O&G Magazine

Vol 18 No 2 Winter 2016

Genetics

The Royal Australian and New Zealand College of Obstetricians and Gynaecologists

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Genetics

Women's health

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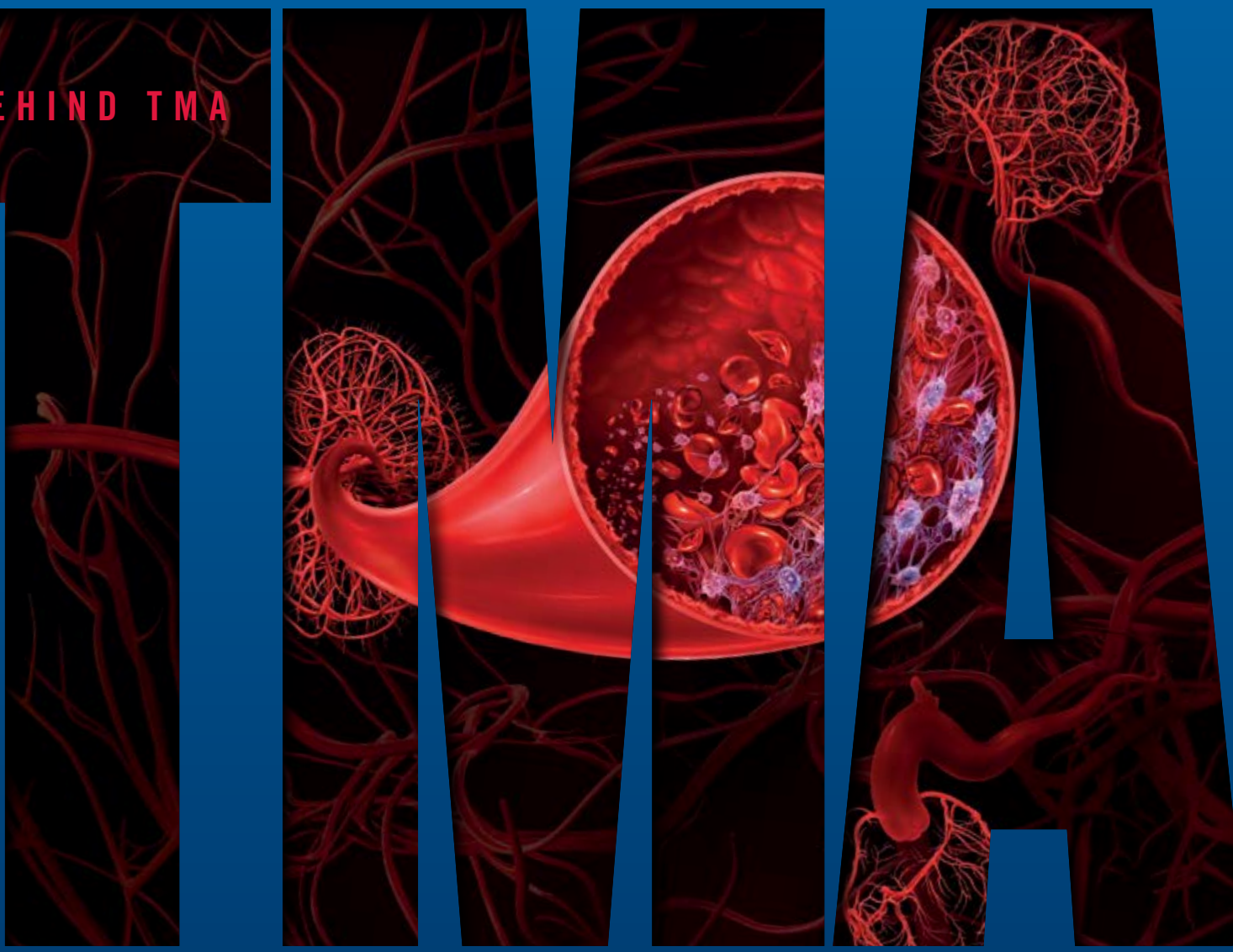

In the diagnosis of postpartum Thrombotic Microangiopathy (TMA), the first 48 hours can be decisive [1]

- HELLP syndrome and pregnancy-related atypical Haemolytic Uraemic Syndrome (P-aHUS) can have similar clinical presentations [2]
- The onset of postpartum HELLP syndrome is usually within 48 hours of birth [1]
- In contrast, P-aHUS can occur at any time in the 4 months following birth [2]
- Prompt, accurate diagnosis is essential as there is a high (62%) risk of the development of End Stage Renal Disease (ESRD) within the first month of aHUS manifestation [2]

It is recommended that any patient suspected of having aHUS be referred to a nephrologist [3]
For more information please call 1800 788 189 or visit PREGNANCYRELATEDAHUS.COM.AU

ALEXION

HELLP = Haemolytic Anaemia, Elevated Liver Enzymes, and Low Platelets. References 1. Cakmak B, et al. *Journal of Clinical and Diagnostic Research.* 2013;7(12):2998-2999. 2. Fakhouri F, et al. *J Am Soc Nephrol.* 2010;21:859-867. 3. Salvadori M, Bertoni E. *World J Nephrol.* 2013;2(3):56-76. ACN 132 343 036. Suite 401, Level 4, Building A, 20 Rodborough Rd, Frenchs Forest NSW 2086 Australia. May 2016 AU/UNB-aHUS/16/0002

From the President

Prof Michael Permezel
President

Let me open this report with congratulations to Prof Stephen Robson on his election in March to President of the Tenth RANZCOG Council. Steve has made enormous contributions to the College, in almost every area of College activity – from examinations through to women's health and publications. He will take over as President at the November Annual General Meeting. The new Board will be elected in July from the current Council and a new Council will be elected by the Fellowship in September.

Workforce

Compulsory blood-borne virus testing

Communicable Diseases Network Australia (CDNA) is a government advisory body comprising appropriate experts and representatives of the states and territories. It has had many roles, including working with the College in developing guidelines in response to the Zika virus outbreak.

Among the areas under consideration is a proposal that health professionals exposed to blood-borne viruses undertake regular (for example, annual) assessments of their blood-borne virus status. The implications of a positive result are not what they once were. Affected practitioners can expect a return to surgical practice when effective antiviral therapy has reduced the viral load to a level that is considered to have virtually no risk of transmission to patients. In addition, early diagnosis and treatment of a positive result will likely be profoundly beneficial to all affected practitioners in terms of their long-term health.

While not yet government policy, the rights to privacy of an affected individual will be paramount. There is an expectation that test results will be the sole property of the individual and any registration requirement should only be a declaration that the test has been performed.

Bullying and harassment

The College will soon circulate a survey to Fellows and trainees in relation to bullying and harassment in the workplace. While the recent focus in the media has largely been on surgeons, it is important to establish the extent of this issue within our own specialty, develop strategies for improvement and, subsequently, monitor progress. Bullying and harassment cannot be allowed to persist in the workplace. My own impression is that such episodes are very uncommon. However, it is timely for the College to review this important area.

Selection for FRANZCOG Training

At the time of writing, the New Zealand selection process is well underway and Australian applications close in mid-May. It is expected that the College will receive another record number of applications. Unfortunately, many outstanding potential future specialists will miss out on selection, largely because there are simply too many good applicants, but we must also recognise that perfection in selection is extremely difficult.

The College continues to explore methods of ensuring that the most-suited applicants emerge from the selection process as the new trainees. In striving for the most efficacious selection tools, the College is exploring the use of situational judgement tests (SJTs), which have been used extensively in the UK. Although they will not count toward the selection score this year, it is hoped that SJTs will prove to be a tool that further enhances the selection process. The College will continue to explore the possible future use of other tools and methods that can capture the desired skills and attributes. For example, it seems surprising that, in a predominantly surgical discipline, there is little in the selection score that reflects surgical aptitude.

Many Fellows and registrars are understandably frustrated by the fact that performance in the hospital before application does not appear to influence the selection process. It would seem reasonable to assume that current performance is a good predictor of future performance and that hospitals are in a valuable position to provide such information. Applicant-nominated referees almost invariably submit very good references and therefore are almost consistently unhelpful as a discriminator. Institutional references do discriminate between applicants and allow the selection process to incorporate important traits not otherwise captured, including surgical aptitude and professional attributes such as diligence and reliability. At this point, institutional references do not count in the total selection points that determine who is selected and who is not. They will continue to be evaluated for future use.

Selection for FRANZCOG training must also ensure diversity across the profession, from generalist practice to each of the five subspecialties, so that the needs of women can be met. It must also recognise workforce issues in provincial areas and indigenous communities in Australia and New Zealand.

> ‘Only when clinicians own a guideline is there a reasonable expectation that the contents are relevant to contemporary practice.’

While a specialty full of academics would be unwelcome, a clinical academic O&G workforce has become progressively more important as our practice becomes increasingly dictated by guideline-development committees. It is so much better if membership of these committees comprises clinical peers practising in the discipline in question – as exemplified by the College’s Women’s Health Committee. The evidence itself almost never dictates a guideline. It is the interpretation of all available evidence in the relevant clinical context that culminates in a clinical guideline. Only when clinicians can be said to own a guideline is there a reasonable expectation that the contents are relevant to contemporary practice.

Women’s health

Clomiphene citrate

Fellows and trainees will have been surprised to hear of an impending shortage of clomiphene citrate, which may prove to be long-lasting. My thanks to the Reproductive Endocrinology and Infertility Subspecialty Committee assisting me in formulating a

response to this unfortunate situation. The College has submitted a request to the Therapeutic Goods Administration (TGA) that letrozole be licensed for ovulation induction.

Zika virus

Evidence that the Zika virus poses a genuine threat to the fetus is accumulating, although quantifying the risk continues to prove challenging, with a recent publication suggesting the fetal risk may be as high as 29 per cent in the presence of maternal infection during pregnancy. All health professionals caring for women who are, or may become, pregnant should have ready access to information regarding Zika-affected areas, management of a woman returning from a Zika-affected area and care of a pregnancy in which a Zika infection may have occurred. All this information is available on the College's women's health webpages, with links to the various information sheets.

PGF2 alpha

After initially receiving advice that hospital pharmacies should be able to access overseas product, following the discontinuation of locally produced prostaglandin F2 (PGF2) alpha, this is proving more difficult than expected. Hospitals may find they need to switch to 15-methyl-PGF2 alpha (carboprost; Prostinfenem). As the recommended routes and doses of PGF2 alpha and 15-methyl-PGF2 alpha are different, all obstetricians should familiarise themselves with the recommended dose and route for carboprost administration. Details are available in the modified statement on the Management of Postpartum Haemorrhage (C-Obs 43) on the College website.

> 'Recommendations from the Obstetric Clinical Committee, as outlined in previous reports, remain in progress...Ultrasound has predictably proved challenging, with some expected differences in perspective between the imaging specialists and clinicians providing ultrasound at the point of care.'

Medical Benefit Schedule Revision

Recommendations from the Obstetric Clinical Committee, as outlined in previous reports, remain in progress. Although there are two general practitioner (GP) members of the Obstetric Clinical Committee, additional feedback has been sought from shared-care GPs, given suggested changes to items that may affect these practitioners.

Ultrasound has predictably proved challenging, with some expected differences of perspective between the imaging specialists and clinicians providing ultrasound at the point of care. An ultrasound performed in the rooms of a GP or specialist has obvious advantages to the patient: convenience, timeliness and, usually, a significantly lower cost. There is also an opportunity for counselling by the usual provider at the time of imaging. On the other hand, few would dispute the high-level skill of the obstetric sonologist in the diagnosis of complex fetal anomalies. To further complicate the issue, many GPs and specialists have not sought accreditation of the ultrasound performed in their rooms, with the consequence that they are unable to use the non-referred item numbers for O&G ultrasound. These GPs and specialists must currently build the costs of this service into other items in their fee structure. Ensuring quality ultrasound both at the point of care and in imaging practices are high priorities and any changes suggested must not adversely affect patient care.

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From the CEO

Alana Killen
CEO

Reports of adverse outcomes for mothers and/or babies are always upsetting. Recent events in Victoria concerning infant deaths at a regional hospital have attracted significant media attention, resulting in a Department of Health investigation and the reporting of three of the deaths to the State Coroner. The Coroner's Report found significant errors were made in the care of the babies during labour and birth. She described the handling of care in each case as 'sub-optimal', with the misinterpretation of the cardiotocograph (CTG) a feature common to each. While there were other causal factors indicated in the report, including entrenched cultural behaviours and attitudes, the identification of poor CTG knowledge as a common element is significant.

The Fetal Surveillance Education Program (FSEP) was funded in 2004, by the Victorian Managed Insurance Association (VMIA) and the Department of Human Services (DHS), and developed by RANZCOG in collaboration with Southern Health, the Mercy Hospital for Women, the Royal Women's Hospital and the Victorian branch of the Australian College of Midwives (ACM). The pilot program was developed in response to a review of nearly 400 cases of obstetric medico-legal claims in Victoria, arising between 1993 and 1998, that identified inadequate or inappropriate use of intrapartum fetal surveillance as a major contributor to the claims burden. Since its introduction, the program has continued to grow in demand and gain acceptance as an evidence-based, standardised educational program designed for multidisciplinary teams.

Research is currently being undertaken to determine the efficacy of the FSEP in terms of neonatal mortality and whether the introduction of the program can be attributed to a reduction in adverse outcomes for newborns and mothers. Regardless of any associated impact, however, the FSEP has provided a range of health practitioners, including obstetricians and trainees, with

training in CTG interpretation, which has been identified as a critical skill for those caring for women during delivery.

Community awareness

RANZCOG, as a provider of education and training in women's health, has been a proactive leader in Australia, New Zealand and the Pacific region for many years. Fellows of the College provide many hours of their valuable time to help train and mentor future O&G specialists in numerous ways. For example, in early May, College House welcomed a parade of pregnant women who were on site to assist trainees learning the skills of ultrasound under the expert guidance of volunteer Fellows. In mid-May, an Obstetric Skills Workshop was conducted at the Royal Women's Hospital in conjunction with a meeting of the Pacific Society of Reproductive Health, which involves a number of College Fellows. This work is all undertaken pro bono and out of a desire to contribute to the provision of excellence in women's health.

This is hardly news to most of you; however, the broader community are largely unaware of the role of RANZCOG and how many of them have likely directly benefited from the work of the College – through giving birth, being born or being cared for by a specialist O&G or GP obstetrician in the region. Medical colleges have generally steered away from self-promotion, preferring to get on with the job of caring for patients. However, in the increasingly crowded healthcare space, it is becoming apparent that raising awareness of what we do should be a priority if RANZCOG wishes to be the pre-eminent source of information for women.

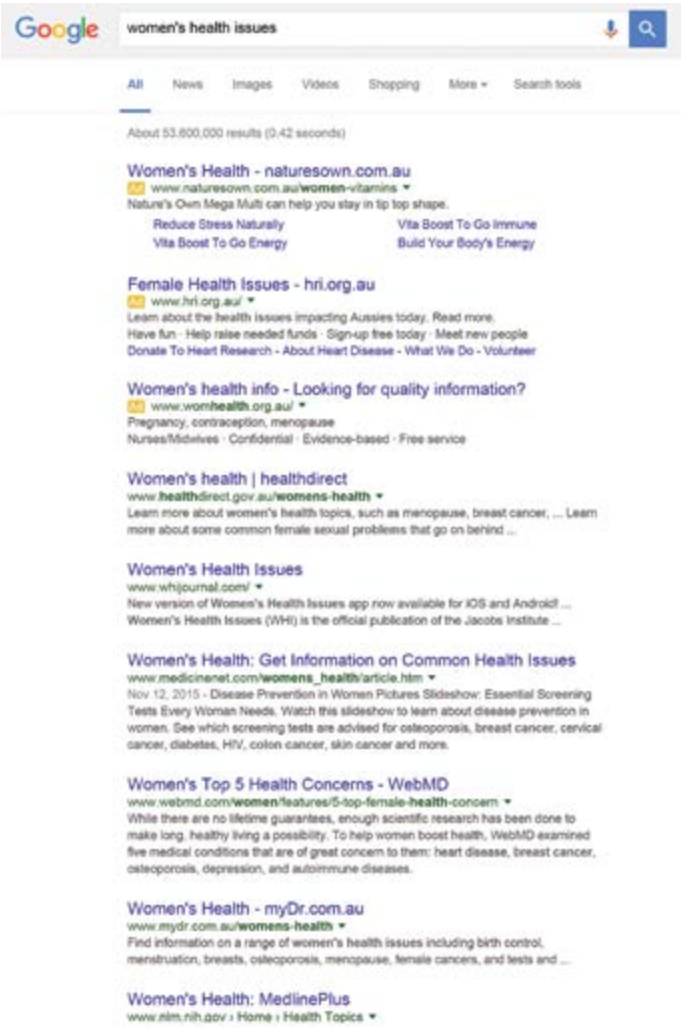

Figure 1. When Googling 'women's health issues', where is the College in these results? The first College webpage search result is the last entry on page seven of the Google search.

> 'As part of RANZCOG's Strategic Plan for 2015–19, the Board expressed the desire for the College to be the go-to authority on women's health issues.'

Regarding the 'M' word

FSEP, Obstetric and Surgical Skills Workshops, PROMPT, C-QuIP and the Nuchal Translucency Ultrasound Education and Monitoring Program are just a few of the activities run by RANZCOG, many in collaboration with other professional groups, to upskill and train clinicians working in women's health. The breadth and depth of knowledge that exists within the College membership is extensive and yet in a quick Google search of 'women's health issues' the College is notable only by its absence (see Figure 1).

This was highlighted to the College Board and Council at a recent presentation regarding social media, with the inclusion of a number of 'doctor' websites quite revealing. This led to the suggestion that all doctors should be provided with a mug bearing the legend 'please do not confuse your Google search with my medical degree' upon graduation (perhaps you already have one).

As part of RANZCOG's Strategic Plan for 2015–19, the Board expressed the desire for the College to be the go-to authority on women's health issues. We are hopeful that recent staff changes, including the recruitment of a Membership and Marketing Director, will assist in this goal, while ensuring that we retain the traditions, history and integrity that Fellows, trainees and members value. Already work has commenced on a new website and the development of a suite of patient information resources that are user-friendly and accessible. We are moving into the 'twittersphere' and are introducing apps to make our members' busy lives easier. It is an evolutionary process, but we are excited about the future growth and ongoing development of RANZCOG as a contemporary education provider and advocate for excellence in women's health.

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Editorial

The best of men cannot suspend their fate: The good die early, and the bad die late.
— Daniel Defoe. Character of the Late Dr S. Annesley (1697).

Dr Brett Daniels
FRANZCOG

It is human nature to want to know why things are the way they are now and what will they be like in the future. In our lifetime, advances in the science of genetics have helped to provide the explanation for what we see in ourselves and a crystal ball into our biological future. As we all know – from the first time we heard the phrase 'nature versus nurture' – genetics doesn't provide the whole answer to the puzzle, but increasingly it is being used to assist our patients. As an obstetrician and gynaecologist, genetics has become an everyday part of my practice. In the past few weeks I have counselled a couple on prenatal testing for aneuploidies, then on interpretation of an inconclusive result and explained the limitations of determining the risk of autism in their fetus. For a patient planning hysterectomy, a discussion of family history and risk of genetic cancer syndromes was a part of taking her medical history and informed her decision to opt for ovarian preservation.

While it may seem that the furthering of our knowledge of the inheritance of disease can only be positive, one does not have to search for long before reaching a dilemma. Perhaps one of the starkest examples of the benefits and problems of genetic advances in medicine is that of Huntington's disease. In the case of this condition, genetic testing can inform an asymptomatic patient that they are definitely going to develop an incurable disease or, alternatively, that they will never develop the disease. Such life-altering information has the potential to have a profound effect on relationships, careers and health insurance. If it was you, would you want to know?

In obstetrics, we have recently seen the emergence of non-invasive prenatal testing (NIPT). On the positive side, NIPT has allowed the earlier detection of fetal aneuploidies, with a reduced need for invasive testing and the possibility of earlier termination of the pregnancy, if that is chosen. Currently, NIPT is performed at ten weeks gestation, but it is conceivable that it will be possible to perform the test at an earlier gestation in the future. For aneuploidies this may well be advantageous, but NIPT can also reveal the sex of the fetus. Until very recently, the majority of pregnant women could not find out the sex of their baby until ultrasound at about eighteen weeks gestation, a time after which termination for maternal choice is unavailable. As NIPT and similar technologies become more easily available, and one can only imagine it becoming cheaper and less well-regulated on a worldwide basis, it is perhaps inevitable that they will be used for sex selection in the first trimester. One could suggest not reporting sex chromosomes on NIPT unless there was an abnormality, but it could be argued that this restricts a woman's choice to know all the available information about her pregnancy.

This issue of *O&G Magazine* explores the theme of genetics with a range of articles on topics from the state of the art in the genetics of gynaecological cancer and prenatal screening through to that of sex selection, while an article on genetic counselling outlines the support our colleagues can provide in this important area.

Genetics: an introduction

Dr Katie Ellis
Genetic Counsellor (FHGSA)
Genea – World Leading Fertility

Although Johann Gregor Mendel (1822–1884) is considered the father of modern genetics, observations on the inheritance of physical traits in humans can be found as early as 300CE. The Charaka Samhita, commonly held to be a foundational text for Ayurvedic medicine, says a child's characteristics are determined by four major factors:

1. the characteristics from the mother's reproductive material;
2. the characteristics from the father's reproductive material;
3. the diet of the pregnant mother; and
4. the soul that enters the fetus.

Although point four is more controversial in today's world, together these observations are rather remarkable, even including the effects of the mother's diet. Further observations on the inheritance of physical traits in humans date back to ancient Greek literature.

Back to Mendel, the first to describe the fundamentals of inheritance, his epic pea plant experiments on 10 000 pea plants over eight years gave us Mendel's Laws of Inheritance. Mendel studied the inherited traits in the pea plants (such as colour and shape) over generations and found traits are inherited from parents in set patterns. His First Law (the Law of Segregation) states that each inherited trait is defined by a gene pair. The offspring inherits one allele from each parent when the sex cells unite in fertilisation.

The Second Law (the Law of Independent Assortment) states that genes for different traits are sorted separately from one another so that the inheritance of one trait is not dependent on the inheritance of another. Finally, the Law of Dominance states that an organism with alternative forms of a gene will express the form that is dominant.

Sadly for Mendel, his extensive and meticulous work went largely unnoticed or misunderstood until the 1900s. There are a number of critical contributors to the history of genetics, but there isn't the space in this article to highlight them all. Numerous individuals expanded on the work of previous contributors to describe and prove the basis of heredity as we now know it. I will outline a few of the key players.

Essential contributions in the 1860s were made by Walter Flemming, who discovered chromosomes (although that term wasn't used until later). He recognised that chromosomal movement during mitosis offered a mechanism for the distribution of nuclear material during cell division. In 1969, Friedrich Miescher, a Swiss biochemist, was the first to isolate DNA inside the nuclei of human white blood cells. He named it 'nuclein', but later changed it to 'nucleic acid' and, finally, deoxyribonucleic acid (DNA).

Phoebus Levine, a Russian biochemist, made several breakthroughs in 1919. He was the first to discover the order of the three major components on a single nucleotide (phosphate-sugar-base), the first to discover the carbohydrate component of RNA (ribose), the first to discover the carbohydrate component of DNA (deoxyribose) and the first to correctly identify the way RNA and DNA molecules are put together.

In 1944, Oswald Avery (among others) reported that DNA was the substance that transferred genetic material. Erwin Chargaff, another biochemist, was able to draw two major conclusions in 1950:

- that nucleotide composition of DNA varies among species; and
- the total number of purines (adenine and guanine) in a DNA molecule is always equal to the total number of pyrimidines (thymine and cytosine) – this is known as Chargaff's Rule.

Chargaff's Rule and Rosalind Franklin's work on x-ray diffraction studies of DNA, which provided images of the helical structure of DNA fibres in 1951, were pivotal in helping James Watson and Francis Crick to determine the molecular structure of DNA in 1953.

From that point in history, the developments in our understanding of human genetics have been rapid and numerous. Most publicised, perhaps, is the Human Genome Project that told us that we have approximately 30 000 genes on our chromosomes (far fewer than we suspected) and that mutations in our DNA are very common. Some mutations have an effect on an individual, whereas others seem to be inconsequential. Understanding variations in the genome and their affects gave rise to genomics and this seems to be where the future of genetics is heading. Let us turn for a moment to where all this work fits into practice.

Patterns of inheritance

Following Mendel's work, we have the general patterns or rules of inheritance. Of course, we need to remember that sometimes rules are made to be broken. The work above means we now know that humans carry 46 chromosomes in 23 pairs (one of each from our mother, one of each from our father). Of these 23 pairs, the first 22 pairs are the same regardless of sex and the 23rd pair determines the sex. Two X

chromosomes make a female, an XY makes a male. In essence, having a Y chromosome makes one male and the lack of a Y results in a female. The first 22 pairs are known as 'autosomes' and the final pair as 'sex chromosomes'.

Autosomal dominant inheritance

Autosomal dominant (AD) inheritance means that the gene is carried on one of the autosomes, so affects males and females in equal proportion, and means that an alteration of one gene is sufficient to cause disease. Although the diagnosis is the same in family members with AD conditions, the disease can be very variable within families. Some individuals may have a mild form of disease, whereas others can be severely affected. Even siblings can vary in their presentation. This can make counselling rather complex for practitioners and families, particularly with prenatal diagnosis, as predictions of disease severity can be difficult.

Some clues that the patient's family may have an AD disease are: there are approximately equal proportions of males and females affected, each generation has affected individuals, and all forms of transmission through the generations are seen (male-male, male-female, female-female, female-male). Some examples of AD disease often encountered in practice include Marfan syndrome, myotonic dystrophy, BRCA 1 breast cancer and Von Willebrand disorder.

Autosomal recessive inheritance

Autosomal recessive (AR) inheritance means an individual needs to carry two mutated copies of the gene to exhibit the disease. We all carry a number of recessive mutations that remain hidden in the family. Most carriers of recessive conditions have no symptoms of the condition and, importantly, no family history, as the disease is only revealed when the carrier meets another carrier. Generally speaking, AR conditions are often severe in nature and present similarly within a family. In each pregnancy, the chance of having a child affected with the disease if both parents are carriers is 25 per cent. There is a 50 per cent chance (with each pregnancy) of producing a carrier, and a 25 per cent chance of having an unaffected child (who is also not a carrier).

Common AR conditions include cystic fibrosis (CF), congenital adrenal hyperplasia (CAH) and phenylketonuria (PKU). Interestingly, given medical advances, it is now not unusual to encounter a pregnant woman who is affected with CF (or CAH or PKU) herself. Obviously, the chance of her offspring being affected with any of these disorders is dependent on her partner's carrier status. However, each of her offspring will be carriers, regardless of sex.

