Safety Assessment of Sodium Dehydroacetate and Dehydroacetic Acid as Used in Cosmetics

Status:	Re-Review for Panel Consideration
Release Date:	August 18, 2023
Panel Meeting Date:	September 11 – 12, 2023

The Expert Panel for Cosmetic Ingredient Safety members are: Chair, Wilma F. Bergfeld, M.D., F.A.C.P.; Donald V. Belsito, M.D.; David E. Cohen, M.D.; Curtis D. Klaassen, Ph.D.; Allan E. Rettie, Ph.D.; David Ross, Ph.D.; Thomas J. Slaga, Ph.D.; Paul W. Snyder, D.V.M., Ph.D.; and Susan C. Tilton, Ph.D. The Cosmetic Ingredient Review (CIR) Executive Director is Bart Heldreth, Ph.D., and the Senior Director is Monice Fiume. This report was prepared by Priya Cherian, M.S., Senior Scientific Analyst/Writer, CIR.

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Memorandum

To: Expert Panel for Cosmetic Ingredient Safety Members and Liaisons
From: Priya Cherian, M.S., Senior Scientific Analyst/Writer, CIR
Date: August 18, 2023
Subject: Re-Review of the Safety Assessment of Sodium Dehydroacetate and Dehydroacetic Acid

The Expert Panel for Cosmetic Ingredient Safety (Panel) first published a review of the safety of Sodium Dehydroacetate and Dehydroacetic Acid in 1985 (identified as *originalreport_SodiumDehydroacetate_092023* in the pdf), with the conclusion that these ingredients are safe as cosmetic ingredients in the present practices of use and concentration, as stated in that report. The Panel previously considered a re-review of this report in 2003 and re-affirmed the 1985 conclusion, as published in 2006 (*rereview2006_SodiumDehydroacetate_092023*).

Because it has been 15 years since the previous re-review was published, in accordance with Cosmetic Ingredient Review (CIR) Procedures, the Panel should consider whether the safety assessment of Sodium Dehydroacetate and Dehydroacetic Acid be re-opened. In July 2023, an extensive search of the world's literature was performed for studies dated 2000 forward. An historical overview, comparison of original and new use data, the search strategy used, and a synopsis of notable new data are enclosed herein (*newdata_SodiumDehydroacetate_092023)*.

New studies were found for several toxicological endpoints (e.g., absorption, distribution, metabolism, and excretion, acute oral toxicity, repeated-dose toxicity, developmental and reproductive toxicity, genotoxicity, dermal irritation, dermal sensitization, and ocular irritation). Of note are hypersensitivity case reports of patients reporting adverse effects following use of creams containing Sodium Dehydroacetate (for ulcer treatment). In addition, one study was found suggesting the photoisomerization potential of Sodium Dehydroacetate and Dehydroacetic Acid.

Also included for your review are current and historical use data (*usetable_SodiumDehydroacetate_092023).* The frequency of use of both Sodium Dehydroacetate and Dehydroacetic Acid have significantly increased according to 2023 FDA VCRP data. In 2002, Sodium Dehydroacetate and Dehydroacetic Acid were reported to be used in 325 and 88 formulations, respectively. In 2023, Sodium Dehydroacetate and Dehydroacetic Acid are reported to be used in 1233 and 833 formulations, respectively. The 2023 reported concentrations of use for both ingredients (maximum concentrations of 0.6% for Sodium Dehydroacetate and 0.7% for Dehydroacetic Acid) are the same maximum concentrations as reported in 2003.

According to the web-based *International Cosmetic Ingredient Dictionary and Handbook*, as in the original report, both ingredients are reported to function as a preservative in cosmetics. It should be noted that European Union regulations state that Sodium Dehydroacetate and Dehydroacetic Acid may be used as a preservative in cosmetics at up to 0.6% (as acid); however, these ingredients should not be used in aerosol dispensers (sprays). (It is not indicated why these ingredients should not be used in sprays.) According to 2023 VCRP data, Dehydroacetic Acid is used at 0.000008% in an aerosolized hair spray.

If upon review of the new studies and updated use data the Panel determines that a re-review is warranted, a Draft Amended Report will be presented at an upcoming meeting.

Re-Review - Sodium Dehydroacetate and Dehydroacetic Acid - History and New Data

(Priya Cherian – September 2023 Meeting)

Ingredients (2)	Citation	Conclusion	Use - New Data	Results	Use - Existing Data	Results	Notes
Sodium Dehydroacetate **Dehydroacetic Acid**	JACT 4(3):123-59, 1985 IJT 25(Suppl. 2): 65-8, 2006	safe as used not re-opened	Sodium Dehydroacetate frequency of use (2023) conc of use (2023) Dehydroacetic Acid frequency of use (2023) conc of use (2023)	 1233 ≤ 0.6% 833 ≤ 0.7%	Sodium Dehydroacetate frequency of use (2002) conc of use (2003) Dehydroacetic Acid frequency of use (2002) conc of use (2003)	 325 ≤ 0.6% 88 ≤ 0.7%	 significant increase in frequency of use no change in concentration of use significant increase in frequency of use no change in concentration of use

NOTABLE NEW DATA			
Publication	**Study Type**	**Results – Brief Overview**	**Different from Existing Data?**
21CFR172.130	Non-Cosmetic Use	Sodium Dehydroacetate and Dehydroacetic Acid may be used as food additives when proper criteria are met (melting point of 109-111°C; minimum purity of 98% (on dry basis)); these ingredients are used as preservatives for cut or peeled squash (not more than 65 ppm Dehydroacetic Acid may remain in or on prepared squash); proper labeling required	This CFR citation was not provided in original report; however, use as food additive is mentioned in report
EU law - EUR-Lex (europa.eu)	Non-Cosmetic Use	According to EU regulations, Sodium Dehydroacetate and Dehydroacetic Acid may be used as a preservative in cosmetics at up to 0.6% (as acid); these ingredients should not be used in aerosol dispensers (sprays)	This citation provides updated EU regulation information. The original report states the limitation of 0.6% as a preservative, but notes that higher concentrations may be used for specific purposes. Current regulations do not indicate permitted uses above 0.6%. In addition, current regulations state the prohibited use of these ingredients in aerosolized products.
Zhang Y, Ying D, Liu H, Yu Z, Han L, Xie J, Xie Y. Serum pharmacokinetics and coagulation aberration induced by sodium dehydroacetate in male and female Wistar rats. Sci Rep. 2017 Apr 7;7:46210.	ADME	Sodium Dehydroacetate (50 - 200 mg/kg) administered to Wistar rats orally (method of oral administration not stated); serum levels of Sodium Dehydroacetate measured (details not provided in study); on days 7 - 23, Sodium Dehydroacetate concentration in males was 26 - 36 mg/l in the 200 mg/kg group, and 20-25 mg/l in the 150 mg/kg group; in females, the respective values were 33 - 48 mg/l and 41 - 40 mg/l	Elevated absorption in females, compared to males, not stated in previous report

NOTABLE NEW DATA			
Publication	**Study Type**	**Results – Brief Overview**	**Different from Existing Data?**
Chen X, Hao F, Zhang M, Xiao J, Zhao W, Zhao Z, Zhang Y. Sex Metabolic Differences and Effects on Blood Coagulation Among Rats Exposed to Sodium Dehydroacetate. Front Pharmacol. 2021 Sep 14;12:727084.	ADME	Sodium Dehydroacetate (200 mg/kg) administered via gavage for 9 - 11 d to rats (6/sex/group; strain not stated); blood samples collected at different time intervals; serum and tissue levels were significantly higher in females; half-life in females: 23.22 ± 4.55; half-life in males: 18.94 ± 4.27; clearance in females: 0.01 l/h/kg; clearance in males: 0.01 l/h/kg; lungs were the tissue with the highest reported concentration of the test substance: 50.95 ± 0.55 mg/kg (average in males and females 7 d after treatment)	Elevated absorption in females not stated in previous report
Zhang Y, Ying D, Liu H, Yu Z, Han L, Xie J, Xie Y. Serum pharmacokinetics and coagulation aberration induced by sodium dehydroacetate in male and female Wistar rats. Sci Rep. 2017 Apr 7;7:46210..	Acute Toxicity - Oral	Sodium Dehydroacetate (50-200 mg/kg) given to Wistar rats orally (method of oral administration not stated); significant decrease in body weight/weight gain in treated animals at all dose levels in females, and in all dose levels in male excluding 50 mg/kg; prothrombin time and partial thromboplastin time prolonged in treated animals; congestion in hepatic sinusoids, renal tubules, and spleen, as well as hemorrhage in lung alveoli, gastric mucosa, intestinal mucosa, and cardiac muscle cells observed in treated animals; no details provided regarding testing methods	Parameters evaluated in this study were not evaluated in studies currently in the report
Registration Dossier - ECHA (europa.eu)	Acute Toxicity – Oral	OECD TG 401; Dehydroacetic Acid in propylene glycol (up 1000 – 6400 mg/kg) fed to albino rats (5/sex/group); female LD_{50} of 1480 mg/kg bw; male LD_{50} of 1620 mg/kg bw; clinical signs observed include lethargy, lack of motor control, hyperactive responses, convulsions, and hemorrhaging from nose and mouth	No
Du HJ, Tong GH, Ning JY, Gao S, Yang Q, Feng Y, Zhang P, Zhang W, Jing HM, Li GJ. A repeated dose 28-day oral toxicity study of sodium dehydroacetate (DHA-S) in Wistar rats. Regul Toxicol Pharmacol. 2023 Jan;137:105313.	Repeated Dose Toxicity – Oral	OECD TG 407; Sodium Dehydroacetate (0, 50, 100, or 200 mg/kg bw/d) was given to Wistar rats (10/sex/group) for 28 d via gavage; controls given water; body weight and food consumption significantly reduced at high dose levels; decreased organ weights noted in high-dosed treated groups; at the 200 mg/kg bw dose, the blood coagulation activities were significantly reduced in females; NOAEL determined to be 50 mg/kg bw/d	Coagulation parameters not evaluated in studies provided in original report
Fang J, Liu HB, Zhi Y, Feng YQ, Wang HL, Cui WM, Zhang JY, Wang HL, Yu Z, Jia XD. Subchronic oral toxicity evaluation of Sodium Dehydroacetate: A 90-day Repeated Dose Study in Rats. Biomed Environ Sci. 2022 Apr 20;35(4):296-311. Erratum in: Biomed Environ Sci. 2023 Mar 20;36(3):304.	Repeated Dose Toxicity – Oral	OECD TG 408; Sodium Dehydroacetate (0, 31, 62, and 120 mg/kg bw/d) was orally administered to Sprague-Dawley rats (5 - 10/sex/group), via gavage, for 90 d; 4-wk recovery period for control group and 120 mg/kg bw/d group; body weight significantly decreased in animals treated with 62 and 124 mg/kg bw/d; significant increase in thyroid-stimulating hormones at 124 mg/kg bw/d; reduction in organ weights observed in treated animals	No

NOTABLE NEW DATA			
Publication	**Study Type**	**Results – Brief Overview**	**Different from Existing Data?**
Registration Dossier - ECHA (europa.eu)	DART	Wistar rats (22/sex/dose) treated with Sodium Dehydroacetate in water (up to 100 mg/kg bw/d), via gavage, on days 6-17 of gestation; maternal toxicity noted at highest dose level (reduced food intake, lower body weight, early resorptions); fetuses from high-doses displayed lower body weight and decreased ossification	Maternal toxicity not evaluated in studies provided in original report
Zeiger E, Anderson B, Haworth S, Lawlor T, Mortelmans K, Speck W. Salmonella mutagenicity tests: III. Results from the testing of 255 chemicals. Environ Mutagen. 1987;9 Suppl 9:1-109.	Genotoxicity – In Vitro	OECD TG 471; Ames assay in several *Salmonella typhimurium* strains; testing Sodium Dehydroacetate at up to 1820 µg/plate; performed with and without metabolic activation; negative results obtained; controls gave expected results	No
Registration Dossier - ECHA (europa.eu)	Genotoxicity – In Vitro	OECD TG 473; in vitro mammalian chromosome aberration assay; testing Sodium Dehydroacetate (up to 14.8 µg/ml) in 1% dimethyl sulfoxide; performed with and without metabolic activation; negative results obtained; controls gave expected results	No
Registration Dossier - ECHA (europa.eu)	Genotoxicity – In Vitro	OECD TG 490; in vitro mammalian cell gene mutation test; test performed using Sodium Dehydroacetate (up to 10 mM); cell culture medium used as vehicle; on mouse lymphoma L5178Y cells; negative results obtained; controls gave expected results	No
Registration Dossier - ECHA (europa.eu)	Genotoxicity – In Vivo	Micronucleus test performed on male ddy mice (2 - 6/group); animals treated with Sodium Dehydroacetate in water or saline at concentrations of up to 1250 mg/kg bw/d; administrations via both intraperitoneal injection and gavage; positive results during intraperitoneal testing; negative results with gavage testing; study authors state that the appropriate route of exposure is most likely oral, rather than intraperitoneal	No
Izawa T, Nakayama K, Uchida N, Nojima K. Photoreactivities of the Antiseptics Dehydroacetic Acid and Sodium Dehydroacetate Used in Cosmetics. Chem Pharm Bull (Tokyo). 2018 May 1;66(5):581-584.	Other Relevant Studies – Photoreactivity	Dehydroacetic Acid was found to induce photoisomerization, converting aldrin and dieldrin into photoaldrin and photodieldrin, respectively, when irradiated with both artificial light and sunlight; Sodium Dehydroacetate induces both photoisomerization and photoepoxidation; photo-erethism may occur due to the isomerization and epoxidation properties of the compound	This information was not included in the original assessment. However, cosmetic products containing Sodium Dehydroacetate (0.1%) were considered to be non-phototoxic and non-photosensitizing in several studies.
Registration Dossier - ECHA (europa.eu)	Dermal Irritation – Animal	OECD TG 404; undiluted Dehydroacetic Acid applied to skin of albino rabbits (n =5; sex not stated); occlusive conditions; non-irritating	No
Registration Dossier - ECHA (europa.eu)	Dermal Sensitization – In Vitro	OECD TG 429; LLNA; Dehydroacetic Acid (5, 10, and 20%) in propylene glycol; 4 female CBA/Ca mice/group; no skin sensitizing potential according to CLP/EU GHS criteria	No

NOTABLE NEW DATA			
Publication	**Study Type**	**Results – Brief Overview**	**Different from Existing Data?**
Registration Dossier - ECHA (europa.eu)	Ocular Irritation – In Vitro	OECD TG 438; Isolated chicken eye test method; treatment with 0.03 g undiluted Sodium Dehydroacetate; incubation for up to 240 min; test material not shown to be eye irritant according to CLP/EU GHS criteria;	No
Foti C, Antelmi A, Guida S, Romita P, Bonamonte D. Sodium dehydroacetate: an emerging allergen. Dermatitis. 2012 Sep-Oct;23(5):243.	Clinical Assessment of Safety – Case Report	A 42-year-old nonatopic woman presented with eczematous lesions on the legs, mainly surrounding malleolar ulcers. The patient had been treating the ulcers with a hyaluronic acid-based cream containing Sodium Dehydroacetate as a preservative. Patch tests were performed on all topical drugs applied to ulcers within a year. Patch tests were positive for nickel sulfate and one cream. Individual ingredients of this cream were then patch tested. Positive results were observed for Sodium Dehydroacetate and an emulsion consisting of cetearyl alcohol, sodium lauryl sulfate, and sodium cetearyl sulfate.	No clinical case reports on topical use provided in original report.
Milpied B, Collet E, Genillier N, Vigan M. Allergic contact dermatitis caused by sodium dehydroacetate, not hyaluronic acid, in Ialuset® cream. Contact Dermatitis. 2011 Dec;65(6):359-61.	Clinical Assessment of Safety – Case Report	Nine patients with contact eczema caused by hyaluronic acid cream containing Sodium Dehydroacetate were evaluated. (All had positive patch tests.) The components of the cream were tested in 7 patients. Four patients were found to be positive for Sodium Dehydroacetate (3%).	No clinical case reports on topical use provided in original report.
Foti C, Romita P, Ettorre G, Angelini G, Bonamonte D. Allergic contact dermatitis caused by resorcinol and sodium dehydroacetate in a patient with leg ulcers. Contact Dermatitis. 2016 Jun;74(6):383-4	Clinical Assessment of Safety – Case Report	A 64-year-old non-atopic man presented with eczematous lesions that had developed around leg ulcers, which he had been treating with several creams containing Sodium Dehydroacetate. Three months following the resolution of the dermatitis, patch tests were performed using the creams he used to treat ulcers. Patch tests were then performed on the individual components of the creams in which the patient showed a positive response. Positive reactions were observed for 3% Sodium Dehydroacetate (and 2% resorcinol).	No clinical case reports on topical use provided in original report.
Valois A, Waton J, Avenel-Audran M, Truchetet F, Collet E, Raison-Peyron N, Cuny JF, Bethune B, Schmutz JL, Barbaud A; Dermatology and Allergy group (GAD) of the French Society of Dermatology. Contact sensitization to modern dressings: a multicentre study on 354 patients with chronic leg ulcers. Contact Dermatitis. 2015 Feb;72(2):90-6.	Clinical Assessment of Safety – Multicenter Study	A prospective multicenter study was carried out in patients with chronic leg ulcers in 5 French dermatology departments (n = 354). Patch tests were performed with the European baseline series, 27 allergens, and 10 modern dressings. Forty-five patients showed positive responses to a cream containing Sodium Dehydroacetate. According to study authors, in the majority of these cases, sensitization to this cream was caused by Sodium Dehydroacetate.	No multicenter studies on topical use provided in original report.
Canavez ADPM, de Oliveira Prado Corrêa G, Isaac VLB, Schuck DC, Lorencini M. Integrated approaches to testing and assessment as a tool for the hazard assessment and risk characterization of cosmetic preservatives. J Appl Toxicol. 2021 Oct;41(10):1687-1699	Risk Assessment	Dehydroacetic Acid: MOS of 84.88 (Based on SED: 1.614 mg/kg bw/d; 100% dermal absorption; 60% max concentration; 269 mg/kg bw/d estimated daily exposure; and 137 mg/kg bw/d NOAEL (based on subchronic oral toxicity study; method of oral administration not stated))	MOS calculation not provided in original report

