Folic acid & Ferrous ascorbate Tablets

Analytical Profile No.: FFT 074/075/ AP022

Folic acid & Ferrous ascorbate Tablets contains not less than 90% of the stated amount of folic acid and not less than 90% and not more than 110% of the stated amount of elemental iron.

1. Identification:

1.1. Folic acid

The retention time of the standard and sample preparation of assay preparation should resemble each other.

1.2 Ferrous Ascorbate

Dissolve a quantity of the powdered tablets containing 10 mg of Iron in 2 ml of water, add 1 ml of potassium ferricyanide solution; a blue precipitate is formed that does not dissolve on addition of 5 ml of dilute hydrochloric acid.

2. Assay:

2.1 Folic acid

Determine by liquid chromatography, as described in the Uniformity of Content.

2.2 Ferrous ascorbate equivalent to elemental iron: Determine by UV Spectroscopy

2.2.1 Test Solution

Weigh 20 tablets individually and crush the tablet in the fine powder. Weigh powder eq. to 100 mg of elemental iron and transfer into 100 ml volumetric flask. Add about10 ml of dilute sulphuric acid and heat in water bath until dissolves. Cool and make up the volume to 100 ml with water. Filter the solution and dilute 2 ml of the resulting solution to 50 ml with water.

2.2.2 Reference Solution

Weigh accurately about 100 mg of working standard of ferrous ascorbate and transfer in 100 ml of volumetric flask. Dissolve in 10 ml of dilute sulphuric acid and heat in water bath until

it dissolves. Cool and make up the volume to 100 ml with water. Dilute 5 ml of the filtrate to 50 ml with water.

2.2.3 Procedure:

Pipette 5 ml of the test and reference solution, add 2 ml of 0.1 % w/v sodium acetate solution, 4 ml of 0.25 % w/v hydroquinone solution and add 4 ml of 0.25 % w/v of 1, 10 phenanthroline, allow to stand for 1 hour. Prepare blank in the similar manner except sample solution. Make up the volume to 50 ml with water. Measure the absorbance of the solution at 515 nm. Calculate the content of elemental iron per tablet.

3. Uniformity of content (Folic acid): Determine by liquid chromatography

3.1 Solvent mixture: Prepare a mixture of 80 volumes of 0.57 % w/v solution of dipotassium hydrogen orthophosphate and 13.5 volumes of methanol.

3.2 Test Solution:

Weigh individually 10 tablets. Transfer one tablet individually into ten 100 ml amber volumetric flask. Disperse the tablet with solvent mixture, add about 70 ml of solvent mixture, sonicate for about 15 minutes and make up the volume to 100 ml with solvent mixture. Centrifuge the resulting solution and dilute 2 ml of the solution to 20 ml with solvent mixture. Filter the final solution through 0.2 µm membrane filter.

3.3 Reference Solution:

Weigh accurately about 25 mg of working standard of folic acid and transfer into 100 ml amber volumetric flask. Dissolve with solvent mixture and make up the volume to 100 ml with solvent mixture. Dilute 5 ml of the resulting solution to 50 ml with solvent mixture. Again dilute 5 ml of the resulting solution to 100 ml with solvent mixture. Filter the final solution through 0.2 µm membrane filter.

3.4 Chromatographic system

- **Column:** C18, (250*4.6 mm), 5 µm
- **Flow rate:** 1.0 ml/min

- **Wavelength:** 277 nm
- **Injection volume:** 50 µl
- **Column Temperature:** Ambient
- **Detector:** UV Detector

Buffer: Solution containing 0.938 % w/v of sodium perchlorate and 0.0075 % w/v of potassium dihydrogen orthophosphate.

Mobile Phase: A mixture of 135 volumes of methanol and 800 volumes of buffer and pH adjusted to 7.2 and adjust the volume to 1000 ml with HPLC grade water. Filter through 0.45 µm membrane filter.

3.5 Procedure:

Inject 50µl of standard solution of folic acid as per above mentioned chromatographic condition. In the chromatogram obtained from the standard preparation, the column efficiency determined from the major peak should not be less than 2000 theoretical plates, the tailing factor should be not more than 2.0 and the relative standard deviation of replicate injections should not more be than 2.0 %. Inject 20µl of the sample preparation and chromatograph as per above mentioned chromatographic condition. Calculate the content of folic acid in the tablet.

3.6 Limit: 85-115% of the average content.

4. **Other tests:** As per pharmacopoeial requirement