



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

OFFICE OF PESTICIDES AND TOXIC SUBSTANCES

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DEC 5 1980

Memorandum

SUBJECT: PP#OF2353. Triforine in or on almonds and apples.  
Evaluation of analytical methods and residue data.

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EM Laboratories, Inc., proposes the establishment of tolerances for residues of the fungicide triforine, N,N'-[1,4-piperazinediyl-bis-(2,2,2-trichloroethylidone)]-bis-[formamide] in or on the following raw agricultural commodities:

Almonds - 0.01 ppm  
Apples - 0.01 ppm

At present, permanent tolerances at 0.1 ppm have been established on blueberries, cranberries and peaches (40 CFR 180.382).

There are two triforine petitions (Nos. OF2351 and OF2352) co-pending, and there are also under review two associated inert petitions (Nos. OF2384 and OF2386).

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Conclusions

1) Clearance for three inerts. [REDACTED]

[REDACTED] - see TOX reviews of PP Nos. OF2351, OF2352 and OF2353 - 9/9/80 - C.C. Frick) is needed before there should be favorable recommendations for the proposed tolerances.

2a) From the proposed use, the parent compound per se is the residue of concern on apples and almonds.

2b) The nature of the residue in animals is not adequately understood. We restate our previous conclusion reached in PP#9E2184 (6/11/79 - M. Nelson) that if feed items are involved (Apple pomace and almond hulls are feed items.) a large animal (lactating ruminant) and if applicable, poultry ---radiotracer metabolism study in which the nature and extent of triforine metabolites in tissues, milk/eggs, and excreta are clearly identified, and feeding studies, will be needed.

3a) The submitted methodology is suitable for triforine residues in plant commodities.

3b.) If the large animal metabolism studies should show other residues of concern which need to be regulated, then additional methodology will be needed.

4a) None of the submitted residue data reflect the proposed use parameters in Section B. The petitioner needs to rerun residue studies in the states of Washington, Michigan and New York. These studies should reflect the proposed use. If any of the residue data shows detectble residues in apples, then a new fractionation study may be necessary. However, this time, the petitioner should report data on fresh apples, apple sauce, wet apple pomace and dry apple pomace originating from the same apple composite. If residues in any of the preceding fractions exceed those in apples, then food additive tolerances will need to be proposed.

4b) The petitioner has proposed that triforine may be used on apples as a tank mix with Glyodin. We will need some residue data on triforine that reflects the proposed use.

4c) We notice that the label differs from Section B in that the label does not prescribe the number of applications or a PHI. We ask the petitioner to correct this difference.

5a) The geographical representation for almonds is satisfactory.

5b) Although the data bank is very limited, it is possible to conclude that triforine residues on almond meat should not exceed the 0.01 ppm tolerance; the studies were allowed to run the full course of the recommended minimal PHI. However, we can not recommend establishment of the proposed tolerance until the inert clearances have been resolved.

5c) We are requesting the petitioner to submit a revised Section F in which a tolerance proposal of 0.1 ppm triforine on almond hulls has been included.

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6) Apple pomace and almond hulls are feed items. Thus, we are requesting that a large animal (lactating ruminant) feeding study be carried out. This conclusion is consistent with the recommendations proposed in our previous review of PP#9E2184 (6/11/79 - M. Nelson), i.e., if feed items are involved, a large animal (lactating ruminant) - and, if applicable, poultry-radiotracer metabolism study in which the nature and extent of triforine metabolites in tissues, milk/eggs, and excreta are clearly identified, and feeding studies, will be needed.

#### Recommendations

We recommend against the establishment of the proposed tolerances for the reasons cited in conclusions 1, 2b, 3b, 4a, 4b, 4c 5b, 5c and 6.

#### Detailed Considerations

##### Manufacture and Formulation

We will not discuss further the manufacturing process for technical triforine since it was discussed favorably in our review of PP#7F1921 (8/18/77 - M. Nelson).

The technical product is formulated for use as Funginex Emulsifiable Concentrate (EPA Reg. No. 21137-4) and as Funginex Wettage Powder (no established EPA number).

Funginex Emulsifiable Concentrate (formulation) contains 1.6 lbs. of triforine as a.i./gal. This formulation has been discussed previously in PP#9E2184 (see our review of 6/11/79 - M. Nelson). There are 3 inerts in this formulation that have not been cleared for the proposed use (see TOX Review of 9/9/80 - C. Frick). Clearance for these inerts is needed before favorable recommendations for the proposed tolerances can be made.

##### Proposed use

To control scab, powdery mildew and rust, on apples, apply 10 fl. oz. of Funginex per 100 gallons of water or 40 fl. oz. per acre dilute or apply as a tank mix of 6 fl. oz of Funginex plus 1 pint of Glyodin per 100 gallons of water. First application is made at 1/2 inch green tip and can be repeated every 7 days up to a maximal of 5 applications. Do not apply Funginex after petal fall. PHIs should be >120 days.

