Newborn screening in Australia over the last 50 years

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Australia
50 years of Newborn Screening in Victoria

Early diagnosis of treatable disorders by newborn screening has saved thousands of babies around the world from mental and physical disability or death

Something to celebrate!
50 years of Newborn Screening in Victoria

- The history of newborn screening begins with PKU
- PKU – phenylketonuria – is an enzyme deficiency
- Levels of an amino acid, phenylalanine, normal in utero, rise after birth and cause severe brain damage
- Very early dietary treatment enables normal intellectual development
- Autosomal recessive inheritance
First screening for PKU

- The disorder was uncovered in 1934 (Følling)
  - A urine test (ferric chloride) was developed
- Stimulus for screening was the discovery of dietary treatment in the 50s

First proof of efficacy of treatment of PKU

Bickel, Gerrard and Hickmans. Lancet, 1953
The experiment

• “A girl aged 2 years.....was an idiot, unable to stand or talk ... spent her time groaning, crying, banging her head”

• Dr Bickel diagnosed PKU. Her mother pressed him to find some treatment

• After experimental near-removal of phenylalanine from the diet, she gradually improved, no longer cried, started to crawl, stand and climb

• Secret re-introduction of phenylalanine caused loss within days of all her developmental gains
The first trial of treatment for PKU:
A cross-over design
How well did the treatment work?

• Babies and children needed a very low protein diet (vegetarian) with the special formula

• The treated children improved a lot, but were still intellectually retarded

• Treatment started at birth was very successful in siblings of known cases

• The race was on to find a test to screen for PKU in all newborns

• An early urine test led to some hospital based screening but was only partially successful in case-finding
  – Dr Willard Centerwell; several others
The first report of the new PKU test

JAMA Nov 25th 1961
Robert Guthrie PhD, MD

Recently, with the cooperation of Dr. William Welch, Director of the Laboratory at Newark State School, Newark, N.Y., we have had the opportunity to develop and evaluate a simplified blood phenylalanine-agar diffusion test by screening a large population of mentally retardedes.

finding permitted the development of a convenient agar diffusion microbial assay, employing small filter paper discs impregnated with blood serum on the agar surface. This assay has been used successfully in the determination of blood phenylalanine levels
Front and back of an original card used for testing in Guthrie’s Buffalo laboratory
From Jean Koch’s book:
“Robert Guthrie, the PKU story”

A PKU test plate - from our laboratory in the 70’s
The first trial of PKU screening

- Bob Guthrie had developed his test by 1961. He received a grant from the Children’s Bureau in 1962 to mount a study of 400,000 infants, with 40 cases projected to be found.

- On May 25th 1962 all US states invited to participate.
- 29 states + Puerto Rico contributed samples by Dec 31st 1963.

- 14.1% infants from participating states tested – 404,568.
- 275 “presumptive positive” (0.067%). 37 had PKU. (PPV 13%)
  - One infant with PKU not tested (discharged before d 3). One had a result 4-6mg% and was picked up on a test at 4w. 100-fold variation among states for presumptive positive rate.
Significant findings in the report
Guthrie and Whitney  Children’s bureau publication 419 1964

• At least 26 of 37 PKU cases (maybe more) were on diet by 1 month
• Birth incidence 1:10,374
• Cases of “occult PKU” (mild or delayed Phe rise)
• Older PKU-affected siblings discovered
• Case of maternal PKU. “It is hoped all laboratories will test mothers” of non-PKU infants with elevated Phe levels
• Suggested protocols for intermediate Phe levels
• Early discharge seen as a problem – milk feed thought necessary
• “This procedure (screening for PKU) is recommended as a public health procedure”

• “Current investigation.......is directed towards a “multiple test” for a number of rare inherited conditions”
How well did things work for PKU screening?

• Pretty well
  – By 1965, 32 states in USA had laws about screening, mandatory in 27 states

• Very little adverse effect (Brosco et al 2006)
  – Early dietary struggles; cases with phenylalanine deficiency symptoms; need for confirmation of diagnosis before instituting treatment
  – “Atypical” PKU due to pterin disorders not appreciated early

• Factors aiding success:
  – The natural history was well known
  – The surrogate effects of treatment (blood phe level) easy to measure
  – Mild cases thus fairly easy to identify and manage

• Persisting problems: We still don’t know for sure –
  – What exact blood levels to aim for at different ages
  – How to treat better – in a more acceptable way – (or how to cure)
What was happening in Australia?
PKU screening

Victoria: The father of Victorian screening – Dr David Pitt
- 1964: screening of children in institutions for PKU underway:
- 1966: Start of newborn screening for PKU
5th April, 1963

Dr. Robert Guthrie,
Department of Paediatrics,
University of Buffalo,
BUFFALO. U.S.A.

