

2009

UWOMJ Volume 78, Issue 3

Western University

Follow this and additional works at: <https://ir.lib.uwo.ca/uwomj>



Part of the [Medicine and Health Sciences Commons](#)

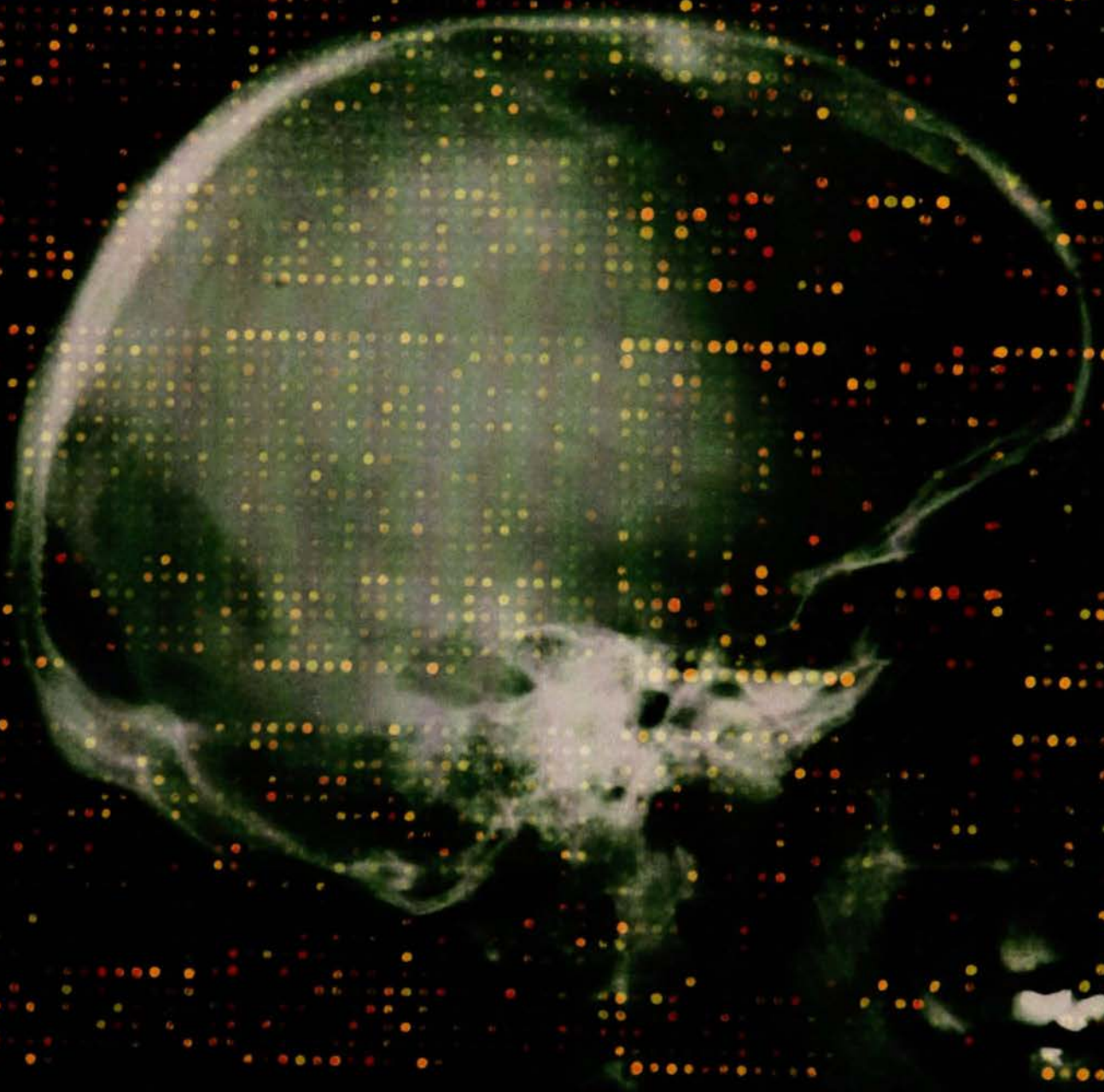
Recommended Citation

Western University, "UWOMJ Volume 78, Issue 3" (2009). *University of Western Ontario Medical Journal*. 18.
<https://ir.lib.uwo.ca/uwomj/18>

This Book is brought to you for free and open access by the Digitized Special Collections at Scholarship@Western. It has been accepted for inclusion in University of Western Ontario Medical Journal by an authorized administrator of Scholarship@Western. For more information, please contact tadam@uwo.ca, wlsadmin@uwo.ca.

UWOMJ

The University of Western Ontario Medical Journal
Volume 78, Number 3



GENETICS

CONTENTS

EDITORIAL

- Editorial 4 A Bump in the Road for the UWOMJ 5
Wendy Ng and Amber Menezes *Wendy Ng*

DEPARTMENTAL ARTICLES

- Clinical Procedures** 8 **Interdisciplinary Collaboration** 33
 Nailfold Capillaroscopy and Hereditary Multidisciplinary Management at Key Stages in the Huntington's Disease Neurodegenerative Process
Edward Weiss and Paul Lau *Abhijat Kitchlu and Allanah Li*
- Diagnostic Review** 13 **Medicine and the Law** 37
 Osteoarthritis: An Old Disease Cast in a New Light through Greater Understanding of the Human Genome
Rachel Bevan and Jason Essue *Who's in Your Genes: A Physician's "Duty to Warn" Patients' Relatives about Genetic Risk*
Colin Meyer-MacAulay
- Ethics** 18 **Medicine and Technology** 41
 Ethical Implications of Germ Line Genetic Engineering
Hassan Mir and Christina Morgan *Personalized Cancer Management*
Jenny Shu and Pencilla Lang
- Health Promotion** 22 **Thinking on Your Feet** 45
 The Blameworthy Gene
Jennifer N. Bondy and Laura Hinz *Marfan Syndrome*
Kalpa Shah and Aiman Alak
- History of Medicine** 27 **Zebra Files** 50
 Vampire Projects or Long Ago Person Found? A History of Genetic Research in First Nations Communities
Kate MacKeracher and Michael Livingston *Human Statues: Challenges in Management of Patients with Fibrodysplasia Ossificans Progressiva*
Anna Burianova, Ashley Brown, and Jenna Ashkanase

FEATURE ARTICLES

- Quality of Life Consequences of Long QT Syndrome 53 Nuchal Translucency and Prenatal Diagnosis of Congenital Heart Disease: Progress and Future Directions
Ishvinder Chattha and Caleb Zelenietz *Xiangning Fan*
- Genetic Susceptibility in Rheumatoid Arthritis 58 The Legacy of a Life Lived: Trans-Generational Epigenetics and Cancer Development
Kimberley Colangelo and Caleb Zelenietz *Mark Kirchhof*

CONTENTS

FEATURE ARTICLES

- | | | | |
|--|----|---|----|
| Amish, Mennonite and Hutterite Genetic Disorder Database
<i>Michael Payne</i> | 71 | Amniocentesis: Safety, Reliability and Alternatives
<i>Caleb Zelenietz, Kimberley Colangelo, and Ishvinder Chattha</i> | 79 |
| Hemochromatosis – Screening for a Common Condition
<i>Nick Sunderland</i> | 75 | | |

CASE REPORT

- | | |
|---|----|
| Ductal Carcinoma in situ in a 25-Year-Old Male with Unilateral Gynecomastia
<i>Christopher J. Coroneos</i> | 83 |
|---|----|

COVER ART

Front: Edward Weiss, Meds 2012.

In honour of UWOMJ's inaugural genetics issue, I thought it would be interesting to juxtapose the old and the new -- the x-ray and the microarray. Each represents an attempt to peer inside the inner workings of the human machine and divine its mysteries. Of course, while the Roentgenesque head can be easily identified as such, the radiograph gives us little information about the health and function of what lies beneath. The microarray, on the other hand, resembles nothing human at all, yet provides more information than we can really handle all at once. The challenge for medical genetics in the future will be the integration of these multiple modalities -- the shaping and moulding of mountains of data into findings we can see, feel, and hear.

Back: Julie Huang, Meds 2012.

Michelangelo's *The Creation of Man*, found on the ceiling of the Sistine Chapel depicts the hand of Adam reaching out to the hand of God. The cover references this iconic work, and brings into question the ways in which science and medicine use genetics as determinants of human creation.

EDITORIAL BOARD AND SUPPORT

Editors-in-Chief

Amber Menezes and Wendy Ng, Meds 2009

Managerial Stream

Managing Editor

Wendy Ng, Meds 2009

Senior Associate Editor

Tiffany Kwok, Meds 2010

Junior Associate Editor

Laura Hinz, Meds 2011

Contracts and Awards Stream

Executive Associate Editor

Renata Villela, Meds 2009

Senior Associate Editor

Jean Chen, Meds 2010

Junior Associate Editor

Pasquale Montaleone, Meds 2011

IT Director

Ishvinder Chattha, Meds 2011

Layout Editor

Wang Xi, Meds 2011

DEPARTMENTAL EDITORS

Clinical Procedures

Sr: Paul Lau, Meds 2011

Jr: Edward Weiss, Meds 2012

Diagnostic Review

Sr: Jason Essue, Meds 2011

Jr: Rachel Bevan, Meds 2012

Ethics

Sr: Christina Morgan, Meds 2011

Jr: Hassan Mir, Meds 2012

Health Promotion

Sr: Laura Hinz, Meds 2011

Jr: Jennifer N. Bondy, Meds 2012

History of Medicine

Sr: Michael Livingston, Meds 2011

Jr: Kate MacKeracher, Meds 2012

Interdisciplinary Collaboration

Sr: Abhijat Kitchlu, Meds 2011

Jr: Allannah Li, Meds 2012

Medicine and Technology

Sr: Pencilla Lang, Meds 2011

Jr: Jenny Shu, Meds 2012

Medicine and the Law

Sr: Abdullah Alabousi, Meds 2011

Jr: Colin Meyer-MacAulay, Meds
2012

Profiles

Sr: Emma Love, Meds 2011

Jr: Julie Hughes, Meds 2012

Thinking on your Feet

Sr: Aiman Alak, Meds 2011

Jr: Kalpa Shah, Meds 2012

Zebra Files

Sr: Jenna Ashkanase, Meds 2011

Ashley Brown, Meds 2011

Jr: Anna Burianova, Meds 2012

UWOMJ ADVISORY BOARD

Dr. Lois Champion

Dr. David Colby

Dr. Kellie Leitch

Dr. Jeffrey Nisker

Dr. Douglas Quan

Dr. Faisal Rehman

Dr. Michael Rieder

Dr. Jim Silcox

Dr. Wayne Weston

Dr. Ron Wexler

PRINTER

InPrint Design: University Students' Council, University of Western Ontario

The mapping of human genes has been an important step in the development of medicines and other aspects of health care, emphasized by the worldwide interest in the Human Genome Project that began in 1990 in the United States at the National Institutes of Health. It is with this deep respect for the profound importance of genetics that we are proud to present our newest issue of the University of Western Ontario Medical Journal. Case reports, reviews, and in-depth discussions cover a variety of topics on this fascinating theme. Major implications for the future, present and past underlie each of these articles.

The future is a focal point of both speculation and hope when it comes to the detection of blood vessel anomalies and microvascular disease, as described in an enlightening article within the Clinical Procedures departmental section herein. Prenatal genetic diagnosis helps us predict the future of an unborn child, as highlighted in a feature article in this issue. Meanwhile, on a broader scale, the endless possibilities behind the cutting edge technology in personalized cancer management are also explored in the Medicine and Technology departmental section. A feature article discusses how our seemingly innocuous current environmental exposures can even affect our future generations!

And yet, despite all its predictive power, genetic diseases continue to burden society in the present time. What are the implications of long QT syndrome on quality of life? What role does genetics play for patients diagnosed with rheumatoid arthritis? How should we screen for hemochromatosis? Our authors address these issues. Far more uncommon conditions, such as

fibrodysplasia ossificans progressa, can be detected early with genetic screening today, as described in the Zebra Files. Our Interdisciplinary Collaboration authors weigh in on the necessity of support when it comes to Huntington's Disease. But the follies of genetics are also addressed – what does it mean to label one another as ill or healthy, particularly when it comes to alcoholism? This dilemma is eloquently outlined in our Health Promotion departmental article.

It is also essential to look back in time. Our contributors do not forget to take an in depth look at the past. As our History of Medicine departmental article demonstrates, genetic research has affected indigenous peoples, and ethical considerations have arisen from historical challenges, leading to stricter implementation of informed consent principles today.

Genetics connects all of humanity, from past to present to future, and underscores the similarities and differences between us all. Understanding our past allows us to plan for the future. The recent period of volatility for the University of Western Ontario Medical Journal is highlighted in this issue, with a reflection piece that addresses the major changes that have occurred within the journal's structure, operations, and content, ultimately producing the impressive collection of quality articles that you see now.

We hope that you find this issue relevant in looking towards the future, keeping up with the present, and understanding the past when it comes to genetics and its wide-ranging impacts. Enjoy!

- Wendy Ng and Amber Menezes
Editors-in-chief

A Bump in the Road for the UWOMJ

In 2005, the University of Western Ontario Medical Journal had not printed an actual issue in two years, despite its long, successful history as Canada's second oldest medical student journal. At that point, the only printed issue that had been distributed in recent memory was the *Obstetrics and Gynecology* issue. Meanwhile, the *Pediatrics* issue had been laid out in full – with cover art, student articles and advertisements included – but there were no funds for printing, nor a printing service set up. However, there were still bills to be paid for the layout of the *Pediatrics* issue. At the same time, some articles had already been collected for the *History of Medicine* issue, but there was no realistic plan in place for the issue to go forward.

My four years as the managing editor of the UWOMJ have been extremely rewarding. When I both started medical school and joined the UWOMJ in 2005, the position of “Managing Editor” was newly created and loosely defined – my original job description was merely to implement a peer or faculty review process for student articles – but I learned quickly there was so much more to be done, and I vastly expanded my role.

The company that had taken care of UWOMJ's layout, printing and distribution in previous years reportedly went bankrupt and fled North America. My phone calls and emails were not returned. The UWOMJ was caught unprepared in a confusing state of affairs. Many students were frequently asking me when their articles would be printed and questioning when the next issues would be coming out. Multiple advisors recommended to me that I should simply put a stop to printing the journal at all, and that I resort to online copies only. But this was not an acceptable answer to me – it was of utmost importance that the UWOMJ remain in print.

It was with this realization that I began to seek out alternatives for both advertising and printing. Individually soliciting advertisements for the journal was not at all an easy task – I came to this discovery through trial and error. I contacted CU Advertising, and with the help of my colleague Renata Villela, we signed our first contract with them for the *Oncology* issue. At the same time, I looked for a new printing company. I visited several, collecting proofs and comparing prices, before I finally decided to work with Imprint. I was confident that their staff would ensure that the journal was printed to our high standards.

The *Oncology* issue was the first edition of the UWOMJ to accept only articles reviewed and approved by faculty at UWO. Recruiting contributors was a challenge in itself, and I designed an original new layout for the issue. I contacted dozens of faculty in 2005, asking them to join our faculty review board. Articles that did not meet rigorous review standards were rejected. I designed and sent out a peer review form to reviewers and requested countless revisions from authors. I acted as a mediator between all authors and reviewers to maintain absolute anonymity in the process.

Finally, I needed a new process for distribution of the journal. I put together a long list of mailing addresses, and we had our first “mailing label sticker party” in the UWOMJ office. Amber and I carried the journals down to the mailroom, and the bulk of our very first new journals were on their way!

However, a sustainable structure for the UWOMJ was sorely needed. Renata offered to take care of the contracts, while I worked on the journal itself. This was how the two divisions of the Editorial Board of the journal were born: Renata took charge of the Contracts and Awards

Division, while I headed up the Managerial stream. The backlogged *Pediatrics* and *History of Medicine* issues were published online.

When Amber Menezes helped collect articles for the subsequent *Family Medicine* issue, authors were responsible for ensuring that their articles were indeed reviewed and approved by UWO physician faculty members before submission. This process has proven sustainable and has maintained the high quality of articles appearing in the UWOMJ.

The first online summer supplement of the UWOMJ was put together by Stephen Chihrin in 2005. This has become an annual tradition. Tiffany Kwok continued to produce the summer supplement and put together the *Cardiovascular* issue, our first alternatively funded issue. She also ensured that the UWOMJ office was clean and in a functional state for our editorial staff – a massive task, given the mess that it was in before!

Renata instituted some amazing new changes for the UWOMJ. Recently, she has spearheaded the process for the UWOMJ to be catalogued by PubMed. Notably, she suggested that we recognize our writers' and artists' talents and hard work with departmental and feature article awards, as well as cover art awards. We had our first Awards meeting in the early summer of 2008. Dr. Faisal Rehman, who has been our most active faculty supporter from our rocky start,

provided much-appreciated food and encouragement for this first meeting, and generously funded the awards.

We are incredibly lucky to have a remarkable editorial team taking on the immensely rewarding and challenging experience of running the UWOMJ. Laura Hinz organized our first Awards Ceremony, where plaques were presented to very deserving winning contributors. She also initiated the creation of the new Interdisciplinary Collaboration department of the medical journal, reflecting the growing multidisciplinary nature of health care today.

It has been unmistakably clear that the transition into sustainable operations has progressed spectacularly. We are fortunate to have an extraordinary group working on ensuring that the UWOMJ continues its high-quality publishing and distribution, while consistently exceeding our readership's expectations. The graduating students of the UWO Medicine Class of 2009 have seen our medical journal recover dramatically over the past few years from its lowest point in 2005. I eagerly look forward to following the journal as a UWO alumnus, with such a capable new editorial team at the lead!

Wendy Ng
Managing Editor-in-chief

Join the hospital that's building for the future.

Humber River Regional Hospital is quickly earning a reputation for excellence in the delivery of acute medical care. We offer a full range of medical services, and are working actively to make our hospital a better place – for our patients and our physicians.

NEW HOSPITAL APPROVED

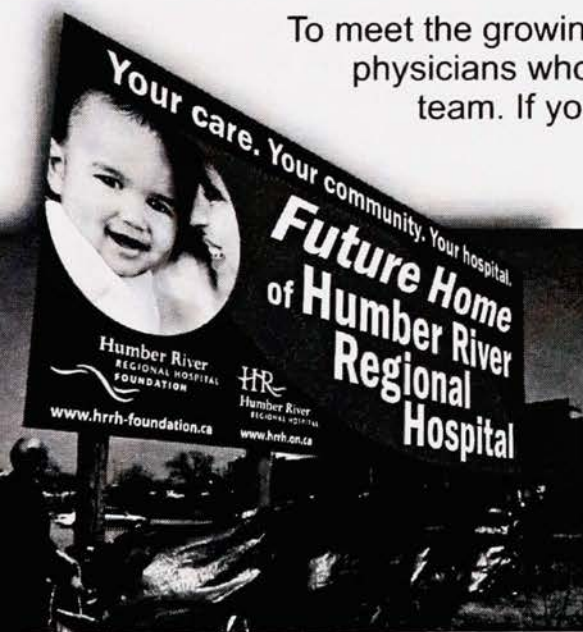
Our redevelopment plan, officially approved by the Ontario government, will see us move to a new showcase acute care facility in 2014. By joining Humber River now, you can be a part of program and facility planning for Ontario's newest hospital project. Help us build our new hospital; one we plan to make the technologically advanced centrepiece of our New Model of Care. Preliminary site work is underway, and planning has already begun.

We offer placements for clinical clerks, residents and clinical fellows. Humber River Regional Hospital is affiliated with University of Toronto and a number of HRRH physicians hold teaching appointments at the University.

EXCITING CAREER OPPORTUNITIES

Fundamental to the success of our hospital are the remarkable individuals who comprise our medical team. Their expertise, commitment and caring are integral to fulfilling our vision as *the hospital of choice ... committed to patient and family needs.*

To meet the growing community demand for our services, we are looking for physicians who demonstrate excellence in clinical care to join our medical team. If you are interested contact us by email: mlatter@hrrh.on.ca



Nailfold Capillaroscopy and Hereditary Disease: Current Applications and Future Prospects

Edward Weiss (Meds 2012) and Paul Lau (Meds 2011)

Faculty Reviewer: Dr. Janet Pope

Nailfold capillaroscopy is a proven non-invasive method of examining the peripheral microvasculature. Primarily used to assess rheumatological disease, it is being increasingly recognized as a versatile tool that can detect blood vessel abnormalities in a wide range of conditions, including a number of hereditary diseases. As a complement to genetic testing and other clinical examinations, capillaroscopy can often yield important diagnostic clues, and has the potential to aid research in hereditary conditions as disparate as telangiectasia, glaucoma, and schizophrenia.

Introduction

At an estimated length of between 50,000 and 100,000 miles, the vascular system of the human body holds the distinction of being longer than the road networks of many small countries. As a conduit for metabolites, hormones, electrolytes, and drugs, it excels in its function and is exquisitely sensitive to constant changes in supply and demand. Its very pervasiveness can, however, act as a double-edged sword – pathologies that affect one area of the vasculature, such as atherosclerosis or Marfan's syndrome, are likely to affect other areas as well.

