World Health Organization

DIETHYLCARBAMAZINE DIHYDROGEN CITRATE

(Diethylcarbamazini dihydrogenocitras)

Draft proposal for inclusion in *The International Pharmacopoeia*

(May 2024)

DRAFT FOR COMMENTS

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For any technical questions, you may contact **Dr Herbert Schmidt**, Technical Officer, Norms and Standards for Pharmaceuticals, Technical Standards and Specifications (schmidth@who.int), with a copy to Ms Sinéad Jones (jonessi@who.int, nsp@who.int).
Comments should be submitted through the online platform on or by **31 July 2024**. Please note that only comments received by this deadline will be considered for the preparation of this document.
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SCHEDULE FOR THE ADOPTION PROCESS OF DOCUMENT QAS/21.896:

DIETHYLCARBAMAZINE DIHYDROGEN CITRATE

(DIETHYLCARBAMAZINI DIHYDROGENOCITRAS)

Description	Date
Monograph revision drafted following up on information received from the custodian centre for the establishment, storage and distribution of ICRS, the European Directorate for the Quality of Medicines and HealthCare.	July 2021
Discussion at the Consultation on Quality Control and Pharmacopoeial Specifications	April 2024
Draft revision sent out for public consultation	June – July 2024
Presentation to the 58th WHO Expert Committee on Specifications for Pharmaceutical Preparations.	October 2024
Further follow-up action as required.	

*[**Note from the Secretariat.** It is proposed to revise the monograph on Diethylcarbamazine dihydrogen citrate to avoid the use of organoleptic tests and of toxic solvents.]*

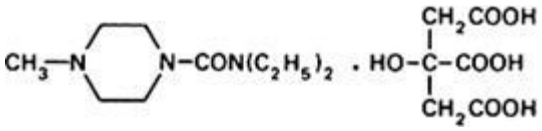
DIETHYLCARBAMAZINE DIHYDROGEN CITRATE

(DIETHYLCARBAMAZINI DIHYDROGENOCITRAS)

Molecular formula. $C_{10}H_{21}N_3O,C_6H_8O_7$ or $C_{16}H_{29}N_3O_8$

Relative molecular mass. 391.4

Graphic formula.



Chemical name. *N*,*N*-Diethyl-4-methyl-1-piperazinecarboxamide citrate (1:1); *N*,*N*-diethyl-4-methyl-1-piperazinecarboxamide 2-hydroxy-1,2,3-propanetricarboxylate (1:1).

CAS Registry Number. 1642-54-2.

Description. A white, crystalline powder~~; odourless or almost odourless~~.

Solubility. Very soluble in water R; soluble in 35 parts of ethanol (~750 g/L) TS; practically insoluble in acetone ~~ether~~ R.

Category. Filaricide.

Storage. Diethylcarbamazine dihydrogen citrate should be kept in a tightly closed container, protected from light.

Additional information. Diethylcarbamazine dihydrogen citrate is hygroscopic~~; it has an acid and bitter taste~~. Even in the absence of light, Diethylcarbamazine

dihydrogen citrate is gradually degraded on exposure to a humid atmosphere, the decomposition being faster at higher temperatures.

Requirements

Definition. Diethylcarbamazine dihydrogen citrate contains not less than 98.0% and not more than 101.0% of $C_{10}H_{21}N_3O,C_6H_8O_7$, calculated with reference to the anhydrous substance.

Identity tests

- Either tests A and D, or tests B, C and D may be applied.

A. Carry out the test as described under *1.7 Spectrophotometry in the infrared region*. The infrared adsorption spectrum is concordant with the spectrum obtained from diethylcarbamazine dihydrogen citrate RS or with the reference spectrum of diethylcarbamazine dihydrogen citrate. ~~Dissolve 0.05 g in 25 mL of water. Add 1 mL of sodium hydroxide (~80 g/l) TS and 4 mL of carbon disulfide R, and shake for 2 minutes. Separate the aqueous layer. Centrifuge the lower layer if necessary, and filter through a dry filter, collecting the filtrate in a small flask provided with a glass stopper. Carry out the examination of the filtered solution using carbon disulfide R as the blank as described under 1.7 Spectrophotometry in the infrared region. The infrared absorption spectrum is concordant with the spectrum obtained from diethylcarbamazine dihydrogen citrate RS treated similarly or with the *reference spectrum* of diethylcarbamazine base~~.