Dear Doctor Guthrie,

I am writing from a Mental Deficiency Institution of 730 patients, where we are building up a research programme. We have 13 cases of Phenylketonuria in the Institution, and there are screening programmes in progress throughout the State of Victoria in other Institutions and in well Baby Clinics (Infant Welfare Centres).

In addition we are establishing a Central Registry of Phenylketonuria.

We propose to introduce the Guthrie Test, at first here, and possibly later in other Institutions and Hospitals, and I am writing to ask you whether the particular culture of Bacillus subtilis ATCC 6051 is the only strain which gives the necessary inhibition reactions. If so I will be glad if you could tell me where we could obtain this particular strain, which is unobtainable in Australia at present.

Yours sincerely,

Dr. David Pitt,
Paediatrician
What was happening in Australia?  
PKU screening

Victoria:
• 1964: screening of children in institutions for PKU underway:
• 1966: Start of newborn screening for PKU
• 1973: Screening universal throughout Victoria

SA:
• 1966: PKU screening started; Universal by 1974 (or maybe earlier)
• 1974-77 BIA for 4 other disorders (CBS def, His, Tyr 1, MSUD) + gal α-1AT

WA:
• 1968: pilot PKU screen
• 1969: universal

Qld:
• 1973: PKU screening (state health dept microbiology lab)

NSW:
• 1964: urine chromatography c. 80% coverage achieved in a few years
• 1969: Guthrie blood-spot testing started. Universal by 1973
NSW: BEGINNINGS AT THE OLIVER LATHAM LABORATORY 1964 +
Urine chromatography

Brian Turner
Don Brown
Radial chromatography
Eluting urine

Paper prepared for chromatography
The finished chromatogram
Laboratory book from 1964

The first case of PKU identified by mass screening in New South Wales.

“Phenylalanine present. PKU reported by phone”

This was urine screening at 6+ weeks, done via baby health centres. Coverage about 80%

Blood –spot screening didn’t start in earnest until 1969
The time course of newborn screening

- **1963** Guthrie’s seminal paper; PKU screening starts
  - Other BIAs for MSUD, homocystinuria (CBS), histidinaemia etc
  - Galactosaemia
- **1975** – T4 assay for hypothyroidism
  - Soon overtaken by TSH as primary assay in Australia and elsewhere
Screening for hypothyroidism

- **1971-72: Treatment at <3 months prevented significant intellectual disability** *(Raiti & Newns, Klein et al)*
- **1971-72: Dussault & Laberge develop an RIA for T4 in dried blood spots**
  - Abstract for International Endocrine Soc meeting REJECTED
  - Pilot in Quebec: 30,000. 4 cases of CH detected
  - Preliminary report REJECTED by Clin Chem & NEJM (“irrelevant”)
  - Their first paper published in French (1973)
- **1974 Routine screening for CH added to Phe & Tyr in Quebec**
  - Late 1974 prelim. report – American Thyroid Association (last paper on last day)
  - Computerisation in the screening lab; tight collaboration of endocrinologists
  - Screening in Oregon, New England, France
  - 1977 Screening started in New South Wales, Victoria
The time course of newborn screening

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- **1980’s** – Cystic Fibrosis – blood-spot method; not well taken up
  - CAH; biotinidase; haemoglobinopathies; DMD

- **Newborn screening was a quiet back-water for 30 years**
Screening in Australia until 1998

- NSW
- QLD
- SA
- VIC
- WA

- CF
- GAL
- CH
- PKU

1960 1980 2000
Many thanks for a copy of your report on three years experience of screening for cystic fibrosis. This is a very good paper…………………….Your conclusion is that the programme advantages outweigh the disadvantages and accordingly the programme should be continued. We have at the present time no formal mechanism for considering such issues...........
1990’s: A pivotal change

- 1991 Tandem mass spectrometry (MSMS) becoming possible
- A method of separating compounds by both mass and structure, and using linked scanning techniques to observe fragments
- Can be used to detect many different classes of compound
- MULTIPLEX TESTING – one test, many analytes, many disorders.
  - aminoacid disorders
  - organic acid disorders
  - fatty acid oxidation disorders
- Discussed at the 1991 International screening meeting in Leura, NSW technology still too slow for large numbers, but getting there
- 1996 N Carolina and Pennsylvania: some screening via private laboratory
- 1998 Routine MSMS screening started in NSW
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1990’s: A pivotal change

