UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES
AND TOXIC SUBSTANCES

NOV 27 2000

CERTIFIED MAIL 7000 0600 0023 8260 0635

Carl D. D'Ruiz
Ciba Specialty Chemicals Corporation
4090 Premier Drive
High Point, N.C. 27261-2444

SUBJECT: Metabolism Toxicology Data
Irgasan
EPA Registration 70404-2

Dear Mr. D'Ruiz:

The following toxicology data have been reviewed and a brief summary is provided for:

MRID 149464 (TRID 460065-05), submitted March 1973 is not acceptable for purposes of satisfying the guideline requirement, OPPTS 870.7485 - Guideline 85-1, for a General metabolism study. However, if the deficiencies cited in the enclosed review can be corrected, these data might upgraded to acceptable.

MRID 68161 (TRID 2401510-03), submitted February 1977 is not acceptable for purposes of satisfying the guideline requirement, OPPTS 870.7485 - Guideline 85-1, for a General metabolism study. However, if the deficiencies cited in the enclosed review can be corrected, these data might upgraded to acceptable.

MRID 68163 (TRID 2401510-05), submitted February 1977 is not acceptable for purposes of satisfying the guideline requirement, OPPTS 870.7485 - Guideline 85-1, for a General metabolism study. These data cannot be upgraded.

MRID 68162 (TRID 2401510-04), submitted June, 1976 is not acceptable for purposes of satisfying the guideline requirement, OPPTS 870.7485 - Guideline 85-1, for a General metabolism study. These data cannot be upgraded.

MRID 79590 (TRID 2401510-02), submitted March 1973 is not acceptable for purposes of satisfying the guideline requirement, OPPTS 870.7485 - Guideline 85-1, for a General metabolism study. However, if the deficiencies cited in the enclosed review can be corrected, these data might upgraded to acceptable.

The Agency believes it is unlikely that existing data can be upgraded to satisfy the guideline requirement and would prefer that a new metabolism study using the mouse as the test species be conducted and submitted in accordance with the revised guideline requirement 85-1 (OPPTS 870.7485). This data requirement can not be waived and surrogate data will not be accepted. You must reply to this Office within 90 days of your receipt of this letter as to how you will satisfy the guideline requirement. If you fail to reply as required, the Agency will pursue appropriate regulatory action, including issuance of a Notice of Intent to Suspend.

Copies of our reviews are enclosed. Please refer to them for additional information regarding the evaluation of these data. If you have any questions regarding this letter, please contact Tom Luminello of my staff at (703) 308-8075.

Sincerely yours,

Robert S. Brennis
Product Manager (32)
Regulatory Management Branch II
Antimicrobial Division (7510-C)

Enclosure

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

NOV - 2 2000

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: 5-Chloro-2-(2,4-dichlorophenoxy)phenol (Triclosan): Review of Metabolism data.

EPA Identification Numbers:

P.C. Code: 054901 MRID's: 149464; 79590; 68163; 68162; 68161
DP Barcode:D266143 Submission: none

TO: Connie Welch, Chief
Robert Brennis, Product Manager / PM Team 32
Regulatory Management Branch II
Antimicrobials Division (7510C)

FROM: Timothy F. McMahon, Ph.D. 10/25/00
Senior Toxicologist
Risk Assessment and Science Support Branch (RASSB)
Antimicrobials Division (7510C)

THRU: Najm Shamim, Ph.D. Acting Team Leader 10/26/2000
Team Two
Risk Assessment and Science Support Branch (RASSB)
Antimicrobials Division (7510C)

Norm Cook, Chief 11.02.00
Risk Assessment and Science Support Branch (RASSB)
Antimicrobials Division (7510C)

Action Requested: Review of metabolism studies conducted with Triclosan in various species.

Background

The Risk Assessment and Science Support Branch (RASSB), Antimicrobials Division (AD), is in possession of Toxicology data with regard to the disposition and metabolism of Triclosan. These data have not been previously reviewed. This memorandum addresses the results of these reviews.

1) Metabolism and Pharmacokinetics - Multiple Species

CITATION: Stierlin, H. (1972). GP 41 353: Study of pharmacokinetics and metabolism in mouse, rat, rabbit, and dog. Pharma Research, Pharmacological Chemistry. CIBA-GEIGY, Ltd. Basle. Project No. GP 41 353. Report No. 33/1972. MRID 149464. December 1, 1972. Unpublished.

SPONSOR: CIBA-GEIGY Limited, Pharma Research, Pharmacological Chemistry, Basle.

EXECUTIVE SUMMARY: In a metabolism study (MRID 149464), rats, mice, rabbits, and dogs were administered [^{14}C]-GP 41 353 or [^{3}H]-GP 41 353 (radiochemical purity 99%; chemical purity 99.5%; batch/lot nos. not provided) intravenously, intraduodenally, or orally (gavage) at doses of 10 mg/kg (mice), 0.4 mg/kg (rats), 5 mg/kg (rats, rabbits, dogs), or 50 mg/kg (rabbits). Radioactivity levels in the blood, tissues, and excreta were measured for time intervals up to 168 hours. Additionally, biliary elimination was also analyzed in rats given a single intraduodenal dose.

There was no indication of any toxic effects in the test animals. Recovery of administered radioactivity at 2, 4, 6, and 8 hours ranged from 99.67 to 104.53 % in rats given a single 5 mg/kg oral dose of [^{3}H]-GP 41 353. Data were unavailable in the study report to accurately determine radioactivity inventory for the other species tested.

Absorption of the test material was reported by the study author as 70-80% (for rats) as determined by comparisons of areas under-the-blood concentration curve for oral and intravenous administrations. These data were, however, unavailable for the preparation of this data evaluation report. Absorption in rats could also be estimated based upon biliary and urinary elimination data. Over a 7.5 to 10-hour period, biliary elimination accounted for 62.5% of a 5 mg/kg gavage dose and 67% of a 5 mg/kg intraduodenal dose while urinary excretion at 6 and 8 hours accounted for 76.60% and 5.17%, respectively, of a 5 mg/kg gavage dose. These data

suggest that absorption is in excess of 70%. Absence of biliary elimination data for the other test species precluded assessment of the extent of absorption in those species; however, biliary excretion data in rats and mice suggest enterohepatic recirculation of the test material in both species. Time-course concentration data revealed that peak blood levels occurred within 30 minutes in rats following a single oral or intravenous dose of 5 mg/kg and at 2-4 hours for dogs.

Tissue distribution patterns were similar among mice and rats, and exhibited only slight quantitative variability for intravenous versus oral dosing. Following a single oral dose in rats, radioactivity in tissues was low (generally <1 μg/g tissue) with the exception of blood and the organs associated with excretory function (e.g., liver, gall bladder, kidneys). Following an intravenous dose to rats and mice, tissue levels were also greatest in highly perfused organs or those associated with excretory function. Based upon data from rats, tissue burdens declined appreciably over 24 hours with no indication of accumulation/sequestration.

Excretion of GP 41 353 was examined in two strains of rats, rabbits, and dogs. Biliary elimination was also assessed in rats. Urinary excretion appeared to be a minor route of elimination in rats and dogs, accounting for 3-17% of the administered oral dose in rats over a 168-hour period, and 8.3-8.8% in dogs over a 120-hour period. Urinary elimination in dogs was somewhat greater following intravenous administration; 12.9-17.7% over 120 hours. For rabbits, urinary excretion was a significant route of elimination and accounted for 74.1% of a single 50 mg/kg oral dose and 60.4% of a single 5 mg/kg oral dose over a 72-hour period. The biliary secretion data in rats showed that most of the fecal radioactivity could be attributed to biliary elimination products rather than unabsorbed test material. Biliary elimination was also affirmed by data from the mouse showing very high concentrations of radioactivity in the gall bladder at 5 minutes to two hours following a 10 mg/kg, i.v. dose.

Analysis of bile samples from the rats indicated that the test material underwent Phase II biotransformation. Treatment of samples with β-glucuronic acid revealed that most of the biliary product was glucuronide conjugates while some was unchanged parent compound.

The results of this multi-species study indicated that at least 70% of an oral dose of GP 41 353 is absorbed from the gastrointestinal tract and that biliary excretion and subsequent fecal elimination is a major excretory route in the rat and dog. Urinary excretion appeared to be a major route of elimination in the rabbit. Tissue accumulation was minimal after a single dose and was primarily associated with highly perfused tissues and organs with excretory function, although a companion study (MRID 68161) shows distribution of triclosan-derived radioactivity into the pituitary gland, optic nerve, and sciatic nerve at concentrations equivalent to those found in highly perfused organs at the same dose level (5 mg/kg). Repeated oral dosing at 5 mg/kg leads to accumulation of radioactivity in the blood, plasma, pituitary gland, sciatic nerve, and kidneys (MRID 68161). Metabolite data in rats revealed glucuronide conjugates and unchanged parent compound as biliary metabolites.

This metabolism study is **unacceptable/upgradable** and does not currently satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Numerous deficiencies were observed in this report, including information regarding dose confirmation, homogeneity, and stability, environmental and housing conditions of the animals used in this study, and a lack of statements regarding compliance with Good Laboratory Practice and Quality Assurance. If the missing information can be provided, this study can be upgraded. Otherwise, a new guideline metabolism study will need to be conducted. Despite the deficiencies, the data do provide useful information on the disposition of Triclosan in multiple species and can be considered in conjunction with an acceptable metabolism study if such study is necessary.

2) Metabolism and Pharmacokinetics - Human

EXECUTIVE SUMMARY: In a metabolism and disposition study (MRID 68163), ^{14}C-labelled Triclosan (batch/lot nos. not provided; >99% radiochemical purity) was administered as a single oral dose (200.3 mg/kg; 100 μCi) to a human volunteer (male, 48 years of age).

Recovery of administered radioactivity and evaluation of mass balance was not possible because tissue burdens and overall body burden could not be assessed and biliary contribution to excretion were not measured. Recovery of radioactivity in the urine implied 73.79% elimination of the administered dose over 72 hours. The majority of urinary elimination occurred during the first 24 hours and appeared to be nearly complete by 72 hours. Assuming residual radioactivity residing in the blood and other tissues at the terminal time point, absorption of >74% may be inferred for the 200 mg/kg dose.

Treatment of plasma and urine with β-glucuronidase and arylsulfatase revealed that the majority of the radioactivity in these matrices could be attributed to conjugation products rather than unchanged parent compound. For urine, 93.85% of the recovered radioactivity could be attributed to glucuronide conjugates while approximately 2.3% could be attributed to sulfate conjugates. For plasma, 51.87- 60.62 % of the radioactivity could be attributed to glucuronide conjugates while enzymatic hydrolysis revealed that the sulfate conjugate represented 31.61-43.50% of the plasma radioactivity. Minor changes in the relative contributions of these metabolites over time were detected but of uncertain significance.

In summary, this report provides anecdotal data in humans indicating that orally administered triclosan undergoes Phase II metabolism to conjugation product metabolites (sulfate and glucuronide conjugates) which are eliminated via the urine within 72 hours.

This metabolism study using a human volunteer is **Acceptable/Nonguideline** but does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Although properly conducted and providing data regarding the metabolism of triclosan in a human volunteer following a single oral dose, the data were anecdotal, the protocol did not include tissue distribution assessments, and no information was provided regarding dose stability and confirmation or homogeneity. Therefore, the study was not consistent with an 85-1 Guideline study. The results, however, do affirm findings of companion studies (MRID 149464, 79590) in other animal species and provide important information based upon human data.

3) Metabolism and Pharmacokinetics - Human

CITATION: Stierlin, H., R. Murbach, W. Theobald (1976). GP 41 353: Pharmacokinetic and metabolic studies in man following oral administration of a ^{14}C-labelled preparation. Pharma Research, Pharmacological Chemistry. CIBA-GEIGY, Ltd. Basle. Report No. B 6/1976. MRID 68162. January 27, 1976. Unpublished

SPONSOR: CIBA-GEIGY Limited, Pharma Research, Pharmacological Chemistry, Basle.

EXECUTIVE SUMMARY: In a metabolism and disposition study (MRID 68162), ^{14}C-labelled Triclosan (batch/lot nos. not provided; >99% radiochemical purity) was administered as a single oral dose (203.57 mg/kg; 100 μCi) to a human volunteer (male, 43 years of age).

Recovery of administered dose was 90% based upon radioactivity in the urine and feces. A complete assessment was not possible in the absence of tissue distribution/burden data. Absorption could be estimated as being at least 57% based upon urinary excretion but is not definitive in the absence of biliary secretion data. Recovery of radioactivity in the urine indicated that 57 % of the administered radioactivity was excreted over 144 hours. The majority of urinary elimination (69%) occurred during the first 24 hours and appeared to be nearly complete within 120 hours. Fecal elimination accounted for 33.5% of the administered dose over a 144-hour period with most elimination (90%) occurring within 48 hours.

For both plasma and urine, very little (1-3%) of the radioactivity was associated with free triclosan. The majority of the radioactivity could be attributed to glucuronide conjugates or other nonspecified conjugates as determined by β-glucuronidase hydrolysis or acid hydrolysis, respectively.

7

In summary, this study provides anecdotal information regarding urinary and fecal excretion, metabolite characterization, and blood/plasma time-course of triclosan in a human volunteer following a single oral dose. The results of both the excretion assessment and metabolite quantitation/characterization studies are consistent with other reports in a human subject (MRID 68163) or in laboratory species (MRID 149464, 79590).

This metabolism study using a human volunteer is **Acceptable/Nonguideline** but does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Although properly conducted and providing data regarding the excretion, metabolism, and plasma/blood kinetics of triclosan in a human volunteer following a single oral dose, there was no statistical base and the protocol was not consistent with a 85-1 Guidelines (no tissue distribution data). The results, however, do affirm findings of companion studies (MRID 149464, 79590) in animal species and another study (MRID 68163) with a human volunteer.

4) Metabolism and Pharmacokinetics - Rats, Dogs, and Baboons

CITATION: Stierlin, H., K. Schmid, A. Sutter (1977). GP 41 353: Comparison of pharmacokinetic and metabolic parameters of triclosan and HCP in the mouse, rat, beagle dog and baboon. Part A. Survey of findings. Part B. Detailed account of the study. Pharma Research, Pharmacological Chemistry. CIBA-GEIGY, Ltd. Basle. Report No. B 1/1977. MRID 68161. January 27, 1977. Unpublished

SPONSOR: CIBA-GEIGY Limited, Pharma Research, Pharmacological Chemistry, Basle.

EXECUTIVE SUMMARY: In a metabolism study (MRID 68161), ^{14}C-triclosan (>98% radiochemical purity, lot/batch no. not specified) was administered to male rats by gavage at doses of 5 or 30 mg/kg (single or 14-day repeated). In addition, two male beagle dogs and two male baboons received single 5 mg/kg doses in gelatine capsules and 10 male mice were given a single 10 mg/kg intravenous dose. Blood levels were monitored in mice up to 2 hours, in rats at 24 hours postdose, in the dogs up to 72 hours and in monkeys up to168 hours postdose. Urinary and fecal excretion was monitored in monkeys and dogs for 6-10 days. Tissue distribution was assessed in mice and rats.

There were no test article-related toxic effects reported. Recovery of administered radioactivity was marginal; 86.9% and 74.1% for each of two dogs, and 83.1% and 80.5% for each of two monkeys. Administered radioactivity was widely distributed among tissues/organs in mice after an intravenous injection (10 mg/kg) and in rats following a single oral administration or the last dose of a 14-day repeated oral administration (5 or 30 mg/kg). Based on the radioactivity

distribution data, there was no evidence indicating sequestration of the test material or its metabolites in either mice or rats.

Time-course analysis of blood/plasma radioactivity in rats revealed that peak concentrations were attained within three hours after a single oral dose of 5 mg/kg and that the concentrations declined approximately five-fold within 24 hours. For dogs, somewhat greater blood t_{max} values were observed but were variable (2- 8 hours) for the two dogs tested. Generally, the test article did not exhibit especially rapid partitioning into or clearance from the blood for either species. Time-course analysis of tissues from mice given a single intravenous dose (10 mg/kg) of ^{14}C-triclosan showed that the highest concentrations of radioactivity were initially associated with highly perfused organs/tissues. These levels significantly declined within 30 minutes but the decline was somewhat less rapid for organs /tissues associated with metabolism and elimination and notably increased for the gall bladder.

