CASE STUDY 3:
PSA-STANDARDISATION & VARIABILITY
COMPARATIVE STUDY IN HOSPITALS

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University Hospital Limerick,
IRELAND

JCTLM Meeting
BIPM, Sevres, France
4th December 2013
Prostate Cancer in Ireland

• Most common non-cutaneous cancer in males
• 1 in 11 Irish men diagnosed in their lifetime
• Between 1994 to 2009 – increased incidence in all age groups
  • 55 to 64 years (134 cases to 859)
Prostate Cancer in Ireland

• Incidence is predicted to increase by 140% during 2008-2030
• Incidence is rising and survival is increasing, more men are living longer with PCa
• Over 2,500 men are diagnosed with prostate cancer each year.
• The cumulative risk of a man developing prostate cancer before the age of 50 is 1 in 485 and before the age of 70 is 1 in 13
Each year approximately 20,000 Irish people develop cancer and 7,500 die of the disease.

Recommended that Cancer Centres should be networked together in Managed Cancer Control Networks.

The National Cancer Control Programme (NCCP) was set up to provide a comprehensive programme of cancer control in Ireland, to transform how cancer care is delivered, ensure that cancer services meet the highest standards.

8 Specialist Cancer Centres were set up and networked within each of the four HSE administration regions.

Patients suspected of having PCa are assessed and diagnosed through a single integrated care pathway.
Specialist Cancer Centres

- Each Specialist Cancer Centre must serve a population of at least 500,000
- Rare and complex cancers should be treated by a subset of the eight cancer centres
- Cancer Centres must be well supported
GP Electronic Cancer Referral

• Electronic referral for Breast, Prostate and Lung cancer is available free for GPs using the following ICGP accredited software systems:
  • Socrates, Complete GP, Helix Practice Manager & HealthOne
Prostate cancer is the leading cause of cancer in men (excluding skin cancer). Over 2,500 men are diagnosed with prostate cancer in Ireland each year. The cumulative risk of a man developing prostate cancer before the age of 50 is 1 in 485 and before the age of 70 is 1 in 13.

**Risk Factors:** Family history of prostate cancer, age (risk of prostate cancer increases after 50 years), and men of African ethnicity.

**Prostate Specific Antigen (PSA) Testing**
- PSA testing of asymptomatic men or PSA screening is not national policy
- Prostate assessment consists of a digital rectal examination (DRE) and a PSA test
- PSA testing should only be carried out after full advice and provision of information.
  (Patient information leaflet about prostate assessment is available from the National Cancer Control Programme on (01) 8287100 or can be downloaded by logging onto www.cancercontrol.hse.ie)
- All men with an abnormal DRE should be referred to a urologist regardless of PSA results

**GENERAL RECOMMENDATIONS**
A patient who presents with symptoms or signs suspicious of prostate cancer should be referred for rapid access prostate assessment. Primary healthcare professionals should encourage all men over 50 years of age, or men over 40 who have a first degree relative with prostate cancer or those of African ethnicity to be aware of prostate health issues, in order to minimise delay in presentation of disease.

To make a referral, **FAX** or **POST** a **NATIONAL RAPID ACCESS PROSTATE CLINIC REFERRAL FORM** or submit an electronic prostate cancer referral form via [healthlink.ie](http://healthlink.ie). Electronic referral systems are currently being developed, go to the following website www.healthlink.ie for further updates.

Additional prostate cancer referral forms can be obtained by ringing the National Cancer Control Programme on (01) 8287100 or by logging onto [www.cancercontrol.hse.ie](http://www.cancercontrol.hse.ie).