As a diagnostic tool, the tight integration of the vasculature with all other body organs and systems can often be used to the clinician and patient's advantage, since signs observed at one point in the vessel network can indicate pathology at a distant site. The presence of caput medusa around the umbilicus, for example, is useful in diagnosing portal hypertension and inferior vena cava obstruction. Often, however, abnormalities in the vascular system are more subtle and require suitable clinical skill to ascertain them.

Nailfold capillaroscopy (NC) is the *in vivo* microscopic examination of the capillaries found in the skin just proximal to the nail. The capillaries in this area run parallel to the surface of the skin, and can be easily visualized with

appropriate lighting and magnification. Using only a magnifying glass, the 19th century Italian physician Giovanni Rasori was able to describe abnormal capillaroscopic findings and related them to conjunctival inflammation.¹ In the twentieth century, the practice of NC was refined and now it is used most prominently to detect markers of rheumatological disease, such as systemic sclerosis. However, there are indications that NC may also be helpful in the diagnosis and prognosis of various hereditary conditions that involve the microvasculature. In this article, we hope to illustrate a general approach to NC as a tool in assessing rheumatological pathologies as well as its potential utility in the context of genetic disease.

Procedure

Traditionally, NC has been performed with a light microscope. The patient is seated and acclimatized to the ambient temperature of the examining room. Immersion oil or clear gel is applied to the nailfold area to enhance transparency of the skin and make the capillaries more visible, and the light source is positioned at a 45 degree angle to the nailfold. A green LED or green-filtered light is often used to further enhance visibility, as light in the blue-green part of the spectrum is preferentially absorbed by hemoglobin. A wide view of the terminal capillaries can be obtained with a 10x-60x

magnification, and more detailed views can easily be achieved with higher magnification (250x-1000x).² Photomicrographs can be taken at the time of examination for further study or archival purposes.

The capillaries can also be examined with hand-held instruments, such as an ophthalmoscope or a dermatoscope, although these often make it more difficult to spot areas of diminished vascularity.³ More recently, specialized videocapillaroscopic systems have been developed to facilitate the analysis of capillary abnormalities, although the expense of these systems may be prohibitive for some clinicians.

The more common findings in patients with microvascular abnormalities include the presence of large or giant capillaries, disorganization of the vascular array, tortuous vessels, microhemorrhage, loss of capillaries, and ramified, or "bushy," capillaries.⁴

Rheumatological Findings

Classically, nailfold capillaroscopy has been utilized to differentiate patients presenting with isolated Raynaud's phenomenon (RP) from those who may have underlying connective tissue disorders causing secondary RP. The nailfold capillaries of healthy patients and isolated RP patients appear as parallel hairpin loops with visible afferent, transitional and efferent portions (Figure 1). In contrast, secondary RP patients may possess deviatory capillary patterns directing the clinician to investigate for certain connective tissue or autoimmune rheumatic diseases.⁵

The scleroderma pattern has been extensively characterized and represents a multitude of nailfold capillary changes seen in patients with systemic scleroderma.⁶ Three variations of the scleroderma pattern have led to the categorization into early, active and late phases (Figure 2). Specific findings that have been described include giant capillaries proposed to be a result of an autoregulatory response to tissue hypoxia, capillary microhemorrhages due to early vascular damage, avascular fields due to

increasing hypoxia and architectural disruption of capillaries.

Similar to systemic sclerosis, variations of the scleroderma pattern are also seen in patients with other connective tissue diseases such as dermatomyositis, systemic lupus erythematosus, antiphospholipid syndrome, Sjogren's syndrome and occasionally rheumatoid arthritis.⁷



Figure 1. Normal nailfold capillaries. (Courtesy of Dr. Joerg Piper)

Nailfold Capillaroscopy in Hereditary Hemorrhagic Telangiectasia

An autosomal dominant disorder characterized by easily bleeding telangiectases on the skin and mucosal surfaces, hereditary hemorrhagic telangiectasia (HHT; OMIM 187300) can lead to arteriovenous malformations in many organs and paradoxical septic emboli.⁸ Therefore, early diagnosis is important for better prognosis. Currently, three typical telangiectases are required to make the diagnosis of HHT, with genetic testing only positive for a small subset of families. If not diagnosed, arteriovenous malformations and septic emboli may be missed. One study demonstrated that over 80 percent of patients with HHT show enlarged afferent portions of the capillary loop, a finding that was not evident in matched patients without HHT.⁸ More importantly, five of nine patients without visible macroscopic cutaneous telangiectases but having a clinical diagnosis of HHT showed enlarged afferent capillary loops. Although further studies are needed to validate this, nailfold capillaroscopy appears promising as a diagnostic aid when a definitive diagnosis of HHT can't be

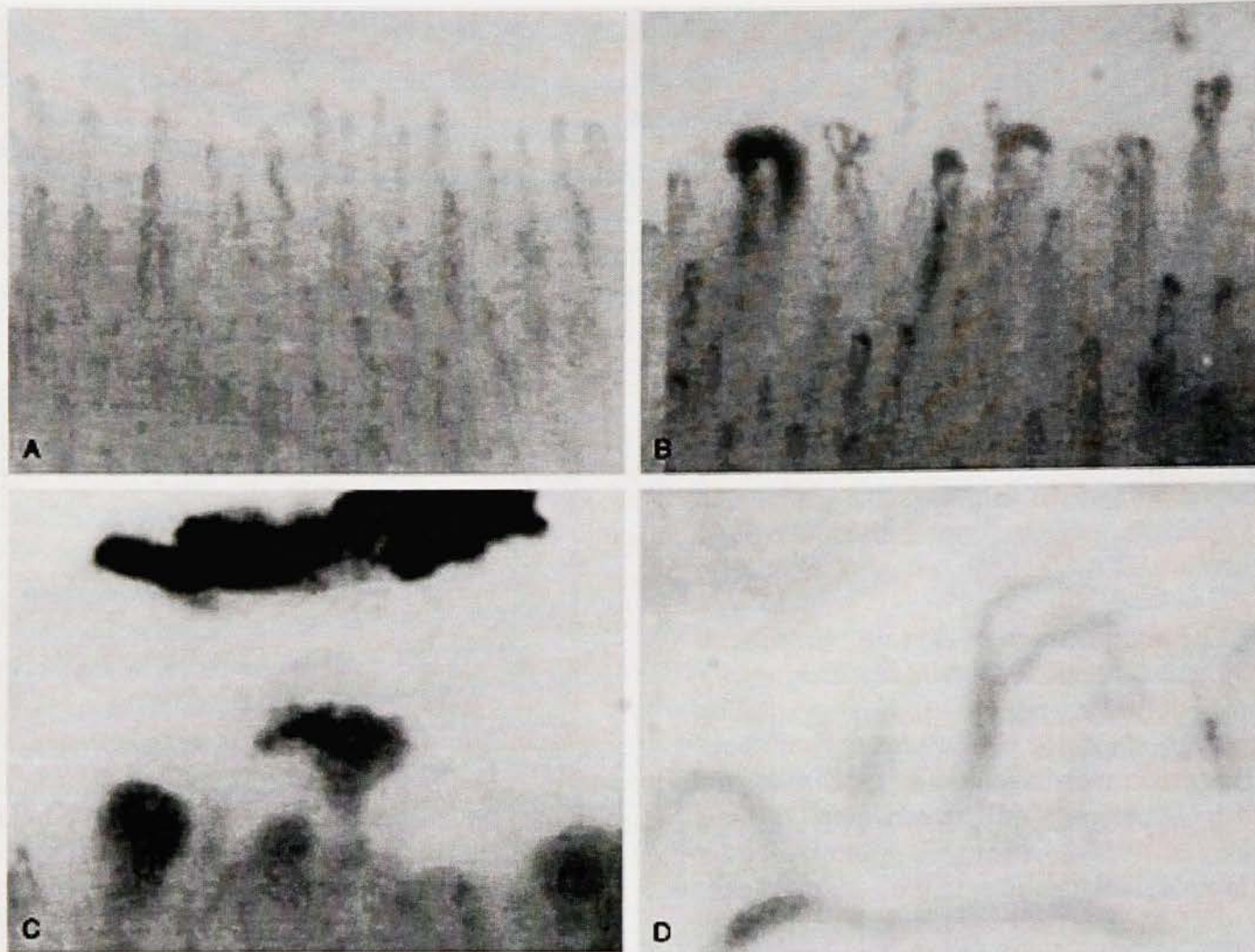


Figure 2. Capillaroscopy findings in systemic sclerosis. (A) Normal nailfold pattern. (B) Early scleroderma pattern, with dilated and giant capillaries. (C) Active scleroderma pattern shows frequent giant capillaries and hemorrhages. (D) Late scleroderma pattern shows severe capillary architecture disorganization. (Courtesy of Dr. Soumya Chatterjee)

made based on clinical information alone (for example, if it is suspected but not yet proven).

Capillary Blood Flow Measurements and Glaucoma

As one of the leading causes of blindness in the world, glaucoma represents a significant threat to ocular health and day-to-day function. One of the most common subtypes, primary open-angle glaucoma (POAG; OMIM 137760), is known to have a significant genetic component, as the prevalence of POAG among those with a positive family history is 5- to 20-fold greater than the normal population.⁹ The progression of POAG is often insidious, and it is estimated that more than half of those with the disease remain unaware of their condition until their vision begins to

deteriorate.¹⁰ Thus, screening measures for those who have a family history of POAG and others at increased risk have an important role in preventing the progression of glaucoma to visual damage and blindness. Unfortunately, the classic sign of developing glaucoma, a high intra-ocular pressure, is only detectable in about half of those with the disease.¹¹

Although the precise pathophysiology of POAG is complex and still not completely understood, it has been noted that aberrations in systemic blood flow are often correlated with the incidence of glaucoma.¹² One such abnormality is the degree of peripheral capillary vasospasticity after a period of cooling. Using capillaroscopy, it is possible to measure blood flow in the nailfold vessels and estimate the extent to which flow

stops in response to cold. Patients with POAG often show an exaggerated vasospastic response relative to normal controls, which may indicate a systemic abnormality of blood flow regulation.¹² Thus, capillaroscopic examination in patients with risk factors for POAG may be useful in research and eventually in screening for this disease, especially when an elevated intra-ocular pressure is not detectable.

Capillary Abnormalities and Psychiatry

One of the more intriguing domains of medicine that can be examined with capillaroscopy relates to hereditary mental illness, namely schizophrenia. As early as 1939, researchers discovered that a disproportionate number of patients with schizophrenia exhibited abnormalities of the nailfold capillaries and increased visibility of the nailfold plexus, the dense subcutaneous network of capillaries found proximally adjacent to the fingernail, which is generally not visible past puberty.¹³ Typical findings upon examination of the nailfold include a highly visible capillary plexus (Figure 3), and oddly-patterned individual capillaries with numerous horizontal anastomoses.¹⁴

Subsequent studies in the 1960's and onwards aimed to establish an objective scale for measuring visibility of the plexus and concurrently found evidence that the endophenotype of high plexus visibility was positively correlated with familial, rather than sporadic, schizophrenia,¹⁴ as well as more severe and more negative symptoms.¹⁵ Conversely, it is estimated that 70% of patients with familial schizophrenia have an elevated plexus visibility.¹⁶ This demonstrates that schizophrenia is a more complex disease, with abnormalities that extend beyond the brain in many patients.

Taken together, these data suggest that an elevated plexus visibility has the potential to be a suitable non-invasive biomarker for those at risk of developing hereditary schizophrenia or schizotypic personality features.¹⁷ Given the young age at which many patients with schizophrenia begin to show symptoms, the ability to screen for nailfold abnormalities could

possibly enable families with a history of schizophrenia to be mindful of early manifestations of mental illness and seek appropriate treatment. Active research in this area is underway, and it is hoped that being able to identify a distinct subclass of people with schizophrenia may assist in future genetic studies aimed at discovering the underlying etiology for a truly debilitating disease.

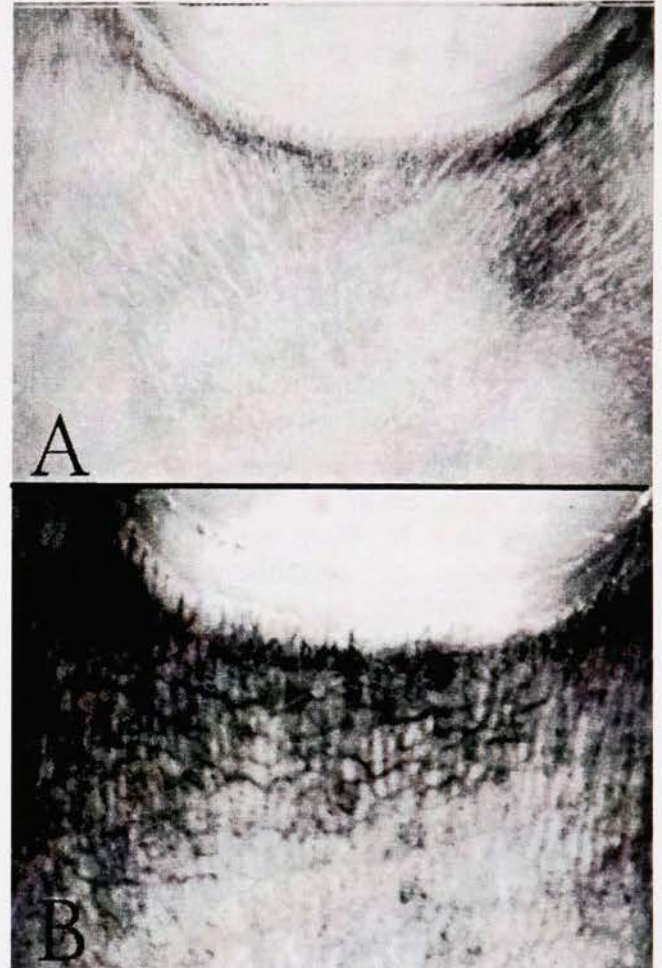


Figure 3. Nailfold venous plexus images with associated Maricq plexus visibility scores (0 = no visible plexus, 4 = highly visible plexus). (A) Normal, 0/4 on Maricq scale. (B) Abnormal, 4/4 on Maricq scale. (Courtesy of Dr. John Vuketich)

Conclusion

Nailfold capillaroscopy is a tried and true clinical method that has applications to many disparate pathologies. While the usual investigations that comprise genetic workups cannot be replaced by a simple observation of nailfold capillaries, a thorough examination of the microvasculature can often yield clues that aid in

diagnosis and prognosis, with a minimum of expense and invasiveness. It is important to be aware that capillary abnormalities and other vascular signs can be missed if not sought and that magnification of the nailbeds in patients with many diseases can be a useful and relatively easy skill to acquire.

Acknowledgements

We thank Drs. Joerg Piper (<http://www.joerg-piper.com>), Soumya Chatterjee, and John Vuketich for contributing helpful images of nailfold findings.

References

1. Cutolo M, Grassi W, Matucci Cerinic M. Raynaud's Phenomenon and the Role of Capillaroscopy. *Arthritis Rheum* 2003;48(11):3023-3030.
2. Barker JH, Anderson GL, Menger MD. Clinically applied microcirculation research. Boca Raton, FL: CRC Press, 1995:325.
3. Baron M, Bell M, Bookman A, *et al*. Office capillaroscopy in systemic sclerosis. *Clin Rheumatol* 2007;26:1268-1274.
4. Cutolo M, Pizzorni C, Sulli A. Capillaroscopy. *Best Pract Res Clin Rheumatol* 2005;19(3):437-452.
5. Blockmans D, Beyens G, Verhaeghe R. Predictive value of nailfold capillaroscopy in the diagnosis of connective tissue disease. *Clin Rheumatol*. 1996;15(2):148-53.
6. Cutolo M, Sulli A, Secchi ME, Olivieri M, Pizzorni C. The contribution of capillaroscopy to the differential diagnosis of connective autoimmune diseases. *Best Pract Res Clin Rheumatol*. 2007;21(6):1093-1108.
7. Cutolo M, Sulli A, Secchi ME, Paolino S, Pizzorni C. Nailfold capillaroscopy is useful for the diagnosis and follow up of autoimmune rheumatic diseases. A future tool for the analysis of microvascular heart involvement? *Rheumatology (Oxford)*. 2006;45 Suppl 4:iv43-6.
8. Mager JJ, Westermann CJ. Value of capillary microscopy in the diagnosis of hereditary hemorrhagic telangiectasia. *Arch Dermatol*. 2000;136:732-4.
9. Tsai JC, Forbes M. *Medical Management of Glaucoma*. West Islip, NY: Professional Communications, 2008:18.
10. Lieberman MF. Glaucoma in the World: The elusive challenges of a major cause of blindness. *Cataract and Refractive Surgery Today*. 2005 Oct;62-63.
11. Harasymowycz P, Fansi AK, Papamatheakis D. Screening for primary open-angle glaucoma in the developed world: are we there yet? *Can J Ophthalmol* 2005;40:477-486.
12. Pache M, Dubler B, Flammer J. Peripheral vasospasm and nocturnal blood pressure dipping—two distinct risk factors for glaucomatous damage? *Eur J Ophthalmol*. 2003;13(3):260-5.
13. Olkon D. Capillary structure in patients with schizophrenia. *Arch Neurol and Psychiat* 1939;42:652.
14. Maricq HR. Capillary Pattern in Familial Schizophrenics: A Study of Nailfold Capillaries. *Circulation* 1963; 27:406-413.
15. Poole JH, Maricq HR, Alson E, Willerman L. Negative symptoms in schizophrenia and nailfold plexus visibility. *Biol Psychiatry* 1991;29(8):757-73.
16. Maricq HR. Familial schizophrenia as defined by nailfold capillary pattern and selected psychiatric traits. *J Nerv Ment Dis* 1963;136:216-26.
17. Vuchetich JP, Liska JL, Dionisio DP, Stanwyck JJ, McGuire KA, Sponheim SR. Elevated nailfold plexus visibility aggregates in families and is associated with a specific negative symptom pattern in schizophrenia. *Psychiatry Res* 2008;160(1):30-7.

Osteoarthritis: An Old Disease Cast in a New Light through Greater Understanding of the Human Genome

Rachel Bevan (Meds 2012) and Jason Essue (Meds 2011)

Faculty Reviewer: Dr. Andrew Leask

The successful mapping of the human genome offers the potential to greatly advance our understanding of various disease processes. Osteoarthritis is a disease of particular interest because there is significant morbidity and societal burden associated with this condition. Furthermore, the prevalence of this disease is expected to rise dramatically with the aging of the baby-boomer generation (individuals born between 1946 and 1964), with a concomitant elevation in health care costs in Canada and abroad. Osteoarthritis was previously viewed as a commonplace disease resulting from environmental 'wear and tear', that was an inevitable part of aging. However, new advances have demonstrated that osteoarthritis is a surprisingly complex multi-factorial disease that is affected by both environmental and genetic variables. This review discusses some of the environmental and genetic factors that influence the progression of osteoarthritis. Recent advances in biomarkers that are associated with increased disease risk are also discussed. Finally, the potential impact of genetics on both early diagnosis of disease, and prediction of how a patient will respond to a particular pharmacological treatment is presented.

Introduction

Ever since the discovery of the three-dimensional structure of DNA¹ the field of genetics has intrigued both scientists and the general public alike for its potential impact on disease diagnosis. Indeed, the concept of genetics as a primary player in the role of diseases is assumed by most of the lay public today. One of the goals of understanding genetic influences in the disease process is to aid in treatment of patients through the development of a personalized medical approach. Predictive genetic testing is one area that has garnered great interest in the past decade, especially since the sequencing of the human genome.² The goal of predictive genetic testing is to determine if an individual is likely to develop a disease state based upon the presence/absence of particular genes or biomarkers. Although challenging, determining how best to treat a patient based on their genetic make-up is simplified in the cases where a particular gene or mutation is the direct cause of particular disease. For example, cystic fibrosis is

an autosomal recessive disease; for the disease to occur, each copy of the CFTR gene must have a mutation that renders the resultant protein non-functional.^{3,4}

However, in many cases the disease process is not straightforward; the presence of a particular genetic variant doesn't necessarily lead to the disease. Instead, diseases, especially those of a chronic nature, are multi-factorial, resulting from a complex set of interactions among many different genetic and environmental factors. In general, there is no single gene that causes the disease, but genetic factors are likely to contribute to an increased probability of developing the condition. Thus, it is imperative to develop methods to identify individuals at risk.

Osteoarthritis (OA) is one example of a multi-factorial disease that is influenced by genetic and environmental variables. OA was previously viewed as a commonplace disease resulting from environmental 'wear and tear' that was an inevitable part of aging.⁵ However, new

advances have demonstrated that OA is a surprisingly complex multi-factorial disease that is affected by both environmental and genetic variables.^{5,6} This review discusses some of the environmental and genetic factors that influence the progression of OA. Recent advances in biomarkers that increase disease risk are also discussed. Finally, the potential impact of genetics on both early diagnosis of disease, and prediction of how a patient will respond to a particular pharmacological treatment are presented.

