its use in food packaging and it is a multiple-purpose GRAS food substance when used in accordance with good manufacturing practices [21CFR182.90; 21CFR182.1320]. Also, glycerin is approved for use in over-the-counter drugs, such as anorectal drug products, dermal protectants (up to 45%), in ophthalmic drug products (up to 1%), and in oral health-care products [21CFR346.14; 21CFR347.10; 21CFR349.12].

Glycerin functions as a humectant, solvent, cake icing component, confectionary component, bodying agent, and plasticizer for foods.[3]

Glycerin has been administered orally and/or intravenously to reduce intracranial pressure caused by various medical conditions.[15] Glycerin has been used to reduce brain volume for neurosurgical procedures. It is also used as the active ingredient in laxative products (ie, glycerin suppositories).

Glycerin is used in paints, lacquers, and varnishes; polymers; tobacco products; absorbents and adsorbents; adhesives and binding agents; antifreezing agents; cleaning agents and disinfectants; explosives; heat transferring agents; pesticides; and softeners.[3] It is an intermediate and monomer in resins, polyols, and polyurethanes.[2]

Toxicokinetics

Absorption, Distribution, Metabolism, and Excretion

Glycerin is rapidly absorbed from the intestine and the stomach, distributed throughout the extracellular space, and excreted renally.[2,16,17] Glycerin is phosphorylated to α-glycerophosphate by glycerol kinase, predominantly in the liver (80%-90%) and kidneys (10%-20%), and incorporated via the standard metabolic pathways to form glucose (gluconeogenesis) and glycogen.[16,18] Glycerin kinase is also found in intestinal mucosa, brown adipose tissue, lymphatic tissue, lung, and pancreas. Glycerin may also combine with free fatty acids in the liver to form triglycerides (lipogenesis) that can be distributed to adipose tissues. The glycerin turnover rate is directly proportional to plasma glycerin levels.[19]

Free glycerin is naturally present in human plasma.[20] Normal serum levels in adult humans range from 0.05 to 0.1 mmol/L. Urinary glycerol excretion is associated with plasma glycerol concentrations >0.327 ± 0.190 mmol/L.

Dermal/percutaneous. Data on dermal absorption, distribution, metabolism, and excretion of glycerin were not found in the published literature nor were unpublished data provided.

Oral. Orally administered glycerin is rapidly absorbed from the gastrointestinal tract, and peak serum concentrations occur within 60 to 90 minutes.[15] Glycerin is distributed throughout the blood. Glycerin generally does not appear in ocular fluids; however, it may enter the orbital sac when the eye is inflamed.

Table 3. Code of Federal Regulations Listings Concerning Glycerin From FDA, EPA, and ATF.

Citation	Regulation
Food additive	
21CFR172.866	FOOD ADDITIVES PERMITTED FOR DIRECT ADDITION TO FOOD FOR HUMAN CONSUMPTION Synthetic glycerin produced by the hydrogenolysis of carbohydrates may be safely used in food, subject to the provisions of this section: (a) It shall contain not in excess of 0.2% by weight of a mixture of butanetriols. (b) It is used or intended for use in an amount not to exceed that reasonably required to produce its intended effect.
21CFR182.1320	SUBCHAPTER B—FOOD FOR HUMAN CONSUMPTION SUBSTANCES GENERALLY RECOGNIZED AS SAFE Subpart B—Multiple-Purpose GRAS Food Substances (a) Product. Glycerin. (b) Conditions of use. This substance is generally recognized as safe when used in accordance with good manufacturing practice.
Indirect food additive	
21CFR175.300	INDIRECT FOOD ADDITIVES: ADHESIVES AND COMPONENTS OF COATINGS Resinous and polymeric coatings may be safely used as the food contact surface of articles intended for use in producing, manufacturing, packing, processing, preparing, treating, packaging, transporting, or holding food, in accordance with the following prescribed conditions: (1) The coating is applied as a continuous film or enamel over a metal substrate, or the coating is intended for repeated food contact use and is applied to any suitable substrate as a continuous film or enamel that serves as a functional barrier between the food and the substrate. The coating is characterized by one or more of the following descriptions: (1) Coatings cured by oxidation. (2) Coatings cured by polymerization, condensation, and/or cross-linking without oxidation. (3) Coatings prepared from prepolymerized substances. (b) The coatings are formulated from optional substances that may include: (1) Substances generally recognized as safe in food. (3) Any substance employed in the production of resinous and polymeric coatings that is the subject of a regulation in subchapter B of this chapter and conforms with any specification in such regulation. Substances named in this paragraph (b)(3) and further identified as required: (*b*) Rosin esters formed by reacting rosin (paragraph (b)(3)(v)(*a*) of this section) with: Glycerol (*c*) Polyhydric alcohols: Glycerol Trimellitic anhydride adducts of ethylene glycol and glycerol, prepared by the reaction of 1 mole of trimellitic anhydride with 0.4 to 0.6 mole of ethylene glycol and 0.04 to 0.12 mole of glycerol, for use only as a cross-linking agent at a level not to exceed 10% by weight of the cured coating, provided that the cured coating only contacts food containing not more than 8% alcohol. Glycerol (ii) Reconstituted oils from triglycerides or fatty acids derived from the oils listed in paragraph (b)(3)(i) of this section to form esters with: (iv) Natural fossil resins, as the basic resin: Glycerol ester of damar, copal, elemi, and sandarac. (v) Rosins and rosin derivatives, with or without modification by polymerization, isomerization, incidental decarboxylation, and/or hydrogenation, as follows: (*b*) Rosin esters formed by reacting rosin (paragraph (b)(3)(v)(*a*) of this section) with: Glycerol. (*c*) Polyhydric alcohols: Glycerol.
21CFR178.3500	INDIRECT FOOD ADDITIVES: ADJUVANTS, PRODUCTION AIDS, AND SANITIZERS Synthetic glycerin may be safely used as a component of articles intended for use in packaging materials for food, subject to the provisions of this section: (a) It is produced by the hydrogenolysis of carbohydrates and shall contain not in excess of 0.2% by weight of a mixture of butanetriols. (b) It is used in a quantity not to exceed that amount reasonably required to produce its intended physical or technical effect, and in accordance with any limitations prescribed by applicable regulations in parts 174, 175, 176, 177, 178, and 179 of this chapter. It shall not be intended to, nor in fact accomplish, any direct physical or technical effect in the food itself.
21CFR182.90	SUBSTANCES GENERALLY RECOGNIZED AS SAFE Substances migrating to food from paper and paperboard products. Substances migrating to food from paper and paperboard products used in food packaging that are generally recognized as safe for their intended use, within the meaning of section 409 of the Act, are as follows: Glycerin

(continued)

Table 3. (continued)

Citation	Regulation
Drug	
21CFR346.14	ANORECTAL DRUG PRODUCTS FOR OVER-THE-COUNTER HUMAN USE Active ingredients Protectant active ingredients. (a) The following active ingredients may be used as the sole protectant active ingredient in a product if the ingredient as identified constitutes 50% or more by weight of the final product. In addition, the following active ingredients may be used in concentrations of less than 50% by weight only when used in combinations in accordance with 346.22 (a), (b), or (n). (3) Glycerin in a 20% to 45% (wt/wt) aqueous solution so that the final product contains not less than 10 and not more than 45% glycerin (wt/wt). Any combination product containing glycerin must contain at least this minimum amount of glycerin.
21CFR347.10	SKIN PROTECTANT DRUG PRODUCTS FOR OVER-THE-COUNTER HUMAN USE Active ingredients Skin protectant active ingredients. The active ingredients of the product consist of any of the following, within the concentration specified for each ingredient: (h) Glycerin, 20% to 45%.
21CFR349.12	OPHTHALMIC DRUG PRODUCTS FOR OVER-THE-COUNTER HUMAN USE Active ingredients The active ingredients of the product consist of any of the following, within the established concentrations for each ingredient: (d) Polyols, liquid: (1) Glycerin, 0.2% to 1%. Demulcents.
Agriculture	
21CFR582.1320	SUBCHAPTER E—ANIMAL DRUGS, FEEDS, AND RELATED PRODUCTS SUBSTANCES GENERALLY RECOGNIZED AS SAFE General-purpose food additives (a) Product. Glycerin. (b) Conditions of use. This substance is generally recognized as safe when used in accordance with good manufacturing or feeding practice.
40CFR180.1250	Glycerin is used in several pesticide applications, including those applied to food and feed crops.
40CFR180.910 40CFR180.920 40CFR180.930 40CFR180.950	Glycerin is exempted from the requirement of a tolerance when used in accordance with good agricultural practice as inert (or occasionally active) ingredients in pesticide formulations applied to growing crops only.
Other	
27CFR21.58	Alcohol, Tobacco Products, and Firearms. CHAPTER I—ALCOHOL AND TOBACCO TAX AND TRADE BUREAU, DEPARTMENT OF THE TREASURY ALCOHOL. PART 21—FORMULAS FOR DENATURED ALCOHOL AND RUM. Subpart D—Specially Denatured Spirits Formulas and Authorized Uses (a) Formula. To every 100 gallons of alcohol add: One hundred pounds of glycerin (glycerol), USP., and 20 pounds of hard soap, NF XI. (b) Authorized uses. (1) As a solvent: 113. Lotions and creams (hands, face, and body). 131. Tooth paste and tooth powder. 141. Shampoos.
27CFR21.151	Title 27—Alcohol, Tobacco Products, and Firearms. CHAPTER I—ALCOHOL AND TOBACCO TAX AND TRADE BUREAU, DEPARTMENT OF THE TREASURY. SUBCHAPTER A—ALCOHOL. PART 21—FORMULAS FOR DENATURED ALCOHOL AND RUM. Subpart G—Denaturants Authorized for Denatured Spirits. List of denaturants authorized for denatured spirits. Context: Glycerin (glycerol), USP; Specially Denatured Alcohol. 31-A.

