5.11 FENAZAQUIN (297)

TOXICOLOGY

Fenazaquin is the ISO–approved common name for 4-tert-butylphenethyl-quinazolin-4-yl-ether (IUPAC name), with the CAS number 120928-09-8. It is an acaricide and has a quinazoline structure.

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

All critical studies contained statements of compliance with GLP and were conducted in accordance with relevant national or international test guidelines, unless otherwise indicated.

Biochemical aspects

In studies conducted using [^{14}C]fenazaquin, C_{max} was reached in plasma at 8 hours after a single oral dose of 1, 10 or 30 mg/kg bw to rats. The absorption in rats was estimated to be 60% of 1 mg/kg bw dose. Fenazaquin and/or its metabolites did not accumulate in tissues. The major route of excretion was via faeces (71.9–88.9% after 7 days). Excretion in the urine ranged from 16.4% to 20.9%. The majority of fenazaquin was excreted within 48 hours after dosing. The biotransformation of fenazaquin in the rat was predominantly by cleavage of the ether bond, resulting in the formation of the respective alcohol (4-hydroxyquinazoline or 4-OH) and carboxyl acid (AN-1) derivatives. Other biotransformation reactions included oxidation of one of the methyl groups on the alkyl side chain to produce either alcohol (F-1) or carboxylic acid (F-2) metabolites. Finally, hydroxylation at the *O*-ether alkyl moiety of F-1 or F-2 resulted in the metabolites F-1A and 2-hydroxyfenazaquin acid (F-3), respectively. Major metabolites were AN-1, NN-2 complex and NN-3 complex in urine and F-2 and F-3 in faeces (>7% for all metabolites).

Fenazaquin was more rapidly absorbed in mice and hamsters than in rats and more rapidly excreted by mice than by rats and hamsters. The half-life in mice was about one tenth to one twentieth of that in rats or hamsters, respectively.

Toxicological data

The oral LD_{50} of fenazaquin was 134 mg/kg bw in rats, 1480 mg/kg bw in mice and 812 mg/kg bw in hamsters. The dermal LD_{50} in rabbits was more than 5000 mg/kg bw. The LC_{50} in rats was greater than 1.9 mg/L. Fenazaquin was not irritating in rabbit skin and eyes. Fenazaquin was not sensitizing in guinea-pigs.

The main toxic effects of fenazaquin in short- and long-term toxicity studies in hamsters, rats and dogs were decreased body weight gain and feed consumption. Testicular toxicity was observed in hamsters.

In a 90-day oral toxicity study in hamsters, fenazaquin was administered by gavage at doses of 0, 5, 25, 50 (females only), 75 (males only), 100 (females only) or 150 (males only) mg/kg bw per day. The NOAEL was 25 mg/kg bw per day based on lower body weights and body weight gains at 50 mg/kg bw per day.

Two 90-day oral toxicity studies were conducted in rats. In the first, fenazaquin was administered by gavage to rats at doses of 0, 1, 3, 10 or 30 mg/kg bw per day. The NOAEL was 10 mg/kg bw per day based on decreased body weight, body weight gain and feed consumption at 30 mg/kg bw per day.

In the second 90-day study in rats, fenazaquin was administered in the diet at 0, 15, 45, 150 or 450 ppm (equal to 0, 1.0, 3.0, 9.6 and 28.7 mg/kg bw per day for males and 0, 1.2, 3.5, 11.5 and 33.0 mg/kg bw per day for females, respectively). The NOAEL was 150 ppm (equal to 9.6 mg/kg bw per day) based on decreased body weight, body weight gain and feed consumption at 450 ppm (equal to 28.7 mg/kg bw per day).

In a 90-day oral toxicity study in dogs, fenazaquin was administered by gavage at 0, 1, 5 and 15 mg/kg bw per day. The NOAEL was 5 mg/kg bw based on decreased body weights and feed consumption at 15 mg/kg bw.

In a 12-month oral toxicity study in dogs, fenazaquin was administered by gavage at 0, 1, 5 and 12 mg/kg bw per day. The NOAEL was 5 mg/kg bw per day based on decreased body weight gain and feed consumption at 12 mg/kg bw.

The overall NOAEL for oral toxicity in dogs was 5 mg/kg bw per day.

In a 78-week carcinogenicity study in hamsters, fenazaquin was administered by gavage at 0, 2, 15, 30 (for males) and 35 mg/kg bw per day (for females). The Meeting considered the data on females insufficiently reliable to assess toxicity and carcinogenicity because of the high mortality of controls. The study was also considered confounded due to amyloidosis and the long-term use of vancomycin hydrochloride to treat a serious *Clostridium difficile* infection. The NOAEL for systemic toxicity in male hamsters was 2 mg/kg bw per day based on lower body weights and decreased body weight gain at 15 mg/kg bw per day. The NOAEL for carcinogenicity in male hamsters was 30 mg/kg bw per day, the highest dose tested.

In a combined chronic toxicity and carcinogenicity study in rats, fenazaquin was administered in the diet at 0, 10, 100, 200 or 400 (males)/450 (females) ppm (equal to 0, 0.46, 4.5, 9.2 and 18.3 mg/kg bw per day for males and 0, 0.57, 5.7, 11.5 and 25.9 mg/kg bw per day for females, respectively) for 24 months. The NOAEL for long-term toxicity was 100 ppm (equal to 4.5 mg/kg bw) based on significantly decreased body weight gain at 200 ppm (equal to 9.2 mg/kg bw). The Meeting considered the changes in hepatocellular atypia (currently termed basophilic altered focus) of uncertain significance as the incidence of this lesion is highly variable in this strain and no other pathological changes were observed. The NOAEL for carcinogenicity in rats was 18.3 mg/kg bw per day, the highest dose tested.

The Meeting concluded that fenazaquin is not carcinogenic in rats and male hamsters.

Fenazaquin was tested for genotoxicity in an adequate range of in vitro and in vivo assays. It gave negative or equivocal responses in most of the in vitro studies and was negative in the in vivo studies.

The Meeting concluded that fenazaquin is unlikely to be genotoxic in vivo.

In view of the lack of genotoxicity in vivo and the absence of carcinogenicity in rats and male hamsters, the Meeting concluded that fenazaquin is unlikely to pose a carcinogenic risk to humans.

In a two-generation reproductive toxicity study in rats, fenazaquin was administered by gavage at 0, 1, 5 or 25 mg/kg bw per day. The NOAEL for parental toxicity was 5 mg/kg bw per day based on decreased body weight gain and body weights and clinical signs at 25 mg/kg bw per day. The NOAEL for effects on reproduction and on offspring was 25 mg/kg bw per day, the highest dose tested.

In a developmental toxicity study in rats, fenazaquin was administered by gavage at 0, 3, 10 and 40 mg/kg bw per day. The NOAEL for maternal toxicity was 10 mg/kg bw per day based on decreased body weight gain and feed consumption at 40 mg/kg bw per day. The NOAEL for embryo/fetal toxicity was 40 mg/kg bw per day, the highest dose tested. There was no evidence of teratogenicity.

In a developmental toxicity study in rabbits, fenazaquin was administered by gavage at 0, 3, 13 and 60 mg/kg bw per day. The NOAEL for maternal toxicity was 13 mg/kg bw per day based on lower feed consumption on gestation days 6–12 at 60 mg/kg bw per day. The NOAEL for embryo/fetal toxicity was 60 mg/kg bw per day, the highest dose tested. There was no evidence of teratogenicity.

The Meeting concluded that fenazaquin is not teratogenic.

In an acute neurotoxicity study in rats, a single dose of fenazaquin was administered by gavage at 0, 20, 65 or 130 mg/kg bw in males and 0, 20, 60 and 120 mg/kg bw in females. The

NOAEL for acute neurotoxicity in rats was 120 mg/kg bw, the highest dose tested. The NOAEL for systemic toxicity was 20 mg/kg bw based on decreased body weight and feed consumption at 60 mg/kg bw.

The Meeting concluded that fenazaquin is not neurotoxic..

In an immunotoxicity study in rats, fenazaquin was administered by gavage at 0, 15, 30 or 37.5/45 (from day 8) mg/kg bw per day for 28 days. The NOAEL for immunotoxicity was 37.5 mg/kg bw, the highest dose tested.

The Meeting concluded that fenazaquin is not immunotoxic.

Toxicological data on metabolites and/or degradates

4-Hydroxyquinazoline (4-OH)

4-OH is a minor metabolite in rats and a plant metabolite. Oral LD_{50} of 4-OH in rats was 300 mg/kg bw,

In a 4-week oral toxicity study in rats administered 4-OH at 0, 10, 30 and 100 mg/kg bw by gavage, the NOAEL was 30 mg/kg bw per day based on slight reductions in body weight gains at 100 mg/kg bw per day.

4-OH was negative in an in vitro bacterial reverse mutation assay.

Based on the available data and comparisons of toxicity with the parent fenazaquin, the Meeting concluded that 4-OH was unlikely to be of greater toxicity than fenazaquin. The Meeting concluded that the ADI and ARfD of fenazaquin can be applied to 4-OH.

