FLUSILAZOLE (165)

EXPLANATION

Flusilazole was previously reviewed for residues by the 1989, 1990 and 1991 Meetings. An outstanding requirement from the 1991 evaluation was "Information on GAP and additional supervised trials data (including data on major metabolites) for nectarines and peaches reflecting that GAP from additional countries." The current MRLs are temporary pending the availability of this information.

Desirable information comprised:

1. Additional information on GAP for flusilazole on grapes in Europe.

2. Details of the wheat grain freezer storage stability study (Guinivan, 1987) and information on the relevance thereof to the storage of samples in the supervised trials.

3. Information on the stability of flusilazole metabolites (especially the silanol IN F7321) in cereal grains and/or plant parts under freezer storage conditions.

4. Submission of the hen metabolism study by Smyser (1990) which had been cited but not provided.

5. Information on residues of the major flusilazole metabolites (especially the silanol) in processed fractions of cereal grains from field-treated cereals containing measurable residues.

6. Submission of final reports of soil studies of which the interim reports were reviewed by the 1989 JMPR.

The Meeting received and reviewed information on all these items. Additional residue data were also submitted on pome fruit, grapes, barley, rye and wheat, although there were no outstanding requirements for trials on these commodities (they all have CXLs). Data on sugar cane were provided for the first time.

USE PATTERN

Information on current GAP in the use of flusilazole was provided for stone fruit, cereals, pome fruit, grapes and sugar cane, in some cases from countries whose previously submitted GAP has changed (Table 1). In cases where conflicting information was provided, that supported by labels or English translations of labels was given preference. The Meeting was informed that the product is not registered in The Netherlands owing to environmental persistence.

Table 1. Registered and approved uses of flusilazole on selected crops.

Crop Country	Application	PHI, days	Notes

	Form.	g ai/hl (g ai/ha)	No.	Int., days		
Cereals						
Belgium barley* rye & winter wheat	SC[1]	(160-200) (160)	 >1 >1		42	* winter or spring
Germany wheat	EC[2]	40-80 (160)	2		49	
winter wheat, barley, rye	SC[3]	≤75 (300)	1		56	
Italy barley, wheat	20DF	(120-160)	-	-	-	proposed use
Grapes						
Australia	DF	2 or (10)** (15)*** (20)****	*	14-21	14	* as needed ** early season *** mid-season ****late season (pre-flowering to bunch closing)
France	EW	(30)	2-3	14	*	*unspecified
Spain	40EC	1.6-3.2* 3.2-4.8*** (16-48)	5**		14	* normal infestation ** 10 cm buds to ripening, 2 appl. after flowering *** heavy infestation
Pome fruits						
Spain	40EC	2.4-4.8 (3.6-7.2)	4	10-14	14	
Australia Apple	DF	2-3*	>1**	10-14*	14	* pest pressure-dependent ** until term. growth ceases
Pear	DF	2	2	10-14	14	
Stone fruits						
France peach, nectarine	EW	4	3-4	12-14*	12-14*	* label interval, whether between or after appls. not specified

Table 1 (contd.).

Crop Country	Application				PHI, days	Notes
	Form.	g ai/hl (g ai/ha)	No.	Int., days		
Stone fruits						
Italy plum, peach, apricot, cherry	20DF	4	2-3		10	proposed use
Spain peach, apricot	40EC	4[4]	>1*	-	7	* start at petal fall
Greece peach	40EC	3-4 (600-800)	-	*	*	* pending use, no PHI proposed (PHIs of other countries will be considered)
Sugar cane Australia	Liq. 1.6 gai/L	0.2	*	*	*	* Set treatment in spray or dip planter

[1]Mixed formulation, 80 g flusilazole and 200 g chlorothalonil/l
[2]Mixed formulation, 160 g flusilazole and 350 g tridemorph/l
[3]Mixed formulation, 250 g flusilazole and 125 g carbendazim/l
[4]From manufacturer's label. Spanish summary gives 2 applications at 2.4-4.8 g ai/hl (26-53 g ai/ha).

RESIDUES RESULTING FROM SUPERVISED TRIALS

Cereals. Results of additional supervised trials were provided although there were no outstanding requirements for them and although there are CXLs for barley, rye and wheat at 0.1 mg/kg and for their straws and fodders (dry) at 2 mg/kg. Results are summarized in Table 2. All flusilazole residues in the grain were below the CXL (maximum 0.05 mg/kg in wheat and spring barley) and in the straw were ≤1.7 mg/kg except one residue of 2.3 mg/kg in wheat straw. This residue was found 56 days after the last of 3 post-emergence broadcast applications of a mixed suspension concentrate formulation (the first at 200 g ai/ha and the other two at 160 g ai/ha) in 1986 UK trials (Du Pont, 1993). Both the application rate and PHI are consistent with current GAP in other European countries and with UK GAP summarized by the 1989 JMPR.

The Meeting noted from data submitted, but not included in Table 2, that residues in straw and particularly forage could significantly exceed the 2 mg/kg CXL for dry fodders and straws at shorter PHIs and generally higher application rates than those which are reported as European GAP.

Table 2. Residues of flusilazole and metabolites on cereals resulting from supervised trials.

Crop Country Year	Application			Residue, mg/kg				Ref.[4]
	Rate g ai/ha	No.	PHI (days)	Flusilazole	F-7321[1]	H-7169[2]	TA[3]	
Wheat Italy 1987	175	1	34	0.05	<0.01			1
Winter wheat grain Germany 1986-8, USA 1983-4, UK 1986 straw	70-300	1-3	26-140 42-138	0.02, ≤0.014 (91)[5] 2.3, 1.7, 1.4, ≤1.1 (73)	0.02, ≤0.01 (7) 9.9, ≤6.6 (16)	≤0.01 (8) ≤0.3 (16)	2.5, ≤0.6 (10)	2
Spring wheat grain USA 1983-4 straw	70-140	2	19	0.013, <0.01 0.34, 0.3				3
Spring barley grain USA 1983-4 UK 1986 straw USA UK	70-140 160 70-160 160	2 2	12 63-69 12 63-69	0.01, 0.05 0.05, 0.01 1.2, 1.1, 1, 0.5	 3.9, 3.4 (2), 3.1	 ≤ 0.12 (4)	≤0.13 (4)	4 5
Winter barley grain Germany 1986-8, USA 1983-4, UK 1986 straw	140-300	1-2	44-88	0.02, ≤0.01 (17) 1.1-1.6 (5) ≤0.7 (20)	 7.7, 5.9, ≤4.8 (12)		≤0.31 (12)	6
Winter rye grain Germany 1986-8 straw	160-300 300	1-2	82-83 120	<0.01 (3) <0.01				7

