5.37 TRINEXAPAC-ETHYL (271)

TOXICOLOGY

Trinexapac-ethyl is the ISO-approved common name for 4-(cyclopropyl-α-hydroxymethylene)-3,5-dioxo-cyclohexanecarboxylic acid ethyl ester (IUPAC), with CAS No. 95266-40-3. Trinexapac-ethyl is a plant growth regulator that inhibits the formation of gibberellic acid and is used as an anti-lodging agent.

Trinexapac-ethyl has not previously been evaluated by JMPR and was reviewed by the present Meeting at the request of CCPR.

All critical studies contained statements of compliance with GLP.

Biochemical aspects

In studies conducted in rats using [^{14}C]trinexapac-ethyl, the time to reach the maximum plasma and tissue concentration of radioactivity was 15 minutes following a single gavage dose of 1 or 200 mg/kg bw. Gastrointestinal absorption was at least 96%. The plasma elimination half-life of radioactivity was less than 1 hour. Radioactivity was rapidly eliminated from tissues; mean first-phase tissue half-lives ranged from 0.2 to 0.5 hour at 1 mg/kg bw and from 0.5 to 0.9 hour at 200 mg/kg bw, whereas the slower second-phase elimination ranged from 1.6 to 3.2 hours at 1 mg/kg bw and from 3.2 to 11.7 hours at 200 mg/kg bw. There was no evidence of accumulation of radioactivity in any tissue. Excretion of radioactivity was predominantly via the urine (≥ 90% of the administered dose), with the majority of this occurring within 24 hours of dosing. Low levels of radioactivity were detected in faeces and bile (up to approximately 2.4% and 3.3% of the administered dose, respectively). Trinexapac-ethyl undergoes limited metabolism in the rat, involving predominantly ester hydrolysis of trinexapac-ethyl to trinexapac acid. The predominant urinary metabolite was trinexapac acid (up to 100% of total urinary radioactivity), with low levels of a conjugated derivative of trinexapac acid detected only in the urine of bile duct–cannulated rats (6.3% of the administered dose). In faeces, the parent compound accounted for 5–22% of total faecal radioactivity (1–2.5% of the administered dose), with the balance comprising trinexapac acid. Bile contained mainly a conjugated derivative of trinexapac acid (2.9% of the administered dose), with low levels of the parent compound also detected (0.2% of the administered dose).

Toxicological data

The oral LD_{50} in rats was greater than 2000 mg/kg bw. In rats, the dermal LD_{50} was greater than 4000 mg/kg bw, and the LC_{50} was greater than 5.3 mg/L. Trinexapac-ethyl was neither a skin nor an eye irritant in rabbits. In a guinea-pig maximization test, no skin sensitization occurred.

In repeated-dose toxicity studies in rats and dogs, the main target organ was the kidneys. In rats, increased kidney weight and accompanying histopathological changes (focal tubular basophilia, tubular hyaline droplets and pigment depositon) occurred. Additional treatment-related effects in the brain were observed in dog studies.

In a 4-week gavage study in rats, which tested doses of 0, 10, 100 and 1000 mg/kg bw per day, the NOAEL was 100 mg/kg bw per day for effects on the liver and kidneys at 1000 mg/kg bw per day.

In a 13-week dietary toxicity study in rats, which tested concentrations of 0, 50, 500, 5000 and 20 000 ppm (equal to 0, 3, 34, 346 and 1350 mg/kg bw per day for males and 0, 4, 38, 395 and 1551 mg/kg bw per day for females, respectively), the NOAEL was 500 ppm (equal to 34 mg/kg bw per day) for histopathological findings in the kidney in males at 5000 ppm (equal to 346 mg/kg bw per day).

In a 7-week, non-guideline study in dogs that tested dietary concentrations of 0, 500, 5000, 15 000, 30 000 and 50 000 ppm (equal to an average of 0, 23, 217, 683, 734 and 965 mg/kg bw per day in both sexes, respectively), the NOAEL was 15 000 ppm (equal to 683 mg/kg bw per day), based on a range of effects that occurred at 30 000 ppm (equal to 734 mg/kg bw per day), including body weight loss, lower feed consumption, increased serum cholesterol, increased kidney weight and histopathological findings in the kidney.

In a 13-week study in dogs, which tested dietary concentrations of 0, 50, 1000, 15 000 and 30 000 ppm (equal to 0, 2, 35, 516 and 927 mg/kg bw per day for males and 0, 2, 40, 582 and 891 mg/kg bw per day for females, respectively), the NOAEL was 15 000 ppm (equal to 516 mg/kg bw per day) for reduced body weight gain and feed consumption at 30 000 ppm (equal to 927 mg/kg bw per day). Reduced blood glucose and focal vacuolation in the brain occurred in one male dog at 30 000 ppm.

In a 52-week toxicity study in dogs, which tested dietary concentrations of 0, 40, 1000, 10 000 and 20 000 ppm (equal to 0, 1.6, 32, 366 and 727 mg/kg bw per day for males and 0, 1.4, 40, 357 and 793 mg/kg bw per day for females, respectively), the NOAEL was 1000 ppm (equal to 32 mg/kg bw per day) for cerebral vacuolation at 10 000 ppm (equal to 357 mg/kg bw per day) in the absence of neurodegenerative or inflammatory histopathological changes or neurological signs.

In a 78-week study in mice, which tested dietary concentrations of 0, 7, 70, 1000, 3500 and 7000 ppm (equal to 0, 0.9, 9, 131, 451 and 912 mg/kg bw per day for males and 0, 1.1, 11, 154, 539 and 1073 mg/kg bw per day for females, respectively), the NOAEL for chronic toxicity and carcinogenicity was 7000 ppm (equal to 912 mg/kg bw per day), the highest tested dietary concentration.

In a 104-week study in rats, which tested dietary concentrations of 0, 10, 100, 3000, 10 000 and 20 000 ppm (equal to 0, 0.4, 4, 116, 393 and 806 mg/kg bw per day for males and 0, 0.5, 5, 147, 494 and 1054 mg/kg bw per day for females, respectively), the NOAEL for chronic toxicity was 3000 ppm (equal to 116 mg/kg bw per day) for histopathological lesions in the kidneys at 10 000 ppm (equal to 393 mg/kg bw per day). The NOAEL for carcinogenicity was 20 000 ppm (equal to 806 mg/kg bw per day), the highest tested dietary concentration.

The Meeting concluded that trinexapac-ethyl is not carcinogenic in mice or rats.

Trinexapac-ethyl was tested in an adequate range of in vitro and in vivo genotoxicity tests. No evidence of genotoxicity was found.

The Meeting concluded that trinexapac-ethyl is unlikely to be genotoxic.

Given the lack of genotoxicity and the absence of carcinogenicity in mice and rats, the Meeting concluded that trinexapac-ethyl is unlikely to pose a carcinogenic risk to humans.

In a two-generation reproductive toxicity study in rats, which tested dietary concentrations of 0, 10, 1000, 10 000 and 20 000 ppm (equal to 0, 0.57, 58.6, 570.5 and 1166 mg/kg bw per day for males and 0, 0.73, 73, 721.5 and 1427 mg/kg bw per day for females, respectively), there was no evidence of reproductive toxicity up to the highest tested dietary concentration of 20 000 ppm (equal to 1166 mg/kg bw per day). The NOAEL for parental toxicity was 1000 ppm (equal to 58.6 mg/kg bw per day) for reduced body weight gain and feed consumption at 10 000 ppm (equal to 570.5 mg/kg bw per day). The NOAEL for offspring toxicity was 10 000 ppm (equal to 570.5 mg/kg bw per day) for reduced survival and body weight in the F_1 and F_2 generations at 20 000 ppm (equal to 1166 mg/kg bw per day).

In a rat developmental toxicity study, which tested doses of 0, 20, 200 and 1000 mg/kg bw per day, the NOAEL for maternal and embryo/fetal toxicity was 1000 mg/kg bw per day, the highest dose tested.

In a rabbit developmental toxicity study, which tested doses of 0, 10, 60 and 360 mg/kg bw per day, the NOAEL for maternal toxicity was 60 mg/kg bw per day for deaths of several dams at

360 mg/kg bw per day. The NOAEL for embryo and fetal toxicity was also 60 mg/kg bw per day for increased post-implantation losses and a reduction in mean number of live fetuses at 360 mg/kg bw per day.

