

PHOSMET (addendum)

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Explanation

Phosmet was evaluated toxicologically by the Joint Meeting in 1978 (Annex 1, reference 30), when a temporary ADI of 0–0.005 mg/kg bw was allocated. It was re-evaluated in 1979 (Annex 1, reference 32) when additional data on teratogenicity became available, and an ADI of 0–0.02 mg/kg bw was established. Further data were made available to the 1994 JMPR (Annex 1, reference 73), when an ADI of 0–0.01 mg/kg bw was allocated on the basis of the NOAEL in a multigeneration study of reproductive toxicity in rats, with a 100-fold safety factor. At that meeting, further information was requested, namely a further long-term study in dogs, studies on DNA binding *in vivo*, and a study of the potential of phosmet to cause delayed polyneuropathy in hens at an appropriately high dose, with an estimate of neuropathy target esterase. The last study was supplied but not the requested study of repeated dosing in dogs; moreover, a study of unscheduled DNA synthesis in rat liver *in vivo* was submitted rather than a study of DNA binding *in vivo*. The new studies are reviewed below.

Evaluation for Acceptable Daily Intake**1. Delayed neuropathy**

A study of the ability of phosmet to cause delayed polyneuropathy in Lohmann Brown hens (*Gallus gallus domesticus*) was conducted with phosmet of a purity of 97.4%. In a preliminary study, the LD₅₀ of phosmet given orally was determined in seven groups of 10 birds. It was estimated to be 580 mg/kg bw (95% confidence interval, 410–770). A dose of 600 mg/kg bw was therefore given to 24 birds; 12 further birds received the vehicle (corn oil), and a further 12 were given tri-*ortho*-cresyl phosphate at 1 g/kg bw as positive controls. All birds were injected subcutaneously with atropine sulfate at 20 mg/kg bw just before treatment, and further injections were given to birds that showed severe clinical signs. The birds were observed for adverse clinical signs, ataxia, and effects on body weight. After 48 h, three birds from the groups receiving phosmet, the vehicle or the positive control were killed, and brain acetylcholinesterase and brain and spinal cord neuropathy target esterase activities were estimated. After 21 days, six birds from each group were sacrificed and fixed by perfusion, and the head, spinal column, and dissected sciatic and tibial nerves were taken and stored in 10% buffered formalin. Sections of fore-, mid-, and hindbrain; upper and lower cervical, mid-thoracic, and lumbo-sacral spinal cord; and proximal and distal sciatic and tibial nerves were examined histopathologically.

No ataxia was observed in the birds treated with phosmet, while 8/12 birds treated with tri-*ortho*-cresyl phosphate developed clinical ataxia, commencing at 11–18 days. Brain acetylcholinesterase activity was reduced in birds given phosmet to 37% of the value in concurrent vehicle controls, but the neuropathy target esterase activity in both brain and spinal cord was similar to that of controls. In contrast, in birds given tri-*ortho*-cresyl phosphate, the acetylcholinesterase activity was similar to that of vehicle controls, whereas the neuropathy target esterase activity was markedly inhibited, being 9.6% of the value for concurrent controls in brain and 20% that of concurrent controls in spinal cord. Vehicle controls gained weight normally, while those treated with tri-*ortho*-cresyl phosphate lost weight. With phosmet, initial weight loss was followed by recovery. Histopathological examination showed no evidence of the characteristic changes of delayed neuropathy in the phosmet-treated or vehicle control hens, whereas birds that had received tri-*ortho*-cresyl phosphate showed minimal axonal degeneration in the cerebellum and minimal or moderate axonal degeneration at one or more levels of the spinal cord and in some peripheral nerve sections (Johnson, 1997).

2. Unscheduled DNA synthesis *in vivo*

The ability of phosmet (purity, 96.4%) to induce unscheduled DNA synthesis in male Alpk:AP_rSD rats was determined by an autoradiographic technique. Five rats received phosmet orally at a dose of 32 (two rats) or 50 mg/kg bw (three rats) in corn oil, the higher dose being the maximum tolerated dose of phosmet in this strain of rat. Hepatocytes were isolated and prepared at 2 and 16 h; two independent experiments were carried out at each time. Groups of two animals received the vehicle or dimethylhydrazine dihydrochloride as a positive control. The mean net nuclear grain counts and the percentages of cells in repair were recorded. Phosmet did not induce DNA repair at either dose or time, whereas the positive control produced marked unscheduled DNA synthesis in comparison with vehicle controls (Mackay, 1996).

Comments

In the study of delayed polyneuropathy, a dose of 600 mg/kg bw was given to 24 hens, this dose being greater than the experimentally determined LD₅₀. There was no evidence that phosmet could produce clinical signs of delayed polyneuropathy or significantly inhibit neuropathy target esterase.

The ability of phosmet (96.4% pure) to induce unscheduled DNA synthesis in the liver of male rats *in vivo* was determined with doses of 32 or 50 mg/kg bw, the higher dose being the maximum tolerated dose of phosmet. Unscheduled DNA synthesis was not observed. The Meeting noted that the study on DNA binding had not been provided, but it concluded that no further characterization of mutagenicity was required. The Meeting considered that a further study in dogs would be unlikely to affect the overall evaluation.

The ADI of 0–0.01 mg/kg bw allocated by the 1994 JMPR, which was based on a NOAEL of 1.3 mg/kg bw per day in a multigeneration study in rats and a safety factor of 100, was confirmed.

An acute RfD of 0.02 mg/kg bw was allocated on the basis of a NOAEL of 2 mg/kg bw per day in a study of developmental toxicity in rabbits (fetotoxicity) and a safety factor of 100.

Toxicological evaluation

Levels that have no toxic effect (from 1994 monograph)

Mouse:	25 ppm, equivalent to 4 mg/kg bw per day (two-year study of carcinogenicity)
Rat:	40 ppm, equal to 1.8 mg/kg bw per day (two-year study of toxicity and carcinogenicity)

- Rat: 20 ppm, equal to 1.3 mg/kg bw per day (two-generation study of reproductive toxicity)
5 mg/kg bw per day (maternal toxicity in study of developmental toxicity)
15 mg/kg bw per day (study of developmental toxicity)
- Rabbit: 5 mg/kg bw per day (maternal toxicity in study of developmental toxicity)
2 mg/kg bw per day (study of developmental toxicity)

Estimate of acceptable daily intake for humans

0–0.01 mg/kg bw

Estimate of acute reference dose

0.02 mg/kg bw

Studies that would be useful for continued evaluation of the compound

Further observations in humans

References

- Johnson, A.J. (1997) Phosmet acute delayed neurotoxicity study in the domestic hen. Unpublished report No. CTL/C/3123, dated 23 January 1997, from Huntingdon Life Sciences, Huntingdon, Cambridgeshire, United Kingdom. Submitted to WHO by Gowan Co., Yuma, Arizona, USA.
- Mackay, J.M. (1996) Phosmet: *in vivo* rat liver unscheduled DNA synthesis assay. Unpublished report No. CTL/P/5090, dated 23 September 1996, from Central Toxicology Laboratory, Alderley Park, Cheshire, United Kingdom. Submitted to WHO by Gowan Co., Yuma, Arizona, USA.
- WHO (1996) *The WHO Recommended Classification of Pesticides by Hazard and Guidelines to Classification 1996–1997* (WHO/PCS/96.3), International Programme on Chemical Safety, Geneva.

