

VersaFlash® Application Report 254

Raw Data File Name: RBI/BN-021104

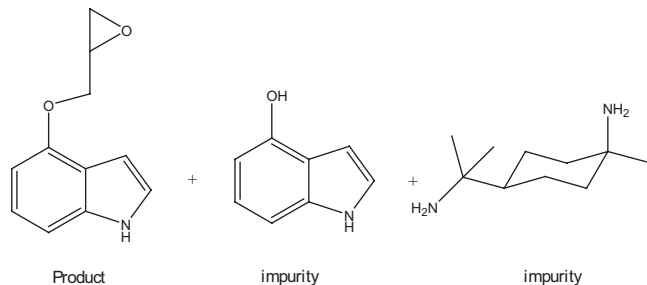
Step Gradients

Step gradients are commonly used in HPLC but can also be very useful in flash chromatography, especially in situations where the target product elutes between two different impurities. VersaPak® cartridges can be packed in many different combinations to achieve the optimal length for the purification.

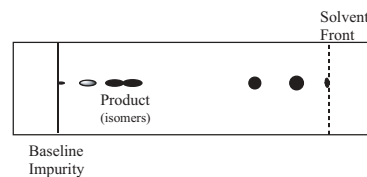
Key Words

step gradients, difficult separations

Reaction Product



TLC Plate (crude)



G002959

G002960

TLC Information

TLC plate: silica
Solvent: $i\text{-C}_3\text{H}_7\text{OH}:\text{NH}_4\text{OH}$ 97:3
Rf values:
Product: 0.32
Impurity: 0.85
Impurity: 0.65
Impurity: 0.25

HTEP Conditions

VersaPak® cartridge size: 3 stacked 40 mm x 75 mm cartridges
cartridge media: silica
sample size: 11 g
loading method: direct
mobile phase: 1. DCM:MeOH: NH_4OH 98.5:1:0.5
2. $i\text{-C}_3\text{H}_7\text{OH}:\text{NH}_4\text{OH}$ 99:1
3. $i\text{-C}_3\text{H}_7\text{OH}:\text{NH}_4\text{OH}$ 97:3
flow rate: 40 mL/min.

Results

volume collected: 6.4 L
product percent purity: >95 by TLC
amount pure product: 4.7 g
percent of crude: 43%
total time: 3.5 hours

Discussion

Once the two early impurity bands had eluted, the mobile phase was strengthened to move the product away from the trailing impurities. A final, slightly stronger solvent composition was applied once the product band was fully separated from the trailing bands. The unique ability to stack multiple VersaPak cartridges provided the versatility to devise the optimal length (225 mm) for this separation.

VersaFlash and VersaPak are registered trademarks of Sigma-Aldrich Co.