The Meeting concluded that trinexapac-ethyl is not teratogenic in rats or rabbits.

In an acute neurotoxicity study in rats that tested doses of 0, 500, 1000 and 2000 mg/kg bw per day, the NOAEL was 2000 mg/kg bw, the highest dose tested.

In a subchronic neurotoxicity study in rats that tested dietary concentrations of 0, 3750, 7500 and 15 000 ppm (equal to 0, 233, 464 and 948 mg/kg bw per day for males and 0, 294, 588 and 1171 mg/kg bw per day for females, respectively), the NOAEL was 15 000 ppm (equal to 948 mg/kg bw per day), the highest tested dietary concentration.

Toxicological data on metabolites and/or degradates

The Meeting noted the formation of two processing degradates of trinexapac acid, CGA 113745 and CGA 313458, not detected in rat metabolism studies. Based on a structural assessment of these degradates and an estimate of the levels of chronic dietary intake, the Meeting concluded that they are unlikely to pose a dietary risk.

Human data

There were no reports submitted on adverse health effects in workers involved in the manufacture or use of trinexapac-ethyl. No cases of human poisonings have been reported.

The Meeting concluded that the database on trinexapac-ethyl was adequate to characterize the potential hazards to fetuses, infants and children.

Toxicological evaluation

The Meeting established an ADI of 0–0.3 mg/kg bw per day, expressed as trinexapac acid equivalents[1], based on a NOAEL of 32 mg/kg bw per day for trinexapac-ethyl (equivalent to 29 mg/kg bw per day expressed as trinexapac acid equivalents) for cerebral vacuolation in male and female dogs following 52 weeks of dietary exposure, with the application of a 100-fold safety factor. In the absence of information to the contrary, including mechanistic data, the cerebral vacuolation observed in dogs was considered relevant to humans.

The Meeting concluded that it is not necessary to establish an ARfD for trinexapac-ethyl in view of its low acute oral toxicity and the absence of developmental toxicity or any other toxicological effects that would be likely to be elicited by a single dose.

A toxicological monograph was prepared.

[1] To cover the possible dietary exposure to a range of salts, esters and conjugates of trinexapac, it is appropriate to express the ADI as trinexapac acid equivalents using a conversion factor of 0.9 based on differences in molecular weight between trinexapac-ethyl and trinexapac acid.

Levels relevant to risk assessment of trinexapac-ethyl

Species	Study	Effect	NOAEL	LOAEL
Mouse	Two-year study of toxicity and carcinogenicity[a]	Toxicity	7000 ppm, equal to 912 mg/kg bw per day[b]	—
		Carcinogenicity	7000 ppm, equal to 912 mg/kg bw per day[b]	—
Rat	Thirteen-week study of toxicity[a]	Toxicity	500 ppm, equal to 34 mg/kg bw per day	5000 ppm, equal to 346 mg/kg bw per day
	Two-year study of toxicity and carcinogenicity[a]	Toxicity	3000 ppm, equal to 116 mg/kg bw per day	10 000, equal to 393 mg/kg bw per day
		Carcinogenicity	30 000 ppm, equal to 806 mg/kg bw per day[b]	—
	Two-generation study of reproductive toxicity[a]	Reproductive toxicity	20 000 ppm, equal to 1166 mg/kg bw per day[b]	—
		Parental toxicity	1000 ppm, equal to 59 mg/kg bw per day	10 000 ppm, equal to 571 mg/kg bw per day
		Offspring toxicity	10 000 ppm, equal to 571 mg/kg bw per day	20 000 ppm, equal to 1166 mg/kg bw per day
	Developmental toxicity study[c]	Maternal toxicity	1000 mg/kg bw per day[b]	—
		Embryo and fetal toxicity	1000 mg/kg bw per day[b]	—
Rabbit	Developmental toxicity study[c]	Maternal toxicity	60 mg/kg bw per day	360 mg/kg bw per day
		Embryo and fetal toxicity	60 mg/kg bw per day	360 mg/kg bw per day
Dog	One-year study of toxicity[a]	Toxicity	1000 ppm, equal to 32 mg/kg bw per day	10 000 ppm, equal to 357 mg/kg bw per day

[a] Dietary administration.

[b] Highest dose tested.

[c] Gavage administration.

Estimate of acceptable daily intake

0–0.3 mg/kg bw per day

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 trinexapac-ethyl

Absorption, distribution, excretion and metabolism in mammals	
Rate and extent of oral absorption	Rapid and complete
Distribution	Widespread tissue distribution
Potential for accumulation	No potential for accumulation
Rate and extent of excretion	Rapid and complete
Metabolism in animals	Limited; mainly hydrolysis to trinexapac acid
Toxicologically significant compounds in animals, plants and the environment	Trinexapac-ethyl, trinexapac acid
Acute toxicity	
Rat, LD_{50}, oral	> 2000 mg/kg bw
Rat, LD_{50}, dermal	> 4000 mg/kg bw
Rat, LC_{50}, inhalation	> 5.3 mg/L
Rabbit, dermal irritation	Non-irritating
Rabbit, ocular irritation	Non-irritating
Dermal sensitization	Non-sensitizing (maximization test)
Short-term studies of toxicity	
Target/critical effect	Kidney and brain
Lowest relevant oral NOAEL	32 mg/kg bw per day (dog)
Lowest relevant dermal NOAEL	1000 mg/kg bw per day (rat)
Lowest relevant inhalation NOAEC	No data
Long-term studies of toxicity and carcinogenicity	
Target/critical effect	Kidney
Lowest relevant NOAEL	116 mg/kg bw per day (rat)
Carcinogenicity	Not carcinogenic
Genotoxicity	
	Not genotoxic
Reproductive toxicity	
Reproduction target/critical effect	No evidence of reproductive toxicity
Lowest relevant parental NOAEL	59 mg/kg bw per day
Lowest relevant offspring NOAEL	571 mg/kg bw per day
Lowest relevant reproduction NOAEL	1166 mg/kg bw per day, the highest dose tested
Developmental toxicity	

Developmental target/critical effect	Post-implantation losses at maternally toxic doses (rabbit)
Lowest maternal NOAEL	60 mg/kg bw per day (rabbit)
Lowest embryo/fetal NOAEL	60 mg/kg bw per day (rabbit)
Neurotoxicity	
Acute and subchronic neurotoxicity	Not neurotoxic
Medical data	
	No data

Summary

	Value	Studies	Safety factor
ADI	0–0.3 mg/kg bw	One-year toxicity study in dogs	100
ARfD	Unnecessary	—	—

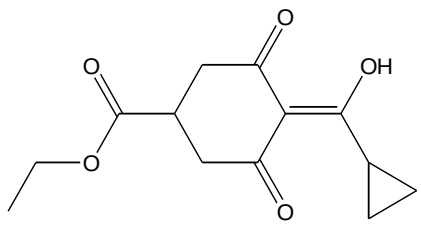
RESIDUES AND ANALYTICAL ASPECTS

At the 44th session of the CCPR (2012), trinexapac-ethyl was scheduled for evaluation as a new compound by 2013 JMPR.

Trinexapac-ethyl is a synthetic plant growth regulator that is derived from cyclohexanecarboxylate. It is applied as a foliar spray, post-emergence and is approved for use on cereal crops such as barley, durum wheat, oats, rye, triticale and wheat, oilseed rape and sugarcane as well as on grassland pastures.

The manufacturer supplied information on identity, metabolism, methods of residue analysis, freezer storage stability, registered use patterns, supervised residue trials, fates of residues in processing and farm animal feeding studies.

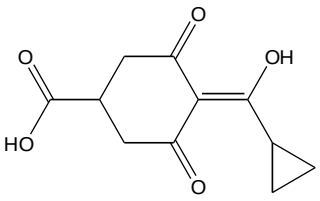
The IUPAC name is 4-(cyclopropyl-hydroxy-methylene)-3,5-dioxo-cyclohexanecarboxylic acid ethyl ester.



Trinexapac-ethyl

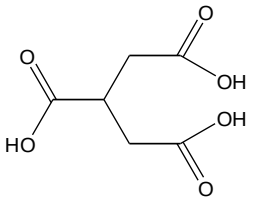
The 2013 JMPR established an ADI for trinexapac-ethyl of 0–0.3 mg/kg bw trinexapac acid equivalents.