• Screening now interesting to scientists and physicians
  – New technology exciting and flexible;
  – And DNA could be extracted from dried blood spots easily

• Public pressure in the US

• 2005 HHS Secretary’s Advisory Committee on Heritable Disease recommends Uniform Screening Panel: 29 disorders (now 32)
  ACMG. Newborn screening: toward a uniform screening panel and system. *Genet Med.* 2006;8 Suppl 1:1S-252S

• By 2006: MSMS screening widespread

• Increasing consideration of other disorders for multiplex tests: eg Lysosomal storage disorders
What NSW currently screens for by tandem mass spectrometry (One 3mm blood spot)

Except for disorders considered benign or for technical reasons not screened

Other states similar

<table>
<thead>
<tr>
<th>Class</th>
<th>Disorder</th>
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</thead>
<tbody>
<tr>
<td>Amino acids</td>
<td>Argininaemia/arginase deficiency</td>
</tr>
<tr>
<td></td>
<td>Argininosuccinic aciduria (ASA lyase deficiency)</td>
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<td></td>
<td>Citrullinaemia (argininosuccinate synthase deficiency, citrin deficiency)</td>
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<tr>
<td></td>
<td>Citrin deficiency</td>
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<tr>
<td></td>
<td>Fumaryl acetoacetase deficiency (tyrosinaemia type I)*</td>
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<td></td>
<td>Homocystinuria (cystathionine β-synthase deficiency)</td>
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<td></td>
<td>Maple syrup urine disease</td>
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<td></td>
<td>Phenylketonuria</td>
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<td>Pterin defects</td>
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<td>Tyrosine aminotransferase deficiency</td>
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<td>Organic acid disorders</td>
<td>Beta-ketothiolase deficiency (mitochondrial acetoacetyl-CoA thiolase deficiency)</td>
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<td>Cobalamin C defect (homocystinuria with methylmalonic aciduria)</td>
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<td>Glutaryl CoA dehydrogenase deficiency (glutaric acidemia type I)</td>
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<td>Holocarboxylase synthetase deficiency</td>
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<td>3-hydroxy-3-methylglutaryl-CoA lyase (HMGCoA lyase) deficiency</td>
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<tr>
<td></td>
<td>Isobutyryl-CoA dehydrogenase deficiency†</td>
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<td>Isovaleric acidemia</td>
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<td></td>
<td>Methylmalonic acidurias (mutase deficiency, CblA and CblB defects)</td>
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<td>Propionic acidemia</td>
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<td></td>
<td>3-methylcrotonyl-CoA carboxylase deficiency</td>
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<td>2-methylbutyryl-CoA dehydrogenase deficiency</td>
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<td></td>
<td>3-methylglutaconyl-CoA hydratase deficiency</td>
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<td>Fatty acid oxidation</td>
<td>Carnitine/acylcarnitine translocase deficiency</td>
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<td>Carnitine transporter defect</td>
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<tr>
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<td>CPTI (carnitine palmitoyl transferase deficiency type I)</td>
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<tr>
<td></td>
<td>CPTII (carnitine palmitoyl transferase deficiency type II)</td>
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<tr>
<td></td>
<td>LCHAD (3-hydroxy long chain acyl-CoA dehydrogenase deficiency)</td>
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<tr>
<td></td>
<td>MCAD (medium chain acyl-CoA dehydrogenase deficiency)</td>
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<tr>
<td></td>
<td>MADD (multiple acyl-CoA dehydrogenase deficiency)</td>
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<tr>
<td></td>
<td>SCAD (short chain acyl-CoA dehydrogenase deficiency)†</td>
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<tr>
<td></td>
<td>SCHAD (short chain hydroxy acyl-CoA dehydrogenase deficiency)†</td>
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<td>TFP (trifunctional protein deficiency)</td>
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<td></td>
<td>VLCAD (very long chain acyl-CoA dehydrogenase deficiency)</td>
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</tbody>
</table>
A quiet endeavour for 33 years, then a huge expansion.
## Current screening in Australasia

