

SELECTIVE ANALYSIS OF QUININE AND QUINIDINE IN SERUM/PLASMA BY FAST HPLC

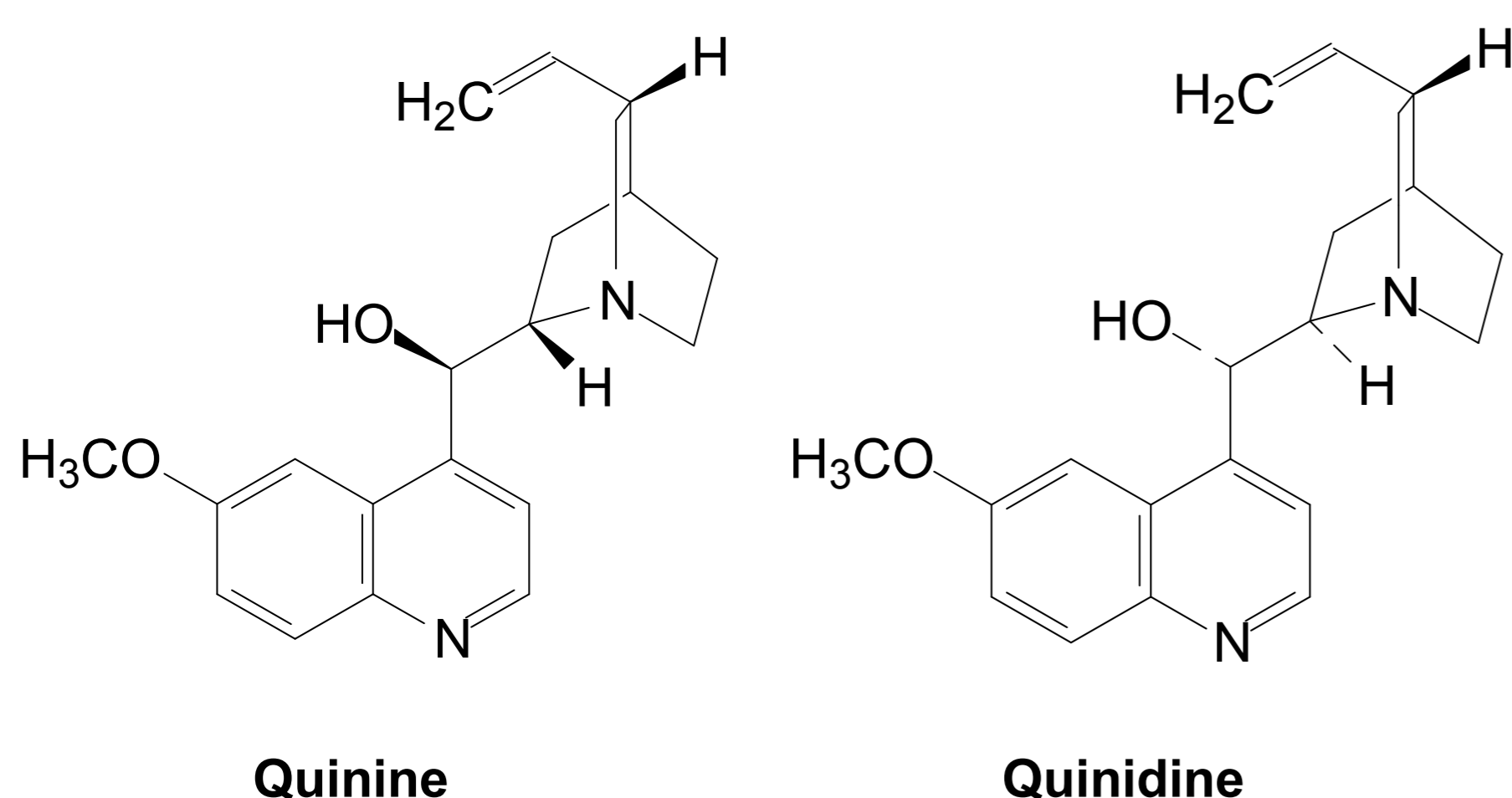
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Introduction

The diastereoisomers quinine and quinidine are used in the treatment of *Plasmodium falciparum* infection, as well as nocturnal cramp (quinine) and cardiac arrhythmia (quinidine). The ability to distinguish between these compounds may be important in cases of suspected poisoning, and is usually achieved using reversed-phase HPLC¹. However, poor peak shapes and complex extraction procedures are typical. Using strong cation-exchange (SCX)-modified 5 µm packings with non-aqueous eluents gives good peak shapes, and allows direct injection of sample extracts². Traditional 'analytical' SCX columns (150 x 4.6 mm i.d.) cannot differentiate between quinine and quinidine under these conditions³. However, using a 'fast LC' column (100 x 2.1 mm i.d.) packed with 5 µm SCX particles and low dead-volume LC equipment allows resolution of quinine and quinidine.

Figure 1. Chemical structures of quinine and quinidine.