Tertiary butylphenylethanol (TBPE)

The oral and dermal LD_{50} of TBPE, a plant metabolite, was greater than 2000 mg/kg bw in rats. TBPE was a slightly irritating to the skin and severely irritating to the eye of rabbits. TPBE was not sensitizing in the skin of guinea-pigs.

In a 4-week oral toxicity study in rats administered TBPE at 0, 20 150 and 400 mg/kg bw per day, the NOAEL was 20 mg/kg bw per day based on toxic effects on the kidney, testis and liver at 150 mg/kg bw per day.

TBPE was not genotoxic in vitro and in vivo.

Based on the available data and comparisons of toxicity with the parent compound, the Meeting concluded that TBPE was unlikely to be of greater toxicity than fenazaquin. The Meeting concluded that the ADI and ARfD of fenazaquin can be applied to TBPE.

Fenazaquin dimer

The Meeting concluded that a fenazaquin dimer, a photoproduct, was unlikely to be of toxicological concern given its higher molecular weight leading to expected low absorption.

2-Hydroxyfenazaquin acid (F-3)

Although no toxicity studies on the livestock metabolite F-3 were submitted, it is present as a major metabolite in rat faeces. It is therefore covered by studies of the parent compound, fenazaquin.

2-Oxy-fenazaquin

Although no toxicity studies on 2-oxy-fenazaquin, a plant and soil metabolite, were submitted, oxidative metabolism and the presence of F-3, a faecal metabolite in rats, suggests the formation of 2-oxy-fenazaquin in the rat. The toxicity of 2-oxy-fenazaquin should therefore be covered by studies of the parent compound, fenazaquin.

Human data

There were no reports of fenazaquin-associated adverse effects in manufacturing plant personnel.

The Meeting concluded that the existing database on fenazaquin was adequate to characterize the potential hazards to the general population, including fetuses, infants and children.

Toxicological evaluation

The Meeting established an ADI for fenazaquin of 0–0.05 mg/kg bw based on the NOAEL of 4.5 mg/kg bw per day for decreased body weight gains in males at 9.2 mg/kg bw per day in a combined chronic toxicity and carcinogenicity study in rats. A safety factor of 100 was applied.

This ADI was supported by the overall NOAEL of oral toxicity studies in dogs and the NOAEL of the two-generation reproductive toxicity study in rats.

The lowest NOAEL in the database was 2 mg/kg bw per day for decreased body weight gains at 15 mg/kg bw per day in male hamsters in an 18-month carcinogenicity study. The Meeting noted that the lower LOAEL of 9.2 mg/kg bw per day in the database was obtained from the long-term study in rats, the species most sensitive to fenazaquin toxicity. The Meeting concluded the long-term study in rats was more appropriate for establishing the ADI for fenazaquin.

The Meeting established an ARfD for fenazaquin of 0.1 mg/kg bw based on the NOAEL of 10 mg/kg bw per day for decreased body weight gains at 40 mg/kg bw per day early in a rat developmental toxicity study. A safety factor of 100 was applied.

The Meeting concluded that the ADI and ARfD for fenazaquin could be applied to TBPE and 4-OH.

A toxicological monograph was prepared.

Levels relevant to risk assessment of fenazaquin

Species	Study	Effect	NOAEL	LOAEL
Hamster	Eighteen-month study of toxicity and carcinogenicity[a]	Toxicity	2 mg/kg bw per day	15 mg/kg bw per day
		Carcinogenicity	30 mg/kg bw per day[b] (males only)	–
Rat	Two-year study of toxicity and carcinogenicity [c]	Toxicity	100 ppm, equal to 4.5 mg/kg bw per day	200 ppm, equal to 9.2 mg/kg bw per day
		Carcinogenicity	400 ppm, equal to 18.3 mg/kg bw per day[b]	–
	Two-generation study of reproductive toxicity[a]	Reproductive toxicity	25 mg/kg bw per day[b]	–
		Parental toxicity	5 mg/kg bw per day	25 mg/kg bw per day
		Offspring toxicity	25 mg/kg bw per day[b]	–
	Developmental toxicity study [a]	Maternal toxicity	10 mg/kg bw per day	40 mg/kg bw per day
		Embryo/fetal toxicity	40 mg/kg bw per day[b]	–

	Acute neurotoxicity study[a]	Neurotoxicity	120 mg/kg bw per day[b]	–
Rabbit	Developmental toxicity study[a]	Maternal toxicity	13 mg/kg bw per day	60 mg/kg bw per day
		Embryo/fetal toxicity	60 mg/kg bw per day[b]	–
Dog[e]	Ninety-day and 1-year studies of toxicity[c,d]	Toxicity	5 mg/kg bw per day	12 mg/kg bw per day

[a] Gavage administration.

[b] Highest dose tested.

[c] Dietary administration.

[d] Two or more studies combined.

Estimate of acceptable daily intake (ADI; applies to fenazaquin, TBPE and 4-OH)

0–0.05 mg/kg bw

Estimate of acute reference dose (ARfD; applies to fenazaquin, TBPE and 4-OH)

0.1 mg/kg bw

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 fenazaquin

Absorption, distribution, excretion and metabolism in mammals	
Rate and extent of oral absorption	60% absorbed; T_{max} within 8 h at low dose
Dermal absorption	<1%
Distribution	Widely distributed
Potential for accumulation	No indication of accumulation in tissues
Rate and extent of excretion	Rapidly excreted (>90% within 48 h at low dose)
Metabolism in animals	Oxidation + hydroxylation
Toxicologically significant compounds in animals and plants	Fenazaquin, TBPE, 4-OH
Acute toxicity	
Rat, LD_{50}, oral	134 mg/kg bw
Rabbit, LD_{50}, dermal	>5 000 mg/kg bw[a]
Rat, LC_{50}, inhalation	>1.9 mg/L
Rabbit, dermal irritation	Non-irritating
Rabbit, ocular irritation	Non-irritating
Guinea-pig, dermal sensitization	Non-sensitizing (modified Buehler and Magnusson–Kligman)
Short-term studies of toxicity	
Target/critical effect	Decreased body weight gain (hamster, rat, dog)

Lowest relevant oral NOAEL	5 mg/kg bw per day (dog)
Lowest relevant dermal NOAEL	1 000 mg/kg bw per day (rabbit)[a]
Lowest relevant inhalation NOAEC	No data
Long-term studies of toxicity and carcinogenicity	
Target/critical effect	Decreased body weight gain (rat)
Lowest relevant NOAEL	4.5 mg/kg bw per day (rat)
Carcinogenicity	Not carcinogenic in hamster (male only) or rat[b]
Genotoxicity	
	No evidence of genotoxicity in vivo[b]
Reproductive toxicity	
Target/critical effect	Body weight, clinical signs (rat)
Lowest relevant parental NOAEL	5 mg/kg bw per day (rat)
Lowest relevant offspring NOAEL	25 mg/kg bw per day (rat)[a]
Lowest relevant reproductive NOAEL	25 mg/kg bw per day (rat)[a]
Developmental toxicity	
Target/critical effect	Body weight effect (rat)
Lowest relevant maternal NOAEL	10 mg/kg bw per day (rat)
Lowest relevant embryo/fetal NOAEL	40 mg/kg bw per day (rat)[a]
Neurotoxicity	
Acute neurotoxicity NOAEL	120 mg/kg bw (rat)[a]
Subchronic neurotoxicity NOAEL	No data
Developmental neurotoxicity NOAEL	No data
Other toxicological studies	
Immunotoxicity NOAEL	37.5 mg/kg bw per day (rat)[a]
Studies on toxicologically relevant metabolites	
4-OH	Oral LD_{50} = 300 mg/kg bw (rat) Four-week oral toxicity study NOAEL: 30 mg/kg bw based on effects on body weights at 100 mg/kg bw per day (rat) No genotoxicity in vitro
TBPE	Oral LD_{50}: >2 000 mg/kg bw (rat)[a] Severely irritating to eye Four-week oral toxicity study NOAEL: 20 mg/kg bw based on effects on kidney and liver at 150 mg/kg bw per day (rat) No genotoxicity in vitro and in vivo
Human data	
	–

[a] Highest dose tested.

[b] Unlikely to pose a carcinogenic risk to humans via exposure from the diet.

Summary

	Value	Study	Safety factor
ADI[a]	0–0.05 mg/kg bw	Two-year study of toxicity and carcinogenicity (rat)	100
ARfD[a]	0.1 mg/kg bw	Developmental toxicity study (rat)	100

[a] Applies to fenazaquin, TBPE and 4-OH, expressed as fenazaquin.