Both urinary and fecal elimination were identified as major routes of excretion. The relative contribution of each to overall elimination of administered radioactivity exhibited species variability. For monkeys, urinary excretion accounted for 56.69% of the administered dose and fecal elimination accounted for 25.15%. For dogs, urinary and fecal elimination accounted for 12.16% and 68.30%, respectively, of the administered dose. In both species, most of the urinary and fecal excretion occurred within 48 hours.

Definitive characterization of metabolites was not performed. Preliminary investigations using acid and enzyme hydrolysis, indicated that very little (<1%) of the blood/plasma radioactivity was associated with unchanged triclosan. In the brains of rats, however, 35-50% of the radioactivity was attributed to parent compound.

Overall, this study demonstrated that ^{14}C-triclosan is readily absorbed from the gastrointestinal tract, has a potential wide volume of distribution, and can cross the blood-brain barrier. Blood absorption and clearance is not especially rapid, but the compound does not appear to undergo sequestration in the species tested. Most of the circulating radioactivity was attributed to metabolism products (probably conjugation products based upon preliminary experiments using acid and enzymatic hydrolysis). Both the urine and the feces are significant routes of excretion with the relative importance appearing to be species dependent.

This metabolism study in multiple species, which predates GLP guidelines, is **Unacceptable/ upgradable** and does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. There are numerous deficiencies in this study , including test article characterization, environmental and housing conditions of the experimental animals, dose solution information (stability, homogeneity, confirmation of doses received), and lack of mass balance information. Additionally, a quality assurance statement was not provided. If these data can be provided, the study can be upgraded to acceptable. Otherwise, a new guideline metabolism study will need to be conducted.

5) Metabolism and Pharmacokinetics - Dogs and Baboons

CITATION: Stierlin, H. (1976). GP 41 353: Isolation and identification of the main metabolites in the blood of the beagle and baboon and in the urine of the latter following oral administration of ^{14}C-labelled triclosan. Pharma Research, Pharmacological Chemistry. CIBA-GEIGY, Ltd. Basle. Report No. B 14/1976. MRID 79590. March 16, 1976. Unpublished.

SPONSOR: CIBA-GEIGY Limited, Pharma Research, Pharmacological Chemistry, Basle.

EXECUTIVE SUMMARY: In a metabolism study (MRID 79590), two baboons and two dogs were administered a single oral dose (5 mg/kg) of [^{14}C]-GP 41 353 (radiochemical purity 99%; chemical purity 99.5%; batch/lot nos. not provided). Blood samples were taken at 3 hours postdosing from one dog and at 8 and 12 hours postdosing from one baboon. Another dog and baboon were killed at 6 and ~7 hours postdosing to obtain sufficient blood samples for metabolite characterization. Urine samples were collected from the dog and baboon that were not sacrificed for blood sample acquisition.

There were no adverse effects associated with the test article. At 3 hours postdosing, total radioactivity was 6.08 μg eq./mL blood in the dog. For the baboon total radioactivity was 1.24 and 1.03 μg eq./mL blood, respectively at 8 and 12 hours postdosing. For the dog and baboon terminated for sample acquisition, total blood radioactivity was 4.86 μg eq./mL blood for the dog at 6 hours and 2.29 μg eq./mL blood for the baboon at ~7 hours postdosing. Urinary elimination accounted for 32% of the administered dose to the surviving baboon by 72 hours. Urinary excretion by the dog was minimal and accounted for "only a few percent of the administered dose".

Hydrolysis of the biological samples with arylsulfatase and β-glucuronidase resulted in the release of free triclosan indicating that the measured radioactivity was associated with sulfate and glucuronide conjugation products. For the dog, glucuronide conjugates accounted for 7% of the blood radioactivity and sulfate conjugates accounted for 88% of the radioactivity sampled at three hours. At six hours, 7.2% of the radioactivity in the blood was associated with glucuronide conjugates, and the remaining percentage was not identified except through combined glucuronidase/sulfatase hydrolysis (86.3%). For the baboon, glucuronide conjugates represented about 25% and sulfate conjugates represented about 33% of the circulating radioactivity at 8 hours postdosing. Similar analysis with blood collected at 12 hours postdosing, revealed somewhat greater amounts of sulfate conjugates (~43.9%) and less glucuronide conjugate (10.6%). For the baboon, analysis of 0-72 hour urine samples revealed that approximately 6% of the urinary radioactivity was due to unchanged triclosan. Up to 75% of the urinary radioactivity

underwent spontaneous hydrolysis presumably due to endogenous urinary β-glucuronidase. No analyses were conducted for the dog due to the minimal urinary elimination of radioactivity

In summary the results of this study showed that the major blood metabolites in the baboon and beagle dog were sulfate and glucuronide conjugation products. The major urinary metabolite in the baboon was a glucuronide conjugate but analysis of urinary metabolites in the dog was precluded by negligible urinary products as determined by radioactivity.

This metabolism study is **unacceptable/upgradable** and does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Although the study appears to have been properly conducted and provided data regarding the characterization of blood and urinary metabolites in the baboon and blood metabolites in the dog following a single oral dose, several deficiencies exist, including lack of test article characterization, lack of animal husbandry and environmental control information, lack of dosing administration, and lack of dose solution stability, homogeneity, and dose confirmation. If these data can be provided, this study can be upgraded to acceptable. Otherwise, a new metabolism study must be conducted.

Conclusions

RASSB has reviewed submitted studies on the metabolism and disposition of Triclosan in various species. Various aspects of the metabolism and disposition of ^{14}C- triclosan in multiple species including human volunteers were evaluated: MRID 149464 (rats, mice, rabbits, dogs), MRID (baboons and dogs), MRID 68161 (baboons, dogs, rats, mice), MRID 68162 (human volunteer), MRID 68163 (human volunteer). Tritiated triclosan was also utilized in MRID 149464.

Species variability was observed in primary excretory routes and the relative contributions of the two routes to overall elimination of administered radioactivity. Based upon biliary excretion data in rats, 63-67% of the radioactivity in feces can be attributed to biliary secretion products. The data also revealed that the test material and/or metabolites has a large volume of disposition following oral or intravenous administration but there was no evidence of sequestration in any tissues or organs. In the human, rat, dog, and monkey, triclosan appears to be metabolized primarily to glucuronide and sulfate conjugates.

Taken in total, the data generated from these studies reports do not satisfy the requirements for a metabolism/pharmacokinetics study. Numerous deficiencies exist with these data. If the deficiencies cannot be addressed, new metabolism data will be required in order to satisfy the guideline. Despite the deficiencies, the data do provide useful insights into the dispositional behavior of triclosan in several species including humans. Of note is the ability of triclosan to cross the blood brain barrier, the apparent accumulation of triclosan in certain tissues after repeated oral dosing, and the enterohepatic recirculation of the chemical in rats and mice.

It should be noted that there is a relative lack of information regarding the disposition of triclosan in the mouse, a species which has shown a positive carcinogenic response to this chemical. However, data are available to allow some comparison of plasma levels of total triclosan in mice (from an FDA review of a mouse carcinogenicity study with triclosan) and rats (EPA reviewed data summarized above) after comparable doses (10 mg/kg and 5 mg/kg dose levels respectively). Mouse data were obtained as a single undefined time point (blood obtained after 6 or 18 months of treatment with the chemical), while rat data were obtained as time-course data after a single oral dose. Whole blood radioactivity was determined in rats through combustion of blood samples, while the technique used to determine radioactivity in mouse plasma was not stated. After oral administration of a 10 mg/kg dose in mice in the diet for 6 months, the mean plasma concentration of total triclosan was 16.8 μg/ml in males and 21.9 μg/ml in females, while in rats, blood concentration was highest at 30 minutes after oral administration and was reported as 4.0 μg/ml. While not conclusive, these results point to potentially significant differences in the blood and/or plasma levels of triclosan-derived radioactivity between rats and mice, with mice showing much higher levels of the compound than rats.

There are unanswered questions with regard to disposition and metabolism of triclosan which would provide information helpful to determining the potential species difference in carcinogenic response. Specifically, time course data on fecal and urinary elimination of triclosan in the strain of mouse showing a carcinogenic response (CD-1 strain) would assist in comparing to rats and other species. Rate(s) of biotransformation in liver and specific metabolic products in the liver and blood would also provide significant information to assist in the determination of the apparent sensitivity of mice to the carcinogenic effects of triclosan. This information is important, as previous work on the biochemical toxicology of triclosan (MRID #'s 44389702; 44389703) has shown similar responses of the liver of rats and mice to administration of triclosan, while the carcinogenic response appears to be confined to mice of the CD-1 strain based on available information. Limited data on comparison of blood and plasma data in rats and mice suggest significant differences in levels of triclosan-derived radioactivity in the blood and/or plasma which could be related to the difference in carcinogenic response of rats and mice. However, these types of data are not part of a standard metabolism study, and unless obtained through a specific request by the Agency, inferences will have to be made from the existing metabolism data.

DATA EVALUATION REPORT

TRICLOSAN

Study Type: METABOLISM AND PHARMACOKINETICS
[OPPTS 870.7485 (§85-1)]
MRID 68161

Prepared for

Antimicrobials Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. T07

Primary Reviewer: Robert A. Young, Ph.D., D.A.B.T.	Signature:	Robert A. Young
	Date:	OCT 0 6 2000
Secondary Reviewers: H. T. Borges, MT(ASCP),Ph.D., D.A.B.T.	Signature:	H. T. Borges
	Date:	OCT 0 6 2000
Robert H. Ross, M.S., Group Leader	Signature:	Robert H. Ross
	Date:	OCT 0 6 2000
Quality Assurance: Lee Ann Wilson, M.A.	Signature:	L. A. Wilson
	Date:	OCT 0 6 2000

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UTBattelle, LLC, for the U.S. Dept. of Energy under contract DEAC0500OR22725

EPA Reviewer: Tim McMahon, Ph.D.
Antimicrobials Division

_____, Date 10/25/00

DATA EVALUATION RECORD

STUDY TYPE: Metabolism - Multiple Species; OPPTS [870.7485 (§85-1)]

DP BARCODE: D266143
P.C. CODE: 054901

SUBMISSION CODE: None
TOX. CHEM. NO.: None

TEST MATERIAL (PURITY): Triclosan (radiochemical purity >99%)

SYNONYMS: GP 41 353; Triclosan;5-chloro-2-(2,4-dichlorophenoxy)phenol

CITATION: Stierlin, H., K. Schmid, A. Sutter (1977). GP 41 353: Comparison of pharmacokinetic and metabolic parameters of triclosan and HCP in the mouse, rat, beagle dog and baboon. Part A. Survey of findings. Part B. Detailed account of the study. Pharma Research, Pharmacological Chemistry. CIBA-GEIGY, Ltd. Basle. Report No. B 1/1977. MRID 68161. January 27, 1977. Unpublished

SPONSOR: CIBA-GEIGY Limited, Pharma Research, Pharmacological Chemistry, Basle.

EXECUTIVE SUMMARY: In a metabolism study (MRID 68161), ^{14}C-triclosan (>98% radiochemical purity, lot/batch no. not specified) was administered to male rats by gavage at doses of 5 or 30 mg/kg (single or 14-day repeated). In addition, two male beagle dogs and two male baboons received single 5 mg/kg doses in gelatine capsules and 10 male mice were given a single 10 mg/kg intravenous dose. Blood levels were monitored in mice up to 2 hours, in rats at 24 hours postdose, in the dogs up to 72 hours and in monkeys up to168 hours postdose. Urinary and fecal excretion was monitored in monkeys and dogs for 6-10 days. Tissue distribution was assessed in mice and rats.

There were no test article-related toxic effects reported. Recovery of administered radioactivity was marginal; 86.9% and 74.1% for each of two dogs, and 83.1% and 80.5% for each of two monkeys. Administered radioactivity was widely distributed among tissues/organs in mice after an intravenous injection (10 mg/kg) and in rats following a single oral administration or the last dose of a 14-day repeated oral administration (5 or 30 mg/kg). Based on the radioactivity distribution data, there was no evidence indicating sequestration of the test material or its metabolites in either mice or rats.

Time-course analysis of blood/plasma radioactivity in rats revealed that peak concentrations were attained within three hours after a single oral dose of 5 mg/kg and that the concentrations declined approximately five-fold within 24 hours. For dogs, somewhat greater blood t_{max} values

/ 5

were observed but were variable (2- 8 hours) for the two dogs tested. Generally, the test article did not exhibit especially rapid partitioning into or clearance from the blood for either species. Time-course analysis of tissues from mice given a single intravenous dose (10 mg/kg) of ^{14}C-triclosan showed that the highest concentrations of radioactivity were initially associated with highly perfused organs/tissues. These levels significantly declined within 30 minutes but the decline was somewhat less rapid for organs /tissues associated with metabolism and elimination and notably increased for the gall bladder.

Both urinary and fecal elimination were identified as major routes of excretion. The relative contribution of each to overall elimination of administered radioactivity exhibited species variability. For monkeys, urinary excretion accounted for 56.69% of the administered dose and fecal elimination accounted for 25.15%. For dogs, urinary and fecal elimination accounted for 12.16% and 68.30%, respectively, of the administered dose. In both species, most of the urinary and fecal excretion occurred within 48 hours.

Definitive characterization of metabolites was not performed. Preliminary investigations using acid and enzyme hydrolysis, indicated that very little (<1%) of the blood/plasma radioactivity was associated with unchanged triclosan. In the brains of rats, however, 35-50% of the radioactivity was attributed to parent compound.

Overall, this study demonstrated that ^{14}C-triclosan is readily absorbed from the gastrointestinal tract, has a potential wide volume of distribution, and can cross the blood-brain barrier. Blood absorption and clearance is not especially rapid, but the compound does not appear to undergo sequestration in the species tested. Most of the circulating radioactivity was attributed to metabolism products (probably conjugation products based upon preliminary experiments using acid and enzymatic hydrolysis). Both the urine and the feces are significant routes of excretion with the relative importance appearing to be species dependent.

This metabolism study in multiple species, which predates GLP guidelines, is **Unacceptable/ upgradable** and does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. There are numerous deficiencies in this study , including test article characterization, environmental and housing conditions of the experimental animals, dose solution information (stability, homogeneity, confirmation of doses received), and lack of mass balance information. Additionally, a quality assurance statement was not provided. If these data can be provided, the study can be upgraded to acceptable. Otherwise, a new guideline metabolism study will need to be conducted.

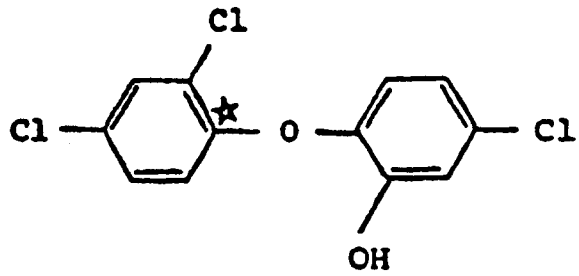
COMPLIANCE: Signed and dated, Quality Assurance, and Data Confidentiality statements were not included with all studies. Flagging statements were not included. The study predates Good Laboratory Practice guidelines.

I. MATERIALS AND METHODS

A. MATERIALS

1. Test compound

Radiolabeled: [^{14}C]-GP 41 353 (sp. act. not specified)
Batch No.: not provided
Purity:
Radiochemical purity: >98%
Chemical purity: not provided
Description: not available
Contaminants: none noted
CAS No.: not available
Structure:



[^{14}C]-GP 41 353

2. Vehicle

For mice, the test article was dissolved in 0.1N sodium bicarbonate for intravenous administration. For gavage dosing in rats, triclosan was suspended in 2% carboxymethylcellulose. For oral dosing of dogs and baboons, the test article was administered in a gelatin capsule.