Abbreviations: ADME = absorption, distribution, metabolism, and excretion; CLP = classification, labeling, and packaging; DART = developmental and reproductive toxicity; EU = European Union; GHS = globally harmonized system; LD_{50} = median lethal dose; MOS = margin of safety; NOAEL = no-observed-adverse-effect-level; OECD TG = Organisation for Economic Co-operation and Development Test Guidelines; SED = systemic exposure dose

Search (from 2000 on)
PubMed
(((“Sodium Dehydroacetate”) OR (4418-26-2[CAS Number])) AND (("2000"[Date - Publication] : "2023"[Date - Publication]))

(((“Dehydroacetic Acid”) OR (16807-48-0 [CAS Number])) OR (520-45-6 [CAS Number])) OR (771-03-9 [CAS Number])) AND (("2000"[Date - Publication] : "2023"[Date - Publication]))

Table 1. Frequency (2023/2002) and concentration (2023/2003) of use according to likely duration and exposure and by product category

	Sodium Dehydroacetate				Dehydroacetic Acid			
	# of Uses		*Max Conc of Use (%)*		*# of Uses*		*Max Conc of Use (%)*	
	2023[1]	**2002[2]**	**2023[3]**	**2003[2]**	**2023[1]**	**2002[2]**	**2023[3]**	**2003[2]**
Totals*	**1233**	**325**	**0.000005 – 0.6**	**0.00003 – 0.6**	**833**	**88**	**0.0000029 – 0.7**	**0.007 – 0.7**
summarized by likely duration and exposure**								
Duration of Use								
Leave-On	*1191*	*293*	*0.000005 – 0.6*	*0.00003 – 0.6*	*637*	*68*	*0.0000029 – 0.5*	0.01 – 0.7
Rinse-Off	*42*	*32*	*0.00003 – 0.2*	*0.0001 – 0.3*	*190*	*19*	*0.0000042 – 0.7*	*0.007 – 0.1*
Diluted for (Bath) Use	*NR*	*NR*	*NR*	*NR*	*6*	*1*	*NR*	*NR*
Exposure Type								
Eye Area	331	110	0.0015- 0.6	0.0006 – 0.5	145	9	0.015 – 0.5	0.1 – 0.3
Incidental Ingestion	5	1	0.2 – 0.5	0.3 – 0.5	61	NR	0.064	NR
Incidental Inhalation-Spray	3; 469[a]; 96[b]	52[a]; 24[b]	0.000005[a]	0.001 – 0.5’ 0.001 – 0.5[a]; 0.00003 – 0.5[b]	4; 131[a]; 166[b]	18[a]; 20[b]	0.0000029 – 0.000008; 0.0000042 – 0.64[a]; 0.062[b]	0.03 - 0.2[a]; 0.01 – 0.08[b]
Incidental Inhalation-Powder	83; 96[b]; 2[c]	34; 24[b]	0.2 – 0.5; 0.0005 – 0.6[c]	0.05 – 0.4; 0.00003 – 0.5[b]; 0.6[c]	24; 166[b]; 2[c]	3; 20[b]	0.0019 – 0.5; 0.062[b]; 0.00008 – 0.048[c]	0.7; 0.01 – 0.08[b]
Dermal Contact	1152	293	0.00004 – 0.6	0.00003 – 0.6	690	85	0.000021 – 0.7	0.007 – 0.7
Deodorant (underarm)	NR	2[a]	0.00004	NR	5[a]	NR	NR	NR
Hair - Non-Coloring	31	7	0.000005 – 0.05	0.2	76	1	0.0000029 – 0.08	0.02 – 0.03
Hair-Coloring	NR	3	0.1	NR	NR	NR	0.00003	NR
Nail	2	5	NR	0..02 – 0.2	NR	2	NR	NR
Mucous Membrane	9	3	0.00058 – 0.5	0.0001 – 0.3	99	1	0.000043 – 0.85	0.03
Baby Products	2	NR	0.05 – 0.5	0.6	7	NR	0.048 – 0.071	NR
as reported by product category								
Baby Products								
Baby Shampoos	NR	NR	0.05	NR	3	NR	NR	NR
Baby Lotions/Oils/Powders/Creams	2	0.48	NR	0.6	2	NR	0.048	NR
Other Baby Products	NR	0.071	0.5	NR	2	NR	0.071	NR
Bath Preparations (diluted for use)								
Bath Oils, Tablets, and Salts	NR	NR	NR	NR	NR	1	NR	NR
Bubble Baths	NR	NR	NR	NR	5	NR	NR	NR
Other Bath Preparations	NR	NR	NR	NR	1	NR	NR	NR
Eye Makeup Preparations								
Eyebrow Pencil	3	NR	NR	0.2 – 0.3	3	NR	NR	NR
Eyeliner	36	4	0.34 – 0.51	0.05 – 0.5	3	NR	0.15	0.1
Eye Shadow	204	74	0.3 – 0.6	0.05 – 0.3	99	4	0.05 – 0.5	0.3
Eye Lotion	8	3	0.0015 – 0.2	NR	16	NR	0.1	0.2
Eye Makeup Remover	1	1	NR	0.05	4	5	0.084	0.1
Mascara	43	16	0.3 – 0.5	0.001 – 0.4	5	NR	0.015 – 0.02	0.2
Other Eye Makeup Preparations	36	12	0.32 – 0.45	0.0006 – 0.4	15	NR	NR	NR
Fragrance Preparations								
Cologne and Toilet Water	1	NR	NR	0.001 – 0.5	NR	NR	NR	NR
Powders (dusting/talcum, excl aftershave talc)	NR	3	NR	NR	NR	NR	NR	NR
Other Fragrance Preparation	2	NR	NR	NR	3	NR	NR	NR
Hair Preparations (non-coloring)								
Hair Conditioner	10	NR	0.00003 – 0.015	0.2	27	NR	0.0000042 – 0.08	NR
Hair Spray (aerosol fixatives)	NR	NR	NR	NR	1	NR	0.0000029 - 0.000008	NR
Shampoos (non-coloring)	8	2	0.00003 – 0.0005	0.2	33	NR	0.000007 – 0.079	0.02 – 0.03
Tonics, Dressings, and Other Hair Grooming Aids	5	1	0.000005	NR	5	1	0.0000042 – 0.064	NR
Other Hair Preparations	2	4	NR	NR	6	NR	0.048 – 0.08	NR
Hair Coloring Preparations								
Hair Dyes and Colors (all types requiring caution statements and patch tests)	NR	NR	0.1	NR	NR	NR	0.00003	NR
Hair Tints	NR	12	NR	NR	NR	1	NR	NR
Other Hair Coloring Preparation	NR	2	NR	NR	NR	2	NR	NR
Makeup Preparations								
Blushers (all types)	69	15	0.3 – 0.45	0.1 – 0.4	31	1	NR	0.05 – 0.2
Face Powders	83	31	0.2 – 0.5	0.05 – 0.4	24	3	0.5	0.7
Foundations	70	10	0.00049 – 0.5	0.0001 – 0.4	1	3	NR	0.1
Leg and Body Paints	NR	NR	NR	0.1	NR	NR	NR	NR
Lipstick	5	1	0.2 – 0.5	0.3	60	NR	0.064	NR
Makeup Bases	5	6	0.00049	0.1	2	NR	NR	NR
Rouges	1	NR	0.35	NR	NR	1	NR	NR

Table 1. Frequency (2023/2002) and concentration (2023/2003) of use according to likely duration and exposure and by product category

	Sodium Dehydroacetate				Dehydroacetic Acid			
	# of Uses		*Max Conc of Use (%)*		*# of Uses*		*Max Conc of Use (%)*	
	2023[1]	**2002[2]**	**2023[3]**	**2003[2]**	**2023[1]**	**2002[2]**	**2023[3]**	**2003[2]**
Makeup Fixatives	NR	1	NR	NR	1	NR	NR	NR
Other Makeup Preparations	15	4	0.45	0.0003 – 0.2	27	NR	NR	0.07
Manicuring Preparations (Nail)								
Basecoats and Undercoats	NR	NR	NR	0.02	NR	NR	NR	NR
Cuticle Softeners	NR	2	NR	NR	NR	1	NR	NR
Nail Creams and Lotions	NR	3	NR	NR	NR	NR	NR	NR
Nail Polish and Enamel	1	NR	NR	0.2	NR	1	NR	NR
Other Manicuring Preparations	1	NR	NR	0.2	NR	NR	NR	NR
Oral Hygiene Products								
Other Oral Hygiene Products	NR	NR	NR	NR	1	NR	NR	NR
Personal Cleanliness Products								
Bath Soaps and Detergents	2	2	0.00058	0.0001	16	NR	0.000043 – 0.07	0.03
Deodorants (underarm)	NR	2	0.00004	NR	5	NR	NR	NR
Douches	NR	NR	NR	NR	1	NR	NR	NR
Feminine Deodorants	NR	NR	NR	NR	NR	NR	0.062	NR
Other Personal Cleanliness Products	2	NR	NR	NR	15	NR	0.064 – 0.085	0.03
Shaving Preparations								
Aftershave Lotion	5	1	NR	0.0003	1	NR	0.000066	NR
Beard Softeners	NR	NR	NR	NR	2	NR	NR	NR
Shaving Cream	4	4	NR	NR	1	NR	0.000024 – 0.003	NR
Other Shaving Preparations	NR	1	NR	NR	NR	NR	NR	NR
Skin Care Preparations								
Cleansing	8	13	0.0003 – 0.2	0.0003 – 0.3	65	8	NR	0.007 – 0.02
Face and Neck (exc shave)	96	4	0.0005 – 0.5	0.008 – 0.2	102	11	NR	0.01 – 0.08
Body and Hand (exc shave)	19	20	0.3 – 0.6	0.00003 – 0.5	64	9	NR	0.03 – 0.05
Moisturizing	455	39	0.1	0.001 – 0.3	96	10	NR	NR
Night	5	5	NR	0.003 – 0.2	20	20	NR	0.03
Paste Masks (mud packs)	1	6	0.00027 – 0.1	0.03 – 0.2	23	6	NR	NR
Skin Fresheners	4	2	NR	NR	7	NR	NR	NR
Other Skin Care Preparations	15	25	NR	0.00003 – 0.1	31	16	NR	0.03
Suntan Preparations								
Suntan Gels, Creams, and Liquids	NR	1	0.01 – 0.05	0.2	2	NR	0.05	0.2
Indoor Tanning Preparations	NR	2	NR	0.4	NR	5	NR	NR
Other Suntan Preparations	NR	2	0.5	0.1	1	NR	NR	NR

NR – not reported

*Because each ingredient may be used in cosmetics with multiple exposure types, the sum of all exposure types may not equal the sum of total uses.

**likely duration and exposure are derived based on product category (see Use Categorization https://www.cir-safety.org/cir-findings)

[a] It is possible these products are sprays, but it is not specified whether the reported uses are sprays.

[b] Not specified whether a spray or a powder, but it is possible the use can be as a spray or a powder, therefore the information is captured in both categories

[c] It is possible these products are powders, but it is not specified whether the reported uses are powders.

1. US Food and Drug Administration (FDA) Center for Food Safety & Applied Nutrition (CFSAN). 2023. Voluntary Cosmetic Registration Program - Frequency of Use of Cosmetic Ingredients. (Obtained under the Freedom of Information Act from CFSAN; requested as "Frequency of Use Data" January 4, 2023; received February 2, 2023).

2. Andersen FA (ed). Annual review of cosmetic ingredient safety assessments - 2004/2005. *IJT*. 2006;25:65-68.

3. Personal Care Products Council. 2023. Concentration of use by FDA product category: Sodium Dehydroacetate and Dehydroacetic Acid. Unpublished data submitted by Personal Care Products Council on February 23, 2023.

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4

Final Report on the Safety Assessment of Sodium Dehydroacetate and Dehydroacetic Acid

Sodium Dehydroacetate and Dehydroacetic Acid are used as preservatives in cosmetic formulations at concentrations of 1.0 percent or less. Both compounds are rapidly absorbed when administered orally or on the skin of test animals.

Acute toxicity studies indicate that Sodium Dehydroacetate and Dehydroacetic Acid are slightly toxic when administered orally to rats. Neither compound was an irritant when applied to rabbit skin. Sodium Dehydroacetate was found to exhibit minimal eye irritation. Subchronic and chronic studies reveal various toxic effects, primarily due to the incurred lack of appetite and weight loss. No evidence of mutagenicity was reported for either ingredient use. No evidence of tumor induction by Dehydroacetic Acid was detected in a 64-week study. Dehydroacetic Acid had an inhibitory effect on hepatoma induction in rats when fed 4-(dimethylamino)azobenzene. A teratogenicity study in mice revealed no significant findings when compared to untreated controls.

Sodium Dehydroacetate, Dehydroacetic Acid, and cosmetics containing these ingredients were found practically nonirritating, nonsensitizing, nonphotosensitizing, and nonphototoxic in numerous clinical tests. On the basis of the available animal and clinical data, it is concluded that Sodium Dehydroacetate and Dehydroacetic Acid are safe as cosmetic ingredients in the present practices of use and concentration.

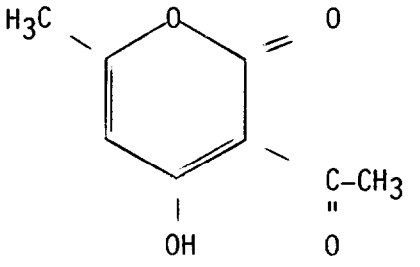
INTRODUCTION

Sodium Dehydroacetate is the sodium salt of dehydroacetic acid. The acid is used as a cosmetic preservative. However, because of its limited aqueous solubility, the freely soluble sodium salt may be used in manufacturing processes.[1] Consequently, these two compounds have been tested concurrently and at times used interchangeably.

CHEMISTRY

Structure and Preparation

Sodium Dehydroacetate is the sodium salt of the cyclic ketone Dehydroacetic Acid. Dehydroacetic Acid conforms to the following structural formula[2]:



Dehydroacetic Acid

Dehydroacetic Acid can be prepared by the action of N-bromosuccinimide on ketene dimer and by strong heating of acetoacetic ester.[3,4]

Description and Properties

Sodium Dehydroacetate occurs as a tasteless, odorless, white powder. It is soluble in water, propylene glycol, glycerin, and methanol but insoluble in most organic solvents.[3,5]

Dehydroacetic Acid is a white to cream crystalline powder with practically no odor or taste. Although only slightly soluble in water, Dehydroacetic Acid dissolves in aqueous solutions of fixed alkalies, acetone, benzene, ether, and methanol.[5,6] Table 1 summarizes the physicochemical properties of Dehydroacetic Acid and its sodium salt.

