# *In Vivo* Metabolism of SALATRIM Fats in the Rat

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SALATRIM fats are structured triacylglycerols that contain at least one short-chain fatty acid (SCFA) and a saturated long-chain fatty acid, usually stearic. SALATRIM $^{14}C$-labeled in either acetate, propionate, stearate, or glycerol and triolein $^{14}C$-labeled in either oleate or glycerol were administered to male rats by gavage. Rats were fed basal diet or basal diet containing 10% (w/w) SALATRIM 23CA lot A014 for 14 days before administration of radiolabeled fats. Acetate and propionate from SALATRIM are rapidly converted to carbon dioxide, which indicates the triacylglycerols are cleaved by lipases in the stomach and upper small intestine. The stearate of SALATRIM is hydrolyzed more slowly than the SCFA but almost identically to the oleate of triolein. Long-chain fatty acids of SALATRIM and triolein are predominantly metabolized to $CO_2$. Stearic acid is more poorly absorbed than oleic acid. Of the absorbed stearate in fat stores, 57.2% had been converted to oleate. The biodisposition of glycerol from SALATRIM or triolein is essentially identical. The study supports the hypothesis that SALATRIM biodisposition is predictable and similar to that of other fats. Feeding SALATRIM 23CA lot A014 for 14 days did not significantly alter SALATRIM or triolein biodisposition.

## INTRODUCTION

SALATRIM fats are a family of triacylglycerols with lower caloric availability (4.5–6 kcal/g) than fats such as corn oil (9 kcal/g) (Finley et al., 1994). Although their basic chemical structure is similar to that of other fats, SALATRIM fats differ from other fats with respect to their content of short-chain fatty acids (SCFA). SALATRIM fats are produced by interesterification among triacetin, tripropionin, tributyrin, and hydrogenated vegetable fats containing high quantities of stearic acid. The ratio of SCFA to long-chain fatty acids (LCFA) is higher in SALATRIM fats than in other fats containing SCFA, such as milk fat. Varying the specific SCFA and the SCFA/LCFA ratio produces SALATRIM fats with various physical characteristics and functionalities.

Because of their similarity to other fats, SALATRIM fats should be digested, absorbed, metabolized, and excreted in a highly predictable manner. Fatty acids at the 1- and 3-positions of the glycerol of SALATRIM should be hydrolyzed by lipases to yield free fatty acids and the 2-monoacylglycerol. The free LCFA can become associated with bile salt micelles and be absorbed by the intestinal mucosa along with the 2-monoacylglycerols. Longer chain free fatty acids, especially stearic acid, are more poorly absorbed and are excreted in the feces as free fatty acids and calcium and magnesium salts (Abe et al., 1993; Mattson, 1979). Therefore, the stearic acid from SALATRIM fats should be poorly absorbed. The SCFA of SALATRIM fats should be rapidly absorbed in their protonated form. After absorption, the components of SALATRIM fats should be metabolized in a highly predictable manner. The majority of the SCFA should be oxidized to $CO_2$, yielding energy. A smaller proportion of the SCFA should be incorporated into anabolic metabolism to be used as precursors of more complex biomolecules. The LCFA should, in part, be metabolized to $CO_2$ and, in part, enter into anabolic metabolism by well-known pathways. A portion of the LCFA may become part of the stored fat.

This study was designed to test the hypothesis that SALATRIM fats will be digested, absorbed, metabolized, and excreted as predicted from the literature on fat metabolism. SALATRIM fats radiolabeled ($^{14}C$) in either the SCFA, LCFA, or glycerol were administered to rats, and the fate of radiolabel was monitored over a 72-h time course. Triolein radiolabeled in either the LCFA or glycerol was similarly administered to rats to serve as a reference fat for comparison to SALATRIM. An additional hypothesis, that prior consumption of SALATRIM fats should not alter the metabolism of SALATRIM or other fats, was also tested. To test this hypothesis, rats were prefed either a diet containing 10% (w/w) SALATRIM 23CA lot A014 or a basal diet for 14 days prior to administration of radiolabeled fat and the metabolism of radiolabeled SALATRIM and radiolabeled triolein was compared between the two dietary groups.

## MATERIALS AND METHODS

**Radiolabeled SALATRIM Fats.** Radiolabeled SALATRIM fats that mimic SALATRIM 23CA lot A014 were synthesized and purified by Sigma Chemical Co., St. Louis, MO. Synthesis was by random interesterification of triacetin, tripropionin, and tristearin at a ratio of 2.5:2.5:1.0, respectively. The resulting radiolabeled fats were designated SALATRIM APS. Upon receipt, the radiolabeled SALATRIM fats were stored at 4 °C until used. The radiolabeled SALATRIM fats were assayed by thin-layer chromatography (TLC) to confirm purity before use. The labeled SALATRIM fats and their radiopurity were (1) [$^{14}C$]-SALATRIM $APS_A$ ($^{14}C$ in the acetate carbonyl) (radiopurity = 95%),; (2) [$^{14}C$]SALATRIM $APS_P$ ($^{14}C$ in the propionate carbonyl) (radiopurity = 96.5%), (3) [$^{14}C$]SALATRIM $APS_S$ ($^{14}C$ in the stearate carbonyl) (radiopurity = 94.6%), and (4) [$^{14}C$]-SALATRIM $APS_G$ ($^{14}C$ in the 1- and 3-positions of glycerol) (radiopurity ≥ 98%). TLC suggested that the impurities were mono-, di-, and triacylglycerols and, in some cases, the labeled free fatty acid.

**Radiolabeled Reference Fat.** Radiolabeled [$^{14}C$]triolein was used as a reference fat to allow a comparison between its metabolism and that of SALATRIM. Triolein ($^{14}C$ in the oleic carbonyl) having a radiopurity of 95% was obtained from DuPont/

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Table 1. Total Fatty Acid Profile for SALATRIM 23CA Lot A014[a,b]

| fatty acid name | fatty acid designation | wt % |
|---|---|---|
| stearic (octadecanoic) | C18:0 | 57 ± 1 |
| acetic | C2:0 | 21.1 ± 0.1 |
| propionic | C3:0 | 2.58 ± 0.03 |
| palmitic (hexadecanoic) | C16:0 | 2.37 ± 0.04 |
| arachidic (eicosanoic) | C20:0 | 1.50 ± 0.03 |
| behenic (docosanoic) | C22:0 | 0.668 ± 0.007 |
| oleic (9-octadecenoic) | C18:1 | 0.572 ± 0.005 |
| lignoceric (tetracosanoic) | C24:0 | 0.335 ± 0.001 |
| linoleic (9,12-octadecadienoic) | C18:2 | 0.066 ± 0.001 |
| lauric (dodecanoic) | C12:0 | 0.007 ± 0.000 |

[a] Data represent the mean ± standard deviation for triplicate determinations. [b] Data developed at EPL-BAS, Decatur, IL.

NEN (Boston, MA). Triolein ($^{14}C$ in the 1- and 3-positions of glycerol) with a radiopurity of 94.8% was obtained from Sigma. The radiolabeled triolein was stored at 4 °C until used.

**Nonradiolabeled SALATRIM.** Nonradiolabeled SALATRIM 23CA Lot A014 was used in this study to dilute the radiolabeled SALATRIM and triolein to the appropriate specific activity. It was also used to prepare diets intended to assess the effect of SALATRIM prefeeding on the metabolism of SALATRIM and triolein. This SALATRIM was provided by the Nabisco Foods Group, East Hanover, NJ. SALATRIM 23CA lot A014 was stored at −20 °C until used.

**Chemical Characterization of Nonradiolabeled SALATRIM.** SALATRIM 23CA lot A014 contains acetic acid and propionic acid as the SCFA and stearic acid as the LCFA. Total fatty acids profile analysis of SALATRIM 23CA lot A014 was conducted at EPL Bio-Analytical Services, Inc. (EPL-BAS), Decatur, IL; the results are presented in Table 1. Total fatty acids profile data for >C12:0 and higher were obtained by saponification of the triacylglycerol mixture with methanolic sodium hydroxide followed by esterification with methanolic boron trifluoride. Methyl esters of the fatty acids were quantitated by gas chromatography. The SCFA profiles were obtained by saponification of the sample with sodium hydroxide followed by acidification with concentrated hydrochloric acid. The fatty acids were directly quantitated by gas chromatography. Standard curves were constructed by bracketing the concentration level of the analyte. Internal standards were methyl pentadecanoate for the LCFA and butyric acid for the SCFA.

**Animals and Housing.** Male Sprague-Dawley rats were obtained from Harlan Sprague-Dawley, Inc. (Madison, WI). At arrival, the rats were 3–6 weeks old and were 6–10 weeks of age at dosing. The rats were acclimated to the laboratory for a minimum of 1 week before study initiation. Rats were maintained in compliance with the standards outlined in the *Guide for the Care and Use of Laboratory Animals* (NIH Publication 86-23, 1985). They were individually housed in stainless steel, suspended, wire-bottom cages until dosing. After dosing with radiolabeled fat, they were individually housed in glass metabolism cages designed for collection of expired $CO_2$, urine, and feces. Throughout the study, the animal rooms were set to maintain a temperature of 22 ± 3 °C, a relative humidity of 50% ± 20%, and a 12-h light/dark cycle. Rats were examined daily during acclimation for clinical abnormalities and twice daily during the study periods for moribundity and mortality.

**Diets.** During acclimation, all rats received a basal diet of powdered Purina Rat and Mouse Ration 5018 from Purina Mills, Inc. (St. Louis, MO), which is equivalent to the NIH-07 rodent diet (Rao and Knapka, 1987). After the acclimation period, rats were assigned randomly by body weight to feeding and treatment groups. Prefed rats were fed the basal diet containing 10% (w/w) SALATRIM 23CA lot A014 for 14 days, and nonprefed rats were maintained on the basal diet for 14 days before dosing with radiolabeled fat. All rats were fasted for approximately 4 h before and after dosing with radiolabeled fat. Except for this fasting period, the appropriate feed was provided *ad libitum*. Water was available *ad libitum* throughout the study. Feed consumption was recorded weekly during the 14-day predose period and after the 72-h postdosing period.

**Diet Preparation and Analysis.** The diet containing 10% (w/w) SALATRIM 23CA lot A014 was prepared as previously described (Hayes et al., 1994). The homogeneity of the SALATRIM fat in the diet was confirmed by gravimetric analysis. The percent dietary SALATRIM was confirmed by supercritical fluid chromatography. These analyses indicated that the diet mix contained 9.59% SALATRIM 23CA lot A014 that was homogeneously distributed in the diet.

**Preparation, Analysis, and Administration of Radiolabeled Fat Doses.** SALATRIM 23CA lot A014 was weighed into glass vials and the appropriate quantity of radiolabeled SALATRIM APS or radiolabeled triolein added. A magnetic stirring bar was added and the fat heated to 35–40 °C and stirred for 30 min. All dosing solutions were stored at 4 °C until used. Each radiolabeled SALATRIM APS and triolein dose was analyzed in triplicate by a liquid scintillation spectrometer (Packard Instrument Co., Palo Alto, CA) to determine the quantity of radioactivity and specific activity. Scintillation counting was automatically corrected for counting efficiency by external standardization using a quench curve generated from sealed quenched standards.

