LC/MS Method for the Determination of Stable Isotope Labeled Promethazine in Human Plasma

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Purpose: Promethazine (PMZ) is taken by astronauts orally (PO), intramuscularly (IM) or rectally (PR) for space motion sickness. We developed an LC/MS method with off-line solid phase extraction to measure plasma concentrations of PMZ given as stable isotope-labeled (SIL) formulations by the three different routes of administration simultaneously.

Method: Samples (0.5ml) were loaded on to Waters Oasis HLB co-polymer cartridges and eluted with 1.0 mL methanol. HPLC separation of the eluted sample was performed using an Agilent Zorbax SB-CN column (50 x 2.1 mm) at a flow rate of 0.2 mL/min for 6 min. Acetonitrile/ ammonium acetate (30 mM) in water (3:2, v/v), pH 5.6 ± 0.1, was used as the mobile phase for separation. Concentrations of PMZ, PMZ-d4 and PMZ-d7 and chlorpromazine (internal standard) were determined using a Micromass ZMD single quadrupole mass spectrometer with Electrospray Ionization (ESI). ESI mass spectra were acquired in positive ion mode with selected ion monitoring of $[M+H]^+$.

Results: The method is rapid, reproducible and the assay specific parameters are listed in the following table.

	Linear Range	**LOQ**	**CV (%)**	
Analyte	(ng/ml)	(ng/ml)	Inter-day	Intra-day
Promethazine-d0	1-100	1.00	< 5.4	< 4.5
Promethazine-d4	1-100	1.00	< 6.6	< 6.8
Promethazine-d7	0.5-100	0.50	< 6.5	< 7.5

Conclusion: We report a novel, sensitive and specific method for the measurement of PMZ and SIL PMZ in human plasma.