

Garlic Delayed-Release Tablets

Garlic Delayed-Release Tablets are prepared from Powdered Garlic or Powdered Garlic Extract and contain not less than 90.0 percent and not more than 140.0 percent of the labeled amount of alliin ($C_6H_{11}NO_3S$) and not less than 90.0 percent and not more than 140.0 percent of the labeled amount of potential allicin ($C_6H_{10}OS_2$).

Packaging and storage— Preserve in tight containers.

Labeling— The label states the Latin binomial and, following the official name, the article from which the Tablets were prepared. Label it to indicate the amount of total alliin, in μg per Tablet, and the amount of potential allicin, in μg per Tablet.

Identification—

A: Transfer an amount of pulverized Tablets, equivalent to about 30 mg of alliin, to a 100-mL volumetric flask. Add 70 mL of a mixture of methanol and water (1:1), shake, and centrifuge. Concentrate to a small volume (about 5 mL), using a rotary evaporator. Continue as directed in Identification test A under [Garlic](#).

B: The retention times of the alliin diastereomer peaks in the chromatogram of the Test solution correspond to those in the chromatogram of the Standard solution, as obtained in the test for Content of alliin.

Allicin release— Proceed as directed for Method A in Apparatus 1 and Apparatus 2, Delayed-Release Dosage Forms under Dissolution <711>. Place a number of Tablets, equivalent to about 5 mg of potential allicin, in each vessel.

Apparatus 2: 100 rpm.

Time: 60 minutes for the Buffer stage.

Mobile phase, Crude alliinase solution, Blank solution, and Chromatographic system— Proceed as directed in the test for Content of potential allicin.

Standard solution— Dissolve an accurately weighed quantity of USP Alliin RS in water, and dilute quantitatively, and stepwise if necessary, with water to obtain a solution having a known concentration of about 50 μg per mL. Transfer 1.0 mL of this solution to a 5-mL volumetric flask containing 100 μL of Crude alliinase solution, mix, and allow to stand for 5 minutes at room temperature. Dilute with water to volume, and pass through a membrane filter having a 0.45- μm or finer porosity.

Test solution— Transfer 1.0 mL of the solution under test to a test tube containing 50 µL of 0.21 M carboxymethylamine hemihydrochloride. [note—The solution must be transferred immediately upon removal from the dissolution vessel in order to inhibit the alliinase enzyme.]

Procedure— [note—Do not perform the allicin determination in the Acid stage.] Determine the amount of allicin released by injecting equal volumes (about 100 µL) of the Standard solution and the Test solution into the chromatograph, record the chromatograms, and measure the responses for the allicin peaks. Calculate the amount, in µg, of allicin released in the Buffer stage by the formula:

$$1050C(162.26/354.42)(r_U / r_S)$$

in which C is the concentration, in µg per mL, of USP Alliin RS in the Standard solution; 162.26 is the molecular weight of allicin; 354.42 is twice the molecular weight of alliin; and r_U and r_S are the peak responses for allicin obtained from the Test solution and the Standard solution, respectively. [note—Q is the percentage of the labeled amount of potential allicin released only in the Buffer stage.]

[Weight variation <2091>](#): meet the requirements.

Content of alliin—

0.045 M Phosphate buffer, 0.05 M Phosphate buffer, 0.01 M Carboxymethylamine hemihydrochloride solution, Derivatization reagent, Mobile phase, Standard solution, and

Chromatographic system— Proceed as directed in the test for Content of alliin under [Garlic](#).

Test solution— Pulverize an accurately counted number of Tablets, equivalent to about 50 mg of alliin, with a mortar and pestle. Transfer an accurately weighed amount of the powder, equivalent to 5 mg of alliin, to a 100-mL volumetric flask, add about 70 mL of 0.01 M Carboxymethylamine hemihydrochloride solution, and shake for 1 minute. Dilute with 0.01 M Carboxymethylamine hemihydrochloride solution to volume, and mix. Using a volumetric syringe, transfer 0.1 mL of this solution to a septum-capped vial, add 0.5 mL of the Derivatization reagent, and mix. Allow a reaction time of not less than 2 minutes before injection into the chromatograph.

Procedure— Proceed as directed in the test for Content of alliin under [Garlic](#). Calculate the quantity, in µg, of alliin in the portion of Tablets taken by the formula:

$$600C/(r_U / r_S)$$

in which C is the concentration, in μg per mL, of USP Alliin RS in the Standard solution; and rU and rs are the sums of the peak responses for the alliin diastereomers obtained from the Test solution and the Standard solution, respectively.