The structures of the key metabolites discussed are shown below:



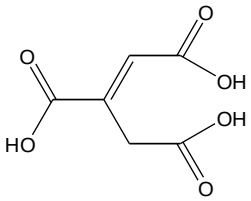
CGA 179500

Trinexapac acid



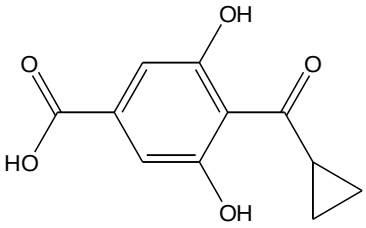
CGA 275537

Tricarballylic acid

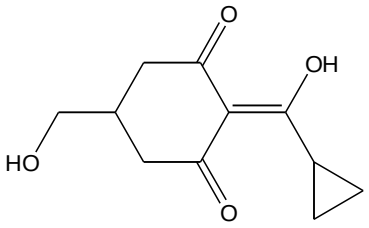


CGA 312753

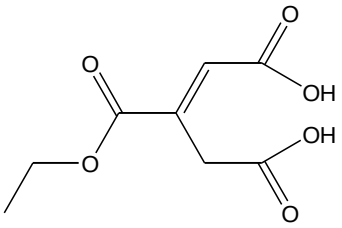
Trans aconitic acid



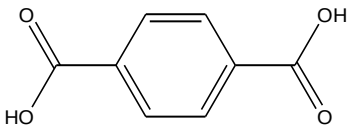
CGA 329773



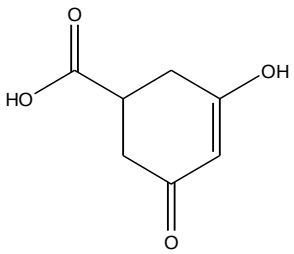
CGA 351210



CGA 312753 mono-ethyl ester



Terephthalic acid



CGA113745

Animal metabolism

The Meeting received animal metabolism studies with trinexapac-ethyl in rats, hens and goats.

Rats

Evaluation of the metabolism studies in rats was carried out by the WHO Core Assessment Group.

Trinexapac-ethyl undergoes limited metabolism in the rat, involving predominantly ester hydrolysis of trinexapac-ethyl to trinexapac acid. The predominant urinary metabolite was trinexapac acid (up to 100% of total urinary radioactivity), with low levels of a conjugated derivative of trinexapac acid detected only in the urine of bile-duct cannulated rats (6.3% of the administered dose). In faeces, the parent compound accounted for 5–22% of total faecal radioactivity (1–2.5% of the administered dose), with the balance comprising trinexapac acid. Bile contained mainly a conjugated derivative of trinexapac acid (2.9% of the administered dose), with low levels of the parent compound also detected (0.2% of the administered dose).

Goats

[1, 2-^{14}C-cyclohexyl] trinexapac-ethyl was orally administered via capsule to lactating goats at 7.2 ppm or 694 ppm in the feed (or 0.20 or 19.9 mg/kg bw/day respectively) for 4 consecutive days. Milk was sampled twice daily, in the morning and afternoon. Animals were sacrificed approximately 4 hours after the last dose.

The low dose goat eliminated 16% and 50% via faeces and urine of the total administered dose respectively at 76 hours in the study, while the corresponding values of the high dosed goat were 19% and 62%. In milk, 0.02% of the administered dose was recovered at both dose levels. A plateau was reached after 2 days (low dose-0.002 mg/kg equiv (am) and 0.007 mg/kg equiv. (pm)) or 3 days (high dose-0.22 mg/kg equiv (am) and 0.83 mg/kg equiv. (pm)) after administration.

The tissue residues of the low dosed goat were 0.035–0.043 mg/kg equiv. in muscles, 0.017–0.094 mg/kg equiv. in fat, 0.25 mg/kg equiv. in liver and 0.50 mg/kg equiv. in kidney. In the high dosed goat the residues were correspondingly higher, i.e., 1.90–2.49 mg/kg equiv. in muscles, 1.20–1.55 mg/kg equiv. in fat, 12.1 mg/kg equiv. in liver and 41.9 mg/kg equiv. in kidney.

Residues in muscle and kidney were mostly trinexapac acid (81–90% TRR). In milk, trinexapac acid accounted for 46–76% TRR. In liver and fat trinexapac acid was 31–67% TRR, although for fat this represented 87–96% of the extracted radioactivity. For liver this represented only 34–47% of the extracted radioactivity.

In another study [1, 2, 6-^{14}C-cyclohexyl] trinexapac-ethyl was orally administered by gelatine capsule to two lactating goats at 100 ppm in the feed (or 3.1 mg/kg bw/day) for 4 consecutive days. Milk was sampled twice daily. Animals were sacrificed approximately 6 hours after the last dose.

The goats had eliminated 3% and 81% via faeces and urine of the total administered dose respectively at 78 hours in the study while 0.05% of the administered dose was recovered in milk. Residues in the Day 2 pm sample of milk were 0.076 mg/kg equiv.

The tissue residues were 0.28 mg/kg equiv. in muscles, 0.11 mg/kg equiv. in fat, 0.80 mg/kg equiv. in liver and 5.90 mg/kg equiv. in kidney.

No parent was observed in tissues or milk. Residues in liver, kidney, muscle and fat were mostly trinexapac acid (CGA179500) (66–97% TRR). In milk, trinexapac acid accounted for 85% TRR. In liver, kidney and fat the metabolite CGA113745 accounted for 6–16% TRR. This metabolite was not detected in either muscle or milk.

Hens

[1, 2-^{14}C-cyclohexyl] trinexapac-ethyl was orally administered daily to laying hens using gelatine capsules at a low dose level (2 hens, 0.4 mg/kg body weight equivalent to 3.8 ppm in feed) or a high dose level (4 hens, 20.3 mg/kg body weight equivalent to 180 ppm in feed) for 4 consecutive days. Eggs were collected throughout the day and in the morning before subsequent administration. Animals were sacrificed approximately 4 hours after the last dose.

Over the period of the experiment (76 hours), 85–89% (high and low doses respectively) of the total administered dose was eliminated in excreta. Transfer of radioactivity into eggs accounted for only 0.01% and 0.02% of the total administered dose for the low and high dosed hens, respectively. A plateau was reached at Day 2 of the dosing period. Concentrations of radioactivity in egg yolk were generally less than in egg whites. Concentrations of radioactivity in egg white and egg yolk in the low-dosed hens did not exceed 0.007 and 0.002 mg/kg equiv., respectively. In the high dosed hens the corresponding values were 0.33 and 0.055 mg/kg equiv.

Mean radioactive residues in tissues of the low-dosed hens were 0.002 mg/kg equiv. in lean meat (0.12 mg/kg equiv. high dose), 0.011 mg/kg equiv. in skin (including attached fat) (0.37 mg/kg equiv.), 0.003 mg/kg equiv. in peritoneal fat (0.18 mg/kg equiv.), 0.013 mg/kg equiv. in liver (0.60 mg/kg equiv.) and 0.043 mg/kg in kidney (1.77 mg/kg equiv.).

Radioactive residues in muscle (lean meat), liver, kidney and fat samples of low and high-dosed hens were predominantly trinexapac acid (44–84% TRR). In skin including attached fat in which the extractability was low, trinexapac acid was again the major metabolite accounting for 64–80% of the extracted radioactivity.

In the yolk the major metabolite was trinexapac acid accounting for 57–76% of the extracted radioactivity (0.0003–0.011 mg/kg equiv.), while parent trinexapac-ethyl was present at only 10–25% (< 0.0001–0.005 mg/kg equiv.). In egg white the major residue was parent accounting for 64–78% of the extractable TRR (0.0017–0.124 mg/kg equiv.). Trinexapac acid was either not detected (high dose) or accounted for only 13% (0.0003 mg/kg equiv.) of the extractable TRR (low dose).

In another hen metabolism study [1, 2, 6-^{14}C-cyclohexyl] trinexapac-ethyl was orally administered daily to 5 laying hens by gelatine capsule at 8.1–10.4 ppm in feed for 10 consecutive days. Eggs were collected at 24 hour intervals throughout the morning before subsequent administration. Animals were sacrificed approximately 22 hours after the last dose.

