5.18 MEPTYLDINOCAP (244)

TOXICOLOGY

Meptyldinocap is the International Organization for Standardization (ISO)–approved name for 2-(1-methylheptyl)-4,6-dinitrophenyl crotonate (International Union of Pure and Applied Chemistry [IUPAC]), with Chemical Abstracts Service (CAS) No. 131-72-6. Meptyldinocap is a new dinitrophenolic fungicidal compound, which acts by uncoupling mitochondrial oxidative phosphorylation. Meptyldinocap was reviewed for the first time by the Joint FAO/WHO Meeting on Pesticide Residues (JMPR) on the request of the Codex Committee on Pesticide Residues (CCPR).

Meptyldinocap is one of the six structural analogues present in the existing active substance dinocap. Dinocap was evaluated previously by the JMPR in 1969, 1974, 1989, 1998 and 2000. In 1998, the acceptable daily intake (ADI) and the acute reference dose (ARfD) for dinocap were established at 0–0.008 mg/kg body weight (bw) and 0.008 mg/kg bw, respectively. In 2000, two ARfDs were established for dinocap, one for women of childbearing age, at 0.008 mg/kg bw, and another for the general population, at 0.03 mg/kg bw. Dinocap contains approximately 22% meptyldinocap.

The database supporting meptyldinocap consists of some new studies performed with meptyldinocap together with earlier studies performed with dinocap. Previously evaluated studies with dinocap were reviewed at the Meeting but are summarized only briefly. Most of the pivotal studies met the basic requirements of the relevant Organisation for Economic Co-operation and Development (OECD) or national test guidelines, although the level of detail in some of the older reports of studies performed with dinocap did not always meet current requirements. A number of studies using dinocap did not contain certificates of compliance with good laboratory practice (GLP).

Biochemical aspects

No new absorption, distribution, metabolism and excretion (ADME) studies on meptyldinocap in mammals have been conducted. However, in a number of the ADME studies with dinocap, the radiolabel was present on the methylheptyl analogue, which is the primary component of meptyldinocap. The Meeting considered that these ADME studies were applicable to meptyldinocap.

Meptyldinocap is relatively well absorbed, with approximately 60–70% of the radiolabel absorbed in rabbits. Absorption is rapid, with peak plasma radioactivity seen 1–6 h after oral administration. Radiolabel was widely distributed, with tissue levels generally low and below those in blood. The compound did not tend to concentrate in any particular organ or tissue; highest levels were found in the liver, kidneys and skin. Metabolism was extensive, consisting of hydrolytic cleavage to release the crotonate moiety and subsequent oxidation of the methylheptyl chain. The basic metabolic pathways are similar in rats and mice. Excretion of radiolabel was extensive via urine (39–58% in mice; 31–50% in rats) and faeces and mainly occurred within 48 h.

Toxicological data

Meptyldinocap is of low acute toxicity when administered orally or dermally (median lethal dose [LD_{50}] > 2000 mg/kg bw) but is of moderate toxicity by inhalation (median lethal concentration [LC_{50}] 1.2 mg/L). Meptyldinocap is a slight irritant to skin and a moderate irritant to the eye; it has been found to produce skin sensitization in a local lymph node assay in mice.

Short-term studies of toxicity with meptyldinocap were performed in mice, rats and dogs. Yellow urine staining was a consistent finding, but this is considered not to be adverse, as it is associated with the excretion of coloured metabolites of meptyldinocap. In a 28-day dietary study of meptyldinocap in mice, there were increases in liver weight (approximately 10–15%) at 750 ppm (equal to 126 mg/kg bw per day), with a NOAEL of 200 ppm (equal to 33 mg/kg bw per day). In a

90-day dietary study of meptyldinocap in rats, altered clinical chemistry parameters and mononuclear cell infiltration of the lacrimal glands were seen at 2000 ppm (equal to 122 mg/kg bw per day), with a NOAEL of 650 ppm (equal to 40 mg/kg bw per day).

In a 90-day dietary study, groups of dogs were exposed to meptyldinocap at 0, 15, 60 or 120 ppm, with a positive control group receiving 60 ppm dinocap. Reduced body weight gain was seen in males after the first week of dosing with 120 ppm (equal to 3.9 mg/kg bw per day), following a gradual introduction to the treated diets. These initial body weight effects showed no consistency between animals or with food consumption patterns. The body weight gain over 90 days was 41% lower in males receiving 120 ppm than in controls. Ocular changes seen in the dinocap-exposed group were not evident in the meptyldinocap-treated groups. The NOAEL for meptyldinocap was 60 ppm (equal to 1.6 mg/kg bw per day), based on the effects on body weight gain over the duration of the study. In an extension to this 90-day study, a satellite group was exposed to meptyldinocap for 1 year at 120 ppm (equal to 3.5 mg/kg bw per day). The examinations were limited to tibial nerves, eyes and heart. This segment of the study showed that there were no significant eye, heart or nerve lesions evident after exposure to meptyldinocap for 1 year. The reduced body weight gain seen in the 90-day phase was not evident over the extended dosing period. The 1-year study was not designed to permit the identification of a NOAEL.

