The European Agency for the Evaluation of Medicinal Products
Veterinary Medicines and Information Technology

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COMMITTEE FOR VETERINARY MEDICINAL PRODUCTS

DICYCLANIL

SUMMARY REPORT (2)

1. Dicyclanil is a pyrimidine-derived insect growth regulator which is used in veterinary medicine for the prevention of myiasis or fly-strike. The target species is sheep with a recommendation for one application per season at a therapeutic dose of 30 to 100 mg/kg bw. The compound is applied topically in a suspoemulsion pour-on formulation, with a dicyclanil concentration of 50 mg/ml.

   Dicyclanil is currently entered in Annex III of Council Regulation No 2377/90 as follows:

Pharmacologically active substances(s)	Marker residue	Animal species	MRLs	Target tissues	Other provisions
Dicyclanil	Sum of dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile	Ovine	200 µg/kg 50 µg/kg 400 µg/kg 400 µg/kg	Muscle Fat Liver Kidney	Not for use in animals from which milk is produced for human consumption. Provisional MRLs expire on 1.7.2000

   Further data have now been provided to support the establishment of final MRLs for dicyclanil.

2. Dicyclanil has a long lasting action which interferes with moulting and pupation in dipteran species. However, the precise mode of action of this compound on ectoparasites is not known.

   A variety of *in vitro* and *in vivo* pharmacodymanic tests were performed with dicyclanil in rats, mice and guinea pigs. *In vitro*, dicyclanil was devoid of significant effects on the neuromuscular junction at concentrations below 3 mM. Slight antagonistic effects on smooth muscle were reported at concentration levels above 0.1 mM. Dicyclanil caused a statistically significant increase in heart rate, tidal volume and minute volume at a dose level of 100 mg/kg bw in studies on the cardiovascular and respiratory systems in the rat. In various *in vivo* studies, dicyclanil was devoid of significant effects on behavior at a dose level of 1 mg/kg bw in mice.

3. In the rat, the absorption, distribution, metabolism and excretion profile of ^{14}C-labelled dicyclanil was investigated following repeated oral administration of test compound at dose levels of either 0.5 or 20 mg/kg bw for 7 consecutive days. At least 80% of the administered dose was absorbed from the gastrointestinal tract independent of sex or dose level. Approximately 93% of the dose was excreted in the first 7 days of dosing (80% via the kidneys). In the following 48 hours, only an additional 2 to 3% was excreted. Biotransformation was initiated by oxidative cyclopropyl-ring opening at various positions, with further oxidation occurring in some cases. Most of the metabolites were the result of more than one transformation step. The cyano group of this compound was metabolically stable. The major metabolite recorded in urine, accounting for approximately 50 to 55% of the administered dose (low and high dose levels) was a secondary propionic acid amide - N-(4,6-diamino-5-cyano-pyrimidin-2-yl)-propionamide. The metabolite 2, 4, 6-triamino-pyrimidine-5-carbonitrile accounted for approximately 11% of the dose, whilst

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unchanged dicyclanil represented 2% (low dose) and 7% (high dose) of the administered dose. Faecal metabolites appeared in 11 fractions including unchanged dicyclanil, but no single fraction accounted for more than 3% of the total dose. Besides polar metabolites 2, 4, 6-triamino-pyrimidine-5-carbonitrile was the major metabolite recorded in liver and kidney, with lower amounts of parent compound and probably N-(4,6-diamino-5-cyano-pyrimidin-2-yl)-propionamide. The pattern was similar for muscle and fat, except that these latter tissues contained more non-polar metabolites as a percentage of the total residue, especially so in the case of fat.

Tissue residue levels were measured 24 and 72 hours following the completion of 7 consecutive daily oral doses of radiolabelled dicyclanil at dose levels of either 0.5 or 20 mg/kg bw. Twenty-four hours following the final dose (0.5 mg/kg bw treatment group) the mean residue concentrations for dicyclanil were below 4 μg dicyclanil equivalents/kg, except in the liver (300 μg equivalents/kg), blood (200 μg equivalents/kg), kidney (40 μg equivalents/kg) and residual carcass (20 μg/kg). The residual radioactivity in blood was associated with red blood cells. At the higher dose level (20 mg/kg bw), the tissue residues were accordingly higher (10 to 60 times) and demonstrated a similar distribution pattern. Again, the highest residues were recorded in liver (approximately 5000 μg dicyclanil equivalents/kg). Within 72 hours following the last dose, the residues declined to values of about 30 to 80% of those at the first time point, except for whole blood, where the residue levels remained nearly constant during this period.