To control brownrot blossom blight on almonds, apply 12 fl. oz. of Funginex per 100 gallons of water or 36 fl. oz. per acre dilute. First application is made at pink bud and repeat at 50-100% bloom.

Do not graze animals in treated apple and almond orchards.

We notice that the label differs from Section B in that the label does not prescribe the number of applications or a PHI. We ask the petitioner to correct this difference.

### Nature of the Residue

Plants. To support this petition, EM Laboratories, Inc., has resubmitted the plant (apples, barley) radioactive metabolism studies which were originally submitted in PP#7F1921, and they were subsequently discussed in our (M. Nelson) reviews of 8/8/77 and 6/12/78.

The compiled plant metabolism data suggest that triforine is degraded by hydrolysis to N-(1-formamido-2,2,2-trichloroethyl) piperazine, then to piperazine and then to oxalic acid, a naturally occurring compound in plants. Also, triforine intermediates containing the formyl or glyoxyl group are incorporated into natural constituents of plants. No appreciable accumulation of any metabolite was reported. Current plant metabolism data support our conclusion that the parent compound per se is the residue of concern from the proposed use. However, we do not foreclose the possibility that more plant metabolism data will be needed to support any future petition for other crops/usages.

Animals. Two studies on the metabolism of radioactive triforine in rats were submitted in PP#7F1921; we (M. Nelson - 8/8/77) have already reviewed these studies. In brief, greater than 90% of the administered radioactivity was found in the urine: feces at a 4:1 ratio. Mostly unchanged triforine was excreted in rat feces; no metabolites were characterized. However, a metabolite, N-[1-formamido-2,2,2-trichloroethyl]piperazine, found in the rat urine was characterized. No appreciable quantities of radioactivity were found in any of the rat tissues. No livestock metabolism studies are submitted.

Conclusion. The nature of the residue in animals is not adequately understood. We restate our previous conclusion reached in PP#9E2184 (6/11/79 - M. Nelson) that if feed items are involved (Apple pomace and almond hulls are feed items.), a large animal (lactating ruminant)---radiotracer metabolism study in which the nature and extent of triforine metabolites in tissue, milk/eggs, and excreta are clearly identified, and feeding studies, will be needed.

Analytical Methods. Two analytical methods are submitted with this petition: (1) "Method for residue analysis of Cela W 524[triforine] in apples and cucumbers" 7/26/71, and (2) "Determination of triforine residues" 12/7/76. RCB has previously reviewed both procedures (see our review of PP#7F1921, 8/8/77, Dr. M. Nelson). The petitioner has stated that the former method (7/26/71) was used to analyze samples before 1977; the latter method (12/7/76) was used to analyze samples in 1977 and thereafter. Our Method Evaluation Unit, CBIB, BFGD, has successfully conducted a method trial on the 12/7/76 method. Peaches were fortified with triforine at 5 and 10 ppm; recoveries ranged from 85-94%. Blueberries were fortified at 0.1 and 0.2 ppm; recoveries ranged from 63-73%.

The two methods differ procedurally in the choice of toluene or ethylacetate as a petition solvent. The residue is extracted from macerated plant samples with acetone, filtered, and precleaned by partitionings with first a H<sub>2</sub>O-saturated NaCl solution and then ethyl acetate (1971 procedure)

or toluene (1976 procedure). The solvent is evaporated/distilled off and the residue remaining is mixed with dilute  $H_2SO_4$  which, upon heating results in formation of chloral hydrate (by cleavage) which is distilled into  $H_2O$ , partitioned into ethyl formate, and determined by EC-GLC. Residues are reported as ppm triforine (with the aid of a 1.31 conversion factor and an internal standard). The limit of detection appears to be 0.01 ppm judging from the chromatograms.

Validation data for the parent compound triforine are summarized below:

Commodities	Fortification levels, ppm	Recoveries, %	Controls, ppm
apples	0.1-1.0	75-118	<0.01
apple juice	not given	85	0
apple sauce	not given	90	0
apple pomace	not given	100	0
almond hulls	0.5	100	<0.01
almond meat	0.5	91	<0.01
almond shells	0.5	86	<0.01

The submitted methodology is suitable for determining and enforcing a tolerance for triforine residues in plant commodities. However, if the large animal metabolism study (see the Nature of the Residue section of this review) should show other residues of concern which need to be regulated, then additional methodology will be needed.

#### Residues Data

Apples. The residue data on apples grown in 6 states (R.I., N.Y., N.C., Mo., Va., and Mich.) and Canada were submitted. Samples from apple trees receiving 1-11 applications [6-64 fl.oz. of formulation CME-74770 (20EC), CA 73021 or 70203 (IX=40 fl.oz./A) at 0-day PHI contained 0.02-0.63 ppm triforine residues. Samples receiving 1-14 applications [6-64 fl.oz. of formulation CME-74770 (20EC), CA 73021 or CA 70203] at PHIs of 1-56 days contained ND (none detected amounts)-0.71 ppm triforine residues. Only two samples with a PHI of 56 days (the maximum PHI observed) were reported; both contained 0.06 ppm triforine.

