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PHARMACEUTICAL COMPOSITIONS CONTAINING SALTS OF CHOLESTEROL HEMISUCCINATE

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A61K 031/415

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(71) Applicant(s)
THE LIPOSOME COMPANY, INC.

(72) Inventor(s)
ANDREW STUART JANOFF; CHRISTINE E. SWENSON; PAUL A. TREMBLAY; MIRCEA CONSTANTINE POPESCU; ALAN LEE WEINER; LOIS E. BOLCSAK

(74) Attorney or Agent
PHILLIPS ORMONDE & FITZPATRICK, 367 Collins Street, MELBOURNE VIC 3000

(56) Prior Art Documents
US 4040784
US 4042330
US 4183847

(57) Claim

1. A composition comprising the tris(hydroxymethyl) aminomethane salt of cholesteryl hemisuccinate and an antifungal compound adapted for administration to animals, including humans.

27. A composition comprising the tris-(hydroxymethyl) aminomethane salt of cholesteryl hemisuccinate and a peptide adapted for administration to animals, including humans.

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COMPLETE SPECIFICATION

(ORIGINAL)

Class Int. Class

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Priority

Related Art:

APPLICANT'S REF.: Div. of 49528/85

Name(s) of Applicant(s): THE LIPOSOME COMPANY, INC.

Address(es) of Applicant(s): One Research Way, Princeton Forrestal Center, Princeton, New Jersey 08540, United States of America

This document contains the amendments made under Section 49 and is correct for printing

Actual Inventor(s): ANDREW STUART JANOFF
MIRCEA CONSTANTINE POPESCU
ALAN LEE WEINER
LOIS E. BOLCSAK

Address for Service is:

PHILLIPS, ORMONDE AND FITZPATRICK
Patent and Trade Mark Attorneys
367 Collins Street
Melbourne, Australia, 3000

Complete Specification for the invention entitled:

"Pharmaceutical Compositions Containing Salts of Cholesterol Hemisuccinate".

The following statement is a full description of this invention, including the best method of performing it known to applicant(s):

AUSTRALIAN PATENT OFFICE LS

P19/3/84

FIELD OF THE INVENTION

This application is a divisional application of Australian Patent Application No 49528/85 the entire contents of which are incorporated herein by reference.

The present invention relates to novel and useful pharmaceutical compositions containing the tris(hydroxymethyl) aminomethane salt (tris-salt) form of cholesterol hemisuccinate.

SUMMARY OF THE INVENTION

In one embodiment the present invention is directed to a composition comprising the tris(hydroxymethyl) aminomethane salt of cholesteryl hemisuccinate and an antifungal compound, particularly when the antifungal agent is miconazole, terconazole or econazole, isoconazole, tioconazole, bifonazole, clotrimazole, ketoconazole, butaconazole, itraconazole, oxiconazole, fenticonazole, nystain, naftifine, amphotericin B, zinoconazole or ciclopirox olamine. The composition can be used to treat a fungal infection and can be administered topically including orally or intravaginally.

In a further embodiment the present invention includes a composition comprising a tris(hydroxymethyl) aminomethane salt of cholesteryl hemisuccinate and a peptide, particularly a hydrophobic peptide, human growth hormone, bovine growth hormone, porcine growth hormone or insulin. The composition can be administered to increase milk production or to increase or initiate growth of a mammal.

DETAILED DESCRIPTION OF THE INVENTION

CHS-tris Antifungal Compositions

The tris(hydroxymethyl)aminomethane salt of cholesteryl hemisuccinate ("CHS-tris") forms a semi-solid gel when dissolved at a high concentration (for example, about 100 mg/ml or greater) in hot organic solvents such as ethanol and allowed to cool to about 25°C. The resulting gel is more

uniform in appearance, if the host solvent CHS-tris solution is sonicated briefly in a bath sonicator. The solvent in the gel can be removed by evaporation in air or under vacuum. The gel (before or after removal of the solvent) disperses as small vesicles, upon hydration.

Bioactive agents, for example, antifungal compounds can be incorporated into the gel, prior to gel formation, and the gel can then be administered intravaginally,

intrarectally, topically, or orally. Antifungal compounds which may be present in the formulations of the instant invention include miconazole, terconazole, econazole, isoconazole, tioconazole, bifonazole, butaconazole, itraconazole, oxiconazole, fenticonazole, nystatin, naftifine, ketoconazole, ciclopirox olamine, clotrimazole, zinoconazole, amphotericin B, and the like. It is to be understood that free bases or pharmaceutically acceptable salts of the antifungal compounds are within the scope of the present invention. A pharmaceutically acceptable salt is one which is nontoxic and does not cause unacceptable side effects. Generally, any antifungal compound compatible with the formulations may be employed. Othe ingredients can be added to the ethanol solution prior to gelling to give the final samples desirable physical characteristics such as softness, lubricity, stability, scent, taste and the like. Useful additional ingredients include waxes, vegetable butters such as either hard butter triglycerides of lauric acid derived from vegetable fats, or cocoa butter, and phospholipids such as egg phosphocholine. Organic carboxylic acids which are weak to moderately acidic such as lactic acid can be added to lower the pH and/or increase the solubility of basic drugs. Preferably the organic carboxylic acid has up to 12 carbon atoms, more preferably up to 6 carbon atoms. Improved solubility of polar, basic antifungals, such as terconazole in CHS-tris, occurs when about 5-15 percent by weight of an organic carboxylic acid, such as lactic acid, is present in the formulation. Certain compounds such as lactic acid may also be desirable due to their ability to hold water and therefore "soften" the formulation. The additional ingredients or excipients should be stable, pharmaceutically acceptable, i.e., nontoxic, should not adversely interfere with the efficacy and safety of the bioactive agent, and should be appropriate for the mode of administration, such as intravaginally.

Suppositories

The gel can be formed in any suitable container. For suppositories, the gels may be formed in molds of appropriate size and shape and administered in the form of an intravaginal suppository which will slowly "disperse" to form liposomes in vivo. The gel can also be hydrated prior to use to form a cream or suspension which are administered topically or intravaginally. The hydrated gels have been shown by x-ray diffraction to consist of multilamellar stuctures typical of liposomes. Alternatively, the gel can be formed as a cream or suspension by other methods known to those skilled in the art.

When making suppositories, CHS-tris gels are generally difficult to mold because of the lack of adhesion of the gel particles to each other. Therefore, a wax, a vegetable butter, phospholipid or other molding agent can be included. For hard butters such as Wecobee M, the weight-to-weight ration of CHS-tris to hard butter is between about 0.15:1 and 4:1. Remington's Pharmaceutical Sciences, 16th Ed., Mack Publishing Co., Easton, PA, 1980, pages 1530-1533, discusses suppository formulations and is incorporated herein by reference.

