European Medicines Agency
Veterinary Medicines and Inspections

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COMMITTEE FOR MEDICINAL PRODUCTS FOR VETERINARY USE

BECLOMETASONE DIPROPIONATE

SUMMARY REPORT

1. Beclometasone dipropionate (9α-chloro-11β, 17α, 21-trihydroxy-16β-methylpregna-1, 4-diene-3, 20-dione 17, 21-dipropionate; CAS No. 5534-09-08) is a synthetic glucocorticoid with anti-inflammatory and anti-allergic properties. In veterinary medicine, beclometasone dipropionate is used for the treatment of recurrent airway obstruction (RAO) in the horse. It is administered by the inhalation route at a maximum recommended therapeutic dose of 500 µg/animal twice daily for 10 days. In human medicine, beclometasone dipropionate is indicated for the treatment of bronchial asthma in adults and children at a maximum daily dose of 400 µg/patient twice daily via the inhalation route. This latter dose in man may be modified upwards or downwards according to patient response. Beclometasone dipropionate is additionally used in human medicine as a nasal spray (maximum 400 µg/patient/day) for allergic and non-allergic rhinitis, and as a topical formulation (0.25%) for the treatment of various skin disorders.

2. The pharmacological activity of beclometasone dipropionate closely resembles that of other synthetic glucocorticoids. Beclometasone dipropionate has marked anti-inflammatory activity with a low potency for sodium retention and liver glycogen deposition. Studies in the rat have demonstrated that the glucocorticoid activity of beclometasone dipropionate was less than or equal to 10% that of dexamethasone in terms of suppression of the hypothalamic-pituitary-adrenal (HPA) axis. Suppression of the hypothalamic-pituitary-adrenal axis in healthy human volunteers and asthmatic patients was not significant at dose rates up to 1.0 mg/patient/day by the inhalation route. Beclometasone dipropionate has a 7-fold higher affinity for the human glucocorticoid receptor than cortisol, whilst the corresponding value for the 17-monopropionate metabolite is 192-fold higher. The effects of beclometasone dipropionate on tyrosine aminotransferase activity and corticosterone levels in the rat were evaluated in a GLP-compliant study. Beclometasone dipropionate produced no significant increase in tyrosine aminotransferase activity nor any significant alteration in corticosterone concentrations following daily oral administration of test compound for 7 days at dose rates of 0.0005, 0.001, 0.0015, 0.002 and 0.004 mg/kg bw. The pharmacological NOEL was therefore set at 0.004 mg/kg bw.

3. The pharmacokinetics of beclometasone dipropionate have been studied in rats, horses and humans. One significant limitation to the data relating to the rat was the use of tritium labels in the older studies available. Following oral administration of beclometasone dipropionate to rats at a dose rate of 4 mg/kg bw, T_{max} occurred at 3 hours. A second plasma peak occurred at approximately 6-7 hours, probably indicating biliary re-absorption. Most radioactivity was concentrated in liver, gastrointestinal tract, kidneys and adrenals. After 72 hours, 92% and 8% of the administered dose was eliminated in faeces and urine, respectively. No parent compound was detected in urine, faeces or bile. Levels of radioactivity excreted in faeces and urine increased in a linear fashion following daily oral dosing of rats at 1 mg/kg bw for 17 to 24 days. Blood concentrations gradually increased until day 14. Tissue radioactivity 24 hours after the last dose was highest in the liver, kidneys and adrenals.

7 Westferry Circus, Canary Wharf, London, E14 4HB, UK
Tel. (44-20) 74 18 84 00 Fax (44-20) 74 18 84 47
E-mail: mail@emea.eu.int http://www.emea.eu.int

A dose of 50 µg of tritium-labelled beclometasone dipropionate was administered by inhalation to rats. The results obtained were compared with tritium-labelled beclometasone dipropionate (1.5 mg/kg bw) administered by the intravenous and oral routes. At 3 minutes, 87% of the beclometasone dipropionate dose was present in the bronchi, with 2% present in lung. Approximately 28 to 38% of the administered beclometasone dipropionate dose was still present in lung and bronchi at the 8-hour post dose time point. Beclometasone dipropionate levels in plasma peaked at 20 minutes and then reached a plateau for several hours. In total, 88% of the administered dose was recovered in faeces. Beclometasone dipropionate was primarily metabolised to beclometasone monopropionate. A second, unidentified metabolite (less than 10% of tissue radioactivity) was also detected in lung. However, unchanged parent compound was not detected in plasma following oral treatment. The elimination half-life was in the order of 5-6 hours.

