Automated Sample Preparation Methods for Content Uniformity and Assay of Rosuvastitin Tablets

Application Note

Authors

Sven Saladin
Product & Application Specialist
accroma labtec Ltd.

Abstract

Manual sample preparation methods for Content Uniformity and Assay of Rosuvastatin Calcium Tablets were automated on the accroma® samplePrep system. The goal was to reduce extraction and overall sample preparation hands-on times, while improving data quality and reducing human errors. Rosuvastatin drug contents were determined using an online HPLC interfaced to accroma. The reference standard, 10 individual tablets for Content Uniformity, and 5 tablets for Assay were automatically prepared. The extraction was done by vertical shaking of accroTubes with 16mm steel balls as grinding and mixing aids. The measured contents of all samples were all within 102% and 105%, which are within specification. The relative standard deviation (RSD) of the ten Content Uniformity measurements was calculated to 0.98%. The extraction times could be reduced by 60% for Assay and 80% for Content Uniformity. Since shorter milling and extraction times were not tested, the extraction time could be potentially shortened even more. The estimated overall hands-on time could be reduced by 83% for Content Uniformity and by 87.5% for Assay without counting documentation efforts. The results demonstrate the successful automation of sample preparation methods for Content Uniformity and Assay testing.

Introduction

The key attributes in quality control of oral solid dosage forms (OSDs) are identity, strength, and purity. [1] Contents of Active Pharmaceutical Ingredients (API), in tablets are assessed in Content Uniformity and Assay tests. While the Uniformity of Dosage Units test USP<905> requires sample preparation and analysis of ten individual tablets, multiple tablets are pooled for Assay tests. Preparing samples manually is time-consuming, expensive, and prone to errors. In chromatography, sample preparation requires 61% of lab technician's time, accounts for a minimum of 30% of all errors and adds up to more than 50% of the total lab costs. [2] As documentation is done manually, traceability is difficult to achieve. Therefore, automated sample preparation is key to improve productivity and compliance.

Manual sample preparation method

Stock solution (500 µg/ml) of rosuvastatin calcium reference standard was prepared by transferring 25 mg, accurately weighed, into a 50 ml volumetric flask and adding 20 ml water/acetonitrile (50:50, v/v). The mixture was sonicated for 2 min to dissolve the rosuvastatin calcium and the solution was then diluted to volume with the same solvent mixture.

Standard solution (50 µg/ml) was prepared by diluting 5 ml standard stock solution to 50 ml, in a volumetric flask, with the same solvent mixture. To prepare stock solution (500 µg/ml) for assay, 20 tablets were weighed and mixed. An aliquot of powder equivalent to the weight of 5 tablets was accurately weighed and transferred to a 100 ml volumetric flask. 60 mL of water/acetonitrile (50:50, v/v), was added to the flask and the mixture was mixed for 10 min with normal hand shaking. The contents of the flask were then left to return to room temperature and diluted to volume with the same solvent mixture. This solution (10 ml) was filtered through a 0.45-µm nylon syringe filter.

To prepare test solution (50 µg/ml) for assay, 5 mL of test stock solution was transferred to a 50 mL volumetric flask and diluted to volume with water/acetonitrile (50:50, v/v). To prepare the test solution (50 µg/ml) for determination of content uniformity, one tablet was accurately weighed and transferred to a 200 mL volumetric flask. Water-acetonitrile (50:50, v/v) 100 mL was added to the flask and the mixture was mixed for 10 min with normal hand shaking. The contents of the flask were left to return to room temperature, then diluted to volume with the same solvent mixture and filtered through a 0.45 µm nylon syringe filter. Twenty tablets were weighed, and the average weight of a tablet was used for assay calculation. [3]

Experimental

Chemicals and reagents

- Rosuvastatin-Mepha tablets 10 mg (PC: 07680664170047, SN: 10063858391625, Lot: 15456, EXP: 03/2023
- Working standard Rosuvastatin Calcium (purity 96.1%)
- Water, HPLC grade
- Acetonitrile, HPLC grade
- Phosphoric acid 85%
- Mobile phase
  - Acetonitrile (50 parts)
  - Water (50 parts)
  - Adjusted to pH 3.5 with Phosphoric acid
- Extraction solvent (diluent)
  - Acetonitrile (50 parts)
  - Water (50 parts)

Instrumentation

accroma samplePrep system

- Analytical balance Mettler-Toledo WMS 404C-L/11
- Shaker module
- Liquid- & Filtration module
- Agilent Online analysis interface
- accroLab software

Agilent 1260 Infinity II HPLC (400 bar)

- 1260 Infinity II Quat Pump VL (G7111A)
- 1260 Infinity II Vial sampler (G7129A) with Integrated Column Compartment and External Tray
- 1260 Infinity II Variable Wavelength Detector (G1314F)
- Chromatographic Data System (CDS): Chromeleon 7.3

