ASSAY

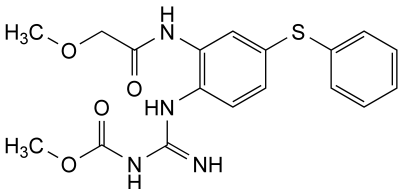
Liquid chromatography (*2.2.29*) as described in the test for related substances with the following modification.

Injection: test solution (b) and reference solution (b).

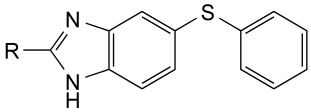
Calculate the percentage content of $C_{20}H_{22}N_4O_6S$ from the declared content of *febantel CRS*.

IMPURITIES

Specified impurities: A, B, C.



A. methyl [[2-[(methoxyacetyl)amino]-4-(phenylsulphanyl)phenyl]carbamimidoyl]carbamate,



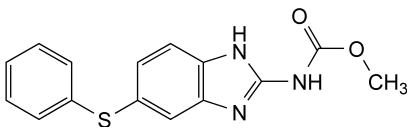
B. R = CH_2-OCH_3: 2-(methoxymethyl)-5-(phenylsulphanyl)-1*H*-benzimidazole,

C. R = NH-CO-OCH_3: methyl [5-(phenylsulphanyl)-1*H*-benzimidazol-2-yl]carbamate (fenbendazole).

01/2008:1208
corrected 6.0

FENBENDAZOLE FOR VETERINARY USE

Fenbendazolum ad usum veterinarium



$C_{15}H_{13}N_3O_2S$ M_r 299.4
[43210-67-9]

DEFINITION

Methyl [5-(phenylsulphanyl)-1*H*-benzimidazol-2-yl]carbamate.

Content: 98.0 per cent to 101.0 per cent (dried substance).

CHARACTERS

Appearance: white or almost white powder.

Solubility: practically insoluble in water, sparingly soluble in dimethylformamide, very slightly soluble in methanol.

IDENTIFICATION

Infrared absorption spectrophotometry (*2.2.24*).

Preparation: discs.

Comparison: *fenbendazole CRS*.

TESTS

Related substances. Liquid chromatography (*2.2.29*).

Test solution. Dissolve 50.0 mg of the substance to be examined in 10.0 ml of *hydrochloric methanol R*.

Reference solution (a). Dissolve 50.0 mg of *fenbendazole CRS* in 10.0 ml of *hydrochloric methanol R*. Dilute 1.0 ml of this solution to 200.0 ml with *methanol R*. Dilute 5.0 ml of the solution to 10.0 ml with *hydrochloric methanol R*.

Reference solution (b). Dissolve 10.0 mg of *fenbendazole impurity A CRS* in 100.0 ml of *methanol R*. Dilute 1.0 ml of this solution to 10.0 ml with *hydrochloric methanol R*.

Reference solution (c). Dissolve 10.0 mg of *fenbendazole impurity B CRS* in 100.0 ml of *methanol R*. Dilute 1.0 ml of this solution to 10.0 ml with *hydrochloric methanol R*.

Reference solution (d). Dissolve 10.0 mg of *fenbendazole CRS* and 10.0 mg of *mebendazole CRS* in 100.0 ml of *methanol R*. Dilute 1.0 ml of this solution to 10.0 ml with *hydrochloric methanol R*.

Column:

– *size*: *l* = 0.25 m, Ø = 4.6 mm;
– *stationary phase*: *octadecylsilyl silica gel for chromatography R* (5 µm).

Mobile phase:

– *mobile phase A*: *anhydrous acetic acid R, methanol R, water R* (1:30:70 *V/V/V*);
– *mobile phase B*: *anhydrous acetic acid R, water R, methanol R* (1:30:70 *V/V/V*);

Time (min)	Mobile phase A (per cent *V/V*)	Mobile phase B (per cent *V/V*)
0 - 10	100 → 0	0 → 100
10 - 40	0	100
40 - 50	0 → 100	100 → 0

Flow rate: 1 ml/min.

Detection: spectrophotometer at 280 nm.

Injection: 10 µl.

Retention time: fenbendazole = about 19 min.

System suitability: reference solution (d):

– *resolution*: minimum 1.5 between the peaks due to fenbendazole and mebendazole.

Limits:

– *impurity A*: not more than 2.5 times the area of the corresponding peak in the chromatogram obtained with reference solution (b) (0.5 per cent);
– *impurity B*: not more than 2.5 times the area of the corresponding peak in the chromatogram obtained with reference solution (c) (0.5 per cent);
– *any other impurity*: for each impurity, not more than twice the area of the principal peak in the chromatogram obtained with reference solution (a) (0.5 per cent);
– *total*: not more than 4 times the area of the principal peak in the chromatogram obtained with reference solution (a) (1 per cent);
– *disregard limit*: 0.2 times the area of the principal peak in the chromatogram obtained with reference solution (a) (0.05 per cent).

Heavy metals (*2.4.8*): maximum 20 ppm.

1.0 g complies with test C. Prepare the reference solution using 2 ml of *lead standard solution (10 ppm Pb) R*.

Loss on drying (*2.2.32*): maximum 1.0 per cent, determined on 1.000 g by drying in an oven at 105 °C for 3 h.