**NATIONAL RAPID ACCESS PROSTATE CLINICS (please refer to only one clinic)**

<table>
<thead>
<tr>
<th>Clinic</th>
<th>Tel:</th>
<th>Fax:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Beamount Hospital, Dublin 9</td>
<td>(01) 809 3485</td>
<td>(01) 809 3488</td>
</tr>
<tr>
<td>Cork University Hospital</td>
<td>(021) 492 2113</td>
<td>(021) 492 2391</td>
</tr>
<tr>
<td>Galway University Hospital</td>
<td>(091) 542 053</td>
<td>(091) 542 092</td>
</tr>
<tr>
<td>Mid Western Regional Hospital, Limerick</td>
<td>(061) 585 637</td>
<td>(061) 482 572</td>
</tr>
<tr>
<td>Mater Hospital, Dublin 7</td>
<td>(01) 803 2646 / 2295</td>
<td>(01) 803 4036</td>
</tr>
<tr>
<td>St. James's Hospital, Dublin 8</td>
<td>(01) 416 2850</td>
<td>(01) 428 4090</td>
</tr>
<tr>
<td>St. Vincent's University Hospital, Dublin 4</td>
<td>(01) 221 3056</td>
<td>(01) 221 4318</td>
</tr>
<tr>
<td>Waterford Regional Hospital</td>
<td>(051) 842 044</td>
<td>(051) 848 844</td>
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</tbody>
</table>

A Digital Rectal Examination (DRE) should be performed on every patient who is having a prostate assessment.

**Patient Advice:**
- Prostate assessment involves a blood test and a rectal examination
- A normal assessment does not rule out cancer
- A biopsy can be uncomfortable. Side effects such as bleeding, infection or urinary retention may occur but less than 1% require hospital admission

**Guidance on PSA Testing**
- Patients should be counselled before they have a PSA test
- Patients with an abnormal PSA result should have a repeat PSA at six weeks. If the patient also has an abnormal DRE, the PSA test does not need to be repeated and they should be referred directly
- Finasteride/dutasteride reduce PSA results by 50%, therefore the PSA result should be doubled in these patients
- DRE performed before the PSA does not raise the result

This guideline represents the view of the NCCP, which was arrived at after careful consideration of the evidence available. Health professionals are expected to take fully into account when exercising their clinical judgment.

The guidance does not, however, override the individual responsibility of health professionals to make decisions appropriate to each patient. This guideline will be reviewed as new evidence arises, and supersede all previous HSE/NCCP prostate cancer GP referral guidelines. Version 4 – Date: January 2012
REFERRALS FOR SUSPECTED PROSTATE CANCER

PATIENTS SHOULD RECEIVE FULL ADVICE PRIOR TO PSA TESTING

Patient is aged from 50 to 70 years (or from 40 to 70 years if he has a first degree relative with prostate cancer or is of African ethnicity).

ASYMPTOMATIC MEN

Advise patients on the advantages and disadvantages of PSA testing

If prostate assessment requested perform the following:
- DRE – Digital Rectal Examination
- PSA – Prostate Specific Antigen

SYMPTOMATIC MEN

Male patient presents with:
Any of the following features when unexplained:
- Lower urinary tract symptoms e.g. dysuria, urgency, nocturia
- Unexplained back pain

RECOMMENDED INVESTIGATIONS

- DRE – Digital Rectal Examination
- PSA – Prostate Specific Antigen
- Creatinine
- Hb
- Urinalysis

If normal DRE and PSA manage symptoms in Primary Care or refer to urology clinic as clinically indicated.

Refer Patient to Rapid Access Clinic if he has
- A second abnormal PSA at 6 weeks after the first PSA test
- Abnormal hard Prostate on DRE

PSA ADVICE

WHEN TO DELAY PSA TEST
PSA test should be delayed by 6 weeks if patient has any of the following:
- active urinary tract infection, prostate biopsy, TURP or prostatitis.

WHEN TO REPEAT PSA TEST
- Repeat an abnormal PSA test at 6 weeks before referral. The result can vary by up to 30%

HOW THE NORMAL PSA RAISES WITH AGE

<table>
<thead>
<tr>
<th>Age</th>
<th>PSA Caucasian Reference Ranges</th>
</tr>
</thead>
<tbody>
<tr>
<td>40-49 years</td>
<td>0.2-5.5ng/ml</td>
</tr>
<tr>
<td>50-59 years</td>
<td>0.3-5.5ng/ml</td>
</tr>
<tr>
<td>60-69 years</td>
<td>0.4-5.5ng/ml</td>
</tr>
<tr>
<td>70-79 years</td>
<td>0.6-5.5ng/ml</td>
</tr>
</tbody>
</table>

Corresponding reference ranges for men of African ethnicity are 0-2.0ng/ml(40-49yrs), 0.4-5.5ng/ml(50-59yrs), 0.7-5.5ng/ml(60-69yrs) and 0.5-5.5ng/ml(70-79yrs).