Procedure

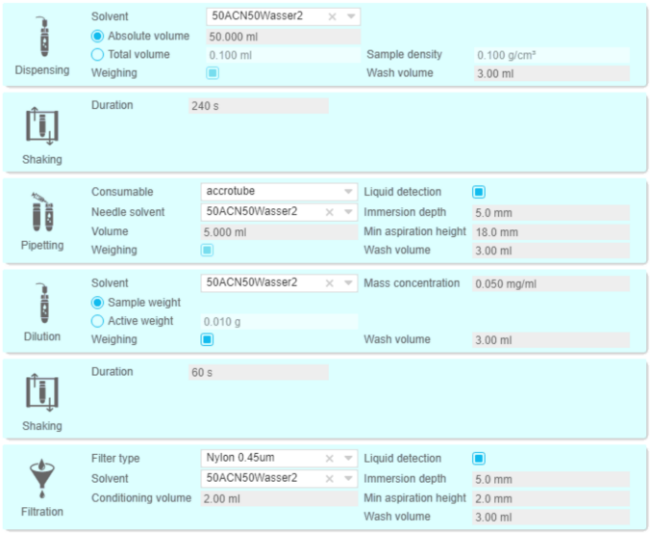
Chromatography

Parameter	Value
Mobile Phase A	Water/Acetonitrile 50:50 pH 3.5
Flow Rate	1.5 ml/min
Injection Volume	20 µl
Column Temperature	25°C
UV Wavelength	242 nm
Gradient	**Time** **Min** **%A** 0 100 6.00 100
Column	Zorbax Extend C18, (150 x 4.6) mm, 5µm

Sample injections (1-10) were bracketed between two working standard injections.

Standard preparation

25.1mg of working standard (Purity: 96.1%) was weighed and transferred into an accroTube containing a steel ball (16mm). The accroTube was placed into the accroma system and the following workflow was started.

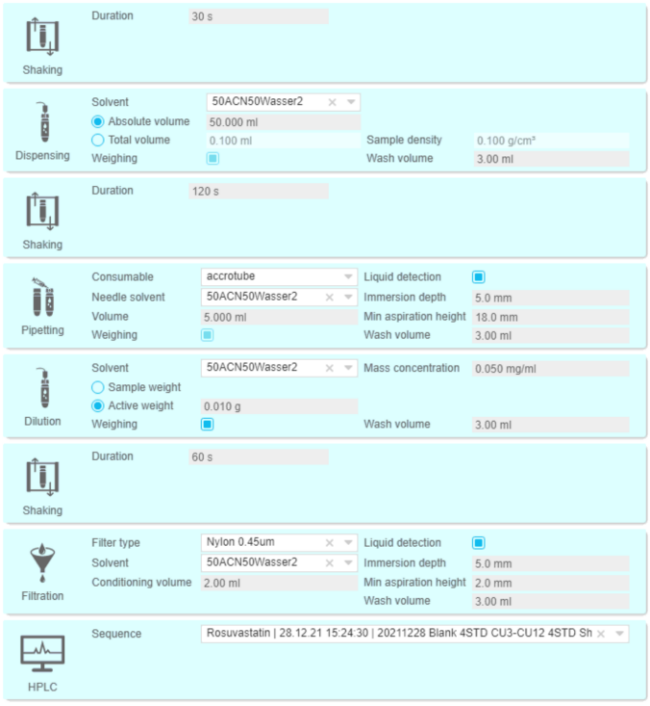


Standard preparation workflow on the accroma.

Automated content uniformity samplePrep method

Compared to the extraction in the manual sample preparation where the samples were first ground, the extraction was performed by vertical shaking of the accroTubes.

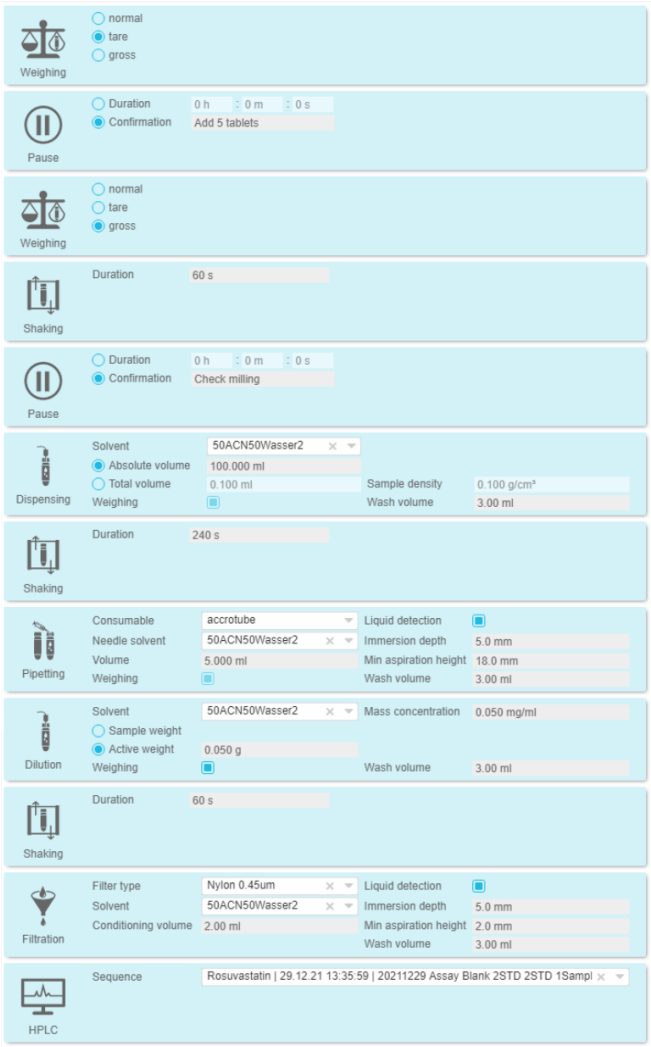
One Rosuvastatin tablet (10 mg) was weighed and transferred into an accroTube with a steel ball (16mm). The closed accroTube was placed into the accroma system and the following workflow was started.