Reactivity/Stability

Dehydroacetic Acid is a reactive, unstable, and combustible compound.[3,7] Profoundly affected by pH, the acid is active only in its undissociated state; 65 percent is undissociated at pH 5 in comparison to 6 to 7 percent at pH 6.5. The degree of effectiveness of Dehydroacetic Acid and Sodium Dehydroacetate against various bacteria and fungi is greatest in acidic media.[13–17] The minimum inhibitory concentration (MIC) of Dehydroacetic Acid against *Escherichia coli* was found to increase sharply with increasing pH.[1] However, Dehydroacetic Acid retains partial antimicrobial activity under alkaline conditions, although the corresponding MICs are much greater.[18,19]

Sodium Dehydroacetate is stable at room temperature and in solution for 1 hour at temperatures up to 130°C. The salt is subject to discoloration by iron.[20]

Aqueous solutions of Dehydroacetic Acid (0.05 percent) stored at room temperature in polyethylene, polyvinyl chloride, and glass containers were found to be unstable as determined by spectrophotometry, with concentration decreases of 63 percent, 21 percent, and 11 percent after 12, 12, and 6 weeks, respectively. Solutions of 0.08 percent Dehydroacetic Acid with pHs of 3.87 (aqueous), 5.5

TABLE 1. Physicochemical Properties of Sodium Dehydroacetate and Dehydroacetic Acid

Property	Value: Sodium Dehydroacetate	Value: Dehydroacetic Acid	Reference
Empirical formula	$C_8H_7NaO_4$	$C_8H_8O_4$	8
Molecular weight	208.15 (hydrated)	168.16	4,6
Boiling point (°C)	–	270 at 760 mm Hg	4
	–	132-3 at 5 mm Hg	4
Melting point (°C)	–	109–111 (Sublimes)	5,4
Vapor pressure (mm)	–	1 at 91.7°C	8
Vapor density	–	5.8	8
Heat of vaporization (gram calories per gram mole)	–	14,663.8	4
λ max (nm)	–	308 (10 to 35 percent alcohol); 312	9,10
Sadtler Standard Spectra Reference No.			
Infrared (IR) Prism	–	7172	4
NMR	–	V504	4
Solubility (g per 100 g solvent at 25°C)			5,4
Acetone	0.2	22	
Benzene	<0.1	18	
Carbon tetrachloride	<0.1	3	
95 percent ethanol	1	3	
Ether	<0.1	5	
Glycerin	15	<0.1	
n-Heptane	<0.1	0.7	
Methanol	14	5	
Olive oil	<0.1	1.6	
Propylene glycol	48	1.7	
Water	33	<0.1	
Assay			
As ingredient (anhydrous basis)	98.0 percent minimum $C_8H_7NaO_4$	98.0 percent minimum $C_8H_8O_4$	6,11
Arsenic as As*	3 ppm	3 ppm	
Heavy Metals as Pb*	10 ppm; 20 ppm	10 ppm; 20 ppm	
Sulfated ash*	–	0.10 percent	11
Water	8.5–10 percent	–	6
Residue on ignition*	–	0.1 percent	6,12
Loss on drying*	8.3–9.5 percent	1.0 percent	12,11

*Maximum recommended.

(buffered), and 6.5 (buffered) showed degradations of 53 percent, 58.8 percent, and 18.5 percent, respectively, after a storage period of 32 weeks.[1]

Aqueous solutions of Dehydroacetic Acid (0.1 percent) subjected to gamma-radiation for periods of 22.25 and 44.5 hours had degradations of 76 percent and 75 percent, respectively.[21] In a study of the effects of heating on preservatives,

Sodium Dehydroacetate had a decrease in MIC of 860 ppm after heating with the spores of *Bacillus subtilis* at 85°C for 30 minutes. No change occurred in the MIC when spores and preservative were heated separately.[16]

Potential Interaction with Other Cosmetic Ingredients

Studies of the effect of cosmetic nonionic surfactants on Sodium Dehydroacetate and Dehydroacetic Acid have produced conflicting results. Patel[22] concluded that Dehydroacetic Acid was an effective preservative in the presence of the nonionic emulsifiers, cetomacrogol 1000 and polysorbate 80, after finding that even in a medium (pH 6.1) containing 5 percent of either nonionic, the MIC of Dehydroacetic Acid against *Aspergillus niger* increased by only 0.005 percent (from 0.040 percent to 0.045 percent). The corresponding MIC against *Aerobacter aerogenes* increased by only 0.025 percent (from 0.075 percent to 0.10 percent). Barr and Tice[23] studied the effect of 20 percent sorbitol solutions on Dehydroacetic Acid (as Sodium Dehydroacetate) at a concentration of 0.4 percent. The growth of *Staphylococcus aureus, B. subtilis, Pseudomonas aeruginosa,* and *Candida albicans* was inhibited, although growth of *A. niger* and *Penicillium notatum* was not. McCarthy[1] has questioned the high concentration of Dehydroacetic Acid used by Barr and Tice and suggested that this could have been necessary due to the deleterious effects of autoclaving sorbitol and Dehydroacetic Acid, the presence of the sorbitol, or the use of particularly resistant organisms.

Wedderburn[13] found that at a concentration of 0.3 percent (pH of 5.5), Dehydroacetic Acid was completely inactivated by 2 percent concentrations of nonionic surfactants, including polysorbate 80. Her findings have been supported by the work of DeNavarre.[14] Wallhaeusser[24] has also reported the inactivation of Dehydroacetic Acid by nonionics, anti-inhibition agents used in preservation stress tests, and high microbial counts.

Sodium Dehydroacetate had an additive antimicrobial effect in combination with the cosmetic preservatives, sodium benzoate and butyl *p*-hydroxybenzoate,[25] and with butyl *p*-hydroxybenzoate alone.[26]

Lowe[27] documented the synthesis of 4-hydroxy-5-hydroxyimino-7-methyl-5H-pyrano-2,3-bpyridine 8-oxide by the reaction between Dehydroacetic Acid, hydroxylamine, and N,N-dimethyl-formamide dimethyl acetal. This indicates that Dehydroacetic Acid could potentially react with the hydroxylamine group of hydroxylamine hydrochloride and hydroxylamine sulfate. Both of these hydroxylamines are listed as cosmetic ingredients; however, their use was not reported in the 1981 FDA product formulation data voluntarily submitted by industry.[28]

Analytical Methods

Qualitative and/or quantitative determinations of Sodium Dehydroacetate and Dehydroacetic Acid have been made by thin-layer chromatography,[9,29-30] gas chromatography with or without mass spectrometry,[29] infrared spectrometry,[11,29] fluorometry,[29,31,32] UV spectrophotometry,[9,10,33] partition chromatography,[9] and colorimetry.[10] Identification can be made of both Sodium Dehydroacetate and Dehydroacetic Acid by comparison to standard infrared spectra[11] and by various physical and chemical assays.[6,12]

Analytical methods for the determination of clarity and color, arsenic con-

tent, heavy metal content, presence of readily carbonizable substances, and loss on drying have been described for Sodium Dehydroacetate and Dehydroacetic Acid.[6,12] Additionally, assays are defined for measuring the melting point and residue on ignition for Dehydroacetic Acid and water, alkali, chloride, and sulfate content for Sodium Dehydroacetate.

USE

Purpose in Cosmetics

Sodium Dehydroacetate and Dehydroacetic Acid are used as preservatives and antimicrobial agents in cosmetic products at concentrations of 1 percent or less.[28] Both are active at low concentrations, with an optimal pH of 2 to 4.[14,34]

Scope and Extent of Use in Cosmetics

Table 2 presents the FDA product formulation data for Sodium Dehydroacetate and Dehydroacetic Acid.[28] This cosmetic product formulation computer printout is made available by the FDA and is compiled through voluntary filing of such data in accordance with Title 21, part 720.4 of the Code of Federal Regulations.[35] Ingredients are listed in prescribed concentration ranges under specific product type categories. Since certain cosmetic ingredients are supplied by the manufacturer at less than 100 percent concentration, the value reported by the cosmetic formulator may not necessarily reflect the actual concentration found in the finished product; the actual concentration in such a case would be a fraction of that reported to the FDA. The fact that data are only submitted within the framework of preset concentration ranges also provides the opportunity for overestimation of the actual concentration of an ingredient in a particular product. An entry at the lowest end of a concentration range is considered the same as one entered at the highest end of that range, thus introducing the possibility of a two- to ten-fold error in the assumed ingredient concentration.

Sodium Dehydroacetate and Dehydroacetic Acid are used in a wide variety of products, including bath, skin care, suntan, fragrance, and hair preparations, eye and facial makeups. Sodium Dehydroacetate is additionally used in shaving and manicuring preparations.[28] The literature contains some references to the use of Sodium Dehydroacetate in toothpaste and a mouthwash[3,36] and the use of Dehydroacetic Acid in toothpastes.[4] In 1981, Sodium Dehydroacetate and Dehydroacetic Acid were used in a total of 260 and 139 formulations, respectively. All of these uses were at concentrations of 1 percent or less.[28]

Dehydroacetic Acid and its salts have been approved as provisionally permitted substances on the European Economic Community (EEC) proposed cosmetic preservative list. The maximum concentration, expressed as the acid, is 0.6 percent; however, it is noted that higher concentrations may be used for specific purposes.[37]

Frequency of Use and Surfaces to which Applied

Products containing Sodium Dehydroacetate and Dehydroacetic Acid may contact all areas of the skin, the hair, ocular mucosa, and nails with possible inci-

TABLE 2. Product Formulation Data[28]

Product Category*	Total No. Containing Ingredient	No. Product Formulations Within Each Concentration Range (percent)*	
		>0.1–1	≤0.1
Sodium Dehydroacetate			
Bath oils, tablets, and salts	1	–	1
Eyeliner	2	1	1
Eye shadow	56	53	3
Mascara	13	11	2
Other eye makeup preparations	4	4	–
Fragrance powders (dusting and talcum, excluding aftershave talc)	1	1	–
Tonics, dressings, and other hair grooming aids	1	–	1
Blushers (all types)	22	17	5
Face powders	23	19	4
Makeup foundations	8	7	1
Makeup bases	14	14	–
Rouges	2	1	1
Other makeup preparations (not eye)	2	2	–
Cuticle softeners	4	4	–
Nail creams and lotions	2	1	1
Other manicuring preparations	1	1	–
Aftershave lotions	1	–	1
Shaving cream (aerosol, brushless, and lather)	1	1	–
Other shaving preparation products	1	1	–
Skin cleansing preparations (cold creams, lotions, liquids, and pads)	23	19	4
Face, body, and hand skin care preparations (excluding shaving preparations)	24	17	7
Moisturizing skin care preparations	27	24	3
Night skin care preparations	7	–	7
Paste masks (mud packs)	4	3	1
Skin lighteners	2	1	1
Skin fresheners	2	2	–
Wrinkle smoothers (removers)	1	1	–
Suntan gels, creams, and liquids	5	5	–
Indoor tanning preparations	3	1	2
Other suntan preparations	3	3	–
1981 TOTALS	260	214	46

TABLE 2. (Continued)

*Product Category**	*Total No. Containing Ingredient*	*No. Product Formulations Within Each Concentration Range (percent)**	
		>0.1–1	*≤0.1*
Dehydroacetic Acid			
Bath oils, tablets, and salts	1	–	1
Bubble baths	2	–	2
Eyeliner	1	1	–
Eye shadow	11	6	5
Eye makeup remover	8	6	2
Mascara	1	1	–
Other eye makeup preparations	9	1	8
Colognes and toilet waters	4	–	4
Perfumes	4	–	4
Hair shampoos (noncoloring)	2	–	2
Tonics, dressings, and other hair grooming aids	2	1	1
Blushers (all types)	5	4	1
Face powders	6	1	5
Makeup foundations	13	12	1
Lipstick	1	–	1
Makeup bases	1	–	1
Rouges	1	1	–
Other makeup preparations (not eye)	1	–	1
Skin cleansing preparations (cold creams, lotions, liquids, and pads)	15	2	13
Face, body, and hand skin care preparations (excluding shaving preparations)	16	4	12
Moisturizing skin care preparations	10	3	7
Night skin care preparations	5	2	3
Paste masks (and packs)	3	2	1
Skin fresheners	2	–	2
Wrinkle smoothers (removers)	2	–	2
Other skin care preparations	9	4	5
Suntan gels, creams, and liquids	3	3	–
Other suntan preparations	1	1	–
1981 TOTALS	139	55	84

*Preset product categories and concentration ranges in accordance with federal filing regulations (21 CFR 720.4); see Scope and Extent of Use in Cosmetics.

dental exposure of the vaginal mucosa (Table 2). These products may be used daily or occasionally over a period of up to several years. The frequency and length of application could result in continuous exposure.

Noncosmetic Uses

Sodium Dehydroacetate and Dehydroacetic Acid are listed as indirect food additives for use as preservatives for adhesives in food packaging.[35] Both ingredients are also listed as direct food additives for use as preservatives for cut or peeled squash, with the stipulation that no more than 65 ppm expressed as Dehydroacetic Acid remains in or on the treated squash.[38] Sodium Dehydroacetate has been found to be effective in preventing microbial decay of strawberries, raspberries (Ireland), sweet potatoes (US), mango pulp (India), dates (Iraq), and grapes (Dehydroacetic Acid also, Japan), in addition to the discoloration and decay of shredded lettuce and snap beans.[15,39-47] Dehydroacetic Acid is also used as a mold inhibitor on hams and bacons in the Philippines.[48] Sodium Dehydroacetate is used as a preservative in Japan for cheese, butter, and margarine.[49]

Sodium Dehydroacetate and Dehydroacetic Acid have limited use as pharmaceutical preservatives.[1,3] In a Japanese study, "prohibited" Dehydroacetic Acid was found in a stimulating "drink."[50] Both ingredients are used as preservatives, bactericides, and fungicides in veterinary drugs.[3,51] Rossoff[51] states that Dehydroacetic Acid is a natural product of adrenal steroid metabolism and prevents poultry death caused by exposure to extreme hot and cold temperatures.

Sodium Dehydroacetate and Dehydroacetic Acid are widely used as industrial preservatives and antimicrobials; specifically cited uses include starch glue, cleaning solutions, and deodorants used in toilets and for human waste treatment.[3,4,52-57] Both also are used as plasticizers and are compatible with nitrocellulose, polystyrene, methacrylate, and vinylite resins.[3,4]

Sodium Dehydroacetate and Dehydroacetic Acid are effective pesticides and insecticides against slugs, ants, mites, and cockroaches.[58-60]

GENERAL BIOLOGY

Absorption, Metabolism, and Excretion

Absorption occurs rapidly when Sodium Dehydroacetate and Dehydroacetic Acid are administered orally to man, monkey, dog, and rat, as determined in an extensive series of absorption and distribution studies conducted by Woods et al.[10] Three men ingested a daily dose of 500 mg of Dehydroacetic Acid for 115 or 118 days, after which the dosage was raised to 750 mg per day for 35 or 38 days. A slight increase in the plasma Dehydroacetic Acid concentration occurred, but plasma Dehydroacetic Acid remained relatively constant (with constant dose) at 10 to 15 mg/100 ml after approximately 15 to 20 days. Plasma concentrations increased slightly with increased doses. In a study on the effect of alcohol on the absorption of Dehydroacetic Acid, 3 women and 7 men (five groups of 2) ingested daily 380 ml of wine containing 0 (two groups), 225 (one group), and 445 ppm (two groups) Dehydroacetic Acid. The plasma concentration of Dehydroacetic Acid was directly related to the amount ingested. Also, the investigators concluded that absorption was unaffected by alcohol.