Rats were administered a target dose of 1.4 g of fat/kg of body weight of either radiolabeled SALATRIM APS or radiolabeled triolein as a single oral dose by gavage. The dose administered was based upon the weight of the rat on the day of dosing. The individual dose administered to each rat was determined by weighing the gavage syringe before and after dosing. The mean dose administered was 1.37 ± 0.07 g/kg with a coefficient of variation of 8.2% (specific activity 134 μCi/g). To determine if microcrystallization took place in the dosing solutions, which would result in nonhomogeneity of the doses, predose and postdose samples were compared with respect to specific activity. The data indicated close agreement between pre- and postdose specific activity.

Radiolabeled triolein was diluted with unlabeled SALATRIM 23CA lot A014 instead of unlabeled triolein to yield a fat matrix equivalent to the matrix containing radiolabeled SALATRIM. Since the physical properties of triolein and SALATRIM 23CA differ, the initial physical states of a pure triolein matrix in the stomach may differ significantly from that of SALATRIM. The physical characteristics of the fat, e.g., droplet size, hydrophobicity, and other factors, may influence lipase activity. This could result in differences in *in vivo* metabolism based upon physical characteristics. Matrix effects were minimized by placing the reference fat in a matrix identical to that of the fat under investigation.

**Study Design.** To determine if consumption of SALATRIM fats alters the metabolism of SALATRIM and other fats, rats were fed either the basal diet or basal diet containing 10% (w/w) SALATRIM 23CA lot A014 for 14 days prior to administration of the radiolabeled fats. Each group of five rats was administered one of the following radiolabeled fats by gavage: SALATRIM $APS_A$; SALATRIM $APS_P$; SALATRIM $APS_S$; SALATRIM $APS_G$; triolein radiolabeled in the oleate carbonyl; and triolein radiolabeled in the glycerol backbone. Expired $CO_2$ was collected in 2-ethoxyethanol/ethanolamine solutions for the following periods after radiolabeled fat administration: 0–3, 3–6, 6–12, 12–24, 24–48, and 48–72 h. Urine and feces were collected separately at the same time periods as $CO_2$. At termination of the sample collection period, the rats were removed from the metabolism cages and each cage was washed and wiped to determine residual radioactivity. Radioactivity remaining in the cages was added to that from the other samples to obtain total radiolabel recovery. In an additional experiment using SALATRIM $APS_A$, $CO_2$ was collected during the following periods: 0–15, 15–30, and 30–60 min and 1–1.5, 1.5–2, 2–3, 3–4, 4–5, 5–6, 6–8, 8–10, 10–12, 12–18, and 18-24 h postadministration. In this experiment, the rats were not prefed a SALATRIM-containing diet and feces, urine, and tissues were not collected.

**Tissue Collection.** At the termination of the 72-h sample collection period, the rats were anesthetized with halothane and sacrificed. The following tissues were collected: blood, serum, liver, and epididymal fat. The carcass was frozen with solid $CO_2$ and retained at freezer temperature for determination of total carcass radioactivity. Radiolabel in all tissues, urine, feces, and $CO_2$ was determined on duplicate samples.

**Radiolabel Determination Methods.** Radiolabeled $CO_2$ was determined by weighing the 2-ethoxyethanol/ethanolamine trapping solutions, removing approximately 1-g samples, and adding them to 5 mL of methanol and 10 mL of Perma-Fluor V scintillation cocktail (Packard Instrument Co., Palo Alto, CA). Radiolabel in feces was determined by homogenization of samples collected during each time period in 2 volumes (w/v) of water. Approximately 0.2 g of the homogenized sample was combusted in a Packard Model 306 or Model 307 sample oxidizer (Packard Instrument Co.) and the liberated $^{14}CO_2$ trapped and counted in Perma-Fluor V scintillation cocktail. Urinary radiolabel was determined in duplicate by weighing the total urine sample from each time period. Approximately 0.2 g of urine was counted in 5 mL of Ultima-Gold scintillation cocktail (Packard Instrument Co.).

Samples of approximately 0.25 g of blood were combusted in the sample oxidizer, and $^{14}CO_2$ was trapped and counted in Perma-Fluor V scintillation cocktail. Radiolabel in liver tissue was determined by homogenization of liver samples (0.2 g) followed by combustion of the homogenate in the sample oxidizer. The $^{14}CO_2$ was trapped and counted in Perma-Fluor V scintillation cocktail. Fat was homogenized in a Virtis homogenizer (Virtis Co., Gardiner, NY) followed by solubilization and counting of 0.2 g of the homogenate in Ultima-Gold scintillation cocktail. Total carcass radioactivity was determined from carcasses frozen with solid $CO_2$ followed by freezing at liquid nitrogen temperature. The frozen carcass was ground in a Wiley mill (Thomas Wiley Scientific Products) precooled with solid $CO_2$. Samples of approximately 0.5 g were combusted in the sample oxidizer, and $^{14}CO_2$ was trapped and counted in Perma-Fluor V scintillation cocktail.

**Determination of the Conversion of Stearic Acid to Oleic Acid.** Epididymal fat samples (1 g) from rats dosed with SALATRIM $APS_S$ were saponified by refluxing in 20 mL of 3 M potassium hydroxide for 12 h. The fatty acids were extracted by acidifying the cooled reflux mixture to pH 2 with 3 M hydrochloric acid followed by four extractions with 20 mL of ethyl ether/hexane (1:1). The extracts were pooled, the volume was reduced under a vacuum, and the samples were lyophilized. The samples were purified by column chromatography. The chromatography column consisted of 10 g of aminopropyl packing (Alltech, Deerfield, IL, 30–70 μm) conditioned with 25 mL of hexane. The extracted fatty acids were dissolved in 2 mL of chloroform and placed on the column. The column was rinsed with 25 mL of 2-propanol/chloroform (1:2 v/v), and the fatty acids were eluted with 75 mL of 2% acetic acid in ethyl ether. The ether fraction was reduced in volume under a vacuum and lyophilized. The samples were derivatized with 5 mL each of a 20 mg/mL solution of 2-bromoacetophenone in acetone and a 20 mg/mL solution of triethylamine in acetone. The samples were placed in tightly sealed vials and placed in a boiling water bath for 1 h. After cooling, 8 mL of acetic acid solution in acetone (4 mg/mL) was added and the solvents were evaporated under nitrogen. The resulting residue was resuspended in 0.5 mL of methanol and subjected to high-performance liquid chromatographic (HPLC) analysis on a Hewlett-Packard HP 1050 liquid chromatograph equipped with a Waters Model U6K injector, a Waters Model 486 UV detector, and a Radiomatic A-280 flow-through scintillation detector. The HPLC column was a Nucleosil $C_{18}$ 5 μm (250 × 4.6 mm) using a Nucleosil 100 $C_{18}$ 5 μm (10 × 4.6 mm) guard column. An initial 40% acetonitrile, 40% methanol, 20% water gradient moving to 100% methanol over 100 min at a flow rate of 1.0 mL/min was used to separate stearate and oleate.

**Fecal Stearate Analysis.** Fecal samples from rats fed SALATRIM $APS_S$ were homogenized in 2 volumes (w/v) of water, and approximately 1 mL of 1 M NaOH was added to 0.5 g of the homogenate. One microcurie of $^{14}C$-radiolabeled myristic acid was added to the sample as an internal standard. The homogenate was extracted three times with 3 mL of hexane. The homogenate was vortexed and centrifuged at 2000 rpm for 10 min. The supernatant was acidified with 2 mL of 1 M HCl and extracted three times with 3 mL of ethyl ether. The extracts were pooled and evaporated to dryness under nitrogen. Methyl ester derivatives of the free fatty acids were formed by adding 1 mL of boron trifluoride/methanol (14% w/v) to the sample and heating at 60 °C for 15 min. Two milliliters of a saturated NaCl solution was then added to the sample. The sample was extracted three times with 2 mL of hexane, and the extracts were combined. Hexane was removed by evaporation under a stream of nitrogen. The sample was resuspended in 2 mL of 2-propanol/acetonitrile (1:3 v/v) for HPLC analysis. HPLC conditions were similar to those for analysis of stearate and oleate in fat except an isocratic mobile phase consisting of 2-propanol/acetonitrile (1:3) was used.

**Table 2. Total Percent of Radiolabeled Dose Recovered**

| treatment group | total % of dose[a] |
|---|---|
| SALATRIM $APS_A$ | |
| non-SALATRIM prefed | 94.4 |
| SALATRIM prefed | 93.0 |
| SALATRIM $APS_S$ | |
| non-SALATRIM prefed | 89.2 |
| SALATRIM prefed | 91.5 |
| SALATRIM $APS_G$ | |
| non-SALATRIM prefed | 94.6 |
| SALATRIM prefed | 94.5 |
| SALATRIM $APS_P$ | |
| non-SALATRIM prefed | 97.0 |
| SALATRIM prefed | 96.7 |
| triolein (oleate carbonyl labeled) | |
| non-SALATRIM prefed | 95.0 |
| SALATRIM prefed | 96.5 |
| triolein (glycerol labeled) | |
| non-SALATRIM prefed | 96.8 |
| SALATRIM prefed | 94.7 |

[a] Data represent the total percent of the dose recovered calculated by the addition of radioactivity recovered from the blood, carcass, fat, liver, $CO_2$, feces, urine, cage wipes, cage wash, and $CO_2$ backup.

**Statistical Analysis.** Selected data were subjected to statistical analysis by a two-tailed, unpaired $t$-test. Variances between groups were compared with the two-tailed $F$-test. If the variances between groups differed significantly ($p \leq 0.05$), then a Wilcoxon signed rank test was conducted. Differences were considered to be statistically significant at $p \leq 0.05$. Statistical analysis was conducted with StatView (Abacus Concepts, Inc., Berkeley, CA) statistical analysis program.

## RESULTS AND DISCUSSION

**Chemical Characterization of SALATRIM 23CA Lot A014.** Chemical characterization data for SALATRIM 23CA lot A014 are presented in Table 1. The total fatty acid profile after saponification of this fat indicates that the predominant fatty acids are stearic acid (57 ± 1 wt %), acetic acid (21.1 ± 0.1 wt %), propionic acid (2.58 ± 0.03 wt %), and palmitic acid (2.37 ± 0.04 wt %).

**Total Percent Radioactivity Recovered.** To determine the total percent radioactivity recovered in each treatment group, the radioactivity from $CO_2$, feces, urine, blood, fat, liver, carcass, cage wipes, cage wash, and $CO_2$ backup was totaled. These data are summarized in Table 2. Recovery of radioactivity ranged from 89.2% to 97.0%, with the mean recovery being 94.5%.

**Feed Consumption and Body Weights.** There were no compound-related differences in feed consumption during the 2-week feeding period prior to administration or after administration of the radiolabeled fats (data not shown).

Body weights of the rats ranged from 216 to 303 g at the time of administration of the radiolabeled fats (data not shown). There were no statistically significant differences in body weights of the rats fed the 10% (w/w) SALATRIM 23CA lot A014 diet compared with the rats administered the basal diet with the exception of the SALATRIM $APS_G$ group. The body weights of the rats fed the 10% (w/w) SALATRIM 23CA lot A014 diet and administered SALATRIM $APS_G$ were slightly, but statistically, less than those fed the basal diet. The reason for the difference in this pair of treatment groups was not apparent but does

not appear to be related to SALATRIM consumption because it was only seen in this one group.

