NATAMYCIN (300)

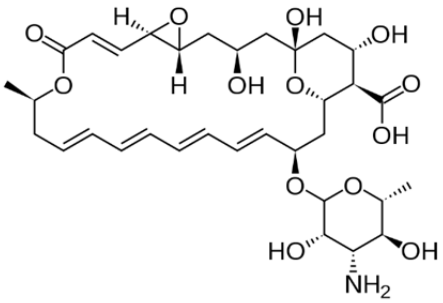
The first draft was prepared by Dr M Doherty, United States Environmental Protection Agency, Washington, DC, USA

EXPLANATION

Natamycin (WHO-approved name; no ISO common name) is a contact fungistat belonging to the polyene macrolide class of compounds. The pesticidal mode of action is binding to ergosterol in the cell membrane resulting in prevention of fungal spore germination. Natamycin is registered only for indoor use (in mushroom houses) and post-harvest treatment (various fruits) in Canada and the United States for control of fungal diseases. Natamycin was scheduled for evaluation at the 48th Session of the CCPR (2016) and considered for the first time for toxicology and residues by the 2017 JMPR.

Note: Throughout this document, values are listed to the precision provided in the submitted reports, except for values calculated by the JMPR (2 significant figures). All rounding was in accordance with ISO standards.

IDENTITY

WHO-approved name	Natamycin
Chemical Name	
IUPAC	(8E,14E,16E,18E,20E)-(1R,3S,5R,7R,12R,22R,24S,25R,26S)-22-(3-amino-3,6-dideoxy-β-D-mannopyranosyloxy)-1,3,26-trihydroxy-12-methyl-10-oxo-6,11,28-trioxatricyclo[22.3.1.05,7]octacosa-8,14,16,18,20-pentaene-25-carboxylic acid
CAS	(1R,3S,5R,7R,8E,12R,14E,16E,18E,20E,22R,24S,25R,26S)-22-[(3-amino-3,6-dideoxy-β-D-mannopyranosyl)oxy]-1,3,26-trihydroxy-12-methyl-10-oxo-6,11,28-trioxatricyclo[22.3.1.05,7]octacosa-8,14,16,18,20-pentaene-25-carboxylic acid
CIPAC No.	None
CAS No.	7681-93-8
Structural Formula	
Molecular formula	C33H47NO13
Molecular mass	665.7

PHYSICAL AND CHEMICAL PROPERTIES

Table 1 Physical and chemical properties of natamycin

Property	Guideline and method	Findings	Reference/ Remarks
Technical Grade Active Ingredient (as trihydrate, 91.02% nominal purity; 7.0% water by weight)			
Appearance (physical state, color, odor)	Not Specified	White, powder; odorless to lightly acidulous	Jovanovich, A.P. *et al*. 2010
Partition coefficient		Log K_{ow} = -3.67	

Property	Guideline and method	Findings	Reference/ Remarks
Stability		Natamycin was found stable at 54 °C for 14 days. Long-term stability demonstrated 2–5 years.	
Vapor pressure and volatility		Not available	--
Pure Active Ingredient ("highly pure technical material")			
Melting Point	Not Specified	Does not melt; darkens at 200 °C; vigorously decomposes at 280–300 °C	Brik, H. 1981.
Relative density		Loose bulk density <3,300 mL/kg; Tapped bulk density >1,700 mL/kg	
Solubility of purified active substance		30–50 ppm @ 20-25 °C and pH 5–7.5; Very soluble at pH ≥ 10 or pH ≤ 2 but rapidly degrades	
Solubility in organic solvents		Methanol = 0.3 %; Ethanol = 40 ppm; Acetone = 10 ppm; Ethyl acetate = 10 ppm; Acetic acid, glacial = 25%	

Formulations

Natamycin is registered as a suspension concentrate (SC) formulations containing 43 or 111 g ai/L.

METABOLISM AND ENVIRONMENTAL FATE

The Meeting received information depicting the fate of natamycin in rats and the toxicity of natamycin and acid-hydrolyzed natamycin in rats. Information regarding breakdown products from acid and alkaline hydrolysis and from UV photolysis was also provided, though these were not guideline studies. Studies investigating the metabolism of natamycin in laboratory animals, plants, and livestockwere not provided to the Meeting. Similarly, studies depicting the environmental fate of natamycin were not provided.

Laboratory animals

A series of investigations using multiple techniques were used to examine the absorption, distribution, and elimination of ^{14}C-labelled natamycin in rats. In total, the data from rats indicates that natamycin is poorly absorbed following oral exposure. Degradation of the parent compound, presumably by bacterial flora, occurs in the intestine with the majority of biotransformation occurring in the large intestine and resulting in products that were more hydrophobic than parent compound. Studies comparing fate of radioactivity following oral or intraperitoneal administration of either parent compound or acid-hydrolysed natamycin (simulating hydrolysis in the stomach) demonstrated that uptake of both the hydrophobic breakdown products and the hydrophilic hydrolysis products was minimal.

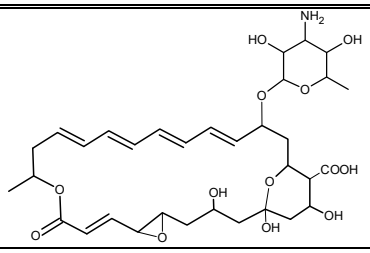
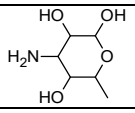
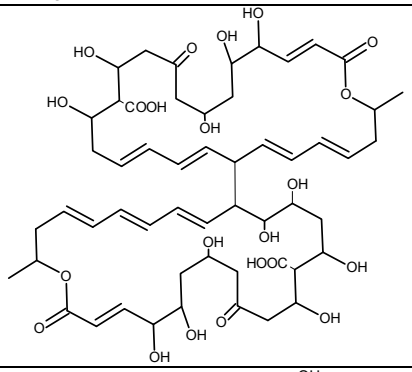
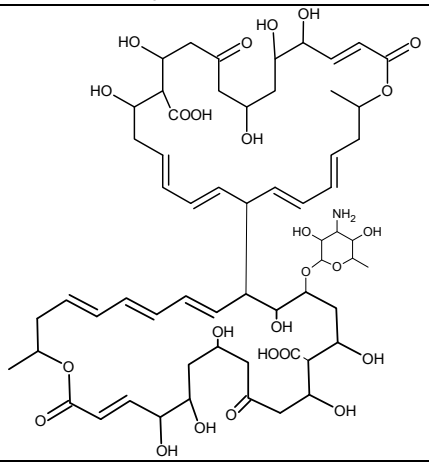
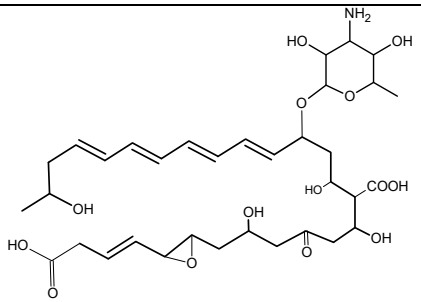
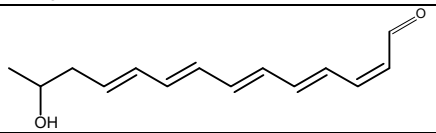
Hydrolysis

Under acid conditions (pH < ~3), the mycosamine moiety is cleaved resulting in an unstable aglycone. The aglycone can react with another aglycone or an intact natamycin molecule to form dimers of either natamycinolidediol or aponatamycin, respectively. Under alkaline conditions (pH > ~9), the lactone moieties are saponified, resulting in formation of natamycoic acid. Natamycoic acid may undergo further breakdown to a long-chain aldehyde, acetone, acetaldehyde, and ammonia (Table 2).

