APPLICATIONS

USP Dissolution Test 3 for Metformin Hydrochloride Tablets

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Outside of the lab, Laura enjoys spoiling her dog Maggie and subjecting her husband to novel methods of torture, such as end-less playlists of sad songs and long walks on the beach to catch Pokémon.

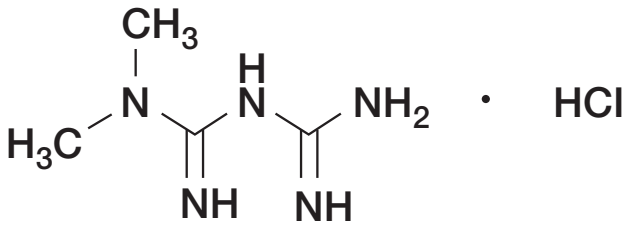
Overview

Metformin is a drug approved by U.S. FDA to treat Type 2 diabetes (also known as non-insulin-dependent diabetes). With the increasing diagnosis of Type 2 diabetes worldwide, it may be expected that treatment with metformin will continue to increase. Quality control testing of metformin oral dose drug products will then take on increased importance, and analytical testing based on USP methods such as dissolution and related bioequivalence (BE) studies need to be more robust and efficient.

Consistent with common bioequivalence studies used by generic pharmaceutical drug companies, we have demonstrated the chromatography for the dissolution of metformin hydrochloride tablets per the USP method (dissolution test 3) using the recommended Prodigy™ ODS-3 5 µm 250 x 4.6 mm column. Comparable chromatography on a Kinetex® core-shell column with the same particle size and dimension has also been demonstrated in this study. As expected, the Kinetex column exhibited improved speed and sensitivity than the column described in the USP monograph because of the inherent benefits derived from the Kinetex core-shell particle.

Material

Metformin Hydrochloride
Molecular Formula: $C_4H_{11}N_5 \bullet HCl$



Metformin Hydrochloride standard and all other reagents and chemicals were purchased from Sigma-Aldrich.

Experimental Conditions

Dissolution procedure

The dissolution batch and samples were prepared based on the USP monograph for Metformin Hydrochloride Standard and Tablets (dissolution test 3). The dissolution medium was prepared by dissolving 1.38 g of monobasic sodium phosphate in about 1800 mL of water; 3.484 g of 1-pentanesulfonic acid sodium salt was added, then adjusted to a pH of 3.00 ± 0.05 with diluted phosphoric acid and brought to 2000 mL in a volumetric flask. 500 mg Metformin Hydrochloride Tablets were dissolved in the pH 6.8 phosphate buffer dissolution medium.

Apparatus: basket, 100 RPM, 1000 mL

Time points: a single time point was taken at 60 minutes

Standard solution: 0.05 mg/mL in medium

The sample was filtered through a 0.45 µm Phenex Nylon syringe filter (Part No. AF0-2107-12). The sample can be diluted with medium, if necessary, to obtain a solution with a concentration similar to that of the Standard solution

Mobile phase preparation

The mobile phase is acetonitrile and buffer (1:19). The buffer was prepared as above in the dissolution procedure.

LC-UV conditions

Column 1:	Prodigy 5 µm ODS-3, 250 x 4.6 mm
Part No.:	00G-4097-E0
Column 2:	Kinetex 5 µm C18, 250 x 4.6 mm
Part No.:	00G-4601-E0
Mobile Phase:	Acetonitrile and Buffer (1:19)
LC condition:	Isocratic
Flow rate:	1.0 mL/min
Injection Volume:	40 µL
Temperature:	Ambient
Detection:	UV @ 230 nm
HPLC System:	Agilent® 1100 (Agilent Technologies®, Santa Clara, CA, USA)
Run time:	30 minutes

Figure 1.
Representative chromatogram of metformin standard (0.05 mg/mL)

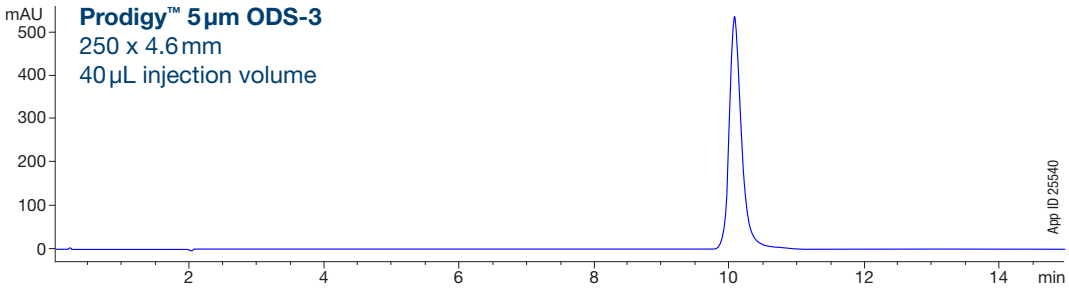


Figure 2.
Representative chromatogram of 500 mg tablet dissolved in 1L of buffer, 10 x dilution (0.05 mg/mL)