3. Test species

Species: mouse, rat, dog, monkey (all males)
Strain: mouse (MA 01 SPF); rat (RA 25); dog (beagle), monkey (baboon)
Age and weight at study initiation: mice (20 g); rats (160-210 g); dogs (11 kg); monkeys (6 and 7 kg); ages of animals not specified
Source: Sisseln breeding unit (mice, rats), not specified (dogs, monkeys)
Housing: no information provided
Diet: no information provided
Water: no information provided
Environmental conditions:
Temperature: no information provided
Humidity: no information provided
Air changes: no information provided
Photoperiod: no information provided
Acclimation period: no information provided

4. Preparation of dose

For the i.v. administration in mice, the test article (0.2 mg) was dissolved in 0.1 mL of 0.1N sodium bicarbonate. For the gavage dose in rats, triclosan was suspended in carboxymethylcellulose to provide doses of 5 or 30 mg/kg in 0.5 mL volume. For the dogs and monkeys, the triclosan was administered in gelatin capsules.

Results –
Homogeneity: No data provided.

Stability: No data provided.

Dose confirmation: No data provided.

B. STUDY DESIGN AND METHODS

1. Group arrangements

The study were groups were established as shown in Table 1. No information was provided regarding assignment of animals to the groups.

TABLE 1. Study design			
Experimental group	**Dose (mg/kg)**	**Number/Sex**	**Remarks**
Mice; i.v.	10	2 males/time period	tissue distribution, whole-body autoradiography; two mice per time period (1 min., 5 min., 30 min., 2 hrs)
Rats; oral	5 30 30 5 5	4 males 4 males 4 males 4 males 3 males/time period	all animals in both dose groups killed at 24 hours to assess tissue distribution 14-day repeated dose study; all animals in both dose groups killed 24 hours after last dose for tissue distribution assessment 3 animals killed at 3, 6, and 24 hours for assessment of brain and plasma concentrations
Monkeys; oral	5	2 males	Blood collected at 2, 8, 12, 24, 48, 74 and 96 hours for radioactivity and metabolite assessments; urine collected for 10 days and feces collected for 11 days;
Dogs; oral	5.0	2 males	Blood collected at 1, 2, 8, 12, 24, 48, 72 and 168 hours for radioactivity and metabolite assessments; urine and feces collected daily for 6 days

Information taken from pp. 23-24, MRID 68161

2. Dosing and sample collection

The following samples were collected:

Blood - Blood was collected from the monkeys at 2, 8, 12, 24, 48, 74 and 96 hours and from the dogs at 1, 2, 4, 8, 12, 24, 48, 72 and 168 m hours for radioactivity and metabolite assessments. For the comparison of radioactivity levels in the blood and brain of rats, blood samples were taken by cardiac puncture at 3, 6, and 24 hours. No further details were provided.

Urine - Urine was collected daily from dogs for six days and daily from monkeys for 10 days.

Feces - Feces were collected from the monkeys for 11 days and from dogs for six days.

3. Sample preparation/analysis

Blood/plasma - Radioactivity was determined in all samples. The 2, 4, 8, and 24-hour samples were analyzed for free and bound triclosan and also subjected to enzyme hydrolysis for assessment of conjugation products. Blood/plasma samples were oxidized prior to scintillation counting. Radioactive compounds from plasma samples

were adsorbed on Amberlite XAD-2 neutral resin and subsequently analyzed as described below.

Tissues - Brain tissue was homogenized in 50 mL of water and 0.5 mL aliquots were incubated with 1 mL Soluene for 2 hours at 40°C.

Urine - Radioactivity was determined in all urine samples. Urine samples up to 72 hours were used for assessment of unchanged and conjugation product metabolites. Individual fractions were also extracted and chromatographed. Samples were added directly to the scintillation fluid and counted. Urinary metabolites were separated and concentrated using gel filtration as described below.

Feces - Fecal samples were lyophilized and analyzed for radioactivity by LSC.

4. Analytical techniques

Liquid scintillation counting (LSC) - LSC was performed using Diotol scintillation fluid and a Packard Model 3380 scintillation counter. Blood/plasma and feces were combusted prior to counting. Tissues were dissolved in Soluene and, where necessary, bleached with hydrogen peroxide. No information was provided regarding quench correction, counting efficiency, background count limits, or number of samples counted.

Derivatization and structural analysis - Enzymatic hydrolysis of samples with β-glucuronidase and arylsulfatase was used to generate free triclosan.

5. Statistics and calculations

No information was provided regarding statistical analysis.

II. RESULTS

A. DISTRIBUTION/EXCRETION STUDIES

1. Mass balance

Data were not available to allow for an accurate accounting of administered radioactivity. Based on radioactivity excreted in the urine and feces, recovery (% of administered dose) was 86.9% and 74.1% for each of two dogs, and 83.1% and 80.5% for each two monkeys.

2. Absorption

An incomplete estimate of absorption may be implied from urinary excretion data following oral dosing in the dogs and monkeys. In these experiments, urinary

excretion in dogs accounted for 10.22% and 14.17% of the administered dose over a 240- hour period. In monkeys, urinary excretion accounted for 53.05% and 60.33% of the dose over a 264-hour period. It is likely that at least some fecal excretion of radioactivity may represent absorbed dose due to the presence of conjugation products but accurate assessment of the contribution is not possible without complementary biliary secretion data.

Based upon blood concentration data in mice, the absorption rate is rapid as notable levels of radioactivity were detected at 1-5 minutes that represented compound concentrations of 13 μg/g.

3. Excretion

Both the urine and feces were notable routes of excretion in the dogs and monkeys. Excretion time-course data for two dogs and two monkeys each given a single oral dose of 5 mg/kg are shown in Table 2. Although some individual variability was detected between individuals of a given species, it could be attributed to normal variability in physiologic function. There appeared to be a species-related quantitative difference in excretory pattern; urinary elimination was more prevalent in monkeys while fecal elimination was more prevalent in dogs. For both species, elimination was not rapid. Although most (~52-73%) of the elimination occurred within 48 hours, levels of radioactivity were detectable in both urine and feces of both dogs at 96 hours and in the urine of monkeys up to 168 hours post dosing.

21

TABLE 2. Time course for excretion of radioactivity (% of administered dose[a]) by dogs and monkeys following a single oral dose (5 mg/kg) of ^{14}C-triclosan.

Time interval (hrs)	Monkeys		Dogs	
	Urine	Feces	Urine	Feces
0-24	43.36	6.81	4.09	25.50
24-48	8.84	14.35	3.73	18.87
48-72	3.20	4.00	3.04	13.88
72-96	0.73	-	0.82	7.54
96-120	0.25	-	-	-
120-144	0.20	-	0.51 (96-144 hrs)	2.47
144-168	0.12	-	-	-
168-192	-	-	-	-
192-216	-	-	-	-
216-240	-	-	-	-
240-268	-	-	-	-
Total	56.69	25.15	12.20	68.30

[a] Average of two animals

Data taken from Tables 8 and 10, pp. 37 and 40, MRID 68161.

4. Tissue distribution

Tissue distribution was assessed in mice following a single intravenous administration of a single 10 mg/kg dose of ^{14}C-triclosan and in the rat following a single 5 mg/kg dose and a 14-day repeated 5 mg/kg dose. The mouse data in this review are the same data cited in MRID 149464. For mice, the administered radioactivity was widely distributed within 1 minute after dosing (Table 3; not all tissues are shown). Greater levels were detected in organs/tissues associated with metabolism/elimination (i.e., liver, kidneys, gall bladder) and highly perfused organs (heart, lungs). It is evident that initially, highly perfused tissues such as the liver, lungs, heart, and kidney are associated with the highest concentrations of triclosan-derived radioactivity. Relative to the other organs measured in this study, this relationship is maintained up to the 2 hour post-dose time period which is when monitoring in mice was stopped. Of interest is the rapid distribution into the brain as well as "spinal marrow" (presumably, spinal fluid) and the accumulation of triclosan-derived radioactivity in the gall bladder over the 2 hour time frame.

TABLE 3. Time course for tissue concentrations (μg eq. /g) in mice following a single intravenous dose (10 mg/kg) of ^{14}C-triclosan[a].				
Organ/tissue	1 min	5 min	30 min	2 hrs
Blood	13.8	13.0	12.1	9.5
Heart	47.6	16.7	5.7	3.1
Lungs	40.6	20.3	14.7	8.3
Thyroid	48.3	38.3	13.2	5.4
Adrenal	105.7	43.9	18.8	4.7
Liver	43.8	41.1	26.2	15.9
Kidneys	38.0	27.9	32.2	13.3
Gall bladder	36.5	136.6	368.2	850.5
Fat	24.4	21.9	6.0	2.1
Brain	16.9	13.0	1.9	0.3
Spinal marrow	15.1	11.9	3.2	0.5

[a] Average of two animals
Data extracted from Table 1, p. 31, MRID 68161.

Tissue distribution was also assessed in rats given a single or 14-day repeated dose of ^{14}C-triclosan (5 or 30 mg/kg). The 24 hour post-dose pattern of distribution following these oral doses was similar to that observed for the intravenous dose in mice, with the lungs, liver, and kidneys observed with the highest concentration of triclosan-derived radioactivity. Of interest are the levels of radioactivity detected in the pituitary gland, optic nerve, and sciatic nerve after 24 hours. These concentrations were equivalent to those observed in the highly perfused tissues. Although the distribution pattern was qualitatively similar for the high dose (30 mg/kg) rats, somewhat greater amounts of test material radioactivity (7 to 10-fold on a μg/g basis) than expected for a 6-fold dose increase were detected in tissues of the high dose rats than for the low-dose groups. Repeated low dose (5 mg/kg) administration of triclosan to rats was associated with increases in tissue levels of radioactivity for the blood, plasma, pituitary gland, sciatic nerve, and kidneys relative to single dose administration (Table 4 of this review). Repeated oral dosing at the 30 mg/kg dose level resulted in notable increases in triclosan-derived radioactivity in the plasma, sciatic nerve, liver, and kidneys.

TABLE 4. Comparison of Triclosan-derived radioactivity (μg/g tissue) in selected tissues 24 Hours after single and repeated low (5 mg/kg) and high (30 mg/kg) oral doses in rats of ^{14}C-triclosan[a]. N = 4				
	5 mg/kg single dose	**5 mg/kg repeat dose**	**30 mg/kg single dose**	**30 mg/kg repeat dose**
Organ/tissue				
Blood	0.34±0.08	0.64±0.08	2.74±0.66	3.69±0.52
Plasma	0.55±0.13	1.14±0.14	4.58±1.07	6.46±1.07
Lungs	0.28±0.06	0.33±0.08	2.60±0.61	3.30±0.43
Pituitary gland	0.25±0.037	0.77±0.06	1.73±0.27	1.79±0.27
Optic nerve	0.28±0.12	0.20±0.08	1.79±0.42	1.03±0.35
Trigeminal nerve	0.16±0.04	0.20±0.08	1.52±0.46	0.94±0.27
Sciatic nerve	0.31±0.06	1.02±0.12	1.75±0.21	3.70±0.68
Thyroid	0.15±0.03	0.32±0.08	1.37±0.21	1.33±0.19
Liver	0.27±0.05	0.39±0.07	2.70±0.57	4.60±0.37
Kidneys	0.46±0.06	0.76±0.06	3.10±0.74	4.81±0.31

[a]data from Tables 3 and 4, pages 33- 34 of the report.

B. PHARMACOKINETIC STUDIES

Time-course for radioactivity in the blood was provided for rats (Table 4) and dogs (Table 5) given oral doses of the test material. Additionally, time course data on radioactivity in the brain of rats was also reported (Table 4). At the time periods tested, concentrations in brain tissue were considerably less than detected in blood and plasma. The available data indicated relatively rapid and complete elimination of test article radioactivity from all three tissues. Peak concentration in the blood and plasma was attained within three hours and had declined ~5-fold at 24 hours. Concentrations in the brain were considerably less and declined about 3-fold over 24 hours. The investigators noted that very little (<1%) of the blood/plasma radioactivity represented unchanged triclosan. For the brain, however, ~35-50% of the radioactivity was attributed to parent compound.

Blood radioactivity in rats following a single or 14-day repeated oral dose of 5 mg/kg was about two-fold greater (0.34 vs 0.64 μg/g) for the repeated dose regimen. The blood concentrations for rats receiving a single vs 14-day repeated dose of 30 mg/kg dose were 8- and 5-fold greater, respectively, relative to the 5 mg/kg groups. It is of interest to note that relative to brain, peripheral nerve (sciatic) showed a higher concentration of triclosan-derived radioactivity at the same time point (24 hours) at the same dose level (5 mg/kg; Table 5 vs. Table 4 of this review).

TABLE 5. Time course of ^{4}C-triclosan (μg eq./mL) in blood, plasma, and brain of rats following a single oral dose (5 mg/kg).

Time (hrs)	Blood	Plasma	Brain
3	2.48	4.71	0.077
6	1.76	3.8	0.060
24	0.44	0.85	0.024

Data taken from Tables 5 and 6, p. 35, MRID 68161.

For the two dogs tested, considerable variability in blood concentration time-course was observed. Although peak concentrations were similar (8.59 and 7.63 μg eq./mL), these values were attained at 2 and 8 hours, respectively, post dosing. By 48 hours, blood concentration values for the two dogs were similar. Similar to rats, unchanged triclosan represented ≤1.6% of the radioactivity.

TABLE 5. Time course of ^{4}C-triclosan (μg eq./ml) in the blood of two dogs following a single oral dose (5 mg/kg).		
Time (hrs)	**Dog a**	**Dog b**
1	5.43	0.10
2	8.59	0.36
4	7.59	5.36
8	6.87	7.63
24	3.87	4.94
48	2.24	3.07
72	0.83	-

Data taken from Table 7, p. 36, MRID 68161.

C. METABOLITE CHARACTERIZATION STUDIES

Definitive characterization of metabolites in urine and feces was not performed. The available data indicated that very little of the radioactivity in the blood could be attributed to unchanged triclosan. Preliminary investigations using dog blood samples subjected to enzymatic hydrolysis indicated that at least a portion of the radioactivity (~10-20%, data not shown) could be attributed to glucuronide conjugates.

III. DISCUSSION

A. DISCUSSION

In a metabolism study (MRID 68161), ^{14}C-triclosan (>98% radiochemical purity, lot/batch no. not specified) was administered to male rats by gavage at doses of 5 or 30 mg/kg (single or 14-day repeated). Two male beagle dogs and two male baboons received single 5 mg/kg doses in gelatine capsules and male mice were given a single 10 mg/kg intravenous dose. Blood levels were monitored in mice up to 2 hours, in rats at 24 hours postdose, in the dogs up to 72 hours and monkeys up to 168 hours postdose. Urinary and fecal excretion was monitored in monkeys and dogs for 6-10 days. Tissue distribution was assessed in mice and rats.

There were no test article-related toxic effects reported. Recovery of administered radioactivity was 86.9% and 74.1% for each of two dogs, and 83.1% and 80.5% for each of two monkeys. Administered radioactivity was widely distributed among tissues/organs in the mice as assessed at 1, 5, 30, and 120 minutes after an intravenous injection (10 mg/kg) and at 24 hours in rats following a single oral administration or the last dose of a 14-day repeated oral administration (5 or 30 mg/kg). For mice, the administered

radioactivity was widely distributed within 1 minute after intravenous dosing (Table 3 of this review). Greater levels were detected in organs/tissues associated with metabolism/elimination (i.e., liver, kidneys, gall bladder) and highly perfused organs (heart, lungs). It is evident that initially, highly perfused tissues such as the liver, lungs, heart, and kidney are associated with the highest concentrations of triclosan-derived radioactivity. Relative to the other organs measured in this study, this relationship is maintained up to the 2 hour post-dose time period which is when monitoring in mice was stopped. Of interest is the rapid distribution into the brain as well as "spinal marrow" (presumably, spinal fluid) and the accumulation of triclosan-derived radioactivity in the gall bladder over the 2 hour time frame.

