Rec'd 10-14-94

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

OCT 14 1994

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: Chlorothalonil Reregistration: List A Case No. 0097: Chemical No. 081901: ISK Biotech Submission Concerning Meat and Milk Issues: CBRS No 13275: DP Barcode D199685.

FROM: William O. Smith, Ph.D., Chemist
Chemistry Pilot Review Team
Chemistry Branch II-Reregistration Support (CBRS)
Health Effects Division (7509C)

THROUGH: Edward Zager, Chief
Chemistry Branch II-Reregistration Support
Health Effects Division

TO: Walter Waldrop/Andrew Ertman - PM 71
Reregistration Branch
Special Review & Reregistration Division (7508W)

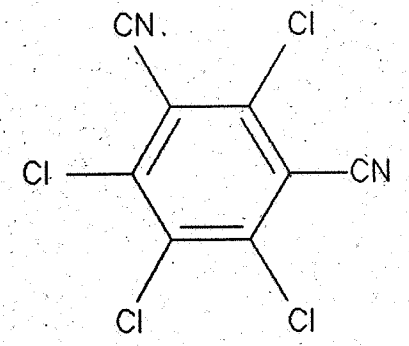
The registrant is proposing to modify a livestock feeding study, analyzing only for the 4-OH metabolite of chlorothalonil (SDS-3701) in meat and milk commodities. They also propose to use the results of this feeding study to set tolerances on meat and milk for residues of SDS-3701 only, excluding chlorothalonil from the tolerance expression.

BACKGROUND

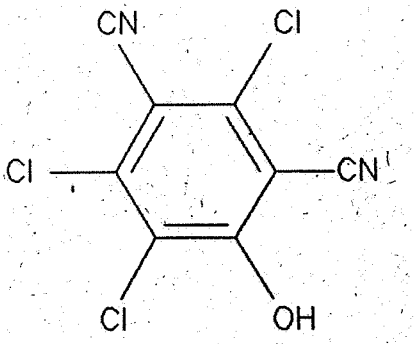
Tolerances have been established for the combined residue of chlorothalonil (2,4,5,6-tetrachloroisophthalonitrile) and its metabolite (4-hydroxy-2,5,6-trichloro-isophthalonitrile) in or on numerous raw agricultural commodities ranging from 0.05 ppm to 15 ppm (see Fig 1 for structures). No tolerances have been established for meat, milk, poultry, or eggs.

In response to the Chlorothalonil Registration Standard the registrant has agreed to conduct a ruminant feeding study to determine appropriate tolerance levels for livestock commodities. A protocol has been submitted to the Agency (see review by D. McNeilly, 7/11/91, D162243). It was agreed that the registrant would dose the animals with both chlorothalonil and its 4-OH metabolite (SDS-3701).

Fig. 1. Chlorothalonil Residues Regulated in Plant Commodities.



SDS-2787
CHLOROTHALONIL



SDS-3701

Information from a several sources indicates that chlorothalonil itself is very rapidly metabolized once ingested by livestock while SDS-3701 is not; therefore, the major component of residues in meat and milk is proposed to be SDS-3701. The current submission contains a summary of data presented by the registrant as justification for exclusion of chlorothalonil from meat and milk tolerances.

CBRS CONCLUSIONS

1. We accept the registrant's argument that detectable residues of chlorothalonil are not likely to transfer from the diet of livestock to meat or milk commodities.

2. The requirement for analysis of meat and milk commodities for chlorothalonil in the ruminant feeding study will be waived. These commodities need only be analyzed for SDS-3701.

3. In conformance with Branch policy, chlorothalonil must remain in the tolerance expression and enforcement methodology for both chlorothalonil and SDS-3701 must be validated for animal commodities.

4. For purposes of assessing dietary risk from the transfer of chlorothalonil residues to animal commodities, the Branch uses anticipated residues of the parent and its metabolite separately. In so doing we take into consideration the dietary risks that are inherent to each component of residue.

DETAILED CONSIDERATIONS

The registrant has submitted information (ISK Biotech Document # RC-94-RPB-001-001; No MRID # assigned) that was originally presented by representatives of ISK Biotech Corporation and Ricerca, Inc. at a January 6, 1994 meeting with representatives of SRRD and CBRS.

The basic premise forwarded by the registrant is that there is no reasonable expectation that finite residues of parent chlorothalonil will be present in meat or milk; therefore, tolerances on these commodities should exclude the parent and only cover the 4-hydroxy metabolite, SDS-3701. In support of this premise, discussions were presented concerning goat metabolism studies, in vitro incubations of chlorothalonil with bovine tissues and preliminary frozen storage stability studies. These presentations are summarized below.

Goat Metabolism Studies

The registrant summarized recently accepted (P. Deschamp, D174779, 11/4/93) goat metabolism studies, which included separate studies with ^{14}C-chlorothalonil and ^{14}C-SDS-3701. When ^{14}C-chlorothalonil was fed at the exaggerated level of 30 ppm in the diet, no chlorothalonil was detected in milk or tissue samples. The radioactive residues present were polar metabolites or non-extractable residues. When ^{14}C-SDS-3701 was fed at 2 ppm, considerably higher levels of radioactivity were found in milk and meat and SDS-3701 comprised 90-100% of the total radioactive residue. These data, among others, support the registrant's conclusion that chlorothalonil is extensively metabolized, probably to multiple glutathione conjugates, in animal tissues while SDS-3701 is transferred to tissues intact.