Abbreviations: ATF, Alcohol and Tobacco Tax and Trade Bureau; CFR, Code of Federal Regulations; EPA, Environmental Protection Agency; GRAS, generally recognized as safe; USP, US Pharmacopeia.

It is not known if glycerin is distributed into milk. The elimination half-life of glycerin is approximately 30 to 45 minutes. Most orally administered glycerin is incorporated into body fat or brought into glycolysis or gluconeogenesis pathways (principally in the liver) via glycerokinase-catalyzed phosphorylation and oxidation to dihydroxyacetone phosphate, with ultimate conversion to carbon dioxide and water in the former pathway, and glucose and glycogen synthesis in the latter. Glycerin can also combine with free fatty acids to form triglycerides. Approximately 80% of glycerin metabolism

Table 4. Highest NOAEL Reported in a Survey of 4 Research Organizations for Glycerin Used as a Vehicle.[22]

Animal	Route	Duration	Dose	Comments
Rat	Oral	Not provided	1,000 mg/kg	Well tolerated
		1 month	15 g/kg	Reduced adrenal weight
		1 month	1,000 mg/kg	Well tolerated
Guinea pig	Oral	1 month	500 mg/kg	Well tolerated
Mouse	Oral	90 days	50 mg/kg	Depression and reduced respiration
	Intravenous	1 month	100 mg/kg	Well tolerated
	Subcutaneous	Acute	10 mg/kg	Well tolerated
	Intraperitoneal	1 month	250 mg/kg	Well tolerated
Rabbit	Intravenous	Not provided	10 mg/kg	Well tolerated

Abbreviation: NOAEL, no-observed-adverse-effect level.

takes place in the liver and approximately 10% to 20% in the kidney. The metabolism of glycerin to carbohydrate produces 4.3 cal/g glycerin. Most of an oral dose of glycerin is metabolized within 2.5 hours. Approximately 7% to 14% of an oral dose of glycerin is excreted unchanged in urine during this time.

Orally administered glycerin elevates the osmotic pressure of the plasma to such an extent that water from the extravascular spaces is drawn into the blood. The osmotic effect of glycerin produces a decrease in intraocular pressure (IOP) by reducing the volume of intraocular fluids in a manner completely independent of the normal ocular fluid inflow and outflow mechanisms. The extent of IOP reduction varies with the dose of glycerin and the etiology and degree of the increased pressure. Reduction in IOP reaches its maximum within 30 minutes to 2 hours and may persist for 4 to 8 hours. In general, reduction in IOP is greatest when the pretreatment IOP is high. The osmotic effect of glycerin may also produce tissue dehydration and a decrease in cerebrospinal fluid pressure. Glycerin produces only very slight diuresis in healthy individuals receiving a single oral dose of 1.5 g/kg or less.[15]

Acute ingestion of glycerin (1 mL/kg in water) in human male patients led to an increase in plasma glycerides. In human female patients, the oral administration of glycerin (1 mL/kg in water) resulted in no change in plasma glyceride concentrations. When glycerin (1 mL/kg/d in 3 doses) was orally administered for 42 days, increased serum glyceride concentrations were observed in both sexes; however, the increase was greater in men.[21]

Toxicological Studies

In a survey of 4 laboratories of the use of vehicles for in vivo experiments, the highest no-observed-adverse-effect levels (NOAELs) of various routes of administration were assembled.[22] The oral NOAELs for glycerin were 15 g/kg for rats and 500 mg/kg for guinea pigs for 1 month and 500 mg/kg for 90 days for mice (Table 4). The highest subcutaneous NOAELs were 10 mg/kg in rats and mice.

Acute Toxicity

Nonhuman. Reported oral median lethal dose (LD_{50}) values of glycerin ranged from 2,530 to 58,400 mg/kg in rats; there were no deaths at 24,000 mg/kg in one study (Table 5).[3,18,23-27] Reported oral LD_{50} values of glycerin were 4,090 to 38,000 mg/kg in mice, 27,000 mg/kg in rabbits, and 77,500 mg/kg in guinea pigs.[3,18,23,24,26-29] The dermal LD_{50} value of glycerin was reported to be >21,900 mg/kg in rats and >18,700 mg/kg in rabbits.[3,26] The approximate value for the median lethal time (time until death) after exposure (Lt_{50}) for rats was determined to be 423 minutes for exposure to glycerin vapors at 11.0 mg/L.[4] Reported intraperitoneal LD_{50} values of glycerin were 4,420 to 10,100 mg/kg in rats and 8,600 to 9,500 mg/kg in mice.[3] Reported subcutaneous LD_{50} values of glycerin were 100 mg/kg in rats and 91 to 100 mg/kg in mice.[3,29] Reported intravenous LD_{50} values of glycerin were 5,200 to 6,600 mg/kg in rats, 4,250 to 6,700 mg/kg in mice, and 53,000 mg/kg in rabbits.[3]

Oral—Human. The oral lowest lethal dose (LD_{LO}) of glycerin was reported to be 1,428 mg/kg for humans.[3] There were no signs of toxicity when patients (n = 10 men, 4 women) were orally administered glycerin (30 mL; 95% purity administered in orange juice) after each of 3 meals in 1 day.[2]

When used as a drug, it was reported that the adverse effects following the oral administration of glycerin (dose not provided) include mild headache, dizziness, nausea, vomiting, thirst, and diarrhea.[15] Headache may result from cerebral dehydration, which may be prevented or relieved by having the patient lie down during and after treatment. Hypotonic fluids relieve thirst and headache caused by the dehydrating action of glycerin.

Repeated Dose Toxicity

Oral—Nonhuman. Repeated dose toxicity studies are summarized in Table 6. Undiluted glycerin caused a dose-dependent increase in the number of animals showing hyperemia, petechial hemorrhage, and erosions in the gastrointestinal tract.[30] In short-term feeding experiments, glycerin at 20% for 4 weeks in feed produced no adverse effects, but at 53.4%, an increase in

Table 5. Acute Toxicity Studies of Glycerin.