Beclometasone diproprionate was administered by intravenous infusion (1 mg), orally (4 mg), intranasally (1.34 mg) and by inhalation (1 mg) to twelve healthy human volunteers. The contribution of the lung, nose and gut to systemic exposure was assessed by repeating the inhaled, intranasal and oral dosing regimes together with activated charcoal to block oral absorption. Intravenous administration of beclometasone dipropionate (half life ($T_{1/2}$) equal to 0.5 h) was associated with rapid conversion to beclometasone 17-monopropionate, which was eliminated more slowly (half life ($T_{1/2}$) equal to 2.7 h). Beclometasone dipropionate was not detected in plasma following oral or intranasal dosing. The mean absolute bioavailability of inhaled beclometasone dipropionate was 2%; this was not reduced further by the co-administration of charcoal. The mean percentage bioavailability of the 17-monopropionate metabolite was 41%, 44% and 62% for oral, intranasal and inhaled dosing without charcoal, respectively. The corresponding estimates of nasal and lung absorption, based on the co-administration of charcoal, were less than 1% and 36%, respectively.

Four horses (2 male and 2 female) were administered 19 doses (2 doses per day) of beclometasone dipropionate by inhalation at the clinically recommended dose rate of 500 µg /animal in a GLP-compliant study. Following suitable washout periods in between each step (7 days), the horses were subsequently administered beclometasone dipropionate at a single low dose level (300 µg/animal), a single high dose level (1000 µg/animal) and finally a single intravenous dose of 500 µg/animal. Beclometasone dipropionate was not quantifiable in almost all plasma samples taken on day 1 and day 10 in the initial repeat-dose inhalation study, indicating the rapid metabolism of the parent molecule. Furthermore all plasma samples analysed for beclometasone-21-monopropionate and beclometasone were below the limit of quantification of the analytical technique (0.05 µg/l). Plasma beclometasone-17-monopropionate concentrations were invariably below 1 µg/l following administration of the clinically recommended dose of 500 µg beclometasone-17, 21-dipropionate by the inhalation route. Beclometasone-17-monopropionate was only quantifiable in plasma up to 8 hours after the last administration of 500 µg beclometasone-17, 21-dipropionate. Neither the 17-monopropionate metabolite nor parent compound were detected in urine at any time point in this study.

4. The acute oral LD_{50} values for beclometasone dipropionate were reported to be more than 3750 mg/kg bw in rats and more than 3000 mg/kg bw in mice. Subcutaneous acute LD_{50} values were more than 300 mg/kg bw in rats and more than 750 mg/kg bw in rabbits. The intraperitoneal acute LD_{50} was more than 1500 mg/kg bw in the rat. Asthenia was the major clinical sign observed in such studies.

5. Two pre-GLP repeated dose oral toxicity studies were performed in rats. In the first study, daily doses of 0.16, 1.6, 8, 16 and 32 mg/kg bw were administered for 26 weeks. A reduction in body weight gain was observed at dose rates above or equal to 1.6 mg/kg bw, although a true dose-response relationship was lacking. A dose-dependent reduction in the white blood cell count, and a dose-dependent increase in the red blood cell count and haemoglobin concentrations (females) were evident in all treated groups. Alopecia was noted in all female treated groups. A significant reduction (with occasional exceptions) was seen in the weights of the thymus and spleen in both sexes at all treatment levels, and uterine weights at dose rates of more than or equal to 8 mg/kg bw. Significant decreases in organ weights were also observed in the liver (more than or equal to 0.16 mg/kg bw) and lungs (more than or equal to 32 mg/kg bw).

