

Appendix Q Supplementary blood analytes

Sonja Nicholson, Lorna Cox, Polly Page, Chris Bates and Ann Prentice

Q.1 Introduction

This appendix presents descriptive statistics for a number of additional blood analytes, considered of less public health significance, than those reported in Chapter 6 (see Table 1 below):

Table 1: Blood analytes presented in this appendix

Appendix section	Blood analyte
Q.2	Haematocrit
Q.3	Serum high sensitivity C-reactive protein
Q.4	Plasma soluble transferrin receptors
Q.5	Plasma pyridoxic acid
Q.6	Plasma total homocysteine
Q.7	Plasma retinyl palmitate
Q.8	Plasma γ -tocopherol
Q.9	Serum triglycerides

Details of the blood protocols and methodology, including priority order for analysis, quality control measures, weighting the data, treatment of results below the limit of detection and limitations of the data presented are included in other chapters and appendices (see Chapters 2 and 6; Appendices B and N to P). Notional values were assigned to results below the limit of detection. These were calculated by dividing the analytical limit of detection by the square root of two. This method is consistent with that used in NHANES and has been described by Hornung and Reed (1990).¹ Results are presented for the age groups 1.5 to 3 years, 4 to 10 years, 11 to 18 years, 19 to 64 years and 65 years and over and are split by sex, except data for the age group 1.5 to 3 years which are presented as combined sex only. The values at the upper and lower 2.5th percentiles have been provided in Table Q.1 for each age/sex group. It should be noted that cell sizes for children aged 1.5 to 3 years are quite small and this should be borne in mind when interpreting the data for this age group. Where there are threshold concentrations for an analyte the percentage of participants above or below the threshold (ie where a low status is indicated) has been provided in Table Q.1.

Q.2 Haematocrit (packed cell volume – PCV) (*litres/litre fractional volume, L/L*)

Haematocrit is the proportion of the blood volume taken up by the red cells and is determined by the cell size and number. A low haematocrit may

indicate abnormal cell development, as shown by abnormally small red blood cells (microcytosis) as occurs in iron-deficiency anaemia. Haematocrit for men aged 16 years and over is usually between 0.40L/L and 0.50L/L, whilst for women aged 16 years and over it is usually between 0.36L/L and 0.46L/L.² WHO lower limits for haematocrit levels below which anaemia is indicated are 0.33L/L for children aged 1.5 to 4 years, 0.34L/L for children aged 5 to 11 years, 0.36L/L for children aged 12 to 14 years and non-pregnant women aged 15 years and over, and 0.39L/L for men aged 15 years and over.³

Table Q.1 presents descriptive statistics for haematocrit and Table 6.1 in Chapter 6 presents descriptive statistics for haemoglobin concentration and plasma ferritin, which are key measures of iron status.

(Table Q.1)

Q.3 Serum high sensitivity C-reactive protein (*milligrams/litre, mg/L*)

High sensitivity C-reactive protein (Hs-CRP) is CRP measured by a high-sensitivity assay. CRP is an acute phase protein, the serum levels of which rise during the initial stages of a general, non-specific response to infections and non-infectious inflammatory processes such as rheumatoid arthritis, and also as a result of cardiovascular disease and peripheral vascular disease.⁴ The presence of an acute phase reaction can confound the interpretation of some markers of nutritional status, eg ferritin and retinol. The NDNS RP is the first time Hs-CRP has been measured in NDNS. Alpha-1-antichymotrypsin was used in previous NDNS as the acute phase marker. Reference ranges for Hs-CRP used in clinical practice, eg for stratification of cardiovascular risk, are not appropriate for identification of the subclinical acute phase for nutritional biomarker assessment. A threshold of 5mg/L can be used as an indication of the presence of an acute phase reaction.⁵ Notional values were assigned to results below the limit of detection (see Introduction).¹

Table Q.1 presents descriptive statistics for CRP.

(Table Q.1)

Q.4 Plasma soluble transferrin receptors (*micrograms/millilitre, µg/mL*)

The transferrin receptor is a transmembrane protein present in all cells throughout the body which facilitates the uptake and internal transport of iron bound to circulating transferrin especially during the production of erythrocytes (red blood cells) in the bone marrow. The cellular content of transferrin receptor varies with the iron needs of the tissue and measurements of soluble transferrin receptor (sTfR) in plasma provide a useful clinical index of tissue iron deficiency. The concentration of sTfR circulating in the blood is elevated when there is increased demand for iron in the tissues. This may be

attributable to low intracellular iron concentrations or to physiological factors such as growth or increased haematopoiesis. An advantage of measuring sTfR is that the concentration varies little with age, sex, or pregnancy and unlike ferritin the concentration of sTfR is not affected by the acute phase reaction.⁶ Its measurement facilitates the discrimination between iron deficiency anaemia and anaemia caused by chronic illness or inflammation. Because antibodies used in kits measuring sTfR differ between manufacturers, there is currently no internationally recognised reference range for sTfR concentration.