<table>
<thead>
<tr>
<th>Programme</th>
<th>Annual births</th>
<th>Disorders screened</th>
</tr>
</thead>
<tbody>
<tr>
<td>NSW + ACT</td>
<td>97,000</td>
<td>PKU, CH, CF, gal, MSMS (~29)</td>
</tr>
<tr>
<td>Queensland + NT</td>
<td>65,000</td>
<td>PKU, CH, CF, gal, MSMS (~29)</td>
</tr>
<tr>
<td>Victoria</td>
<td>79,000</td>
<td>PKU, CH, CF, MSMS (~29)</td>
</tr>
<tr>
<td>S. Australia +NT+Tas</td>
<td>28,000</td>
<td>PKU, CH, CF, gal, MSMS (~29)</td>
</tr>
<tr>
<td>W. Australia</td>
<td>35,000</td>
<td>PKU, CH, CF, gal, MSMS (~29)</td>
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<tr>
<td><strong>Total</strong></td>
<td><strong>300,000</strong></td>
<td></td>
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<tr>
<td>[New Zealand]</td>
<td><strong>57,000</strong></td>
<td>PKU, CH, CF, CAH, Biot, Gal, MSMS</td>
</tr>
</tbody>
</table>
Screening in Australia over time
Nothing new since MSMS in 1998-2004

1960                    1980                   2000

NSW
QLD
SA
VIC
WA

MSMS
CF
GAL
CH
PKU
What have we achieved in Australia?

• **Very well run programs:**
  – Good performance: high specificity and sensitivity
    (Low false positive rates, accurate diagnosis)
  – Careful consideration of benefits and harms

• **Coordinated programs:**
  – Screening laboratories now closely linked with diagnostic laboratories and clinical service - rare in the world!

• **Measuring benefit:**
  – World leaders in measuring benefit especially for cystic fibrosis screening and tandem mass spectrometry
What are the problems that newborn screening faces, world-wide and in Australia

- **Over-diagnosis: Not too bad here. Bad in US**
  - Huge lack of differentiation between very mild clinically insignificant “biochemical” disease and the real thing

- **Expensive confirmatory testing: Pretty good in Australia**

- **Lack of proper follow-up: We have been good at this**
  - It is vital to know if screening is beneficial overall – or not

- **Assessing programs overall and stopping ineffective ones: again we have been good at this:**
  - SA stopped early efforts to screen for α1AT, histidinaemia, tyrosinaemia, MSUD and CBS deficiency
  - NSW stopped neuroblastoma, biotinidase def.

- **Using good evidence to include new disorders: We haven’t had an opportunity to do this**
What is holding us back now?

- No new disorder has been added to the screening panel since MSMS came in from 1998 onward.
- We are not funded to screen for several disorders which are widely screened for around the world (e.g. CAH). A huge increase in birth-rate has not seen funding increases.
- USA screens for 32 “core disorders” plus a number of others. Illinois currently claims 58: Many of these do NOT appear wise inclusions, but some are clearly beneficial.
- We do not yet have any Australia–wide policy (but wait for Professor Craig White to update you on this later today!)
What is on the horizon?
New testing being performed or seriously considered world wide

Performed in several places, or being piloted

- **SCID** – (severe combined immunodeficiencies) 11 US states, 1:58,000
  New Zealand has approved this
- Lysosomal storage disorders
  - Pompe, Gaucher, Fabry, Krabbe, Niemann-Pick B, MPS I etc
- **XALD** – started in New York State

Being considered

- Disorders that can take advantage of new molecular-specific treatments (stop-codon read-through, exon-skipping, chemical chaperones etc) eg Duchenne muscular dystrophy
- And many, many more:
  - Rett syndrome, FraX, Long QT, Diabetes Mellitus etc etc etc
- Whole genome, exome, or targeted sequencing
Next generation sequencing is being widely touted

- The ultimate multiplex test
- The almost certain way to find many “cases” that do not need any intervention, ever
- “$100 test, $100,000 interpretation”
Next generation sequencing – a whole new ball-game

• **Whole genome, exome, or targeted sequencing**
  – Detection of mild or insignificant “cases” bound to be a major problem BUT

• **NGS almost certain to be the way of the future**
  – Despite worries about cost, accuracy, interpretation, clinical validity and utility, iterative nature of the test, etc, etc, etc

• **NIH (US) research programmes**
  “We are not ready now to deploy whole genome sequencing on a large scale but it would be irresponsible not to study the problem” using the time before the technology matures to hash out difficult issues. “There will be an industry setting up around this.” Dr Eric Green, Human Genome Research Institute NIH.
But for now:

- We can look with satisfaction on what we’ve achieved
- We can look forward to an Australian policy and groundwork for some expansion
- Perhaps we can work to publish Australia–wide results – not done since David Pitt’s series of communications between 1970 and 1981
- And we must thank Victorians for being such great colleagues in all newborn screening endeavours—especially Ivan, for 50 years service to newborn screening