Significant increases in relative organ weight were recorded in the brain, kidney, testes and epididymis in males at dose rates of more than or equal to 1.6 mg/kg bw. Histological changes correlated to the dose group were observed in the thymus (atrophy at all dose rates), adrenal gland (congestion at dose rates of more than or equal to 8 mg/kg bw), spleen (atrophy) and uterus (atrophy at dose rates more than or equal to 8 mg/kg bw). A NOEL could not be retained from this study. In the second study, beclometasone dipropionate was administered orally to groups of rats (n equal to 15 animals/group) at dose rates of 0, 1.6 and 8 mg/kg bw each day for 3 months. Reductions in food consumption and body weight gains were evident from the second week of treatment. A reduction in the total white blood cell count, accompanied by neutrophilia and lymphopoenia, was recorded at the end of the treatment period in both dosage groups. Significant alterations in the serum concentrations of glucose, lactate dehydrogenase (females), chloride (females) and various globulin fractions were observed at both treatment levels. The absolute weights of the thymus, spleen, uterus, liver (males) and kidneys (females) were significantly reduced in both beclometasone treated groups. Histological examination revealed atrophy of the thymus and uterus, along with congestion in the adrenal glands of females. Following a 1-month recovery period, some of the lesions observed had regressed. Nevertheless, a NOEL could not be retained from this study.

6. Beclometasone dipropionate was administered by inhalation to five groups of Sprague-Dawley rats (24 rats/sex/group) for 1 hour/day for 90 consecutive days. Three groups were exposed to aerosols of the beclometasone dipropionate formulation at target concentrations of 0.001, 0.005 and 0.025 mg/l. The two remaining groups were exposed to either the placebo formulation or filtered air. Only minor clinical signs of red material around the nose and eyes were recorded. Concentration-related decreases in mean body weights, which were generally statistically significant, were observed in all three beclometasone-exposed groups, beginning after approximately 6 weeks of exposure and continuing for the duration of the 90-days. Mean cumulative body weight gains were significantly decreased from the third week and for the duration of the 90-day exposure phase. Exposure-related decreases in absolute thymus, spleen and liver (males only) weights were observed in all three beclometasone-treated groups. Haematological effects were limited to significantly decreased white blood cell counts in rats exposed to the high concentration of beclometasone dipropionate. This effect was most likely related to the decreased number of lymphocytes present. Many of the changes noted in organ weights and haematological values had disappeared following an 8-week recovery period. An increased mean corpuscular haemoglobin concentration was observed in high concentration-exposed males and females, as well as in placebo-exposed females, at the end of the eight-week recovery period. Test article-related depletion of thymic lymphocytes was observed microscopically at the mid- and high-dose rates. A NOEL could not be established from this study.

7. Beclometasone dipropionate was administered to beagle dogs (8 animals/group) at nominal aerosol dose levels of 0.05, 0.16 and 0.50 mg/kg/day over a maximum of 52 weeks. Additional control groups were exposed to a chlorofluorocarbon (CFC)-beclometasone formulation at a dose level of 0.50 mg/kg/day, placebo formulation or air. A total of 9 animals receiving the higher dose levels were euthanized due to the onset of severe secondary demodicosis. Treatment related clinical signs that were dose-related also included abdominal distension, excess body fat and hair loss. Premature decedents receiving high doses of beclometasone dipropionate had body weight stasis or body weight loss prior to sacrifice. Surviving males in the mid- and high-dose groups showed a gradual weight loss over the final 10 to 12 weeks of treatment. Haematological examination revealed significant changes in mean cell haemoglobin, mean cell volume, mean cell haemoglobin concentration and reticulocyte percentages. Changes in white blood cell parameters were evident in the high-dose group. Significant biochemical changes (principally in the high-dose group) included increases in urea, alkaline phosphatase, total protein, albumin, globulin, triglycerides and reductions in aspartate aminotransferase and creatinine. Glucose levels were lower in beclometasone treated dogs. Organ weight changes included reduced adrenal weights for all treated groups, as well as increased liver weights for males (mid- and high-dose). Histological examination revealed changes in the adrenal glands (atrophy), liver (clear vacuolation), lymphoid tissue (lymphoid depletion and germinal centre atrophy), skin (parasitic dermatitis) and bone marrow (hypoplastic haematopoietic component).