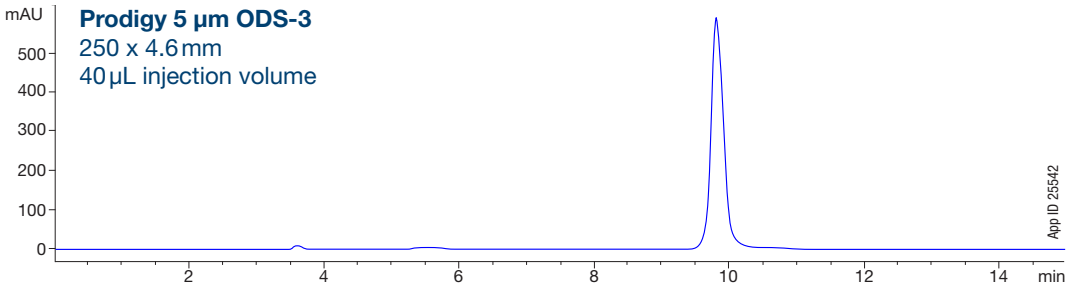


Figure 3.
Representative chromatogram of metformin standard (0.05 mg/mL)

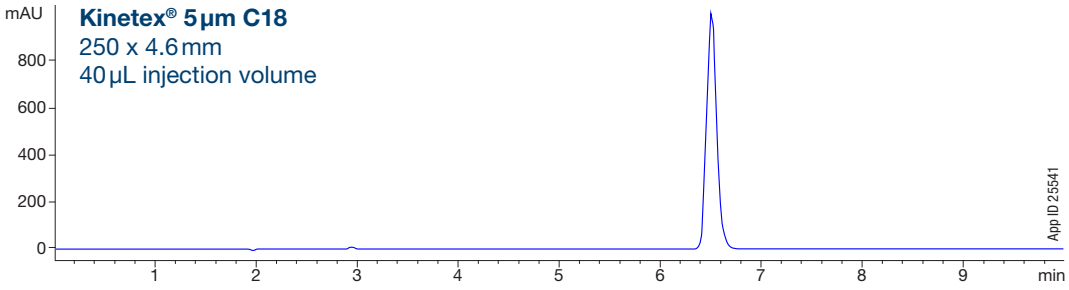
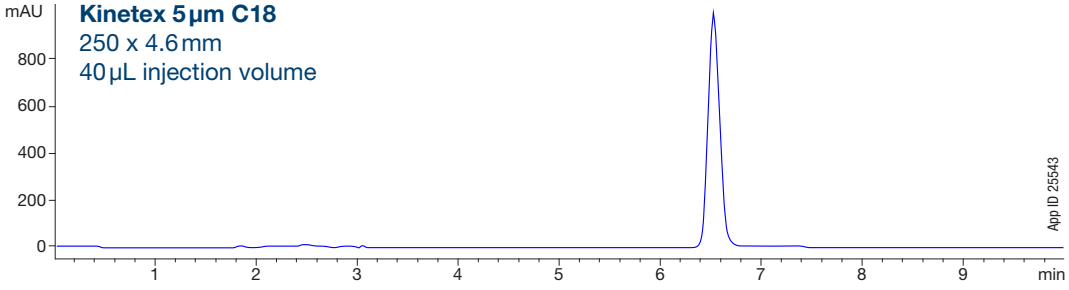


Figure 4.
Representative chromatogram of 500 mg tablet dissolved in 1L of buffer, 10 x dilution (0.05 mg/mL)



Comparative separations may not be representative of all applications.

LC-UV conditions for all columns:

Column 1:	Prodigy 5 µm ODS-3, 250 x 4.6 mm
Part No.:	00G-4097-E0
Column 2:	Kinetex 5 µm C18, 250 x 4.6 mm
Part No.:	00G-4601-E0
Mobile Phase:	Acetonitrile and Buffer (1:19)
LC condition:	Isocratic
Flow rate:	1.0 mL/min
Injection Volume:	40 µL
Temperature:	Ambient
Detection:	UV @ 230 nm
HPLC System:	Agilent® 1100 (Agilent Technologies®, Santa Clara, CA, USA)
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Metformin Hydrochloride

Molecular Formula: $C_4H_{11}N_5 \bullet HCl$

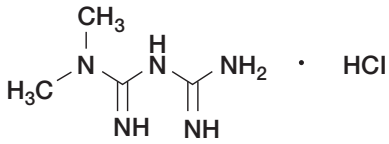

Table 1.