Animal (n, if provided)	Results and notes	Reference
Oral		
Rat	$LD_{50} > 10,000$ mg/kg	3
Rat	$LD_{50} = 12,600$ mg/kg	3
Long-Evans, rat, female (12)	$LD_{50} = 27,200$ mg/kg for both natural and synthetic glycerin. Purity of both test materials = 99.5% administered neat.	26
	Clinical signs included muscle spasms and convulsions. Survivors appeared normal within 2.5 hours of dosing. Number of deaths was not reported. Macroscopic examination of decedents and survivors showed hyperemia of the pylorus, small intestine and cerebral meninges (3 rats), and congestion of the lungs and pale spleen	
Sprague Dawley rat (10)	$LD_{50} > 2,530$ mg/kg	23
Fischer 344 rat, female (5)	$LD_{50} > 24,000$ mg/kg glycerin/water mixture of unknown composition	25
	No deaths at 48 hours	
Wistar rat, male (10)	$LD_{50} = 27,500$ mg/kg	27
Rat	$LD_{50} > 25,000$ mg/kg	18
Rat	$LD_{50} = 58,400$ mg/kg	24
NMRI mouse, male and female (10)	$LD_{50} = 37,950$ mg/kg	23
Mouse	$LD_{50} = 4,090$ mg/kg	3
Mouse	$LD_{50} \sim 26,000$ mg/kg. LD_{50} for natural glycerin = 20.65 mL/kg; LD_{50} for synthetic glycerin = 20.81 mL/kg. Purity of the synthetic glycerin = 99.8%	28
Mouse	$LD_{50} = 38,000$ mg/kg	23
Swiss mouse, male	$LD_{50} = 23,000$ for both natural and synthetic glycerin. Purity of both test materials = 99.5% administered neat	26
	Body tremors, erection of the tail, and generalized clonic convulsions preceded all observed deaths of mice	
Mouse	$LD_{50} = 4,250$ mg/kg	3
Mouse	$LD_{50} > 38,000$ mg/kg	18
Mouse	$LD_{50} = 37,763$ mg/kg	24
Mouse	$LD_{50} = 25,888$ mg/kg	24
Mouse	$LD_{50} = 12,500$ mg/kg	29
Mouse	$LD_{50} = 25,000$ mg/kg	29
Rabbit	$LD_{50} = 27,000$ mg/kg	3
Guinea pig (9-10)	$LD_{50} = 10,000 \pm 130$ mg/kg for natural glycerin and $11,500 \pm 2,800$ mg/kg for synthetic glycerin. Purity of both test materials = 99.5%; administered neat.	26
	Tremors of the head and body, initiated by auditory stimuli, occurred immediately after injection. Death was usually preceded by tremors, but not all guinea pigs with tremors died.	
Guinea pigs (10)	$LD_{50} = 77,500$ mg/kg	27
Dermal		
Rat	$LD_{50} > 21,900$ mg/kg. 2.52 g of the neat liquid (21,900 mg/kg) for more than 20 minutes produced excretion of hemoglobin in the urine of male rats, indicating red blood cell damage	3
	No deaths	
Rabbit (6)	$LD_{50} > 18,700$ mg/kg for both natural and synthetic glycerin. Purity for both natural and synthetic glycerin = 99.5%. Glycerin administered under occlusion for 8 hours	26
	No clinical signs were observed for either synthetic or natural glycerin	
Guinea pig (1, 8, 5, 5)	LD_{50} was determined to be 45 mL/kg. Glycerin (16, 40, 50, and 60 ml/kg) remained in contact for 3 to 4 days under semiocclusion. Animals were observed for 2 weeks after removal of the dermal bandage	4
	After about 12 hours, the animals became accustomed to the restriction of the bandage and ate and in general behaved as usual, those having large doses being weak with low body temperature and dying; those with small doses apparently not being affected. Survival: 16 mL/kg, 0 or 1; 40 mL/kg, 3 of 8; 50 mL/kg, 4 of 5; 60 mL/kg, 3 of 5	
Inhalation		
Rat, male (5)	Based on the time-to-death data, an approximate Lt_{50} was determined to be 423 minutes for exposure calculated to be 11.0 mg/L.	4

(continued)

Table 5. (continued)

Animal (n, if provided)	Results and notes	Reference
	Exposed to vapors produced by heating glycerin to 200°C for 1, 2, or 7 hours. During the initial 4 hours, no adverse effects were observed. After approximately 240 minutes, clinical signs were mild irritation, indicated by hyperemia of the eyelids and nasal discharges. These signs became more severe as the exposure progressed. The rats eventually salivated profusely and exhibited dyspneic respiratory patterns. At approximately 360 minutes, all rats began gasping for air and lay in prostration. Two animals died at approximately the 419-minute time point. The third one died shortly after the termination of the exposure. The fourth animal succumbed at approximately 30 minutes postexposure. The remaining animal succumbed on the fourth postexposure day.	
Intraperitoneal		
Rat	LD_{50} = 7,500-10,100 mg/kg	3
Rat	LD_{50} = 4,420 mg/kg	3
Mouse	LD_{50} = 8,600-9,500 mg/kg	3
Mouse	LD_{50} = 8,700 mg/kg	3
Subcutaneous		
Rat	LD_{50} = 100 mg/kg	3
Mouse	LD_{50} = 91 mg/kg	3
Mouse	LD_{50} = 100 mg/kg	29
Intravenous		
Rat	LD_{50} = 5,200-6,600 mg/kg	3
Rat	LD_{50} = 5,566 mg/kg	3
Mouse	LD_{50} = 5,700-6,700 mg/kg	3
Mouse	LD_{50} = 4,250-4,370 mg/kg. LD_{50} for natural glycerin = 4.37 g/kg; LD_{50} for synthetic glycerin = 4.25 g/kg. Purity of the synthetic glycerin = 99.8%.	3
Mouse	LD_{50} = 4,250 mg/kg	3
Mouse	LD_{50} = 6,000 mg/kg	29
Rabbit	LD_{50} = 53,000 mg/kg	3

kidney weights and increased liver enzymes were observed.[4,30-32] The NOAEL was between 115 and 2,300 mg/kg when administered in water to rats for 44 days.[33] Calcified masses were observed in kidney tubules between the cortex and medulla in 3 of 5 rats administered either natural or synthetic glycerin (3,335 mg/kg/d) in drinking water for 6 months.[28] When glycerin was administered in the diet for 2 years, feed consumption was slightly increased in males fed synthetic glycerin versus those fed naturally sourced glycerin. There were no treatment-related effects in organ weights and gross pathology.[26]

The no-observed-effect-level (NOEL) in mongrel dogs was 950 mg/kg/d when glycerin was orally administered for 3 days (Table 6). At 3,800 mg/kg/d, the mucosa of the stomach was severely hyperemic with petechial hemorrhages.[34] Mongrel dogs experienced weight loss after 36 weeks when glycerin (35%) was incorporated into their feed. The weight loss continued when the glycerin content was reduced by 50% to 80% for the remainder of a 50-week study.[30]

There were no pathological changes in guinea pigs (n = 10) orally administered glycerin (6,300 mg/kg/d) for 30 to 40 days (Table 6).[35]

Oral—Human. There were no signs of toxicity or effects on blood or urine production when patients (n = 10 males, 4 females) were orally administered glycerin (approximately 1.3-2.2 g/kg/d; glycerin in orange juice with meals) for 50 days.[3] The NOAEL was $\geq$2.2 g/kg/d. No further information was provided. There were no adverse effects observed in patients (n = 14) administered glycerin (30 mL, neat) 3 times daily with each meal for 50 days.[16]

Dermal—Nonhuman. There were no treatment effects when glycerin (100%; 0.5-4 mL) was administered to 30% of the body surfaces of rabbits 5 d/wk for 45 weeks (Table 6).[26]

Inhalation—Nonhuman. The inhalation lowest-observed-adverse-effect-level (LOAEL) was 1,000 mg/L for glycerin administered nose only 6 h/d, 5 d/wk for 2 weeks in Crl: DCD Sprague Dawley rats, based on local effects on the epithelium of the upper respiratory tract (Table 6).[36]

The inhalation NOAEL was 0.167 mg/L for glycerin-administered nose only for 5 h/d, 5 d/wk for 13 weeks in Crl: DCD Sprague Dawley rats (Table 6).[36] There was minimal squamous metaplasia of the epiglottis in 2 of 25, 1 of 19, 4 of 20, and 10 of 21 rats at 0, 33, 167, and 662 mg/L, respectively; 1 male in the high-dose group showed mild squamous metaplasia.

Reproductive and Developmental Toxicity

In a 2-generation reproductive study in rats (n = 10 per sex), the administration of glycerin (0%, 20%; $\sim$2,000 mg/kg/d in drinking water) for 8 weeks before mating until weaning of pups produced no adverse effects on the reproductive efficiency of the parents (F_0 generation), or the growth, fertility,

Table 6. Repeated-Dose Toxicity Studies.

