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## Quantification of water soluble vitamins in natural health products and dietary supplements using Liquid Chromatography: Mass Spectrometry

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A liquid chromatography triplex tandem mass spectrometry (LC-MS/MS) method has been developed for the quantitative determination of water-soluble vitamins in a dietary supplement and dietary dosage forms (tablets, softgels and powders). The water-soluble vitamins include thiamine, riboflavin, niacinamide, niacin, pantothenate, pyridoxine, folic acid, biotin, methylcobalamin and cyanocobalamin. This method consisted of initial extraction with methanol or methanol: water (50:50 v/v) containing 10 mM ammonium formate and 0.1% butylated hydroxyl toluene in an ultrasonic water bath for 30 min depends on the sample matrixes. This method involves the following simple pre-treatment procedures, centrifugation and filtration after an extraction step, whereas extract samples were diluted prior to injection. Chromatography separation was performed on a Waters Acquity BEH C18, 2.1×100 mm, 1.7 μm column. Mobile phases consist of (A) 0.1% formic acid, 5 mM ammonium formate in and (B) methanol. Mobile phase flow was 0.35 mL/min at 40 °C with a run time of 5 min. The amount of compounds present is determined with a calibration curve consisting of sample extracts from a dietary supplement that were spiked at three different level including 75%, 100% and 125% of the label claim. The spike recovery (n=3) for the three levels ranged from 70-120 (RSD 11.5-20.5%). The effect of the matrix on the ionization process in ESI was evaluated by analyzing the extracted sample, spiked extract sample and standard solution at the same concentration and the absolute matrix effect was from 80-120%. The method was applied to identify and quantify the vitamins in commercial natural health products and dietary supplement.

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