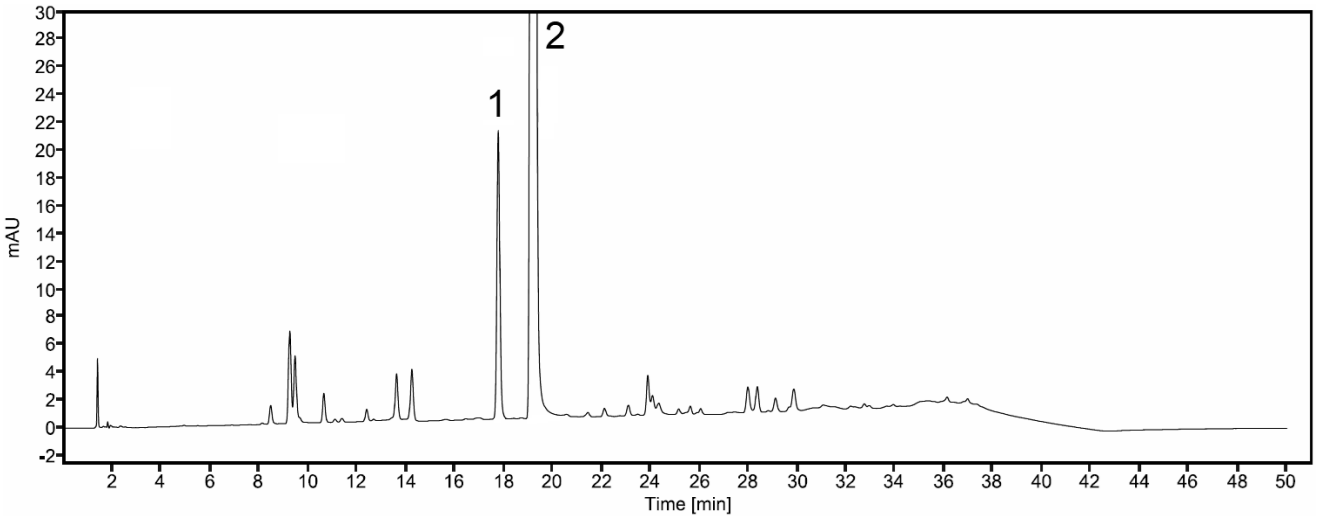
Flucloxacillin Injection – BP 2025

These chromatograms are provided for information only as an aid to analysts and are intended as guidance for the interpretation and application of BP monographs.

Typical chromatogram for solution (3) from the Related Substances test for Flucloxacillin Injection as published in BP 2025.

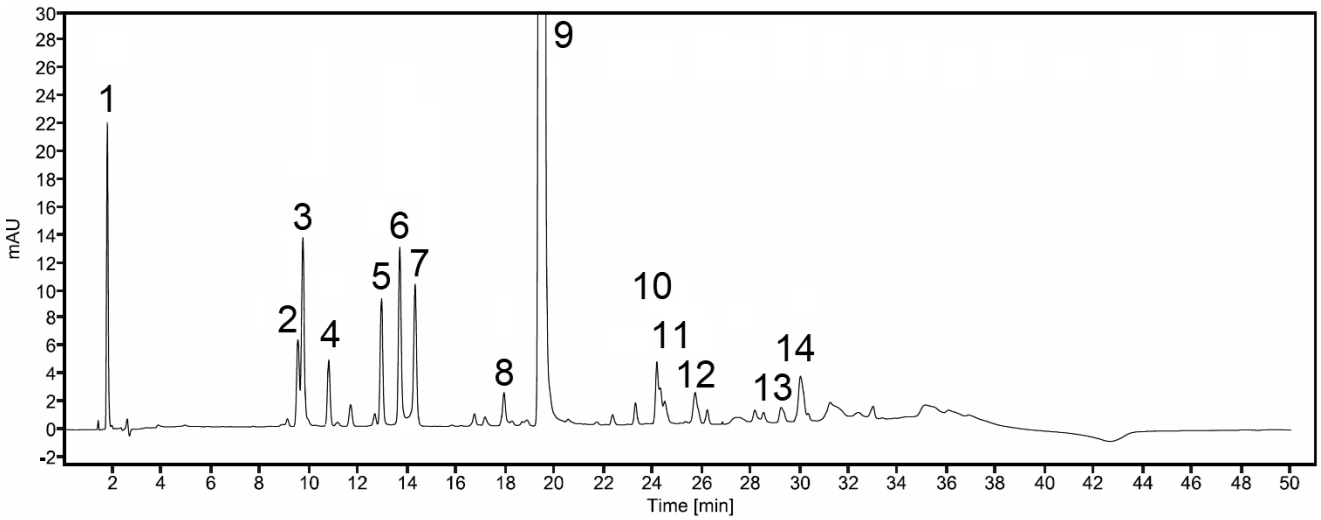


Peak ID: 1: Impurity D. 2: Flucloxacillin.

