Food and Agriculture Organization of the United Nations

World Health Organization

Specification Monograph prepared by the meeting of the Joint FAO/WHO Expert Committee on Food Additives (JECFA), 87th Meeting 2019

BRILLIANT BLACK PN

This monograph was also published in: Compendium of Food Additive Specifications. Joint FAO/WHO Expert Committee on Food Additives (JECFA), 87th meeting 2019. FAO JECFA Monograph 23 (2019)

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Prepared at the 87th JECFA and published in JECFA Monograph 23 (2019) superseding specifications prepared at the 28th JECFA (1984), published in FNP 31/1 (1984) and in FNP 52 (1992). Metals and arsenic specifications revised at the 59th JECFA (2002). An ADI of 0-1 mg/kg bw was established at the 25th JECFA (1981) and confirmed at the 87th JECFA (2019).

SYNONYMS INS No. 151, CI Food Black 1, CI (1975) No. 28440, Black PN, Brilliant Black BN

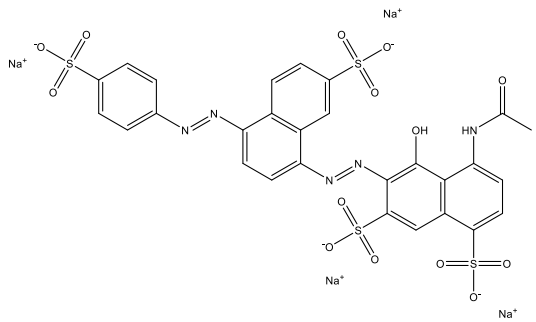
DEFINITION Brilliant Black PN consists of tetrasodium 4-(acetylamino)-5-hydroxy-6-[2-[7-sulfo-4-[2-(4-sulfophenyl)2pprox.2]-1-naphthalenyl]2pprox.2]-1,7-naphthalenedisulfonate and subsidiary colouring matters, as well as sodium chloride and/or sodium sulfate as the principal uncoloured components. Brilliant Black PN is manufactured by diazotizing 4-aminobenzenesulfonic acid (sulfanilic acid), coupling with 8-aminonaphthalene-2-sulfonic acid (1,7-Cleve's acid), diazotizing the product, and coupling with 4-(acetylamino)-5-hydroxy-1,7-naphthalenedisulfonic acid (N-acetyl K acid).
Brilliant Black PN may be converted to the corresponding aluminium lake in which case only the requirements in the *General Specifications for Aluminium Lakes of Colouring Matters* apply.

Chemical name Tetrasodium 4-acetamido-5-hydroxy-6-[7-sulfonato-4-(4-sulfonato-phenylazo)-1-naphthylazo]-1,7-naphthalenedisulfonate
Tetrasodium salt of 4-(acetylamino)-5-hydroxy-6-[[7-sulfo-4-[(4-sulfophenyl)azo]-1-naphthalenyl]azo]-1,7-naphthalenedisulfonic acid
Tetrasodium;(6E)-4-acetamido-5-oxo-6-[[7-sulfonato-4-[(4-sulfonatophenyl)2pprox.2]naphthalen-1-yl]hydrazinylidene]naphthalene-1,7-disulfonate

C.A.S. number 2519-30-4

Chemical formula $C_{28}H_{17}N_5Na_4O_{14}S_4$

Structural formula



Formula weight	867.69
Assay	Not less than 80% total colouring matters
DESCRIPTION	Black powder or granules
FUNCTIONAL USES	Colour
CHARACTERISTICS	
IDENTIFICATION	
Solubility (Vol. 4)	Soluble in water, sparingly soluble in ethanol.
Spectrophotometry (Vol. 4)	Maximum wavelength approximately 572 nm Determine the UV-visible absorption spectrum of the sample dissolved in water.
PURITY	
Loss on drying, chloride and sulfate as sodium salts (Vol. 4)	Not more than 20% Determine chloride as sodium chloride, sulfate as sodium sulfate, and loss on drying (135°, 6 h) as described in Volume 4 (under “Specific Methods, Food Colours”).
Water insoluble matter (Vol. 4)	Not more than 0.2%
Lead (Vol. 4)	Not more than 2 mg/kg Determine using a method appropriate to the specified level. The selection of sample size and method of sample preparation may be based on the principles of the method described in Volume 4 (under “General Methods, Metallic Impurities”).
Subsidiary colouring matters	Not more than 4% See description under TESTS
Organic compounds other than colouring matters	Not more than 0.8% sum of 4-(acetylamino)-5-hydroxy-1,7-naphthalenedisulfonic acid, 4-amino-5-hydroxynaphthalene-1,7-disulfonic acid, 8-amino-2-naphthalenesulfonic acid, sulfanilic acid, and 4,4’-(diazoamino)dibenzenesulfonic acid See description under TESTS
Unsulfonated primary aromatic amines (Vol. 4)	Not more than 0.01% calculated as aniline

