

2. In § 91.14, paragraphs (a)(11) and (a)(12) are revised to read as follows:

**§ 91.14 Ports of embarkation and export inspection facilities.**

(a) \* \* \*

(11) *New Jersey.*

(i) Elizabeth—ocean port.

(A) Tolleshunt Horse Farm (horses only), 10 Island Road, Box 469, Whitehouse, NJ 08888-0469, (908) 534-7738.

(B) The U.S. Equestrian Team's headquarters (horses only), Pottersville Road, Gladstone, NJ 07934, (908) 234-1251.

(ii) Newark International Airport.

(A) Tolleshunt Horse Farm (horses only), 10 Island Road, Box 469, Whitehouse, NJ 08888-0469, (908) 534-7738.

(B) The U.S. Equestrian Team's headquarters (horses only), Pottersville Road, Gladstone, NJ 07934, (908) 234-1251.

(iii) Salem—ocean port.

(A) Mannington Meadows Farm (horses only), 60 Oechsle Road, Woodstown, NJ 08098, (609) 769-2009.

(12) *New York.*

(i) New York—airport and ocean port.

(A) Tolleshunt Horse Farm (horses only), 10 Island Road, Box 469, Whitehouse, NJ 08888-0469, (908) 534-7738.

(B) The U.S. Equestrian Team's headquarters (horses only), Pottersville Road, Gladstone, NJ 07934, (908) 234-1251.

(C) Vetport, Inc., Bldg. 189, J.F. Kennedy International Airport (Cargo Area), Jamaica, NY 11430, (212) 656-6042.

\* \* \* \* \*

Done in Washington, DC, this 28th day of May 1999.

**Bobby R. Acord,**

*Acting Administrator, Animal and Plant Health Inspection Service.*

[FR Doc. 99-14186 Filed 6-3-99; 8:45 am]

BILLING CODE 3410-34-P

**DEPARTMENT OF HEALTH AND HUMAN SERVICES**

**Food and Drug Administration**

**21 CFR Part 172**

[Docket No. 91F-0228]

**Food Additives Permitted for Direct Addition to Food for Human Consumption; Sucrose Acetate Isobutyrate**

**AGENCY:** Food and Drug Administration.

**ACTION:** Final rule.

**SUMMARY:** The Food and Drug Administration (FDA) is amending the food additive regulations to provide for the safe use of sucrose acetate isobutyrate (SAIB) as a stabilizer of emulsions of flavoring oils used in nonalcoholic beverages. This action is in response to a petition filed by Eastman Chemical Co.

**DATES:** Effective June 4, 1999; written objections and requests for a hearing by July 6, 1999. The Director of the Office of the Federal Register approves the incorporation by reference in accordance with 5 U.S.C. 552(a) and 1 CFR part 51 of certain publications in § 172.833(b) (21 CFR 172.833(b)), effective June 4, 1999.

**ADDRESS:** Written objections may be sent to the Dockets Management Branch (HFA-305), Food and Drug Administration, 5630 Fishers Lane, rm. 1061, Rockville, MD 20852.

**FOR FURTHER INFORMATION CONTACT:** Blondell Anderson, Center for Food Safety and Applied Nutrition (HFS-206), Food and Drug Administration, 200 C St. SW., Washington, DC 20204, 202-418-3106.

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**I. Introduction**

In a notice published in the **Federal Register** of September 5, 1991 (56 FR 43927), FDA announced that a food additive petition (FAP 1A4266) had been filed by Eastman Chemical Co. (Eastman), P.O. Box 511, Kingsport, TN 37662, proposing that the food additive regulations be amended in part 172 (21 CFR part 172) to provide for the safe use of SAIB as a stabilizer of emulsions of flavoring oils used in nonalcoholic carbonated and noncarbonated beverages.

SAIB is the chemical *alpha*-D-glucopyranoside, O-acetyl-tris-O-(2-methyl-1-oxopropyl)-*beta*-D-fructofuranosyl, acetate tris(2-methyl propanoate). It is also referred to as sucrose diacetate hexaisobutyrate, sugar esters of fatty acids, and sucrose esters of fatty acids.

SAIB is a slightly yellow, clear, viscous liquid, practically odorless, with a bitter taste (not apparent at the levels used in the regulated application). The compound is produced by reaction of food grade sucrose with acetic anhydride and isobutyric anhydride in the presence of a catalyst. The product is purified by molecular distillation.

In support of safety for the proposed use of SAIB, Eastman submitted toxicity studies performed in a variety of species. Those studies included: Absorption, metabolism, and elimination studies (rats, dogs, rabbits, monkeys, and humans); short-term (7 to 56 days) studies (rats, dogs, and monkeys); a palatability study (mice); subchronic (90 days) studies (rats and dogs); chronic studies (rats and monkeys); carcinogenicity studies (rats and mice); reproduction studies (rats); teratology studies (rats and rabbits); genotoxicity tests; liver function studies (rats, dogs, monkeys, and humans); and clinical studies (humans).

The one concern raised by FDA's evaluation of the SAIB data base was some liver effects, which were observed in the short-term and subchronic studies. These effects were observed primarily in SAIB-treated dogs; for example, decreased clearance rates for bromosulphophthalein (BSP) and indocyanine green (ICG) from the blood, and increased serum alkaline phosphatase. To further evaluate these liver effects, the petitioner performed special liver function tests (BSP and ICG clearance tests) in rats, dogs, monkeys, and humans. The BSP clearance test was also performed in monkeys and rats after exposure to SAIB for 1 year in order to demonstrate that the liver effects were not observed in these SAIB-

treated animals after long-term repeated exposure. The results from these studies and results from other studies that were pivotal to the safety decision for the proposed use of SAIB in beverages are discussed in section II.B of this document.

## II. Evaluation of Safety

In order to establish, with reasonable certainty, that a new food additive is not harmful under its intended conditions of use, FDA considers the projected human dietary exposure to the additive, the additive's toxicological data base, and other relevant information (such as published literature) available to the agency.

### A. Estimated Daily Intake for SAIB

In determining whether the proposed use of an additive is safe, FDA typically compares an individual's estimated daily intake (EDI) of the additive to the acceptable daily intake (ADI) established by the toxicological database. The EDI is determined by projections based on the amount of the additive proposed for use in particular foods and on data regarding the consumption levels of these particular foods.

The proposed levels of use for SAIB in beverages (up to 300 parts per million (ppm)) are supported by functionality and stability data presented in the petition. The agency commonly uses the EDI for the 90th percentile consumer of a food additive as a measure of high chronic exposure. For the requested food use of SAIB, the agency has estimated the lifetime exposure for 90th percentile consumers, 2 years old and older (all ages), to be 0.17 gram per person per day (g/p/d). The corresponding mean intake is 0.082 g/p/d (Ref. 1).

### B. Evaluation of Safety Studies on SAIB

The principal studies relevant to the safety evaluation of the petitioned use of SAIB were performed in several animal species as mentioned in section I of this document. The individual studies are identified by an Appendix number in this document, as designated by Eastman in the SAIB petition.

#### 1. Pharmacokinetics and Metabolism Studies (Appendices 16, 17, 18, 19, 22, 23, 24, 25, 26, 27, 29, and 31)

The pharmacokinetics and metabolism studies on SAIB were performed with rats, dogs, and humans in order to compare the absorption, metabolism, and excretion of the food additive in animal models to that seen in humans. Results from these studies showed the following similarities and

differences in the pharmacokinetics and metabolism of SAIB in the test subjects:

(1) There were quantitative differences in the amounts of administered SAIB that were absorbed by rats, dogs, and humans. Rats and humans absorbed greater amounts of SAIB from the gastrointestinal tract compared to dogs. In rats and humans, the majority of the orally administered SAIB was eliminated in expired air, whereas in dogs, the majority of SAIB was eliminated in the feces;

(2) Dogs excreted a greater proportion of the absorbed SAIB in the bile compared to rats. The excreted materials in the bile of the dog were identified as either unchanged SAIB or higher acylated sucrose molecules. Lower acylated sucroses were identified in the bile of rats; and

(3) The urinary metabolites of SAIB in rats and humans were more similar qualitatively than those between dogs and humans. Higher acylated sucroses were identified as the primary metabolite in the urine of dogs. In the urine of rats and humans, only lower acylated sucroses and free sucrose were identified. Free sucrose was not found in the urine from dogs. These data show that more deacylation of SAIB occurs in rats and humans than in dogs. The patterns of absorption, metabolism, and elimination are more similar for rats and humans than for dogs and humans. Therefore, the agency concludes that the rat is more appropriate than the dog to model the metabolic disposition and fate of SAIB in humans (Refs. 2, 3, 4, and 5).