Animal	n	Dose	Results and notes	References
Oral				
Charles River rat, female	10, 20 controls	0, 0.75, 1.5, or 3.0 mg/kg glycerin; 100% by stomach tube; or 3.00 mg/kg at 20%, 40%, 60%, or 80% 3 times/day for 3 days.	Undiluted glycerin caused a dose-dependent increase in the number of animals showing hyperemia, petechial hemorrhage, and erosions in the gastrointestinal tract. Dilution with distilled water of glycerin reduced the effects in a dose-dependent manner.	30
Wistar rat, male	24, 18 controls 3 control and 4 treatment rats were killed and necropsied at 6 times	Glycerin replaced the 53.4% carbohydrate in feed for 20 days. Controls had stock carbohydrate or were fed a stock diet calculated to deliver the same calories as the glycerin diet. A rat was necropsied each day at the same time. Several enzymes, including glycerol kinase, were assayed.	Controls gained weight at 6% per day, glycerin-fed and pair-fed controls gained 3% per day. Livers were 6% body weight in the glycerin-fed rats and 4.6% in the other 2 groups. Kidneys of the glycerin-fed rats were 45% heavier than normal; the fat pad was normal. Enzymes (eg, pyruvate kinase, phosphofructokinase) increased and remained high in activity in the kidneys and livers of the glycerin-fed rats. At the end of the experiment, 95% of the ingested glycerin was being retained and blood glucose and liver glycogen were above normal. Water intake and urine production in the glycerin-fed rats were approximately 5 times higher than the normal.	31
Carworth rat, male	Not specified	Diet containing 20% glycerin (8,824 mg/kg body weight/d) for 4 weeks. At the end of 4 weeks, 5 rats were killed and necropsied. Both epididymis fat pads were excised, dried, and weighed. Liver total lipids and cholesterol were assessed.	No adverse effects were observed with regard to weight gain, feed intake, epididymal adipose tissue dry weight, and total liver lipids and cholesterol.	32
Rat, male	20	0, 1%, 5%, 10%, 20% (115, 575, 1,150, and 2,300 mg/kg aqueous); 1 mL; for 44 days	No adverse effects were observed for growth curves, lethality, and histological examination of the kidneys, livers, and bladders. Mortality was 15% in all groups.	33
Rat, male and female	10/sex	Diet containing 5% or 20% for 90 days from 3 different manufacturers	5% glycerin was tolerated without any evidence of adverse effect; 20% glycerin showed slight pathological changes in the liver. In general, all the groups of treated rats grew faster and bigger than the controls. No adverse effects were noted in the low-dose group from any source. In the male rats in the high-dose group, there was an increase in the final liver/body weight ratio, and upon microscopic examination, generalized cloudy swelling and hypertrophy of the parenchymal cells was observed. Female rats in this group exhibited some generalized cloudy swelling upon microscopic examination of the liver.	4
Rat, female	5	5% natural and synthetic glycerin in drinking water (3,335 mg/kg/d) for 6 months.	No effects on growth, red blood cells, and hemoglobin. Macroscopic incidental findings were a small thymus in 2 rats and slight interstitial pneumonia in one on natural glycerin and small spleen (with small lymph nodes and moderate hemosiderin deposits) and thymus atrophia in one animal that died on synthetic glycerol. Calcified masses in kidney tubules between cortex and medulla in 3/5 rats on natural glycerin and 3/5 rats on synthetic glycerin.	28
Long-Evans rat, male and female (22/sex; 26 controls)	6-7	Diet containing 0%, 5%, 10%, or 20% natural or synthetic glycerin for 2 years. Purity 99.5%.	Feed consumption was increased in males at 5% and 10% natural glycerin. No treatment-related effects in hematology, urinalysis, albumin, organ weights, gross pathology, and liver glycogen and lipids. Incidental bronchiectasis, pneumonia, pulmonary abscesses, *taenia* infestation of the liver, hydronephrosis, and pyelonephritis (27 rats total).	26

(continued)

Table 6. (continued)

Animal	n	Dose	Results and notes	References
Rabbit	4	0% or 50%, 10 mL in saline or saline by stomach tube or from a drinking cup daily for 30 to 40 days.	No adverse effects. Well tolerated. Necropsy at the end of the experiment showed no gross pathological changes. Neither the plasma nor the red blood cell cholesterol levels showed any consistent changes which could be attributed to glycerin.	35
Mongrel dog, male and female	Not specified	950, 1,900, and 3,800 mg/kg 3 times/day for 3 days	NOEL = 950 mg/kg. At 950 mg/kg body weight, no abnormalities. At 1,900 mg/kg, stomach mucosa was severely hyperemic with petechial hemorrhages. At 3,800 mg/kg, stomach mucosa was slightly to severely hyperemic with areas with petechial hemorrhages or erosions; duodenum appeared normal or with hyperemic areas.	30
Dog	Not specified	0% or 35% in feed for 50 weeks, then reduced to 50% to 80% of previous dose.	Body weight similar between groups until week 36, then after week 36 weight loss (16%, 1.8 kg) in dogs on glycerin-rich diet, but not in controls. Erythrocyte counts were similar between groups.	34
Guinea pig	10	0% or 50% in saline (=6,300 mg/kg/d) by stomach tube or from a drinking cup daily for 30 to 40 days.	Guinea pigs administered >5 mL of the 50% glycerin solution by stomach tube died with acute symptoms. Necropsies revealed no pathological changes. Plasma cholesterol levels had no changes attributable to glycerin. Red blood count of 3 guinea pigs (2 stomach tube, 1 drinking water) indicated a probable anemic effect.	35
Dermal				
Rabbit	12	90 administrations of 0.5 or 4 mL of either natural or synthetic glycerin (8 h/d 5 d/wk, 45 weeks). Administered to 30% of the body surface. Purity of both = 99.5%.	No treatment-related effects at 100% with regard to weigh gain, urinalysis, blood counts, skin condition, or grow and microscopic examination at necropsy.	26
Inhalation				
Sprague Dawley Crl: CD Rat, male/female	10/sex	0, 1,000, 2,000, 4,000 mg/L for 6 h/d, 5 d/wk for 2 weeks. Nose-only exposure. Particle size = <1.5 µm.	LOAEL = 1,000 mg/m^3 based on local effects on the epithelium of the upper respiratory tract. Two males at 1,000 mg/m^3 and 1 male and 1 female at 2,000 mg/m^3 died. No clinical signs observed. Body weight gains were decreased in males and females at all concentrations (28%-58% in females). Glucose decreased in females at all concentrations (19%-28%). No treatment-related effects for hematology, organ weights, and gross pathology. Histopathology: Minimal-to-mild squamous metaplasia of the epiglottis in males and females (1/10, 13/18, 16/19, and 13/14, respectively). No dose-related increase in the frequency, but the incidence of mild metaplasia was greatest in the high dose (7 animals with minimal and 6 with mild).	36
Sprague Dawley Crl: DCD rat, male/female	15/sex	0, 0.033, 0.167, 0.662 mg/L for 5 h/d, 5 d/wk, for 13 weeks. Purity >99.8%, particle size <2.0 µm. Nose-only study.	NOAEL = 0.167 mg/L. Minimal-to-mild squamous metaplasia of the epithelium lining the base of the epiglottis at the high dose. Three per sex necropsied at 10 and 13 weeks to examine lungs with electron microscope. No clinical signs or mortalities. No treatment-related effects for body weights, clinical chemistry, hematology, organ weights, and gross pathology. Histopathology at 13 weeks: Minimal squamous metaplasia of the epiglottis in 2/25, 1/19, 4/20, and 10/21 rats, respectively; 1 male at 662 mg/L showed mild squamous metaplasia. No differences in morphology of the Clara cells in control and high-dose rats at histopathological examination of the lungs.	36

Abbreviations: LOAEL, lowest-observed-adverse-effect-level; NOEL, no-observed-effect-level.

or reproductive performance of the untreated F_1 generation.[2] No histological changes occurred in the tissues of either the F_1 or F_2 generations. The onset of estrus cycles, weight gain, and microscopic observations of the endocrine organs were comparable to those of the controls in both the F_1 and the F_2 generation. In the F_0 generation, all 10 females became pregnant with similar litter size as controls (9.0 vs 8.1). In the F_1 generation, 9 of 10 females became pregnant.

When glycerin (13.1, 60.8, 282, and 1,310 mg/kg/d) was administered by gavage to Wistar rats (n = 25-28) on days 6 through 15 of gestation, there were no adverse effects observed in the dams.[37] The NOAEL for maternal toxicity and teratogenicity was 1,310 mg/kg/d. The numbers of pregnancies were 23 of 25, 24 of 25, 22 of 28, and 22 of 25 for 13.1, 60.8, 282, and 1,310 mg/kg/d, respectively, and 21 of 25 for controls. The number of implantations, resorptions, litter sizes, weights, and sex ratio were similar among groups, as were the incidences of external, visceral, and skeletal abnormalities.

When glycerin (12.8, 59.4, 276, and 1,280 mg/kg/d) was administered by gavage to CD-1 mice (n = 25) on days 6 through 15 of gestation, there were no adverse effects observed in the dams.[37] The NOAEL for maternal toxicity and teratogenicity was 1,280 mg/kg/d. The numbers of pregnancies were 14 of 15, 12 of 15, 10 of 18, 13 of 20, and 13 of 15 for controls, 12.8, 59.4, 276, and 1,280 mg/kg, respectively. The number of implantations, resorptions, litter sizes, weights, and sex ratio were similar among groups as were external, visceral, and skeletal abnormalities.