No evidence of carcinogenicity was seen in long-term studies of toxicity and carcinogenicity with dinocap at the highest doses tested, 150 ppm (equal to 23 mg/kg bw per day) in mice and 2000 ppm (equal to 71 mg/kg bw per day) in rats. The NOAEL for general toxicity in the chronic study in mice with dinocap was 15 ppm (equivalent to 2.8 mg/kg bw per day), based on body weight deficits in females. In the 30-month study of dinocap in rats, there was a significant increase in survival in both sexes at the top dose of 2000 ppm (equal to 71 mg/kg bw per day), which had an impact on the incidences of a number of age-related changes. The NOAEL for general toxicity of dinocap was 200 ppm (equal to 6.4 mg/kg bw per day).

The potential genotoxicity of meptyldinocap has been investigated in an adequate range of tests in vitro and in vivo. No evidence of mutagenicity or clastogenicity was noted.

The Meeting concluded that meptyldinocap is unlikely to be genotoxic.

The Meeting concluded that dinocap is not carcinogenic and that this conclusion could be extrapolated to meptyldinocap.

No effects on fertility, reproductive parameters, sperm or reproductive tissues were seen in a two-generation dietary study with dinocap at doses up to 400 ppm (equal to 27 mg/kg bw per day), the highest dose tested over both generations. At 1000 ppm (equal to 65 mg/kg bw per day), reduced pup survival in the first generation led to a reduction in the dose level to 400 ppm, which was without effect on pups in the second generation. The NOAEL for pup development and parental toxicity was 200 ppm (equal to 13 mg/kg bw per day).

In a developmental toxicity study in mice investigating effects on the palate and inner ear, meptyldinocap did not produce any such effects on fetuses at the highest dose of 500 mg/kg bw per day, whereas a dinocap dose of 25 mg/kg bw per day produced cleft palate in nearly all fetuses and had marked effects on otoconia formation. Additional studies showed that the teratogenicity of dinocap in mice was associated with the 4-propylpentyl analogue and not the methylheptyl analogue present in meptyldinocap. In a developmental study in rats, marked maternal toxicity and marked reductions (approximately 50%) in food consumption were seen with a meptyldinocap dose of 500 mg/kg bw per day, such that this dose level had to be terminated by gestation day 11. At the next highest dose level of 150 mg/kg bw per day, there were reductions from gestation days 6 to 9 in food consumption (approximately 18 g per rat) and maternal body weight gain (approximately 14 g per rat) at the start of the dosing period. The body weight deficit and increased absolute liver weights (23%) were evident at the end of the study, but there were no indications of fetotoxicity. The NOAEL for maternal toxicity was 50 mg/kg bw per day, with the NOAEL for fetotoxicity being 150 mg/kg bw per day. In a rabbit developmental toxicity study with meptyldinocap, maternal body weight loss was

seen in several dams before dosing commenced and in half the dams exposed at 48 mg/kg bw per day early in the dosing period. From gestation days 7 to 10, dams in the top dose group had a mean deficit of 44 g in body weight gain relative to controls; although this was similar in magnitude to the decrease in food consumption over the same period, there was no apparent consistency in individual body weight gains or food consumption. Over the remainder of the study, the body weight gain in the top dose group was similar to that in controls, and from gestation days 20 to 28 (during which dosing continued), the body weight gain was greater than that in controls. There were no effects on the fetuses at any dose level. The maternal NOAEL was 12 mg/kg bw per day, and the NOAEL for fetotoxicity was 48 mg/kg bw per day.

The Meeting concluded that meptyldinocap did not induce developmental toxicity and that it was not teratogenic.

There are no specific neurotoxicity studies on meptyldinocap, but there were no indications of neurotoxicity in routine studies, including a 90-day study in rats that included a functional observational battery.

No information on medical surveillance or poisoning incidents was available.

The Meeting concluded that the existing database on dinocap and meptyldinocap was adequate to characterize the potential hazards of meptyldinocap to fetuses, infants and children.