Table Q.1 presents descriptive statistics for sTfR and Table 6.1 in Chapter 6 presents descriptive statistics for haemoglobin concentration and plasma ferritin which are key measures of iron status. Plasma sTfR concentration was not measured for participants aged 1.5 to 6 years because the small volume of blood taken from these children precluded including these analyses (see section 6.1.1 in Chapter 6), therefore plasma sTfR concentration is provided for children aged 7 to 10 years rather than for children aged 4 to 10 years.

(Table Q.1)

Q.5 Plasma pyridoxic acid (*nanomoles/litre, nmol/L*)

Vitamin B6 comprises pyridoxal, pyridoxine, pyridoxamine and their 5'-phosphates, which are metabolically interconvertible. Pyridoxic acid (PA) is a breakdown product of pyridoxal which is excreted by the kidneys, the clearance of which is dependent on kidney function. Plasma PA is less sensitive to variations in acute phase status but more sensitive to variations in kidney function than plasma PLP.⁷ It should be noted that PA and PLP were not measured in previous NDNS.^{8,9,10,11} Previous NDNS surveys measured erythrocyte aspartate aminotransferase activation coefficient (EAATAC) as an index of vitamin B₆ status. There is currently no internationally recognised normal range for PA concentration.

Table Q.1 presents descriptive statistics for PA and Table 6.2 in Chapter 6 presents descriptive statistics for pyridoxal-5-phosphate (PLP).

(Table Q.1)

Q.6 Plasma total homocysteine (*micromoles/litre, µmol/L*)

Homocysteine is an amino-acid which can be recycled into methionine, a process requiring both folate and vitamin B₁₂. Plasma total homocysteine (tHcy) is therefore sensitive to changes in folate and vitamin B₁₂ status, and, because of a role in folate metabolism, riboflavin status can also influence plasma tHcy. Additionally, because of another, vitamin B₆-dependent, turnover pathway, plasma tHcy can become sensitive to changes in vitamin B₆ status. For these reasons, plasma tHcy is sometimes used as a biomarker of

adequacy of some B vitamins. Previous studies have suggested an association between relatively high plasma tHcy concentrations and increased risk of vascular diseases, although the findings have been inconsistent. Plasma tHcy concentration less than or equal to 12µmol/L is considered normal for adults but a concentration below 10µmol/L is considered optimal.^{12,13}

Data for plasma tHcy are presented in Table Q.1 and data for vitamin B₆ and B₁₂ status are presented in Table 6.2 (Chapter 6). Plasma tHcy concentration was not measured for participants aged 1.5 to 6 years because the small volume of blood taken from these children precluded including these analyses (see section 6.1.1 in Chapter 6), therefore plasma tHcy concentration is provided for children aged 7 to 10 years rather than for children aged 4 to 10 years.

(Table Q.1)

Q.7 Plasma retinyl palmitate (*micromoles/litre, µmol/L*)

Plasma retinyl palmitate can be used as a marker for assessing high use of vitamin A supplements and, if detected, is seen at very low concentrations in plasma. Notional values were assigned to results below the limit of detection (see introduction).¹

Table Q.1 presents descriptive statistics for retinyl palmitate and Table 6.3 in Chapter 6 presents descriptive statistics for retinol.

(Table Q.1)

Q.8 Plasma γ-tocopherol (*micromoles/litre, µmol/L*)

Plasma γ-tocopherol concentration can be used as a measure of vitamin E status. Alpha-tocopherol is the predominant form of vitamin E in human tissues and has the highest biological activity of the tocopherols, with γ-tocopherol accounting for up to 10% of the tocopherol concentration. Increased concentration of plasma lipids appears to cause tocopherols to partition out of cellular membranes, thus increasing plasma concentrations of tocopherols and resulting in a correlation between tocopherols and total lipid in the blood, particularly with the cholesterol fraction. For this reason plasma tocopherols can be usefully expressed as a ratio to plasma total cholesterol (µmol/mmol), enabling comparisons to be made between groups with different plasma lipid levels. Some researchers attribute a different metabolic and health significance to γ-tocopherol from that attributed to α-tocopherol. For adults, a total concentration of plasma tocopherols below 11.6µmol/L, of which approximately 10% would be γ-tocopherol, or a plasma tocopherols to cholesterol ratio of below 2.25µmol/mmol, tends to cause red blood cells to haemolyse after exposure to oxidising agents *in vitro*, which is a

functional test for vitamin E deficiency. This is sometimes considered to be an indicator of biochemical deficiency but is not indicative of a clinical deficiency of vitamin E. There is currently no established normal range for plasma γ -tocopherol concentration. The Committee on Medical Aspects of Food and Nutrition Policy (COMA) Panel on Dietary Reference Values considered a tocopherol to cholesterol ratio of 2.25 μ mol/mmol to be the lowest satisfactory value for adults.¹⁴

Table Q.1 presents descriptive statistics for γ -tocopherol and Table 6.3 in Chapter 6 presents descriptive statistics for α -tocopherol.