Of the administered dose 89% was recovered in the excreta. Residue levels in the various tissue samples were all < 0.01 mg/kg equiv. Residue levels in egg yolk were very low, ranging from < 0.003–0.009 mg/kg equiv. Egg white residues ranged from 0.005–0.031 mg/kg equiv. The maximum residue levels of 0.009 and 0.031 mg/kg equiv. were reached by Day 8 of the dosing period. Overall retention of the radioactivity in eggs was < 0.1% of the administered dose.

TRR values for edible tissues and egg yolks were below 0.01 mg/kg equiv., so no further analysis was undertaken. The major metabolites identified in the organosoluble residue of the composite egg white sample were parent trinexapac-ethyl and trinexapac acid which accounted for 31% TRR and 20% TRR respectively.

Summary of animal metabolism

The metabolic pathways of trinexapac-ethyl observed in rats, goats and poultry are similar. Animal metabolism studies showed that parent was rapidly absorbed and almost completely hydrolysed to trinexapac acid before excretion. No accumulation of residues was observed in any organ, tissue or animal commodity.

Plant metabolism

Paddy rice, spring wheat, spring rape and grass (foliar treatment) metabolism studies were carried out with [1, 2-^{14}C-cyclohexyl] trinexapac-ethyl or [1, 2, 6-^{14}C-cyclohexyl] trinexapac-ethyl.

[1, 2, 6-^{14}C-cyclohexyl] trinexapac-ethyl was applied by foliar methods to rice at rates of 40 and 160 g ai/ha. TRRs in samples taken at maturity (60 days after treatment at 160 g ai/ha) were 1.58 mg/kg equiv. for straw, 2.22 mg/kg equiv. for husks and 1.07 mg/kg equiv. for grain. Trinexapac acid was the only identified compound present in rice grain at greater than 10% TRR (36% TRR) and was the major identified component of rice husks (30% TRR). It was present in rice straw at 9% TRR. Tricarballylic acid (CGA 275537) was present at 14% TRR in straw. Some parent compound was observed in grain, husks and straw (all < 10% TRR). Metabolite CGA 329773 was a minor metabolite in grain, husks and straw (< 3% TRR).

[1, 2-^{14}C-cyclohexyl] trinexapac-ethyl was applied to spring wheat by foliar spraying at a rate of 150 g ai/ha. Trinexapac acid was the only identified component in wheat grain (24% TRR) and was the only identified component of both wheat husks and wheat straw observed at > 10% of the TRR (15 and 21% TRR respectively).

A stem-injection experiment was conducted on six week old wheat plants grown under greenhouse conditions. Trinexapac acid was the major identified component of wheat grain (27.9% TRR, 0.129 mg/kg equiv.) and was the only identified component of both wheat husks and wheat straw observed at > 10% of the TRR (17% (0.073 mg/kg equiv.) and 13% TRR (0.069 mg/kg equiv.) respectively). Metabolites CGA329773 and CGA275537 were present in grain at 11% (0.050 mg/kg equiv.) and 3% TRR (0.014 mg/kg equiv.) respectively and in straw at 3 and 2% TRR respectively.

[1, 2, 6-^{14}C-cyclohexyl] trinexapac-ethyl was applied to spring rape under greenhouse conditions in plastic containers filled with soil. Trinexapac acid (free and conjugated) was the major metabolite observed in rape meal (31% TRR, 0.647 mg/kg equiv.), rape whole seeds (30% TRR, 0.431 mg/kg equiv.), pods (19% TRR, 1.06 mg/kg equiv.) and stalks (10% TRR, 0.304 mg/kg equiv.) and was also observed in oil (4% TRR, 0.004 mg/kg equiv.). Metabolite CGA 351210 was observed in oil at 16% TRR but at only 0.02 mg/kg equiv. This metabolite, only observed in rape, accounted for 16–28% TRR in pods and stalks (0.918 mg/kg equiv. and 0.869 mg/kg equiv. respectively).

[1, 2, 6-^{14}C-cyclohexyl] trinexapac-ethyl was applied by foliar methods to tall fescue grass at 560 g ai/ha. TRRs of 5.45 mg/kg equiv., 7.13 mg/kg equiv. and 4.78 mg/kg equiv. were observed for seeds, seed screenings and for straw respectively. TRRs of 2.03 mg/kg equiv. and 0.054 mg/kg equiv. were observed for 22 day forage and 105 day forage respectively. In grass, trinexapac acid was the major identified component of 22 day forage (16% TRR), straw (22% TRR) and regrowth forage (10% TRR) and was also a major component of seeds (15% TRR) and seed screenings (13% TRR). Also observed in all matrices were CGA 275537 (9-17% TRR) and terephthalic acid (7–12% TRR) the latter metabolite only observed in grass.

Summary of plant metabolism

Parent compound was completely degraded and was not detectable (< 0.001/< 0.002 mg/kg) in any plant part of wheat, rapeseed and grass at harvest time. In rice only traces (0.001 and 0.003 mg/kg) were observed in straw and husks respectively.

Trinexapac acid was therefore the major metabolite which was present in all species and all plant parts at harvest. Sugar conjugates of trinexapac acid were hydrolysed in some crop parts to yield free acid which, in some cases, significantly increased the levels of trinexapac acid recovered. Trinexapac acid sugar conjugates accounted for an increase in trinexapac acid of 11.6% TRR in rice grain and 10% and 14% TRR in grass forage and straw, respectively.

Rotational Crops

Studies of residues in confined rotational crops have been submitted in which soil was treated at 350 g ai/ha, followed by soil aging at 30, 120 and 270 days. Representative succeeding crops of lettuce (leafy vegetable), radish (root vegetable) and wheat (cereal grain) were planted at the above intervals to determine whether trinexapac-ethyl residues or degradates appear in follow crops.

The uptake of residues by the rotational crops lettuce, radish and wheat planted or sown after several intervals after application of trinexapac-ethyl to bare ground was very low. No accumulation was observed. Residues of trinexapac-ethyl in the rotational crop RACs were below the LOQ (< 0.001 mg/kg). The residues of trinexapac acid and CGA 312753 (trans aconitic acid) were very close to or below the LOQ (< 0.001–0.002 mg/kg). The very limited uptake of radioactive material in succeeding crops clearly indicates the lack of systemic behaviour of trinexapac-ethyl. It was concluded that residues in rotational crops are negligible.

The Meeting received information on field rotational crop studies for trinexapac-ethyl which were conducted in both the USA and Switzerland.

In a study conducted in the USA in the 2004–2005 growing season, a 250 g/L EC formulation of trinexapac-ethyl was applied once as a broadcast spray to wheat at 203 g ai/ha. Radish, wheat and spinach were planted after three different plant back intervals (14, 30 and 45 days). The results showed that rotational crops sown 14, 30 and 45 days after application of trinexapac-ethyl to the target crop, are very unlikely to contain residues of trinexapac-ethyl as its main metabolite trinexapac acid, above the LOQ of 0.05 mg/kg.

A trial was conducted in Switzerland during 1989 to investigate residues of trinexapac-ethyl in succeeding crops, grown in soil previously treated with [14C]cyclohexyl trinexapac-ethyl. A 250 g/L EC formulation of trinexapac-ethyl was applied once to bare soil at a rate of 150 g ai/ha.

Treated plots were planted after four different time intervals (69, 119, 299 and 338 days after treatment). The crops investigated were lettuce, winter wheat, sugar beet and maize (corn).

The uptake of residues by the rotational crops lettuce, winter wheat, sugar beet and maize planted or sown after several intervals after application of trinexapac-ethyl to bare ground was therefore very low. No accumulation was observed. Residues of trinexapac-ethyl in the rotational crop RACs were below the LOQ (< 0.001 mg/kg) so no characterization was possible. The very limited uptake of radioactive material in succeeding crops clearly indicates the lack of systemic behaviour of trinexapac-ethyl. It was concluded that the residues situation in rotational crops is negligible.

The confined and field rotational crop studies suggest that residues of trinexapac acid are unlikely to occur in succeeding crops.

