5.19 THIABENDAZOLE (065)

TOXICOLOGY

Thiabendazole (2-(4-thiazolyl)-1*H*-benzimidazole) was evaluated by JMPR in 2006, when an ADI of 0–0.1 mg/kg bw was established. The 2006 Meeting also established an ARfD of 1 mg/kg bw for the general population and an ARfD of 0.3 mg/kg bw for women of childbearing age (Annex 5, reference *109*).

Following a request for additional maximum residue levels by CCPR, thiabendazole was placed on the agenda of the present Meeting, which assessed additional toxicological information available since the last review.

Several toxicological studies on thiabendazole were submitted to the present Meeting, including an acute neurotoxicity study, a 90-day neurotoxicity study and an immunotoxicity study.

All critical studies contained statements of compliance with GLP and were conducted in accordance with relevant national or international test guidelines, unless otherwise specified. One additional study that complemented the toxicological information submitted for the current assessment was identified from a literature search and was included in the evaluation.

Toxicological data

In a chronic toxicity and carcinogenicity study, mice were administered thiabendazole in the diet at 0, 310, 1250 or 5000 ppm (equal to 0, 33.2, 146 and 605 mg/kg bw per day for males and 0, 40.0, 179 and 615 mg/kg bw per day for females, respectively) for 78 weeks. The NOAEL for long-term toxicity in mice was 310 ppm (equal to 33.2 mg/kg bw per day), based on body weight suppression and an increased incidence of nephrosis at 1250 ppm (equal to 146 mg/kg bw per day). No carcinogenicity was observed.

In an acute neurotoxicity study in rats treated with thiabendazole as a single dose of 0, 50, 200 or 2000 mg/kg bw by gavage, the NOAEL for systemic toxicity was 50 mg/kg bw, based on decreases in mean rearing counts in females, body weight loss secondary to reduced feed consumption, and lower ambulatory locomotor activity counts at time of peak effect on study day 0 in both sexes at 200 mg/kg bw. There was no clear evidence that thiabendazole was acutely neurotoxic.

In a 90-day neurotoxicity study in rats treated with thiabendazole in the diet at 0, 200, 750 or 1500 ppm (equal to 0, 13, 47 and 95 mg/kg bw per day for males and 0, 15, 54 and 108 mg/kg bw per day for females, respectively), the NOAEL for systemic toxicity was 750 ppm (equal to 47 mg/kg bw per day), based on findings of decreased body weight gain, depressed body weights and lower feed consumption at 1500 ppm (equal to 95 mg/kg bw per day). The NOAEL for neurotoxicity was 1500 ppm (equal to 95 mg/kg bw per day), the highest dose tested.

The Meeting concluded that thiabendazole is not neurotoxic.

In an immunotoxicity study in female mice treated with thiabendazole in the diet at a concentration of 0, 100, 1000 or 5000 ppm (equal to 0, 20.9, 205.6 and 1027.0 mg/kg bw per day, respectively) for 28 days, the NOAEL for immunotoxicity was 1000 ppm (equal to 205.6 mg/kg bw per day), on the basis of lower total spleen activity measured as immunoglobulin M antibody-forming cells per spleen at 5000 ppm (equal to 1027.0 mg/kg bw per day). The NOAEL for systemic toxicity was 1000 ppm (equal to 205.6 mg/kg bw per day), based on reduced body weight and marked increases in liver weights at 5000 ppm (equal to 1027.0 mg/kg bw per day).

The Meeting concluded that thiabendazole is not immunotoxic in the absence of systemic toxicity.

Toxicological evaluation

The Meeting concluded that no revision of the ADI or ARfDs was necessary. The Meeting noted that the NOAEL for systemic toxicity from the acute neurotoxicity study (50 mg/kg bw) is lower than the

NOAEL from the study currently used in the derivation of the ARfD for the general population (100 mg/kg bw). However, as the lowest-observed-adverse-effect level (LOAEL) for both studies was 200 mg/kg bw, and as the findings in both studies were similar, the Meeting concluded that there was no reason to revise the ARfD for the general population.