Photolysis

Natamycin is inactivated as a fungistat under ultraviolet radiation (as cited by Brik, 1981). This is likely through oxidation of the tetraene structure. Where oxygen is present, this likely results in the addition of oxygen to the conjugated carbons, leading to the formation of a higher number of hydroxylation sites and natamycin polymers similar to those observed following acid hydrolysis.

Table 2 Acid and alkaline hydrolysis products of natamycin

Name	Structure
Natamycin (8*E*,14*E*,16*E*,18*E*,20*E*)-(1*R*,3*S*,5*R*,7*R*,12*R*,22*R*,24*S*,25*R*,26*S*)-22-(3-amino-3,6-dideoxy-β-D-mannopyranosyloxy)-1,3,26-trihydroxy-12-methyl-10-oxo-6,11,28-trioxatricyclo[22.3.1.$0^{5,7}$]octacosa-8,14,16,18,20-pentaene-25-carboxylic acid	
Acidic Conditions	
Micosamine 4-amino-6-methyltetrahydro-2*H*-pyran-2,3,5-triol	
Natamycinolidediol	
Aponatamycin	
Alkaline Conditions	
Natamycoic acid	
13-Hydroxytetradeca-2,4,6,8,10-pentaenal	
Acetone	

Name	Structure
Acetaldehyde	
Ammonia	NH_3

Conclusions – Metabolism and environmental fate

Given the registered pesticidal uses of natamycin, the plant, livestock, and environmental fate studies typically required by the Meeting are not particularly relevant for assessing natamycin.

Natamycin is stable for several years as the trihydrate dry powder, and aqueous solutions between pH 5 and pH9 are "quite stable" if stored in the dark (as cited by Brik, 1981). Decomposition products that may form following post-harvest treatment are likely to be the same as those observed following hydrolysis and photolysis.

Data are not available to ascertain identity and relative levels of breakdown products that would be expected in mushrooms following application of natamycin to the growth media.

RESIDUE ANALYSIS

Summary of analytical methods

Table 3 Overview of the analytical methods submitted for natamycin.

Report ID *Method ID*	Matrix	Analytes	Extraction	Clean-up	Separation/ Analysis/[LOQ] [a]
Data Gathering					
Marin, 2010 *PTRL 1869W* Cassidy, 2013 029035	Mushroom Mushroom compost Mushroom casing Pineapple (whole, rind, wet bran, meat, and juice)	Natamycin	Methanol	None	HPLC-MS/MS (solvent-based standard for mushroom and pineapple, matrix-matched standard for compost and casing) [0.01 mg/kg mushroom, pineapple] [0.1 mg/kg mushroom compost, casing]
Ferguson, 2016 034208-1 Schreier, 2016 PSM-15-03-01	Citrus (whole fruit, peel, and flesh)	Natamycin	Methanol	Solid-phase extrac'n (flesh only)	HPLC-MS/MS (solvent-based standards) [0.01 mg/kg flesh] [0.1 mg/kg whole fruit, peel]

[a] Defined by the lowest limit of method validation

Mushroom and Pineapple

A single method was used for all residue studies with mushroom and pineapple, and is described in Marin, 2010; PTRL West Study No. 1869W). Prior to extraction, samples are homogenized in the presence of dry ice. Residues are extracted with methanol, filtered, and analysed by HPLC-MS/MS. Natamycin is isolated using reverse-phase chromatography using a C8 (mushroom) or C18 (pineapple) column. For mushroom matrices, the mobile phase was water and methanol running on a gradient starting with 80% water followed by a rapid shift to 2% water; for pineapple, the mobile phase water and acetonitrile running on a linear gradient from 95% water to 0% water. Mass transitions are m/z 666.3 to 503.8 ($m+H^+$) for quantification and m/z 666.3 to 485.5 and m/z 666.3 to 467.4 for confirmation. Recoveries of natamycin are summarized in Table 4. Limits of quantification in the table are based on the lowest level of fortification for which acceptable recoveries were achieved.

Table 4 Recoveries for natamycin using Method PTRL 1869W

Sample material	Fortification level, mg/kg	Recovery, %	Mean recovery, %	RSD, %	Mean overall	RSD overall	LOQ, mg/kg
Method Validation (Report 1915W)							
Mushroom	0.01	74, 94, 92, 90, 71	84	13	89	12	0.01
	0.1	111, 88, 87, 93, 92	94	11			
Compost	0.01	77, 81, 69, 55, 61	69	16	59	22	--
	0.1	57, 55, 48, 46, 41	49	14			
Casing	0.01	69, 80, 89, 68, 55	72	18	70	16	0.1
	0.1	60, 74, 72, 74, 57	67	12			
Whole pineapple	0.01	84, 86, 84	85	1.5	84	3.3	0.01
	1	81, 81, 88	83	1.6			
Pineapple rind	0.01	76, 75, 80	77	3.1	80	5.4	0.01
	1	84, 82, 86	84	2.6			
Pineapple juice	0.01	101, 98, 95	98	2.9	99	3.1	0.01
	1	100, 103, 96	100	3.5			
Concurrent Fortifications (Report 1915W)							
Mushroom	0.01	91, 114, 81	95	18	98	15	0.01
	0.1	90, 119, 94	11	16			
Compost	0.1	84, 108	96	18	93	12	0.1
	1	86, 93	90	5.5			
Casing	0.1	74, 73	74	0.96	76	8.9	0.1
	1	86, 71	78	14			
Whole pineapple	0.01	87, 85, 99, 87	90	7.2	93	7.1	0.01
	1	96, 92, 104, 95	97	5.3			
Pinapple rind	0.01	84	84	--	88	5.7	0.01
	1	91	91	--			
Pineapple wet bran	0.01	72	72	--	75	5.7	0.01
	1	78	78	--			
Pineapple meat	0.01	76	76	--	78	3.6	0.01
	1	80	80	--			
Pineapple juice	0.01	73	73	--	76	5.6	0.01
	1	79	79	--			
Method Validation (Report 029035)							
Mushroom	0.01	94, 94, 101, 98, 101	98	3.6	95	8.8	0.01
	0.1	83, 86, 89, 92, 91	88	4.2			
	1.0	87, 116, 103, 89, 95	98	12			
Compost	0.01	79, 80, 84, 79, 76	80	3.6	99	17	0.01
	0.1	135, 114, 111, 99, 107	113	12			
	1.0	104, 105, 101, 104, 105	104	1.6			
Casing	0.01	87, 92, 91, 106, 82	92	9.8	95	7.6	0.01
	0.1	89, 97, 90, 94, 94	93	3.5			
	1.0	100, 103, 107, 103, 94	101	4.8			

Citrus

The method was used for examination of residue levels in citrus is described in Ferguson, 2016 (Ricera Study No. 034208-1) and in Schreier, 2016 (Study No. PSM-15-03-01). Prior to extraction, samples are homogenized in the presence of dry ice. For whole fruit and peel, residues are extracted with methanol, centrifuged, and analysed by HPLC-MS/MS. Residues in flesh are extracted with methanol and cleaned-up by solid-phase extraction (Waters HLB®), and analysed by HPLC-MS/MS. Natamycin is isolated using reverse-phase chromatography using a C18 column in a gradient mobile phase consisting of 40% water and 60% methanol (with 0.1% acetic acid) transitioning to 95% methanol over three minutes. Mass transitions are m/z 666.3 to 503.8 ($m+H^+$) for quantification and m/z 666.3 to 485.2 and m/z 666.3 to 467.2 for confirmation. Recoveries of natamycin are summarized in Table 5.