Tissue distribution was also assessed in rats given a single or 14-day repeated dose of ^{14}C-triclosan (5 or 30 mg/kg). The 24 hour post-dose pattern of distribution following these oral doses was similar to that observed for the intravenous dose in mice, with the lungs, liver, and kidneys observed with the highest concentration of triclosan-derived radioactivity. Of interest are the levels of radioactivity detected in the pituitary gland, optic nerve, and sciatic nerve after 24 hours. These concentrations were equivalent to those observed in the highly perfused tissues. Although the distribution pattern was qualitatively similar for the high dose (30 mg/kg) rats, somewhat greater amounts of test material radioactivity (7 to 10-fold on a μg/g basis) than expected for a 6-fold dose increase were detected in tissues of the high dose rats than for the low-dose groups. Repeated low dose (5 mg/kg) administration of triclosan to rats was associated with increases in tissue levels of radioactivity for the blood, plasma, pituitary gland, sciatic nerve, and kidneys relative to single dose administration (Table 4 of this review). Repeated oral dosing at the 30 mg/kg dose level resulted in notable increases in triclosan-derived radioactivity in the plasma, sciatic nerve, liver, and kidneys. Thus, the available data suggest some potential for accumulation in certain tissues after repeated exposure to Triclosan, but further work is necessary to confirm this observation. It is especially of interest to pursue the apparent accumulation in sciatic nerve and thyroid and pituitary.

Time-course analysis of blood/plasma radioactivity in rats revealed that peak concentrations were attained within three hours after a single oral dose of 5 mg/kg and that the concentrations declined approximately five-fold within 24 hours. For dogs, somewhat longer blood t_{max} were observed but were variable (2-8 hours). For one dog, blood concentration were approximately one fifth of maximum at 72 hours. Generally, the test article did not exhibit especially rapid partitioning into or clearance from the blood for either of the two species tested. Relative to blood concentrations, levels of radioactivity in the brains of rats were notably lower (18 to 30-fold) at the tested time periods (3, 6, and 24 hours), but levels in sciatic nerve were equivalent. Time-course analysis of tissues from mice given a single intravenous dose (10 mg/kg) of ^{14}C- triclosan showed that the highest concentrations of radioactivity were initially (1 minute) associated with highly perfused organs/tissues (i.e., heart, lungs, liver, kidneys, thyroid gland). These levels significantly declined within 30 minutes but was somewhat less rapid for organs /tissues associated with metabolism and elimination and notably increased for the gall bladder.

Both urinary and fecal elimination were identified as major routes of excretion. The relative contribution of each to overall elimination of administered radioactivity exhibited species variability. For monkeys, urinary excretion accounted for 56.69% (average for the two monkeys tested) of the administered dose and fecal elimination accounted for 25.15%. For dogs (average of the two animals tested), urinary and fecal elimination accounted for 12.16% and 68.30%, respectively, of the administered dose. In both species, most of the urinary and fecal excretion occurred within 48 hours.

Definitive characterization of metabolites was not performed. Preliminary investigations using acid and enzyme hydrolysis, indicated that very little (<1%) of the blood/plasma radioactivity was associated with unchanged triclosan. In the brains of rats, however, 35-50% of the radioactivity could be attributed to unchanged parent compound.

Overall, this study demonstrated that ^{14}C-triclosan is well-absorbed from the gastrointestinal tract, has a potential wide volume of distribution, can cross the blood-brain barrier, and potentially accumulates in specific tissues after repeated oral exposure. Absorption into the blood and clearance from the blood is not especially rapid, but the compound does not appear to undergo sequestration in the species tested. Most of the circulating radioactivity was attributed to metabolism products (probably conjugation products based upon preliminary experiments using acid and enzymatic hydrolysis). Both the urine and the feces are significant routes of excretion with the relative importance appearing to be species dependent.

This metabolism study in multiple species, which predates GLP guidelines, is **Unacceptable/ upgradable** and does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. There are numerous deficiencies in this study , including test article characterization, environmental and housing conditions of the experimental animals, dose solution information (stability, homogeneity, confirmation of doses received), and lack of mass balance information. Additionally, a quality assurance statement was not provided. If these data can be provided, the study can be upgraded to acceptable. Otherwise, a new guideline metabolism study will need to be conducted.

B. STUDY DEFICIENCIES

No quality assurance statement was available and the report lacked information regarding dose confirmation, homogeneity, and stability. Additionally, details regarding the animals and the test article (e.g., no lot or batch numbers) were sketchy, and overall recoveries of administered radioactivity were marginal.

DATA EVALUATION REPORT

TRICLOSAN

Study Type: METABOLISM AND PHARMACOKINETICS – HUMAN
[OPPTS 870.7485 (§85-1)]
MRID 68162

Prepared for

Antimicrobials Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. T07

Primary Reviewer:
Robert A.Young, Ph.D., D.A.B.T. Signature: Date: OCT 0 6 2000

Secondary Reviewers:
H. T. Borges, MT(ASCP),Ph.D., D.A.B.T. Signature: Date: OCT 0 6 2000

Robert H. Ross, M.S., Group Leader Signature: Date: OCT 0 6 2000

Quality Assurance:
Lee Ann Wilson, M.A. Signature: Date: OCT 0 6 2000

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UTBattelle, LLC, for the U.S. Dept. of Energy under contract DEAC0500OR22725

EPA Reviewer: Tim McMahon, Ph.D.
Antimicrobials Division

_____, Date 10/23/00

DATA EVALUATION RECORD

STUDY TYPE: Metabolism - Human Volunteer; OPPTS [870.7485 (§85-1)]

DP BARCODE: D266143
P.C. CODE: 054901

SUBMISSION CODE: None
TOX. CHEM. NO.: None

TEST MATERIAL (PURITY): Triclosan (radiochemical purity >99%)

SYNONYMS: GP 41 353; Triclosan;5-chloro-2-(2,4-dichlorophenoxy)phenol

CITATION: Stierlin, H., R. Murbach, W. Theobald (1976). GP 41 353: Pharmacokinetic and metabolic studies in man following oral administration of a ^{14}C-labelled preparation. Pharma Research, Pharmacological Chemistry. CIBA-GEIGY, Ltd. Basle. Report No. B 6/1976. MRID 68162. January 27, 1976. Unpublished

SPONSOR: CIBA-GEIGY Limited, Pharma Research, Pharmacological Chemistry, Basle.

EXECUTIVE SUMMARY: In a metabolism and disposition study (MRID 68162), ^{14}C-labelled Triclosan (batch/lot nos. not provided; >99% radiochemical purity) was administered as a single oral dose (203.57 mg/kg; 100 μCi) to a human volunteer (male, 43 years of age).

Recovery of administered dose was 90% based upon radioactivity in the urine and feces. A complete assessment was not possible in the absence of tissue distribution/burden data. Absorption could be estimated as being at least 57% based upon urinary excretion but is not definitive in the absence of biliary secretion data. Recovery of radioactivity in the urine indicated that 57 % of the administered radioactivity was excreted over 144 hours. The majority of urinary elimination (69%) occurred during the first 24 hours and appeared to be nearly complete within 120 hours. Fecal elimination accounted for 33.5% of the administered dose over a 144-hour period with most elimination (90%) occurring within 48 hours.

For both plasma and urine, very little (1-3%) of the radioactivity was associated with free triclosan. The majority of the radioactivity could be attributed to glucuronide conjugates or other nonspecified conjugates as determined by β-glucuronidase hydrolysis or acid hydrolysis, respectively.

In summary, this study provides anecdotal information regarding urinary and fecal excretion, metabolite characterization, and blood/plasma time-course of triclosan in a human volunteer following a single oral dose. The results of both the excretion assessment and metabolite

quantitation/characterization studies are consistent with other reports in a human subject (MRID 68163) or in laboratory species (MRID 149464, 79590).

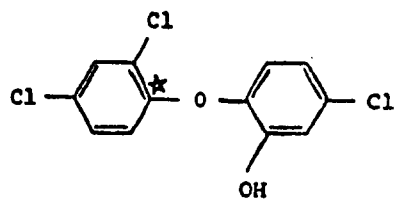
This metabolism study using a human volunteer is **Acceptable/Nonguideline** but does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Although properly conducted and providing data regarding the excretion, metabolism, and plasma/blood kinetics of triclosan in a human volunteer following a single oral dose, there was no statistical base and the protocol was not consistent with a 85-1 Guidelines (no tissue distribution data). The results, however, do affirm findings of companion studies (MRID 149464, 79590) in animal species and another study (MRID 68163) with a human volunteer.

COMPLIANCE: Signed and dated, Quality Assurance, and Data Confidentiality statements were not included with all studies. Flagging statements were not included. A Good Laboratory Practice statement was not relevant.

I. MATERIALS AND METHODS

A. MATERIALS

1. Test compound
   Radiolabeled: [^{14}C]-GP 41 353 (sp. act. 0.50 μCi/mg)
   Batch No.: not provided
   Purity:
   Radiochemical purity: >99%
   Chemical purity: not provided
   Description: not available
   Contaminants: none noted
   CAS No.: not available
   Structure:



[^{14}C]-GP 41 353

2. Vehicle
   The test article was administered in a gelatine capsule; there was no vehicle noted.

3. Test species

Species: human
Strain: NA
Age and weight at study initiation: 43 years; 78 kg
Source: NA
Housing: NA
Diet: no information provided
Water: no information provided
Environmental conditions:
Temperature: no information provided
Humidity: no information provided
Air changes: no information provided
Photoperiod: no information provided
Acclimation period: NA

4. Preparation of dose

The test article was administered in a gelatine capsule. No additional information was provided.

Results –
Homogeneity: No data provided.

Stability: No data provided.

Dose confirmation: No data provided.

B. STUDY DESIGN AND METHODS

1. Group arrangements

A human volunteer (male) was given a single 203.57 mg/kg oral dose (100 μCi) of the test material. This was followed by a meal two hours later.

2. Dosing and sample collection

Following administration of the 203.57 mg/kg dose in a gelatine capsule, the following samples were collected:

Blood - Blood was collected prior to dosing and at 0.5, 1, 2, 4, 8, 24, and 72 hours postdose.

Urine - Urine was collected at 0-8 hours, 8-24 hours, and daily for an additional five days.

Feces - Feces were collected for six days.

3. Sample preparation/analysis

Blood/plasma - Radioactivity was determined in all samples. The 2, 4, 8, and 24-hour samples were analyzed for free and bound triclosan and also subjected to enzyme hydrolysis for assessment of conjugation products. Blood/plasma samples were oxidized prior to scintillation counting. Radioactive compounds from plasma samples were adsorbed on Amberlite XAD-2 neutral resin and subsequently analyzed as described below.

Urine - Radioactivity was determined in all urine samples. Urine samples up to 72 hours were used for assessment of unchanged and conjugation product metabolites. Individual fractions were also extracted and chromatographed. Samples were added directly to the scintillation fluid and counted. Urinary metabolites were separated and concentrated using gel filtration as described below.

Feces - Total radioactivity was measured in all samples.

4. Analytical techniques

Liquid scintillation counting (LSC) - LSC was performed using Diotol scintillation fluid and a Packard Model 3380 scintillation counter. No information was provided regarding quench correction, counting efficiency, background count limits, or number of samples counted.

Mass spectrometry (MS) - For final identification of metabolites, MS was performed using a Varian MAT CH5-DF mass spectrophotometer equipped with an FI/FD/EI-ion source.

Thin-layer Chromatography (TLC) - Metabolite concentrates from resin filtration were chromatographed two dimensionally on Antec silica gel plates (solvent of ethylenechloride/methanol/diethylamine/water; 72:25:3:1). TLC plates were then exposed to an X-ray film for 2-3 days. No further details were provided.

Gel filtration - For isolation of urinary metabolites, a 2000 mL urine sample (0-24 hour) was extracted three times with diethylether. Upon drying, the resulting extract was reconstituted in 4 mL of water and separated on a Sephadex LH 20 column using methanol/water (1:1) mobile phase. A similar protocol utilizing a 75:25 water/methanol mobile phase was used for metabolite purification. Amberlite XAD-2 neutral resin was used to adsorb radioactive compounds from plasma samples. The fractions were eluted from the resin using a gradient of methanol and water.

Derivatization and structural analysis - Enzymatic hydrolysis of samples with β-glucuronidase and arylsulfatase was used to generate free triclosan.

5. Statistics and calculations

No information was provided regarding statistical analysis.

II. RESULTS

A. DISTRIBUTION/EXCRETION STUDIES

1. Mass balance

It was not possible to accurately assess mass balance based upon the available data and the experimental protocol (i.e., no tissue burden data). Based upon radioactivity in the plasma and recovery of radioactivity in the urine and feces, overall recovery was at least 91%. Although not representing a totally accurate mass balance, this recovery is acceptable.

2. Absorption

Absorption may be implied from the urinary excretion data and assumed to be at least 57% of the administered dose. In the absence of biliary secretion data, it is not possible to definitively attribute any of the fecal radioactivity to an absorbed dose or simply to radioactivity unabsorbed from the gastrointestinal tract.

3. Excretion

Urinary excretion over a 144-hour period accounted for 57% of the administered radioactivity (Table 1). Approximately 42% of the urinary excretion (23.77% of the total dose) occurred during the first eight hours.

Table 1. Time-course for urinary excretion of ^{14}C-triclosan in a human volunteer following a single oral administration (203 mg/kg)		
Time Interval (hrs)	**% administered dose**	**Concentration (μg/mL)**
0-8	23.77	118.06
8-24	15.82	71.61
24-48	11.24	16.59
48-72	3.56	4.05
72-96	1.73	2.27
96-120	0.64	1.10
120-144	0.34	0.61
Total (0-144)	57.10	14.94

Data taken from Table 1, p. 8, MRID 68162.

Fecal excretion accounted for 33.50% of the administered dose over a 144-hour period (Table 2). Fecal elimination was approximately 90% complete within 48 hours.

Table 2. Time-course for fecal excretion of ^{14}C-triclosan in a human volunteer following a single oral administration (203 mg/kg)	
Time Interval (hrs)	**% administered dose**
0-24	10.84
24-48	19.41
48-72	1.27
72-96	1.21
96-120	0.57
120-144	0.20
Total (0-144)	33.50

Data taken from Table 1, p. 8, MRID 68162.

4. Tissue distribution

Tissue distribution was not determined.

B. PHARMACOKINETIC STUDIES

Time-course concentrations (expressed as μg eq./mL blood) in blood and plasma are shown in Table 3. Peak blood/ plasma concentrations were achieved at approximately four hours after dosing and appeared to decline gradually over 48 hours. Binding of radioactivity to erythrocytes appeared to occur initially but was not sustained beyond two hours.

Table 3. Time-course for blood and plasma concentrations of ^{14}C-triclosan in a human volunteer following a single oral administration (203 mg/kg)

Time (hrs)	Blood (μg/mL)[a]	Plasma (μg/mL)[a]	Erythrocyte bound (%)
0.5	0.03	0.10	-
1	0.15	0.32	9.2
2	0.58	1.11	0
4	5.34	10.54	0
8	4.32	8.28	0
24	1.36	2.45	0
72	0.26	0.48	-

[a] Values are expressed as unchanged triclosan
Data taken from Table 2, p. 9, MRID 68162.

C. METABOLITE CHARACTERIZATION STUDIES

1. Metabolites in urine
The majority (~75 to 100%) of the radioactivity in the urine was associated with a glucuronide conjugate as determined by enzymatic or acid hydrolysis, respectively (Table 4). Free triclosan represented very little of the urinary radioactivity (maximum of ~3.8% at 8-24 hours).

Table 4. Metabolite quantitation in urine from a human volunteer following a single oral administration (203 mg/kg) of triclosan						
Time (hrs)	Unconjugated triclosan µg/mL	% dose	Glucuronide conjugate µg/mL	% dose	Acid hydrolysis product µg/mL	% dose
0-8	0.41	0.35	117.49	99.5	118.89	110.7
8-24	2.70	3.78	61.44	85.8	62.00	86.6
24-48	0.13	0.81	14.14	85.2	14.51	87.5
48-72	0.04	0.99	3.02	74.6	2.64	69.2
Total (0-72)	-	-	24.14*	85.8	23.91*	85.0

* Values are expressed as unchanged triclosan
Data taken from Table 5, p. 11, MRID 68162.

2. Metabolites in blood/plasma
Similar to urine, free triclosan represented very little (<1%) of the radioactivity in the plasma at any sample time (Table 5). Enzymatic hydrolysis of plasma samples with β-glucuronidase resulted in significant amounts of free triclosan indicating that much (up to 50%) of the plasma radioactivity was a glucuronide conjugate. Release of even greater amounts of free triclosan following acid hydrolysis suggests that additional conjugation products were present and that these accounted for up to 90% of the plasma radioactivity.