Residue data were also collected on some apple fractions; the apple trees received 9 applications of 32-64 fl. oz. of formulation CA 70203/A. These data are summarized below:

<u>Fraction</u>	<u>PHI</u>	<u>Triforine residues, ppm</u>
apples	no value	no values reported
apple juice	51 days	0.01-0.04
apple sauce	51 days	0.01-0.02
apple pomace	51 days	0.14-0.37

### Conclusions

None of the submitted residue data reflect the proposed use parameters in Section B, i.e., a use pattern containing 5 blossom applications of 8 oz. a.i./A and PHIs greater than 120 days. The petitioner did not allow any of his studies to go beyond 56 days at which time triforine residues (only two values reported) were found at a level of 0.06 ppm. From the submitted data, no assumption can be made that fresh apples would contain less than 0.01 ppm triforine residue at a PHI of 120 days. At a 1 day PHI in the Virginia study, the residue values ranged from a none detectable amount to 0.71 ppm. In brief, the residue patterns are not clear enough to make extended residue predictions at any set PHI beyond 59 days.

In view of the above, we are requesting that the petitioner submit additional residue data on apples grown in the states of Washington, Michigan and New York. The additional data should reflect the proposed use on the label. If detectable residues are found in apples, then a new fractionation study may be necessary. However, this time, the petitioner should divide the apple sample chosen for the fractionation study into subsamples. At least one subsample of fresh apples should be analyzed for triforine residues (no values were reported in the above study). The remaining subsamples should be used for obtaining triforine residues on apple juice, apple sauce, wet apple pomace and dry apple pomace. If there are triforine residues in any of the preceding fractions that exceed the level in the apples, then food additive tolerances need to be proposed.

Finally, we notice that triforine is beng proposed for use as a tank mix with Glyodin. We will also need some residue data on triforine reflecting this use.

Almonds. All of the residue data submitted were obtained on almonds grown in the state of California. Within the United States, almost all of our almonds are produced in California; therefore, we consider the geographical representation to be adequate.

The data submitted reflected only 3 triforine analyses on each, almond shells, hulls and meat. Triforine formulated as CME 74770 (20EC), CME-10225 (50WP) or CME-10224 (80F) was applied to almond trees at rates of 0.15-0.25 lb. a.i./100 gal. Triforine residues on the almond shells, hulls and meat ranged from 0.005 to 0.008 ppm, 0.019 to 0.087 ppm and 0.001 to 0.007 ppm, respectively. Although the data bank is very limited, it is possible to conclude that triforine residues on almonds should not exceed the proposed 0.01 ppm tolerance since the studies were allowed to run (203 days) slightly beyond the minimal proposed PHI of 200 days. In the case of apples above, the minimal proposed PHI was never reached. In view of the above data, however, we must request from the petitioner a revised Section F in which there should be a tolerance proposal of 0.1 ppm triforine on almond hulls. Almond hulls are used as a feed item.

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Meat, Milk, Poultry and Eggs Residues

The possible feed items involved in this petition would be apple pomace and almond hulls. Depending on the animal, the apple pomace dietary intakes for cattle, swine, horses and lambs may vary from 25 to 50%; for poultry, dietary intakes may vary from no use to 5%. With regards to almond hulls, the dietary intakes for the above large animals may vary from 10 to 50%; almond hulls are usually not used as a poultry feed item.

The petitioner has reported that apple pomace from apple harvested at a PH1 of 51 days had 0.14-0.37 ppm triforine residues; the raw data sheets indicated that these results reflect only wet pomace. He has also reported that almond hulls contained between 0.019 to 0.087 ppm (rounded off to 0.1 ppm) triforine residues. In consequence, we are requesting that a large animal (lactating ruminant) feeding study be carried out.

This conclusion is consistent with the recommendations in our previous review of PP# 9E2184(6/11/79-M. Nelson), i.e., if feed items are involved, a large animal (lactating ruminant)--and, if applicable, poultry---radiotracer metabolism study in which the nature and extent of triforine metabolites in tissues, milk/eggs, and excreta are clearly identified, and feeding studies, will be needed.

Therefore, without a large animal (lactating ruminant) feeding study, we are unable to predict if problems with secondary residues will arise in meat and milk. However, we are able to conclude that at this time there should not be any problems with secondary residues in poultry and eggs as a result of the proposed use.

INTERNATIONAL RESIDUE LIMIT STATUS

CHEMICAL Triforine

PETITION NO. 0F2353

CCPR NO. 116

REVIEWER: J. Onley

Codex Status

Proposed U.S. Tolerances

☐ No Codex Proposal  
Step 6 or above

Residue (if Step 9): Determined as  
chloral hydrate & expressed as triforine

Residue: Triforine

Crop(s)	Limit ( mg/kg)
apples	2 mg/kg Step 5

Crop(s)	Tol. (ppm)
Apples	0.01
Almonds	0.01

CANADIAN LIMIT

Residue: triforine

MEXICAN TOLERANCIA

Residue: None

Crop      Limit ppm

Crop      Tolerancia (ppm)

None on these commodities

Notes: