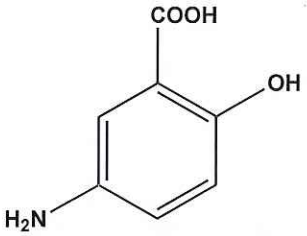
WS Batch: P03673

Certificate of Analysis

ISO GUIDE 34
ACLASS Cert# AR-1470

ISO/IEC 17025
ACLASS Cert# AT-1467

MESALAMINE
(Mesalazine)
CERTIFIED REFERENCE MATERIAL



CERTIFIED PURITY:

99.9%, U_{crm} = ±0.1% k = 2 (Mass Balance/dried basis)

99.8%, U_{crm} = ±0.1% k = 2 (Mass Balance/as is basis)

NOMINAL PACKAGE SIZE: 1g

CATALOG #: PHR1060 **LOT #:** LRAA7173

CERTIFICATE VERSION: LRAA7173.1 **ISSUE DATE:** 19 January 2015

Note: Certificates may be updated due to Pharmacopeial Lot changes or the availability of new data. Check our website at: www.sigma-aldrich.com *for the most current version.*

CRM EXPIRATION: 31 December 2019 (Proper Storage and Handling Required).

RECEIPT DATE: _____
Note: this space is provided for convenience only and its use is not required.

STORAGE: Store at Room Temperature/Protect from Light, keep container tightly closed. Attachment of a 20 mm aluminum crimp seal recommended for unused portions.

CHEMICAL FORMULA: $C_7H_7NO_3$ **MW:** 153.1

PHYSICAL DESCRIPTION: White powder in amber vial **CAS #:** 89-57-6

HAZARDS: Read Safety Data Sheet before using. All chemical reference materials should be considered potentially hazardous and should be used only by qualified laboratory personnel.

Fluka Analytical SIGMA-ALDRICH

INSTRUCTIONS FOR USE: For USP and EP applications, dry under vacuum at 105°C for 3 hours. For BP applications, use on the as is basis. The internal pressure of the container may be slightly different from the atmospheric pressure at the user's location. Open slowly and carefully to avoid dispersion of the material. This material is intended for R&D use only. Not for drug, household or other uses.

TRACEABILITY ASSAY

Comparative assay demonstrates direct traceability to Pharmacopeial Standards
Specification: 98.5–101.5% (USP)

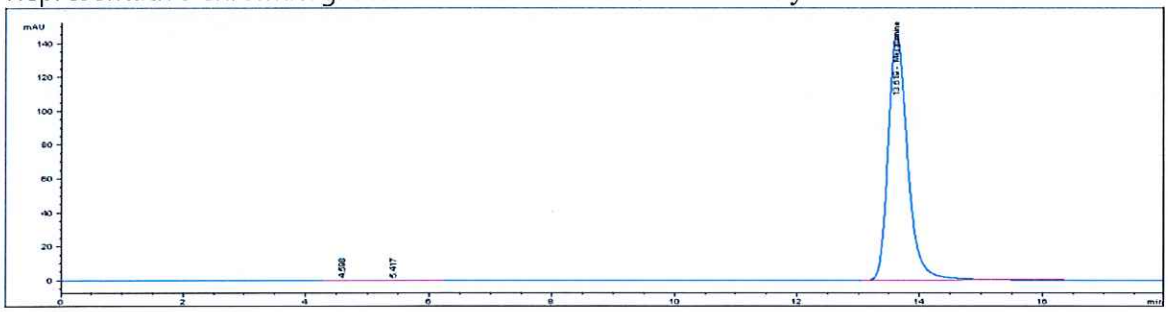
METHOD: HPLC (ref.: Mesalamine USP32)

ASSAY vs. USP REFERENCE STANDARD (dried basis)

ASSAY VALUE	vs. USP LOT
100.1%	**I0F121**
	Labeled Content = 1.000mg/mg

Column: Wakosil C18, 4.6 x 250mm, 5µm
Mobile Phase: Buffer, methanol; (85:15) Buffer: dibasic sodium phosphate, monobasic sodium phosphate, tetrabutylammonium hydroxide in methanol, water
Flow Rate: 2mL/min
Column Temperature: 27°C
Injection: 20µl
Detector: 254nm