Toxicological evaluation

The Meeting established an ADI of 0–0.02 mg/kg bw on the basis of the NOAEL of 1.6 mg/kg bw per day in the 90-day dietary study in dogs, based on reduced body weight gain in males at 3.9 mg/kg bw per day. A safety factor of 100 was applied.

The Meeting concluded that an ARfD was unnecessary, as there were no effects that could be attributed to a single exposure. Meptyldinocap did not produce neurotoxicity, fetotoxicity or reproductive effects and has an oral LD_{50} of greater than 2000 mg/kg bw. The Meeting reviewed in depth the reduced body weight gains and food consumption seen in the early stages of the 90-day study in dogs and the developmental toxicity studies in rats and rabbits. In the rat developmental toxicity study, the body weight deficits were considered secondary to reduced food consumption, which was probably associated with palatability issues. The Meeting concluded that the body weight and food consumption patterns seen in the early stages of the dog and rabbit studies were not consistent between individual animals. The findings in these three studies did not provide an appropriate basis for establishing an ARfD for meptyldinocap.

A toxicological monograph was prepared.

Levels relevant to risk assessment

Species	Study	Effect	NOAEL	LOAEL
Mouse	Twenty-eight-day study of toxicity with meptyldinocap[a]	Toxicity	200 ppm, equal to 33 mg/kg bw per day	750 ppm, equal to 126 mg/kg bw per day
	Seventy-eight-week study of toxicity and carcinogenicity with dinocap[a]	Toxicity	15 ppm, equal to 2.8 mg/kg bw per day	100 ppm, equal to 18 mg/kg bw per day
		Carcinogenicity	150 ppm, equal to 23 mg/kg bw per day[b]	—
Rat	Ninety-day study of toxicity with meptyldinocap[a]	Toxicity	650 ppm, equal to 40 mg/kg bw per day	2000 ppm, equal to 122 mg/kg bw per day
	Thirty-month study of toxicity and carcinogenicity	Toxicity	200 ppm, equal to 6.4 mg/kg bw per day	2000 ppm, equal to 71 mg/kg bw per day

Species	Study	Effect	NOAEL	LOAEL
	with dinocap[a]	Carcinogenicity	2000 ppm, equal to 71 mg/kg bw per day[b]	—
	Multigeneration study of reproductive toxicity with dinocap[a]	Reproductive toxicity	400 ppm, equal to 27 mg/kg bw per day[b]	—
		Parental toxicity	200 ppm, equal to 13 mg/kg bw per day	400 ppm, equal to 27 mg/kg bw per day
		Offspring toxicity	200 ppm, equal to 13 mg/kg bw per day	400 ppm, equal to 27 mg/kg bw per day
	Developmental toxicity study with meptyldinocap[c]	Maternal toxicity	50 mg/kg bw per day	150 mg/kg bw per day
		Embryo and fetal toxicity	150 mg/kg bw per day[b]	—
Rabbit	Developmental toxicity study with meptyldinocap[c]	Maternal toxicity	12 mg/kg bw per day	48 mg/kg bw per day
		Embryo and fetal toxicity	48 mg/kg bw per day[b]	—
Dog	Ninety-day study of toxicity with meptyldinocap[a]	Toxicity	60 ppm, equal to 1.6 mg/kg bw per day	120 ppm, equal to 3.9 mg/kg bw per day[b]

[a] Dietary administration.

[b] Highest dose tested.

[c] Gavage administration.

Estimate of acceptable daily intake for humans

0–0.02 mg/kg bw

Estimate of acute reference dose

Unnecessary

Information that would be useful for the continued evaluation of the compound

Results from epidemiological, occupational health and other such observational studies of human exposure

Critical end-points for setting guidance values for exposure to meptyldinocap

Absorption, distribution, excretion and metabolism in mammals	
Rate and extent of oral absorption	Rapid; moderately well absorbed (60–70%)
Distribution	Widely distributed
Potential for accumulation	None
Rate and extent of excretion	Relatively rapid
Metabolism in animals	Extensively metabolized, initially hydrolysis to remove the crotonate side-chain and then via oxidation of the methylheptyl chain
Toxicologically significant compounds (animals, plants and the environment)	Meptyldinocap
Acute toxicity	
Rat, LD_{50}, oral	> 2000 mg/kg bw