4. Radiolabelled pharmacokinetic studies were conducted in sheep using both a jetting technique and a pour-on formulation. Dicyclanil was applied as a single topical application (jetting technique) to Oxford Down sheep at a dose level of 35 mg/kg bw. Approximately 37 to 59% of the total dose remained on the animals, the remainder was collected as "run-off". Dermal absorption was approximately 2% of the retained radioactivity. Peak whole blood levels occurred at 4 to 6 hours post dose. There was no evidence of continued absorption from the application site. By 7 days post dose, 0.83% and 1.05% of the retained dose were excreted in urine and faeces respectively. Radioactivity was detected in bile. The urinary metabolite pattern consisted of five fractions, each less than 0.2% of the retained dose. Parent compound and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were amongst these five fractions. The faecal metabolite pattern consisted predominately of unchanged dicyclanil. High levels of radioactivity were associated with the wool and did not decrease considerably over time.

   In a further radiolabelled study, ^{14}C-dicyclanil was applied topically as a pour-on to Greyface sheep at a dose level of approximately 35 mg/kg bw. The maximum blood concentrations were reached 12 to 48 hours post dose. Approximately 4% of the applied dose was absorbed over a 7 day period. There was an indication for continuous dermal absorption at a low level, a factor not recognised in the previous jetting study. Absorbed radioactivity was widely distributed throughout the body. Considerable inter-animal variation was seen, but the overall picture was one of slow depletion of residues from the tissues. Radioactivity was excreted in both urine and faeces. The metabolites and metabolic pathways were similar to the results of the previous jetting experiment, although the ratios of certain metabolites to total radioactivity present did vary somewhat between the two studies.

5. Dicyclanil was investigated in a series of acute toxicity studies. The acute LD_{50} by the oral route in the rat was 520 mg/kg bw. The acute LD_{50} by the dermal route was greater than 2000 mg/kg bw in the rat. An acute inhalation toxicity study also conducted in the rat showed the acute LC_{50} to be 3,184 mg/m3 of air. The toxic effects were dyspnoea and reduced locomotor activity.

6. Repeated dose toxicity studies were performed in the rat and dog. Groups of 20 rats (10 of either sex) per dose group were fed dicyclanil for 90 days at dietary levels of 0, 5, 25, 125 and 500 mg/kg feed/day corresponding to 0, 0.31, 1.6, 8.05, and 32.7 mg/kg bw in males and 0, 0.31, 1.65, 8.38 and 34.1 mg/kg bw in females. Body weight, food consumption, biochemical parameters (decrease in plasma glucose) and significant variations in organ weights were reported at doses of 125 and 500 mg/kg feed/day. A statistically significant increase in epididymus to body weight ratios was recorded in the 25 mg/kg feed/day treatment group. However, absolute epididymus weights were within the normal physiological range and no abnormal findings were present on histopathology of this tissue. Consequently, the dose level of 25 mg/kg feed/day (equivalent to 1.6 mg/kg bw/day) can be retained as a NOEL for this study.

A 3-month dietary toxicity study was performed in beagle dogs. Eight animals per dose group were fed dicyclanil at dietary levels of 0, 20, 100, 500 and 1500 mg/kg feed/day corresponding to 0, 0.61, 2.67, 13.9 and 41.5 mg/kg bw/day in males and 0, 0.71, 3.45, 17 and 41.8 mg/kg bw/day in females. Significant increases in the relative and absolute liver weights were noted in females at the 3 highest doses and liver microscopic changes were reported at all doses. Moreover, at 100, 500 and 1500 mg/kg feed/day, effects on male gonads were described. A NOEL could not be set for this study.