Sex-linked conditions

Sex-linked conditions, as the name suggests, are carried by either the X or Y chromosome. X-linked conditions have been given more press and attention over the years, owing to the sheer number of genes, and therefore disorders, linked to the X chromosome. Traditionally, we've talked of conditions where the mother is the carrier and her male children exhibit symptoms of the disease. Female carriers of an X-linked condition have a 50 per cent chance of their male children being

affected with the disease. Female offspring will have a 50 per cent chance of being carriers of the condition.

It is not unusual for a carrier of an X-linked condition to have some symptoms of the condition itself over time. Carriers of haemophilia, for instance, can also have altered clotting factors. However, the symptoms are generally far less severe than in affected males. The reason for this phenomenon is X-inactivation.

X-inactivation is a process whereby one of the female's X chromosomes is switched off. Generally this is a random process in each cell, so either the paternal or maternal X is silenced. Skewed X-inactivation can also occur, whereby a disproportionate amount of either the maternal or paternal X chromosome is silenced. If the functionally normal gene is switched off in higher proportions or in cells reliant on the function of that gene, symptoms of the diseased gene will appear. Common X-linked conditions include haemophilia and Duchenne muscular dystrophy.

An interesting X-linked disease is Fragile X syndrome. Fragile X syndrome is the most common form of inherited intellectual impairment. The FMR-1 gene is composed of a triplet repeat sequence (CGG) that, when expanded to a critical level, impairs production of a protein vital for brain development. Fragile X was the first time we saw that healthy males could in fact pass on the condition to their daughters (who are all carriers). Previously, the assumption was that if a male was asymptomatic for a condition, he was unaffected and therefore would not pass the condition on. The condition then follows the general rules of X-linked inheritance in that the carrier daughters have a 25 per cent chance of having an affected son in each pregnancy and a 25 per cent chance of having a carrier (or affected) daughter in each pregnancy. Importantly, female pre-mutation carriers of Fragile X syndrome have a 20 per cent chance of premature ovarian failure before the age of 40.

Both X-linked recessive and X-linked dominant conditions exist. In very general terms, female carriers of X-linked recessive conditions are less likely to exhibit symptoms (although these include muscular dystrophy and Fragile X syndrome). X-linked dominant inheritance is very rare, with Rett syndrome being one example.

Further study on the Y chromosome and its relationship with disease came to the fore during the new millennium. Micro-Y deletions can cause lowered fertility or infertility in the male. Intracytoplasmic sperm injection (ICSI) can be performed as part of an IVF cycle to allow the individual to have biological children. It should be noted that all his male children will also have the deletion and subsequent infertility.

Other modes of inheritance

There are other modes of inheritance, not originally described by Mendel, but nonetheless very important.

Chromosomal inheritance

Approximately 1:500 individuals carry a balanced translocation in their chromosomes. This means that although they have all the genetic material and the standard number of chromosomes, they don't carry the information in the standard way. In those carrying a balanced reciprocal translocation, a piece of one chromosome has broken off and swapped positions with another piece from another chromosome. For the individual, problems tend to only arise when they attempt to have children. The implications depend on how large the translocated pieces are, but in practice the carrier may have issues with fertility or recurrent pregnancy loss, as the fetus inherits an unbalanced rearrangement (either too much genetic information or not enough). In some cases, offspring are born with a number of medical issues, including major malformations and mental impairment, owing to the unbalanced material.

Carriers of Robertsonian translocations have 45 chromosomes, but actually have the full complement of genetic material. Robertsonian translocations occur when there is a fusion at the centromere of two acrocentric chromosomes. In humans, it is seen most commonly involving chromosomes 13, 14, 15, 21 and 22. Other forms do not lead to a viable outcome. Robertsonian translocation carriers are also at increased risk of infertility, miscarriage and children with unbalanced chromosomes.

Mitochondrial inheritance

While most of our genes are contained on the chromosomes inside the nucleus of the cell, some genes are located in the mitochondria of the cell. The mitochondria and the DNA inside it are passed on through the mother's eggs. In simple terms, the role of mitochondria in the cell is to produce energy for the cell and therefore the rest of the body. The amount of mitochondria in each cell is variable.

A mitochondrial mutation can result in biochemical problems, owing to the absence or impairment of enzymes involved in the respiratory chain. This leads to a reduction in the supply of the energy source adenosine triphosphate (ATP) that drives various reactions essential for function and growth.

Generally mitochondrial disorders are progressive and often crippling, involving many body systems. Counselling is complex, owing to the diversity of disease and variability within families, due to the variable mitochondrial load in each cell.

What else, what next?

In addition to the basic inheritance patterns, we cannot forget the interaction between genes and environment. There are a number of conditions that are due to multifactorial inheritance and there are likely to be many more reported as we enter the age of genomics. We know that some mutations confer a susceptibility to a disease and that other factors can modify that risk further. One dramatic example of this is the reduction in neural tube defects with an increase in folate consumption.

Epigenetics occurs when there are heritable changes to the gene expression, without causing a change in the DNA itself. Epigenetic changes alter the way a gene is switched on or off. Genetic imprinting and X-inactivation are the best examples of epigenetics and disease, instances of these types of disease include Prader-Willi syndrome and Angelman syndrome.

Looking to the future, genomics is now moving to the fore with the advent of whole genome sequencing (WGS) and whole exome sequencing (WES). These advances are likely to bring both exciting and daunting changes to genetics and medicine. While a patient's entire genome can be sequenced relatively easily, we are yet to understand all the variations that are found. Variants of unknown significance will be revealed as will unexpected findings. The complexity of understanding and explaining that information could be difficult, but not insurmountable!

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A Breakthrough Treatment For Vulvo-Vaginal Atrophy And Dyspareunia

Non-Surgical, Non-Hormonal Treatment With No Downtime

MonaLisa Touch is a five minute, non-hormonal treatment for symptoms often related to Vulvo-Vaginal Atrophy. There is no downtime following the procedure and it is suitable for patients who suffer from symptoms including:

- Vaginal Dryness
- Dyspareunia
- Loss Of Lubrication
- Vaginal Itching
- Vaginal Burning
- Vaginal Laxity

The treatment can be used in conjunction with Hormone Therapy, as an alternative treatment for patients who are not suitable for Hormone Therapy, or for those patients who have found Hormone Therapy to be ineffective.

Histological preparation of vaginal mucosa sections stained with haematoxylin and eosin (H&E)

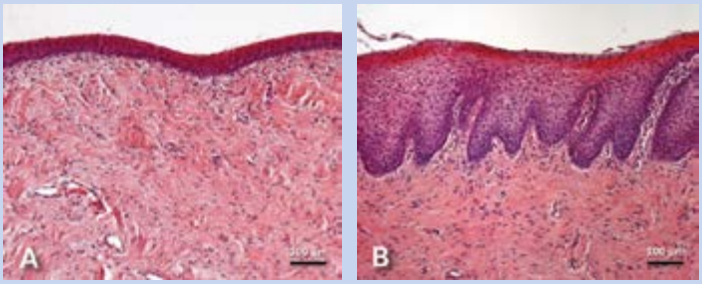
Patient data
Age: 59
Age at menopause: 48

Note: The patient was not treated with HRT.

Courtesy of: Prof. A. Calligaro, MD Professor of Histology and Embryology at the University of Pavia, Italy

(A) Vaginal mucosa in the basal condition with a thinner epithelium typical of atrophic vaginitis. Never treated with HRT.

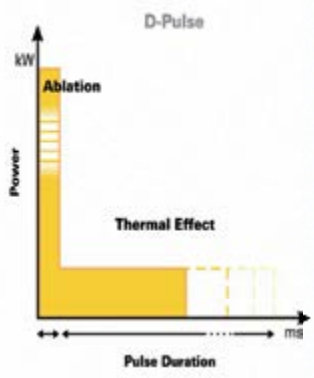
(B) Same magnification two months after one MonaLisa Touch® session showing significantly thicker epithelium of the mucosa.



Histological Observations Post-Treatment [2]

- A thicker (non-atrophic) epithelium with a basal layer of closely packed (proliferating) cells.
- Significant storage of glycogen in the large epithelial cells forming the intermediate and superficial layers.
- A high degree of epithelial exfoliation of superficial cells filled with glycogen.
- Increased synthesis of the molecular components of the extracellular matrix.
- A rich content of blood vessels in the connective tissue stimulating and supporting the activity of fibroblasts and capillaries.

Technology Unique To MonaLisa Touch™



The Deka Pulse is unique to MonaLisa Touch and is required to achieve tissue changes observed in the published research on MonaLisa Touch. [1, 2, 3]

This feature maximises the treatment effect while minimising patient downtime.

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A user's guide to the human genome

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The human genome can be described by considering its origin. For sexually reproducing species, the genome originates at the moment of conception; for humans that is when the 23 chromosomes from the egg cell of the mother combine with the 23 chromosomes from a sperm cell of the father. Together, those 46 chromosomes make up the human genome. Each chromosome is made from DNA, which is a polymer comprising nucleotides represented by the letters A, T, C and G. Our genome contains more than six billion of such letters, divided between the 46 chromosomes.

Our genome contains all the information required by life to program the single-celled embryo that is created at the moment of conception to divide in a perfectly coordinated fashion into hundreds, thousands and, eventually, billions of cells as the fetus develops in utero into a human being. Every tissue and structure in an individual's body – such as blood cells, muscles, nervous system, heart, lungs, kidneys, bones – are all programmed and operated by the information in their genome. The remarkable precision of this program is dramatically illustrated in cases of identical twins, where it can run with almost complete reproducibility – not just eye and hair colour, but even the finest details of how our bodies develop, age and respond to disease and the environment; including, for example, where and when our hair greys, the complexion of our skin and where we accumulate fat and muscle.

Genomic variation

The information in our DNA makes us different. Virtually every disease, or our response to disease, is encoded in our genomes. Although the majority of the genome among humans is identical, of the six billion letters of information, approximately three million differ between two unrelated individuals. Among these millions of differences or 'variants', the majority are considered 'benign' because they have no damaging effect in the way our bodies work and are simply part of what creates the variation in our physical and biochemical make up. However, a small number of such variants can cause or predispose to disease. These 'pathogenic' variants may change the meaning of the genetic instructions by altering or removing letters in a segment directly translated by the cell, or a region that regulates gene activity. These changes can cause a cell or tissue to work poorly or not at all.

Genetic variation can involve a single DNA nucleotide or lead to the loss or rearrangement of huge chunks of DNA. Techniques such as karyotyping and fluorescent in-situ hybridisation have provided high-level resolution of large chromosomal duplications or rearrangements. Chromosomal microarrays have provided information about deletion and duplication of large segments of DNA, or copy number variants (CNVs). Genomic sequencing is now able to provide high-resolution information on small and large variants – including substitutions, deletions, insertions and complex rearrangements, such as inversions of large pieces of DNA.

We previously referred to benign and pathogenic variants as polymorphisms and mutations, but it has become clear over recent years that these lie on a continuum. Our understanding of the genome is still limited: there are many variants of unknown significance (VUS) that lie between our knowledge of benign and pathogenic. To complicate matters further, the severity or frequency of disease (so-called penetrance) that manifests from even well-characterised pathogenic variants can also be highly variable between individuals.[1] This variable penetrance further confounds the interpretation of genomic information and limits the potential to predict disease from genomic information alone. Accordingly, accurate genomic interpretation is undertaken in the context of family history and patient presentation. Nevertheless, despite these complications, our existing knowledge of genomic information can already have powerful applications in clinical practice.

Revolutions in genetics

Our knowledge of the human genome builds on major leaps in the 19th and 20th centuries, particularly those of: defining the principles of heredity; resolving DNA's double helix structure and the genetic code; and developing the first sequencing chemistries that could determine the order of nucleotides along a length of DNA. However, the last two decades have seen an unprecedented leap forward in information and insight into the human genome.

More than ten years ago, the 'reference' human genome sequence was published: a virtual tome that provided global infrastructure for biology.[2] As a result, researchers around the world could access a common collection of genomic data and

overlay information about DNA variation and function. Scientists could begin to compare DNA sequences between humans and other organisms to identify sequences common to all living things.

The original reference sequences lay the foundations for genetic exploration of the approximately 21 000 genes in the human genome, which act as the templates for producing proteins and other molecules. However, the smaller-than-expected number of genes hinted at the hidden complexity of the human genome and the regulatory mechanisms that affect the activity of genes. The observation that many of the conserved regions lay in areas between genes – sequences not directly involved in producing proteins – also suggested that protein-coding genes were not the only vital components of the genome.

Decoding the genome

International projects in the last ten years have attempted to add flesh to the bare bones of the genome sequence. The introduction of increasingly high-throughput technologies, such as microarrays and automated sequencing, provided a means to capture snapshots of genetic activity, which led to more detailed descriptions of which parts of the genome are used in different cells and at different times. These deeper explorations of the human genome led to a number of unexpected observations, such as that a single gene can be differentially spliced to encode multiple forms of a protein that have subtly different functions and the identification of genes that do not encode proteins at all, but rather function to produce other classes of regulatory molecules (referred to as noncoding RNAs).

As research into the activity of the genome has blossomed, so has research into the 'epigenome', which are tissue- and cell-type modifications to the biochemical environment of the DNA or direct chemical modifications to the DNA itself. These modifications, in turn, affect the activity of the genome and, ultimately, define how the genome behaves in different cells. Each new study answers more questions about how different cells in our body use the information encoded in the genome and how our characteristics are mediated by the environment we inhabit.[3]

As advances in sequencing technologies accelerated in the mid-2000s, the genomic data available to researchers grew from a trickle to a flood. These technologies were employed to create catalogues of variation in different human populations[4] and thousands of people's genomes from a cross-section of cultures have now been sequenced and released as global resources.[5,6] Lower sequencing costs have also enabled researchers to ask health-related questions on a massive scale: some studies have catalogued mutations that drive a tumour's growth by comparing the genomes of tumour and normal cells. Others have focused on mapping the functionality of significant proportions of the human genome or seeking DNA differences between people with and without complex diseases.

These projects have provided a far more

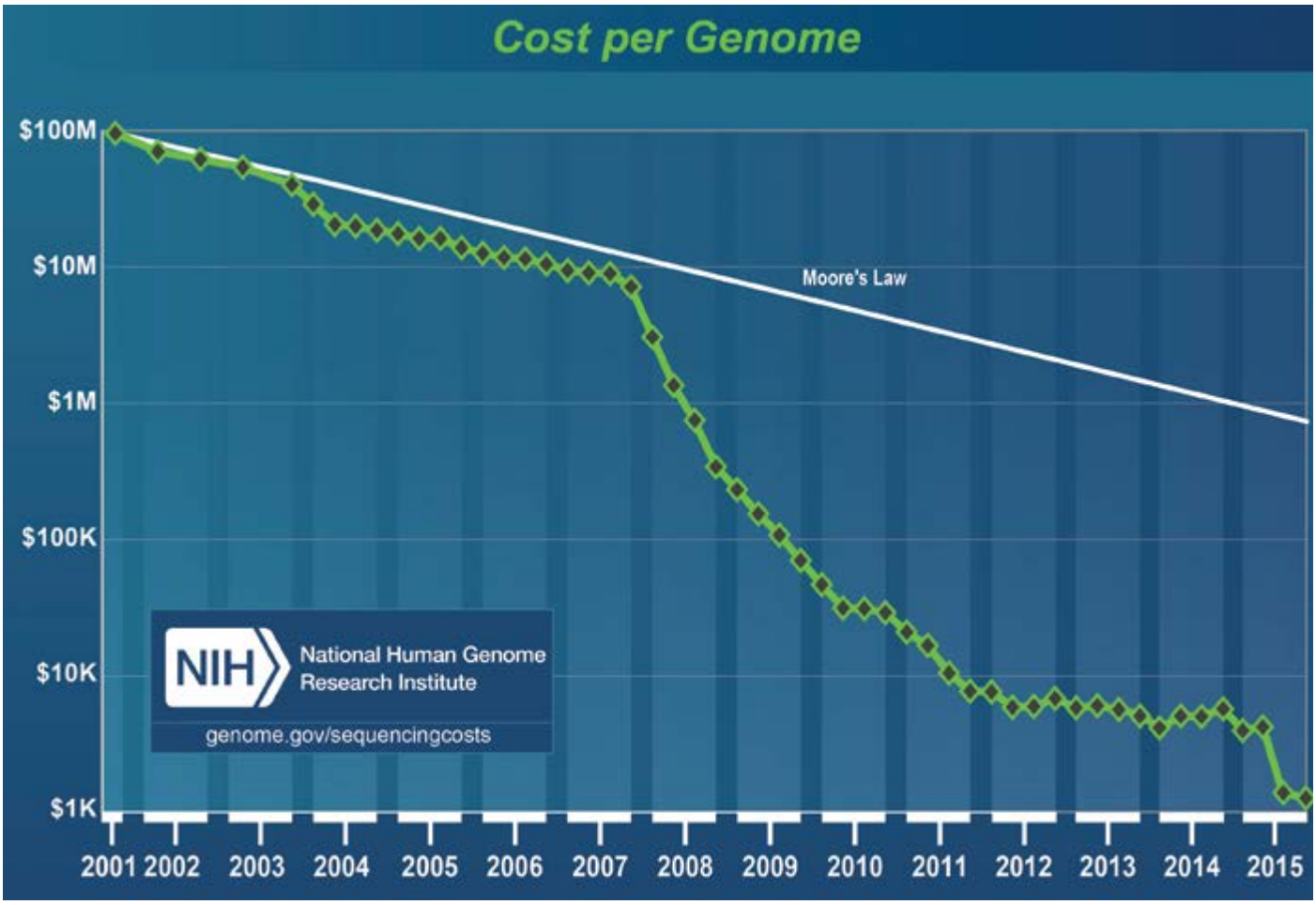

Figure 1. The cost of genome sequencing has declined at a hyper-exponential rate over the past decade, exceeding the rate of advance in semiconductor technology as predicted by Moore's Law. Credit: Wetterstrand KA. DNA Sequencing Costs: Data from the NHGRI Genome Sequencing Program (GSP) Available at: www.genome.gov/sequencingcosts. Accessed 04/2016.

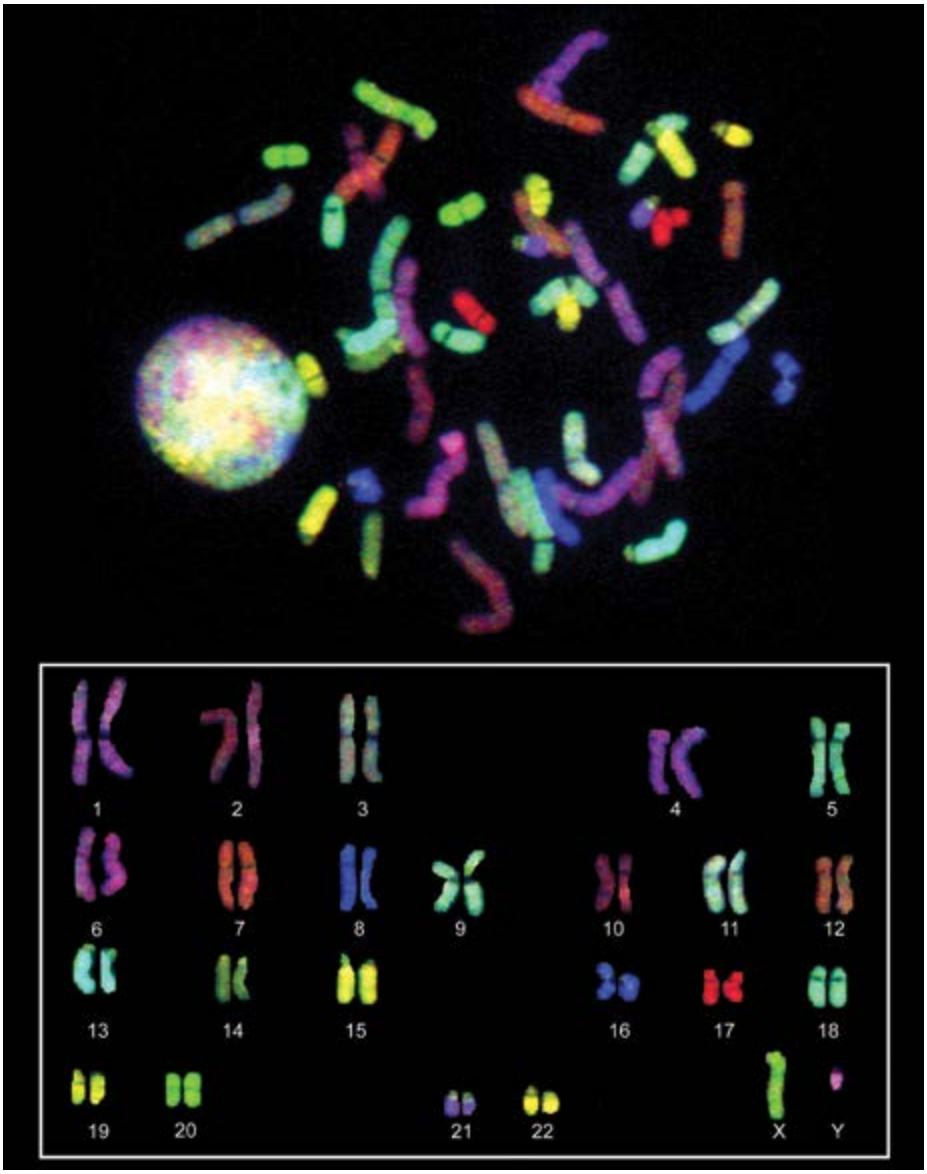

Figure 2. A spectral karyotype of chromosome pairs. Credit NHGRI, www.genome.gov.

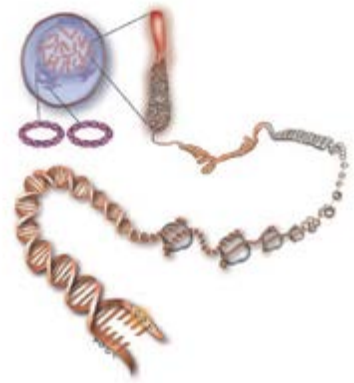

Figure 3. Chromosome organisation. Credit: Kate Patterson/Garvan.

Genome organisation

We each have about two metres of DNA crammed into the nuclei of our cells. The 3D structure of the human genome goes far beyond DNA's double helix shape. When cells divide, the DNA is tightly wound around protein scaffolds into structures called chromosomes. The DNA is coiled systematically around histone proteins, packed into fibres and tied up into scaffolds with specific organisation. Throughout the cell cycle, threads of DNA coil and uncoil, depending on which pieces of genetic information are being used.

Mitochondrial DNA (mtDNA) sits outside the nucleus in a cell, and is inherited solely from the mother. Numbers of mitochondria in a cell vary, as do the numbers of copies of mitochondrial chromosomes, but each cell is thought to carry on average 1000 copies of the mtDNA, contributing around 0.1 per cent of total cellular DNA.

Anatomy of a genome

Although definitions vary according to the scientific circumstance, the human genome is typically defined as the full complement of genetic material in a cell. In cells other than germ cells (and mature red blood cells that lack most cellular organelles), genetic material is organised into 23 pairs of chromosomes inside the nucleus, and in the many copies of a small ring of DNA inside the cell's energy-producing mitochondria.

The nuclear chromosomes are numbered in approximate order of size – from 1 to 22 – with the sex chromosomes (X&Y). One of each chromosome pair is inherited from an individual's mother, and the other one from their father.

nuanced view of genomic variation across the genome. Genetic variants across the genome may be inherited, induced by environmental factors, or arise from errors made while copying DNA for cell division.

Following a decade of rapidly declining costs of genomic sequencing, in parallel with increasingly sophisticated knowledge of how the genome works, genome sequencing is now making a transition from an almost exclusively research-based discovery tool to a routine clinic-based diagnostic tool.

Clinical implications of sequencing

The value of genome sequencing in the clinic – known as genomic medicine – is in using timely genetic diagnoses and better characterisation of disease to provide critical information to doctors and families. Information that predicts an individual's risk of developing a disease, or their response to treatment, can optimise medical practice.

Genomics is already affecting clinical practice through the diagnosis of heritable disease.[7] Whole-genome sequencing (WGS) is transforming the diagnosis of thousands of rare, usually paediatric, diseases. Even for diseases that have not been described before, unbiased WGS can provide information that makes it possible to diagnose and define new genetic diseases.

Genomics is also starting to change oncology, by guiding the management and

treatment of cancer. Cancer is ultimately a genetic disease: there are many ways that our genome can be mutated to cause cancer. Traditionally, cancer was treated according to its tissue of origin, but by understanding the genetic basis, treatment can be prescribed to target the specific molecular pathway individualised to that cancer.[8] This 'precision medicine' approach attempts to apply the right drug in the right dose to the right patient at the right time.

Beyond diagnosis and guiding patient management, the most profoundly transformative application for genomics in healthcare is anticipated to be in disease risk stratification, early disease detection, and, ultimately, prevention of disease occurrence. The more we learn about what the information in our genomes means for health, the more accurately we can predict our health futures. As well as detecting disease predisposition, genomics also has the potential to anticipate adverse drug reactions, assess carrier status for recessive disease and predict the body's response to different sorts of environmental exposures. The collective value of the health information derived from the genome is anticipated to eventually reach a tipping point where it will be advantageous for every individual to undergo genome sequencing and to incorporate this information as part of their health record.

The future

By reading the information that forms the very essence of our bodies, genomics reveals the inner workings of our cells, in both health and disease. The power of sequencing derives from volume and the capacity to be able to compare newly sequenced genomes with many others to pinpoint the relevant variations. Genomic information will only gain interpretive power as it is combined with information about patients' observable characteristics (their 'phenomes') and as more variants are understood and classified. Genomics is having an increasing impact in the clinic; and has the potential to reconfigure the nature of the interaction between the patient and the practitioner.

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Epigenetics 101: what it is and why it's important

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Knowing the human genome sequence does not automatically mean we have the information required to fully understand human biology and disease.[1] Rather, human development and disease are the result of complex processes that emerge and are modified by interactions between an individual's genotype and the environment. An individual's genotype is complex and, contrary to popular opinion, is not fixed. Instead, there is considerable variation owing to copy number variation, deletion and insertion within an individual's non-inherited somatic cells.[2] Moreover, a significant contribution to the overall dynamics of an individual's genome comes from the microbes that make up the microbiome.[3] So, how does the environment interact with the genome?

Environment-genome interactions occur through numerous mechanisms: epigenetic, metabolic or even directly by physical mechanisms[4], all of which are modulated by the set of genetic variations present in the individual's genome. Of course, different combinations of these environment-genome interactions occur over the entire life-course of an individual and while they are important modifiers of disease risk at all stages of life, the signalling that occurs during early development (including pregnancy, birth and early infancy) sets the scene and may be a primary driver of later disease risk.