Although histological evidence of adrenal pathology was only detected in the mid- and high-dose groups, a NOEL was not retained from this study due to the significant decline in adrenal weights in males at the lowest dose tested.

8. A tolerance study was conducted in the target species in which beclometasone dipropionate was administered by inhalation to groups of horses (6 animals/group) at dose levels of 5, 15 or 25 puffs twice daily (representing 1 time, 3 times and 5 times the recommended therapeutic dose). However, subsequent analysis revealed that the aerosol device delivered less medication than originally anticipated. Treatment was administered for 42 days. A control group (*n* equal to 6) received 25 puffs of aerosol placebo twice a day. There were no drug-related clinical observations noted during the study. Haematology revealed some treatment-related effects at the higher dose levels tested, whilst blood glucose levels were increased in the 5 times dose group. Cortisol levels were significantly reduced in a dose-dependent manner in the 3 times dose and 5 times treatment groups. Whilst slight reductions in cortisol levels were noted in the 1 time dose group, most values remained within the normal range. Following completion of a recovery period, cortisol values were generally comparable to original baseline values. After adenocorticotropic homone (ACTH) stimulation, increases in cortisol values were considerably lower amongst the 3 times and 5 times dose treated group animals when compared to pre-study baseline and control values. Reduced effect of adenocorticotropic homone (ACTH) stimulation was noted as well on day 29 and 43 amongst the group 3 and 4 animals. No treatment-related gross necropsy findings were noted among the animals sacrificed immediately following study termination. No lesions were present in the nasopharynx or oropharynx of any treated horses. During the recovery period, there were no clinical signs attributable to abrupt steroid withdrawal in the 1 time and 3 times dosing groups.

9. Two pre-GLP reproductive toxicity studies were reported in the rat with beclometasone dipropionate. In the first study, test compound was administered at dose rates of 0.016, 0.16, 1.6 and 16 mg/kg bw to male and female Wistar rats. Males were treated for 62 days prior to mating and females for 14 days. Females continued to receive beclometasone dipropionate until day 7 of gestation. Although systemic treatment-related effects were evident in males even at the lowest dose tested, mating performance and fertility were unaffected at all dose levels employed. The number of pregnant females declined significantly at the 16 mg/kg bw dose rate. Additional effects noted in females and foetuses included reduced food consumption and body weight gain post-gestation, a decrease in the number of implantations and live foetuses (1.6 mg/kg bw group), and delays in ossification of the sixth sternebrae, caudal veretebrae and metacarpals of the front paws (16 mg/kg bw). The NOEL for reproductive toxicity in this study was 0.16 mg/kg bw. In the second report, beclometasone dipropionate was administered orally at dose rates of 0.016, 0.16, 1.6 and 16 mg/kg bw to Wistar rats from day 17 of pregnancy to day 28 post-partum. A non-significant prolongation of the gestation period, a significant reduction in birth rate and a significant increase in the number of dead foetuses/pups were observed at dose rates of more than or above 1.6 mg/kg bw. Affected dams also displayed evidence of reduced motor activity, reduced body weight and adrenal gland pathology. General parameters relating to growth, behaviour and reproductive function in the F_1 generation were unaffected by treatment except that the period for vaginal opening was prolonged at dose rates greater than or above 0.16 mg/kg bw. The dose rate of 0.016 mg/kg bw can be retained as a NOEL for reproductive toxicity in this study.

10. A series of pre-GLP teratogenicity studies were conducted in three separate laboratory species. In rats, beclometasone dipropionate was administered at dose rates of 0.016, 0.16, 1.6 and 16 mg/kg bw from days 7 to 17 of pregnancy. Decreases in foetal body weights and an increase in the percentage incidence of fifth rudimentary sternebrae were observed at dose rates more than or equal to 1.6 mg/kg bw. Suppression of maternal body weight gain and a reduction in the body weights of F_1 progeny at birth were observed at dose rates more than or equal to 16 mg/kg bw, whilst individual maternal organ weights were significantly reduced at dose rates more than or equal to 0.16 mg/kg bw. Congestion of adrenal glands was noted in the progeny of treated dams (16 mg/kg bw). The dose rate of 0.16 mg/kg bw can be set as a NOEL for teratogenicity in this study.