Monkeys (2 per dose group) received oral doses of 50, 100, and 200 mg/kg per day, 5 days per week, for 290 to 397 days. Dehydroacetic Acid was administered in olive oil to 1 monkey in each group and Sodium Dehydroacetate in water to the other. Plasma concentrations of Dehydroacetic Acid were determined at various intervals after dosing. The peak concentration occurred between 4½ and 7 hours after dosing (plasma levels at 4 hours from low dose to high were 15, 26 to 33, and 45 to 51 mg/100 ml, respectively), and diminished rapidly and progressively from high- to low-dose groups. Olive oil apparently had no influence on absorption.[10]

Sodium Dehydroacetate was administered in a single dose of 160 mg/kg (Dehydroacetic Acid) to each of 4 dogs—2 orally, 2 intravenously.[10] The collection and analysis of blood samples revealed rapid absorption, with maximum plasma concentrations (approximately 22 to 26 mg/100 ml) occurring 1½ to 2 hours after administration and diminishing slowly thereafter; traces of Dehydroacetic Acid remained after 3 to 4 days. This slow elimination of Dehydroacetic Acid in the dog was supported by another experiment in which single intravenous doses of 20 to 480 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) were given to 6 dogs. Similar results were obtained; Dehydroacetic Acid was detectable after 3 days at doses of 80 mg/kg and above. Two deaths were reported at 2 and 3 days after dosing, 480 and 320 mg/kg dose levels, respectively.

Rats in four groups of 12 each were fed diets containing 0.02, 0.05, 0.1, and 0.2 percent Dehydroacetic Acid (as Sodium Dehydroacetate).[10] Two rats from each group were killed every 2 days for 12 days, and total Dehydroacetic Acid intake and blood plasma concentrations were determined. Plasma concentrations increased correspondingly with increased dose. Maximum plasma concentrations of Dehydroacetic Acid from low dose to high were approximately 6, 8, 11, and 18 mg/100 ml, respectively. Mean Dehydroacetic Acid plasma concentrations in rats on a 2-year chronic oral toxicity study were directly related to the doses of 0.02, 0.05, and 0.1 percent Dehydroacetic Acid. Mean plasma concentrations (determined after 730 days) for the males from low to high dose were 0.4, 1.6, and 5.8 mg/100 ml, respectively; means for the females were 0.8, 3.2, and 10 mg/100 ml, respectively.

In a study of the effect of caffeine on the gastric absorption of Dehydroacetic Acid in rabbits, Goto et al.[61] found that the absorption rate constants (per hour) for Dehydroacetic Acid alone and for the soluble Dehydroacetic Acid–caffeine complex were 0.98 and 0.40, respectively. The Dehydroacetic Acid absorption rate decreased proportionately to increased concentrations of caffeine.

A skin absorption study in rabbits was conducted by the Draize sleeve technique, with the exception that the animals were not restrained in their cages and the sleeves were wrapped in heavy cloth bandages.[5] After a 24-hour exposure period, the sleeves were removed, and the skin was cleansed with soap and water. All rabbits were observed for 2 weeks afterwards. Dehydroacetic Acid was applied as a 50 percent suspension in a greaseless ointment base at doses of 1.0, 3.0, and 5.0 g/kg. Animals of the 1.0 and 3.0 g/kg groups had only slight and transitory weight loss. One of the two rabbits exposed to the 5.0 g/kg dose died. Sodium Dehydroacetate was also applied to 5 animals at a dose of 5.0 g/kg in a water slurry; no toxic effects were noted. In another study using the same sleeve technique, the absorption of Dehydroacetic Acid and Sodium Dehydroacetate applied in different vehicles to the skin of rabbits was determined.[10] Equimolar

concentrations of both compounds were absorbed to the same degree from a washable base. Absorpton of Dehydroacetic Acid did not differ greatly between washable base or white petrolatum vehicles; approximately 50 percent more Sodium Dehydroacetate was absorbed from the washable base than from the aqueous solution. Doses ranged from 0.01 to 2.0 g/kg, and concentrations of Dehydroacetic Acid applied were 0.1, 1.0, and 10.0 percent. The plasma Dehydroacetic Acid concentrations increased correspondingly with increasing dose and concentration.

The effects of Dehydroacetic Acid on the respiratory metabolism of various rat tissues were determined using the conventional Warburg technique to measure oxygen uptake and anaerobic glycolysis.[62] The lowest concentration at which effects were consistent was 2.3×10^{-3}M. Oxygen consumption of cerebral and kidney mince was inhibited 10 to 20 percent at this concentration and progressively inhibited up to 40 to 60 percent at 9.3×10^{-2}M Dehydroacetic Acid. Liver mince oxygen uptake increased at all concentrations up to 9.3×10^{-2}M. Skeletal muscle mince oxygen consumption was inhibited at the lower concentrations but markedly increased by 60 to 80 percent at the higher concentrations. Liver and cerebrum slices were also studied. Cerebral cortex slices were affected similarly to the mince. Oxygen consumption of liver slices in the absence of substrate was not decreased by 9.3×10^{-3}M Dehydroacetic Acid or less; however, concentrations above this produced increasing degrees of inhibition. The inhibition of oxygen consumption by cerebral slices was not reversible after washing. Dehydroacetic Acid effects on oxygen consumption of rat cerebral mince with or without glucose, lactate, pyruvate, succinate, fumarate, and malate were determined. Concentrations of Dehydroacetic Acid that inhibited total oxygen uptake did not influence substrate oxygen utilization, with the exception of succinate inhibition.[62] Sodium Dehydroacetate also inhibited succinic acid dehydrogenase in rabbit intestine.[63]

In additional experiments conducted by Woods et al.,[10] Dehydroacetic Acid was bound to plasma proteins in rats, humans, and dogs, as determined by ultrafiltration, centrifugation, and dialysis. Approximately 90 and 98 percent of the Dehydroacetic Acid in rat and human plasma were bound to protein at Dehydroacetic Acid plasma concentrations of 5 to 6 and up to 6 mg/100 ml, respectively. The maximum absolute binding in the dog was reached at Dehydroacetic Acid plasma concentrations of 70 to 100 mg/100 ml; higher concentrations had a lower absolute value. The percentage of binding varied inversely with total plasma Dehydroacetic Acid concentration, irrespective of route or duration of administration. Barman et al.[64] later showed, by electrophoresis of human serum incubated with ^{14}C-Dehydroacetic Acid, that Dehydroacetic Acid was mainly associated with serum albumin and to a lesser degree with serum globulins.

Distribution of Dehydroacetic Acid between erythrocytes and plasma was determined in dogs and humans.[10] Samples taken after intravenous administration of 240 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) to dogs had a plasma Dehydroacetic Acid concentration two to two and one-half times greater than that of erythrocytes. In a human ingesting 750 mg Dehydroacetic Acid daily, the ratio of plasma to erythrocytic Dehydroacetic Acid was 1.76:1.

A dog receiving 80 mg/kg per day Dehydroacetic Acid for 49 days gave birth to a litter of pups. All tissues of 1 newborn examined contained a mean of 13 mg/

100 ml Dehydroacetic Acid, demonstrating that the placenta was permeable to Dehydroacetic Acid.[10] The mother and another pup had similar plasma concentrations of Dehydroacetic Acid, 12 and 10 mg/100 ml, respectively.

Dehydroacetic Acid has been detected in the milk of a postpartum dog that had been receiving Dehydroacetic Acid daily and in the spinal fluid of dog and man. A man ingesting 1200 mg Dehydroacetic Acid daily for 6 days had a spinal fluid concentration of 2.3 mg/100 ml. This concentration was comparatively lower than that in dogs (determined to be 19 and 16 mg/100 ml after intravenous and oral administration of Sodium Dehydroacetate, respectively).[10]

Dehydroacetic Acid produced a stimulating effect on drug-metabolizing enzymic activity.[65,66] Takabatake et al.[65] demonstrated this in their study on female rats subcutaneously injected with 200 mg/kg Dehydroacetic Acid daily for 7 or 10 days. The administration of Dehydroacetic Acid shortened the duration of the hypnotic state caused by 100 mg/kg hexobarbital injected intraperitoneally. The Dehydroacetic Acid-treated rats had higher activity of N-demethylation of aminopyrine, hydroxylation of hexobarbital or cyclobarbital, aromatic hydroxylation of aniline, and hydrolysis of parathion by the liver than had controls. Dehydroacetic Acid increased the glycogen content but had no effect on the fat content of the liver.

Dehydroacetic Acid produced a marked increase in succinic acid excretion in 12 rats and 4 dogs receiving oral doses of 600 mg/kg for 2 days and 200 mg/kg for 3 days, respectively.[62] Urinary samples were taken 24 hours after each dose in rats and 24 hours after the first dose and continuing for 8 days in the dogs. The authors stated that increased urinary succinic acid would be expected if the oxidation of succinic acid was inhibited in vivo, as previously proposed.[63]

Multiple studies were conducted by Shideman et al.[67] on the detoxification and renal effects of Dehydroacetic Acid and Sodium Dehydroacetate in dogs, monkeys, and humans. Total urinary excretion was only 15 to 20 percent of single oral and intravenous doses of 160 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) to the dog, approximately half of which was excreted in the first 24 hours. After daily oral and intravenous doses of 80 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) to 4 dogs, excretion was no greater than 20 percent of the daily dose. Monkeys receiving 50, 100, and 200 mg/kg Dehydroacetic Acid or Sodium Dehydroacetate (doses represent weight of Dehydroacetic Acid) by stomach tube 5 days per week had the same renal elimination of Dehydroacetic Acid as the dog. Samples of urine were collected from 3 men ingesting Dehydroacetic Acid at doses ranging from 6.1 to 12.5 mg/kg per day. The percentage of daily dose recovered in the urine varied between 12.4 and 29.2 (mean of 22.2). Comparatively, this was greater than that in monkeys and dogs; however, the investigators commented that the doses for man were much lower, although plasma concentrations were relatively the same. They concluded that the percentage of dose of Dehydroacetic Acid or Sodium Dehydroacetate eliminated in a 24-hour period was dependent on the plasma concentration and, therefore, on daily intake.

Shideman et al.[67] studied the renal clearance rates of Dehydroacetic Acid in dog and man. During continuous intravenous infusion in the dog of 0.5 mg/kg per minute of Dehydroacetic Acid, maximum clearance was 1 ml per minute at plasma concentrations of 5 to 15 mg/100 ml. The authors noted that this low value was misleading due to the fact that calculations were based on total Dehy-

droacetic Acid plasma concentration when a substantial portion was bound to plasma proteins and, therefore, was not available. An experiment on a single dog with calculations based on concentrations of "free" plasma Dehydroacetic Acid demonstrated that 98 to 99 percent of the dose was reabsorbed by the renal tubules. The clearance rates in 2 men ingesting Dehydroacetic Acid daily (1 receiving 200 to 400 mg Dehydroacetic Acid four times per day for 17 days) varied from 0.43 to 2.86 ml per minute (means of 1.49 and 1.14 ml per minute).

Fecal excretion was determined by Shideman et al.[67] in dogs receiving 80 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) per day orally or intravenously. Feces of dogs administered orally contained 5 percent of the daily dose; those administered intravenously, only 1.3 percent. It was suggested that the presence of Dehydroacetic Acid in bile may account for a portion of the excreted Dehydroacetic Acid in those animals receiving intravenous doses.

Degradation of Dehydroacetic Acid at concentrations of 2.3×10^{-3} to 9.3×10^{-2}M was not demonstrated in vitro by rat liver, kidney, or cerebral cortex slices or skeletal muscle mince.[67] However, the investigators believed that it does occur slowly in vivo because 70 to 80 percent of Dehydroacetic Acid administered was not accounted for by excretion.

Barman et al.[64] studied the metabolites of Dehydroacetic Acid labeled with ^{14}C (in four positions) in both rats and rabbits. Oral doses of 20 to 70 mg/kg ^{14}C-Dehydroacetic Acid were not rapidly excreted. Rabbits excreted 70 to 80 percent of the ^{14}C label in the urine, 7 to 10 percent in respiratory CO_2, and 2 to 3 percent in the feces after 3 to 7 days, leaving 8 to 11 percent in the tissues. Rats excreted 20 to 40 percent in the urine, 10 to 25 percent in respiratory CO_2, and 10 to 20 percent in the feces after 4 to 5 days, leaving 5 to 26 percent in the tissues. Three metabolites were identified from rabbit urine: triacetic acid lactone (TAL), a hydroxy-Dehydroacetic Acid, and a compound (designated metabolite X) believed to be the salt of TAL 3-carboxylic acid. Another metabolite (designated Y) was detected, determined not to be a pyrone, but not further identified. Dehydroacetic Acid and hydroxy-Dehydroacetic Acid occurred in the urine both as the free compounds and as the 1'-imino derivatives. Urine from rabbits contained an average of 5 percent Dehydroacetic Acid of the administered dose, 20 percent hydroxy-Dehydroacetic Acid, 10 percent TAL, 0.3 percent urea, and 20 and 15 percent of metabolites X and Y, respectively. Urine from rats contained 5 percent Dehydroacetic Acid, 8 percent hydroxy-Dehydroacetic Acid, 1 percent TAL, and 0.3 percent urea. The percentages of dose excreted as Dehydroacetic Acid and hydroxy-Dehydroacetic Acid markedly increased with increasing doses, while that of X and Y decreased correspondingly. The investigators found that subcutaneous injection of Dehydroacetic Acid resulted in a pattern of metabolite excretion similar to that following oral administration. It also was suggested that Dehydroacetic Acid and hydroxy-Dehydroacetic Acid were excreted in the bile because these two compounds, but not TAL, were isolated from the feces and gut contents of rats after subcutaneous injection. In additional experiments using rats, the bile contained an average of 8 percent of a 6.7 mg/kg intraperitoneally administered dose of ^{14}C-Dehydroacetic Acid. The investigators suggested that Dehydroacetic Acid and hydroxy-Dehydroacetic Acid, by readily combining with amino groups in proteins and other essential compounds, could cause deleterious effects.

In additional studies on dogs, Shideman et al.[67] found no significant effects

of Dehydroacetic Acid on renal blood flow, creatine clearance, or maximal tubular reabsorption of glucose or phosphate. Renal clearance and tubular excretion (both maximums) of *p*-aminohippurate (PAH) were markedly decreased, as were renal clearances of penicillin and phenolsulfonphthalein (PSP).

Antimicrobial Effects

Sodium Dehydroacetate and Dehydroacetic Acid have been extensively tested for their antimicrobial properties. Results of these studies indicate that both ingredients are effective at low concentrations against bacteria and fungi. They are more effective in acidic media and exert a static influence rather than a lethal effect. Dehydroacetic Acid was significantly more effective as an antimicrobial when administered simultaneously with cocyl hydrolyzed peptides.[68] Inactivation of Dehydroacetic Acid may occur in the presence of nonionics, anti-inhibition agents used in preservation stress tests, high microbial counts, and the "presence of organic matter."[13,14,24,69] Both Sodium Dehydroacetate and Dehydroacetic Acid have additive effects when combined with other preservatives.[52-55,70-73] Table 3 summarizes the antimicrobial properties of the acid and salt.

The percentages of antimicrobial activity of Dehydroacetic Acid at various pHs were reported by DeNavarre.[14] These are listed in Table 4 and coincide with the percentage of undissociated acid (Dehydroacetic Acid) at pH 5.0 and pH 6.5 as given by McCarthy.[1] Andersson et al.,[17] who obtained strongly pH-dependent results in their antimicrobial studies, supported the accepted idea that the mechanism for growth inhibition depends on the undissociated acid molecules.

The specific action of Sodium Dehydroacetate on the metabolic activities of five strains of lactic acid bacteria in milk media was studied by Nakae et al.[80] Acid production was inhibited by 0.06 to 0.10 percent concentrations of Sodium Dehydroacetate, most markedly inhibited being *Lactobacillus bulgaricus* at 0.10 percent. Proteolysis with inhibited at concentrations of 0.08 to 0.10 percent; these same concentrations produced almost total inhibition of aroma production.

Dehydroacetic Acid inhibited the growth of a *Lactobacillus casei*-phage J1 system at a concentration four to five times less than the concentration required to inhibit the growth of the host bacteria.[81] In a further study to determine the mechanism of growth inhibition, Dehydroacetic Acid did not appreciably affect the activity of free phage, adsorption of phage onto cells, injection of phage DNA into cells, replication of progeny phage DNA, or the synthesis of phage endolysin and phage-related protein. The authors concluded, after conducting Dehydroacetic Acid-pulse experiments, that Dehydroacetic Acid inhibited the intracellular growth of phage J1 by preventing maturation.[82]

Dehydroacetic Acid was more effective if dissolved in hot water before its use in a pickling solution for hams and bacon. Mold growth was inhibited for 60 to 122 days on hams and 75 to 141 days on bacons. Pastor[48] noted that the residual Dehydroacetic Acid in a product was affected by the cooking process, so that the use of 1.5 percent Dehydroacetic Acid on ham produces a residue within the 0.0065 percent concentration allowed by the US FDA.