An addendum to the toxicological monograph was prepared.

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

Long-term studies of toxicity and carcinogenicity	
Target/critical effect	Body weight suppression, nephrosis
Lowest relevant NOAEL	33.2 mg/kg bw per day (mouse)
Carcinogenicity	No evidence of carcinogenicity
Neurotoxicity	
Acute neurotoxicity NOAEL	2000 mg/kg bw, highest dose tested (rat) (systemic toxicity NOAEL 50 mg/kg bw)
Subchronic neurotoxicity NOAEL	95 mg/kg bw per day, highest dose tested (rat)
Other toxicological studies	
Immunotoxicity NOAEL	205.6 mg/kg bw per day (mouse); not immunotoxic in the absence of systemic toxicity

RESIDUE AND ANALYTICAL ASPECTS

Thiabendazole, a benzimidazole fungicide, was first evaluated by JMPR in 1970, and the latest residue evaluation was conducted in 2006 (T, R).

The Joint FAO/WHO Expert Committee on Food Additives (JECFA) established an ADI of 0–0.1 mg/kg and the 2006 JMPR established an ARfD of 0.3 mg/kg bw for women of childbearing age and of 1 mg/kg bw for the general population.

The residue definitions for thiabendazole are:

Compliance with the MRL and dietary risk assessment for plant commodities: *thiabendazole*

Compliance with the MRL for animal commodities: *sum of thiabendazole and 5-hydroxythiabendazole*

Dietary risk assessment for animal commodities: *sum of thiabendazole, 5-hydroxythiabendazole and its sulfate conjugate*.

The compound was scheduled at the Fiftieth Session of the CCPR for the evaluation of additional uses by the 2019 Extra JMPR. Plant metabolism studies on orange (post-harvest) and maize (seed treatment), analytical methods and residue studies on mango, beans, peas and sweet potato, and processing studies were submitted to the Meeting.

Plant metabolism

[Phenyl-U-^{14}C]-thiabendazole was applied post-harvest to orange fruits in a single dose at 0.2 kg ai/hL prior to storage in the dark at 5°C, and samples were analysed just after application, 8 and 16 weeks later. Radioactivity was extracted from the fruit surface with acetonitrile, and oranges separated into peel and flesh. Radioactivity in orange flesh was < 0.01 mg/kg eq. (0.002-0.007 mg/kg eq) and was not

further investigated. From 94 (day 0) to 73% (16 weeks) of TRR were recovered from the fruit surface. About 98% TRR in the orange peel on day 0 was thiabendazole (5.2 mg/kg), with residues dropping to 90%TRR after 16 weeks (3.7 mg/kg). Only minor metabolites of thiabendazole were observed in orange peel, arising via hydroxylation of the phenyl ring to produce 5-hydroxy-thiabendazole (~0.02 mg/kg eq.), and elimination of the thiazole ring to produce benzimidazole (0.002 mg/kg at 8 weeks) and carboxylated benzimidazole (0.02 mg/kg eq. at 8 weeks).

[Phenyl-U-^{14}C]-thiabendazole was applied to maize seed at 0.09 mg/seed. Treated maize was grown under glasshouse and plants were harvested at stages representing commercial forage, sweet corn and maturity. No residues were found in cobs and kernels. The TRRs of foliage from the sweet corn stage and maturity were 0.005 and 0.002 mg/kg eq., respectively. Only the foliage from the forage stage had TRR > 0.01 mg/kg eq. (0.014 mg/kg eq.), from which 55.5% remained unextracted (0.008 mg/kg eq.). Extracted residues in forage were composed of multiple minor metabolites without the presence of thiabendazole. No further attempt was made to characterise the unextracted residue.

In summary, thiabendazole was the only relevant residue found in orange after post-harvest treatment and no thiabendazole related residues were found in maize commodities after seed treatment.