For phospholipids such as egg phosphocholine, the weight-to-weight ratio of CHS-tris to phospholipid is between about 10:1 to 1:1. When phospholipids such as egg phosphocholine are employed, an organic carboxylic acid having for example, 1-6 carbon atoms such as lactic acid can be added in order to increase the solubility of the antifungal agent if necessary. The amount of lactic acid added to an antifungal, such as miconazole or terconazole base, is preferrably equimolar or less.

Waxes, vegetable butters, and the like are chosen which are compatible with the antifungal CHS-tris formulation, are solid at room temperature and melt at body temperature. Phospholipids are chosen which are compatible with the antifungal CHS-tris formulations and disperse in the vaginal mucosa. Other carriers known in the art may also be employed for the intravaginal administration of the formulations of the present invention.

Creams

CHS-tris cream formulations of the present invention are prepared by dissolving CHS-tris in an organic solvent, preferably between about 50 and 100 mg of CHS-tris per ml of solvent. Useful solvents including alcohols, such as methanol, ethanol and isopropanol, are those in which the antifungal compound or other bioactive agent is soluble. The CHS-tris is added to boiling solvent which is then removed from the heat source. The bioactive agent is then added with gently stirring. The resulting solution is poured into a large container so that there is considerable exposed surface area of the solution in order to facilitate solvent evaporation.

The solution is briefly sonicated for between about 30 and 180 seconds until gelling begins. The terms "gel" and "gelling" refers to a viscous composition which resembles jelly or gelatin in its texture. The colloidal properties of the viscous composition are not known. The container is covered tightly and gelling is completed at about 20-30°C, preferable 25°C in about four hours. The cover is removed and the solvent is allowed to evaporate. The solvent can also be removed *in vacuo*.

The resulting dry gel is solid and can be cut into sections and ground in a blender with frozen carbon dioxide until a fine, granular powder is obtained. The powder is mixed with water until a homogeneous cream having the desired volume is obtained. The consistency of the cream is dependent upon the amount of water added. In general, if the final concentration of CHS-tris is less than about 200 mg/ml, the resulting composition is fluid and could be administered as a douche. Alternatively, a cream can be produced by the addition of waxes and the like which increase the viscosity of the composition. If the concentration is about 200-400 mg/ml, the resulting composition can be administered as a cream. If the concentration is greater than about 450 mg/ml, the resulting composition will be a semi-solid to solid gel that could be administered as a suppository.

Administration

CHS-tris formulations of the present invention which contain antifungal agents such as miconazole, terconazole and the like are useful for treating vaginal infections such as those caused by Candida, for example Candida albicans, in mammals including humans. Infections in other parts of the body such as thrush can also be treated using formulations of the present invention. These formulations are conveniently administered intravaginally as douches, creams or suppositories. The amount of antifungal agent in douche, cream or suppository dosage forms will depend on several factors for example, the potency of the drug, irritation caused by the drug, how quickly the drug is washed out of the vaginal cavity by normal vaginal secretions, etc. but can range from about 10 to 1500 mg. For example, about 50 mg to 1.2 g for miconazole, or about 20 mg to 480 mg for terconazole are useful doses. This amount of drug can be incorporated in a suppository generally weighing

about 5 ml or less. In general, a more concentrated formulation administered in a smaller volume is more desirable from the standpoint of convenience and comfort of the patient.

Often, only one administration of the formulation of the present invention is necessary to cure a vaginal infection susceptible to the antifungal agent. For prior art formulation, generally three to six to as many as fourteen doses are required.

CHS-tris Peptide Compositions

Proteins and other peptides, especially those which are hydrophobic such as growth hormones, insulin, low density lipoproteins and the like, can be included in formulations of the the present invention in order to solubilize, control the rate of release or target the site of action. The peptides are generally administered parenterally, such as intravenously, intramuscularly or intraperitoneally. Growth hormones which can be employed includes human growth hormone, bovine growth hormone and porcine growth hormone. For example, bovine growth hormone can be solubilized in aqueous buffer using CHS-tris. Between about 5 and 170 mg or more, preferably 150-170 mg, of bovine growth hormone per ml of aqueous buffer can be solubilized with CHS-tris, preferably between about 5 and 300 mg of CHS-tris and more preferably between about 25 and 50 mg of CHS-tris per ml of aqueous buffer.

One method of solubilizing proteins and other peptides using CHS-tris is to prepare a multilamellar liposome (MLV) in aqueous buffer. The resulting MLV's can be used as is or sonicated to obtain SUV's. The peptide is suspended in the aqueous buffer-liposome mixture and

partitions into the liposomal bilayers, thereby being solubilized. Alternatively, the solubilized peptide can be employed as the aqueous phase in preparing SPLV's. The resulting solubilized peptide composition can be administered to a mammal, including humans.

Growth hormones can be employed to increase milk production or to increase or initiate growth. For administration of bovine growth hormone to dairy cows to increase milk productions intermuscularly, generally about 10 mg per day is required. For a 30 day controlled release dosage form of the presen invention about 300 to 1200 mg are administered as a single dose.

For controlled release dosage forms of peptides, the amount administered is determined by a veterinarian or physician, as appropriate. The release characteristics of the dosage form, the amount of peptide which can be utilized by the body, toxicological consideration, and the like will determine the actual dose.

The following examples are given for purposes of illustration and not by way of limiting the scope of the invention.

1. MICONAZOLE - CHS-TRIS CREAM

The tris(hydroxymethyl)aminomethane ("tris") salt of cholesterylhemisuccinate ("CHS-tris") (20.64 g), available from Sigma Chemical Co., St. Louis, Missouri, was dissolved in 206 ml of boiling ethanol. When the ethanol-CHS-tris solution was clear, it was removed from the heat and 5.16 grams of miconazole base in 103 ml ethanol was added with gentle stirring. The clear solution was poured into a large, flat dish (243 x 243 x 18mm) and sonicated briefly in a large bath sonicator until gelling began. The dish was then covered tightly and allowed to complete gelling at 25°C for four hours. The cover was removed and the ethanol allowed to evaporate. The dried gel was then cut into sections, placed in a blender with frozen carbon dioxide and ground until it was a fine, granular powder. The resulting powder was mixed with sterile, distilled water until a homogeneous cream with a final volume of 51.6 ml was obtained with a concentration of miconazole of 100 mg/ml.

The procedure was repeated using 10.32 g of miconazole to obtain a cream having a concentration of miconazole of 200 mg/ml.

Additional miconazole-CHS-tris formulations were prepared according to the above described procedures and

materials with the materials having the proportions as follows:

Miconazole (mg)	CHS-tris (mg)	Water (mg)
50	100	850
100	200	700
100	400	400
200	400	500

2. TERCONAZOLE CHS-TRIS CREAM

CHS-tris (2.0 g) was dissolved in 20 ml of boiling ethanol. When the ethanol-CHS-tris solution was clear, it was removed from the heat and 1.0 gm of terconazole (in 50 ml of ethanol) and 83 mg of L (+)-lactic acid (in 2 ml ethanol) was added with gentle stirring. The clear solution was poured into a large beaker and sonicated until "gelling" began. The beaker was covered tightly and allowed to complete gelling at room temperature for four hours. The cover was removed and the ethanol allowed to evaporate in air at 25°C. The dried gel was cut into sections, placed in a blender with frozen carbon dioxide and ground until it was a fine granular powder. This powder was mixed with 10 ml of water containing 6.3 g CHS-tris sonicated vesicles. The resulting suspension was mixed with distilled, sterile water until a homogeneous cream with a final volume of 20.8 ml was obtained having 48 mg terconazole per ml of cream.