#### 2. Genotoxicity Studies (Appendices 88, 89, 90, 91, 92, 93, and 94)

SAIB was subjected to the following battery of studies to evaluate its genotoxic potential in prokaryotic and mammalian species: Ames Test, Chinese Hamster Ovary Cells/HGPRT Forward Mutation Assay, In Vitro Cytogenetic Chromosomal Aberration Assay, Unscheduled DNA Assay, and Dominant Lethal Assay. In the absence of bioassay data, these tests are often used to predict the carcinogenic potential of the test compound. However, in the case of SAIB, carcinogenicity bioassays are also available.

SAIB was shown to be nonmutagenic in the Ames test, with or without metabolic activation (Appendices 88, 89, and 90) (Refs. 6 and 7). The compound did not induce changes in mutation frequency in the Chinese Hamster Ovary Cells/HGPRT Forward Mutation Assay (Appendix 91) (Ref. 8). Chromosomal aberrations were not induced in Chinese hamster ovary cells

(Appendix 92), thereby demonstrating that SAIB is not clastogenic (Ref. 9).

Results from the Unscheduled DNA Assay (Appendix 93) were negative regarding any significant increases in nuclear labeling or unscheduled DNA synthesis in rat primary hepatocytes treated/incubated with SAIB (Refs. 9, 10, 11, and 12). The Dominant Lethal Assay (Appendix 94) did not show any significant effects on early fetal deaths per pregnancy in rats.

Based upon the negative mutagenic and clastogenic findings in the genotoxicity studies, the agency concludes that SAIB is not genotoxic under the test conditions of these studies (Refs. 5, 6, 7, 8, 9, 10, 11, 12, 13, and 14).

#### 3. Reproduction and Developmental Toxicity Studies (Appendices 86 and 87)

The objectives of the reproduction and developmental toxicity studies were to evaluate the toxic potential of SAIB on the reproductive system of mature rats (males and females) as well as postnatal maturation of reproductive functions of offspring through three successive generations. Assessment of the potential effects of the food additive on the developing fetus was the objective of the teratology studies.

a. *Three-generation reproduction with teratology phase in rats (Appendix 86).* In this study, groups of Fischer F344 rats (three generations: F<sub>0</sub>, F<sub>1</sub>, and F<sub>2</sub> males and females) were administered SAIB in the diet at dose levels of 0, 0.5, 1.0, or 2.0 g per kilogram body weight per d (g/kg bw/d). Parental (F<sub>0</sub>) males were fed SAIB for 10 weeks prior to mating; F<sub>0</sub> females were fed SAIB for 2 weeks prior to mating, and throughout mating, gestation, and lactation until the time of necropsy. F<sub>1</sub> and F<sub>2</sub> males and females were exposed to SAIB in utero; during their lactation and weaning periods as well as throughout their mating, gestation, and lactation periods for respective F<sub>2</sub> and F<sub>3</sub> litters. The F<sub>1</sub> males and females were bred twice in succession to produce F<sub>2a</sub> and F<sub>2b</sub> pups. For each generation, the following reproductive parameters were examined: Mating indices, fertility indices, gestation indices, gestation length, number of corpora lutea, implantation efficiency, and number of early or late resorptions. Litters from the F<sub>1</sub> and F<sub>2</sub> generations were examined for the number of dead pups (day 0), number of live offspring per litter, sex ratios, pup survival percentages, pup weights, and physical abnormalities. Macroscopic examinations of the corpora lutea and implantations were performed on the F<sub>2</sub> dams that were

sacrificed on day 14 of gestation period of the F<sub>3</sub> generation. For the teratology phase of this study, macroscopic examination of the number and distribution of fetuses in the uterine horn and the number of resorptions and corpora lutea were performed on F<sub>1</sub> dams that were sacrificed on day 20 (during gestation) of the F<sub>2b</sub> generation. The pups from these dams were examined for any soft-tissue or skeletal malformations.

The agency observed no reproductive or developmental toxicities in three successive generations of rats that were exposed to SAIB at levels up to 2.0 g/kg bw/d. There was a trend towards decreased fertility with increasing dose of SAIB in the females of the F<sub>1</sub> generation during the breeding for the F<sub>2a</sub> litters. The agency does not consider this trend to be treatment-related because there were no significant decreases in fertility observed in the F<sub>0</sub> females during the breeding for the F<sub>1</sub> litters or the F<sub>1</sub> females during the breeding for the F<sub>2b</sub> litters. The agency has determined that the no observed effect level (NOEL) for this study is 2.0 g/kg bw/d, which was the highest dose of SAIB tested in this study (Ref. 13).

b. *Teratology study in rabbits* (Appendix 87). New Zealand White SPF female rabbits were divided into a control group (32 rabbits) and 3 SAIB treatment groups (16 rabbits per group). Control and treated female groups were induced to superovulate by receiving injections of human chorionic gonadotropin 3 weeks prior to insemination. SAIB was administered by oral gavage, twice daily, to the treatment groups at dose levels of 0.50, 0.85, or 1.20 g/kg bw/d on days 7 through 19 of gestation. The control group received only the vehicle (corn oil).

The agency concludes that in this study there were no developmental toxicities observed in rabbits that were exposed to SAIB by gavage at levels up to 1.20 g/kg bw/d during gestation (days 7 to 19). The agency has determined that the NOEL for this study is 1.20 g/kg bw/d for this study (Ref. 13).

c. *Agency conclusions regarding reproduction and developmental toxicity studies on SAIB.* Based on the data obtained from these reproduction and developmental toxicity studies on SAIB (Appendices 86 and 87), the agency concludes that the oral administration of SAIB does not induce reproductive or developmental effects in rats when tested in the diet at doses up to 2.0 g/kg bw/d or developmental effects in rabbits when tested by gavage at doses up to 1.20 g/kg bw/d. Therefore, a NOEL of 2.0 g/kg bw/d is

established for SAIB based upon the highest dose tested in the three-generation rat study (Refs. 5 and 13).

#### 4. Two-Year Carcinogenicity Studies (Appendices 95 and 96)

The objective of the carcinogenicity studies was to study the carcinogenic potential of SAIB when administered to rodents for 104 weeks.

a. *Rat study* (Appendix 95). Fischer F344 (CDF/CR1BR) rats were randomly assigned to 5 groups that were fed a dietary mixture of SAIB at dose levels of 0, 0.50, 1.0, or 2.0 g/kg bw/d for 104 weeks. Two groups of rats served as duplicate controls and were fed an NIH07 diet that had been treated with acetone only. BW data for all of the rats were collected on day 1, at weekly intervals during the study, and on the day of necropsy. Food consumption was recorded weekly. Hematology measurements were performed on all rats prior to the initiation of treatment and at the end of the study at week 104. During necropsy, organ weight data were collected for heart, kidneys, liver, testes, ovaries, and brain of the rats in the two control groups and in each of the SAIB-treated groups. Macroscopic and microscopic examinations were performed at sacrifice (week 104) on representative tissue from a comprehensive selection of organs from all groups of rats.

Survival in the treated rats was not significantly affected by the SAIB treatment for the 2-year exposure duration. The antemortem changes seen in the SAIB-treated groups at termination were similar to those seen in the concurrent control rats and represented typical changes seen in aging rats.

Overall, SAIB did not significantly affect the final mean bw's or food consumption of either the male or female rats during the 104 weeks of the study. The organ weight data showed reduced brain (absolute) weight in the 1.0 g/kg bw/d SAIB-treated females when compared to females in group 1 controls and increased kidney-to-brain ratios in the 1.0 g/kg bw SAIB-treated males when compared to males in group 2 controls. FDA did not consider these weight differences to be treatment-related or toxicologically significant because they occurred sporadically among the treated groups, that is, at only one dose level (1.0 g/kg bw/d dose) or in only one sex. There were tumors or nonneoplastic lesions that occurred in the control and SAIB-treated rats of this study that represented histopathological changes commonly seen in aging rats or represented normal variation of spontaneous tumor

incidences (e.g., testicular interstitial cell tumors, mammary gland fibroadenomas, endometrial stromal polyps, and pituitary hyperplasia). Thus, the histopathology data showed no evidence of male or female SAIB-treated rats with increased incidences of tumors or nonneoplastic lesions at any organ site that were related to the feeding of SAIB (Ref. 15).