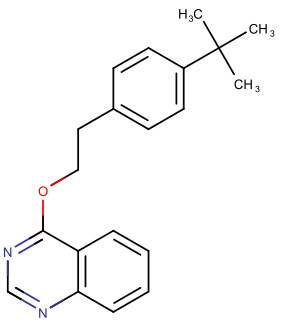
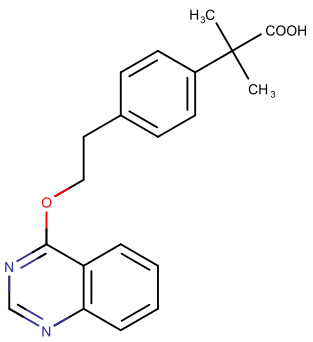
RESIDUE AND ANALYTICAL ASPECTS

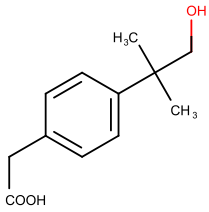
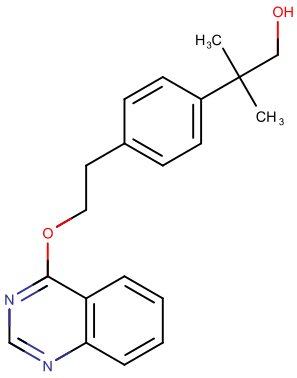
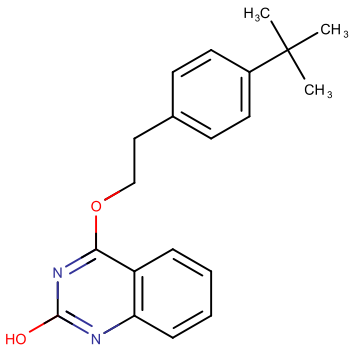
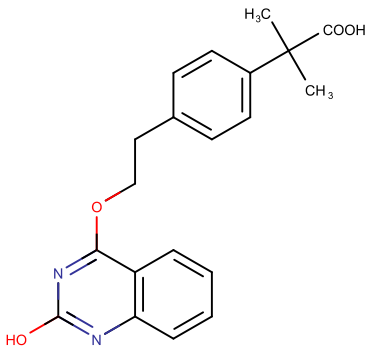
Fenazaquin is a quinazoline insecticide/acaricide which exhibits contact and ovicidal activity against a broad spectrum of mites in grapes, pome fruit, citrus, peaches, cucurbits, tomatoes, cotton and ornamentals. At the 48th Session of the CCPR (2016), it was scheduled for evaluation as a new compound by the 2017 JMPR.

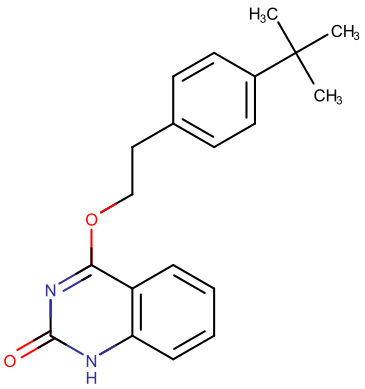
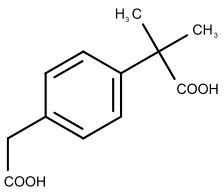
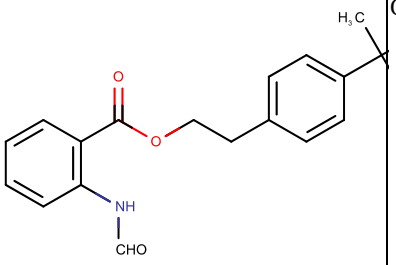
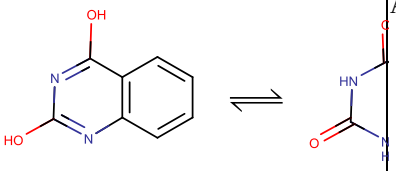
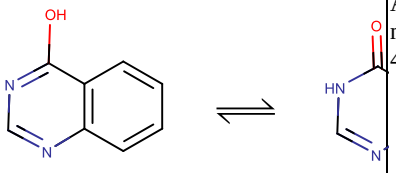
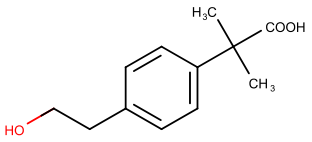
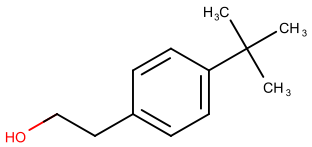
The Meeting received information on the metabolism of fenazaquin in apples, oranges, grapes and corn, lactating goats and laying hens, methods of residue analysis, freezer storage stability, GAP information, supervised residue trials on stone fruits, pineapples, tree nuts (almonds and pecans), hops and processing studies.

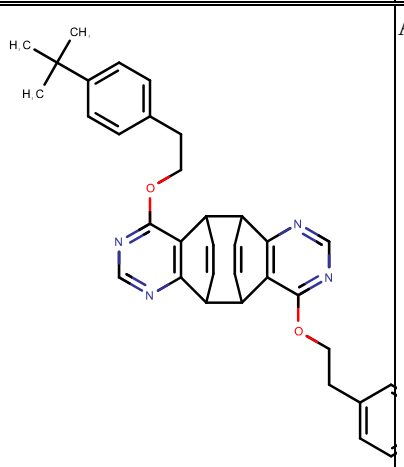
In this document, the common names, chemical structures and chemical names of the metabolites were as follows:

List of metabolites and degradates of fenazaquin

Common name	Chemical name	Chemical structure	Occurrence in
Fenazaquin	4-[2-(4-tert-Butyl-phenyl)-ethoxy]-quinazoline		Apples, oranges, grapes, maize, rat, goats, hens, soil
Fenazaquin acid	2-methyl-2-{4-[2-(quinazolin-4-yloxy)ethyl]phenyl}propanoic acid		Maize, grapes, rat (faeces, F-2), hen, soil

List of metabolites and degradates of fenazaquin			
Common name	Chemical name	Chemical structure	Occurrence in
Metabolite B	2-[4-(1-hydroxy-2-methylpropan-2-yl)phenyl]acetic acid		Rat (urine, AN-1)
Metabolite C	2-Methyl-2-{4-[2-(quinazolin-4-yloxy)-ethyl]-phenyl}-propan-1-ol		Apples, grapes, rat (faeces, F-1)
2-Hydroxy-fenazaquin	4-[2-(4-tert-butylphenyl)ethoxy]quinazolin-2-ol/ Exists in tautomeric equilibrium with 2-oxy-fenazaquin		Maize, goats, soil
2-Hydroxy-fenazaquin acid	2-(4-{2-[(2-hydroxyquinazolin-4-yl)oxy]ethyl}phenyl)-2-methylpropanoic acid		Apples, oranges, goats, rats (faeces, F-3)

List of metabolites and degradates of fenazaquin			
Common name	Chemical name	Chemical structure	Occurrence in
2-Oxy-fenazaquin	4-[2-(4-tert-butylphenyl)ethoxy]-1,2-Dihydroquinazolin-2-one Exists in tautomeric equilibrium with 2-hydroxy-fenazaquin		Maize, soil
Metabolite G	2-[4-(carboxymethyl)phenyl]-2-methylpropanoic acid		Goats, soil
Metabolite H	2-(4-tert-butylphenyl)ethyl 2-formamidobenzoate		Grapes, soil
Dihydroxyquinazoline	Equilibrium exists between the two tautomeric forms: quinazoline-2,4-diol and 1,2,3,4-tetrahydroquinazoline-2,4-dione		Apples, grapes
4-Hydroxyquinazoline	Equilibrium exists between the two tautomeric forms: quinazoline-4-ol and 3,4-dihydroquinazolin-4-one		Apples, grapes, maize, rats (urine, 4-OH), goats, soil
Metabolite K	2-[4-(2-hydroxyethyl)phenyl]-2-methylpropanoic acid		Grapes
Tertiarybutylphenylethanol (TBPE)	2-(4-tert-butylphenyl)ethan-1-ol		Apples, grapes, maize, soil

List of metabolites and degradates of fenazaquin			
Common name	Chemical name	Chemical structure	Occurrence in
Fenazaquin Dimer	7,15-bis[2- (4- tert-butylphenyl)ethoxy]-4,6,12,14-tetraazapentacyclo[8.6.2.2^{2,9}.0^{3,8}.0^{11,16}]icosa-3(8),4,6,11(16),12,14,17,19-octaene		Apples

Plant metabolism

Apples

Study 1

Four semi-dwarf *Golden Delicious* apple trees, grown outdoors, were treated with a single foliar application of ^{14}C-fenazaquin labelled in the phenyl ring of the tert-butyl phenyl portion of the molecule (P-Label) and with ^{14}C-fenazaquin uniformly labelled in the phenyl portion of the quinazoline ring (Q-Label). Two trees (one per radiolabel) were sprayed at a rate of 0.45 kg ai/ha in late June when the apples were 2–3 cm in diameter (early season application), while the remaining two trees were sprayed at the same rate approximately 4–5 weeks prior to harvest when the apples were 6–7 cm in diameter (late season application).

In order to study the effect of photolysis on the decline of the fenazaquin residues, six apples on the tree receiving the late season application of the P-label were covered shortly after application with bags made of a white muslin cloth.

Apples from trees receiving the early season application were sampled at 0, 4, 7, 14, 29, 57, and 92 days after treatment (DAT), while apples from trees receiving the late season application were sampled at 0, 7, 14, 28, and 42 DAT. The covered apples from the photolysis study were sampled at 7 and 14 DAT.

