Editor’s Notes

Dear all, this Spring edition of Pathology Newsletter has a main article written by Dr Chris Jenkins (Consultant Haematologist for ABHB) and colleagues which describes the MGUS monitoring undertaken in the Health Board and includes what we hope are some helpful guidelines and explanations.

There are a few current issues pertaining to the Pathology service which are worthy of note:

1) PTH & Vitamin D

Change of sample type. Most will be aware that there has been a change in sample type for these tests. An additional EDTA (lavender top) tube is now required for these tests. The reason for this change is due to the improved stability that has been demonstrated for these analytes in EDTA in comparison to the previous SST (red/yellow top) tubes that were used.

2) Hypopituitarism

If you have such patients let us know their identity by emailing Dr Dave Hampton at RGH (david.hampton@wales.nhs.uk). Though this is a fairly rare condition there have been occasions when thyroid function results can be misinterpreted due to the lack of awareness that a patient has this condition. If we are notified we can note hypopituitarism for these patients on our computer record so that relevant clinical comments can be added to future reports thus reducing the risk of error.

3) Albumin/creatinine ratio (ACR) also known as microalbumin vs. Protein/creatinine ratio (PCR)

There is always some confusion about what to request where these tests are concerned. Guidelines recommend that PCR (lab code RUPROT) is tested for renal NSF and that ACR (lab code MALB) for Diabetes. Using the lab codes shown would be helpful to our registration staff (particularly RUPROT as PCR can be confused with meaning Polymerase Chain Reaction).

4) Changes to reference ranges

Please note that the reference ranges for many of our tests have changed due to the all Wales harmonisation that is taking place in preparation for the Welsh Clinical Portal and the new laboratory computer systems. Most changes are slight and as always will appear on all reports alongside the results.

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Provision of Andrology Services within Aneurin Bevan Local Health Board

Andrology services for ABHB encompass male fertility screening and post vasectomy semen analysis. Since February 2009 all of the Andrology services are provided by the Department of Microbiology. Now two years have passed, I would like to take the opportunity to bring you all up to date with the service, to inform you of changes to the assessment of the samples and how this will affect you.

The service over the last 12 months for which we have data show a total workload of 891 fertility assessments and 904 post vasectomy semen analyses. 72% of fertility tests and 99% of post vasectomy tests are performed at Nevill Hall Hospital and the balance at Caerphilly and District Miners Hospital.

The service, subject to staffing, is all day Monday to Thursday and Friday morning at NHH, and two Wednesdays a month at CDMH. This is an appointment only service. Please be aware that if a patient arrives without an appointment the sample will not be processed and they will be referred back to their requesting doctor. We expect the majority of patients to use the service at NHH, whilst the CDMH service is primarily for patients based in the Rhymney valley area.

Since we have centralised this service the department is now fully CPA accredited for andrology. Three members of staff, including myself have completed the Association of Biomedical Andrologists portfolio in Basic Semen Analysis.

In 2009 we prepared two user surveys for andrology, one for the GP surgeries and the other for the patient themselves. The aim was to determine if the service we provided met with our users’ expectations. An issue raised by some patients was the distance they had to travel to deliver the specimen and the time taken. In response to this there is now a room available on Monday and Thursday afternoons which patients are able to book, to allow them to produce the specimen onsite at NHH. We also have a direct telephone line for Andrology at NHH, 01873 733062, this should allow patient easier access to book appointments and faster response to calls from clinicians. This should be your first point of contact for enquiries about andrology reports, Dr Neil Carbins or I are happy to answer your questions.

The routine semen analysis includes a general macroscopic appearance, pH, volume, motility, concentration, morphology and vitality. As many of you may be aware the service has been reporting to WHO 1999 guidelines, recently the WHO has released the 2010 guidelines. It is our intention that as of April we will be reporting to the 2010 guidelines. You will notice some changes in the way we reports some of the parameters, and some of the reference ranges may change. Below I will explain each of the parameters and expand on the changes.