From this study, the agency concludes that SAIB did not induce any tumors in Fischer 344 rats that were fed diets containing up to 2.0 g/kg bw/d of SAIB for 104 weeks. No SAIB-related histopathological lesions were observed in the SAIB-fed rats. Thus, the NOEL for this study is 2.0 g/kg bw/d (Refs. 5, 14, and 15).

b. *Mouse Study* (Appendix 96). In this study, groups of B6C3F<sub>1</sub>/Cr1BR mice (50 per sex per group) were fed SAIB at concentrations of 1.25, 2.5, or 5.0 g/kg bw/d in an NIH07 diet for 104 weeks. Two groups of mice served as controls and were fed an NIH07 diet that had been treated with acetone only. BW data were collected on day 1, at weekly intervals during the study, and on the day of necropsy. Food consumption was recorded weekly. Hematology measurements were performed on mice in the control and 5.0 g/kg bw group only; 10 mice per sex prior to the initiation of treatment and 15 mice per sex during weeks 28, 53, 79, and 105. During necropsy, organ weight data were collected for the kidneys, liver, gall bladder, and lungs of all mice. Macroscopic and microscopic examinations were performed at sacrifice (week 104) on representative tissue from a comprehensive selection of organs from all groups of mice.

The study results revealed no treatment-related effects on the survival of SAIB-treated mice in this study. All antemortem observations seen in the SAIB-treated mice were comparable to those seen in the concurrent controls.

Organ-to-bw ratios of the liver and lungs of the SAIB-treated mice were not different from the respective weight ratios in the control mice. There were some differences in the relative kidney weights in the SAIB-treated mice compared to controls; however, these differences were not associated with any treatment-related kidney histopathology.

The histopathology data showed an increased incidence of SAIB-treated male mice with bronchiolar/alveolar adenomas and an increased incidence of SAIB-treated male mice with (combined) bronchiolar/alveolar adenomas or carcinomas when compared to control group males (Refs. 14 and 15). The incidences of SAIB-

treated females with bronchiolar/alveolar adenomas or carcinomas were comparable to incidences in control females. According to historical control incidence data from the National Toxicology Program data base, these incidences are within the range commonly seen in aged B6C3F1 mice. Therefore, FDA concludes that the increased incidences of SAIB-treated mice with this tumor represent expected variations in spontaneous incidences and were not related to the SAIB treatment (Refs. 14 and 15). At the other organ sites, there was no evidence of increased incidences of mice with tumors or nonneoplastic lesions that were related to the feeding of SAIB (Refs. 14 and 15). From this study, the agency concludes that SAIB did not induce tumors at any organ site in B6C3F1 mice that were fed diets containing SAIB up to 5.0 g/kg bw /d for 104 weeks. No SAIB-related nonneoplastic lesions were observed in the SAIB-fed mice, nor was there other evidence of adverse effects in the SAIB-fed mice at any of the tested doses. Thus, the NOEL for this study is 5.0 g/kg bw/d (Refs. 5, 14, and 15).

#### 5. Concerns Regarding Altered Liver Function

During the early reviews of the petition, the agency raised a concern regarding liver effects that were observed in the SAIB-treated animals in short-term toxicity studies (rats, dogs, and monkeys) and in subchronic toxicity studies (rats and dogs), especially in the SAIB-treated dogs. However, the agency could not easily determine whether the liver effects observed in these SAIB-treated rats and monkeys were treatment-related because of certain inadequacies in the studies, their limited experimental designs, and the studies' short exposure durations. These studies are discussed in section II.B.5.a of this document. The agency also raised a concern that there were no chronic (1 year or longer) toxicity studies on SAIB in dogs that further examined the liver function effects.

To address these concerns, the petitioner performed BSP and ICG clearance tests, which are specific liver function tests, with rats, dogs, and monkeys. In addition, to address the concern regarding possible altered liver function in chronically-exposed animals, the petitioner performed a 1-year oral toxicity study on SAIB in monkeys; this study included BSP clearance tests and measurements of clinical chemistry parameters relevant to liver toxicity. The petitioner also performed BSP clearance tests in humans that were administered doses of

SAIB up to 0.02 g/kg bw/d for 14 days to evaluate any potential effects of SAIB on liver function in humans. These studies and the agency's conclusions regarding them are discussed in sections II.B.5.b and II.B.5.c of this document.

a. *Liver Effects in the SAIB-treated Animals. i. Short-term Studies* (Appendices 60, 66, 71, 72, 73, 74, and 77). The following short-term studies were designed to provide data on the short-term oral toxicity of SAIB in rats, dogs, and monkeys with regard to potential target organs of SAIB, as well as to determine appropriate doses for the subchronic and chronic studies.

*Rat Studies* (Appendices 66 and 74). In a short-term study (Appendix 66), SAIB was fed to groups of male and female rats at levels of 1.0, 2.0, or 4.0 percent (equivalent to 1.0, 2.0, or 4.0 g/kg bw/d) in the diet for 28 or 56 days. Levels of serum alkaline phosphatase (SAP), glucose, ornithine carbamyl transferase, triglyceride, cholesterol, and blood urea nitrogen were examined. Organ weight data were collected only on the liver.

The limited clinical chemistry data from this study showed decreases in blood glucose levels in female rats fed SAIB at levels of 2.0 and 4.0 percent in the diet for 56 days. The glucose levels in the treated males were not different from comparable levels in controls for the 56-day duration. SAP levels were not affected in the SAIB-treated rats. There were no effects on bw or bw gain in the SAIB-treated rats. Liver weights in these SAIB-treated rats were similar to control rats. Also, the levels of glucose in the treated groups were not different from controls (Ref. 16).

In another short-term study (Appendix 74), groups of rats (15 per sex per group) were fed diets containing 0, 5,000, or 50,000 ppm (equivalent to 0, 0.50, or 5.0 g/kg bw/d) SAIB for 3 weeks. Organ weight data on livers from the male and female SAIB-treated rats (five per sex per group) revealed no evidence of liver enlargement at either of the doses of SAIB. In addition, SAIB did not affect bw gain or food consumption in this study (Ref. 3).

*Dog Study* (Appendix 77). In this study (Appendix 77), six male beagle dogs were initially fed a ground chow diet without SAIB (control diet) daily for 3 weeks. For the next 3 to 4 weeks, the six male dogs were fed a ground chow containing 5-percent (equivalent to 1.25 g/kg bw/d) SAIB. ICG clearance tests were performed on four of the six dogs at week 3 of this 5-percent SAIB feeding period. After the 3 or 4 weeks feeding period of SAIB, the dogs were returned to control diet for an additional 8 weeks (91st day). ICG clearance tests

were performed on week 3 and 6 of this 8-week control diet feeding period. On the 88th day, 4 of the 6 dogs were returned to a diet containing 5-percent SAIB for 1 day. After this 1-day SAIB feeding, SAP measurements and ICG clearance tests were performed on the six dogs. This study did not have a group of dogs that served as concurrent controls nor were pretest ICG baseline values determined. Instead, the data from this study were compared to previously reported laboratory data for ICG clearance in normal beagle dogs.

The results of this study showed decreased clearance of serum ICG (half-lives ( $t_{1/2}$ )<sup>1</sup> of 17.0 to 40.0 minutes) in dogs that were fed 5-percent SAIB for 3 weeks compared to ICG clearance in normal dogs ( $t_{1/2}$  of 4.2 to 8.1 minutes). ICG clearance in the SAIB-treated dogs had returned to normal by day 84 after these dogs were returned to control diets without SAIB. Five of six dogs had increased SAP levels at the end of the 4-week SAIB feeding period that were four to seven times greater than pretest values.

Blood glucose levels decreased (25- to 57-percent reductions) in all of the dogs at the end of the 4-week SAIB treatment period compared to pretest average values. However, blood glucose levels monitored at the end of the recovery phase of the study were reversed and were comparable to the pretest values. Ornithine carbamyl transferase and blood cholesterol levels also increased during the SAIB exposure period. Other blood parameters measured in these dogs (hemoglobin, hematocrit, white blood cell counts (five out of six dogs), serum protein, and blood urea nitrogen) were not affected by the 5-percent SAIB treatment. The 5-percent SAIB treatment had no effect on body weight, food consumption, or organ weights (only liver and kidney were measured) in the dogs for the 4-week period (Ref. 3).