Sodium Dehydroacetate and Dehydroacetic Acid were tested against five

TABLE 3. Antimicrobial Effectiveness of Sodium Dehydroacetate and Dehydroacetic Acid (DHA)

		Effective Concentration (percent)		
Microorganism	*Species*	*Sodium Dehydroacetate*	*DHA*	*Reference*
Fungi	*Aspergillus niger*	0.10, 0.048–0.10*	0.04	74 70,71 52–54 22
	A. usamii	0.05	0.05	75
	Botrytis cinerea	0.01	0.01	45
	Brewer's yeast	0.10	0.02	34
	Chaetomium globosum	0.048*	–	52,53
	Cladosporium herbaum	0.048*	–	52,53
	C. resinae	0.05–0.10*	–	54,70,71
	Epicocum sp.	0.05*	–	52
	Mold (unspecified)	0.2–0.3, 0.4*	0.10–1.5 (residue of 0.0065)	41,77,55,34,48
	Monascus anka	0.20	0.10	75
	Mucor racemosus	0.05	0.05	75
	Penicillium citrinum	0.048–0.10*	–	70,71,52–54
	P. digitatum	–	0.02–0.20	69
	P. roquetorti	0.05	0.05	75
	Rhizopus javanicus	0.05	0.05	75
	R. nigricans	–	0.02–0.20	69
	Saccharomyces cerevisiae	0.10	0.02–0.20	74,69
	Trichoderma viridis	0.05–0.10*	–	70,71,54
Bacteria	*Aerobacter aerogenes*	–	0.075, 0.02–0.20	22,69
	Bacillus anthracis	–	1.0	76
	B. subtilis	0.025*	–	54
	Bacterium flexneri	–	<1.0	76
	B. friedlanderi	–	<1.0	
	B. prodigiosum	–	<1.0	
	B. shigae Behring	–	<1.0	
	B. shigae Parker	–	<1.0	
	Corynebacterium diphtheriae	–	0.05	76
	Eberthella typhosa	–	<1.0	76
	Enterococcus sp.	–	<1.0	76
	Escherichia coli	1.0, 0.025*	<1.0	49,54,76
	Lactobacillus sp. (4 species)	0.06–0.10	–	80
	t. casei	0.80	–	81
	t. casei, phage J1	0.20	–	81
	Neisseria sp.	–	<1.0	76
	Pseudomonas aeruginosa	0.025*	–	54
	P. fluorescens	–	0.035	17
	P. pyocyanea	–	<1.0	76
	Salmonella paratyphi	–	<1.0	76
	S. typhimurium	–	<1.0	76
	S. typhosa	–	0.02–0.20	69
	Sarcina lutea	–	<1.0	76

TABLE 3. (Continued)

Microorganism	Species	Effective Concentration (percent)		Reference
		Sodium Dehydroacetate	DHA	
	Staphylococcus albus	–	<1.0	76
	S. aureus	1.0, 0.025*	0.02–0.20, <1.0	49,54,69,76
	S. citreus	–	<1.0	76
	Streptococcus anhemolyticus	–	<1.0	76
	S. hemolyticus	–	<1.0	76
	S. lactis	0.08	–	80
	S. pneumoniae, Type I	–	<1.0	76
	Vibrio cholerae	–	1.0	76
	V. metchnikowii	–	0.1	
	Unspecified species	0.20	–	41

*When used with a synergist.

Note: The literature contains references to the effectiveness of Sodium Dehydroacetate and DHA against other microbials; however, these had no given concentrations.[72,77–79]

TABLE 4. Antimicrobial Activity of DHA as Affected by pH[14]

pH	Antimicrobial Activity (~ percent)
2.0	100
4.0	95
5.0	65
5.5	33
6.0	16
6.5	6–7
7.0	0–1

strains of fungi inoculated in margarine and kept at 25°C for 30 days.[75] With the exception of *Monascus anka*, both ingredients prevented fungal growth at 0.05 percent but produced some off-flavor. Growth of *M. anka* was inhibited at 0.20 percent, with no accompanying off-flavor.

Biochemical Effects

Sodium Dehydroacetate altered protein biosynthesis in cell-free systems of rat liver.[83] The incorporation of ^{14}C-leucine into the total protein by postmitochondrial supernatant was stimulated slightly by 0.11 and 0.21 percent Sodium Dehydroacetate, but 90 percent inhibition occurred at concentrations above 0.63 percent. Similarly, ^{14}C-leucine incorporation in microsome and cell sap systems was little affected by 0.11 and 0.32 percent Sodium Dehydroacetate; complete inhibition occurred at 1.1 percent. In a further study, 0.32 to 1.1 percent Sodium Dehydroacetate stimulated attachment of ^{14}C-leucine to tRNA and inhib-

ited incorporation of 15 amino acids from tRNA to the total protein by microsome and cell sap systems at concentrations greater than 0.32 percent.[84]

Hisatsune[85,86] studied the mode of action of Sodium Dehydroacetate. In aqueous solution, Sodium Dehydroacetate and flavin interacted to form a complex that inhibited the fluorescence and photodecomposition (in phosphate-buffered solution, 37.8 percent stabilized) of the flavin, shifted the UV absorption spectrum to a longer wavelength region with a concomitant loss in intensity, and markedly increased solubility of riboflavin. The enolic hydroxyl occupying the 4-position of the Dehydroacetic Acid molecule pyrone ring was considered the site of interaction with flavin. Furthermore, using enzyme reaction kinetics, Hisatsune[86] found that Sodium Dehydroacetate inhibited the flavin enzyme D-amino acid oxidase in two ways: by the direct combination of Sodium Dehydroacetate and coenzyme flavin adenine dinucleotide (FAD) and a competitive reaction between the same in a saturated substrate system.

In a study on the effect of metabolic inhibitors on the incorporation of ^{14}C-acetate into aflatoxins by *Aspergillus parasiticus*, Dehydroacetic Acid decreased the specific activities of aflatoxins at all concentrations tested (0.00017 to 0.17 percent). In a select case (0.17 percent) where Dehydroacetic Acid was added to the growth medium, toxin production was drastically reduced, although actual growth of the organism was hardly affected.[87]

Dehydroacetic Acid (0.05 percent) inhibited the activity of lipase at pH 8.0 but not to an extent that would sufficiently explain the lower lipolytic activity of *Pseudomonas fluorescens* found in a study on preservative effects.[17] The authors suggested that Dehydroacetic Acid may inhibit lipase production or the release of lipase by *Ps. fluorescens* and also that the inhibition of lipase activity seen was the result of competition between Dehydroacetic Acid and the lipase at the water–lipid interface.

The effects of Dehydroacetic Acid on the succinoxidase and other enzyme systems were studied by Seevers et al.[62] Succinate oxidation by rat brain mince was inhibited by Dehydroacetic Acid. The degree of inhibition and the log of the Dehydroacetic Acid molar concentration exhibited a linear relationship. There was no lag period, no increase in inhibition with time, and no effect on the cytochrome–cytochrome oxidase fraction of the system. Inhibition was reversed by washing although not by sulfhydryl compounds, and no relationship existed between degree of inhibition and substrate concentration (indicating a noncompetitive basis). To test other possible mechanisms of inhibition of succinic dehydrogenase, Dehydroacetic Acid was added to a urease test system (sensitive to sulfhydryl reactive groups). Urease was not inhibited, and carbon dioxide production actually increased. The chance that Dehydroacetic Acid might be interfering with the action of calcium and aluminum ions, essential for maximum activity of succinic dehydrogenase, was tested by addition of 10 times the normal concentration of these ions. The inhibitory action of Dehydroacetic Acid was not altered.

Seevers et al.[62] also investigated the effects of Dehydroacetic Acid on cholinesterase in vitro and in vivo. Dehydroacetic Acid up to 0.02 M failed to inhibit dog serum cholinesterase. A dose of 600 mg/kg body weight administered intravenously in the dog produced no significant changes in serum cholinesterase activity.

Cellular Effects

The toxic effects of Sodium Dehydroacetate on cultured human amniotic FL-cells were studied.[49] Treatment with 0.05 percent Sodium Dehydroacetate had no discernible effect on the growth of FL-cells. However, 1 percent treatment for 5 minutes depressed growth approximately 95 percent in comparison to controls. Degenerative changes, including swelling of mitochondria and fragmentations of nuclear envelope and plasma membrane, were noted.

Male mice were injected intraperitoneally with 120, 200, or 300 mg/kg Sodium Dehydroacetate in 0.9 percent saline for 3 or 7 consecutive days.[88] Controls received an identical volume of 0.9 percent saline. Twenty-four hours after the last injection, the animals were killed, and sections of liver were prepared. The number and size of lipid droplets increased with increasing doses of Sodium Dehydroacetate. Rough or smooth endoplasmic reticulum often encircled the lipid droplets, whereas, in controls, only a few droplets were partially surrounded. There appeared to be no proliferation of the smooth endoplasmic reticulum and no effects on glycogen and glucose 6-phosphatase activity. Takabatake et al.,[65] on the other hand, by electron microscopy found that the hepatic cells from female rats subcutaneously injected with 200 mg/kg Dehydroacetic Acid daily for 7 or 10 days had a marked proliferation of smooth endoplasmic reticulum. Dehydroacetic Acid also increased the glycogen content of the liver but had no effect on the fat content.

Tissue Effects

Sodium Dehydroacetate was dissolved in propylene glycol and added to a medium containing protoscoleces of *Echinococcus granulosus*.[89] Effective concentrations of 1, 10, and 100 μg Sodium Dehydroacetate were tested in triplicate and observed for 10 days. Percent survival (with respect to controls) was used to compute regression curves and their respective quadratic equations. From these, times of exposure necessary to reach 50 and 90 percent mortality rates were estimated. Sodium Dehydroacetate had low scolicidal activity.

The effect of Sodium Dehydroacetate on the spontaneous contractile activity of rabbit intestine was studied by Weeks et al.[63] In a substrate-free medium containing mounted intestinal strips, the normal stimulatory action of sodium acetate and sodium pyruvate was blocked by 10 mM but not by 1 mM Sodium Dehydroacetate. Subsequent addition of glucose increased contractile activity, but only to about one-half (amplitude height) that of controls. In a series of tests with 10 mM Sodium Dehydroacetate, glucose added before sodium acetate exerted its normal stimulatory effect; sodium acetate was ineffective. The inhibitory action of Sodium Dehydroacetate is much weaker in the presence of substrate, although it is more pronounced in acetate substrate than in glucose. The authors suggest that the substrate inhibition results from blockage of succinic acid dehydrogenase by Sodium Dehydroacetate, thus preventing oxidation.

Physiological Effects

Blood pressure, heart rate, femoral blood flow, and respiration effects of Sodium Dehydroacetate were determined in 7 dogs anesthetized with sodium thio-

pental and sodium barbital.[62] Single, intravenous doses of 100 mg/kg Dehydroacetic Acid administered as Sodium Dehydroacetate produced only slight and transient increases in femoral blood flow; the other parameters were unaffected. This same dose of Sodium Dehydroacetate did not change the responses to standard intravenous test doses of acetylcholine, epinephrine, and histamine. Doses of 300 and 400 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) resulted in small, momentary decreases in blood pressure and heart and respiratory rates. Death resulted after an approximate accumulation of 600 mg/kg Dehydroacetic Acid administered continuously at 300 mg/kg per hour (as Sodium Dehydroacetate). Small decreases, followed by marked increases, in blood pressure and respiratory and heart rates were observed, terminating in vascular collapse and respiratory and cardiac failure.

Plasma carbon dioxide concentrations and pH values were determined after administration of single oral and intravenous doses of Sodium Dehydroacetate to dogs.[62] A single intravenous dose of 300 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) given to each of 4 dogs induced a respiratory alkalosis noted for 48 hours; recovery occurred in 72 hours. Daily oral doses of 200 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) given to each of 4 dogs produced respiratory alkalosis, followed by metabolic acidosis, leading to tremors, convulsions, and death in 2 to 4 days. An inverse relationship between plasma Dehydroacetic Acid and plasma bicarbonate and carbon dioxide tension was noted. No significant changes in plasma pH were observed until after the onset of convulsions. Similarly, daily oral doses of 60 mg/kg Dehydroacetic Acid (just above maximum tolerated dose [MTD]) given to each of 4 dogs resulted in the development of metabolic acidosis, which had the same inverse relationship, not only to plasma Dehydroacetic Acid but also of the Dehydroacetic Acid dose, with plasma bicarbonate and carbon dioxide tension. Plasma pH values remained unchanged.

ANIMAL TOXICOLOGY

Acute Toxicity

Toxic doses of Sodium Dehydroacetate and Dehydroacetic Acid administered orally or intravenously affect primarily the central nervous system in dogs.[62] Salivation, retching, and vomiting proceed to ataxia, weakness and stupor, muscle twitching, and convulsions, ending in respiratory failure. Depending on size and route of administration, death may occur anywhere from 24 to 72 hours after receiving a single dose. Seevers et al.[62] noted that these effects compare very similarly to results of extreme metabolic acidosis.

Oral

The acute oral LD_{50} of the compound in the dog has not been determined. Seevers et al.[62] reported that a dose of 400 mg/kg Dehydroacetic Acid administered as Sodium Dehydroacetate consistently produced death up to 72 hours later. Onset of ataxia, vomiting, and other symptoms may not occur until several hours after administration. A single oral dose of 250 mg/kg to the dog produced only temporary scratching. One animal had no ill effects for 96 hours after a dose of 500 mg/kg Dehydroacetic Acid, but death occurred 10 hours after administra-

tion of a second comparable dose. A single dose of 1 g/kg to a dog produced the first signs of ataxia in 4 hours; death followed at 29 hours. Four dogs receiving 200 mg/kg per day died in 2 to 4 days; however, 4 dogs in an excretion study received 200 mg/kg per day for 3 successive days and were observed for 5 additional days with no report of toxic effects or death.

Single doses of Dehydroacetic Acid in a 10 percent olive oil solution were given to 130 rats at seven dosages ranging from 0.70 to 2.00 g/kg. One hundred rats received single doses of Sodium Dehydroacetate in a 10 percent olive oil solution at five dosages ranging from 0.40 to 0.81 g/kg (acid weight). All doses were administered by stomach tube, usually at volumes of 2 ml or less, but never exceeding 6 ml. All surviving rats were observed for at least 2 weeks. The LD_{50}s for Dehydroacetic Acid and Sodium Dehydroacetate (acid equivalent) were determined to be 1.00 g/kg and 0.57 g/kg, respectively.[5] These values were also reported by Schmidt[90] and Kobayashi et al.[91] Similarly, LD_{50}s of 0.50 and 0.58 g/kg for Sodium Dehydroacetate were found in the literature.[90,92] These LD_{50}s fall into the "slightly toxic" category, according to Hodge and Sterner.[93]

The acute oral LD_{50} for the mouse was reported as 1.05 g/kg Sodium Dehydroacetate.[91]

In two different studies, a paste mask and a rouge, each containing 0.2 percent Sodium Dehydroacetate, were administered by stomach tube to 5 female rats at doses of 15 and 5 g/kg body weight, respectively.[94,95] No deaths resulted during the following 7 days; all animals appeared normal, and no gross changes were observed at necropsy. The acute oral LD_{50}s for the paste mask and rouge were greater than 15 and 5 g/kg, respectively. Another rouge containing 0.1 percent Sodium Dehydroacetate, tested according to 16 CFR 1500.3, gave an LD_{50} greater than 5 g/kg body weight.[96]

Dermal

A dermal toxicity test of a rouge containing 0.1 percent Sodium Dehydroacetate was conducted on rabbits according to 16 CFR 1500.40.[96] The LD_{50} for the rouge was determined to be greater than 2 g/kg body weight, and it was concluded to be nontoxic.