Method

Sample preparation:

- 100 µL serum/plasma
- 25 µL internal standard (aqueous imipramine, 25 mg/L)
- 100 µL Tris buffer (2 mol/L, pH 10.6)
- 125 µL methyl *tert*-butyl ether
- Vortex 20 s, centrifuge (4 min, 10,000 rpm, Hettich Mikro 200)
- Inject 50 µL of extract (upper layer)

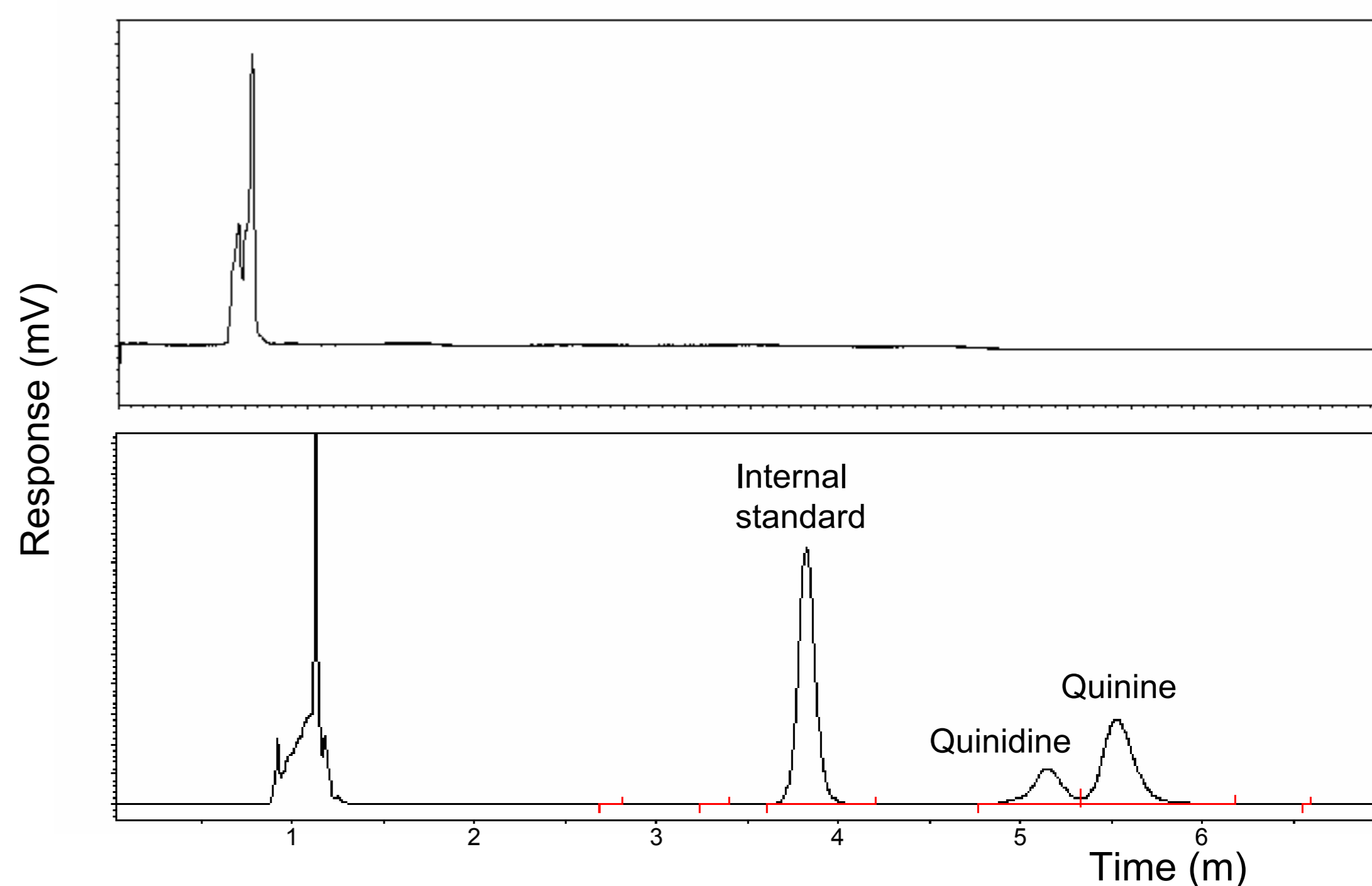
HPLC:

- 100 x 2.1 mm i.d. Waters Spherisorb S5SCX column (35 °C)
- 50 mmol/L ammonium perchlorate in methanol/water (95+5 v/v), pH* 6.7, flow rate 0.5 mL/min
- Detection: UV, 250 nm

Results

- Simple sample preparation and fast analysis (< 6 min).
- Calibration standards (prepared in newborn calf serum) were linear over the range 2–24 mg/L for quinine, and 0.5–8 mg/L for quinidine.
- Analysis of internal QC solutions, prepared in pooled human serum (5 and 15 mg/L quinine, 2 and 5 mg/L for quinidine) gave: intra-assay CV < 2 % (n = 5 each concentration), inter-assay CV < 4 % (n = 6 each concentration).
- Limit of detection 0.2 mg/L both analytes.
- No significant interferences were noted.
- The method is suitable for monitoring plasma/serum concentrations during therapy, and in cases of acute poisoning.
- Following ingestion of 28 x 200 mg tablets of quinine sulfate by a female patient (43 years old), measured plasma quinine concentrations were 8.4, 5.7, and 4.4 mg/L at 7.5, 12.5, and 22.5 h post ingestion, respectively.

Figure 2. Chromatograms following extraction of blank serum (top), and a serum calibration standard (bottom) containing quinidine (3 mg/L) and quinine (8 mg/L).



Conclusion

The assay developed is selective, rapid, reproducible and robust, requires little specialist equipment, and is ideal for TDM of quinine or quinidine, and for use in the diagnosis of acute poisoning.

Advances in column technology can produce faster analyses with enhanced sensitivity/selectivity. A smaller size SCX packing should enable even greater gains in these areas.

References

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3. Croes, McCarthy, Flanagan. Simple and rapid HPLC of quinine, hydroxychloroquine, chloroquine, and desethylchloroquine in serum, whole blood, and filter paper-adsorbed dry blood. *J Anal Toxicol* 1994; 18:255-260