*Monkey Studies* (Appendices 60, 71, 72, and 73). In a short-term study (Appendix 60), SAIB was administered by oral intubation (in an orange juice concentrate) to four monkeys (two per sex) as a single dose that started at a dose of 1.25 g/kg bw, increased by increments of 2-fold (72-hour intervals between doses), and ended at a dose of 20 g/kg bw over a dosing period of 14 days. All of the SAIB-dosed monkeys survived the study. Slight to moderate watery, yellow stools were observed in some of the monkeys administered SAIB at doses of 1.25 g/kg bw (one male, two females), 2.5 g/kg bw (one male, one female), and 5.0 g/kg bw (one female).

<sup>1</sup> Half-life( $t_{1/2}$ ) is the time required for the serum ICG concentrations to be reduced by one half.

Large amounts of watery yellow stools and emesis were observed in a monkey that received a SAIB dose of 5.0 g/kg bw dose. Gross postmortem examinations of the four monkeys after the last dosing of SAIB revealed no effects that were attributable to the SAIB administration (Refs. 2 and 17).

In a two-part range-finding study (Appendices 71 and 72), SAIB was administered by oral intubation to groups of monkeys (one per sex per group) at dose levels of 0, 0.5, 1.0, 2.0, 5.0 or 10.0 g/kg bw/d for 15 days. Incidences of soft, loose stools were observed in the SAIB-dosed groups (1.0, 2.0, and 10.0 g/kg bw/d doses), as well as in the control male and female groups. At the termination of the study, SAP levels in the males of the 10.0 g/kg bw/d dose group and the females of the 5.0 and 10.0 g/kg bw/d dose groups were increased compared to their respective controls. Pretest alkaline phosphatase levels in the SAIB-dosed groups were also higher than pretest levels of the controls. Decreased BSP clearance was observed in 8 out of the 10 treated monkeys. Electron microscopy was performed only on the livers of the control group and the high-dose group in this study. Results from the ultrastructural analyses of the livers from the SAIB-treated monkeys revealed increased glycogen, large glycogen aggregations surrounded by scant smooth endoplasmic reticulum, and decreases in the amounts of smooth endoplasmic reticulum (Refs. 2, 3, and 17). While these effects in the SAIB-dosed monkeys suggest suppressed liver function, the agency could not determine the toxicological significance of these effects because of the small group sizes (Refs. 2, 3, and 17).

In another exploratory study (Appendix 73), groups of monkeys (one per sex per group) were administered SAIB (in corn oil) orally by gavage at doses of 0.50, 1.45, or 2.40 g/kg bw/d for 4 weeks. Control monkeys received only the vehicle (corn oil) by gavage. BW gains were comparable in all of the groups except for the high-dose female monkey, who lost weight (12-percent loss) over the 4-week study duration. Reduced food consumption was reported for this high dose female monkey. SAP levels were increased 8-to 78-percent in the treated groups for both sexes except for the one male in the high-dose (10 g/kg bw dose) group. Values reported for erythrocyte counts, hemoglobin, and hematocrits were low for all of the females in both the treatment groups and the control group. BSP clearance rates in these monkeys were normal. Clinical biochemistry parameters related to liver and kidney

functions were also normal in the dosed monkeys (Refs. 3 and 17).

*Agency conclusions regarding short-term studies on SAIB.* The agency's overall review of the data from the preceding short-term studies (see section II.B.5.a.i of this document) established the following: (1) Decreased glucose levels in rats that were fed SAIB at levels of 2.0 and 4.0 g/kg bw/d for 56 days; (2) decreased ICG clearance rates, increased SAP levels, and decreased blood glucose levels in dogs that were fed 1.25 g/kg bw/d SAIB for 4 weeks; and (3) increased BSP retention and increased SAP levels in monkeys that were administered SAIB by gavage at dose levels of 5 and 10 g/kg bw/d for 15 days. Based on these observed effects, the agency concludes that the liver is a target organ for the toxicity of SAIB. However, because of the short exposure durations and limited experimental designs of these studies, the agency concludes that these studies are inadequate to resolve concerns regarding the observed liver effects (Refs. 3 and 5).

ii. *Subchronic oral toxicity studies on SAIB (Appendices 63, 64, 65, 67, 68, 69, and 70).* The following subchronic oral toxicity studies were performed in rats and dogs to examine the general systemic toxicity of SAIB and to investigate further the liver effects of SAIB that were observed in the short-term SAIB studies.

*Rat Studies (Appendices 63, 64, and 65).* In a 90-day study (Appendix 63), groups of rats (25 per sex per group) were fed SAIB in the diet at 0, 1, or 5 percent (equivalent to 0, 1.0, or 5.0 g/kg bw/d). This study showed an increase (7.4 percent) in the relative liver weights of the 5-percent SAIB-treated female rats compared to the control females; liver weights in SAIB-treated males were not affected. Kidney weights in the SAIB-treated groups were not different from the kidney weights of the control rats. Final bw's were slightly decreased (3 to 4 percent) in only the males of the 5-percent dose group. No differences were observed in the final bw's of the males in the 1-percent dose group or the females in all of the dose groups when compared to respective controls. BW gain in all of the female treatment groups was comparable to the female control groups. Overall, feed intakes and feed efficiencies appeared to be similar across treatment and control groups for both sexes (Ref. 16).

In another 90-day study (Appendix 64), groups of rats (10 per sex per group) were fed SAIB in the diet at levels of 0, 0.38, 1.88, or 9.38 percent (equivalent to 0, 0.38, 1.88, or 9.40 g/kg bw/d). A slight increase in the mean hemoglobin values

and a tendency toward leukocytosis (increased white corpuscle counts) were observed in treated rats relative to control rats. SAP levels and BSP clearance rates were not evaluated in this study. BW gains in the SAIB-treated males were slightly decreased (8 to 11 percent) compared to control males; in treated females, bw gain was not affected. Liver, kidney, lung, gonad, spleen, and heart weights (relative and absolute weights) of the SAIB-treated rats were not significantly different from the respective organ weights of the control rats.

Data from the limited histopathological analyses showed an increased incidence of clear vacuoles (fat vacuoles) in the livers of all of the SAIB-treated rats with the greatest increase being seen in the 1.88-percent SAIB group (Ref. 16).

In a 12-week study (Appendix 65), groups of rats (20 per sex per group) were fed SAIB at doses of 2.5, 5.0, or 10 percent (equivalent to 2.50, 5.0, or 10.0 g/kg bw/d) in the diet. SAIB-treated male rats in this study showed decrements in weight gain at all dose levels compared to controls; weight gains in the SAIB-treated female rats were not affected. There was a significant decrease in SAP levels in females treated with 10-percent SAIB. Urinary ascorbic acid levels were substantially decreased (47 percent in males and 64 percent in females) in the 10-percent SAIB group relative to controls. There were no increases in carboxyl esterase levels in any of the SAIB-treated rats. Neither liver weights nor the ultrastructure of the livers in the SAIB-treated rats were affected during the study. Biochemical analyses performed on the livers of rats in the control and 10-percent SAIB groups showed increases in liver glycogen in the 10-percent SAIB group (in both sexes) as well as significant increases in the water content of the livers in the males of the 10-percent SAIB group (Ref. 16).

Because of inadequacies in data analyses and reporting (e.g., limited statistical analyses and incomplete histopathology data) in the subchronic rat studies, the agency could not reach a conclusion as to whether there were treatment-related liver effects in the SAIB-fed rats of these studies. The results from these studies did show: (1) Significantly increased (relative to controls) relative liver weights in rats (females only) that were fed 5-percent SAIB, and (2) increased glycogen content and increased water content (males only) in the livers of rats (both sexes) fed 10-percent SAIB relative to controls (Ref. 5).