**Metabolism of SALATRIM and Triolein to $CO_2$.** SALATRIM fats, like other fats, will enter the stomach in the form of small fat droplets. Depending upon the dietary source of the SALATRIM fat, these droplets may be either composed predominantly of SALATRIM or consist of mixed-fat droplets with fats from other sources. Fat droplets of the appropriate size will interact with lingual lipase and gastric lipase to initiate gastric triacylglycerol hydrolysis (Carey et al., 1983; Gargouri et al., 1986).

Lingual lipase shows preference for triacylglycerols with short- and medium-chain triacylglycerols (Staggers et al., 1981). Fatty acids at the 1- and 3-positions of glycerol are preferentially cleaved compared to that at the 2-position. The fatty acid at the 3-position is preferentially cleaved compared with the fatty acid at the 1-position (Paltauf et al., 1974). Gastric lipase appears to hydrolyze SCFA at a faster rate than LCFA (Gargouri et al., 1986). The SCFA released by lipases in the stomach will enter the bulk aqueous medium. Because of the low pH of the stomach contents, the SCFA will be protonated and rapidly absorbed by the stomach mucosa (Rombeau and Kripke, 1990). Most LCFA released by lipolysis in the stomach will generally partition back into the fat droplets (Friberg et al., 1971; Carey et al., 1983). Once the SCFA enter the mucosa, either they are used as substrates for energy production within the mucosa with concomitant release of $CO_2$ or they enter the portal circulation to be transported to the liver where they can serve as an energy source (Aw and Grigor, 1980; Bugaut, 1987).

The above indicates that in the stomach the SCFA components of SALATRIM would be more rapidly cleaved from the glycerol backbone than the LCFA. The $^{14}C$ in the SCFA should rapidly appear as $^{14}CO_2$ in the expired breath. Little or no stearic acid should be absorbed by the gastric mucosa. This is borne out by the data in this study. Figure 1 illustrates the percent of dose of SALATRIM containing either labeled acetate or labeled propionate that is excreted as $^{14}CO_2$ in the expired breath. Maximal metabolism of both SCFA to $CO_2$ had occurred by the first time point (3 h after administration). In contrast, only a small portion of the stearate derived from SALATRIM appeared to be absorbed and oxidized to $CO_2$ at 3 h postadministration. More oleate from triolein was absorbed and metabolized to $CO_2$ than stearate from SALATRIM, supporting the predicted poor absorption of stearate compared to that of oleate. Maximal oleate conversion to $CO_2$ occurred at 6 h postadministration. These data indicate that SCFA are more rapidly hydrolyzed from the fat, absorbed, and metabolized to $CO_2$ than are the LCFA, as predicted.

Absorption of the glycerol backbone of either SALATRIM or triolein can take place by at least two mechanisms. The fatty acids could be completely hydrolyzed from the fat, resulting in free glycerol and its subsequent absorption, or glycerol can be absorbed as the 2-monoacylglycerol. This latter mechanism is generally accepted as the predominant mechanism for glycerol absorption. Once absorbed, the 2-monoacylglycerol is hydrolyzed by cellular lipases to free glycerol and a fatty acid. The liberated glycerol can either be reesterified to form a triacylglycerol or enter cellular anabolic and/or catabolic metabolism. Glycerol that enters these pathways can ultimately be metabolized to $CO_2$.

The metabolism of SALATRIM and triolein radiolabeled in the glycerol backbone to expired $^{14}CO_2$ is illustrated in Figure 1. Between 3 and 12 h postadministration, the metabolism of fat-derived glycerol to $CO_2$ appears to be relatively constant. From 12 to 24 h postadministration, there is a gradual decline in glycerol-derived $CO_2$, followed by a low rate of $^{14}CO_2$ production between 24 and 72 h. A comparison of the metabolism of SALATRIM-derived glycerol and triolein-derived glycerol to $CO_2$ indicates little or no qualitative or quantitative difference between the two fats.

These data do not address the question of whether the metabolism and absorption took place in the stomach or small intestine at the 3-h time point because transit times were not determined. Varga (1976) has presented data that indicate the gastric half-time for rats of this age may be between 0.50 and 0.60 h. However, gastric emptying may be modified by a number of factors (Brown et al., 1987). Data obtained at 3 h postadministration of the radiolabeled fats can be assumed to have resulted from events that took place in the stomach and upper small intestine.

To determine the absorption of SCFA from SALATRIM fats at time points earlier than 3 h postadministration, SALATRIM $APS_A$ was administered to rats in a manner identical to that in the earlier study. Expired $^{14}CO_2$ was collected at time intervals from 15 min to 24 h. The data from this study are presented in Figure 2. As early as 15 min postadministration, $^{14}CO_2$ derived from acetate-labeled SALATRIM was detected in expired $CO_2$, indicating the rapid hydrolysis, absorption, and metabolism of the SCFA in the stomach. There was a rapid rise in radiolabel between 15 min and 1 h. Labeled $CO_2$ expiration was relatively constant between 1 and 3 h postadministration. By 1 h postadministration, gastric emptying would have been initiated with the fat beginning to enter the upper quarter of the small intestine. By 3 h postadministration, most of the fat would have entered the small intestine. This suggests that hydrolysis and absorption of the SCFA begins in the stomach and continues in the upper region of the small intestine. The study design does not allow a differentiation between the percent absorbed in the stomach and that in the small intestine.

In the first quarter of the small intestine, lipolysis will be predominantly by pancreatic lipase and generally proceeds at a faster rate than gastric hydrolysis. Pancreatic lipase, like lingual and gastric lipase, hydrolyzes SCFA more quickly than it does LCFA. It shows positional specificity toward ester bonds at the 1- and 3-positions of glycerol. This suggests the SCFA of SALATRIM fats will be more rapidly cleaved than the stearate portion and the oleic acids of triolein. Indeed, this is evident in the data. The SCFA of SALATRIM are rapidly hydrolyzed, absorbed, and metabolized to $CO_2$ during the first 1–3 h postadministration followed by a rapid decline in $^{14}CO_2$ expiration between 3 and 12 h. This rapid decline in $^{14}CO_2$ probably represents exhaustion of the SCFA available for metabolism.

In contrast to the rapid hydrolysis, absorption, and metabolism of SCFA esterified to glycerol in SALATRIM fats, the hydrolysis of the LCFA occurs at a slower rate. For instance, as shown in Figure 1, $^{14}CO_2$ production from triolein (oleic carbonyl labeled) is maximal at 6 h postadministration, when $^{14}CO_2$ production from SALATRIM-derived acetate and propionate is rapidly declining. Carbon dioxide production from SALATRIM-derived stearate is also maximal at 6 h, although the $^{14}CO_2$ production from stearate is much less than that from oleate because of the poor absorption of stearate by the intestinal mucosa.

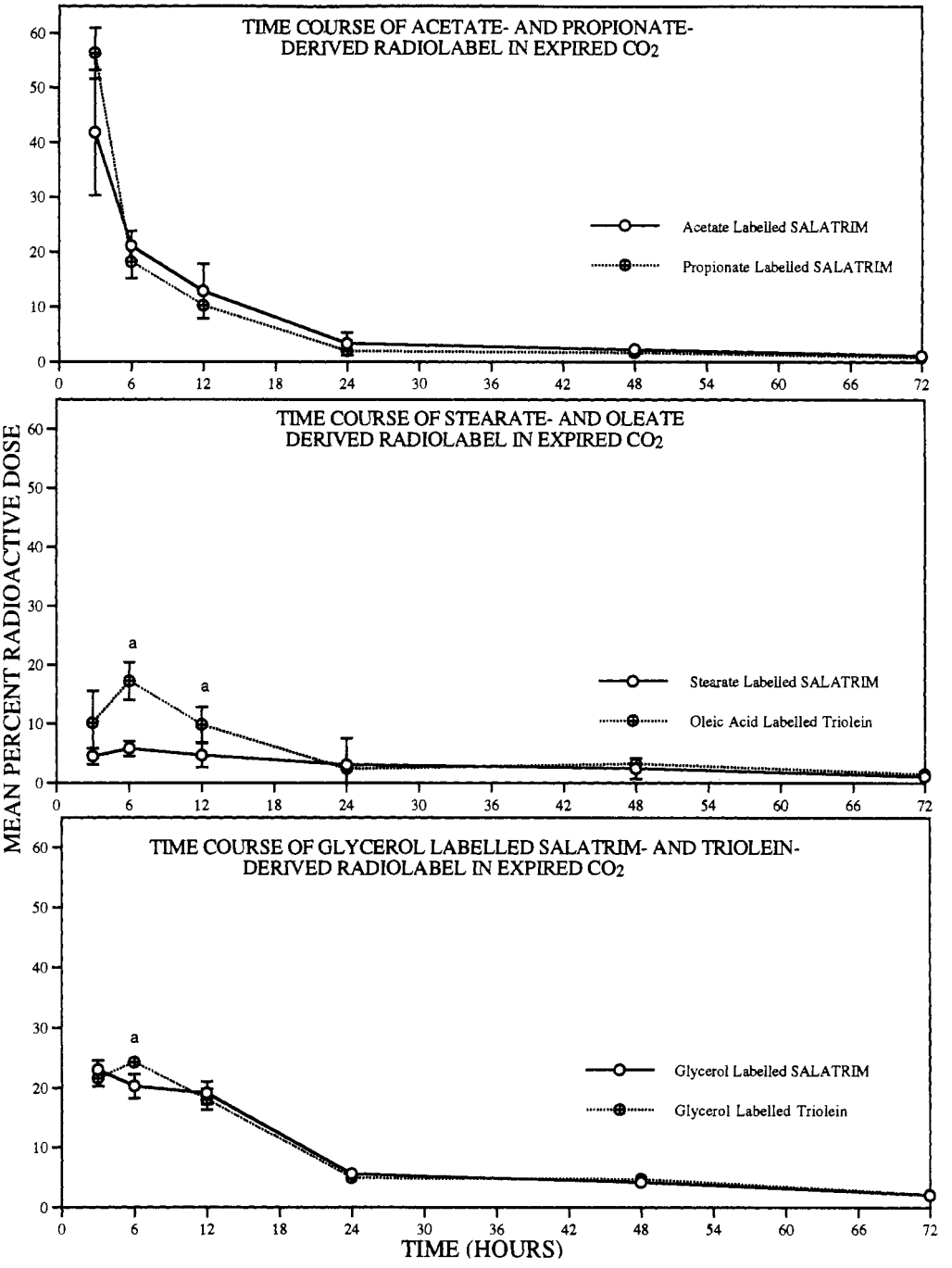


**Figure 1.** Metabolism of SALATRIM and triolein to $CO_2$ by male rats over a 72-h time course. The SALATRIM and triolein were administered as a single oral gavage. Data represent the mean ± standard deviation for five rats at each time point. Data significantly different at each time point are marked with an "a".

Further indication that most of the absorption and metabolism of the LCFA occurs in the upper small intestine is evident in the time course of $^{14}CO_2$ production from the fats with radiolabeled glycerol. There are no reports of free glycerol from fat lipolysis occurring in the stomach. Also, the di- and monoacylglycerols produced by lingual and gastric lipase activity in the stomach are generally not absorbed and partition back into the fat droplet where they aid emulsification of the droplet. Therefore, the $CO_2$ produced from glycerol most likely results from monoacylglycerol absorption in the small intestine. This indicates that the fats have reached the small intestine by 3 h postingestion.