- Double the PSA result if the patient is on finasteride / dutasteride (These drugs halve the PSA level)
- Please refer to your local PSA reference ranges as some assays give slightly different results

REFERRAL
Major inter-laboratory variations in PSA testing practices: results from national surveys in Ireland in 2006 and 2007

F. J. Drummond · L. Sharp · H. Comber

The number of tPSA tests continues to rise and variation in testing practices persists: a survey of laboratory services in Ireland 2008–2010

F. J. Drummond · E. Barrett · R. Burns · C. O’Neill · L. Sharp
Table 1: PSA workload in laboratories in Ireland, 2006 and 2007

<table>
<thead>
<tr>
<th></th>
<th>No. labs Measuring PSA</th>
<th>Mean no. tests/lab N (range) 2006</th>
<th>No. tests January–March 2006</th>
<th>Total no. tests 2006</th>
<th>Mean no. tests/lab N (range) 2007</th>
<th>No. tests January–March 2007</th>
<th>Total no. tests 2007</th>
</tr>
</thead>
<tbody>
<tr>
<td>tPSA</td>
<td>36/55</td>
<td>2,656 (100–11,000)</td>
<td>95,622</td>
<td>382,488</td>
<td>2,843 (25–12,900)</td>
<td>102,345</td>
<td>409,380</td>
</tr>
<tr>
<td>fPSA</td>
<td>14/55</td>
<td>865 (10–5,500)</td>
<td>12,116</td>
<td>48,464</td>
<td>637 (25–4,600)</td>
<td>8,923</td>
<td>35,692</td>
</tr>
</tbody>
</table>

a measured between 1 January and 31 March 2006
b measured between 1 January and 31 March 2007
c Estimated annual workload was extrapolated from the responses on numbers of tests conducted during the first quarter of each year

Reference ranges

- 56% (n=19) tPSA ≥4 ng/mL
- 3% (n=1) tPSA ≥3.2 ng/mL; 3% (n=1) tPSA ≥3.1 ng/mL
- 38% (n=13) age-specific normal values
- 9% (n=3) unknown

Calibration

- 26% (n=11) implemented the WHO First International Standards (IRR96/670)
- 2 / 11 reduced reference limit to ≥3.2 ng/mL
Imprecision of tPSA and fPSA assays

Coefficient of Variation (CV)

Mean tPSA (ng/mL)

30% > 5%

Coefficient of Variation (CV)

Mean fPSA level (ng/mL)

38% > 5%
• Most PSA tests originate from GPs
• Opportunistic case finding has led to a decrease in age at PCa diagnosis and a shift towards more localised disease at diagnosis
Trends in prostate specific antigen testing in Ireland: lessons from a country without guidelines

F. J. Drummond · A.-E. Carsin · L. Sharp · H. Comber

- In Ireland there are no national guidelines on PSA testing.
- In 2006 the National Cancer Forum recommended against the introduction of population-based prostate cancer screening.
• There was a 19-fold increase in the number of PSA tests performed, 1994–2005.
• The rate of PSA testing increased by 39% in men younger than 50 years and by 25% annually in men aged 50 years and older.
• Men outside the recommended age groups (<50 and >70 years) are having regular PSA tests, despite the fact that this has not been shown to be clinically beneficial.
Sources of Variation

• Assays used
• Reference ranges
• Calibration methods
• Turnaround times
• Imprecision
• Workload
Standardization of assay methods reduces variability of total PSA measurements: an Irish study

James C. Forde*, Laure Marignol†, Ophelia Blake‡, Ted McDermott*, Ronald Grainger*, Vivien E. Crowley‡ and Thomas H. Lynch*

*Department of Urology, St James’s Hospital, †Prostate Molecular Oncology Research Group, St James’s Hospital and Trinity College, and ‡Department of Clinical Biochemistry, St James’s Hospital, Dublin, Ireland