*Dog Studies (Appendices 67, 68, 69, and 70).* In a 12-week study (Appendix 67), groups of dogs (four per sex per group) were fed diets containing 0, 0.2, 0.6, or 2.0 percent (equivalent to 0, 0.05, 0.15, or 0.5 g/kg bw/d) SAIB. This study showed increases in SAP levels in the SAIB-treated male dogs, with a two-fold increase in the 2.0-percent dose group. At the end of the study, relative liver weights of male and female dogs fed SAIB at the 0.6-percent and 2.0-percent dose levels increased compared to the respective control groups. Relative weights of the other organs that were examined in the study (kidney, spleen, brain, gonads, adrenals, thyroids, and pituitary) did not differ significantly from respective relative organ weights of controls. Survival, hematology parameters, and urine parameters tested in the SAIB-treated dogs were also not significantly different from controls (Ref. 18).

In another subchronic study (Appendices 68 and 69), groups of dogs (six per sex per group) were fed dog chow containing 0-, 0.5-, 1.0-, 2.0-, or 4.0-percent (equivalent to 0, 0.13, 0.25, 0.50, or 1.0 g/kg bw/d) SAIB for 12 weeks followed by a 3-week recovery period, during which the dogs were fed a chow diet that did not contain SAIB. During the 12-week treatment period and the 3-week recovery period of the study, the control group received a basal chow meal without SAIB. During the 12-week exposure period, all of the dogs in this study that were fed SAIB (all doses) exhibited significant increases (3- to 7-fold) in serum BSP concentrations compared to control dogs. BSP retention data collected during the 3-week recovery period without SAIB showed a reduction in BSP plasma levels in the 4-percent SAIB-treated dogs to levels that were similar to pretest values and those seen in control dogs (Appendix 69).

Relative liver weights increased in the male dogs fed SAIB at levels of 1.0 and 2.0 percent in the diet; relative liver weights in the 0.5-percent SAIB-treated males were not different from controls. Relative liver weights in the SAIB-treated female dogs (all groups) were not significantly different from control females. Absolute liver weights were significantly increased in SAIB-treated males at dose levels of 0.5, 1.0, and 2.0 percent. Liver weights of the 4.0-percent male dose group were not analyzed at the time that the 1.0 and 2.0-percent male dose groups were analyzed; instead, this dose group was held for 3 additional weeks for a recovery phase of the study. At the end of the 3-week period (recovery phase), absolute and relative liver weights of the 4-percent male dose group were also significantly

increased when compared to control liver weight values measured at the end of the 12-week treatment phase. This 3-week recovery phase of the study did not include a comparable control group of dogs that was held for the additional 3 weeks after the treatment phase.

Data from liver biochemistry analyses showed significantly increased liver glycogen in all of the SAIB-treated groups, significantly increased liver lipid content in all of the dogs fed 2.0-percent SAIB, and significantly increased liver carboxyl esterase levels in all of the dogs fed 4.0-percent SAIB. Total protein levels in the liver were greatly reduced in all of the SAIB-treated groups compared to controls. Alkaline phosphatase, adenosine triphosphatase, and glucose-6-phosphatase levels in the bile canaliculi of the livers in all of the dose groups increased relative to controls.

Results from the microscopic (light and electron) analysis of liver tissue samples showed dilation of the bile canaliculi, liver hypertrophy and enlargement (males only), increased bile pigment granules, increases in the smooth endoplasmic reticula, and prominent Golgi bodies in the dogs fed 2-percent SAIB in the diet (Appendix 69). In addition, the distribution and arrangement of the smooth and rough endoplasmic reticula were altered in the 2-percent SAIB-treated dogs (Ref. 18).

In a 91-day study (Appendix 70), a group of five dogs were fed dog chow containing 5-percent (equivalent to 1.25 g/kg bw/d) SAIB. A second group of five dogs served as controls and was fed dog chow containing 5-percent corn oil for the study duration. This study demonstrated that SAIB significantly affected liver function in the five SAIB-treated dogs, causing moderate elevations in SAP levels, prolonged ICG clearance, and increases in the absolute and relative liver weights. Hematological or clinical chemistry parameters examined in this study, other than SAP, were not affected by the SAIB treatment (Ref. 3).

Based upon the data in the subchronic studies in dogs, the agency concludes that SAIB affected liver function in SAIB-treated dogs at all of the tested dose levels. As noted, the liver effects observed in the SAIB-treated dogs were: (1) Increased BSP retention at SAIB doses as low as 0.13 g/kg bw/d and up to a dose of 1.0 g/kg bw/d, (2) increased SAP levels at SAIB doses of 0.05 g/kg bw/d and higher, (3) increased liver weights at doses of 0.13 g/kg bw/d and higher, and (4) liver ultrastructural changes in the 0.5 g/kg bw/d dose group (liver enlargement/hypertrophy, increased liver glycogen deposition,

increased liver carboxyl esterase activity, and proliferation of smooth endoplasmic reticulum). Because effects were observed at the lowest tested dose, the agency could not establish a NOEL for the observed liver effects in the SAIB-treated dogs in the subchronic studies (Refs. 3, 5, and 18).

*Agency Conclusions Regarding Subchronic Studies on SAIB.* The agency concludes from the subchronic studies that SAIB affected liver function in dogs when fed SAIB at doses of 0.13 g/kg bw/d up to 1.0 g/kg bw/d.

The subchronic studies in rats also suggested apparent liver effects in rats that were fed SAIB at dose levels of 5.0 g/kg bw/d and higher. However, because of study limitations (e.g., incomplete histopathology data and inadequate statistical analyses), the agency could not determine from the subchronic rat studies whether the liver effects seen in the SAIB-treated rats were caused by the treatment with SAIB (Refs. 3, 5, 16, and 18).

In order to investigate further the effects of SAIB on liver function in different species, the petitioner performed specific liver function tests in rats, dogs, monkeys, and humans. The results from these tests are discussed in sections II.B.5.a.iii. and II.B.5.b.ii of this document.

iii. *Specific liver function tests (Appendices 75, 76, 78, 80, and 81).* BSP and ICG clearance tests were performed by the petitioner in rats, dogs, and monkeys. In these tests, BSP or ICG is administered by injection and the clearance of these dyes from the blood is analyzed spectrophotometrically at various time intervals up to 48 hours. In normal subjects, generally 95 percent of the injected dye is cleared from the blood through the liver within 30 minutes. Retention of BSP in the blood is indicative of some form of liver dysfunction such as hepatic degeneration/inflammatory changes, hepatic fibrosis, hepatic cholestasis, or depressed hepatic blood flow (Refs. 19, 20, 21, 22, 23, and 24).

*Rat Tests (Appendices 75 and 76).* In a 36-day study (Appendix 75), two groups of rats (17 males per group) were fed a chow diet containing either 4.0-percent (equivalent to 4.0 g/kg bw) SAIB in 5.0-percent corn oil or only 5.0-percent corn oil. On days 1, 3, 5, 8, 10, 22, 26, and 36, after the start of these diets, 2 rats from each group were selected for ICG clearance testing. ICG clearance rates in SAIB-treated rats were not significantly different from control rats at any of the time intervals (Ref. 3).

In a 7-day study (Appendix 76), 15 rats (5 males per group) were fed a rodent diet containing 4-percent

(equivalent to 4.0 g/kg bw/d) SAIB. BSP clearance was measured in these rats at 0, 24, and 48 hours posttreatment with SAIB. SAIB had no effect on BSP clearance from the liver in these rats when fed for 7 days (Ref. 3).

**Dog Tests (Appendices 76 and 78).** In an intermittent dosing study (Appendix 76), two male and two female dogs were serially provided, on one dose per week, laboratory dog chow ration containing SAIB at increasing concentrations of 0.1, 0.3, and 0.5 percent (equivalent to dose levels of 0.03, 0.08, or 0.13 g/kg bw). The animals were fed dog chow without SAIB on days between each dosing. BSP clearance rates for the 4 dogs were evaluated at 24 and 48 hours following each dosing. BSP clearance rates were also measured in each of the dogs prior to the start of the study to determine pretest baseline values. Results from this study showed increased BSP retention at the 24-hour time interval in the dogs at all treatment levels (Ref. 3).

Results from another study in dogs (Appendix 78) showed that BSP retention increased (up to seven-fold) in both male and female dogs administered SAIB as single (oral gavage) doses ranging from 0.005 g/kg bw to 2.0 g/kg bw. Initial increases of BSP levels were observed within 4 to 6 hours posttreatment with SAIB (Refs. 2 and 3).