Disease Burden of Osteoarthritis

OA affects 10-12% of the adult population in North America and is a leading cause of pain, physical disability, and use of health care services.^{7,8} OA is a degenerative disease characterized by the early deterioration of articular cartilage from within the joint, and later by complete loss of articular cartilage, damage to the subchondral bone, severe deformities, and disabling pain.^{5,9,10} The consequences of disease are detrimental to overall quality of life¹¹ in many ways, including fatigue, reduced income due to impaired labour force engagement, hospitalization, surgery, medication side effects, family instability, and decreased social participation.⁸ Alarming, the number of people affected by OA is expected to increase dramatically as the baby-boomer generation ages. For instance, in Canada approximately 3 million people are currently living with OA. This figure is expected to rise to 5 million by the year 2026.¹² Unless better methods are developed to diagnose and treat OA, the direct and indirect costs associated with caring for such individuals will rise substantially with broader implications for the overall economic burden.

Risk Factors for Osteoarthritis

The risk factors for OA can be classified as either systemic or local-mechanical.¹³ The local-mechanical risk factors are then further classified as intrinsic or extrinsic to the joint in question. In accordance with this model, OA is widely believed to be the result of local risk factors acting within the context of systemic

susceptibility.¹³ The intrinsic local-mechanical risk factors for OA include joint alignment, muscle weakness, and proprioception.^{5,13} Extrinsic local-mechanical risk factors include physical activity, occupations involving strenuous repetitive motions, obesity, and joint injury.^{5,13} A number of systemic risk factors have been reported, including age, sex, ethnicity, bone density, nutritional factors, and genetic factors.⁵

Biomarkers in Osteoarthritis: Targets for Personalized Medicine

At present, the best way to characterize OA involves measuring joint space narrowing on radiographs, evaluating clinical symptoms suggestive of OA (i.e. pain), and direct arthroscopic visualization of the articular surfaces within the joint capsule (particularly in the case of knee OA).^{10,14} However, these methods are often only able to detect OA after irreparable joint damage has occurred. Thus, there is a need to identify more sensitive methods to detect OA prior to irreversible joint damage, in order to improve morbidity outcomes. In particular, there is great interest in identifying specific biological markers that will reflect the biological changes that occur within the joint during the early phases of the OA disease process. Since OA primarily affects bone, cartilage, and synovial tissue, it is logical to consider the structural molecules derived from these tissues as potential biological markers of OA.¹⁴ Notable candidate biological markers include:

1. N-terminal cross-linked telopeptide of type I collagen (NTX-I) as a marker of bone degradation;
2. C-terminal cross-linked telopeptide of type II collagen (CTX-II) as a marker of cartilage degradation;
3. Glucosyl-galactosyl-pyridinoline (Glc-Gal-PYD) as a marker of synovium degradation.^{14,15}

It should also be noted that the biomarkers described above represent the net outcome of disease and do not indicate how the disease originated, nor do they represent targets for drug intervention in OA. However, these biomarkers

are useful to clinicians to stage the disease process in patients. Furthermore, biological markers may enable clinicians to differentiate patients based on risk of experiencing rapid progression of OA as these individuals will be in greatest need of targeted early intervention.¹⁶ Biomarkers are also expected to advance the process of drug development by providing cost-effective and sensitive indicators of a drug's effectiveness.¹⁵ Unfortunately, a single specific biological marker has yet to emerge to fulfill these lofty objectives, and it seems likely that it will be necessary to use a combination of markers in order to adequately characterize OA. Additionally, practical issues such as tissue specificity, clearance rates, potential circadian variations,¹⁴ and differences due to gender, ethnicity, and age need to be resolved prior to widespread acceptance of candidate biological markers.

It is also important to highlight that recent genome-wide linkage studies have identified several gene variants that appear to predispose to OA.^{14,17} In these studies, researchers relied upon microarrays capable of assessing 300,000 or more single-nucleotide polymorphisms (SNPs) in a given DNA sample. These microarrays examined interpersonal differences in inherited genetic variability by comparing the prevalence of gene variants among patients who have a given disease with controls who do not have the disease.¹⁸

Several chromosome regions and genes have been identified that are associated with OA prevalence (FRZB, BMP2, CD36, PTGS2, and NCOR2) or OA progression (CILP, TNFRSF11B, and ESR1; ADAM12 is associated with both prevalence and progression).^{14,17} Most of the genes identified encode proteins that are involved in signal-transduction pathways.¹⁴ This work should help to clarify the relationship between genetic susceptibility, the genomic expression of aberrant genes that predispose to OA via biological markers, and the actual manifestation and rate of onset of the disease process. More significantly, further clarification of such pathways in preclinical and clinical models of OA will lead not only to the identification of new clinically relevant biological markers, but help

achieve the ultimate goal of OA researchers in both academia and industry, which is to develop novel drug targets and therapies to combat this debilitating condition.^{14,15,17}

Predictive Genetic Testing

OA is a good example of a multi-factorial disease with a large societal burden for which predictive genetic testing could play a role in the future. In general, the goal of predictive genetic testing is to identify asymptomatic individuals at risk for disease based on particular biomarkers and/or genes that have been linked to predisposition to a particular disease. This differs from the majority of current medical tests, which are diagnostic, and thus seek to determine the etiology of a patient's *current* disease state. Thus, there is a fundamental uncertainty as to whether or not the disease state will develop and how severe the disease will manifest if it does in fact develop. This is especially true in complex multi-factorial diseases such as OA where it is difficult to make accurate predictions due to the influence of environmental factors.²

Pharmacogenomics

Genetic diagnosis and biomarker testing is important not only for current and future disease states, but also in predicting how an individual might respond to particular medications. The study of the genetic basis of differential drug response has been termed 'pharmacogenomics'. In general, many genes play a role in pharmacokinetics and pharmacodynamics of various drug responses. Pharmacogenomics research seeks to elucidate the genetic basis of how individuals or sub-populations respond to different drugs in terms of side-effects, toxicity, and drug efficacy. Such knowledge can lead to the development of biomarker tests to predict which individuals will respond well to particular drugs.^{19,20} It might also help in targeting drug development to specific sub-populations that may be more likely to respond to a particular therapy.²¹ In general, knowledge of the genetic factors that relate to drug response will provide a powerful tool for treatment in the future.^{19,20}

However, the development of detailed genetic tests that can be linked to a population-specific drug response is quite complex. A number of steps are involved, including:

1. Identification of genes that are involved in the drug response (e.g. pharmacokinetics and pharmacodynamics of the drug);
2. Determining differing variants of these genes;
3. Determining whether or not these different genetic variants correlate with a differential drug response.

One recent success of the field is the drug warfarin, whereby specific variants of particular genes were found to correlate with response to warfarin at particular doses.²² As of August 2007, the FDA has updated prescribing information for warfarin to include the information that the genetic make-up of a patient may influence how they respond to warfarin.²³ However, it is not clear that the benefits outweigh the costs. In a recent study on non-valvular atrial fibrillation, it was determined that there is little benefit to genetic testing for warfarin dosing, based on an estimated cost of \$400 (US) per genetic test, except in patients at the highest risk for hemorrhage.²⁴ This is likely to change as the costs of genetic testing decrease with the advancement of sequencing technologies²⁵, however it does call into question the cost-effectiveness of personalized medicine.²⁶

Personalized Medicine: Is it Realistic?

The goal of personalized medicine is to provide individualized treatment to patients, based partially on the knowledge of the genetic profile of an individual (including genes/biomarkers of disease state) and knowledge of how patients might respond to different medical treatments.^{27,28}

The complexities of moving towards a personalized medicine approach are immense. Cost-effective genome sequencing techniques²⁵, methods for analyzing genome-wide gene/protein/mRNA expression profiles and interaction related to disease states²³, and the

complete human haplotype mapping project (the HapMap project which provides a complete listing of human SNPs)^{27,30} will help in the acquisition of knowledge about the genetic basis for human disease. However, even these advanced techniques are not sufficient to deal with problems like incomplete penetrance, whereby presence of the disease allele(s) does not necessarily mean that that disease will occur.³¹ Furthermore, models for the genetic basis of disease should account for the effect of epigenetic control of gene expression.²⁴ Epigenetic control of gene expression (and problems with epigenetic control) have been shown to play a fundamental role in both Prader-Willi and Angelman diseases³³, and is thought to be implicated in the disease process for many diseases, including autoimmune diseases and psychiatric disorders³⁴.

Realizing the full benefits of personalized medicine and predictive genetic testing is still far in the future.²⁹ Nonetheless, it is important to be aware of these rapidly evolving fields as they hold the potential to revolutionize medicine.

References

1. Watson JD, Crick FH. The structure of DNA. Cold Spring Harbor Symposium on Quantitative Biology 1953;18:123-31.
2. Evans JP, Skrzynia C and Burke Wylie. The complexities of predictive genetic testing. British Medical Journal. 2001;322: 1052-1056.
3. Kerem B, Rommens JM, Buchanan JA, Markiewicz D, Cox TK, Chakravarti A, Buchwald M and Tsui LC. Identification of the cystic fibrosis gene: genetic analysis. Science. 1989;245: 1073-1080.
4. Gene Gateway – Exploring Genes and Genetic Disorders. A web companion to the human genome landmarks poster. http://www.oml.gov/sci/techresources/Human_Genome/posters/chromosome/cftr.shtml.
5. Felson DT, Lawrence RC, Dieppe PA, Hirsch R, Helmick CG, Jordan JM, *et al*. Osteoarthritis: New insights. part 1: The disease and its risk factors. Annals of Internal Medicine 2000;133(8):635-646.
6. Andriacchi TP, Mundermann A, Smith RL, Alexander EJ, Dyrby CO, Koo S. A framework for the in vivo pathomechanics of osteoarthritis at the knee. Annals of Biomedical Engineering 2004;32(3):447-457.
7. Cunningham LS, Kelsey JL. Epidemiology of musculoskeletal impairments and associated

- disability. *American Journal of Public Health* 1984;74(6):574-579.
8. Health Canada. Arthritis in Canada: An ongoing challenge No. Cat. #H39-4/14-2003E). Ottawa: Health Canada 2003.
 9. Brage ME, Draganich LF, Pottenger LA, Curran JJ. Knee laxity in symptomatic osteoarthritis. *Clinical Orthopaedics & Related Research* 1994;304: 184-189.
 10. Dieppe PA. Recommended methodology for assessing the progression of osteoarthritis of the hip and knee joints. *Osteoarthritis & Cartilage* 1995;3(2):73-77.
 11. Marks PH, Droll KP, Cameron M. Does ACL reconstruction prevent articular degeneration? The ACL risk equation. In R. J. Williams, & D. P. Johnson (Eds.), *Controversies in knee surgery* (pp. 15-36). Oxford: Oxford University Press 2004.
 12. Canadian Orthopaedics Association. Canada in motion: Mobilizing access to orthopaedic care. 2005. Retrieved June/06, 2007, from http://www.coa-aco.org/library/health_policy/canada_in_motion.html
 13. Sharma L. Local factors in osteoarthritis. *Current opinion in rheumatology* 2001;13(5):441-446.
 14. Rousseau J, Delmas PD. Biological markers in osteoarthritis. *Nature clinical practice rheumatology* June 2007;3(6):346-356.
 15. Kraus VB. Biomarkers in osteoarthritis. *Curr Opin Rheumatol* 2005;17:641-646.
 16. Lohmander LS, Felson D. Can we identify a 'high risk' patient profile to determine who will experience rapid progression of osteoarthritis? *OsteoArthritis and Cartilage* 2004;12:S49-S52.
 17. Slagboom E, Meulenbelt I. Genetics of osteoarthritis: early developmental clues to an old disease. *Nature Clinical Practice Rheumatology* November 2008;4(11):563.
 18. Hunter, DJ, Khoury MJ, Drazen JM. Letting the genome out of the bottle – Will we get our wish? *New England Journal of Medicine*. 2008;358(2):105-107.
 19. Evans WE and Johnson JA. Pharmacogenomics: The Inherited Basis for Interindividual Differences in Drug Response. *Annual Review of Genomics and Human Genetics*. 2001: 2: 9-39.
 20. Evans WE and Relling MR. Pharmacogenomics: Translating Functional Genomics into Rational Therapeutics. *Science*. 1999;286: 487 - 491
 21. An audience with: Jonathan K. C. Knowles discusses the impact of pharmacogenomics on market segmentation. *Nature Reviews Drug Discovery*. 2004: 3:822
 22. Sang-Seop L and Jae-Gook S. Current warfarin pharmacogenomics research for personalized medicine: status and perspectives. *Personalized Medicine*. 2009;6:13-14.
 23. US Food and Drug Administration. FDA Approves Updated Warfarin (Coumadin) Prescribing Information. <http://www.fda.gov/bbs/topics/news/2007/new01684.html>.
 24. Eckman MH., Rosand J, Greenberg SM, and Gage BF. Cost-Effectiveness of Using Pharmacogenetic Information in Warfarin Dosing for Patients With Nonvalvular Atrial Fibrillation. *Annals of Internal Medicine*. 2009;150:73-83.
 25. Wheeler DA, Maithreyan S, Egholm M, Shen Y, Che L, McGuire A *et al.* The complete genome of an individual by massively parallel DNA sequencing. *Nature*. 2008;452: 872-876.
 26. Ioannidis JPA. Personalized Genetic Prediction: Too Limited, Too Expensive, or Too Soon? *Annals of Internal Medicine*. 2009;150:139-141.
 27. The International HapMap Consortium. A haplotype map of the human genome. *Nature*. 2005;437:1299-1320.
 28. Weston AD and Hood L. Systems biology, proteomics and the future of health care: toward predictive, preventative and personalized medicine. *Journal of Proteome Research*. 2004;3;179-196.
 29. Gomez A. and Ingelman-Sundberg M. Pharmacoeugenetics: Its role in interindividual differences in drug response. *Clinical Pharmacology & Therapeutics*. Advance Online Publication February 25, 2009.
 30. The International HapMap Consortium. A second generation human haplotype map of over 3.1 million SNPs. *Nature*. 2007;449:851-861.
 31. Holtzman NA. and Marteau TM. Will genetics revolutionize medicine. *New England Journal of Medicine*. 2000;343:141-144.
 32. Lesko LJ. Personalized Medicine: Elusive Dream or Imminent Reality? *Clinical Pharmacology & Therapeutics*. 2007;81:807-816.
 33. Adams J. Imprinting and genetic disease: Angelman, Prader-Willi and Beckwith-Weidemann syndromes. *Nature Education* 2008;1(1).
 34. Rodenhiser D and Mann M. Epigenetics and human disease: translating basic biology into clinical applications. *Canadian Medical Association Journal*. 2006;174:341-347.

Ethical Implications of Germ Line Genetic Engineering

Hassan Mir (*Meds 2012*) and Christina Morgan (*Meds 2011*)
 Faculty Reviewer: Dr. Lois Champion

Following the recent decision by the Obama administration to lift strict limitations on embryonic stem cell research, genetic engineering has once again come to the forefront of scientific discussion. An upcoming FDA-approved trial using human embryonic stem cells in recently paralyzed individuals has further prompted an analysis of the legal and ethical issues surrounding the use of germ line genetic engineering. Though prohibited globally due to inadequate safety and effectiveness, it is inevitable that these concerns will one-day be met by continual technological advancements. Nevertheless, there remains a plethora of issues such as equal accessibility for all socioeconomic groups, autonomy of descendants, and effect on the human gene pool to name a few. In this discussion, it is important to make the distinction between germ line genetic engineering used for therapeutic purposes (altering DNA to correct a genetic defect before it manifests itself as a disease) and enhancement purposes (altering DNA to improve an individual above “normal” functioning). Although the use of this technology could be justified for therapeutic purposes, it is very difficult, if not impossible, to morally defend interventions for enhancement purposes.

Introduction

Genetic engineering has once again come to the forefront of scientific discussion following President Obama’s recent decision to lift strict limitations on human embryonic stem cell research set by the previous administration in America. Coinciding with this was the approval of a phase I stem cell trial by the Food & Drug Administration aimed primarily at testing the safety of this therapy in eight to ten completely paralyzed patients with severe spinal injuries.¹ The stem cells, which were obtained from embryos that would have otherwise been discarded, will also be used to test for any signs of functional recovery in these patients.¹

A lot of controversy has surrounded alterations made specifically in the germ line (sperm or egg), since they affect every cell in the child and would be transmitted to all future descendants.² It is important to make the distinction between germ line genetic engineering used for therapeutic purposes (altering DNA to correct a genetic defect before it manifests itself as a disease) and enhancement purposes (altering

DNA to improve an individual above “normal” average functioning).²

Currently, many laws around the world prohibit the use of germ line genetic engineering due to safety concerns and the possible implications for future generations. In addition, the four principles of bioethics,³ – beneficence, non-maleficence, autonomy, and justice – influence the debate regarding the future use of this technology. This article evaluates some relevant legal and ethical arguments that are important in determining whether germ line genetic engineering should be utilized in humans in the future.

Current Legislation

There are well defined Canadian and international laws governing the use of germ line genetic engineering. Canada’s Assisted Human Reproduction Act (2004) prohibits the alteration of “the genome of a cell of a human being or *in vitro* embryo such that the alteration is capable of being transmitted to descendants.”⁴ The prohibition is largely due to the questionable

nature of the safety, effectiveness, appropriateness and efficiency of the technology.⁵ This view is shared internationally,^{6, 7} and thus there are currently no legal cases pertaining to this topic around the world.

While the law dictates what can and cannot be done, the technology's controversial nature makes it necessary to assess the many ethical issues involved. Laws often reflect changing societal values, and therefore are constantly evolving. As a result, ethical analysis becomes invaluable for future policy development.

The Costs and Benefits of Germ Line Genetic Engineering

The principles of beneficence and non-maleficence constitute the backbone of bioethics.^{8,9} Beneficence asserts that there is an obligation to treat and improve the condition of others when it is possible to do so. Hence, altering the genome for therapeutic purposes on a permanent and heritable basis to prevent future genetic diseases becomes an obligation. This suggests that even within our limited capabilities, it is our duty to prevent any unnecessary harm and suffering and to improve the quality of life for future generations. The promise of enhancement to increase human functioning to above normal levels may seem very alluring and advantageous. Potentially favourable outcomes include a dramatic increase in life expectancy, a delay in the natural aging process, and increased tolerance and functioning of our immune system.^{10,11} It is also fair to argue in favour of enhancements that improve health and mental acuity in general. Such enhancements will increase survivorship, quality of life as well as the life expectancy for future generations.¹²

Nevertheless, the majority of techniques involved in this technology are in their early testing stages. This imposes numerous technical barriers that jeopardize the effectiveness and safety of such procedures, hence challenging the principle of non-maleficence; this principle states that a therapy should have a net benefit, loosely translating to "do no harm".⁹ Additionally, germ

line genetic engineering experiments involving animal models are highly inefficient and produce greatly variable offspring. The process requires breeding to be repeated through several generations until it results in a stable and permanent animal line with the desired properties. Not only would this be highly dangerous in humans, but because of the threat this presents to human dignity, it would also be impossible to justify morally.¹⁰ Insertion or modification of certain genes may also have unknown and potentially harmful interactions with other genes in the recipient genome. Similarly, the removal of disease-linked genes may remove the beneficial effects of those genes.¹⁰ Due to the unpredictable and largely unknown hazards of such procedures in humans, taking such risks is difficult to justify despite the potential benefits.

Respecting the Right to Self-Governance

Autonomy is the right to self-determination, which embodies freedom and the ability to determine one's own future.¹⁰ In germ line genetic engineering, one must consider autonomy of the parent, the child, and his or her progeny. According to the principle of reproductive autonomy, parents have the right to use whatever therapeutic means available to ensure that they have a normal pregnancy and a healthy baby. However, this principle excludes the parents' attempts to enhance the traits of their genetically normal offspring. Similar to situations where parental autonomy can be taken away in cases of social concern (such as child neglect), the American Association for the Advancement of Science has argued that strict legislation should regulate and differentiate between germ line genetic engineering for therapeutic versus enhancement applications.¹⁰

The autonomy of the child and subsequent progeny must also be considered. Upon reaching adulthood, a child is considered a rational agent and it is therefore safe to assume that he or she would consent to most genetic augmentations.¹³ While this argument may apply for therapeutic purposes, autonomy for enhancement ordeals is more complicated. Traits that a parent may choose to improve (such as improved

mathematical versus physical abilities) may not be what the child or their progeny would have chosen to enhance if given the choice as rational adults. In such instances, issues of informed consent and violation of autonomy arise, leading to the possibility of legal cases that challenge the reproductive autonomy of the parents versus the autonomy of the child.

The Role of Justice

Justice is another fundamental ethical concept and requires that harms and benefits be distributed equally among the whole population.³ If certain procedures become classified as therapeutic, it is reasonable for individuals to ask that such services be made universally available. It can be argued that germ line therapy is analogous to largely accessible public health initiatives aimed at preventing heart disease and therefore should also be accessible to the majority of the population. Nevertheless, even if therapeutic germ line genetic engineering becomes covered under a universal health plan, such as Medicare in Canada, long waiting lists will likely pose a myriad of problems.

On the other hand, the prospect of germ line genetic engineering for enhancement purposes becoming covered under public health insurance is highly unlikely.¹⁰ Thus, the wealthy would have greater access to this technology, further expanding the divide between different socioeconomic groups in society. This skewed access raises a few critical concerns. Those unable to afford this procedure would likely have some undesirable traits and could be viewed as abnormal.¹⁴ In contrast, those able to afford it would have greater control over shaping the human gene pool by removing undesirable traits while enhancing beneficial ones, leading to a form of eugenics within society. Additionally, alterations in the germ line would be carried on throughout the progeny of an individual, maintaining this inequality throughout future generations.