Table 5 Recoveries for natamycin from citrus matrices using Method 034208-1

Sample material	Fortification level, mg/kg	Recovery, %	Mean recovery, %	RSD, %	Mean overall	RSD overall	LOQ, mg/kg
Method Validation Study 034208-1							
Grapefruit Whole fruit	0.1	111, 112, 109, 106, 102	108	4.1	108	4.1	0.1
	10	116, 110, 104, 109, 106	109	4.6			
Grapefruit Peel	0.1	83, 86, 85, 79, 86	84	2.9	91	9.0	0.1
	10	99, 103, 99, 97, 89	97	5.2			
Grapefruit Flesh	0.01	98, 92, 104, 90, 99	97	5.6	96	10	0.01
	0.1	87, 120, 89, 91, 90	95	14			
Concurrent Recovery Study PSM-15-03-01							
Grapefruit Whole fruit	0.1	71	71	--	76	7.8	0.1
	10	82	82	--			
Grapefruit Peel	0.1	0.0816	82	--	83	1.4	0.1
	10	84	84	--			
Grapefruit Flesh	0.01	76	76	--	79	4.2	0.01
	0.1	82	82	--			
Lemon Whole fruit	0.1	88	88	--	87	1.4	0.1
	10	86	86	--			
Lemon Peel	0.1	0.099216	92	--	92	0.71	0.1
	10	91	91	--			
Lemon Flesh	0.01	71	71	--	76	7.1	0.01
	0.1	81	81	--			
Orange Whole fruit	0.1	72, 83	78	10	84	12	0.1
	10	82, 101	92	15			
Orange Peel	0.1	79, 71	75	7.5	84	12	0.1
	10	85, 99	92	11			
Orange Flesh	0.01	80, 79	80	0.89	81	2.9	0.01
	0.1	85, 79	82	5.2			

Conclusions – Residue Analysis

For mushrooms, pineapple, and citrus fruit matrices, recoveries from method validation and concurrent fortification samples were within the generally acceptable range of 70–120% and relative standard deviations were less than 20%. For mushroom compost and casing, method performance was not adequate for the method validation samples. Overall, Method PTRL 1869W and Method 034208-1 are very similar, and both are suitable for analysis of natamycin residues in mushroom and fruit matrices.

Stability of residues in stored samples

The Meeting received storage stability studies for natamycin that were conducted concurrently with the residue trials for mushrooms (1915W) and pineapple (197SRUS12R-1) and as a separate study for citrus (grapefruit, orange, and lemon) matrices. For all matrices, control samples were fortified with natamycin at 10× the LOQ (0.01 mg/kg) and placed into frozen storage. For mushroom matrices, 0-day samples were not analysed. Results from storage stability samples are summarized inTable 6.

Table 6 Storage stability results for natamycin in mushroom, pineapple, and citrus matrices

Report	Matrix	Storage time, days	Natamycin, % of nominal application	Average, %	RSD, %	Average concurrent recovery, %	Average, % (normalized to Day 0)
1915W	Mushroom	93	73, 69	71	4.0	95	--
	Mushroom compost	98	96, 64	80	28	104	--
	Mushroom casing	98	70, 38	54	42	72	--
197SRUS12R-1	Whole pineapple	0	83, 83	83	--		100

Report	Matrix	Storage time, days	Natamycin, % of nominal application	Average, %	RSD, %	Average concurrent recovery, %	Average, % (normalized to Day 0)
		48	72, 72	72	--		87
	Pineapple rind	0	77, 77	77	--		100
		48	62, 65	64	3.3		83
	Pineapple juice	0	90, 90	90	--		100
		48	81, 80	80	0.88		89
034209-1	Grapefruit whole fruit	0	76, 75, 77	76	1.0	77	100
		78	84, 79, 83	82	2.6	85	108
	Grapefruit peel	0	80, 82, 80	81	1.2	80	100
		78	76, 75, 78	76	1.5	80	95
	Grapefruit flesh	0	104, 103, 100	102	2.1	99	100
		78	80, 79, 83	81	2.1	92	79
	Lemon whole fruit	0	83, 83, 79	82	2.3	83	100
		78	81, 79, 78	79	1.5	81	97
	Lemon peel	0	94, 93, 90	92	2.1	93	100
		78	83, 82, 82	82	0.58	86	89
	Lemon flesh	0	95, 96, 92	94	2.1	92	100
		78	69, 75, 75	73	3.5	82	77
	Orange whole fruit	0	75, 74, 76	75	1.0	75	100
		78	82, 83, 79	81	2.1	81	108
	Orange peel	0	84, 80, 80	81	2.3	81	100
		78	76, 76, 78	77	1.2	77	94
	Orange flesh	0	102, 104, 102	103	1.2	101	100
		78	78, 82, 78	79	2.3	96	77

Conclusions – Storage Stability

Natamycin is stable for at least 93 days in mushroom, for at least 48 days in pineapple commodities, and for at least 78 days in citrus commodities. The stability of natamycin in mushroom compost or casing over the 98 days of storage cannot be determined based on the data provided.

USE PATTERN

Natamycin is registered for use on indoor-grown mushroom in Canada and the US, and for post-harvest treatment of various fruits in the US. Information on registered uses that was provided to the meeting is summarized in Table 7.

Table 7 Summary of registered use patterns for natamycin

Crop	Country	Site	Formulation		Application			Applic. rate per trtmt.			PHI	Remarks
			Type	Conc., g ai/L	Method	Max No.	RTI, days	Conc., g ai/L	Spray rate, L/ha	Rate, kg ai/ha		
Fruits												
Citrus [Calamondin (*Citrus mitis*, *Citrofortunella mitis*), Citrus citron (*Citrus medica*), Citrus hybrids (*Citrus spp.*) (includes chironja, tangelo, tangor), Grapefruit (*Citrus paradise*), Kumquat (*Fortunella spp.*), Lemon (*Citrus jambhiri, Citrus limon*), Lime (*Citrus arantiifolia*), Mandarin (tangerine) (*Citrus reticulate*), Orange, sour (*Citrus aurantium*), Orange, sweet (*Citrus sinensis*), Pummelo (*Citrus grandis*, *Citrus maxima*), Satsuma mandarin (*Citrus unshiu*), and all cultivars and hybrids.]												
Citrus	USA	Po	SC	111	In-line dip or drench	1	--	0.99	--	--	--	Treat fruits for 10 seconds and drain
					In-line aqueous or fruit coating spray	1	--	0.99	--	--	--	--
Assorted tropical and sub-tropical fruit - inedible peel												
Pineapple	USA	Po	SC	43	Dip, pour, or cascade	1	--	11	--	--	--	After treatment, allow fruits to dry, then spray peduncle.

Crop	Country	Site	Formulation		Application			Applic. rate per trtmt.			PHI	Remarks
			Type	Conc., g ai/L	Method	Max No.	RTI, days	Conc., g ai/L	Spray rate, L/ha	Rate, kg ai/ha		
Edible fungi												
Mushroom	Canada	In	SC	111	Surface drench	2	NS	NS	NS	2.22	6 hours	Apply once at casing and once at pinning.
	USA	In	SC	111	Surface drench	4	NS	NS	NS	2.23	6 hours	After casing: Apply any time after the casing layer has been applied and before flushing. After flushing: Apply any time after flushing up until 6 hours before picking begins. Between breaks: Apply any time between each set of breaks, up until 6 hours before picking begins. Do not apply more than one time to mushrooms remaining between each set of breaks.