**Monkey Tests (Appendices 80 and 81).** In a study (Appendix 80), a group of monkeys (three males) were administered 1.0 g/kg bw of SAIB in cottonseed oil by gavage as a single dose. A second group of monkeys (three males) received no treatment and served as controls. After this dosing of SAIB, BSP clearance tests were performed. The three SAIB-treated monkeys were given a second 1.0-g dose of SAIB after a 7-day rest period followed by additional BSP clearance testing. The first SAIB dosings showed an increase in the BSP level in one of the three treated monkeys, while the second SAIB dosing resulted in an increase in the BSP levels in a different treated monkey (Ref. 3). FDA concluded that these results are inconclusive because of the equivocal BSP results and the small group sizes.

In another study (Appendix 81), a group of monkeys (four males) was administered SAIB orally by gavage at a dose of 5 g/kg bw. Another group of four males was gavaged with corn oil and served as a control group. BSP clearance was tested in the control and SAIB-treated monkeys 5 hours after the SAIB dosing. The group mean BSP level in the treated monkeys was comparable to that in the control group (Ref. 3). Based upon the results from this study, which tested a higher dose of SAIB and had a

larger group size than the above 1.0 g/kg bw monkey study (Appendix 80), FDA concludes that BSP clearance was not affected in monkeys that were orally gavaged with SAIB as a single dose of 1 or 5 g/kg bw (Ref. 5).

Based upon FDA's reviews of these liver function tests, the agency concludes that liver function in dogs was clearly affected by SAIB regardless of the doses tested (0.005 to 2.0 g/kg bw). From these studies the agency also concludes that liver function was not affected in either rats or monkeys at SAIB doses up to 5 g/kg bw. However, because of the short duration of these studies, the agency was unable to determine whether liver function would be affected in rats or monkeys upon chronic exposure to SAIB.

In response to this concern of FDA, the petitioner conducted two 1-year feeding studies (rats and monkeys), in which test animals were subjected to specific liver function tests following a continuous SAIB exposure for 1 year. Results from these 1-year studies are discussed in section II.B.5.b.i of this document. In addition to the 1-year studies in rats and monkeys, the petitioner conducted three human clinical studies to investigate whether the liver function effect that was consistently demonstrated in SAIB-treated dogs could also occur in humans upon oral ingestion of SAIB. Results from the three human clinical studies are also discussed in section II.B.5.b.ii of this document.

**b. Studies resolving the altered liver function issue.** The petitioner performed two 1-year chronic toxicity studies (rats and monkeys) and the human clinical studies in an effort to resolve the concern regarding liver function. These investigations are discussed in sections II.B.5.b.i. and II.B.5.b.ii. of this document.

**i. One-Year Chronic Toxicity Studies (Appendices 83 and 84).** The 1-year chronic toxicity studies were performed in rats and in monkeys in order to evaluate any general toxicological effects of SAIB in these animals and to investigate whether there were effects on liver function in rats and monkeys chronically-exposed to SAIB.

**Rat Study (Appendix 83).** Groups of male and female Charles River rats (20 per sex per group) were fed SAIB in the diet at dose levels of 0, 0.5, 1.0, or 2.0 g/kg bw/d for 52 weeks. The control group was fed the diet minus SAIB for the same duration. BSP clearance tests were performed during weeks 23 and 48 on all control and high-dose rats after an overnight fast. Ophthalmic examinations were performed at weeks 0, 26, and 52 of the study. Selected

hematology and clinical chemistry tests were performed on 10 animals prior to dosing and on all animals at weeks 27 and 53. Histopathological examinations were performed on tissue from liver, kidneys, lungs, and all lesions from all dose groups. Liver sections were also processed for electron microscopy.

A small bw gain decrement (10.3 percent) was observed in the high-dose (2.0 g/kg bw/d) SAIB-treated females. The mean final bw in the high-dose females was also significantly decreased by 6.4 percent, compared to controls. The decreased bw gain in the high-dose females was mostly accounted for by decreased food intake (4-percent reduction). BW gains in the SAIB-treated females at the mid and low doses were not different from control females. The decreases in bw gain that were sporadically seen in the SAIB-treated males in the short-term studies were not observed in the males during this 1-year chronic study. Because the bw gain decrement observed in the high-dose females was small, and because it was not observed in either the low- or mid- dose females or in treated males, and was partially accounted for by decreased food intake in females, the agency concludes that this effect is not toxicologically significant.

No differences were observed in BSP clearance between the SAIB-treated rats and the control rats at 23 or 48 weeks. Other clinical chemistry parameters measured in the SAIB-treated rats at week 53 were comparable to values in control rats.

An increased incidence of high-dose female rats with hepatocellular adenomas (2 out of 19) was observed in this study but was not seen in the longer-term (2-year) rat carcinogenicity study on SAIB, indicating that this effect was not treatment related (see section II.B.4 of this document). Therefore, the agency concludes that there are no indications of liver toxicity or other toxicologically significant effects seen in rats chronically exposed to SAIB for 1 year. The NOEL for this study is 2.0 g/kg bw/d, the highest dose tested (Refs. 5, 16, and 25).

**Monkey Study (Appendix 84).** In this study, groups of *Cynomolgus* monkeys (four per sex per group; young adults, age unknown) were administered SAIB in corn oil by gavage at doses of 0, 0.50, 1.45, or 2.40 g/kg bw/d for 1 year. The control group was administered only corn oil in a similar manner for the same duration. Ophthalmic, hematological, and clinical chemistry examinations were performed at pretest and at months 3, 6, 9, and 12 of the study. BSP clearance tests were performed to assess liver function in all



animals at pretest and at months 3, 6, 9, and 12 of the study. Organ weight data (absolute and relative) were collected on brain, thyroid/parathyroid, heart, kidney, liver, testis, spleen, ovary, pituitary, and adrenals for all monkeys after week 52 of the study. Macroscopic and microscopic examinations were performed at sacrifice (week 52) on representative tissue from a comprehensive selection of organs. Liver sections from the monkeys in the control and high-dose group were processed for electron microscopy.

The survival, bw, ophthalmoscopic, and hematological data showed no findings that were toxicologically significant or SAIB-related. There were some differences noted between the SAIB-treated and control monkeys for some of the clinical chemistry parameters, but these were sporadically expressed and thus were not toxicologically significant.

Data from the clinical chemistry parameters that assessed hepatobiliary function did not reveal any effects that could be attributed to the administration of SAIB. The percentages of BSP excretion seen 30 minutes after BSP dye injection in the SAIB-treated monkeys at 3, 6, and 12 months, were similar to those seen in controls at comparable time intervals. There were no differences between SAIB-treated monkeys and control monkeys with respect to SAP levels, cholesterol, bile acids, bilirubin, and gamma glutamyl transpeptidase. Organ weight data for SAIB-treated monkeys were comparable to control monkeys except for some occasional differences in the combined weights of the thyroid and parathyroid glands (absolute and relative) in the low- and mid-dose male monkeys and in the absolute and relative ovary weights in high-dose female monkeys. The liver weights (absolute or relative weights) of the dosed monkeys were not different from the liver weights in control monkeys. The agency concludes that none of these changes are toxicologically significant.

Electron micrographs of liver tissue from the SAIB-treated monkeys (high-dose group, four per sex) showed no difference from controls in the quantity of smooth endoplasmic reticulum in their livers. Compared to the controls, there were no ultrastructural changes in either the mitochondria or their associated rough and smooth endoplasmic reticula or any evidence of peroxisomal proliferation in the liver of the SAIB-treated monkeys. Based upon these findings, the agency concludes that there was no evidence of abnormalities in the livers of the SAIB-treated monkeys compared to livers

from control monkeys that would indicate an SAIB-induced effect on liver function.

Based upon FDA's review of the data in this 1-year chronic study, the agency concludes that SAIB does not affect the function or ultrastructure of the liver in monkeys when orally administered at doses up to 2.40 g/kg bw/d for 1 year. No other SAIB-related histopathological lesions were observed in the SAIB-treated monkeys, nor was there other evidence of adverse effects in the SAIB-gavaged monkeys at any of the administered doses. Therefore, the agency has determined that the NOEL for this study is 2.40 g/kg bw/d (Refs. 5 and 17).

ii. *Human clinical studies* (Appendices 97, 98, and 99). The primary objective of the human clinical studies was to evaluate any potential effects of SAIB on liver function in humans when administered as a single daily dose for 14 days.