Methods of analysis

The Meeting received information on analytical methods suitable for the determination of residues of trinexapac acid in plant matrices and animal matrices. The methodology involves quantification of free trinexapac acid (plant and animal matrices) or quantification of both free and conjugated trinexapac acid with the inclusion of an acid hydrolysis step (plant matrices). The methods used are based on HPLC-MS/MS (LOQs of 0.01 or 0.05 mg/kg for trinexapac acid in plant matrices and 0.02 mg/kg for animal matrices (0.01 mg/kg for milk)) and HPLC-UV (LOQs of 0.02 mg/kg for trinexapac acid in plant matrices or 0.01 mg/kg in animal matrices (0.005 mg/kg for milk)).

Details of HPLC-UV methods for determining residues of parent trinexapac-ethyl have also been submitted (LOQs of 0.02 or 0.04 mg/kg).

Stability of pesticide residues in stored analytical samples

The Meeting received information on the freezer storage stability of trinexapac acid in plant and animal commodities.

Freezer storage stability studies indicate that trinexapac acid is stable for at least 24 months in wheat grain and rapeseed and up to 24 months in wheat straw and for at least 12 months in wheat processed fractions. The stability of trinexapac acid in extracts of animal matrices was tested in the dairy cattle and laying hen feeding studies. No significant degradation of residues of trinexapac acid was observed in cattle matrices (83–121 days) or in poultry matrices (31–82 days).

The periods of storage stability studies cover the sample storage intervals of residue trials.

Definition of the residue

Animals

In the goat metabolism studies, no parent was observed in tissues and milk. Trinexapac acid accounted for 81–97% of the total residues in muscle and kidney, 31–84% of the TRRs in liver and fat and 46–85% of the TRRs in milk. In one study the metabolite CGA 113745 accounted for 6–16% TRR in liver, kidney and fat, but was not detected in either muscle or milk.

In the hen metabolism studies residues in muscle (lean meat), liver, kidney and fat samples of low and high-dosed hens were dominated by trinexapac acid (44–84% of TRR). Only in the skin (with subcutaneous fat) was the major portion of the total radioactivity (70–86%) not extractable. However trinexapac acid was the dominant metabolite in the extract (64–80% of the extractable radioactivity). The major metabolites identified in the organosoluble residue of egg whites were parent trinexapac-ethyl and trinexapac acid which accounted for 31% TRR and 20% TRR respectively.

The observed metabolic pathway of trinexapac-ethyl in livestock is comparable to that observed for the rat in which trinexapac acid is the major and only residue component of significance.

The ratio of trinexapac acid residues in muscle and fat observed in the livestock metabolism and feeding studies support the conclusion that trinexapac is not fat soluble. There is no evidence to suggest that there is significant potential for bioaccumulation in fat tissues.

It is considered that a residue definition of "Trinexapac (acid)" is appropriate for commodities of animal origin for compliance with MRLs (enforcement) and for risk assessment.

Plants

In the rice study trinexapac acid was the major identified component in rice grain (36% TRR) and rice husks (30% TRR). In the spring wheat study, trinexapac acid was the major identified component of wheat grain (28% TRR) and was the only identified component of both wheat husks and wheat straw observed at > 10% of the TRR. It was the major metabolite observed in rape meal, rape whole seeds and pods (19–31% TRR) and was also observed in oil and stalks (4–10% TRR). In grass, trinexapac acid was the major identified component of 22 day forage, straw, straw and regrowth forage (10–22% TRR) and was also a major component of seed screenings and seeds (13–15% TRR). Sugar conjugates of trinexapac acid were hydrolysed in some crop parts to yield free acid.

Trinexapac acid was present in all species and all plants parts at harvest and was generally the main component of the TRR. A definition of trinexapac acid is therefore considered suitable as the residue definition for compliance. Since conjugates of trinexapac acid were sometimes significant in edible commodities and are supposed to be of similar toxicity, the Meeting decided to include conjugates of trinexapac acid in the residue definition for risk to the acid assessment. A suitable method is available for determining residues of free and conjugated trinexapac acid.

It is therefore considered that a residue definition of "Trinexapac (acid)" is appropriate for plant commodities for compliance with MRLs (enforcement). It is considered that a residue definition of "Trinexapac and its conjugates, expressed as trinexapac acid" is appropriate for plant commodities for risk assessment.

Definition of the residue (for compliance with the MRL for plant and animal commodities and for estimation of dietary intake for animal commodities): Trinexapac (acid)

Definition of the residue (for estimation of dietary intake for plant commodities): Trinexapac and its conjugates, expressed as trinexapac acid

The residue is considered not fat soluble.

Results of supervised residue trials on crops

Supervised trials were available for the use of trinexapac on barley, wheat, oilseed rape and sugarcane.

In some studies (barley, wheat and sugarcane) residues have been determined as total trinexapac (includes free trinexapac acid and conjugates of trinexapac acid) while in other studies (wheat, sugarcane and rape seed) residues have been determined as trinexapac (free trinexapac acid only). In both cases trinexapac means trinexapac acid. For dietary intake assessment (risk assessment) residues expressed as total trinexapac acid, where available, have been considered. For maximum residue level estimation (compliance) residues expressed as trinexapac acid, where available, have been considered.

Product labels were available from Belgium, France, Germany and the United States of America.

Cereals - wheat (free trinexapac acid)

Residue trials were conducted in wheat in the USA according to the critical GAP in the USA (1 application at 0.123 kg ai/ha, 45-day PHI).

For the estimation of maximum residue levels the ranked order of residues of trinexapac acid in wheat grain from supervised trials according to the GAP in the USA was 0.10, 0.25, 0.32, 0.34, 0.35, 0.46, 0.49, 0.55, 0.55, 0.57, 0.77, 0.88, 0.91, 0.98, 0.99, 1.05, 1.35 and 1.95 mg/kg.

The Meeting estimated a maximum residue level for trinexapac acid in wheat of 3 mg/kg.

The Meeting recognized that wheat (spring wheat, winter wheat and durum wheat) and triticale, barley and oats have similar GAPs and normally show comparable residues after early treatment. As application was before flowering, the Meeting decided to extrapolate the MRL estimated for wheat grain to barley, oats and triticale.

Cereals - barley

Residue trials were conducted in barley in the USA according to the GAP in the USA (1 application at 0.123 kg ai/ha, 45-day PHI).

For dietary intake purposes the ranked order of total residues of trinexapac acid in barley grain from supervised trials according to the GAP in the USA was 0.03, 0.08, 0.44, 0.50, 0.52, 0.53, 0.60, 0.72, 0.76, 0.83, 1.0 and 1.2 mg/kg.

Cereals - wheat (total residues of trinexapac acid)

Residue trials were conducted in wheat in the USA according to the GAP in the USA (1 application at 0.123 kg ai/ha, 45-day PHI).

For dietary intake purposes the ranked order of total residues of trinexapac acid in wheat grain from supervised trials according to the GAP in the USA was 0.07, 0.15, 0.27, 0.31, 0.32, 0.40, 0.45, 0.47, 0.53, 0.77, 0.78, 0.82, 0.85, 0.99, 1.01, 1.14, 1.64 and 3.32 mg/kg.

The Meeting noted that the USA GAP is the same for barley and wheat. The Meeting noted that the populations of residues data for barley and wheat matching USA GAP resulted in similar distributions of residues for barley and wheat (e.g. medians do not differ by more than 5×). Given the similarity of the datasets (confirmed by the Mann-Whitney U test), the Meeting decided to combine the datasets for barley and wheat to give a larger dataset for estimation of dietary parameters for the purposes of determining an STMR residue level for barley grain and wheat grain.

For dietary intake purposes residues were 0.03, 0.07, 0.08, 0.15, 0.27, 0.31, 0.32, 0.40, 0.44, 0.45, 0.47, 0.50, 0.52, 0.53, 0.53, 0.60, 0.72, 0.76, 0.77, 0.78, 0.82, 0.83, 0.85, 0.99, 1.0, 1.01, 1.14, 1.2, 1.64 and 3.32 mg/kg.

The Meeting estimated an STMR of 0.57 mg/kg.

The Meeting recognized that wheat (spring wheat, winter wheat and durum wheat), triticale, barley and oats have similar GAPs and normally show comparable residues after early treatment. As application was before flowering, the Meeting decided to extrapolate the STMR estimated for barley grain and wheat grain to oats and triticale.

Grasses for sugar or syrup production - sugarcane (free trinexapac acid)

Residue trials were conducted in sugarcane in the USA according to the critical GAP in the USA (1 application at 0.347 kg ai/ha, 28-day PHI).