The ^{14}C levels in the peel, pulp, and whole fruit of apples from the late season application (1.92–2.47 mg eq/kg, 0.05–0.06 mg eq/kg, 0.37–0.49 mg eq/kg, respectively) were up to 3 times greater than the levels in the corresponding fractions of apples receiving the early season application (0.65–0.80 mg eq/kg, 0.03 mg eq/kg, 0.14–0.16 mg eq/kg, respectively). The radioactivity in peel was consistently higher than that in pulp, demonstrating limited penetration of the radioactivity.

Once harvested, fruits were rinsed with various organic solvents, The levels of fenazaquin in the surface washes declined relatively rapidly from 85–90% of the TRR on day 0 to 2–6% of the TRR on day 92. The decline of residues in the washes appeared to be associated with a corresponding increase in peel and pulp residues. In apples that were covered from the sun, the decline in fenazaquin residues was insignificant as was the accumulation of residues in pulp and peel. Photolysis appears to be a major route of degradation of fenazaquin.

Characterization of the residues in peel of washed mature fruit showed 30–53% of the TRR to be extracted using acetonitrile:water (75:25, v:v) with 38–57% of the TRRs remaining unextracted. Analysis of the peel extracts, showed fenazaquin to be the major component (up to 20% of the total peel residue at harvest). No metabolites with the intact ether bridge were present at measurable levels in the peel. All of the metabolites found were confirmed as being cleavage products of fenazaquin. Since cleavage of the fenazaquin ether bridge is known to occur as a result of photolysis, it was assumed that most of these metabolites were photoproducts. A total of six or more cleavage products were observed in the peel extracts, with 4-hydroxyquinazoline and TBPE tentatively identified as the two major cleavage constituents (2–5% of the TRR). All of the other observed cleavage metabolites accounted for <3% of the TRR. Therefore, the structure of these could not be elucidated further. The unextracted residues in peel appeared to be cleavage products which had been incorporated into natural products.

Characterization of the residues in apple pulp showed 14–40% of the TRR to be extracted using acetonitrile:water (75:25, v:v) with 5–13% of the TRR being unextracted. Analysis of the extracted residues showed no presence of fenazaquin in the pulp. All of the observed metabolites were confirmed as being cleavage products, most of which were different from those observed in the peel. No individual metabolite in the pulp represented more than 3% of the whole fruit residue. As with the peel metabolites, none of the pulp metabolites were present at levels high enough to facilitate isolation and identification.

Study 2

Fenazaquin, labelled in the phenyl ring (P-Label) or in the quinazoline ring (Q-Label), and formulated as a suspension concentrate, was applied to *Golden Delicious* apple trees, maintained outdoor, at total seasonal application rates of 3.3 g ai/hL or 13.3 g ai/hL. The first application was made to apple trees when fruits had reached an average size of 2 cm in diameter.

Fruits were harvested on the day of application, 7, 14, 28 days after treatment (DAT) and at maturity (105 DAT). Five weeks later, a separate cluster of apple trees were treated at the low and high application rates. Apples were harvested immediately after treatment (0-DAT) and at maturity (70 DAT). In a separate experiment, a single application of P-labelled fenazaquin was made to one tree at the low rate. Following treatment, the fruit were covered with aluminum foil-lined plastic plant pots, the open end being covered with mesh to exclude light but allow air exchange.

In all scenarios, the total radioactivity in fruits declined relatively rapidly over the duration of the study.

Following the early season application, the TRRs removed by surface washing were greater than 50% of the TRR at all sampling intervals except at 105 DAT where surface washing removed 20–40% of the TRR from fruit treated at the low rate. Similarly, at the 70-DAT interval following the higher application rate, 26–35% of the TRR were removed by surface washing. After the surface washes, most of the radioactivity remaining in the fruit was found in the peel for both the low and high rates, following early and late season applications, demonstrating limited penetration.

Fenazaquin accounted for greater than 92% of the TRR in whole fruits collected 0-DAT and declined rapidly thereafter following low and high rates of application made early season and late season. Starting on day 7, the fenazaquin dimer was a major metabolite accounting for 7–33% of the TRR, following early and late season applications at the low and high rates. The Metabolites C/TBPE, which could not be separated either by HPLC or TLC, accounted for up to 2.3% of the TRR for both radiolabels at the low and high rates following early and late season applications. As Metabolites C and TBPE were present predominantly in the P-labeled samples, the Meeting concluded that it likely represents TBPE. Several other unknown metabolites were characterized, none of which exceeded 5% of the TRR.

For the mature 14 DAT covered fruits, surface residues were comprised solely of fenazaquin, and peel extracts were fenazaquin and Metabolites C/TBPE. The absence of the dimer on these samples tends to confirm that it is a product of photochemistry.

Oranges

^{14}C-Phenyl-fenazaquin (P-label) and ^{14}C-quinazoline-fenazaquin (Q-label) were formulated as emulsifiable concentrates (EC) and were separately applied at 0.45 kg ai/ha to orange trees, variety *Valencia*, grown outdoors. Applications were made approximately 6 months prior to fruit maturity (early season application, when fruit were 3 cm in diameter) and approximately 2 months prior to fruit maturity (late season application when fruit were 6 cm in diameter).

Fruits were collected at 0, 28, 112 and 191 days after treatment (DAT) following the early season application and at 0, 19 and 63 DAT following the late season application.

In an effort to examine the effect of photolysis on the nature of the residues, some of the fruit from the late season application were covered with muslin cloth shortly after the application of fenazaquin. Samples of covered fruit were collected at 9, 19 and 63 DAT.

Following the early season application, the TRRs in whole fruit averaged 2.3 mg eq/kg at 0 DAT and declined rapidly to an average of 0.77 mg eq/kg by 28 DAT, and then declined more slowly to an average of 0.34 mg eq/kg by fruit maturity (191 DAT). Following the late-season application, TRRs in whole fruit were 0.53 mg eq/kg at 0 DAT and remained relatively unchanged (P label), or increased slightly (Q label), until maturity, at 63 DAT (0.45–0.90 mg eq/kg). Regardless of the application timing or sampling interval, the two radiolabels yielded similar TRRs. There was no apparent decline in TRRs in the covered fruit from the late season application.

The distribution of the TRRs between peel and pulp fractions was similar for mature fruits for both ^{14}C-labels. For the early and late season applications, the peel accounted for 86–99% of the TRR in mature fruits, and the pulp accounted for less than 14% of the TRR. These findings confirmed that there was limited penetration from the peel into the pulp.

Analyses of surfaces washes and solvent extracts of the mature fruit (191 DAT) from the early season application demonstrated that parent, fenazaquin, was the major residue in the fruit (39–52% of the TRR; 0.13–0.19 mg eq/kg). Minor amounts of 2-hydroxy-fenazaquin (5–8% of the TRR; 0.02–0.03 mg eq/kg) were also identified with the remaining radioactivity comprised largely of unknowns, each present at < 5% TRR.

For the late season application, TLC analyses of solvent washes and fruit extracts of mature fruit (63 DAT), identified parent as the primary residue (55–66% of the TRR; 0.30–0.50 mg eq/kg), along with minor amounts of 2-hydroxy-fenazaquin (≤ 1% of the TRR; ≤0.008 mg eq/kg).

For the mature covered fruits (late season application; 63 DAT), the concentrations of the parent compound were higher (81–84% TRR; 0.15–0.46 mg eq/kg) than in the uncovered fruit. Levels of 2-hydroxy-fenazaquin in the mature covered fruit were comparable to those in the uncovered mature fruits (≤1% of the TRR; 0.001–0.006 mg eq/kg). The limited degradation of the parent molecule that occurred in/on covered fruits suggests that photolysis of surface residues plays an important role in the degradation of fenazaquin residues on the surface of these fruits.

Grapes

Field grown grapes, variety *Cabernet Sauvignon,* were treated with single applications of an EC formulation of [^{14}C]fenazaquin labelled in the phenyl ring of the molecule (P-label) or in the quinazoline portion of the molecule (Q-label) at 15 g ai/hL. An early season application was made to grape bunches approximately two to three weeks after the end of flowering (BBCH 68). A late season application was made to separate grape bunches approximately 7 weeks later (nine to ten weeks after the end of flowering). To facilitate the characterisation of any fenazaquin metabolites, applications at 150 g ai/hL were also made to 10 grape bunches spread over three to five vines at the late application time point with both P- and Q-labelled fenazaquin.

Grape bunches from the early season application were sampled at 0, 49 and 76 days after treatment (DAT) while grape bunches from the late season application were sampled at 0 and 28 DAT.

To determine the potential for fenazaquin and its metabolites to translocate from the site of application, individual branches on two separate vines were treated with ^{14}C-fenazaquin at the time of the early season application.

Following early season application, the levels of radioactivity, removed following the surface washes decreased from 78–81% of the TRR of day 0 to 29–34% of the TRR on day 76. Conversely, the amount of radioactivity extracted using acetonitrile:water (9:1, v/v) increased with increased duration from 16–18% of the TRR on day 0 to 39–45% of the TRR on day 76 as did the radioactivity in unextracted residues, from 3.2–5.0% of the TRR on day 0 to 22–32% of the TRR on day 76. No distinct differences between P- and Q-labelled fenazaquin were observed.