The new guidelines are more prescriptive in the way in which tests are performed and are designed to ensure greater levels of accuracy and reproducibility. All results will have a sampling error of ≤ 5% unless otherwise stated.

**Macroscopic appearance**

The overall appearance of the sample, including liquefaction, viscosity, presence of mucus or blood. Odour will also be reported if it is abnormal.

**Volume**

This will now be calculated by the weight of the sample, assuming a semen density of 1g/ml. It is important that only pre-weighed sample containers are used, these are provided as part of the semen collection pack and are the ONLY acceptable containers; any other container will invalidate the analysis. Semen collection packs are available from the testing laboratory. The previous method of measurement was subject to error as significant quantities of the seminal fluid would adhere to the containers and this would result in a low estimation of volume. Patients will also be asked if they have collected the whole sample. As semen is not a homogeneous sample if part is lost it may significantly alter the result.

**Aggregation and agglutination**

Aggregation, the adherence of immotile sperm to each other or motile sperm to mucus strands, non-sperm cells or debris. Agglutination, motile sperm that adhere to each other and may be suggestive of the presence of anti-sperm antibodies.

**Cells**

“Round cells” this refers to leukocytes and immature germ cells, epithelial cells may also be present.

**Motility**

Using WHO 1999, this was reported as 4 grades. Under the WHO 2010 guidelines motility is reported as 3 grades. PR – progressive motility, this incorporates both the rapid progressive and sluggish progressive from WHO 1999. NP – non-progressive motility and IM – immotile, these definitions remain the same.

**Vitality**

This is estimated using a dye exclusion test. It is used to estimate if the non-motile sperm are viable. This is reported as a percentage.

**Sperm concentration**

The methodology for this test has changed, however this will not affect the concentration. The reporting of the result has changed, the lowest concentration to which sperm will routinely be assessed is 2x106/ml unless there is a clinical reason to accurately determine a lower concentration. All concentrations will be reported to 2 significant figures, and a total number of sperm per ejaculate will also be reported.

**Morphology**

This is the assessment of “normal” sperm. The method has not changed, however a reference range now exists (see below).

**Anti-sperm antibodies (ASA)**

This is a new part of the analysis for this laboratory. We will be looking for IgG only, this test will be performed on the first sample submitted by the patient. We will not retest on subsequent sample for a period of two years, unless the patient presents with a clinical history suggestive of ASA e.g. trauma, mumps, genito urinary infection.

This is a summary of the new guidelines, below I have included two tables taken from the WHO 2010 manual. If you require further information on the new guidelines or andrology in general please contact me, David Sanders, David.Sanders@wales.nhs.uk, 01873 733062.reb
Reference Values (5th centiles and their 95% confidence intervals) – World Health Organisation, 2010

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Lower reference limit</th>
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<tbody>
<tr>
<td>Semen volume (ml)</td>
<td>1.5 (1.4-1.7)</td>
</tr>
<tr>
<td>Total sperm number (10^6 per ejaculate)</td>
<td>39 (33-46)</td>
</tr>
<tr>
<td>Sperm concentration (10^6 per ml)</td>
<td>15 (12-16)</td>
</tr>
<tr>
<td>Total motility (PR+NP, %)</td>
<td>40 (38-42)</td>
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<tr>
<td>Progressive motility (PR, %)</td>
<td>32 (31-34)</td>
</tr>
<tr>
<td>Vitality (live sperm, %)</td>
<td>58 (55-63)</td>
</tr>
<tr>
<td>Sperm morphology (normal forms, %)</td>
<td>4 (3.0-4.0)</td>
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<tr>
<td>Other consensus threshold values</td>
<td></td>
</tr>
<tr>
<td>pH</td>
<td>≥7.2</td>
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<tr>
<td>MAR test (motile sperm with bound particles, %)</td>
<td>&lt;50</td>
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</table>