Representative Chromatogram from Lot: LRAA7173 USP Analysis



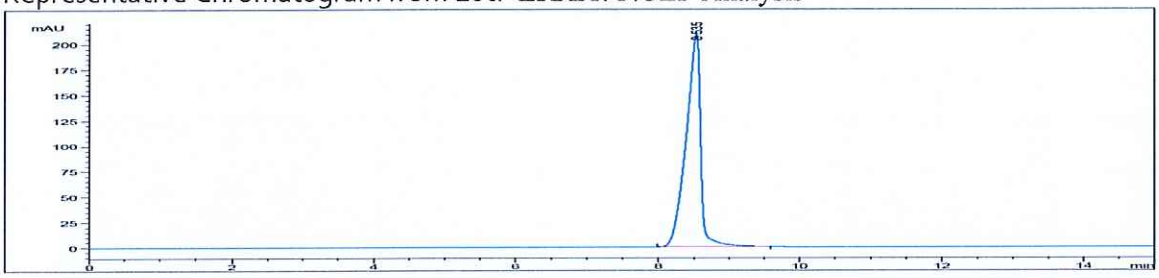
ASSAY vs. EP CRS (dried basis)

ASSAY VALUE	vs. EP BATCH
100.4%	**2.0**
	Labeled Content = None
	Assigned Content = 100.6%*

*The assigned content of the EP CRS was determined by assay against the USP Reference Standard

Column: ProteCol-GP C18 125, 4.6 x 250mm, 5µm
Mobile Phase: Buffer, methanol; (85:15) Buffer: dibasic sodium phosphate, monobasic sodium phosphate, tetrabutylammonium hydroxide in methanol, water
Flow Rate: 2mL/min
Column Temperature: 30°C
Injection: 20µl
Detector: 254nm

Representative Chromatogram from Lot: LRAA7173EP Analysis



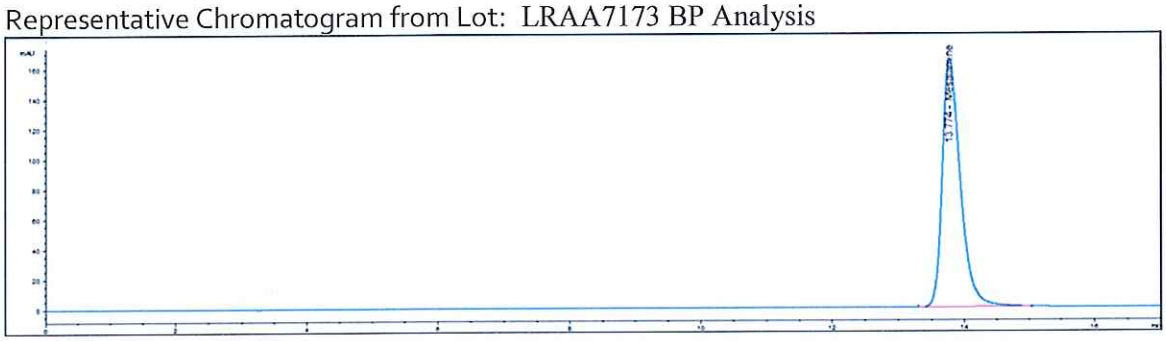
METHOD: HPLC (ref.: Mesalamine USP34)

ASSAY vs. BP CRS (as is basis)

ASSAY VALUE	vs. BP Batch
100.0%	**3391**
	Labeled Content = 99.2%

Column: Supelcosil LC-18, 4.6 x 250mm, 5µm
Mobile Phase: Buffer, methanol; (85:15) Buffer: dibasic sodium phosphate, monobasic sodium phosphate, tetrabutylammonium hydroxide in methanol, water
Flow Rate: 1.5mL/min
Column Temperature: 30°C
Injection: 15µl
Detector: 254nm

