Status: Currently Official on 17-Feb-2025
Official Date: Official as of 01-May-2024
Document Type: USP Monographs
DocId: GUID-01C7EB67-898E-488B-A031-8E50B55F1AFA_3_en-US
DOI: https://doi.org/10.31003/USPNF_M88620_03_01
DOI Ref: u3mt9

Vitamin E Capsules

Change to read:

DEFINITION

Vitamin E Capsules contain Vitamin E or Vitamin E Preparation, where Vitamin E is a form of alpha tocopherol ($C_{29}H_{50}O_2$) that includes *RRR*- or *all-rac*-alpha tocopherol ($C_{29}H_{50}O_2$), *RRR*- or *all-rac*-alpha tocopheryl acetate ($C_{31}H_{52}O_3$), and *RRR*- or *all-rac*-alpha tocopheryl acid succinate ($C_{33}H_{54}O_5$) and where Vitamin E Preparation is a combination of a single form of Vitamin E with one or more inert substances. Vitamin E Capsules contain NLT 95.0% and NMT 120.0% of the labeled amount of vitamin E ▲as 2*R*-alpha tocopherol ($C_{29}H_{50}O_2$).▲ (USP 1-May-2024)

IDENTIFICATION

• **A.**

[Note—Use low-actinic glassware.]

Sample solutions

Alpha tocopherol: Nominally 1 mg/mL in dehydrated alcohol

Alpha tocopheryl acetate: Transfer 220 mg of *RRR*- or *all-rac*-alpha tocopheryl acetate from the Capsule contents to a round-bottom, glass-stoppered, 150-mL flask, and dissolve in 25 mL of dehydrated alcohol. Add 20 mL of diluted sulfuric acid in alcohol (1 in 7) and reflux in an all-glass apparatus for 3 h, protected from sunlight. Cool, transfer to a 200-mL volumetric flask, and add diluted sulfuric acid in alcohol (1 in 72) to volume.

Alpha tocopheryl acid succinate: [Caution—Wear safety goggles.] Transfer an amount of Capsule contents, equivalent to 200 mg of alpha tocopherol, to a round-bottom, glass-stoppered, 250-mL flask; dissolve in 50 mL of dehydrated alcohol; and reflux for 1 min. While the solution is boiling, add, through the condenser, 1 g of potassium hydroxide pellets one at a time to avoid overheating.

Continue refluxing for 20 min and, without cooling, add 2 mL of hydrochloric acid dropwise through the condenser. [Note—This technique is essential to prevent oxidative action by air while the sample is in an alkaline medium.]

Cool, and transfer the contents of the flask to a 500-mL separatory funnel, rinsing the flask with 100 mL each of water and ether and adding the rinsings to the separator. Shake vigorously, allow the layers to separate, and collect each of the two layers in individual separatory funnels. Extract the aqueous layer with two 50-mL portions of ether and add these extracts to the main ether extract. Wash the combined ether extracts with four 100-mL portions of water, then evaporate the ether solution on a water bath under reduced pressure or in an atmosphere of nitrogen until about 7–8 mL remain. Complete the evaporation, removing the last traces of ether without the application of heat. Immediately dissolve the residue in diluted sulfuric acid in alcohol (1 in 72), transfer to a 200-mL volumetric flask, and dilute with diluted sulfuric acid in alcohol (1 in 72) to volume.

Analysis

Sample: Use the appropriate *Sample solution*.

To 10 mL of the *Sample solution* add 2 mL of nitric acid, with swirling, and heat at about 75° for 15 min.

Acceptance criteria: A bright red or orange color develops.

Change to read:

• **B.** ▲Optical Rotation ⟨781S⟩, Procedures, Specific Rotation▲ (USP 1-May-2024)

Sample solutions

Alpha tocopherol: Dissolve an amount of the sample equivalent to 100 mg of alpha tocopherol in 50 mL of ether.

Alpha tocopheryl acetate: Transfer a volume of the *Sample solution* for *Alpha tocopheryl acetate* from *Identification A*, equivalent to 100 mg of the test article, to a separatory funnel, and add 200 mL of water. Extract with ether, first with 75 mL, then with 25 mL, and combine the ether extracts in another separatory funnel.