[1] F7321: bis(4-fluorophenyl)(methyl)silanol.
[2] H7169: [bis(4-fluorophenyl)(methyl)silyl]methanol.
[3] TA: 3-(1H-1,2,4-triazol-1-yl)alanine.
[4] Numbers refer to Du Pont, 1993, Table 2.
[5] Number in parentheses following a residue indicates the number of residues at that level.

Grapes. The 1991 JMPR considered additional information on European GAP desirable because it had been suggested that the 1989 estimate of 0.5 mg/kg was unnecessarily high. It was based on German trials but the GAP of other European countries, and was obviously rounded up since the 1989 Meeting concluded that residues would not be likely to exceed 0.3 mg/kg. There was an outstanding question of whether the 5-8 applications in the German trials was GAP in Europe (the use was not registered in Germany). However, there were no requirements for additional data and the 0.5 mg/kg limit is now a CXL.

Information provided to the Meeting included the updated GAP of Spain (essentially unchanged), and France (essentially unchanged, still no PHI specified) and new information on GAP in Australia (Table 1). It confirms the appropriateness of the 14-day PHI used by the 1989 JMPR. Spain (1993) also provided data from trials in the USA (4 studies, 14 trials) and summary data on 6 grape (and 2 processing) trials (apparently French, no details, formulation unspecified). Results of one Australian trial were available (Australia, 1993). Residues in grapes did not exceed 0.15 mg/kg in the French trials (8 applications of an unspecified formulation at 20 g ai/ha, PHI unspecified), 0.08 mg/kg in the US trials (no US GAP) or 0.22 mg/kg from treatments reflecting GAP in the Australian trial.

Pome fruit. The CXL is 0.2 mg/kg. Although there were no outstanding requirements, the Meeting was provided with current GAP for Spain, Australian trials data (Australia, 1993) and a summary from Spain of trials in Italy, Germany, Belgium and France (Spain, 1993). In the Australian trials maximum residues in apples were 0.2 mg/kg at the Australian 14-day PHI from a double application rate and 0.07 mg/kg at the recommended rate. No residues (<0.02 mg/kg) were detected in pears after 21 days at 1 to 1.5 times the GAP application rate of 2 g ai/hl and up to 0.03 mg/kg at a threefold rate. The highest residues reported in the Spanish submission were 0.16 mg/kg from GAP and exaggerated application rates.

Stone fruit. The 1991 JMPR recommended a temporary MRL of 0.1 mg/kg for peaches and nectarines, based on GAP in New Zealand and Spain and data from France and New Zealand, pending the availability of additional data on residues of flusilazole and its metabolites from treatments according to GAP. New or updated information on GAP for peaches and nectarines or stone fruit in France, Italy, Spain and Greece is summarized in Table 1. Two or more applications at 3-4 g ai/hl and PHIs of 7 to 14 days appear to be usual for these countries, although a PHI is not specified for France or Greece. These practices are comparable to those recorded by the 1991 JMPR for New Zealand and Spain and are in accord with the 7-day PHI on which the 1991 MRL recommendation was based.

Data from supervised trials in Italy, Australia, France, the USA, and Greece were provided (Table 3) although no information on GAP was available for the USA or Australia. Two of the three French trials on peaches which were reported were reviewed by the 1991 JMPR and are not included in the Table. In two additional 1986-87 French studies on peaches (not in Table 3) no residues (<0.05 mg/kg) of triazolylalanine were detected 1 to 26 days after as many as 9 treatments with an EC formulation at application rates up to 30 g ai/ha (Du Pont, 1993, Reports BG-88-01 and BG-88-07).

Table 3. Residues of flusilazole and metabolites in stone fruits resulting from supervised trials. Underlined residues are from treatments according to GAP.

Crop Country Year	Application			Residues, mg/kg at intervals after last application	Ref.
	Form	No.	Rate g ai/ha (g ai/hl)		
Apricots			Days	0 1347 10 106	
France 1989	CE	4	30 (3)	<0.01	1
1991	10EC	8	24 (4)	0.30.20.090.080.05	2
				Controls ≤0.01	
1991	10EC	4	40 (14.3)	0.030.040.040.050.03	2
				Controls <0.01	
1991	10EC	6	40 (14.3)	0.080.10.070.060.06	2
				Controls 0.01	
Cherries					