Characterisation of the residues in the surface washes of samples collected following the early season application showed the major component was fenazaquin, ranging from 24–29% of the TRR (0.22–0.31 mg eq/kg). Analysis of the extracts showed the presence of three metabolites derived from the intact fenazaquin molecule: fenazaquin acid, Metabolite C and Metabolite H. The remaining metabolites were products formed as a result of the cleavage of the ether bridge linking the quinazoline and the ethylphenylbutyl portions of the molecule. These minor metabolites, identified as dihydroxyquinazoline, 4-hydroxyquinazoline, Metabolite K and TBPE, each accounted for 1–6% of the TRR (0.004–0.06 mg eq/kg). A total of 8 unknown metabolites were characterized, two of which contained both the P- and Q-labels, four contained only the P-label and two included the Q-label only. In total, unknowns accounted for 6–9% of the TRR (0.06–0.09 mg eq/kg).

Following the late season application (28 DAT), the radioactivity recovered in the surface washes ranged from 61–71% of the TRR, corresponding to lower radioactivity in the extracts (21–27% of the TRR) and unextracted residues (8–12% of the TRR). Fenazaquin was the predominant residue in the P- and Q-labelled surface washes (46–64% of the TRR; 0.96–1.19 mg eq/kg). Five metabolites were also identified: Metabolite C, Metabolite H, 4-hydroxyquinazoline, dihydroxyquinazoline, and TBPE) in amounts of 0.4 to 3% of the TRR (0.01 to 0.07 mg eq/kg).

In the extracts, the parent fenazaquin, TBPE, dihydroxyquinazoline and 4-hydroxyquinazoline were detected, each accounting for < 6% of the TRR (< 0.16 mg eq/kg).

The translocation experiment showed that, following application of fenazaquin to branches, no radioactivity was found in grape bunches despite the measurable TRRs in the sprayed branches (10 mg eq/kg), thus confirming that translocation of fenazaquin and/or any degradation products beyond the site of application did not occur.

Treatment of grape bunches at the higher application rate (150 g ai/hL) were included in the experiment to assist in the identification of metabolites. The distribution of radioactivity in the higher rate samples was similar to that observed with the late season application samples with surface washes removing up to 60% of the TRR. The radioactivity in the washed fruit samples accounted for 40–44% of the TRR. Analysis of the surface washes showed the presence of the parent compound, fenazaquin which represented 38% of the TRR and a number of unknown components, each accounting for less than 4% of the TRR. Analysis of the extracts showed the presence of more components than in the washes but fenazaquin was still the predominant analyte, accounting for an average of 14% of the TRR.

An additional experiment was conducted to assess whether the radioactivity was mainly associated with the grape skin. Grapes were peeled and the peel and pulp analysed separately. The results showed that 68% of the TRR was associated with the grape skin confirming that the predominant route of fenazaquin degradation is due to photolytic breakdown as the majority of the radioactivity had not yet reached the pulp tissue where metabolic processes are more likely to occur.

Maize

Applications of [^{14}C]Fenazaquin labelled in the phenyl ring (P-label) and in the quinazoline portion (Q-label) as aqueous suspensions were made at an average rate of 0.55 kg ai/ha by foliar application to maize (*Zea mays*) plants at the soft dough stage of development. The plants were grown in above

ground wooden boxes located outdoors. Maize grain (corn-on-the-cob) and stover were harvested 20 DAT.

Acetonitrile extraction of the Q-label maize grain released 46% of the TRR, with fenazaquin identified as the major residue (23% of the TRR, 0.003 mg eq/kg). Fenazaquin dimer was also found at 7.7% of the TRR (0.001 mg eq/kg).

Extraction of the stover with acetonitrile:water followed by tetrahydrofuran released a minimum of 94% of the TRR . The major components identified in these combined extracts were fenazaquin (30–49% of the TRR; 2.0–3.0 mg eq/kg) and fenazaquin dimer (20–54% TRR; 1.2–3.6 mg eq/kg)). Minor components identified included 2-hydroxy-fenazaquin (0.5–1.0% of the TRR; 0.03–0.07 mg eq/kg)), TBPE (2% of the TRR; 0.12 mg eq/kg), fenazaquin acid (0.5% of the TRR; 0.03 mg eq/kg) and 4-hydroxyquinazoline (7% of the TRR; 0.42 mg eq/kg).

Fenazaquin did not readily translocate, as evidenced by the low levels of radioactive residue in the grain as compared to the higher levels in stover. The major route of transformation was conversion to the fenazaquin dimer. The presence of the minor identified metabolites, 4-hydroxyquinazoline and TBPE, suggested cleavage of the ether linkage. The intact fenazaquin appeared to have also been oxidized on the quinazoline ring to yield an alcohol or oxidized on the tert-butyl group to yield a carboxylic acid.

In summary, the metabolism of fenazaquin in fruits and cereals is well understood. The majority of the radioactive residues were located on the surface of the crops with limited penetration from the peel to the pulp. Furthermore, there was no evidence of translocation from the site of application to the untreated parts of the plant. While photolysis was the major route of degradation, the metabolic pathways of fenazaquin include: 1) formation of the dimer; 2) cleavage of the ether linkage to form 4-hydroxyquinazoline and TBPE; 3) oxidation of the quinazoline ring to yield 2-hydroxy-fenazaquin or oxidation on the tert-butyl group to yield fenazaquin acid followed by subsequent hydroxylation of these various metabolites. The metabolites, fenazaquin acid and 2-hydroxy-fenazaquin acid were identified as major metabolites in rats while 4-hydroxyquinazoline was a minor metabolite.

Animal metabolism

Lactating goats

The metabolism of fenazaquin was investigated in two lactating goats (*Capra hircus,* Alpine breed), weighing on average 42 kg, dosed orally once daily for 5 consecutive days, by a balling gun, with [^{14}C-quinazoline]fenazaquin (Q-label) or [^{14}C-phenyl]fenazaquin (P-label) at a dose level of 34 mg/day equivalent to 14 ppm feed. At sacrifice (within 22 hours after the final dose) samples of liver, kidney, muscle (loin and leg), fat (omental and perirenal), blood, bile and GI tract were collected.

The major route of elimination of the radioactivity was via the feces which accounted for 64–91% of the total administered dose (AD), while urine accounted for approximately 5% of the AD and milk accounted for 0.1% of the AD. The tissue burden was low (<1% of the AD). The overall recovery of the administered dose averaged 94% for both radiolabels.

The total radioactive residues (TRR) were highest in liver (0.41–0.79 mg eq/kg) followed by fat (0.09–0.12 mg eq/kg for composite fat and 0.07–0.15 mg eq/kg for P-label renal, omental and subcutaneous fat), kidney (0.04–0.09 mg eq/kg) and muscle (0.005 mg eq/kg for Q-label composite muscle and 0.007–0.03 mg eq/kg for P-label flank and loin muscle). The tissues from goats administered the [^{14}C-phenyl]fenazaquin had consistently higher concentrations of radioactive residues.

Greater than 88% of the TRR in milk was extracted following sequential extraction with acetonitrile and acetonitrile:water (1:1, v:v). Fenazaquin was detected as a predominant component of the residue in both P-label and Q-label milk samples (15–47% of the TRR; 0.004–0.02 mg eq/kg). 4-Hydroxyquinazoline (23% of the TRR; 0.006 mg eq.kg) was found in the Q-label milk sample only.

The phenyl label sample was separated into milk fat and skim milk before analysis. The fenazaquin in this sample was found almost entirely in the milk fat portion of the whole milk sample.

Liver samples from both goats were extracted twice with acetonitrile:water (1:1), and once with acetonitrile. The solids remaining after the acetonitrile:water extraction of the Q-label liver sample were further extracted with methanol:water. The combined extracts accounted for 42–61% of the TRR. While fenazaquin was not detected in either sample of liver, 2-hydroxy-fenazaquin acid was the predominant residue representing 14–15% of the TRR (0.05–0.11 mg eq/kg). 4-Hydroxyquinazoline was the only metabolite identified in the hydrolyzed quinazoline PES sample (9% of the TRR; 0.04 mg eq/kg), while in the acid hydrolysate from the P-label sample, only Metabolite G and other unidentified components were detected, yet none exceeded 6% of the TRR.

The kidney samples were extracted in a similar manner to liver samples, with 88–89% of the TRR being extracted. No fenazaquin was detected. The identified metabolites were 2-hydroxy-fenazaquin acid (25–29% of the TRR; 0.01–0.02 mg eq/kg), Metabolite G (9% of the TRR; 0.01 mg eq/kg: P-label only) and 4-hydroxyquinazoline (6% of the TRR; 0.002 mg eq/kg: Q-label only). Unidentified residues accounted for 54% of the TRR (≤ 0.05 mg eq/kg). These accounted for extracted components observed on HPLC that did not match available reference standards (no more than 4 components; none of which exceeded 25% of the TRR [≤ 0.023 mg eq/kg]), unknowns (no more than 6 accounting for a total of 14% of the TRR [0.013 mg eq/kg]) and the remaining methanol:water extract which was not characterized or identified.