NCCP - Working Group on PSA Harmonisation – using patient samples to compare variability across the country
PSA study – St James’s Hospital

• Between July and December 2009
• 84 male patients attending the Urology OPD Clinic
• Blood sample collected and serum dispensed into 9 aliquots within 2 hours of venesection
• All aliquots stored at -20°C
• An aliquot was sent to each of the Cancer designated Laboratories throughout the country
• One spare aliquot was retained in the host Lab.
PSA study – St James’s Hospital

• Samples were transported and stored at -20°C
• All samples were thawed on ice and analysed within 1 hour
• Six different methods in use in the 9 laboratories
  • Beckman Coulter (Hybritech, WHO calibrated)
  • Tosoh AIA 1800
  • Roche E170 (4 laboratories)
  • Abbott AxSym
  • Immulite 2500 (2 versions 2nd Gen & 3rd Gen)
  • Siemens Advia Centaur
Results – all tPSA results (0.5-30 µg/L, N=84)

- Differences between the different methods were statistically significant (ANOVA, P<0.001)
- Differences in tPSA values were >10µg/L at the upper range
Bland–Altman plots demonstrating the agreement between the tPSA assays used by each Hospital and the reference method (Beckman Access Hybritech, WHO)

Best agreement-Hospital 6 (bias: $0.65 \pm 1.6 \mu g/L$)
Poorest agreement-Hospital 8 (bias: $2.2 \pm 2.4 \mu g/L$)
Results - TPSA (3-7 μg/L, n=25)

• Mean and SD of 5.2 ± 1.3 μg/L
• Differences between the means were statistically significant (ANOVA, P<0.001)
• Minimum variability in PSA values was 0.86 μg/L
• Largest variability in PSA values was 4.3 μg/L
Results: PSA 3 - 7μg/L

- The difference in tPSA between the two methods increased as the mean tPSA increased in all Hospitals
- The range in individual tPSA values was
  - <1μg/L for 2/25 (8%) patients
  - between 1 and 2 μg/L for 11/25 (44%) patients
  - Excess of 2 μg/L for 12/25 (48%) patients
Results – same method

- Four hospitals used the same assay (Roche E170)
- Mean tPSA value measured by E170 in this cohort was not statistically significant (ANOVA, P=0.990)
- Agreement was excellent between these Laboratories (Bias <0.2 μg/L)
Results – same assay method

• For tPSA ranging from 3 to 7 μg/L
  • Minimum variability in PSA is ± 0.16 μg/L
  • Largest variability in tPSA is ± 1.77 μg/L

• Range in individual tPSA values was <0.5 μg/L for 13/25 (52%) patients
Conclusion

• PSA values varied significantly throughout the nine hospitals involved in the study
• Using the same assay method reduces this variation considerably
• Despite the availability of the WHO reference material for assay calibration, significant differences exist
• Number of PSA assays currently in use throughout the country needs to be reduced
• A significant number of patients in Ireland would be referred for biopsy simply based on the inherent variability of the assay
Setting quality specifications for PSA assay performance

- Serum/Plasma PSA: unit of measurement
- Calibration of PSA assays
- Reference values/Clinical cutoffs
- Internal Quality Control (IQC) Targets
- External Quality Assessment (EQA) targets
- Harmonisation of pre-analytical requirements
- Biological variation
# Reference Standards for PSA Assays

<table>
<thead>
<tr>
<th>Standards traceable to</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Hybritech Standard</strong></td>
<td>Tandem-R assay, first FDA approved PSA assay in 1986. Using this assay a multicentre prospective study (J Urol 1994; 152: 2037-42) validated a clinical decision point of 4.0 μg/L for early detection of prostate cancer. Many assays whose standardisation has been closely aligned to the Hybritech assay have been developed promoting the 4.0 μg/L cut-off value.</td>
</tr>
<tr>
<td><strong>WHO International Standard</strong></td>
<td>Released in 1999 with the expectation that this standard would lead to greater consistency of PSA results as manufacturers began to use it to calibrate PSA assays.</td>
</tr>
</tbody>
</table>