3. MICONAZOLE - CHS-TRIS SUPPOSITORY

CHS-tris (400 mg) was dissolved in 4 ml of boiling ethanol. 100 mg miconazole base in 1 ml ethanol and 1.2 g of a melted hard butter triglyceride of lactic acid derived from vegetable fats (Wecobee M, PVO Internation, Boonton, New Jersey), was added with stirring. The resulting clear solution was aliquotted into 1.5 ml polypropylene microfuge

tubes, sonicated briefly (approximately 60 second) to "gel" the CHS-tris and left at room temperature for four hours to solidify completely. The polypropylene "mold" was removed and the semi-solid gels were put in a vacuum dessicator overnight to remove the residual ethanol.

Other miconazole CHS-tris suppository formulations prepared according to the above described procedure are listed below.

Miconazole (mg)	CHS-tris (mg)	Wecobee M (mg)	Lactic Acid (mg)	Egg Phosphocholine (mg)
3.4	6.8	80.3		
3.4	13.6	20.5		
6.9	13.8	80.3		
6.9	27.6	44.0		
10.3	20.6	80.3		
20.0	41.0	12.0		
20.6	41.2			
20.6	41.2		2.2	10.3
20.6	41.2	79.4		

4. IN VIVO ACTIVITY FOR VAGINAL CANDIDA INFECTIONS

Ovariectomized rats (Charles River Breeding Laboratories) were treated weekly with beta-estradiol valerate to induce a state of constant estrus (and susceptibility to vaginal Candida infection). The rats were inoculated, intragavaginally with 5×10^6 CFU of Candida albicans. Starting on the third day after inoculation, infected rats were treated, intravaginally, with 2% weight to volume miconazole nitrate cream twice a day for three days, 12% miconazole nitrate cream administered once on the third day post-inoculation, or with one of the CHS-tris suppository or cream preparations of the present invention administered only once on the third day post-inoculation. Some rats

received no treatment. On the sixth or tenth day after inoculation, the vaginas of all rats were sampled for Candida albicans. Those rats with fewer than 25 CFU/vaginal swab were considered cured.

The claims defining the invention are as follows:

1. A composition comprising the tris(hydroxymethyl) aminomethane salt of cholesteryl hemisuccinate and an antifungal compound adapted for administration to animals, including humans.

2. A composition according to claim 1 wherein the antifungal compound is miconazole.

3. A composition according to claim 1 wherein the antifungal compound is terconazole.

4. A composition according to claim 1 wherein the antifungal compound is econazole, isoconazole, tioconazole, bifonazole, clotrimazole, ketoconazole, butaconazole, itraconazole, oxiconazole, fenticonazole, nystatin, naftifine, amphotericin B, zinoconazole or ciclopirox olamine.

5. A method for treating a fungal infection in a subject comprising the step of administering an antifungal effective amount of a composition according to any one of claims 1 to 4.

6. A method according to claim 5 wherein the administration is topically.

7. A method according to claim 5 wherein the administration is orally.

8. A method according to claim 5 wherein the administration is intravaginally.

9. A method according to claim 7 wherein the antifungal compound is miconazole.

10. A method according to claim 1 wherein the antifungal compound is terconazole.

11. A method according to claim 1 wherein the antifungal compound is econazole, isoconazole, tioconazole, bifonazole, clotrimazole, ketoconazole, butaconazole, itraconazole, oxiconazole, fenticonazole, nystatin, naftifine, amphotericin B, zinoconazole or ciclopirox olamine.

12. A method according to claim 5 wherein the antifungal compound is miconazole.

13. A method according to claim 5 wherein the antifungal compound is terconazole.

14. A method according to claim 5 wherein the antifungal compound is econazole, isoconazole, tioconazole, bifonazole, clotrimazole, ketoconazole, butaconazole, itraconazole,

oxiconazole, fenticonazole, nystatin, naftifine, amphotericin B, zinoconazole or ciclopirox olamine.

15. A cream comprising a composition according to any one of claims 1 to 4.

16. A cream according to claim 15 wherein the antifungal compound is miconazole.

17. A cream according to claim 15 wherein the antifungal compound is terconazole.

18. A method for treating a fungal infection in a subject comprising the step of administering an antifungal effective amount of a cream according to claim 15.

19. A method according to claim 18 wherein the administration is topically.

20. A method according to claim 18 wherein the administration is orally.

21. A method according to claim 18 wherein the administration is intravaginally.

22. A suppository comprising a composition according to any one of claims 1 to 4.

23. A suppository according to claim 22 wherein the antifungal compound is miconazole.

24. A suppository according to claim 22 wherein the antifungal compound is terconazole.

25. A method for treating a fungal infection in a subject comprising the step of administering an antifungal effective amount of a suppository according to claim 22.

26. A method according to claim 25 wherein the administration is intravaginally.

27. A composition comprising the tris-(hydroxymethyl) aminomethane salt of cholesteryl hemisuccinate and a peptide adapted for administration to animals, including humans.

28. A composition according to claim 27 wherein the peptide is hydrophobic.

29. A composition according to claim 27 wherein the peptide is human growth hormone.

30. A composition according to claim 27 wherein the peptide is bovine growth hormone.

31. A composition according to claim 27 wherein the peptide is porcine growth hormone.

32. A composition according to claim 27 wherein the peptide is insulin.

33. A method of increasing milk production or of increasing or initiating growth of a mammal comprising the step of administering an effective amount of a composition according to claim 27 wherein the peptide is a growth hormone.

34. A method according to claim 33 wherein the growth hormone is bovine growth hormone.

35. A composition as claimed in claim 1 or claim 27 substantially as hereinbefore described with reference to any one of the examples.