Methods of analysis

Additional methods of analysis and validation data for crop commodities were submitted to the Meeting. In general, samples are extracted with ethyl acetate, cleaned-up with cation exchange SPE and analysed by LC-MS/MS with a LOQ of 0.01 mg/kg. In another LC-MS/MS method, conjugates of thiabendazole or benzimidazole are extracted with ethyl acetate following addition of glucosidase enzyme to the aqueous phase (LOQ of 0.01 mg/kg). The efficiency of ethyl acetate extraction was confirmed with orange (whole fruit)

treated post-harvest from the metabolism study. Additionaly, the QuEChERS method was validated for thiabendazole in crop commodities and for thiabendazole and 5-hydroxy thiabendazole in animal commodities, with a LOQ of 0.01 mg/kg in all cases.

Storage stability of residues under frozen conditions

Stability studies conducted with beans (dry seed), soya beans, spinach, barley and oranges showed that residues were stable under frozen conditions (-20 ºC) for at least 24 months.

Results of supervised residue trials on crops

Mango

Thiabendazole is registered for post-harvest use in a dip solution at a concentration of 0.24 kg ai/hL in Central American countries and 0.19 kg ai/hL in Brazil. In four trials conducted in Brazil according to central American GAP, residues were 2.4, 2.6, 3.4 and 4.5 mg/kg in the whole fruit and 0.01, 0.012, 0.023, and 0.027 (highest individual level of 0.030) mg/kg in the pulp.

The Meeting agreed that four trials were enough to make a recommendation for mango due to the lower variability of the residues in post-harvest treatment, using the mean + 4×SD approach.

The Meeting estimated a maximum residue level of 7 mg/kg (Po), a STMR of 0.0175 mg/kg and a HR of 0.030 mg/kg for thiabendazole in mango.

Succulent beans and peas subgroups

Thiabendazole is registered in the USA as a seed treatment in beans (succulent and dry, except soya bean) at 0.55 kg ai/tonne seed. The GAP for soya bean is 0.20 kg ai/tonne seed. In seven bean trials conducted in the USA approximating the GAP, residues in beans with pods were < 0.01 (6) mg/kg and residues in bean without the pods in one trial were < 0.01 mg/kg.

The GAP rate for peas (succulent and dry) in the USA is 0.33 kg ai/tonne seed. In nine trials conducted in peas at about 3–4 times the GAP rate, residues in the peas without the pods were < 0.01 (9) mg/kg.

As the trials conducted with beans at GAP and the trials conducted with peas at a rate higher than the GAP gave no quantified residues, and the GAP for soya bean is lower, the Meeting agreed that the residue data provided support a recommendation for the subgroups of succulent beans and peas.

The Meeting estimated a maximum residue level of 0.01(*) mg/kg, a STMR and HR of 0 mg/kg for thiabendazole for the subgroups of Beans with pods, Peas with pods, Succulent beans without pods and Succulent peas without pods

Dry beans and peas, subgroups

Thiabendazole is registered in the USA as a seed treatment in beans (succulent and dry, except soya bean) at 0.55 kg ai/tonne seed. The GAP for soya bean is 0.20 kg ai/tonne seed. In nine trials conducted approximating the GAP of the USA, residues in dry beans were < 0.01 (9) mg/kg.

The GAP rate for peas (succulent and dry) in the USA is 0.33 kg ai/tonne seed. In 10 trials conducted with peas using at least 2.4 times the GAP rate, residues in dry peas were < 0.01 (5) and < 0.05 (5) mg/kg.

As the trials conducted with beans at GAP and the trials conducted with peas at a higher rate than the GAP gave no quantified residues, and the GAP for soya bean is lower, the Meeting agreed that the residue data provided support a recommendation for the subgroups of dry beans and peas.

The Meeting estimated a maximum residue level of 0.01(*) mg/kg and a STMR of 0 mg/kg for thiabendazole for the subgroups of Dry beans and Dry peas.