Alpha tocopheryl acid succinate: Transfer a volume of the *Sample solution* for *Alpha tocopheryl acid succinate* from *Identification A*, equivalent to 100 mg of the test article, to a separatory funnel, and add 200 mL of water. Extract with ether, first with 75 mL, then with 25 mL, and combine the ether extracts in another separatory funnel.

Analysis

Sample: Use the appropriate *Sample solution*.

To the entire volume of a *Sample solution* add 20 mL of a solution (1 in 10) of potassium ferricyanide in sodium hydroxide solution (1 in 125) and shake for 3 min. Wash the ether solution with four 50-mL portions of water, discard the washings, and dry over anhydrous sodium sulfate. Evaporate the dried ether solution on a water bath under reduced pressure or in an atmosphere of nitrogen until 7–8 mL remain, then complete the evaporation, removing the last traces of ether without the application of heat. Immediately dissolve the residue in 5.0 mL of 2,2,4-trimethylpentane, ▲transfer into a sample cell, and record the observed rotation in degrees (°). For *RRR*-isomers, calculate the specific rotation using *c* as the concentration of alpha tocopherol determined in the appropriate *Assay*.▲ (USP 1-May-2024)

Acceptance criteria

For Capsules labeled to contain *RRR*-isomers: NLT +24°

For Capsules labeled to contain *all-rac* forms: -0.01° to +0.01°

Change to read:

• **C.** The retention time of the major peak ▲▲ (USP 1-May-2024) of the *Sample solution* corresponds to that of the *Standard solution*, as obtained in the *Assay*.

ASSAY

Change to read:

• **Alpha Tocopherol**

[Note—Use low-actinic glassware.]

Internal standard solution: 10 mg/mL of squalane in cyclohexane

System suitability solution: 0.1 mg/mL each of USP Alpha Tocopherol RS and USP Alpha Tocopheryl Acetate RS in cyclohexane

Standard solution: 10 mg/mL of USP Alpha Tocopherol RS in *Internal standard solution*

Sample solution: Weigh NLT 10 Capsules in a tared weighing bottle. With a sharp knife or by other appropriate means, carefully open the Capsules, without loss of the shell material, and transfer the combined Capsule contents to a 100-mL beaker. Remove any adhering substance from the emptied Capsules by washing with several small portions of *n*-hexane. Discard the washings and allow the empty Capsules to dry in a current of dry air until the odor of *n*-hexane is no longer perceptible. Weigh the empty Capsules in the original tared weighing bottle and calculate the average net weight/Capsule. Dissolve a portion of the combined Capsule contents in the *Internal standard solution* to prepare a vitamin E (*RRR*- or *all-rac*-alpha tocopherol) solution with a nominal concentration of 10 mg/mL.

Chromatographic system

(See *Chromatography ⟨621⟩, System Suitability*.)

Mode: GC

Detector: Flame ionization

Column: 0.25-mm × 30-m fused silica capillary; bonded with a 0.25-µm film of phase G2

Temperatures

Injection port: 290°

Column: 280°

Detector: 290°

Carrier gas: Helium

Flow rate: 1 mL/min

Injection type: Split, split ratio 100:1

Injection volume: 1 µL

System suitability

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

Suitability requirements

Resolution: NLT 3.5 between alpha tocopherol and alpha tocopheryl acetate, *System suitability solution*

Relative standard deviation: NMT 2.0% for peak response ratios of alpha tocopherol to the internal standard from replicate injections, *Standard solution*

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of the labeled amount of vitamin E ▲as 2*R*-alpha tocopherol ($C_{29}H_{50}O_2$)▲ (USP 1-May-2024) in the portion of Capsule contents taken:

$$\text{Result} = (R_U/R_S) \times (C_S/C_U) \text{ ▲} \times F \text{▲ (USP 1-May-2024)} \times 100$$

R_U = peak response ratio of alpha tocopherol to the internal standard from the *Sample solution*