Ether extractable matter (Vol. 4) — Not more than 0.2%

TESTS

PURITY TESTS

Subsidiary colouring matters

Determine subsidiary colouring matters content by reversed-phase HPLC (Vol. 4) using the following conditions:

- Column: Atlantis T3 RP18 (4.6 mm x 150 mm, 3 µm particle size) or equivalent
- Eluent A: 0.04 M ammonium acetate in water
- Eluent B: methanol
- Injection volume: 20 µl
- Column temperature: 35°
- Detector: UV-visible/diode array at 572 nm
- Flow rate: 0.8 ml/min

Gradient:

Elution time (min)	Eluent A (%)	Eluent B (%)
0	98	2
15	60	40
30	60	40
35	10	90
35.1	98	2
45	98	2

Reagents: HPLC grade

Standard:

- Brilliant Black PN (C.A.S. No. 2519-30-4) – USP Brilliant Black PN RS or equivalent

Prepare standard solutions as required using 0.1 M ammonium acetate in water as the solvent.

Sample solution (0.1 mg/ml):
Weigh accurately 100±2 mg of sample into a 100 ml volumetric flask and dilute to volume with 0.1 M ammonium acetate in water. Dilute the solution, if required, to separate subsidiary colours from the primary colour component in order to improve their resolution.

Procedure:
Inject the standard and sample solutions. Integrate all peaks in the chromatogram of the sample solution. Identify the peak of Brilliant Black PN from the chromatogram of the standard solution. Determine the ratio of the sum of all peak areas not corresponding to Brilliant Black PN to the sum of all peak areas. Calculate the result for subsidiary colours as a percentage of the sample weight.

Organic compounds other than colouring matters

Determine organic compounds other than colouring matters content by reversed-phase HPLC (Vol. 4) using the following conditions:

- Column: Atlantis T3 RP18 (4.6 mm x 150 mm, 3 µm particle size) or equivalent
- Eluent A: 0.04 M ammonium acetate in water
- Eluent B: methanol
- Injection volume: 20 µl
- Column temperature: 35°
- Detector: UV-visible/diode array at 254 nm
- Flow rate: 0.8 ml/min

Gradient:

Elution time (min)	Eluent A (%)	Eluent B (%)
0	98	2
15	60	40
30	60	40
35	10	90
35.1	98	2
45	98	2

Reagents: HPLC grade

Standards:

- 4-(Acetylamino)-5-hydroxy-1,7-naphthalenedisulfonic acid (N-acetyl K acid) (C.A.S. 6409-21-8) – ChemTik, Cat. No. CTK2F3097 or equivalent
- 4-Amino-5-hydroxynaphthalene-1,7-disulfonic acid (K acid) (C.A.S. 130-23-4) – BOC Sciences, Cat. No. 130-23-4 or equivalent
- 8-Amino-2-naphthalenesulfonic acid (1,7-Cleve's acid) (C.A.S. 119-28-8) – TCI Cat. No. A0356 or equivalent
- Sulfanilic acid (4-aminobenzenesulfonic acid) (C.A.S. 121-57-3) – Sigma, Cat. No. 251917 or equivalent
- 4,4'-(Diazoamino)dibenzenesulfonic acid (DAADBSA) (C.A.S. 17596-06-4) – Wako Cat. No. 040 33231 or equivalent

Prepare standard solutions as required using the following solvents:

- Dissolve N-acetyl K acid, K acid, and sulfanilic acid in water
- Dissolve 1,7-Cleve's acid in methanol/water (1:1)
- Dissolve DAADBSA in water containing 1 drop of 50% sodium hydroxide in water

Sample preparation:
Weigh accurately 100±2 mg of sample into a 100 ml volumetric flask and dilute to volume with 0.1 M ammonium acetate in water.

Procedure:
Inject the standard solutions. Integrate the chromatogram peaks obtained for N-acetyl K acid, K acid, 1,7-Cleve's acid, sulfanilic acid, and DAADBSA. Construct the relevant standard curves. Inject the sample solution and determine the concentration of each analyte from its respective standard curve.Calculate the percentage of each analyte in the sample and calculate their sum.

METHOD OF ASSAY Determine total colouring matters content by spectrophotometry using Procedure 1 in Volume 4 (under "Specific Methods, Food Colours") and an appropriate solvent. Analyze immediately after preparation.
Using water as the solvent: absorptivity (a) = 53.0 l/(g·cm) and wavelength of maximum absorbance = 572 nm.