In the 1940s, Waddington introduced the concept of epigenetics to describe the interaction between the development of a phenotype and the sum of genetic expression and environmental (tissue) interactions.[5] Epigenetics, as a term, has since been defined and redefined and has a certain degree of ambiguity associated with it.[6] Throughout this article we will discuss epigenetics in terms of changes in gene function that cannot be explained by changes in DNA sequence. Prions and non-coding RNAs are considered to be epigenetic. However, we will focus on post-translational modifications to DNA and histones that cause a change in gene expression and form what is known as the epigenetic code (see Figure 1).[7]

The epigenetic code is made up of a series of modifications to DNA and proteins related to the control of DNA transcription (for example, histones; see Figure 1) that don't change the underlying sequence of the DNA, but rather they alter the way a genome functions by changing how accessible it is to the enzymes that are necessary to decode it.[6] The action of security guards at a factory can be used as an analogy to illustrate this. Specifically, at the beginning and end of each day, security guards lock and unlock doors in response to signals from their boss (the environment). In this way, they limit the areas of the factory that the workers can access – just like epigenetic marks on the genome. If the security guards are instructed to change their routine then they can alter worker access to store rooms and, in so doing, alter the final products that are produced by the factory.[8] Such a change can be positive or negative by promoting the inclusion of inferior parts, or even absence of parts, that limit the lifetime of the final products.

Epigenetics and the Liggins Institute

Increasing epidemiological and experimental evidence links perinatal factors to later cardiometabolic disease risk – a phenomenon preferentially termed 'developmental programming'. This is consistent with the evidence that both fixed genomic and epigenetic variation contribute to later disease risk. Despite this, our understanding of the underlying mechanisms remains poorly defined. Data from human cohorts and animal models suggest that epigenetic processes are an important link between the early-life environment, such as maternal diet, and altered metabolism and body composition in offspring in later life.[9]

There is a strong record of research at the Liggins Institute into the epigenetic processes that underpin developmental programming. As an example, a longstanding and highly productive collaboration between the Liggins Institute and the University of Southampton, UK, demonstrated the importance of the developmental contribution to later adiposity by showing that methylation (see Figure 1) of the retinoid-X receptor-alpha (RXRA) gene at birth was associated with the child's later adiposity.[10] Subsequent work has also identified a link between methylation of the RXRA gene at birth and childhood bone mineral content.[11] These

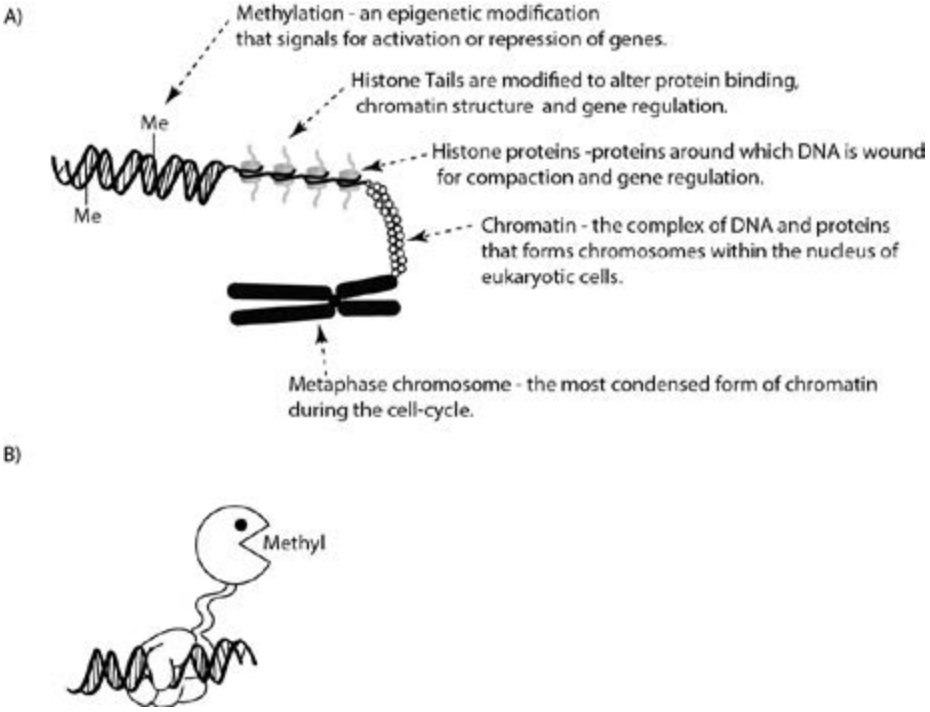

Figure 1. A) Cartoon illustrating features of DNA organisation. An epigenetic modification in the form of methylation (Me) is shown on the double-stranded DNA. Histone tails are subjected to a number of post-translational epigenetic modifications. B) Epigenetic engineering uses composite enzymes that have 1) a DNA binding domain (hand; for example, an enzymatically inactive CAS9 [reviewed in[26]]) which binds to a specific DNA sequence and 2) an enzyme component (Pac-Man™) that catalyses a specific epigenetic modification (for example, methylation) to turn on or off a specific gene(s).

findings were replicated in independent cohorts, highlighting the robustness of the observations. Interestingly, recent observations have identified a genetic and epigenomic component to post-term birth that may contribute to the increased risks of subsequent life-long complications for these individuals (Prof Wayne Cutfield, Liggins Institute: personal communication). Research at the Liggins Institute has also examined epigenetic profiles in twin births and the potential for epigenetic effects in offspring arising as a result of IVF. Work by Miles et al showed that IVF resulted in altered methylation of genes that appeared to favour childhood growth and metabolism.[12] Further, in addition to outcomes linking the early life environment and later obesity and cardiometabolic disorders, work undertaken at the Liggins Institute has also examined effects on later cancer risk and potential mediation by epigenetic processes. Of note, Perry et al showed that similar epigenetic mechanisms underpinned both cancer tumour progression and implantation in human pregnancy.[13] Collectively, these studies are consistent with a substantial component of metabolic disease risk having a prenatal developmental basis. Therefore, perinatal epigenetic analysis may have utility in identifying individual vulnerability to later obesity and metabolic disease.

Preclinical models have enormous potential for examining epigenetic mechanisms underpinning developmental programming, transgenerational effects and also opportunities for intervention. Small and large animal models of altered early-life nutrition and direct cell-based platforms have been used by Liggins Institute researchers to characterise a number of epigenetic mechanisms associated with a poor start to life. Rodent models have shown that maternal obesity leads to hypomethylation and enhanced activity of an important regulator of cell growth (in other words, the cell-cycle; P21 gene). Hypomethylation of P21 is present in offspring of obese mothers at the time of birth and likely primes the hepatic dysfunction and later steatosis observed in these animals.[14]

In sheep, periconceptional undernutrition causes epigenetic changes in offspring in key hypothalamic genes related to energy expenditure (for example, proopiomelanocortin and neuropeptide Y) and glucose homeostasis.[15,16] Work by Frank Bloomfield and colleagues has shown that these epigenetic changes are likely to explain the predisposition of these lambs to become obese and suffer from related metabolic disorders.[15,16]

Changes in developmental trajectory arising from developmental programming were once thought to be permanent. However, research by one of the authors (Mark Vickers) and colleagues was among the first to show that the effects of a poor start to life were indeed reversible when intervention strategies were targeted to particular critical times during development.[17] As an example, treatment of neonates with leptin was shown to be able to reverse the adverse metabolic sequalae in rodent offspring following maternal undernutrition. Importantly, the leptin treatment effects highlighted that metabolic plasticity in early development was both sex-specific and directionally dependent upon prior maternal nutritional status.[18,19] These observations have now been replicated in a series of independent studies around the world, including in other model species.

Biological data generated by '-omics' platforms are commonly confounded by the nature of sample frequency distributions, which can adversely affect the accuracy of functional inference between biological measure and phenotype.[20] Researchers at the Liggins Institute are undertaking different approaches to address this issue. Firstly, Allan Sheppard and colleagues have developed statistical tools that enable improved confidence of functional inference and, in the setting of epigenomic data, also reveal otherwise cryptic information.[20] Secondly, novel methods have been developed by one of the authors (Justin O'Sullivan) and colleagues that enable the visualisation of the epigenome in three-dimensions.[21] Crucially, this work is identifying spatial associations that help to explain how the information within the genome is accessed to ultimately result in the phenotype that we observe.[22]

Although most work in the area of epigenetics has focused on changes in DNA methylation, recent work has also examined the role of microRNAs (miRNAs) and histone modifications in gene control. Allan Sheppard and colleagues recently described specific miRNAs associated with steatosis/non-alcoholic fatty liver disease and highlighted biomarkers that may underpin diagnostic profiles and thereby identify at-risk individuals

without the need for invasive biopsy, for example, the use of blood plasma.[23] Consistent with this, one of the authors (Mark Vickers) and his colleagues have shown that growth hormone treatment of maternal malnutrition-induced hypertension and cardiac hypertrophy in offspring is associated with changes in the Let-7 family of miRNAs.[24] Notably, these Let-7 miRNAs have been previously linked to inflammation and cardiovascular development.

Epigenetics and obesity

The role of epigenetics in predisposing individuals to obesity in later life is still being determined. However, work completed to date has a number of important implications. Firstly, the effect sizes associated with epigenetics are considerably greater than those associated with birthweight or maternal body composition.[10] This suggests that epigenetic measurements made in the maternal tissues/neonate may be useful predictors of later obesity and other phenotypic outcomes. Secondly, the association between CpG methylation and a child's later adiposity operates within the normal ranges of maternal nutritional state and birth size. This supports the argument that developmental programming is the consequence of an evolved and potentially adaptive process involving the mechanisms of developmental plasticity.[25] Indeed, the data provide strong evidence supporting a role for developmental plasticity in determining individual risk of metabolic disease. Thirdly, the epigenetic data suggest that developmental factors may make a much more significant contribution to phenotypic variation and disease risk than is generally considered. Despite the fact that many of the studies that have been performed to date were limited to identifying correlations and not causative interactions, epigenetic modifications hold great promise across a wide range of metabolic disorders as early biomarkers to predict at-risk individuals.

The future: epigenetics in O&G

The use of drugs that inhibit methyltransferases (for example, azacitidine, decitabine) and histone deacetylases (for example, vorinostat, romidepsin) to modify specific epigenetic marks within the epigenome as therapeutic interventions is already occurring. While these approaches are useful as part of combination therapies, they can have significant side effects as individual treatments. These side effects can be overcome by the development of epigenetic engineering approaches that use new, targeted techniques to manipulate the epigenetic marks at known genes (see Figure 1[26,27]). Such approaches have applications across a wide variety of clinical research domains (for example, cancer, protein aggregation diseases, metabolic diseases, neurological and psychiatric diseases) and metabolic diseases where manipulating the expression of a single or small subset of genes can overcome

Box 1. A glossary of terms

Copy number variation – the number of copies of a piece of DNA varies from one cell to the next.
Development – the process of growing to maturity.
Developmental programming – a stimulus or insult operating at a critical or sensitive period of development that can result in a longstanding or life-long effect on the structure or function of the organism.
DNA – a chemical contained in our cells that carries information to build the body. DNA itself has a double helix structure: the information is encoded in the sequence of chemical units along complementary DNA strands. The chemical units are known by the first letters of their names: A, C, G and T. Segments of DNA (genes) act as templates for producing different molecules. DNA information is a unique combination inherited from the mother and father.
Epigenetics – the study of heritable traits that are not caused by changes in the DNA sequence.
Epigenome – the spatial arrangement of the total set of all the epigenetic marks that are present in a genome.
Epigenetic code – a code consisting of epigenetic marks that is dictated in response to environmental signals.
Epigenetic engineering – targeted epigenetic modification of specific genes.
Epigenetic marks – there are three basic classes of epigenetic marks:
1. post-translational modifications that occur on the proteins around which the DNA is wrapped (histones);
2. methylation of the deoxyribonucleotide bases within DNA; and
3. hydroxymethylation of DNA.

Gene – segments of DNA that encode the instructions to build molecules (usually proteins) that make our cells and keep them functioning.
Genome – all of the genes or genetic material present in cell or organism.
Genotype – the set of genetic variations present in the individual's genome. This includes all the changes from the reference genome that are unique to the individual.
Histones – a group of proteins that form a complex (the nucleosome) about which DNA is wound inside cells with nuclei.
Histone deacetylases – these remove acetyl marks (a post-translational modification) from histone proteins.
Hypomethylation – a measurable reduction in the amount of methylation at a site in the DNA or histones.
Leptin – a 'satiety' hormone made primarily by fat cells that helps regulate energy balance.
Methylation – an epigenetic modification that involves the addition of a methyl group to the DNA or histone.
Methyltransferases – enzymes that add methyl groups to their target substrate (for example, DNA or histone proteins).
Microbiome – the collection of micro-organisms present on/in an individual.
microRNAs (miRNA) – a group of small RNAs that do not code for proteins, but act to regulate genes and gene expression.
Nucleotide – the chemical units of DNA, known by the first letters of their names: A, C, G and T. Each nucleotide is made up of a nitrogenous base (for example, adenine, thymine, guanine or cytosine) together with sugar and phosphate groups. The order of As, Cs, Ts and Gs along a DNA strand encodes information; segments of DNA act as templates for producing different molecules.
Somatic cell – any non-inherited, non-reproductive cell in an organism.
Undernutrition – a deficiency of one or more essential nutrients or calories.
Variant – a DNA difference or change on a continuum from benign to pathogenic. Variants can range from large rearrangements, deletions or duplications of massive segments of DNA (copy number variants, or CNVs) right down to single nucleotide changes (single nucleotide variants, or SNVs).

programmed or genetic changes that cause changes to the phenotype of the cell or organism. However, the successful application of epigenetic engineering requires a full understanding of the genome biology in the targeted cells. This understanding must include the inter-relationships between the epigenome and genotype.[21,22,28]

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Sex chromosome disorders

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Sex chromosome disorders are one group within the classification of disorders of sex development (DSD). The two other main subcategories are the 46XY DSD group – which includes androgen insensitivity syndrome and pure gonadal dysgenesis – and the 46XX DSD group, of which congenital adrenal hyperplasia and Müllerian agenesis are examples.[1] Sex chromosome disorders may be further classified into numerical abnormalities (aneuploidies) or structural defects.[2] The latter include chromosome deletions, duplications and translocations. These defects present with a variable phenotype, which may include hypotonia, intellectual disability and seizures.

The most common sex chromosome aneuploidy in females is 47XXX (triple X), with an incidence of one in 1000. Diagnosis is usually an incidental finding, if made at all. Features include tall stature and a higher incidence of development delay. The majority have normal puberty and fertility, though premature ovarian insufficiency can occur. There is no increased risk of chromosomal abnormality in offspring. The incidence of males with a 47XYY karyotype is around one in 2000. Affected men have tall stature, delay in speech and motor development, and behavioural issues. Pubertal development is normal and most are fertile. The majority of these men also remain undiagnosed. Cases of 49XXXXY, 49XXXXX and 49XYYYY have also been reported. Severity of phenotype appears to worsen with increasing numbers of chromosomes.

Turner syndrome

Turner syndrome occurs in one in 2500 liveborn females. Karyotype may show 45XO, a 45XO/46XX mosaic, or a 45XO/46XY mosaic in a small number of cases, with or without a partial deletion of the Y chromosome. A small number have a structural abnormality of the second X chromosome, such as a deletion of the short arm. Most cases are sporadic mutations rather than inherited. The presence of Y material is associated with the development of gonadoblastoma and dysgerminoma in up to 12 per cent of cases, so prophylactic gonadectomy is indicated.

The phenotypic presentation of Turner syndrome is broad, ranging from complete ovarian failure in childhood through to an absence of clinical features and normal reproductive outcomes. The severity of features in patients with mosaicism is dependent upon the percentage of abnormal cells in relevant tissues. Features include: short stature, broad chest, webbed neck, low-set ears and micrognathia. Intelligence is usually normal, but there may be difficulties with spatiotemporal processing.

Associated abnormalities are common. Cardiac abnormalities occur in one-third of cases and include bicuspid aortic valve, aortic coarctation and aortic dilatation with risk of dissection. Renal anomalies, such as horseshoe kidney, occur in around 50 per cent of cases. Hearing impairment, otitis media, scoliosis, hypertension, osteoporosis, diabetes and coeliac disease are also more common. Screening for these conditions is part of the initial management.

Treatment of Turner syndrome includes growth hormone where indicated to achieve maximal height. If spontaneous pubertal development does not occur, pubertal induction is required, with ongoing hormone replacement in adulthood. Spontaneous ovulation and pregnancy can still occur, so contraception is advised. There is an increased risk of adverse pregnancy outcome, including a two per cent risk of maternal mortality from aortic dissection. There is a higher incidence of chromosomal abnormality in offspring, including Turner syndrome and Trisomy 21. Lifelong follow up by a multidisciplinary team is warranted.[3]

Klinefelter syndrome

Klinefelter syndrome affects one in 500 to 1000 liveborn males and is the most common congenital cause of primary hypogonadism. The 47XXY karyotype is a result of nondisjunction of the sex chromosomes during meiotic division and can be maternal or paternal in origin. Male infants are phenotypically normal and diagnosis is often delayed until the male is investigated for infertility. Tall stature occurs secondary to an increased length of the long bones of the leg owing to testosterone deficiency.

Other features include small, firm testes, gynaecomastia and very low sperm counts or azoospermia. Lutenising hormone (LH) and follicle-stimulating hormone (FSH) are elevated and testosterone levels are lower than normal. There is a higher incidence of cryptorchidism, which further affects these parameters. There is an increased incidence of mental health disorders, autism spectrum disorder and social difficulties, marked by a lack of insight, poor judgment and an impaired ability to learn from adverse experience.[4] There is impairment in higher levels of language competence and difficulty in sustaining attention without impulsivity.

Klinefelter syndrome is also associated with an increased risk of other morbidities, including the following:

- pulmonary disease, such as bronchiectasis and emphysema;

- cancer, specifically breast cancer and non-Hodgkin lymphoma;
- systemic lupus erythematosus; and
- diabetes.

The mortality from breast cancer in this group is much higher than in the general population. Treatment is with testosterone replacement, which results in significant improvement in quality of life. Fertility may be achieved by testicular aspiration and intracytoplasmic sperm injection (ICSI). There is a higher incidence of chromosomal abnormalities in the offspring of men with Klinefelter syndrome.[5]

Mixed gonadal dysgenesis

Mixed gonadal dysgenesis is characterised by asymmetric reproductive anatomy. There is usually a normal or dysgenetic testis and Wolffian ducts on one side, and a streak ovary with underdeveloped Müllerian structures on the other. All patients have a Y chromosome, most commonly with a mosaic karyotype of 45X/46XY. There is a variable phenotype of genital atypia, which reflects the level of function of the dysgenetic testis. Half of patients have ambiguous genitalia at birth, which may be asymmetric with an enlarged labioscrotal fold on the side of the testis. Asymmetry of external genitalia should raise suspicion of this condition.

The gender of rearing is dependent on the external genitalia and capacity for future coital function. The majority of people affected are raised as female. Once gender has been assigned, the gonad that conflicts with the assignment is removed. The risk of a germ cell tumour is around 30 per cent in the setting of a dysgenetic gonad and a Y chromosome. The risk is higher if the gonad is within the abdomen compared with the scrotum. Intra-abdominal dysgenetic gonads should be removed at the time of diagnosis. A dsygenetic testis that has descended in a child with a male sex assignment may be left in place, with regular surveillance for tumour development.[6]

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Summary

Sex chromosome disorders are a group of conditions within the DSD classification, and include both numerical and structural abnormalities. There is a broad range of severity of phenotype, depending on the number of additional chromosomes or extent of the structural defect. Management is individualised, depending on clinical features and presence of other morbidities. Removal of dysgenetic gonads containing Y material is indicated, owing to the significantly increased risk of malignancy.

Genetic counselling couples before pregnancy: a guide

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Genetic counselling is the communication process that helps patients understand and adapt to the implications of health conditions that have a genetic contribution. Genetic counselling should take place, at least to some extent, in the context of all pregnancies. The ideal time is before conception, but when this is not possible, early pregnancy is preferable. Complex cases, such as when there is a known family history of a hereditary disorder, are best referred to a clinical genetics service; however, most pre-pregnancy genetic counselling is performed by general practitioners, midwives and obstetricians. The key components of genetic counselling are summarised in Table 1.

Pre-pregnancy genetic counselling should include open-ended enquiry about any concerns that the prospective parents may have about having a child with a genetic condition or congenital abnormality. In some situations, such concerns may be unfounded and reassurance may be all that is required. Alternatively, further enquiry and investigation may be necessary. It is important to recognise that attitudes of prospective parents towards disability and prenatal testing vary greatly; it is helpful to clarify the views and beliefs of the prospective parents early in the counselling process.

It is also important to recognise that some congenital abnormalities are not genetic in origin. In addition, many disorders that are genetic are not inherited from a parent; but rather occur as the result of new gene mutations arising in the child. This highlights that prior to pregnancy it is not possible to predict, or prevent, all genetic disorders or congenital abnormalities. Table 2 provides a summary of the types of genetic disorders that are potentially identifiable before pregnancy. The availability of pre-implantation genetic diagnosis (PGD) now provides a greater incentive to identify at-risk couples prior to pregnancy.

Despite the increasing profile of new, powerful genetic-testing technologies, the personal and family histories of the prospective parents remain an important screening tool. It is helpful to draw a three-generation family tree, and ask focused questions about any history of genetic disorders, stillbirths, miscarriages, consanguinity and the birth of children with physical or intellectual disability. A history of affected males on the maternal side of the family is a warning sign that an X-linked disorder may be present.

The pedigree may indicate the presence, or raise suspicion, of a genetic disorder in the family, particularly if there is evidence of autosomal dominant (AD) or X-linked transmission. Common examples of AD disorders include neurofibromatosis, Marfan syndrome and familial cancer syndromes (for example, familial breast and ovarian cancer caused by mutations in the genes BRCA1 and BRCA2). Frequently encountered X-linked disorders are Fragile X syndrome, haemophilia A and Duchenne muscular dystrophy. Autosomal recessive disorders, such as cystic fibrosis (CF), are seldom detectable by family history, but a family history of the more common autosomal recessive disorders should prompt consideration of carrier testing in the couple.

Some prospective parents may raise concerns about the risk of a disorder that has complex, or polygenic, inheritance, particularly if one of the parents is affected by the condition. Examples include autoimmune disease, diabetes and major psychiatric disorders. Genetic testing is usually not helpful in this setting; however, empiric risk figures are available and can be used in counselling. Even when one parent is affected by the polygenic condition, the risk to their offspring is typically low (less than five per cent).

Frequently, prospective parents are concerned about the effect of their age on the risk of having a child with a genetic disorder. Most women have a good understanding that the risk of having a baby with Down syndrome increases with maternal age, and that there is a parallel increase in the risks of miscarriage and of other chromosome aneuploidies. There is also increasing recognition that the risk of single gene disorders, caused by de novo gene mutations, increases with paternal age. In recent years, it has become evident that a substantial proportion of genetic conditions occur as a result of de novo mutations, examples of which include achondroplasia and craniosynostosis syndromes.

Genetic carrier screening

Autosomal recessive disorders occur when

both parents are carriers of a faulty gene that, when present in a 'double dose' in a child, results in the disorder. There are literally thousands of autosomal recessive conditions and, in the vast majority of cases, an affected child is born into a family with no family history of the condition. It is now recognised that all people are carriers of multiple recessive genes; these cause no harm to the individual, but pose a risk if their partner carries a fault in the same recessive gene. Of course, statistically, this is unlikely to occur in an individual couple, but collectively autosomal recessive disorders result in very significant morbidity and mortality. Therefore, there is considerable interest in whether these disorders can be prevented by identifying at-risk couples prior to pregnancy.

The best-studied example of population-based carrier screening in Australia is CF. CF is the most common severe autosomal recessive disorder in the Australian population and one-in-25 Caucasians are carriers. Importantly, 94 per cent of babies with CF are born into families where there is no known family history and so a screening approach where only those with a family history of CF are offered testing is not sufficient.[1] Population screening (offering carrier testing to prospective parents regardless of family history) for CF is made more straightforward by the fact that a small number of recurrent gene mutations account for most children born with CF, and has been available in Australia for more than ten years, with strong endorsement from the CF support groups and the Human Genetics Society of Australasia.[2] Usually CF population screening is performed as a two-step process: one partner is screened first, and the other partner is tested only if a mutation is detected in the first partner. This process has been shown to be effective in identifying carrier couples, and most carrier couples detected choose either preimplantation genetic diagnosis or prenatal diagnosis by chorionic villus sampling.[3] Similar outcomes have been demonstrated for thalassaemia screening[4] and for Tay-Sachs disease screening in the Ashkenazi Jewish community.[5]

Expanded carrier screening

Although carrier screening for common recessive disorders (such as CF and thalassaemia) is of proven benefit, the overall impact of screening is limited by the fact that these disorders represent only a small fraction of the thousands of rare autosomal recessive disorders. It is now known that we are all carriers of approximately five to ten autosomal recessive disorders; if this carrier status could be detected, then the overall health burden of autosomal recessive disease might be reduced substantially. The advent of next-generation gene sequencing technology offers the potential to extend the CF screening model to include hundreds or even thousands of diseases.[6]

During the last five years, proof-of-principle studies of expanded carrier screening have been published[7,8], and commercial screening tests are currently being offered in Australia. Expanded carrier screening tests typically offer screening for more than 100 different recessive disorders, with selection of disorders for testing based on their prevalence, seriousness and the ability of carrier status to be determined with accuracy.[8,9] Testing for Fragile X syndrome, the most common inherited form of intellectual disability, is added as a separate test because it requires a different testing technology. Although there are clear potential benefits of these tests, their clinical utility is currently limited by lack of knowledge regarding the pathogenicity of many sequence variants. As a result, for some of the conditions tested, the sensitivity of these tests (in other words, the proportion of carrier couples detected by the test) is less than ten per cent. Other criticisms of these tests include the lack of supportive guidelines from professional organisations, marketing of tests directly to patients, the cost of follow-up testing and counselling and the absence of public funding.[9,10] In spite of these valid concerns, as the technology matures over the next decade, it is likely that these tests will become widely used, and may eventually become standard care.

Table 1. Essential components of pre-pregnancy genetic counselling.

Medical history of prospective parents, with particular focus on disorders with possible genetic aetiology
Drawing of three-generation family tree
Discussion of concerns regarding the risk of having a child with a disability or genetic disorder
Basic advice on folate supplementation and the avoidance of potential teratogens
Discussion of the availability of carrier screening for CF and other recessive conditions
Discussion of genetic testing relevant to specific ethnic backgrounds
Provision of information about screening and diagnostic testing options during pregnancy

Table 2. Types of genetic disorder that are potentially identifiable before pregnancy.

Inheritance	Able to detect at-risk couples before pregnancy	Able to detect during pregnancy
Inherited AD	+	+
New AD	-	+
Autosomal recessive	+	+
X-linked	+	+
Chromosomal	+/-	+
Polygenic	-	+/-

Concluding comments

All couples hope for a healthy baby, and the advent of new genetic-testing technologies provides the opportunity to reduce the risk of having a child with a serious disability. The evolution of these new tests means that pre-pregnancy counselling is more important, but also more complex, than ever before.