For the estimation of maximum residue levels the ranked order of residues of trinexapac acid in sugar cane from supervised trials according to the GAP in the USA was < 0.05, 0.06, 0.09, 0.12, 0.22, 0.23 and 0.25 mg/kg.

The Meeting estimated a maximum residue level for trinexapac acid in sugarcane of 0.5 mg/kg.

Grasses for sugar or syrup production—sugarcane (total residues of trinexapac acid)

Residue trials were conducted in sugarcane in the USA according to the critical GAP in the USA (1 application at 0.347 kg ai/ha, 28-day PHI).

For dietary intake purposes the ranked order of total residues of trinexapac acid in sugar cane from supervised trials according to the GAP in the USA was < 0.01, 0.04, 0.06, 0.08, 0.17 and 0.42 mg/kg.

The Meeting estimated an STMR value for trinexapac acid in sugarcane of 0.07 mg/kg.

Oilseeds - Rape

As conjugates of trinexapac acid were not significant in the submitted rape metabolism studies, residue levels from the submitted rape seed studies, which were determined as trinexapac only, have been considered suitable for estimation of a maximum residue level and also for estimation of dietary intake parameters.

Residue trials were conducted in winter rape (canola) in Germany according to the critical GAP in Germany for winter rape (1 application at 0.375 kg ai/ha).

For the estimation of maximum residue levels and for dietary purposes the ranked order of residues of trinexapac acid in winter rape seed from supervised trials according to GAP was 0.04, 0.10, 0.10, 0.13, 0.15, 0.15, 0.16, 0.24, 0.24, 0.26, 0.29, 0.31, 0.64, 0.64, 0.90 and 1.0 mg/kg.

The Meeting estimated maximum residue level and STMR values for trinexapac acid in rape seed of 1.5 and 0.24 mg/kg respectively.

Animal feeds

The Meeting received supervised trials data for barley hay and straw, wheat forage, hay and straw and rape seed forage.

Moisture content percentages for animal feeds have not been determined. The values from the FAO Manual on the Submission and Evaluation of Pesticides Residues Data for the feeds, have been used to convert wet weight or 'as received' residues values to dry weight residues values.

Forage - Wheat (residues of free and conjugated trinexapac acid)

Wheat forage was collected in the trials carried out in the USA according to GAP in the USA (1 application at 0.123 kg ai/ha).

For the calculation of the livestock animal dietary burden the ranked order of total residues in wheat forage samples collected 30 days after application (wet weight) at the GAP application rate was 0.02, 0.03, 0.04, 0.06, 0.07, 0.08, 0.08, 0.09, 0.10, 0.10, 0.12, 0.17, 0.17, 0.22, 0.23, 0.33, 0.38 and 0.94 mg/kg.

For the calculation of the livestock animal dietary burden the ranked order of total residues in wheat forage samples collected 30 days after application (dry weight) at the GAP application rate was 0.08, 0.12, 0.16, 0.24, 0.28, 0.32, 0.32, 0.36, 0.40, 0.40, 0.48, 0.68, 0.68, 0.88, 0.92, 1.32, 1.52 and 3.76 mg/kg.

The Meeting estimated median and highest residue values for trinexapac acid in wheat forage (dry weight) of 0.40 and 3.76 mg/kg respectively.

The Meeting recognized that wheat (spring wheat, winter wheat and durum wheat) and triticale, barley and oats have similar GAPs and normally show comparable residues after early treatment. The Meeting decided to extrapolate the dietary parameters estimated for wheat forage to barley, oats and triticale.

Hay and Straw - Barley and Wheat

Data for wheat hay and straw were collected in the trials carried out in the USA which approximate USA GAP (1 application at 0.123 kg ai/ha).

For the estimation of maximum residue levels the ranked order of residues in wheat hay samples collected 30 days after application (wet weight) at the GAP application rate was < 0.05, < 0.05, < 0.05, 0.05, 0.06, 0.07, 0.07, 0.08, 0.08, 0.09, 0.11, 0.11, 0.13, 0.15, 0.18, 0.19, 0.19 and 0.75 mg/kg.

The ranked order of residues in wheat hay samples converted to dry weight basis at the GAP application rate was < 0.06, < 0.06, < 0.06, 0.06, 0.07, 0.08, 0.08, 0.09, 0.09, 0.10, 0.13, 0.13, 0.15, 0.17, 0.20, 0.22, 0.22 and 0.85 mg/kg.

For the estimation of maximum residue levels the ranked order of residues in wheat straw samples collected 45 days after application (wet weight) at the GAP application rate was < 0.05, < 0.05, < 0.05, < 0.05, < 0.05, < 0.05, < 0.05, < 0.05, < 0.05, 0.06, 0.06, 0.07, 0.07, 0.08, 0.10, 0.11 and 0.12 mg/kg.

The ranked order of residues in wheat straw samples (dry weight) at the GAP application rate was < 0.06, < 0.06, < 0.06, < 0.06, < 0.06, < 0.06, < 0.06, < 0.06, < 0.06, < 0.06, 0.07, 0.07, 0.08, 0.08, 0.09, 0.11, 0.13 and 0.14 mg/kg.

The Meeting used the wheat hay data for the free trinexapac acid to estimate a maximum residue level in wheat straw and fodder (dry) of 0.9 mg/kg. The Meeting recognized that wheat (spring wheat, winter wheat and durum wheat), triticale, barley and oats have similar GAPs and normally show comparable residues after early treatment. It was therefore decided to apply the maximum residue level recommended for trinexapac acid on wheat straw and fodder (dry) to barley, oats and triticale.

For the calculation of the livestock animal dietary burden the ranked order of total residues in barley hay samples collected 30 days after application (wet weight) at the GAP application rate was < 0.01, 0.03, 0.05, 0.10, 0.13, 0.15, 0.17, 0.18, 0.25, 0.33, 0.40 and 0.48 mg/kg.

The ranked order of total residues in barley hay samples converted to a dry weight basis at the GAP application rate was < 0.011, 0.03, 0.06, 0.11, 0.15, 0.17, 0.19, 0.20, 0.28, 0.38, 0.45 and 0.55 mg/kg.

For the calculation of the livestock animal dietary burden the ranked order of total residues in barley straw samples collected 45 days after application (wet weight) at the GAP application rate was < 0.01, 0.02, 0.07, 0.08, 0.08, 0.09, 0.11, 0.12, 0.14, 0.17, 0.20 and 0.24 mg/kg.

The ranked order of total residues in barley straw samples converted to a dry weight basis at the GAP application rate was 0.01, 0.02, 0.08, 0.09, 0.09, 0.10, 0.12, 0.13, 0.16, 0.19, 0.22 and 0.27 mg/kg.

For the calculation of the livestock animal dietary burden the ranked order of total residues in wheat hay samples collected 30 days after application (wet weight) at the GAP application rate was 0.03, 0.04, 0.04, 0.06, 0.09, 0.11, 0.11, 0.14, 0.17, 0.19, 0.24, 0.30, 0.31, 0.41, 0.50, 0.59, 0.78 and 1.18 mg/kg.

The ranked order of total residues in wheat hay samples collected 30 days after application (dry weight) at the GAP application rate was 0.03, 0.05, 0.05, 0.07, 0.10, 0.13, 0.13, 0.16, 0.19, 0.22, 0.27, 0.34, 0.35, 0.47, 0.57, 0.67, 0.89 and 1.34 mg/kg.

For the calculation of the livestock animal dietary burden the ranked order of total residues in wheat straw samples collected 45 days after application (wet weight) at the GAP application rate was 0.01, 0.03, 0.04, 0.04, 0.06, 0.09, 0.11, 0.11, 0.15, 0.15, 0.17, 0.20, 0.23, 0.28, 0.33, 0.46, 0.59 and 0.60 mg/kg.

The ranked order of total residues in wheat straw samples collected 45 days after application (dry weight) at the GAP application rate was 0.01, 0.03, 0.05, 0.05, 0.07, 0.10, 0.13, 0.13, 0.17, 0.17, 0.19, 0.23, 0.26, 0.32, 0.38, 0.52, 0.67 and 0.68 mg/kg.

