



Waters Application Library

Identification of Minor Impurities in
Pharmaceutical Compounds

LC/MS Application Library

Compound: Trimethoprim
Type: Antibacterial Agent
Matrix:
Secondary Matrix:

Conditions:

Column / Capillary: Waters Symmetry C8
Column / Capillary Dimensions: 150 X 2.1 mm
Column / Capillary Part Number:
Flow Rate / Voltage: 0.25 ml/min
Temperature: Ambient
Injection Volume / Type:
Injection Conditions:
Sample Concentration:
Sample Preparation:
Run Time:

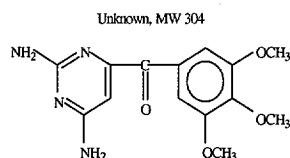
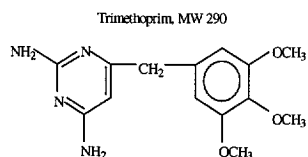
Mobile Phase / Electrolyte: 80% Water 20% Acetonitrile
Gradient Conditions: None
Detection (Primary):
Detection (Secondary):

Instrumentation /System:

2690/996/TMD

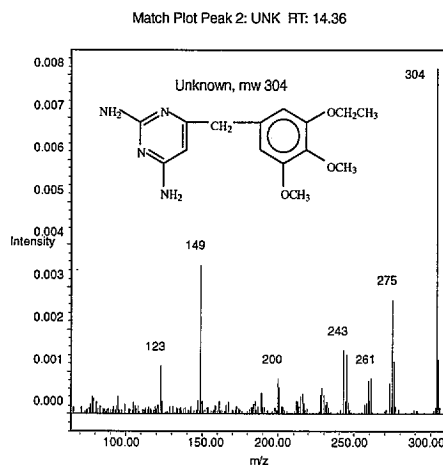
Chromatogram / Electropherogram:

Possible Impurity



Possible oxidation product of Trimethoprim
More polar than Trimethoprim

Trimethoprim Impurity



- Less polar than Trimethoprim
- Logical loss of m/z 29 - (CH_2CH_3)
- Ethoxy-dimethoxy benzaldehyde is a known impurity in the synthesis of Trimethoprim

Objectives:

Impurity identification is becoming increasingly significant in regulated environments, and detection and identification of impurities at the ~0.1% level is critical. Since compound identification based solely on retention time is not absolute, the dual detection capabilities of the Integrity System is extremely useful for the absolute identification of impurities in pharmaceutical materials. Low dispersion chromatography is also becoming more and more desirable in the pharmaceutical industry.

Details:

The overlay of the TIC from the mass detector with the UV chromatogram @ 287 nm is shown. The major peak was Trimethoprim and the goal was to identify the impurity at approximately 14 min. Note there is almost no loss of chromatographic resolution when going from the PDA detector through the ThermaBeam interface. The UV spectrum of Trimethoprim was added to a user built library of UV spectra. The overlay of the UV spectrum from the impurity and the library spectrum of Trimethoprim is shown. While the match is not a good match, we can clearly see that the spectra are related and, therefore, the impurity is probably a related compound. Positive compound identification of minor impurities is easily achieved using the Integrity LC/MS System. The chromatographic information for Trimethoprim indicates that the impurity must be less polar than Trimethoprim; the PDA information shows similarity in structure; the MS spectrum of the impurity confirms its structure. Simultaneous acquisition of PDA and MS data on a single injection and seamless processing and reporting of both channels of data as well as automated library search capabilities offer confidence in peak identification.

Ordering Information:

Part Number	Description	Quantity