B. Carry out the test as described under *1.14.1 Chromatography*, Thin-layer chromatography, using the conditions given under "Impurities A and B" with the following modifications. Use solution (A) as described. For solution (B), prepare a solution containing 50 mg of diethylcarbamazine dihydrogen citrate RS per mL of methanol R. The principal spot in the chromatogram obtained

with solution (A) corresponds in position, appearance and intensity with the spot due to diethylcarbamazine in the chromatogram obtained with solution (B). ~~Dissolve 0.5 g in 10 mL of water, add 10 mL of sodium hydroxide (1 mol/l)VS, and extract with 4 successive quantities, each of 5 mL of chloroform R. Retain the aqueous layer for test C. Wash the combined chloroform extracts with water, filter through a plug of cotton wool, and evaporate the chloroform. Add 1 mL of ethyl iodide R to the residue, and heat gently under a reflux condenser for 5 minutes. Cool, separate the viscous yellow oil, and dissolve it in ethanol (~750 g/l) TS. Add, with continuous stirring, sufficient ether R to precipitate the quaternary ammonium salt, and filter. Dissolve the precipitate in ethanol (~750 g/l) TS, reprecipitate with ether R, and dry at 105°C; melting temperature, about 152°C (1-diethylcarbamoyl-4-methylpiperazine ethiodide).~~

C. Dissolve 0.1 g of the test substance in 5 mL of water R. The solution ~~aqueous layer from test B~~ yields reaction B described under *2.1 General identification tests* as characteristic of citrates.

D. Melting temperature, after drying at 80 °C, about 137 °C.

Heavy metals. Use 1.0 g for the preparation of the test solution as described under *2.2.3 Limit test for heavy metals*, Procedure 1: determine the heavy metals content according to Method A; not more than 20 µg/g.

Sulfated ash. Not more than 1.0 mg/g.

Clarity and colour of solution. Dissolve 2.5 g in carbon-dioxide-free water R and dilute to 25 mL with the same solvent. The solution not more intensely coloured than reference solution BY6 when compared as described under *1.11.2 Degree of coloration of liquids*.

~~**Water.** Determine as described under *2.8 Determination of water by the Karl Fischer method*, Method A, using about 1 g of the substance; the water content is not more than 10 mg/g.~~

Loss on drying. Dry 1.000 g of the test substance at 60 °C for 4 hours; it loses not more than 5 mg/g.

~~**pH value.** pH of a 30 mg/mL solution, 3.5-4.5.~~

~~***N*-Methylpiperazine.** Carry out the test as described under *1.14.1 Thin-layer chromatography*, using silica gel R1 as the coating substance and a mixture of 6 volumes of ethanol (~750 g/L) TS, 3 volumes of glacial acetic acid R and 1 volume of water as the mobile phase. Apply separately to the plate 5 µl of each of 2 solutions in methanol R containing (A) 50 mg of the test substance per mL and (B) 0.050 mg of *N*-methylpiperazine R per mL. After removing the plate from the chromatographic chamber, allow it to dry in air, spray with a mixture of 3 volumes of platinic chloride (60 g/L) TS, 97 volumes of water and 100 volumes of potassium iodide (60 g/L) TS, and examine the chromatogram in daylight. The spot obtained with solution B is more intense than any spot, corresponding in position and appearance, obtained with solution A.~~

Impurities A and B. Carry out the test as described under *1.14.1 Chromatography*, Thin-layer chromatography, using silica gel R5 as the coating substance and a mixture of 5 volumes of ammonia (~260 g/L) TS, 30 volumes of methyl ethyl ketone R and 65 volumes of methanol R as the mobile phase. Develop the plate for 2/3 of its height. Apply separately to the plate 10 µl of each of 3 solutions in methanol R containing (A) 50 mg of the test substance per mL, (B) 0.10 mg of *N*-methylpiperazine R (impurity A) per mL and (C) 0.10 mg of dimethylpiperazine R (impurity B). After removing the plate from the chromatographic chamber, dry it at 100-105 °C and place it in a chamber with iodine vapour of 30 minutes. Examine the plate in daylight.