DATED: 1 February 1991

PHILLIPS ORMONDE & FITZPATRICK
Patent Attorneys for:
THE LIPOSOME COMPANY, INC David B Fitzpatrick

FIG. 1

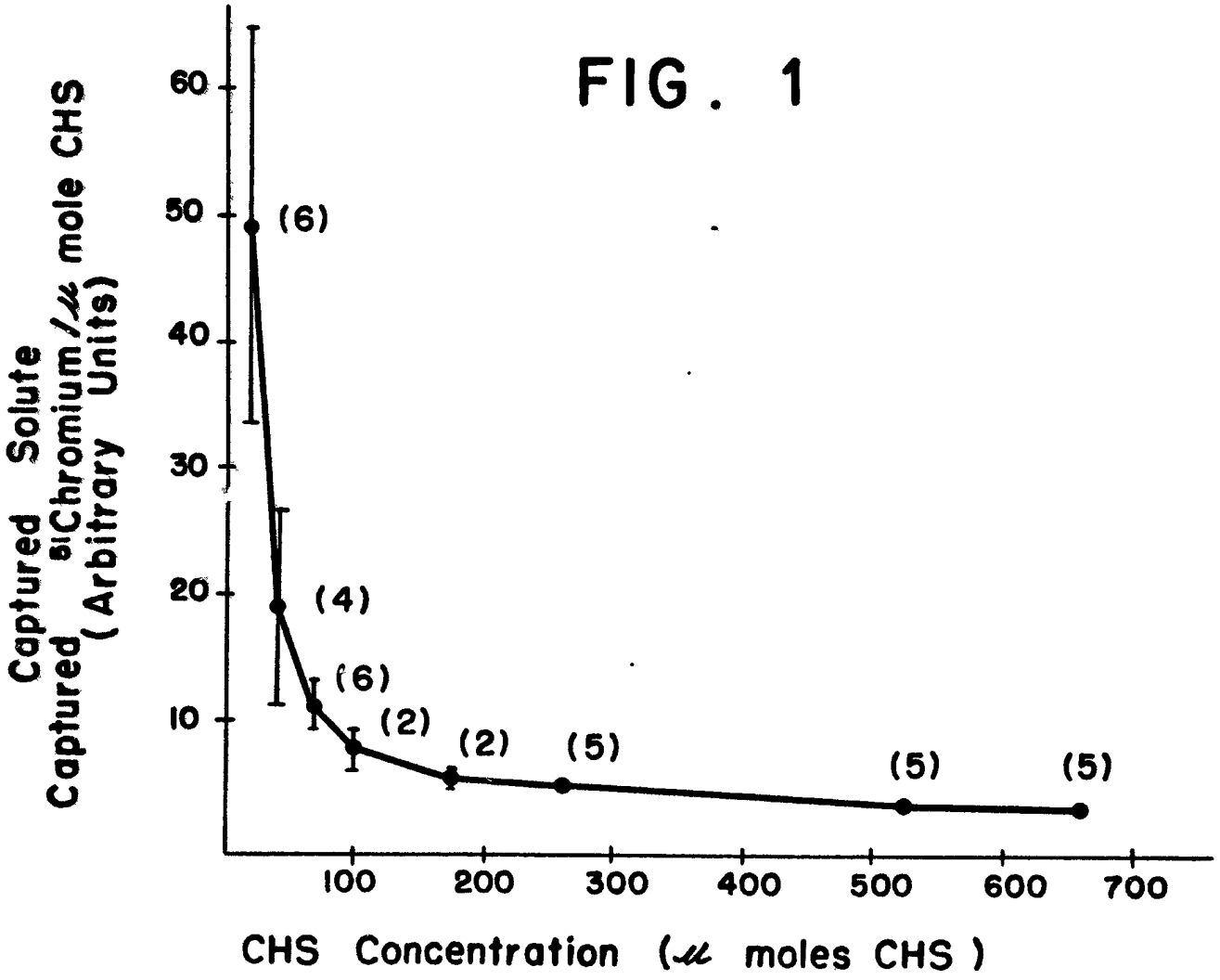


FIG. 2

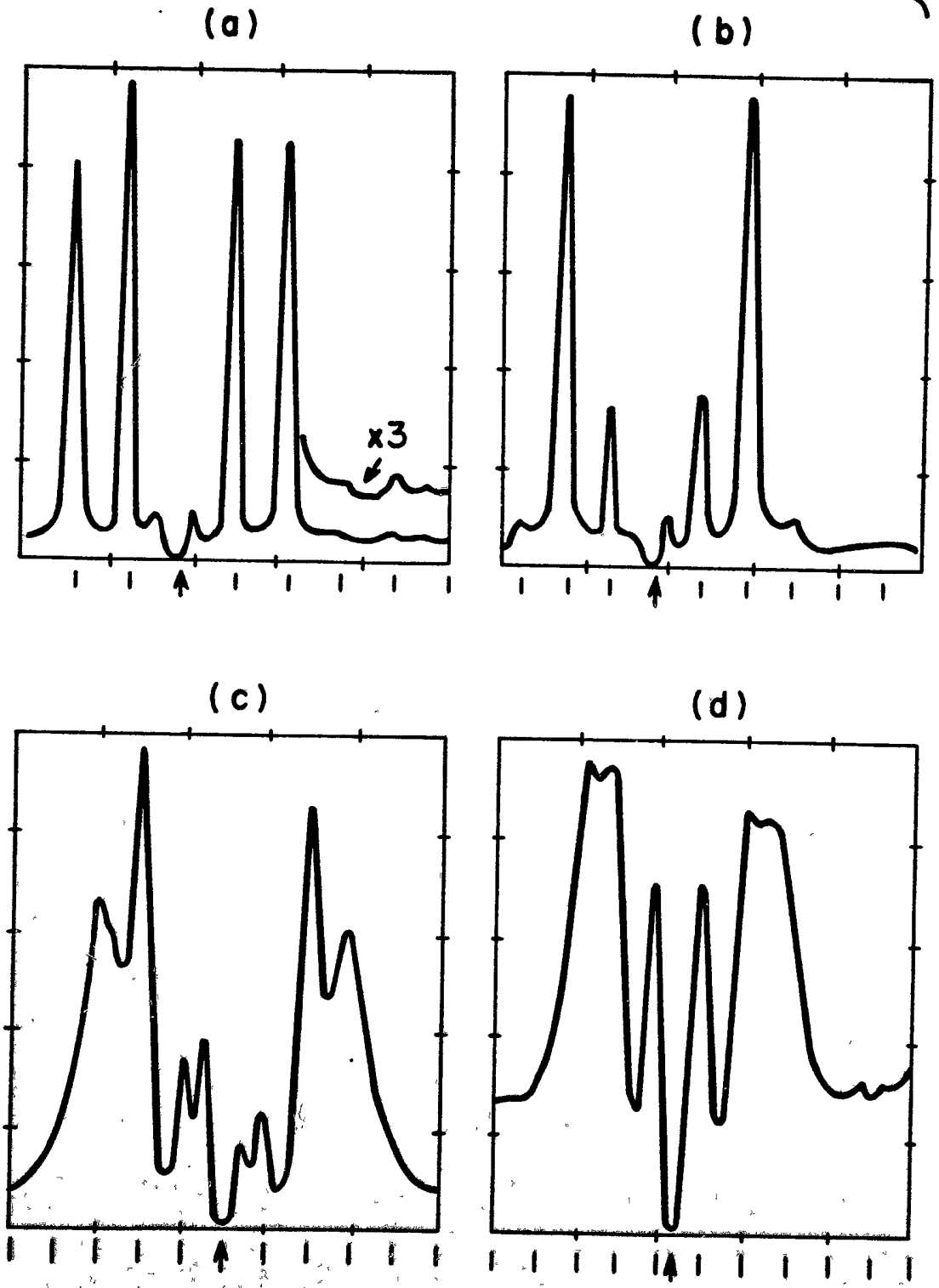
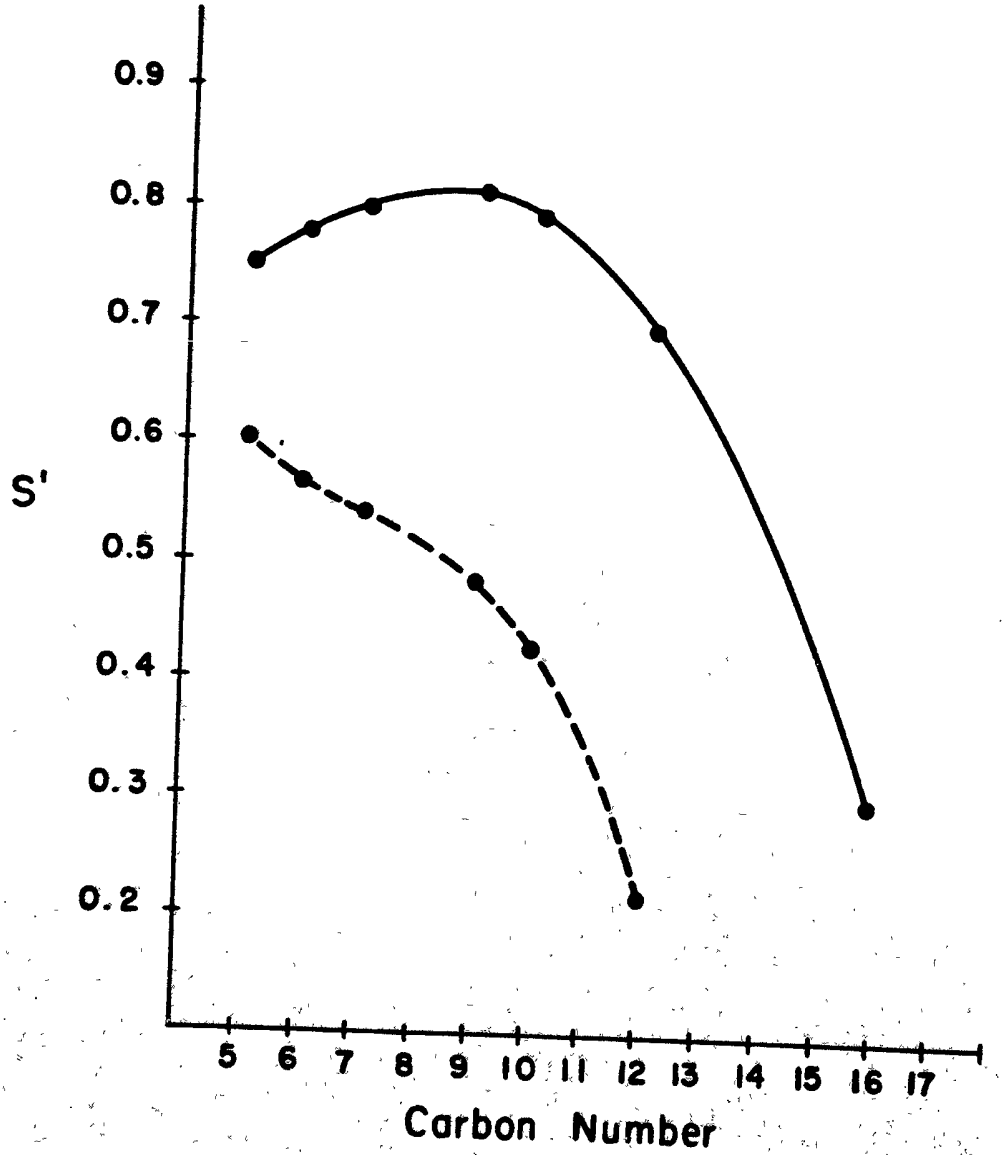


FIG. 3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0.7
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0.5
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0.3
0.2
S'
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Legend
CHS
EPC