Representative Chromatogram from Lot: LRAA7173 BP Analysis



PURITY DETERMINATION BY MASS BALANCE

CHROMATOGRAPHIC IMPURITY ANALYSIS

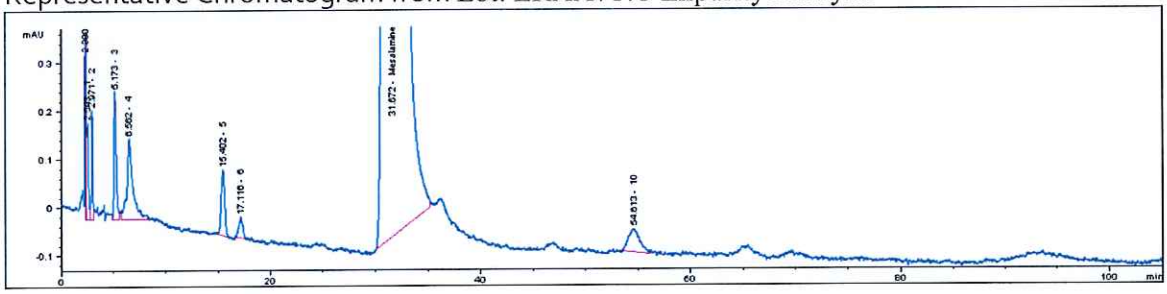
METHOD 1: HPLC (ref.: Mesalamine, USP32)

Column: Wakosil II C8, 4.6 x 250mm, 5µm
Mobile Phase: Buffer, methanol, acetonitrile; (89:8:3) Buffer: monobasic potassium phosphate, sodium 1-octanesulfonate, water, pH 2.2 with phosphoric acid
Flow Rate: 1mL/min
Column Temperature: 27°C
Injection: 20µl
Detector: 220nm

Impurities Detected:

Impurity 1:	**0.006%**	Impurity 2:	**0.006%**
Impurity 3:	**0.01%**	Impurity 4:	**0.02%**
Impurity 5:	**0.01%**	Impurity 6:	**0.005%**
Impurity 7:	**0.02%**	Impurity 8:	**0.01%**
2-Aminosalicylic acid:	**0.005%**		
Total Impurities:	**0.09%**		

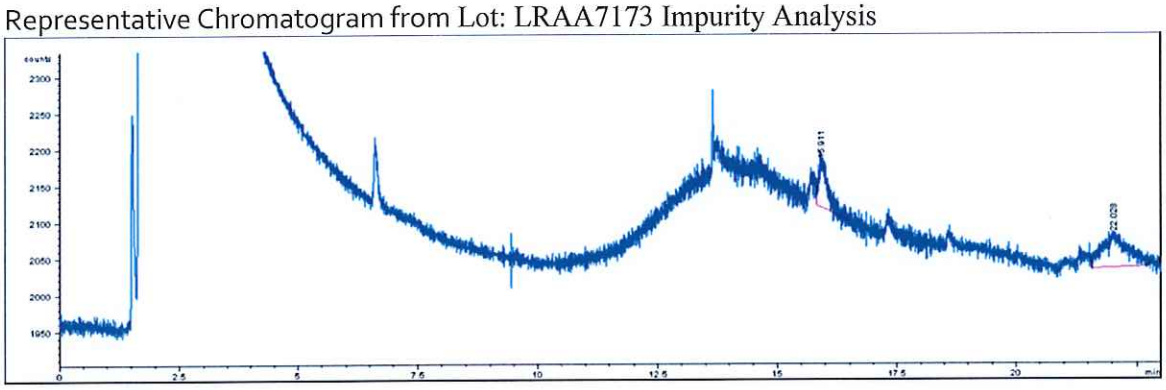
Representative Chromatogram from Lot: LRAA7173 Impurity Analysis



METHOD 2: GC (ref.: Mesalamine, USP32)
Column: AT-5MS, 0.25mm x 30m x 0.25µm
Carrier: He
Flow: 2mL/min
Split Ratio: 1:50
Temperature Program: 40°C/10min, 20°C/min to 200°C, hold 5min
Injection: 2µ/280°C
Detector: FID/300°C

Impurities Detected: **None Detected**

Representative Chromatogram from Lot: LRAA7173 Impurity Analysis



RESIDUAL SOLVENTS
Method: GC-MS Headspace (ref.: Residual Solvents <467>, USP34)
Column: DB-1301
Carrier gas: He
Flow: 1.2mL/min
Split Ratio: 1:5
Injection/Temperature: 1µl/250°C
Temperature Program: 40°C for 20min, 10°C/min to 240°C, hold 20min

Solvents Detected: None

LOSS ON DRYING/VOLATILES
Method: Oven at 105°C
Mean of three measurements, Loss = **0.1%**

RESIDUE ANALYSIS
Method: Sulfated Ash
Sample Size: ~1g
Mean of three measurements, Residue = **0.03%**