The repeated dose dermal toxicity of dicyclanil was investigated in a 28-day study in rats. Dicyclanil was applied on gauze patches at dose levels of 0, 5, 30, 300 and 1000 mg/kg bw/day. Decreased body weight gains and food intake were observed in male animals at dose levels greater than or equal to 300 mg/kg bw. Mean absolute and relative liver weights were slightly increased for females up to 300 mg/kg bw and hypertrophy of hepatocytes was recorded in high dose males and females. At doses greater than or equal to 30 mg/kg bw, a significant increase in brain weights was recorded for females. The NOEL from this study was 5 mg/kg bw.

7. A 12-month dietary toxicity study was also performed in beagle dogs. Dicyclanil was administered at dietary levels of 0, 5, 25, 150 and 750 mg/kg feed/day (equivalent to average intakes of 0, 0.16, 0.71, 4.38 and 22.5 mg/kg bw for males and 0, 0.15, 0.77, 5.06 and 22.7 mg/kg bw/day for females) to groups of 4 to 6 animals per dose group. Increased absolute and relative liver weights were recorded at the 750 mg/kg feed/day dose level. At 150 and 750 mg/kg feed/day, reversible biochemical and haematological changes (increase in plasma cholesterol levels in males, decrease in platelet count and minimal increase in plasma calcium in females) were noted. A NOEL of 25 mg/kg feed/day (corresponding to a mean daily intake of 0.71 and 0.77 mg/kg bw/day for males and females, respectively) was set from this study.

8. A tolerance study was performed in the target species whereby dicyclanil was applied topically as a pour-on formulation on three occasions at weekly intervals. The test compound was administered at 1, 3 and 10 times the recommended therapeutic dose of 42 mg/kg bw and 8 animals were employed per dose group. Ten times the recommended therapeutic dose was associated with significant increases in liver and spleen weights. Overall, 3 times the recommended therapeutic dose was well tolerated and was devoid of any significant signs of systemic toxicity.

9. A 2-generation reproductive toxicity study was conducted with dicyclanil in the rat. Groups of 60 rats per dose level (30 male and 30 female) were exposed to dicyclanil at nominal concentrations of 0, 5, 30, 200 and 500 mg/kg feed/day. Treatment began 10 weeks prior to mating and was continued until the end of the lactation period. Food consumption and bodyweight gains were reduced at dose levels greater than or equal to 200 mg/kg feed/day.

   However, male and female mating and fertility indices, maternal gestation and parturition indices and the duration of pregnancy were unaffected by treatment at either the first or second matings of both the F_0 and F_1 parent generations. No adverse effects of treatment were noted in the F_1 or F_2 progeny in terms of the sex ratios, the development of physical landmarks, clinical signs or macroscopic findings at post-mortem. At the 500 mg/kg feed/day dose level for both the F_0 and F_1 generations, there was a reduction in mean pup weight at birth and a retardation of bodyweight gain in pups during lactation. Thus, pup body weights were reduced at weaning at this treatment level. An increase in pup mortality was recorded between days 0 to 4 at the 30 mg/kg feed/day dose level in the F_{1b} generation and at 500 mg/kg feed/day dose level in the F_{1a} generation. Litter loss in the F_{1b} generation did not show a dose-response relationship and was mainly attributable to total litter loss in two litters. The pup mortality in the high dose group in the F_{1a} generation was again primarily confined to two specific litters. There was no other evidence of reproductive toxicity for this compound. Overall, the NOEL for this study can be set at 30 mg/kg feed/day (equivalent to approximately 1.5 to 6.0 mg/kg bw/day).

10. Teratogenicity data were available from studies conducted with dicyclanil in the rat and the rabbit. In the rat oral teratogenicity study, test compound was administered by gavage at daily dose levels of 0, 1, 5, 25 and 75 mg/kg bw between days 6 and 15 of gestation. Body weights, body weight gains and food intake were reduced in the 75 mg/kg bw group. A smaller but not statistically significant decrease in such parameters was evident at the 25 mg/kg bw dose level. Foetal weights were significantly reduced at the 75 mg/kg bw dose level. A slight delay in skeletal development in progeny of dams from the high dose group was observed. A significantly increased incidence of skeletal anomalies was reported for foetuses of dams treated at the 5 mg/kg bw dose level. There was no evidence of any teratogenic effect. The NOEL was 25 mg/kg bw for dams and 1 mg/kg bw for foetuses.

