VITAMIN D AND A CAN BE SUCCESSFULLY MEASURED BY LC-MS/MS IN CORD BLOOD DILUTED PLASMA, COMPARED TO SERUM

A Albarhani1,2, F Collier3, R Geaves1,4, A-L Ponsonby5,5, K Allen4,5, P Vuillermin3,6, P Roche1, M Clarke2, BIS Steering Committee

1School of Medical Sciences, RMIT University, Victoria, Australia; 2Security Forces Hospital, Dammam, Saudi Arabia; 3Deakin University, Victoria, Australia; 4Murdoch Children’s Research Institute, Melbourne, Australia; 5Department of Paediatrics, University of Melbourne, Victoria, Australia; 6Child Health Research Unit, Barwon Health, Victoria Australia; 7University of Western Australia, Western Australia, Australia

Introduction

In widely used protocols for the collection and isolation of cord blood mononuclear cells, investigators are left with substantial volumes of diluted plasma which could be used for other measurements. The aim of this study was to ascertain the validity of umbilical cord blood (UCB) diluted plasma samples for vitamin D, A and E analysis compared to UCB serum samples.

Method

Twenty UCB matched samples of diluted plasma and serum were collected. The samples were analysed by two liquid chromatography-tandem mass spectrometry (LC-MS/MS) methods (labs A and B) on two separate occasions.

Results

The results of 25(OH)D3 obtained by the two laboratories demonstrated close agreement with a mean difference of 0.14 nmol/L [95% confidence interval (95% CI): -0.8 to 7.1]. Both methods demonstrate close agreement for 25(OH)D3 in UCB serum versus diluted UCB plasma; mean differences 2.2 nmol/L [95% CI: -9.5 to 13.9] and 4.1 nmol/L [95% CI: -14.5 to 6.1] for the results from lab A and lab B, respectively. Vitamin A was quantified by lab A in UCB serum and diluted UCB plasma; mean difference between the results was -0.07 μmol/L [95% CI: -0.41 to 0.28]. Results of 25(OH)D3 epimer and vitamin E in the diluted UCB plasma were below the limit of quantification, and could not be compared with UCB serum.

Conclusion

Diluted UCB plasma can be used for the quantification of retinol and 25(OH)D3 by LC-MS/MS. By contrast, measurement of 25(OH)D3 epimer and vitamin E in diluted UCB plasma is not supported by this study due to limitations in analytical sensitivity for quantification.

Acknowledgement

• This work is supported by NHMRC Grant ID 1029927.
• The Barwon Infant Study Steering Committee consists of Katie Allen, David Burgner, John Carlin, Terry Deyer, Anne-Louise Ponsonby, Sarah Ranganathan, Richard Saffery, Mimi Tang and Peter Vuillermin. Anne-Louise Ponsonby held an NHMRC Senior Research Fellowship.
• The work performed at RMIT University was conducted in the RMIT-Agilent Clinical Biochemistry Mass Spectrometry collaboration laboratory.

Box plots show UCB serum versus diluted UCB plasma results of 25(OH)D3 obtained by Labs A (p=0.032) and B (p=0.205); and vitamin A measured by lab A (p=0.224). Method LoQ is 3.5 nmol/L (lab A) and 2.0 nmol/L (lab B) for both 25(OH)D3 and 0.16 μmol/L (lab A).