Po = post-harvest, In = Indoor, NS = Not Specified

RESULTS OF SUPERVISED RESIDUE TRIALS ON CROPS

The Meeting received data from supervised residue trials reflecting use on growing mushrooms and post-harvest use on citrus and pineapple.

The field trial reports included method validation data, as recoveries from spiked samples at levels reflecting those observed in the field trial samples; dates from critical events during the study, including application, harvest, storage, and analysis; as well as detailed information on the treatment parameters. Analytical reports were sufficiently detailed and included example chromatograms and example calculations. Samples were analysed by Method PTRL 1869W described above.

The field trial study designs included control sample, for which residues were < 0.01 mg/kg (i.e. < LOQ) and are not included in the summary tables in this evaluation.

In the summary tables, values used for making maximum residue level recommendations are underlined and highest individual values for estimating dietary intake are bolded. Trials that were determined not to be independent are surrounded by a heavy border (Table 9, for example).

Supervised trials for natamycin:

Category	Crop	Table
Fruits	Citrus (FC 0001)	8
Trop. and sub-trop. fruit – inedible peel	Pineapple (FI 0353)	9
Fruiting veg., other than cucurbits	Mushrooms (VO 0450)	10

Citrus Fruits

As part of a study investigating the efficacy of various fungicides, including natamycin, to control green mold and sour rot (Smilanick *et al.*; Report 160901), treated lemons and oranges were analysed for residues of natamycin. The study is not a supervised residue trial, and details regarding fungicide treatments, storage durations and conditions, and analytical methods were not provided.

Results list treatments as either by recirculating flooder or 30-second dip with natamycin at concentrations of either 0.5 or 1 g ai/L, alone or in combination with other fungicides.

In a second study investingating residues of natamycin residues in citrus (Schreier, 2016; Report PSM-15-03-01), natamycin was applied as a post-harvest aqueous dip to control green mold and sour rot. Grapefruits, lemons, and oranges were harvested at maturity and shipped to treatment facilites in Florida (grapefruit) or California (lemon, orange) where they were dipped for 10–15 seconds in a treatment solution containing ca. 1000 ppm natamycin. The treated fruits were allowed to dry and placed into frozen storage. Fruits from one location were divided into batches, treated as previously described, and placed into refrigerated storage in order to determine residue decline. At the end of the decline period, the samples were transferred to frozen storage prior to analysis.

Residues in whole fruit, peel, and flesh were extracted with methanol, cleaned up by solid-phase extraction (flesh only), and analysed by LC-MS/MS.

Table 8 Results of natamycin residue trials with citrus

Study No. Trial No.	Crop *Variety*	Application				Matrix	Days after last application	Natamycin, mg/kg [mean]
		No.	Timing	Method (Formul'n)	g ai/L			
GAP USA	Citrus	1	Post-harvest	Dip, drench, or spray	0.99	--	Not Specified	--
060901 2	Lemon	1	Post-harvest	Flood (SC)	0.5	Fruit	Not reported	0.53
060901 3	Lemon	1	Post-harvest	Dip (SC)	0.5	Fruit	Not reported	1.14
060901 4	Lemon	1	Post-harvest	Flood (SC)	1.0	Fruit	Not reported	1.02
060901 5	Lemon	1	Post-harvest	Flood (SC)	1.0	Fruit	Not reported	0.28
060901 6	Lemon	1	Post-harvest	Flood (SC)	1.0	Fruit	Not reported	0.49
060901 7	Lemon	1	Post-harvest	Dip (SC)	1.0	Fruit	Not reported	1.94
060901 13	Lemon	1	Post-harvest	Flood + FDL [a] (SC)	0.5	Fruit	Not reported	0.48
060901 14	Lemon	1	Post-harvest	Flood + FDL [a] (SC)	1.0	Fruit	Not reported	1.77
060901 15	Lemon	1	Post-harvest	Flood + IMZ [a] (SC)	1.0	Fruit	Not reported	0.78
060901 16	Lemon	1	Post-harvest	Flood + PYR [a] (SC)	1.0	Fruit	Not reported	1.02
160901 17	Lemon	1	Post-harvest	Flood + TBZ [a] (SC)	1.0	Fruit	Not reported	0.40
060901 2	Orange	1	Post-harvest	Flood (SC)	0.5	Fruit	Not reported	0.57
060901 3	Orange	1	Post-harvest	Dip (SC)	0.5	Fruit	Not reported	1.83
060901 4	Orange	1	Post-harvest	Flood (SC)	1.0	Fruit	Not reported	1.11
060901 5	Orange	1	Post-harvest	Flood (SC)	1.0	Fruit	Not reported	0.28
060901 6	Orange	1	Post-harvest	Flood (SC)	1.0	Fruit	Not reported	0.40

Study No. Trial No.	Crop *Variety*	Application				Matrix	Days after last application	Natamycin, mg/kg [mean]
		No.	Timing	Method (Formul'n)	g ai/L			
060901 7	Orange	1	Post-harvest	Dip (SC)	1.0	Fruit	Not reported	1.83
060901 13	Orange	1	Post-harvest	Flood + FDL [a] (SC)	0.5	Fruit	Not reported	0.55
060901 14	Orange	1	Post-harvest	Flood + FDL [a] (SC)	1.0	Fruit	Not reported	1.59
060901 15	Orange	1	Post-harvest	Flood + IMZ [a] (SC)	1.0	Fruit	Not reported	1.02
060901 16	Orange	1	Post-harvest	Flood + PYR [a] (SC)	1.0	Fruit	Not reported	0.95
160901 17	Orange	1	Post-harvest	Flood + TBZ [a] (SC)	1.0	Fruit	Not reported	0.30
PSM-15-03-01 01	Grapefruit *Texan Star*	1	Post-harvest	Dip (SC)	1.0	Peel	0	1.70, 1.78 [1.7]
						Flesh	0	0.0848, 0.0302 [0.058]
						Fruit	0	1.10, 0.97 [1.0]
PSM-15-03-01 02	Grapefruit *Red Flame*	1	Post-harvest	Dip (SC)	1.0	Peel	0	1.75, 2.00 [1.9]
						Flesh	0	0.0365, 0.0262 [0.031]
						Fruit	0	0.998, 1.09 [1.0]
PSM-15-03-01 03	Grapefruit *Red Flame*	1	Post-harvest	Dip (SC)	1.0	Peel	0	1.67, 1.41 [1.5]
						Flesh	0	0.0137, 0.0246 [0.019]
						Fruit	0	0.760, 0.868 [0.81]
PSM-15-03-01 04	Lemon *Lisbone*	1	Post-harvest	Dip (SC)	1.0	Peel	0	1.56, 1.99 [1.85]
						Flesh	0	0.0721, 0.139 [0.11]
						Fruit	0	1.59, 1.79 [1.7]
PSM-15-03-01 05	Lemon *Lisbone*	1	Post-harvest	Dip (SC)	1.0	Peel	0	1.68, 1.65 [1.7]
						Flesh	0	0.079, 0.042 [0.060]
						Fruit	0	1.44, 1.59 [1.5]
PSM-15-03-01 06	Lemon *Lisbone*	1	Post-harvest	Dip (SC)	1.0	Peel	0	3.01, 2.66 [2.8]
						Flesh	0	0.168, 0.023 [0.096]
						Fruit	0	1.92, 1.58 [1.8]
PSM-15-03-01 07	Orange *Navel*	1	Post-harvest	Dip (SC)	1.0	Peel	0	2.14, 1.36 [1.8]
							7	2.28, 2.18 [2.2]
							14	1.60, 1.56 [1.6]
							28	2.90, 2.56 [2.7]
							56	2.96, 2.84 [2.9]
						Flesh	0	0.0187, 0.0433 [0.031]
							7	0.046, 0.115 [0.080]
							14	0.0375, 0.0502 [0.044]
							28	0.0379, 0.0658 [0.052]
							56	0.0466, 0.0353 [0.041]
						Fruit	0	2.11, 1.56 [1.8]
							7	1.97, 1.88 [1.9]
							14	1.89, 1.88 [1.9]
							28	1.59, 1.78 [1.7]
							56	1.62, 1.83 [1.7]
PSM-15-03-01 08	Orange *Bricks*	1	Post-harvest	Dip (SC)	1.0	Peel	0	1.61, 1.97 [1.8]
						Flesh	0	0.0723, 0.0792 [0.076]
						Fruit	0	1.30, 1.23 [1.3]
PSM-15-03-01 09	Orange *Mandarin*	1	Post-harvest	Dip (SC)	1.0	Peel	0	5.94, 4.86 [5.4]
						Flesh	0	0.104, 0.024 [0.064]