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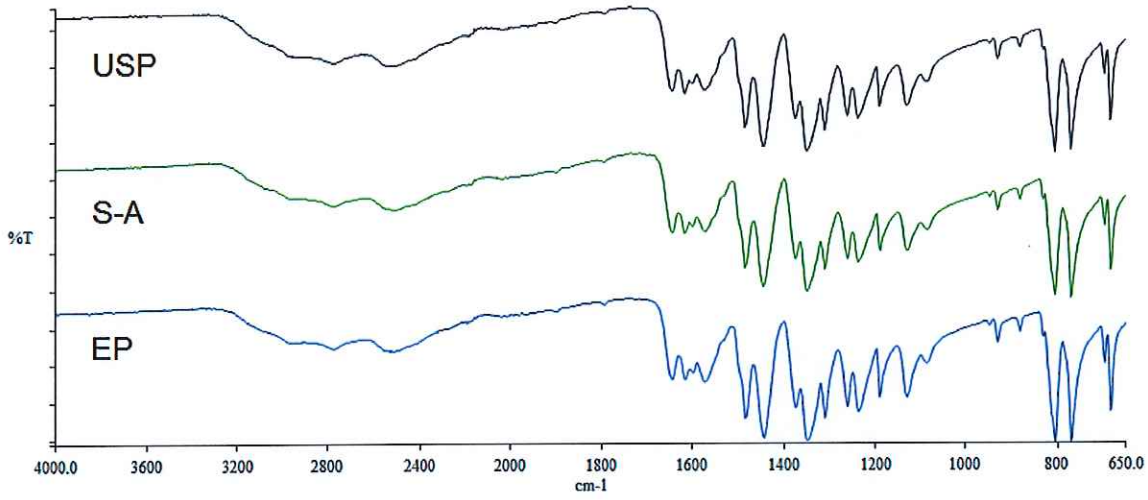
CERTIFIED PURITY BY MASS BALANCE [100% - Impurities (normalized)]

99.9% U_{crm} = ±0.1%, k = 2 (dried basis)
99.8% U_{crm} = ±0.1%, k = 2 (as is basis)

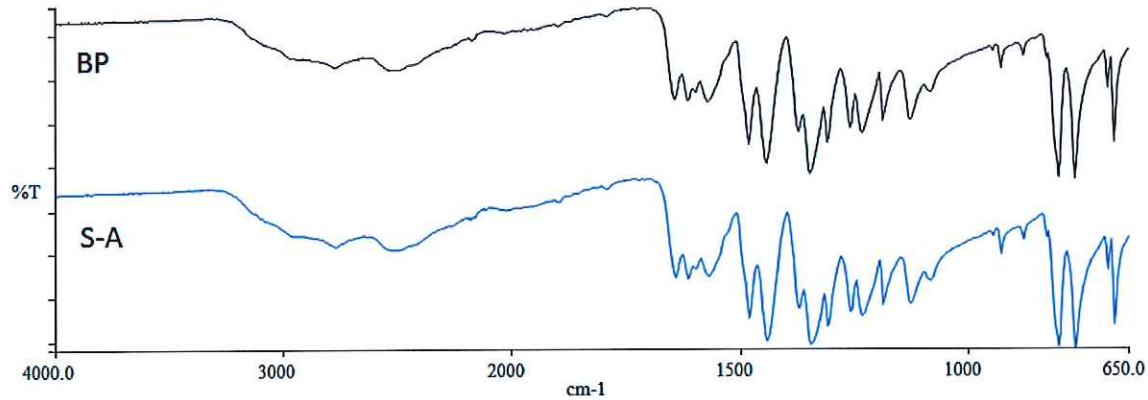
IDENTIFICATION TESTS

INFRARED SPECTROPHOTOMETRY (Comparative identification analysis demonstrates direct traceability to Pharmacopeial standards)