Total PSA: 96/670
Free PSA: 96/668
<table>
<thead>
<tr>
<th>Beckman Coulter Immunodiagnostics PSA Assay (Beckman Coulter Data)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Total PSA (μg/L)</strong></td>
</tr>
<tr>
<td>Using Hybritech calibration</td>
</tr>
<tr>
<td>4.0</td>
</tr>
</tbody>
</table>

*Note: Sensitivity (81.6%) and specificity (48.0%) maintained at these cut-offs*
Age related Reference Intervals

- Oesterling et al showed an age related increase in serum PSA in normal men
- This increase was mostly explained by the age related increase in prostate size
- Distribution was found to be skewed (log normal); so for a 95\textsuperscript{th} percentile (one tailed) cutoff for PSA, 5\% of normal men will have a PSA above the cutoff (ie 95\% specificity at any age)
<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Total PSA (µg/L)</th>
<th>Total PSA (µg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Oesterling et al. 1993</td>
<td>If recalculated to WHO</td>
</tr>
<tr>
<td></td>
<td>in current NCCP Guidelines</td>
<td>96/670</td>
</tr>
<tr>
<td>40 – 49</td>
<td>0 – 2.5</td>
<td>0 – 1.9</td>
</tr>
<tr>
<td>50 – 59</td>
<td>0 – 3.5</td>
<td>0 – 2.7</td>
</tr>
<tr>
<td>60 – 69</td>
<td>0 – 4.5</td>
<td>0 – 3.5</td>
</tr>
<tr>
<td>70 – 79</td>
<td>0 – 6.5</td>
<td>0 – 5.0</td>
</tr>
</tbody>
</table>
Setting a target for IQC

- **Option 1**: Set as target that which is consistently attained by 80% of the participants.

- 79% of CVs are less than 4%
<table>
<thead>
<tr>
<th>IQC CV (%)</th>
<th>Number of values</th>
<th>Percent of all values</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 2</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>&lt; 3</td>
<td>15</td>
<td>52</td>
</tr>
<tr>
<td>&lt; 4</td>
<td>23</td>
<td>79</td>
</tr>
<tr>
<td>&lt; 5</td>
<td>26</td>
<td>90</td>
</tr>
<tr>
<td>&lt; 6</td>
<td>29</td>
<td>100</td>
</tr>
</tbody>
</table>
Setting a target for IQC

- **Option 2:**
  - Set target for analytical precision \((C_{Va})\) in relation to the within subject biological variation (References: Callum Fraser).
  
  - *Within subject biological coefficient of variation \((CV_i)\) for PSA = 14.0%*
  
  - *(From Carmen Ricós and associates).*

- **Callum Fraser’s proposals:**
  - Desirable performance: \(CV_a < 0.5\ C_v\) \[ < 7.0\% \]
  - Optimum performance: \(CV_a < 0.25\ C_v\) \[ < 3.5\% \]
  - Minimum performance: \(CV_a < 0.75\ C_v\) \[ < 10.5\% \]
The goal adopted is such that over 80% of laboratories can achieve the performance.

This target encourages further refinement of methods particularly to achieve the tighter monitoring goals.

The format of the ALP is $\pm x$ from the target value where $x$ may be expressed as a percent, an absolute value, or an absolute value up to a certain target value and then a percent above that value.
### Allowable Limits of Performance

Reviewed January 2012

<table>
<thead>
<tr>
<th>Total PSA</th>
<th>± 0.4 μg/L for values up to 5.0 μg/L</th>
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<tbody>
<tr>
<td></td>
<td>8% for values greater than 5.0 μg/L</td>
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</table>
Other issues

• Existence of antibodies to PSA in the serum of 5% of sexually active women as well as men with P Ca
• Heterophilic antibodies will affect PSA assays using the respective animal antibody
• Form of PSA used for calibrations (90:10, 80:20 or 70:30)
• Matrix of the calibrator (PBS or female sera)
• Ratio of Free to Total PSA varies in patient samples – assays with equimolar reactivity is required
• Assay architecture (monoclonal/polyclonal or monoclonal/monoclonal Abs)
Thank You

• Acknowledgements:
  • Dr James Forde
  • Dr Ned Barrett (IEQAS & NCCP)