Content of potential allicin—

0.01 M Carboxymethylamine hemihydrochloride solution— Prepare as directed in the test for Content of alliin under [Garlic](#).

Mobile phase— Prepare a filtered and degassed mixture of methanol and water (60:40). Make adjustments if necessary.

Crude alliinase solution— Homogenize about 5 g of raw garlic cloves with 25 mL of water. Filter, and extract three times with 50 mL of tert-butyl methyl ether. Discard the organic phase, and remove the residual solvent from the aqueous phase by rotary evaporation in vacuum for 5 minutes. Filter, and store frozen in small vials. [note—This solution is stable for 6 months when stored as directed.] Thaw at room temperature just before use.

Blank solution— Dilute 100 μL of Crude alliinase solution with water to 1 mL.

Standard solution— Dissolve an accurately weighed quantity of USP Alliin RS in water, and dilute quantitatively, and stepwise if necessary, with water to obtain a solution having a known concentration of about 50 μg per mL. Transfer 1.0 mL of this solution to a 5-mL volumetric flask containing 100 μL of Crude alliinase solution, mix, and allow to stand for 5 minutes at room temperature. Dilute with water to volume, and pass through a filter having a 0.45- μm or finer porosity.

Test solution— Weigh and finely powder not fewer than 20 Tablets. Transfer an accurately weighed quantity of the powder, equivalent to about 5 mg of potential allicin, to a 200-mL volumetric flask, add 25 mL of water, and mix. Incubate at room temperature for exactly 30 minutes. Stop the enzymatic reaction by diluting with 0.01 M Carboxymethylamine hemihydrochloride solution to volume, and mix. Centrifuge a portion of this solution, transfer 1.0 mL of the supernatant to a 5-mL volumetric flask, dilute with water to volume, and mix.

Chromatographic system— The liquid chromatograph is equipped with a 240-nm detector and a 4.6-mm \times 25-cm column that contains packing L1. The flow rate is about 1.0 mL per minute. Chromatograph the Standard solution and the Blank solution, and record the peak responses as directed for Procedure: the allicin peak is identified by comparing the chromatograms of the Blank

solution and the Standard solution; and the relative standard deviation for replicate injections is not more than 2.0%. Chromatograph the Test solution, and record the peak responses as directed for Procedure: the resolution, R, between the allicin peak and the preceding peak at a relative retention time of 0.80 (allyl methyl thiosulfonates) is not less than 2.0.

Procedure— Inject equal volumes (about 100 µL) of the Standard solution, the Blank solution, and the Test solution into the chromatograph, record the chromatograms, and measure the peak responses for allicin. Calculate the amount of potential allicin, in µg, in the portion of Tablets taken by the formula:

$$1000C(162.26/354.42)(r_U / r_S)$$

in which C is the concentration, in µg per mL, of USP Alliin RS in the Standard solution; 162.26 is the molecular weight of allicin; 354.42 is twice the molecular weight of alliin; and r_U and r_S are the responses for the allicin peaks, corrected by the response of the blank, obtained from the Test solution and the Standard solution, respectively.

Alliinase activity—

0.045 M Phosphate buffer, 0.05 M Phosphate buffer, 0.01 M Carboxymethoxylamine hemihydrochloride solution, Derivatization reagent, Mobile phase, Standard solution, and

Chromatographic system— Proceed as directed in the test for Content of alliin under [Garlic](#).

Test solution— Weigh and finely powder not fewer than 20 Tablets. Transfer an accurately weighed quantity of the powder, equivalent to about 5 mg of alliin, to a 100-mL volumetric flask, add 25 mL of water, and mix. Incubate at room temperature for exactly 5 minutes. Stop the enzymatic reaction by diluting with 0.01 M Carboxymethoxylamine hemihydrochloride solution to volume, and mix. Centrifuge a portion of this solution, and, using a volumetric syringe, transfer 0.1 mL of the supernatant to a septum-capped vial. Add 0.5 mL of the Derivatization reagent, and mix. Allow a reaction time of not less than 2 minutes before injection into the chromatograph.

Procedure— Proceed as directed in the test for Content of alliin under [Garlic](#). The area of the alliin peak obtained from the Test solution is not more than 1% of the area of the alliin peak obtained from the Standard solution.