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Pre-implantation testing: what you need to know

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Pre-implantation genetic testing (PGT) is used within an IVF cycle. There are two types: pre-implantation genetic diagnosis (PGD) and preimplantation genetic screening (PGS). PGD is indicated for the diagnosis of genetic disease in early embryos before implantation. PGS tests embryos for euploidy (the correct number of chromosomes) before implantation. Both types of PGT are embryo-selection tools to enable couples to choose an embryo that is more likely to develop into a healthy child.

PGD was first reported by Alan Handysides in 1990, using polymerase chain reaction in order to sex embryos to prevent sex-linked disease[1], and then in 1992[2] for cystic fibrosis that ultimately resulted in the birth of a healthy girl. Before the development of PGD, couples at risk of having an affected child could have chorionic villus sampling or amniocentesis in order to detect the genetic disease. Both procedures carry risks to the pregnancy and require parents to face the challenging decision whether to continue the pregnancy if the child is affected. PGD offered the option of screening the embryos before implantation, so parents could be confident that their child was free of the genetic illness. PGD also meant that tested embryos could be frozen for future use.

The PGD process

PGD as a process has evolved since it was first described, both in terms of the stage that embryos are biopsied and the technology used to detect the genetic issues. The woman goes through an IVF cycle in which her ovaries are stimulated to produce a number of oocytes. The oocytes are collected using a transvaginal-ultrasound-guided procedure, injected with sperm and the resulting embryos are cultured for several days. The embryos are biopsied and the cells sent to a genetics lab for analysis. Unaffected embryos are then transferred into the woman's uterus.

PGD can be performed on the polar bodies from the oocyte – day-three embryos that typically contain six to ten cells – or from the trophectoderm of a day-five or -six blastocyst containing 50–100 cells. Polar body biopsies can be performed either before or after fertilisation. Polar bodies have no role in embryo development and so their removal is not thought to compromise the embryo's development. Polar body biopsy also means there is a reasonable interval in which to obtain testing results before embryo transfer on day five. The big disadvantages are that it gives no information about the genetics of the sperm or mitotic errors that occur after the embryo starts growing. Today, polar body biopsy is rarely performed.

PGD was initially performed on day-three embryos at around the six to ten cell stage and one or two cells were removed. Since total culture time is five to six days before embryos must be replaced or frozen, biopsying on day three allows two or three days for results to be available before the transfer of the embryo. There were some concerns that day-three biopsies could lower the chances of a pregnancy and, in 2013, a randomised study was published confirming that biopsying embryos on day three reduced their implantation potential from 50 per cent to 30 per cent, whereas

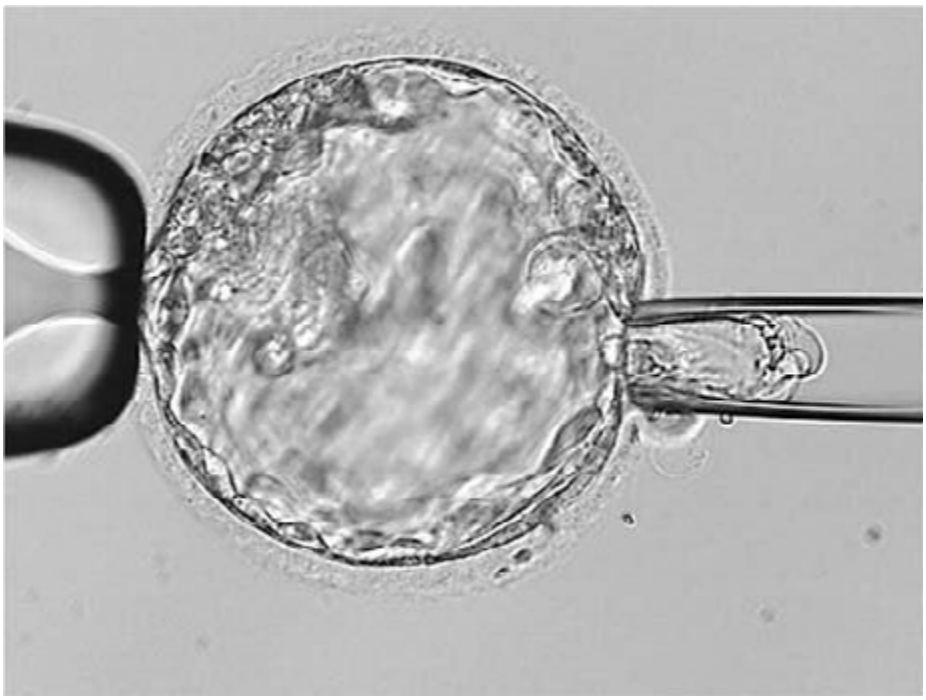

Figure 1. Biopsy of a day-five blastocyst.

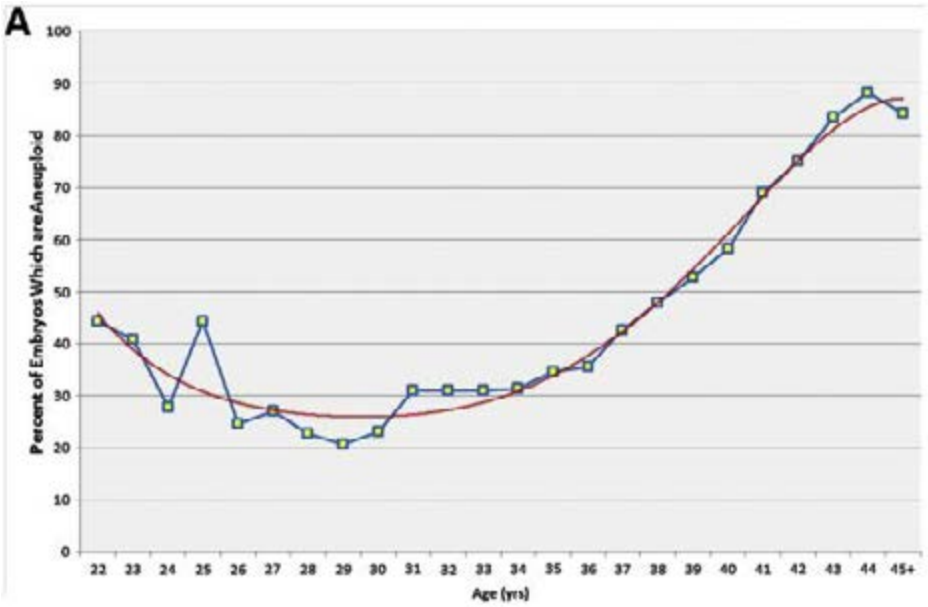

Figure 2. Aneuploidy rate by maternal age.

day-five trophectoderm biopsy had no negative impact.[3]

Lowered implantation potential along with the limited amount of genetic material available for analysis means that PGD is now more commonly performed on day-five or -six embryos at the blastocyst stage. Embryo freezing technology using vitrification enables embryos to be frozen without compromising pregnancy rates while genetic testing is performed.

In order to obtain cells from the trophectoderm, a laser is used on the zona pellucida on day three of development to produce a hole that trophectoderm cells protrude through on day five or six. Once an embryo has reached the expanded blastocyst stage, a laser is used to obtain between five and ten cells. Unlike day-three biopsy, which uses only one or two cells, trophectoderm biopsy provides more cells for testing and thus improves the diagnostic accuracy of the genetic testing. Moscaism can mean that there is a discordance between cells from the inner cell mass and those obtained from the trophectoderm; however, more recent studies suggest that the majority of cells are concordant. This means that the results obtained from the trophectoderm largely reflect the genetic make-up of the inner cell mass.

What are the indications for PGD?

PGD is offered to couples who carry a genetic disease with known mutations, making it suitable for genetic analysis. The most common diseases that PGD is used for include cystic fibrosis, Huntington's disease, spinal muscular atrophy, BRCA genes, haemophilia and parental translocations. PGD may also be used for human leukocyte antigen testing, when a baby could be a stem cell donor for a relative. There are now more than 300 monogenic conditions for which PGD has been used, with the number continuing to expand. There have been thousands of healthy children born following PGD.

Some couples choose carrier testing before starting to conceive a family. Carrier screening often occurs where there is no family or personal history of a genetic illness; however, both partners are screened to determine whether they are carriers of particular genetic diseases. Often these couples will then choose PGD if they are found to be at risk. The most common carrier screening programs test for cystic fibrosis, spinal muscular atrophy and Fragile X syndrome.

PGS

Maternal ageing is associated with increasing oocyte aneuploidy, making embryonic aneuploidy the most common reason IVF fails in older patients. PGS is often offered to older women, couples with recurrent pregnancy loss and those who have experienced failed IVF cycles.

The first PGS cycle was reported in 1993[4] using fluorescent in situ hybridisation (FISH) to screen cells from day-three embryos. The major drawback of this technology was that only a limited number of FISH probes could be used simultaneously; probes were chosen to test for the chromosomes most commonly linked to miscarriage (X, Y, 13, 18, 21), thus missing the impact of aneuploidies from chromosomes not tested. A randomised controlled trial then showed no benefit with day-three PGS and, indeed, fewer babies from an IVF cycle were reported than replacing embryos based on morphological appearances.

Array comparative genomic hybridisation (aCGH) and, more recently, next-generation sequencing (NGS) allows analysis of all of the chromosomes, which, combined with trophectoderm biopsy, has made PGS an effective testing option. In a select population, livebirth rates of around 70 per cent are observed when a single euploid embryo is transferred. Though these results are promising, the challenge is that a couple may not have any embryos to biopsy or any that are euploid.

PGS is a tool for embryo selection and does not increase the number of babies obtained from an IVF cycle. PGS does, however, reduce miscarriage rates, as aneuploid embryos are not replaced, thus reducing the number of thaw cycles a patient must go through to get to a normal embryo. PGS is also being used in the USA to reduce the number of embryos being transferred, with some insurance companies funding PGS so that a single embryo is transferred, hence reducing the costs and risks associated with multiple pregnancies. PGS is also highly informative for the parents and clinician. Where IVF has been previously unsuccessful, information that all embryos turn out to be aneuploid helps some couples consider gamete donation.

Analysis of the biopsied cells

The most common technology used to analyse the cells for PGS is aCGH. The DNA is extracted, then amplified and labelled with a fluorescent green label. The reference DNA is also amplified and labelled with a red label. Both the test and reference DNA are then hybridised on to normal metaphase chromosomes. The green/red ratio is analysed to determine the number of chromosomes. The use of aCGH has some drawbacks, as it does not detect triploidy or balanced translocations.

Single gene disorders are mostly detected using single-cell multiplex PCR. Karyomapping is a new technique being developed where single nucleotide polymorphisms (SNP) can be analysed within the informative loci using aCGH,

without the need for the development of costly probes.

Many embryos tested for a genetic disease and found to be unaffected do not develop into an ongoing pregnancy, likely owing to aneuploidy. It is now possible to combine PGD with PGS, enabling only unaffected euploid embryos to be selected.

Non-invasive embryo selection

Within the IVF lab, there is a huge focus on whether euploid embryos can be selected using non-invasive (not biopsying) techniques. Time-lapse monitoring involves culturing embryos in an undisturbed environment with frequent photography to aid the selection of embryos more likely to result in a healthy live birth.[5] A study by Rubio el al suggested that ongoing pregnancy rates may be increased by nine per cent (51 per cent versus 42 per cent) using time-lapse technology, in part by decreasing miscarriage rates. Other non-invasive technologies, metabolomics and transcriptomics, involve analysis of the fluid surrounding the embryo or the cells nurturing the oocytes in order to gain more information about the embryo.

What does the future look like?

PGD and PGS are selection tools to enable the IVF clinician to choose the best embryo. There are many PGD and PGS cycles where there are no normal embryos available. There are also parents who do not wish to discard their affected embryos. The future may be about fixing the embryo. In a recent report from China, gene-editing techniques were attempted on human embryos with thalassaemia.[6] We are still a long way from knowing whether this is possible, safe or useful.

In conclusion, PGD and PGS have become robust technologies that continue to improve. Nonetheless, most couples using assisted reproduction do not require or benefit from either PGD or PGS.

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Genetic testing in early pregnancy

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Genetic testing identifies changes in genes or chromosomes. These changes may result in structural abnormalities or have functional implications for the developing fetus or the child after birth. An abnormal result may allow parents to prepare for a child who may require additional medical or social needs and to allow maternity caregivers to optimise the delivery. Prenatal diagnosis may be regarded as an important bridge between obstetrics and paediatrics. In other situations, an abnormal result may allow parents the opportunity to make reproductive choices, increasing their autonomy, and consider termination of pregnancy. Over the past two decades, genetic testing has rapidly shifted from the second to the first trimester, owing to the advancement of high-resolution ultrasound and genetic analysis technology.

Screening versus diagnostic testing

Offering a genetic test should be nondirective and it should be clear that testing is voluntary. Informed consent via pre-test counselling should be obtained to enable parents to balance the risk, limitations and benefits of a test. It is important to clarify to parents the differences between the screening and diagnostic nature of the tests offered. The purpose of a screening test is to detect potential indicators of a condition and a positive result indicates a suspicion of a condition. A prenatal screening test is non-invasive and, therefore, procedure-related complications can be avoided whereas a diagnostic test, which involves an invasive procedure, is to establish the presence or absence of the condition.

In accordance with the RANZCOG statement, it is recommended that all pregnant women should be provided with the opportunity to discuss[1]:

- the range of chromosomal abnormalities together with the characteristics of the available prenatal screening and diagnostic tests; and
- prenatal diagnostic testing for other genetic conditions.

Screening tests

The following screening test options are available and meet the performance standard for the common chromosomal abnormality (trisomies 13, 18, 21 and sex chromosomes).[2-5]

Combined first trimester test (11+0 to 13+6 weeks)

A combination of maternal age, sonographic measurement of nuchal translucency (NT) and biochemical markers – pregnancy-associated plasma protein-A (PAPP-A) and chorionic gonadotropin (hCG) – is used to calculate a risk (low or increased). Most developed countries have adopted the combined first trimester test as the standard of care.

Advantages include the following:

- early detection;
- high sensitivity of around 85 per cent, given a false positive rate of five per cent for trisomy 21;
- at time of early NT scan, the incorporation of extra sonographic markers (such as the assessment of nasal bone, ductus venosus waveform and tricuspid regurgitation) allows better detection to 96 per cent and a reduction of false positive rate to 2.5 per cent for trisomy 21; and
- abnormal maternal serum markers, for example, a decreased level of PAPP-A (<0.4 MoM) and hCG (<0.5 MoM) may be associated with increased frequency of adverse obstetric outcome. The optimal method to manage them is unclear; however, educating women about the signs and symptoms of certain complications such as preterm labour, vaginal bleeding, pre-eclampsia and decreased fetal movements may be of value.[6]

Disadvantages include the following:

- NT – variability in quality of measurement; and
- barriers to access – cost and availability.

Second trimester quadruple test (15–20 weeks)

A combination of maternal age and the measurement of maternal serum alpha-fetoprotein (AFP), unconjugated oestriol, hCG and inhibin-A level.

Advantages include the following:

- available for women who present late in the second trimester; and
- an unexplained elevation of maternal serum AFP (>2.5 MoM), hCG (>4 MoM), inhibin-A (>2 MoM) or decreased level of unconjugated oestriol (<0.5 MoM) may be

associated with an increased frequency of adverse obstetric outcomes. Again, no clear management strategy is available, with some authors suggesting closer fetal/maternal surveillance, and its benefit is still subject to debate.[6]

Disadvantages include the following:
- later detection; and
- lower detection rate of 80–83 per cent, given a false positive rate of five per cent for trisomy 21.

Fully integrated test
This is an integration of first and second trimester maternal serum screen, NT and maternal age: a single result is provided after all tests are completed. The fully integrated test offers the benefit of a higher detection rate of 96 per cent, given a false positive rate of five per cent for trisomy 21, but has the disadvantage of later detection.

Step-wise sequential test
This is a modified version of the fully integrated test. The result is available after the first combined screening test, so an early diagnostic test can be offered for those with a high-risk result (for example, a less than one-in-50 risk). Those who are not at the highest risk go on to the second trimester testing to complete the integration. The advantage of this is that women at the highest risk benefit from early detection while women at a lower risk benefit from the high detection rate.

Integrated and sequential screening strategies are not funded and not routinely used in Australia and New Zealand.

Cell-free fetal DNA testing
In cell free fetal DNA (cffDNA) testing, fragments of cffDNA released from the placenta into the maternal blood are used for analysis. Quantitative difference in the DNA sequences that map to individual chromosomes can be used to distinguish fetuses with trisomy and those without. It can be used as either a primary screening test or as a secondary test after a high-risk first trimester screening result.

Advantages include the following:
- early detection – from ten weeks onward; and
- a superior detection rate, with a sensitivity of 99.5 per cent and specificity of 99.8 per cent for trisomy 21. Numbers of procedure-related miscarriage can thus be reduced.

Disadvantages include the following:
- up to five per cent inconclusive result, largely owing to inadequate fetal fractions in maternal blood sample (for example, sample taken at early gestation or maternal obesity);
- cost for patients;
- an NT scan is not included in the test, so there is a lack of information on anatomical abnormality in fetus; and
- a false positive/negative result could be owing to confined placental mosaicism.

According to the RANZCOG statement, first trimester combined screening remains the recommended modality for twin pregnancy screening. Its sensitivity range is 72–80 per cent for trisomy 21 (without incorporation of nasal bone assessment). Second trimester maternal serum screening can also be offered, if women have missed the opportunity in the first trimester. Owing to the smaller number included in the studies, cffDNA testing in twin pregnancies has not been evaluated as extensively. Sensitivity for trisomy 21 is only 90 per cent and the failure rate can be more than five per cent. Therefore, it is important women take these limitations into account when choosing a screening test. For higher order multiple pregnancies, serum markers and cffDNA testing cannot be used, therefore only ultrasound markers in the first trimester can be offered.

Diagnostic tests
Moving on to diagnostic testing, some women who are at a higher risk for aneuploidy or other genetic conditions may choose to go straight to this option for a definitive result. Diagnostic testing can also be offered after a high-risk screening result or in a fetus with major structural anomalies.

Chorionic villi sampling (11–14 weeks)
Ultrasound-guided placental biopsy via a needle aspiration for chromosome or DNA analysis, chorionic villi sampling (CVS) is usually performed via a transabdominal approach, although some practitioners may still use a transcervical approach.

Advantages include the following:
- early diagnosis; and
- an abnormal result allows women the options of surgical or medical termination of pregnancy.

Disadvantages include the following:
- the weighted polled procedure-related rate of miscarriage is 0.22 per cent (much lower than previously quoted)[7];
- there is a one to two per cent risk of confined placental mosaicism, which requires further testing, such as amniocentesis, to exclude fetal mosaicism; and
- a posterior placenta may not be accessible, making CVS not possible.

Amniocentesis (15 weeks gestation onward)
Amniotic fluid is withdrawn via needle under ultrasound guidance. Amniotic fluid contains fetal urine, secretions, exfoliated cells and transudate that can be used for chromosome and DNA analysis. Advantages include a lower procedure-related miscarriage rate (0.11 per cent), lower than previously quoted or that of CVS[7], and it is not restricted by the location of placenta. However, there is the disadvantage of later diagnosis.

Laboratory testing techniques[8-9]
Quantitative fluorescent polymerase chain reaction
Quantitative fluorescent polymerase chain reaction (QF-PCR) technique consists of amplifying polymorphic markers located on the chromosomes of interest to determine the number of copies present per cell. Usually only chromosomes 13, 18, 21 and sex chromosomes are tested. Only a small amount of DNA is required for analysis and the result is available within 24 hours. Other merits include its ability to detect maternal cell contamination, triploidy and mosaicism. (This test is not available in our unit.)

Fluorescence in situ hybridisation
This is another technique for rapid aneuploidy detection (chromosome 13, 18, 21 and sex chromosomes); the result can be available within 48 hours. Fluorescence in situ hybridisation (FISH) uses fluorescently labelled probes targeted to a unique sequence of DNA on the chromosomes of interest. These probes selectively bind and the cells are examined under a microscope to determine gain or loss of that specific chromosomal region of interest. Like QF-PCR, it can also detect mosaicism, triploidy and other common chromosomal microdeletions, such as 22q11 deletion. The main drawback, in comparison to QF-PCR, is the cost and it is a more labour-intensive process.

G-band karyotyping
This involves culturing cells in vitro and harvesting chromosomes for analysis. This is a low-resolution, whole genome study that can detect abnormality at a resolution of approximately 10MB, that is major chromosomal aneuploidies. The advantage of this test, compared to the

rapid aneuploidy tests, is that all of the chromosomes are examined so that whole or partial chromosome aneuploidy will be detected. Balanced translocations will also be detected. However, it is limited by its low resolution and is a labour-intensive test. Results are usually available within ten to 14 days.

Chromosomal microarray

Many units nowadays across Australasia have shifted from conventional karyotyping to chromosomal microarray analysis, particularly in the setting of fetal structural anomalies identified on ultrasound. This is a high-resolution whole genome study, with a resolution of approximately 25KB. It can identify aneuploidy as well as the location and type of specific genetic changes that are too small to be detected by conventional karyotype. Therefore, it can yield more genetic information.

The result is usually available within two weeks. Two types of technologies are used: comparative genomic hybridisation (CGH) and single-nucleotide polymorphism (SNP). CGH detects copy number variants, such as trisomy, but it cannot detect triploidy. SNP detects homozygosity or heterozygosity (identical or different segment of DNA) and therefore can detect triploidy and uniparental disomy. Unlike conventional karyotyping, it cannot detect balanced translocation. One drawback is that it may detect too much genetic information, including CNV of unknown or uncertain clinical significance, which may lead to substantial patient anxiety. Therefore, pre-test counselling and informed consent is essential before women undergo this test.

Testing for single gene disorders

If there is a high clinical suspicion of a single gene disorder based on ultrasound findings or when a specific gene mutation associated with a single gene disorder has been identified in the family and the fetus is known to be at risk, fetal DNA can be analysed. In the case of a suspected single gene disorder a specific gene or genes of interest are analysed in an attempt to identify a pathogenic mutation. In the case of a known familial mutation, targeted analysis of part of the gene is performed to determine whether the mutation is present or absent. This type of testing is usually done in consultation with the clinical genetic service.

The role of the clinician

Parents can choose to gather more information about their unborn child in early pregnancy by means of genetic testing. All of these tests, either screening or diagnostic, involve limitations, risks and benefits. It is our duty to provide women with accurate information so that they can make informed choices for a test that will best suit their needs.

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The problem with sex selection

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Sex selection is a controversial technology that allows people to select their biological children based on their sex. While NHMRC guidelines recommend against sex selection for non-medical reasons, these guidelines are currently under review, making a debate about this issue even more timely. (Throughout this article I shall use 'sex selection' to refer to sex selection for non-medical reasons.) It has been available for some time now and is possible via three methods:

1. sex determination of the fetus followed by abortion if it is an undesired sex;
2. IVF followed by pre-implantation genetic diagnosis, wherein only embryos of the desired sex are transferred to the uterus of the prospective mother; or
3. sperm sorting/flow cytometry that separates X-bearing sperm from Y-bearing sperm by their slight differences in weight, and uses either the X-enriched or Y-enriched sperm to fertilise the egg.

Many countries in Asia and Eastern Europe have seen a drastic drop in the number of female babies born compared with male babies.[1,2] India has laws that prohibit sex selection, but they are not strictly enforced.[3] Similarly, while sex selection is illegal in China, the practice is widespread. In certain parts of India the sex ratio is 120 males for every 100 females and in parts of China the ratio is more than 130 males for every 100 females.[4] The skewed sex ratios began with the introduction of sex selection technology and have been growing worse in most of these countries ever since.[2]

This trend has had dire consequences. Countries with a gender imbalance are showing an increase in sexual violence, human trafficking, bride kidnapping and crime, with political unrest as a result.[2,5] It is no coincidence that the increase in gang rapes in India coincides with the decrease in the number of females born. Therefore, women in such countries now face multiple pressures to avoid having girls: from their families, with psychological and/or physical abuse if they do not practice sex selection, or their own desire to avoid having a child who will suffer the egregious effects of being born female in a deeply sexist society. (While the sexism in countries such as India is quite extreme, it is worth remembering that no country has yet achieved gender equality.) When stuck between a rock and a hard place, such choices are not truly free. The choice to avoid having a daughter also places the mother in the position of participating in a practice that causes the conditions for women like her to go from bad to worse.

It is easy to see why sex selection in such a context is sexist, but in Western countries there is not a strong son preference. Instead, sex selection in the West is more commonly requested for 'gender balancing' (when all current children are of one sex and the parent wishes to have a child of the 'opposite' sex).[6,7] It is worth remembering in this context that although humans are usually thought of as either male or female, there are a number of babies born intersex, with 'ambiguous' genitalia/sex chromosomes. Thus, the binary model of sex does not reflect reality. The problem with sex selection in the West is thus not as obvious as it is in countries that manifest a son preference. Nevertheless, a number of scholars assert that parents who undergo sex selection for gender balancing do so because they are heavily invested in having a child who will conform to the stereotypical traits, norms and behaviours associated with children of the sex they do not have.[8-10] The evidence bears this out. Studies of parents in the US, the UK and Australia who wish to use sex selection show that those who want a daughter want a child who will conform to the gender roles, norms and stereotypes associated with being female, such as playing with dolls, dressing in pink frilly dresses and going to ballet lessons.[7,11,12] They assume she will be heterosexual, get married and have children, without considering that she may be lesbian, a tomboy or may not want children. Many are mothers who want to enjoy talking, shopping and having a close mother-daughter bond[11,13] – all of which they assume they cannot do with a son. On the other side are parents who wish to have a son in order to enjoy activities such as fishing, sports or to have a child who will pass on the family name[7,11,12] – all of which they assume they cannot have with a daughter.

Yet the evidence available so far does not support these assumptions. Of course, we know that there is nothing essential about being male that causes males to pass on the family name: it is not as though it is inscribed in the Y chromosome. We also know that girls are perfectly capable of playing sport and going fishing, and that boys are capable of talking and playing with dolls. But what is commonly assumed is that girls/women and boys/men are naturally inclined to certain activities and have different aptitudes and abilities.[12,14] It appears, however, that this assumption is unfounded. A/Prof Cordelia Fine (a psychologist) and A/Prof Lise Eliot (a neuroscientist), are among the scholars who have critically examined the studies claiming to show that gender differences in psychological characteristics (for example, behaviours, roles and tendencies) are explained by differences in the brain. Their books detail the flaws – ranging from problems with the reasoning on which the studies are based, to methodological flaws

and very small studies whose findings are not replicated – that lead them to conclude that despite over a hundred years of searching, we do not have evidence to support the claim that psychological gender differences are directly caused by differences in male and female brains.[15,16] This is not to say that it is not possible for evidence of neurological causes of gender differences to emerge in the future. Scientists may yet devise an experiment that could somehow separate the effects of nature from nurture, yet short of raising babies in a vacuum, it would likely be hard to accomplish. This means, according to the evidence we currently have (or lack thereof), the assumptions that underlie parents' reasons for undergoing sex selection are not based on fact.

