Calculate the percentage content of alpha-lactose:

$$\text{Result} = S_a/(S_a + S_b) \times 100$$

S_a = area of the peak due to alpha-lactose
S_b = area of the peak due to beta-lactose

Calculate the percentage content of beta-lactose:

$$\text{Result} = S_b/(S_a + S_b) \times 100$$

S_a = area of the peak due to alpha-lactose
S_b = area of the peak due to beta-lactose

✦■2S (*NF30*)

IMPURITIES

- ✦**HEAVY METALS,** *Method II* ⟨**231**⟩**:** NMT 5 ppm✦

Change to read:

- **RESIDUE ON IGNITION** ⟨**281**⟩**:** NMT 0.1% ■■2S (*NF30*)

SPECIFIC TESTS

Change to read:

- **CLARITY AND COLOR OF SOLUTION**

  ■**Hydrazine sulfate solution:** Dissolve 1.0 g of hydrazine sulfate in water, and dilute to 100.0 mL. Allow to stand for 4–6 h.

  Hexamethylenetetramine solution: In a 100-mL ground-glass stoppered flask dissolve 2.5 g of hexamethylenetetramine in 25.0 mL of water.

  Primary opalescent suspension: To the *Hexamethylenetetramine solution* in the flask add 25.0 mL of the *Hydrazine sulfate solution*. Mix and allow to stand for 24 h. This suspension is stable for 2 months, provided it is stored in a glass container free from surface defects. The suspension must not adhere to the glass and must be well mixed before use.

  Standard opalescence: Dilute 15.0 mL of the *Primary opalescent suspension* to 1000.0 mL with water. This suspension is freshly prepared and may be stored for up to 24 h.

  Reference suspension: To 5.0 mL of the *Standard opalescence* add 95.0 mL of water. Mix and shake before use.

  Reference solution: To 6.0 mL of ferric chloride CS, 2.5 mL of cobaltous chloride CS, and 1.0 mL of cupric sulfate CS add hydrochloric acid (10 g/L HCl) to make 1000 mL.

  Sample solution: 1 g in 10 mL of boiling water. Allow to cool.

  Instrumental conditions

  Mode: Vis

  Analytical wavelength: 400 nm

  Acceptance criteria: NMT 0.04 for the absorbance divided by the path length in centimeters; and the clarity of the *Sample solution* is the same as that of water or its opalescence is not more pronounced than that of the *Reference suspension*, and it is not more colored than the *Reference solution*.■2S (*NF30*)

- **LOSS ON DRYING** ⟨**731**⟩

  Analysis: Dry a sample at 80° for 2 h.

  Acceptance criteria: NMT 0.5%

- **WATER DETERMINATION,** *Method I* ⟨**921**⟩

  Sample solution: Anhydrous Lactose in a mixture of methanol and formamide (2:1)

  Acceptance criteria: NMT 1.0%

- **MICROBIAL ENUMERATION TESTS** ⟨**61**⟩ and **TESTS FOR SPECIFIED MICROORGANISMS** ⟨**62**⟩**:** The total aerobic microbial count is NMT 10^2 cfu/g and ✦the total combined molds and yeasts count is NMT 50 cfu/g✦. It meets the requirements of the test for absence of *Escherichia coli*.

- **PROTEIN AND LIGHT-ABSORBING IMPURITIES**

  (See *Spectrophotometry and Light-Scattering* ⟨851⟩.)

  Sample solution: 1% solution (w/v)

  Instrumental conditions

  Mode: UV

  Wavelength range: 210–300 nm

  Acceptance criteria: NMT 0.25 for the absorbance divided by the path length in centimeters at 210–220 nm; NMT 0.07 for the absorbance divided by the path length in centimeters at 270–300 nm

Change to read:

- **ACIDITY OR ALKALINITY**

  Sample solution: Dissolve 6 g by heating in 25 mL of carbon dioxide-free water, cool, and add 0.3 mL of phenolphthalein TS.

  Acceptance criteria: The solution is colorless, and NMT 0.4 mL of 0.1 N sodium hydroxide is required to produce a ■pink or■2S (*NF30*) red color.

- **OPTICAL ROTATION,** *Specific Rotation* ⟨**781S**⟩

  Sample solution: Dissolve 10 g by heating in 80 mL of water to 50°. Allow to cool, and add 0.2 mL of 6 N ammonium hydroxide. Allow to stand for 30 min, and dilute with water to 100 mL.