When glycerin (11.8, 54.8, 254.5, and 1,180 mg/kg/d) was administered by gavage to Dutch-belted rabbits (n = 25) on days 6 through 18 of gestation, there were no adverse effects found in the dams.[37] The NOAEL for maternal toxicity and teratogenicity was 1,180 mg/kg/d. The numbers of pregnancies were 22 of 25, 23 of 25, 20 of 25, 22 of 25, and 21 of 25 for controls, 11.8, 54.8, 254.5, and 1,180 mg/kg/d, respectively. The number of implantations, resorptions, litter sizes, weights, and sex ratio were similar among groups, as were external, visceral, and skeletal abnormalities.

Male Fertility—Nonhuman

Glycerin injected into the testes of rats (50-200 µL and 862 mg/kg body weight) and monkeys (119 mg/kg body weight) suppressed spermatogenesis (Table 7).[38-40]

Male Fertility—Human

In a fertility study of male employees (n = 64) who manufacture synthetic glycerin, there were no differences observed in sperm counts and percentage normal forms compared with a control group (n = 63) who did not work with glycerin (Table 7).[41]

Genotoxicity

In vitro

Glycerin was not genotoxic in multiple Ames tests using multiple strains of *Salmonella typhimurium* up to 50 mg/plate (Table 8).[25,42-46] It was not genotoxic in a cytogenetic assay, X-linked hypoxanthine-guanine phosphoribosyl transferase (HGPRT) assay, sister chromatid exchange assay using Chinese hamster ovary (CHO) cells, unscheduled DNA synthesis assay using rat hepatocytes, or chromosome aberration test using CHO cells; up to 1.0 mg/mL was tested in these studies.[42,44]

In Vivo

In a bone marrow chromosomal aberration assay, glycerin was not genotoxic when administered by injection into the abdomens of rats at 1,000 mg/kg (Table 8).[47]

Carcinogenicity

Glycerin administered in the feed of rats at concentrations up to 20% for 1 year or up to 10 g/kg for 2 years did not increase the incidence of tumors (Table 9).[26]

Glycerin administered in drinking water, up to 5% in as little as 4 weeks, had a synergistic effect with 4-nitroquinoline 1-oxide (4NQO) in mice (Table 9).[48-50] There was an increased number of pulmonary tumor-bearing mice in the treated mice compared to controls.

Irritation and Sensitization

Irritation

Dermal—Nonhuman. Glycerin was not dermally irritating in rabbits at concentrations up to 100% (Table 10).[25,26,51] Glycerin was a mild dermal irritant at 100% in guinea pigs.[29]

Dermal—Human. Glycerin (50% in water) was not irritating to patients with dermatitis (n = 420) when administered for 20 to 24 hours under occlusion.[52] One patient had a positive reaction. She reported using a mixture of glycerin (1 part) and 70% ethanol (9 parts) applied on the hands after washing with soap and water. She was tested with glycerin (1%, 5%, and 10% in water) and her glycerin–ethanol mixture (100%), resulting in +++ reactions for both test substances 48 and 72 hours after exposure. There were negative results in a routine test series. The eczema resolved after she stopped using the ethanol–glycerin mixture.

Glycerin (10%; 0.05 mL) was slightly irritating in a 48-hour occlusive patch test.[3] The irritation score was 4 of 9 on day 14 of observation. No further information was provided.

In a patch test, glycerin (25%; 0.2 mL) was not an irritant when administered to patients (n = 33) for 24 hours.[4]

Table 7. Fertility Studies of Glycerin in Males.

Test animal (n)	Concentration; route	Results; notes	Reference
Sprague Dawley rat; age 48, 69, 90-95 days old (12)	0, 50-200 μL; 2 intratesticular injections 7 days apart into right testes; left was control	Testis treated with 50 μL glycerin decreased in weight (45%-60% within 2 weeks) compared to control side for all ages and complete loss of spermatogenic cells. Testis treated with 200 μL had decreased weights of prostate and seminal vesicles over 73 days. Number of sperm/epididymis declined rapidly, reduced by 99.99% (of controls) after the third mating. Females were added in weeks 2, 3, 4, 5, and 6. Treated males mated with virgin females at same frequency as controls but all were infertile after third mating and remained infertile for the duration of the tests (21 weeks after treatment). No resumption of spermatogenesis.	40
Rat (not provided)	862 mg/kg; intratesticular injection 1 day prior to mating	Suppressed spermatogenesis (meiosis). No evidence of toxic or endocrine effects.	38
Saimiri sciureus, squirrel monkey (10)	119 mg/kg; intratesticular injection 1 day prior to mating and sampled for 22 months	Suppressed spermatogenesis (meiosis) to near 0 in 2 months and remained at 0 for the rest of the study. No evidence of toxic or endocrine effects.	39
Human (64; control, 63)	Exposure through working in a factory manufacturing glycerin; synthetic manufacturing process	No differences observed between sperm counts and percent normal forms compared with a control group. Patients were exposed to other chemicals: epichlorohydrin-allyl chloride and allyl chloride-1,2-dichlorophopene.	41

Table 8. Genotoxicity Assays of Glycerin.

Assay	Concentration	Result; comments	Reference
In vitro			
Ames test using *Salmonella typhimurium* (strain TA100)	0.1 and 1 mmol/plate	Negative with and without metabolic activation	45
Ames test using *S typhimurium* (strains TA98, TA100, TA1535, TA1537, and TA1538)	0.2, 0.4, 0.6, 0.8, and 1.0 mg/plate	Negative with and without metabolic activation	42
Ames test using *S typhimurium* (strains TA98, TA100, TA1535, and TA1537)	10 mg/plate	Tested in 3 laboratories. 2 laboratories had negative results with and without metabolic activation. One lab had ambiguous results	43
Ames test using *S typhimurium* (strains TA98, TA100, TA1537, and TA1538)	10 mg/plate	Negative with and without metabolic activation	25
Ames test using *S typhimurium* (strain TA100)	0.5 mg/plate	Negative with and without metabolic activation	46
Ames test using *S typhimurium* (strains TA94, TA98, TA100, TA1535, and TA1537)	50 mg/plate	Negative with and without metabolic activation	44
Ames test using *S typhimurium* (strains TA98, TA100, TA1535, and TA1537)	1-10 μg/plate; glycerin/water mixture of unknown composition	Negative with and without metabolic activation	25
Cytogenetic assay using CHO cell line WBL	0.1, 0.2, 0.3, 0.6, 0.8, and 1.0 mg/mL	Negative with and without metabolic activation	42
HGPRT assay using CHO (K1 and BH4 cell lines)	0.1, 0.2, 0.4, 0.6, 0.8, and 1.0 mg/mL	Negative with and without metabolic activation	42
Sister chromatid exchange assay using CHO (cell line WBL)	0.2, 0.4, 0.6, 0.8, and 1.0 mg/mL	Negative with and without metabolic activation; purity >99.5%	42
Unscheduled DNA synthesis using rat hepatocytes	0.1, 0.25, 0.5, 0.75, and 1.0 mg/mL	Negative without metabolic activation; purity >99.5%	42
Chromosomal aberration test using CHO cells	1 mg/mL	Negative 100 metaphases analyzed	44
In vivo			
Chromosomal aberration assay using male white rats (10)	1,000 mg/kg by injection into the abdomen	Increase in number of metaphases with agerrations + gaps 3.8% Cytogenic analysis was performed in 500 metaphases	47

Abbreviations: CHO, Chinese hamster ovary; HGPRT, hypoxanthine-guanine phosphoribosyl transferase; WBL, Wolff/Bloom/Litton.

Table 9. Carcinogenicity Studies of Glycerin.