    In a study in rabbits, five groups of 19 inseminated female rabbits were dosed by gavage from day 7 to day 19 of gestation with daily dose levels of 0, 1, 3, 10 and 30 mg/kg bw. Maternal toxicity was evident at dose levels greater than or equal to 10 mg/kg bw and was chiefly manifested by reduced bodyweight gains. Foetal weights were significantly reduced at the 30 mg/kg bw treatment level. Skeletal variations indicating a slight delay in ossification were observed in progeny from dams treated at this high dose level (30 mg/kg bw). The NOEL was 3 mg/kg bw for dams and 10 mg/kg bw for foetuses. There was no indication of any teratogenic potential.

11. Dicyclanil was devoid of mutagenic activity in four *in vitro* test (*Salmonella*-microsomal assay, *in vitro* DNA repair on rat hepatocytes, in the gene mutation test in V79 Chinese hamster cells, in the *in vitro* cytogenetic test on Chinese hamster ovarian cells) and in one *in vivo* test (micronucleus test in mice). There was no evidence of mutagenicity.

12. Carcinogenicity data for dicyclanil was available from a 24-month study in rats and an 18-month study in mice.

    The rat 24-month long-term toxicity/oncogenicity study employed dose levels of 0, 5, 25, 125 and 500 mg/kg feed/day corresponding to average intakes of 0, 0.19, 0.97, 4.83 and 21.8 mg/kg bw/day for males and 0, 0.23, 1.18, 6.01 and 26 mg/kg bw/day for females. Bodyweight gains and food intake were significantly depressed (exceeding the maximum tolerable dose) at the 500 mg/kg feed/day dose level. Increases in relative liver weights and inorganic phosphorous concentrations were recorded in males treated at the 125 and 500 mg/kg feed/day dose levels. An increased incidence of hepatic cysts in females and pancreatic masses in males was recorded at the 500 mg/kg feed/day dose level. On microscopic examination, the liver cysts were seen to be biliary in nature, whilst the pancreatic nodules were classified as hyperplastic. An increased incidence of pigmentation of the olfactory epithelium was observed in males at 25 mg/kg feed/day and in both sexes at higher dose levels. The pigmentation appeared to be concentrated in the sustentacular or supporting cells of the olfactory epithelium and in Bowmans glands. There was no evidence of increased pigmentation in the neuronal cells in the epithelial lining, in olfactory nerve bundles or in the olfactory bulbs in the brain. This increase in pigmentation in the olfactory epithelium was not considered to be toxicologically significant. There was no evidence of a carcinogenic effect. A NOAEL of 25 mg/kg feed/day (equivalent to 1.0 mg/kg bw/day for males and 1.2 mg/kg bw/day for females) can be set for this study.

    An 18-month oncogenicity study in mice incorporated 120 animals per dose group at treatment levels of 0, 10, 100, 500 and 1500 mg/kg feed/day corresponding to average intakes of 0, 1.11, 11.3, 57.7 and 210 mg/kg bw/day for males and 0, 1.08, 11.5, 63.2 and 199 mg/kg bw for females. The high dose group was sacrificed early due to a higher than expected mortality rate. Survival and clinical signs were not affected by any other dose level. The maximum tolerable dose was exceeded at 1500 mg/kg feed/day for males and at 500 and 1500 mg/kg feed/day for females. Benign hepatomas were observed in females at 500 mg/kg feed/day and both benign hepatomas and hepatocellular carcinomas were recorded in females at 1500 mg/kg feed/day. Other signs of liver pathology were evident in males at treatment levels greater than or equal to 100 mg/kg feed/day. Hepatocellular hypertrophy was seen in males at levels greater than or equal to 500 mg/kg feed/day. Increased incidences of pigmentation of the olfactory epithelium were seen in both sexes at 100 and 500 mg/kg feed/day treatment levels. A tumorigenic effect was seen in the liver of treated females (greater than or equal to 500 mg/kg feed/day), but the exact mechanism of such a finding was not clear, although the dose needed for such an effect exceeded

the maximum tolerable dose for this sex. A NOEL of 10 mg/kg feed/day (equivalent to 1.1 mg/kg bw/day) can be retained on the basis of the hepatic effects seen at higher dose levels.