(Table Q.1)

Q.9 Serum triglycerides (millimoles/litre, mmol/L)

Triglycerides (triacylglycerols) are composed of three molecules of fatty acids esterified with one molecule of glycerol and are the form in which dietary fatty acids are transported in the bloodstream for storage in adipose tissue and for metabolism by various organs. Fasting serum triglyceride concentration is subject to considerable biological variation within an individual. Triglyceride concentration is included in the Friedewald equation to calculate LDL cholesterol concentration.¹⁵

Table Q.1 presents descriptive statistics for fasted triglyceride concentration for those aged 4 years and over and Table 6.3 in Chapter 6 presents descriptive statistics for total, HDL and LDL-cholesterol.

(Table Q.1)

¹ Hornung, RW, Reed, LD. Applied Occupational and Environmental Hygiene, 1990, 5: 46-51.

² Dacie JV, Lewis SM. Practical Haematology. 9th Edition. Churchill Livingstone (Edinburgh, 2001).

³ World Health Organization. Iron Deficiency Anaemia; Assessment, Prevention, and Control: A guide for programme managers. WHO (Geneva, 2001).

⁴ Scientific Advisory Committee on Nutrition. *Iron and Health*. The Stationery Office (London, 2010).

⁵ Thurnham DI, McCabe GP, Northrop-Clewes CA, Nestel P. Effects of subclinical infection on plasma retinol concentrations and assessment of prevalence of vitamin A deficiency; meta-analysis. *Lancet*, 2003; **362**, 2052-2058

⁶ Beerenhout C, Bekers O, Kooman JP, van der Sande FM, Leunissen KM: A comparison between the soluble transferrin receptor, transferrin saturation and serum ferritin as markers of iron state in hemodialysis patients. *Nephron*, 92: 32– 35, 2002.

⁷ Bates CJ, Pentieva KD, Prentice A, Mansoor MA, Finch S. Plasma pyridoxal phosphate and pyridoxic acid and their relationship to plasma homocysteine in a representative sample of

British men and women aged 65 years and over. *British Journal of Nutrition*, 1999; 81: 191–201.

⁸ Rustin D, Hoare J, Henderson L, Gregory J, Bates CJ, Prentice A, Birch M. National Diet and Nutrition Survey: adults aged 19 to 64 years. Volume 4: Nutritional status (anthropometry and blood analytes), blood pressure and physical activity. TSO (London, 2004).
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(accessed 31/03/14).

⁹ Gregory JR, Collins DL, Davies PSW, Hughes JM, Clarke PC. National Diet and Nutrition Survey: children aged 1 ½ to 4 ½ years. Volume 1: Report of the diet and nutrition survey. HMSO (London, 1995). Gregory J, Lowe S, Bates CJ, Prentice A, Jackson LV, Smithers G, Wenlock, R, Farron M. National Diet and Nutrition Survey: young people aged 4 to 18 years. Volume 1: Report of the diet and nutrition survey. TSO (London, 2000).

¹⁰ Finch S, Doyle W, Lowe C, Bates CJ, Prentice A, Smithers G, Clarke PC. National Diet and Nutrition Survey: people aged 65 years and over. Volume 1: Report of the diet and nutrition survey. TSO (London, 1998).
<http://www.esds.ac.uk/findingData/snDescription.asp?sn=4036> (accessed 31/03/14).

¹¹ Gregory JR, Collins DL, Davies PSW, Hughes JM, Clarke PC. National Diet and Nutrition Survey: children aged 1 ½ to 4 ½ years. Volume 1: Report of the diet and nutrition survey. HMSO (London, 1995).

¹² Bates CJ, Mansoor MA, Gregory J, Pentieva K, Prentice A. Correlates of plasma homocysteine, cysteine and cysteinyl-glycine in respondents in the British National Diet and Nutrition Survey of Young People Aged 4-18 Years, and a comparison with the Survey of People Aged 65 Years and Over. *British Journal of Nutrition*, 2002; 87: 71–79.

¹³ Malinow MR, Bostom AG, Krauss RM. Homocyst(e)ine, diet, and cardiovascular diseases: a statement for healthcare professionals from the Nutrition Committee, American Heart Association. *Circulation*, 1999; 99: 178–182.

¹⁴ Committee on Medical Aspects of Food Policy, *Dietary Reference Values for Food Energy and Nutrients for the United Kingdom*, Department of Health Report on Health (1991).

¹⁵ Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clinical Chemistry* 1972;18: 499-502.