Figure 1 indicates that by 24 h postadministration, $^{14}CO_2$ production from SALATRIM-derived acetate and propionate had reached a low level and continued to decrease slowly up to 72 h postadministration. This indicates the vast majority of the SCFA esterified to the glycerol of SALATRIM fats have been hydrolyzed, absorbed, and metabolized by 24 h postadministration. A portion of the $^{14}CO_2$ being expired during the latter hours of the study probably is generated by catabolism of substrates produced by anabolic metabolism of radiolabeled acetate and propionate. After 24 h postadministration, expiration of $^{14}CO_2$ from stearate derived from SALATRIM and oleate derived from triolein exhibits trends similar to the SCFA, indicating that hydrolysis, absorption, and metabolism of the LCFA to $CO_2$ have essentially ceased. By that time, the unmetabolized fats should be predominantly in the cecum and large intestine, where metabolism and absorption of LCFA are minimal (Varga, 1976).

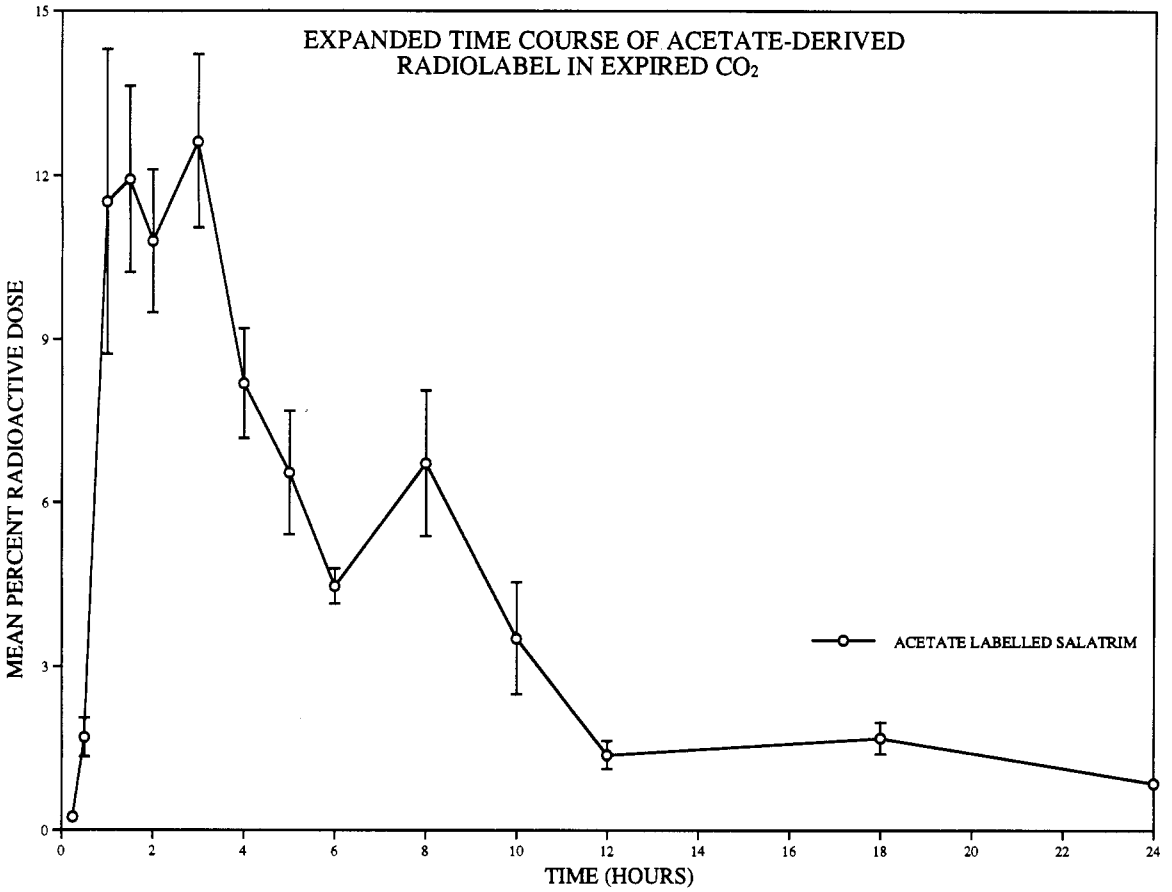


**Figure 2.** Metabolism of SALATRIM $APS_A$ to $CO_2$ over a 24-h time course. The SALATRIM was administered as a single oral gavage. Data represent the mean ± standard deviation for five male rats at each time point. Data significantly different at each time point are marked with an "a".

**Comparison of $CO_2$ Production from Rats Prefed a SALATRIM 23CA-Containing Diet.** The quantity and trends of $^{14}CO_2$ production between rats fed basal diet and rats fed basal diet containing 10% (w/w) SALATRIM 23CA can be seen by comparing Figure 1 with Figure 3. Production of $^{14}CO_2$ from SALATRIM-derived SCFA did not differ between the two dietary groups. This was also true for a comparison between $^{14}CO_2$ production from stearate derived from SALATRIM and oleate derived from triolein (Figures 1 and 3). With respect to the metabolism of glycerol derived from SALATRIM and triolein to $^{14}CO_2$, little or no effect of prefeeding either the basal diet or basal diet containing 10% (w/w) SALATRIM 23CA was evident. This indicates that feeding SALATRIM 23CA to rats affects neither its own metabolism nor the metabolism of other fats to $CO_2$. Additional support for this conclusion is provided by the lack of alteration of intestinal microflora in rats consuming 10% dietary SALATRIM for at least 90 days (Scheinbach et al., 1994).

**Excretion of SALATRIM- and Triolein-Derived Radiolabel in Urine.** Urinary radiolabel detected after administration of radiolabeled fats can have a number of sources. For instance, the $CO_2$ produced from the catabolic metabolism of either the fatty acids or glycerol from the fat will be excreted in the expired breath and a much smaller fraction as bicarbonate in the urine. Also, a portion of the radiolabeled carbon in the fats will be assimilated into normal cellular anabolic pathways and be distributed into biological molecules. As these molecules undergo normal catabolic reactions, a portion of this radiolabeled carbon will be excreted in the urine as a component of these reaction products. Because the components of the SALATRIM fats are normal dietary constituents and normal constituents of the body, no attempt was made to determine the chemical structure associated with fat-derived $^{14}C$ in these studies.

As shown in Figure 4, radiolabeled carbon could be detected in the urine by 3 h postadministration of SALATRIM labeled in the carbonyl of acetate or propionate. In contrast to expiration of $^{14}CO_2$, the quantity of label detected was low. After 3 h postadministration, there was a rapid rise in radiolabel in the urine until the maximal level was reached at 12 h postadministration. After the 12-h peak, urinary radiolabel dropped rapidly until 24 h postadministration and then declined gradually. The rapid rise in radiolabel between 3 and 12 h occurred during the period of rapid decline of expired radiolabeled $CO_2$. This indicates that much of the radiolabel detected in the urine after 3 h was not associated with bicarbonate but associated with the products of normal catabolic metabolism. The decrease in radiolabeled carbon in the urine after 12 h is indicative that the concentration of SCFA available as a metabolic substrate had decreased. Although there was a trend toward slightly more radiolabel derived from propionate than acetate between 6 and 12 h postadministration, after 12 h, there was no difference in the radiolabel derived from these SCFA.

The urinary radiolabel derived from the stearate of SALATRIM and the oleate from triolein followed trends similar to those of the SCFA. Radiolabel could be detected as early as 3 h postadministration with maximal urinary radioactivity detected at 12 h, after which time urinary radiolabel slowly declined. Quantitatively, the concentration of radiolabel in the urine from the LCFA was lower than that produced by the SCFA. This would be expected

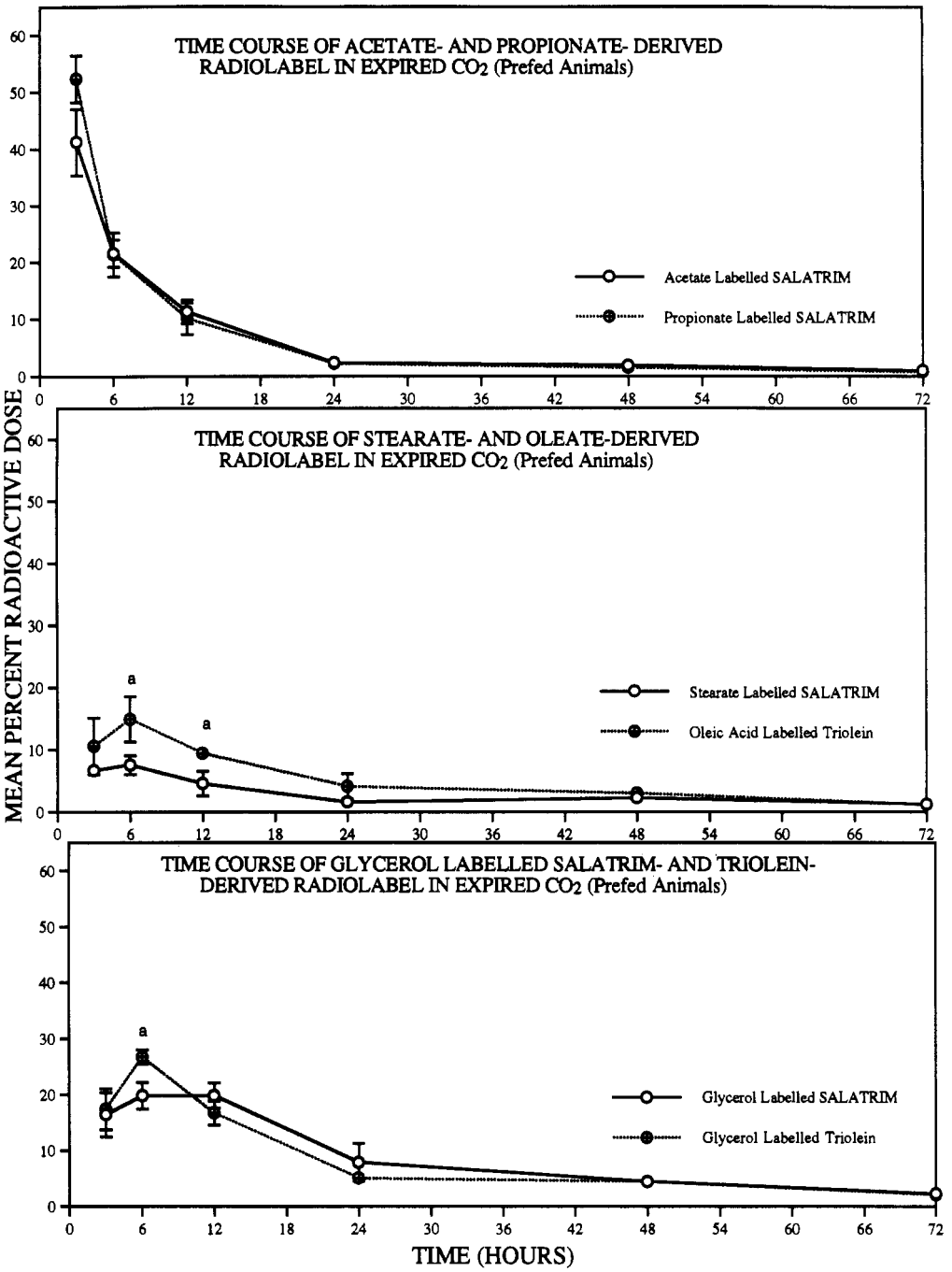


**Figure 3.** Metabolism of SALATRIM and triolein to $CO_2$ by male rats fed the basal diet containing 10% (w/w) SALATRIM 23CA lot A014 for 14 days. The SALATRIM and triolein were administered as a single oral gavage. Data represent the mean ± standard deviation for five male rats at each time point. Data significantly different at each time point are marked with an "a".

because of the more rapid hydrolysis of the SCFA and their more complete metabolism compared to that of the LCFA. Urinary radiolabeled carbon derived from the oleate of triolein tended to be higher than that derived from the stearate of SALATRIM during the period of rapid hydrolysis and absorption of LCFA. This would be predicted because of the poorer absorption of free stearate compared to that of free oleate. The urinary radiolabeled carbon derived from stearate may represent stearate that was absorbed as the 2-monoacylglycerol.

