IGF-1 in blood using microsampling for monitoring patients with acromegaly

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Introduction
Acromegaly is a condition which results in the development of abnormal soft tissue and skeletal growth2. Prevalence of Acromegaly is 60 per million and there’s a high mortality rate from sufferers2. Acromegaly is caused by abnormally high serum levels of growth hormone (GH) due to oversecretion by a tumour of the pituitary gland5. When GH binds to GH receptors in liver and cartilage, Insulin-like Growth Factor-1 (IGF-1) is secreted. IGF-1 promotes cell proliferation, which causes abnormal bone muscle growth and apoptosis inhibition - resulting in acromegaly. Transsphenoidal surgery is the recommended primary therapy with biomarker post-operatively to help ascertain cure of the disease3. IGF-1 is usually measured in serum to diagnose and postoperative monitoring of the disease1. IGF-1 secretion is more continuous than GH and it’s usually measured in serum or plasma by immunoassay or mass spectrometry in specialist labs. Immunoassay is simpler and faster but it is important to use an antibody with a high degree of specificity.

Currently, patients attend clinics to have their IGF-1 levels checked however, a far more convenient and inexpensive alternative would be to sample blood at home and post the sample to the lab for testing. Volumetric Absorptive Microsampling (VAMS™) offers such a solution. The Mitra sampler using VAMS technology rapidly absorbs an accurately fixed volume of blood (10 µL or 20 µL) from a finger-prick. IGF-1 was measured using IDS-iSYS immunoassay analyser. The original whole blood was centrifuged and the plasma was measured directly on the analyser to compare the result with VAMS extracted whole blood.

Materials and Methods
Comparison of IGF-1 concentration in plasma and whole blood extracted with 10 µL Mitra tips
20 whole blood (EDTA) samples of IGF-1 concentration ranging from 50.8 to 515.4 ng/mL were measured to compare the IGF-1 results of the plasma and the whole blood extracted using VAMS. VAMS was used to sample the whole blood and air dried at room temperature for 2 hours. 2 Mitra tips were pooled into micro container filled with 150 µL of distilled water and rotated on a rotating chamber for 1 hour. IGF-1 was measured using IDS-IYS immunoassay analyser. The original whole blood samples were centrifuged and the plasma was measured directly on the analyser to compare the result with VAMS extracted whole blood.

Comparison of IGF-1 concentration of serum and whole blood extracted with 10 µL Mitra tips
10 paired whole blood (EDTA) and serum samples of IGF-1 concentration ranging from 96.9 to 515.4 ng/mL were measured to compare the IGF-1 results of the whole blood extracted using VAMS. Mitra tips were used to sample the whole blood and air dried at room temperature for 2 hours. 2 Mitra tips were pooled into micro container filled with 150 µL of distilled water and rotated on the rotating chamber for 1 hour. IGF-1 was measured using IDS-IYS immunoassay analyser.

Stability experiment
16 VAMS sampled mid-level of IGF-1 of both serum and plasma of the same patient (serum IGF-1 of 160 ng/mL). Another 16 VAMS sampled high-level of IGF-1 of both serum and plasma of the same patient (serum IGF-1 of 577.5 ng/mL). The samples were dried in room temperature and extracted after 2 hr, 24 hr, 48 hr and 96 hr. The samples were measured to compare the IGF-1 results of the plasma and the whole blood extracted from Mitra con- trasted very well with both wet plasma and serum samples. However, new reference ranges would be needed to be created due to the volumetric presence of the haemato- crit. Furthermore, it was noted that the very low level concentrations of IGF-1 were too dilute from Mitra and so efforts would be needed to reduce the amount of dilution or modifications would be needed to be done to the instrument to boost sensitivity. Never- theless, the results show that as a convenient at home monitoring option, Mitra microsam- pling devices demonstrate great promise.

Conclusion
The results shown in this investigation give very encouraging results that Mitra can be used for analysis of biomarkers on immunoassay instruments. Moreover, for IGF-1, dried whole blood extracts from Mitra cor- related very well with both wet plasma and serum samples. However, new reference ranges would be needed to be created due to the volumetric presence of the haemato- crit. Furthermore, it was noted that the very low level concentrations of IGF-1 were too dilute from Mitra and so efforts would be needed to reduce the amount of dilution or modifications would be needed to be done to the instrument to boost sensitivity. Never- theless, the results show that as a convenient at home monitoring option, Mitra microsam- pling devices demonstrate great promise.

Results and Discussion
Results from both the serum and plasma experiments demonstrated that IGF-1 was detected from reconstituted blood from the Mitra tips. The Mitra tips contained very well when compared with both the plasma (R² = 0.9524) and serum (R² = 0.9893) extractions. Due to the fact that only 20 µL blood was taken per Mitra experiment allowed to dry and recon- stituted in 150 µL, a dilution factor of 7.5 was applied to the data (Figures 3 and 4). Moreover, the data showed a positive bias for the serum (y = 0.7332) and plasma (y = 0.762) to the volumetric presence of the haemato- crit. Furthermore, it was noted that the very low level concentrations of IGF-1 were too dilute from Mitra and so efforts would be needed to reduce the amount of dilution or modifications would be needed to be done to the instrument to boost sensitivity. Nevertheless, the results show that as a convenient at home monitoring option, Mitra microsam- pling devices demonstrate great promise.

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Results from both the serum and plasma experiments demonstrated that IGF-1 was detected from reconstituted blood from the Mitra tips. The Mitra tips contained very well when compared with both the plasma (R² = 0.9524) and serum (R² = 0.9893) extractions. Due to the fact that only 20 µL blood was taken per Mitra experiment allowed to dry and recon- stituted in 150 µL, a dilution factor of 7.5 was applied to the data (Figures 3 and 4). Moreover, the data showed a positive bias for the serum (y = 0.7332) and plasma (y = 0.762) to the volumetric presence of the haemato- crit. Furthermore, it was noted that the very low level concentrations of IGF-1 were too dilute from Mitra and so efforts would be needed to reduce the amount of dilution or modifications would be needed to be done to the instrument to boost sensitivity. Nevertheless, the results show that as a convenient at home monitoring option, Mitra microsam- pling devices demonstrate great promise.

References