Conclusions

As we venture into the twenty-first century, technological advancements continue to expand the horizons of human potential, while also raising important ethical issues; germ line genetic engineering is one such advancement. Although these procedures are legally prohibited due to their lack of safety and effectiveness, it is important to develop an understanding of the implications on society to help make more informed decisions about their future uses. Though by no means exhaustive, this paper aimed to analyze and discuss the four major principles of bioethics - beneficence, non-maleficence, autonomy, and justice - as related to germ line genetic engineering. Through this analysis, it seems that though the use of this technology could be justified for therapeutic purposes, it is very difficult, if not impossible to defend morally for enhancement purposes.

Among other concerns, the availability of this therapy to the public would require extremely close monitoring of its health effects (harms and benefits in both the short and long term), as well as ensuring equal access for everyone in a timely manner regardless of their socioeconomic status. Failure to do so could result in deleterious situations from dire health effects for patients and their progeny to discrimination based on ones genetic make-up. Though there are endless possibilities in improving both the quality and quantity of human life, it is critical to ensure that genetic engineering is safe, effective, reasonable for future generations, and equally accessible before it can become a viable component of both the scientific and global community.

References

1. Falco, M. FDA approves human embryonic stem cell study [Internet]. 2009 [cited 2009 Mar 07]. Available from: <http://www.cnn.com/2009/HEALTH/01/23/stem.cell/index.html>.
2. Glannon, W. Genes, Embryos, and Future People. *Bioethics*. 1998; 12(3): 187-211.

3. Hebert, P.C. *Doing Right: A Practical Guide to Ethics for Medical Trainees and Physicians*. Brett Miller, eds. Don Mills, Ontario: Oxford University Press; 1995. 5-13 p.
4. Department of Justice, Canada. *Assisted Human Reproduction Act [Internet]*. 2004 [cited 2009 Mar 15]. Available from: <http://laws.justice.gc.ca/en/A-13.4/2389.html>.
5. Bernier, L. and Gregorie, D. *Reproduction and therapeutic cloning, germline therapy, and purchase of gametes and embryos: comments on Canadian legislation governing reproduction technologies*. *Journal of Medical Ethics*. 2004; 30(6): 527-532.
6. Institute on Biotechnology and the Human Future. *National Legislation Concerning Human Cloning and Germline Manipulation [Internet]*. No date [cited 2009 Mar 05]. Available from: www.thehumanfuture.org/documents/Intl_Legis_clon_germline_text.pdf.
7. The Council of Europe. *About the Council of Europe [Internet]*. 2006 [cited 2009 Mar 13]. Available from: http://www.coe.int/T/e/Com/about_coe/.
8. Rancich, A.M., Perez, M.L., Morales, C., and Gelpi, R.J. *Beneficence, Justice, and Lifelong Learning Expressed in Medical Oaths*. *Journal of Continuing Education in the Health Professions*. 2005; 25(3): 211-220.
9. McCormack, P. *Quality of Life and the Right to Die: an Ethical Dilemma*. *Journal of Advanced Nursing*. 1998; 28(1): 63-69.
10. Frankel, M.S., and Chapman, A.R. *Human Inheritable Genetic Modifications: Assessing Scientific, Ethical, Religious, and Policy Issues*. The American Association for Advancement of Science [Internet]. 2000 [cited 2009 Mar 05] Available from: <http://www.aaas.org/spp/sfirl/projects/germline/report.pdf>.
11. Loftis, J.R. *Germ-Line Enhancement of Humans and Nonhumans*. *Kennedy Institute of Ethics Journal*. 2005; 15(1): 57-76.
12. Mwase, I.M.T. *Genetic Enhancement and the Fate of the Worse Off*. *Kennedy Institute of Ethics Journal*. 2005; 15(1): 83-89.
13. Farrelly, C.P. *Justice in a Genetically Transformed Society*. *Kennedy Institute of Ethics Journal*. 2005; 15(1): 91-99.
14. Grey, W. L. *The Ethics of Human Genetic Engineering*. *Australian Biologist*. 1996; 9(1): 50-56.



IMMEDIATE NEEDS:

- Family Practice
- Internal Medicine
- Psychiatry
- GP/Anesthesia
- GP/Obs
- General Surgery
- Radiology

VISIT OUR SOUTH-WESTERN ONTARIO COMMUNITIES SERVED BY THE FOLLOWING HOSPITALS:

- Clinton Public Hospital
- Goderich Alexandra Marine & General Hospital
- Seaforth Community Hospital
- St. Marys Memorial Hospital
- Stratford General Hospital

WHY OUR MEDICAL COMMUNITY?

World-class theatre, fine dining, beautiful beaches, agricultural landscapes & welcoming individuals surround the healthcare community. Incentives include:

- Under-serviced designation benefits
- Six new Family Health Teams
- OMA Rural Locum Program – FFS, FHN, FHG
- Opportunities in group medical clinics – locum or full-time
- All emergency departments have 24/7 guaranteed hourly stipend
- Resident/medical students welcome for electives – housing available
- Proximity to University of Western Ontario & University of Waterloo Campuses

GWEN DEVEREAUX
 Recruitment Development Leader
 Phone: 519.527.8403
 Cell: 519.870.3667
 Fax: 519.527.8414

Email: gwen.devereaux@hpha.ca or gwen.devereaux@amgh.on.ca
 Web: www.healthforceontario.ca; www.hpha.ca



Delhi Community Health Centre
 105 Main Street
 Delhi, ON N4B 2L8
 Phone: (519) 582-2323

We offer:

- Your choice of practice model:
 - (1) Family Health Team (Blended Salary)
 - (2) Family Health Group (or)
 - (3) Family Health Organization
- Fully Electronic Medical Record
- Two sites (Port Rowan or Delhi)
- On-site laboratory and pharmacy
- Two full-service hospitals 15 minutes away

For more information contact:
Linda VanLondersele
 519-582-2323 ext. 239
info@dchc.ca

Delhi Community Health Centre

The Blameworthy Gene

Jennifer N. Bondy (*Meds 2012*) and Laura Hinz (*Meds 2011*)

Faculty Reviewer: Dr. Raj Harricharan

With improved techniques of genetic analysis, medicine is becoming increasingly reliant on genes to diagnose and treat disease. However, a genetic diagnosis is a value-laden entity with significant potential to change the way we categorize people as ill or healthy, flawed or normal, and responsible for or a hapless victim of disease. Nowhere are these value judgements more prevalent than the field of mental health. This article will examine the meaning and implications of genetic diagnoses, and apply the theories to the example of alcoholism.

“Genetic Diagnosis” Defined and Clarified

“Help us find the gene for *insert disease here.*” Slogans such as this have become increasingly common since the human genome was sequenced. These slogans seem to imply that once we have found the gene, the cure will naturally follow. They also imply that we cannot be truly certain of a diagnosis until it is proven by genetic technology. These attitudes have the potential to modify the way we conceptualize both the disease and the patient. Is a person with a genetic diagnosis but no symptoms a patient? Is a patient with symptoms but no gene defect simply a malingerer? The answers to these questions have the potential to change the way the person (or patient, depending on the interpretation) is categorized, palliated, and stigmatized.

The term ‘genetic diagnosis’ is oft used in the literature, but requires several clarifications. The first is the timing of the diagnosis. One may have a disease and *later* discover that it has a genetic component. This impacts the patient by a process that has been termed ‘geneticization’: the patients are relieved of blame for their conditions, they are simply victims of faulty genes.¹⁸ Such a diagnosis, however, does not prevent susceptible individuals from looking ahead; there are implications for family planning as the condition could potentially be passed on to offspring.^{13,14} In contrast, one may receive genetic testing *before* the onset of symptoms. When the genetic

precedes the clinical diagnosis, the individual may be prematurely thrust into the sick role.¹⁴ This broadened definition of the sick role carries with it societal repercussions, as it increases the number of patients requiring intervention.¹⁴ A genetic diagnosis can carry permanent stigma and may damage one’s feeling of personal control.^{13,14}

Secondly, it is important to keep in mind which disease is being examined with the ‘genetic diagnosis’. Certain diseases have more significant genetic contributions, thus affecting the potential for remedy.¹⁴ For instance, a genetic diagnosis of Huntington’s disease correlates well with clinical progression to the disorder whereas depression has approximately 40% heritability.¹⁴ Mental illnesses carry both a high social stigmatization and a low genetic determination, two conditions that Spriggs *et al* alleged require stringent justification for genetic testing.¹⁴ However, the low genetic determination means that identification of a gene carrier need not be a diagnosis, rather it may be an opportunity for prophylactic intervention.¹⁴ This has been termed the ‘genetic window’.¹⁷

The final clarification is the distinction between pathology and variation. There will naturally be variations in genes, but these differences need not be correlated with disease.¹² Therefore, a genetic diagnosis may inappropriately label an individual as “ill.” The antithesis of pathologizing the healthy is

legitimizing the plight of the ill. Mental health is often viewed as a disorder, something that is the responsibility of the affected person or the consequence of poor moral fibre.¹⁴ Once there is a genetic diagnosis, the disorder may qualify as a legitimate disease, one that is now the responsibility of the health care system.^{14,16} Thus, genetic diagnoses must be approached with caution: they have equal potential to inappropriately or appropriately cast individuals in the sick role.

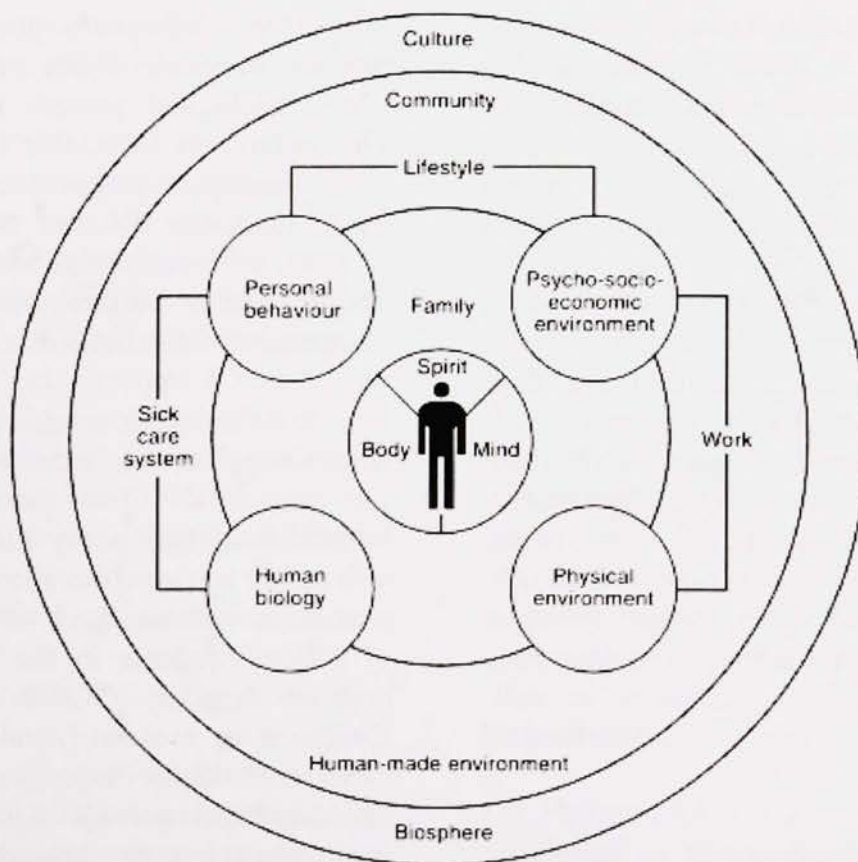
Nature Versus Nurture

The distinction between a clinical and genetic diagnosis hinges on the multi-factorial nature of disease. Specifically, the expression of genes can be influenced by the individual's environment. In a document classic in the realm of health promotion, Hancock summarized these multiple influences in the Mandala of Health (Figure 1). The individual is viewed as the centre

of a web of influence, with each spoke having the potential to create or treat disease.¹⁵ Therefore, while genes themselves cannot be altered by promoting healthy living, there is the potential to mitigate disease through environmental and lifestyle interventions.^{12,14}

Reactions to a Genetic Diagnosis

With the meaning of "genetic diagnosis" clarified, the *reaction* to such diagnoses can be examined. A genetic diagnosis can decrease blame and stigma associated with disease.¹² In an era of preventive health and personal responsibility, Minkler summarized these value judgements as "when ill is redefined as being guilty".¹⁶ In the past, sufferers of mental illness were the victims of societal stigma largely because mental pathology does not fit into the classic Western biophysical approach as there is often not one easily delineated causal mechanism. If there is a causal gene identified, however, the disorder becomes a biophysical disease and the



THE MANDALA OF HEALTH

Figure 1. Hancock's Mandala of Health. The individual is viewed as the centre of a series of spheres of influence, each providing an opportunity for health promotion interventions.¹⁵

person now has legitimate claim to the sick role. Notwithstanding this positive reaction to a genetic diagnosis, there can also be negative ramifications.¹³ While there was nothing you *could have done* to prevent the disease, there is nothing you *can do now*. Minkler warns that a genetic diagnosis may be the same as labelling someone as fatally flawed.¹⁴ However, this fatalistic approach disregards the multi-factorial nature of most mental diseases.¹³ The same argument applies to those who receive their genetic diagnosis before the onset of clinical symptoms - the optimism of environmental modification should temper the doom of one's biological lot.

Given this spectrum of possible reactions, the impact of a genetic diagnosis must be considered at three levels: the patient, the health care system, and society. An individual receiving a genetic diagnosis is now a candidate for early intervention and a legitimate actor in the sick role. Conversely, the genetic diagnosis may turn a healthy individual into a ticking time bomb, waiting anxiously for the onset of symptoms that now seems inevitable.¹⁴ This emphasizes the importance of not viewing a genetic diagnosis as a label of defectiveness nor as an immutable entity. These issues can now be considered in the context of a specific illness - alcoholism.

Genetic Diagnoses in Practice: Alcoholism

A number of studies demonstrate that children of alcoholics suffer from a variety of behavioural and psychopathological problems, such as substance abuse, anxiety, depression, conduct disorders and delinquency.¹⁰ This type of behaviour, however, is not consistent across all children of alcoholics. By developing a better understanding of these variations, clinicians can attempt to ensure that children of alcoholics will not succumb to potentially avoidable health problems.

The mode of inheritance of an alcoholic "gene" is far from established, yet there is evidence to suggest that alcoholism is indeed a disease of both genetic and environmental etiology.¹ Past research has demonstrated an

increased risk of alcohol abuse in the children of alcoholics.^{2,3,4} It is possible that this is due to "vertical cultural transmission"⁵. This theory describes the transmission of disorders, or traits which lead to increased susceptibility to a disease, through parental-offspring learned behaviour. However, studies have demonstrated that behavioural transmission is not the only contributing factor when the children of alcoholics develop drinking problems of their own.

For example, a twin-family study by Kendler *et al* examined the mode by which alcoholism was passed from parents to daughters.⁶ The results indicated that a solely environmental etiology was insufficient to explain alcoholism. Furthermore, the researchers found that in the best-fitting model, susceptibility to alcoholism was due in large part to a genetic predisposition.

In a cohort study by Goodwin *et al*, the prevalence of alcoholism was compared in two groups of adoptees: a group whose biological parents were alcoholics, and a control group whose biological parents were not alcoholics.⁷ The researchers found that 18% of those adoptees whose biological parents were alcoholics suffered from the same disease; nearly four times the prevalence found in the control group. These findings offer further evidence of a genetic component of alcoholism.

As there has not yet been a single culpable gene identified, one cannot be "diagnosed" with the potential for developing alcoholism. Notwithstanding, family history may be utilized as a proxy by which to identify targets for health promotion interventions. For example, in a study of college students in the U.S., the children of problem drinkers (COPDs) were identified and compared to a control group, and COPDs were 17% more likely to engage in heavy episodic drinking than non-COPDs.⁸ Additionally, they were approximately three times more likely than non-COPDs with similar drinking habits to seek help for their drinking problems. This was particularly true of students who had previously consumed alcoholic beverages but had abstained

from drinking in the past year. In light of these results, it stands to reason that when possible, COPDs, particularly those who have recently attempted to curtail their own drinking, should be targeted for counselling and treatment in order to prevent future alcohol dependency.

Walker and Lee, however, caution that clinicians should not pathologize the children of alcoholics (COAs).⁹ They indicate that COAs who come from “famil[ies] with strong emotional bonds and ... warm, supportive environment[s]”, are able to maintain caring and empathetic interpersonal relationships, and will not necessarily develop psychiatric disorders, as has been suggested to be typical of COAs. The authors elaborate, explaining that alcoholic families may have “reservoirs of strength”, which can come in a variety of forms, which clinicians should seek out and draw on in order to provide treatment. For example, sibling-sibling relationships within alcoholic families may be the only instance in which a family member is consistently emotionally available, and thus these relationships should be encouraged in an effort to promote healthy living and an avoidance of alcohol dependency.

Walker and Lee emphasize the plasticity of human development and the fact that it can drastically influence the qualities exhibited by COAs; specifically, the exhibition of resilient or maladaptive behaviours.⁹ The key to this process is to determine which relationships (i.e. marital, parent-child, sibling-sibling) within the family are resilient, and then encourage the fostering of these relationships, the affirmation of belief systems, and the improvement of communication. Resilient relationships within families may be the counterbalance to genetic predisposition.

Werner¹¹ adds that COAs who do not develop serious problems tend to have good communication skills, are goal-oriented, have a positive concept of self, and believe in self-help. While these determinants may possess a somewhat innate component, they are also qualities which can be addressed in counselling. By encouraging the development of the aforementioned attributes, it is possible that

susceptible individuals may alter their environments in such a way as to not fall victim to a genetic predisposition to alcoholism.

Impact of a Genetic Diagnosis - Conclusions

Genetic diagnosis can be costly not only in terms of the gene test itself, but in terms of treating the patients we have created. Before the advent of gene testing, these asymptomatic individuals did not consume health care resources. Ethical and political considerations arise of whether we have an obligation to treat those that we have identified as being ill or at risk, regardless of a lack of clinically recognizable disease. An additional societal consideration is the creation of the “other” - a group of genetically distinct individuals, almost a separate species. This us-versus-them attitude stems from the view of a genetic diagnosis as a fatal flaw and has the potential to perpetuate rather than reduce stigma.^{13,14}

On the other hand, early identification of at-risk individuals opens the possibility of health promotion and disease prevention interventions. Alcoholism is but one example of diseases which are all too frequently attributed to a lack of “will power” on the part of the ill. By acknowledging the scientifically demonstrated genetic component of this disease we can play a decisive role in the prevention of alcoholism in those who are most susceptible to the disease.

Being cognizant of the issues discussed in this article is an important step in improving our understanding of the individual and societal implications of genetic testing. Furthermore, recognition that phenotypic expression of genotype is environmentally mediated provides an opportunity for health promotion interventions.

References

1. Ferguson RA, Goldberg DM. Genetic markers of alcohol abuse. *Clinica Chimica Acta*. 1997;257:199-250.
2. Cotton NS. The familial incidence of alcoholism: a review. *J Stud Alcohol*. 1979;40:89-116.

3. Merikangas KR. The genetic epidemiology of alcoholism. *Psychol Med.* 1990;20:11-22.
4. Pollock VE, Schneider LS, Gabrielli WF, Goodwin DW. Sex of parent and offspring in the transmission of alcoholism: a meta-analysis. *J Nerv Ment Dis.* 1987;175:668-673.
5. Kendler KS. Indirect vertical cultural transmission: A model for nongenetic parental influences on the liability to psychiatric illness. *The American Journal of Psychiatry.* 1988;145(6):657-665.
6. Kendler KS, Neale MC, Heath AC, Kessler RC, Eaves LJ. A population-based twin study of alcoholism in women. *J Am Med Assoc.* 1994;151(5):707-715.
7. Goodwin DW, Schulsinger F, Hermansen L, Guze SB, Winokur G. Alcohol problems in adoptees raised apart from alcoholic biological parents. *Arch Gen Psychiatry.* 1973;28:238-243.
8. Weitzman E, Wechsler H. Alcohol use, abuse, and related problems among children of problem drinkers: Findings from a national survey of college alcohol use. *The Journal of Nervous and Mental Illness.* 2000;188(3):148-154.
9. Walker JP, Lee RE. Uncovering strengths of children of alcoholic parents. *Contemporary Family Therapy.* 1998;20(4):521-538.
10. Steinhausen HC. Children of alcoholic parents. A review. *European Child and Adolescent Psychiatry.* 1995;4(3):143-152.
11. Werner EE. Resilient offspring of alcoholics – A longitudinal study from birth to age 18. *Journal of Studies on Alcohol.* 1986;47:34-40.
12. Castiel LD, Guilam MCR, Vasconcellos-Silva PR, Sanz-Valero J. Genomic risk and personal responsibility in health. *Panamerican Journal of Public Health.* 2006; 19(3): 189-197.
13. Phelan JC. Genetic bases for mental illness- a cure for stigma? *Trends in Neurosciences.* 2002; 25(8): 430-431.
14. Spriggs M, Olsson CA, Hall W. How will information about the genetic risk of mental disorders impact on stigma? *Australian and New Zealand Journal of Psychiatry.* 2008; 42(3): 214-220.
15. Schatz P, Dzvimbo KP. The adolescent sexual world and AIDS prevention: a democratic approach to programme design in Zimbabwe. *Health Promotion International.* 2001; 16(2): 127-136.
16. Minkler M. Personal responsibility for health? A review of the arguments and the evidence at century's end. *Health Educ Behav.* 1999; 26: 121-141.
17. Hood KK, Johnson SB, Carmichael SK, Laffel LMB, She JX, Schatz DA. Depressive symptoms in mothers of infants identified as genetically at risk for Type I Diabetes. *Diabetes Care.* 2005; 28: 1898-1903.
18. Arribas-Ayllon M, Sarangi S, Clarke A. Managing self-responsibility through other-oriented blame: Family accounts of genetic testing. *Social Science and Medicine.* 2008; 66: 1521-1532.