The Meeting noted that the USA GAP is the same for barley and wheat. The Meeting also noted that the populations of residues data for barley and wheat hay matching USA GAP gave higher residues than the straw and resulted in similar distributions of residues (e.g., medians do not differ by more than 5×). Given the similarity of the datasets (confirmed by the Mann-Whitney U test), the Meeting decided to combine the datasets for barley and wheat hay (dry weight) to give a larger dataset for estimation of dietary parameters for the purposes of determining median and highest residue levels for barley straw and fodder (dry) and wheat straw and fodder (dry).

For the calculation of the livestock animal dietary burden the ranked order of total residues of trinexapac acid in barley and wheat hay (dry weight) collected 30 days after application (dry weight) from supervised trials according to the GAP in the USA was < 0.011, 0.03, 0.03, 0.05, 0.05, 0.06, 0.07, 0.10, 0.11, 0.13, 0.13, 0.15, 0.16, 0.17, 0.19, 0.19, 0.20, 0.22, 0.27, 0.28, 0.34, 0.35, 0.38, 0.45, 0.47, 0.55, 0.57, 0.67, 0.89 and 1.34 mg/kg.

The Meeting used the barley and wheat hay data to estimate median and highest residue values for trinexapac acid in barley straw and fodder (dry) and wheat straw and fodder (dry) of 0.19 and 1.34 mg/kg respectively, based on the combined dataset for barley and wheat hay.

The Meeting recognized that wheat (spring wheat, winter wheat and durum wheat) and barley have similar GAPs to oats and triticale and normally show comparable residues after early treatment. The Meeting decided to extrapolate the dietary parameters estimated for wheat and barley to oats and triticale.

Rape seed forage

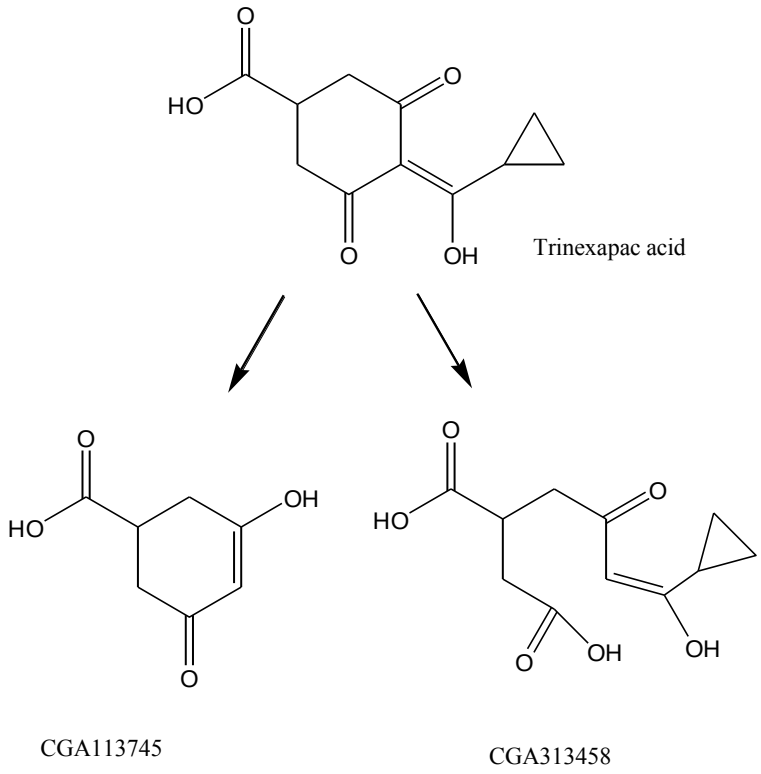
In four trials conducted according to GAP in Germany, residues in rape remaining plant type (forage) at PHIs from 49–68 days were < 0.04, < 0.04, < 0.04 and 0.04 mg/kg.

The Meeting agreed that four trials according to GAP were not sufficient to estimate a highest residue for trinexapac in rape forage.

Fate of residues during processing

High temperature hydrolysis of residues of trinexapac-ethyl and trinexapac acid under varying conditions have been reported. The [^{14}C]-labelled compounds were dissolved in aqueous buffer; at pH 4 and heated for 20 or 25 minutes at 90 ºC to simulate pasteurisation, at pH 5 and refluxed at 100 ºC for 60 minutes to simulate baking, brewing and boiling and at pH 6 at about 120 ºC in an autoclave for 20 minutes to simulate sterilisation. Trinexapac-ethyl was not degraded. Trinexapac acid underwent limited degradation and was the major component of the radioactive material at the end of the hydrolysis reactions (51–59%). It degraded into the compounds CGA113745 (10–12%) and CGA313458 (16–21%).

CGA113745 was observed as a goat metabolite in one metabolism study, while CGA313458 was observed as a rapeseed and rice metabolite. Based on a structural assessment of CGA 113745 and CGA 313458 and the estimated levels of chronic dietary intake, the Meeting concluded that these metabolites are unlikely to pose a dietary risk.



Hydrolysis of trinexapac acid under processing conditions

The Meeting also received processing studies for barley, wheat, sugarcane and rape seed. The table below summarizes STMR-P values calculated on the determined processing factors. In addition the following maximum residue levels were estimated.

Barley

Based on the total trinexapac processing factor of 1.9 for barley bran (in the absence of a trinexapac processing factor) and the barley grain MRL of 3 mg/kg, the calculated expected highest residues in barley bran are 5.7 mg/kg. The Meeting estimated an MRL for trinexapac in barley bran of 6 mg/kg.

Wheat

Based on the trinexapac processing factor of 2.3 for wheat bran and the wheat grain MRL of 3 mg/kg the calculated expected highest residues in wheat bran are 6.9 mg/kg. The Meeting estimated an MRL for trinexapac in wheat bran of 8 mg/kg.

The processing factors derived from the processing studies and the resulting recommendations for STMR-Ps (and maximum residue levels) are summarized in the table below.

Processing Factors from the Processing of Raw Agricultural Commodities (RACs) with Field-Incurred Residues from Foliar Treatment with Trinexapac-ethyl

RAC	Processed Commodity	Best Estimate Processing Factor	RAC MRL	RAC STMR	Processed Commodity STMR-P
Barley	Pearled barley	1.2	3	0.57	0.68
	Bran	1.9			1.08
	Flour	0.43			0.25
Wheat	Aspirated Grain Fractions	0.55	3	0.57	0.31

RAC	Processed Commodity	Best Estimate Processing Factor	RAC MRL	RAC STMR	Processed Commodity STMR-P
	Bran	1.9			1.08
	Flour	0.43			0.25
	Middlings	0.55			0.31
	Shorts	0.46			0.26
	Germ	1.1			0.63
Sugarcane	Molasses	5.8	0.5	0.07	0.40
	Refined sugar	0.15			0.01
Rapeseed	Press cake	1.2	1.5	0.24	0.32
	Refined oil	0.05			0.01

Except for the rapeseed study (in which processing factors were determined based on residues of trinexapac), processed commodity STMR-Ps were calculated on the basis of the total trinexapac acid processing factors.

Residues in animal commodities

Estimated maximum and mean dietary burdens of farm animals

Dietary burden calculations for beef cattle and dairy cattle and poultry are provided below. The dietary burdens were estimated using the OECD diets listed in Appendix IX of the 2009 edition of the FAO Manual.

Potential cattle feed items include: wheat, barley, oat and triticale grain, straw, forage, hay and silage, wheat milled by-products (bran), wheat aspirated grain fractions, barley bran fractions, sugarcane molasses and bagasse and rape forage and rape seed meal.

Summary of livestock dietary burden for trinexapac (ppm of dry matter diet)

	US-Canada		EU		Australia		Japan	
	max	mean	max	mean	max	mean	max	mean
Beef cattle	0.98	0.87	1.76	0.82	3.76 [a]	0.88	0.97	0.97 [c]
Dairy cattle	1.76	0.77	1.76	0.76	3.76 [b]	0.86 [d]	1.00	0.83

[a] Highest maximum beef or dairy cattle dietary burden suitable for HR and MRL estimates for mammalian meat

[b] Highest maximum dairy cattle dietary burden suitable for HR and MRL estimates for mammalian milk

[c] Highest mean beef or dairy cattle dietary burden suitable for STMR estimates for mammalian meat

[d] Highest mean dairy cattle dietary burden suitable for STMR estimates for mammalian milk

Potential poultry feed items include: wheat, barley, oat and triticale grain, straw, forage, hay and silage, wheat milled by-products (bran), wheat aspirated grain fractions, barley bran fractions, sugarcane molasses and bagasse and rape forage and rape seed meal.