Table 5. Metabolite quantitation in plasma from a human volunteer following a single oral administration (203 mg/kg) of triclosan						
Time (hrs)	Unconjugated triclosan µg/mL	% dose	Glucuronide conjugate µg/mL	% dose	Acid hydrolysis product µg/mL	% dose
2	0.008	0.71	0.56	50.4	1.00	90.1
4	0.018	0.17	4.68	44.4	8.30	78.7
8	0.016	0.19	2.73	33.0	7.03	84.9
24	0.014	0.57	1.12	45.7	1.96	80.0

Data taken from Table 3, p. 10, MRID 68162.

3. Metabolites in feces
Metabolite characterization in this matrix was not conducted.

III. DISCUSSION

A. DISCUSSION

In a metabolism and disposition study (MRID 68162), ^{14}C-labelled Triclosan (batch/lot nos. not provided; >99% radiochemical purity) was administered as a single oral dose (203.57 mg/kg; 100 μCi) to a human volunteer (male, 43 years of age). Blood was collected prior to dosing and at 0.5, 1, 2, 4, 8, 24, and 72 hours postdose, and urine and feces collected over six days The objective of this experiment was to assess excretion, time-course in blood/plasma, and to identify the major urinary and plasma metabolites of the test article.

Recovery of administered radioactivity was 90% based upon radioactivity in the urine and feces. A complete assessment was not possible in the absence of tissue distribution/burden data. Absorption could be estimated as being at least 57% based upon urinary excretion but is not definitive in the absence of biliary secretion data. Radioactivity in the urine indicated that 57 % of the dose was excreted over 144 hours. The majority of urinary elimination (69%) occurred during the first 24 hours and appeared to be nearly complete within 120 hours. Fecal elimination accounted for 33.5% of the administered dose over a 144-hour period with most elimination (90%) occurring within 48 hours.

For both plasma and urine, very little (1-3%) of the radioactivity was associated with free triclosan. The majority of the radioactivity could be attributed to glucuronide conjugates or other nonspecified conjugates as determined by β-glucuronidase hydrolysis or acid hydrolysis, respectively.

In summary, this study provides anecdotal information regarding the urinary and fecal excretion, metabolite characterization, and blood/plasma time-course of triclosan in a human volunteer following a single oral dose. The results of both the excretion assessment and metabolite quantitation/characterization studies are consistent with other reports in a human subject (MRID 68163) or in laboratory species (MRID 149464, 79590).

This metabolism study using a human volunteer is **Acceptable/Nonguideline** but does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Although properly conducted and providing data regarding the metabolism of triclosan in a human volunteer following a single oral dose, there was no statistical base and the protocol was not consistent with 85-1 Guidelines (no tissue distribution/burden data). The results, however, do affirm findings of companion studies (MRID 149464, 79590) in animal species.

The data are anecdotal. Although the findings indicate that there are two major conjugation product metabolites which are excreted in the urine, the experiment could not address species or dose related variability in this process.

There was no quality assurance statement and no information regarding dose confirmation, homogeneity or stability.

DATA EVALUATION REPORT

TRICLOSAN

Study Type: METABOLISM AND PHARMACOKINETICS – HUMAN
[OPPTS 870.7485 (§85-1)]
MRID 68163

Prepared for

Antimicrobials Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. T07

Primary Reviewer: Robert A. Young, Ph.D., D.A.B.T.	Signature: Date:	Robert A. Young OCT 0 6 2000
Secondary Reviewers: H. T. Borges, MT(ASCP),Ph.D., D.A.B.T.	Signature: Date:	H T Borges OCT 0 6 2000
Robert H. Ross, M.S., Group Leader	Signature: Date:	Robert H. Ross OCT 0 6 2000
Quality Assurance: Lee Ann Wilson, M.A.	Signature: Date:	L. A. Wilson OCT 0 6 2000

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UTBattelle, LLC, for the U.S. Dept. of Energy under contract DEAC0500OR22725

EPA Reviewer: Tim McMahon, Ph.D.
Antimicrobials Division

_____, Date 10/23/00

DATA EVALUATION RECORD

STUDY TYPE: Metabolism - Human Volunteer; OPPTS [870.7485 (§85-1)]

DP BARCODE: D266143
P.C. CODE: 054901

SUBMISSION CODE: None.
TOX. CHEM. NO.: None

TEST MATERIAL (PURITY): Triclosan (radiochemical purity >99%)

SYNONYMS: GP 41 353; Triclosan;5-chloro-2-(2,4-dichlorophenoxy)phenol

CITATION: Stierlin, H., H.P. Kriemler, W. Theobald (1976). GP 41 353: Identification of conjugated metabolites in plasma and urine of man after oral administration of ^{14}C-labelled ..[illegible copy]...tion. Pharma Research, Pharmacological Chemistry. CIBA-GEIGY, Ltd. Basle. Report No. B 56/1976. MRID 68163. December 20, 1976. Unpublished

SPONSOR: CIBA-GEIGY Limited, Pharma Research, Pharmacological Chemistry, Basle.

EXECUTIVE SUMMARY: In a metabolism and disposition study (MRID 68163), ^{14}C-labelled Triclosan (batch/lot nos. not provided; >99% radiochemical purity) was administered as a single oral dose (200.3 mg/kg; 100 μCi) to a human volunteer (male, 48 years of age).

Recovery of administered radioactivity and evaluation of mass balance was not possible because tissue burdens and overall body burden could not be assessed and biliary contribution to excretion were not measured. Recovery of radioactivity in the urine implied 73.79% elimination of the administered dose over 72 hours. The majority of urinary elimination occurred during the first 24 hours and appeared to be nearly complete by 72 hours. Assuming residual radioactivity residing in the blood and other tissues at the terminal time point, absorption of >74% may be inferred for the 200 mg/kg dose.

Treatment of plasma and urine with β-glucuronidase and arylsulfatase revealed that the majority of the radioactivity in these matrices could be attributed to conjugation products rather than unchanged parent compound. For urine, 93.85% of the recovered radioactivity could be attributed to glucuronide conjugates while approximately 2.3% could be attributed to sulfate conjugates. For plasma, 51.87- 60.62 % of the radioactivity could be attributed to glucuronide conjugates while enzymatic hydrolysis revealed that the sulfate conjugate represented 31.61-43.50% of the plasma radioactivity. Minor changes in the relative contributions of these metabolites over time were detected but of uncertain significance.

In summary, this report provides anecdotal data in humans indicating that orally administered triclosan undergoes Phase II metabolism to conjugation product metabolites (sulfate and glucuronide conjugates) which are eliminated via the urine within 72 hours.

This metabolism study using a human volunteer is **Acceptable/Nonguideline** but does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Although properly conducted and providing data regarding the metabolism of triclosan in a human volunteer following a single oral dose, the data were anecdotal, the protocol did not include tissue distribution assessments, and no information was provided regarding dose stability and confirmation or homogeneity. Therefore, the study was not consistent with an 85-1 Guideline study. The results, however, do affirm findings of companion studies (MRID 149464, 79590) in other animal species and provide important information based upon human data.

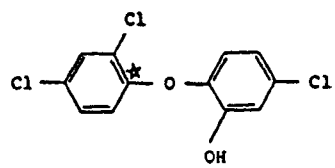
COMPLIANCE: Signed and dated, Quality Assurance, and Data Confidentiality statements were not included with all studies. Flagging statements were not included. A Good Laboratory Practice statement was not relevant.

I. MATERIALS AND METHODS

A. MATERIALS

1. Test compound

Radiolabeled: [^{14}C]-GP 41 353 (sp. act. 0.50 μCi/mg)
Batch No.: not provided
Purity:
Radiochemical purity: >99%
Chemical purity: not provided
Description: not available
Contaminants: none noted
CAS No.: not available
Structure:



2. Vehicle

The test article was administered in a gelatine capsule; there was no vehicle noted.

3. Test species

Species: human
Strain: NA
Age and weight at study initiation: 48 years; 83 kg
Source: NA
Housing: NA
Diet: no information provided
Water: no information provided
Environmental conditions:
Temperature: no information provided
Humidity: no information provided
Air changes: no information provided
Photoperiod: no information provided
Acclimation period: NA

4. Preparation of dose

The test article was administered in a gelatine capsule. No additional information was provided.

Results –
Homogeneity: No data provided.

Stability: No data provided.

Dose confirmation: No data provided.

B. STUDY DESIGN AND METHODS

1. Group arrangements

A human volunteer (male) was given a single 200.3 mg/kg oral dose of the test material and radioactivity determined in blood and urine.

2. Dosing and sample collection

Following administration of the 200.3 mg/kg dose in a gelatine capsule, the following samples were collected:

Blood - Blood was collected prior to dosing and at 2, 4, 8, and 24 hours postdose.

Urine - Urine was collected daily for 72 hours.

3. Sample preparation/analysis

Urine - Radioactivity was determined in the 24-hour urine samples. A 0-72 hour pooled sample was used for assessment of metabolites. Samples were added directly to the scintillation fluid and counted. Urinary metabolites were separated and concentrated using gel filtration as described below.

Blood/plasma - Samples were oxidized prior to scintillation counting. Radioactive compounds from plasma samples were adsorbed on Amberlite XAD-2 neutral resin and subsequently analyzed as described below.

4. Analytical techniques

Liquid scintillation counting (LSC) - LSC was performed using Diotol scintillation fluid and a Packard Model 3380 scintillation counter. No information was provided regarding quench correction, counting efficiency, background count limits, or number of samples counted.

Mass spectrometry (MS) - For final identification of metabolites, MS was performed using a Varian MAT CH5-DF mass spectrophotometer equipped with an FI/FD/EI-ion source.

Thin-layer Chromatography (TLC) - Metabolite concentrates from resin filtration were chromatographed two dimensionally on Antec silica gel plates (solvent of ethylenechloride/methanol/diethylamine/water; 72:25:3:1). TLC plates were then exposed to an X-ray film for 2-3 days. No further details were provided.

Gel filtration - For isolation of urinary metabolites, a 2000 mL urine sample (0-24 hour) was extracted three times with diethylether. Upon drying, the resulting extract was reconstituted in 4 mL of water and separated on a Sephadex LH 20 column using methanol/water (1:1) mobile phase. A similar protocol utilizing a 75:25 water/methanol mobile phase was used for metabolite purification. Amberlite XAD-2 neutral resin was used to adsorb radioactive compounds from plasma samples. The fractions were eluted from the resin using a gradient of methanol and water.

Derivatization and structural analysis - Enzymatic hydrolysis of samples with β-glucuronidase and arylsulfatase was used to generate free triclosan.

5. Statistics and calculations

No information was provided regarding statistical analysis.

II. RESULTS

A. DISTRIBUTION/EXCRETION STUDIES

1. Mass balance

It was not possible to assess mass balance based upon the available data and the experimental protocol. Based upon radioactivity in the plasma and urine, overall recovery was >74%.

2. Absorption

Absorption of the test material from the gastrointestinal tract may be implied from the urinary excretion data. Over a 72-hour period, 73.79% of the administered radioactivity was recovered in the urine. However, data were not available to assess that portion of the administered dose which may have been eliminated via the bile and feces or which may have been retained in the tissues. Therefore, an accurate assessment of absorption is not possible based upon the available data.

3. Excretion

Time-course and urinary concentration data for urinary excretion by a human subject following a single oral dose of triclosan is shown in Table 1. The majority of urinary elimination occurred during the first 24 hours and appeared to be nearly complete by 72 hours. Urinary elimination over three days accounted for 73.79% of the administered radioactivity. The concentration of the test article and its metabolites paralleled the elimination of radioactivity. Analysis of urine indicated that most of the radioactivity was associated with conjugation products (primarily glucuronides).

Table 1. Time-course for urinary excretion of ^{14}C-triclosan in a human volunteer following a single oral administration (200 mg/kg)

Time Interval (hrs)	% administered dose	Concentration (μg/mL)
0-24	62.91	62.07
24-48	8.14	4.75
48-72	2.74	1.47
Total (0-72)	73.79	16.08

Data taken from Table 2, p. 11, MRID 68163.

4. Tissue distribution

Tissue distribution was not determined.

B. PHARMACOKINETIC STUDIES

A definitive assessment of pharmacokinetic parameters was not a study objective. Time-course concentrations (expressed as μg eq./mL blood) in blood and plasma are shown in Table 2.

Table 2. Time-course for blood and plasma concentrations of ^{14}C-triclosan in a human volunteer following a single oral administration (200 mg/kg)

Time (hrs)	Blood (μg/mL)[a]	Plasma (μg/mL)[a]
2	2.42	5.05
4	3.71	7.41
8	2.85	5.91
24	0.66	1.27

[a] Values are expressed as unchanged triclosan; most of the radioactivity actually represents glucuronide conjugates of triclosan.
Data taken from Table 1, p. 10, MRID 68163.

C. METABOLITE CHARACTERIZATION STUDIES

Metabolite characterization was conducted for urine and blood/plasma samples. For both matrices, unchanged triclosan represented only a small fraction (< 1% for blood/plasma; 3.40 % for urine) of the recovered radioactivity.

1. Metabolites in urine

Hydrolysis of urine samples with β-glucuronidase and arylsulfatase revealed that 93.85% of the urinary radioactivity could be attributed to glucuronide conjugates while approximately 2.3% could be attributed to sulfate conjugates. Following gel filtration, spectrophotometric analysis revealed that the major urinary metabolite was a glucuronide and two-dimensional TLC identified a secondary metabolite as the ester sulfate of triclosan.

2. Metabolites in plasma

Similar to urine, the majority of the radioactivity in blood plasma could be attributed to glucuronide and sulfate conjugation products; < 1% of the radioactivity was

associated with unchanged triclosan. Hydrolysis of plasma samples with β-glucuronidase and arylsulfatase revealed that 51.87- 60.62 % of the plasma radioactivity could be attributed to glucuronide conjugates. Only minor differences in this percentage were observed when comparing samples of different collection times. Generally, the glucuronide percent decreased slightly with time but this observation is based on an insufficient sample to assign any significance. Enzymatic hydrolysis revealed that the sulfate conjugate represented 31.61- 43.50% of the plasma radioactivity.

III. DISCUSSION

A. DISCUSSION

In a metabolism and disposition study (MRID 68163), ^{14}C-labelled Triclosan (batch/lot nos. not provided; >99% radiochemical purity) was administered as a single oral dose (200.3 mg/kg; 100 μCi) to a human volunteer (male, 48 years of age). Blood was collected prior to dosing and at 2, 4, 8, and 24 hours postdose, and urine was collected daily for three days. The objective of this experiment was to identify the major urinary and plasma metabolites of the test article.

Recovery of administered radioactivity and evaluation of mass balance was not possible because tissue burdens and overall body burden could not be assessed and biliary contribution to excretion were not measured. Recovery of radioactivity in the urine indicated that 73.79% of the administered radioactivity was excreted over 72 hours. This implies that absorption of the 200 mg/kg dose was likely >74%. The majority of urinary elimination occurred during the first 24 hours and appeared to be nearly complete by 72 hours.

Treatment of plasma and urine with β-glucuronidase and arylsulfatase revealed that the majority of the radioactivity in these matrices could be attributed to conjugation products rather than unchanged parent compound. For urine, 93.85% of the recovered radioactivity could be attributed to glucuronide conjugates while approximately 2.3% could be attributed to sulfate conjugates. For plasma, 51.87- 60.62 % of the radioactivity could be attributed to glucuronide conjugates while enzymatic hydrolysis revealed that the sulfate conjugate represented 31.61- 43.50% of the radioactivity. Minor changes in the relative contributions of these metabolites over time were detected but were of uncertain significance.

In summary, the report provided anecdotal data showing that orally administered triclosan undergoes Phase II metabolism to conjugation product metabolites (sulfate and glucuronide conjugates) which are eliminated via the urine within 72 hours.

This metabolism study using a human volunteer is **Acceptable/Nonguideline** but does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Although properly conducted and providing data regarding the

44

metabolism of triclosan in a human volunteer following a single oral dose, the data were anecdotal, the protocol did not include tissue distribution assessments, and no information was provided regarding dose stability and confirmation or homogeneity. Therefore, the study was not consistent with an 85-1 Guideline study. The results, however, do affirm findings of companion studies (MRID 149464, 79590) in other animal species and provide important information based upon human data.