Vampire Projects or Long Ago Person Found? A History of Genetic Research in First Nations Communities

Kate MacKeracher (Meds 2012) and Michael Livingston (Meds 2011)

Faculty Reviewer: Dr. Michelle Hamilton

Genetic research of Indigenous populations has been fruitful for scientists and the wider public, but has it benefited Indigenous communities themselves? We explore past (ab)uses of genetic research in Canadian First Nations communities through case studies. Although early projects were largely researcher-driven, the Human Genome Diversity Project of the 1990s catalysed a change towards participatory, community-controlled genetic research in Canadian Aboriginal communities.

Introduction

Indigenous* peoples have often been subjects for biomedical research, and the benefits to mainstream society include improved vaccines and better understanding of type 2 diabetes pathophysiology.¹ Genetic studies of Aboriginal populations have yielded dissertations and scholarly articles on subjects ranging from rheumatoid arthritis,² to breast cancer,³ to evolution and migration of human populations. But to what extent have Indigenous communities themselves benefited from this research? Through selected case studies, we explore the history of genetic research in Canadian First Nations communities, with a particular emphasis on the degree of community control.

Why Community Control?

In the late 1960s, Vine Deloria (Standing Rock Sioux) drew attention to research in Aboriginal communities that was driven more by researchers' interests than by "the needs of the people."⁴ Given that these communities are often struggling for survival against the political, economic, and social consequences of colonialism, Deloria condemns "pure research" of Indigenous peoples that benefits only the

researcher: "We should not be objects of observation for those who do nothing to help us."⁴ Indeed, power disparities between a community and outside researchers can make the research process into a form of exploitation,⁵ undermining Aboriginal knowledge and sovereignty: "The Indian explanation is always cast aside as a superstition," Deloria observes, "Indians must simply take whatever status they have been granted by scientists."⁶

Furthermore, research ethics in North America have traditionally focussed on individual rights, neglecting collective rights of groups.^{7,8,9} Yet many Indigenous communities consider information about the group to be communal property.¹⁰ Even something considered so individualistic, at least in mainstream Canadian society, as DNA may be seen as a common resource, as Debra Harry (Northern Paiute) describes: "We're talking about something that has existed collectively. It doesn't belong to the present generation."⁷ Outsider researchers may lack the knowledge to negotiate these political, cultural, and ethical complexities, and so the long-term distribution of benefits and harms from a project will be influenced by the degree of community control.^{9,11}

* We use "Indigenous" and "Aboriginal" interchangeably. In a Canadian context, "Aboriginal peoples" include First Nations, Metis, and Inuit populations.

“He Used Us like Guinea Pigs”: Two Cases of Genetic Research

In 1989, the Havasupai people of Arizona gave blood to researchers from Arizona State University, seeking to understand the high prevalence of diabetes in their community. In the early 2000s they realised the blood had also been used for unrelated studies by several researchers.¹¹ Of particular concern to some donors was the use of their DNA in population evolution studies that contradicted their oral history; Havasupai Chairwoman Carletta Tilousi explains, “They challenged our identity and our origins with our own blood without telling us what we were doing.”⁷ Anishinaabeg scholar Winona LaDuke argues that such studies have political, as well as cultural, significance. Governments or others with interest in Aboriginal peoples’ traditional lands, she suggests, may use the authority of “genetic evidence” to portray Indigenous groups as migratory “settlers” with no higher claim to land rights.⁷ The Havasupai Tribal Council and several community members have filed multi-million-dollar lawsuits against the researchers and their university.¹²

Members of Nuu-chah-nulth First Nations in British Columbia gave their blood to Ryk Ward, a geneticist at the University of British Columbia (UBC), in the mid-1980s, hoping to uncover the reason for their community’s high rates of inflammatory arthritis.¹³ They heard nothing more until 2000, when the Nuu-chah-nulth learned that Ward failed to find significant results in his arthritis study and had taken their blood with him to new appointments at Utah State University and Oxford University. He and colleagues received funding for further, unrelated analyses of the blood, including population evolution studies.¹³ Nuu-chah-nulth people, led by Larry Baird, one of the blood donors, have successfully lobbied for the return of the blood, and it is now stored at UBC.¹⁴ Baird explained his efforts to retrieve the blood from Ward in a Nuu-chah-nulth-run newspaper: “He profited at our expense. He published over 200 papers and became top guru in his field because he was carrying our blood around with him. He used us like cheap guinea pigs.”¹⁴

Note the diversity of communities’ responses. Baird objected to a researcher profiting from his community without reciprocity; Havasupai donors protest the use of their DNA in population studies that contradict their oral history; others, such as members of the Amazonian Yanomami people, have cultural beliefs against the preservation of blood;¹⁵ and different groups within communities may have conflicting responses to the secondary use of their samples.⁹

What these cases have in common are researchers who stored, shared, and re-used genetic samples without donors’ knowledge. Geneticists treated the samples much as biologists treat bacterial cultures: as scientific resources to be studied and shared. But individual donors did not consent to these uses; and since geneticists were naming a specific Indigenous community in their analyses, research was also being done on the community as a whole, without its consent. Perhaps for these researchers, operating relatively early in the genetic era, complex ethical reasoning had not yet caught up with scientific technique and curiosity. In an interview with *Nature*, Ward implies his secondary use of Nuu-chah-nulth samples was simply the “way people operated at the time... it didn’t cross anyone’s mind -- we didn’t mean to be evil, and we are more careful now.”¹³

“The Vampire Project”

The increasing vigilance of Indigenous communities, and corresponding tendency of geneticists to be “more careful now,” developed over the 1990s in the context of the Human Genome Diversity Project (HGDP) controversy. Conceived as a supplement to the Human Genome Project, the not-for-profit HGDP aimed to collect samples from genetically isolated Indigenous communities around the world, create immortalized cell lines, and make the DNA available to not-for-profit scientists. The initial goal was to facilitate study of population evolution and migration,¹⁶ although later representations of the project emphasised potential health research.¹⁷

An Indigenous resistance movement to the project developed very quickly. By 1993, the World Council of Indigenous Peoples had christened it “The Vampire Project” for the emphasis on retrieving blood, and the term stuck in North American Aboriginal newspapers.¹⁸⁻²⁰ HGDP organizers attempted to mitigate criticism by forming a North American Regional Committee with two Aboriginal members, and by drafting a Model Ethical Protocol for collecting samples.²¹ Nevertheless, Canadian and American government agencies ultimately declined to fund the project, and it has stalled in North America.²²

Why did a project considered innocuous by its originators²² generate such resistance? Representatives of Indigenous organizations from the Americas outlined common problems with the HGDP in 1995, including spiritual objections to immortalized cell lines, potential military or commercial abuses, and objectification of Indigenous peoples “to satisfy scientific curiosity” without benefit to the peoples themselves.²³ Was true informed consent from individuals and communities possible, given linguistic and cultural barriers?²⁴ Would genetic research support population migration theories that dismissed community origin stories and undermined struggles for land and sovereignty rights?²⁵

One of the most frequent objections stemmed from the language of the HGDP’s draft project proposal; genetically distinctive Indigenous populations are referred to as “isolates of historical interest” whose genetic resources need to be collected immediately, before the populations die out or disappear.^{18,25-27} LaDuke explains many Aboriginal individuals’ outrage at the idea of salvaging genes instead of saving people: “Why would so many resources be involved in collecting the genetic materials from ‘vanishing populations’ rather than working to preserve those peoples and their cultures?”⁷

As evidenced by the presence of lawyer Catherine Twinn (Sawridge First Nation) and anthropologist Russell Thornton (Cherokee) on the HGDP’s North American Regional

Committee, Indigenous opposition to the project has not been universal. Defences from Aboriginal persons in scholarly literature or the popular press, however, are difficult to find. The most prolific Indigenous writer on the HGDP in academia has been Frank Dukepoo, a Hopi/Lacuna geneticist. Citing the moral naiveté of the project’s originators, the real risks and uncertain benefits to Indigenous peoples, and the lack of community control inherent in open-ended gifts of genetic resources, he has generally argued against the project.²⁸ Yet he asserts that not all Indigenous individuals and communities reject genetic research *per se*; the HGDP’s mistake, he suggests, is its “paternalistic” approach to conducting “research *on* rather than *with* indigenous people.”²⁹

Participatory Research

Early in the 1990s, the people of Oji-Cree Sandy Lake First Nation (Ontario/Manitoba border) decided to address their community’s high prevalence of type 2 diabetes. They formed a partnership with two physician-researchers that “incorporates the principles of participatory research,” according to the project website.³⁰ Community leaders and researchers established goals – determining community-specific prevalence and risk factors, implementing primary and secondary prevention programmes – and continue to discuss all aspects of the project, from protocol designs to dissemination of results.³⁰ Protocols must be approved by the Band Council, and research is conducted by trained community members.³⁰ Prevalence of type 2 diabetes was found to be five times the Canadian average, and a range of environmental and lifestyle risk factors were isolated; based on these findings, the project partners began to develop community-run, culturally-specific intervention programmes in 1995.³¹

The following year, the project organizers invited researchers from Robarts Research Institute at UWO to seek possible genetic components to diabetes in Sandy Lake First Nation.³² Analysis of DNA from 728 community members showed that a mutation unique to this community in gene *HNFI1A* increased the risk of

developing type 2 diabetes by up to 15 times in homozygous individuals, compared with community members homozygous for the normal allele.³² The clinical applicability of testing for this mutation was examined as a means for directing enhanced prevention support to genetically susceptible individuals.³³ The relative importance of genetic and environmental risk factors for early onset type 2 diabetes in community members was quantitatively assessed, with the conclusion that “changes in environment, at the level of lifestyle, could overturn genetic susceptibility, probably rapidly, and in a ‘low-tech’ manner.”³²

These genetic studies have not occurred in reductionist isolation, as all too often happens in basic science research, but rather are embedded in a community-controlled research project that includes problems, causes, and community-specific solutions in its scope of inquiry, and that requires results to be relevant and meaningful for all parties. Canadian Mohawk scholar Marlene Brant Castellano observes that this sort of collaboration makes for more effective research, and cites the Sandy Lake First Nation diabetes project as evidence that “Holistic awareness and highly focussed analysis are complementary, not contradictory.”¹⁰

Long Ago Person Found

In 1999, the body of a young man who died hundreds of years ago was found preserved in a glacier on traditional Champagne Aishihik First Nations (CAFN) land (BC/Yukon border).³⁴ The First Nations assumed responsibility for the remains, in keeping with a 1995 treaty affirming their control over cultural resources on their land.³⁴ CAFN Elders named the remains Kwäday Dän Ts’inchj (Long Ago Person Found),³⁵ and together with the Band Council and other community members, decided to investigate his origins.³⁴

After consulting with neighbouring First Nations, the CAFN Band Council signed a management agreement with the BC government to allow for research on the remains.³⁴ Researchers would not be able to own genetic or

other materials gathered at the discovery site and would return all samples after analysis.³⁴ The research included a comparison of mitochondrial DNA (mtDNA) in the remains to nearly 250 Aboriginal volunteers from the area.³⁵ This genetic test suggested Long Ago Person Found has living relatives in CAFN and neighbouring Aboriginal communities.³⁵ Since the relatives identified by maternally-inherited mtDNA are all members of the Wolf/Eagle clan, Elders concluded their ancestor likely was as well, given the matrilineal clan structure of their nations.³⁵ This identification allowed for a memorial potlatch to be planned by Elders of the correct clan.³⁵

Symposia at the Royal BC Museum and the CAFN reserve in 2008 and 2009 made information gathered about the remains accessible to local Indigenous people and the general public. This included scientific and cultural talks, traditional accounts of lost travellers and trade routes to complement forensic analyses of clothing material and stomach contents, and local genealogies to fill in the gaps of high specificity/low sensitivity mtDNA testing.³⁵ Requested by the community and contextualized with oral history and other studies, this DNA analysis demonstrates that community-controlled genetic research can sometimes support Aboriginal peoples’ psychosocial, as well as biomedical, well-being.

In this case, the community’s desire to know more about their ancestor coincided with outside researchers’ curiosity, and DNA testing was deemed acceptable by Band Council, Elders, and blood donors. Not all Aboriginal peoples will make similar decisions about remains found in their land, however, and interested geneticists must not suppose that DNA analysis will always be welcomed or acceptable in these situations.³⁶

Conclusions

In response to the increasing power and voice Indigenous peoples have negotiated for themselves about research in their communities, Canadian government funding agencies have begun requiring special ethical considerations, in

addition to usual practices, from those seeking grants for research with Aboriginal peoples.^{8,9} For example, the 2007 CIHR Guidelines for Health Research Involving Aboriginal People emphasises community/group consent, participatory research, ownership of biological specimens and data by the community, and new informed consent before any secondary use of samples and data, among other requirements.⁹

Although these regulations apply only to CIHR-funded researchers, Canadian First Nations communities are beginning to establish their own codes of conduct and research ethics boards.¹⁰ These efforts flow from Canadian Aboriginal peoples' constitutional right to self-government, according to a report by the National Aboriginal Health Organization (NAHO), and reflect the principles of "OCAP": community ownership, control, access, and possession of research and biological samples.³⁷

When they learned how Ward had used their samples, the Nuu-chah-nulth Tribal Council formed a Research Ethics Committee chaired by Larry Baird, the community member most active in repatriating the blood. "We're not closing the door on research," Baird explains, noting that some Nuu-chah-nulth people are interested in renewing inquiry into the arthritis that still plagues their communities.¹⁴ But the "way people operated" when Ward took their blood in the 1980s will not be tolerated, Baird warns: "From now on our eyes are wide open."¹⁴

References

1. Rhoades ER, Rhoades DA, Freeman W. Research Ethics and the American Indian. In: American Indian Health. Ed Rhoades ER. Baltimore: John Hopkins University Press; 2000. 426-33.
2. Oen K, El-Gabalawy HS, Canvin JM, Hitchon C, Chalmers IM, Schroeder M, Jacobson K, Reed M, Wood S, and Cheang M. HLA associations of seropositive rheumatoid arthritis in a Cree and Ojibwe population. *Journal of Rheumatology*. 1998 25(12): 2319-23.
3. Liede A, Jack E, Hegele RA, Narod SA. A BRCA1 mutation in Native North American families. *Human Mutation*. 2002 19(4):460.
4. Deloria V. *Custer died for your sins*. London: Macmillan Company; 1969.
5. Schnarch B. Ownership, control, access, and possession (OCAP) or self-determination applied to research. *Journal of Aboriginal Health*. 2004 Jan;1(1):80-95.
6. Deloria V. *Red earth, white lies: Native Americans and the myth of scientific fact*. Toronto: Scribner; 1995.
7. LaDuke, W. *Recovering the sacred: The power of naming and claiming*. Cambridge, MA: South End Press; 2005.
8. Canadian Institutes of Health Research, National Sciences and Engineering Research Council of Canada, Social Sciences and Humanities Research Council of Canada. Tri-council policy statement: Ethical conduct for research involving humans: Research involving Aboriginal Peoples [Internet]. 2005 [cited 7 March 2009]. Available from: <http://www.pre.ethics.gc.ca/english/policystatement>.
9. CIHR (Canadian Institutes of Health Research). Guidelines for Health Research Involving Aboriginal People [Internet]. Ottawa 2007 [cited 7 March 2009]. Available from: http://www.cihr-irsc.gc.ca/e/documents/ethics_aboriginal_guidelines_e.pdf.
10. Castellano, MB. Ethics of Aboriginal research. *Journal of Aboriginal Health*. 2004 Jan; 1(1):98-114.
11. Noe TD, Manson SM, Croy CD, McGough H, Henderson JA, Buchwald DS. In their own voices: American Indian decisions to participate in health research. In: *The handbook of ethical research with ethnocultural populations and communities*. Eds Trimble JE and Fisher CB. Thousand Oaks, CA: Sage Publications; 2006. 77-92.
12. Shaffer M. Havasupai blood samples misused. *Indian Country Today*. 2004 May 10 23(39):B1.
13. Dalton R. Tribe blasts 'exploitation of blood samples'. *Nature*. 2002 Nov; 420:111.
14. Wiwchar D. Nuu-chah-nulth blood returns to west coast. *Ha-Shilth-Sa. Ahousaht*; 2004 Dec 16 31(25): 1,3.
15. Martin LL. Yanomani blood samples: A question of ethics. *Native Americas*. 2001 Dec 13 8(3/4):6.
16. Marks J. The trouble with the Human Genome Diversity Project. *Molecular Medicine Today*. 1998 4(6):243.
17. Cavalli-Sforza LL. *The Human Genome Diversity Project: Past, present, and future*.

- Nature Reviews: Genetics. 2005 6:335-40.
18. 'Vampire Project' draws fire from Indigenous groups. Cherokee Observer. Parkhill, Oklahoma; 1995 April 20 3(4): 17.
 19. Smoke-Asayenes D. The commodification of life: The Human Genome Diversity Project. News from Indian Country. Hayward, Wisconsin; 2001 Aug 15 15(15):4A.
 20. Wendell J. Tracking the Vampire Brigade. Akwesane Notes. Rooseveltown; 1996 March 31 2(1): 73.
 21. Morrison Institute for Population and Resource Studies. Human Genome Diversity Project [Internet]. Stanford University; 1999 [cited 5 March 2009]. Available from: <http://www.stanford.edu/group/morrinst/hgdp.html>.
 22. Lindee MS, Goodman AH, Heath D. Introduction. In: Genetic Nature/Culture. Eds. AH Goodman AH, Heath D, Lindee MS. Berkeley, CA: University of California Press; 2003. 1-19.
 23. Indian leaders from North, Central, and South America meet to declare opposition against genetic manipulation and the Human Genome Diversity Project. News from Indian Country. Hayward, Wisconsin; 1995 April 30 9(8):6.
 24. Oros T, Gray S. Blood money. The Circle: News from an American Indian Perspective. Minneapolis; 1994 Nov 30 15(11):7.
 25. Harry D, Dukepoo FC. Indians, genes and genetics: What Indians should know about the new biotechnology [Internet]. Nixon, Nevada: Indigenous Peoples Coalition Against Biopiracy; 1998 [cited 9 March 2009]. Available from: http://www.ipcb.org/pdf_files/primer.pdf.
 26. Indigenous Peoples Council on Biocolonialism. Declaration of Indigenous Peoples of the western hemisphere regarding the Human Genome Diversity Project [Internet]. Phoenix, Arizona; 1995 Feb 19 [cited 9 March 2009]. Available from: http://www.ipcb.org/resolutions/htmls/dec_phx.html.
 27. Lone Dog L. Whose genes are they? The Human Genome Diversity Project. Journal of Health & Social Policy. 1999; 10(4): 51-66.
 28. Dukepoo FC. The trouble with the Human Genome Diversity Project. Molecular Medicine Today. 1998; 4(6):242-3.
 29. Dukepoo FC. It's more than the Human Genome Diversity Project. Politics and the Life Sciences. 1999; 18(2):293-7.
 30. Sandy Lake Health and Diabetes Project [Internet]. 2006 [cited 5 March 2009]. Available from: <http://www.sandylakediabetes.com/?q=node/1>.
 31. Black J. Scientists find diabetes link in Oji-Cree. Windspeaker. Edmonton, Alberta; 1999 Apr; 16(12):1.
 32. Hegele RA, Zinman B, Henley AJG, Harris SB, Barrett PH, and Cao H. Genes, environment, and Oji-Cree type 2 diabetes. Clinical Biochemistry. 2003; 36: 163-70.
 33. Hegele RA, Cao H, Hanley AJG, Zinman B, Harris SB, Anderson CM. Clinical utility of *HNFI1A* genotyping for diabetes in Aboriginal Canadians. Diabetes Care. 2000; 23(6): 775-8.
 34. Hunter T. Discovery: They call him Long Ago Person Found. Raven's Eye. Edmonton; 31 Dec 2000; 4(8): 2.
 35. McKinnon A. CAFN Newsletter [Internet]. 2008 June-July [cited 7 March 2009] Available from: http://www.cafn.ca/pdfs/June-July_2008_Newsletter_144_dpi.pdf.
 36. Weaver J. Indian presence with no Indians present: NAGPRA and its discontents. Wicazo Sa Review. 1997; 12(2): 13-30.
 37. First Nations Centre, AHO (National Aboriginal Health Organization). First Nations Conceptual Frameworks and Applied Models on Ethics, Privacy, and Consent in Health Research and Information: Summary Report [Internet]. 2006 November [cited 23 March 2009] Available from: http://www.naho.ca/english/pub_research.php.