Crop Country Year	Application			Residues, mg/kg at intervals after last application	Ref.
	Form	No.	Rate g ai/ha (g ai/hl)		
Australia 1989	20DF	3[1]	(2, 3, 4, or 8)	Not detected (<0.05 mg/kg) at 43 days	3
Peaches					
Australia 1990	20 DF	3[1]	(2, 3, 4, or 8)	Not detected (<0.05 mg/kg) at 148 days	3
				Daysflusilazole IN-F7321[2]IN-H7169[3]	
France 1986	40EC	3-4	23 (3)	1 0.060.03<0.01	4
(nectarines)			30 (4)	1 0.040.01<0.01	
			23 (3)	17 0.070.01<0.01	
			30 (4)	17 0.050.01<0.01	
				Controls <0.01 <0.01<0.01	
		9	10.5 (3)	8 0.02<0.01<0.01	4
			14 (4)	8 0.550.07<0.01	
				Controls 0.1,<0.01<0.01	
				0.05	
Italy 1988	20DF	3[4]	60 (4)	<0.01 at 102 days	5
				Control <0.01	
Greece			Days	0 3 7 14 21	
1992	40EC	5	40 (4)	0.1, 0.10.2, 0.2 0.05	6
			80 (8)	0.3, 0.20.07, 0.05 0.04	
				Controls 0.01	
1992	40EC	6	40 (4)	0.3, 0.2 0.090.02 0.03	6
			80 (8)	0.2, 0.2 0.070.05 0.04	
				Control <0.01	
USA 1984				5 days12 days 5 applcns. 3 applcns.	
	EC	3 or 5	70 (1.2) 140 (2.4) 280 (4.8) 140 (2.4)	0.050.1 0.20.06 0.30.3 0.10.1	7
			Days	712 14 15 22 29	8
U.S.A. 1984	40EC	3	70 (1.2)	0.040.030.02<0.01 0.01 0.01	
			140 (1.2)	0.050.070.05 0.05 0.020.03	
			280 (4.8)	0.20.10.1 0.09 0.050.03	
				Controls <0.01	
			Days	0 4 6 7 14 21	8
1984	40EC	5	70 (1.2)	0.050.060.030.04 0.050.03	
			140 (2.4)	0.050.080.090.09 0.080.06	
			280 (4.8)	0.20.50.30.2 0.2-	
Plums					
Australia 1990	20DF	3[5]	(2 to 8)	No residues (<0.05 mg/kg) at 119 days	
Australia 1989	20DF	3[5]	(7.5 to 30)	No residues (<0.05 mg/kg) at 121 days	3

Crop Country Year	Application			Residues, mg/kg at intervals after last application	Ref.
	Form	No.	Rate g ai/ha (g ai/hl)		
1989	20DF	3[5]	(2, 3, or 8)	No residues (<0.05 mg/kg) at 117 days	3
			(4)	0.07 at 117 days	

[1] The last at petal fall
[2] F7321: bis(4-fluorophenyl)(methyl)silanol
[3] H7169: [bis(4-fluorophenyl)(methyl)silyl]methanol
[4] Applications at bloom, beginning of fruit set and end of fruit set

Sugar cane. No limit has been proposed for sugar cane. The Meeting was provided with information on Australian GAP and data from trials in Australia in which sugar cane was grown from cane sets dipped at concentrations of 1, 2 (GAP rate), 5 and 10 mg flusilazole/l (Australia, 1993). No residues (<0.02 mg/kg) of flusilazole were detected in cane juice extracted 11 months after the dip treatments. The stalks were not analyzed.

FATE OF RESIDUES

In animals

The 1991 JMPR requested the submission of the report of a hen metabolism study which was cited and referenced, but not provided, in a 1991 submission to FAO (Wustner, 1991). In response the manufacturer re-submitted all available reports of hen metabolism studies, including the one requested (Du Pont, 1993).

The reports:
1. AMR-245-84 (Bodden and Kneeland, 1984); phenyl label. Reviewed by the 1989 JMPR (omitted from 1989 monograph references).
2. Supplement to AMR-245-84 (Stadalius, 1984). Not previously provided. 3. AMR-638-86-1 (Lin, 1988a); phenyl label. Reviewed by 1989 JMPR.
4. Supplement to AMR-638-86-1 (Lin, 1988b). Not previously provided.
5. AMR-638-86-2 (Lin, 1988c); triazole label. Reviewed by the 1989 JMPR.
6. Supplement to ARM-638-86-2 (Lin, 1988d). Not previously provided.
7. AMR-638-86, Supplement 2 (Smyser, 1990). The requested report.

Report AMR-245-84, reviewed by the 1989 JMPR, left questions on the nature of the residues in poultry. The situation was clarified by AMR-638-1 and -2 which were also reviewed in 1989. The Supplement to AMR-245-84 is merely a response to a US EPA requirement for a hen metabolism study with the triazole label and cites AMR-638-86-2 as fulfilling this requirement. The supplements to AMR-638-86-1 and -2, not previously provided to the JMPR, respond to EPA requirements for more details of the raw data and the calculation of the distribution of residues in the original reports. This leaves only the requested report for consideration by the Meeting.

The report (Smyser, 1990) is a response to EPA requirements for chromatograms showing the co-chromatography of metabolites and analytical standards, and for the expression of metabolite concentrations as a percenage of the total residue in Reports AMR-638-86-1 and -2. Table 4 of the 1991 JMPR monograph, taken from AMR-638-86-1, showed the metabolites in liver, kidney, muscle and fat (but not eggs) as a percentage of the total residues found with the phenyl label. The resiudes from the triazole-labeld compound were not shown. They are given in Table 4 below, which is taken from AMR-638-86-2.

Tabe 4. HPLC quantification of flusilazole and its metabolites in extracts of tissues from laying hens administered [triazole-3-^{14}c]flusilazole for 14 days at 3 ppm in the diet (Smyser, 1990).

Component	Component as % of total radioactivity in						
	Liver	Kidney	Thigh muscle	Breast muscle	Fat	Eggs[1] 4-day	12-day
Flusilazole	5	5	-	1	68	1	2
Triazole	76	79	75	86	14	83	77
Thymine	8	7	11	6	3	5	9
Other	6	9	3	2	15[2]	11	2

[1] 4 and 12 days chosen as representative of the 2-, 4-, 6-, 8-, 10-, and 12-day samples which were analysed.
[2] Radioactivity associated with three HPLC peaks. No single component exceeded 2%.

Triazole is the predominant residue in all samples except fat where flusilazole is greater.

When chickens were fed for 14 days at a 3 ppm dietary level with phenyl-labelled flusilazole, residues in 12-day eggs (chosen to be representative) were as follows (Smyser, 1990):

	% of total radioactivity
IN-F7321 (methyl silanol)	32
IN-37738 (silyl phenol)	5
Phosphate conjugate of IN-37738 at the 3-hydroxy position	3
[(4-fluorophenyl)methyl]silanediol	38
Flusilazole	4
P5 (unidentified)	5
P11 (at least 3 unidentified metabolites	3
Lipophilics	7
Other	3

In plants

No new information.

In soils

The 1989 and 1991 Meetings requested submission of the final reports on two field soil dissipation studies reviewed by the 1989 JMPR at an interim stage (AMR-556-86, Stadalius, 1986; AMR-791-87, Fujinari, 1986b). The completed study AMR 556-86 (Smyser, 1993) was submitted, but the Meeting was informed that report AMR-791-87 had not yet been completed.