In the chromatogram obtained with solution (A)

- any spot due to impurity A is not more intense than the corresponding spot in the chromatogram obtained with solution (B) (0.2 per cent);
- any spot due to impurity B is not more intense than the corresponding spot in the chromatogram obtained with solution (C) (0.2 per cent).

Related substances. Carry out the test as described under *1.14.1 Chromatography*, High-performance liquid chromatography, using a stainless steel column (15 cm x 3.9 mm) packed with end-capped particles of silica gel, the surface of which has been modified with chemically-bonded octadecylsilyl groups (5 µm).·As the mobile phase use a mixture of 100 volumes of methanol for chromatography R and 900 volumes of a 10 g/L solution of potassium dihydrogen phosphate R (*V*/*V*).

Operate with a flow rate of 0.8 mL per minute. As a detector use an ultraviolet spectrophotometer set at a wavelength of 220 nm.

Prepare the following solutions:

For solution (A), dissolve 31.2 g of potassium dihydrogen phosphate R in water R and dilute to 1000 mL with the same solvent.

For solution (1), suspend 0.30 g of the test substance in solution (A) and dilute to 100 mL with solution (A). Filter or centrifuge and use the clear filtrate or supernatant.

For solution (2), dilute 1.0 mL of solution (1) to 100.0 mL with solution (A).

For solution (3), dilute 5.0 mL of solution (2) to 100.0 mL with solution (A).

For solution (4), dissolve 10 mg of citric acid R in solution (A) and dilute to 10 mL with solution (A).

For solution (5), add 0.5 mL of hydrogen peroxide (~330 g/L) TS to 3 mL of solution (1). Maintain the solution at 80 °C for 3 h. Dilute to 100 mL with solution (A).

Inject 20 µL each of solution (1), (2), (3), (4) and (5) and record the chromatograms for about twice the retention time of diethylcarbamazine.

Use the chromatogram obtained with solution (4) to identify the peak due to citrate and the chromatogram obtained with solution (5) to identify the peak due to the degradation product. The impurities are eluted, if present, at the following relative retention with reference to diethylcarbamazine (retention time about 7 minutes): citrate about 0.2; degradation product about 1.6.

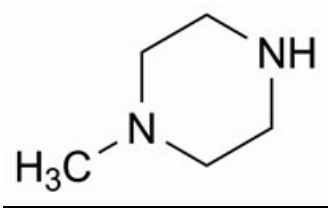
The test is not valid unless in the chromatogram obtained with solution (4) the resolution between the peaks due to diethylcarbamazine and the degradation product is at least 5. Also, the test is not valid unless in the chromatogram obtained with solution (3) the peak due to diethylcarbamazine is detected with a signal-to-noise ratio of at least 10.

In the chromatogram obtained with solution (1):

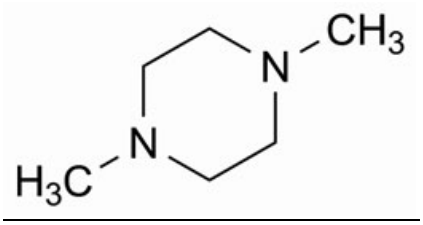
- the area of any impurity peak is not greater than 0.1 times the area of the peak due to dimethylcarbamazine in the chromatogram obtained with solution (2) (0.10 %).
- The sum of the areas of all impurity peaks is not greater than 0.5 times the area of the peak due to dimethylcarbamazine in the chromatogram obtained with solution (2) (0.5 %). Disregard any peak with an area less than the area of the peak due to dimethylcarbamazine in the chromatogram in the chromatogram obtained with solution (3) (0.05%). Disregard the peak due to citrate.

Assay. Dissolve about 0.350 g~~, accurately weighed,~~ in 30 mL of glacial acetic acid R1, and titrate with perchloric acid (0.1 mol/L) VS as described under *2.6 Non-aqueous titration*, Method A. Each mL of perchloric acid (0.1 mol/L) VS is equivalent to 39.14 mg of $C_{10}H_{21}N_3O,C_6H_8O_7$.

Impurities



A. 1-methylpiperazine,



B. 1,4-dimethylpiperazine.

Reference substance:

Diethylcarbamazine dihydrogen citrate RS

Established ICRS. Changes in the intended uses to be verified.

Infrared reference spectrum:

Reference spectrum of diethylcarbamazine dihydrogen citrate to be recorded.

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