The European Agency for the Evaluation of Medicinal Products
Veterinary Medicines Evaluation Unit

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COMMITTEE FOR VETERINARY MEDICINAL PRODUCTS

D-PHE^6-LUTEINIZING-HORMONE-RELEASING-HORMONE

SUMMARY REPORT

1. D-Phe^6-luteinizing-hormone-releasing-hormone (D-Phe^6-LHRH) is a synthetic analogue of the naturally occurring gonadotropin releasing-hormone gonadorelin (GnRH; LHRH). Both substances are linear decapeptides, the difference between them being the amino acid at position 6: gonadorelin - glycine, D-Phe^6-LHRH-phenylalanine.

2. GnRH with the same chemical structure has been isolated from man, sheep, cattle pigs and rats. The substance regulates the production and release of follicle stimulating hormone (FSH) and luteinizing hormone (LH) from the adenohypophysis. Substitution at position 6 confers agonistic activity, while substitution at position 2 or 3 gives rise to substances with GnRH-antagonistic effects. Substitution is done with lipophilic and hydrophilic amino acids. The amino acid at position 6 affects the affinity of the molecule to the receptors.

3. In veterinary medicine GnRH and its synthetic analogues are employed mainly because of their influence on the release of LH. Indications are the correction of certain reproductive disorders in production animals of the female gender, e.g. delayed ovulation and ovarian cysts. In addition, the substances are used for zootechnical purposes. Recommended doses of D-Phe^6-LHRH for horses, cattle and swine are 25-100 µg, for smaller ruminants 20 µg, and for rabbits 10 µg, administered either subcutaneously, intramuscularly or intravenously.

4. Substitution at position 6 does not change the general pharmacodynamic characteristics of the molecule. However, substitution may cause minor quantitative changes, i.e. in the duration and/or intensity of FSH/LH release.

   As regards secondary pharmacodynamic effects no behavioral changes were observed in mice at doses up to 1000 µg D-Phe^6-LHRH/kg subcutaneous. In similar dose design, increases were observed in heart rate, while blood pressure decreased at 1000 µg/kg; no effects were observed at 50 µg/kg. Intravenous administration of 1 mg/kg to rats caused a slight increase in diuresis.

5. At normal doses peptides are broken down in the gastro-intestinal tract. Therefore, GnRH and its synthetic analogues are administered by parenteral injection.

6. D-Phe^6-LHRH is quickly absorbed following subcutaneous administration. The plasma half-life of GnRH and its synthetic analogues, including D-Phe^6-LHRH, is short, varying from a few minutes for GnRH to up to half an hour for some of the analogues.

7. Following parenteral administration GnRH as well as all the analogues so far examined are rapidly degraded enzymatically. Biotransformation takes place in the plasma and in several organs/tissues. The primary step involves cleavage of peptide bonds resulting in the formation of various oligopeptides with a maximum of five amino acids. The peptides exert no GnRH activity. The degradation products are either excreted via the kidneys (a small amount of unchanged GnRH is also excreted via the kidney) or are further degraded in the liver, where the remains enter the general pool of aminoacids. Therefore, no accumulation of D-Phe^6-LHRH will take place in treated animals.

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8. The intravenous LD_{50} of D-Phe^6-LHRH in juvenile female rats was 15.4 mg/kg. GnRH caused no fatalities up to the highest dose examined (50 mg/kg).

9. In repeated-dose toxicity studies in rats (0, 2, 10, 50 µg D-Phe^6-LHRH/kg subcutaneously daily for four weeks) only minor substance-associated changes were observed with few of them, e.g. bodyweight increase in females at 50 µg /kg, being statistically significant.

   In a reproduction study with female mice, 3 generations were treated immediately before mating with doses of up to 37.8 µg DPhe^6-LHRH/kg subcutaneously. The only substance-related effect observed was a slight dose-independent decrease in foetal weight.

10. In a teratogenicity study employing the same dose regimen as used in the three-generation reproduction study an increase in the incidence of skeletal malformations was observed. The increase was statistically significant at 37.8 µg/kg. The specific nature of malformations was not revealed in the report.

11. The mutagenic potential of D-Phe^6-LHRH was investigated in the Ames' test and in an *in vivo* test for chromosomal abnormalities in rat bone marrow cells. No results indicated the presence of mutagenic potential.

Conclusions and recommendation

Based on the documentation available concerning D-Phe^6-LHRH itself as well as the documentation available for GnRH and other synthetic GnRH analogues the Committee considers that there is no need to establish an MRL for D-Phe^6-LHRH and recommends its inclusion into Annex II of Council Regulation (EEC) No 2377/90 in accordance with the following table:

Pharmacologically active substance(s)	Animal species	Other provisions
D-Phe^6-luteinizing-hormone-releasing-hormone	All food producing species	