13. Evidence of atrophy of lymphatic tissue was seen in the 90-day repeated-dose oral toxicity study in dogs (dose levels greater than or equal to 13.9 mg/kg bw/day). Dicyclanil can be considered non-irritant to the eye and skin. Dicyclanil was shown to have a low sensitisation potential in the guinea pig (optimisation test) following epidermal challenge after intravenous exposure).

14. No information is available on the microbiological properties of residues of dicyclanil.

15. Dicyclanil is not used in human medicine and therefore no information is available on observations in man.

16. Taking the NOEL of 0.7 mg/kg bw/day observed in the 12-month dietary toxicity study in the dog and applying a safety factor of 100, an ADI of 0.007 mg/kg bw, equivalent to 0.42 mg per person, is established.

17. Initial radiolabelled studies were performed in sheep by a jetting or a pour-on technique in the target species. In the first experiment, radiolabelled dicyclanil was applied topically at a single dose of 35 mg/kg bw by jet application. The highest residues were generally detected one day post-dose, with the highest values being recorded in liver and subcutaneous fat. At this time point, mean residue concentrations of 39, 234, 289 and 71 µg dicyclanil equivalents/kg were recorded for muscle, subcutaneous fat, liver and kidney respectively. At the 14-day time point post dose, mean residue concentrations had declined to values of 7, 43, 37 and 10 µg dicyclanil equivalents/kg for muscle, subcutaneous fat, liver and kidney respectively. The major residue detected in muscle and fat was unchanged parent compound, with lower amounts of 2, 4, 6-triamino-pyrimidine-5-carbonitrile and N-(4,6-diamino-5-cyano-pyrimidin-2-yl)-propionamide (muscle). Individual metabolites depleted at about the same rate from fat and muscle tissues, with half life times of approximately 2 to 5 days. The major residue in liver and kidney was 2, 4, 6-triamino-pyrimidine-5-carbonitrile. Smaller amounts of dicyclanil/or N-(4,6-diamino-5-cyano-pyrimidin-2-yl)-propionamide were also present. A further unidentified metabolite, representing 7 to 11% of total residues, was also present in kidney.

    In the second radiolabelled study, dicyclanil was applied topically as a pour-on suspoemulsion at a dose level of 35 mg/kg bw on a single occasion. Levels of radioactivity in muscle, fat, liver and kidney declined from 227, 44 to 225, 454 and 78 µg dicyclanil equivalents/kg, respectively, at the three day time point post dose to levels of 33, 14 to 71, 454 and 54 µg dicyclanil equivalents/kg at the 21 day time point post dose for these respective tissues. Residues in muscle and fat were predominantly unchanged dicyclanil, whilst in liver and kidney dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were identified. There was evidence of retention of radioactivity in kidney at a low level. Depletion half lives for plasma, whole blood, liver and kidney were approximately 8, 9, 13 and 10 days respectively. Those for muscle and fat were within the range of 2 to 11 days.

    In a third radiolabelled study, dicyclanil was applied topically as a pour-on suspoemulsion at a dose level of 100 mg/kg bw on a single occasion. Levels of radioactivity declined from 2955, 431, 2646 and 762 µg dicyclanil equivalents/kg in muscle, fat, liver and kidney respectively at the 7-day time point post dose to levels of 880, 208, 1475 and 230 µg dicyclanil equivalents/kg at the 21-day time point post dose for these respective tissues. Residues in muscle and fat were predominantly unchanged dicyclanil (85% or greater), whilst in liver and kidney dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were the major residues accounting for 23% and 43% of total residues at 7 days and 13% and 24% at 21 days.

    As both dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were significant residues in different tissues in the above studies, the sum of dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile could be retained as the marker residue (expressed as dicyclanil equivalents/kg). Muscle, fat, liver and kidney should all be retained as marker tissues.

18. A total of seven non-radiolabelled residue depletion studies were performed in sheep. Differences in the studies involved dose levels, formulations and methods of application, breed and fleece length. Differences also existed in the specific metabolite fractions assayed in the studies.