In the AMR-556-86 study [phenyl(U)-^{14}C]flusilazole was applied to Delaware silt loam soil (in 38 cm x 10 cm i.d. steel cylinders driven into the ground) at the nominal rate of 105 g ai/ha four times a year for 3 years. That rate would be comparable to current GAP for many crops and higher than that for others. The 1989 JMPR noted that the half-life of flusilazole was <12 months and that flusilazole and its silanol metabolite (IN-F7321) were the main identified residues. It also noted that the radioactive residues increased with the number of applications, with 90% of the total radioactivity in the top 0 to 8 cm of the soil and with maximum soil residues of flusilazole

of 0.22 mg/kg after 2 years. Only 62% of the applied radioactivity was recovered after 2 years. Losses were attributed to photolysis and microbial attack (1989 JMPR monograph).

The interim report summarized results for 697 days and the final report for the entire 1092 days of the completed study. The percentages of the radioactivity recovered in the three cylinders at different soil depths in each of the three years, with a cumulative rainfall 325 cm, was as follows (from Table V of the report):

Soil depth (cm)	year 1	year 2	year 3
Day->	0-368	326-697	694-1092
0-8	97.3-100	92.6-99	92.6-98.6
8-16[a]	ND-0.9	0.7-5.6	0.9-4.8
16-24[b]	ND-0.5	0.2-1.4	0.4-2.3
24-36[c]	ND	ND-0.3[d]	0.1-0.9

[a] peak residue on day 633
[b] peak residue on day 737 (12th and last application)
[c] Nominal 38 cm cylinders, but 2-3 cm left above ground
[d] Radioactivity first detected after 368 days

Calculations indicate a flusilazole average half-life of 251 days. The statistical evaluation of the data given in the report predicts that flusilazole residues would reach a steady state of approximately 57% of the yearly application under worse-case application conditions.

The residues of flusilazole and IN-F7321 (expressed as mg/kg flusilazole in the 0-8 cm soil segment after each of 12 applications (from Table VI of the report) were:

Application	Day	Flusilazole	IN-F7321	Total
1	0		0.06	<0.01
0.06				
2	14		0.12	<0.01
0.12				
3	28	0.16	<0.01	0.16
4	42	0.22	0.01	0.23
5	326	0.22	0.04	0.26
6	339	0.26	0.04	0.3
7	354	0.31	0.04	0.35
8	368	0.31	0.05	0.36
9	694	0.26	0.05	0.31
10	708	0.36	0.08	0.44
11	722	0.39	0.06	0.45
12	737	0.34	0.07	0.41

In the 8-16 cm soil segments radioactivity did not exceed 0.01 mg/kg flusilazole equivalents until after 483 days (after the 6th application at 0.016 mg/kg), with maximum residues of 0.03 mg/kg after the 12th application. In the 16-24 cm segments residues did not exceed 0.01 mg/kg until immediately after the 12th application at 737 days (0.015 mg/kg), but were below 0.01 mg/kg after 844 days. In the 24-36 cm segments no residues exceeded 0.01 mg/kg throughout the study although, as noted in the Table showing percentage radioactivity above, some radioactivity was recorded in this segment after 368 days.

In water/sediment systems

No new information.

In storage and processing

Storage No information.

Processing. The 1991 Meeting requested information on residues of the major flusilazole metabolites (especially IN-F7321, the silanol) in processed fractions field-treated cereals with measurable residues in the grain. A processing study on barley (Guinivan and Desmond, 1993) was provided.

A 91-kg sample of barley grain was taken at maturity 47 days after the second of two over-crop tractor spray applications (at a two-week interval) to a 6 x 156 m barley plot at 420 g ai/ha (3 times the proposed US GAP rate). Bulk samples were shipped at ambient temperatures (a small sample also frozen) for simulated commercial processing at Texas A&M University. Samples were stored frozen until processing approximately 3 months later. The pre-milling steps included aspiration, screening, dehusking and husk removal. Pearled grain was milled (broken and sieved) to remove bran and further milled (reduction and sieving) for shorts, low-grade flour and patent flour.

Fractions were analyzed for flusilazole and its major phenyl metabolites by Du Pont method AMR 2126-91 (Koch, 1993; see "Methods of residue analysis") 16-20 months after processing. Validation data and sample chromatograms were provided. Percentage recoveries from the various fractions (mostly at 0.05 to 0.2 mg/kg fortifications) were flusilazole 90±14, IN-7321 89±19, IN-G7072 107 ±22, IN-37722 95±22, and IN-37738 91±15. The results are summarized in Table 5 (an the metabolites defined in its footnotes). Flusilazole residues were not concentrated in milling fractions which showed ratios of residue in fraction to residue in grain of 0.8 in bran, 0.6 in shorts 0.4 in low grade and 0.5 in patent flour. They were concentrated in normal pre-milling fractions: 9.2-fold in light impurities and 2.4-fold in huskes. A similar pattern was for metabolite IN-F7321. Factors could not be estimated for the other metabolites because the grain did not have measurable residues, although some concentration in the light impurities from pre-milling were observed.

Table 5. Residues of flusilazole and metabolites in barley grain and its processed fractions from simulated commercial processing (Guinivan and Desmond, 1993).