Multidisciplinary Management at Key Stages in the Huntington's Disease Neurodegenerative Process

Abhijat Kitchlu (Meds 2011) and Allanah Li (Meds 2012)

Faculty Reviewers: Dr. Shannon Venance and Dr. Christopher Hyson

Introduction

Huntington's Disease (HD) is an adult-onset neurodegenerative disorder with significant motor, cognitive, and psychiatric manifestations. Symptoms are progressive, with no effective treatments currently available and death occurring 15-20 years after onset.¹ HD is inherited in an autosomal dominant manner. Affected individuals display an expanded CAG trinucleotide repeat in the HD gene on chromosome 4.¹ Given the devastating natural history of the disease and its hereditary basis, HD can present many challenges for patients, their families, and healthcare providers. In this article, we describe some of the complex issues that arise during key stages of the HD neurodegenerative process and emphasize the importance of comprehensive, interdisciplinary team management at each stage.

Predictive Testing for HD

Predictive testing for HD based on trinucleotide repeats has been available since 1993, with a less definitive linkage analysis test available in the mid-1980s. While some argue that the test can relieve uncertainty and enable planning for the future, only 4-24% of those at risk for HD go through with testing.² Decisions regarding testing have many ethical, legal, and psychosocial implications and at-risk patients must find a way to navigate through these concerns. Patients must consider the psychosocial effects of declining or postponing testing, as well as the effects of undergoing testing and receiving either a positive or negative result. Moreover, the results of testing can have a significant effect on the patient's family, who may become future

caregivers or future patients due to their at-risk status.³ Patients also face the possibility of discrimination based on genetic information, particularly regarding eligibility for disability or life insurance and obtaining employment. While there are only anecdotal reports of genetic discrimination in Canada, the fear of discrimination remains a very powerful influence on patients and physicians.^{4,5}

The healthcare team at this stage takes a primarily counselling and supportive role. Genetic counselling, often done by genetic counsellors or medical geneticists, is particularly important during this time. Counselling provides patients with disease information, takes them through the implications of testing, and offers psychosocial support.⁶ Counselling must provide the patient with all necessary and relevant information, because the far-reaching consequences of testing necessitate truly informed consent for the procedure. Genetic counsellors must present information regarding HD and predictive testing in a balanced and non-directive manner, ultimately respecting the patient's autonomy.⁵ Patients must be assured of privacy and confidentiality. In order to support the patient through their decision of whether or not to undergo testing, other services that may become involved at this stage include social work, family support and counselling, psychology, psychiatry, neurology, and family medicine. Management should be specific to the individual patient, while taking the family and cultural contexts into account.

After Undergoing Testing for HD – Living with the Results

The results of predictive testing for HD can have a profound impact on patients and their families. A Canadian study examining the psychological consequences of predictive testing found that although psychological well-being was significantly improved for the majority of participants, 6.9% experienced clinically significant adverse psychological events, including diagnosed clinical depression and suicide attempts.⁷ These adverse events occurred in individuals given positive and negative test results. The paradoxical reaction of some patients with a negative test result (noncarriers of the HD gene) has been called “survivor’s guilt” and involves psychological distress from being spared from HD.¹ Patients receiving a positive test result (carriers of the HD gene) must come to terms with the meaning for their own health, as well as implications for their family, career, and life plans. Although fear, anxiety, and depression may be understandable responses to a positive test result, these are also possible psychiatric manifestations of HD and patients must be followed for early onset of symptoms.³ A positive result can also have a tremendous impact on family dynamics, with studies showing higher divorce rates among carriers than noncarriers in the 6 months following test results, and parents possibly experiencing guilt for having transferred risk status to their offspring.² Thus for HD and other genetic diseases, the unit of care is often the family rather than the patient.³

Following predictive testing for HD, patients are faced with new challenges that may require multidisciplinary involvement. It is clear that psychological counselling and follow-up must be available to all patients undergoing predictive testing regardless of test result, and psychiatric care may also be required at this stage. Support and education should be offered to the patient’s family and may require the help of social workers, nurses, psychologists, and therapists. Early collaboration between the family doctor and a specialist in HD can monitor for onset of disease symptoms. Physicians must also formulate a plan with the patient for future

management.³ In addition to the formal healthcare team, patients and their families may benefit from joining a support network like the Huntington Society of Canada.

Reproductive Decisions

HD presents complex dilemmas for patients and their families with respect to reproductive decision-making. Although HD follows autosomal dominant inheritance, the penetrance of the disease varies with the extent of the CAG triplet expansion. Triplet repeats of 40 or more are fully penetrant, those 36 – 39 are thought to be variably penetrant and those less than 35 repeats do not typically manifest as disease. The expansion (and, in some cases, contraction) of the repeats during gametogenesis further complicates reproductive choices and can present a challenge for health care professionals when communicating risks.⁸

Qualitative research has identified several difficult decisions facing patients and their healthcare teams: whether to have children at all, whether to undergo prenatal testing or pre-implantation genetic diagnosis, and whether to abort gene-positive fetuses. Many patients who wish to have children express opposition to giving birth without attempting to prevent passing on the disease; however, this concern is often superseded by patients’ fears of their illness preventing them from providing for and raising children.⁸ For this reason, patients must be counselled with consideration and potential involvement of their partners, families, and wider social support networks. This in turn can be a concern for patients, as many report the influence of others as a major factor in decision-making.⁹ This includes the influence (whether or not intentional) of the healthcare team, which is an important consideration in light of a recent study which indicated that 38% of Mexican neurologists, psychologists and psychiatrists felt that those with the HD mutation should not have offspring.¹⁰

Prenatal testing for HD currently exists via amniocentesis or chorionic villus sampling. However, testing rates for those at risk for HD remain low; within the United Kingdom and

Australia only 5 – 25% of at risk populations undergo prenatal testing.⁸ Within Canada, a survey examining 15 of the 22 centres offering testing found that only 15 prenatal tests were completed between 1987 and 2000. The 12 families who underwent these tests were estimated to represent approximately 0.1% of the at-risk population.¹¹ Women were shown to be more likely to request prenatal testing as well as predictive testing for themselves.¹¹ More recently, pre-implantation genetic diagnosis has become available to ensure only mutation-negative embryos are implanted. This process, however, can be difficult for many couples and may not be an option for many due to personal beliefs and preferences.¹² Pre-implantation genetic diagnosis is now also offered without informing the parents of their own HD gene status, which is preferable to some patients who desire certainty that they are not passing on the disease to their children, but do not want to determine their own gene status.

Despite these difficulties, many HD patients and carriers do opt to have children. A European study demonstrated that 14% of HD carriers had subsequent pregnancies following predictive testing, compared to 28% of non-carriers.¹² Furthermore, many patients consider adoption as a potential alternative depending on their expectations of disease onset.⁸

Interdisciplinary Care of the Symptomatic HD Patient

The management of symptomatic HD patients is a broad topic, and in its entirety is beyond the scope of this overview; instead, a brief outline of the interdisciplinary nature of HD care is presented here.

The nature of HD requires a team-based approach to symptom management. An optimal healthcare team includes leadership not only from a neurologist or psychiatrist specializing in HD, but also a family physician to monitor complications in the late stages of the disease.³ Allied health care professionals, including nurses, dentists, dieticians, physical, occupational and speech therapists, psychologists, and social workers also play major roles. Advance care

planning is also extremely important in establishing quality of life goals and end-of-life care directives.

The stages of symptomatic HD dictate the need for each of these team-members. The Shoulson-Fahn scale, originally designed for research, has become useful in evaluating the severity of HD. The five stage scale is based on a Total Functional Capacity (TFC) score from 0 to 13 that assesses the patient's functional skills and ability to carry out activities of daily living. In stages 1 through 3, the focus of care includes treatment of chorea, sleep disturbance, depression, anxiety, impulsivity, irritability and other psychiatric symptoms.^{3,13} In these stages the physician members of the team are foremost. However, the role of psychologists and social workers may be crucial in the transition from working life to disability status for some patients and their families. It has been suggested that one of the most prevalent deficiencies in HD treatment is disproportionate focus on minor findings of chorea, without proper management of depression and deterioration of family relationships.¹³ Medical therapy including antidepressants and mood stabilizers may be of value and neuroleptics may be warranted in cases of more aggressive behaviour.¹³

Mid- to late-phase HD requires follow-up visits to physicians to assess for complications, but also increased involvement of other healthcare professionals. Occupational therapists may be involved in assessing driving ability and determining necessary restrictions. Patients often have large appetites, and may have difficulty meeting nutritional requirements given increasing dysphagia and lack of coordination (aspiration and subsequent pneumonia are often the terminal events for HD patients). For this reason, dieticians' expertise and nursing or personal support worker assistance with feeding can be of great benefit. Physician follow-up may involve the reduction of medication for chorea, which paradoxically decreases in the late stages and is supplanted by rigidity and dystonia. These symptoms are often worsened by excess pharmacotherapy for chorea, which must become more judicious.¹³ Physicians must also help

patients and families deal with increasing cognitive impairment and mood alterations, and as such, behavioural counselling is often required.³ In the end stage of the disease, hospice nursing and palliative care specialists ensure patients are as comfortable as possible and assist families with the patients' death.

Conclusion

Huntington's disease presents significant challenges for the healthcare team. Despite the paucity of treatments available to delay disease progression, a well-coordinated, multidisciplinary team can assist patients and their families with managing the evolving nature of their illness and help them make informed decisions. Through appropriate collaboration, care can be optimized and the burden of this multi-faceted disease can be reduced.

References

1. Wahlin T-BR. To know or not to know: A review of behaviour and suicidal ideation in preclinical Huntington's disease. *Patient Education and Counselling*. 2007;65:279-87.
2. Tibben A. Predictive testing for Huntington's disease. *Brain Research Bulletin* 2007;72:165-71.
3. Nance MA. Comprehensive care in Huntington's disease: A physician's perspective. *Brain Research Bulletin*. 2007;72:175-8.
4. Harper PS, Gevers S, de Wert G, Creighton S, Bombard Y, Hayden MR. Genetic testing and Huntington's disease: issues of employment. *THE LANCET Neurology*. 2004;3:249-52.
5. Ensenauer RE, Michels VV, Reinke SS. Genetic Testing: Practical, Ethical, and Counseling Considerations. *Mayo Clinic Proceedings*. 2005;80(1):63-73.
6. Mularczyk M, Decruyenaere M, Denayer L, Evers-Kiebooms G. A Theoretical Psychological Perspective on Predictive Testing for Late Onset Hereditary Diseases. *Genetic Counseling*. 2007;18(4):367-78.
7. Almqvist E, Brinkman R, Wiggins S, Hayden M. Psychological consequences and predictors of adverse events in the first 5 years after predictive testing for Huntington's disease. *Clinical Genetics*. 2003;64:300-9.
8. Klitzman R, Thorne D, Williamson J, Chung W, Marder K. Decision-making about reproductive choices among individuals at-risk for Huntington's disease. *Journal of Genetic Counselling*. 2007;16(3):347-62.
9. Klitzman R, Thorne D, Williamson J, Chung W, Marder K. Disclosures of Huntington disease risk within families: patterns of decision-making and implications. *American Journal of Medical Genetics Part A*. 2007;143A(16):1835-49.
10. Vilatela M, Morales A, de la Cadena C, Lopez I, Aranda C, Villa A. Predictive and prenatal diagnosis of Huntington's disease: Attitudes of Mexican neurologists, psychiatrists, and psychologists. *Archives of Medical Research*. 1999;30(4):320-4.
11. Creighton S, Almqvist E, MacGregor D, *et al*. Predictive, prenatal and diagnostic genetic testing for Huntington's disease: The experience in Canada from 1987 to 2000. *Clinical Genetics*. 2003;63:462-75.
12. Evers-Kiebooms G, Nys K, Harper P, *et al*. Predictive DNA-testing for Huntington's disease and reproductive decision making: a European collaborative study. *European Journal of Human Genetics*. 2002;10(3):167-76.
13. Walker F. Huntington's Disease. *Seminars in Neurology*. 2007;27(2):143-50.

Who's in Your Genes: A Physician's "Duty to Warn" Patients' Relatives about Genetic Risk

Colin Meyer-MacAulay (*Meds 2012*)

Faculty Reviewer: Dr. Victoria Siu

The genetic information of a patient as it relates to disease risk may prove invaluable to blood relatives who may wish to use it to make informed decisions about health care or reproduction. On the other hand, diagnosis of a genetic disorder may carry with it a certain social stigma, as well as concerns about discrimination with regards to employment and life insurance. Furthermore, genetic risk cannot be altered nor is it caused by the actions of the patient. A physician's "duty to warn" individuals who may be put at risk by the actions of a patient has been recognized in the context of both infectious disease and psychiatric illness. However, it is unclear whether these same precedents may apply in the context of genetic information. Nonetheless, a number of lawsuits in the United States have been brought against physicians for their alleged failure to warn relatives of genetic risk. Canadian Policy decisions on this issue must be made based on an understanding the benefits and limitations of genetic medicine, as well as a clear appreciation of the physician's dual duties to maintain confidentiality and prevent harm. In light of this, this article reviews the available literature to address legal and ethical issues as well as potential policy directions surrounding a physician's "duty to warn" patients' relatives about genetic risk.

Introduction

Recently, well-known Harvard professor and bestselling author of *The Stuff of Thought*, Steven Pinker, allowed his entire genome not only to be sequenced, but also to be posted on the Internet for the world to see.¹ In so doing he has given celebrity status to key questions in the era of personalized genomics. These include whether one has a responsibility to disclose the results of genetic tests to family members who might likewise be affected,² and who should protect the consumers of the health care system from what has become known as "genetic discrimination".³

To date, instances of so-called genetic discrimination have been scarce in the United States³ and nearly unheard of in Canada, aside from the odd account of discrimination with respect to life insurance. Furthermore, this topic has been well covered by this publication in a previous article and will not be expanded upon further.⁴ However, an issue that does warrant further investigation is whether physicians have

an ethical and legal obligation to warn the relatives of patients with a positive genetic test if that information may benefit them. A thorough understanding of the theoretical benefits and limitations of genetic testing, as well as how these translate to our patients in practice, will ultimately have to guide policy in this matter.

Genetic Testing

According to Knoppers *et al.* (2004), genetic tests can be defined in a legal sense as either those tests that are based on "the presence or absence of specific genetic abnormalities" or more broadly, those that include "tests based on the end products of most genes." Knoppers *et al.* argues that in the latter case, genetic risk factors amount to nothing more than probabilities, and thus are difficult to distinguish from other routine predictive tests performed by a family physician.⁵ Some diseases can be conferred by a single gene mutation, or the inheritance of two mutated genes at the same genetic locus. In contrast, many

common diseases such as diabetes, cancer, and heart disease have a multifactorial etiology, involving the interaction between genes and the environment. Up to now, most genetic tests have been aimed at detecting single gene disorders, and often provided definitive diagnoses when the clinical picture was suggestive of genetic disease. With the advent of whole genome sequencing, tests that enable clinicians to assess genetic risk in much the same way as a family history are increasingly available.⁶

Genetic Medicine

Medical uses of genetic information are nearly as diverse as the definition of genetic testing is broad. Physicians can use genetic information to make diagnoses in newborns (e.g. phenylketonuria, PKU), to identify future health risks (e.g. BRCA1 and 2 mutations), and to predict drug responses or even to predict health risks to children not yet born.⁶ Most pertinent to the current discussion are those genetic diagnoses or predictive genetic tests that may indicate a familial risk of disease. Often some of this risk can be mitigated through early recognition of a genetic risk factor. Conversely however, there are genetic diseases where pre-symptomatic diagnosis confers no prognostic advantage, but which may carry a significant associated burden of disease, such as Huntington disease (HD).⁶

Central to the ethical dilemma of predictive genetic testing is what the information will actually mean to the patient in terms of health care options. In fact, according to Wylie Burke only about 20% of those at risk for development of HD in the UK had opted for predictive testing as of 2002.⁶ This reluctance to pursue predictive testing for an incurable disease may lie in the psychosocial consequences of an unfavorable result. One study by Giargiulo *et al.* found that of 119 patients who underwent predictive testing for HD, a significantly greater number of pre-symptomatic carriers experienced depressive episodes than those found to be non-carriers.⁷ Those people who do choose to pursue predictive testing often do so in order to make informed decisions about reproduction, rather than for the hope of treatment.^{7,8} In this sense, predictive

genetic testing can be used to make decisions that limit the possibility of birthing a child with significant risk of disease development. On the other hand, many genetic disorders show incomplete penetrance, with some individuals having an altered genotype but showing no evidence of disease.⁶

Confidentiality and the Duty to Warn

Physicians have a duty to protect the genetic information of a patient from unauthorized disclosure to any third party. However, under certain circumstances confidentiality may be justifiably breached, as is the case when legislation mandates it, or when it is breached with the aim of preventing harm to a third party.⁹ Numerous legal precedents, including *Tarasoff v. Regents of the University of California*¹⁰ and *Pitman Estate vs. Bain*¹¹ recognize a physician's "duty to warn" under circumstances where "disclosure is essential to avert danger to others".⁹ Furthermore, the Personal Health Information Protection Act provides that physicians or other health care workers "may disclose personal health information if there are reasonable grounds to believe that the disclosure is necessary to eliminate or reduce a significant risk of serious bodily harm to a person or a group of persons."¹²

It is important to understand this distinction when considering whether or not a family member may be entitled to know about a patient's genetic information. An excusable breach of confidentiality is technically illegal, though the courts may choose not to mandate compensation or punishment based on well-intentioned motives.⁹ Thus, a patient's right to confidentiality is legally protected in general, though the courts recognize that under certain circumstances it may be ethically necessary for a physician to breach this confidentiality. By contrast, a physician's "duty to warn" refers to a legal obligation to inform a third party of imminent risk to their well-being, regardless of a patient's right to confidentiality.^{2,9,13} Whether or not such a breach of confidentiality might be justified in the case where disclosure of genetic information to family members may enable them to decrease their risk of disease development remains to be determined

by the courts.^{2,9} On the other hand where genetics is concerned, physicians become privy to information that has implications to both the patient and their relatives. In this sense, the calculation of genetic risk in one person may violate another's right to personal autonomy where health care decisions are concerned.⁹ Thus Dickens *et al.* argue that health care professionals may take on a new and very real set of duties as it pertains to sensitive information that may be very beneficial to individuals who may in fact not be their actual patients.⁹

While a "duty to warn" has been recognized in the context of both psychiatric illness¹⁰ and infectious disease,¹¹ Canadian courts have yet to set similar legal precedents where genetic information is concerned.² Whether or not the aforementioned precedents might apply to cases where disclosure of genetic information may benefit a third party is questionable. Disease risk conferred by your genotype is not preventable, nor can relatives be viewed as potential victims of the patient (i.e. the patient does not directly cause the risk).^{2,9} Finally, less than 1% of physicians surveyed in the US believed a "duty to warn" was reasonable in instances where no valid medical therapy exists.¹³

Notwithstanding, as of 2004 failure to warn a relative of genetic risk had already resulted in 3 lawsuits against physicians in the United States.¹³ In all three of these cases, State Appellate courts ruled in favor of the plaintiffs, asserting that physicians must ensure that immediate family members are warned of impending genetic risk (though in one case the actual jury eventually decided in favor of the physician).¹³ Currently in Canada there is no obligation to warn family members of genetic risk, and disclosure could even potentially be punishable by courts or professional regulatory bodies.² Some strategies aimed at resolving this issue include: strict confidentiality, a universal duty to warn, and informed consent or as Offit *et al.* would say a "genetic Miranda warning."^{2,13} This third strategy involves physicians informing their patients of the circumstances under which they would be obliged to disclose information prior to genetic testing. While the first strategy

does not adequately protect the interests of the family members, the latter two strategies are coercive and do not respect the patient's right to make autonomous decisions about health care. In addition, it is likely that they would adversely affect patient utilization of predictive testing because of perceived genetic discrimination.^{2,13} Thus, the currently accepted strategy is to adopt an "intermediate position" whereby confidentiality is respected as a rule, but it is recognized that circumstances exist in which disclosure of genetic information may be justifiable.^{2,9}

Conclusion

While no formal legal duty to warn currently exists in Canada, there is a general consensus that physicians should make a valiant effort to inform their patients that genetic information may prove invaluable or even life saving to their relatives. In cases where penetrance is high, risk of imminent and severe harm is great, and viable treatment options do exist, the benefits of disclosure to a third party may so far outweigh the potential harm to the patient as to make it justifiable in a court of law.^{2,9,13} Thus the physician's dual duties to maintain confidentiality while simultaneously acting in a spirit of beneficence continue to be at odds in this matter. Unfortunately it is likely that they will remain so until more is known about the circumstances under which knowledge of their relative's genetic risk might actually prove beneficial to an individual.