This is where the problem with sex selection and the struggle for gender equality intersect – both are based on an assumption that boys/men and girls/women are naturally good at, or inclined to, different things. For instance, the assumptions that women should bear the lion's share of housework and caring responsibilities because they are more nurturing by nature and that it makes sense for industries such as finance and engineering to be male dominated because they are more mathematically inclined by nature are both unfounded. Yet they are the sorts of beliefs that underlie sexism. Studies that show how males and females are conditioned to behave in certain ways and view their abilities in certain ways have been replicated time and again in different contexts[17-22], whereas the studies that claim to show that these behaviours and abilities are biologically hard-wired or at least precede socialisation have not been replicated. Yet the latter belief is what keeps men and women 'in their places' and sexism entrenched. It is clear, then, that if we should not hold such beliefs about adults, we should not hold such beliefs about children either. Since the drive for sex selection is premised on sexist beliefs regardless of the selection made, it undermines our fight for gender equality.

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Who's your daddy? Paternity testing in the DNA era

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In their book, Sex at Dawn, the authors Ryan and Jethá examine a number of assumptions about parenting and paternity.[1] They write that a cornerstone of the standard narrative of human reproductive behaviour is that it can be viewed from a perspective of economics and game theory. By maximising the number of your offspring who survive and themselves reproduce, your chances of a genetic legacy are greatest, says the theory. This is no guarantee of happiness though, as Robert Wright points out in his book, The Moral Animal. He writes: 'We are built to be effective animals, not happy ones… the frequent absence of happiness is what keeps us pursuing it, and thus makes us productive.'[2]

Taking an evolutionary approach to reproductive behaviour leads to important considerations of paternity and concerns about it, at least for men. The issue of paternity is at the core of much of men's behaviour – and for good evolutionary reasons. 'In our…past, men who invested in children which were not their own would, on average, have left fewer descendants than those who reared only their own genetic offspring. As a consequence men were, and continue to be, preoccupied with paternity.'[3]

Prof Robert Brooks, an evolutionary biologist at the University of New South Wales, has this to say:

> Questions of paternity are built over the deepest well of human insecurity, for children searching to know who they are, for fathers wanting to know whose kids they are raising and for mothers uncertain about the strength of the bonds holding their families together. […] If you have read, heard or watched anything on this question, you will have encountered many estimates, from 9% to more than 30%. The idea that almost one in three people might be the result of what we biologists rather matter-of-factly call 'extra-pair copulations' titillates and horrifies in equal measure.[4]

Sexual economics

A figure of 30 per cent of offspring resulting from 'extra-pair copulation' seems extraordinary. Ryan and Jethá point out that, from a man's perspective, the genetic worst-case scenario is to spend time and resources raising another man's child (or children) and thus propel somebody else's genes into the future at the expense of one's own.[1] However, they question the accepted assumption that male sexual jealousy has the evolutionary underpinning it is assumed to have – paternity certainty. For example, if the fundamental basis of this behaviour (both for men and for women) is a concern that an individual's genes are propagated, then:

> A man should be far less concerned about his wife having sex with his brothers – who share half his genes – than with


The Daily Telegraph

EXCLUSIVE

DNA test reveals Archbishop of Canterbury's astonishing family past

My secret father

Table of Allele Inheritance

Charles Moore, The Daily Telegraph UK, 8 April 2016.

On hearing the results of the DNA test, the Archbishop said: 'My own experience is typical of many people. To find that one's father is other than imagined is not unusual. To be the child of families with great difficulties in relationships, with substance abuse or other matters, is far too normal.' And he said that he found who he was in his religious faith, 'not in genetics'.

> unrelated males. Gentlemen, would you be far less upset to find your wife in bed with your brother than with a total stranger? Ladies, would you prefer your husband have an affair with your sister? Didn't think so.[1]

When the argument is put in these terms, it does indeed seem a little shaky. There is clear evidence though that interest in paternity is strong in the community (at least in the male members of the community) and online paternity testing services, using newly available DNA sequencing technologies, have ramped up this demand.

For most of human history, paternity testing was based on assessment of physical features and eye colour, which was clearly in the eye of the beholder. By the 1920s, advances in the understanding of physiology allowed for more objective tests of paternity using methods such as ABO blood group typing. By the 1930s, pattern analysis of other proteins and enzymes was established and in 1970[2] human leukocyte antigen (HLA) status was used. With the development of Sanger sequencing and now massive parallel sequencing, DNA testing has become the only formal and exact method for paternity testing.[5] Such testing requires an easily obtained cheek-swab saliva sample.

Rapid genetic profiling of the child and putative father can be undertaken with 99.999 per cent probability of a match, making for very accurate results. The most common techniques compare DNA from father and offspring using polymerase chain reaction, short tandem repeat and restriction fragment length polymorphism methodologies. Results for a DNA paternity match can be available within a few days from most paternity testing laboratories. Cheek-swab saliva testing can be done as soon as immediately after birth and can either exclude or confirm the identity of the biological father with great accuracy.

Garbage in, garbage out

So what about the estimate that 30 per cent of paternity assumptions (or claims) are wrong? According to Brooks[4]:

> The problem with most data on paternity is the near impossibility of obtaining an unbiased sample. A paternity clinic, for example, is a bad place from which to estimate the rate of misattributed paternity. Many clients are there because at least one party isn't convinced. Likewise, any study recruiting families – however randomly – might have more success recruiting mothers who harbour no doubts about their children's paternity.

Prof Brooks details how Swinburne University scientist Dr Michael Gilding researched the origins of the popular belief that as many as 30 per cent of paternities are misattributed and was able to trace the source of the high estimate. It appears that British obstetrician Dr Elliot Philipp mentioned this at a meeting in 1972, as an estimate from a small sample of parents. The findings were never formally published and it remains unclear as to how paternity was established and from what population the estimate was made.

Home delivery

Marketing of home DNA paternity tests that play on insecurity has proven to be a business bonanza, but firms draw a very clear distinction between home tests and legally acceptable chain-of-custody paternity tests, where correct identification of the samples is critical. Forensic tests are typically collected by an independent doctor or nurse, who must take legal responsibility for sample handling and can act as a witness to the sample collection. A typical explanation of this critical difference from an Australian website runs like this:

> A legal paternity test is used when court admissible results are required, typically in cases of child custody, child support or paternity disputes. Home tests are informative DNA tests of which results are used for peace of mind or just for reassurance. They have no legal validity.[6]

The advent of cell-free DNA isolation and massive parallel sequencing has opened new business opportunities for paternity entrepreneurs. Here is a typical example of web content from a prenatal paternity testing service that advertises online:

> Prenatal DNA paternity testing can be performed after the 14th week of gestation. Our company will provide you with a special sample collection kit which will contain everything needed to collect the blood sample. We provide within it blood collection tubes for the mother and swabs for the alleged father, the instructions as well as forms to be filled out. Included in the kit is also a prepaid FedEx courier pouch to send your samples back to our laboratory. Also, we will arrange your blood collection at a local laboratory or hospital. Full confidentiality of our testing: the mother can submit different samples from the alleged father(s) without asking him. Our laboratory accepts hair samples, toothbrushes, semen stain and other forensic samples collected from the alleged father.[7]

Law, but not as we know it

As part of a two-year investigation into handling of sensitive genetic information, the Australian Law Reform Commission[8] raised a number of concerns about direct-to-public paternity testing. The report notes that information revealed by paternity testing is particularly sensitive; going beyond 'familial information' into the 'very nature and identity of the family itself'. To make things worse, the context in which the information is revealed is often highly emotionally charged. Where paternity has been misattributed, perhaps for many years, there may be issues of betrayal, revenge and the search for resolution.

Furthermore, the Commission emphasised that DNA paternity testing differs from many other kinds of genetic testing in a fundamental way. Whereas for many medical purposes, useful information can be obtained by testing the genetic material of a single person – who may be shown to have (or not to have) a particular genetic mutation with potential clinical consequences – paternity testing is relationship testing and requires the participation of two or more individuals in order to reveal useful information about their biological relationship.

In most cases, one of the individuals whose genetic sample is required for testing will be a child. In such instances, the Commission pondered who should make the decision on behalf of the child about whether they should submit a genetic sample for testing. The authors noted that the question is particularly difficult when those who have parental responsibility for the child (who in other circumstances would make important decisions affecting the child's welfare) are directly affected by the outcome of the testing procedure. Issues of special concern were identified in the Commission's report:

1. the possibility of error and fraud where the genetic sample is collected without independent supervision;
2. the possibility of sample contamination because the sample may not have been stored correctly or shipped to the laboratory under optimal conditions, or because the chain of custody of the sample cannot be verified; and
3. the possibility that appropriate informed consent may not have been obtained from the person.

How much does all of this actually matter? Prof Brooks says:

> Many men make magnificent fathers to children that do not bear their DNA. But…insecurity over paternity has tectonically shaped much that is least admirable about male behaviour and [has] twisted societies [...] Interweaving strands of evolutionary research suggests that paternity confidence forms part of the glue bonding men to their children and to the women who bore them. Undermine that confidence and men invest less readily in the subsistence and safety of their families, and become more likely to abscond.

So what of that statistic that up to 30 per cent of paternity is incorrectly assigned? The DNA era has allowed us to look at this afresh and, using the very technology that has caused so much of the insecurity, it seems more likely that the true rate is somewhere between one and three per cent across a broad range of societies.[4]

Perhaps this should provide some comfort, but as Prof Brooks reminds us:

> Even a one per cent rate of misattributed paternity still adds up to millions of individual children, worldwide, each part of an interesting, sometimes tenuous and often heart-breaking story.

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Genetics and gynaecological cancer

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In a move that was described as 'courageous and influential', Hollywood actor and UN ambassador Angelina Jolie famously made the decision to have a risk-reducing bilateral salpingo-oophorectomy (RR BSO) at the age of 39. This was two years after a bilateral mastectomy. Her actions ignited worldwide discussion about genetic cancers and prophylactic treatment. Jolie wrote an editorial piece in the *New York Times* about her decision.[1] She carries a mutation in the BRCA 1 gene, giving her a significant risk of developing early ovarian and breast cancer. Her editorial captured the attention of worldwide media and suddenly we were all talking about genetic cancers and risk reduction. According to the Tasmanian Clinical Genetic Service, referrals to genetic counsellors saw a marked increase.

Approximately five per cent of endometrial cancers and ten per cent of ovarian cancers can be attributed to an inherited predisposition.[2] Hereditary breast and ovarian cancer (HBOC) syndrome and hereditary non-polyposis colorectal cancer (HNPCC or Lynch syndrome) account for most inherited gynaecological cancers. Gynaecologists and gynaecological oncologists have a major role to play in not only identifying women at risk of inherited cancer syndromes, but also managing them appropriately.

However, the issue is much more complex than just referring women for genetic testing and offering them prophylactic treatment. The genetics of hereditary gynaecological cancer is continually evolving and our understanding of the molecular basis of inherited susceptibility to gynaecological cancer has improved considerably.[3] Thus, it is the responsibility of the general gynaecologist to keep up to date with advances in this area so as to support patients to make informed decisions.

Basic genetics

Under normal circumstances, the body makes proteins, coded for in the DNA, that control functions within a cell. For example, how quickly it grows, how often it divides or how long it lives. When one or more genes in a cell are mutated, errors in cell regulation can result. If an error causes a defect in the ability of a cell to repair DNA damage, then tumour genesis may occur.[4]

Cancer cells all share several essential properties, including: self-sufficiency in growth signals; insensitivity to antigrowth signals; evasion of apoptosis; limitless replicative potential; sustained angiogenesis; tissue invasion and metastasis; and the development of genomic instability.[5] It is the combination of these properties, which are normally under tight genetic control, that causes the development of a malignancy.

Genetic testing and counselling

Genetic testing is more complex than simply referring a patient for a blood test. In the first instance, there should be a carefully made decision about who is likely to benefit from genetic testing. The approach should be multidisciplinary: involving experts in clinical genetics, oncology, gynaecology and psychology.

Initially, genetic risk assessment should be performed to determine whether a family history is suggestive of an inherited cancer syndrome. Many people who are referred for genetic testing will not qualify for testing once a genetic risk assessment is performed. Genetic counselling is recommended before testing occurs and once results are available. Appropriate counselling includes assessing the patient's understanding of testing for cancer risks and the benefits and limitations of the tests. Patients should be fully aware of possible test outcomes including: the possibility of finding a gene mutation; finding genetic variations that may or may not be cancer-causing; and finding no mutation, which may or may not mean there is a risk of a hereditary cancer syndrome. In the case of the latter, a false negative result could occur because limitations in testing have not detected the genetic mutation or an as-yet-unknown gene is involved. The impact on the patient's ongoing healthcare and the implications for their family will also need to be discussed.

Breast and ovarian cancer

HBOC syndrome is caused by mutations in the BRCA1 or BRCA2 genes. BRCA1 is localised to chromosome 17q, whereas BRCA2 is localised to chromosome 13q. Both are tumour suppressor genes that play a role in DNA repair. The frequency of mutations in the general population is estimated to be one in 300 to one in 800; however, this is higher in some populations (for example, the Ashkenazi Jewish population).[6] Mutations in BRCA1 confer a lifetime risk of epithelial ovarian cancer of 20–60 per cent. The lifetime risk of epithelial ovarian cancer associated with BRCA2 mutations is estimated between 10–20 per cent.[7] BRCA-related ovarian cancers are exclusively epithelial, most are of high-grade serous or undifferentiated histology and tend to be diagnosed younger.

Standard criteria exist for identifying women at risk of a BRCA mutation and include the following:

- early onset breast cancer (less than 45 years old);
- two breast primaries in one individual;

- breast and ovarian cancer in the same individual;
- breast cancer and more than one close blood relative with breast cancer less than age 50 years, or more than one close relative with ovarian cancer at any age, or more than two close relatives with breast cancer at any age;
- breast or ovarian cancer at any age and Ashkenazi Jewish ancestry;
- a family history of male breast cancer;
- ovarian cancer and family history of breast or ovarian cancer; and
- women from families that meet the above criteria.

Maternal and paternal sides of families should be considered independently. A three-generational history is the standard for determining risk. Close attention should be paid to the type of cancer, bilaterality, age of diagnosis and history of chemoprevention or risk-reducing surgeries.

When carrying out genetic testing, individuals with affected family members who have early onset disease, bilateral disease or multiple primaries are preferred as they are more likely to have an identifiable mutation. Negative tests in an unaffected family member are less informative as there is an absence of a known mutation. Thus, women should be fully informed of these limitations before testing.

Once a mutation is known, what should we do about it? Combined oral contraceptive pill (COCP) use by women with BRCA mutation has been shown to be associated with a lower risk of ovarian cancer.[7] An estimated 50 per cent reduction has been observed. There has been no proven association with increased breast cancer in mutation carriers that used oral contraception. Transvaginal ultrasound and serum CA 125 levels as a means of surveillance are associated with high false-positive rates and show no proven benefit. In fact, it is associated with late-stage presentation in affected women. Currently there are no recommendations for its use. RR BSO greatly reduces the risk of ovarian cancer and mortality. Risk reduction is estimated to be 85–95 per cent, especially if performed by the age of 40.[8] In BRCA-positive women who have not previously been diagnosed with breast cancer, hormone replacement therapy can be considered for quality-of-life issues.

Women undergoing RR BSO have an approximately three per cent risk of having a tubo-ovarian malignancy at the time of their surgery.[9] Women need to be counselled about this risk and their options at the time of surgery, in other words to proceed with a full staging operation or to establish the diagnosis and further counsel the patient. Some women want a 'one-stop shop' and request definitive surgery in this situation. Others need time with their families to consider their options. This counselling is best done by a gynaecological oncologist and careful consideration needs to be given to this risk before performing the surgery.

Lynch syndrome

HNPCC or Lynch syndrome is the most common cause of hereditary endometrial cancer, accounting for approximately three per cent of all endometrial cancers. It is the second most common cause of inherited epithelial ovarian cancer.[10] Lynch syndrome is an AD condition that predisposes to colorectal, endometrial and ovarian malignancies, as well as hepatobiliary, urinary, small bowel, brain and sebaceous tumours. It is caused by mutations in multiple genes that produce proteins responsible for mismatch repair, these include: MLH1, MSH2, MSH6 and PMS2. The lifetime risk of endometrial and ovarian cancers in women with Lynch syndrome is 20–60 per cent and approximately ten per cent, respectively. Women typically have an earlier age of onset of disease and are more likely to have multiple primary cancers.

Identifying women for whom genetic risk assessment is recommended is done using the Modified Bethesda Guidelines, as follows:

- Colorectal cancer (CRC) or endometrial cancer diagnosed before age 50;
- CRC or endometrial cancer with a synchronous, metachronous or other Lynch-associated malignancy at any age;
- CRC characterised by tumour-infiltrating lymphocytes, peritumoral lymphocytes, Crohn-like lymphocytic reaction, mucinous/signet-ring differentiation, or medullary growth pattern diagnosed before age 50 years;
- CRC or endometrial cancer and a first-degree relative diagnosed with CRC, endometrial or a Lynch-associated malignancy diagnosed before age 50 years; or
- CRC or endometrial cancer diagnosed at any age with two or more first or second-degree relatives with Lynch-associated malignancies diagnosed at any age.

Prophylactic hysterectomy and RR BSO should be discussed with women with Lynch syndrome who have completed child bearing. However, women should be made aware that surgery will significantly reduce, but not eliminate, their risk. Low dose oestrogen-only hormone therapy can be used to control menopausal symptoms. For younger women, COCP, systemic progesterone and progesterone intrauterine devices have been shown to reduce the risk of developing endometrial cancer in the general population.[11] COCP use is also associated with a reduced risk of ovarian epithelial cancers in low-risk populations. The American College of Obstetricians and Gynecologists recommends endometrial biopsy starting at age 30–35 years and repeating every one to two years. Women should also keep a menstrual diary and promptly report any abnormal uterine bleeding. Neither ultrasound nor CA 125 levels are recommended for screening for ovarian cancer for reasons discussed above.

Peutz-Jeghers syndrome

Peutz-Jeghers syndrome (PJS) is rare, with an estimated prevalence of one in 8 000 to one in 200 000. Males and females are equally affected. It is an AD disorder that is most often owing to germline mutations in the STK11 (LKB1) gene mapped to chromosome 19p13.3. PJS is characterised by pigmented mucocutaneous macules (melanin spots), multiple gastrointestinal polyps and an increased risk of gastrointestinal and non-gastrointestinal cancer. Melanin spots are present in more than 95 per cent of patients and gastrointestinal hamartomatous polyps are present in most patients. The most common sites of gastrointestinal tract malignancy are the colon and pancreas. The most common site of extraintestinal tract cancer is the breast.

Women have an increased lifetime risk of gynaecological cancers. Specifically, women with PJS have a 21 per cent lifetime risk of ovarian cancer and a ten per cent lifetime risk of cervical cancer.[12] Sex cord tumours with annular tubules (SCTAT tumours) occur commonly and are often associated with signs of hyperoestrogenism such as sexual precocity. Cervical tumours include cervical adenoma malignum, a highly differentiated mucinous adenocarcinoma.

Cowden syndrome

Cowden syndrome was first reported in 1963. It is also a rare AD inherited disease, with an estimated prevalence of one in 200 000 to 250 000. Cowden syndrome appears in patients with germline mutations in the PTEN gene (a tumour suppressor gene) located on chromosome

10q23. Clinical manifestations include hamartomatous tumours in multiple organ systems, both mucocutaneous and extracutaneous, and an increased risk for malignancy. Nearly every internal malignancy has been reported in the setting of Cowden syndrome, the most common is breast cancer. Skin and oral findings are distinctive, common and are often the initial finding that leads to the diagnosis.

The lifetime risk of endometrial cancer is reported to be 13–28 per cent for women with Cowden syndrome. There are rare reports of endometrial cancer occurring in adolescents as young as 14. The National Comprehensive Cancer Network recommendations are to educate women with Cowden syndrome regarding endometrial cancer and prompt response to symptoms, to consider annual random endometrial biopsies and transvaginal ultrasound at age 30–35 years and to discuss options of risk-reducing hysterectomy.

Li-Fraumeni syndrome

Li-Fraumeni syndrome is an AD disorder that is manifested by a wide range of malignancies that appear at an unusually early age. It is also known as the sarcoma, breast, leukaemia and adrenal gland (SBLA) cancer syndrome. It results from germline mutations in the tumour protein p53 gene located on chromosome 17p13.1. A wide variety of malignancies have been reported and patients who develop cancer are at markedly increased risk of developing a second malignancy.

For women with Li-Fraumeni syndrome, the lifetime risk of cancer approaches 100 per cent, and has been estimated to be about 90 per cent by 60 years of age. Specifically, women are at markedly increased risk of premenopausal breast cancer at an early age. Gynaecological malignancies are not common, although the most frequently diagnosed gynaecological cancer associated with Li-Fraumeni syndrome is ovarian adenocarcinoma, with an average age of onset of 39.5 years.

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Summary

Patients with possible or known hereditary syndromes with increased cancer risk can be difficult to manage. Patients often want clear information about exactly what their risk is and how best to reduce that risk. Often the answers are unknown and the soundest advice is to tailor a management strategy that best suits each individual patient. While there has been considerable advance in our knowledge of inherited cancer syndromes in recent times, there are still no comprehensive guidelines on the most appropriate action of management for these patients.

Realising the promise of precision oncology

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There is now compelling evidence that the molecular heterogeneity of cancer leads to disparate molecular phenotypes with variable disease outcomes and responses to therapy in histologically indistinguishable cancers.[1] Moreover, therapy induces selection pressures in the heterogenenous tumour environment, leading to either the rapid emergence of resistant clones and/or the acquisition of mutations that confer resistance and alter the molecular phenotype of the tumour. Knowledge of the molecular phenotype has the potential to improve therapeutic selection and, hence, the early delivery of the optimal therapeutic regimen, which would improve overall outcomes and minimise treatment-related morbidity and cost by avoiding ineffective therapies. Although these advances are creating substantial opportunities for improved treatment outcomes, we are faced with significant challenges in implementing precision oncology strategies.[2]

We have made some spectacular advances in some cancer types – or, rather, subgroups of some cancer types. Targeted therapeutics include: anti-oestrogen therapy in oestrogen receptor-positive breast cancer; anti-human epidermal growth receptor 2 (HER2) therapy in HER2 amplified breast and gastric cancer; anti-epidermal growth factor receptor (EGFR) therapy in EGFR mutant lung cancer; imatinib therapy that targets BCR-ABL fusion positive CML and KIT mutant gastrointestinal stromal tumours (GIST); BRAF inhibitors in BRAFV600E mutant melanoma; cetuximab for KRAS wild type colon cancer; and crizotinib for EML-ALK fusion-positive lung cancer. However, we are yet to experience significant improvements in outcome for most cancer types.[3] Although clinical trials of targeted therapies in unselected patients have shown positive signals in several cancer types, the improvements have generally been made in small increments, leading to a lack of adoption and relatively few approvals (for example, erlotinib in pancreatic cancer).

Large-scale genomic sequencing efforts, such as the International Cancer Genome Consortium (ICGC)[4] and The Cancer Genome Atlas (TCGA)[5], are unveiling marked heterogeneity and diversity in previously indistinguishable cancers and, as a consequence, it is perhaps not surprising that therapies designed to target specific molecular aberrations have not had broad success. The appreciation of this diversity, with potential gains in smaller and smaller subgroups, is putting enormous pressure on therapeutic development strategies and health systems to modify processes, from individual patient care through to regulatory approval mechanisms.

In order to advance precision oncology strategies, there is now the need to molecularly characterise both the patient and the tumour they develop in a meaningful way and to match the hypothesised right treatment to the right patient.[2] In principle, this molecular marker selection of potential responders based on the known mechanism of a specific drug appears a relatively straightforward approach; however, our ever-increasing appreciation of the complexity and diversity of cancer makes this challenging to implement in drug development, let alone in routine healthcare.

Progress to date has been made where the proportion of a responsive subgroup of patients within a traditional clinical trial has been substantial enough to generate a detectable signal. For example, the proportion of BRAFV600E mutant melanoma approaches 40 per cent. In contrast, studies such as the ICGC and TCGA are showing that most mutated genes occur at a prevalence of less than five per cent. As a consequence, clinical trials would not detect a signal even if all five per cent of these patients had an excellent response to the treatment. In response, the drug development paradigm is shifting towards a need to select patients based on a marker that reflects the drug's mechanism of action.

This presents significant logistical hurdles, particularly in 'finding' patients that satisfy the molecular criteria for a specific study. Take, for example, a molecular subtype that exists at two per cent (an average proportion in many cases), then with attrition owing to assay failure and for clinical reasons (each of 15 per cent) one would need to screen 78 patients to find one eligible for the study. This attracts a significant cost to drug development, but more importantly is a poor experience for an individual patient and their clinician, since 98 per cent of

patients are effectively told that they are not eligible for the study and need to move on. This further delays therapy as there is a need for additional assays in order to satisfy recruitment criteria for other trials.

These challenges have seen the emergence of molecular screening programs and infrastructure in many countries. These include the US National Cancer Institute's NCI-MATCH program and cancer-specific programs such as Pancreatic Cancer Network's Know Your Tumor program, also in the US, and SpectaCOLOR and SpectaLUNG in Europe. These programs screen patients for molecular markers and then allocate them to clinical trials. This emerging model of finding the trial for the patient, rather than the traditional model of finding the patient for the trial is gaining traction with many stakeholders.

A recent success using molecular selection of patients for treatment is evident in ovarian cancer. The standard of care for advanced disease is a platinum-taxane combination. Although the precise mechanism of action of these therapeutics is not known, they are thought to target dividing cells through damaging DNA (platinums) and cell division (taxanes).

Large-scale genomic studies have revealed that a significant proportion of ovarian cancers harbour defects in genes involved in the DNA damage response, supporting the efficacy of platinum agents in this subgroup. Based on these data, a recent study (Ariel2) used rucaparib, an inhibitor of the enzyme poly ADP ribose polymerase (PARP), which is important in the DNA damage response.

This approach exploited a therapeutic strategy called synthetic lethality. Synthetic lethality means that in normal cells the drug has minimal effect, yet in cells that have specific abnormalities, the drug combines with this defect and is effective at killing these cells. Tumours that had defects in DNA damage response (specifically homologous recombination, in most cases caused by either inherited or acquired mutations in BRCA or related genes) made these cells dependent on PARP for repairing their DNA. Inhibiting the enzyme with rucaparib increased DNA damage, killing the cells.