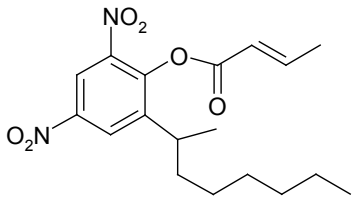
Rat, LD_{50}, dermal	> 5000 mg/kg bw
Rat, LC_{50}, inhalation	1.2 mg/L (4 h, nose only)
Rabbit, dermal irritation	Slight
Rabbit, ocular irritation	Moderate
Mouse, dermal sensitization	Sensitizer (local lymph node assay)
Short-term studies of toxicity	
Target/critical effect	Body weight gain (males)
Lowest relevant oral NOAEL	Dog: 1.6 mg/kg bw per day (meptyldinocap)
Lowest relevant dermal NOAEL	No data
Lowest relevant inhalation NOAEC	No data
Long-term studies of toxicity and carcinogenicity	
Target/critical effect	Body weight
Lowest relevant NOAEL	Mouse: 2.8 mg/kg bw per day (dinocap)
Carcinogenicity	Not carcinogenic in rats or mice
Genotoxicity	
	Not genotoxic in vitro or in vivo
Reproductive toxicity	
Reproduction target/critical effect	Pup survival
Lowest relevant reproductive NOAEL	Rat: 13 mg/kg bw per day (dinocap)
Developmental target/critical effect	None
Lowest relevant developmental NOAEL	Rabbit: 48 mg/kg bw per day (meptyldinocap)
Neurotoxicity/delayed neurotoxicity	
	No indications in routine studies
Other toxicological studies	
	No data
Medical data	
	No data

Summary

	Value	**Study**	**Safety factor**
ADI	0–0.02 mg/kg bw	Ninety-day study of toxicity in dogs	100
ARfD	Unnecessary		

RESIDUE AND ANALYTICAL ASPECTS

Meptyldinocap is a protectant and curative fungicide for the control of powdery mildew diseases. As a new compound it is evaluated at the first time by the JMPR. The meptyldinocap is the single isomer [2,4-dinitro-6-(1-methylheptyl)phenyl crotonate] of the existing active substance dinocap.



The 2,4-dinitro-6-(1-ethylhexyl)phenyl crotonate, present in the technical meptyldinocap in 1.5% concentration, is considered as impurity. The dinocap is a mixture of 2,4-dinitro-6-octylphenyl crotonates and 2,6-dinitro-4-octylphenyl crotonates. The 'octyl' being a mixture of 1-methylheptyl, 1-ethylhexyl and 1-propylpentyl groups. Approximately 22% of dinocap is meptyldinocap. Dinocap was last evaluated as new compound by the 1998 (R) and for some additional commodities by the 2001 Meetings of the JMPR. Presently both dinocap and meptyldinocap are marketed, but the manufacturers intend to gradually replace dinocap with meptyldinocap.

The manufacturer submitted information on metabolism in plants, analytical methods and residues in/on pome fruits, stone fruits, grapes, strawberries, cucurbits with edible and inedible peel which were evaluated by the present Meeting.

The studies evaluated by the present Meeting were conducted either with meptyldinocap or dinocap. The typical composition of the test substances are given below:

Isomers	Meptyldinocap	Dinocap
Meptyldinocap, 2,4-dinitro-6-(1-methylheptyl)phenyl crotonate	98.5 %	22 %
2,6-dinitro-4-(1-methylheptyl)phenyl crotonate	0 %	11 %
2,4-dinitro-6-(1-ethylhexyl)phenyl crotonate[1]	1.5 %	22 %
2,6-dinitro-4-(1-ethylhexyl)phenyl crotonate	0 %	11 %
2,4-dinitro-6-(1-propylpentyl)phenyl crotonate	0 %	22 %
2,6-dinitro-4-(1-propylpentyl)phenyl crotonate	0 %	11 %

Animal metabolism

The intended use for meptyldinocap is on vines, cucurbits and strawberries, which are not fed to animals. Therefore, no animal metabolism studies were provided for evaluation.

Farm animal metabolism studies evaluated by previous Meetings of the JMPR indicated that no radioactive residues were detectable in milk or tissues when lactating caws were fed with dinocap at 0.1, 0.3 and 1 ppm dose levels.

Plant metabolism

The plant metabolism studies on apples, cucumber and squash submitted to the current meeting had already been evaluated by the 1998 JMPR, and they were a re-evaluated by the present Meeting. The metabolism studies were carried out with meptyldinocap. A single major metabolite [2, 4-dinitro-6-(1-methylheptyl) phenol] was identified and it is referred to as 2,4-DNOP.

An apple tree was treated with a single foliar application of an EC formulation containing 45.6% ai at a rate equivalent to 1.96 kg ai/ha, four times the normal maximum application rate (0.49 kg ai/ha). Apple and leaf samples were taken on the day of application, both before and after treatment, and after 7, 14, and 21 days. Half of each fruit sample was analysed as whole fruit, and the other half peeled and the peel and pulp analysed separately.

