Melbourne Pathology approaches to vitamin B_{12} deficiency, Dr Ken Sikaris

Melbourne Pathology is one of the many Sonic Health Laboratories across the world. Melbourne Pathology has one core and five satellite laboratories. It employs 400 staff in the core laboratory, 400 pathology collectors and 100 couriers. Together they serve the 4 million population of Melbourne, Australia and receive almost 8,000 patient samples each day.

Dr Zhong Lu and Dr Ken Sikaris are the Chemical Pathologists working at Melbourne Pathology. Dr Zhong Lu has a strong interest in clinical nutrition and she has obtained a PhD in nutrition in addition to her qualifications in Medicine and Chemical Pathology. Dr Ken Sikaris has a special interest in analytical quality and is currently Chair of the International Federation of Clinical Chemistry's Committee on Analytical Quality.

Melbourne Pathology have been using Abbott instrumentation for over 20 years and currently have two Architect 2000SR's, one Architect 4000 and one AxSYM performing a wide variety of tests including vitamin B_{12}, hormones and tumour markers.

Vitamin B_{12} is one of the common tests that clinicians request for investigation of possible causes underlying a variety of symptoms that their patients have presented with. The symptoms can be non-specific, such as tiredness and memory loss. Currently, there is no gold standard available for diagnosis of vitamin B_{12} deficiency. Until recently, diagnosis has been predominantly based on direct measurement of total vitamin B_{12} in the blood and the results are methodology-dependent. Different laboratories use different methods which can produce very different results for the same patient. In addition, the results can also be affected by the concentration of the carrier proteins for B_{12} in the blood. These can lead to discordance between the total vitamin B_{12} result and the clinical status of an individual. Both Dr Lu and Dr Sikaris are constantly being called by clinicians about the interpretation of total vitamin B_{12} levels. Clinicians and pathology specialists have long been waiting for an assay that can better assess B_{12} status of an individual.

The new holotranscobalamin ("Active-B12") assay from Abbott Diagnostics (developed and manufactured by Axis-Shield under the AxSYM ***xtra*** programme) may provide the answer for us. Vitamin B_{12} in our blood binds to two proteins: predominantly to haptocorrin (holo-haptocorrin, non-active biologically) with a smaller proportion (<30%) to transcobalamin (holo-transcobalamin, active biologically). Thus by measuring "Active-B12", the variable effect of non-active carrier protein on the total B_{12} concentration is minimised.

Since May 2007 Melbourne Pathology has performed over 25,000 "Active-B12" analyses. The protocol in place offers the test to every patient with a total B_{12} below 200 pmol/L as we are over 95% confident that patients with a total B_{12} greater than 200 pmol/L would not have a low "Active-B12". The test is also offered to any patient regardless of their total B_{12} level if the request was made by a haematologist. From our experience reviewing these cases, "Active-B12" has advantages over total B_{12} in assessing the B_{12} status of an individual. "Active-B12" was helpful in excluding B_{12} deficiency falsely 'identified' by low total B_{12} such as in pregnancy. It

is also helpful in confirming true B_{12} deficiency missed by a normal total B_{12} in patients such as those with iron deficiency and hypothyroidism. An example is illustrated below:

History: A 51 year old lady, vegetarian, was being followed up for persistent iron deficiency.

	22/3/2006	***01/07/2006***	***05/01/2008***	***Unit***	***Reference Intervals***
Serum iron studies					
Iron	7	5	3	umol/L	9 - 31
TRF*	3.8	3.5	3.9	g/L	2.2 – 3.0
TRF Saturation	7	6	3	%	15 – 50
Ferritin	22	15	12	nmol/L	30 – 250
Full blood Examinations					
Hb			96	g/L	115 - 160
Haematocrit			0.31		0.36 – 0.47
RBC			4.6	$x10^{12}$/L	3.8 - 5.4
MCV			67	fL	80 – 100
MCH			21	pg	27 – 32
MCHC			315	g/L	310 – 360
RDW			20.3		11 – 17
Serum vitamin B_{12}					
Total B_{12}			175	pmol/L	132 – 650
Active-B12			12	pmol/L	23 – 100

*TRF: transferrin

Her results show that she is still iron deficient. Although her total B_{12} level appeared normal, the "Active-B12" level was very low. The patient is microcytic (small red blood cells) rather than macrocytic (large red blood cells) because of the iron deficiency, but the RDW (red cell distribution width) shows a very wide variation in red cell sizes. Our experience is that when both iron deficiency and vitamin B_{12} deficiency co-exist in a patient, microcytosis remains more common than macrocytosis until the "Active-B12" level is extremely low. More specifically, when "Active-B12" is below about 8 pmol/L, macrocytosis is more likely than microcytosis.

While we know that iron deficiency and B_{12} deficiency often co-exist in vegetarians, the case illustrates how easy it is to overlook significant vitamin B_{12} deficiency if using only the total B_{12} assay.

"Active-B12" has fulfilled our expectations in being a robust assay that improves the assessment of vitamin B_{12} status. It provides reassurance to us that we have contributed to improved patient care. The routine availability of this assay differentiates our laboratory as a progressive provider of quality pathology.