Pregnant mice were administered beclometasone dipropionate orally at dose rates of 0, 0.08, 0.8, 8 and 64 mg/kg bw from the day 7 to 15 of pregnancy. A suppression of maternal body weight gain, an increase in the number of dead foetuses and a decrease in birth rate were evident at dose rates more than or equal to 8 mg/kg bw. A significant decrease in foetal body weights at birth was evident in all treated groups. A dose-related increase in the frequency of cleft palate was observed at treatment rates more than or equal to 0.8 mg/kg bw. In addition, a dose-dependent decline in the number of ossified caudal vertebrae was evident in foetuses from all treated groups. Various post-natal developmental abnormalities relating to reproductive function were observed at later time points (descent of testes, opening of vagina). A NOEL for teratogenic effects could not be set in this study.

Pregnant rabbits were administered beclometasone dipropionate orally at dose rates of 0, 0.04, 0.08, 0.16 and 0.4 mg/kg bw from the day 7 to 18 of pregnancy. An increase in the number of dead foetuses and a decrease in body weights of live foetuses were observed at dose rates of more than or equal to 0.16 mg/kg bw. This latter effect on body weights was also evident in female foetuses at the lowest dose tested. A dose-dependent increase in the incidence of cleft palate was observed in all treatment groups, although the increase was not statistically significant at the lowest dose tested. Significant declines in the presence of the thirteenth rib, ossification of sternebrae and the number of caudal vertebrae were noted at dose rates more than or equal to 0.16 mg/kg bw. An increase in keratolysis of the head and open eyelids was observed at the highest dose tested. Due to the dose-dependent increase in the incidence of cleft palate, a NOEL for teratogenic effects was not established in this study.

11. A series of GLP-compliant mutagenicity studies were conducted with beclometasone dipropionate. Negative results were obtained in an *in vitro* assay for gene mutation in *Salmonella typhimurium* TA98, TA100, TA102, TA1535 and TA1537 using test concentrations of 30 to 300 µg/plate with and without metabolic activation. In an *in vitro* chromosomal aberration assay using cultured human lymphocytes, test concentrations of 10 to 250 µg beclometasone dipropionate/ml did not induce a reproducible increase in structural chromosomal aberrations when tested in the presence and absence of metabolic activation. In addition, no signs for a treatment-related induction of polyploid cells were evident in this assay. An *in vivo* micronucleus assay was conducted in the mouse in which beclometasone dipropionate was administered by two intraperitoneal injections (24 hours apart) to groups of male mice (*n* equal to 5 animals/group) at test concentrations of 500, 1000 and 2000 mg/kg bw. There was a statistically significant decrease in polychromatic erythrocytes in all dose groups compared to negative controls, which was most likely related to bone marrow toxicity indicating that beclometasone dipropionate is distributed into this tissue. No consistent increase in the number of polychromatic erythrocytes containing micronuclei (0.19 to 0.25%) was observed after dosing with beclometasone dipropionate compared to the corresponding negative control (0.17%). The mid-dose group showed a slight but statistically significant increase in polychromatic erythrocytes containing micronuclei (0.25%) compared to controls. However, due to the lack of a dose-response relationship, this finding was considered incidental. The data available from this series of *in vitro* and *in vivo* studies indicate that beclometasone dipropionate is not mutagenic.

12. No carcinogenicity data were submitted for beclometasone dipropionate. However, in keeping with previous assessments for other synthetic glucocorticoids, the CVMP considered that carcinogenicity studies were unnecessary in light of the absence of any structural alerts for this family of compounds, the negative mutagenicity data and the absence of any carcinogenic concerns following the long-term topical and inhalation use of this substance in man.

13. No data were provided on the potential microbiological properties of residues of beclometasone dipropionate, including potential effects on the human gut flora and micro-organisms used in industrial food processing. However, taking into account the chemical properties of this compound, no such data were considered necessary.