The P-label flank muscle sample was extracted twice with acetonitrile:water (1:1) and once with acetonitrile, resulting in 84% of the TRRs being extracted. Fenazaquin was not detected in flank muscle. The predominant metabolite observed was 2-hydroxy-fenazaquin acid (20% of the TRR; 0.005 mg eq/kg). Unidentified residues accounted for 64% of the TRR (≤ 0.02 mg eq/kg). Similar to kidney, these accounted for extracted components observed on HPLC that did not match available reference standards (no more than 3 components none exceeding 36% of the TRR [≤ 0.009 mg eq/kg]), unknowns (no more than 6 accounting for a total of 12% of the TRR [0.003 mg eq/kg]) and the remaining methanol:water extract which was not characterized or identified.

Greater than 92% of the TRR in P- and Q-label composite fat samples were extracted, following two extractions with acetone:hexane (1:4, v:v) and a third extraction with 100% acetone. The primary component of the residue was fenazaquin (77–83% of the TRR; 0.07–0.08 mg eq/kg). A trace of 2-hydroxy-fenazaquin was detected (1% of the TRR; 0.001 mg eq/kg) in both fat samples, and a trace of 2-hydroxy-fenazaquin acid (< 1% of the TRR; < 0.001 mg eq/kg) was detected in the phenyl label sample.

Laying hens

Twenty laying hens (*Gallus gallus domesticus*), weighing on average 1.4 kg, were dosed orally once daily for seven consecutive days with [^{14}C-quinazoline]fenazaquin at a dose level of 12.3 ppm feed, or [^{14}C-phenyl]fenazaquin at a dose level of 12.4 ppm feed. The test substances were administered using cellulose-filled gelatin capsules. The animals were sacrificed approximately 21–23 hours after the last dose and the liver, breast and thigh muscle, omental fat, subcutaneous fat and gastrointestinal tracts (with contents) were collected and pooled by treatment group.

Approximately 90% of the administered dose (AD) was recovered, most of which (an average of 89% of the AD) was excreta-related. Total radioactive residues (TRR) in eggs accounted for 0.04–0.3% of the AD. The tissue burden was low (< 0.1% of the AD). Highest concentrations of radioactivity were found in fat (0.16–0.18 mg eq/kg) followed by liver (0.06–0.1 mg eq/kg) and muscle (0.005–0.07 mg eq/kg).

Acetonitrile:water extracted 74–91% of the TRR in P- and Q-label eggs. The major residues in P-label eggs were fenazaquin (13% of the TRR; 0.003 mg eq/kg) and fenazaquin acid (13% of the TRR; 0.003 mg eq/kg). None of the individual unknowns accounted for more than 4% of the TRR. While 26% of the TRR (0.006 mg eq/kg) remained in the P-label PES, there was insufficient

radioactivity for any further work-up. Fenazaquin and fenazaquin acid were also present in Q-label eggs (2% of the TRR [0.003 mg eq/kg] and 1% of the TRR [0.002 mg eq/kg], respectively), however, benzoyleneurea was the major residue in eggs, accounting for 82% of the TRR (0.12 mg eq/kg).

All of the radioactivity in the omental and subcutaneous fats was readily extracted with acetone and hexane (> 98% of the TRR). The predominant residue was fenazaquin (83–95% of the TRR; 0.134–0.158 mg eq/kg). While no other residue was identified in P-label fats, benzoyleneurea was detected in the Q-label fats (3–15% of the TRR; 0.005–0.02 mg eq/kg). The PES contained < 2% of the TRR.

In the P-label thigh and breast muscles, 75–80% of the TRR were extracted using acetonitrile:water. Fenazaquin (20–69% of the TRR; 0.001–0.01 mg eq/kg) and fenazaquin acid (20% of the TRR; 0.001 mg eq/kg) were the only residues detected. In the Q-label thigh and breast muscles, 72–76% of the TRR was extracted, with benzoyleneurea identified as the major residue (63–70% TRR; 0.04 mg eq/kg) and fenazaquin present at lower concentrations (≤ 5% of the TRR; < 0.003 mg eq/kg).

Extraction with acetonitrile:water and acetonitrile released 40–79% of the TRR in liver. No fenazaquin was detected. Moreover, in the P-label liver, fenazaquin acid was the only residue identified (7% of the TRR; 0.003 mg eq/kg), while in the Q-label liver, benzoyleneurea was the predominant metabolite accounting for 53% of the TRR (0.05 mg eq/kg).

The Meeting concluded that, in all species investigated (goats, hens and rats), the total administered radioactivity was predominantly eliminated in excreta. The metabolic profiles differed quantitatively between the species, yet qualitatively there are no major differences with the exception that the metabolism in goats was more extensive. The routes and products of metabolism were similar across all animals, resulting from oxidation of the tert-butyl group giving rise to the hydroxyl-fenazaquin (goats only), fenazaquin alcohol (postulated), fenazaquin acid and its hydroxylated form. The methylene group adjacent to the oxygen of the ether is also oxidized to release the 4-hydroxyquinazoline that is further oxidized to benzoyleneurea (laying hens only).

Environmental fate

The FAO Manual (FAO, 2016) explained the data requirements for studies of environmental fate. The focus should be on those aspects that are most relevant to MRL setting. For fenazaquin, supervised residue trials were received for foliar spray applications on permanent and semi-permanent crops. Therefore, according to the FAO Manual, only studies on rotational crops (confined, field), aerobic degradation, hydrolysis and soil photolysis are required.

Confined and field rotational crops

Confined/field rotational crop studies were not provided to the Meeting as cherries, pineapples, almonds and hops were considered permanent/semi-permanent crops.

In the future, should maximum residue levels be requested for non-permanent crops, the Meeting recommends that confined and field rotational crop studies should be provided.

Aerobic degradation in soil

The degradation of ^{14}C-fenazaquin, uniformly labelled in the tert-butylphenyl ring (P-label) was studied in four soil types in the laboratory under aerobic conditions. The soils were incubated in the dark at 20 °C. Fenazaquin was applied to each soil subsample at a rate of 0.27 mg ai/kg.

The predominant degradation product in all soils was 2-oxy-fenazaquin which reached a maximum of 9% of the applied radioactivity (AR). Fenazaquin acid reached its maximum concentration (2% of the AR) at 14 DAA. In addition to these two minor metabolites, Metabolites G, H and additional unknowns were also detected at < 3% of the AR by the end of the study.

The half-lives (DT_{50}) determined using single first-order (SFO) kinetics ranged from 48 days to 114 days depending on the soil type. The results of the sterilised control samples showed that the degradation of fenazaquin was mainly microbiological.

The rate of degradation of ^{14}C-fenazaquin was also investigated in a laboratory where ^{14}C-quinazoline labelled (Q-label), ^{14}C-phenyl labelled (P-label) and unlabelled fenazaquin were individually applied to a sandy loam soil at 0.56 g ai/ha. In addition, soil samples maintained in an open system, were treated with the same mixture at an exaggerated application rate (56 g ai/ha) for the structural determination of metabolites. Less than 0.2% of the AR was lost by day 365 as a result of volatilization, indicating that it was not a significant dissipation pathway for fenazaquin or its metabolites. During incubation, the mineralisation to CO_2 continuously increased until the end of incubation ($\leq$ 21% of the AR).

No single degradate exceeded 10% of the AR at the lower application rate, however, at the higher application rate, the major metabolite was 2-oxy-fenazaquin, reaching a maximum level of 8% at day 168. The amount of applied fenazaquin decreased steadily from 99% (day 0) to 13% of the AR (day 336), resulting in a calculated DT_{50} of 51 days, determined using SFO kinetics.

In summary, the degradation of fenazaquin in soil appeared to proceed via hydrolysis of the ether linkage between the quinazoline ring system and the tertiary butyl-phenyl portions of the parent molecule, hydroxylation of the quinazoline ring system, and oxidation of the tertiary butyl group and phenylethyl alcohol, formed following hydrolysis of fenazaquin.

Field dissipation

The dissipation of ^{14}C-fenazaquin in soil under field conditions was investigated at one USA test site. ^{14}C-Quinazoline or ^{14}C-phenyl labelled fenazaquin, formulated as emulsifiable concentrate formulations, were applied to bare soil at an application rate of 0.224 kg ai/ha.

The amount of applied fenazaquin decreased from 93–96% of the AR to 9–11% of the AR by day 112, resulting in DT_{50} values of 34 to 36 days, applying single first order kinetics.

The dissipation of fenazaquin in soil under field conditions was also investigated in Germany and Italy. A suspension concentrate of fenazaquin was applied to bare soil at application rates of 0.15 and 0.20 kg ai/ha, respectively. The amount of applied fenazaquin declined relatively rapidly where the residues of fenazaquin reached non-quantifiable levels (< 0.005 mg/kg) 3 months after application.

The DT_{50} values of fenazaquin were calculated to range from 26–75 days applying 1st-order reaction kinetics.

The Meeting concluded that fenazaquin is moderately persistent in soil.

Photolysis - Soil

The photodegradation of ^{14}C-fenazaquin was investigated under laboratory conditions using a sandy loam soil. Soil samples were treated separately with 40 µg of ^{14}C-quinazoline labelled fenazaquin (Q-label) or ^{14}C-phenyl labelled fenazaquin (P-label). Immediately after dosing, the treated soils were exposed to simulated summer sunlight for up to 30 days.