Study No. Trial No.	Crop *Variety*	Application				Matrix	Days after last application	Natamycin, mg/kg [mean]
		No.	Timing	Method (Formul'n)	g ai/L			
						Fruit	0	2.21, 2.48 [2.3]

[a] FDL = fludioxanil, IMZ = imazalil, PYR = pyrimethanil, TBZ = thiabendazole

Assorted tropical and sub-tropical fruit – inedible peel

Pineapple

Pineapples were collected from plantations in Costa Rica and transported to Biotech CR GRM S.A. in Cartago, Costa Rica for storage and treatment (Report 197SRUS12R-1). Four trials were conducted. Trials A and C used the MD-2 variety of pineapple and Trials B and D used the Montelirio variety. Samples from Trial A were used to determine residue decline; samples from Trials B, C, and D were used for analysis of residues in whole fruits and processed commodities (see Fate of Residues in Storage and Processing, below, for further discussion).

Treatment solutions were prepared as an aqueous-wax mixture using Zivion A or Zivion P formulations of natamycin. Fresh treatment solution was prepared for each of the four trials. Each solution had a nominal concentration of 400 ppm natamycin. For each trial, fourty pineapples were treated by pouring a sufficient volume of the treatment solution over each fruit to ensure good coverage. The fruits were allowed to drain and air dry, and then the peduncles were sprayed with 1 mL of treatment solution, constituting a single post-harvest treatment. The fruits were again allowed to dry.

Trial A: Four fruits were selected at random following treatment and designated as 0-time samples. For these fruits, the crowns were removed and the fruits were quartered. Opposite quarters were retained, composited, and frozen within two hours of collection. The 0-time samples were shipped on dry ice to the analytical laboratory and placed into frozen storage upon receipt. The remaining pineapples from Trial A, designated for residue-decline analysis, were maintained whole and with crowns intact, and were shipped at ambient temperature to the analytical laboratory. At the analytical laboratory, fruits were stored in a cold room (3–9 °C) for their designated residue-decline interval, at which point they were transferred to frozen storage. The duration of frozen storage for all samples (0-day and residue-decline samples) was no more than 24 days. Each sample was a composite of four fruits and consisted of alternate quarters per fruit.

Trials B, C, and D: Fruit samples were maintained at ambient temperature following collection and during shipment to the processing laboratory (elapsed time 3 days). Prior to shipment, the crowns were removed by twisting off; otherwise, fruits remained whole. Samples were kept in cold storage at the processing facility from receipt until processing. The time intervals between treatment and frozen storage ranged from 11 to 16 days, and samples were kept in frozen storage for approximately 1 month (27–31 days).

Table 9 Results of natamycin residue trials with pineapple

Study No. Trial No. Country, Region, Year	Crop *Variety*	Application				Matrix	Days after last application	Natamycin, mg/kg [mean]
		No.	Timing	Method (Formul'n)	g ai/L			
GAP USA	Pineapple	1	Postharvest	Dip, pour, drench	11	--	Not Specified	--
197SRUS12R-1 A-02 Costa Rica America, Central 2012	Pineapple *MD-2*	1	Postharvest	Drench (Zivion A)	10	fruit	0	0.87, 0.73 [0.80]
							4	0.46, 0.50 [0.48]
							7	0.43, 0.39 [0.41]

Study No. Trial No. Country, Region, Year	Crop *Variety*	Application				Matrix	Days after last application	Natamycin, mg/kg [mean]
		No.	Timing	Method (Formul'n)	g ai/L			
							14	0.32, 0.32 [0.32]
							21	0.35, 0.25 [0.30]
197SRUS12R-1 A-03 Costa Rica America, Central 2012	Pineapple *MD-2*	1	Postharvest	Drench (Zivion P)	11	fruit	0	0.78, 0.94 [0.86]
							4	0.62, 0.58 [0.60]
							7	0.47, 0.45 [0.46]
							14	0.41, 0.41 [0.41]
							21	0.46, 0.46 [0.46]
197SRUS12R-1 B-02 Costa Rica America, Central 2012	Pineapple *Montelirio*	1	Postharvest	Drench (Zivion A)	10	fruit	14	0.19, 0.12 [0.16]
197SRUS12R-1 B-03 Costa Rica America, Central 2012	Pineapple *Montelirio*	1	Postharvest	Drench (Zivion P)	11	fruit	15	0.22, 0.20 [0.21]
197SRUS12R-1 C-02 Costa Rica America, Central 2012	Pineapple *MD-2*	1	Postharvest	Drench (Zivion A)	10	fruit	16	0.34, 0.35 [0.34]
197SRUS12R-1 C-03 Costa Rica America, Central 2012	Pineapple *MD-2*	1	Postharvest	Drench (Zivion P)	11	fruit	16	0.53, 0.55 [0.54]
197SRUS12R-1 D-02 Costa Rica America, Central 2012	Pineapple *Montelirio*	1	Postharvest	Drench (Zivion A)	10	fruit	11	0.14, 0.15 [0.14]
197SRUS12R-1 D-03 Costa Rica America, Central 2012	Pineapple *Montelirio*	1	Postharvest	Drench (Zivion P)	11	fruit	14	0.24, 0.17 [0.20]

Fruiting vegetables, other than Cucurbits

Mushroom

The Meeting received a study investigating residues of natamycin in treated white button mushrooms and in steam-treated mushroom casing and compost following either two or four applications of natamycin (Report 1915W). Mushrooms were grown in trays under normal cultivation practices. Applications consisted of 2 mL of natamycin formulation applied in 250 mL of water to 1 square meter either at casing and pinning (Treatment A), or at casing, pinning, between 1st and 2nd breaks, and between 2nd and 3rd breaks (Treatment B). The treatment rate is equivalent to 2.2 kg/ha per application.

Mushrooms were harvested three times. Relative to treatments, harvests were 9 (1st break), 15 (2nd break), and 23 days (3rd break) after the last application for Treatment A, and 9 (1st break), 4 (2nd break), and 5 days (3rd break) after the last application for Treatment B. Duplicate samples of ca.

100 g each were harvested per tray. In addition, duplicate samples of casing and compost were collected at each sampling time. All samples were frozen immediately and shipped on dry ice to the analytical laboratory. Samples were stored frozen for up to 84 days for mushrooms, up to 99 days for compost, and up to 100 days for casing. Results from Treatment A were reported for the 1st break samples only. Following steam treatment (substrate maintained at ca 65 °C for 24 hrs), residues of natamycin were <LOQ in all samples of casing and compost.