The Ariel2 study used a new strategy to identify women with recurrent ovarian cancer who might benefit from rucaparib treatment. In addition to mutations in BRCA1 and BRCA2, which occur in about 20 per cent of cases of high-grade ovarian cancer, Ariel2 investigated whether a molecular signature of defective DNA damage repair could be used as a predictive biomarker of sensitivity to rucaparib. The signature was based on the ability to use next-generation genomic sequencing to detect scars in tumour DNA caused by imperfect repair. The Ariel2 investigators hypothesised that tumours with large amounts of scarring would respond better than those with low levels of scarring.

More than 200 women were enrolled in the study and the results, presented at the American Society of Clinical Oncology Annual Meeting in 2015, confirmed the hypothesis. Overall, response rates were highest in tumours with mutations in BRCA1 or BRCA2, with over 80 per cent of patients responding to treatment. However, the main focus of the study was on those patients with no mutations in BRCA1 or BRCA2. Response rates were 45 per cent in those with high levels of DNA scarring; double that seen in patients with low levels of scarring (21 per cent). This is the first time that a predictive signature of response to any treatment has been successfully applied in ovarian cancer. Further validation of this DNA scarring signature is still required.

More recently, there have been dramatic successes using immune therapies to inhibit tumour mechanisms that evade immune destruction. These include checkpoint inhibitors that inhibit PD-1 and other mechanisms. Similar to other targeted agents, immune therapies will also require selection markers, such as the mutation load within tumour cells, where high mutation burdens, such as those seen in mismatch repair deficient colon cancer and melanoma, are associated with responsiveness.[6]

Is summary, we have made some significant advances in implementing precision oncology strategies for some cancer types (and some molecular subtypes) yet there are significant challenges ahead if we are to fully realise the promise of personalised medicine.

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The genetic basis to cerebral palsy: seek and ye shall find

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The 'cerebral palsies' are a diverse group of pathologies with heterogeneous causes. The umbrella clinical term cerebral palsy (CP) is given when a child exhibits non-progressive difficulty in movement control, but it does not signify any particular cause and, until recently, most causes and pathways to CP were unknown. Research into causation has been held back by the long-held and non-evidence-based belief that most cases were owing to acute severe hypoxia at birth. The undefined labels 'birth asphyxia' and 'hypoxic-ischaemic encephalopathy' are still sometimes given to infants in poor condition at birth when there is no direct evidence of an intrapartum hypoxic sentinel event, a severe metabolic acidosis at birth or brain ischaemia. The condition of these infants at birth sometimes reflects other pathology extending well before labour and possibly back to conception.

Epidemiological research groups, led by Dr Karin Nelson in the USA and Prof Fiona Stanley in Australia in the 1980s, clearly showed most cases of CP were not associated with any putative intrapartum hypoxic event and many cases were associated with clinical risk factors for CP (see Table 1).[1,2,3]

In later epidemiological studies, our Australian Collaborative Cerebral Palsy Research Group confirmed these risk factors and, in a systematic review of the world literature, showed that neither elective nor emergency caesarean delivery reduced CP risk.[4,5] Indeed, a telling statistic is that, over 40 years, the incidence of CP has remained the same at around two per 1000 births, despite the caesarean delivery rate in Australia rising from five to 34 per cent.[6] A major component of this rise is defensive obstetrics for fear of CP litigation and the high false-positive rate of intrapartum cardiotocography. Electronic fetal heart rate monitoring has not been shown to reduce the incidence of CP.[7]

It became clear in the 1980s that fear of CP litigation, the difficulties in using the fetal heart rate as a very poor and indirect surrogate marker of brain health and the rising caesarean rate together were having an untoward effect on obstetrics.[8] Since then, four international CP consensus conferences have been convened to help define known clinical risk factors for CP and, in particular, criteria to define the few cases that may have had an acute hypoxic intrapartum cause and timing.[9,10,11,12] In many cases, it is easier in retrospect to define that uncommon cause with objective criteria than speculate about the many unknown causes. All obstetricians should gather evidence after a neonate is delivered in potentially poor condition.[13] Arterial cord gases, placental pathology and customised weight for gestation to assess potential growth restriction may suggest long-term fetal compromise rather than recent severe hypoxia.[6]

For decades, we have suspected that some of the cerebral palsies have a genetic basis. In families with an affected singleton child, there is a nine-fold risk of CP in a subsequent sibling and affected parents have a 6.5-fold increased risk of an affected child.[14,15] This reflects inherited risk and different causative genes have been found in families with more than one affected member.[16,17]

Spontaneous de novo mutations causing CP are harder to identify prospectively. Over the last few years, there have been great advances in genetic methodologies that are now uncovering a genomic basis to many neurodevelopmental disorders. CP shares genetic similarities to several neurodevelopmental disorders, including autism, intellectual disability and epilepsy.[18,19,20] These are often co-

Table 1. Epidemiological risk factors for cerebral palsy.

Preterm delivery
Co-existing congenital anomaly (maldevelopment)
Probable genetic causes
Bacterial and viral intrauterine infection
Altered fetal inflammatory or thrombophilic response (perinatal stroke)
Fetal growth restriction
Higher order pregnancy; risk greater with monozygosity and IVF
Tight nuchal umbilical cord
Prolonged shoulder dystocia
Placental pathology, for example, chorioamnionitis, funisitis, villitis
Inborn errors of metabolism
Male to female ratio 1.3:1

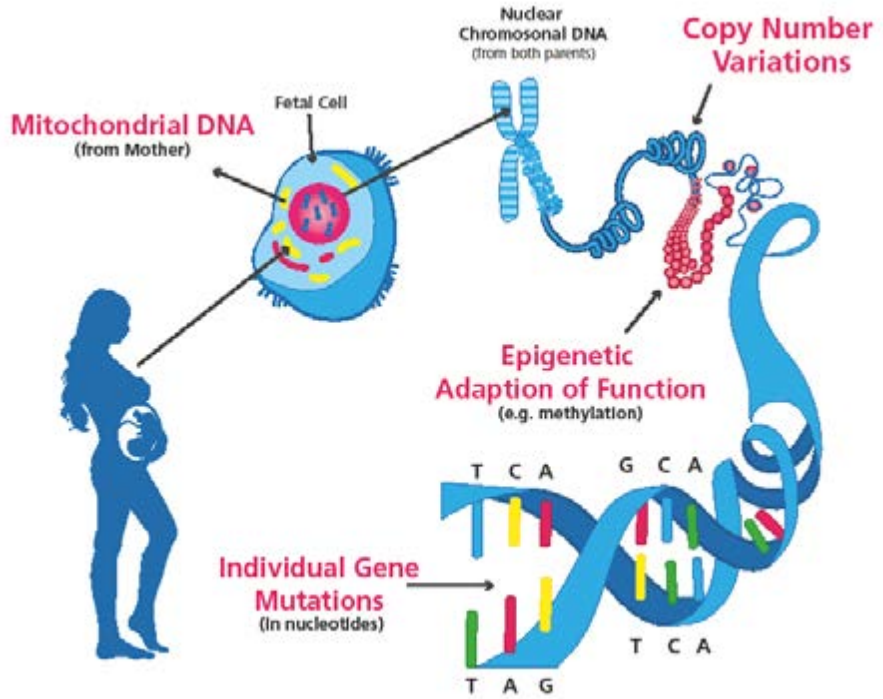

Figure 1. Main sites and types of genetic variation in cerebral palsy.

morbidities with CP. Research on these other neurodevelopmental disorders has progressed more quickly than on CP. The Australian Collaborative Cerebral Palsy Research Group has built up a large biobank of DNA from more than 800 CP patients – including 400 trios of affected child, mother and father – linked to an extensive clinical database.

Initially, the methodology of the era was to conduct candidate or genome-wide association studies that look for genetic markers with higher frequency in CP cohorts than in control groups.[21] This proved a not so suitable methodology, probably because of very high underlying genetic heterogeneity of CP, requiring potentially very large numbers (in 1000s) of CP cases to be investigated. There appears to be a large number of mutations that can disrupt the pathways to brain development or the brain's defence mechanisms; for example, the fetal inflammatory response or the brain's defence against thrombosis and fetal stroke.[22,23,24] There are a great many putative genetic variants that can directly cause or may contribute to CP through susceptibility triggered by epigenetic (environmental triggers changing gene function) factors.[25,26]

Genetic variation can be of different types and can occur in nuclear or in the mitochondrial DNA. Genetic variation affecting larger chunks of DNA is referred to as copy number variation (CNV), while that involving individual nucleotides is referred to as single nucleotide variation (SNV). There is also variation that does not affect the nucleotides, but their modifications; that is, epigenetic variation can affect function (see Figure 1).

Two of the main variants are CNVs and SNVs. CNVs consist of submicroscopic structural variations (duplications, deletions or inversions occurring at meiosis) of at least 1kb in size and usually involve many genes. CNVs usually lead to gene dosage imbalance. Single nucleotide variants (SNVs) are a variation in a single or small number of nucleotides. This variation also involves single nucleotide polymorphisms (SNPs); that is, variation found in the populations with a frequency higher that one per cent. Genome-wide CNVs and SNPs can be detected by array-based comparative genomic hybridisation techniques while SNVs – and, in particular, those that are rare (less than 0.1 per cent) or unique – require sophisticated DNA sequencing techniques such as whole exome (WES) and whole genome (WGS) sequencing. Both types of genetic variation can be either inherited or occur de novo in an individual. Most of the CNV, SNP and many SNV variants are benign, unless they happen to hit a developmentally and functionally important area of the genome, in which case they become pathogenic.

In our first such study using WES, of nearly 200 unselected CP cases at least 14 per cent were found to have SNPs with characteristics suggestive of pathogenicity.[26] We also have found inherited CNVs in 20 per cent of sporadic CP cases.[27] A recent Israeli study has shown that 31 per cent of CP cases had CNVs that were likely to be pathogenic.[28] WGS will allow the study of non-coding regions outside the exome that may control gene expression and protein production. Variants affecting movement control may also reside in mitochondrial DNA, which is passed on to the child only from the mother. All these variants require further study. There are different modes of inheritance, variants may have different penetrance. Some disease-causing variants might be X-chromosome linked, which could partly explain the increased burden of CP in hemizygous males.

Genetic research into other, similar and often co-morbid, neurodevelopmental disorders is more advanced than in CP, but similar rates of potentially causative genetic variants have been found (see Table 2).

Next-generation sequencing techniques (WES and WGS) reveal many genetic variants in each individual; many will have no pathogenic effect and will be irrelevant to CP causation. The challenge is to find the best and most effective strategies to prioritise the potentially relevant variants according to their genetic characteristics.[26,29] The candidate variants found can be classified as likely pathogenic because of their de novo nature, type (stop gain, splice and missense), evolutionary conservation of the mutated residue, known disease association and so forth. These combined characteristics help to prioritise the variants for further investigation.[29] However, it is still necessary to demonstrate

Table 2. Prevalence to date of reported potentially causative genetic variants SNVs and CNVs found through WES and array-comparative genomic hybridisation in neurodevelopmental disorders.

Disorder	CNVs	SNVs	Total variants
Intellectual disability	15–25%	16%	31–41%
Autism spectrum disorder	10–20%	14%	24–34%
Epilepsy	10%	10%	20%
Cerebral palsy	20%	14–33%	34–53%

– using animal, cellular, molecular or in silico models – a functional effect of such a candidate variant in order to resolve its pathogenic (or not) nature. One of the models we are currently exploring is zebrafish, in which we effectively ablate the expression of a specific candidate CP gene and subsequently observe its consequences on fish movement (for example, difficulty swimming away from an adverse stimulus).[30,31] Other function studies can be conducted using manipulation of ex vivo cultured primary neurons, for example, mouse hippocampal neurons. Stem cell models also offer a great potential, whether these are patient-derived induced pluripotent stem cells (iPS) or mouse-derived and genetically engineered mouse stem cells.[32]

Manipulating gene expression can be performed in culture to determine the effect of the predicted causative gene variant on neuron morphology, movement or function. To complement these animal function studies, investigations of global gene expression using RNA sequencing of patient-derived cells allow the study of changes in RNA abundance caused by DNA variants, coding or non-coding. Such transcriptome data (the array of RNA transcripts produced in a particular tissue) also help to identify pathways and networks that might be perturbed[33] and further assist in the prioritisation of variants where the functional impact of the mutation may be unclear.

It is likely that, over the coming years, more and more causative genetic variants involved with CP will be found and validated. There are multiple pathways that develop, control and protect movement control and many places that a spontaneous mutation can occur with varying effects on neural function – a great many variants could be involved. Some variants will have a direct effect on movement control, while others will increase susceptibility to CP that may, for example, require an environmental trigger. It is here that the identification of clinical risk factors in epidemiological studies will help direct epigenetic studies. For example, intrauterine infection may, in genetically susceptible fetuses, affect gene expression that controls the normal fetal inflammatory response, either by allowing neurotropic infections to damage the fetal brain or by resulting in a failure to control an exaggerated cytokine response, which also can be neurotoxic.[34,35,36] It will take the study of large numbers of cases and their families and much research funding to unravel this complicated story. Genetic factors will certainly not be the only cause of this heterogenetic disorder, but there is a growing understanding of the role they may play.

In the long term, we shall be able to identify common susceptibility genes and gene families leading to CP that can be targeted by personalised therapies. Soon it will be possible to screen the embryo, fetus and newborn for known and validated CP genes, but many genes and genetic variants will still be undiscovered. Again, in the long term, when a known pathogenic gene is identified as early as possible, then gene therapy in the form of gene silencing or gene editing could be possible. It's been a long road and there is much farther to travel, but now we can see where we should be going.

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Conflicts of interest
The author reports no conflict of interest.

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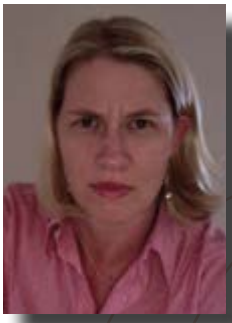
Q&a

For the broader *O&G Magazine* readership, balanced answers to those curly-yet-common questions in obstetrics and gynaecology.

Q *'A 36-year-old woman presents at 28 weeks gestation in her second pregnancy with fatigue and shortness of breath. Her haemoglobin is 95g/L and her ferritin is 5µg/L. She is taking a pregnancy multivitamin. She has an 18-month-old child who was delivered as an emergency caesarean section (CS) for fetal distress at a gestation of 40 weeks and three days. This was complicated by a 1000ml blood loss. Postpartum, she was treated for iron deficiency anaemia with oral iron tablets that caused constipation and exacerbated her haemorrhoids. It is planned that she will have a repeat CS at 39+ weeks. Does she need an iron infusion?'*



Dr Briony Cutts
MD, FRACP, FRCPA

A Iron deficiency anaemia (IDA) affects nearly a quarter of all pregnancies in the Western world, due to the increasing iron demands of each subsequent trimester. By the third trimester, iron demands are three-times greater than in the non-pregnant menstruating woman, owing to increases in red cell mass to enable adequate fetal oxygenation.[1] Iron demands continue to be elevated if women breastfeed. Symptoms of IDA in pregnancy include fatigue, light-headedness, palpitations and shortness of breath. Extreme anaemia can present with chest pain. It is unknown at what haemoglobin level maternal mortality increases; however, maternal morbidity from untreated IDA includes susceptibility to infection and postpartum disturbances to cognition and emotions.[2,3] The recently published National Blood Authority (NBA) 'Patient Blood Management (PBM) Guidelines: Module 5 – Obstetrics and Maternity' provides guidance and recommendations to clinicians for prevention and management of iron deficiency anaemia in pregnancy.

There is no nationally agreed definition for IDA in pregnancy; however, most maternity hospital laboratories have established haemoglobin reference ranges for each trimester. The World Health Organization (WHO) defines anaemia in pregnancy as Hb <110g/L in the first trimester and <105g/L in the second and third trimesters.[4] While haemoglobin is the best measurement of anaemia, inadequate iron stores are confirmed by measuring ferritin, as it is depleted before a fall in haemoglobin is seen. A ferritin level of <15µg/L is diagnostic of iron deficiency while a level of 15–30µg/L is highly suggestive. Women noted to have a ferritin level of 80µg/L or above in the first trimester are likely to have adequate iron stores for their pregnancy.[5]

An investigation for IDA should consist of a full blood examination (FBE) and blood film looking for microcytic hypochromic anaemia at booking. If IDA is suspected, a confirmatory ferritin level should then be performed. Simultaneous testing of an FBE with ferritin should only be performed in women considered at risk of IDA so that early treatment can occur.[6] This group includes indigenous Australian and New Zealand women, teenagers, vegetarians, women with inflammatory bowel disease or previous bariatric/bowel surgery, a past history of postpartum haemorrhage and when there has been less than 12 months duration between pregnancies. Ferritin can be spuriously elevated in infective and inflammatory states. If this is the case, complete iron studies can be done to diagnose iron deficiency. Determining the aetiology of IDA is important. The majority of maternity patients will have IDA owing to inadequate dietary iron intake in the face of the increasing physiological iron demands of pregnancy. However, other aetiology – such as previous heavy menstrual loss or gastrointestinal blood loss – should be considered in women presenting with IDA in early pregnancy or beyond six weeks postpartum.

The majority of pregnant women will have adequate iron stores to deal with the physiological demands of pregnancy and, for this reason, in the PBM Module 5 guidelines, the routine use of iron supplementation in pregnancy is not recommended.[7] In women who require iron supplementation for IDA, first-line treatment requires an oral iron preparation containing at least 100mg of elemental iron.[7] Given iron supplements do not require prescription, clinicians can recommend iron preparations and encourage women to check the iron content of the pregnancy supplement they purchase, as many preparations contain inadequate elemental iron to sufficiently treat IDA. Ideally, preparations will also contain vitamin C or women can be counselled to take a source of vitamin C, such as orange juice, with their iron supplement to aid iron absorption. Oral iron is cheap (one month costs less than $20) and readily available; however, unfortunately, oral iron can exacerbate adverse gastrointestinal effects of pregnancy, especially constipation, haemorrhoids and nausea. Trialling a lower dose of iron, such as 80mg daily, may help reduce these side effects.[7] Relief of symptoms and an increase in haemoglobin by approximately 20g/L should occur after three to four weeks of compliant therapy.[6]

If oral iron has been inadequate, not tolerated or patients are noncompliant, then using intravenous iron may be appropriate. Other indications to use intravenous iron upfront include women diagnosed with IDA in late pregnancy (more than 38 weeks gestation); women with IDA with a high risk of antepartum blood loss, for example, women with placenta percreta; or symptomatic postpartum women that have suffered moderate blood loss (Hb <80g/L).[7] The total iron deficit must be calculated before the administration of intravenous iron. In most cases, 1000mg of iron replacement is adequate. Iron carboxymaltose is now superseding both iron polymaltose and iron sucrose as the preparation of choice for infusion. Mainly because an equivalent dose of 1000mg of iron can be given over 15 minutes, rates of anaphylaxis during infusion are lower and incorporation into red cells with symptomatic relief occurs in a short timeframe (48–72 hours). Iron carboxymaltose has also been

used in randomised controlled trials in pregnant and postpartum women with maternal and fetal comorbidity equivalent to oral iron.[8] The main drawback is cost: upfront iron carboxymaltose is approximately ten-times more expensive than iron polymaltose. However, the cost of the drug is offset by a cheaper inpatient stay, as beds and staffing are required for much shorter periods of time. Iron carboxymaltose is very easy to use to treat IDA intravenously and hospitals need to have robust clinical practice guidelines and auditing tools in place to ensure that its administration is not misused and that quality assurance processes are met.

The patient in this scenario meets the definition of IDA. Her risk factors include less than 12 months between pregnancies and a previous postpartum haemorrhage. She has had gastrointestinal side effects from oral iron in the past, which may make her reluctant to take oral iron again. She requires definitive treatment, given her iron stores will continue to fall as she enters the final trimester of pregnancy. In accordance to the PBM Module 5 guidelines, it is important to treat her anaemia and optimise her red cell mass before her repeat CS to reduce the transfusion risk in the setting of potential blood loss. In this woman an iron infusion is reasonable, but when is administration appropriate? If she is amenable, there is time to trial a reduced dose of oral iron for four weeks with a clinical review and follow-up. However, if she is already suffering from constipation and haemorrhoids she is unlikely to submit to an oral iron trial. In this setting, it is reasonable to offer an iron infusion with iron carboxymaltose upfront.

Iron infusions should be offered to women who have IDA, not iron deficiency alone. While untreated iron deficiency will result in anaemia with time, there is no evidence that treating iron deficiency without the presence of anaemia (in other words, treating a low ferritin level <30μg/L and normal Hb level >110g/L), reduces maternal or fetal morbidity. These patients, who are unlikely to be symptomatic, should be monitored and offered a trial of oral iron.

The PBM Module 5 guidelines indicate that in an obstetric and maternity setting, blood transfusion is usually appropriate if a patient's haemoglobin is less than 70g/L. With intravenous iron so easy to administer, clinical trials are needed in the maternity setting to determine at what haemoglobin level intravenous iron is ineffective and blood transfusion more appropriate.

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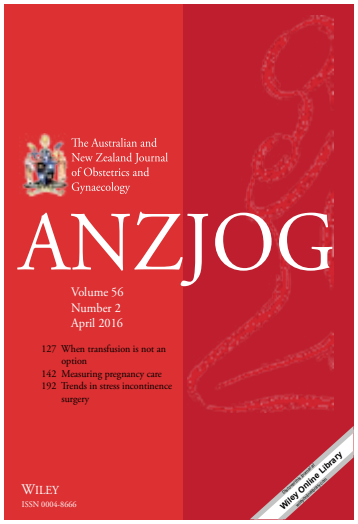
FROM THE EDITOR'S DESK

This is the first of what will be a regular feature in *O&G Magazine*, written with the aim of linking the *Australian and New Zealand Journal of Obstetrics and Gynaecology (ANZJOG)*, the College's academic journal, with *O&G Magazine* which, while it also publishes clinical articles, aims to provide opinion, news and comment as well.



April issue – 56(2)

The April 2016 issue of *ANZJOG* (56:2) kicks off with a review from Kidson-Gerber and five other experts titled, 'Caring for pregnant women for whom transfusion is not an option. A national review to assist in patient care'. Every practising obstetrician will have come across this challenge, which is associated with increased rates of maternal morbidity and mortality. While conceding that high-quality evidence is difficult to find, the authors manage as comprehensive a look as possible at what is available from cohort studies, case series and reports together with basic physiological principles, and have produced a pragmatic approach to the management of the problem. They strongly recommend a multidisciplinary ante- and perinatal plan, including attention to pre-delivery haemoglobin levels and iron stores, minimising blood loss, haemorrhage control and postnatal management. This is important reading for all obstetricians.

A second useful review, by Wong and Merkur, explores the anatomy of the inferior epigastric artery in relation to the surface anatomy, and will be of interest to everyone performing, or learning to perform, abdominal surgery.

New original research

Among the original articles in obstetrics are contributions from two very different places – the first from Monash, the second from Papua-New Guinea – on the assessment of pregnancy care. Sinni et al evaluate a clinical audit tool to measure the quality of antenatal and intrapartum care in their Melbourne hospital; Tanimia et al look at the value of WHO guidelines for 'near-miss' situations in obstetric practice in Port Moresby General Hospital. Dipping in further you will find an article by Kohlhoff et al that highlights the relative lack of screening for postnatal depression among women attending for private obstetric care in Australia; also articles investigating intravenous iron infusions, and the use of vitamin and herbal supplements in pregnancy.

Under original articles in gynaecology is a study by Brown and King of age-stratified changes over 20 years in surgical procedures for urinary stress incontinence. They identify the mid-urethral sling as now being the most commonly performed procedure. Blain et al look at the feasibility of 'see-and-treat' for high-grade Pap smears, and Sozen et al recommend combined adjuvant radiotherapy and chemotherapy in early uterine cancers.

June issue – 56(3)

In the upcoming June issue you will find four articles and an invited editorial dealing with the reproductive health of Aboriginal and Torres Strait Islander women. There are some positive findings among these, but also much evidence to show that we are far from Closing this Gap. There are also, among many other topics, an interesting look at gestational surrogacy in Australia; an article recommending more communication and consistency among those responsible for giving information to the parents of very preterm babies; and a paper detailing the prevalence of thyroid dysfunction among women attending a private obstetric practice.

Expert reviews

Wherever possible, each issue of *ANZJOG* will contain at least one, and sometimes more, expert reviews on topics relevant to current obstetric and gynaecological practice. In general, reviews are by editorial invitation only, but readers are invited to submit suggested topics for editorial consideration.

Opinion pieces

While many opinion pieces appear in *O&G Magazine*, there is also a place in *ANZJOG* for the reasoned debate of clinical controversies in our specialty. Such pieces may include both sides of an argument in the one article or there can be articles from

Prof Caroline de Costa
FRANZCOG
Editor-in-Chief
ANZJOG

two authors, each exposing a viewpoint. Opinion pieces submitted to *ANZJOG* are subject to the same intellectual rigour applied by the peer-review process as original research.

Letters to the Editor
These are always welcome. In general they will be responses to recent articles, and will add something more to the topic. When submitted in a timely manner, it is possible for letters to the editor to appear in the issue subsequent to the one in which the original article appears.

> **“Opinion pieces submitted to *ANZJOG* are subject to the same intellectual rigour applied by the peer-review process as original research.”**

Peer reviewers
We are constantly seeking enthusiastic reviewers for submissions. More than 200 manuscripts are considered for publication annually, which means we need at least 400+ reviews. Almost all practising obstetricians and gynaecologists should be able to review from time to time. Without reviewers, *ANZJOG* would not exist. So if you are interested, contact Sarah Ortenzio via: anzjog@ranzcog.edu.au.

Data repositories
A number of journals now require authors to submit all data sets related to their research for permanent storage and access. Other researchers then have free access in order to verify or attempt to duplicate research findings, and to extend further research based on these findings. Wiley, the publisher of *ANZJOG*, has embarked on a pilot data repository project with other journals, but not at present with *ANZJOG*. One of the concerns is who will pay for these repositories; overseas, applicants for research grants are now starting to require funds from major grant bodies to include a component for continued data storage. However, for the clinicians conducting smaller projects within their own institutions or practices that comprise a reasonable number of *ANZJOG* contributors, the way forward is not yet clear. Further news is awaited!

Drug and alcohol policy in Australia

Dr Alex Wodak AM
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Visiting Fellow
Kirby Institute, UNSW
President
Australian Drug Law Reform Foundation
Director
Australia21

It is hard to find a family in Australia that is not troubled by at least one member with a serious alcohol or drug problem. Yet trying to get expert help for a loved one with a serious substance abuse issue is never easy in Australia. Also, whatever branch of medicine we practise, we still run into many patients struggling with major dependance problems. So, either because of your professional work or because of your family, the reasons why Australia responds to alcohol and drug problems so poorly is relevant to all of us.

No other branch of medicine or health policy is as politicised as the alcohol and drug field. The way our community responds to alcohol is dominated by what is – and what is not – acceptable to the drinks industry. When responding to illicit drugs, poor policy has for decades been great politics. But the fact that the public health David was able to eventually overcome the tobacco industry Goliath suggests our terrible alcohol and illicit drug policies are not immutable.