The samples were extracted with methanol which recovered more than 90% of the radioactivity from the day 0 samples and 40–60 % from the aged samples. More than 92% of the radioactivity at each PHI was associated with the peel. The total radioactivity recovered in the neutral and alkaline methanolic extracts was more than 80% in all cases.

Two compounds present in the apple fruit have been identified by coelution with standards in both normal phase TLC and reversed phase HPLC. These are the parent meptyldinocap and its corresponding phenol, 2,4-DNOP. The parent compound was present in all of the treated samples, On the day of treatment, meptyldinocap was present at 2.12 mg/kg. After seven days the level had fallen to 0.52 mg/kg, representing 23% of the total radioactivity. After 14 days, the concentration of meptyldinocap had decreased to 0.25 mg/kg (11% of the total radioactivity), and after 21 days it was present at 0.12 mg/kg (8% of the total radioactivity). The half-life of meptyldinocap was calculated to be 5.2 days, The single major metabolite, 2,4-DNOP, was present at lower levels: it comprised roughly 24% of the total radioactivity in the aged samples (0.03–0.08 mg/kg, expressed as parent equivalent).

Five minor metabolites could be identified by gas chromatography with mass spectrometric detection: [2-methyl-5-nitro-7-(2-octyl) benzoxazole, 2-(hydroxymethyl)-5-nitro-7-(2-octyl) benzoxazole, 4-(1-propenyl)-5-nitro-7-(2-octyl)benzoxazole, 5-nitro-7-(octyl) benzoxazole and 2-hydroxymethyl-5-nitro-(2-octyl)-phenyl crotonamide]. The corresponding concentrations ranged from 0.001–0.007 mg/kg.

In cucumber, the distribution and rate of decrease of residues after a single treatment with ^{14}C-meptyldinocap at 0.56 kg ai/ha were studied. The residues of ^{14}C-meptyldinocap dissipated rapidly from the cucumber leaves and stems. The half lives of radioactive residues on leaves and stems were 11.8 days and 18.8 days, respectively. The ^{14}C residues on the leaves decreased from 38.2 mg/kg immediately after application to 1.4 mg/kg at final harvest. The ^{14}C residues in the stems decreased from 3.6 mg/kg immediately after application to 0.5 mg/kg at final harvest, 65 days after last application.

The whole mature fruit harvested 48 days after application contained ^{14}C residues of 0.16 mg/kg and whole mature fruit harvested 63 days after application contained 0.09 mg/kg. Of the whole mature fruit harvested 48 days after application, the peels contained 0.15 mg/kg and the flesh contained 0.11 mg/kg. The proportion of parent compound and 2, 4-DNOP was about the same in cucumber fruits at days 48 and 63.

Of the residues extracted from the leaves by acetone, only one metabolite (2,4-DNOP) occurred in significant (> 10%) quantity. The metabolism of ^{l4}C-meptyldinocap in cucumber leaves was extensive, leading to 18 minor metabolites. Only 2,4-DNOP could be identified amounting to 2.4% of TRR at day 8, while the unextractable residues accounted for 58% of TRR.

The half-life of the radioactivity was 8 days in the squash leaves treated with ^{14}C-meptyldinocap two times at a rate of 0.56 kg ai/ha. 2,4-DNOP was the main metabolite in the leaves and was also found in the fruit. About 6 unidentified metabolites were found in the fruit and 10 in the leaves, none of which individually accounted for more than 10% of the TRR. The 2,4-DNOP, meptyldinocap, organic soluble metabolites and water soluble polar metabolites and unextractable ^{14}C-residues amounted to 1.3%, 5.9%, 5.5%, 21% and 57.6% of TRR, respectively.

Photolysis, under natural daylight conditions, played a major role in the rapid dissipation of meptyldinocap from plant foliage. The concentration of meptyldinocap did not change on covered leaves over 27 hours following the foliar treatment with ^{14}C-meptyldinocap, while it decreased to 39% on leaves exposed to natural light. The extract of 27-hour uncovered leaves contained 53% polar photoproducts.

In summary, the metabolism of meptyldinocap is complex resulting in a large number of metabolites present at low concentrations. The metabolism of 2, 4- meptyldinocap appears to proceed by relatively rapid hydrolysis of the crotonate ester (half lives are about 5, 8 and 11.8 days on apple fruits, quash and cucumber leaves, respectively) to the corresponding phenol (2,4-DNOP). The

phenol is then further metabolised rapidly to a large number of more polar compounds, none of which is present in a significant amount. The proposed pathway for the formation of minor metabolites involves reduction of a nitro group to the amine. Metabolites are then formed by reaction of the amine with formic or acetic acid to form amides, or by intramolecular transfer of the crotonyl group to form the crotonamide. Ring closure of the amides then forms benzoxazoles. Individual metabolites could not be isolated. The amines could readily form conjugates with acids to form amides. Further degradation led to small carbon units which were subsequently incorporated into a number of natural products including cutin, lignin and other constituents that make up the acid detergent fibre.