An initial non-GLP compliant study involved the topical application by jetting of dicyclanil at a dose rate of 29 to 44 mg/kg bw onto sheep with 8 weeks wool growth (2 to 3 cm in length). No residues of dicyclanil were recorded at the 48-hour time point post dose in muscle, fat, liver or kidney. The limit of quantification in this study was 10 µg/kg. Only the parent compound dicyclanil was assayed. Two further non-GLP compliant studies were performed with a pour-on formulation on sheep shorn 2 weeks before treatment. Dicyclanil alone was assayed in the various tissues. Following the topical application of the test compound at a dose level of 45 mg/kg bw, significant levels of dicyclanil were detected in all edible tissues. Residue levels increased in tissues between days 14 and 21, such that the mean dicyclanil concentrations on day 21 were 100, 640, 140 and 120 µg dicyclanil/kg for muscle, fat, liver and kidney, respectively. Thereafter, mean residue concentrations declined so that at day 35 values of 40, 90, 30 and 40 µg dicyclanil/kg were recorded for muscle, fat, liver and kidney respectively. A further decline in residue concentrations occurred by day 42. Only 3 animals per time point were sacrificed.

The second of these non-GLP compliant pour-on studies utilized a dose level of 90 mg/kg bw. This dose level is within the recommended therapeutic dose of 30 to 100 mg/kg bw. Very high residue concentrations were still detectable at day 21 in this study; mean values of 580, 200, 680 and 570 µg dicyclanil/kg were recorded in muscle, fat, liver and kidney respectively. No further data were available from this study beyond the 21 day time point post dose. Only 3 animals were sacrificed per time point.

The fourth study was GLP compliant and involved the topical application of dicyclanil as a pour-on suspoemulsion to groups of 6 unshorn animals per time point at dose levels of either 99 mg/kg bw (maximum recommended therapeutic dose) or 199 mg/kg bw (twice the maximum recommended therapeutic dose). Dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were both assayed. At the 99 mg/kg bw dose level, only very low concentrations of dicyclanil were detected in the various edible tissues, whereas 2, 4, 6-triamino-pyrimidine-5-carbonitrile was present to a slightly higher extent, particularly in kidney but also in muscle and liver. The highest level of 2, 4, 6-triamino-pyrimidine-5-carbonitrile was detected in kidney on day 14 post treatment (110 µg/kg); this declined to 40 µg/kg on day 28. At the 199 mg/kg bw dose level, low levels of dicyclanil were detectable in fat and kidney up to day 7 post treatment (approximately 20 µg/kg). Dicyclanil could also be detected in muscle and liver up to day 28 post treatment (30 µg/kg). Significant levels of 2, 4, 6-triamino-pyrimidine-5-carbonitrile were present in muscle (20 µg/kg), liver (90 µg/kg) and kidney (80 µg/kg) on day 28 post treatment.

In a study on Merino sheep 1 day or 6 weeks off shears and treated with either 100 mg/kg bw or 200 mg/kg bw, residues of dicyclanil and metabolite 2, 4, 6-triamino-pyrimidine-5-carbonitrile were analysed following slaughter at 7, 14, 21, 28 and 56 days. For the low dose group (1 day off shears), maximum individual residues of dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were respectively, 760 and 190 µg/kg in muscle, 970 and 500 µg/kg in kidney, 1130 and 360 µg/kg in liver, 280 and 60 µg/kg in subcutaneous fat and 130 and 30 µg/kg in kidney fat.

For the high dose group (1 day off shears), maximum individual residues of dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were respectively, 1180 and 560 µg/kg in muscle, 1580 and 630 µg/kg in kidney, 1830 and 600 µg/kg in liver, 3290 and 70 µg/kg in subcutaneous fat and 200 and 60 µg/kg in kidney fat. For the low dose group (6 weeks off shears), maximum individual residues of dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were respectively, 320 and 130 µg/kg in muscle, 360 and 300 µg/kg in kidney, 450 and 240 µg/kg in liver, 620 and 20 µg/kg in subcutaneous fat and 80 and 10 µg/kg in kidney fat. For the high dose group (6 weeks off shears), maximum residues of dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were respectively, 950 and 440 µg/kg in muscle, 1220 and 980 µg/kg in kidney, 1380 and 680 µg/kg in liver, 3860 and 80 µg/kg in subcutaneous fat and 140 and 70 µg/kg in kidney fat.