In a second study (Cassidy, 2013; Report No. 029035), Natamycin was applied to mushrooms (and casing and compost) at different growth stages: at casing, at pinning, between first and second breaks, and between second and third breaks. Mushrooms were harvested at breaks 2 and 3, each at 6, 12, 24, and 48 hours after application. In addition, casing and compost samples were collected pre-steam-off and after 8, 12, and 16 hours of steaming. Residues of natamycin were analysed by the method described previously.

Table 10 Results of natamycin residue trials with mushrooms (Study 1915W)

Trial No. Country Region Year	Crop *Variety*	Application				Matrix	Days after last application	Natamycin, mg/kg [mean]
		No.	Timing	Method (Formul'n)	kg ai/ha			
GAP USA	Mushroom	4	After casing, After flushing, Break 1-2, and Break 2-3	Surface drench	2.2 2.2 2.2 2.2	--	0.25 (6 hrs)	--
GAP Canada	Mushroom	2	At casing At pinning	Surface drench	2.2 2.2	--	0.25 (6 hrs)	--
Study 1915W								
1869W Ontario, Canada America, North 2009 Tray 29	Mushroom *White button*	2	At casing At pinning	Surface drench (Delvocid L)	2.2 2.2	Mushroom	9 (1st break)	0.059, 0.089 [0.074]
Tray 30							9 (1st break)	0.027, 0.028 [0.028]
Tray 31							9 (1st break)	0.029, 0.018 [0.024]
Tray 32							9 (1st break)	0.018, 0.019 [0.018]
1869W Ontario, Canada America, North 2009 Tray 1	Mushroom *White button*	4	At casing At pinning Break 1-2	Surface drench (Delvocid L)	2.2 2.2 2.2	Mushroom	9 (1st break)	0.023, 0.022 [0.022]
Tray 2							9 (1st break)	0.037, < 0.01 [0.019]
Tray 3							9 (1st break)	0.027, 0.034 [0.031]
Tray 4							9 (1st break)	0.034, 0.027 [0.031]
Tray 1			Break 2- 3		2.2		4 (2nd break)	0.24, 0.14 [0.19]
Tray 1							5 (3rd break)	0.041, 0.15 [0.096]
Study 029035								
Kittanning, PA USA America, North	Mushroom *White button*	4	At casing At pinning Break 1-2	Surface drench (Zivion M)	2 2 2	Mushroom	-- -- 0.25	3.94, 3.47, 3.48 [3.6]
							0.5	1.75, 2.29, 1.73 [1.9]
							1	2.98, 2.87, 3.55 [3.1]
							2	1.85, 2.10, 2.05 [2.0]
			Break 2- 3		2	Mushroom	0.25	4.97, 5.43, 3.65 [4.7]
							0.5	3.41, 4.15, 3.63 [3.7]
							1	4.07, 4.46, 3.79 [4.1]
							2	2.90, 2.18, 2.89 [2.7]
						Casing (pre-steam)	0	3.04, 4.36, 5.58 [4.3]
						Casing (post-steam)	0.33	0.52, 1.67, 0.28 [0.82]
							0.5	0.12, 0.73, 0.22 [0.33]
							0.67	0.07, 0.17, 0.08 [0.11]

Trial No. Country Region Year	Crop *Variety*	Application				Matrix	Days after last application	Natamycin, mg/kg [mean]
		No.	Timing	Method (Formul'n)	kg ai/ha			
						Compost (pre-steam)	0	0.044, 0.025, 0.098 [0.060]
						Compost (post-steam)	0.33	0.001, 0.002, 0.031 [0.010]
							0.5	0.024, < 0.003, 0.016 [0.010]
							0.67	< 0.003, < 0.003, < 0.003 [< 0.003]

FATE OF RESIDUES IN STORAGE AND PROCESSING

Residues after processing

Citrus

Fruit samples from the second citrus study described above (Report PSM-15-03-01) were separated into peel and flesh. Residues in those fractions and in the whole fruit are summarized in Table 11.

Table 11 Summary of residues in citrus peel, flesh, and whole fruit from Study PSM-15-03-01

Crop	Trial ID	Matrix	Mean natamycin, mg/kg	Processing factor
Grapefruit	PSM-15-03-01 1	Whole fruit	1.0	--
		Peel	1.7	1.7
		Flesh	0.058	0.058
	PSM-15-03-01 2	Whole fruit	1.0	--
		Peel	1.9	1.9
		Flesh	0.031	0.031
	PSM-15-03-01 3	Whole fruit	0.81	--
		Peel	1.5	1.9
		Flesh	0.019	0.023
Lemon	PSM-15-03-01 4	Whole fruit	1.7	--
		Peel	1.85	1.1
		Flesh	0.11	0.065
	PSM-15-03-01 5	Whole fruit	1.3	--
		Peel	1.7	1.3
		Flesh	0.06	0.046
	PSM-15-03-01 6	Whole fruit	1.8	--
		Peel	2.8	1.6
		Flesh	0.096	0.053
Orange	PSM-15-03-01 7	Whole fruit	1.8	--
		Peel	1.8	1.0
		Flesh	0.031	0.017
	PSM-15-03-01 8	Whole fruit	1.3	--
		Peel	1.8	1.4
		Flesh	0.076	0.058
	PSM-15-03-01 9	Whole fruit	2.3	--
		Peel	5.4	2.3
		Flesh	0.064	0.028

Pineapple

Fruit samples from Trials B, C, and D described above were designated for processing. Whole fruits from these trials were maintained at ambient temperature following collection and during shipment to the processing laboratory (elapsed time 3 days). Prior to shipment, the crowns were removed by twisting off; otherwise, fruits remained whole. Upon receipt at the processing laboratory, the

pineapples were maintained at 2–7 °C until processing. Fruits were processed into flesh (edible fruit), rind, juice, and wet bran using procedures that simulated commercial processing, and mass balance data were provided for all samples. Samples were stored for no more than 31 days and residues were analysed using Method PTRL 1869W.

Table 12 Effect of processing on residues of natamycin in pineapple (Study 197STUS12R-1)

Plot Year	Formulation	Matrix	Natamycin, mg/kg [mean]	Processing factor
B-02 2012	Zivion A	Fruit	0.19, 0.12 [0.16]	--
		Flesh	< 0.01, < 0.01 [< 0.01]	< 0.062
		Juice	< 0.01, < 0.01 [< 0.01]	< 0.062
		Rind	0.25, 0.21 [0.23]	1.4
		Bran	0.020, 0.025 [0.022]	0.14
B-03 2012	Zivion P	Fruit	0.22, 0.20 [0.21]	--
		Flesh	< 0.01, < 0.01 [< 0.01]	< 0.048
		Juice	0.011, < 0.01 [< 0.01]	< 0.048
		Rind	0.69, 0.55 [0.62]	3.0
		Bran	0.032, 0.031 [0.032]	0.15
C-02 2012	Zivion A	Fruit	0.34, 0.35 [0.34]	--
		Flesh	< 0.01, < 0.01 [< 0.01]	< 0.029
		Juice	0.018, 0.012 [0.015]	0.044
		Rind	0.68, 0.67 [0.67]	2.0
		Bran	0.054, 0.052 [0.053]	0.16
C-03 2012	Zivion P	Fruit	0.53, 0.55 [0.54]	--
		Flesh	0.013, 0.013 [0.013]	0.024
		Juice	0.068, 0.069 [0.069]	0.13
		Rind	1.1, 1.2 [1.1]	2.0
		Bran	0.18, 0.17 [0.17]	0.31
D-02 2012	Zivion A	Fruit	0.14, 0.15 [0.14]	--
		Flesh	< 0.01, < 0.01 [< 0.01]	< 0.071
		Juice	< 0.01, < 0.01 [< 0.01]	< 0.071
		Rind	0.42, 0.34 [0.38]	2.7
		Bran	0.024, 0.017 [0.020]	0.14
D-03 2012	Zivion P	Fruit	0.24, 0.17 [0.21]	--
		Flesh	< 0.01, < 0.01 [< 0.01]	< 0.048
		Juice	< 0.01, < 0.01 [< 0.01]	< 0.048
		Rind	0.52, 0.43 [0.47]	2.2
		Bran	0.024, 0.034 [0.029]	0.14

Mushroom

Samples from Treatment B and harvested after the second and third breaks were analysed before and after gentle washing. The results are summarized below.