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FIG. 4

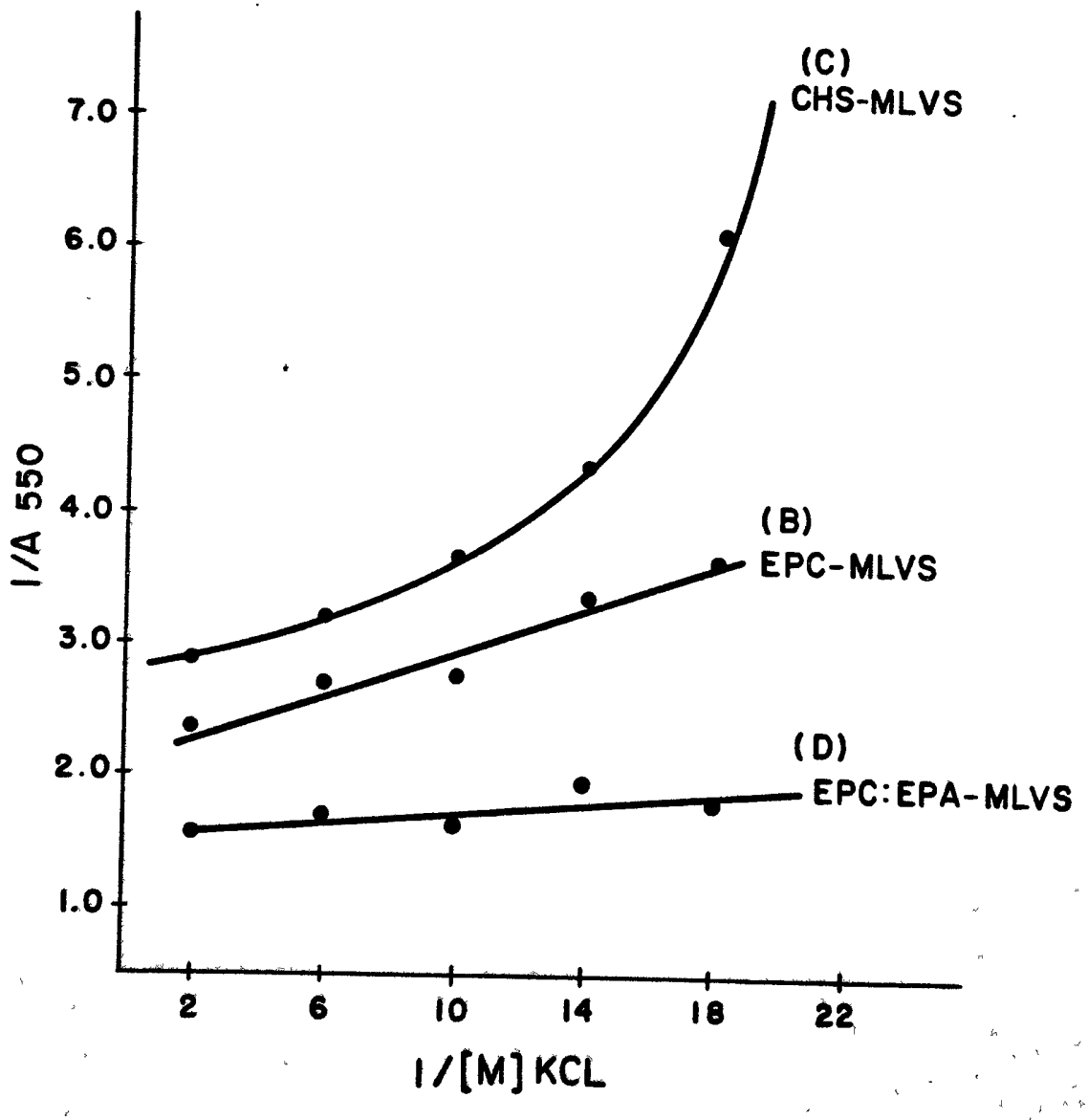

FIG. 5

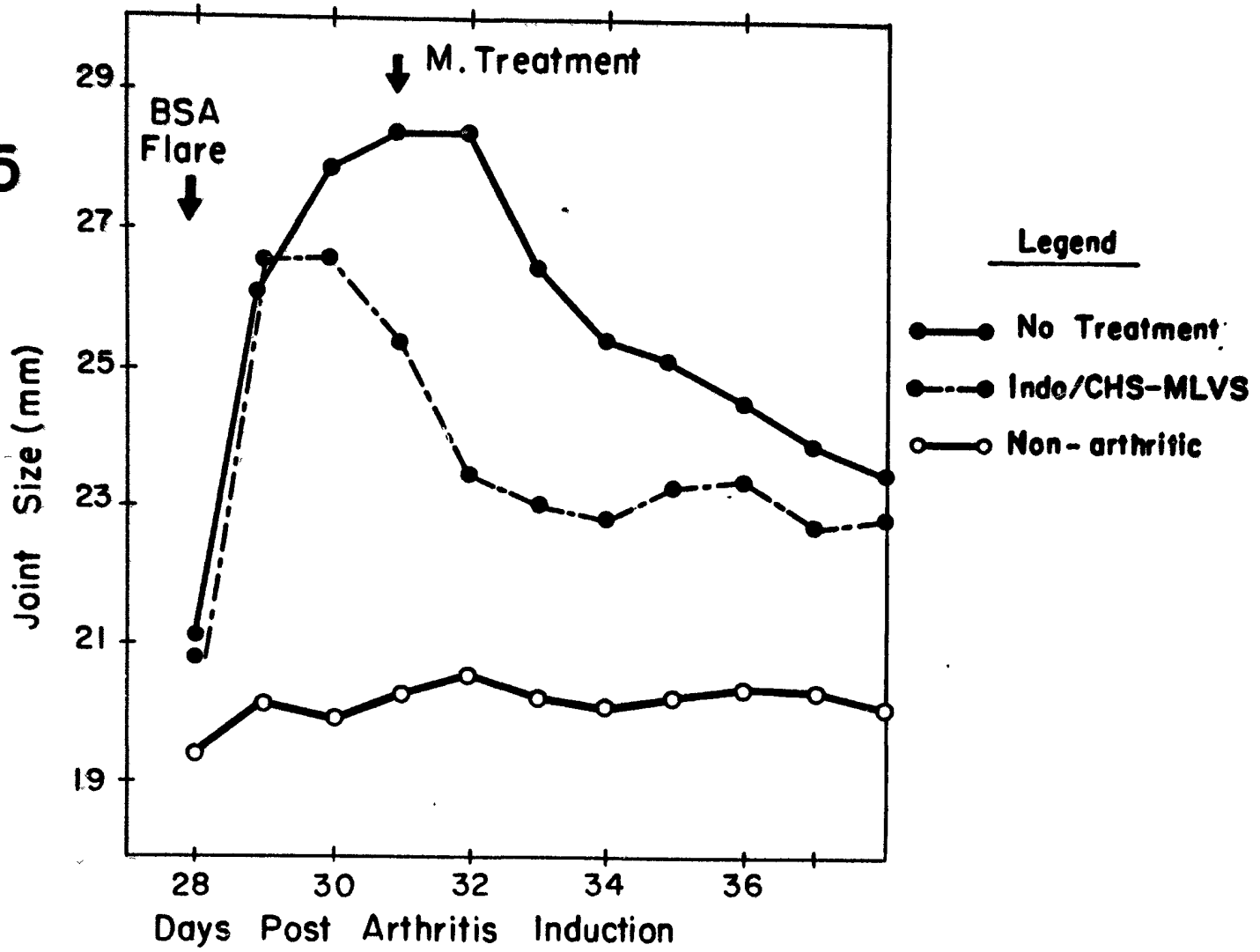

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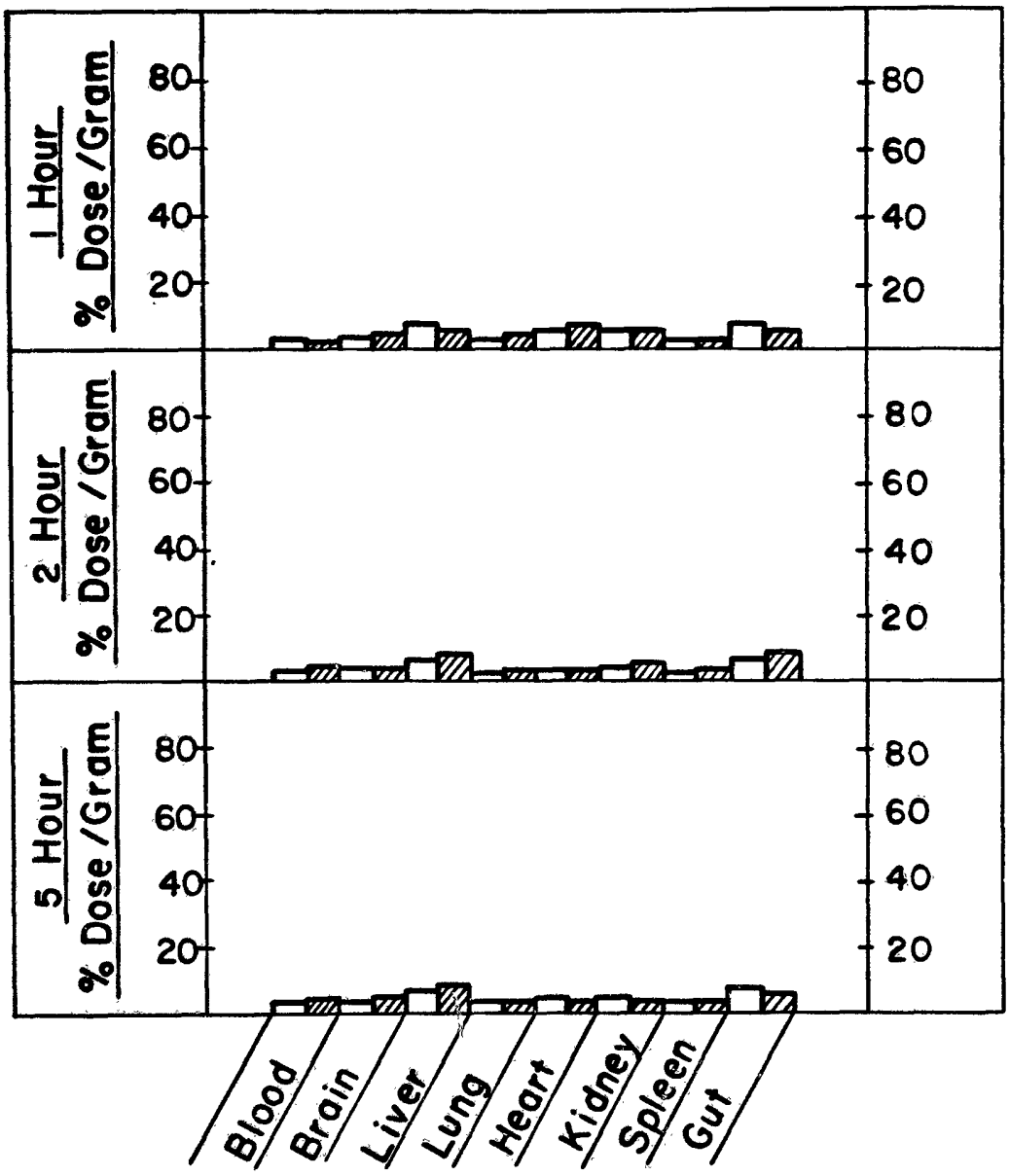
FIG. 6