Environmental fate in soil

Soil metabolism, degradation, leaching, rotational crop studies are not requires for compounds with intended use of foliar application only on permanent crops with no crops planted in rotation.

As part of the plant metabolism studies soil samples were also taken and analysed for meptyldinocap residues.

The ^{14}C residues in the top 2.5 cm of soil of cucumber plot decreased from 0.45 mg/kg immediately after application to 0.31 mg/kg after 63 days. The ^{14}C residues in the 2.5–7.6 cm soil depth never exceeded 0.02 mg/kg and the 7.6–15.2 cm soil depth residues never exceeded 0.007 mg/kg.

The ^{14}C residues in the top section of the soil (0–2.5 cm) of quash plot decreased from 0.43 mg/kg after the last treatment to 0.40 mg/kg 63 days later. The ^{14}C residues in the other soil sections were low.

The residues of [^{14}C]meptyldinocap in the soil in which the cucumber or squash were grown dissipate at a much slower rate than from the plants. There appeared to be no significant leaching of [^{14}C]-meptyldinocap residues into the lower depths of the soil.

Metabolism in rotational crops

The studies evaluated by the 1998 JMPR indicated that when beans, oats and turnips were grown in soil in which cucumber and squash were treated with [^{14}C]meptyldinocap 250 days earlier, the radioactive residues in samples taken until maturity of crops were at or below 0.02 mg/kg. Consequently, residues in follow up crops are unlikely to occur in measurable concentration.

Methods of analysis

The analytical methods used for determination of meptyldinocap residues in supervised trials were principally the same as those evaluated by the 1998 JMPR. Following the solvent extraction, the residues are converted to the corresponding phenols and determined by GC after methylation or analysed directly by HPLC-MS/MS. The validated limit of quantification for the meptyldinocap was 0.025 mg/kg and for combined residues 0.05 mg/kg. The average recoveries ranged from 80 to 104% with relative standard deviation of 7–14%. The concurrent recoveries obtained during the analysis of samples were in the same range.

The DFG S-19 multi residue method was found to be suitable for the determination of meptyldinocap residues in apples, barley grain, grapes and soya bean flour over in the concentration range from 0.05 mg/kg to 1.0 mg/kg with a validated limit of quantitation (LOQ) of 0.025 mg/kg for parent compound. The meptyldinocap peak was well separated from the dinocap isomers under the gas chromatographic conditions applied. The independent laboratory validation trials were conducted to satisfy the relevant requirements of the European Commission and the US EPA Guidelines.

Stability of residues in stored analytical samples

The stability of residues were tested in apples and grapes using 97.5% pure 2,4-dinitro-6-octylphenyl crotonates (2,4-DNOPC isomers) and 2,6-dinitro-4-octylphenyl crotonate isomer mixtures. The 'octyl' being a mixture of 1-methylheptyl, 1-ethylhexyl and 1-propylpentyl groups. In separate set of experiments the untreated samples were spiked at 1 mg/kg level. The overall mean procedural recoveries for 2,4-dinitro-6-octylphenyl crotonates in grapes and apples were 94.5% (RSD: 5.25%) and 88.6% (RSD: 10.3%) the average of residues remained over the period of 24 months were 89.8% (RSD: 10.13%) 71.5% (RSD: 10.5%), respectively. The results indicate that residues of the 2,4-DNOPC isomers are stable in apples, grapes, tomatoes, peaches and strawberries stored frozen up to 24 months. The stability of the meptyldinocap alone during deep-frozen storage could not be determined from these studies. However, it may be assumed to be similar to the other isomers.

Definition of the residue

Results of metabolism studies on fruits and fruiting vegetables indicate that the parent compound, meptyldinocap, forms the main residue remaining in the plant tissues at harvest. The major metabolite, the corresponding phenol, 2,4-DNOP, showed concentrations of 2–10% of total radioactivity only. The concentration of the major metabolite, the corresponding phenol (2, 4-DNOP) had not changed with time after application of the pesticide, indicating that that further metabolism to a number of minor compounds occurred relatively quickly. Initially the meptyldinocap amounted to the major portion of the TRR. The proportion of 2, 4-DNOP gradually increased with time and it was present in about the same concentration as meptyldinocap 48–63 days after the treatment of cucumber. The parent/2, 4-DNOP ratio was about 4 in apples 21 days after application.