As would be expected, the time course of urinary labeled carbon derived from the glycerol of SALATRIM and triolein was identical to that followed by the LCFA. Most LCFA are absorbed as the 2-monoacylglycerol, and little free glycerol is formed in the intestine. Therefore, the glycerol moiety and fatty acid moiety will be absorbed together and the time courses of their absorptions should be identical. There were no quantitative differences between urinary labeled carbon from glycerol derived from SALATRIM and glycerol derived from triolein. This is in contrast to the trend for higher concentrations of labeled carbon from oleate derived from triolein compared to stearate derived from SALATRIM. If the glycerol moiety and fatty acid moiety are absorbed together, the trends should be quantitatively identical. However, the data for oleate-derived radioactivity include radiolabel from some oleate absorbed in its free form, while the data for stearate-derived radiolabel does not because of the poor absorption of free stearate. Overall, the data for urinary radiolabeled carbon follow predictable patterns and indicate that SALATRIM is digested, absorbed, and metabolized similarly to other fats.

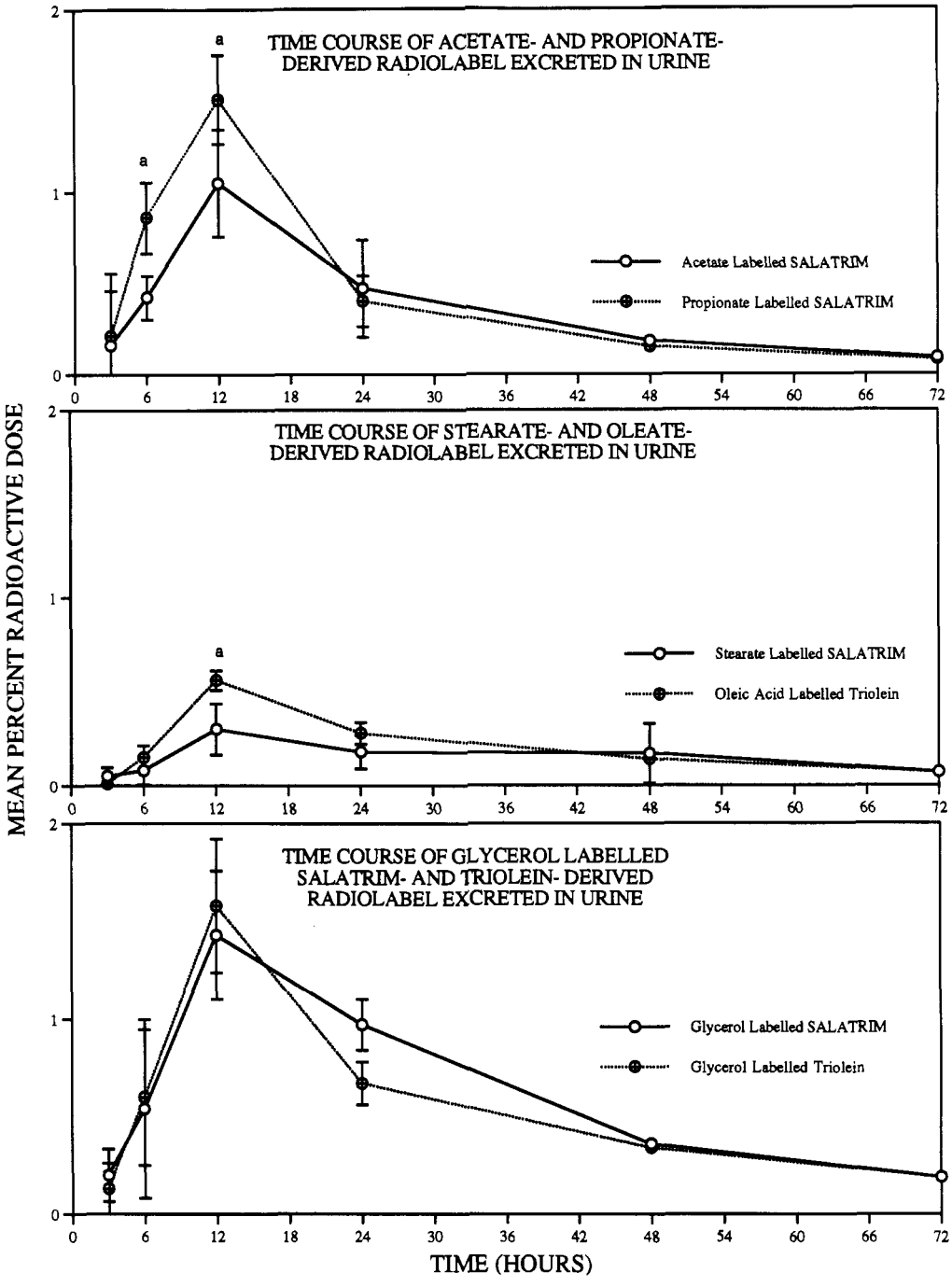


**Figure 4.** Excretion of radiolabel derived from SALATRIM and triolein in the urine of male rats over a 72-h time course. The SALATRIM and triolein were administered as a single oral gavage. Data represent the mean ± standard deviation for five rats at each time point. Data significantly different at each time point are marked with an "a".

**Comparison of Urinary Labeled Carbon from Rats Prefed with SALATRIM 23CA and Basal Diets.** Urinary excretion of radiolabeled carbon from rats fed SALATRIM 23CA lot A014 is shown in Figure 5. Comparison of urinary radiolabeled carbon derived from the SCFA and the LCFA indicates that prefeeding SALATRIM 23CA for 2 weeks prior to administration of the radiolabeled fats did not alter the metabolism of either the SALATRIM fat or triolein.

There appears to be a trend toward delay in the appearance of labeled carbon from glycerol in the urine of rats fed the 10% SALATRIM 23CA diet compared to the basal diet. However, considering the experimental error and the lack of difference in other variables between the SALATRIM prefed rats and the rats fed the basal diet, the biological significance of this is questionable.

**Excretion of SALATRIM- and Triolein-Derived Radiolabel in Feces.** Data for the time course of fecal excretion of radiolabeled carbon from the radiolabeled fats are presented in Figure 6. As would be expected, very little labeled carbon derived from SCFA is excreted in the feces. Because of the apparent hydrolytic specificity of lipases for the SCFA esterified to glycerol and their rapid absorption and metabolism, only small quantities would be expected to reach the large intestine and occur in the feces.

In contrast to the SCFA, a significant quantity of LCFA appears in the feces. A portion of radiolabel derived from the stearate of SALATRIM and the oleate of triolein probably remains esterified to glycerol as mono-, di-, and triacylglycerols. However, the amounts of esterified LCFA must be low because the amount of glycerol in the feces

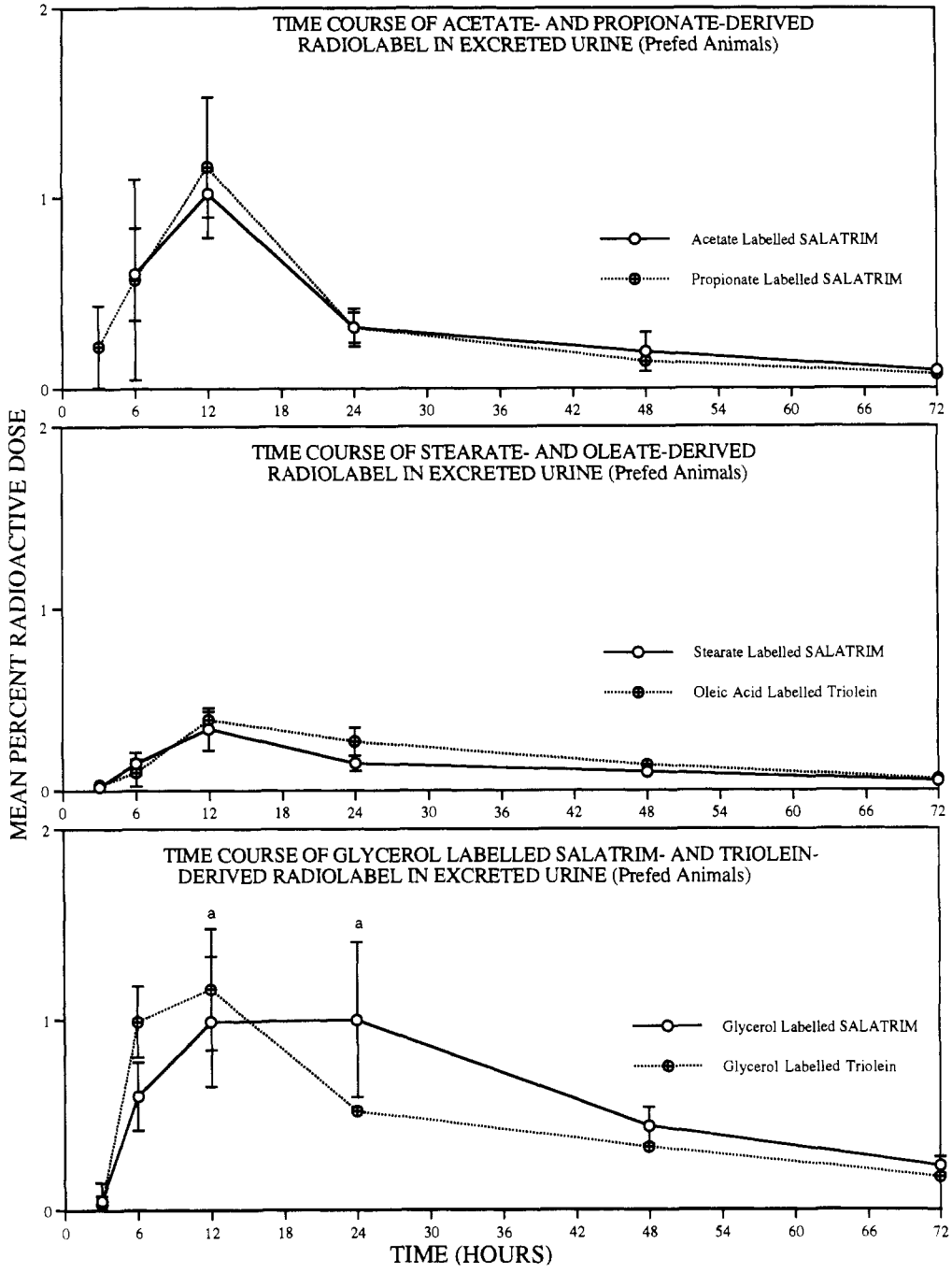


**Figure 5.** Excretion of radiolabel derived from SALATRIM and triolein in the urine of male rats fed the basal diet containing 10% (w/w) SALATRIM 23CA lot A014 for 14 days. The SALATRIM and triolein were administered as a single oral gavage. Data represent the mean ± standard deviation for five male rats at each time point. Data significantly different at each time point are marked with an "a".

is low compared to the LCFA. This indicates that the majority of the LCFA occur in their nonesterified form. Although the experimental error associated with LCFA-derived radiolabel in the feces is too large for a definitive conclusion, the trend is toward higher concentrations of stearate-derived radiolabel compared to oleate-derived radiolabel during the first 24 h of the study. This is consistent with the other aspects of this study that indicate poor absorption of stearate compared to oleate.