B. STUDY DEFICIENCIES

The data are anecdotal. Although the findings indicate that there are two major conjugation products which are excreted in the urine, the experiment could not address species or dose related variability.

There was no quality assurance statement and no information regarding dose confirmation, homogeneity or stability.

DATA EVALUATION REPORT

TRICLOSAN (GP 41 353)

Study Type: METABOLISM AND PHARMACOKINETICS
[OPPTS 870.7485 (§85-1)]
MRID 79590

Prepared for

Antimicrobials Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. T07

Primary Reviewer: Robert A. Young, Ph.D., D.A.B.T.	Signature:	Robert A. Young
	Date:	OCT 0 6 2000
Secondary Reviewers: H. T. Borges, MT(ASCP),Ph.D., D.A.B.T.	Signature:	H T Borges
	Date:	OCT 0 6 2000
Robert H. Ross, M.S., Group Leader	Signature:	Robert H. Ross
	Date:	OCT 0 6 2000
Quality Assurance: Lee Ann Wilson, M.A.	Signature:	L. A. Wilson
	Date:	OCT 0 6 2000

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UTBattelle, LLC, for the U.S. Dept. of Energy under contract DEAC0500OR22725

EPA Reviewer: Tim McMahon Ph.D.
Antimicrobials Division (7510C)

_____, Date 10/25/00

DATA EVALUATION RECORD

STUDY TYPE: Metabolism - OPPTS [870.7485 (§85-1)]

DP BARCODE: D266143
P.C. CODE: 054901

SUBMISSION CODE: Not available
TOX. CHEM. NO.: None

TEST MATERIAL (PURITY): Triclosan (chemical purity 99.5%)

SYNONYMS: GP 41 353; Triclosan;5-chloro-2-(2,4-dichlorophenoxy)phenol

CITATION: Stierlin, H. (1976). GP 41 353: Isolation and identification of the main metabolites in the blood of the beagle and baboon and in the urine of the latter following oral administration of ^{14}C-labelled triclosan. Pharma Research, Pharmacological Chemistry. CIBA-GEIGY, Ltd. Basle. Report No. B 14/1976. MRID 79590. March 16, 1976. Unpublished.

SPONSOR: CIBA-GEIGY Limited, Pharma Research, Pharmacological Chemistry, Basle.

EXECUTIVE SUMMARY: In a metabolism study (MRID 79590), two baboons and two dogs were administered a single oral dose (5 mg/kg) of [^{14}C]-GP 41 353 (radiochemical purity 99%; chemical purity 99.5%; batch/lot nos. not provided). Blood samples were taken at 3 hours postdosing from one dog and at 8 and 12 hours postdosing from one baboon. Another dog and baboon were killed at 6 and ~7 hours postdosing to obtain sufficient blood samples for metabolite characterization. Urine samples were collected from the dog and baboon that were not sacrificed for blood sample acquisition.

There were no adverse effects associated with the test article. At 3 hours postdosing, total radioactivity was 6.08 μg eq./mL blood in the dog. For the baboon total radioactivity was 1.24 and 1.03 μg eq./mL blood, respectively at 8 and 12 hours postdosing. For the dog and baboon terminated for sample acquisition, total blood radioactivity was 4.86 μg eq./mL blood for the dog at 6 hours and 2.29 μg eq./mL blood for the baboon at ~7 hours postdosing. Urinary elimination accounted for 32% of the administered dose to the surviving baboon by 72 hours. Urinary excretion by the dog was minimal and accounted for "only a few percent of the administered dose".

Hydrolysis of the biological samples with arylsulfatase and β-glucuronidase resulted in the release of free triclosan indicating that the measured radioactivity was associated with sulfate and glucuronide conjugation products. For the dog, glucuronide conjugates accounted for 7% of the blood radioactivity and sulfate conjugates accounted for 88% of the radioactivity sampled at

50

three hours. At six hours, 7.2% of the radioactivity in the blood was associated with glucuronide conjugates, and the remaining percentage was not identified except through combined glucuronidase/sulfatase hydrolysis (86.3%). For the baboon, glucuronide conjugates represented about 25% and sulfate conjugates represented about 33% of the circulating radioactivity at 8 hours postdosing. Similar analysis with blood collected at 12 hours postdosing, revealed somewhat greater amounts of sulfate conjugates (~43.9%) and less glucuronide conjugate (10.6%). For the baboon, analysis of 0-72 hour urine samples revealed that approximately 6% of the urinary radioactivity was due to unchanged triclosan. Up to 75% of the urinary radioactivity underwent spontaneous hydrolysis presumably due to endogenous urinary β-glucuronidase. No analyses were conducted for the dog due to the minimal urinary elimination of radioactivity.

In summary the results of this study showed that the major blood metabolites in the baboon and beagle dog were sulfate and glucuronide conjugation products. The major urinary metabolite in the baboon was a glucuronide conjugate but analysis of urinary metabolites in the dog was precluded by negligible urinary products as determined by radioactivity.

This metabolism study is **unacceptable/upgradable** and does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Although the study appears to have been properly conducted and provided data regarding the characterization of blood and urinary metabolites in the baboon and blood metabolites in the dog following a single oral dose, several deficiencies exist, including lack of test article characterization, lack of animal husbandry and environmental control information, lack of dosing administration, and lack of dose solution stability, homogeneity, and dose confirmation. If these data can be provided, this study can be upgraded to acceptable. Otherwise, a new metabolism study must be conducted.

COMPLIANCE: Good Laboratory Practice (not applicable), Quality Assurance, and Data Confidentiality statements were not included with the study report. Flagging statements were not included.

I. MATERIALS AND METHODS

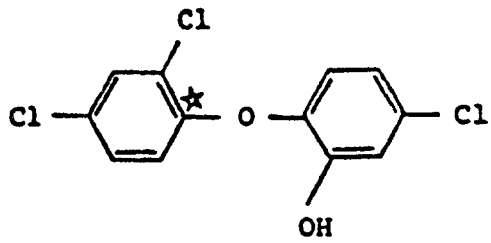
A. MATERIALS

1. Test compound

Radiolabeled: [^{14}C]-GP 41 353 (sp. act. not specified)
Batch No.: not provided
Purity:
  Radiochemical purity: 99.5%
  Chemical purity: 99.5%
Description: not available in report
Contaminants: none noted
CAS No.: 3380-34-5
Stability: report provided no information

57

Structure:



[^{14}C]-GP 41 353

Non-radiolabeled: none indicated
Purity: NA
Batch No.:NA
Description: NA
Contaminants: NA
CAS No.: NA
Stability: NA

2. Vehicle
No information provided

3. Test animals

Species: dog, monkey
Strain:
Dog: beagle (male)
Monkey: baboon (male)
Age and weight at study initiation:
Dog: 11.3 and 10.0 kg; age not specified
Baboon: 8 kg and 12 kg; age not specified
Source: sources of test animals were not specified
Housing: no information provided
Diet: no information provided
Water: no information provided
Environmental conditions:
Temperature: not provided
Humidity: not provided
Air changes: not provided

Photoperiod: not provided
Acclimation period: not provided

4. Preparation of dosing solution

No information was provided.

Results –
Homogeneity: no data provided

Stability: no data provided

Dose confirmation: no data provided

B. STUDY DESIGN AND METHODS

1. Group arrangements

The experiments utilized two baboons and two dogs, each receiving a single oral dose of 5 mg ^{14}C-triclosan/kg body weight (Table 1).

TABLE 1. Study design

Experimental group	Dose (mg/kg)	Number/Sex	Remarks
Dog	5	2 males	blood samples taken at 3 hrs for one dog; for second dog, blood concentrations were monitored hourly to 6 hrs (T_{max}) at which time the dog was killed by exsanguination for isolation of metabolites; urine was collected for 3 days from first dog
Baboon	5	2 males	blood taken at 8 and 12 hrs post-dose and urine at 24-hr intervals for 72 hrs from one baboon; blood collected at 7 hrs from second baboon; no urine collected from second baboon.

Information taken from p. 17, MRID 79590.

2. Dosing and sample collection

Animals were given the test article orally; no further details were provided (i.e., gavage, gastric intubation, capsule).

Expired air - Elimination via expired air was not assessed.

Blood - A 25 mL blood sample was taken from one dog (11.3 kg) three hours postdose for metabolite analysis. To monitor radioactivity levels, blood was sampled every hour for six hours in the second dog after which the dog was killed and a 700 mL sample taken. Blood samples were taken from the baboon at eight and 12 hours

53

postdose. For isolation of metabolites, blood from one dog and one monkey was diluted with water (1:5), and metabolites separated using various gel filtration and chromatographic protocols.

Urine - Urine was collected over three days from the 11.3-kg dog. Urine was collected over ice from the baboon at 24-hr intervals for 72 hours. Urine was stored "deep frozen" prior to analysis by gel filtration and chromatography processes.

Feces - Feces were not collected.

Cage wash - There was no indication that cage wash was collected.

Tissues - Tissues were not examined.

3. Sample preparation/analysis

Urine - For determination of radioactivity, urine samples were measured directly following addition of scintillation fluid. For metabolite analysis, urine sample were adjusted to a pH of 1-2 with 1 N hydrochloric acid, and extracted several times in petroleum ether. Preparative gel filtration (described below) was also used for concentrating metabolites. The extracts were analyzed by TLC using a reference standard. Determination of glucuronide conjugates was performed by incubating urine samples at pH 6.8 at 37°C for 24 hours with β-glucuronidase.

Blood - Blood samples (0.5 mL) were prepared for radioactivity determination as described under LSC techniques. Preparative gel filtration (described below) was also used for concentrating metabolites. For metabolite determination, blood samples were subjected to various hydrolytic processes for determination of conjugates. These included enzymatic hydrolysis using β-glucuronidase/arylsulfatase, β-glucuronidase alone, arylsulfatase alone, and acid hydrolysis.

4. Analytical techniques

Liquid scintillation counting (LSC)
Quench-corrected LSC was performed using a Packard Model 3380 counter. Urine samples were counted directly following addition to Diotol scintillation fluid. Blood samples were dissolved in Soluene and bleached with hydrogen peroxide or combusted prior to counting.

Thin-layer chromatography (TLC)
TLC utilized Antec SL 254 plates scanned with a Berthold Radioactivity Scanner or a Packard Actigraph. The solvent system was chloroform/methanol/ acetic acid (70:25:1) or chloroform/acetone/acetic acid (95:10:2).

Preparative gel filtration

54

Prior to chromatographic analyses, preparative filtration (Amberlite XAD resin, Sephadex LH20, Merck Silica gel G, Merck Lichrosorb) was performed on blood and urine samples. Flow charts delineating the procedures were provided that adequately described the analytical protocols.

High Performance Liquid Chromatography (HPLC)
HPLC (Chromatronix equipment) was used for isolation of metabolites from samples. No further details were provided in the study report.

Electrophoresis
Paper electrophoresis was performed on Whatman No.1 paper using a 0.1N veronal buffer (pH 8.0) at 220-300v, 10 mA for 2-4 hours. Resulting profiles were compared radiographically with labeled reference compounds.

Mass Spectrometry (MS)
For final identification of metabolites, MS was performed using a Varian MAT CH5-DF mass spectrophotometer (70 eV, 200°C ion source temperature). Infrared spectra were recorded with a Perkin Elmer Model 221 spectrophotometer and NMR spectra were recorded using a Varian HA 100 spectrophotometer.

Derivatization and structural analysis
Hydrolysis of samples with β-glucuronidase and arylsulfatase was used to generate free triclosan. To obtain the methylester derivative, samples were esterified with 1% 1-methyl-3-p-tolyl-triazine in ether for 4 hours at 30°C and extracted with ether.

5. Statistics and calculations

No statistical methods or calculations were described.

II. RESULTS

A. DISTRIBUTION/EXCRETION STUDIES

1. Mass balance

Because the goal of this study was metabolite characterization, mass balance data were incomplete. However, a companion study in rats, mice, rabbits and dogs (MRID 149464) indicated acceptable recoveries of radioactivity following oral or intravenous administration.

2. Absorption
Determination of absorption was not a component of the experimental protocol.

3. Excretion

Although definitive determination of excretion was not a component of the experimental protocol, 32% of the administered dose to the baboons was recovered in the urine by 72 hours. Urinary excretion in the dog was minimal ("only a few percent of the administered dose").

4. Tissue distribution
A complete assessment of tissue distribution was not a component of the experimental protocol. Concentration of ^{14}C-compounds in the blood was 6.1 μg/mL for the dog at three hours and 1.2 μg/mL for the baboon at eight hours.

B. PHARMACOKINETIC STUDIES

Determination of pharmacokinetic parameters was not a protocol element.

C. METABOLITE CHARACTERIZATION STUDIES

1. Blood
Based upon results of hydrolysis using β-glucuronidase and arylsulfatase in the dog after 3 hours post-dose (5 mg/kg), 7.3% of the dose was reported as the glucuronide conjugate and 88.6% as the sulfate conjugate. In baboons, after 8 hours, 24.5% was reported as the glucuronide conjugate and 33.2% as the sulfate conjugate. Similar analysis with blood collected at 12 hours postdosing in the baboon revealed somewhat greater amounts of sulfate conjugates (43.9%) and less glucuronide conjugate (10.6%). It is noted that the percentages cited in the baboon leave a significant percentage of the total blood radioactivity unaccounted for in the baboon.

After 6 hours from a 5 mg/kg dose in a single dog, 7.2% of the dose was identified as the glucuronide conjugate of triclosan and no percentage was given for the sulfate conjugate. In the baboon, after 7 hours, 13.3% of the dose was identified as the glucuronide conjugate and 87.7% as the sulfate conjugate.

2. Urine
An analysis of 0-72 hour urine samples from the baboon revealed that approximately 6% of the urinary radioactivity was due to unchanged triclosan. Up to 75% of the urinary radioactivity underwent spontaneous hydrolysis presumably due to endogenous urinary β-glucuronidase.

For the dog, no analyses were conducted due to the minimal urinary elimination of radioactivity.

D. HISTOPATHOLOGY

Histopathologic assessment was not a component of the reviewed study.

56

III. DISCUSSION

A. DISCUSSION

In a metabolism study (MRID 79590), two baboons and two dogs were administered a single oral dose (5 mg/kg) of [^{14}C]-GP 41 353 (radiochemical purity 99%; chemical purity 99.5%; batch/lot nos. not provided). Blood samples were taken at 3 hours postdosing from one dog and at 8 and 12 hours postdosing from one baboon. Another dog and baboon were killed at 6 and ~7 hours postdosing to obtain sufficient blood samples for metabolite characterization. Urine samples were collected from the dog and baboon that were not sacrificed for blood sample acquisition.

There were no adverse effects attributed to the test article administration. At 3 hours postdosing, total radioactivity was 6.08 μg eq./mL blood in the dog. For the baboon total radioactivity was 1.24 and 1.03 μg eq./mL blood, respectively at 8 and 12 hours postdosing. For the dog and baboon terminated for sample acquisition, total blood radioactivity was 4.86 μg eq./mL blood for the dog at 6 hours and 2.29 μg eq./mL blood for the baboon at ~7 hours postdosing. Urinary elimination accounted for 32% of the administered dose to one of the baboons by 72 hours. Urinary excretion in the dog was minimal and accounted for "only a few percent of the administered dose".

Hydrolysis of samples with arylsulfatase and β-glucuronidase resulted in the release of free triclosan indicating that the measured radioactivity was associated with sulfate and glucuronide conjugation products. For the dog, glucuronide conjugates accounted for 7% of the blood radioactivity and sulfate conjugates accounted for 88% of the radioactivity sampled at three hours. At six hours, 7.2% of the radioactivity in the blood was associated with glucuronide conjugates, and the remaining percentage was not identified except through combined glucuronidase/sulfatase hydrolysis (86.3%). For the baboon, glucuronide conjugates represented about 25% and sulfate conjugates represented about 33% of the circulating radioactivity at 8 hours postdosing. Similar analysis with blood collected at 12 hours postdosing, revealed somewhat greater amounts of sulfate conjugates (~43.9%) and less glucuronide conjugate (10.6%). For the baboon, analysis of 0-72 hour urine samples revealed that approximately 6% of the urinary radioactivity was due to unchanged triclosan. Up to 75% of the urinary radioactivity underwent spontaneous hydrolysis presumably due to endogenous urinary β-glucuronidase. No analyses were conducted for the dog due to the minimal urinary elimination of radioactivity.