References

1. My Genome, My Self. Steven Pinker. New York, USA: New York Times. January 11, 2009. MM24 (Accessed at <http://www.nytimes.com/2009/01/11/Genome-t.html?pagewanted=all> on March 23, 2009).
2. Lacroix M, Nycum G, Godard B, Knoppers BM. Should physicians warn patients' relatives of genetic risks? CMAJ. 2008;178:503-5.
3. Geely, H. Banning Genetic discrimination. N Eng J Med. 2005;353:865-7.
4. Slatnik M, Fortin C. BRCA Genetic Testing: Ethical, Legal and Policy Considerations. UWOMJ 2007;76:26-29.

5. Knoppers *et al.* Genetics and Life Insurance in Canada: Points to Consider. CMAJ. 2004;170(online):1-3.
6. Burke W. Genetic testing. 2002. N Engl J Med. 2002;347:1867-75.
7. Gargiulo M, Lejeune S, Tanguy M-L, Lahlou-Laforêt K, Faudet A, Cohen D, Feingold J, Durr A. Long-term outcome of presymptomatic testing in Huntington disease. Eur J Hum Genet. 2009;17:165-171.
8. Decruyenaere M, Evers-Kiebooms G, Boogaerts A, Philippe K, Demyttenaere K, Dom R, Vandenberghe W, Fryns J-P. The complexity of reproductive decision-making in asymptomatic carriers of the Huntington mutation. Eur J Hum Genet. 2007;15:453-462.
9. Dickens B, Pei N, Taylor K. Legal and ethical issues in Genetic Testing and counseling for susceptibility to Breast, Ovarian and Colon Cancer. CMAJ. 1996;154:6813-18.
10. *Tarasoff v. Regents of California* (1976), 551 P 2nd 334 (sup ct).
11. *Pitman Estate v. Bain* (1994), 112 DLR (4th) 257 (Ont Gen Div).
12. Cavoukian A. A Guide to the Personal Health Information Protection Act. Information and Privacy Commissioner/Ontario. 2004.
13. Offit K, Groeger E, Turner S, Wadsworth E, Weiser M. The "Duty To Warn" a Patient's Family Members About Hereditary Disease Risks. JAMA. 2004;292:1469-73.



SARSTEDT

Your partner in medicine and research worldwide

The SARSTEDT Group develops, manufactures and sells equipment and consumables in the field of medicine and research. Ever since it was set up in 1961 the company has continued to grow to the point where it now employs a workforce of 2,500.



6373 Des grandes prairies
Montreal, QC
H1P 1A5

Tel: 1-888-727-7833
Website: www.sarstedt.com

CU advertising



*Rolling out the Red Carpet
for all your Advertising
needs....*



www.cu-ads.com 1.866.362.3331

Personalized Cancer Management

Jenny Shu (Meds 2012) and Pencilla Lang (Meds 2011)

Faculty Reviewer: Dr. Robert Hegele

The aggression of cancer is characterized by its multigenic and heterogeneous nature. With the completion of the Human Genome Project in 2003, there has been growing interest in the field of personalized medicine for cancer management, using an individual's genetic composition to direct prevention, detection, prognostic, and therapeutic efforts. This review will provide an overview of some of the molecular profiling techniques that are currently in use, including transcriptomics, proteomics, and genomics using assessment of single nucleotide polymorphisms (SNPs), copy number variation (CNV) and high-throughput gene sequencing. Some recent examples of personalized medicine applications in the screening, diagnosis, tumor classification, and targeted therapy of various types of cancer will also be discussed. Although more validation and consolidation of these research studies is required before personalized medicine is widely used for cancer management, its potential benefits hold implications for pharmaceutical companies, diagnostic agencies, healthcare providers, and most importantly, patients.

Introduction

A diagnosis of 'cancer' is a life-changing event for patients. An estimate of over 10 million people globally will die annually as a result of cancer in the year 2020.¹ It is not surprising that there is a drive to further our understanding of the disease mechanisms, pathophysiology, and treatment of cancer. The multigenic and heterogeneous nature of cancer has long been recognized, and this heterogeneity can determine the aggressiveness of cancer in an individual patient.^{2,3} The completion of the Human Genome Project in 2003^{4,5} has brought forth the hope of advancing medical practice into a "genomic era" that will identify key therapeutic targets and disease biomarkers for cancer. Despite these advancements, much of the systemic therapy for cancer patients is still empirical⁶ and two patients who may be at apparently 'identical' stages of cancer and harbor the 'same' tumor type will exhibit significantly different clinical outcomes as measured by survival and therapy response.⁷

Instead of continuing to progress down the traditional 'trial and error' method of testing for new therapeutic agents, the personalized medicine model has been proposed because of its

theoretical appeal: in this model, an individual's genetic composition helps focus and direct prevention, detection, prognostic, and therapeutic efforts.⁷ This article will provide an overview of the current status of molecular profiling technologies and their current applications in personalized medicine, from the initial stages of identifying significant causative and modulating genes to the integration of multiple genes within a personalized profile that will direct therapy in the sub-population of cancer patients.

Molecular Profiling Techniques

The main molecular platforms currently widely used in personalized medicine are transcriptomics, proteomics and genomics, which includes the analysis of single nucleotide polymorphisms (SNPs), copy number variations (CNVs) and high-throughput gene sequencing.⁷ Of course, these are often not mutually exclusive and one of the current goals in personalized medicine is to develop an approach to utilize and integrate the information from different techniques.

Transcriptomics, the study of altered expression of the levels of messenger RNA (mRNA) within cancer tissue samples or biopsies, has led to the development of Internet-based resources such as OncoPrint Research Platform⁸ that maintains a catalogue of published transcriptome profiles so that clinicians and scientists can easily access the information. Although microarray technology, the main means of transcriptome profiling, is cost-effective and accurate, it is still inhibited by sample heterogeneity and the absence of a high-quality multi-institutional tumor tissue library that would be necessary to cater to a large population.

Proteomics also explores genetic expression, but at the level of translated protein quantity, structure and function within cancer tissue samples. While technically more difficult than transcription profiling, proteomic analysis has the advantage of more directly examining molecular machinery of cell physiology, post-translational modifications, and protein-protein complexes.⁹ Mass spectrometry and protein microarrays have also been used to target specific biomarkers in signaling cascades, although there are some limitations with respect to difficulties in large-scale target identification as the exact number of polypeptides produced is still uncertain.¹⁰

Finally, there is a wide range of genomic DNA variations that predispose or propagate the development of cancer. Examples include single nucleotide polymorphisms (SNPs), which are genetic variations in the DNA sequence due to the difference of one nucleotide among individuals. Copy number variations (CNVs) refer to larger-scale changes, including gene duplication, deletion, inversion or translocation events. Both SNPs and CNVs have been linked to a wide range of different physiological phenotypes and diseases. However, this is only an initial step; the examination of genomic DNA in a patient's germline or in somatic cells taken from cancerous tissue can stratify risk and identify new causative genes and pathways, but this information alone is not sufficient to define mechanisms of disease or therapeutic approaches.

The ultimate genomic assessment is whole-genome sequence analysis, in which all 3 billion nucleotides of genomic DNA from an individual patient or the cancer tissue sample are analyzed using automated nucleotide sequencers to determine potential functional changes from a normal reference sequence. There have been many initiatives such as the Human Cancer Genome Project and Cancer Genome Anatomy Project to use high throughput gene sequencing to identify novel disease-associated mutations at a genome-wide level.¹¹ Much of the attention has shifted from mastering the technologies themselves to extracting biologically relevant information – a field called “bioinformatics” – such as pairing screen results with functional assessments of a specific neoplastic process.⁷ Despite the more comprehensive sequence information at the expense of a higher cost, there have been some applications of genomic and bioinformatic analysis in tumor classification.¹²

Screening and Diagnostics

Preventive measures taken against cancer can use personalized medicine to screen for one's predisposition to cancer, which is important in providing a wider range of therapeutic options for the patient and guiding in clinical decision making. Genetic screening is particularly useful for individuals with a family history of cancer, where the presence of oncogenic mutations such as *APC* (adenomatous polyposis coli), *BRCA1* (breast cancer 1, early onset), and *BRCA2* (breast cancer 2, early onset) can be used as risk factors before any symptoms or even cellular dysplasia occurs.¹³ Celecoxib, the current chemopreventive agent of choice for colorectal cancer, is only recommended for high-risk patients with familial adenomatous polyposis linked to the *APC* oncogene mutation.¹³ There is also a drive to utilize personalized medicine in clinical diagnostics. Although the prostate-specific antigen (PSA) is the gold-standard for detecting prostate cancer, greater diagnostic accuracy in unscreened populations can be achieved if biologically-based prediction models are used to link biomarker levels to other individual factors such as age.¹⁴

Prognosis and Tumor Classification

Another important goal in personalized medicine is to consolidate information from traditional prognostic factors and genetic profiles in order to optimize treatment for patients. An important prognostic biomarker for breast cancer is the human epidermal growth factor receptor 2 (Her2), which encodes a transmembrane receptor protein that is overexpressed in breast cancer cells.¹⁵ Specifically, these levels can be used to predict clinical outcomes; serial changes in the circulating Her2 extracellular domain (ECD) levels have paralleled the clinical course of disease regardless of the treatment. Specifically, higher ECD levels have been correlated with earlier recurrence of breast cancer.¹⁵ There is also a 186-gene “invasiveness” gene signature (IGS) generated from differentially expressed genes that has been found to be strongly associated with metastasis-free survival and overall survival of medulloblastoma, lung cancer, prostate cancer, and especially breast cancer.¹⁶ Hence, bioinformatics-centric prediction models have been gaining ground in predicting the therapeutic response of common tumors based on their gene expression profiles.

Targeted Therapy

Current research has been progressing towards a consolidation of our current knowledge on diseases, genetic alterations, and drug responses, allowing both the creation of new chemotherapeutic drugs and expanding existing ones on the market. For instance, imatinib mesylate (Gleevec) is a drug that was initially approved by the Food and Drug Administration in 2001 to treat chronic myelogenous leukemia (CML) by targeting the BCR-ABL protein.¹⁷ Later, Gleevec became the first specific effective treatment against advanced gastrointestinal stromal tumors (GISTs) after researchers found that the link between the mutant KIT protein, which shows similarities to the BCR-ABL protein, and GIST aggression.¹⁸ However, researchers are still looking into ways to probe into individual resistance mechanisms to combat the issue of Gleevec resistance in patients.¹⁹ Gefitinib (Iressa) is another anticancer drug that

targets the epidermal growth factor receptor (EGFR), an important component leading to lung cancer aggression. The concept of personalized medicine may explain why there were largely negative results of clinical trials as only 10-15% of the test patient population carried the EGFR gene mutation or gene amplification.²⁰ Hence, clinicians are looking into the possibility of conducting more targeted trials that restrict eligibility of subjects to those whose molecular profile predicts a better response to a particular chemotherapeutic agent.

Challenges and the Future of Personalized Medicine

Although there is much hope regarding the potential of personalized medicine, there are still many barriers preventing its widespread usage. Duffy *et al.*⁶ illustrate some of the key challenges in the field, including underpowered studies, invalidated results from independent patient populations or prospective trials, multiple end points and subsets of patients used, and poor quality of design and unreliable data. These problems can be addressed in part by better coordinated multi-centered prospective studies using much larger patient cohorts than have been studied to this point. Nonetheless, the potential benefits of personalized medicine – with respect to improving positive outcomes and minimizing side effects and costs of treatment - are worth taking the time and effort to pursue. This holds implications for pharmaceutical companies, diagnostic agencies, members of the healthcare profession, and most importantly, patients. Indeed, there is a movement towards focus on the individual and not just the disease—what has been a barrier to treatment in the past may very well be the key to unlocking mechanisms of disease to combat cancer in the future.

References

1. Parkin DM, Bray F, Ferlay J, Pisani P. Global cancer statistics, 2002. *CA Cancer J Clin.* 2005; 55: 74-108.
2. Kopnin BP. Targets of oncogenes and tumor suppressors: key for understanding basic

- mechanisms of carcinogenesis. *Biochemistry (Mosc)*. 2000; 65:2-27.
3. Croce CM. Oncogenes and cancer. *N Engl J Med* 2008; 358:502-511.
 4. Duffy MG. Role of tumor markers in patients with solid cancers: a critical review. *Eur J Intern Med*. 2007 May;18(3):175-184.
 5. [no authors listed]. International Germ Cell Consensus Classification: a prognostic factor-based staging system for metastatic germ cell cancers. International Germ Cell Cancer Collaborative Group. *J Clin Oncol*. 1997 Feb;15(2):594-603.
 6. Duffy MJ, Crown J. A personalized approach to cancer treatment: how biomarkers can help. *Clin Chem*. 2008 Nov;54(11):1770-9. Epub 2008 Sep 18.
 7. Overdevest JB, Theodorescu D, Lee JK. Utilizing molecular gateway : the pathway to personalized cancer management. *Clin Chem*. 2009 Feb 26. [Epub ahead of print]
 8. Compendia Bioscience. Cancer Profiling Database [Internet]. Oncomine Research; 2007. [cited 2009 March 20]. Available from: www.oncomine.org/.
 9. Koomen JM, Haura EB, Bepler G, Sutphen R, Remily-Wood ER, Benson K, Hussein M, Hazlehurst LA, Yeatman TJ, Hildreth LT, Sellers TA, Jacobsen PB, Fenstermacher DA, Dalton WS. Proteomic contributions to personalized cancer care. *Mol Cell Proteomics*. 2008 Oct;7(10):1780-94. Epub 2008 Jul 29.
 10. Aebersold R, Mann M. Mass spectrometry-based proteomics. *Nature*. 2003 Mar 13;422(6928):198-207.
 11. Strausberg RL, Simpson AJG, Woster R. Sequence-based cancer genomics; progress, lesions and opportunities. *Nat Rev Genet*. 2003; 4: 409-18.
 12. Ley TJ, Minx PJ, Walter MJ, Ries RE, Sun H, McLellan M, *et al*. A pilot study of high throughput sequence-based mutational profiling of primary human acute myeloid leukemia cell genomes. *Proc Natl Acad Sci USA*. 2003; 100-14275-180.
 13. Ponder B. Genetic testing for cancer risk. *Science*. 1997; 278: 1050-4.
 14. Shariat SF, Scardino PT, Lilja H. Screening for prostate cancer: an update. *Can J Urol*. 2008 Dec;15(6):4363-74.
 15. Carney WP, Neumann R, Lipton A, Leitzel K, Ali S, Price CP. Monitoring the circulating levels of the HER2/neu oncoprotein in breast cancer. *Clin Breast Cancer*. 2004 Jun;5(2):105-16.
 16. Liu R, Wang X, Chen GY, Dalerba P, Gurney A, Hoey T, Sherlock G, Lewicki J, Shedden K, Clarke MF. The Prognostic Role of a Gene Signature from Tumorigenic Breast-Cancer Cells. *N Engl J Med*. 2007 Jan 18;356(3):217-26.
 17. Wei G *et al*. Gene expression-based chemical Optimizing Imatinib Mesylate Treatment in Gastrointestinal Stromal Tumors. *Gastrointest Cancer Res*. 2008 Sep;2(5):245-50.
 18. Paez JG, Janne PA, Lee JC, *et al*. *EGFR* mutations in lung cancer: Correlation with clinical response to gefitinib therapy. *Science* 2004; 304:1497-500 genomics identifies rapamycin as a modulator of MCL1 and glucocorticoid resistance. *Cancer Cell*. 2006 Oct;10(4):331-42.
 19. O'Brien SG, Guilhot F, Larson RA *et al*. Imatinib compared with Interferon and low-dose cytarabine for newly diagnosed chronic-phase chronic myeloid leukemia. *N Engl J Med*. 2003 Mar 13;348(11):994-1004.
 20. Paez JG, Janne PA, Lee JC, *et al*. *EGFR* mutations in lung cancer: Correlation with clinical response to gefitinib therapy. *Science*. 2004; 304:1497-500.

Marfan Syndrome

Kalpa Shah (Meds 2012) and Aiman Alak (Meds 2011)

Faculty Reviewer: Dr. Victoria Siu

Marfan syndrome was first described in 1896 by Antonine-Bernard Marfan, Professor of Pediatrics in Paris. It is a relatively common inherited connective tissue disorder with an incidence of 1 in 10,000 individuals. The condition is inherited in an autosomal dominant pattern, with an equal distribution in males and females.

Case

The following case is meant to serve as a prompt to guide the discussion on the clinical aspects of Marfan Syndrome.

An 18 year old man develops sudden chest pain while playing basketball. He is noted to be quite tall at 6'5" and has long fingers and toes. Does he have Marfan syndrome? What are the diagnostic criteria? If he has Marfan syndrome, what are 3 possible causes of his chest pain? It would be relevant to note whether the patient wears contact lenses as individuals with Marfan syndrome are often near- or far-sighted and/or have other eye abnormalities.

Clinical Presentation

Most patients who have Marfan syndrome are usually diagnosed incidentally. Typically, patients with Marfan syndrome present with tall stature, lens dislocation (ectopia lentis), aortic root dilatation and a positive family history. Less common presentations include:

- Dominant ectopia lentis with variable skeletal and negligible cardiac involvement
- Mitral valve prolapse without skeletal features
- Dominant aortic aneurysm without skeletal and ocular features

- Presentation at birth, rapid aortic dilatation, deformities (such as pectus deformities, where the breastbone protrudes inward or outward), and death
- Sudden death¹

Pathogenesis

The genetic basis for Marfan syndrome is a mutation in the gene encoding fibrillin-1 (FBN-1) found on chromosome 15. Fibrillin is a glycoprotein that is an integral part of microfibrils found in the connective tissue of the body. Microfibrils function as the main component of elastic fibres, anchoring fibres between the dermis and epidermis, as well as in the lens of the eye. Classically, it was thought that the ubiquitous features of Marfan syndrome could be attributed to weak or deformed connective tissue.² However, recent research suggests that a defect in the fibrillin-1 structure reduces its ability to bind to the cytokine TGF-B, which results in increased expression of TGF-B. In mouse models, increased TGF-B signaling was associated with myxomatous mitral valve leaflets and aortic dilatation, both features of Marfan syndrome. Furthermore, by administering an antibody that binds TGF-B, it was shown that these common features could be prevented.³ Thus, it is now thought that it is the over-expression of the cytokine TGF-B, due to the inability of the defective fibrillin-1 that would in normal circumstances bind TGF-B, that results in the features of Marfan syndrome.

Marfan syndrome has diverse manifestations that can affect many organ systems such as the skeletal, ocular, cardiovascular systems and can also involve the skin, lungs and muscle tissue - an example of pleiotropy.

Clinical Diagnosis

The diagnosis of Marfan syndrome (MFS) is usually based on clinical features and family history.⁴ The features of Marfan syndrome may overlap with other connective tissue disorders and the presentation of Marfan syndrome varies widely, even among family members. Nonetheless, the development of a set of guidelines, known as the Ghent criteria, has brought clarity to the issue.⁴

- If an individual has a first-degree relative affected by Marfan syndrome, a diagnosis of Marfan syndrome requires major involvement in one organ system (skeletal, cardiovascular, or ocular) and minor involvement of a second organ system.
- In the absence of a family history for Marfan syndrome (or ambiguous family history), a diagnosis of Marfan syndrome requires major criteria in two different organ systems and minor involvement of a third.

Please refer to Table 1 attached at the end of the article for more details on the clinical features that would constitute major or minor involvements in different organ systems.

As a simplified guideline, for a patient suspected of having Marfan syndrome some investigations that are helpful in making a clinical diagnosis include:

- Examination by an ophthalmologist to look for lens dislocation or subluxation; often patients with Marfan syndrome are near-sighted (myopia) and at increased risk for developing cataracts and glaucoma

- Referral to a cardiologist for an echocardiogram to assess if there are any cardiovascular abnormalities such as mitral valve prolapse, aortic root dilatation or aortic dissection
- Physical exam to assess skeletal features: ratio of arm span-to-height ratio which is considered significant if it is greater than 1.05, or wrist and thumb signs. (A positive wrist sign is present if the thumb and fifth finger overlap when encircling the opposite wrist. A positive thumb sign is present if the entire thumbnail protrudes beyond ulnar border of hand when the thumb is adducted across the palm.)