Fenazaquin decreased from 91–93% to 35–42% of the AR during 30 days of irradiation. In the dark control samples the amount of fenazaquin decreased slightly from 91–95% at day 4 to 68–87% of the AR at day 30. In the Q-labelled study, up to 6 minor degradation products were detected, none of which exceeded 3% of the AR in the irradiated and dark control samples. One major degradation product was 4-hydroxyquinazoline (irradiated samples: 0.4 to 37% of the AR; dark control samples: 1% to 7% of the AR). In the P-labelled study, two degradation products were identified, 4-tert-butylphenylacetic acid and TBPE. The amount of 4-tert-butylphenylacetic acid increased continuously during irradiation to 7.3% of the AR and did not exceed 0.1% in dark control samples. The amount of TBPE increased continuously from 1.4 to 18% of the AR (irradiated samples) and from 2% to 8% of the AR (dark control samples) immediately after application to day 30,

respectively. The metabolite 4-tert-butylstyrene was also detected and accounted for a maximum level of 6% of the AR. No other volatiles were detected in significant amounts.

The half-life of fenazaquin on the soil surface was calculated to be 15 days in summer sunlight.

The Meeting concluded that the photolysis of fenazaquin in soil, under sunlight conditions, was an important degradation pathway. This is further supported by the photolysis experiments of the plant metabolism studies which demonstrated that the degradation of fenazaquin was enhanced in the presence of sunlight.

Photolysis – Aqueous Solution

The photolytic stability of ^{14}C-fenazaquin was studied in a laboratory using distilled water. Stock solutions of ^{14}C-quinazoline and ^{14}C-phenyl labelled fenazaquin in acetonitrile were prepared at a concentration of 2.5 g ai/hL and applied to distilled water, resulting in a nominal concentration of 0.01 g ai/hL. Test vessels containing the test solutions were exposed to simulated summer sunlight for up to 32 days at 25 ± 1 °C.

Three degradation products of fenazaquin were detected in the distilled water of both light exposed and dark control samples and identified as 4-hydroxyquinazoline, TBPE and 4-tert-butylstyrene. In light exposed samples treated with the ^{14}C-quinazoline label, the amount of 4-hydroxyquinazoline increased continuously reaching a maximum level of 32% of the AR after 32 days of exposure. In dark control samples, 4-hydroxyquinazoline reached 9% of the AR after 32 days. In light exposed samples treated with the ^{14}C-phenyl label, the amount of TBPE and 4-tert-butylstyrene increased during exposure time and peaked at 19% and 9% of the AR at days 32 and 28, respectively. In dark control samples, TBPE and 4-tert-butylstyrene reached 11% and 0.2% of the AR after 32 days, respectively.

The DT_{50} value of fenazaquin was calculated to be 15 days.

Similar to the soil photolysis study, fenazaquin degraded rapidly in the presence of sunlight.

In summary, the Meeting concluded that fenazaquin is moderately persistent in soil under field conditions (DT_{50s} ranging from 26–114 days). Under laboratory conditions, the photolytic degradation of fenazaquin in soil was enhanced under sunlight as was its degradation in distilled water.

Methods of analysis

Methods have been reported in the scientific literature for the analysis of fenazaquin in food, including multi-residue methods. These methods do not involve a hydrolysis step, therefore, the measured residue is reported as fenazaquin, *per se*.

The Meeting received the description and validation data for various analytical methods capable of quantifying fenazaquin in plant commodities. All residue analytical methods rely on GC-MS or LC-MS/MS. The typical LOQ achieved for the plant commodities is 0.01 mg/kg. Methods were successfully validated by independent laboratories, demonstrating good reproducibility.

The Meeting also received the description and validation data for an LC-MS/MS analytical method capable of quantifying fenazaquin in animal commodities (muscle, fat, liver, milk and eggs). The LOQ achieved for all animal commodities is 0.01 mg/kg. The method was successfully validated by an independent laboratory.

Stability of pesticide residues in stored analytical samples

The Meeting received storage stability studies on fenazaquin and the fenazaquin dimer in tomatoes, corn (stover, forage and grain), mint and almond nutmeat. Individual samples were fortified with both analytes at a concentration of 0.1 mg/kg/analyte and stored frozen at -25 °C to -10 °C. Samples were taken for analysis at intervals up to 26 months.

Storage stability studies showed that fenazaquin was stable for 7 months in tomatoes (high water), 26 months in maize stover, 6 months in maize forage (high water) and 3.5 months in maize grain (high starch). The fenazaquin dimer was observed to be stable for 1.5 months in tomatoes, 18 months in maize stover, 17 months in maize forage, 3.5 months in maize grain and 7 months in almond nutmeats. The Meeting could not determine the demonstrated storage stability interval for fenazaquin and its dimer in mint or the stability of fenazaquin in almond nutmeats as > 30% dissipation was observed within the first 3 months of storage in these commodities.

Since livestock feeding studies were not provided to the Meeting, neither was a freezer storage stability study for fenazaquin and metabolite residues in animal commodities.

In the future, should maximum residue levels be requested for crops, from which may be derived feed items, that are likely to contribute significantly to the livestock diet, the Meeting recommends that freezer storage stability studies for animal commodities should be provided, if samples of animal matrices in the livestock feeding studies are stored for greater than 30 days.

Definition of the residue

The nature of the fenazaquin residues was investigated in apples, oranges, grapes and maize following foliar application.

The predominant residue in edible parts of the mature food crops was the parent, fenazaquin, accounting for 10–66% of the TRR. The fenazaquin dimer was also observed in mature apples (8–32% of the TRR) and maize grain (8% of the TRR). Several minor metabolites were observed, none of which exceeded 10% of the TRR. Under simulated processing conditions, quinazoline ring-labelled fenazaquin was degraded to 4-hydroxyquinazoline, the major hydrolysis product formed under pasteurisation conditions (pH 4, 90 °C, 20 minutes), accounting for 61% of the applied radioactivity (approximately 2-fold higher than that of the parent).

Confined and field rotational crop studies were not required as the uses being considered by this Meeting are semi-permanent/permanent crops.

Fenazaquin was the only analyte present as a major compound in all tested matrices. Suitable analytical methods are available to analyse the parent compound.

The Meeting considered that fenazaquin was a suitable marker for enforcement of MRLs for fruits and cereal crops.

From the dietary risk perspective, the Meeting considered the potential exposure and toxicity of the fenazaquin dimer and 4-hydroxyquinazoline. Levels of these metabolites in apples, grapes and maize grain, sampled 7–28 DAT, 28 DAT and 20 DAT, respectively, in the metabolism studies, were considered as these encompassed the range of PHIs of the critical GAPs (1–14 days) provided to the Meeting. In the orange metabolism study, fruits were collected 63 and 191 DAT, therefore, this study was not considered further, as the residues of these metabolites would likely overestimate those expected at shorter PHIs. The percentage of the fenazaquin dimer in comparison to the parent in whole apples (7–14 DAT) ranged from 13–57%, and was 27% in maize grain (20 DAT). No dimer was observed in grapes. The metabolite 4-hydroxyquinazoline accounted for 0.5–9% and 4% of the fenazaquin concentrations in whole apples (7–28 DAT) and grapes (28 DAT), respectively. 4-Hydroxyquinazoline was not observed in maize.

While the fenazaquin dimer, a photoproduct, was not observed in rats, the metabolite is unlikely to be readily absorbed due to its high molecular weight and lipophilic properties and is thus not likely to be more toxic than the parent compound. The 4-hydroxyquinazoline metabolite was observed in rats and based on the metabolite-specific toxicity studies, it was determined to be no more toxic than the parent.

In light of the residue profile, measurable residues of the predominant metabolites, fenazaquin dimer and 4-hydroxyquinazoline, are not expected in crops harvested at short PHIs. Considering that these compounds are not more toxic than parent fenazaquin, the Meeting concluded that fenazaquin is the only residue pertinent for assessing dietary risk.

The nature of the fenazaquin residues was investigated in livestock following oral administration of the test substance.

In the lactating goat metabolism study, fenazaquin and 4-hydroxyquinazoline were the predominant residues in milk (15–47% of the TRR and 23% of the TRR, respectively) while fenazaquin was the major residue in fat (77–83% of the TRR). The parent was not detected in liver, kidney or muscle. In these tissues, 2-hydroxy-fenazaquin acid was observed as the major metabolite accounting for 14–29% of the TRR.

In the laying hen metabolism study, fenazaquin was a major residue (13–71% of the TRR) in eggs and tissues (except liver) while fenazaquin acid was the predominant metabolite in eggs and muscle (13–20% of the TRR). In Q-label eggs, fat, muscle and liver, the cleavage product, benzoyleneurea, was the major component (15–82% of the TRR).

Fenazaquin is the predominant residue in milk and fat of lactating goats and is detectable in eggs and all poultry tissues. The parent compound was not detected in kidney, liver and muscle of lactating goats. In these tissues, the 2-hydroxy-fenazaquin acid was identified as the principal metabolite. A valid enforcement analytical method is available for fenazaquin; however, a method for the 2-hydroxy-fenazaquin acid was not provided.