**Comparison of Fecal Excretion of Labeled Carbon from Rats Prefed with SALATRIM 23CA and Basal Diets.** Fecal excretion of radiolabeled carbon from the SCFA, LCFA, and glycerol of either SALATRIM or triolein was not altered by prefeeding 10% (w/w) SALATRIM 23CA lot A014 for 2 weeks, compared to animals not fed SALATRIM, as illustrated in Figure 7.

**Total Distribution of Radiolabel Derived from SALATRIM and Triolein.** Figure 8 illustrates the total disposition of acetate and propionate moieties (carbonyl labeled) of SALATRIM at 72 h postadministration in rats fed either the basal diet or basal diet containing 10% (w/w) SALATRIM 23CA.

Most of the SCFA esterified to glycerol in the SALATRIM fat are metabolized to $CO_2$ (82.2% for acetate and 89.3% for propionate). For the most part, the $CO_2$ is produced through energy-yielding metabolism in the gastrointestinal mucosa and liver and subsequent excretion in expired breath. The next largest pool of radiolabeled

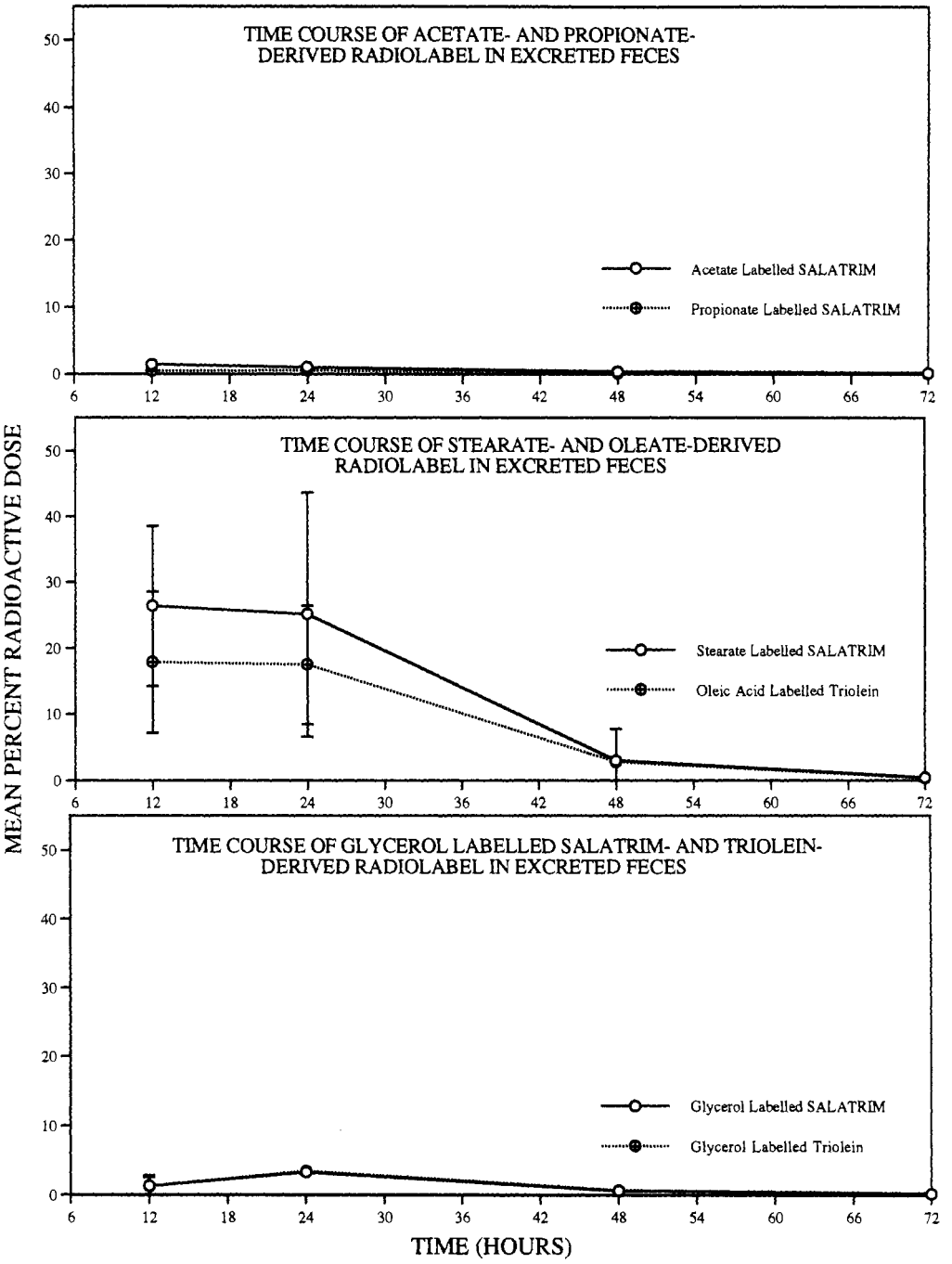


**Figure 6.** Excretion of radiolabel derived from SALATRIM and triolein in the feces of male rats over a 72-h time course. The SALATRIM and triolein were administered as a single oral gavage. Data represent the mean ± standard deviation for five rats at each time point. Data significantly different at each time point are marked with an "a".

carbon is in the rats' carcasses (6.0% for acetate and 2.6% for propionate). Although the biochemical nature of this radiolabel was not determined, it probably represents radiolabeled carbon that has entered normal anabolic metabolism and has been incorporated as a component of a wide array of biomolecules. Fecal and urinary excretions represent the next largest pool of radiolabel derived from the SCFA of SALATRIM fats (5.3% combined urine and feces for acetate and 4.4% for propionate). The liver contains a small amount of radiolabel derived from the SCFA of SALATRIM (0.7% from acetate and 0.7% from propionate). This is expected because of high catabolic and biosynthetic metabolism in the liver. Small quantities of acetate- and propionate-derived radiolabel are found in the blood 72 h postadministration (0.1% for acetate and 0.08% for propionate). This label probably represents bicarbonate, dissolved $CO_2$, and products of biodegradation and biosynthesis. As would be expected, very small quantities of radiolabel occur in the fat (0.05% for acetate and 0.02% for propionate). This radiolabel could represent a low level of fatty acid synthesis and other biosynthetic products.

To test the hypothesis that consuming high dietary concentrations of SALATRIM fats would not alter fat metabolism, rats were prefed 10% (w/w) SALATRIM in their diets and fat disposition was compared to that of rats fed the basal diet. As can be seen in Figure 8, feeding 10% (w/w) SALATRIM 23CA produced no major changes in the disposition of acetate and propionate derived from SALATRIM. There appeared to be a slight increase in the total radiolabel excreted in the feces of rats prefed with SALATRIM. This may result from competition for metabolism between the administered dose of radiolabeled

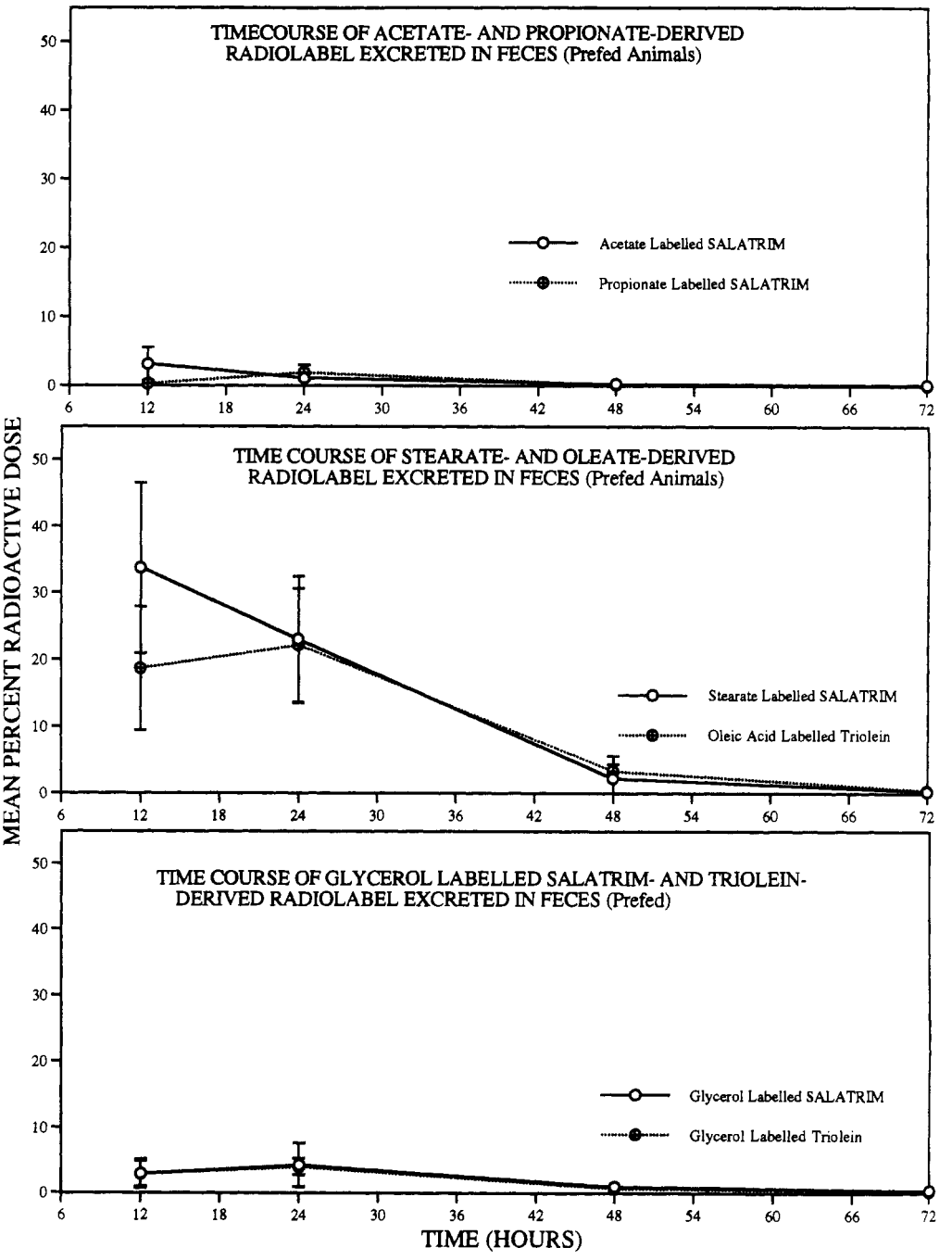


**Figure 7.** Excretion of radiolabel derived from SALATRIM and triolein in feces of male rats fed the basal diet containing 10% (w/w) SALATRIM 23CA lot A014 for 14 days. The SALATRIM and triolein were administered as a single oral gavage. Data represent the mean ± standard deviation for five rats at each time point. Data significantly different at each time point are marked with an "a".

