



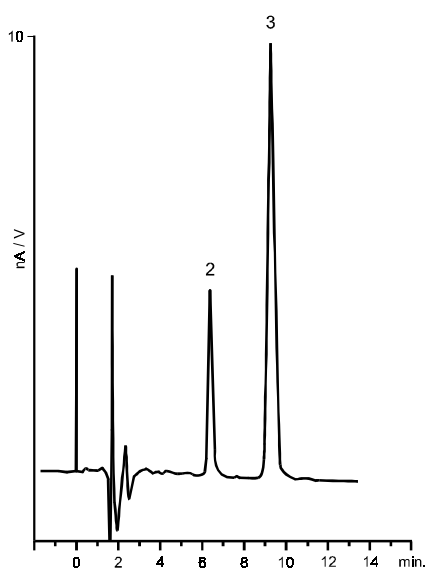
Separation of Morphine and Metabolites Extracted from Blood Plasma

Application
Pharmaceutical
Robert Ricker

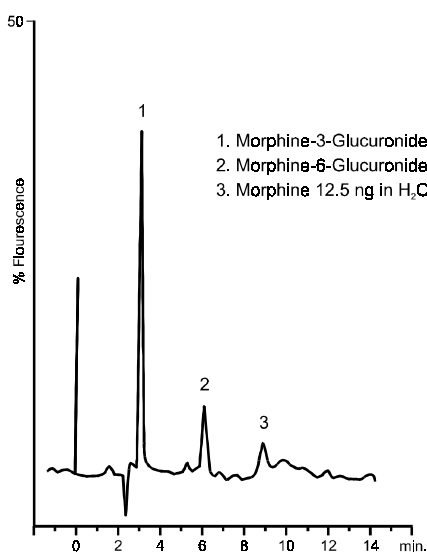
Morphine is an important alkaloid component of opium that acts as an analgesic and narcotic. Detection and quantitation of morphine and its metabolites in blood is important for both clinical and forensic applications.

Highlights

- Good resolution and peak shape for morphine and two metabolites using ZORBAX SB-C18 at moderate pH.
- Electrochemical detection sensitively detects morphine and the metabolite morphine-6-glucuronide; while fluorescence allows sensitive detection of morphine-3-glucuronide.



A) electrochemical detect. (BAS Unijet)
720 mV



B) fluorescence detect. (Waters 470)
exc. = 285 nm, em. = 352 nm

Courtesy of J. Visser, Center for Pharmacy, University of Groningen, The Netherlands

Conditions:
ZORBAX SB-C18 (3.5µm; 4.6 x 150 mm) (Agilent P/N: 863953-902)
Mobile Phase: (97 : 3) 70mM KH₂PO₄ + 1mM EDTA : ACN, pH 4.5
Injection volume 50µL, 1.5 mL/min



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SAMPLE PREPARATION

1. Blood plasma (0.5 mL) is mixed with 0.1 ml internal standard, 0.5 mL H₂O, 1 ml of 0.2M borate buffer pH 9, and 0.2 mL of 0.1M pentane-1-sulphonate. After each addition, the mix should be vortexed. The pentane-1-sulphonate solution must be prepared fresh weekly.
2. A solid phase C18 cartridge is activated with 3 x 1 mL MeOH followed by 3 x 1 mL H₂O. The mixture above is then added to the cartridge and flushed with 3 x 1 mL H₂O. Elution follows with 1 mL MeOH. This fraction is dried under nitrogen at 60° C, dissolved in 200µL of solvent, and vortexed. 50µL is injected onto the column.

NOTE: For Investigational / Research only.
The performance characteristic for this procedure has not been established. Not for *in vitro* diagnostic procedures.

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