Reactivity of Chlorothalonil to Bovine Tissues in vitro

The registrant submitted results from experiments conducted to determine the reactivity of chlorothalonil with glutathione and with components of bovine tissues. Glutathione or tissue homogenates were incubated with ^{14}C-chlorothalonil in pH 7 phosphate buffer at 37 C. The reaction was stopped by addition of perchloric acid and the mixtures extracted with ethyl acetate followed by centrifugation. The radioactivity in the ethyl acetate phase was assumed to be chlorothalonil. Residues in the supernatant fraction, i.e., water soluble radioactivity, were analyzed by HPLC and it was suggested that these residues consisted mostly of diglutathionyl adducts of chlorothalonil. Radioactivity in the pellet fraction was assumed to be bound to proteinaceous material, presumably by reaction with cysteinyl residues in proteins. This was supported by the fact that radioactivity could be solubilized in sodium dodecyl sulfate and was found to migrate on a Sephadex G-100 column in a manner

suggesting a high molecular weight (>2000).

The reactivity of chlorothalonil in the different test systems is illustrated in the following table taken from the registrant's presentation.

Table 1. The stability of chlorothalonil in vitro in bovine tissues.

TISSUE/TEST SYSTEM	HALF LIFE (minutes)	NATURE OF REACTION PRODUCTS
Control (5 mM Glutathione)	10-15	Water soluble
Meat	0.5	Mostly water soluble
Rumen Fluid	360	Mostly in bound residue
Serum	3-5	Mostly in bound residue
Plasma	2-4	Mostly in bound residue
Fat	30	Mostly water soluble
Kidney (cortex)	0.5	
Kidney (medulla)	1	
Liver	1	Mostly water soluble
Mammary Tissue (lactating)	30	Mostly in bound residue
Mammary Tissue (nonlactating)	15	Mostly water soluble
Epithelium (intestinal)	3	
Epithelium (rumen)	9	Mostly water soluble
Epithelium (tripe)	3	Mostly in bound residue

Stability of Chlorothalonil in Tissues Under Frozen Storage

Preliminary storage stability studies were conducted for chlorothalonil and SDS-3701 in milk and animal tissues. Selected bovine tissues and milk were ground or blended, fortified with chlorothalonil or SDS-3701, stored frozen for various time intervals and then assayed. The SDS-3701 metabolite was stable in all tests while the loss of chlorothalonil was very rapid in all tissues except fat (Table 2).

Table 2. Recovery of Chlorothalonil From Bovine Tissues Stored Frozen at -18 to -23 C.

Days Stored	% Recovery of Chlorothalonil After Frozen Storage				
	Milk	Muscle	Liver	Kidney	Fat
1	43	8	2	4	96
ca. 7	2	7	2	2	81
ca. 14	1	5	1	2	73

The registrant's conclusions, based on the three lines of evidence above, are as follow:

- No detectable residues of ^{14}C-chlorothalonil were found in the goat metabolism study. SDS-3701 was the major residue in ^{14}C goat studies.

- The 4-hydroxy metabolite, SDS-3701, is essentially not metabolized by livestock.

- Chlorothalonil is not stable in milk and tissues in frozen storage but SDS-3701 is stable.

- Chlorothalonil reacts very rapidly with blood and animal tissues. Chlorothalonil in the livestock diet will not remain intact through transmittal to meat, milk or edible tissues.

- Taken together, these data confirm that there is no reasonable expectation of finite residues of chlorothalonil in milk, meat or other food products derived from livestock.

- There is no justification for including chlorothalonil analyses in meat and milk feeding study.

- SDS-3701 is the only suitable analyte to monitor food products derived from animals and should be the basis for an enforcement method.

CBRS COMMENTS

We accept the registrant's argument that detectable residues of chlorothalonil are not likely to transfer from the diet of livestock to meat or milk commodities. Therefore, the requirement for analysis of chlorothalonil in meat and milk commodities from the ruminant feeding study will be waived. These commodities need only be analyzed for SDS-3701. However, in conformance with Branch policy, chlorothalonil must remain in the tolerance expression and enforcement methodology for chlorothalonil on animal commodities must be validated. Based on the data submitted by the registrant, this should not be a problem because these data include analyses of meat and milk tissues for chlorothalonil to levels as low as 0.01 ppm.

One of the registrant's arguments for exclusion of chlorothalonil from the tolerance is that chlorothalonil is classified as a B2 carcinogen and SDS-3701 is not carcinogenic; therefore, inclusion of the parent in the tolerance expression leads to an unjustified implication of a cancer dietary risk from animal commodities. We note that the purpose for inclusion of the parent in the tolerance is primarily for enforcement purposes. In assessing dietary exposure the Branch uses anticipated residues of the parent and its metabolite separately. In so doing we take into consideration the dietary risks that are inherent to each component of residue.

cc: W. Smith, Chlorothalonil Reg. Std. File, SF, RF, circulation.

7509C:CB-II:WOS:wos:Rm805A:CM#2:X5353:10/12/94

RDI: P. Deschamp 10/12/94, T. Edwards 10/12/94, S. Knizner 10/12/94. C. Olinger 10/12/94, M. Metzger 10/13/94, E.Zager 10/13/94.