R_S = peak response ratio of alpha tocopherol to the internal standard from the *Standard solution*

C_S = concentration of USP Alpha Tocopherol RS in the *Standard solution* (mg/mL)

C_U = nominal concentration of vitamin E as *RRR*- or *all-rac*-alpha tocopherol in the *Sample solution* (mg/mL)

▲F = conversion factor for the content of alpha tocopherol to 2*R*-alpha tocopherol equivalent: 1/2 for products labeled to contain *all-rac* alpha tocopherol or 1 for products labeled to contain *RRR*-alpha tocopherol▲ (USP 1-May-2024)

Acceptance criteria: 95.0%–120.0% of the labeled amount of vitamin E as ▲2*R*-alpha tocopherol▲ (USP 1-May-2024)

Change to read:

• **Alpha Tocopheryl Acetate:** Proceed as directed in the *Assay* for *Alpha Tocopherol* except as follows. For the *Standard solution* and *Sample solution*, substitute alpha tocopheryl acetate for alpha tocopherol, and substitute USP Alpha Tocopheryl Acetate RS for USP Alpha Tocopherol RS.

▲**Analysis**

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of the labeled amount of vitamin E as 2*R*-alpha tocopherol ($C_{29}H_{50}O_2$) in the portion of Capsule contents taken:

$$\text{Result} = (R_U/R_S) \times (C_S/C_U) \times F \times 100$$

R_U = peak response ratio of alpha tocopheryl acetate to the internal standard from the *Sample solution*

R_S = peak response ratio of alpha tocopheryl acetate to the internal standard from the *Standard solution*

C_S = concentration of alpha tocopherol from USP Alpha Tocopheryl Acetate RS in the *Standard solution* (mg/mL)

C_U = nominal concentration of vitamin E as *RRR*- or *all-rac*-alpha tocopherol in the *Sample solution* (mg/mL)

F = conversion factor for the content of alpha tocopherol to 2*R*-alpha tocopherol equivalent: 1/2 for products labeled to contain *all-rac* alpha tocopherol or 1 for products labeled to contain *RRR*-alpha tocopherol

▲ (USP 1-May-2024)

Acceptance criteria: 95.0%–120.0% of the labeled amount of vitamin E as ▲2*R*-alpha tocopherol▲ (USP 1-May-2024)

Change to read:

• **Alpha Tocopheryl Acid Succinate**

Internal standard solution, System suitability solution, Chromatographic system, ▲and▲ (USP 1-May-2024) **System suitability**▲▲ (USP 1-May-2024) **:** Proceed as directed in the *Assay* for *Alpha Tocopherol*.

Standard solution: Transfer 30.0 mg of USP Alpha Tocopheryl Acid Succinate RS into a 20-mL vial. Add 2.0 mL of methanol, 1.0 mL of 2,2-dimethoxypropane, and 0.1 mL of hydrochloric acid to the vial. Cap tightly and sonicate. Allow to stand in the dark for 1 h ± 5 min. Remove from the dark, uncap, and evaporate just to dryness on a steam bath with the aid of a stream of nitrogen. Add 3.0 mL of the *Internal standard solution* and mix on a vortex mixer to dissolve.

Sample solution: Weigh NLT 10 Capsules in a tared weighing bottle. With a sharp knife or by other appropriate means, carefully open the Capsules, without loss of the shell material, and transfer the combined Capsule contents to a 100-mL beaker. Remove any adhering substance from the emptied Capsules by washing with several small portions of *n*-hexane. Discard the washings and allow the empty Capsules to dry in a current of dry air until the odor of *n*-hexane is no longer perceptible. Weigh the empty Capsules in the original tared weighing bottle and calculate the average net weight/Capsule. Transfer a portion of the combined Capsule contents, equivalent to 30.0 mg of vitamin E (*RRR*- or *all-rac*-alpha tocopheryl acid succinate), into a 20-mL vial. Add 2.0 mL of methanol, 1.0 mL of 2,2-dimethoxypropane, and 0.1 mL of hydrochloric acid to the vial. Cap tightly and sonicate. Allow to stand in the dark for 1 h ± 5 min. Remove from the dark, uncap, and evaporate just to dryness on a steam bath with the aid of a stream of nitrogen. Add 3.0 mL of the *Internal standard solution* and mix on a vortex mixer to dissolve.