  Acceptance criteria: +54.4° to +55.9°, calculated on the anhydrous basis, at 20°

ADDITIONAL REQUIREMENTS

- ✦**PACKAGING AND STORAGE:** Preserve in tight containers.
- **LABELING:** Where the labeling indicates the relative quantities of alpha- and beta-lactose, determine compliance using *Content of Alpha and Beta Anomers*. Where the labeling states the particle size distribution, it also indicates the d_{10}, d_{50}, and d_{90} values and the range for each.
- **USP REFERENCE STANDARDS** ⟨**11**⟩

  USP Dextrose RS
  USP Fructose RS
  USP Anhydrous Lactose RS
  USP Sucrose RS✦

Add the following:

■Polyoxyl Stearate

$R\text{-}CO\text{-}(OCH_2CH_2)_n\text{-}OH$

$R\text{-}CO\text{-}(OCH_2CH_2)_n\text{-}OOC\text{-}R$

$R = CH_3(CH_2)_{16}$ or $CH_3(CH_2)_{14}$

n = 6–100

Polyethylene glycol stearate;
Polyethylene glycol monostearate;
Poly(oxy-1,2-ethanediyl), α-hydro-ω-hydroxyoctadecanoate [9004-99-3].

DEFINITION

Polyoxyl Stearate is a mixture of monoesters and diesters of mainly stearic (octadecanoic) acid and/or palmitic (hexadecanoic) acid and polyethylene glycols. The fatty acids may be of vegetable, animal, or synthetic origin. Polyoxyl Stearate Type I or Type II differs in its content of stearic acid. It may contain free polyethylene glycols. The average polymer length is equivalent to 6–100 ethylene oxide units per molecule (nominal value).

IDENTIFICATION

• **A. Infrared Absorption** ⟨**197A**⟩
Sample: Use an undried specimen.
• **B.** It meets the requirements of the test for *Content of Stearic Acid and Palmitic Acid*.

ASSAY

• **Content of Stearic Acid and Palmitic Acid**
Polyoxyl Stearate exhibits the composition profiles of fatty acids shown in *Table 1* below, as determined in *Fats and Fixed Oils* ⟨401⟩, *Fatty Acid Composition*.

Table 1

	Content of Stearic Acid and Palmitic Acid
Polyoxyl Stearate Type I	Stearic Acid: 40.0%–60.0%; sum of Palmitic and Stearic acids: NLT 90.0%
Polyoxyl Stearate Type II	Stearic Acid: 90.0%–99.0%; sum of Palmitic and Stearic acids: NLT 96.0%

• **Content of Free Polyethylene Glycols**
[Note—This test is for Polyoxyl 40 Stearate only.]
Sample: 6 g of Polyoxyl 40 Stearate
Analysis: Transfer the *Sample* to a 500-mL separator containing 50 mL of ethyl acetate. Dissolve completely, then add 50 mL of sodium chloride solution (29 in 100), shake vigorously for 2 min, and allow to separate for 15 min. If separation is incomplete, carefully insert the separator into the well of a steam bath for short time intervals. Repeat this technique as many times as necessary to ensure the complete separation of the two phases. Cool, and drain the lower, aqueous phase into a second 500-mL separator. Extract the upper layer with a second 50-mL portion of sodium chloride solution (29 in 100). Repeat the separation as before, including the steam bath technique, to facilitate complete separation. To the combined aqueous layers add 50 mL of ethyl acetate, shake vigorously for 2 min, and allow to separate as before. Drain the lower, aqueous phase into a third 500-mL separator, and extract it with two 50-mL portions of chloroform, shaking for 2 min each time. Repeat the steam bath technique to ensure complete separation.
Evaporate the combined chloroform extracts in a 150-mL beaker on a steam bath, with the aid of a stream of nitrogen, to apparent dryness.
Redissolve in about 15 mL of chloroform, and filter, collecting the filtrate in a weighed 150-mL beaker. Record the weight of the empty 150-mL beaker, W_1, in g. Rinse the funnel with several small portions of chloroform, and evaporate the combined filtrate and rinsings, as described above, to remove chloroform or ethyl acetate.
Dry in vacuum at 60° for 1 h. Cool in a desiccator, and weigh. Record the weight, W_2, in g.
Calculate the percentage of free polyethylene glycols in Polyoxyl 40 Stearate taken:

$$\text{Result} = [(W_2 - W_1)/W] \times 100$$

W = weight of Polyoxyl 40 Stearate (g)
Acceptance criteria: 17%–27% of free polyethylene glycols for Polyoxyl 40 Stearate only