Distribution of values for semen parameters from men whose partners became pregnant within 12 months of discontinuing contraceptive use – World Health Organisation, 2010

<table>
<thead>
<tr>
<th>Parameter (units)</th>
<th>N</th>
<th>2.5</th>
<th>5</th>
<th>10</th>
<th>25</th>
<th>50</th>
<th>75</th>
<th>90</th>
<th>95</th>
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<tbody>
<tr>
<td>Semen volume (ml)</td>
<td>1941</td>
<td>1.2</td>
<td>1.5</td>
<td>2.0</td>
<td>2.7</td>
<td>3.7</td>
<td>4.8</td>
<td>6.0</td>
<td>6.8</td>
<td>7.6</td>
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<tr>
<td>Total sperm number (10^6/ejaculate)</td>
<td>1859</td>
<td>23</td>
<td>39</td>
<td>69</td>
<td>142</td>
<td>255</td>
<td>422</td>
<td>647</td>
<td>802</td>
<td>928</td>
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<tr>
<td>Sperm concentration (10^6/ml)</td>
<td>1859</td>
<td>9</td>
<td>15</td>
<td>22</td>
<td>41</td>
<td>73</td>
<td>116</td>
<td>169</td>
<td>213</td>
<td>259</td>
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<tr>
<td>Total motility (PR=NP, %)</td>
<td>1781</td>
<td>34</td>
<td>40</td>
<td>45</td>
<td>53</td>
<td>61</td>
<td>69</td>
<td>75</td>
<td>78</td>
<td>81</td>
</tr>
<tr>
<td>Progressive motility (PR, %)</td>
<td>1780</td>
<td>28</td>
<td>32</td>
<td>39</td>
<td>47</td>
<td>55</td>
<td>62</td>
<td>69</td>
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<td>75</td>
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<tr>
<td>Non-progressive motility (NP, %)</td>
<td>1778</td>
<td>1</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>5</td>
<td>9</td>
<td>15</td>
<td>18</td>
<td>22</td>
</tr>
<tr>
<td>Immotile (IM, %)</td>
<td>1863</td>
<td>19</td>
<td>22</td>
<td>25</td>
<td>31</td>
<td>39</td>
<td>46</td>
<td>54</td>
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<tr>
<td>Vitality (%)</td>
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<td>58</td>
<td>64</td>
<td>72</td>
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<tr>
<td>Normal form (%)</td>
<td>1851</td>
<td>3</td>
<td>4</td>
<td>5.5</td>
<td>9</td>
<td>15</td>
<td>24.5</td>
<td>36</td>
<td>44</td>
<td>48</td>
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MGUS (Monoclonal Gammopathy of Undetermined Significance)

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Introduction

Plasma cells are a normal constituent of the human bone marrow, and are derived from lymphocyte precursor cells. Their role is to produce antibodies (immunoglobulins) of which there are five classes, namely IgG, A, M, D and E. Antibodies are part of the humoral immune system and identify foreign objects such as bacteria or viruses via their different antigens. A temporary increase in antibody levels is common with infections, and the antibodies coated the pathogens which are then targeted for destruction by macrophages. Similarly, polyclonal increases in immunoglobulins can be the result of inflammation and non-haematological neoplasia and these do not require haematology referral. However there are also a number of disorders of the plasma cells which lead to dysregulation of the antibody production. In certain haematological malignancies clonal plasma cells may proliferate in the bone marrow. These cells will produce a marked increase in an identical antibody molecule, and these can be detected as a discrete band on serum electrophoresis. The conditions that are associated with a paraprotein production include myeloma, Waldenstrom’s macroglobulinaemia, amyloidosis and certain types of non-Hodgkin lymphoma.

What is MGUS?