Sample	Residue, mg/kg				
	Flusila-zole	IN-7321[1]	IN-G7072[2]	IN-37722[3]	IN-37738[4]
Treated grain					
Shipped at ambient temp.	0.12, 0.14	0.05, 0.05	<0.05,	<0.05	<0.05
Shipped frozen	0.13, 0.06	0.1, 0.13	<0.05	<0.05	<0.05
Pre-milling fractions					
Light impurities	1.2, 0.82	0.26, 0.58	0.02, 0.11	0.09, 0.12	<0.05, 0.12
Screenings	0.09	0.05	<0.05	<0.05	<0.05
Husks	0.33, 0.19	0.18, 0.24	<0.05, 0.07	0.04, <0.05	0.03, 0.04
Milling					

Sample	Residue, mg/kg				
	Flusila-zole	IN-7321[1]	IN-G7072[2]	IN-37722[3]	IN-37738[4]
fractions					
Bran	0.09	0.07	<0.05	<0.05	<0.05
Shorts	0.07, 0.06	0.04, 0.07	<0.05	<0.05	<0.05
Low-grade flour	0.04	0.03	<0.05	<0.05	<0.05
Patent flour	0.09, 0.03	<0.05, <0.05	<0.05	<0.05	<0.05

[1] IN-7321 = bis(4-fluorophenyl)(methyl)silanol (I)
[2] IN-G7072 (disiloxane) = 1,1,3,3-tetrakis(4-fluorophenyl)-1,3-dimethyldisiloxane (V)
[3] IN-37722 = 2-fluoro-5-[(4-fluorophenyl)(methyl)(1-*H*-1,2,4-triazol-1-ylmethyl)silyl]phenol (IV)
[4] IN-37738 = 2-fluoro-5-[(4-fluorophenyl)(hydroxy)(methyl)silyl]phenol (XII).

Roman numerals refer to structures in Fig. 1, 1989 monograph.

Stability of pesticide residues in stored analytical samples

The 1989 JPMR required the submission of freezer storage stability studies for flusilazole and its metabolites in wheat. The 1991 JMPR reviewed a summary of a stability of flusilazole which reportedly demonstrated that residues in wheat grain were stable up to 36.5 months. The 1991 Meeting considered details of this summary as well as information on freezer storage stability of metabolites to be desirable. Instead of providing details of the previous summary, the manufacturer supplied an entirely new study (Desmond, 1993). Two 2-g samples of wheat or straw were fortified with each compoud and stored for intervals up to 11 months at -20◦C. Results (apparently uncorrected for procedural recoveries) are summarized in Table 6.

The study began before the development of analytical method AMR 2126-91. Although sample chromatograms were provided, they were not sufficiently labelled to be of much use. Control values were recorded as <0.05 mg/kg for all compounds in each matrix. Freshly fortified samples were anaylysed at the same time as the stored samples and results were generally similar for fresh and stored samples throughout the test period and showed similar variation. Differing recoveries therefore appear to have been more the result of analytical variation than of the effects of the storage.

Table 6. Average recoveries [1] of flusilazole and metabolites from wheat grain and straw after laboratory fortification and freezer storage (Desmond, 1993).

Storage Interval (Months)[2]	Percentage recovery from sample, stored/freshly fortified

	Flusilazole 0.27 mg/kg	IN-F7321[3] 0.26 mg/kg	IN-G7072[4] 0.26 mg/kg	IN-37722[5] 0.35 mg/kg	IN-37738[6] 0.27 mg/kg
Grain					
0	87/85	127/123	81/73	69/77	95/93
3	87/100	94/100	89/104	74/80	92/78
4	91/85	140 /123	79/73	74/77	102 /93
5	83/93	79/81	96/108	83/91	78/78
6	70/74	106 /58	73/77	68/86	119 /81
9	78/89	83/92	87/100	77/77	69/63
11	95/96	73/81	106/100	106/126	87/100
Straw					
0	81/85	79/77	92/92	97/69	85/81
3	100 /100	83/85	96/104	91/100	107/107
4	83/85	95/77	88/102	83/69	98/81
5	78/93	94/96	75/88	70/66	96/89
6	85/85	79/119	115/104	103 /109	96/137
9	80/85	125 /135	100/112	83/77	115 /115
11	74/78	81/62	89/92	107/80	83/63

[1]Values for stored samples are averages of two samples. Values for fresh fortification recoveries are single determinations.
[2]Interval from start of freezer storage to extraction.
[3]IN-7321 = bis(4-fluorophenyl)(methyl)silanol
[4]IN-G7072 (disiloxane) = 1,1,3,3-tetrakis(4-fluorophenyl)-1,3-dimethyldisiloxane
[5]IN-37722 = 2-fluoro-5-[(4-fluorophenyl)(methyl)(1-*H*-1,2,4-triazol-1-ylmethyl)silyl]phenol
[6]IN-37738 = 2-fluoro-5-[(4-fluorophenyl)(hydroxy)(methyl)silyl]phenol

Residues in the edible portion of food commodities

The reduction of flusilazole residues in barley milling fractions is discussed in "Fate of residues in processing" above.

RESIDUES IN FOOD IN COMMERCE OR AT CONSUMPTION

No new information.

METHODS OF RESIDUE ANALYSIS

Most of the methods used for residue analyses have been described in earlier monographs, except du Pont methods AMR-115-85 and AMR 2126-91. Method AMR-115-85 is probably a modification of AMR-115-83, previously described. AMR-2126-91 (Koch, 1993) was used for the cereal analyses. It is capable of determining flusilazole and its major phenyl metabolites in wheat grain, straw and forage. It consists in homogenization of the hydrated sample with NaOH and 10% dichloromethane/ethyl acetate, centrifugation, concentration and the determination of flusilazole and the metabolite IN-G7072 by GLC with a mass-selective detector (GC-MSD). Another aliquot is reacted with diazomethane and analyzed by GC-MSD for IN-F7321, IN-37738 and IN-37722. In the case of forage an acetonitrile partition step is added after the initial extraction.

Average percentage recoveries from wheat samples fortified with flusilazole or metabolites at levels from 0.03 to 0.7 mg/kg average recoveries were:

	Forage	Grain	Straw
Flusilazole	97±21%	92±19%	81±13%

IN-G7072	94±11	107±19	100±18
IN-F7321	100±18	97±14	100±12
IN-37738	82±11	83±9	100±21
IN-37722	90±14	99±23	92±13

Sample chromatograms suggest that determinations of 0.05 mg/kg of flusilazole and the metabolites should be feasible in these samples, except possibly IN-F7321 in forage (owing to an interference peak) and flusilazole, IN-G7072 and IN-37722 in straw where 0.1 mg/kg may be more realistic for routine analyses. These judgements are based on a comparison of sample and control chromatograms at the lowest fortification levels. Detection is possible at lower levels.