In a 14-day study (Appendix 97), SAIB was administered to 20 human subjects (10 per sex) daily as a single dose of 0.01 g/kg bw/d. In a second 14-day study (Appendix 98), groups of human subjects (4 per sex) were administered daily a carbonated beverage containing SAIB at either a dose of 0.007 g/kg bw or 0.20 g/kg bw. A third group (four per sex) served as a control and were administered daily a carbonated beverage without SAIB. In a third 14-day study (Appendix 99), groups of 13 human male and 14 human female human subjects were administered daily orange juice containing SAIB at a dose of 0.02 g/kg bw/d. In each of these clinical studies, hematology and clinical chemistry parameters were measured prior to the SAIB dosing on day 0, during the study on day 7, and at the end of the study on day 14 or 18. BSP clearance tests were performed prior to the SAIB dosing and postdosing on day 15.

None of these studies showed any SAIB-related abnormalities in any of the hematology or clinical chemistry parameters measured in these studies, including those clinical chemistry parameters that assessed hepatobiliary function (i.e., SAP levels, alanine amino transferase, aspartate amino transferase, lactate dehydrogenase, gamma glutamyl transferase, bile acids, and total bilirubin). BSP retention in all of the SAIB-treated human subjects was normal compared to pretest values or control values.

Based upon the data in these studies, the agency concludes that SAIB is not toxic in humans and does not induce liver toxicity at doses up to 0.02 g/kg bw/day for 14 days. The 0.02 g/kg bw

SAIB dose is equivalent to exposures resulting from drinking 4 liters per day of a beverage containing SAIB at its assumed maximum allowable use level of 300 milligrams/liter (mg/L) (Refs. 5, 26, 27, and 28).

c. *Agency conclusions regarding altered liver function issue.* During the initial safety review of SAIB, FDA raised a concern that, regardless of the tested dose or study duration, treatment-related liver effects were consistently noted in SAIB-treated dogs. In response to this concern, the petitioner provided a significant amount of pharmacokinetics and metabolism data on SAIB in various species, including humans. Based on these data, FDA finds that there appear to be greater quantitative differences in the absorption and metabolism of SAIB between dogs and humans than between the other tested species and humans. To evaluate further the significance of the liver effects to the overall safety of SAIB for human consumption, the agency carefully considered the test results with monkeys, a nonhuman primate species that is phylogenetically closest to humans, as well as liver function data collected directly from human subjects in the three clinical studies.

Unlike the liver effects seen in SAIB-treated dogs, there was no evidence of liver effects in the specific liver function tests with monkeys that received acute oral doses of SAIB as high as 5 g/kg bw (Appendix 84). Data also demonstrate a lack of treatment-related liver effects in monkeys that were exposed continuously to SAIB at dose levels up to 2.4 g/kg bw/d over a 1-year treatment period. Importantly, this dose level of 2.4 g/kg bw/d is nearly one thousand fold the anticipated 90th percentile human exposure of SAIB in the daily diet.

FDA's review of the human clinical studies (Appendices 97, 98, and 99) further support the agency's conclusion regarding the significance of the liver effects. In all three clinical studies, no SAIB-induced effects on liver function were observed in either male or female subjects. While the duration of the human studies was relatively short (14 days), the highest dose used (0.02 g/kg bw/d) provided reasonable assurance, in conjunction with the chronic monkey study data (Appendix 84), that the liver effects seen in SAIB-treated dogs will not occur in humans that ingest SAIB. The highest dose tested in the human clinical studies is equivalent to an exposure resulting from the drinking 4 L/d of a beverage containing SAIB at its proposed maximum allowable use level of 300 mg/L.



Based upon FDA's reviews of the nonhuman primate data and the direct human data provided in the SAIB data base, the agency concludes that the liver function effect seen in SAIB-treated dogs is not determinative of the overall safety evaluation of SAIB for human consumption. The agency further concludes that there is reasonable certainty that the adverse liver effects seen in the SAIB-treated dogs will not occur in humans that consume SAIB at the anticipated levels of dietary intake.

### C. Acceptable Daily Intake for SAIB

As discussed in section II.B.5.c of this document, FDA has relied on the monkey and human data to resolve questions concerning the altered liver function observed in SAIB-treated dogs. To support the overall safety of SAIB for human consumption and to establish an ADI, FDA has relied on data from rat studies of SAIB because the most complete toxicological profile of SAIB was established in this rodent species. The rat studies in the SAIB data base assess both the potential carcinogenicity and the reproductive/developmental toxicity of SAIB. In addition, because of their duration and size, the chronic rat studies had greater sensitivity and thus, were more likely to manifest treatment-related chronic effects. Furthermore, the available absorption and metabolism data demonstrated substantial similarities, both qualitative and quantitative, between rats and humans in the metabolic handling of SAIB following oral ingestion.

Based on the 1- and 2-year rat studies, FDA determined that the highest dose tested in both studies (2.0 g/kg bw/d) was the NOEL for SAIB. Based on this NOEL and the use of a safety factor of 100, FDA calculated an ADI of 0.02 g/kg bw/d or 1.20 g/p/d for SAIB (Ref. 5). The EDI exposure for SAIB is 0.17 g/p/d (90th percentile, all ages) which is 14 percent of the ADI calculated for the additive.

### III. Conclusion

Based on all the SAIB data reviewed by the agency, FDA concludes that there is a reasonable certainty that no harm will result from the use of SAIB as an emulsion stabilizer for flavoring oils in nonalcoholic beverages, and thus, SAIB is safe for its proposed use. Therefore, the agency concludes that the food additive regulations should be amended as set forth in this document.

In accordance with § 171.1(h) (21 CFR 171.1(h)), the petition and the documents that FDA considered and relied upon in reaching its decision to approve the petition are available for inspection at the Center for Food Safety

and Applied Nutrition by appointment with the information contact person listed above. As provided in § 171.1(h), the agency will delete from the documents any materials that are not available for public disclosure before making the documents available for inspection.

### IV. Environmental Effects

The agency has determined under 21 CFR 25.32(k) that this action is of a type that does not individually or cumulatively have a significant effect on the human environment. Therefore, neither an environmental assessment nor an environmental impact statement is required.

### V. Paperwork Reduction Act of 1995

This final rule contains no collection of information. Therefore, clearance by the Office of Management and Budget under the Paperwork Reduction Act of 1995 is not required.

### VI. References

The following references have been placed on display in the Dockets Management Branch (address above) and may be seen by interested persons between 9 a.m. and 4 p.m., Monday through Friday. **Note:** References with an asterisk are not on display; they are available generally because they are published articles or books.

1. Memorandum from DiNovi, Division of Product Manufacture and Use, to Anderson, Direct Additives Branch, FDA, July 31, 1991.
2. Memorandum from Taylor, Additives Evaluation Branch, to McLaughlin, Direct Additives Branch, FDA, November 4, 1985.
3. Memorandum from Pellicore, Additives Evaluation Branch, to Anderson, Direct Additives Branch, FDA, September 29, 1992.
4. Memorandum from Pellicore, Additives Evaluation Branch, to Anderson, Novel Ingredients Branch, FDA, February 5, 1993.
5. Memorandum from Whiteside, Division of Health Effects Evaluation, to Anderson, Division of Product Policy, FDA, August 27, 1998.
6. Memorandum from Donnelly, Genetic Toxicity Branch, to Dunkel, Genetic Toxicity Branch, FDA, September 23, 1985.
7. Memorandum from Prival, Additives Evaluation Branch #1, to Whiteside, Additives Evaluations Branch #2, FDA, March 14, 1996.
8. Memorandum from Moreland, Genetic Toxicity Branch, FDA, August 12, 1985.
9. Memorandum from Lavappa, Genetic Toxicity Branch, to Chief, Genetic Toxicity Branch, FDA, September 9, 1985.
10. Memorandum from Bradlaw, Genetic Toxicity Branch, to Dunkel, Genetic Toxicity Branch, FDA, May 24, 1985.
11. Memorandum from Bradlaw, Genetic Toxicity Branch, to Dunkel, Genetic Toxicity Branch, FDA, August 15, 1985.
12. Memorandum from Bradlaw, Genetic Toxicity Branch, to Lin, Additives Evaluation Branch, FDA, September 6, 1989.

13. Memorandum from Welsh, Additives Evaluation Branch #2, to Whiteside, Additives Evaluations Branch #2, FDA, August 28, 1995.

14. Memorandum from Whiteside, Division of Health Effects Evaluation, to Lorentzen, Cancer Assessment Committee, FDA, March 18, 1997.