Australia's drug policy begins

Australia's drug policy resulted from a combination of elements. Our first drug laws specifically only prohibited the smoking of opium. These laws were passed in South Australia, Victoria and New South Wales during the gold rush period. Chinese people working on the goldfields were the only people in Australia at the time to smoke opium. California, USA, and British Colombia, Canada, passed similar laws around the same time and for the same reason: racism. Edible opium was lawful, taxed and regulated in Australia until it was prohibited in 1906, but edible opium was consumed by many mainstream Australians.

The failure of the edible opium ban was quickly recognised. The Commonwealth Comptroller-General of Customs, HNP Wollaston, stated in his report to the Commonwealth Parliament in 1908: 'it is very doubtful if such prohibition has lessened to an extent the amount which is brought in to Australia'. He added: 'owing to total prohibition, the price of opium has risen enormously … the Commonwealth gladly gave up about £60,000 revenue with a view to a suppression of the evil, but the result has not been what has been hoped for. What now appears to be the effect of total prohibition is that, while we have lost the duty, the opium is still imported pretty freely.'[1]

The origins of drug prohibition

Momentum for global drug prohibition started building in the early 20th century. The US convened the International Opium Commission, a meeting of 13 nations in Shanghai, in 1909, to discuss a global ban on opium. For many years, American missionaries in China had been reporting to Washington DC their observation of the immense cruelty of the powerful British, forcing opium on to citizens of much weaker China. Chinese opposition to this policy led to two opium wars in the 19th century against a much more powerful UK. The UK seized Hong Kong for 150 years to punish China for waging war and losing.

The International Opium Commission led to other meetings and agreements, culminating in three United Nations international drug treaties (1961, 1971 and 1988). Virtually all countries have signed and ratified these treaties, committing these countries to pass laws imposing criminal sanctions on persons convicted of selling, buying or possessing prohibited drugs.

The UN has a system for selecting which drugs are to be banned. At present, about 250 drugs are on this list. Following UN pressure, Australia banned the production and importation of heroin in 1953. Australia's problems with heroin began after and not before the Commonwealth government banned heroin. The leaders of the Australian medical profession opposed the decision to ban heroin, including the British Medical Association (this was before the Australian Medical Association had been established).

Scrutiny of our national drug policy

For a number of decades, drug prohibition in Australia, as in other countries, was strongly supported, rarely questioned and politically beyond reproach. Problems started appearing slowly in the 1970s, with a burgeoning heroin problem. An Australian government has initiated a major enquiry or Royal Commission almost every year since. In the US, President Nixon's declaration of a war against drugs in 1971 helped him to win the 1972 presidential elections in a landslide. Nixon won 49 of the 50 states, despite the hugely unpopular Vietnam War. Politicians in other countries took note of this apparent political 'Magic Pudding'.

In the 1980s, cracks in our drug policy started appearing and getting wider. In 1985, the then Prime Minister (Bob Hawke) convened a meeting of all state Premiers and the NT Chief Minister that approved 'harm minimisation' becoming Australia's official national drug policy. HIV spreading among and from people who inject drugs was becoming an increasing concern. Calls for the establishment of needle syringe programs to stem the spread of HIV were rejected. After a pilot project was established in Sydney as an act of civil disobedience, the NSW government decided to allow needle syringe programs. These clearly helped to avert an HIV epidemic, saving thousands of lives and billions of dollars. The effectiveness, safety and cost effectiveness of harm reduction contrasted with the lack of evidence for drug law enforcement and its often-severe collateral damage and high cost.

The winds of change start to blow

In 2011, the Global Commission on Drug Policy was established with about 20 former Presidents, Prime Ministers and other senior leaders from a number of countries. The Commission's questioning of the continuation of global drug prohibition attracted considerable international media. The momentum for change has been gathering pace ever since. In 2012, Australia21, a small independent think tank, issued two reports that also attracted considerable media coverage. In November 2012, majorities of voters in Colorado and Washington in the US created a milestone in global drug policy by supporting ballot initiatives to begin taxing and regulating cannabis. In July 2013, 119 of the 120 members of the New Zealand parliament voted in favour of a bill to regulate certain new psychoactive substances. Although the bill was suspended after ten months, the clear benefits and lack of serious negative consequences were evident.[3]

In recent decades, Australian governments have generously funded drug law enforcement and often used harsh rhetoric to refer to people who use drugs. Health and social interventions have been starved of funds and rarely mentioned by government. Meanwhile the illicit drug market continued to expand with increasing numbers of consumers and a growing array of new drug types.[2] The price of drugs continued to fall and 70–90 per cent of drug users reported that obtaining drugs was 'easy' or 'very easy'. Deaths, disease, crime, corruption and violence increased. The number of heroin overdose deaths increased from six in 1964, to peak at 1116 in 1997, before settling at about 400 a year for the early 21st century and then starting to rise again.

The (then) Prime Minister Tony Abbott admitted in 2014 that the war on drugs was a war that could not be won, but could be lost. Serving and retired police commissioners started to acknowledge that Australia could not arrest or imprison its way out of its drug problems.

> 'The paramount need is to re-define drugs as primarily a health and social issue, rather than a primarily criminal justice issue.'

What is to be done?

The paramount need is to re-define drugs as primarily a health and social issue, rather than a primarily criminal justice issue. Criminal sanctions need to be reduced and, where possible, be eliminated ('decriminalisation'). Governments will also have to try to regulate as much of the drugs market as they can. It will never be possible to regulate all of the market. There will always be some black market. Drug treatment has to be expanded and improved so that it becomes like any other health service.

Australia is not unusual in facing a difficult and complicated drug policy problem. Most countries are in a similar predicament. Global drug prohibition is slowly unravelling. The federal system makes it harder to develop a consistent national approach, but also enables states or territories to undertake experiments.

A number of specific drug policy issues are now being discussed. After several years of debate, Australia is likely to make medicinal cannabis lawful in 2016. In many countries, a few years after medicinal cannabis became lawful, a debate developed regarding the taxing and regulating of recreational cannabis. The deaths of a number of young Australians after taking drugs at youth music events in recent years has provoked a debate about continuing the current policy of saturation policing with sniffer dogs or perhaps evaluating pill testing. This has been available in some European countries for a quarter century and seems to have reduced the number of such deaths. Each state and territory in Australia now has laws enabling random roadside saliva tests of drivers for cannabis, amphetamine and ecstasy. The offence is to have detectable levels in oral fluids. Though introduced as a road-safety measure, the drugs tested for only make a modest contribution to the risk of a road crash death while other drugs, which pose a much greater risk, are not tested for. Our drug-driving laws, based on poor science, are now coming under increasing scrutiny and rightly so.

The establishment of Sydney's Medically Supervised Injecting Centre in Kings Cross in 2001 has clearly saved lives and dollars, referred many clients to health and social services and improved the amenity of the neighbourhood; however, it is still the only such centre in the country. The former Victorian Premier and others have been advocating for such a centre to be established in Melbourne. Meanwhile, the increasing number of drug overdose deaths continues to be ignored. Although alcohol is responsible far greater health, social and economic costs than illicit drugs, and the means of reducing these harms are well known (increasing price, reducing availability, regulating advertising), there is little change in policy.

Since becoming Prime Minister, in 2015, Malcolm Turnbull has often emphasised the need for Australia to become more nimble, agile and innovative. There are few areas where Australia needs these changes more than in the alcohol and drug field.

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Maternity care for Syrian refugees in Lebanon

Dr Kate Tyson
MBBS, FRANZCOG Trainee

Conditions for Syrian refugees in Lebanon are dire and their needs vast. More than a million Syrian refugees have sought refuge here seeking shelter where they can. If it's winter, they must contend with frigid nights and heavy snowfalls that often collapse their flimsy tents. In summer, they're exposed to extreme, arid heat. Rains at any time bring floods and mud as well. And, regardless of the month, they have little access to the sort of healthcare so many of them urgently need.

Priced out of care

Of particular concern is the lack of access to free, high-quality healthcare for the tens of thousands of Syrian women giving birth in Lebanon. The Lebanese health system is entirely privatised and highly interventionist. Although the refugees, most of whom live in extreme poverty on less than AU$5 a day, receive a 75 per cent subsidy from UNHCR, the $65 fee they are left with for a straightforward delivery is devastating.

And 'normal' deliveries are by no means the norm. More than half of deliveries in most Lebanese clinics are caesarean sections, even if the mother has had previous vaginal deliveries. At a subsidised cost of around $200 or more, the tendency towards caesareans, not uncommon worldwide, results in significant burden for the patient. If she cannot pay, she might be refused access to the hospital or have her refugee card confiscated, which often means no access to food vouchers until the hospital bill is cleared.

The result is that families are faced with a debt they can ill afford, leaving them to borrow from friends and family, send their children out to work instead of school or opt to avoid the clinics altogether and risk unattended homebirths or private midwives with no training. Such stories are very upsetting to me, because I know how dangerous it is and how awful it must be for a mother to give birth scared and alone.

Opening a basic maternity clinic

To respond to this need, Médecins Sans Frontières (MSF) decided to open a second basic emergency obstetric and newborn care (BEmONC) clinic in the Bekaa Valley, close to the single border crossing that allows Syrians to pass into Lebanon, and in an area home to up to 90 000 refugees. I was responsible for all aspects of setting up the clinic: recruiting the staff, training the team and implementing and adjusting the protocols in coordination with the medical advisors at headquarter level, so that the clinic would be ready to receive patients. All contingencies had to be planned for, so I also would wake up at 4am thinking: 'I need to talk to logistics and pharmacy about the back-up plan for the cold chain' or 'what are the pros and cons of keeping a single unit of O-negative blood on site?'

My logistics colleagues renovated an existing primary health centre on the outskirts of the town, making the downstairs area suitable for births. As a BEmONC facility we would be offering basic obstetric and neonatal services only, meaning we would not have an operating theatre and would need to transfer complicated cases. Babies born in our unit requiring simple interventions (for example,

A pregnant woman peers out from her tent home as freezing fog surrounds the Khoder Hawash makeshift settlement for refugees on the outskirts of Baalbek in Lebanon's Bekaa valley. ©Ghazal Soutoudeh.

Setting up the clinic involved training local staff and establishing appropriate evidence-based protocols. ©Jinane Saad/MSF.

intravenous antibiotics or management of hypoglycaemia) could also be managed, but more complicated cases would still have to be referred. However, the success of a project like this depends on building a good referral system between lower and higher levels of care. To achieve this, we not only coordinated free transport with the Lebanese Red Cross, but we also provided vouchers for the patients needing transfers, to reduce the barrier of cost for their advanced care.

As it was winter, with snow all around, and the Lebanese electricity supply is highly unpredictable, a large generator and oil burners were essential. Looking out the clinic window at the snowy peaks, I often wondered how our patients survived their pregnancies with nothing but a bit of plastic sheeting to protect them from the cold.

Training and protocols

Preparing the team and cementing protocols for the clinic involved revisiting my training and what was considered standard practice back home. In Australia we work very hard, with big financial investments, to make small improvements. This is because the standard of care is already so good, patient expectations are high and neonatal mortality is so low. In Lebanon the challenges were different. The national staff midwives are all used to working in a hospital system where only doctors do deliveries. Caesarean sections are common and there is a lot of monitoring and ultrasound. I would consider many of these practices to be outside of evidence-based medicine.

The MSF approach is somewhere in between. MSF midwives are trained to perform the whole delivery, including perineal repair, postnatal care and discharge of the mother and baby once safety criteria are met. They are expected to be able to detect risk factors and call the doctor when complications occur. Overall, there is a strong push towards achieving safe, normal delivery with the introduction of simple and effective interventions, resulting in significant reductions in maternal and neonatal mortality.

Being able to call on my training to implement measures with a high impact was incredibly rewarding. In Australia, routine management of the third stage of labour requires an injection of oxytocin and controlled cord traction. This simple intervention halves the chances of having a postpartum haemorrhage. It is standard, evidence-based practice in Australia and also for MSF, but it wasn't part of hospital policy in Lebanon. It was both a pleasure and a challenge for me to guide the introduction of this to our clinic and demonstrate to our team the impact it had.

I decided to become an obstetrician because to me it was the perfect way to combine medical skills with humanity and kindness. Aid work presented an opportunity to apply these skills to help people when they are most vulnerable. My first patient in the newly opened maternity clinic was a

Hasnaa, who is 16 years old, and her newborn resting at home after delivery at MSF's new maternal and child centre in Majdal Anjar, Lebanon. ©MSF.

16-year-old Syrian refugee. I know it sounds clichéd, but seeing that baby being born on our first Saturday morning meant everything to me. It wasn't just the success of the birth – a vaginal delivery resulting in a healthy baby girl – it was seeing our team of national staff in action. They were incredible. Clearly all those weeks of training and practice paid off.

In Lebanon, we had two months to open a clinic and get the team working to MSF standards of care. This required them adapting to new ways of working and letting go some of the old. However, when I walked in to the clinic that first Saturday morning to find everything set up perfectly from the neonatal resuscitation unit and the notes on the whiteboard, I could see how far we had come. Not only did I feel privileged to contribute to offering health and dignity to Syrian refugees, I also felt confident that this dedicated team would give women a quality of intrapartum care I would be proud of here in Australia.

About MSF in Lebanon

MSF first started working in Lebanon in 1976, providing medical assistance in response to the outbreak of civil war. It currently provides primary healthcare – including treatment for acute and chronic diseases – in Tripoli, the Bekaa Valley, Beirut and Sidon. MSF also operates mother-and-child services in the Bekaa Valley and the Shatila Palestinian refugee camp. MSF's services are open to Syrian refugees, vulnerable Lebanese, Lebanese returnees from Syria, and Palestinian refugees from Syria. MSF treats all Syrian refugees irrespective of their registration status with UNHCR.

Working for MSF

MSF Australia is currently looking for qualified gynaecologists and obstetricians who are able to commit to a minimum of six weeks to work in the field. For more information, please visit: www.msf.org.au/recruitment.

Trainees (PGY3+) are eligible for general medical doctor roles. These roles require a minimum availability of nine months. Completion of specialty training is not mandatory before applying and there is potential for some placements to be accredited towards training (fully or partially) with reference to the FRANZCOG Training in Resource Limited Settings Guidelines: http://www.ranzcog.edu.au/additional-information-on-training-a-training-posts/franzcog-training-in-resource-limited-settings-overseas-guidelines.html. Applications for MSF field placements to be accredited towards training are assessed on a case-by-case basis by Chair of the relevant Regional TAC and the Chair of the RANZCOG TAC.

Management of hypothyroidism in pregnancy

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Metabolic requirements dramatically increase in pregnancy and thyroid hormone dosing needs to increase proportionately. Insufficient thyroxine replacement can have serious affects on the growing fetus, including increased risk of premature birth, low birthweight and miscarriage.[1] These affects are seen especially in the first trimester, as the fetus is relying solely on transplacental free T3 and T4 until it develops its own thyroid gland at 11–12 weeks gestation. Furthermore, the fetal thyroid does not fully mature and secrete adequate thyroid hormone until 16 weeks.[2] The first trimester of pregnancy tends to be managed entirely by general practitioners, necessitating clear guidelines for investigation, treatment and monitoring of hypothyroidism in such a critical trimester.

Thyroid physiology undergoes many changes during pregnancy. In the first trimester, beta HCG rises. TSH and HCG share similar beta subunits, allowing the HCG to directly stimulate thyroid tissue, this leads to increased fT3 and fT4 and, therefore, TSH suppression may occur in the first trimester. As beta HCG falls in the second and third trimester, TSH normalises. Furthermore, there is:

- increased iodine clearance through increased glomerular filtration and decreased tubular reabsorption;
- increased uptake of T3 and T4;
- increased oestrogen, causing increased thyroxine-binding globulin (TBG) production in the liver, reducing biologically active fT4;
- T4 undergoes transplacental passage to supply the fetus;
- increased renal excretion of T4; and
- breakdown of T4 by placental deiodinases.

In view of these physiological changes, gestation-specific thyroid stimulating hormone (TSH) level standards need to be consulted when interpreting thyroid function tests, as they are different to non-pregnant interval ranges.[14]

Universal screening for thyroid dysfunction in pregnancy is not recommended[4]; however, thyroid function testing is recommended by the American Thyroid Association in the woman[5]:

- from an area with moderate to severe iodine insufficiency;
- with symptoms of hypothyroidism;
- with a family or personal history of thyroid disease;
- with a personal history of thyroid peroxidase antibodies;
- who has type 1 diabetes;
- who has had head and neck radiation;
- who has experienced recurrent miscarriage/reduced fertility; or
- with morbid obesity.

Hypothyroidism may be pre-existing (Hashimoto's thyroiditis, iodine deficiency, previous radiation exposure to the thyroid, previous thyroidectomy or thyroid nodule excision) or newly diagnosed in pregnancy. It can be divided into either:

1. overt hypothyroidism (OH) (affecting 0.5 to one per cent of all pregnancies); or
2. subclinical hypothyroidism (SCH) (two to 15 per cent of all pregnancies). The incidence varies widely across studies depending on which value of TSH is used as the upper limit of normal.[6,7]

Treatment and surveillance

For the treatment of OH or SCH in pregnancy, the optimal agent is oral T4. The goal is to normalise the maternal serum TSH to within the normal trimester-specific pregnancy reference ranges. Thyroxine

Implications for practice

- Pregnancy causes changes in thyroid hormone metabolism.
- Thyroid function screening is suggested in at-risk women, to avoid the detrimental affect on both maternal and fetal outcomes. Studies are ongoing to determine if universal screening should be recommended.
- SCH and OH in pregnancy require monitoring.
- Dietary iodine supplementation should begin at diagnosis of pregnancy.
- A prompt increase in thyroxine dose in OH will decrease adverse events to both mother and fetus.
- Treatment of SCH in pregnancy is controversial; however, the European Thyroid Association recommends treatment. Studies are ongoing and will soon provide definitive evidence.

Table 1. Suggested target TSH levels specific to gestation.

Trimester	TSH level (mIU/L)
First	0.1–2.5
Second	0.2–3.0
Third	0.3–3.0
Note that individual labs may have slightly different pregnancy-specific ranges ranges and it is important to confirm ranges with your local pathologist.	

should be administered first thing in the morning, before food, and at least half an hour before other medications.[8] Patients should be advised to avoid taking drugs that may impair absorption directly post-thyroxine.[9] These include[9]:

- iron;
- calcium-containing supplements; and
- proton pump inhibitors.

Iodine supplementation

Pregnant women are recommended to ingest 220μg of iodine per day; women who are breastfeeding are recommended to ingest 270μg per day.[10] In 2007, a study demonstrated that Australian women are currently mildly iodine deficient, with a mean intake of 100μg per day.[3] Therefore, the recommendation is to take a supplement containing 150μg of iodine each day. This dose is safe and effective for pregnant and breastfeeding women. Pregnant women in search of vitamin and mineral supplements should be advised to check that the formulation includes the recommended amount of iodine.

Newly diagnosed OH

The incidental finding of OH during pregnancy screening should warrant immediate T4 replacement. A recent study suggested the average daily dosing of thyroxine in OH should be approximately 2.3μg per kilogram of body weight.[14] Despite this recent study the usual commencing dose of T4 for newly diagnosed OH would be 100–150μg per day.[7,15] The TSH should then be revaluated within four weeks, and dose adjustments made appropriately.

The diagnosis of OH during the perinatal period should warrant further enquiry:

- focus on patient history, screening for previous thyroid surgery, for treatment with radioactive iodine for previous Graves' disease, dietary iodine deficiency, drug induced hypothyroidism, and other autoimmune disease (particularly type 1 diabetes);
- examine for signs of hypothyroidism, which can affect the cardiovascular, neurologic and musculoskeletal system; and
- investigate for presence of thyroid autoantibodies (anti-TPO and anti-thyroglobulin antibodies, and thyroid receptor antibodies if there is a history of treated Graves' disease) and ongoing thyroid function test monitoring.

Pre-existing diagnosis of OH

There is an increased need for thyroxine replacement in women with pre-existing OH, owing to the normal physiology of pregnancy. In those with pre-existing OH, the thyroxine dose should be increased by 20–40 per cent from very early gestation.[2]

Currently, guidelines recommend optimisation of maternal TSH in the preconception period. After conception, an increase in T4 as soon as possible with the goal of normalising TSH is recommended. An easy approach is to increase total T4 dose by two tablets per week or by 20–40 per cent of baseline when pregnancy is diagnosed.[8,12]

The presence of thyroid-stimulating receptor antibodies (typical of Graves' disease) in a patient treated with complete thyroidectomy or radioactive iodine now requiring thyroxine replacement for treatment of their resultant hypothyroidism, will require specialist input. The persistent circulating maternal TSH receptor antibodies can cross the placenta and potentially have deleterious effects on the fetus, resulting in fetal hyperthyroidism. This could potentially cause fetal anaemia and heart failure. This condition warrants a notification to the obstetrician and referral either to an endocrinologist or to an obstetric medicine physician.

Subclinical hypothyroidism

Approximately 50–60 per cent of women with SCH will have positive antibodies and therefore all women with SCH must be tested for their presence. There are currently discrepancies in the guidelines relating to the treatment of subclinical hypothyroidism, and the role of T4 in reducing adverse outcomes is unclear.[13] However, the European Thyroid Association recommends treating all SCH women (TPO antibody positive and negative) with T4.[16] One approach would be to commence between 50–75μg per day T4 and then retest the thyroid function in four weeks.[7] A recent study which included only 77 women with SCH suggested slightly higher dosing may be needed.[14]

Monitoring

In those with treated OH, serum TSH should be monitored regularly in the first trimester to ensure the patient is euthyroid, and then at least once in the second and third trimesters.[16] The thyroid function tests should be re-checked four weeks after any dosage adjustments in thyroxine, to ensure the patient is euthyroid. Following delivery, the T4 dose should be reduced to the patient's preconception dose if they had been euthyroid on that dosage, with a follow-up TSH measurement approximately four to six weeks postpartum.[15]

Women with SCH in pregnancy who are not initially treated should be monitored throughout their pregnancy to detect

Table 2. Diagnosis and risks associated with subtypes of hypothyroidism in pregnancy.

Type	Lab diagnosis	Risks
Overt hypothyroidism	↑ TSH and ↓ fT4 *OR* TSH >10 (irrespective of fT4)	Fetal: prematurity, low birthweight, perinatal mortality, cognitive impairment and developmental delay[1] Maternal: anovulation, miscarriage risk, increased gestational hypertension, anaemia, postpartum haemorrhage[1]
Subclinical hypothyroidism and thyroid antibody negative	↑TSH and normal fT4 and no auto-thyroid antibodies	Higher risk of pregnancy loss, placental abruption, premature rupture of membranes, and neonatal death[13] Treatment advised by the European Thyroid Association[16], though unclear the value of T4 treatment in preventing adverse outcomes[13]
Subclinical hypothyroidism plus TPO antibody positive	↑TSH and normal fT4	A single study demonstrated reduction of miscarriage and preterm birth with thyroxine treatment[4]

progression to OH. Currently, guidelines suggest this should take place every four weeks during the first trimester of pregnancy, and then at least once in the second and third trimesters.[16] In the postpartum period we would usually cease therapy and recheck thyroid function tests four weeks later. If, however, the TSH was significantly elevated we would consider a more gradual reduction in T4 to ovoid OH, with repeat thyroid function tests every four weeks.

A failure to achieve a sufficient response in TSH after appropriate therapy should spur investigation into causes for lack of T4 absorption. This may represent one of the following: poor compliance; drug interactions; or impaired absorption.

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Tales from Timor: teaching and learning together

Dr Skanda Jayaratnam
FRANZCOG

Timor-Leste, as a nation, was born out of desire for independence – the independence vote in 1999 was unanimous, but the carnage that followed was overwhelming – infrastructure was destroyed and the health system fared no better. Many doctors left the country and the infrastructure, particularly in the regional areas, was close to non-existent. In the 16 years since, external aid, a concerted government investment in healthcare and a characteristic Timorese courage have helped to re-energise the health sector. Today, the beautifully designed Hospital Nacional Guido Valadares (HNGV), funded by international aid donations, is the largest medical facility in the country and treats thousands of patients each year.

The maternity unit at HNGV delivers approximately 4500 babies a year. The hospital also has a busy outpatient department (OPD) and gynaecological unit. The daily work is divided between the OPD – with referrals from clinical health centres (CHCs) – and operating theatres, on-call labour ward cover and daily rounds of O&G inpatients. There are six maternity cubicles, each with two beds, and an acute gynaecology room for presentations – typically ruptured ectopic pregnancies, heavy menstrual bleeding, miscarriage and acute pelvic inflammatory disease (PID). There are a large number of spontaneous vaginal deliveries, as it is the main maternity unit in a country of 1.2 million people. The caesarean section rate at HNGV in 2015 was 21.3 per cent, which reflects the complicated referred cases as well as the differing indications and thresholds for a caesarean section, depending on the background of the consultants in the unit. Instrumental delivery rates were 6.8 per cent in 2015, while there are large numbers of both breech vaginal and twin deliveries.

The O&G unit is staffed by six consultants, eight junior doctors (resident equivalent), five family medicine program (FMP) trainees (intern equivalent) and approximately 20 midwives and nurses. In addition, there is a consultant midwife from St John of God Hospital, Perth, who has become an invaluable member of the maternity unit. A problem faced in Timor-Leste is the number of languages used: there are specialist doctors from the Cuban brigade, the Chinese brigade, a United Nations Population Fund (UNFPA) employee who is Nepalese, two local Timorese consultants (trained in Indonesia) and me (trained in Australia). Spanish, Portuguese (a colonial remnant), Indonesian, Tetun and English are used in conversation daily as well as in the clinical notes, making understanding and conversing between health personnel and patients challenging, to say the least.

The Royal Australasian College of Surgeons (RACS) has had a presence in Timor-Leste since 2002. The project has been conducted under the Australia Timor-Leste Program of Assistance for Secondary Services (ATLASS), funded by the Australian Government and managed by RACS, in collaboration with the Timor Leste Ministry of Health. The project has made a significant contribution to the provision of clinical services and training of doctors in the hospital. Currently, the project involves the provision of five specialists – in the areas of surgery, paediatrics, emergency medicine, anaesthesia and O&G – to help with the training of the next generation of Timorese doctors, with a particular focus on postgraduate training. As there is currently no postgraduate training available in Timor-Leste for O&G, this has become one of the main aims of the RACS initiative here.

On the labour ward at HNGV.

Teaching caesarean section to junior medical officers.

Teaching ultrasound to FMP doctors. The ultrasound machine was kindly donated by Dr B Khara, Royal Darwin Hospital.

Members of the obstetrics and gynaecology unit at HNGV.

The current role of the O&G specialist is to train the FMP doctors, who spend eight to ten weeks rotating around the various specialities. This short but intensive period with the specialist within the O&G department aims to place them in good stead to manage and refer cases once they are sent to the CHCs, many of which are in isolated areas. The teaching is a combination of didactic lectures along with on-the-job training on the wards, OPD and in theatre.