In a study on Cross-bred and Merino sheep treated at a rate of 100 mg/kg bw at 6-week off-shears and sacrificed at 11, 28 and 35 days post treatment maximum and mean residues of dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were generally higher in the Merinos than in Cross-bred sheep. For the Merinos, maximum individual residues of dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were respectively, 100 and 90 µg/kg in muscles, 140 and 280 µg/kg in kidney, 110 and 100 µg/kg in liver, 30 and 20 µg/kg in renal fat. These all occurred within the 11 day post-treatment slaughter group. For the cross-breds, maximum individual residues of dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were respectively, 40 and 50 µg/kg in muscle, 60 and 110 µg/kg in kidney, 70 and 110 µg/kg in liver, and 30 and 20 µg/kg in renal fat. These also occurred within the 11 day post-treatment slaughter group.

In a further study on Merino sheep and cross-bred lambs treated at a rate of 50 and 100 mg/kg bw respectively at 1 day off shears and slaughtered at 7, 28, 56, 84 days and 4 months post treatment, residues of dicyclanil and 2, 4, 6-triamino-pyrimidine-5-carbonitrile were relatively low in both adult Merino sheep and Crossbred lambs, with many animals having no quantifiable residues (below 10 µg/kg). In fatty tissue, the predominant species was the parent compound, whilst in muscle, liver and kidney the metabolite 2, 4, 6-triamino-pyrimidine-5-carbonitrile was the predominant residue metabolite. In Merino sheep, no residues were observed in renal fat, and the maximum individual residues observed were 90 µg/kg dicyclanil in subcutaneous fat at 7 days, and total residues (dicyclanil + 2, 4, 6-triamino-pyrimidine-5-carbonitrile) of 100 µg/kg, 90 µg/kg and 60 µg/kg in kidney, liver and muscle respectively at 56 days. At 4 months post-treatment, traces of metabolite only were found in offal (liver and kidney), but there were no quantifiable residues in the carcass (muscle and fat). In crossbred lambs, no residues were found in the muscle. The maximum individual residues observed were 130 µg/kg dicyclanil in subcutaneous fat and 40 µg/kg 2, 4, 6-triamino-pyrimidine-5-carbonitrile in kidney at 7 days, and 30 µg/kg dicyclanil in renal fat and 30 µg/kg 2, 4, 6-triamino-pyrimidine-5-carbonitrile in liver at 28 days. Traces of 2, 4, 6-triamino-pyrimidine-5-carbonitrile were found at 4 months in the kidneys, but the carcass was free of quantifiable residues.

19. Dicyclanil is not indicated for use in sheep producing milk for human consumption.

20. The analytical method proposed for the routine analytical method is based on HPLC. The method measures both parent compound and the metabolite 2, 4, 6-triamino-pyrimidine-5-carbonitrile in muscle, fat, liver and kidney. The method has been properly validated and presented according to the recommendations of Volume VI of the Rules Governing Medicinal Products in the European Community The values of the limit of quantification was 10 µg/kg for all edible tissues and the limits of detection were 4 µg/kg for muscle, liver and kidney and 3 µg/kg for fat.

Conclusions and recommendation

Having considered that:

- an ADI of 0.42 mg/person was established,
- the sum of dicyclanil and metabolite 2, 4, 6-triamino-pyrimidine-5-carbonitrile was identified as marker residue and considering that the marker residue represents approximately 100%, 100%, 15% and 25% respectively in muscle, fat, liver and kidney at 21 days after treatment,
- a validated analytical method for monitoring is available;

the Committee for Veterinary Medicinal Products recommends the inclusion of dicyclanil in Annex I of Council Regulation (EEC) No 2377/90 in accordance with the following table:

Pharmacologically active substances(s)	Marker residue	Animal species	MRLs	Target tissues	Other provisions
Dicyclanil	Sum of dicyclanil and 2,4,6-triamino-pyrimidine-5-carbonitrile	Ovine	200 µg/kg 50 µg/kg 400 µg/kg 400 µg/kg	Muscle Fat Liver Kidney	Not for use in animals from which milk is produced for human consumption.

Based on these MRL values, the daily intake will represent about 97% of the ADI.