Column	Zorbax SB C18 (250 mm x 4.6 mm, 5 µm)
Method Ref.	Related Substances for the Flucloxacillin Injection monograph from BP 2025
Mobile Phase A	0.118% w/v sodium hexanesulfonate monohydrate dissolved in a mixture of 0.8 volumes of concentrated ammonia and 1000 volumes of water, adjusted to pH 2.9 with orthophosphoric acid
Mobile Phase B	Acetonitrile
Diluent	50% Acetonitrile
Flow rate	Refer to gradient table below
Column Temp	40°C

Injection Volume	10 µL			
Detection	225 nm			
Gradient				
Time (minutes)	**Mobile phase A (% v/v)**	**Mobile phase B (% v/v)**	**Flow rate (mL/min)**	**Comment**
0 – 30	80 → 45	20 → 55	1.5	linear gradient
30 – 35	45 → 35	55 → 65	1.5	linear gradient
35 – 40	35 → 80	65 → 20	1.5	linear gradient
40 – 45	80	20	1.5	re-equilibration

Typical chromatogram for solution (4) from the Related Substances test for Flucloxacillin Injection as published in BP 2025.



Peak ID: 1: Impurity C. 2: Impurity A (isomer 1). 3: Impurity A (isomer 2). 4: Impurity F. 5: Impurity G. 6: Impurity B (isomer 1). 7: Impurity B (isomer 2). 8: Impurity D. 9: Flucloxacillin. 10: Impurity H. 11: Impurity E. 12: Impurity I. 13: Impurity J. 14: Impurity K.

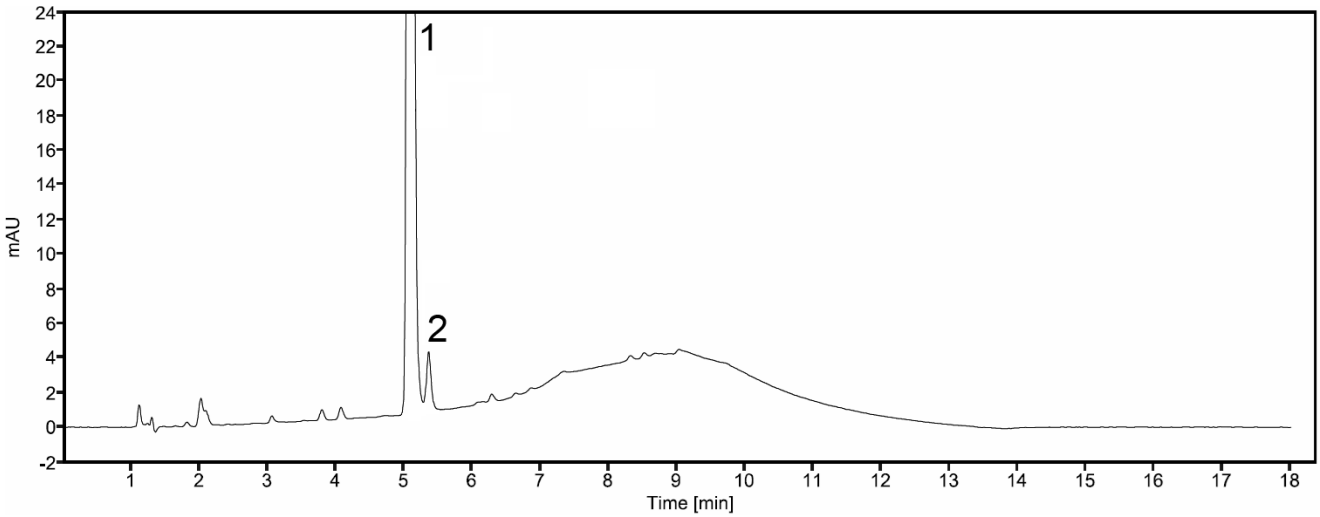
Column	Zorbax SB C18 (250 mm x 4.6 mm, 5 µm)
Method Ref.	Related Substances for the Flucloxacillin Injection monograph from BP 2025
Mobile Phase A	0.118% w/v sodium hexanesulfonate monohydrate dissolved in a mixture of 0.8 volumes of concentrated ammonia and 1000 volumes of water, adjusted to pH 2.9 with orthophosphoric acid
Mobile Phase B	Acetonitrile
Diluent	50% Acetonitrile
Flow rate	Refer to gradient table below
Column Temp	40°C
Injection Volume	10 µL
Detection	225 nm

Gradient

Time (minutes)	Mobile phase A (% v/v)	Mobile phase B (% v/v)	Flow rate (mL/min)	Comment
0 – 30	80 → 45	20 → 55	1.5	linear gradient

30 – 35	45 → 35	55 → 65	1.5	linear gradient
35 – 40	35 → 80	65 → 20	1.5	linear gradient
40 – 45	80	20	1.5	re-equilibration

Typical chromatogram for solution (3) from the Assay test for Flucloxacillin Injection as published in BP 2025.



Peak ID: 1: Flucloxacillin 2: Impurity D.

Column	Zorbax SB C18 (250 mm x 4.6 mm, 5 µm)
Method Ref.	Assay for the Flucloxacillin Injection monograph from BP 2025
Mobile Phase A	0.118% w/v sodium hexanesulfonate monohydrate dissolved in a mixture of 0.8 volumes of concentrated ammonia and 1000 volumes of water, adjusted to pH 3.1 with orthophosphoric acid
Mobile Phase B	Acetonitrile
Diluent	50% Acetonitrile
Flow rate	Refer to gradient table below
Column Temp	40°C
Injection Volume	10 µL
Detection	225 nm

Gradient

Time (minutes)	Mobile phase A (% v/v)	Mobile phase B (% v/v)	Flow rate (mL/min)	Comment
0 – 8	65 → 41	35 → 59	1.8	linear gradient
8 – 12	41 → 65	59 → 35	1.8	linear gradient
12 – 18	65	35	1.8	re-equilibration