Sample preparation workflow content uniformity accroma

Automated assay samplePrep method

One steel ball (16mm) was added to an accroTube. The closed accroTube was placed into the accroma system. After starting the workflow, the accroTube was tared automatically. Subsequently 5 randomly selected Rosuvastatin tablets (10mg) were loaded into the accroTube, placed back into the accroma system and the workflow was continued.



Sample preparation workflow assay accroma

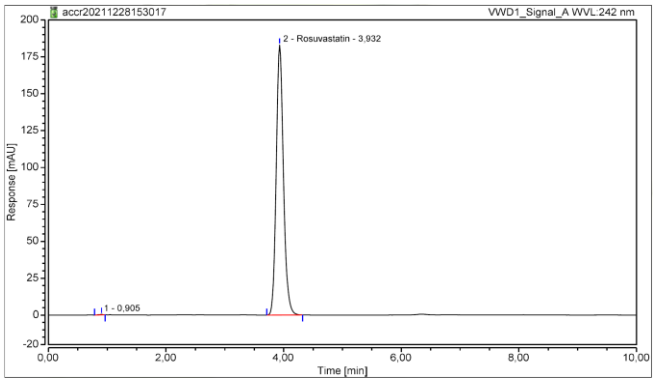
For content uniformity workflow development, two workflow experiments with different milling and extraction times were performed.

Experiment	Milling	Extraction	Total
CU 1	0.5 min	2 min	3 min
CU 2	1 min	4 min	4.5 min

No difference in the results was observed with longer extraction times. The final measurements were therefore carried out with the shorter extraction time.

Results and discussion

The acquired chromatograms of the sample solutions showed peaks between 3.5 and 4.5 minutes. No non-expected peaks were identified comparing the working standard and the samples. Therefore, it was assumed that no degradation took place during the milling and extraction process.



Chromatogram of sample solution prepared by workflow 1

A full comparison of the measured concentrations of the automated sample preparations are shown in the following table:

Experiment	Concentration	Recovery
CU 1	50.2 µg/ml	105.0 %
CU 2	50.2 µg/ml	103.6 %
CU 3	50.3 µg/ml	104.8 %
CU 4	50.2 µg/ml	104.4 %
CU 5	50.3 µg/ml	104.4 %
CU 6	50.3 µg/ml	102.3 %
CU 7	50.3 µg/ml	104.4 %
CU 8	50.3 µg/ml	103.5 %
CU 9	50.3 µg/ml	103.1 %
CU 10	50.3 µg/ml	105.8 %
Assay	50.2 µg/ml	104.2 %

Even though the contents of the tablets were measured slightly higher than labelled, the contents met manufacturer's tolerances and regulatory specifications. A possible explanation for the higher recoveries is incomplete dissolution of the reference standard. The results were not carried out with the original method. Therefore, they may differ from the manufacturer's values.

Conclusion

Extraction times by the accroma samplePrep system were shown to be significantly shorter than by the manual method of ultrasonication.

Parameter	Manual	accroma	Improvement
Extraction time CU	10 min	2 min	80% reduction
Hands-on time CU	30 min*	5 min	83% reduction*
Extraction time Assay	10 min	4 min	60% reduction
Hands-on time Assay	40 min*	5 min	87.5% reduction*

* Estimated value.

The accroma system proved to be suitable for the automated sample preparation of Rosuvastatin Calcium tablets (10 mg) for Content Uniformity and Assay. The extraction time could be reduced by 80%. The overall sample preparation hands-on times could be reduced significantly. The accroma system can automate sample weighing, pipetting, extraction, filtration, and transferring vials into the HPLC for online analysis. The system verifies every added volume gravimetrically, which increases traceability and accuracy. It documents every possible parameter and step of the process. In conclusion, lab efficiency and compliance are significantly improved by using the accroma samplePrep system.

References

[1] Görög S., Drug Safety, drug quality, drug analysis 2008, Journal of Pharmaceutical and Biomedical Analysis, Vol. 48, Issue 2.

[2] Majors R. e., Sample preparation – Fundamentals for chromatography, 2013, p. 4

[3] Kaila H. O. et al., A New Improved RP-HPLC Method for Assay of Rosuvastatin Calcium in Tablets, Indian Journal of Pharmaceutical Sciences, 2010, 72(5): p. 592-598

www.accroma.com

Printed in Switzerland, Juli 2021
9652-3923EN

accroma
accuracy is key