15. Memorandum of Conference, Cancer Assessment Committee Meeting, FDA, October 28, 1996.

16. Memorandum from Raffaele, Additives Evaluations Branch #2, to Whiteside, Additives Evaluations Branch #2, FDA, June 23, 1994.

17. Memorandum from Whiteside, Additives Evaluation Branch #2, to Anderson, Direct Additives Branch, FDA, March 23, 1994.

18. Memorandum from Whiteside, Additives Evaluation Branch #2, to Anderson, Direct Additives Branch, FDA, August 20, 1993.

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\*23. Krasavage, et al., "Indocyanine Green Testing," *Proceedings Society for Experimental Biology of Medicine*, 119: 215, 1965.

\*24. Bonasch, H. and C. E. Cornelius, "Indocyanine Green: A Liver Function Test for the Dog," *American Journal of Veterinary Research*, 25: 254-59, 1964.

25. Memorandum from Alam, Pathology Branch, to Anderson, Novel Ingredients Branch, FDA, July 10, 1997.

26. Memorandum from Hotta, Clinical Nutrition Branch, to Blendermann, Division of Nutrition, FDA, December 31, 1986.

27. Memorandum from Calvert, Clinical Nutrition Branch, to Anderson, Novel Ingredients Branch, FDA, January 21, 1992.

28. Memorandum from Calvert, Clinical Nutrition Branch, to Anderson, Novel Ingredients Branch, FDA, September 2, 1993.

### VI. Objections

Any person who will be adversely affected by this regulation may at any time on or before July 6, 1999, file with the Dockets Management Branch (address above) written objections thereto. Each objection shall be separately numbered, and each numbered objection shall specify with particularity the provisions of the regulation to which objection is made and the grounds for the objection. Each numbered objection on which a hearing is requested shall specifically so state.

Failure to request a hearing for any particular objection shall constitute a waiver of the right to a hearing on that objection. Each numbered objection for which a hearing is requested shall include a detailed description and analysis of the specific factual information intended to be presented in support of the objection in the event that a hearing is held. Failure to include such a description and analysis for any particular objection shall constitute a waiver of the right to a hearing on the objection. Three copies of all documents shall be submitted and shall be identified with the docket number found in brackets in the heading of this document. Any objections received in response to the regulation may be seen in the Dockets Management Branch between 9 a.m. and 4 p.m., Monday through Friday.

#### List of Subjects in 21 CFR Part 172

Food additives, Incorporation by reference, Reporting and recordkeeping requirements.

Therefore, under the Federal Food, Drug, and Cosmetic Act and under authority delegated to the Commissioner of Food and Drugs, 21 CFR part 172 is amended as follows:

#### PART 172—FOOD ADDITIVES PERMITTED FOR DIRECT ADDITION TO FOOD FOR HUMAN CONSUMPTION

1. The authority citation for 21 CFR part 172 continues to read as follows:

**Authority:** 21 U.S.C. 321, 341, 342, 348, 371, 379e.

2. Section 172.833 is added to subpart I to read as follows:

##### § 172.833 Sucrose acetate isobutyrate (SAIB).

Sucrose acetate isobutyrate may be safely used in foods in accordance with the following prescribed conditions:

(a) Sucrose acetate isobutyrate (CAS Reg. No. 27216-37-1), or SAIB, is the chemical *alpha*-D-glucopyranoside, O-acetyl-tris-O-(2-methyl-1-oxopropyl)-*beta*-D-fructofuranosyl, acetate tris(2-methyl propanoate).

(b) SAIB, a pale, straw-colored liquid, meets the following specifications:

(1) Assay: Not less than 98.8 percent and not more than 101.9 percent, based on the following formula:

Assay =  $((SV \cdot 0.10586) \div 56.1) \times 100$   
Where SV = Saponification value

(2) Saponification value: 524–540 determined using 1 gram of sample by the "Guide to Specifications for General Notices, General Analytical Techniques, Identification Tests, Test Solutions, and Other Reference Materials," in the

"Compendium of Food Additive Specifications, Addendum 4, Food and Agriculture Organization of the United Nations (FAO), Food and Nutrition Paper 5, Revision 2" (1991), pp. 203 and 204, which is incorporated by reference, in accordance with 5 U.S.C. 552(a) and 1 CFR part 51. Copies are available from the Office of Premarket Approval, Center for Food Safety and Applied Nutrition (HFS-200), Food and Drug Administration, 200 C St. SW., Washington, DC 20204, or may be examined at the Center for Food Safety and Applied Nutrition's Library, 200 C St. SW., rm. 3321, Washington, DC, or at the Office of the Federal Register, 800 North Capitol St. NW., suite 700, Washington, DC.

(3) Acid value: Not to exceed 0.20 determined using 50 grams of sample by the "Guide to Specifications for General Notices, General Analytical Techniques, Identification Tests, Test Solutions, and Other Reference Materials," in the "Compendium of Food Additive Specifications, Addendum 4, FAO Food and Nutrition Paper 5, Revision 2," p. 189 (1991), which is incorporated by reference; see paragraph (b)(2) of this section for availability of the incorporation by reference.

(4) Lead: Not to exceed 1.0 milligrams/kilogram determined by the "Atomic Absorption Spectrophotometric Graphite Furnace Method, Method I," in the "Food Chemicals Codex," 4th ed. (1996), pp. 763 and 764, with an attached modification to the sample digestion section in Appendix III.B (July 1996), which is incorporated by reference. Copies are available from the National Academy Press, 2101 Constitution Ave. NW., Box 285, Washington, DC 20055 (Internet "http://www.nap.edu"), or may be examined at the Center for Food Safety and Applied Nutrition's Library, 200 C St. SW., rm. 3321, Washington, DC, or at the Office of the Federal Register, 800 North Capitol St. NW., suite 700, Washington, DC.

(5) Triacetin: Not to exceed 0.10 percent determined by gas chromatography as described in the "Guide to Specifications for General Notices, General Analytical Techniques, Identification Tests, Test Solutions, and Other Reference Materials," in the "Compendium of Food Additive Specifications, Addendum 4, FAO Food and Nutrition Paper 5, Revision 2" (1991), pp. 13–26, which is incorporated by reference; see paragraph (b)(2) of this section for availability of the incorporation by reference.

(c) The food additive is used as a stabilizer (as defined in § 170.3(o)(8) of

this chapter) of emulsions of flavoring oils in nonalcoholic beverages.

(d) The total SAIB content of a beverage containing the additive does not exceed 300 milligrams/kilogram of the finished beverage.

Dated: May 27, 1999.

**William K. Hubbard,**

*Associate Commissioner for Policy Coordination.*

[FR Doc. 99–14147 Filed 6–3–99; 8:45 am]

BILLING CODE 4160–01–F

## ENVIRONMENTAL PROTECTION AGENCY

### 40 CFR Part 52

[AL–40–2–9909a; FRL–6352–5]

### Approval and Promulgation of Implementation Plans; Alabama

**AGENCY:** Environmental Protection Agency (EPA).

**ACTION:** Direct final rule.

**SUMMARY:** The EPA is approving a State Implementation Plan (SIP) revision for the State of Alabama. This revision consists of the 1990 base year ozone emission inventory for the Birmingham marginal ozone nonattainment area. The inventory was submitted to satisfy a Clean Air Act (CAA) requirement that states containing ozone nonattainment areas submit inventories of actual ozone precursor emissions in accordance with guidance from the EPA.

**DATES:** This direct final rule is effective August 3, 1999 without further notice, unless EPA receives adverse comment by July 6, 1999. If adverse comment is received, EPA will publish a timely withdrawal of the direct final rule in the **Federal Register** and inform the public that the rule will not take effect.

**ADDRESSES:** All comments should be addressed to Joey LeVasseur at the EPA, Region 4 Air Planning Branch, 61 Forsyth Street, SW, Atlanta, Georgia 30303.

Copies of the state submittal are available at the following addresses for inspection during normal business hours: The interested persons wanting to examine these documents should make an appointment with the appropriate office at least 24 hours before the visiting day. Reference file AL–40–2–9909. The Region 4 office may have additional background documents not available at the other locations.

Environmental Protection Agency,  
Atlanta Federal Center, Region 4 Air  
Planning Branch, 61 Forsyth Street  
SW, Atlanta, Georgia 30303–3104.