In summary the results of this study showed that the major blood metabolites in the baboon and beagle dog were sulfate and glucuronide conjugation products. The major urinary metabolite in the baboon was a glucuronide conjugate but analysis of urinary metabolites in the dog was precluded by negligible urinary products as determined by radioactivity.

This metabolism study is **unacceptable/upgradable** and does not satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Although the study appears to have been properly conducted and provided data regarding the characterization of blood and urinary metabolites in the baboon and blood metabolites in the dog following a single oral dose, several deficiencies exist, including lack of test article characterization, lack of animal husbandry and environmental control information, lack of dosing administration, and lack of dose solution stability, homogeneity, and dose confirmation. If these data can be provided, this study can be upgraded to acceptable. Otherwise, a new metabolism study must be conducted.

B. STUDY DEFICIENCIES

The deficiencies in this study are noted above.

DATA EVALUATION REPORT

TRICLOSAN (GP 41 353)

Study Type: METABOLISM AND PHARMACOKINETICS – MULTIPLE SPECIES
[OPPTS 870.7485 (§85-1)]
MRID 149464

Prepared for

Antimicrobials Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Chemical Hazard Evaluation Group
Toxicology and Risk Analysis Section
Life Sciences Division
Oak Ridge National Laboratory
Oak Ridge, TN 37831
Task Order No. T07

Primary Reviewer:		
Robert A.Young, Ph.D., D.A.B.T.	Signature:	Robert A. Young
	Date:	OCT 0 6 2000
Secondary Reviewers:		
H. T. Borges, MT(ASCP),Ph.D., D.A.B.T.	Signature:	H T Borges
	Date:	OCT 0 6 2000
Robert H. Ross, M.S., Group Leader	Signature:	Robert H. Ross
	Date:	OCT 0 6 2000
Quality Assurance:		
Lee Ann Wilson, M.A.	Signature:	L. A. Wilson
	Date:	OCT 0 6 2000

Disclaimer

This review may have been altered subsequent to the contractor's signatures above.

Oak Ridge National Laboratory, managed by UTBattelle, LLC, for the U.S. Dept. of Energy under contract DEAC05000R22725

EPA Reviewer: Tim McMahon, Ph.D.
Antimicrobials Division

_____, Date _____

DATA EVALUATION RECORD

STUDY TYPE: Metabolism - Multiple Species; OPPTS [870.7485 (§85-1)]

DP BARCODE: D266143
P.C. CODE: 054901

SUBMISSION CODE: None
TOX. CHEM. NO.: None

TEST MATERIAL (PURITY): Triclosan (chemical purity 99.5%)

SYNONYMS: GP 41 353; Triclosan;5-chloro-2-(2,4-dichlorophenoxy)phenol

CITATION: Stierlin, H. (1972). GP 41 353: Study of pharmacokinetics and metabolism in mouse, rat, rabbit, and dog. Pharma Research, Pharmacological Chemistry. CIBA-GEIGY, Ltd. Basle. Project No. GP 41 353. Report No. 33/1972. MRID 149464. December 1, 1972. Unpublished.

SPONSOR: CIBA-GEIGY Limited, Pharma Research, Pharmacological Chemistry, Basle.

EXECUTIVE SUMMARY: In a metabolism study (MRID 149464), rats, mice, rabbits, and dogs were administered [^{14}C]-GP 41 353 or [^{3}H]-GP 41 353 (radiochemical purity 99%; chemical purity 99.5%; batch/lot nos. not provided) intravenously, intraduodenally, or orally (gavage) at doses of 10 mg/kg (mice), 0.4 mg/kg (rats), 5 mg/kg (rats, rabbits, dogs), or 50 mg/kg (rabbits). Radioactivity levels in the blood, tissues, and excreta were measured for time intervals up to 168 hours. Additionally, biliary elimination was also analyzed in rats given a single intraduodenal dose.

There was no indication of any toxic effects in the test animals. Recovery of administered radioactivity at 2, 4, 6, and 8 hours ranged from 99.67 to 104.53 % in rats given a single 5 mg/kg oral dose of [^{3}H]-GP 41 353. Data were unavailable in the study report to accurately determine radioactivity inventory for the other species tested.

Absorption of the test material was reported by the study author as 70-80% (for rats) as determined by comparisons of areas under-the-blood concentration curve for oral and intravenous administrations. These data were, however, unavailable for the preparation of this data evaluation report. Absorption in rats could also be estimated based upon biliary and urinary elimination data. Over a 7.5 to 10-hour period, biliary elimination accounted for 62.5% of a 5 mg/kg gavage dose and 67% of a 5 mg/kg intraduodenal dose while urinary excretion at 6 and 8 hours accounted for 76.60% and 5.17%, respectively, of a 5 mg/kg gavage dose. These data suggest that absorption is in excess of 70%. Absence of biliary elimination data for the other test species precluded assessment of the extent of absorption in those species; however, biliary

60

excretion data in rats and mice suggest enterohepatic recirculation of the test material in both species. Time-course concentration data revealed that peak blood levels occurred within 30 minutes in rats following a single oral or intravenous dose of 5 mg/kg and at 2-4 hours for dogs.

Tissue distribution patterns were similar among mice and rats, and exhibited only slight quantitative variability for intravenous versus oral dosing. Following a single oral dose in rats, radioactivity in tissues was low (generally <1 μg/g tissue) with the exception of blood and the organs associated with excretory function (e.g., liver, gall bladder, kidneys). Following an intravenous dose to rats and mice, tissue levels were also greatest in highly perfused organs or those associated with excretory function. Based upon data from rats, tissue burdens declined appreciably over 24 hours with no indication of accumulation/sequestration.

Excretion of GP 41 353 was examined in two strains of rats, rabbits, and dogs. Biliary elimination was also assessed in rats. Urinary excretion appeared to be a minor route of elimination in rats and dogs, accounting for 3-17% of the administered oral dose in rats over a 168-hour period, and 8.3-8.8% in dogs over a 120-hour period. Urinary elimination in dogs was somewhat greater following intravenous administration; 12.9-17.7% over 120 hours. For rabbits, urinary excretion was a significant route of elimination and accounted for 74.1% of a single 50 mg/kg oral dose and 60.4% of a single 5 mg/kg oral dose over a 72-hour period. The biliary secretion data in rats showed that most of the fecal radioactivity could be attributed to biliary elimination products rather than unabsorbed test material. Biliary elimination was also affirmed by data from the mouse showing very high concentrations of radioactivity in the gall bladder at 5 minutes to two hours following a 10 mg/kg, i.v. dose.

Analysis of bile samples from the rats indicated that the test material underwent Phase II biotransformation. Treatment of samples with β-glucuronic acid revealed that most of the biliary product was glucuronide conjugates while some was unchanged parent compound.

The results of this multi-species study indicated that at least 70% of an oral dose of GP 41 353 is absorbed from the gastrointestinal tract and that biliary excretion and subsequent fecal elimination is a major excretory route in the rat and dog. Urinary excretion appeared to be a major route of elimination in the rabbit. Tissue accumulation was minimal after a single dose and was primarily associated with highly perfused tissues and organs with excretory function, although a companion study (MRID 68161) shows distribution of triclosan-derived radioactivity into the pituitary gland, optic nerve, and sciatic nerve at concentrations equivalent to those found in highly perfused organs at the same dose level (5 mg/kg). Repeated oral dosing at 5 mg/kg leads to accumulation of radioactivity in the blood, plasma, pituitary gland, sciatic nerve, and kidneys (MRID 68161). Metabolite data in rats revealed glucuronide conjugates and unchanged parent compound as biliary metabolites.

This metabolism study is **unacceptable/upgradable** and does not currently satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Numerous deficiencies were observed in this report, including information regarding dose confirmation, homogeneity, and stability, environmental and housing conditions of the animals used in this study, and a lack of statements regarding compliance with Good Laboratory Practice and Quality Assurance. If the missing information can be provided, this study can be upgraded.

61

Otherwise, a new guideline metabolism study will need to be conducted. Despite the deficiencies, the data do provide useful information on the disposition of Triclosan in multiple species and can be considered in conjunction with an acceptable metabolism study if such study is necessary.

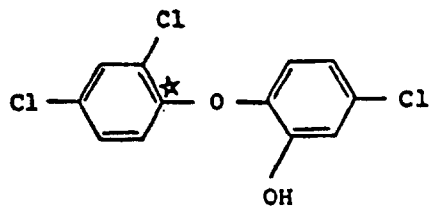
COMPLIANCE: Good Laboratory Practice (not applicable), Quality Assurance, and Data Confidentiality statements were not included with the study report. Flagging statements were not included.

I. MATERIALS AND METHODS

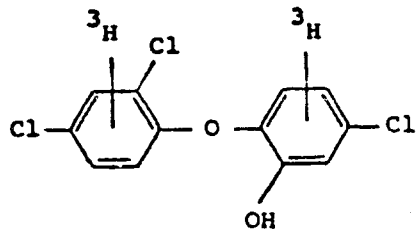
A. MATERIALS

1. Test compound

Radiolabeled: [^{3}H]-GP 41 353 (sp. act. 62.7 μCi/mg)
[^{14}C]-GP 41 353 (sp. act. 13.25 μCi/mg)
Batch No.: not provided
Purity:
Radiochemical purity: 99%
Chemical purity: 99.5%
Description: white crystalline powder
Contaminants: none noted
CAS No.: 3380-34-5
Stability: report noted stability in neutral, acidic, and alkaline media
Structure:



[^{14}C]-GP 41 353



2. Vehicle

For intravenous administration, the test material was administered in 1N sodium carbonate. For gavage administration, the test material was administered in ethanol/water (1:9).

3. Test animals

Species: mouse, rat, rabbit, dog
Strain:
Mouse: male BALB/C x MJW
Rat: male and female Wistar WU and SIV-50
Rabbit: male, mixed breed
Dog: male beagles
Age and weight at study initiation:
Mice: 20±1 g; age not specified
Rats: 200-250 g; age not specified
Rabbits: 2-3 kg; age not specified
Dogs: 10 and 14 kg; age not specified
Source: sources of test animals were not specified
Housing: during the experimental period, animals were kept in metabolism cages (no details provided)
Diet: NAFAG No. 85 (NAFAG, Gossau, Switzerland) was given *ad libitum* to rats and rabbits but withdrawn 15 hours prior to the start of the experiment and resumed two hours after test article administration. Diet regimen not specified for mice and dogs.
Water: tap water *ad libitum*
Environmental conditions:
Temperature: not provided
Humidity: not provided
Air changes: not provided
Photoperiod: not provided
Acclimation period: not provided

4. Preparation of dosing solution

For intravenous administration the test material was dissolved in 1N sodium carbonate to provide a concentration of 0.2 mg/0.1 mL. For gavage administration, the test material was dissolved in ethanol/water (1:9) to allow for dosing at 0.4 or 5 mg/kg. No further details were provided.

Results –
Homogeneity: no data provided

Stability: no data provided

Dose confirmation: no data provided

B. STUDY DESIGN AND METHODS

1. Group arrangements

The brief descriptions of experimental protocol identified the experimental groups shown in Table 1. There was no indication of procedures for group arrangements.

TABLE 1. Study design

Experimental group	Dose (mg/kg)	Number/Sex	Remarks
Mice; i.v.	10	2 males/time period	[^{14}C]-GP 41 353; tissue distribution, whole-body autoradiography; two mice per time period (1 min., 5 min., 30 min., 2 hrs)
Rats; oral	0.4	4 males; 4 females	SIV-50 rats; [^{3}H]-GP 41 353; time course in urine and feces
	5.0	3 females	Biliary elimination Wistar WU; [^{3}H]-GP 41 353; time course in urine and feces
Rats, i.v.	5.0	4 rats/time period	gender and strain not specified; [^{3}H] radiolabled GP 41 353; blood concentration at 0.5, 1, 2, 4, 8, and 24 hrs.
Rats, i.v.	5.0	5 rats/time period	gender and strain not specified; [^{3}H]-GP 41 353; blood concentration at 10 min., 0.5, 1, 2, 4, 8, and 24 hrs.
Rat; intraduodenal	5.0	3 rats	Wistar strain; intraduodenal administration of [^{3}H]-GP 41 353 to assess biliary elimination.
Rabbits; oral	5.0	3 males	[^{3}H]-GP 41 353; time course in urine and feces
	50	3 males	[^{3}H]-GP 41 353; time course in urine and feces
Dogs; i.v./oral	5.0/5.0	2 males	single i.v. dose followed 15 days later with single oral dose; time course in blood, urine, feces

Information taken from pp. 12-14, Tables 1-3, pp. 22-24, Table 8, p. 29, and Tables 9-10, pp. 30-31, MRID 149464.

2. Dosing and sample collection

Animals were given the test article intravenously (tail vein of rats; no details provided for dogs), via gastric intubation, or intraduodenally.

Expired air - Elimination via expired air was not assessed.

Blood - Blood samples were taken from rats via cardiac puncture at 10 minutes (i.v. group only), and 0.5, 1, 2, 4, 8, and 24 hours after administration of the test material. For dogs, blood samples were taken (site not specified) from each dog at 10 min., 30 min., and 1, 2, 4, 8, 24, and 48 hours after the oral dose.

Urine - Urine was collected from low-dose and high-dose rats at 24-hour intervals over 3-7 days. Urine was also collected from rabbits at 24-hour intervals for three days and from dogs at 24-hour intervals for five days.

Feces - Feces were collected on the same schedule as described for urine collection.

Cage wash - Cage wash was not indicated as a collected matrix.

Tissues - Tissues were collected from two mice at four time intervals (2, 4, 8 and 24 hrs.) and from two orally and four intravenously treated rats at the same time points as described for blood collection. Tissues included heart, muscle, brain, bone marrow, testis, lung, adrenal gland, spleen, thyroid, liver, kidney, intestine, gastrointestinal contents, stomach, and carcass.

3. Sample preparation/analysis

Urine - For determination of radioactivity, urine samples were measured directly following addition to scintillation fluid. For metabolite analysis, urine samples were adjusted to a pH of 1-2 with 1 N hydrochloric acid, and extracted several times in petroleum ether. The extracts were analyzed by TLC using a reference standard. Determination of glucuronide conjugates was performed by incubating urine samples at pH 6.8 at 37°C for 24 hours with β-glucuronidase.

Feces - Fecal samples were homogenized and freeze-dried. Aliquots of these samples were combusted and radioactivity determined. Freeze-dried fecal samples were also extracted three times in methanol, twice in methanol/water, once in water, and three times in acetone. The extracts were then analyzed for unchanged parent compound by adjusting the pH to 1-2 with 1 N hydrochloric acid followed by TLC analysis with a reference standard. Determination of glucuronide conjugates was performed by incubating fecal extracts at pH 6.8 at 37°C for 24 hours with β-glucuronidase.

Bile - For radioactivity determination, bile samples were prepared as described for urine samples. For metabolite determination, bile samples (0.1-0.5 mL) were bleached in hydrogen peroxide and dissolved in hyamine hydroxide. The extracts were then analyzed for unchanged parent compound by adjusting pH to 1-2 with 1 N hydrochloric acid followed by TLC analysis with a reference standard.

Blood - Blood samples (0.5 mL) were dried on filter paper, combusted, and the radioactivity determined.

Tissue/organs - Tissue/organs were homogenized and aliquots dissolved in hyamine hydroxide at 45°C for 15 hours or aliquots (100 mg) were combusted and analyzed. The carcass was dissolved in 1 N sodium hydroxide at 70°C, emulsifier (not specified) added, and 1.0 mL aliquots counted for radioactivity.

Metabolite analysis- To assess unchanged parent compound, fecal extracts, bile, and urine samples were adjusted to a pH of 1-2 with hydrochloric acid and repeatedly extracted with ether. To assess glucuronides, urine samples were pH adjusted to 6.8 and incubated with β-glucuronidase at 37°C for 24 hours. Freeze-dried fecal samples were extracted in methanol (3X), methanol/water (2X), water (1X) and acetone (3X) for 15-30 minutes.