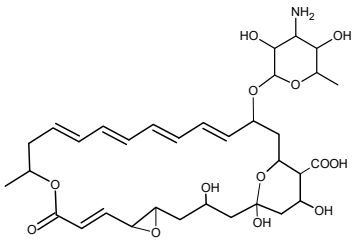
Table 13 Effect of washing on residues of natamycin in mushroom (Study 197STUS12R-1)

Plot Year	Matrix	Natamycin, mg/kg [mean]	Processing factor
Tray 1, 2nd break 2009	Mushroom (unwashed)	0.24, 0.14 [0.19]	--
	Mushroom (washed)	< 0.01, 0.076 [0.036]	0.19
Tray 1, 3rd break 2009	Mushroom (unwashed)	0.041, 0.15 [0.096]	--
	Mushroom (washed)	0.012, 0.022 [0.017]	0.18

APPRAISAL

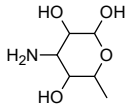
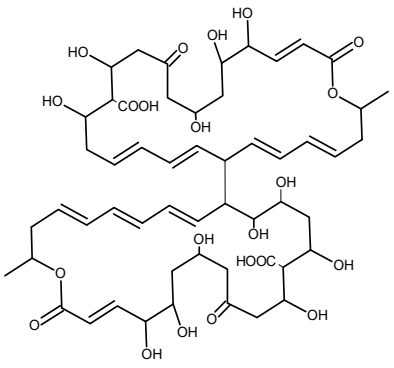
Residue and analytical aspects of natamycin were considered for the first time by the present Meeting. The residue evaluation was scheduled for the 2017 JMPR by the 48th Session of the CCPR. The Meeting noted that natamycin is used as a preservative in sausages and cheese and other dairy products and that the Joint FAO/WHO Expert Committee on Food Additives (JECFA) established a maximum ADI for natamycin of 0.3 mg/kg bw in 1976 and confirmed that ADI in 2006. In addition, natamycin is used to treat fungal keratitis and is on the WHO's List of Essential Medicines.

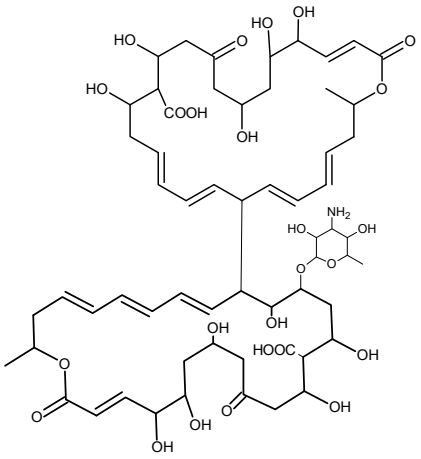
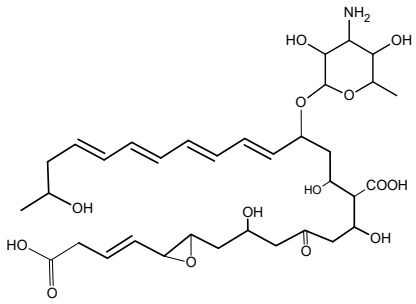
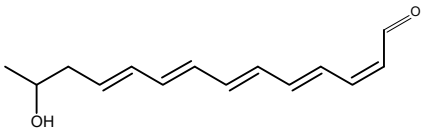
Natamycin is a contact fungistat belonging to the polyene macrolide class of compounds. The pesticidal mode of action is binding to ergosterol in the cell membrane resulting in prevention of fungal spore germination. The uses under consideration by the Meeting are application to mushrooms and post-harvest application to fruits. The Meeting received information on rat metabolism of natamycin, as well as acid and alkaline hydrolysis and UV photolysis. Data depicting the metabolism of natamycin in plants, fungi, livestock, and soils were not provided. Limited residue trial data, analytical methods data, storage stability data, and processing data were provided. Livestock feeding studies were not available.



Natamycin (8*E*,14*E*,16*E*,18*E*,20*E*)-(1*R*,3*S*,5*R*,7*R*,12*R*,22*R*,24*S*,25*R*,26*S*)-22-(3-amino-3,6-dideoxy-β-D-mannopyranosyloxy)-1,3,26-trihydroxy-12-methyl-10-oxo-6,11,28-trioxatricyclo[22.3.1.$0^{5,7}$]octacosa-8,14,16,18,20-pentaene-25-carboxylic acid

In this appraisal, the following abbreviated names were used for metabolites.

Identifier	Chemical Structure
Micosamine 4-amino-6-methyltetrahydro-2*H*-pyran-2,3,5-triol	
Natamycinolidediol	

Identifier	Chemical Structure
Aponatamycin	
Natamycoic acid	
13-Hydroxytetradeca-2,4,6,8,10-pentaenal	
Acetone	
Acetaldehyde	
Ammonia	NH_3

Plant and animal metabolism

The Meeting did not receive metabolism studies for natamycin in laboratory animals, plants (primary or rotational), fungi, or livestock.

Environmental fate in soil and water

The Meeting received information on acid and alkaline hydrolysis, and aqueous photolysis. The information that was provided is from a published book chapter and not from studies conducted according to guidelines. Nevertheless, the information is useful for characterizing the expected behaviour of natamycin under hydrolytic and photolytic conditions.

Under acid conditions, natamycin forms an unstable aglycone molecule which can dimerize with another natamycin aglycone to form natamycinolidediol or combine with an intact natamycin molecule to form aponatamycin. Under alkaline conditions, natamycin undergoes saponification to form natamycoic acid, which may undergo additional breakdown to form aldehydes, ketones, and ammonia.

Ultraviolet irradiation causes natamycin to become inactive as a fungistat. The process is believed to be via oxidation of the tetraene structure leading to formation of polymers similar to those observed during acid hydrolysis.

Methods of analysis

The Meeting received description and validation data for analytical methods for residues of natamycin in tropical fruits (citrus and pineapple) and mushrooms.

For all of the submitted methods, residues of natamycin are extracted with methanol and analysis for residues is by LC-MS/MS. For citrus flesh only, the methanol extracts undergo clean-up by solid-phase extraction prior to analysis. The methods were validated to an LOQ of 0.01 mg/kg, as a lower limit of method validation, in mushroom, pineapple, and citrus flesh, and to 0.1 mg/kg in mushroom casing and compost and citrus whole fruit and peel. Suitable ion transitions are available for residue quantification and confirmation. Mean recoveries from edible commodities (mushrooms and fruits) ranged from 70% to 108% with a maximum relative standard deviation of 15%. The methods have been shown to be suitable for analysis of natamycin in high-acid and high-water commodities.

Stability of residues in stored analytical samples

The Meeting received data on the stability of residues of natamycin in mushroom, pineapple, and citrus matrices. Homogenized control samples of each matrix were fortified with natamycin and placed into frozen storage (≤ -10 °C) concurrently with residue trial samples.