During the application of the method in grain processing studies (see above) analytical recoveries of 90 ± 14% were reported for flusilazole in grain and its processed fractions at fortification levels mostly ranging from 0.05 to 0.2 mg/kg. Similar or better results were reported for the metabolites. Chromatograms suggest that quantification should be generally achievable at about 0.05 mg/kg for flusilazole and 0.1 mg/kg for the metabolites in these samples.

NATIONAL MAXIMUM RESIDUE LIMITS

The following national MRLs were reported to the Meeting:

Crop	Country	MRL, mg/kg
Banana	Australia	0.2
	The Netherlands	0.05 (under consideration)
Grapes	Australia	0.5
	Spain	0.05 (incl. wine grapes)
Pome fruit	Australia	0.2
	Spain	0.2
Stone fruit	Spain	0.1 (peach/apricot) 0.05 (other stone fruit)
Sugar cane	Australia	0.02
Commodities other than banana	The Netherlands	0* (0.05)

APPRAISAL

Flusilazole was previously reviewed for residues by the 1989, 1990 and 1991 Meetings. The present Meeting reviewed information provided in response to the 1991 JMPR requirement for additional GAP and residue data to confirm the 0.1 mg/kg temporary estimate for peaches and nectarines, and information listed as desirable on grapes, details of wheat grain freezer storage studies, stability of metabolites in freezer-stored grain samples, hen metabolism, metabolites in grain processed fractions and soil studies. Additional residue data on pome fruit, grapes and cereals (although there were no outstanding residue data requirements on these commodities) and new data on sugar cane were also provided.

Fate of residues in animals. Several reports on hen metabolism were provided. Some had been submitted before and some, including the requested study by Smyser, were new. Only the Smyser report included data in need of review by the Meeting.

The report basically combines and summarizes information in two previously reviewed reports and provides further clarification of the residues of metabolites, especially in terms of the percentage of the total radioactivity in poultry tissues and eggs, for both the phenyl and triazole labels. It confirms previous JMPR conclusions that bis(4-fluorophenyl)(methyl)silanol (IN-F7321, the methyl silanol) and 4-fluorophenyl(methyl)silanediol are the predominant residues in poultry tissues

and eggs arising from the phenyllabelled compound and that triazole is the main residue from the triazole label, except in fat where flusilazole is the primary residue from the triazole label.

The report does not effectively answer questions raised by the 1989 and 1991 Meetings concerning differences in residues found between ruminant and poultry metabolism and feeding studies. The Meeting noted and agreed with the 1991 JMPR conclusion that these differences probably result largely from the more detailed residue characterization and identification in the poultry studies than in the ruminant studies. The Meeting also agreed with the 1991 JMPR that although all questions have not been completely answered, the nature of the residue in animal products can be considered to be reasonably well understood in view of the low residues expected (especially for flusilazole) in animal products.

Soil dissipation. The Meeting reviewed the final report of a 3-year soil dissipation study (4 applications per year) for which an interim report was reviewed by the 1989 JMPR. It confirms the 1989 observations that over 92% of the radioactivity is confined to the top 8 cm of soil over the test period, and that the predominant residues in this segment are flusilazole and its silanol metabolite IN-F7321. The author cites statistical evaluation of the data to support the view that residues will reach a steady level at 57% of yearly application levels after repeated application levels under worst-case conditions.

The report cites the steady-state conclusion, the strong adsorption to the top layers of soil, the lack of residues exceeding 0.01 mg/kg in the 24-36 cm soil depths and the weak leaching potential indicated in other studies as evidence that residues in ground water were unlikely. While the data indicate that over 92% of the radioactivity remains in the top 8 cm of the silt loam soil investigated, and indeed that residue levels are extremely low in the 24-36 cm depths, it also shows an increasing penetration by low levels of radioactivity over the test period in this soil type. The identity of these residues in the deeper soil segments was not indicated.

While the adsorption of this persistent pesticide to soil is strong, the 1989 JMPR had noted that uptake of low residue levels can occur in rotational crops and that the leaching potential would be less for silt loams (as in this study) than for more sandy soils. Because the silt loam study was under worst-case conditions (bare ground, repeated applications) and was consistent with reassuring findings of a number of other relevant studies, the Meeting accepted that ground water residues from silt loam soils were unlikely.

Freezer storage stability. Instead of details of a previous 36.5-month study for the parent compound only, the Meeting was provided with a new 11-month freezer storage study of flusilazole and its metabolites in wheat grain and straw. While the results suggest that about 30% of 0.3 mg/kg residues of the parent compound and its phenyl metabolites in grain and straw are lost after various storage intervals up to 11 months, the variability in the recoveries of freshly fortified samples indicates that the apparent losses are probably as much the result of analytical variability as actual storage losses. The Meeting concluded that the data demonstrated adequate stability of flusilazole and the metabolites IN-7321, 1,1,3,3-tetrakis(4-fluorophenyl)-1,3-dimethyldisiloxane (IN-G7072), 2-fluoro-5-[(4-fluorophenyl)(methyl)(1-*H*-1,2,4-triazol-1-ylmethyl)silyl]phenol (IN-37722) and 2-fluoro-5-[(4-fluorophenyl)(hydroxy)(methyl)silyl]phenol (IN-37738) (presumably unconjugated) over 11 months under the conditions of the study.

The 11-month storage interval compares with sampling-to-laboratory-receipt intervals ranging from 2 to 15 months in cereal grain trials from which data were reviewed by the 1989 JMPR. The Meeting did not know the actual sampling-to-analysis intervals for the data reviewed in 1989, although according to the 1989 monograph all samples were generally stored at -20°C.

Cereals. The original 1989 JMPR estimates of maximum residue levels of 0.1

and 2 mg/kg respectively for cereal grains and straws or fodders (dry) were based on maximum residues of 0.07 mg/kg in grain and 1.7 mg/kg in straw. Although there were no outstanding requirements for additional supervised trials data, the Meeting received extensive additional cereal grain, plant, forage and straw data from Europe and North America. Because no need for MRL revisions was indicated, the Meeting only briefly summarized the submitted data on grain and straw. It concluded that there was no need to revise the recently adopted limits of 0.1 mg/kg in the grains and 2 mg/kg in the straws and fodders (dry) of barley, rye and wheat at present. This conclusion may need to be reconsidered at a future Meeting in the light of future GAP information.

