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Implementation of Blood Collection Using the Tasso Microsampling Device to Measure Thyroid Hormone Biomarkers

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Introduction

Collection of samples for biomarker analysis is typically conducted by a trained phlebotomist at a clinic or hospital and requires a painful blood draw and several milliliters of blood. The large volume of blood required makes pediatric blood sampling difficult and the travel requirement for the collection can be burdensome for the elderly or critically ill. The Tasso-M20 device enables at-home, automated, self-collection of sample aliquots. The device automatically collects four 17.5 microliter samples in minutes with only a few easy steps and very little pain or blood loss. The accurate quantitation of two thyroid biomarkers extracted from the Tasso-M20 device and analyzed using HPLC-MS/MS was performed in this study. Thyroid hormones Thyroxine (T4) and 3,3',5-triidothyronine (T3) are regulators of metabolism, growth, and development. The levels of T3 and T4 are useful biomarkers of overall thyroid function. Hyperthyroidism or hypothyroidism can result from autoimmune disorders, such as Graves' disease, certain medications, thyroid cancer, and can often occur during pregnancy. Individuals suffering from thyroid dysfunction suffer from a broad range of symptoms including weakness and fatigue.

Methods

A method was developed to extract T3/T4 from blood collected using a Tasso device followed by HPLC-MS/MS analysis. For standard preparation T4 and T3 were spiked into a surrogate matrix and 17.5 microliters of this solution was pipetted onto exposed blank Tasso-M20 tips. The tips were dried at ambient and placed into a DWP 96 and spiked with stable label internal standards and 500 microliters of methanol containing 1% formic acid. After 60 minutes of shaking, 400 microliters of the extract was transferred, evaporated to dryness, and reconstituted with 100 microliters of 80:20 Methanol:H2O containing 1% formic acid. The extract was analyzed on a SCIEX triple quad mass spectrometer. The binary HPLC method utilized an Agilent Pursuit Diphenyl column and Water/Acetonitrile mobile phases containing formic acid.

Results

TASSO samples were collected from 17 individual donors. Sixteen of the donors had no diagnosed thyroid dysfunction and one donor was diagnosed with hyperthyroidism prior to testing. The samples were analyzed in duplicate and T3/T4 was quantified from an eight point calibration curve (0.100-500 ng/mL dynamic range) prepared in surrogate matrix and analyzed in duplicate. The data collected from the assumed 16 healthy

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volunteers had an average T3 value of 0.380 ng/mL and an average T4 value of 24.5 ng/mL. The subject with diagnosed hyperthyroidism was found to have a T3 value of 1.28 ng/mL (over 3X the pool average) and a T4 value of 237 ng/mL (over 9X the pool average). This data suggests that the TASSO collection device coupled with HPLC-MS/MS analysis can be used as a tool to help diagnose T3/T4 thyroid conditions.