Residues of natamycin were stable in mushrooms (frozen, temperature not specified) for at least 93 days, in pineapple commodities for at least 48 days, and in citrus commodities for at least 78 days. The Meeting noted that the mushroom analysis did not include 0-day samples to confirm the fortification level.

Definition of the residue

Plants/fungi

Except for the use on mushrooms, the uses under consideration by the Meeting are post-harvest uses. As such metabolism under typical agricultural field conditions is not particularly germane to determining residue definitions.

Natamycin undergoes breakdown under hydrolytic and photolytic conditions but is considered to be stable under dry, dark conditions. For the post-harvest uses being considered by the Meeting, natamycin is not expected to undergo significant degradation on the surface of the fruits. Residue decline data for citrus support this contention; however, residue decline data from pineapple show an approximately 2-fold reduction in residues between Day 0 and Day 7 after treatment, followed by a residue plateau from post-treatment days 7–21 (study duration 0–21 days). As for mushrooms, mushroom compost and casing soil are typically maintained at near-neutral pH levels[2] and cultivation is generally under low-light conditions. Given the information on hydrolysis and photolysis, these conditions are likely to minimize breakdown of natamycin.

Analytical methods are available that are suitable for analysis of natamycin.

The Meeting agreed that natamycin is a suitable marker for compliance with MRLs in citrus fruits and mushrooms. Furthermore, the Meeting agreed that the residue definition for assessing dietary risk from these commodities is natamycin.

Definition of the residue for plant commodities and fungi (for compliance with the MRL and for dietary risk assessment): *Natamycin*

[2] Allison, W. H. and Kneebone, L. R., 1963, *Influence of Compost pH and Casing Soil pH on Mushroom Production*, International Society for Mushroom Science, Volume 5, Part 1.

Results of supervised residue trials on crops

The Meeting received supervised trial data reflecting application of natamycin to growing mushrooms and post-harvest application to citrus fruits and pineapple.

Labels for end-use products containing natamycin were available from Canada describing the registered use on mushrooms and from the United States of America describing the registered uses on citrus fruits, pineapple, and mushrooms.

For all trials, residues were determined by the methods referenced above (methanol extraction, LC-MS/MS analysis). Analyses were completed within ca. one month for pineapple and mushrooms and within ca. 3 months for citrus. The available storage stability data support the storage durations and conditions from the residue studies.

Citrus fruits

Natamycin is registered in the US for post-harvest use on citrus (including calamondin, citrus citron, grapefruit, kumquat, lemon, lime, mandarin, oranges, and pummelo). The USA GAP is for a single application via in-line dip, drench, or aqueous or fruit-coating spray at a concentration of 0.99 g ai/L. The label does not specify a holding period following treatment; therefore, the Meeting assumed that treated fruits could enter commerce on the day of treatment.

Three supervised trials were conducted according to the USA GAP on each of grapefruit, lemon, and orange, with application made by dipping into an aqueous treatment solution. Residues zero days after application were:

Grapefruit (n=3): 0.81, 1.0 (2) mg/kg,

Lemon (n=3): 1.5, 1.7, 1.8 mg/kg, and

Orange (including mandarin; n=3): 1.3, 1.8, and 2.3 mg/kg.

Based on the similarity of the residue levels, the Meeting decided to combine the data for making residue estimates (n=9):0.81, 1.0 (2), 1.3, 1.5, 1.7, 1.8, 1.9, 2.3 mg/kg.

The Meeting estimated a maximum residue level for residues of natamycin in citrus fruits of 5 (Po) mg/kg

Residues of natamycin in citrus flesh from that same study were (n=9): 0.019, 0.031, 0.058, 0.060, 0.064, 0.076, 0.0800, 0.096, and 0.11 mg/kg.

The Meeting estimated an STMR of 0.064 mg/kg and an HR of 0.11 mg/kg.

Assorted tropical and sub-tropical fruit – inedible peel

Pineapple

Natamycin is registered in the USA for post-harvest use on pineapple. The US GAP is for a single application via dip, pour, or cascade in wax at a concentration of 11 g ai/L. The label specifies applying natamycin to the peduncle after the initial application has dried.

The label does not specify a holding period following treatment; therefore, the Meeting assumed that treated pineapples could enter commerce on the day of treatment. Only one independent trial reflected GAP and reporting residues on the day of treatment was provided. The residue was:

Pineapple (n=1): 0.86 mg/kg.

The Meeting determined that there are insufficient data to estimate a maximum residue level for pineapple.

Edible fungi

Mushrooms

Natamycin is registered in Canada and the USA for use on cultivated mushrooms. The GAP is for surface drench applications at 2.2 kg ai/ha not less than six hours before harvest. The Canadian label allows two applications (at casing and at pinning) whereas the US label allows four applications (after casing and before flushing, after flushing, and any time between breaks).

Only one independent trial reflected the critical GAP and reported residues six hours after treatment was provided. The residue was:

Mushrooms (n=1): 4.7 mg/kg.

The Meeting determined that there are insufficient data to make a recommendation for mushrooms.

RECOMMENDATIONS

Based on the data from supervised trials, the Meeting concluded that the residue levels reported are suitable for establishing maximum residue limits. However, the JMPR couldn't complete the dietary risk assessment as no ADI was set.

Definition of the residue for plant commodities and fungi (for compliance with the MRL and for estimation of dietary intake): *Natamycin.*

DIETARY RISK ASSESSMENT

Long-term dietary exposure

The International Estimated Daily Intakes (IEDIs) of natamycin were calculated for the 17 GEMS/Food cluster diets using STMRs/STMR-Ps estimated by the current Meeting. The calculated IEDIs were up to 0.56 µg/kg bw/day. An ADI was not established.

Short-term dietary exposure

The International Estimated Short-Term Intakes (IESTIs) of natamycin were calculated using HRs/HR-Ps estimated by the current Meeting. The calculated IESTIs were up to 6.4 µg/kg bw. An ARfD was not established.

REFERENCES

Report No.	Author	Year	Title
1869W	Marin, J.E.	2008	Development and Validation of an Analytical Method for the Determination of Natamycin in Mushrooms and Mushroom and Mushroom Compost, Casing and Casing plus Inoculum
1915W	Marin, J.E.	2009	Magnitude of the Residue of Natamycin in Mushrooms
197SRUS12R-1	Devine, J.M., Cenni, M.	2013	Magnitude of Residues of Natamycin in Post-Harvest Treated Pineapples (Decline, Processing)
197SRUS12R-1 (Attachment)	Habeeb, S.B.	2013	Magnitude of Residues of Natamycin in Post-Harvest Treated Pineapples (Decline, Processing)
060109	Smilanick, J., Kim, R., Fassel, B., Corey, D., and Smith, M.	Unkn	Effect of Dose Rates and Natamycin Mixture with Postharvest Fungicides on the Control of Postharvest Disesases in Citrus
034208-1	Feguson, L.	2016	Independent Laboratory Validation (ILV) of Environmental Chemistry Methods for Determination of Natanycin in Citrus Crops
034209-1	Jutson, J.	2016	Storage Stability of Natamycin in Citrus, Citrus Peel, and Citrus

Report No.	Author	Year	Title
			without Peel (Flesh)
029035	Cassidy, P.	2016	Natamycin Residue Analysis in Mushrooms, Compost and Casing
PSM-15-03-01	Schreier, T.	2016	Magnitude of the Residues of Natamycin in/on Citrus Following Treatment with Zivion M