Summary of poultry dietary burden for trinexapac (ppm of dry matter diet)

	US-Canada		EU		Australia		Japan	
	max	mean	max	mean	max	mean	max	mean
Poultry Broiler	0.94	0.94	0.70	0.70	0.34	0.34	0.13	0.13
Poultry Layer	0.94	0.94 [b]	1.08 [a]	0.76	0.34	0.34	0.37	0.37

[a] Highest maximum poultry dietary burden suitable for HR and MRL estimates for poultry meat and eggs

[b] Highest maximum poultry dietary burden suitable for STMR estimates for poultry meat and eggs

Farm Animal Dietary Burden

The Meeting received a lactating dairy cow feeding study which provided information on residues of trinexapac acid arising in tissues and milk when dairy cows were dosed for 29–30 days, at target feeding levels equivalent to 0, 2, 6 and 20 ppm trinexapac acid in the diet.

No quantifiable residues of trinexapac acid were observed in milk at the 0, 2 and 6 ppm dose groups. Residues in the 20 ppm dose group ranged from < 0.005–0.011 mg/kg. Residues reached a plateau after one day.

No quantifiable residues of trinexapac acid were observed in the tissues of the control cow. In the lowest and mid-range dose groups (2 and 6 ppm), no quantifiable residues were detected in muscle, liver and fat, although residues slightly above LOQ (0.02 mg/kg) were observed in kidney (highest (mean in brackets) were 0.03 (0.03) and 0.05 (0.04) mg/kg respectively). No quantifiable residues were detected in muscle in the 20 ppm dose group while the highest residues (mean in brackets) observed in liver, kidney and fat in the 20 ppm dose group were 0.03 (0.03), 0.29 (0.17) and 0.02 (< 0.02) mg/kg respectively.

The Meeting also received information on the residue levels arising in tissues and eggs when laying hens were dosed with trinexapac acid for 28 days, at feeding levels equivalent to 3.7, 10 and 37 ppm in the diet.

No quantifiable residues of trinexapac acid were observed in eggs at any feeding level except in the 37 ppm dose group at 28 days (highest and mean residues were 0.01 mg/kg).

No residues of trinexapac acid above LOQ (0.01 mg/kg) were detected in any muscle (breast and thigh muscle) sample from any treatment group. No residues of trinexapac acid above LOQ were detected in any liver sample from any treatment group apart from the highest dose group (highest and mean residues were 0.02 mg/kg). No residues of trinexapac acid above LOQ were detected in any fat sample from any treatment group apart from the highest dose group (highest and mean residues were 0.03 mg/kg). Highest (mean) residues of trinexapac acid in kidney were 0.08 (0.06), 0.05 (0.04) and 0.54 (0.45) mg/kg in the lowest to highest dose groups respectively.

Animal commodity maximum residue levels

Cattle- STMR, HR and MRLs

For highest residue level estimation, the high residues in the cattle tissues were calculated by interpolating the maximum dietary burden for beef cattle (3.76 ppm) between the relevant feeding levels (2 and 6 ppm) in the dairy cow feeding study and using the highest tissue concentrations from individual animals within those feeding groups. For highest residue level estimation, the high residues in the cattle milk were calculated by interpolating the maximum dietary burden for dairy cattle (3.76 ppm) with the lowest feeding level (2 and 6 ppm) in the dairy cow feeding study and using the highest mean milk concentrations from those feeding groups.

The STMR values for the tissues were calculated by extrapolating the mean dietary burden for beef cattle (0.97 ppm) with the 2 ppm feeding level from the dairy cow feeding study and using the mean tissue concentrations from that feeding group. The STMR values for the milk were calculated by extrapolating the mean dietary burden for dairy cattle (0.86 ppm) with the 2 ppm feeding level from the dairy cow feeding study and using the mean milk concentrations from that feeding group.

Trinexapac Feeding Study	Feed Level (ppm) for milk residues	Residues (mg/kg) in milk	Feed Level (ppm) for tissue residues	Residues (mg/kg)			
				Muscle	Liver	Kidney	Fat
HR Determination (beef or dairy cattle)							
Feeding Study	2	< 0.005	2	< 0.02	< 0.02	0.03	< 0.02
	6	< 0.005	6	< 0.02	< 0.02	0.05	< 0.02
Dietary burden and estimate of highest residue	3.76	< 0.005	3.76	< 0.02	< 0.02	0.04	< 0.02
STMR Determination (beef or dairy cattle)							
Feeding Study	2	< 0.005	2	< 0.02	< 0.02	0.03	< 0.02
Dietary burden and estimate of highest residue	0.86	< 0.0022	0.97	< 0.010	< 0.010	0.015	< 0.010

The Meeting estimated the following STMR values: milk 0 mg/kg; muscle 0 mg/kg; edible offal (based on kidney) 0.015 mg/kg and fat 0 mg/kg.

The Meeting estimated the following maximum residue levels: milk 0.005(*) mg/kg; meat (mammalian except marine mammals) 0.01(*) mg/kg, edible offal (based on kidney) 0.1 mg/kg and mammalian fats (except milk fats) 0.01(*) mg/kg.

Poultry - STMR, HR and MRLs

For highest residue level estimation, the high residues in the hen tissues and eggs were calculated by extrapolating the maximum dietary burden (1.08 ppm) with the lowest feeding level (3.7 ppm) in the laying hen feeding study and using the highest tissue concentrations from individual animals within that feeding group and using the highest mean egg concentration from that feeding group.

The STMR values for the tissues and eggs were calculated by extrapolating the mean dietary burden (0.94 ppm) with the lowest feeding level (3.7 ppm) from the poultry feeding study and using the mean tissue and egg concentrations from that feeding group.

Trinexapac Feeding Study	Feed Level (ppm) for egg residues	Residues (mg/kg) in egg	Feed Level (ppm) for tissue residues	Residues (mg/kg)			
				Muscle	Liver	Kidney	Fat
HR Determination (poultry broiler or layer)							
Feeding Study	3.7	< 0.01	3.7	< 0.01	< 0.01	0.08	< 0.01
Dietary burden and estimate of highest residue	1.08	< 0.003	1.08	< 0.003	< 0.003	0.023	< 0.003
STMR Determination (poultry broiler or layer)							
Feeding Study	3.7	< 0.01	3.7	< 0.01	< 0.01	0.06	< 0.01
Dietary burden and estimate of highest residue	0.94	< 0.0025	0.94	< 0.0025	< 0.0025	0.015	< 0.0025

The Meeting estimated the following STMR values: egg 0 mg/kg; muscle 0 mg/kg; edible offal (based on kidney) 0.015 mg/kg and fat 0 mg/kg.

The Meeting estimated the following maximum residue levels: eggs 0.01(*) mg/kg; poultry meat 0.01(*) mg/kg, poultry edible offal (based on kidney) 0.05 mg/kg and poultry fats 0.01(*) mg/kg.

RECOMMENDATIONS

Definition of the residue (for compliance with the MRL for plant and animal commodities and for estimation of dietary intake for animal commodities): Trinexapac (acid)

Definition of the residue (for estimation of dietary intake for plant commodities): *Trinexapac and its conjugates, expressed as trinexapac acid.*

The Meeting estimated the maximum residue levels and STMR values shown below.

DIETARY RISK ASSESSMENT

Long-term intake

The evaluation of trinexapac has resulted in recommendations for MRLs and STMRs for raw and processed commodities. Consumption data were available for 36 food commodities and were used in the dietary intake calculation. The results are shown in Annex 3. The International Estimated Daily Intakes for the 13 GEMS/Food regional diets, based on estimated STMRs were in the range 0–1% of the maximum ADI of 0.3 mg/kg bw (Annex 3).

The Meeting concluded that the long-term intake of residues of trinexapac from uses that have been considered by the JMPR is unlikely to present a public health concern.

Short-term intake

The International Estimated Short-term Intake (IESTI) for trinexapac was not calculated, as it was not considered necessary to establish an ARfD.

The Meeting concluded that the short-term intake of residues of trinexapac from uses that have been considered by the JMPR is unlikely to present a public health concern.