IMPURITIES

• **Heavy Metals,** *Method II* ⟨**231**⟩**:** NMT 10 ppm
• **Articles of Botanical Origin,** *Total Ash* ⟨**561**⟩**:** NMT 0.3%, determined on 1.0 g
• **Limit of Ethylene Oxide and Dioxane**
Analysis: Proceed as directed in *Ethylene Oxide and Dioxane* ⟨228⟩, *Method II*.
Acceptance criteria
Ethylene oxide: 1 ppm
Dioxane: 380 ppm

SPECIFIC TESTS

• **Alkalinity**
Phenol red solution: Dissolve 100 mg of phenolsulfonphthalein in a mixture of 2.82 mL of 0.1 M sodium hydroxide and 20 mL of alcohol, and dilute with water to 100 mL.
Sample solution: 2.0 g of Polyoxyl Stearate
Analysis: Dissolve the *Sample* in alcohol and dilute with alcohol to 20 mL. To 2 mL of this solution add 0.05 mL of *Phenol red solution*.
Acceptance criteria: The solution does not turn red.
• **Fats and Fixed Oils,** *Acid Value* ⟨**401**⟩**:** NMT 6.0
• **Fats and Fixed Oils,** *Hydroxyl Value* ⟨**401**⟩**:** Within the ranges specified in *Table 2*
• **Fats and Fixed Oils,** *Iodine Value* ⟨**401**⟩**:** NMT 3.0
• **Fats and Fixed Oils,** *Peroxide Value* ⟨**401**⟩**:** NMT 10.0
• **Fats and Fixed Oils,** *Saponification Value* ⟨**401**⟩**:** Within the ranges specified in *Table 2*
• **Melting Range or Temperature** ⟨**741**⟩
Sample: 10 g
Analysis: Melt the *Sample* at 80°–90°. Introduce a sufficient amount of the *Sample* into the tube by capillary action to form a column of the prescribed height in the tube. Allow to stand at 0° for 2 h.
Acceptance criteria: Within the ranges specified in *Table 2*

Table 2

Ethylene Oxide Units/ Molecule (Nominal Value)	Melting Range or Temperature (°)	Hydroxyl Value	Saponification Value
6	26–37	80–110	90–115
8	26–35	80–105	88–100
32	46–50	20–40	30–45
40	Measure Congealing Temperature	25–40	25–35
75	53–59	15–35	8–25
100	48–60	15–30	5–20

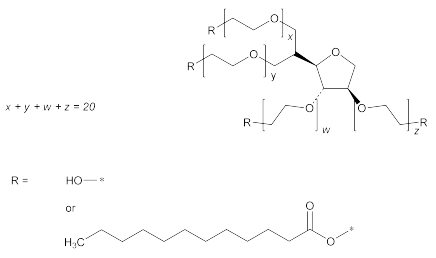
• **Congealing Temperature** ⟨**651**⟩**:** 37°–47° for Polyoxyl 40 Stearate only
• **Water Determination,** *Method I* ⟨**921**⟩**:** NMT 3.0%

ADDITIONAL REQUIREMENTS

• **Packaging and Storage:** Preserve in tight containers, and store at room temperature. Protect from light and moisture.
• **Labeling:** Label it to indicate the number of ethylene oxide units/molecule (nominal value), and the type of Polyoxyl Stearate. Label it to indicate whether the fatty acids are derived from vegetable, animal, or synthetic sources.
• **USP Reference Standards** ⟨**11**⟩
USP Polyoxyl 6 Stearate RS
USP Polyoxyl 8 Stearate RS
USP Polyoxyl 32 Stearate RS
USP Polyoxyl 40 Stearate RS
USP Polyoxyl 75 Stearate RS
USP Polyoxyl 100 Stearate RS■2S (NF30)

Polysorbate 20

Change to read:



■The ratio of the OH group to the ($C_{11}H_{23}COO$) group is mainly 3:1.
Polyethylene glycol 20 sorbitan ether monolaurate;
Polyoxyethylene 20 sorbitan monododecanoate;■2S *(NF30)*
Polyoxyethylene 20 sorbitan monolaurate [9005-64-5].