Intravenous

Single intravenous doses of 160 mg/kg Dehydroacetic Acid as Sodium Dehydroacetate to dogs have produced no ill effects.[62,67] No death occurred after a single dose of 240 mg/kg Dehydroacetic Acid administered to a dog as Sodium Dehydroacetate.[10] Seevers et al.[62] reported that a single dose of 300 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) to 4 dogs produced respiratory alkalosis within 48 hours; however, the dogs recovered in 72 hours. Death was consistently produced by a single dose of 400 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) to dogs. Ataxia and vomiting occurred within minutes of the intravenous injection and were followed by the signs of Dehydroacetic Acid toxicity described previously. Death occurred as late as 72 hours after administration. The rapid death was associated with the sudden upset in acid-base equilibrium caused by the injection of excess base.[62] Two dogs receiving single doses of 320 and 480 mg/kg Dehydroacetic Acid (as Sodium Dehydroacetate) died in 3 and 2 days, respectively.[10]

Continuous intravenous infusion at a dose of 300 mg/kg per hour in anesthetized dogs produced death after 600 mg/kg body weight was given. Slight decreases followed by marked increases in respiratory rate, blood pressure, and heart rate were observed. Vascular collapse, respiratory failure, and cardiac arrest ensued.[62]

Intraperitoneal

In a study designed to determine the effect of hepatic damage on the toxicity of Dehydroacetic Acid in mice, two groups of mice (60 and 57) were administered intraperitoneally six doses of Dehydroacetic Acid ranging from 700 to 1600 mg/kg body weight. One group of the mice received an oral hepatotoxic dose of 0.02 ml/kg of 20 percent carbon tetrachloride (CCl_4) in peanut oil 24 hours before administration of Dehydroacetic Acid. The LD_{50}s of the normal and CCl_4 treated mice were determined to be 1186 and 922 mg/kg, respectively.[67]

The acute lethal dose for mice (20 g body weight) given Dehydroacetic Acid in saline by intraperitoneal injection was 40 mg.[76]

Primary Skin Irritation

In a skin irritation test conducted by a method described by Adams et al.,[97] 10 percent solutions of Dehydroacetic Acid in butylcarbitol acetate and Sodium Dehydroacetate in water were applied to the inner ear surfaces and the shaved abdomens of an undisclosed number of rabbits.[5] The ears were left uncovered, and the applications on the abdomen were made with a cotton pad and covered by a large filter cloth taped in place. Applications were made once daily, 5 days per week for 4 weeks. Dehydroacetic Acid and Sodium Dehydroacetate in their solid forms were applied similarly to the abdomen only. No irritation was observed, and the investigators concluded that neither compound was a primary skin irritant.

Skin irritation was observed in rabbits in a study conducted using the Draize cuff technique.[10] The mixture of either Dehydroacetic Acid or Sodium Dehydroacetate with the vehicle was applied over a shaved area encircling the trunk and representing approximately 19 percent of the total body surface. This area was then covered with plastic tubes fastened with tape at each end. Aqueous solutions of test mixtures were applied by fine needles into a gauze layer covered with rubber dental dam and encompassing a similar shaved area. Sodium Dehydroacetate and Dehydroacetic Acid in a washable base and Dehydroacetic Acid in white petrolatum were applied at concentrations of 0.1, 1.0, and 10.0 percent (acid weight). Sodium Dehydroacetate in distilled water was applied only at the 10 percent (acid weight) concentration. Doses consisted of 0.01, 0.1, and 1.0 to 2.0 g/kg for each concentration, respectively. Two rabbits were used for each dose and concentration, except for the dose of 2.0 g/kg of 10 percent Dehydroacetic Acid in washable base, which was applied to only 1 rabbit. All exposures were for 24-hour periods. Some maceration of skin was recorded for the 2 rabbits receiving a 1.0 g/kg dose of 10 percent Sodium Dehydroacetate (acid weight) in distilled water. No other effects were reported.

A primary skin irritation test was conducted with 100 percent Sodium Dehydroacetate using a modified Draize technique.[98] A 0.1 ml dose was applied to a filter disc placed on the shaved skin of each of 9 albino rabbits, occluded for 24 hours, and scored at 2 and 48 hours. Sodium Dehydroacetate had an average irri-

tation index of 0.25 (max = 4.0) and was only minimally irritating and comparable to the control standard (average irritation index = 0.54).

A rouge and a paste mask, each containing 0.2 percent Sodium Dehydroacetate, were analyzed in modified Draize primary irritation tests.[99,100] A 0.1 ml dose of each product was applied to a filter disc placed on the shaved skin of each of 9 albino rabbits, occluded for 24 hours, and scored at 2 and 24 hours. The highest score was used to calculate the average irritation index. The rouge had an index of 0 and was nonirritating. Animals receiving applications of the paste mask had individual scores of 1 (1 animal), 2 (7 animals), and 3 (1 animal), resulting in an index of 2.00. The investigators concluded that the product was mildly to moderately irritating.

A rouge containing 0.1 percent Sodium Dehydroacetate was also evaluated in a primary irritation test.[96] Patches were applied to the abraded and intact shaved skin of a minimum of 6 albino rabbits, occluded for 24 hours, and scored upon removal and 48 hours later. The rouge gave a Primary Irritation Index (PII) of 0.25 (max = 8) and was nonirritating.

Ocular Irritation

Sodium Dehydroacetate (100 percent) was analyzed in a modified Draize eye irritation test.[101] A 0.1 ml dose was sprayed for 48 seconds from a distance of 6 inches into the conjunctival sac of one eye of each of 6 albino rabbits. Eyes were graded and scored according to Draize at 1, 2, 3, 4, and 7 days or until negative. Sodium Dehydroacetate had average irritation scores of 5, 1, 2, and 0 (max = 110) on Days 1, 2, 3, and 4, respectively. This was considered minimally irritating according to the Draize classification and was comparable to the control standard.

A rouge and a paste mask, each containing 0.2 percent Sodium Dehydroacetate, were evaluated in modified Draize eye irritation tests.[102,103] A 0.1 ml dose of each product was instilled into the conjunctival sac of one eye of each of 6 albino rabbits; the other eye served as the control. Eyes were graded and scored according to Draize at 1, 2, 3, 4, and 7 days or until negative. The rouge had average irritation scores of 1, 1, and 0 (max = 110) on Days 1, 2, and 3, respectively. The paste mask had scores of 1 and 0 on Days 1 and 2, respectively. Both of these products were practically nonirritating by the Draize classification of eye irritation.

A rouge containing 0.1 percent Sodium Dehydroacetate was evaluated for irritation in the eyes of 6 rabbits according to 16 CFR 1500.42.[96] The rouge was placed in the conjunctival sac of one eye of each rabbit; the other eye served as the control. Eyes were examined at 24, 48, and 72 hours. No positive reactions were observed for conjunctival redness and chemosis, keratitis, and iritis; the rouge was nonirritating.

Subchronic Toxicity

Oral

Four groups of 5 male rats each were repeatedly administered Dehydroacetic Acid in olive oil by stomach tube in doses of 0.01, 0.03, 0.10, and 0.30 g/kg.[5] Two control groups of 5 and 6 rats each received only olive oil by stomach tube.

Rats receiving Dehydroacetic Acid at the three lower doses were administered 24 doses over 34 days and appeared in excellent health. These animals were then killed and examined. The investigators concluded that, judging by growth and appearance, body and organ weights, blood urea values, and histopathological results, these rats had no toxic effects at doses of 0.10 g/kg Dehydroacetic Acid or less administered orally 24 times in 34 days. However, those rats receiving 0.30 g/kg had a rapid weight loss, with 2 deaths occurring at 7 and 11 days, 5 and 7 doses, respectively. The remaining animals were killed after the second death at 11 days. A 20 to 30 percent reduction in body weight, emaciation, contracted stomach containing some blood, congested mucosa, and slight hemorrhaging in some areas were observed. No pathological changes were noted except for a slightly swollen liver. Blood urea nitrogen values averaged 18.0 mg/100 ml. Difficulty in separating Dehydroacetic Acid and inanition effects was noted. A second group of 5 male rats was administered doses of 0.30 g/kg Dehydroacetic Acid at intervals such that severe weight loss did not occur. One rat died after 7 doses and the other 4 sustained 11 doses over 22 days. These rats were then killed, and the changes were similar to those observed in the first group, although they were less severe.

Nine dogs received daily oral doses of 80 mg/kg Dehydroacetic Acid administered by stomach tube as Sodium Dehydroacetate in water.[62] Two of the nine dogs were forcefed daily (by stomach tube) for 2 weeks before the start of the study, and this practice continued during administration of Dehydroacetic Acid. Forcefeeding markedly reduced the toxicity of Dehydroacetic Acid. The 2 forcefed dogs appeared in excellent health up to 73 days, at which time they were killed. Only one mild convulsion was observed in 1 dog at Day 46; the other delivered a litter of pups on the forty-ninth day. Those dogs not forcefed developed anorexia, salivation increase, weight loss of 13 to 33 percent, convulsions, and death in 10 to 23 days.

In a study conducted by Miyaki et al.,[104] rats (exact number not given) were fed a diet containing 0.25 percent Dehydroacetic Acid for 60 days. Compared to controls, these rats had approximately a 15 percent reduction in body weight, a marked increase in urine volume (on both a basal diet and a 0.06 percent yellow butter diet, each tested on 4 rats), similar organ weights, and a significant (P = 0.05) increase in hepatic metabolism of 4-(monomethylamino)azobenzene, examined in 6 rats.

Intravenous

Nine dogs received daily intravenous doses of 80 mg/kg Dehydroacetic Acid administered as Sodium Dehydroacetate in water.[62] Four of the nine dogs were forcefed daily by stomach tube for 2 weeks before the start of the study, and this was continued during the administration of Dehydroacetic Acid. Those dogs not forcefed lived 10 to 22 days; the 4 forcefed dogs lived 18 to 41 days before dying from Dehydroacetic Acid toxicity. The investigators concluded that forcefeeding did reduce the toxicity of Dehydroacetic Acid given by intravenous injections, although to a much lesser degree than after oral administration.

Subcutaneous

A dose of 200 mg/kg Dehydroacetic Acid was injected subcutaneously into 12 female rats once daily for 10 days.[65] Eleven rats were kept as controls. Rats

receiving the Dehydroacetic Acid injections gained an average of 3 g over the 10-day period, compared to a 15 g average in the controls. Dehydroacetic Acid treatment also increased the amount of glycogen in the liver; however, it had no effect on total fat. Furthermore, liver weight increased relative to body weight in the treated rats.

Chronic Toxicity

Oral

In a one-year study, monkeys (*Macaca mulatta*) received repeated oral doses by stomach tube of 5 percent Dehydroacetic Acid in olive oil and 10 percent Sodium Dehydroacetate (acid weight) in water.[5] Each compound was administered to 1 monkey at doses of 0.05, 0.10, 0.20, and 0.30 g/kg. Two monkeys received only olive oil and 3 only water as controls. Rapid reduction in appetite and weight loss occurred in the high-dose group. The monkey receiving 0.30 g/kg Dehydroacetic Acid (11 doses in 18 days) had a 23 percent weight loss and was a moribund sacrifice on Day 18. The monkey receiving 0.30 g/kg Sodium Dehydroacetate (19 doses in 24 days) showed a 13 percent weight loss and died after 20 doses at Day 26. Both monkeys suffered from ataxia, vomiting, and convulsions prior to death. Forcefeeding did not lessen the severity of these effects. The Dehydroacetic Acid monkey had moderate degeneration of renal tubular epithelium and a blood urea nitrogen value of 114 mg/100 ml. No pathological changes were observed other than inflammation of portions of the small intestine.

The monkeys administered doses of 0.20 g/kg Dehydroacetic Acid or Sodium Dehydroacetate 5 times per week had lack of appetite and weight loss. If the administration was suspended, the monkeys recovered in 3 days and dosage could be resumed; if continued, ataxia, vomiting, and convulsions ensued. Forcefeeding in the latter case did lessen the toxic effects. By omitting critical doses, the investigators were able to administer 0.20 g/kg Dehydroacetic Acid/Sodium Dehydroacetate three to four times per week for 1 year.[5]

Those monkeys given 0.10 and 0.05 g/kg of either compound received five doses per week for 1 year and had no toxic effects.[5] After several months on test, blood concentrations of nitrogenous compounds in monkeys given 0.20, 0.10, and 0.05 g/kg were similar to controls. No effects related to the compounds were found during intermittent hematological studies.

On examination at the year end, no significant differences were found in the organ weights or urea nitrogen concentrations of monkeys given the 0.20, 0.10 and 0.05 g/kg doses. Total lipid content of the liver of the monkey receiving 0.05 g/kg Dehydroacetic Acid was increased. However, because this was not observed in any other animal, the investigators concluded that it was unrelated to treatment. No lesions were found in the tissues examined.[5]

Four dogs tolerated the oral administration of 50 mg/kg per day of Dehydroacetic Acid 6 days per week for 200 days.[62] No weight loss or other signs of toxicity were observed. The investigators believed that this dose was the approximate maximum tolerated dose. However, with the elimination of anorexia effects by forcefeeding, 80 mg/kg per day (administered by stomach tube) was tolerated.

A 2-year chronic toxicity study was conducted on rats (three groups of 25 each) fed diets containing Dehydroacetic Acid at concentrations of 0.02, 0.05,

and 0.10 percent.[5] A group of 10 female rats fed a basic diet were controls. Records of body weight, general appearance, and estimated food consumption were kept. Periodic hematological studies were conducted. At the end of 2 years, surviving rats were weighed, killed, and examined. Organ weights were taken, urea nitrogen and total lipid content of the liver were determined, and tissue sections of 16 organs were prepared. No appreciable difference between the control group and all three test groups was observed in gross appearance, behavior, growth, mortality, or occurrence of spontaneous diseases. Most of the deaths during the test were attributed to respiratory infections and tumors or infections of the ovaries and uterus. Periodic and terminal hematological values, urea nitrogen concentration, organ weights, incidence of abnormalities of ovary or uterus, and tissues of all the rats were similar to controls. A substantial portion of each group did have some pulmonary congestion or consolidation at necropsy. Body weights were equal to or greater than controls. Total lipid content of the liver was only slightly higher in the 0.10 Dehydroacetic Acid group and was comparable to controls for the other groups. Mild fatty changes were found in the livers of most animals, including controls. These were considered common in older rats and not related to treatment. Hyaline casts were observed in the renal tubules, although not extensively, of 2 or 3 rats of each test group (including those that died). No other pathological changes occurred consistently or appeared associated with treatment.

Ten rats were each fed a maximum total of 1830 mg of Sodium Dehydroacetate over a maximum period of 94 days. No gross abnormalities of the liver were observed, but atrophic degeneration was found at microscopic examination.[105]

Twelve rats were fed a diet containing 0.25 percent Dehydroacetic Acid for 350 days. All animals survived to the end of the study, at which time they were killed. Organs were weighed and fixed, and calculations were made on the total drug intake. Livers were examined microscopically and found free of pathological changes.[104]

Twenty rats received a daily oral dose of 20 mg Dehydroacetic Acid for 15 weeks. Another group of 20 rats was kept as controls. Effects reported include a reduction in growth, decrease in hepatic enzymic activity, hepatic hypertrophy, reduced tubular function, renal atrophy, and increased oxygen:nitrogen ratio.[106]

In a study of synergistic toxicity, male and female Wistar rats were fed Sodium Dehydroacetate and dibutylhydroxy toluene (BHT) for 6 months. Diet concentrations were 0 (control), 0.05, 0.15, and 0.5 percent Sodium Dehydroacetate, 0.1, 0.3, and 1 percent BHT, and combinations of 0.05 percent Sodium Dehydroacetate plus 0.1 percent BHT, 0.5 percent Sodium Dehydroacetate plus 0.1 percent BHT, and 0.05 percent Sodium Dehydroacetate plus 1 percent BHT. Under these experimental conditions, no significant toxic effects were observed in the combination of Sodium Dehydroacetate and BHT.[107]

Teratogenesis

Sodium Dehydroacetate was administered orally to mice at doses of 50, 100, and 200 mg/kg per day on the Days 6 through 15 of gestation.[108] Fetuses were examined on the seventeenth day. High mortality and decreased fetal weight were observed in the high-dose group. Abnormalities found included a four-

teenth rib (in all treated groups), sternebrae deformities, and rib malformation. However, these results were not significant when compared to untreated controls.