Prodigy™ ODS-3, 5 µm, 250 x 4.6 mm (Injection vol. 40 µL)			Kinetex® C18, 5 µm, 250 x 4.6 mm (Injection vol. 40 µL)		
Sample Conc.: 0.05 mg/mL					
USP Tailing factor	Peak Height	Efficiency (plates/meter)	USP Tailing factor	Peak Height	Efficiency (plates/meter)
0.67	536.5	14,361	0.81	999.4	18,731
Standard Conc.: 0.05 mg/mL					
0.784	592.2	12,959	0.83	991.9	15,087

Results and Discussion

Figure 1 and **Figure 2** show the chromatograms for metformin hydrochloride standard and tablet dissolution test with the Prodigy ODS-3 column recommended for USP dissolution test 3. The retention time for metformin is about 10.0 minutes. **Figure 3** and **Figure 4** are the chromatograms for metformin hydrochloride standard and tablet dissolution test with the Kinetex C18 column. The retention time for metformin is about 6.5 minutes.

Unlike the fully porous Prodigy C18 column, the Kinetex C18 column utilizes a core-shell particle composed of a solid silica core surrounded by a thin porous shell of silica. The benefits of the core-shell particle morphology are typically narrower peaks with greater peak height due to the higher efficiency expressed by core-shell particles. In addition, the lower surface area yields shorter retention times and reduced analysis times, without loss in chromatographic separation power. As shown in the figures, the retention time for metformin on the Kinetex C18 column (~6.5 minutes) is much shorter than on the Prodigy C18 column (~10.0 minutes) with the same dimension. As shown in **Table 1**, the column efficiency of Kinetex C18 is much higher than Prodigy ODS-3, and the peak intensity is significantly greater for standards and samples of the same concentration. This indicates that one could inject a lower concentration sample, as in the case for an extended release formulation or for earlier time points in a dissolution analysis; or inject a smaller sample volume while still obtaining good peak height and peak shape.

The system suitability requirements for USP dissolution test 3 for Metformin Hydrochloride Tablets Dissolution are for the tailing factor to be no more than (NMT) 2.0 and column efficiency no less than (NLT) 1500 theoretical plates. As shown in **Table 1**, both Prodigy ODS-3 and Kinetex C18 easily meet these requirements.

Conclusion

In vitro dissolution testing is an important tool that can be used for approval of safe and effective generic drug products. In this tech note, we have demonstrated the USP Dissolution Method 3 for Metformin Hydrochloride Tablets with two different columns, the fully porous column Prodigy ODS-3 as suggested in the USP monograph and core-shell column Kinetex C18 column with the same particle size and dimension. One of the benefits of the Kinetex core-shell column is an increase in chromatographic efficiency, which yields narrower peaks. The lower surface area associated with the Kinetex core-shell particle also contributes to shorter retention time for metformin, which results in a reduction in the total analysis time. The faster analysis achieved using the Kinetex column would be very useful for a laboratory looking to increase sample throughput (for example, where many dissolution samples need to be analyzed), without compromising the results for a bioequivalence (BE) study.

Ordering Information

Prodigy LC Columns

5 µm Analytical Columns (mm)						SecurityGuard™ Cartridges*
Phases	30 x 4.6	50 x 4.6	100 x 4.6	150 x 4.6	250 x 4.6	10/pk
ODS-3	00A-4097-E0	00B-4097-E0	00D-4097-E0	00F-4097-E0	00G-4097-E0	AJ0-9532
						for 3.2-8.0 mm ID

Kinetex Core-Shell LC Columns

5 µm Analytical Columns (mm)					SecurityGuard ULTRA Cartridges‡
Phases	50 x 4.6	100 x 4.6	150 x 4.6	250 x 4.6	3/pk
C18	00B-4601-E0	00D-4601-E0	00F-4601-E0	00G-4601-E0	AJ0-8768
					for 4.6 mm ID

* SecurityGuard Standard Cartridges require holder, Part No.: KJ0-4282.
‡ SecurityGuard ULTRA Cartridges require holder, Part No.: AJ0-9000.

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