▲**Analysis**

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of the labeled amount of vitamin E as 2*R*-alpha tocopherol ($C_{29}H_{50}O_2$) in the portion of Capsule contents taken:

$$\text{Result} = (R_U/R_S) \times (C_S/C_U) \times F \times 100$$

R_U = peak response ratio of alpha tocopheryl acid succinate to the internal standard from the *Sample solution*

R_S = peak response ratio of alpha tocopheryl acid succinate to the internal standard from the *Standard solution*

C_S = concentration of alpha tocopherol from USP Alpha Tocopheryl Acid Succinate RS in the *Standard solution* (mg/mL)

C_U = nominal concentration of vitamin E as *RRR*- or *all-rac*-alpha tocopherol in the *Sample solution* (mg/mL)

F = conversion factor for the content of alpha tocopherol to 2*R*-alpha tocopherol equivalent: 1/2 for products labeled to contain *all-rac* alpha tocopherol or 1 for products labeled to contain *RRR*-alpha tocopherol

▲ (USP 1-May-2024)

Acceptance criteria: 95.0%–120.0% of the labeled amount of vitamin E as ▲2*R*-alpha tocopherol▲ (USP 1-May-2024)

PERFORMANCE TESTS

• **Disintegration ⟨701⟩**

Buffer solution: Dissolve 2.99 g of sodium acetate and 1.66 mL of glacial acetic acid in 1000 mL of water to obtain a pH of 4.50 ± 0.05.

Medium: *Buffer solution*

Time: 45 min

Acceptance criteria: Meet the requirements

• **Uniformity of Dosage Units ⟨905⟩:** Meet the requirements

ADDITIONAL REQUIREMENTS

• **Packaging and Storage:** Preserve in tight containers and store at room temperature. Protect Capsules containing *RRR*- or *all-rac*-alpha tocopherol from light.

Change to read:

• **Labeling:** Label to indicate the chemical form of vitamin E present and to indicate whether the *RRR* or *all-rac* form is present, excluding any different forms that may be introduced as a minor constituent of the vehicle. ▲Express content of vitamin E in mg of alpha tocopherol/Capsule.[1]▲ (USP 1-May-2024)

• **USP Reference Standards ⟨11⟩**

USP Alpha Tocopherol RS

USP Alpha Tocopheryl Acetate RS

USP Alpha Tocopheryl Acid Succinate RS

[1] 1 mg of vitamin E (alpha tocopherol) = 1 mg of *RRR*-alpha tocopherol = 2 mg of *all-rac*-alpha tocopherol; 1 mg of *RRR*-alpha tocopheryl acetate = 0.91 mg of alpha tocopherol equivalent; 1 mg of *RRR*-alpha tocopheryl acid succinate = 0.81 mg of alpha tocopherol equivalent. To convert IU to mg: 1 IU of *RRR*-alpha tocopherol = 0.67 mg of alpha tocopherol; 1 IU of *all-rac*-alpha tocopherol = 0.45 mg of alpha tocopherol.

Auxiliary Information - Please check for your question in the FAQs before contacting USP.

Topic/Question	Contact	Expert Committee
VITAMIN E CAPSULES	Natalia Davydova Scientific Liaison	NBDS2020 Non-botanical Dietary Supplements
REFERENCE STANDARD SUPPORT	RS Technical Services RSTECH@usp.org	NBDS2020 Non-botanical Dietary Supplements

Chromatographic Database Information: Chromatographic Database

Most Recently Appeared In:
Pharmacopeial Forum: Volume No. 48(5)

Current DocID: GUID-01C7EB67-898E-488B-A031-8E50B55F1AFA_3_en-US

DOI: https://doi.org/10.31003/USPNF_M88620_03_01

DOI ref: u3mt9