

Temozolomide Capsules

To view the Notice from the Expert Committee that posted in conjunction with this accelerated revision, please click www.uspnf.com/rb-temozolomide-caps-20220826.

DEFINITION
Temozolomide Capsules contain NLT 90.0% and NMT 110.0% of the labeled amount of temozolomide (C₆H₆N₆O₂).
[CAUTION—Temozolomide is cytotoxic. Great care should be taken to prevent inhaling particles of Temozolomide and exposure to the skin.]

IDENTIFICATION
• **A. [THIN-LAYER CHROMATOGRAPHIC IDENTIFICATION TEST \(201\)](#)**
Standard solution: 1 mg/mL of [USP Temozolomide RS](#) in [acetonitrile](#)
Sample solution: Transfer the contents of Capsules indicated in [Table 1](#) to a glass beaker containing a stir bar. Add [acetonitrile](#) according to [Table 1](#). Stir for NLT 20 min. Sonicate for 10 min followed by a 30-s stirring to disperse any remaining solids. Allow the contents of the flask to settle.

Table 1

Capsule Strength (mg/Capsule)	Acetonitrile (mL)	Number of Capsules
5	50	4
20	50	1
100	100	1
140	140	1
180	180	1
250	250	1

Chromatographic system
(See [Chromatography \(621\), General Procedures, Thin-Layer Chromatography](#).)

Mode: TLC
Adsorbent: 20-cm × 20-cm or 10-cm × 20-cm of 0.25-mm layer of [chromatographic silica gel mixture](#)
Application volumes
Standard solution: 10 µL
Sample solution: See [Table 2](#).

Table 2

Capsule Strength (mg/Capsule)	Sample Solution (µL)
5	25
20	25

Capsule Strength (mg/Capsule)	Sample Solution (μ L)
100	10
140	10
180	10
250	10

Developing solvent system: [Acetone](#) and [toluene](#) (90:15)

Analysis: Spot the *Standard solution* and *Sample solution* approximately 2.5 cm from the bottom of the plate. Evaporate the solvent using a current of air. Place the plate in the developing chamber and allow the *Developing solvent system* to rise to approximately 75% of the plate height. Remove the plate from the developing chamber, mark the position of the solvent front, and allow the plate to dry in the fume hood. Observe the plate under 254 nm of UV light.

Acceptance criteria: The R_f value from the *Sample solution* corresponds to that from the *Standard solution*.

- **B.** The retention time of the major peak of the *Sample solution* corresponds to that of the *Standard solution*, as obtained in the Assay.

ASSAY

• PROCEDURE

Store the solutions containing temozolomide at 4°.

Solution A: 0.5% (v/v) [glacial acetic acid](#) in [water](#)

Mobile phase: *Solution A* and [methanol](#) (96:4), containing 0.94 g/L of [sodium 1-hexanesulfonate](#) (0.005 M)

System suitability stock solution: 0.4 mg/mL of [USP Temozolomide RS](#) in *Mobile phase*

System suitability solution: Transfer 25 mL of [0.1 N hydrochloric acid](#) and 25 mL of *System suitability stock solution* into a container. Heat the container at 80° for 4 h. [NOTE—The preparation forms 2-azahypoxanthine, temozolomide acid, and aminoimidazolecarboxamide.]

Standard solution: 0.1 mg/mL of [USP Temozolomide RS](#) in *Mobile phase* from *System suitability stock solution*

Sample stock solution: Place 10 Capsules in a suitable volumetric flask according to [Table 3](#). Add 80% of the final volume of *Mobile phase* with agitation or stirring for NLT 1 h. Ensure that the Capsules are broken apart into small pieces and that contents are adequately dissolved. Dilute with *Mobile phase* to volume and mix well. Allow to sit for 30 min and mix again. Centrifuge 25 mL of this solution for 10 min or until the solution is clear. Pass the supernatant through a suitable filter of 0.45- μ m pore size and discard the first few milliliters.

Table 3

Capsule Strength (mg/Capsule)	Volumetric Flask Size (mL)	Nominal Concentration (mg/mL)
5	500	0.1
20	200	1.0
100	1000	1.0
140	2000	0.7
180	2000	0.9
250	2000	1.25

Sample solution: Nominally equivalent to 0.1 mg/mL of temozolomide in *Mobile phase* from *Sample stock solution*

Chromatographic system

(See [Chromatography \(621\)](#), [System Suitability](#).)

Mode: LC

Detector: UV 270 nm

Column: 4.6-mm $\times$ 15-cm; 5- μ m packing [L1](#)

Autosampler temperature: 4°

Flow rate: 1 mL/min

Injection volume: 20 µL

System suitability

Samples: *System suitability solution* and *Standard solution*

Suitability requirements

Tailing factor: NMT 1.9 for the temozolomide peak, *System suitability solution*

Relative standard deviation: NMT 1.0%, *Standard solution*

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of the labeled amount of temozolomide ($C_6H_6N_6O_2$) in the portion of Capsules taken:

$$\text{Result} = (r_U/r_S) \times (C_S/C_U) \times 100$$

r_U = peak response of temozolomide from the *Sample solution*

r_S = peak response of temozolomide from the *Standard solution*

C_S = concentration of [USP Temozolomide RS](#) in the *Standard solution* (mg/mL)

C_U = nominal concentration of temozolomide in the *Sample solution* (mg/mL)

Acceptance criteria: 90.0%–110.0%

PERFORMANCE TESTS

Change to read:

- [DISSOLUTION \(711\)](#).

▲Test 1▲ (RB 1-Sep-2022)

Medium: [Water](#); 500 mL (for 5 mg/Capsule), 900 mL (for all other strengths)

Apparatus 1: 100 rpm

Time: 30 min

Determine the percentage of the labeled amount of temozolomide ($C_6H_6N_6O_2$) dissolved by using one of the following procedures.

Spectrophotometric procedure

Standard solution: Prepare a solution of [USP Temozolomide RS](#) in *Medium* according to [Table 4](#).

Table 4

Capsule Strength (mg/Capsule)	Nominal Concentration (mg/mL)
5	0.01
20	0.022
100	0.11
140	0.16
180	0.20
250	0.28

Sample solution: Pass a portion of the solution under test through a suitable filter of 0.8-µm pore size. Discard the first few milliliters of the filtrate and use the remaining filtrate for analysis.

Instrumental conditions

(See [Ultraviolet-Visible Spectroscopy \(857\)](#).)

Mode: UV

Analytical wavelength: Maximum absorbance at about 328 nm