The analytical method, which is used in the residue trials, determined meptyldinocap residues as a sum of the parent and the corresponding phenol. Multi residue methods, based on gas chromatographic and HPLC-MS/MS detection are available for the determination of meptyldinocap alone and have been validated for four representative commodities. Residues deriving from the use of dinocap could be identified based on the presence of 2,6-DNOP isomers provided that the chromatographic system used has sufficient resolution.

The current residue definition of dinocap is dinocap. As meptyldinocap is one isomer of dinocap, it is covered by the current residue definition. Non-selective methods cannot distinguish meptyldinocap from dinocap, but selective methods are available. While meptyldinocap and dinocap are both registered for crop uses, it is preferable, for enforcement purposes, to maintain a single residue definition.

It follows that, at least while dinocap MRLs are maintained, the residue definition for meptyldinocap as "dinocap, sum of all isomers" would be a practical solution.

The present Meeting established an ADI of 0–0.02 mg/kg/bw day. The new ADI is applicable for the sum of meptyldinocap and its corresponding phenol, when only they are present in the commodities analysed.

The Meeting recommended that while dinocap MRLs are maintained, the residue definition for meptyldinocap enforcement purposes should be dinocap, sum of all isomers.

Definition of the residue for dietary exposure assessment: the sum of meptyldinocap and the corresponding phenol, 2,4-DNOP, expressed as the parent meptyldinocap.

A residue definition for animal products is not required as no residue is expected to occur in animal products from the targeted use of meptyldinocap.

Results of supervised trials on crops

All trials were conducted according to GAP and the samples were analysed within the tested deep-frozen storage period. The methods applied for the analyses of samples determine meptyldinocap

residues as a sum of the parent and the corresponding phenol. The validity of the results was confirmed with concurrent recovery tests performed in the same analytical batch.

Cucumber and courgettes

GAP in France, Italy and Slovenia permits maximum 3 applications at 10 days with maximum dosage of 0.21 kg ai/ha and a PHI of 3 days. A total of eight supervised field trials on cucumbers/courgette were conducted according to maximum GAP in greenhouses located in the North and South European zones. Samples collected 3 days after the last application contained residues of < 0.005 (2), 0.01, 0.02 (4) and 0.04 mg/kg.

The Meeting estimated a maximum residue level, STMR value and HP value for Fruiting vegetables, Cucurbits, except melons of 0.07, 0.02 and 0.04 mg/kg.

Melons

GAP in France, Italy and Slovenia permits a maximum of 3 applications at 10 days with a maximum rate of 0.21 kg ai/ha and PHI of 3 days. A total of eight supervised field trials on melons were conducted according to maximum GAP in the North and South European zones. Whole fruit samples collected 3 days after the last application contained residues: < 0.005, 0.008, 0.02 (4), 0.05 and 0.28 mg/kg. No detectable residues were found in pulp samples.

The Meeting estimated a maximum residue level, STMR value and HP value for melons of 0.5, 0.005 and 0.28 mg/kg, respectively. Note: there is no information on pulp residues at high whole fruit residue.

The Meeting recommended to re-evaluate the current CXL of 0.05* for dinocap in fruiting vegetables cucurbits.

Grapes

GAP in France, Greece, Hungary and the UK permits a maximum of 4 applications with a maximum rate of 0.21 kg ai/ha and a PHI of 21 days. A total of eighteen trials were conducted on Grapes in Europe between 2005 and 2007. Eight trials with two formulations side by side containing meptyldinocap alone and the mixture of 2,4-DNPOC and 2,6-DOPOC (three isomers of each compound). In addition, eight trials were conducted in 2006 with a formulation containing meptyldinocap. All trials were performed with the permitted maximum application rate and frequency.

Samples collected at day 21 following the last application of meptyldinocap contained residues: < 0.01 (5), < 0.025 (6), 0.03 (3), 0.06 (2), 0.08, and 0.12 mg/kg.

The Meeting estimated a maximum residue level, STMR value and HP value for grapes 0.2, 0.025 and 0.12 mg/kg, respectively.

The Meeting noted that the current CXL of 0.5 mg/kg for dinocap in grapes covers the residues deriving from the use of meptyldinocap.

Strawberry

The GAP of France, Italy and Slovenia permits a maximum of 3 applications at 10 day intervals with a maximum rate of 0.21 kg ai/ha and a PHI of 3 days. A total of eight supervised field trials on strawberries were conducted according to the maximum GAP in greenhouses located in Northern and Southern Europe. The pesticide treatment was made with a formulation containing meptyldinocap.

