YMC SEPARATION TECHNOLOGY

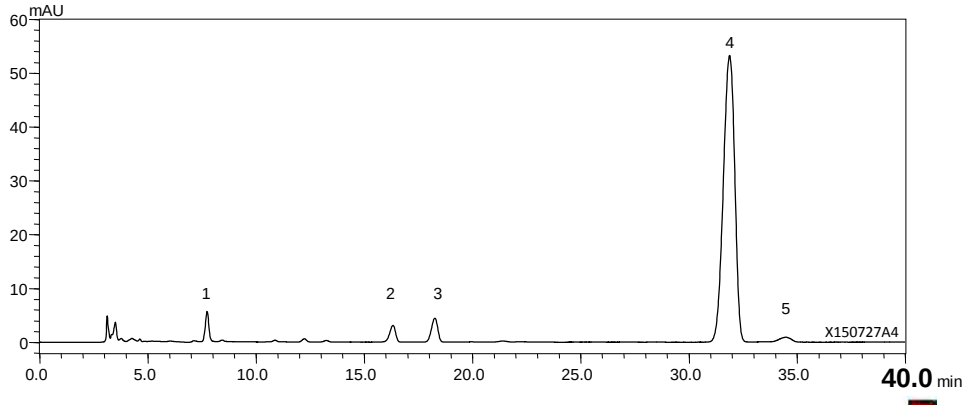
HPLC DATA SHEET

Method Development for Rapid SFC Analysis of Vitamin D_3 and Related Compounds

T150727AE

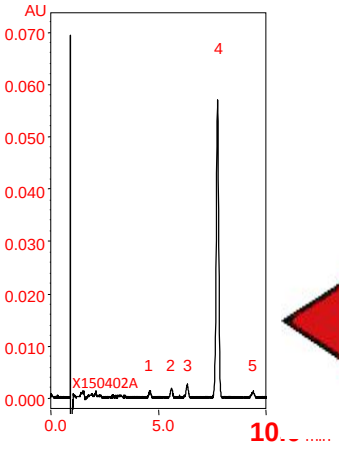
By using the advantages of Supercritical Fluid Chromatography (SFC), namely high permeability and diffusibility, generally higher resolution can be achieved by SFC analysis with shorter run times than by HPLC. Base-line separation of Vitamin D_3 (cholecalciferol), three related compounds (pre-cholecalciferol, 5,6-*trans*-cholecalciferol, tachysterol$_3$) and the antioxidant (α-tocopherol) is achieved using an Alcyon SFC Triart Diol column under SFC conditions with shorter analysis time, which is one quarter the time for the normal phase HPLC method.
Alcyon SFC Triart Diol is based on organic/inorganic hybrid particles and has excellent chemical stability.

Normal Phase HPLC Method



Conditions	
Column:	YMC-Pack SIL (3 µm, 12 nm) 250 x 4.6 mm ID
Part No.:	SL12S03-2546WT
Eluent:	n-hexane/1-pentanol (1000/6)
Flow rate:	1.0 mL/min
Temperature:	25 ℃
Detection:	UV at 254 nm
Injection:	50 µL

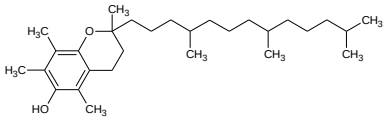
New SFC Method



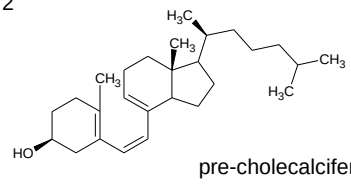
-75%!

Conditions	
Column:	Alcyon SFC Triart Diol (3 µm, 12 nm) 250 x 4.6 mm ID
Part No.:	TDN12S03-2546WTS
Eluent:	CO_2/ ethanol (96/4)
Flow rate:	3.0 mL/min
Temperature:	40 ℃
Detection:	UV at 254 nm
Back pressure:	13.8MPa (2000 psi)
Injection:	20 µL

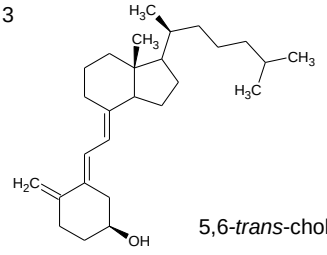
1 dl-α-tocopherol



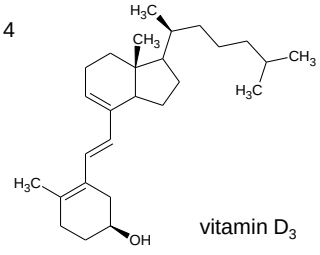
2 pre-cholecalciferol



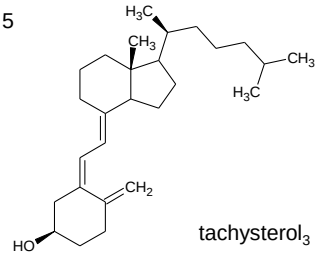
3 5,6-*trans*-cholecalciferol



4 vitamin D_3



5 tachysterol$_3$



Sample: Concentrated cholecalciferol powder, thermally stressed (25 µg/mL)
* Supplied by DSM Nutritional Products
Intentionally prepared to generate the cholecalciferol related compounds, Not commercially available.