A far more common cause of paraprotein production in the body is MGUS (Monoclonal Gammopathy of Undetermined Significance). This is a pre-malignant condition characterised by a stable monoclonal paraprotein in the blood or urine (Bence Jones protein) without any of the underlying features of myeloma or Waldenstrom. It is prevalent in the population, and is found in approximately 1% of people over 60 years old, 3% over 70 years and increases to up to 10% in over 90’s. MGUS by definition is an asymptomatic condition with no abnormal physical findings, and it is usually picked up as an incidental finding. A typical presentation may occur when a patient has LFT’s checked for an unrelated condition, and it is noted that the total protein and globulin level are raised. The biochemistry department then
malignancy is suspected from the blood tests then a letter will be sent out to the team suggesting urgent referral to a haematology clinic if it is considered appropriate. If the patient has any evidence of anaemia, hypercalcaemia or renal failure associated with an IgA or G paraprotein then myeloma should be suspected. Other features of concern include bone pain or lytic lesions on an X-ray. Whilst if the patient has evidence of lymphadenopathy or hepatosplenomegaly with an IgM paraprotein then Waldenstroms is a possibility. However if there are none of these features then the patient may have MGUS. MGUS can occur with any of the immunoglobulin classes; however the level of paraprotein is usually lower than that of myeloma or Waldenstroms. There is also usually no immune paresis in MGUS, i.e. the other classes of immunoglobulins are not suppressed.

How to diagnose MGUS

IgG, A or M paraprotein associated with:

- Paraprotein <30g/L
- No clinical symptoms suggestive of myeloma, Waldenstroms or lymphoma
- No anaemia, hypercalcaemia or renal impairment
- No lytic bone lesions
- <10% plasma cells in the bone marrow

When to refer to a haematologist for a suspected haematological cancer

IgG, A or M paraprotein associated with:

- Hypercalcaemia
- Unexplained renal impairment
- Urinary Bence Jones protein
- Bone pain or pathological fractures
- Anaemia or cytopenias
- Hyperviscosity symptoms (headache, visual loss, acute thrombosis)
- Lymphadenopathy or splenomegaly
- Lymphocytosis

What is the relevance?

Patients with MGUS are asymptomatic and in the majority of the people with the condition the paraprotein will remain stable. The patients can be reassured that most of people with MGUS will never need any treatment for the disorder. However there are a small number of patients which will progress over time from MGUS to a haematological malignancy. In approximately 1% of patients per year the paraprotein level will rise suggesting a possible transformation to myeloma or Waldenstroms. These patients may still be asymptomatic, or may have developed new pain, renal failure or hypercalcaemia. Regular surveillance is therefore recommended to monitor for this progression.

MGUS monitoring Programme

Patients with a newly diagnosed paraprotein are often reviewed in haematology clinic to determine if they have a haematological malignancy. However MGUS patients with a paraprotein but with no suspicious features may be offered a place in the MGUS monitoring system. This is a Gwent haematology initiative which allows the indirect monitoring of the patients with MGUS without them having to attend the hospital. Every six months a series of blood forms are sent out to the patients on the system and they are requested to have a series of blood tests and provide a urine sample at the GP practice. The results are then reviewed by a consultant haematologist who will determine if the results are constant or have changed significantly. If the results are stable the patient and GP are informed and a further automatic recall is instigated for six months later. However a significant rise in the paraprotein, or new anaemia, renal failure or hypercalcaemia could suggest a progression of the MGUS and the patient will be offered a new review in the haematology clinic.

Pathology requesting by GP practice.

As many of you will be aware, Pathology has collected data from our computer system which displays the requesting patterns for each practice in Gwent across the repertoire of pathology tests. This information is now available via a link on the Pathology Homepage (http://howis.wales.nhs.uk/sitesplus/866/page/40759) on the Health Board’s Intranet site in the form of a pivot table. There are separate charts for Biochemistry, Haematology and Microbiology. These can be accessed individually by clicking the relevant tab at the bottom of the spreadsheet.

Hovering the cursor over the bars on the chart will reveal the test and the numbers requested.

The display can be manipulated to show data selectively by test, by practice or by locality (or by a combination of these parameters).