Test animal (n)	Concentration and administration route	Result; comments	Reference
Male and female rats (strain not specified; 24)	5 or 10 g/kg in feed for 2 years	No increase in the incidence of tumors.	26
Male and female Long-Evans rats (22/sex)	0%, 5%, 10%, in diet for 2 years; 20% in diet for 1 year; natural and synthetic glycerin	No increased incidence of tumors following treatment with glycerin. Body weight gain: No differences between treatment and control groups. Histopathology: Malignant neoplasms in 5/26 rats in the control group and 1/22, 5/22, and 0/22 rats for natural glycerin and 0/21, 5/22, and 0/22 for synthetic glycerin, for 5%, 10%, and 20%, respectively. Benign neoplasms in 0/26 rats in the control group and 2/22, 1/22, 0/22 rats for natural glycerin and 4/21, 4/22, and 1/22 for synthetic glycerin, respectively. Among the benign tumors, 3 rats were found with pheochromocytomas, 2 with granulosa cell tumors.	26
Synergistic effects			
ddy mouse (18-20)	0% or 5% in drinking water for 1 to 4 weeks	Increased number of pulmonary tumor-bearing mice and mean number of induced tumors/mouse in mice administered glycerin for 4 to 25 weeks after 4NQO treatment, compared with mice given 4NQO alone. Number of mice with tumors: controls (no 4NQO)—1/20; controls (4NQO)—8/20; 1 week glycerin + NQO1—11/20; 2 weeks glycerin + NQO1—11/19; 3 weeks glycerin + NQO1—7/18; 4 weeks glycerin + NQO1—15/19.	50
Male ddy mice (20)	0%, 5% (approximately 8,350 mg/kg/d) in drinking water for 25 weeks with and without a single injection of 4NQO	Glycerin alone did not result in an increase in the number of mice with tumors compared to untreated controls. Glycerin did have a synergistic effect with 4NQO Two rats in the treatment group died (weeks 25-28) with only fibrosarcomas at injection site, only these had these tumors. Body weight: No treatment-related effects. Pulmonary tumors: Number of mice with tumors: controls—2/20; controls (glycerin)—2/20; treatment (4NQO)—5/20; treatment (4NQO + glycerin)—17/20. Mean number of tumors/mouse: increased after 4NQO + glycerin—2.9 per mouse vs 0.1-0.45 per mouse in the other groups. Histopathology: 4NQO-treated mice all tumors were identified as type II adenomas. In 4NQO + glycerin-treated mice, 52 tumors were identified as type II adenomas and 6 as Clara cell adenomas.	48
Male ddy mice (10)	0%, 5% (~8,350 mg/kg/d) in drinking water for 25 weeks with and without a single injection of 4NQO	Glycerin promoted tumorigenesis when administered after 4NQQ Number of mice with tumors: controls—0/10; controls (glycerin)—0/10; controls (4NQO)—1/10; treatment (4 weeks glycerin)—8/10; treatment (25 weeks glycerin)—8/9; treatment (glycerin week 4-25)—7/10. Mean number of tumors/mouse: controls—0; controls (glycerin 25 weeks)—0; controls (4NQO)—0.1; treatment (4 weeks glycerin)—3.5; treatment (25 weeks glycerin)—2.3; treatment (glycerin weeks 4-25)—1.9. Histopathology: All tumors were adenomas.	49

Abbreviation: 4NQO, 4-nitroquinoline 1-oxide.

Ocular—Nonhuman. Glycerin was not irritating to the eyes of rabbits at concentrations up to 100% (Table 11).[25,26,29,51]

Ocular—Human. Topical administration of anhydrous glycerin to the eyes of human patients with edema of the superficial layers of the cornea resulted in reduced edema and improved visualization.[15] Pain and/or irritation have occurred following administration of glycerin to the eye. No further information was provided.

Glycerin (100%) was reported to be nonirritating when administered to the eyes of human patients (n not specified).[3] There was a strong burning and stinging sensation, with tear production, but no injury was observed. No further information was provided.

Sensitization

Dermal—Nonhuman. Natural and synthetic glycerin were not sensitizing to white male guinea pigs (n = 12).[26] The induction phase consisted of 10 injections of 0.1 mL of a 0.1% solution every other day. The challenge phase consisted of an injection of 0.05 mL of the 0.1% solution after a 2-week resting phase.

Dermal—Human. A moisturizer containing glycerin (65.9%) was not sensitizing in a modified Draize test (n = 48).[53] There were no reactions during either the induction phase or the challenge phase. The test substance was administered 10 times under occlusion for 48 or 72 hours (over the

Table 10. Dermal Irritation Studies in Animals.

Animal (n, if provided)	Results and details	Reference
Rabbit (12)	Not irritating at 100% after 90 days of administration. Draize test of 0.5 to 4 mL of both natural and synthetic glycerin administered to 30% of the body surface for 8 h/d 5 d/wk, 45 weeks. Purity of both = 99.5%.	26
Rabbit, New Zealand female (6)	Not irritating. 0.5 mL glycerin/water mixture of unknown composition. Draize scale score 0.1 for intact and abraded skin.	25
Rabbit, albino male (8)	Not irritating. 0.5 mL administered to 6.25 cm^2 of skin for 24 hours. No signs of irritation at 24 and 72 hours. Draize scale scores 0 to 0.4 compared to a maximum score of 30.	51
Guinea pig (approximately 45)	Mildly irritating; +. 0.1 mL administered to the shaved abdominal skin and observed at 4 and 24 hours.	29

Table 11. Ocular Irritation Studies in Animals.

Animal (n)	Results and details	Reference
Rabbit (6)	Not irritating. 0.1 mL at 100%. No irritation at 1, 24, and 72 hours and 7 days. Overall Draize score 0 to 2 on a scale of 110.	51
Rabbit (4)	Not irritating for both natural and synthetic glycerin with purity of 99.5%. The conjunctiva was irritated in all rabbits 1 hour after treatment. Resolved at 24 hours after treatment.	26
Rabbit, New Zealand White, female (6)	Not irritating. 0.1 mL at 100% glycerin/water mixture of unknown composition. Overall Draize score 0.4 at 1 hour, 0 at 24 to 96 hours.	25
Rabbit (5)	Mildly irritating; + for both edema and hyperemia. Approximately 0.5 mL at 100%.	29

weekend). The challenge patch was in place for 48 hours. The test site was observed at removal and 48 hours after removal.

Patients (n = 15) who worked in a foam rubber factory and were regularly exposed to glycerin were not sensitized to glycerin when patched tested at 100% for 48 hours.[54]

Case Reports

A 29-year-old woman presented with a 7-month history of patchy eczema on her eyelids, face, neck, scalp, and axillae.[55] She was patch tested using the European standard series, and bases, cosmetics, and hairdressers series. She was also patch tested with her own cosmetics and toiletries. She had a + positive reaction on day 4 to dimethylaminopropylamine (1% aqueous) and to her own hand moisturizing cream. Further testing of the ingredients of this cream had a + positive reaction on day 4 to glycerin (1% aqueous). Her eczema resolved when she avoided glycerin-containing cosmetics.

Summary

This is a safety assessment of glycerin, a polyhydric alcohol. Glycerin is reported to function in cosmetics as a denaturant, fragrance ingredient, hair conditioning agent, humectant, oral care agent, oral health-care drug, skin protectant, skin conditioning agent—humectant, and viscosity-decreasing agent.

Impurities were reported to be water, polyglycerol, and diethylene glycol. Phorbol esters may be present if the source material is the *Jatropha* plant.

Glycerin is reported to be used in 15,654 cosmetic products: 10,046 are leave-on products, 5,441 are rinse-off products, and 167 are products that are diluted for the bath. Glycerin is reported to be used at concentrations up to 78.5% in leave-on products, 68.6% in rinse-off products, and 47% in products diluted for the bath.

Glycerin is considered to be GRAS by the FDA for food packaging and as a multiple-purpose food substance. Glycerin is also used as an active ingredient in some over-the-counter drugs.

Glycerin is rapidly absorbed in the intestine and the stomach and distributed throughout the extracellular fluids through much of the body. It is metabolized primarily by the liver and kidneys, with the remainder excreted in urine. Free glycerin is naturally present in humans, primarily in plasma.

The reported oral LD_{50} of glycerin ranged from 2,530 to 58,400 mg/kg in rats, 4,090 to 38,000 mg/kg in mice, 27,000 mg/kg in rabbits, and 77,500 mg/kg in guinea pigs. The dermal LD_{50} of glycerin in rats was reported to be >21,900 mg/kg and >18,700 mg/kg in rabbits. The approximate Lt_{50} for rats was determined to be 423 minutes for exposure to glycerin vapors at 11.0 mg/L.

The oral LD_{LO} of glycerin was reported to be 1,428 mg/kg for humans. There were no signs of toxicity when humans were orally administered 30 mL glycerin. When orally administered as a drug, adverse effects in humans included mild headache, dizziness, nausea, vomiting, thirst, and diarrhea.

In short-term feeding experiments using rats, 20% glycerin for 4 weeks in feed had no adverse effects, but at 53.4%, the kidneys weights were increased and liver enzymes were elevated. When glycerin was administered in the diet for 2 years, feed consumption was slightly increased in males fed synthetic glycerin versus those fed naturally sourced glycerin. There were no treatment-related effects in organ weights and gross pathology. The NOEL in mongrel dogs was 950 mg/kg/d when orally administered for 3 days. At 3,800 mg/kg/d, the mucosa of the stomach was severely hyperemic with petechial hemorrhages. Mongrel dogs experienced weight loss after 36 weeks when 35% glycerin was incorporated into their feed. There were no pathological changes in guinea pigs orally administered 6,300 mg/kg/d glycerin for 30 to 40 days.

There were no signs of toxicity or effects on blood or on urine production when human patients were orally administered approximately 1,300 to 2,200 g/kg/d glycerin for 50 days. The NOAEL was $\geq$2,200 mg/kg/d.