DEFINITION

Change to read:

Polysorbate 20 is a laurate ester of sorbitol and its anhydrides, copolymerized with approximately 20 moles of ethylene oxide for each mole of sorbitol and sorbitol anhydrides. ■The fatty acids may be of vegetable, animal, or synthetic origin.■2S *(NF30)*

IDENTIFICATION

Change to read:

• **A.** ■**INFRARED ABSORPTION** ⟨**197F**⟩■2S *(NF30)*

Change to read:

• **B.** ■It meets the requirements in the *Assay* for *Composition of Fatty Acids.*■2S *(NF30)*

ASSAY

Add the following:

■• **COMPOSITION OF FATTY ACIDS**
Polysorbate 20 exhibits the composition profiles of fatty acids shown in *Table 1*, as determined in *Fats and Fixed Oils* ⟨401⟩, *Fatty Acid Composition*.

Table 1

Carbon-Chain Length	Number of Double Bonds	Percentage (%)
6	0	≤1.0
8	0	≤10.0
10	0	≤10.0
12	0	40.0–60.0
14	0	14.0–25.0
16	0	7.0–15.0
18	0	≤7.0
18	1	≤11.0
18	2	≤3.0

■2S *(NF30)*

IMPURITIES
• **RESIDUE ON IGNITION** ⟨**281**⟩**:** NMT 0.25%
• **HEAVY METALS,** *Method II* ⟨**231**⟩**:** NMT 10 ppm

Add the following:

■• **LIMIT OF ETHYLENE OXIDE AND DIOXANE,** *Method II* ⟨**228**⟩
Acceptance criteria
Ethylene oxide: NMT 1 ppm
Dioxane: NMT 10 ppm■2S *(NF30)*

SPECIFIC TESTS

Add the following:

■• **BACTERIAL ENDOTOXINS TEST** ⟨**85**⟩
For Polysorbate 20 intended for use in the manufacture of injectable dosage forms: The level of bacterial endotoxins is such that the requirement in the relevant dosage form monograph(s) in which Polysorbate 20 is used can be met. Where the label states that Polysorbate 20 must be subjected to further processing during the preparation of injectable dosage forms, the level of bacterial endotoxins is such that the requirement in the relevant dosage form monograph(s) in which Polysorbate 20 is used can be met.■2S *(NF30)*

Change to read:

• ■**FATS AND FIXED OILS,** *Acid Value* ⟨**401**⟩■2S *(NF30)*
Sample: 10.0 g
Analysis: Transfer the *Sample* to a wide-mouth, 250-mL conical flask, and add 50 mL of neutralized alcohol. Heat on a steam bath nearly to boiling, occasionally shaking thoroughly while heating. Invert a beaker over the mouth of the flask, cool under running water, and add 5 drops of phenolphthalein TS. Titrate with 0.1 N sodium hydroxide VS. ■Calculate the acid value as directed in the chapter.■2S *(NF30)*
Acceptance criteria: ■NMT 2.0■2S *(NF30)*
• **FATS AND FIXED OILS,** *Hydroxyl Value* ⟨**401**⟩**:** 96–108

Add the following:

■• **FATS AND FIXED OILS,** *Peroxide Value* ⟨**401**⟩
Sample: 10.0 g
Saturated potassium iodide solution: Prepare a saturated solution of potassium iodide in carbon dioxide-free water. Make sure the solution remains saturated as indicated by the presence of undissolved crystals.
Analysis: Introduce the *Sample* into a 100-mL beaker, and dissolve with 20 mL of glacial acetic acid. Add 1 mL of *Saturated potassium iodide solution*, mix, and allow to stand for 1 min. Add 50 mL of carbon dioxide-free water and a magnetic stirring bar. Titrate with 0.01 M sodium thiosulfate VS, determining the endpoint potentiometrically (see *Titrimetry* ⟨541⟩). Perform a blank titration. Calculate the peroxide value as directed in the chapter.
Acceptance criteria: NMT 10.0
For Polysorbate 20 intended for use in the manufacture of injectable dosage forms: NMT 5.0■2S *(NF30)*
• **FATS AND FIXED OILS,** *Saponification Value* ⟨**401**⟩**:** 40–50
• **WATER DETERMINATION,** *Method I* ⟨**921**⟩**:** NMT 3.0%

ADDITIONAL REQUIREMENTS

Change to read:

• **PACKAGING AND STORAGE:** Preserve in tight containers, ■protected from light and moisture. Store at room temperature.■2S *(NF30)*