The Meeting concluded that fenazaquin and 2-hydroxy-fenazaquin acid are suitable markers for enforcement of MRLs for livestock commodities.

In lactating goats, 2-hydroxy fenazaquin acid may contribute to the consumer exposure as it accounts for the majority of the TRRs in liver (up to 15% of the TRR [0.11 mg eq/kg]), kidney (up to 29% of the TRR [up to 0.02 mg eq/kg]) and muscle (20% of the TRR [0.005 mg eq/kg]), where no parent is present. In milk, 2-hydroxy-fenazaquin acid was up to 25% of the parent residues while in fat, it accounted for less than 2% of the parent residues. The metabolite 4-hydroxyquinazoline is predominantly found in milk accounting for 1.5-fold the fenazaquin residues. In tissues, 4-hydroxyquinazoline was either not detected or detected at lower levels than those of the metabolite 2-hydroxy-fenazaquin acid. The Meeting concluded that the 2-hydroxy-fenazaquin acid and 4-hydroxyquinazoline metabolites are not likely to be more toxic than the parent fenazaquin.

For laying hens, the Meeting considered the potential exposure and toxicity of the metabolites fenazaquin acid and benzoyleneurea, which were significant residues in poultry matrices. Fenazaquin acid occurred at levels similar to parent fenazaquin in eggs, and muscle. Fenazaquin acid was a minor residue in liver, where no fenazaquin was detected. The Meeting determined that fenazaquin acid is a predominant metabolite in rats and no more toxic than the parent compound. The benzoyleneurea was the major residue in eggs (concentration *ca*. 40-fold greater than parent), muscle (*ca*. 13-fold greater than parent), and liver (*ca*. 50% TRR, 0.05 mg eq/kg; no fenazaquin observed). As the benzoyleneurea is structurally similar to the 4-hydroxyquinazoline, it is not expected to be more toxic than the parent fenazaquin. The Meeting noted that, at this time, there are no fenazaquin-treated poultry feed items; therefore, exposure to fenazaquin-related residues from poultry commodities is not expected.

In summary, based on the above, the Meeting recommended the following residue definition for compliance with MRLs and for dietary risk assessment:

Definition of the residue for plant commodities for enforcement of MRLs and for dietary risk assessment: *fenazaquin*

Definition of the residue for animal commodities for enforcement of MRLs and for dietary risk assessment: *sum of fenazaquin and the metabolites 2-(4-{2-[(2-hydroxyquinazolin-4-yl)oxy]ethyl}phenyl)-2-methylpropanoic acid (2-hydroxy-fenazaquin acid) and quinazoline-4-ol and 3,4-dihydroquinazolin-4-one (4-hydroxyquinazoline), expressed as fenazaquin equivalents.*

The Log Kow of fenazaquin is greater than 5, indicating its propensity to sequester into fat. In lactating goats, the ratio of residues (sum of fenazaquin, 2-hydroxy-fenazaquin acid and 4-hydroxyquinazoline) in fat to muscle was greater than 10-fold. Therefore, the Meeting considered the residue fat soluble.

Results of supervised residue trials on crops

Cherries

In the USA, the critical GAP for fenazaquin on sweet and tart cherries is a single foliar spray application at 504 g ai/ha and a PHI of 3 days.

A total of five independent trials were conducted in the USA on sweet and tart cherries during the 2008 and 2009 growing seasons. All five trials were conducted in accordance with the critical GAP. Residues in samples harvested at the PHI of 3 days were: 0.26, 0.47, 0.56, 0.84, 0.91 mg/kg with the highest analytical result reported at 0.965 mg/kg.

The Meeting estimated a maximum residue level of 2 mg/kg, an HR of 0.965 mg/kg and an STMR of 0.56 mg/kg for fenazaquin on the subgroup of cherries.

While supervised residue trials were provided for peaches and plums, in the absence of a critical GAP for these crops, maximum residue levels could not be estimated.

Pineapples

In the Philippines, the GAP for fenazaquin on pineapples is two foliar spray applications of 1.0 kg ai/ha and a minimum PHI of 14 days.

Only one trial in Costa Rica during the 2013 growing season was conducted in accordance with the Philippines GAP, where the average residue was reported to be 0.085mg/kg

The Meeting could not estimate a maximum residue level in the absence of a sufficient number of trials conducted in accordance with the minimum PHI of 14 days prescribed on the Philippine label.

Almonds

In the USA, the critical GAP for fenazaquin on almonds is a single foliar spray application at 504 g ai/ha and a PHI of 7 days.

A total of five independent trials were conducted in the USA on almonds during the 2008 growing season. All five trials were conducted in accordance with the critical GAP in the USA. Almonds were harvested at the 7-day PHI and residues in almond nutmeats were: < 0.01 (4) and 0.01 mg/kg.

As the supervised field trial residues were not supported by the demonstrated storage stability data for almond nutmeats, the Meeting could not estimate a maximum residue level.

Hops

In the USA, the critical GAP for fenazaquin on hops is a single foliar spray application at 504 g ai/ha and a PHI of 7 days.

A total of seven independent trials were conducted in the USA on hops during the 2008 growing season. All seven trials were conducted in accordance with the critical GAP in the USA. Green hop cones were harvested at the 7-day PHI and dried for 4 to 36 hours using a commercial oven maintained at 46 ºC to 66 ºC. Residues in dried hop cones were: 0.72, 1.0, 1.1, 9.0, 9.8, 11 and 12 mg/kg.

No information on the metabolism of fenazaquin in/on leafy vegetables was provided to the Meeting. However, according to the metabolism study in maize, fenazaquin is the predominant residue observed in stover at a preharvest interval (20-DAT) that is almost 3-fold longer than the GAP PHI, demonstrating limited metabolism of the parent compound. Therefore, the Meeting estimated a maximum residue level of 30 mg/kg and an STMR of 9.0 mg/kg for fenazaquin on hops.

Animal feed items

Almond hulls

In the USA, the critical GAP for fenazaquin on almonds is a single foliar spray application at 504 g ai/ha and a PHI of 7 days.

As the supervised field trial residues were not supported by the demonstrated storage stability data for almond hulls, the Meeting could not estimate a maximum residue level.

Fate of residues during processing

High temperature hydrolysis

In the high temperature hydrolysis study, quinazoline ring-labelled fenazaquin was degraded to a significant extent to 4-hydroxyquinazoline under simulated pasteurisation conditions (pH 4, 90 °C, 20 minutes), accounting for 61% of the applied radioactivity at the end of incubation which represented almost two fold the levels detected for the parent, fenazaquin. For processes simulating baking/brewing/boiling (pH 5, 100 °C, 60 minutes) and sterilisation (pH 6, 120 °C and 60 minutes), 4-hydroxyquinazoline represented 22–60% of the parent concentrations, at the end of incubation. The fate of the phenyl ring moiety under processing conditions has not been provided to the Meeting.

Processing

The Meeting received information on the fate of fenazaquin residues during the processing of plums and pineapples. No recommendations could be made for any of the processed commodities as there is no GAP for plums and there were an insufficient number of trials conducted in accordance with the critical GAP for pineapple.

Animal commodities maximum residue level estimation

The information provided to the Meeting on almond hulls, the only potential livestock feed item, could not be considered in the calculation of the dietary burden. Therefore, maximum residue levels for animal commodities were not estimated.

RECOMMENDATIONS

On the basis of the data from supervised trials, the Meeting concluded that the residue levels listed in Annex 1 are suitable for establishing maximum residue limits and for IESTI and IEDI assessments.

Definition of the residue for plant commodities for enforcement of MRLs and for dietary risk assessment: *fenazaquin*

Definition of the residue for animal commodities for enforcement of MRLs and for dietary risk assessment: *sum of fenazaquin and the metabolites 2-(4-{2-[(2-hydroxyquinazolin-4-yl)oxy]ethyl}phenyl)-2-methylpropanoic acid (2-hydroxy-fenazaquin acid) and quinazoline-4-ol and 3,4-dihydroquinazolin-4-one (4-hydroxyquinazoline), expressed as fenazaquin equivalents.*

DIETARY RISK ASSESSMENT

Long-term dietary exposure

The International Estimated Dietary Intakes (IEDIs) of fenazaquin were calculated for the 17 GEMS/Food cluster diets using STMRs estimated by the current Meeting (Annex 3 to the 2017 Report). The ADI is 0–0.05 mg/kg bw and the calculated IEDIs were 0–0.2% of the maximum ADI. The Meeting concluded that the long-term dietary exposure to residues of fenazaquin resulting from the uses considered by the current JMPR is unlikely to present a public health concern.

Short-term dietary exposure

The ARfD for fenazaquin is 0.1 mg/kg bw. The International Estimate of Short Term Intake (IESTI) for fenazaquin was calculated for the food commodities for which STMRs or HRs were estimated by the present Meeting and for which consumption data were available. The results are shown in Annex 4 of the 2017 JMPR Report. The IESTI represented 0–10% of the ARfD for the general population including children. The Meeting concluded that the short-term dietary exposure to residues of fenazaquin, from the uses that have been considered by the present Meeting, is unlikely to present a public health concern.