There were no treatment effects when 100% glycerin was topically applied daily to 30% of the body surfaces of rabbits for 45 weeks.

The inhalation LOAEL was 1,000 mg/m^3 for glycerin administered 6 h/d, 5 d/wk for 2 weeks in rats. The inhalation NOAEL was 0.167 mg/L for glycerin administered for 5 h/d, 5 d/wk for 13 weeks in rats.

No adverse effects were observed in rats administered 20% glycerin in drinking water throughout gestation and nursing of pups. The F_1 generation reproduced normally. The oral NOAEL for maternal toxicity and teratogenicity for rats was 1,310 mg/kg/d. The NOAEL for maternal toxicity and teratogenicity in mice was 1,280 mg/kg/d. The NOAEL for maternal toxicity and teratogenicity in rabbits was 1,180 mg/kg/d.

Glycerin injected into the testes of rats (50-200 µL and 862 mg/kg body weight) and monkeys (119 mg/kg body weight) suppressed spermatogenesis.

Glycerin was not genotoxic in multiple Ames tests using multiple strains of *S typhimurium* at concentrations up to 50 mg/plate. It was not genotoxic in a cytogenetic assay, X-linked HGPRT, sister chromatid exchange assay, unscheduled DNA synthesis assay, and chromosome aberration test at concentrations up to 1.0 mg/mL.

In 2 chromosomal aberration assays, glycerin was not genotoxic when administered orally to rats at 1 mg/kg or by injection into the abdomen at 1 g/kg. In a dominant lethal gene assay using rats, the results were ambiguous.

Glycerin administered in the feed of rats at doses up to 20% in feed for 1 year or up to 10 g/kg for 2 years did not increase the incidence of tumors. Orally administered glycerin, in concentrations up to 5%, had a potentiating effect on the carcinogenicity of 4NQO in mice.

Glycerin was not dermally irritating to rabbits when applied at concentrations up to 100% to up to 30% of the body surface 8 h/d, 5 d/wk for 45 weeks. Glycerin was a mild dermal irritant at 100% in guinea pigs.

Glycerin at 50% was not irritating to patients with dermatitis. Glycerin at 25% was not an irritant when administered to patients in a patch test.

Undiluted glycerin was not irritating when administered to the eyes of human patients. Strong burning and stinging sensation with tear production was reported, but no injury was observed.

Natural and synthetic glycerin were not sensitizing to white male guinea pigs at 0.1%. A moisturizer containing 65.9% glycerin was not sensitizing to humans.

Discussion

When considering the safety of glycerin, the Cosmetic Ingredient Review Expert Panel (Panel) noted that it is naturally occurring and abundant in animal and human tissues, including the skin and blood. The available data demonstrated low systemic oral and dermal toxicity for multiple animal species and humans, in both acute and long-term studies. There were no reproductive or developmental effects observed in oral studies using rats, mice, and rabbits. Glycerin was not genotoxic in multiple in vitro tests and it was not carcinogenetic to rats in a long-term feeding study. This ingredient was not a dermal or ocular irritant and was nonsensitizing to guinea pigs and humans. The Panel also noted the high frequency of use that is reported for glycerin and the low instances of reports of toxicity, irritation, or sensitization in the literature and the fact that glycerin is GRAS.

The source materials and intermediate forms of glycerin (eg, epichlorohydrin) should be completely consumed and/or eliminated in the manufacturing process. The Panel noted FDA's warning that companies should monitor and audit their naturally derived ingredients because of the potential presence of phorbol esters if the source material is the *Jatropha* plant.

The Panel discussed the issue of incidental inhalation exposure from hair sprays (up to 10% in aerosol sprays and 30% in pump sprays), deodorants (up to 4% in a pump spray), face and neck products (up to 10% in aerosol sprays), face powders (up to 15%), body and hand sprays (up to 5% in aerosol sprays), moisturizing products (up to 3.3% in aerosol sprays), and in suntan products (up to 6% in aerosol sprays and 10% in pump sprays). Because the results of a 2-week (up to 1,000 mg/L) and a 13-week (up to 662 mg/L) inhalation study demonstrated little or no toxicity, the Panel concluded that there was little potential for respiratory effects at reported use concentrations. The Panel noted that 95% to 99% of droplets/particles would not be respirable to any appreciable amount. Coupled with the small actual exposure in the breathing zone and the concentrations at which the ingredient is used, the available information indicates that incidental inhalation would not be a significant route of exposure that might lead to local respiratory or systemic effects. A detailed discussion and summary of the Panel's approach to evaluating incidental inhalation exposures to ingredients in cosmetic products is available at http://www.cir-safety.org/cir-findings.

Conclusion

The Panel concluded that glycerin is safe in cosmetics in the present practices of use and concentration described in this safety assessment.

Authors' Note

Unpublished sources cited in this report are available from the Executive Director, Cosmetic Ingredient Review, 1620 L Street, NW, Suite 1200, Washington, DC 20036, USA.

Author Contributions

L. Becker contributed to conception and design, contributed to acquisition, analysis, and interpretation, drafted the manuscript, and critically revised the manuscript. W. Bergfeld, D. Belsito, R. Hill, C. Klaassen, D. Liebler, J. Marks, R. Shank, T. Slaga, P. Snyder, and L. Gill contributed to conception and design, contributed to analysis and interpretation, and critically revised the manuscript. B. Heldreth contributed to analysis and interpretation and critically revised the manuscript. All authors gave final approval and agree to be accountable for all aspects of work ensuring integrity and accuracy.

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References

1. Nikitakis J, Breslawec HP. *International Cosmetic Ingredient Dictionary and Handbook*. 15th ed. Washington, DC: Personal Care Products Council. 2014.
2. Organization for Economic Cooperation and Development (OECD) Screening Information Data Set (SIDS). *SIDS Initial Assessment Report For SIAM 14; Glycerol: CAS No: 56-81-5. Howbery Park, Wallingford UK, Organization for Economic Cooperation and Development (OECD)*. 2002. 1-178. http://www.inchem.org/documents/sids/sids/56815.pdf. Accessed May 5, 2014.
3. European Commission—European Chemicals Bureau. *IUCLID Dataset: Existing Chemical Substance ID: 56-81-5; CAS No. 56-81-5; EINECS Name glycerol; EINECS No. 200-289-5; Molecular Formula C3H8O3*. 2000. 1-173. http://esis.jrc.ec.europa.eu/doc/IUCLID/data_sheets/56815.pdf. Report No. 1.
4. European Chemicals Agency. *ECHA—European Agency (56-81-5, Glycerol)*. http://echa.europa.eu/. Updated 2014. Accessed November 13, 2014.
5. Christoph R, Schmidt B, Ssteinberner U, Dilla W, Karinen R. Glycerol. In: *Ullmann's Encyclopedia of Industrial Chemistry*. Hoboken, NJ. Wiley and Sons, Inc; 2006:67-82.
6. Lewis RJ Sr. *Sax's Dangerous Properties of Industrial Materials*. 10th ed. New York, NY: John Wiley & Sons, Inc, 2000.
7. US Pharmacopeial Convention. *U.S. Pharmacopeia National Formulary*. Rockville, MD: US Pharmacopeial Convention; 2009.
8. US Food and Drug Administration. *FDA Notification to Industry: Products Using Oils, Glycerin, or Protein That Were Derived From the Jatropha Plant May Have Toxic Effects*. http://www.fda.gov/ForIndustry/IndustryNoticesandGuidanceDocuments/ucm391133.htm. Washington, DC: US Food and Drug Administration; 2012. Updated May 1, 2014. Accessed August 6, 2014.
9. US Food and Drug Administration Center for Food Safety & Applied Nutrition (CFSAN). *Voluntary Cosmetic Registration Program—Frequency of Use of Cosmetic Ingredients*. College Park, MD; 2013. Obtained under the Freedom of Information Act from CFSAN; requested as "Frequency of Use Data" January 2013; received February 2013.
10. Personal Care Products Council. *Updated Concentration of Use by FDA Product Category: Glycerin*. Unpublished data submitted by Personal Care Products Council. Washington, DC. 2014.
11. Bremmer HJ, Lodder LCHPH, Van Engelen JGM. *Cosmetics Fact Sheet: To Assess the Risks for the Consumer; Updated Version for ConsExpo 4*. Bilthoven, the Netherlands. Netherlands National Institute for Public Health and the Environment. 2006. 1-77. http://www.rivm.nl/bibliotheek/rapporten/320104001.pdf. Accessed August 24, 2011. Report No. RIVM 320104001/2006.
12. Johnsen MA. The influence of particle size. *Spray Tech Market*. 2004;14(11):24-27. http://www.spraytechnology.com/index.mv?screen=backissues.
13. Rothe H, Fautz R, Gerber E, et al. Special aspects of cosmetic spray safety evaluations: principles on inhalation risk assessment. *Toxicol Lett*. 2011;205(2):97-104. PMID:21669261.
14. Rothe H. *Special Aspects of Cosmetic Spray Safety Evaluation*. Unpublished information presented to the 26 September CIR Expert Panel. Washington, DC. 2011.
15. McEvoy GK.*American Hospital Formulary Service—Drug Information 93. 35th ed*. Bethesda, MD: American Society of Hospital Pharmacists. 1993.
16. Lin EC. Glycerol utilization and its regulations in mammals. *Ann Rev Biochem*. 1977;46:765-795.
17. Tourtellotte WW, Reinglass J, Newkirk T.Cerebral dehydration action of glycerol. I. Historical aspects with emphasis on the toxicity and intravenous administration. *Clin Pharm Ther*. 1972; 13(2):159-171.
18. Tao RC, Kelley R, Yoshimura N, Benjamin F. Glycerol: its metabolism and use as an intravenous energy source. *JPEN*. 1983;7(5): 479-488.
19. Bortz WM, Paul P, Haff A, Holmes W. Glycerol turnover and oxidation in man. *J Clin Invest*. 1972;51(6):1537-1546.
20. Nelson JL, Harmon M, Robergs R. Identifying plasma glycerol concentration associated with urinary glycerol excretion in train humans. *J Analyt Toxicol*. 2011;35(9):617-623.
21. MacDonald I. Effects of dietary glycerol on th eserum glyceride level of men and women. *Br J Nutr*. 1970;24(2):537-543.
22. Gad Shayne C, Cassidy CD, Aubert N, Spainhour B, Robbe H. Nonclinical vehicle use in studies by multiple routes in multiple species. *Int J Toxicol*. 2006;25(6):499-521.
23. Bartsch W, Sponer G, Dietmann K, Fuchs G.Acute toxicity of various solvents in the mouse and rat. LD50 of ethanol, diethylacetamide, dimethylformamide, dimethylsulfoxide, glycerine, N-methylpyrrolidone, polyethylene glycol 400, 1,2-propanediol and tween 20. *Arzneimittelforschung*. 1976;26(8):1581-1583.