MUTAGENESIS, CARCINOGENESIS, AND ANTITUMORIGENESIS

Chromosome aberration tests with Chinese hamster cells in vitro were used to evaluate the mutagenic potential of Sodium Dehydroacetate. Different doses of the compound in saline were applied directly to the cells; chromosome preparations were made 24 and 48 hours later, and aberrations were scored. The maximum tolerated dose was 0.50 mg/ml (2.40×10^{-5}M). Only a 1 percent increase in polyploid production occurred at 48 hours and a 2 percent increase in chromosome aberrations at 24 hours, consisting of chromatid gaps and breaks. The investigators concluded that Sodium Dehydroacetate was nonmutagenic.[109,110]

The potential of Sodium Dehydroacetate to induce chromosomal aberrations and sister chromatid exchanges (SCE) in Chinese hamster cells in vitro was studied. Sodium Dehydroacetate in saline solution was tested at concentrations of 1×10^{-5}, 1×10^{-4}, and 1×10^{-3}M. No significant chromosomal aberrations, SCEs, mitotic inhibition, or dosage effects were noted.[111]

Sodium Dehydroacetate was evaluated for its mutagenic potential in a series of short-term assays using *Salmonella typhimurium* TA100 and TA98 and silkworms for mutations, *B. subtilis* for *rec* assay (without metabolic activation), hamster lung fibroblast cells for chromosomal aberrations and SCEs (without metabolic activation), and rat bone marrow cells for chromosomal aberrations in vivo. Results were negative for all tests; investigators concluded that Sodium Dehydroacetate was nonmutagenic.[112]

Sodium Dehydroacetate in water was evaluated in a modified Ames test and a bacterial repair test. Mutagenic activity was evaluated in *S. typhimurium* strains TA1535, TA100, TA1537, TA1538, and TA98, and *E. coli* WP2 hcr; repair testing (*rec* assay) was conducted with *B. subtilis* H17 and M45. Sodium Dehydroacetate (up to 5 mg/plate) was found nonmutagenic in the presence or absence of polychlorinated biphenyl (PCB)-induced rat liver microsomes (S-9 mix) and gave negative results in the *rec* assay.[113]

Dehydroacetic Acid was assayed for mutagenicity by a simplified paper disc method described by Iyer and Szybalski.[114] Mutagenicity was measured by an increase in the frequency of reversion from streptomycin dependence to independence in *E. coli* strain Sd-4-73. Dehydroacetic Acid produced no evidence of mutagenicity.[115]

A study was conducted in which 6 male rats were given Dehydroacetic Acid in their drinking water for 64 weeks.[116] Dehydroacetic Acid was added to water at a concentration of 10 mg/100 ml and neutralized with sodium bicarbonate to pH 5 in order to bring it into solution. Concentrated stock solutions were stored at 4°C and diluted with tap water to replenish the rats' water bottles. Average weekly intake per rat was 46.3 ml, approximately 6.6 mg Dehydroacetic Acid daily. Animals were examined periodically for the presence of hepatic tumors by laparotomy under ether anaesthesia. An examination for tumors in all organs was made at time of death and on all survivors at 100 weeks; no hepatic or other tumors were found. Hepatic necrosis did occur in a rat alive at 103 weeks, calculated to have ingested approximately 2 g of Dehydroacetic Acid.

Twice weekly subcutaneous injections of 2 mg Dehydroacetic Acid in 0.5 ml arachis oil were made into the right flanks of 6 male rats for 65 weeks.[116] A control group of 6 rats was injected with arachis oil alone. The first local tumor (sarcoma) in the Dehydroacetic Acid group appeared at 37 weeks, all rats being alive at that time. At the end of 85 weeks observation, a total of 5 rats in the Dehydroacetic Acid group had local tumors; some of these were considered malignant due to proliferation and the presence of large multinucleate cells. A large sarcoma-like tumor developed at 81 weeks in 1 control rat, 3 rats surviving at that time. Necropsy did not reveal any other tumors in either the control or Dehydroacetic Acid group. Clayson[117] regarded the induction of localized sarcomas in mice upon repeated subcutaneous injection of test solutions as "notoriously unreliable as an indicator of carcinogenicity." Furthermore, he considered "the results of individual experiments as extremely variable."

Several studies have been conducted on the inhibitory effect of Dehydroacetic Acid on the induction of hepatomas in rats fed the known carcinogen 4-(dimethyl-amino)azobenzene (DAB). The most extensive of these studies involved a set of four groups of rats treated as follows: the first group of 22 rats received 0.06 percent DAB for 126 days, followed by a basal diet for 162 days; the second group of 21 received 0.06 percent DAB for 126 days, followed by 0.25 percent Dehydroacetic Acid in the diet for 162 days; the third group of 16 received 0.06 percent DAB and 0.25 percent Dehydroacetic Acid for 173 days, followed by a basal diet for 77 days; and the fourth group of 12 received only 0.25 percent Dehydroacetic Acid for 350 days. Except for 7 rats from the second group, all survived until the end of the study, at which time they were killed. Liver, spleen, adrenals, kidneys, and testes were weighed and fixed, and livers were examined microscopically. Hepatomas and cholangiocarcinomas were not found in the group receiving DAB and Dehydroacetic Acid simultaneously; however, these occurred in 27.3 percent and 42.9 percent of the rats receiving DAB only and DAB followed by Dehydroacetic Acid, respectively. Of those receiving DAB and Dehydroacetic Acid simultaneously, 62.5 percent did have atypical hepatic cellular growth and one adenoma, although these lesions developed slowly in relation to the development rate in the group receiving only DAB. The incidence of other histological changes, including fibrosis, cholangiocysts, and hyperplasia of bile ducts, was markedly reduced in the rats given both substances at the same time compared to those administered DAB alone or Dehydroacetic Acid after DAB. The livers of the rats receiving Dehydroacetic Acid only had no lesions. Dehydroacetic Acid, if administered simultaneously, delayed induction of hepatomas and cholangiocarcinomas in rats fed DAB. An additional experiment was conducted, confirming this, in which three groups of rats were fed 0.06 percent DAB, the second and third groups receiving an additional 0.1 and 0.25 percent Dehydroacetic Acid in the diet, respectively. These groups were fed until the appearance of hepatomas and cholangiocarcinomas as determined by histopathology on 2 rats from each group killed at 1-month intervals. Tumors were found at 5, 7, and 8 months in the groups fed DAB, DAB with 0.1 percent Dehydroacetic Acid, and DAB with 0.25 percent Dehydroacetic Acid, respectively.[104,105,118,119]

Studies of hepatic microsomal enzymes were conducted throughout the study to determine the mechanism of Dehydroacetic Acid inhibition. Dehydroacetic Acid alone increased the microsomal enzymic activity for metabolizing azo-dyes; however, this increase was negated by simultaneous administration of

DAB. Previously, Dehydroacetic Acid was found not to affect the distribution of DAB in tissues and organs. Based on these two facts, the investigators concluded that Dehydroacetic Acid did not inhibit hepatoma formation by accelerating the carcinogen's metabolism. Further results indicated that the formation of the protein-bound dye was inhibited by Dehydroacetic Acid in the microsomal and supernatant fractions of liver homogenate during the first stages of carcinogenesis. Additionally, Dehydroacetic Acid delayed the decrease in the liver's ability to form this complex, a decrease seen as carcinogenesis proceeds. The inhibition by Dehydroacetic Acid of the formation of the protein-bound dye paralleled its inhibition of hepatoma induction.[104]

CLINICAL ASSESSMENT OF SAFETY

Irritation, Sensitization, Photosensitization, and Phototoxicity

Sodium Dehydroacetate and Dehydroacetic Acid were tested for irritation and sensitization on a randomly selected panel of 100 men and 100 women. Sodium Dehydroacetate was tested as a 60 percent paste of the salt in water; Dehydroacetic Acid was tested as a 65 percent paste of the acid in sesame oil. For both compounds, occlusive patches permeated by the pastes were applied directly to the intact skin of the back. Patches were removed after 5 days and reactions scored. A second application was made in the same manner as the first 3 weeks after patch removal. These patches were removed after 48 hours, and reactions were scored at the time of removal and 3 and 8 days later. No reactions were observed in any of the 200 panelists after the first or second applications. The investigators concluded that Sodium Dehydroacetate and Dehydroacetic Acid were neither primary skin irritants nor sensitizers[5] (Table 5).

Sodium Dehydroacetate as a 0.4 percent aqueous solution was tested for skin irritation on 19 subjects.[120] Patches with a 0.1 ml dose were applied to the volar surface of each forearm or upper arm and occluded for 24 hours. Sixteen subjects had no response, two had "barely perceptible" erythema, and one had marked erythema but was considered atypical, in that similar reactions had been obtained with other materials tested. Sodium Dehydroacetate produced minimal erythema (Table 5).

A 0.1 percent aqueous solution of Sodium Dehydroacetate was tested for photosensitization and phototoxicity in a repeated insult patch test (RIPT). A 24-hour occlusive patch containing 0.1 ml of the test solution was applied to the same site on the back of each subject on Monday, Wednesday, and Friday for 3 consecutive weeks. Challenge patches were applied 2 weeks later to both the original site, and an adjacent site. Identical patches were also applied to the opposite side of the back to serve as nonradiated controls. On the 23 subjects who completed the study, 9 received both UVA and UVB exposure, and 14 received UVA only. The UVA exposure (50 percent at 345 nm) was delivered by four F40 BL fluorescent tubes at 10 cm with a dose of approximately 4.4 $\mu W/cm^2$. Sites were irradiated for 5 minutes after patch removal on each Tuesday and Saturday during the induction phase. Additionally, a 150 W Xenon Arc Solar Simulator was used to deliver twice the individual minimal erythemal dose (MED) of UVB to those designated subjects after each UVA exposure. At challenge, all sites were exposed only to UVA radiation. Sites were scored on a scale of 0 to 4 just prior to each new patch application and 72 hours after the last induction patch.

TABLE 5. Clinical Irritation, Sensitization, and Photosensitization of Sodium Dehydroacetate and Dehydroacetic Acid

Compound Tested	*Type of Test*	*No. of Humans*	*Results/Comments*	*Reference*
Sodium Dehydroacetate–60 percent aqueous paste	Occlusive patch for 5 days; 48-hour occlusive challenge patch 3 weeks later	200	No reactions; nonirritating and nonsensitizing	5
Dehydroacetic Acid–65 percent paste in sesame oil	Occlusive patch for 5 days; 48-hour occlusive challenge patch 3 weeks later	200	No reactions; nonirritating and nonsensitizing	5
Sodium Dehydroacetate–0.4 percent aqueous solution	Occlusive patch for 24 hours	19	No reactions in 16, 2 subjects with "barely perceptible" erythema, 1 subject with marked erythema considered to be atypical; concluded to elicit minimal erythema	120
Sodium Dehydroacetate–0.1 percent aqueous solution	RIPT* with UV exposure	14-UVA only 9-UVA and UVB (2 MED)	Erythematous reactions in all 9 receiving UVA and UVB; one subject initially received an excessive dose of UVB at induction and had a score of 3 (max = 4), but reduced exposure produced no score higher than a 1; no other reaction observed; erythema in the 9 subjects attributed to the 2 MED of UVB; nonphototoxic and nonphotosensitizing	121
Sodium Dehydroacetate–0.2 percent in a rouge	24 hour patch	20	No reactions in 18, 2 subjects with mild erythema; no significant difference in irritancy compared to controls	122
Sodium Dehydroacetate–0.2 percent in a paste mask	24 hour patch	20	No reactions in 19, 1 subject with "barely perceptible" erythema; no significant difference in irritancy compared to controls	123

Sodium Dehydroacetate–0.2 percent in a rouge	RIPT	128	No reactions in 127, 1 subject with minimal and definite erythema 20 minutes and 48 hours after the initial induction patch, respectively (1 and 2 on max scale of 7)–no further reactions seen; causes little or no irritation and no sensitization	124
Sodium Dehydroacetate–0.2 percent in a paste mask	RIPT	112	No reactions in 97, 15 subjects exhibited 22 incidences of minimal erythema and 5 of definite erythema (1 and 2 on max scale of 7) during induction; 3 subjects reacted at challenge–2 with minimal erythema at 1st scoring, none at 2nd scoring, and 1 with definite and minimal erythema at 1st and 2nd scorings, respectively; these were concluded to be due to primary irritation, not sensitization; "essentially nonirritating"	125
Sodium Dehydroacetate–0.1 percent in a rouge	4-week controlled use study	60	No reactions in 58, 2 subjects with weak, nonvesicular reactions at 4 weeks, 1 found to be sensitized to paraben in the rouge; nonirriating	126
Sodium Dehydroacetate–0.1 percent in a rouge	Schwartz-Peck prophetic patch test with UV exposure	102	No reactions in 101, 1 subject with weak reaction under occlusive conditions at induction, none at challenge; nonirritating, nonsensitizing, and nonphotosensitizing	127
Sodium Dehydroacetate–0.1 percent in a rouge	RIPT with UV exposure	53	Total of 8 weak reactions under occlusive conditions at induction, none at challenge; nonirritating, nonsensitizing, and nonphotosensitizing	127

*RIPT, Repeated Insult Patch Test.

Challenge sites were scored at 24, 48, 72, and 96 hours. The combination of UVA and UVB exposure elicited erythematous reactions in all 9 subjects. One subject had an initial "3" reaction at induction; however, it was determined that this subject had received an excessive dose of UVB. With reduced exposure, no score higher than a "1" was elicited. No other reactions were observed in any of the subjects during induction or challenge. It was concluded that the erythema noted in the 9 subjects was due solely to the 2 MED of UVB and that Sodium Dehydroacetate was neither a phototoxic nor photosensitizing agent in humans[121] (Table 5).

A rouge and a paste mask, each containing 0.2 percent Sodium Dehydroacetate, were individually tested for skin irritation on a panel of 20 subjects.[122,123] Patches containing a 0.1 ml dose of each product were applied to the volar surface of the forearm or upper arm and occluded for 24 hours. An unpigmented control and a currently marketed product were used as controls in the rouge and paste mask tests, respectively. Two subjects receiving the rouge application had mild erythema, whereas 18 gave a negative response. One subject receiving the paste mask application had "barely perceptible" erythema, whereas 19 gave a negative response. In both studies there was no significant difference in irritancy potential between the products and the controls (Table 5).

These same rouge and paste mask products (0.2 percent Sodium Dehydroacetate) were tested for irritation and/or sensitization by modified Draize RIPT.[124,125] The rouge was applied to patches and placed on the backs of 128 panelists for 48 hours. Reactions to this initial patch were scored at 20 minutes and 48 and 120 hours after removal. The second patches were applied to previously unpatched sites for 24 hours, and subsequent 24-hour applications were made on the same sites on Mondays, Wednesdays, and Fridays for the next 3 weeks (9 total applications). Challenge patches were applied on Monday of Week 6 to previously unpatched sites for 48 hours and scored 20 minutes and 48 and 120 hours after removal. Minimal erythema at 20 minutes and definite erythema at 48 hours occurred in 1 subject (scores of 1 and 2 on a maximum scale of 7) after the initial induction patch, but no further irritant reactions were observed during the subsequent 3 weeks or at challenge. The rouge was considered to cause little or no irritation and no sensitization under these conditions. The patches dosed with the paste mask were applied on the arm of each of 112 panelists for 24 hours on Monday, Wednesday, and Friday for 3 weeks. Challenge applications were made 2 weeks later on previously unpatched sites for 24 hours; reactions were scored 24 and 72 hours after removal. Twenty-two incidences of minimal erythema and five of definite erythema (scores of 1 and 2 on a maximum scale of 7) were noted throughout the induction period in a total of 15 panelists. Three subjects reacted at challenge: two exhibited minimal erythema at the first scoring with no visible reaction at the second, and one subject exhibited definite and minimal erythema at the first and second scorings, respectively. The investigators considered the latter reaction to be due to primary irritation, not sensitization. The paste mask was concluded to be "essentially nonirritating" throughout the study (Table 5).

A rouge containing 0.1 percent Sodium Dehydroacetate was applied to the facial area or cheek of 60 panelists for 4 weeks in a controlled use study.[126] Two subjects had a weak, nonvesicular reaction at 4 weeks; however, one was sensitized to the paraben in the rouge through further patch testing. The rouge was

nonirritating. This same product was tested in a Schwartz-Peck prophetic patch test and a Shelanski RIPT, each with additional UV exposure procedures.[127] One panelist of 102 in the Schwartz-Peck test had a weak reaction at induction under occlusive conditions; no reactions were observed at challenge. Similarly, only 8 weak reactions were noted under occlusive conditions of the induction patch series administered to 53 panelists in the RIPT; no reactions were noted at challenge. Under the conditions of these studies, the rouge was nonirritating, nonsensitizing, and nonphotosensitizing (Table 5).