14. No data specifically addressing the immunotoxic properties of beclometasone dipropionate were provided. However, reference was made to the effects of beclometasone dipropionate on peripheral leucocyte counts and the weight/morphological appearance of lymphoreticular tissues in the various repeat dose toxicity experiments conducted. In keeping with other members of this family, beclometasone dipropionate has potent immunosuppressive effects when administered at sufficiently high dose rates. However, the effects on leucocyte counts and lymphoreticular tissue in general were often reversible in several of the safety studies in which recovery periods were employed.

15. Beclometasone dipropionate is widely used in human medicine for the treatment of bronchial asthma, rhinitis and various skin diseases. Formulations available include aerosols, inhalation capsules or discs containing powder for inhalation, and topical creams or ointments. Additional indications include adenoidal hypertrophy, bronchopulmonary dysplasia, recurrent cough and inflammatory bowel disease. The maximum dose in man (often divided) is usually below 1000 µg/patient/day. However, the dose rates selected can vary according to severity of disease and patient response. Dose rates for nasal sprays used in the treatment of allergic and non-allergic rhinitis are usually lower (maximum 400 µg/patient/day). The pharmacokinetics of inhaled beclometasone dipropionate in man were addressed previously. Enzymatic hydrolysis of parent compound rapidly produces the 17α-monopropionate metabolite, which is pharmacologically active. Further hydrolytic steps produce the free alcohol, which is virtually devoid of glucocorticoid activity. Most of an administered dose of beclometasone dipropionate is eliminated in faeces, mainly as metabolites. Adverse effects of beclometasone dipropionate in man include adrenal suppression, candidiasis in adults, effects on bone metabolism, as well as those other effects that are common to most members of this family.

    A suppressed growth rate has been reported in asthmatic children (particularly prepubertal children) receiving conventional dose rates of 400 µg beclometasone dipropionate/day for 7-12 months by inhalation. It was noted, however, that this adverse effect on growth was transient with treated individuals attaining an adult height indistinguishable from their predicted height, and no different from that of non-asthmatic individuals. It was further noted that the dose rates required for such an effect were higher than the pharmacological NOEL identified for the induction of tyrosine aminotransferase activity in the rat.

16. No data on the potential neurotoxic effects of beclometasone dipropionate were presented. Although glucocorticoids are not considered to be primarily neurotoxic, the use of such agents has been associated with a very low incidence of paraesthesias, irritation, mental disturbance and intracranial hypertension in man. However, as the doses required for such effects are clearly within the pharmacological range, and as alerts for neurotoxicity were not identified in the safety studies submitted, no additional data were required.

17. A pharmacological ADI of 0.00004 mg/kg bw (i.e. 0.0024 mg/person) was calculated for beclometasone dipropionate by applying a safety factor of 100 to the NOEL of 0.004 mg/kg bw for the induction of tyrosine aminotransferase activity in the rat.

    Although clear NOELs could not be established in many of the repeat dose toxicity studies conducted, most of the effects observed in such studies were primarily attributable to the pharmacological action of beclometasone dipropionate, it is considered appropiate to establish an ADI based on the pharmacological NOEL.

18. No radiometric or cold residue depletion studies were conducted with beclometasone dipropionate in the target species. No development or validation data for a proposed regulatory method to detect residues of beclometasone dipropionate in edible tissues were provided.

Conclusions and recommendation

Having considered that:

- a pharmacological ADI of 0.00004 mg/kg bw (i.e. 0.0024 mg/person) was established for beclometasone dipropionate,
- beclometasone dipropionate will be used in a small number of individual animals for infrequent and irregular treatment,
- the administration of therapeutic doses by the inhalation route results in low systemic exposure (i.e. 1 µg/kg bw),
- pharmacokinetic data in the target species indicate that parent compound is rapidly metabolised, with no bioaccumulation of parent compound or pharmacologically active metabolites following repeat daily dosing by the inhalation route;

the Committee for Medicinal Products for Veterinary Use concludes that there is no need to establish an MRL for beclometasone dipropionate in *Equidae* for inhalation use and recommends its inclusion in Annex II of Council Regulation (EEC) No. 2377/90 in accordance with the following table :

Pharmacologically active substance(s)	Animal species	Other provisions
Beclometasone dipropionate	*Equidae*	For inhalation use only