Mesalamine USP Lot: I0F121
Mesalamine S-A Lot: LRAA7173
Mesalazine EP Batch: 2.0

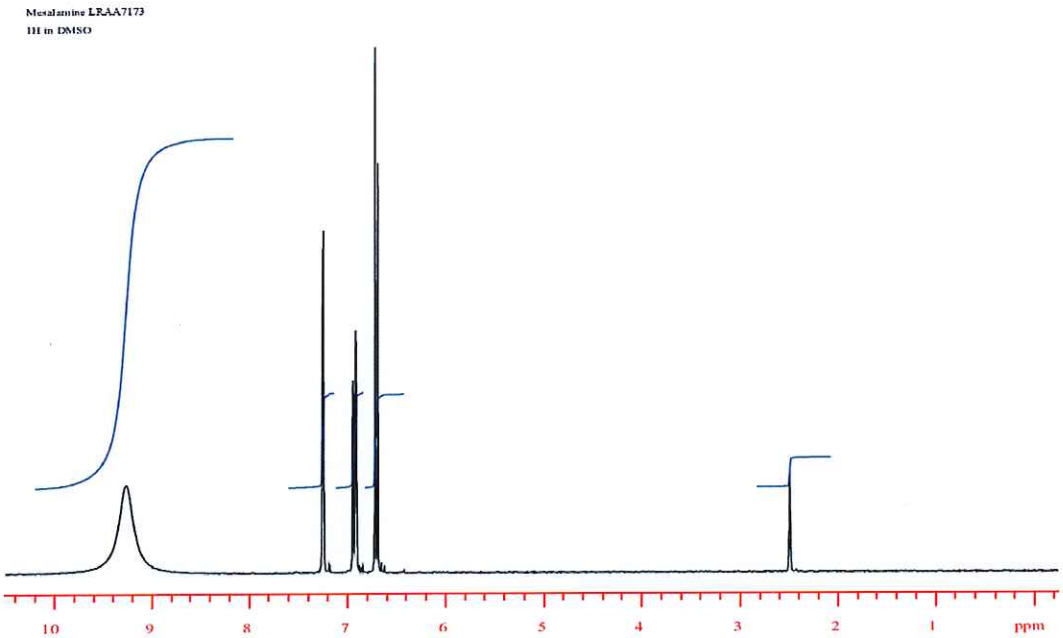
PERKIN ELMER SPECTRUM 100
SAMPLING: UNIVERSAL ATR



Mesalazine BP Batch 3391
Mesalamine S-A Lot LRAA7173

PERKIN ELMER SPECTRUM 100
SAMPLING: UNIVERSAL ATR

^{1}H NMR (Data provided by an external laboratory; not in scope of accreditation)



Consistent with structure

ELEMENTAL ANALYSIS (Data provided by an external laboratory; not in scope of accreditation)

Exeter Analytical 440 Elemental Analyzer

Combustion method

%	Theoretical	Result 1	Result 2	Mean
C	54.90	54.79	54.85	54.82
H	4.61	4.65	4.57	4.61
N	9.15	9.08	8.99	9.04

HOMOGENEITY ASSESSMENT

Homogeneity was assessed in accordance with ISO Guide 35. Completed units were sampled using a random stratified sampling protocol. The results of chemical analysis were then compared by Single Factor Analysis of Variance (ANOVA). The uncertainty due to homogeneity was derived from the ANOVA. Heterogeneity was not detected under the conditions of the ANOVA.

Analytical Method: HPLC Sample size: ~40mg

UNCERTAINTY STATEMENT

Uncertainty values in this document are expressed as Expanded Uncertainty (U_{crm}) corresponding to the 95% confidence interval. U_{crm} is derived from the combined standard uncertainty multiplied by the coverage factor k, which is obtained from a *t*-distribution and degrees of freedom. The components of combined standard uncertainty include the uncertainties due to characterization, homogeneity, long term stability, and short term stability (transport). The components due to stability are generally considered to be negligible unless otherwise indicated by stability studies.

STABILITY ASSESSMENT

Significance of the stability assessment will be demonstrated if the analytical result of the study and the range of values represented by the Expanded Uncertainty do not overlap the result of the original assay and the range of its values represented by the Expanded Uncertainty. The method employed will usually be the same method used to characterize the assay value in the initial evaluation.

Long Term Stability Evaluation - An assessment, or re-test, versus a Compendial Reference Standard may be scheduled, within the 3 year anniversary date of a release of a Secondary Standard. The re-test interval will be determined on a case-by-case basis.

Short Term Stability Study - It is useful to assess stability under reasonably anticipated, short term transport conditions by simulating exposure of the product to humidity and temperature stress. This type of study is conducted under controlled conditions of elevated temperature and humidity.

Operations Manager

QA Supervisor

APPENDIX

Original Release Date: 19 January 2015

Manufactured and certified by Sigma-Aldrich RTC, Inc.
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(Phone): 1-307-742-5452 (Fax): 1-855-831-9212
email: RTCTechGroup@sial.com

ACCREDITED REFERENCE MATERIAL PRODUCER

ACCREDITED TESTING