Toxicity

A hospitalized patient received 200 to 400 mg Dehydroacetic Acid orally four times per day for 17 days with no reported toxic effects. Shideman et al.[67] reported that 6 to 13 mg/kg Dehydroacetic Acid ingested daily for 173 days was tolerated by man. Two patients suffering from blastomycosis developed anorexia at 26 and 48 days, and one was nauseous on Day 13 after daily oral administration of 14 to 17 mg/kg Dehydroacetic Acid. However, any relationship of Dehydroacetic Acid treatment to these symptoms was considered dubious because of the patient's disease and in view of the fact that larger doses of Dehydroacetic Acid have been tolerated without ill effects.[67]

Three normal men ingested 500 mg Dehydroacetic Acid daily in 250 mg tablets at breakfast and dinner. Plasma samples were taken everyday at noon. After 115 days, the dose was increased to 750 mg per day by ingestion of an additional tablet at noon. The study was terminated after 38 more days. Plasma concentrations ranged from 10 to 18 mg/100 ml. No symptoms or toxic effects were evident during or after (approximately 20 days) Dehydroacetic Acid administration.[10]

SUMMARY

Sodium Dehydroacetate is the sodium salt of the cyclic ketone Dehydroacetic Acid, both of which occur as tasteless, odorless, white powders. Sodium Dehydroacetate is soluble in water, propylene glycol, and methanol but insoluble in most organic solvents, whereas Dehydroacetic Acid is soluble in acetone, benzene, ether, and methanol. Dehydroacetic Acid is reactive and relatively unstable, although the sodium salt is fairly stable at room temperature. Profoundly affected by pH, the acid is active only in its undissociated state. Therefore, Dehydroacetic Acid has a much greater degree of effectiveness against bacteria and fungi in acidic media, although partial antimicrobial activity is retained under alkaline conditions. Conflicting results have been reported concerning the inactivation of these compounds by nonionic surfactants.

Sodium Dehydroacetate and Dehydroacetic Acid are used as preservatives in over 139 and 260 cosmetic formulations, respectively, at concentrations of 1.0 percent or less. Products containing these compounds may contact all areas of the skin, the hair, ocular mucosa, and nails, with possible incidental exposure of vaginal mucosa. They may be used daily or occasionally over a period of up to several years, possibly resulting in continuous exposure. Other uses include food preservatives (both direct and indirect additives), limited pharmaceutical use, industrial preservatives and antimicrobials, and insecticides.

Absorption occurs readily when Sodium Dehydroacetate and Dehydroacetic Acid are administered orally, intravenously, or dermally. Dehydroacetic Acid has been shown to bind with plasma proteins, particularly serum albumin. Three metabolites of Dehydroacetic Acid have been identified from rabbit urine: triacetic acid lactone (TAL), a hydroxy-Dehydroacetic Acid, and a compound believed to be the salt of TAL 3-carboxylic acid. Elimination of these compounds is slow.

Sodium Dehydroacetate and Dehydroacetic Acid are effective antimicrobials at low concentrations against both bacteria and fungi. Dehydroacetic Acid has an optimal pH range of 2 to 4 and exerts a more antimicrobial static than lethal effect. Inactivation may occur in the presence of nonionics, anti-inhibition agents, high microbial counts, and organic matter.

Sodium Dehydroacetate and Dehydroacetic Acid both stimulate and inhibit enzyme systems. Dehydroacetic Acid is known to produce a stimulating effect on drug-metabolizing enzyme activity, while inhibiting aflatoxin production and lipase activity in microbial systems and the succinoxidase system in rats. Sodium Dehydroacetate has also been reported to inhibit the flavin enzyme D-amino acid oxidase.

Of the physiological parameters studied, single intravenous doses of Sodium Dehydroacetate or Dehydroacetic Acid administered to dogs produced a slight increase in femoral blood flow and a temporary respiratory alkalosis. Daily oral or continuous IV administration of these compounds (above the MTD) results in respiratory alkalosis, metabolic acidosis, marked increases in blood pressure and respiratory and heart rates, followed by respiratory and cardiac failure.

Acute toxicity studies indicate that Sodium Dehydroacetate and Dehydroacetic Acid are slightly toxic when administered orally to rats. Single IV doses up to 300 mg/kg administered to dogs produced no deaths; however, a dose of 400 mg/kg consistently resulted in death. The intraperitoneal LD_{50} of Dehydroacetic Acid in mice is reported to be 1186 mg/kg. Primary skin irritation studies conducted on rabbits showed both compounds to be practically nonirritating. Sodium Dehydroacetate was found to exhibit minimal eye irritation, whereas cosmetic products containing up to 0.2 percent Sodium Dehydroacetate were nonirritating. Subchronic and chronic studies reveal various toxic effects, primarily due to the incurred lack of appetite and weight loss. These effects have been shown to be reduced by forcefeeding in certain studies.

No evidence of mutagenicity was found in numerous studies on Sodium Dehydroacetate and Dehydroacetic Acid. Rats administered Dehydroacetic Acid in their drinking water for 64 weeks developed no evidence of tumors. Multiple, repeated subcutaneous injections of Dehydroacetic Acid induced local sarcomas in rats but did not produce tumors at distant sites. Dehydroacetic Acid has an inhibitory effect on hepatoma induction in rats fed the known carcinogen 4-(dimethylamino)azobenzene (DAB). A teratogenicity study in mice revealed no significant findings when compared to untreated controls.

Sodium Dehydroacetate, Dehydroacetic Acid, and cosmetics containing these ingredients were found practically nonirritating, nonsensitizing, nonphotosensitizing, and nonphototoxic in numerous clinical tests. Daily ingestion of 500 mg Dehydroacetic Acid for 115 days, followed by 750 mg Dehydroacetic Acid daily for 38 more days, produced no symptoms or toxic effects in 3 men, either during or 20 days after administration.

CONCLUSION

On the basis of the available animal and clinical data, the Panel concludes that Sodium Dehydroacetate and Dehydroacetic Acid are safe as cosmetic ingredients in the present practices of use and concentration.

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SODIUM DEHYDROACETATE AND DEHYDROACETIC ACID

A safety assessment of Sodium Dehydroacetate and Dehydroacetic Acid was published in 1985 with the conclusion that these ingredients are safe as cosmetic ingredients in the present practices of use and concentration (Elder 1985). Studies available since that safety assessment was completed, along with updated information regarding uses and use concentrations were considered by the CIR Expert Panel. The Panel determined to not reopen this safety assessment.

Sodium Dehydroacetate was used in 260 products in 1981, based on voluntary reports provided to FDA by industry; use concentrations ranged from less than 0.1% to 1% (Elder 1985). In 2002 there were 325 uses (FDA 2002) and according to an industry survey the current range of use concentrations is 0.00003% to 0.5% (CTFA 2002).

Dehydroacetic Acid was used in 139 products in 1981, based on voluntary reports provided to FDA by industry; use concentrations ranged from less than 0.1% to 1% (Elder 1985). In 2002 there were 88 uses (FDA 2002) and according to an industry survey the current range of use concentrations is 0.007% to 0.7% (CTFA 2002).

Table 22 presents the available use and concentration information. The most recent information now constitutes the present practices of use.

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SODIUM LAURYL SULFOACETATE

A safety assessment on Sodium Lauryl Sulfoacetate was published in 1987 with the conclusion "On the basis of the available data presented in this report, the Expert Panel concludes that Sodium Lauryl Sulfoacetate is safe as a cosmetic ingredient in the present practices of use and concentration" (Elder 1987). Studies available since that safety assessment was completed, along with updated information regarding uses and use concentrations, were considered by the CIR Expert Panel. After reviewing the available data, the Panel determined to not reopen this safety assessment.

Sodium Lauryl Sulfoacetate was used in 93 products in 1981, based on voluntary reports provided to FDA by industry; use concentrations ranged from >0.1% to >50% (Elder 1985). In 2002 there were 68 uses (FDA 2002) and according to an industry survey in 2004 the current range of use concentrations is 0.6% to 21% (CTFA 2004).

Table 23 presents the available use and concentration information. The most recent information now constitutes the present practices of use.

The CIR Expert Panel did note that Stepan Company had submitted robust summaries and test plans on Sodium Lauryl Sulfoacetate as part of EPA's high production volume chemical testing program. This submission argued that the only missing data were reproductive and developmental toxicity data. The company proposed conducting such a study. Though the Panel noted that there are no data in the published literature,

TABLE 22
Historical and current uses and use concentrations for Sodium Dehydroacetate and Dehydroacetic Acid

Product category	1981 uses (Elder 1985)	2002 uses (FDA 2002)	1981 concentrations (Elder 1985) %	2003 concentrations (CTFA 2003) %
		Sodium Dehydroacetate		
Baby care				
Lotions, oils, powders & creams	—	—	—	0.6
Bath				
Soaps and detergents	—	2	—	0.0001
Oils, tablets, and salts	1	—	≤0.1	—
Eye makeup				
Eyebrow Pencil	—	—	—	0.2–0.3
Eyeliner	2	4	≤0.1–1	0.05–0.5
Eye shadow	56	74	≤0.1–1	0.05–0.3
Eye lotion	—	3	—	—
Eye makeup remover	—	1	—	0.05
Mascara	13	16	≤0.1–1	0.001–0.4
Other eye makeup	4	12	>0.1–1	0.0006–0.4
Fragrances				
Powders	1	3	>0.1–1	—
Colognes and toilet waters	—	—	—	0.001–0.5
Noncoloring hair care				
Conditioners	—	—	—	0.2
Shampoos	—	2	—	0.2
Tonics, dressings, etc.	1	1	≤0.1	—
Other noncoloring hair care	—	4	—	—
Hair coloring				
Tints	—	1	—	—
Other hair coloring	—	2	—	—
Makeup				
Blushers	22	15	≤0.1–1	0.1–0.4
Face powders	23	31	≤0.1–1	0.05–0.4
Makeup foundations	8	10	≤0.1–1	0.0001–0.4
Makeup bases	14	6	>0.1–1	0.1
Leg and body paints	—	—	—	0.1
Lipstick	—	1	—	0.3
Rouges	2	—	≤0.1–1	—
Makeup fixatives	—	1	—	—
Other makeup	2	4	>0.1–1	0.0003–0.2
Nail care				
Basecoats and undercoats	—	—	—	0.02
Nail creams and lotions	—	3	—	—
Cuticle Softeners	4	2	>0.1–1	—
Creams and lotions	2	—	≤0.1–1	—
Polish and enamel	—	—	—	0.2
Other nail care	1	—	>0.1–1	0.2
Personal hygiene				
Underarm deodorants	—	2	—	—
Shaving				
Shaving cream	1	4	>0.1–1	—
Other shaving	1	1	>0.1–1	—
Aftershave lotions	1	1	≤0.1	0.0003

TABLE 22
Historical and current uses and use concentrations for Sodium Dehydroacetate and Dehydroacetic Acid *(Continued)*

Product category	1981 uses (Elder 1985)	2002 uses (FDA 2002)	1981 concentrations (Elder 1985) (%)	2003 concentrations (CTFA 2003) (%)
Skin care				
Skin-cleansing preparations	23	13	≤0.1–1	0.0003–0.3
Face and neck skin care	24*	4	≤0.1–1*	0.008–0.2
Body and hand skin care		20		0.00003–0.5
Moisturizers	27	39	≤0.1–1	0.001–0.3
Night skin care	7	5	≤0.1	0.003–0.2
Paste masks/mud packs	4	6	≤0.1–1	0.03–0.2
Fresheners	2	2	>0.1–1	—
Other skin care	—	25	—	0.00003–0.1
Skin lighteners**	2	—**	≤0.1–1	—**
Wrinkle smoothers**	1	—**	>0.1–1	—**
Suntan				
Suntan gels, creams, and liquids	5	1	>0.1–1	0.2
Indoor tanning preparations	3	2	≤0.1–1	0.4
Other suntan preparations	3	2	>0.1–1	0.1
Total uses/ranges for Sodium Dehydroacetate	**260**	**325**	**≤0.1–1**	**0.00003–0.6**
	Dehydroacetic Acid			
Bath				
Soaps and detergents	—	—	—	0.03
Oils, tablets and salts	1	—	≤0.1	—
Bubble baths	2	1	≤0.1	—
Eye makeup				
Eyeliner	1	—	>0.1–1	0.1
Eye shadow	11	4	≤0.1–1	0.3
Eye lotion	—	—	—	0.2
Eye makeup remover	8	5	≤0.1–1	0.1
Mascara	1	—	>0.1–1	0.2
Other eye makeup	9	—	≤0.1–1	—
Fragrances				
Colognes and toilet waters	4	—	≤0.1	—
Perfumes	4	—	≤0.1	—
Noncoloring hair care				
Shampoos	2	—	≤0.1	0.02–0.03
Tonics, dressings, etc.	2	1	≤0.1–1	—
Makeup				
Blushers	5	1	≤0.1–1	0.05–0.2
Face powders	6	3	≤0.1–1	0.7
Makeup foundations	13	3	≤0.1–1	0.1
Makeup bases	1	—	≤0.1	—
Rouges	1	1	>0.1–1	—
Lipstick	1	—	≤0.1	—
Other makeup	1	—	≤0.1	0.07
Nail care				
Cuticle softeners	—	1	—	—
Polish and enamel	—	1	—	—
Personal hygiene				
Other personal hygiene	—	—	—	0.03

(Continued on next page)

TABLE 22
Historical and current uses and use concentrations for Sodium Dehydroacetate and Dehydroacetic Acid *(Continued)*

Product category	1981 uses (Elder 1985)	2002 uses (FDA 2002)	1981 concentrations (Elder 1985) %	2003 concentrations (CTFA 2003) %
Skin care				
Cleansing creams, lotions, etc.	15	8	≤0.1–1	0.007–0.02
Face and neck skin care	16*	11	≤0.1–1*	0.01–0.08
Body and hand skin care		9		0.03–0.05
Moisturizers	10	10	≤0.1–1	—
Night skin care	5	2	≤0.1–1	0.03
Paste masks/mud packs	3	6	≤0.1–1	—
Skin fresheners	2	—	≤0.1	—
Other skin care	9	16	≤0.1–1	0.03
Wrinkle smoothers**	2	—**	≤0.1	—**
Suntan				
Suntan gels, creams, and liquids	3	—	>0.1–1	0.2
Indoor tanning preparation	—	5	—	—
Other suntan preparations	1	—	>0.1–1	—
Total Uses/Ranges for Dehydroacetic Acid Totals	**139**	**88**	**≤0.1–1**	**0.007–0.7**

*These categories were combined in 1981 but are now separate.
**No longer considered as cosmetic product categories.

which suggest that the reproductive and developmental toxicity potential of Sodium Lauryl Sulfoacetate is an issue, it was agreed that the results of the proposed reproductive and developmental toxicity study would be considered when available.

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[24]Available for review: Director, Cosmetic Ingredient Review, 1101 17th Street, NW, Suite 412, Washington, DC 20036-4702, USA.

SODIUM SESQUICARBONATE, SODIUM BICARBONATE, AND SODIUM CARBONATE

A safety assessment of Sodium Sesquicarbonate, Sodium Bicarbonate, and Sodium Carbonate was published in 1987 with the conclusion that these ingredients are safe as presently used in cosmetic products (Elder 1987). Studies available since that safety assessment was completed, along with updated information regarding uses and use concentrations, were considered by the CIR Expert Panel. After reviewing the available data, the Panel determined to not reopen this safety assessment.

Sodium Sesquicarbonate was used in 111 products in 1981, based on voluntary reports provided to FDA by industry; use concentrations ranged from >1% to 50% (Elder 1985). In 2002 there were 24 uses (FDA 2002) and according to an industry survey in 2004 the current range of use concentrations is 2.0% to 90% (CTFA 2004).

Sodium Bicarbonate was used in 45 products in 1981, based on voluntary reports provided to FDA by industry; use concentrations ranged from less than 0.1% to 50% (Elder 1985). In 2002 there were 70 uses (FDA 2002) and according to an industry survey in 2004 the current range of use concentrations is 0.006% to 95% (CTFA 2004).

Sodium Carbonate was used in 25 products in 1981, based on voluntary reports provided to FDA by industry; use concentrations ranged from less than 0.1% to 25% (Elder 1985). In 2002 there were 21 uses (FDA 2002) and according to an industry survey in 2004 the current range of use concentrations is 0.000002% to 51% (CTFA 2004).

Table 24 presents the available use and concentration information. The most recent information now constitutes the present practices of use.