4. Analytical techniques

Liquid scintillation counting (LSC)
Quench-corrected LSC was performed using Packard Model 4000 and 3380 counters. The following fluids were used: 1) dioxane 1000 mL, glycol monoethylether 200 mL, PPO 12 g, POPOP 0.6 g, and naphthalene 60 g; 2) dioxane 1000 mL, toluene 1000 mL, methanol 600 mL, PPO 13 g, POPOP 0.26 g, naphthalene 208 g, or 3) toluene 1000 mL, butyl BPD 8 g.

Thin-layer chromatography (TLC)
Developed Merck Silicagel F 254 or Antec SL 254 TLC plates were scanned with a Berthold Radioactivity Scanner or a Packard Actigraph. The solvent system was benzene/toluene/methanol/formic acid (50:50:10:5).

Autoradiography
Distribution of radioactivity in mice was determined using whole-body autoradiography. Mice were killed at 1 minute, 30 minutes, 2 hours and 24 hours after administration of 10 mg test article/kg. The mice were frozen at -80°C and sectioned at -20°C. The sections were placed on photographic plates for 15 days at -20°C.

5. Histopathology

Histopathologic evaluations were not conducted.

6. Statistics and calculations

Group means and standard deviations were provided.

II. RESULTS

A. DISTRIBUTION/EXCRETION STUDIES

1. Mass balance

Recovery of administered radioactivity at 2, 4, 6, and 8 hours ranged from 99.67 to 104.53 % in rats given a single 5 mg/kg oral dose of [^{3}H]-GP 41 353. Mass balance data for the other species tested could not be assessed due to an absence of tissue

burden data. Radioactivity accounting data (average for two rats each) at 2, 4, 6, and 8 hours after a single 5 mg/kg oral dose are shown in Table 2.

TABLE 2. Overall recovery of administered radioactivity (% of dose) from rats at various times after a single oral dose of 5 mg GP 41 353/kg

	2 hrs	4 hrs	6 hrs	8 hrs
Expired air	ND	ND	ND	ND
Urine	0.21	0.47	0.60	2.44
Gastrointestinal content	71.91	78.32	76.60	5.17
Feces	-	-	13.42	89.70
Tissues	18.52	12.11	5.68	0.95
Carcass	13.85	8.93	9.54	2.84
Total	104.53	99.67	105.87	101.10

Data taken from Table 6, p. 27, MRID 14 9464.

2. Absorption

Absorption of the test material may be implied from data on blood concentration, renal and biliary excretion, and body burden. At 10 hours following an intraduodenal or oral dose, biliary elimination accounted for 67% and 62.5%, respectively, of the administered dose, indicating significant absorption of the test material. In addition, comparison of blood levels after oral or intravenous administration of a 5 mg/kg dose (Table 3, page 24 of the report) show equivalent blood concentrations, implying significant orla absorption of the test material.

3. Excretion

Time-course data for urinary and fecal excretion by rats over 72 or 168 hours are shown in Tables 3 and 4, respectively. Excretion via the urine was a minor route of elimination for the rat, accounting for 3-17.1% of the administered dose. Based upon the available data, the significance of the apparent gender difference in urinary excretion is uncertain. However, biliary excretion data (Figure 1 and Table 7 of the report) suggest enterohepatic recirculation of the test material in both rats and mice.

Table 3. Time-course for urinary excretion (% of dose) of [^{3}H]-GP 41 353 by rats following oral administration.				
Time (hrs)	Single low dose (0.38 mg/kg) (SIV-50 rats)		Single high dose (5 mg/kg) (Wistar rats)	
	Male	Female	Males	Females
0-4	-	-	0.1±0.07	-
4-8	-	-	0.6±0.4	-
8-24	-	-	1.3±0.2	-
0-24	1.7±0.3	14.1±1.8	-	-
24-48	0.9±0.2	1.9±0.7	0.6±0.1	-
48-72	0.4±0.08	0.7±0.3	0.4±0.1	-
72-96	0.3±0.06	0.2±0.05	-	-
96-120	-	0.1±0	-	-
120-144	-	<0.05	-	-
144-168	-	0.1	-	-
Total	3.3	17.1	3.0±0.09[a]	-

[a] Total is for 0-72 hrs only.
Data taken from Table 2, p. 23, MRID 149464.

Table 4. Time-course for fecal excretion (% of dose) of [^{3}H]-GP 41 353 by rats following oral administration.				
Time (hrs)	**Single low dose (0.38 mg/kg) (SIV-50 rats)**		**Single high dose (5 mg/kg) (Wistar rats)**	
	Male	**Female**	**Males**	**Females**
0-24	52.1±11.8	42.9±3.8	87.4±1.5	-
24-48	22.1±7.9	18.8±7.1	3.8±1.9	-
48-72	3.0±1.2	4.7±1.2	0.1±0.07	-
72-96	0.8±0.3	1.2±0.3	-	-
96-120	-	0.3±0.07	-	-
120-144	-	0.1	-	-
144-168	-	0.1	-	-
Total	78.0	68.1	91.3±2.0[a]	-

[a] Total is for 0-72 hrs only.
Data taken from Table 2, p. 23, MRID 149464.

In the dog, urinary elimination of a single 5 mg/kg dose was minor relative to elimination in the feces regardless of administration route (oral or intravenous; data not tabled in this report). For two male dogs, total urinary elimination over 120 hours

accounted for 12.9% and 17.7% of the administered intravenous dose and 8.3% and 8.8% of the oral dose. For the same two dogs, fecal elimination accounted for 67.4% and 70.1% of the intravenous dose and 69.8% and 71.4% of the oral dose. The majority (66-87%) of the fecal elimination occurred within 24 hours for both administration routes.

Data from three male rabbits given 5 or 50 mg/kg by gavage revealed an excretory pattern differing from that observed for rats and dogs (Table 5). For both dose groups, urinary elimination accounted for a greater proportion of the administered dose than did fecal elimination. The extensive variability about the mean values, however, suggested notable individual variability especially in urinary excretion. In the absence of data on urine volume, it is difficult to determine if the variability was due to excretory transport processes or simply to variability in micturition. The available data for the two dose groups did not imply the presence of saturated absorption/excretion processes.

Table 5. Time-course for fecal and urinary excretion (% of dose) of [^{3}H]-GP 41 353 by male rabbits following oral administration[a].

Time (hrs)	Single low dose (5 mg/kg)		Single high dose (50 mg/kg)	
	Urine	Feces	Urine	Feces
0-24	57.2±12.4	16.3±6.7	38.5±19.6	11.6±2.9
24-48	15.9±9.1	2.9±1.1	21.6±20.8	2.2±0.7
48-72	1.0±0.4	3.2±2.4	0.3±0.1	1.7±1.2
Total	74.1±6.8	22.4±9.6	60.4±6.5	15.5±3.4

[a] Average of three animals
Data taken from Table 8, p. 29, MRID 149464.

Biliary excretion data for rats following intraduodenal or gavage administration are shown in Table 6. Over a period of 7.5-10 hours 67.0 and 62.5%, respectively, of the intraduodenal and gavage dose was eliminated in the bile. Although biliary elimination was marginally slower following gavage administration than for intraduodenal administration, overall biliary elimination was comparable.

Table 6. Time-course of biliary excretion of radioactivity (% of radioactivity administered) by male Wistar rats following intraduodenal or gavage administration of [^{3}H]-GP 41 353.		
Time (hrs)	5 mg/kg intraduodenal	5 mg/kg gavage
0-1	16.1±5.2	9.1±1.0
0-2	36.3±3.3	17.5±2.4
0-4	54.8±6.0	34.4±4.4
0-6	62.3±4.5	48.2±5.6
0-7.5	67.0±3.5	-
0-10	-	62.5±5.0

Data taken from Table 7, p. 28, MRID 149464.

4. Tissue distribution

Tissue distribution data from two strains of rats using test material with different label positions (^{3}H and ^{14}C) revealed that administered radioactivity was consistently associated with organs/tissues having excretory function or those that would be expected to contain residual radioactivity following a specific dose route (e.g., residual radioactivity in the gastrointestinal tract following oral administration, liver, and/or kidneys). For both labels, tissue concentrations were highest within 2 hours of administration. Tissue distribution data revealed radioactivity levels in fat tissue to be equivalent to those in organs/tissue associated with excretory function but the decline in radioactivity over several hours was inconsistent with bioaccumulation. In mice given a single intravenous dose, radioactivity was widely distributed and exhibited initially (1-5 minute) high concentrations in adrenal glands, heart, liver, gall bladder, thyroid, and kidneys that declined up to 10-fold or more within two hours (with the exception of gall bladder radioactivity which increased consistent with biliary elimination).

B. PHARMACOKINETIC STUDIES

Time-course data for plasma radioactivity in rats and dogs revealed that maximum blood concentrations were attained within 30 minutes for both dogs and rats following intravenous administration and at approximately 30 minutes for rats and two to four hours for dogs following oral administration (Table 7). Although a notable decline in blood radioactivity was observed over 24 hours, no elimination half times were available.

Table 7. Time-course of blood radioactivity in rats and dogs following oral or intravenous administration of [^{3}H]-GP 41 353 (rats) or [^{14}C]- GP 41 353 (dogs)[a]

Time	5 mg/kg oral		5 mg/kg i.v.	
	Rats	Dogs[b]	Rats	Dogs[b]
10 min	-	-/-	5.4	4.23/5.69
30 min	4.0	0.15/0	6.0	5.87/9.34
1 hr	3.1	2.05/0.14	5.6	5.65/9.05
2 hrs	2.3	4.18/0.27	4.9	5.31/9.35
4 hrs	2.0	3.11/4.39	4.4	4.28/6.27
8 hrs	3.2	2.42/3.65	3.3	3.75/5.36
24 hrs	1.3	1.33/1.80	0.7	2.44/2.82
48 hrs	-	0.74/0.80	-	1.35/1.44

[a] Expressed as total radioactivity (μg/mL) that includes parent compound and any possible metabolites.
[b] Data are presented for each of two dogs
Data taken from Tables 3 and 10, pp. 24 and 31, MRID 149464.

C. METABOLITE CHARACTERIZATION STUDIES

Metabolite characterization of bile was conducted for rats (data were unavailable for review/analysis). The study report indicated that approximately 50% of the ether extractable dose was unchanged parent compound. It was reported that TLC analysis revealed two or three polar metabolites in the ether extract from the bile samples. Treatment of samples with β-glucuronidase resulted in parent compound, thereby indicating a glucuronide conjugate was present. Approximately 20-30% of the biliary radioactivity was attributed to this conjugate.

D. HISTOPATHOLOGY

Histopathologic assessment was not a component of the reviewed study.

III. DISCUSSION

A. DISCUSSION

In a metabolism study (MRID 149464), rats, mice, rabbits, and dogs were administered [^{14}C]-GP 41 353 or [^{3}H]-GP 41 353 (radiochemical purity 99%; chemical purity 99.5%; batch/lot nos. not provided) intravenously, intraduodenally, orally (gavage) at doses of 10 mg/kg (mice), 0.4 mg/kg (rats), 5 mg/kg (rats, rabbits, dogs), or 50 mg/kg (rabbits). Radioactivity levels in the blood, tissues, and excreta were measured for time intervals up

to 168 hours. Additionally, biliary elimination was also analyzed in the rats given a single intraduodenal dose.

There was no indication of any toxic effects in the test animals. Overall recovery of administered radioactivity in rats ranged from 99.67% to 104.53% at 2 to 8 hours after a single oral dose of 5 mg/kg [^{3}H]-GP 41 353. Data were unavailable in the study report to accurately determine radioactivity inventory for the other species tested.

Absorption of the test material was reported by the study author as 70-80% (for rats) as determined by comparisons of areas under-the-blood concentration curve for oral and intravenous administrations. These data were, however, unavailable for the preparation of this data evaluation report. Absorption in rats could also be estimated based upon biliary and urinary elimination data. Over a 7.5 to 10-hour period, biliary elimination accounted for 62.5% of a 5 mg/kg gavage dose and 67% of a 5 mg/kg intraduodenal dose while urinary excretion at 6 and 8 hours accounted for 76.60% and 5.17%, respectively, of a 5 mg/kg gavage dose. These data suggest that absorption is in excess of 70%. Absence of biliary elimination data for the other test species precluded assessment of the extent of absorption in those species; however, biliary excretion data in rats and mice suggest enterohepatic recirculation of the test material in both species. Time-course concentration data revealed that peak blood levels occurred within 30 minutes in rats following a single oral or intravenous dose of 5 mg/kg and at 2-4 hours for dogs.

Tissue distribution patterns were similar among mice and rats, and exhibited only slight quantitative variability for intravenous versus oral dosing. At two hours after an oral dose to rats, radioactivity in tissues was low (generally <1 μg/g tissue) with the exception of blood and organs associated with excretory function (e.g., liver, gall bladder, kidneys) where levels were as high as 59 μg/g tissue. Over the next 8-24 hours, these levels declined to <1 μg/g tissue. Following an intravenous dose to rats, tissue levels were also greatest in highly perfused organs or those associated with excretory function. Similarly, at two hours after a single intravenous dose to mice, tissue radioactivity was generally <10 μg/g tissue and greatest in highly perfused organs. Based upon data from rats, tissue burdens declined appreciably over 24 hours with no indication of accumulation or sequestration.

Excretion of GP 41 353 was examined in two strains of rats, rabbits, and dogs. Biliary elimination was also assessed in rats. Urinary excretion appeared to be a minor route of elimination in rats and dogs, accounting for 3-17% of the administered oral dose (0.38-5 mg/kg) to rats over a 168-hour period, and 8.3-8.8% by dogs over a 120-hour period. Urinary elimination by dogs was somewhat greater following intravenous administration; 12.9-17.7% over 120 hours. For rabbits, urinary excretion was a significant route of elimination and accounted for 74.1% of a single 50 mg/kg oral dose and 60.4% of a single 5 mg/kg oral dose over a 72-hour period. The biliary elimination data for rats showed that most of the fecal radioactivity could be attributed to biliary elimination products rather than unabsorbed test material. Biliary elimination was also affirmed by data from the mouse showing very high concentrations of radioactivity in the gall bladder at 5 minutes to two hours following a 10 mg/kg, i.v. dose.

Analysis of bile samples from the rats indicated that the test material underwent Phase II biotransformation. Treatment of samples with β-glucuronic acid revealed that most of the biliary product was glucuronide conjugates while some was unchanged parent compound.

The results of this multi-species study indicated that at least 70% of an oral dose of GP 41 353 is absorbed from the gastrointestinal tract and that biliary excretion and subsequent fecal elimination is a major excretory route in the rat and dog. Urinary excretion appeared to be a major route of elimination in the rabbit. Tissue accumulation was minimal after a single dose and was primarily associated with highly perfused tissues and organs with excretory function, although a companion study (MRID 68161) shows distribution of triclosan-derived radioactivity into the pituitary gland, optic nerve, and sciatic nerve at concentrations equivalent to those found in highly perfused organs at the same dose level (5 mg/kg). Repeated oral dosing at 5 mg/kg leads to accumulation of radioactivity in the blood, plasma, pituitary gland, sciatic nerve, and kidneys (MRID 68161). Metabolite data in rats revealed glucuronide conjugates and unchanged parent compound as biliary metabolites.

This metabolism study is **unacceptable/upgradable** and does not currently satisfy the requirements for a Metabolism and Pharmacokinetics study [OPPTS 870.7485 (85-1)]. Numerous deficiencies were observed in this report, including information regarding dose confirmation, homogeneity, and stability, environmental and housing conditions of the animals used in this study, and a lack of statements regarding compliance with Good Laboratory Practice and Quality Assurance. If the missing information can be provided, this study can be upgraded. Otherwise, a new guideline metabolism study will need to be conducted. Despite the deficiencies, the data do provide useful information on the disposition of Triclosan in multiple species and can be considered in conjunction with an acceptable metabolism study if such study is necessary.

B. STUDY DEFICIENCIES

Numerous deficiencies were observed in this study as outlined above. If these deficiencies cannot be addressed, a new study must be performed according to the 870. 7485 guideline.