Sweet potato

Thiabendazole is registered in the USA as a post-harvest treatment as a 0.16 kg ai/hL dip solution or spray (on a conveyor belt) at 0.006 kg ai/tonne.

In seven trials conducted according to the spray GAP, residues were 0.21, 0.26, 0.38, 0.46, 0.51, 0.54 and 1.2 mg/kg.

In eight trials conducted according to the dip GAP, residues were 2.7, 4.4, 4.5, 4.6, 4.8, 5.4, 5.5, and 6.3 (highest individual level of 6.97) mg/kg.

Based on the dip trials, which gives the highest residues, and on the mean + (4×SD) approach, the Meeting estimated a maximum residue level of 9 mg/kg (Po), a STMR of 4.7 mg/kg and a HR of 6.97 mg/kg for thiabendazole in sweet potato.

Animal feedstuffs

The GAP rate for peas (succulent and dry) in USA is 0.33 kg ai/tonne seed. In the trials conducted with pea in the USA at 2.4 times the GAP, residues ranged from < 0.01 to 0.02 mg/kg in the vines and from < 0.01 to 0.08 mg/kg in the hay. In three trials conducted with cowpea beans at 1.4 times the USA GAP rate for beans, residues in vines and hay were < 0.01 mg/kg.

As no trials were conducted according to GAP, no recommendations were made for thiabendazole in legume animal feeds.

Fate of residues during processing

In a study to simulate the hydrolysis of thiabendazole under different temperature/time and pH conditions, 99–103% of the applied radioactivity was recovered.

Sweet potatoes treated post-harvest with a dipping solution were processed to flake, chip, baked and fries. The processing factors and estimated STMRs for the processed commodities are shown below.

Crop	PF	STMR/STMR-P, mg/kg	HR/HR-P, mg/kg
Raw sweet potato	-	4.7	6.97
Baked washed with peel	0.28	1.3	1.95
Chips	0.02	0.094	0.139
Puree	0.02	0.094	0.139
Fries	0.12	0.564	0.836
Flakes	0.08	0.376	0.558

Residues in animal commodities

The estimations conducted by the present Meeting do not impact the previous calculated dietary burden of thiabendazole and do not affect the recommendations made by the JMPR for animal commodities

RECOMMENDATIONS

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

Definition of the residue for compliance with the MRL and dietary risk assessment for plant commodities: *thiabendazole*

Definition of the residue for compliance with the MRL for animal commodities: *sum of thiabendazole and 5-hydroxythiabendazole*

Definition of the residue for dietary risk assessment for animal commodities: *sum of thiabendazole, 5-hydroxythiabendazole and its sulfate conjugate*.

DIETARY RISK ASSESSMENT

Long-term dietary exposure

The ADI for thiabendazole is 0–0.1 mg/kg bw. The International Estimated Daily Intakes (IEDIs) for thiabendazole were estimated for the 17 GEMS/Food Consumption Cluster diets using the STMR or STMR-P values estimated by the JMPR. The results are shown in Annex 3 of the 2019 Extra JMPR Report.

The IEDIs accounted for 2 to 10% of the maximum ADI. The Meeting concluded that the long-term dietary exposure to residues of thiabendazole from uses considered by the JMPR is unlikely to present a public health concern.

Acute dietary exposure

The ARfDs for thiabendazole is 1 mg/kg bw for the general population and 0.3 mg/kg bw for women of child-bearing age. The International Estimate of Short Term Intakes (IESTIs) for thiabendazole were calculated for the food commodities for which HRs/HR-Ps or STMRs/STMR-Ps were estimated by the present Meeting and for which consumption data were available. The results are shown in Annex 4 of the 2019 Extra JMPR Report.

The IESTIs were 0–20% (children) and 0–7% (general population) of the ARfD for the general population; and from 0–9% of the ARfD for women of child bearing age. The Meeting concluded that the acute dietary exposure to residues of thiabendazole from uses considered by the present Meeting is unlikely to present a public health concern.