SALATRIM and SALATRIM 23CA from the diet remaining in the gastrointestinal tract.

The total disposition of radiolabel derived from the stearic acid of SALATRIM and oleic acid derived from triolein is shown in Figure 9. An obvious difference in the disposition of these LCFA is the amount of radiolabel occurring in $CO_2$ from triolein metabolism is approximately twice the amount occurring in $CO_2$ from SALATRIM metabolism (44.3% for oleate from triolein and 21.5% for stearate from SALATRIM). If both fatty acids were absorbed equivalently, their conversion to $CO_2$ should be similar. There was a higher level of stearate-derived radiolabel in the feces compared to oleate (38.4% for oleate derived from triolein vs 54.8% for stearate derived from SALATRIM). These findings support the conclusion that stearate is a poorly absorbed LCFA. To confirm that the radiolabel that appeared in the feces was actually stearate, the quantity of stearate was determined analytically. This indicated 86.2% ± 11.4% of the radiolabel was associated with stearate.

Total body radiolabel was the next largest pool of radiolabel from the LCFA (9.6% for stearate from SALATRIM and 10.08% for oleate from triolein). Because fatty acids can be absorbed as the 2-monoacylglycerol, any stearic acid esterified at the 2-position should be absorbed. Once absorbed, the stearate and oleate at the 2-position of glycerol will be hydrolyzed and enter into normal fatty acid metabolism. Therefore, a portion of the absorbed fats will be metabolized to energy with resultant $CO_2$ production while a portion will serve as precursors for biosynthesis and some will be stored in fat. In fact, 0.36% of the radiolabel from stearate appears in fat, while 0.55% of the radiolabel from oleate appears in fat. A portion of this stearate-derived radiolabel in fat

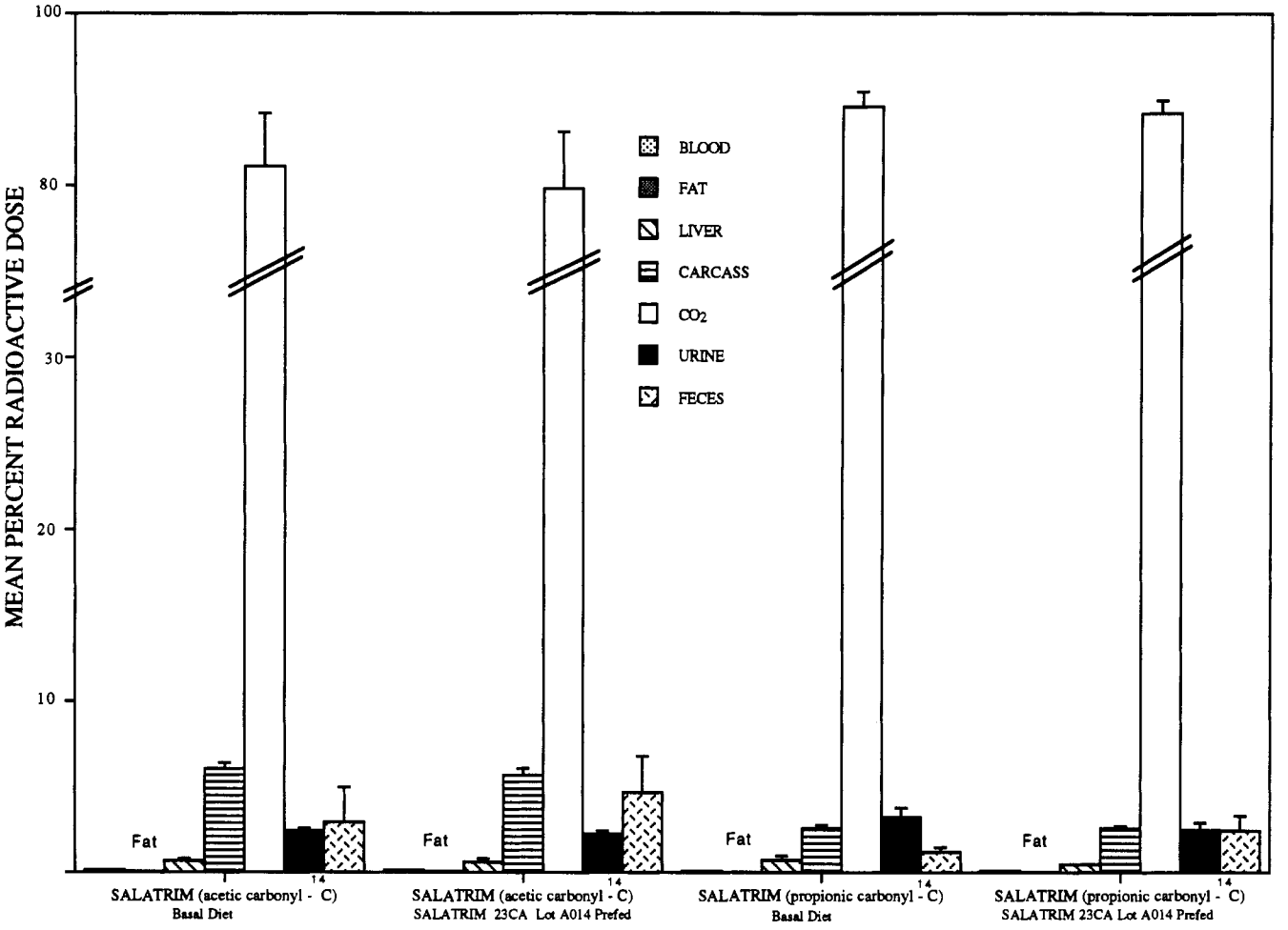


**Figure 8.** Total disposition of radiolabel derived from the acetate and propionate of SALATRIM at 72 h postadministration in male rats fed either the basal diet or the basal diet containing 10% (w/w) SALATRIM 23CA lot A014. Data represent the mean ± standard deviation of five rats.

could be oleate. Bonanome et al. (1992) presented evidence that stearate can be converted to oleate in the body. To determine what percentage of the radiolabel derived from stearate, if any, occurred as oleate, the quantity of radiolabeled oleate was determined in rats treated with SALATRIM containing radiolabeled stearate. These data are presented in Figure 10 and indicate that over half of the radiolabel is represented by oleic acid.

The other pools of radiolabel investigated in this study, i.e., blood, liver, and urine, demonstrate almost identical quantities of radiolabel derived from the stearate of SALATRIM and oleate from triolein (Figure 9). This indicates that the stearate derived from SALATRIM is utilized by the rat in a manner similar to the oleate derived from triolein, with the exception noted above where stearate is converted to oleate.

Overall, these data indicate that the LCFA derived from SALATRIM fats are absorbed and metabolized similarly to the LCFA from other fats. Therefore, the metabolism of these LCFA proceeds by highly predictable and understood pathways.

Comparison of the disposition of LCFA between rats prefed 10% (w/w) SALATRIM and rats prefed basal diet indicates no major differences in the patterns of disposition. However, as seen with the SCFA, the quantity of radiolabel slightly increased in the feces and slightly decreased in the absorption-dependent pools. Again, this may represent some competition for metabolism by the dietary SALATRIM and the radiolabeled SALATRIM administered by gavage.

The disposition of labeled glycerol from the SALATRIM fat and from triolein is illustrated in Figure 11. The glycerol moiety of dietary fats is believed to be absorbed as the 2-monoacylglycerol. Upon entry into the enterocyte of the intestinal mucosa, the 2-monoacylglycerol is hydrolyzed by cellular lipases to yield the free fatty acid and glycerol. The glycerol enters the cellular glycerol pool and can be either used as an energy source, reesterified to fatty acids to form fat, or enter into cellular biosynthetic pathways. As shown in Figure 11, the majority of the glycerol derived from SALATRIM and triolein is used for energy production with the production of radiolabeled $CO_2$ (74.2% for glycerol derived from SALATRIM and 75.8% for glycerol derived from triolein). The second largest pool of radiolabel derived from glycerol is the residual carcass (9.9% for glycerol derived from SALATRIM and 10.2% for glycerol derived from triolein). This pool represents glycerol that has been reesterified with fatty acids to form fats and glycerol that has entered into biosynthetic pathways. The next largest glycerol pool for both fats is the feces (5.34% for glycerol derived from SALATRIM and 5.46% for glycerol derived from triolein). Excretion of glycerol-derived radiolabel in the urine is somewhat less than that in the feces but nearly identical for the two fats (3.8% for glycerol derived from SALATRIM and 3.6% for glycerol derived from triolein). This glycerol-derived radiolabel probably represents bicarbonate from $CO_2$ production and products of anabolic and catabolic metabolism. Liver and blood concentrations of glycerol-associated radiolabel derived from either SAL-