Residues in samples collected 3 days after the final application of meptyldinocap, in ranked order, were: 0.03, 0.06, 0.07, 0.08, 0.09, 0.11, 0.12, 0.13 mg/kg.

The Meeting estimated a maximum residue level, STMR value and HP value for grapes 0.3, 0.085 and 0.13 mg/kg, respectively

The Meeting noted that the current CXL of 0.5 mg/kg for dinocap in strawberries covers the residues derived from the use of meptyldinocap.

The Meeting also noted that the current CXL for dinocap in strawberries included a note that it excludes the glasshouse use. The recommended maximum residue level for meptyldinocap is applicable for both uses.

Fate of residues during processing

Grapes

Dinocap was applied 6–8 times to both red and white grape varieties during the growing season at the recommended or 1.5× rate. Samples were collected for processing at intervals of 14–21 days after the final application. A portion of the collected grape bunches were subjected to vinification similar to commercial practice. The must from the white grape was divided into two equal portions: one of which was processed further without heating, the other was pasteurized for 2 minutes at approximately 85 °C.

The must of the white wine grapes, both pasteurized and non pasteurized, as well as the must of the red wine grapes was processed into wine following the same processing steps: fermentation; clarification (first racking and second racking); filtration, bottling and maturation. The residues were analysed with methods having LOQs of 0.04 and 0.05 mg/kg and an LOD of 0.01 mg/kg. The average recoveries in grapes, must and wine were in the 70–120% range.

Six grape samples taken 20–21 days or at shorter intervals after the last treatment did not contain residues above the LOQ. These trials could not be used for estimation of the processing factor. Other trials on red and white grapes resulted in measurable residues in grapes harvested 14–21 days after last pesticide treatment. The results of processing studies are summarised below:

PHI (days)	14	21	21	21[a]	21[a]
Grape	0.1	0.59	0.33	0.347	0.67
Must	< 0.04	< 0.05	< 0.01	< 0.05	< 0.05
Wine	< 0.04	< 0.01	< 0.01	< 0.01	< 0.01
Pf must/juice	< 0.4	< 0.085	< 0.030	< 0.144	< 0.075
Pf wine	< 0.4	< 0.017	< 0.030	< 0.029	< 0.015

[a] White grapes

The median processing factors for must and wine are < 0.08 and < 0.023 based on samples collected at the recommended PHI. The Meeting estimated STMR values of 0.0020 mg/kg and 0.00072 mg/kg for must and wine, respectively.

Raisins were prepared from the harvested grapes in two trials. However, the results are contradictory (the calculated processing factors were 2.26 and 0.417) and a processing factor could not be calculated.

Strawberry

Strawberry samples, taken 3 days after last pesticide treatment with dinocap, were processed to jam and preserve with a procedure resembling commercial practice. The residues measured in RAC and processed products are summarised below.

Dosage kg ai/ha & appl. No	0.4–0.41 × 6	0.39–0.42 × 6	0.21–0.22 × 3	0.20–0.21 ×3
Strawberry fruits (unwashed)	0.23	0.31	0.07	0.13
Jam	0.079	0.07	< 0.01	0.06
Preserve	< 0.05	0.11	< 0.01	0.11
Pf for jam	0.34	0.23	< 0.14	0.46
Pf for preserve	< 0.22	0.35	< 0.14	0.85

The calculated median processing factor for both jam and preserve is 0.285. The Meeting estimated an STMR value of 0.024 for strawberry jam and preserve.

Residues in animal commodities

Animal metabolism studies performed with dinocap evaluated by previous Meetings of the JMPR revealed that no radioactive residues were detectable in milk or tissues at any dose level (0.1-1 ppm). Consequently animal feeding studies are not required.

DIETARY RISK ASSESSMENT

Long-term intake

The evaluation of meptyldinocap resulted in recommendations for MRLs and STMR values for raw and processed commodities. Where data on consumption were available for the listed food commodities, dietary intakes were calculated for the 13 GEMS/Food Consumption Cluster Diets. The results are shown in Annex 3.

The IEDIs in the thirteen Cluster Diets, based on estimated STMRs were 0 % of the maximum ADI (0.02 mg/kg bw). The Meeting concluded that the long-term intake of residues of difenoconazole from uses that have been considered by the JMPR is unlikely to present a public health concern.

Short-term intake

As the establishment of an ARfD was previously considered unnecessary, the Meeting concluded that the short-term intake of meptyldinocap residues is unlikely to present a public health concern.