24. Bornmann G. Grundwirkungen der glykole und ihre bedeutung für die toxizität. *Arzneimittelforschung*. 1955;4:643-646.
25. Clark CR, Marshall T, Merickel B, Sanchez A, Brownstein D, Hobbs C. Toxicological assessment of heat transfer fluids proposed for use in solar energy applications. *Toxicol Appl Pharmacol*. 1979;51(3):529-535.
26. Hine CH, Anderson H, Moon H, Dunlap M, Morse M. Comparative toxicity of synthetic and natural Glycerin. *Arch Indust Hygiene Occup Med*. 1953;7(4):282-291.
27. Smyth HF Jr, Seaton J, Fischer L. The single dose toxicity of some glycols and derivatives. *J Indus Hygeine Toxicol*. 1941;23(138): 259-268.
28. Anderson RC, Harris P, Chen K. Toxicological studies on synthetic glycerin. *J Am Pharm Assoc*. 1950;39(10):583-585.
29. Latven AR, Molitor H. Comparison of the toxic, hypnotic and irritating properties of eight organic solvents. *J Pharm Exp Therap*. 1939;65(1):89-94.
30. Staples R. Gastrointestinal irritant effects of Glycerin as compared with sorbitol and propylene glycol in rats and dogs. *J Pharm Sci*. 1967;56(3):398-400.
31. Cryer A, Bartley W. Studies on the adaptation of rats to a diet high in glycerol. *Int J Biochem*. 1973;4(21):293-308.
32. Stoewsand GS, Dymsza HA. Synthetic sources of calories in the diets of rats and dogs. *Proceedings of the Seventh International Congress of Nutrition*. Hamburg, Germany; 1966.
33. Fischer L, Kopf R, Loeser A, Meyer G. Chemische konstitution und pharmakologische wirkung der glykole unter besonderer berücksichtigung von 1,3-butylenglykol. *Zeitschr Gesamte Exp Med*. 1949;115(1-2):22-39.
34. Johnson V, Carlson A, Johnson A. Studies on the physiological action of glycerol on the animal organism. *Ame J Physiol*. 1933; 103(3):517-534.
35. Ostwald R. Glycerol intake, blood cholesterol level and anemia in the guinea pig and rabbit. *Exp Biol Med*. 1962;111:632-634.
36. Renne RA, Wehner A, Greenspan B, et al. 2-Week and 13-week inhalation studies of aerosolized glycerol in rats. *Inhal Toxicol*. 1992;4(2):95-111.
37. National Technical Information Service (NTIS). *Teratological Evaluation of Glycerin in Mice, Rats and Rabbits*. Rockville, MD: US Department of Commerce. 1974. 1-48. Report No. PB-234 876/1.
38. Wiebe J, Barr K. Suppression of spermatogenesis without inhibition of stroidogenesis by a 1,2,3-trihydroxypropane solution. *Life Sci*. 1984;34(18):1747-1754.
39. Wiebe J, Barr K, Buckingham K. Sustained azoospermia in squirrel monkey, saimiri sciureus, resulting from a singe intratesticular glycerol injection. *Contraception*. 1989;39(4):447-457.
40. Wiebe JP, Barr K. The control of male fertility by 1,2,3-trihydroxypropane (THP; glycerol): rapid arrest of spermatogenesis without altering libido, acessory organs, gonadal steroidogenesis, and serum testosterone, LH and FSH. *Contraception*. 1984;29(3):291-302.
41. Venable JR, McClimans C, Flake R, Dimick D. A fertility study of male employees engaged in the manufacture of glycerine. *J Occup Med*. 1980;22(2):87-91.
42. Doolittle D. The genotoxic activity of glycerol in an in vitro test battery. *Food Chem Toxicol*. 1988;26(7):631-635.
43. Haworth S, Lawlor T, Mortelmans K, Speck W, Zeiger E. Salmonella mutagenicity test results for 250 chemicals. *Environ Mutagen*. 1983;5(suppl 1):1-142.
44. Ishidate M Jr, Sofuni T, Yoshikawa K, et al. Primary mutagencity screening of food additives currently used in japan. *Food Chem Toxicol*. 1984;22(8):623-636.
45. Stolzenberg SJ, Hine C. Mutagenicity of halogenated and oxygenated three-carbon compounds. *J Toxicol Environ Health*. 1979; 5(6):1149-1158.
46. Yamaguchi T. Mutagenicity of trioses and methyl glyoxal on *Salmonella typhimurium*. *Agric Biol Chem*. 1982;46(3):849-851.
47. Barilyak IR, Kozachuk S. Mutagenic effect of various alcohols in experiment. *Cytol Gen*. 1985;19(6):38-43.
48. Inayama Y. Promoting action of glycerol in pulmonary tumorigenesis model using a single administration of 4-nitroquinoline 1-oxide in mice. *Jpn J Can Res*. 1986;77(4):345-350.
49. Inayama Y, Kitamura H, Kanisawa M. Effects of glycerol on 4-nitroquinoline 1-oxide induced pulmonary tumorigenesis in ddY mice. *Jpn J Can Res*. 1986;77(2):103-105.
50. Nagahara N. Modification by glycerol of the initial process of pulmonary tumorigenesis induced by 4-nitroquinoline 1-oxide in mice. *Yokoham Med Bull*. 1987;38(5-6):141-150.
51. Weil CS, Scala R. Study of intra- and interlaboratory variability in the results of rabbit eye and skin irritation tests. *Toxicol Appl Pharmacol*. 1971;19(2):276-360.
52. Hannuksela M, Förström L. Contact hypersensitivity to glycerol. *Int J Sci*. 1979;1(5):257-263.
53. International Research Services. A study to assess the skin sensitization potential of one test product (moisturizer containing 65.9% glycerin) when applied to the skin of 50 healthy human subjects in a shared panel assay. Unpublished data submitted by personal care products council. 2006.
54. Nagdy AEI, Fahim B. Medicolegal aspects of occupational dermatitis survey in a foam rubber factory. *J Egypt Med Assoc*. 1973; 56(4-5):331-339.
55. Preston PW, Finch T. Allergic contact dermatitis from glycerin in a moisturizing cream. *Contac Dermat*. 2003;49(4):221-222.
56. O'Neil MJ. *The Merk Index—An Encyclopedia of Chemicals, Drugs, and Biologicals*. Whitehouse Station, NJ: Merck and Co, Inc; 2006.