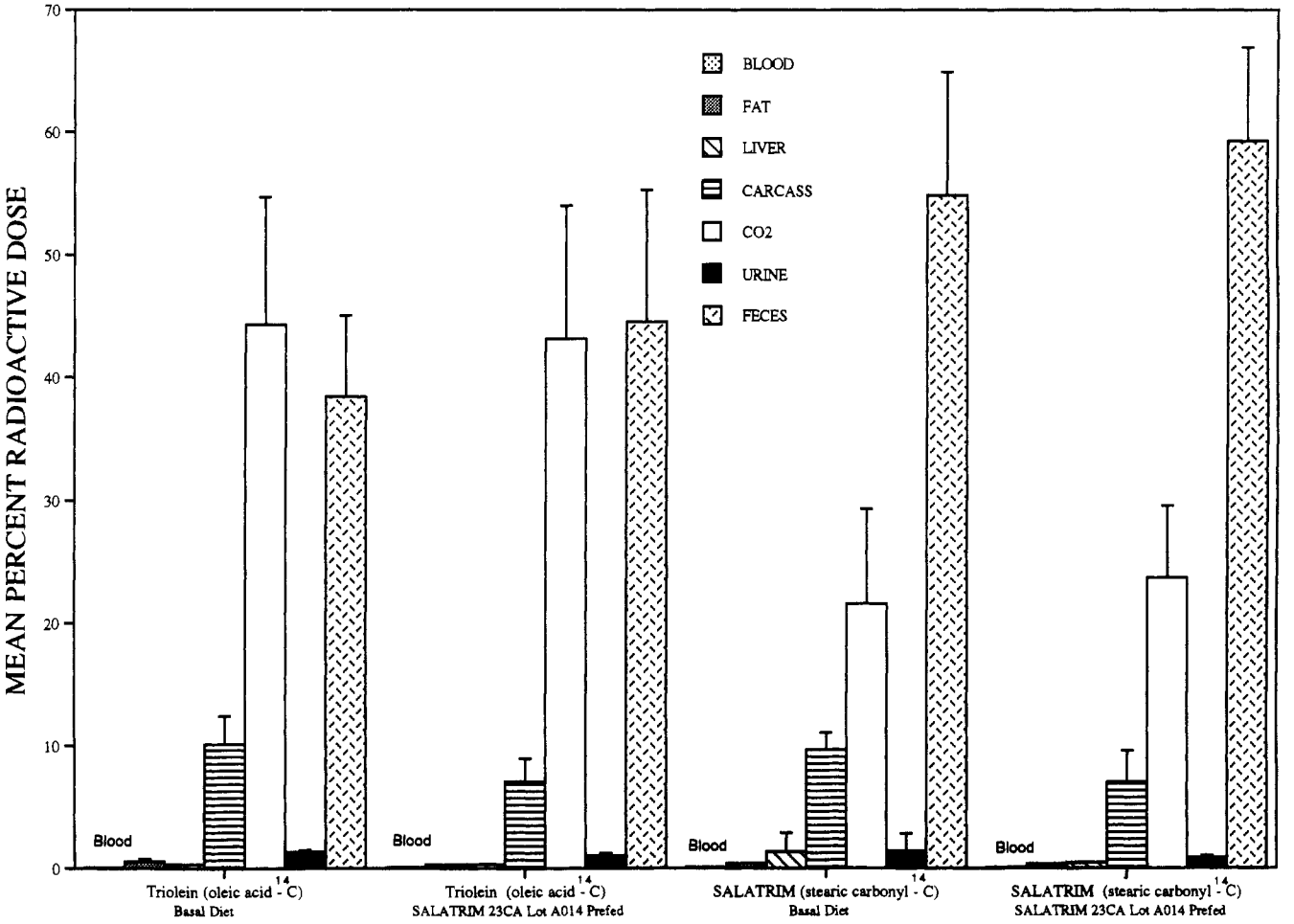


**Figure 9.** Total disposition of radiolabel derived from the oleate of triolein and the stearate of SALATRIM at 72 h postadministration in male rats fed either the basal diet or the basal diet containing 10% (w/w) SALATRIM 23CA lot A014. Data represent the mean ± standard deviation of five rats.

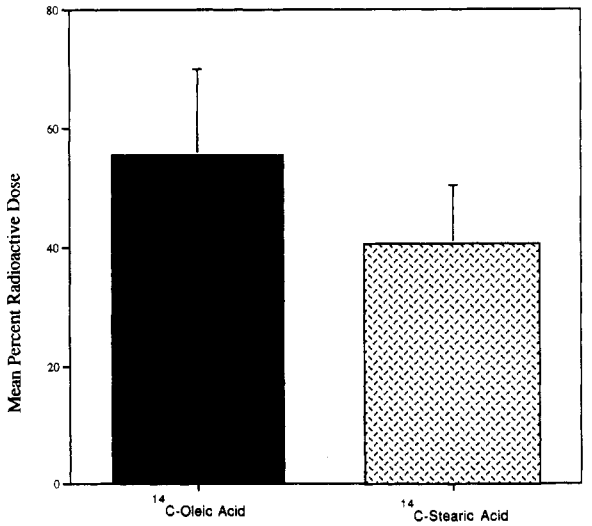


**Figure 10.** Radiolabeled oleic acid derived from stearic acid and the stearic acid content of fat from male rats administered a single dose of SALATRIM (label in carbonyl of stearic acid). Data represent the mean ± standard deviation of 10 rats. Five of the rats were fed the basal diet, and five of the rats were fed the basal diet containing 10% (w/w) SALATRIM 23CA lot A014.

ATRIM or triolein are nearly identical (liver concentrations for radiolabel derived from SALATRIM were 0.96% compared to 0.97% for triolein, and blood radiolabel derived from SALATRIM was 0.25% compared to 0.29% for triolein). Only small quantities of the glycerol-derived radiolabel from either fat reached the fat stores (0.07% for radiolabel derived from SALATRIM and 0.13% for triolein). This indicates that storage of fats produced by the reesterification of glycerol from either SALATRIM or triolein is not a major pathway under the conditions of this study.

Comparison of the disposition of both fats in rats fed either a 10% (w/w) SALATRIM diet or the basal diet indicates that consumption of large quantities of SALATRIM does not alter fat-derived glycerol disposition. No major differences were noted; however, as seen with the other fat components, there were slight differences between the SALATRIM-fed and nonfed rats. As seen with the disposition of the LCFA and SCFA, there was a trend toward slightly higher concentrations of glycerol-derived radiolabel in the feces (5.3% vs 8.5% for glycerol derived from SALATRIM and 5.5% vs 7.8% for glycerol derived from triolein). This slight increase in fecal glycerol was associated with a slight decrease in absorption-dependent glycerol pools. As previously noted, this may result from the competition for metabolism between dietary SALATRIM in the gastrointestinal tract and the fats administered by gavage.

## CONCLUSIONS

The study described in this paper was designed to test the hypothesis that SALATRIM fats will be metabolized by the well-known pathways of fat disposition in a highly predictable manner. This hypothesis was based upon the similarity of the structure of SALATRIM to normal dietary fats. SALATRIM fats are structured triacylglycerols that

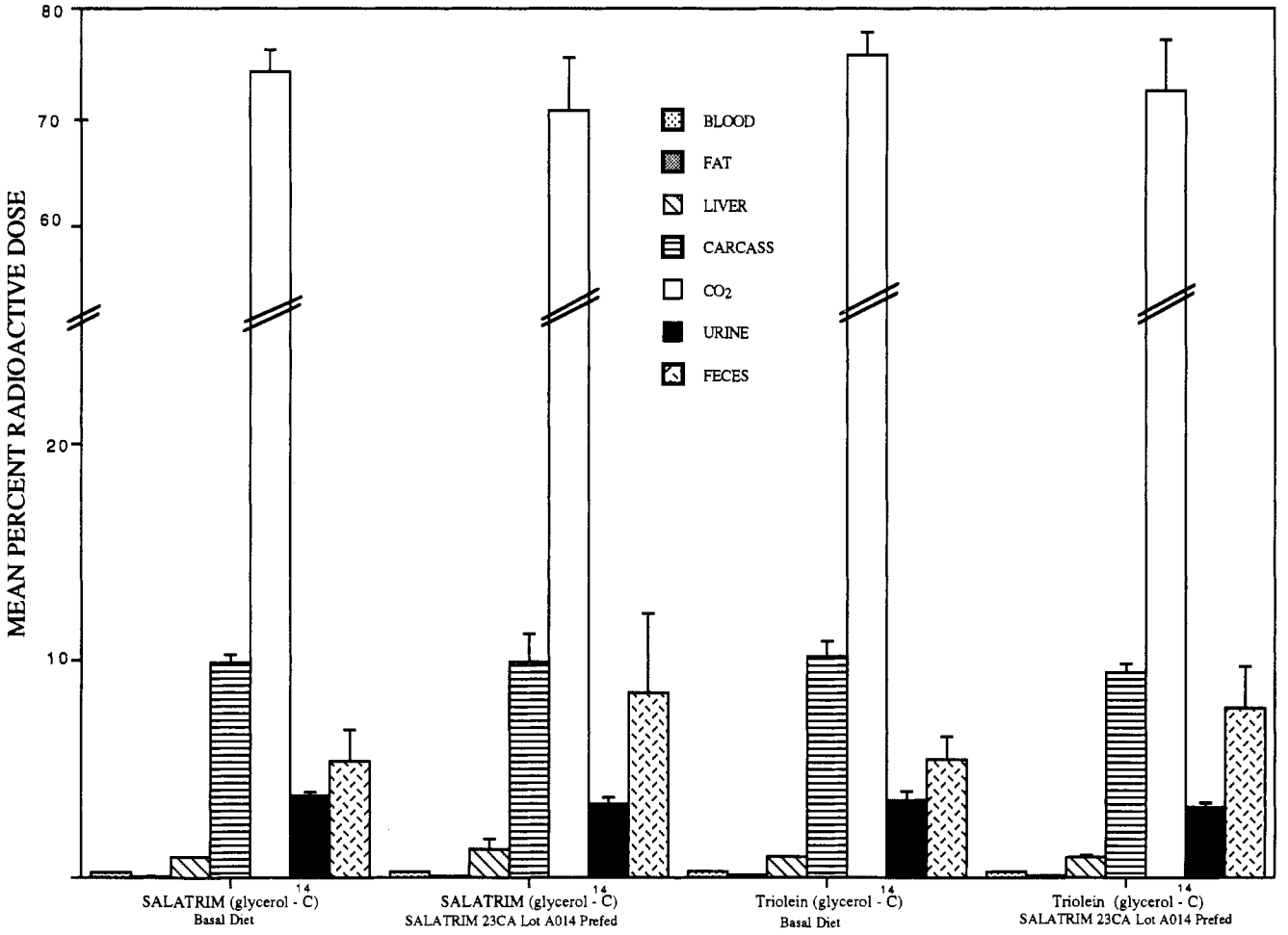


**Figure 11.** Total disposition of radiolabel derived from the glycerol of triolein and SALATRIM at 72 h postadministration in male rats fed either the basal diet or the basal diet containing 10% (w/w) SALATRIM 23CA lot A014. Data represent the mean ± standard deviation of five rats.

differ from most dietary fats in that they have a high proportion of SCFA (acetic, propionic, and/or butyric acid) esterified to glycerol. Milk fats that occur in most dairy products are common dietary fats that contain SCFA. The major LCFA that occurs in SALATRIM is stearic acid, another common component of dietary fats. Therefore, the chemical components that comprise SALATRIM fats are components of dietary fats and should be absorbed, metabolized, and excreted as they are in other fats.

This study indicates the disposition of SALATRIM fats is very similar to that of a model fat, triolein. One difference is the SCFA are more rapidly hydrolyzed from the glycerol backbone of SALATRIM than are the LCFA of triolein. This is predictable because gastrointestinal lipases demonstrate higher lipolysis rates toward SCFA than LCFA. The SCFA produced by the hydrolytic activity of the lipases are rapidly absorbed and used as a substrate for energy production with the concomitant production of $CO_2$.

Another difference between the dispositions of SALATRIM and triolein is that the stearic acid produced by lipase activity is more poorly absorbed than the oleic acid produced from triolein. This is evident by the higher conversion of the oleic acid to $CO_2$ and by the higher concentration of stearic acid in the feces. The poorer absorption of stearic acid and the fewer number of carbon atoms in SCFA compared to the LCFA results in SALATRIM fats having lower caloric availability than fats such as corn oil.

A second hypothesis tested in this study is that prefeeding a SALATRIM fat for as long as 14 days will not alter the disposition of SALATRIM and other fats. The data reported here confirm that hypothesis. No biologically significant differences were observed in the disposition of either SALATRIM or triolein when their disposition in rats fed 10% (w/w) SALATRIM 23CA lot A014 for 14 days was compared to that in rats fed the basal diet for 14 days.

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