Cereal grain processing. The 1991 JMPR reviewed a wheat processing study submitted in response to a 1989 requirement. While no concentration in milled fractions was observed, samples were not analysed for metabolites (especially IN-F7321) and such analysis had been recorded as desirable. A barley grain processing study provided to the Meeting confirmed that no concentration of flusilazole or the major metabolite IN-F7321 occurred in milling fractions.

Grapes. Limited additional information on GAP in Europe and Australia and additional grape data submitted in response to the 1991 requests showed maximum residues reflecting GAP of 0.22 mg/kg compared to the recently adopted CXL of 0.5 mg/kg. A delegation to the CCPR had suggested that a 0.2 mg/kg limit was sufficient. The Meeting confirmed the 1989 JMPR conclusion that residues were unlikely to exceed 0.3 mg/kg.

Pome fruit. Additional GAP information and residue data did not require a revision of the current 0.2 mg/kg limit.

Stone fruit. The 0.1 mg/kg limit for peaches and nectarines recommended by the 1991 JMPR was temporary pending the submission of additional GAP and residue data. It had been based on data from New Zealand and France and GAP from New Zealand and Spain. The Meeting received information on current GAP from Spain, France, Greece (pending) and Italy, and residue data on nectarines from France and on peaches from Australia, Italy, Greece, and the United States. French apricot data were also provided as supporting information. No GAP information was available for Australia or the United States. One to 4 applications at 3-4 g ai/hl and a PHI of 7 to 10 days appears to be usual for countries with established GAP, although in two cases the maximum number of permitted applications was not indicated.

At a 7-day PHI, the new French data or those summarized by the 1991 JMPR which reflect GAP rates showed maximum residues of flusilazole *per se* in peaches of 0.09 mg/kg (1991) or 0.08 mg/kg (1993), except in one trial in the 1993 submission where a residue of 0.55 mg/kg after 8 days was reported from 9 applications at GAP rates. Maximum apricot residues reflecting GAP rates were 0.08 mg/kg after 7 days. Maximum residues in the US trials were 0.09 mg/kg at a 2.4 g ai/hl spray concentration after 7 or 14 days (0.2 mg/kg after 5 days) and 0.3 mg/kg at a 4.8 g ai/hl rate after 12 days. At a pending GAP rate, maximum residues after 7 days in the Greek trials were 0.09 mg/kg. Residues were not detected in the Australian or Italian trials (<0.05 mg/kg and <0.01 mg/kg respectively), but that is not unexpected in view of the long PHIs and the type of application. The Meeting concluded that a 0.5 mg/kg limit was supported for peaches. Observing that GAP for apricots and nectarines is similar to that for peaches, the Meeting concluded that the available data could also mutually support 0.5 mg/kg limits for apricots and nectarines at a 7-day PHI.

Limited data for plums and cherries were insufficient to recommend MRLs.

Sugar cane. No residues (<0.02 mg/kg) were detected in the juice from plants grown after dip treatments of sugar cane sets at fivefold application rates. No stalks were analysed. The Meeting concluded that the data were inadequate to support a limit for sugar cane.

RECOMMENDATIONS

On the basis of the data on residues from supervised trials the Meeting concluded that the residue levels listed below are suitable for establishing maximum residue limits.

Definition of the residue: flusilazole

CCN	Commodity	MRL (mg/kg)		PHI (days), on which recommendation is based
		New	Previous	
FS 0240	Apricot	0.5	-	7
FS 0245	Nectarine	W	0.1 T[1]	7
FS 0247	Peach	0.5	0.1 T[1]	7

[1]Temporary

FURTHER WORK OR INFORMATION

Desirable

1. Submission of analytical method AMR-115-85 cited in Du Pont, 1993, Vol. 1, exhibit 6. Submission of validation information to permit estimation of limits of determination is desirable.

2. On completion, submission of the soil dissipation report AMR-791-87 (Fujinari, 1988). The interim report was reviewed by the 1989 JMPR.

REFERENCES

Australia, 1993. Submission by the government of Australia to the 1993 JMPR. Appendix 24 - Volume 1 of 1 (grapes); Volume 2 of 2 (stone fruit); Volume 2 of 3 (pome fruit); Volume II of III (sugar cane); Appendix 25 -Australian DF label (grapes, apples, pears) and "Liquid Fungicide" label (sugar cane).

Bodden, R. and Kneeland, L. 1984. Metabolism of ^{14}C-DPX-H6573 in Laying Hens. Unpublished Du Pont Report AM-245-84 (see Du Pont 1993, Vol. 5).

Desmond, P.J. 1993. Freezer Storage Stability Study of Flusilazole Fungicide (DPX-H6573) and its Metabolites (IN-F6321, IN-G7072, IN-37722 and IN-37738) in Wheat Grain and Straw. Unpublished Du Pont Report No. AMR 2127-91 completed in response to US EPA data requirements. (See Du Pont, 1993, Vol. II, exhibit 2.

Du Pont, 1993. Supplemental Information on Flusilazole Prepared for the Food and Agriculture Organization:

March 10, 1993 Submission
Vol. I - Residue data and labelling for peaches, nectarines and apricots.
Vol. II - Residue data for cereals.
Vol. III - Residue data for cereals.
Vol. IV - Residue data for cereals.
Vol. V - Chicken metabolism.

April 30, 1993 Submission
Vol. VI - Residue data for peaches and soils and current labelling.

Individual reports are referenced in Table 2 as refs. 1-7 and Table 3 as refs. 1-8 as follows.