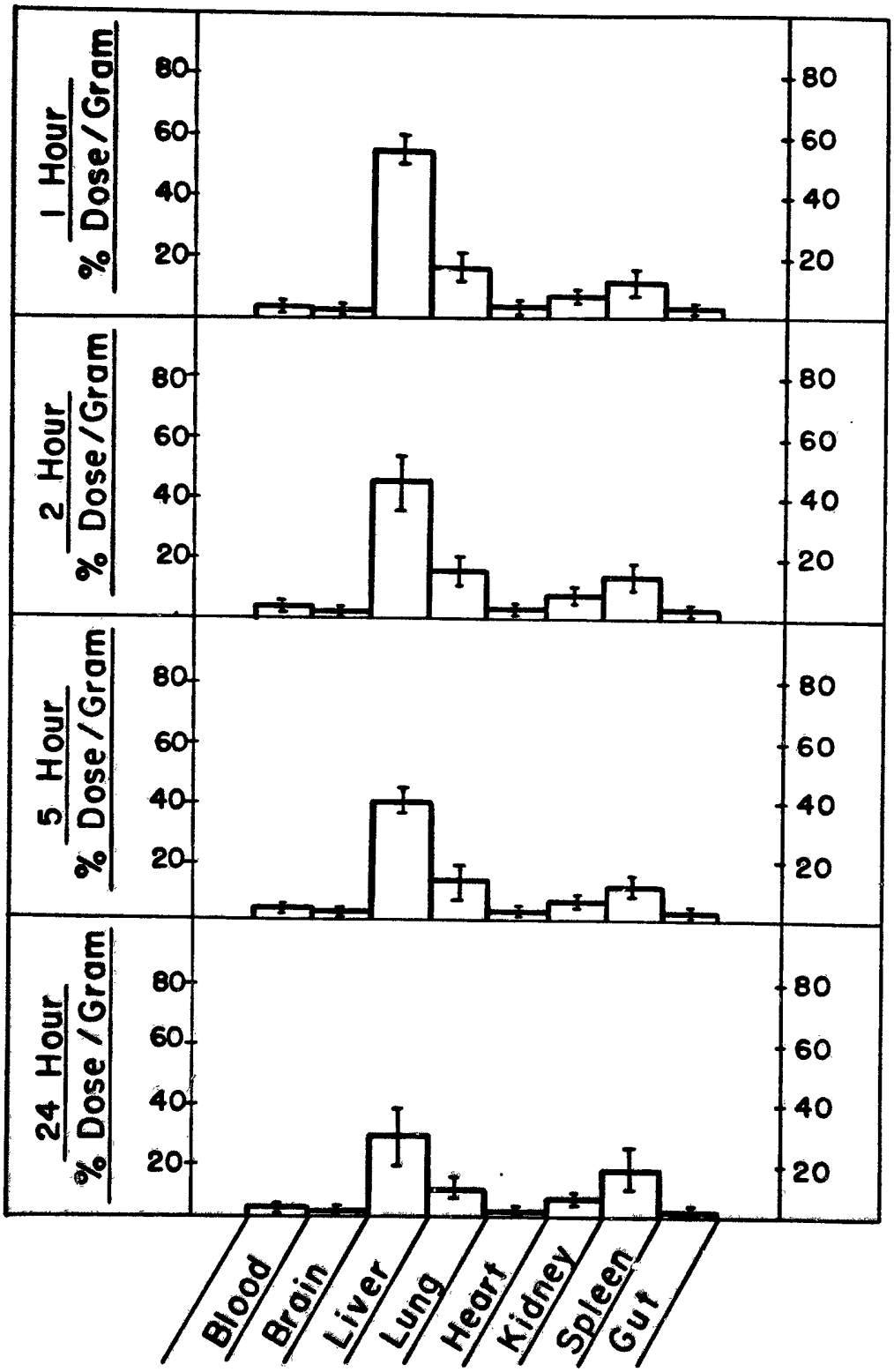
Legend

□ = ^{14}C-Diazepam In CHS-SUVS

▨ = Free ^{14}C-Diazepam

2349-083 (7-3)

FIG. 7A



2349-083 (8-9)