Sulphated ash (*2.4.14*): maximum 0.3 per cent, determined on 1.0 g.

ASSAY

Dissolve 0.200 g in 30 ml of *anhydrous acetic acid R*, warming gently if necessary. Cool and titrate with *0.1 M perchloric acid*, determining the end-point potentiometrically (*2.2.20*).

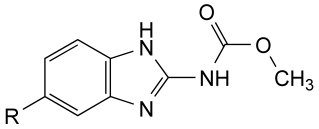
1 ml of *0.1 M perchloric acid* is equivalent to 29.94 mg of $C_{15}H_{13}N_3O_2S$.

STORAGE

Protected from light.

IMPURITIES

Specified impurities: A, B.



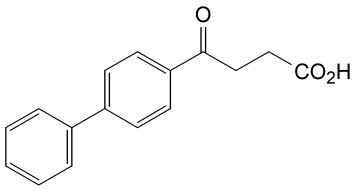
A. R = H: methyl (1*H*-benzimidazol-2-yl)carbamate,

B. R = Cl: methyl (5-chloro-1*H*-benzimidazol-2-yl)carbamate.

01/2008:1209
corrected 6.0

FENBUFEN

Fenbufenum



$C_{16}H_{14}O_3$ M_r 254.3
[36330-85-5]

DEFINITION

4-(Biphenyl-4-yl)-4-oxobutanoic acid.

Content: 98.5 per cent to 101.0 per cent (dried substance).

CHARACTERS

Appearance: white or almost white, fine, crystalline powder.

Solubility: very slightly soluble in water, slightly soluble in acetone, in ethanol (96 per cent) and in methylene chloride.

IDENTIFICATION

First identification: B.

Second identification: A, C.

A. Melting point (*2.2.14*): 186 °C to 189 °C.

B. Infrared absorption spectrophotometry (*2.2.24*).
Comparison: *fenbufen CRS*.

C. Thin-layer chromatography (*2.2.27*).
Test solution. Dissolve 10 mg of the substance to be examined in *methylene chloride R* and dilute to 10 ml with the same solvent.
Reference solution (a). Dissolve 10 mg of *fenbufen CRS* in *methylene chloride R* and dilute to 10 ml with the same solvent.
Reference solution (b). Dissolve 10 mg of *ketoprofen CRS* in *methylene chloride R* and dilute to 10 ml with the same solvent. To 5 ml of this solution, add 5 ml of reference solution (a).
Plate: *TLC silica gel F_{254} plate R*.
Mobile phase: *anhydrous acetic acid R, ethyl acetate R, hexane R* (5:25:75 *V/V/V*).
Application: 10 µl.
Development: over a path of 15 cm.
Drying: in air.
Detection: examine in ultraviolet light at 254 nm.
System suitability: reference solution (b):
– the chromatogram shows 2 clearly separated spots.
Results: the principal spot in the chromatogram obtained with the test solution is similar in position and size to the principal spot in the chromatogram obtained with reference solution (a).

TESTS

Related substances. Liquid chromatography (*2.2.29*).

Solvent mixture: *dimethylformamide R*, mobile phase A (40:60 *V/V*).

Test solution. Dissolve 50.0 mg of the substance to be examined in the solvent mixture and dilute to 10.0 ml with the solvent mixture.

Reference solution (a). Dilute 0.5 ml of the test solution to 50.0 ml with the solvent mixture. Dilute 1.0 ml of this solution to 10.0 ml with the solvent mixture.

Reference solution (b). Dissolve 25 mg of *fenbufen CRS* and 6 mg of *ketoprofen CRS* in the solvent mixture and dilute to 10 ml with the solvent mixture. Dilute 1 ml of this solution to 100 ml with the solvent mixture.

Column:
– *size*: *l* = 0.125 m, Ø = 4.0 mm;
– *stationary phase*: *octadecylsilyl silica gel for chromatography R* (5 µm).

Mobile phase:
– *mobile phase A*: mix 32 volumes of *acetonitrile R* and 68 volumes of a mixture of 1 volume of *glacial acetic acid R* and 55 volumes of *water R*;
– *mobile phase B*: mix 45 volumes of *acetonitrile R* and 55 volumes of a mixture of 1 volume of *glacial acetic acid R* and 55 volumes of *water R*;

Time (min)	Mobile phase A (per cent *V/V*)	Mobile phase B (per cent *V/V*)
0 – 15	100	0
15 – 20	100 → 0	0 → 100
20 – 35	0	100
35 – 40	0 → 100	100 → 0
40 - 45	100	0

Flow rate: 2 ml/min.

Detection: spectrophotometer at 254 nm.

Injection: 20 µl.

System suitability: reference solution (b):
– *resolution*: minimum 5.0 between the peaks due to ketoprofen and fenbufen.

Limits:
– *any impurity*: for each impurity, not more than the area of the principal peak in the chromatogram obtained with reference solution (a) (0.1 per cent);
– *total*: not more than 5 times the area of the principal peak in the chromatogram obtained with reference solution (a) (0.5 per cent);
– *disregard limit*: 0.2 times the area of the principal peak in the chromatogram obtained with reference solution (a) (0.02 per cent).