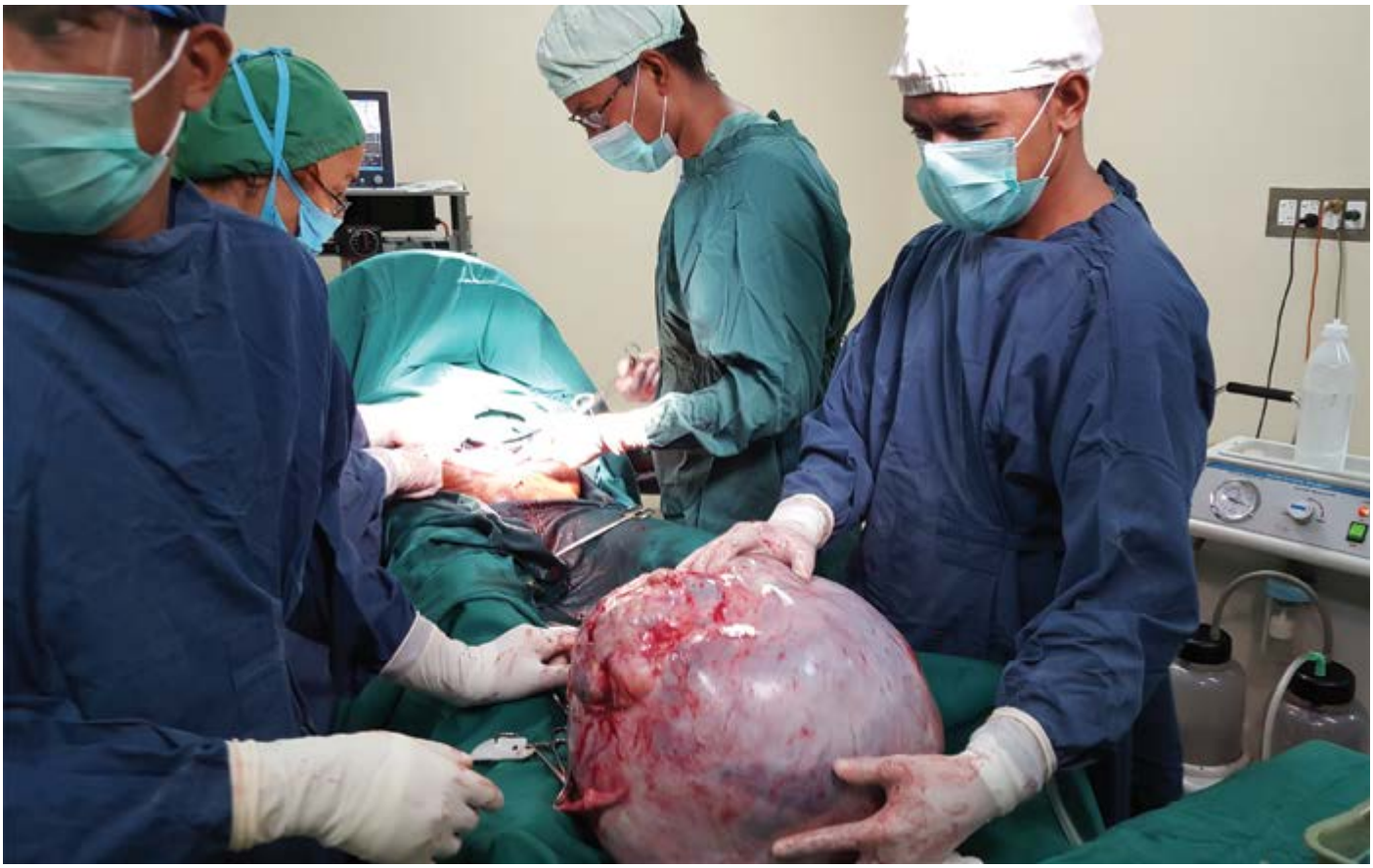
Teaching is not restricted to the FMP doctors. Both formally and informally, teaching is undertaken daily with the current junior medical officers and midwives on a range of topics, including interpretation of cardiotocography, instrumental deliveries, common obstetric emergencies and postoperative care. Daily teaching on ward rounds is also undertaken, according to the caseload available. This teaching is often juxtaposed with more formal multidisciplinary teaching sessions (involving midwives and doctors) on common procedural skills, such as breech extraction, shoulder dystocia management and performance of instrumental deliveries.

Besides the busy labour ward, most of the clinical time is spent in the OPD and operating theatres. The OPD clinic is made up of antenatal referrals and gynaecological presentations. Standard antenatal care is undertaken in the community; however, women with risk factors – large or small for gestational age, pre-eclampsia, twins, unsure dates – are sent to the OPD for a second opinion. Additionally, there is a large gynaecological component in the clinics, with referrals spanning from utero-vaginal prolapse, chronic pelvic pain from PID and endometriosis to heavy and irregular menstrual bleeding.

Ultrasound encompasses a significant amount of the work, as it provides not only the ability to date pregnancies to a reasonable degree, but also diagnose

Table 1. A summary of major procedures undertaken over a seven-month period at HNGV.

Procedure	Number
Caesarean sections	120
Instrumental deliveries	45
Laparotomy (TAH/ cystectomy/oophorectomy)	24
Laparotomy (ectopic pregnancies)	9
Vaginal hysterectomy and repairs	5



An 18.5kg right ovarian tumour removed at laparotomy in a 45-year-old; proceeded to total abdominal hysterectomy, left salpingo-oophorectomy and infracolic omentectomy.

malformations and assess the status of the fetus in utero. Diagnosis of gynaecological conditions, such as large ovarian cysts and uterine myomas, by ultrasound also makes diagnosis and pre-operative planning easier. Discussion and advice on difficult ultrasound interpretation as well as management of specific maternal-fetal medicine cases is often undertaken with colleagues from Australia.

There are three theatres and the O&G unit has three half-day elective lists per week. After-hours access to theatre is good; as theatre staff are present 24 hours a day and anaesthetists are available within 30 minutes. The range of pathology presenting to the OPD allows a relatively large number of vaginal and abdominal operations to be undertaken on the elective lists. As histopathology services are not available in Timor-Leste (and are very expensive for patients to send to Indonesia), anticipatory surgery is often undertaken based on a suspected pre-operative diagnosis.

Though the main aim of the employment by RACS of an O&G specialist was to assist in postgraduate teaching, there is a large service component to the role of consultant, along with providing improvements in the systems within the maternity unit. In seven months, 264 procedures were performed by me, with the major procedures listed in Table 1. The rest of the procedures consisted of vaginal breech and twin deliveries, manual vacuum aspiration/dilatation and curettage, minor vaginal surgery and tubal ligations. This does not include the supervision or assistance of midwives or doctors undertaking complex deliveries or the more than 400 ultrasounds undertaken to date.

There are a number of limited investigations available. Access to x-ray is relatively easy, but attaining computed tomography (CT) imaging is difficult due to the persistent breakdown of the machine. Access to basic laboratory results (full blood count; urea, electrolytes, creatinine; liver function test; and blood sugar level) is relatively easy and quick, though the hospital at times lacks the reagents for serology testing for HIV, hepatitis B and syphilis.

The other main component of assistance is the initiation (or re-initiation) of audits undertaken in the unit and the continued involvement of the unit in hospital multidisciplinary meetings. The main audits include maternal mortality and 'near miss' audits, perinatal mortality audits and caesarean section audits.

In addition to these duties, there is also interaction with various non-governmental organisations within the health sector in Timor-Leste – for example, Bairo Pite clinic and Marie Stopes International – in order to provide assistance in improving the provision of obstetrics and gynaecology care in the community.

Beyond the busy work schedule, there is the possibility to enjoy world-class diving, dining at various restaurants (a remnant of the UN presence here) and hiking/walking the many tracks along the beach and hills surrounding Dili. Visits to the regional towns – such as Baucau and Balibo – and Atauro Island also make for a nice weekend getaway from the busy schedule of the hospital.

Timor-Leste is a safe and friendly place a stone's throw from Australia's northern border and a great first step for anyone considering gaining experience in developing world O&G. Overall, the RACS O&G position at HNGV is an enjoyable, but demanding, job that goes far beyond the postgraduate teaching role that was initially envisaged. There is significant need for care

Members of the RACS team at HNGV.

that allows exposure not often seen in many developed countries and the opportunity to be able to not only provide a valuable service to the community, but also the ability to continually learn and develop one's capacity in this exciting and practical field.

As I complete my assignment in June, the 12-month position is now being advertised for July 2016. I am happy to provide further information on the role and can be contacted via RANZCOG.

The walk up to the 'Cristo Rei' statue is rewarding for the views it provides from the top.

Journal Club

Had time to read the latest journals? Catch up on some recent research by reading these mini-reviews by Dr Brett Daniels.

Robotic hysterectomy

Hysterectomy for benign conditions remains a common gynaecological surgery, with surgeons now having more choices with regard to the route of surgery. This column previously reported a study analysing trends in the route of hysterectomy at a single US hospital from 2004–12, showing an increase in laparoscopic hysterectomy accompanied by a decrease in abdominal hysterectomy over the same period.[1] A new study has compared robotic, laparoscopic, vaginal and abdominal routes in terms of intraoperative and postoperative complications.[2] Interestingly, the authors believed that previous studies analysing robotic hysterectomy may have been adversely affected by the lack of experience of many surgeons with the robotic approach, such that the results compared surgeons still in their learning curve with robotic hysterectomy against surgeons very experienced at laparoscopic, abdominal or vaginal approaches. Consequently, the current study only included cases performed by surgeons who had performed more than 60 previous robotic hysterectomies for benign conditions. The study analysed 2300 robotic-assisted, 9745 abdominal, 8121 vaginal and 11 952 laparoscopic hysterectomies performed across multiple US centres in 2012 and 2013. The robotic-assisted patient cohort had a significantly higher rate of morbid obesity than the vaginal or laparoscopic cohorts, and a significantly higher rate of large uteri (>250g) than the abdominal, vaginal or laparoscopic cohorts. The robotic cohort experienced significantly fewer intraoperative complications (including damage to other organs or blood transfusion) than the abdominal and vaginal routes, and experienced significantly fewer postoperative complications compared with all the other routes. The authors report that an economic analysis of their results is underway. At this stage their results suggest, in experienced hands, robotic-assisted hysterectomy for benign conditions may have benefits compared to the other routes.

1 Daniels B. Journal Club. *O&G Magazine*. 2015; 17(4):79.
2 Lim PC, Crane JT, English EJ, et al. Multicenter analysis comparing robotic, open, laparoscopic, and vaginal hysterectomies performed by high-volume surgeons for benign indications. *International Journal of Gynecology and Obstetrics*. 2016. http://dx.doi.org/10.1016/j.ijgo.2015.11.010.

Levonorgestrel versus copper IUCD

This paper reports on a randomised comparative trial of the levonorgestrel-containing IUCD (Mirena®) and copper containing IUCD (TCu380A ParaGard®) commenced by WHO in 1993, and conducted primarily in developing countries.[1] Approximately 1900 women were allocated to each device and followed up annually for seven years. Data were available at the seven-year follow-up for 682 women in the copper IUCD group and 398 women in the levonorgestrel group. The cumulative seven-year pregnancy rate of the levonorgestrel IUCD was 0.5 pregnancies per 100 women, a significantly lower rate than 2.5 per 100 for the copper IUCD. The discontinuation rates at seven years were 70.6 and 40.8 per cent respectively, although this difference does not achieve statistical significance. The main reasons for discontinuing the levonorgestrel IUCD were amenorrhea and reduced bleeding, particularly in Chinese women. For the copper IUCD the main reason for discontinuation was increased bleeding, especially among non-Chinese women. These results confirm that intrauterine devices are highly effective contraceptives. The changes in bleeding seen with both devices in this study is as would be predicted by most clinicians and there may be some value in ensuring women are aware of these potential changes before inserting IUCDs, and being aware of possible cultural differences in acceptability of these changes.

1 Rowe P, Farley T, Peregoudov A et al. Safety and efficacy in parous women of a 52-mg levonorgestrel-medicated intrauterine device: a 7-year randomized comparative study with the TCu380A. *Contraception*. 2016. (In press) http://dx.doi.org/10.1016/j.contraception.2016.02.024.

Day of delivery and obstetric outcome

All obstetricians are well aware that babies can be born at any time, meaning that obstetric services must function 24 hours a day, seven days a week. Ideally the standard of care and outcome would be uniform across the week but previous studies have shown the presence of a 'weekend effect', with a higher rate of perinatal mortality at weekends compared to weekdays. This study retrospectively analysed data from more than 1.3 million births in England and Wales between 2010 and 2012.[1] The authors reported a significantly higher rate of maternal puerperal sepsis, in-hospital perinatal mortality, neonatal injury and neonatal readmissions within three days, in babies born on weekends compared with babies born on weekdays. The reasons for such results were not able to be determined from the published study, but the authors postulate that levels of staffing and consultant presence may partially explain this result.

1 Palmer WL, Bottle A, Aylin P. Association between day of delivery and obstetric outcomes: observational study. *BMJ*. 2015; 351:h5774.

The Provincial Fellows Clinical Webinar program

Dr Anthony Geraghty
FRANZCOG
Chair
RANZCOG Provincial Fellows Committee

The College's Provincial Fellows Committee is proud to deliver continuing professional development opportunities to a screen near you.

The Provincial Fellows Clinical Webinar Series began in 2013, following on from the long-running and widely attended Clinical Teleconference Series. After the event, the presentation and a recording of the Q&A session is made available to the wider College membership via the RANZCOG CLIMATE eLearning website (www.climate.edu.au).

Each webinar features a keynote speaker, a RANZCOG Fellow and leader in their chosen field, delivering a 15–20 minute slide presentation designed to share knowledge and promote discussion. Keynote speakers prepare pre-reading reference material, which is made available to Provincial Fellows via Climate on registration for each webinar and the wider membership afterwards. A/Prof Ian Pettigrew, champion of the Provincial Fellows Clinical Webinar Series (and previously the Clinical Teleconference Series) introduces each speaker and invites questions, comments and discussion following the presentation.

Continuing professional development (CPD) points are allocated on the basis of one point for attending the webinar and one point for reading the pre-reading/reference material. Each webinar attracts two points. Fellows accessing the webinars via CLIMATE can claim one CPD point per hour under Self Education.

Webinars are an effective way for Provincial Fellows to access professional development in a range of topics important to rural and regional medical practice. Attending a webinar involves no travel time or cost. For further information, please contact Elizabeth Perini at: eperini@ranzcog.edu.au.

The 2016 webinar program.

Date and time	Topic	Speaker
26 April 2016 8pm AEST	Managing male infertility	Prof Steve Robson FRANZCOG
31 May 2016 8pm AEST	Current trends in laparoscopic surgery	A/Prof Anusch Yazdani FRANZCOG, CREI
28 June 2016 8pm AEST	Management of 55-year-old woman with a 6cm ovarian cyst	Prof Yee Leung FRANZCOG, CGO
26 July 2016 8pm AEST	The recent Cochrane review on transvaginal meshes	A/Prof Chris Maher FRANZCOG, CU
30 August 2016 8pm AEST	Ultrasound assessment of the fetus in the third trimester	Dr Patricia Lai FRANZCOG, COGU
27 September 2016 8pm AEST	Induction of labour for the large for gestational age/ macrosomic baby	Prof Susan Walker FRANZCOG, CMFM
25 October 2016 8pm AEST	Menopause management	Prof Martha Hickey FRANZCOG
29 November 2016 8pm AEDT	Obesity and the management of gestational diabetes	Prof Jodie Dodd FRANZCOG, CMFM

Attendee feedback

'I must say I enjoyed immensely my first session with your webinar and certainly benefited a fair bit from the information that was presented. I am sure I will benefit even more with subsequent webinars. Congratulations for a job well done for this webinar.'

'Thank you for the team that has been organising the webinar sessions, which I have found to be better than attending conferences and seminars at convention centres.'

'This last webinar session on urogynaecology was excellent. I shared the sentiments of several of the participants who had commented that it was an excellent session. There were many issues that were raised that were definitely relevant to those practising in the rural and remote regional centres. It is also enlightening to know that the problems that Prof Fraser raised regarding the credentialing of urogynaecology are well known and that attempts are being made to resolve them – certainly not an easy task. There should be more of these sessions so that the generalists in gynaecological practice are aware of the pitfalls in various aspects of subspecialised gynaecology and to tread carefully if they do venture into such areas of gynaecological surgery.'

Introducing the College's RAPID network

Carmel Walker
Senior Coordinator
RANZCOG Global Health Unit

RANZCOG recently launched an online discussion forum, the RAPID network, on the Climate eLearning website.

The purpose of the RAPID (RANZCOG Asia Pacific International Development) network is to forge links between College members with an interest in global health and international development as well as share items of interest and work opportunities.

While RANZCOG's global health involvement focuses on supporting the College's neighbours in the Oceanic region, other global health development and work opportunities can also be promoted on the RAPID network.

The aims of the network are to:

- link those with an interest in global health development, including early-career practitioners;
- provide a platform to share news about recent activities and achievements;
- host requests for, or offers of, services, equipment or other assistance;
- advertise global health development work opportunities, both paid and voluntary assignments;
- share links to useful resources, videos and educational tools;
- provide a forum where trainees from Australia, New Zealand and the Pacific Island Countries (PICs) can discuss issues;
- enable young O&G specialists and trainees in the PICs to seek a mentor, and for experienced O&G specialists with relevant experience to offer their expertise as a mentor to early-career O&G specialists and trainees.

RAPID is not intended for clinical discussion. If submitted, such conversations will be referred to experts for follow up offline.

If you have any questions, suggestions or ideas about the RAPID network, please contact Carmel Walker, Senior Coordinator, Global Health Unit at: cwalker@ranzcog.edu.au. Please contact elearningsupport@ranzcog.edu.au if you have questions about access to the CLIMATE site to enter the RAPID network.

To access the RAPID Network, please visit CLIMATE and login with your username (RANZCOG Member ID) and password. If you haven't visited the CLIMATE site before, click 'Yes' to agree to the RANZCOG eLearning Site Policy. Then type in your enrolment key. If you do not have one, or do not remember it, please contact elearningsupport@ranzcog.edu.au. Once you see the RAPID network landing page, you're in the zone to connect with global health opportunities and news.

There is a two-step process to access all CLIMATE resources: first login with username and password, then select the resource from the dropdown menu.

College Statements Update

March 2016

Prof Stephen Robson
FRANZCOG
Chair, Women's Health Committee

The Women's Health Committee (WHC) reviewed the following statements in March 2016, which were subsequently endorsed by Council. College statements can be viewed on the College website.

New College Statements

The following new statements were approved by RANZCOG Council and Board in March 2016:

- Cross-border reproductive care (C-Gyn 36)

Revised College Statements

The following revised statements were approved by RANZCOG Council and Board in March 2016:

- The use of misoprostol in obstetrics and gynaecology (C-Obs 12)
- Instrumental vaginal birth (C-Obs 16)
- Material group B streptococcus in pregnancy: screening and management (C-Obs 19)
- Collaborative maternity care (C-Obs 33)
- Standards in maternity care in Australia and New Zealand (C-Obs 41)
- Diethylstilboestrol (DES) exposure in utero (C-Obs 56)
- Combined hormonal contraceptives (C-Gyn 28)
- Guidelines for gynaecological examinations and procedures (C-Gyn 30)
- Consent and the provision of information to patients in New Zealand (C-Gen 2b)
- Evidence-based medicine, obstetrics and gynaecology (C-Gen 15)

Retired College Statements

- Rotational forceps (C-Obs 13)

The content of this document has been incorporated into the College Statement Instrumental vaginal birth (C-Obs 16).

College Communiqués

The following Communiqués have been issued:

- Vaginal seeding
- Zika virus

All College Statements and Communiqués can be viewed in the Women's Health section of the RANZCOG website.

Notice of Deceased Fellows

The College was saddened to learn of the death of the following RANZCOG Fellows:

Dr Peter McCormick, New Zealand, 23 October 2015
Prof Colin Peter Wendell Smith, Tas, on 15 December 2015
Dr Gary Basil Hastwell, WA, on 24 December 2015

The Royal Australian and New Zealand College of Obstetricians and Gynaecologists

RANZCOG FOUNDATION

Research Scholarships, Travel Grants & Fellowships 2016

The Royal Australian and New Zealand College of Obstetricians and Gynaecologists (RANZCOG) supports research in the fields of obstetrics, gynaecology, women's health and the reproductive sciences through the awarding of various scholarships, fellowships and grants. The RANZCOG Foundation proudly supports promising Fellows, clinical researchers and scientists undertaking high quality, innovative research and research training at an early stage in their career.

Call for Applications

The RANZCOG Foundation is pleased to announce that the Scholarships, Fellowships and Grants in the accompanying list are available for application in 2016, for commencement in 2017.

Applications must be made on the prescribed application form and submitted to the RANZCOG Foundation by **30 June 2016**. Applicants should ensure that their application is complete and all requested information is provided.

Applicants must also provide proof of Ethics Committee endorsement; however, this can be submitted after the close of applications if not available at the time of application.

Selection & Allocation Process

Applications are assessed by the RANZCOG Research Grants Committee. The selection and allocation process used is modelled on the National Health and Medical Research Council (NHMRC) processes with revisions to ensure, as much as possible, that early career support for high quality researchers with a commitment to women's health is promoted through the selection process.

Further Information

Visit **www.ranzcog.edu.au/foundation**

or contact the RANZCOG Foundation Coordinator
(t) +61 3 9417 1699 (e) foundation@ranzcog.edu.au

Research Scholarships, Fellowships and Grants

- Glyn White Research Fellowship **$60,000** (over two years)
- Luke Proposch Perinatal Research Scholarship **$20,000**
- RANZCOG Fellows' Clinical Research Scholarship **$40,000**
- RANZCOG/OvCan Scholarship **$15,000**
- Robert Wrigley Pain Research Scholarship **up to $24,000** (over two years)
- Taylor-Hammond Research Scholarship **$20,000**
- Mary Elizabeth Courier Research Scholarship **$30,000** (annually up to three years)
- UroGynaecological Society of Australasia (UGSA) Research Scholarship **$20,000**
- RANZCOG NSW Regional Committee Trainee Research Grant **$10,000**
- RANZCOG NSW Regional Committee Fellow Research Grant **$10,000**

Travel Grants and Fellowships

- ASGO International Travelling Fellowship **$10,000**
- ASGO National Travelling Fellowship **$5,000**
- Beresford Buttery Travel Grant **$5,000**
- Brown Craig Travel Fellowship **$5,000**

All amounts in Australian Dollars (AUD$)

Applications Close 30 June 2016

www.ranzcog.edu.au/foundation/research-grants-and-scholarships/

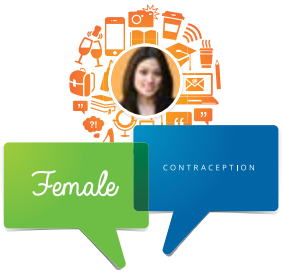
Meet Kim.

Kim is using a low dose oral contraceptive Pill which treats her moderate acne.[1,2]*

*YAZ®/ YAZ® Flex are indicated for the treatment of moderate acne vulgaris in women who seek oral contraception.

YAZ ethinyloestradiol/drospirenone | YAZ Flex ethinyloestradiol/drospirenone

YAZ® / YAZ Flex: 3 mg drospirenone, 20 mcg ethinyloestradiol. **Indications: YAZ / YAZ Flex** - Use as an oral contraceptive. Treatment of moderate acne vulgaris in women who seek oral contraception. **YAZ only** - Treatment of symptoms of premenstrual dysphoric disorder (PMDD) in women who have chosen oral contraceptives as their method of birth control. The efficacy of YAZ for PMDD was not assessed beyond 3 cycles. YAZ has not been evaluated for treatment of PMS (premenstrual syndrome). Refer to full product information. **Dosage and Administration: YAZ** - Take tablets in order directed on package at about same time daily, with liquid as needed. Tablet taking is continuous. Take one tablet daily for 28 consecutive days. **YAZ Flex** - YAZ Flex is an extended cycle oral contraceptive and can only be used in combination with a dedicated CLYK tablet dispenser. CLYK is a tablet dispenser designed to support the user to follow the YAZ Flex regimen. The instructions for use provided with CLYK should be read carefully before and during use. Once the dispenser pack containing the YAZ Flex tablets are inserted into CLYK, follow the prompts displayed on the screen to take YAZ Flex. Tablet taking is continuous. **Contraindications:** Presence or a history of venous or arterial thrombotic/ thromboembolic events (e.g. deep venous thrombosis, pulmonary embolism, myocardial infarction) or of a cerebrovascular accident; prodromi of a thrombosis (e.g. transient ischaemic attack, angina pectoris); presence of a severe or multiple risk factor(s) for venous or arterial thrombosis; diabetes mellitus with vascular involvement; presence or history of severe hepatic disease (as long as liver function values have not returned to normal); liver tumours (benign or malignant); malignant conditions of the genital organs or the breasts (if sex-steroid influenced); migraine (with focal neurological symptoms); pancreatitis or a history thereof if associated with severe hypertriglyceridemia; undiagnosed vaginal bleeding; severe renal insufficiency or acute renal failure; known or suspected pregnancy (Category B3); hypersensitivity to any of the components of YAZ/YAZ Flex. **Precautions:** Circulatory disorders; risk of thrombotic/thromboembolic events (e.g. age, smoking, obesity, family history of venous/arterial thromboembolism, prolonged immobilisation, atrial fibrillation, valvular heart disease); dyslipidaemia; lupus; porphyria; gallstone; hypertension; migraine; neoplasms; chloasma; depression; chronic inflammatory bowel disease (Crohn's disease, ulcerative colitis); lactation. Others: refer to full product information. **Interactions:** Medicines that induce microsomal enzymes (e.g. phenytoin, barbiturates, primidone, carbamazepine, rifampicin, griseofulvin, St John's Wort); medicines that inhibit microsomal enzymes (e.g. azole antifungals, macrolides, verapamil, diltiazem, grapefruit juice); HIV/HCV protease inhibitors and non-nucleoside reverse transcriptase inhibitors; antibiotics; cyclosporin; lamotrigine. Others: refer to full product information. **Adverse Effects:** Nausea, headache (including migraine), depression, depressive mood, breast pain, metrorrhagia, amenorrhoea, emotional lability, unscheduled uterine/genital tract bleeding. Others: refer to full product information. **Date of most recent amendment:** 17 July 2014. **References: 1.** YAZ® Flex Product Information, July 2014. **2.** YAZ® Product Information, July 2014.



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PBS Information: These products are not listed on the PBS.

Please review product information before prescribing. Full Product Information is available on request from Bayer Australia Ltd, or can be accessed from http://www.bayerresources.com.au/resources/uploads/PI/file10266.pdf or http://www.bayerresources.com.au/resources/uploads/PI/file10613.pdf

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