FIG. 7B

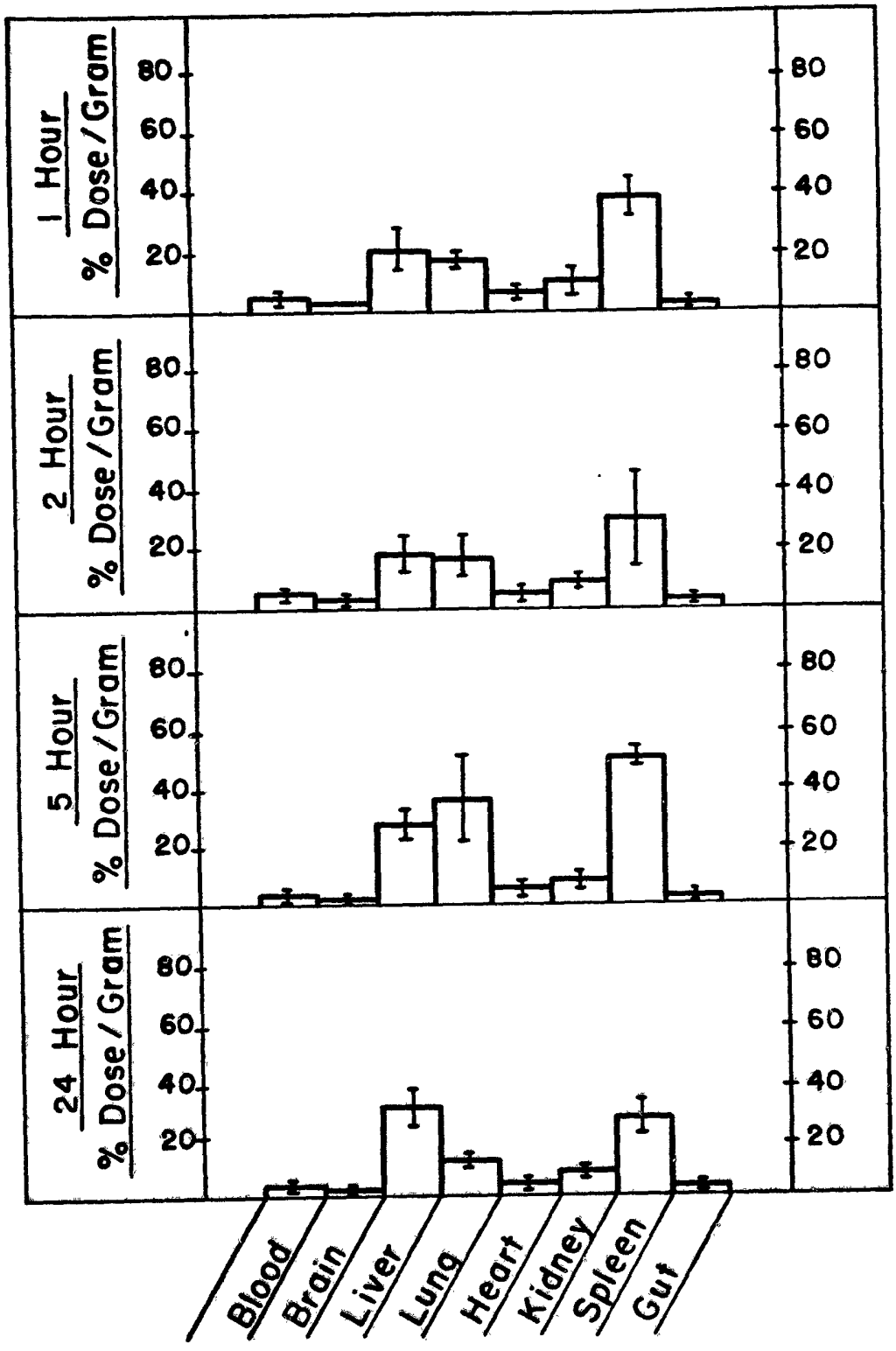
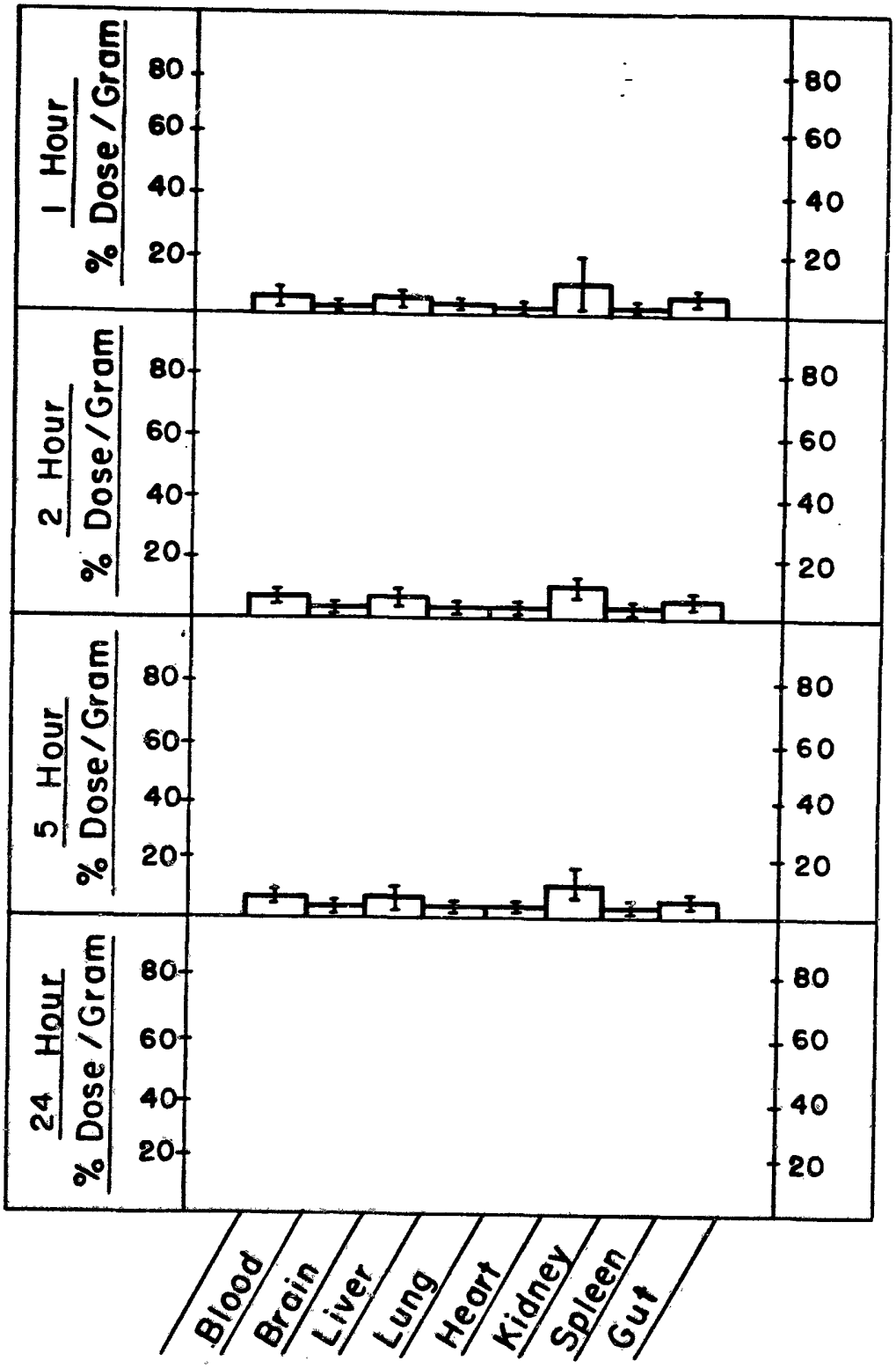


FIG. 7C


1 Hour
% Dose/Gram
2 Hour
% Dose/Gram
5 Hour
% Dose/Gram
24 Hour
% Dose/Gram
80
60
40
20
Blood
Brain
Liver
Lung
Heart
Kidney
Spleen
Gut