Table 2 (Cereals)

Ref.	Subject	Volume	Section	Country
	wheat			
1.	BF-66.630-03-88-19	III	3	Italy
	winter wheat			
2.	BG-BF-88-03	II	5	
	Germany			
	BG-BF-88-04	II	6	Germany
	BE-A-11-89-32-BG	IV	2	UK
	BG-BF-5122-09-89-06	III	3	Germany
	BG-BF-5122-09-89-07	III	4	Germany
	BF-66.630-03/08-88-11	IV	1	Germany
	BF-66.630-03/08-88-01	IV	3	UK
	BF-66.630-03/08-88-08	IV	2	
	Germany			
	BE-A-11-90-02-BG-BF	IV	4	Germany
	BE-A-11-90-03-BG-BF	IV	5	Germany
	AMR-1811-90	II	4	USA
	spring wheat			
3.	AMR-1811-90	II	4	USA
	spring barley			
4.	BE-A-11-89-32-BG	IV	2	
	UK			
	BF-66.630-03-88-01	IV	3	
	UK			
5.	AMR-1811-90	II	4	USA
	winter barley			
6.	BG-BF-88-03	II	5	Germany
	BG-BF-88-04	II	6	
	Germany			
	BE-A-11-89-32-BG	IV	2	
	UK			
	BF-66.630-03-88-01	IV	3	UK
	BF-66.630-03/08-88-08	IV	2	Germany
	BF-66.630-03/08-88-11	IV	1	Germany
	BE-A-11-90-02-BG-BF	IV	2	Germany
	BE-A-11-90-03-BG-BF	IV	5	Germany
	AMR-1811-90	II	4	USA

	winter rye		
7.	BG-BF-88-03	II	5
Germany			
	BG-BF-88-04	II	6
Germany			
	BE-A-11-90-03-BG-BF	IV	5
Germany			

Table 3 (Stone Fruit)

Ref.	Report No./exhibit
1	BE-A-11-89-25-BF/12
2	KO1REO1/13
3	See Australia, 1993. Australian peach data also submitted by du Pont as exhibit 6, Report AMR-1950-91.
4	BF-66.630-03-87-05/8
5	BE-A-11-89-15-BF/5
6	BE-A-11-92-22-BG/7 (du Pont, Vol. VI, 3/30/93).
7	Exhibit 14, Jan. 9, 1985, McIntosh
8	Exhibit 15, Jan. 22, 1985, McIntosh

Guinivan, R.A. and Desmond, P.J., 1993. Magnitude of Residue of Flusilazole in the Processed Fractions of Barley. Unpublished Du Pont Report No. AMR 1972-91 completed in response to US EPA data requirements. (See Du Pont, 1993, Vol. II, exhibit 3).

Koch, S., 1993. Method for the Analysis of Flusilazole and its Major Phenyl Metabolites in Wheat. Unpublished Du Pont Report No. AMR 2126-91, March 25, 1993.

Lin, P., 1988a. Metabolism Study of [Phenyl(U)-^{14}C]DPX-H6573 in Laying Hens. Unpublished Du Pont Report AMR-638-86-1 (see Du Pont, 1993, Vol. V).

Lin, P., 1988b. Supplement to: Metabolism Study of [Phenyl(U)-^{14}C]DPX-H6573 in Laying Hens. Unpublished Du Pont Report AMR-638-86-1 (see Du Pont, 1993, Vol. V).

Lin, P., 1988c. Metabolism Study of [Triazole-3-^{14}C]DPX-H6573 in Laying Hens. Unpublished Du Pont Report AMR-638-86-2 (see Du Pont, 1993, Vol. V).

Lin, P., 1988d. Supplement to: Metabolism Study of [Triazole-3-^{14}C]DPX-H6573 in Laying Hens. Unpublished Du Pont Report AMR-638-86-2 (see Du Pont, 1993, Vol. V).

Smyser, B., 1990. Supplement to: Metabolism Study of [Triazole-3-^{14}-C]DPX-H6573 and [Phenyl(U)DPX-H6573 in Laying Hens. Unpublished Du Pont Report AMR-638-86, Supplement No. 2 (see Du Pont 1993, Vol. V).

Smyser, B., 1993. Long-Term Terrestrial Field Dissipation of [Phenyl(U)-^{14}C]DPX-H6573 at Stine Farm, Delaware. Unpublished Du Pont Report No. AMR 556-86 (See Du Pont, 1993, Vol. VI, exhibit 6).

Stadalius, M., 1984. Supplement to: Metabolism of ^{14}C-DPX-H6573 in Laying Hens. Unpublished Du Pont Report AMR-245-84, MRID 40042130 (see Du Pont, 1993, Vol. V).

Spain, 1993. Documentation, Spanish GAP and Residues Studies in Apples (4), Pears (4), Peaches (7), Grapes (8). Spanish GAP for pome fruit, stone fruit and grapes. June 30, 1993 submission of the Ministerio de Agricultura, Pesca y

Alimentación:

Summary Reports only:

Peaches	Apricots	Grapes
BAT-86-02	BE-A-11-89-25-BF	CTB/E3-5122-02/MC/mf
BE-66.630-03-88-03		
CTB/E3.5122.02/MC/ns		
BE-A-11-89-15-BF		

Apples	Pears
BE-A-11-89-16-BF	BE-66.630-03-88-16
BAT-86-08	BE-A-11-91-03-BF
BE-A-11-89-22-BF	BF-66.630-03-88-18
BE-66.630-03-88-17	BAT-86-04

More Detailed Reports Grapes (US trials):

Test	FWM.JLP.83.8?, Manteca, CA	AAB.K88.83.4?, Madera, CA
	FWM.JLP.83.1?, Sanger, CA	FWM.JLH.83.9?, Riverbank, CA

Wustner, D. 1991. Residue Information on DPX-H-6573 (Flusilazole) Prepared for the World Health Organization, February 1991 - letter: Additional Residue Information for DPX-H6573 Flusilazole (Wustner to Kopisch, January 28, 1991). Exhibit 1 included - "Response to JMPR Review of Flusilazole Animal Metabolism and Feeding Studies (Koch to Wustner, February 27, 1991)" and references: Metabolism Study of Triazole-3-^{14}C-DPX-H6573 and Phenyl(U)-^{14}C-DPX-H6573 in Laying Hens (Smyser, B.P. 1990), unpublished E.I. du Pont de Nemours and Co., Inc. Document No. AMR-638-86, Supplement No. 2. This reference (Smyser, 1990) was requested by the 1991 JMPR).