Differential Diagnosis

The differential diagnosis for MFS includes homocystinuria, congenital contractural arachnodactyly, mild aortic root dilatation-skeletal-skin (MASS) syndrome, Ehlers-Danlos syndrome, Stickler syndrome, congenital bicuspid aortic valve disease, aortic coarctation, Loeys-Dietz syndrome, and familial thoracic aortic aneurysm with aortopathy. The most similar differential is homocystinuria as the two diseases share a similar phenotype. Features that overlap between Marfan syndrome and homocystinuria include ectopic lentis, severe myopia, mitral valve prolapse, and skeletal abnormalities, such as body habitus, chest deformities, and spine deformities. Patients with homocystinuria are at an increased risk for thromboembolic events, but many patients with this condition are responsive to pyridoxine. Thus, it is important to screen for homocystinuria with measurement of urinary amino acids, as there are implications for management.⁵

Management

Overall, the prognosis for Marfan syndrome is relatively favorable. Activity restrictions, medications, monitoring, and elective surgical interventions were associated with an improved life span for MFS patients from 41 years in 1993 to 61 years in 1996.^{6,7}

Aortic disease is the most significant source of morbidity and mortality for patients with MFS. Untreated MFS can be associated with an aortic dissection spanning the entire length of the aorta. Aortic dissection occurs earlier among patients with MFS compared to the other cases of aortic dissection. Assessment for aortic root

dilatation and regurgitation can be performed by thoracic echocardiography; however, if that yields incomplete or insufficient information, transesophageal echocardiography can be used. Routine measurements can be performed annually as long as the aortic root diameter increases proportional to increases in body surface area. If

Table 1. Ghent Criteria for Diagnosis of Marfan Syndrome*

System	Major	Minor
Family/genetic history	<ul style="list-style-type: none"> • Having a first-degree relative who meets these diagnostic criteria • Presence of a mutation in FBN1 known to cause Marfan syndrome 	<ul style="list-style-type: none"> • None
Skeletal	<ul style="list-style-type: none"> • Presence of at least 4 of the following manifestations: • Pectus carinatum • Pectus excavatum requiring surgery • Reduced upper-to-lower segment ratio or arm span-to-height ratio > 1.05 • Wrist and thumb signs • Scoliosis > 20 degrees or spondylolisthesis • Reduced extension at elbows (<170 degrees) • Medial displacement of medial malleolus causing pes planus • Protrusio acetabulare of any degree 	<ul style="list-style-type: none"> • Pectus excavatum of moderate severity • Joint hypermobility • Highly arched palate with crowding of teeth • Facial appearance (dolichocephaly, malar hypoplasia, enophthalmos, retrognathia, down-slanting palpebral fissures)
Ocular	<ul style="list-style-type: none"> • Ectopia lentis (dislocated lens) 	<ul style="list-style-type: none"> • Abnormally flat cornea • Increased axial length of globe
Cardiovascular	<ul style="list-style-type: none"> • Dilatation of ascending aorta with or without aortic regurgitation and involving at least the sinuses of Valsalva or dissection of ascending aorta 	<ul style="list-style-type: none"> • Mitral valve prolapse with or without mitral valve regurgitation • Dilatation of the main pulmonary artery, in the absence of valvular or peripheral pulmonic stenosis or any other obvious cause in patients < 40 years • Calcification of mitral annulus in patients <40 years • Dilatation or dissection of the descending thoracic or abdominal aorta in patients < 50 years
Pulmonary	<ul style="list-style-type: none"> • None 	<ul style="list-style-type: none"> • Spontaneous pneumothorax • Apical blebs
Skin and integument	<ul style="list-style-type: none"> • None 	<ul style="list-style-type: none"> • Stretch marks not associated with weight changes, repetitive stress • Recurrent incisional hernias
Dura	<ul style="list-style-type: none"> • Lumbosacral dural ectasia (by CT or MRI) 	<ul style="list-style-type: none"> • None

* Adapted from Rangasetty UC, Karnath BM. (2006)¹

the increase is disproportional, or if the aortic root diameter is greater than 45 mm, biannual measurements should be performed.

The European Society of Cardiology has published recommendations for the treatment of patients with MFS.⁸ The standard of care involves beta blockers which can delay the progression to aortic dissection. Beta blockers decrease myocardial contractility and pulse pressure and may also improve the elastic properties of the aorta, particularly in patients with an aortic root diameter <40 mm.^{9,10} In a randomized trial, treatment with propranolol was associated with delayed progression of aortic dilatation, and higher survival at approximately five years. Longer term survival differences between propranolol and placebo were less clear.¹¹ No randomized trials have been done to establish the efficacy of beta blockers in children.¹² However, many clinicians would give beta blockers to all children with MFS. In the future, therapy directed at the renin-angiotensin-aldosterone system may prove beneficial, but the role of these drugs in MFS has not yet been established.^{13,14}

Patients with MFS may participate in low to moderate intensity, non-competitive exercise such as bowling, golf, stationary bike, or modest hiking. Strongly discouraged activities include: weight lifting, ice hockey, rock climbing and surfing.¹⁵ The choice of permissible activities requires individual assessment. For children to comply with these activity restrictions, there must be coordination between parents, school officials, and physical educators.

Elective surgical repair for an aortic dilatation is associated with reduced mortality compared to urgent or emergent repairs. For adults and children, surgical repair should be considered at an aortic root diameter of ≥ 50 mm.¹⁶ Uncertainties exist regarding the root diameter for performing elective surgery. The 2006 American College of Cardiology/American Heart Association (ACC/AHA) guidelines recommend elective surgery for an aortic root diameter ≥ 50 mm, while European Society of Cardiology (ESC) recommends surgery for an aortic root diameter of ≥ 45 mm.¹⁷ In addition to

the absolute size of the aortic root diameter, other considerations are a family history of aortic dissection or a rapid increase in aortic size (i.e., ≥ 5 mm per year).¹⁸ Some suggest using aortic root dimensions relative to body surface area. This factor is particularly important for children. For children, the only clear indications for surgical intervention are: an aortic root diameter of ≥ 45 mm ("giant aneurysm"), rapid enlargement of >10 mm/year, or progressive aortic insufficiency. Other cut-offs for aortic root diameter are less clear.

A few options exist regarding the surgical technique and the choice depends on patient factors. One approach involves total replacement of the aortic root.¹⁹ Alternatively, for patients with structurally normal valves the native aortic valve may be retained using remodeling and reimplantation techniques.^{20,21} In this latter option, life-long anti-coagulation is not required; however, repeat operations may be necessary. Antibiotic prophylaxis should be individualized. As other sites throughout the aorta maybe involved, follow-up radiography using MRI or CT angiography is required indefinitely.²²

Ocular and musculoskeletal problems also require attention. Eyeglasses can correct myopia, while artificial lens insertion should only be undertaken when growth of the eye is complete. Further, photocoagulation can correct retinal tears and detachment. If intervention with physical therapy and bracing fails, scoliosis may require surgical stabilization of the spine. Orthotics can correct flat foot, which is associated with muscle cramps and leg fatigue.

Answers to Case

Diagnosis would be made based on Ghent criteria. Possible causes of chest pain are: muscle/ligament strain, aortic root dissection, and pneumothorax.

Glossary

pectus carinatum: deformity of the chest characterized by protrusion of ribs and sternum

pectus excavatum: deformity in which several ribs and the sternum grow abnormally, producing a concave appearance in anterior chest wall

protusio acetabulare: protrusion of the acetabulum (socket that receives femoral head to make the hip joint)

pes planus: flat foot

dolichocephaly: elongated skull

enophthalmos: recession of eyeball within the orbit

sinuses of Valsalva: also known as aortic sinus; the space between each semilunar valve and the wall of the aorta

References

1. Rangasetty UC, Karnath BM. Clinical signs of Marfan syndrome. *Hosp Phys*. 2006; 33-38.
2. Dietz HC, Cutting GR, Pyeritz RE, *et al*. Marfan syndrome caused by a recurrent de novo missense mutation in the fibrillin gene. *Nature*. 1991; 352: 339-10.
3. Pyeritz RE. A small molecule for a large disease. *N Engl J Med*. 2008;358: 2829-2831.
4. Rangasetty UC, Karnath BM. Clinical signs of Marfan syndrome. *Hosp Phys*. 2006: 33-38.
5. Fowler B, Jakobs C. Post- and prenatal diagnostic methods for the homocystinurias. *Eur J Pediatr*. 1998; 157 Suppl 2:S88.
6. Finkbohner R, Johnston D, Crawford S, *et al*. Marfan syndrome: Long-term survival and complications after aortic aneurysm repair. *Circulation*. 1995; 91:728.
7. Silverman DI, Burton KJ, Gray J, *et al*. Life expectancy in the Marfan syndrome. *Am J Cardiol*. 1995; 75:157.
8. Erbel R, Alfonso F, Boileau C, *et al*. Diagnosis and management of aortic dissection. *Eur Heart J*. 2001; 22:1642.
9. Groenink M, de Roos A, Mulder BJ, *et al*. Changes in aortic distensibility and pulse wave velocity assessed with magnetic resonance imaging following beta-blocker therapy in the Marfan syndrome. *Am J Cardiol*. 1998; 82:203
10. Rios AS, Silber EN, Bavishi N, *et al*. Effect of long-term β -blockade on aortic root compliance in patients with Marfan syndrome. *Am Heart J*. 1999; 137:1057
11. Shores J, Berger KR, Murphy E, *et al*. Progression of aortic dilatation and the benefit of long-term beta-adrenergic blockade in Marfan's syndrome. *N Engl J Med*. 1994; 330:1335.
12. Ahimastos AA, Aggarwal A, D'Orsa KM, *et al*. Effect of perindopril on large artery stiffness and aortic root diameter in patients with Marfan syndrome: a randomized controlled trial. *JAMA*. 2007; 298:1539.
13. Brooke BS, Habashi JP, Judge DP, *et al*. Angiotensin II blockade and aortic-root dilation in Marfan's syndrome. *N Engl J Med*. 2008; 358:2787.
14. Maron BJ, Chaitman BR, Ackerman MJ, *et al*. Recommendations for physical activity and recreational sports participation for young patients with genetic cardiovascular diseases. *Circulation*. 2004; 109:2807.
15. Erbel R, Alfonso F, Boileau C, *et al*. Diagnosis and management of aortic dissection. *Eur Heart J*. 2001; 22:1642.
16. Vahanian A, Baumgartner H, Bax J, *et al*. Guidelines on the management of valvular heart disease: The Task Force on the Management of Valvular Heart Disease of the European Society of Cardiology. *Eur Heart J*. 2007; 28:230.
17. Bonow RO, Carabello BA, Chatterjee K, *et al*. ACC/AHA 2006 guidelines for the management of patients with valvular heart disease. A report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (Writing committee to revise the 1998 guidelines for the management of patients with valvular heart disease). *J Am Coll Cardiol*. 2006; 48:e1.
18. Karck M, Kallenbach K, Hagl C, *et al*. Aortic root surgery in Marfan syndrome: Comparison of aortic valve-sparing reimplantation versus composite grafting. *J Thorac Cardiovasc Surg*. 2004; 127:391
19. Kallenbach K, Karck M, Pak D, *et al*. Decade of aortic valve sparing reimplantation: are we pushing the limits too far? *Circulation*. 2005; 112:I253
20. Silverman DI, Gray J, Roman MJ, *et al*. Family history of severe cardiovascular disease in Marfan syndrome is associated with increased aortic diameter and decreased survival. *J Am Coll Cardiol*. 1995; 26:1062
21. Dietz H. Marfan Syndrome [Internet]. Seattle.: University of Washington; 2005 Oct 26 [Cited 2009 Apr 3]. Available from: <http://www.ncbi.nlm.nih.gov/bookshelf/br.fcgi?book=gene&part=marfan>.

Human Statues: Challenges in Management of Patients with Fibrodysplasia Ossificans Progressiva

Anna Burianova (Meds 2012), Ashley Brown (Meds 2011), and Jenna Ashkanase (Meds 2011)

Faculty Reviewer: Dr. Kathleen Hill

Fibrodysplasia ossificans progressiva (FOP) is a rare genetic disorder in which soft tissues transform into bone, causing patients to gradually become entombed in their own bodies. Early symptoms typically involve inflammatory lesions that are often confused with a variety of tumours and other conditions, leading to delays in diagnosis and medical procedures that may cause significant detriment. The recent discovery of the FOP gene may help to alleviate this issue, as DNA testing can allow for earlier identification of the condition. A case study is presented to exemplify the challenges faced when treating a patient with FOP, and possible complications are detailed to emphasize the consequences if precautions are not taken. With a specific gene target in sight, it is hoped that treatments can extend beyond supportive care and be able to prevent or halt this debilitating condition.

Introduction

Metamorphosis is defined as the transformation of one normal tissue or organ system into another through a pathological process.¹ Within its realm, heterotopic ossification (HO) describes the abnormal formation of true bone within extraskelatal soft tissues. Fibrodysplasia ossificans progressiva (FOP) is an exceedingly rare genetic disorder which has, until recently, been one of medicine's most elusive mysteries and the most disabling condition of extraskelatal ossification known in humans.^{2,3}

Genetics

FOP is an extremely rare condition, occurring at a population frequency of about 1 per 2 million with no ethnic, racial, gender or geographic predisposition.^{1,2} The severe disability of FOP results in low reproductive fitness and fewer than ten multigenerational families are known worldwide.¹ Most cases of FOP are sporadic with only one affected individual in a family, though when observed, genetic transmission is autosomal dominant.^{1,2}

A recent breakthrough in the study of FOP was attained with the identification of a recurrent single nucleotide missense mutation in activin receptor IA/activin-like kinase 2 (ACVR1/ALK2).¹ This bone morphogenic protein (BMP) type I receptor causes skeletal morphogenesis, and is therefore the first identified human metamorphogene.^{1,3} The mutation has been reported in all sporadic and familial cases of classic FOP worldwide, making it one of the most highly specific disease-causing mutations in the human genome.³

Clinical Features

The classic presentation of FOP is defined by two clinical features: congenital malformation of the great toes and progressive HO.³ Congenital malformation of the great toes is the earliest phenotypic feature present in all classically affected individuals, who appear otherwise unremarkable at birth.^{1,2} Other skeletal anomalies often associated with FOP include developmental anomalies of the cervical spine, short malformed thumbs, clinodactyly, short broad femoral necks and proximal medial tibial osteochondromas.¹

During the first decade of life, children with FOP develop painful and highly inflammatory soft tissue swellings (or flare-ups) that progressively and permanently transform connective tissues into heterotopic bone.^{1,3} The ossification in FOP progresses in characteristic patterns that mimic normal embryonic skeletal development, with the first episodes typically occurring along the upper back and neck.^{1,2} However, several muscles including the diaphragm, tongue, extraocular and cardiac as well as smooth muscle are enigmatically spared from the FOP process.¹

Interestingly, the clinical features of early involvement in the axial regions differ from those seen in the appendicular regions.^{1,3} Even though swelling appears more rapidly than typically seen with neoplasms, the bulbous lesions which appear on the neck and back are often mistaken for tumours.^{1,3} In the limbs, on the other hand, the swelling is often diffuse and may be mistaken for acute thrombophlebitis.^{1,3}

The natural progression of the disease can also be altered by environmental factors, as any trauma leading to tissue injury has the potential to induce HO. This leads to episodes of explosive and painful new bone growth, with cumulative immobility.^{1,2}

Case Report

An 18-month-old boy from Britain presented with a brainstem lesion in need of neurosurgery. He had recently been diagnosed with FOP, and physical exam prior to surgery revealed heterotopic ossification of the dorsal and lumbar paravertebral muscles, the left sternocleidomastoid, and fusion of the C4-C6 vertebrae. Although the brainstem lesion was in this case unrelated to his FOP, his condition nonetheless complicated the procedure to remove it. His montelukast was switched to prednisone to prevent inflammation 24 hours before surgery and continued until four days post-op. Direct laryngoscopy was not possible due to the fusion of his cervical vertebrae, therefore fiberoptic bronchoscopy via the nostril was necessary to intubate him. Traumatic injury was avoided by

using silicon carpets and a headrest, as well as by padding every point of contact with cotton wool. Because his FOP was recognized and attended to properly, the procedure produced a favourable outcome and the child had no signs of progression of the disease at two months post-op.⁴

Complications of FOP

The case report above illustrates some of the difficulties in managing patients with FOP. Since it is crucial to prevent exacerbations, one of the mainstays of proper care is to avoid the trauma associated with certain medical procedures. Surgeries in particular pose difficulties for patients if, as in the case report, fusion of the cervical vertebrae has occurred and impedes intubation. Anesthesia in an FOP patient can also be extremely difficult if ankylosis of the jaw is present, which could be triggered by something as simple as minor dental procedures.^{4,5} Biopsies are also contraindicated, but since they are the investigation of choice for most tumours, they are often performed before a diagnosis is made.

One of the most severe complications of FOP is cardiopulmonary compromise. FOP can lead to ankylosis of the costovertebral joints, ossification of the intercostal and paravertebral muscles, and progressive spinal deformity such as kyphoscoliosis or thoracic lordosis. Chest expansion is drastically limited, resulting in reduced vital capacity and restrictive lung disease. Right ventricular hypertrophy can also occur due to the thoracic insufficiency.^{5,7}

Patients with FOP are also more prone than the general population to kidney stones as well as conductive hearing loss due to fusion of the bones of the middle ear. They are also prone to fracture of heterotopic bone, which requires closed reduction and splinting, as well as analgesia, as in fracture of normotopic bone in any patient.⁵ Thus, patients should be educated about risk factors for falls to avoid precipitation of a cycle in which a fall leads to further ossification and joint ankylosis, which in turn increases the risk of future falls.^{5,6}

Discussion

FOP is an extremely debilitating disease. Most affected people are wheelchair-bound by their 20s and life expectancy is less than 40, as thoracic insufficiency usually occurs by this point.⁴ Patients must live with the constant knowledge that they have a progressive disease. As more and more joints and muscles become permanently ossified over time, patients with FOP are essentially trapped inside their own bodies until they can no longer move or even breathe.

The recent breakthrough discovery of the FOP gene has identified a specific target for pharmacologic therapy and allowed for the development of a DNA diagnostic test which expedites diagnosis and limits harmful interventions.² However, the opportunities for insight provided by studying the FOP gene extend far beyond this rare disorder alone. That is, characterization of the underlying mechanism causing HO in FOP has much broader implications for patients with more common forms of HO and for developing tissue engineering strategies for skeletal bone and cartilage repair.^{2,3}

Conclusions

Due to the rarity, variable severity and episodic clinical course of FOP, clinicians often fail to associate the rapidly developing axial soft tissue swellings with the distinctive malformed great toes.³ When such associations are not made, FOP is commonly mistaken for other conditions, delaying proper treatment.^{1, 3} Not surprisingly, misdiagnosed individuals often undergo a battery of unnecessary biopsies and tests that exacerbate progression of the condition. Early diagnosis and cautious management of patients with suspected

FOP are therefore very important. Finally, although the current medical management of FOP is largely supportive, the goal of research continues to be the development of treatments that will prevent, halt or even reverse progression of the disease.³

References

1. Kaplan FS, Shen Q, Lounev V, Seemann P, Groppe J, Katagiri T, Pignolo RJ, Shore EM. Skeletal metamorphosis in fibrodysplasia ossificans progressiva (FOP). *J Bone Miner Metab.* 2008;26(6):521-30.
2. Shore EM, Kaplan FS. Insights from a rare genetic disorder of extra-skeletal bone formation, fibrodysplasia ossificans progressiva (FOP). *Bone.* 2008 Sep;43(3):427-33.
3. Kaplan FS, Le Merrer M, Glaser DL, Pignolo RJ, Goldsby RE, Kitterman JA, Groppe J, Shore EM. Fibrodysplasia ossificans progressiva. *Best Pract Res Clin Rheumatol.* 2008 Mar;22(1):191-205.
4. Tumolo M, Moscatelli A, Silvestri G. Anaesthetic management of a child with fibrodysplasia ossificans progressiva. *Br J Anaesth.* 2006;97(5):701-3.
5. The International Clinical Consortium on FOP. The medical managements of fibrodysplasia ossificans progressiva: current treatment considerations. *Clin Proc Intl Clin Consort FOP.* 2008;3(2):1-85.
6. Lanchoney TF, Cohen RB, Rocke DM, Zasloff MA, Kaplan FS. Permanent heterotopic ossification at the injection site after diphtheria-tetanus-pertussis immunizations in children who have fibrodysplasia ossificans progressiva. *J Pediatr.* 1995;126:762-4.
7. Kussmaul WG, Esmail AN, Sagar Y, Ross J, Gregory S, Kaplan FS. Pulmonary and cardiac function in advanced fibrodysplasia ossificans progressiva. *Clin Orthop Relat Res.* 1998;346:104-9.

Quality of Life Consequences of Long QT Syndrome

Ishvinder Chattha (Meds 2011) and Caleb Zelenietz (Meds 2011)

Faculty Reviewers: Dr. Raymond Sy and Dr. Andrew Krahn

Long QT syndrome (LQTS) is a genetic condition characterized by abnormal repolarization of cardiac ventricular myocytes predisposing the individual to ventricular tachyarrhythmias. The first manifestation of LQTS is typically syncope and less frequently cardiac arrest or sudden cardiac death. This article examines the effect of LQTS on patients' quality of life. This includes the psychosocial and physical consequences that may result from both the diagnosis of a potentially lethal genetic disease and its subsequent management. Current guidelines are based on expert consensus and recommend moderate and at times strict exercise restriction, even in asymptomatic patients. Broad exercise restriction affects the social development of children and also removes the protective effect that exercise has against developing both psychological and physical health problems related to inactivity. Due to the potential decline in a patient's quality of life resulting from strict exercise restriction, future research should focus on determining the effectiveness of exercise restriction in the prevention of ventricular arrhythmias.

Introduction

Genetic disorders are unique in their impact on patients. Often incurable, patients not only need to deal with the wide array of feelings surrounding the initial diagnosis, but there is often guilt related to the possibility of passing the disorder onto their children.¹ Given the implications for family members, genetic testing and resultant therapy is offered to relatives. The diagnosis can also influence family planning. These consequences are in addition to the effects of the disease itself.

This is the reality for patients diagnosed with congenital long QT syndrome (LQTS). In addition to the difficulties related to the acceptance of a lifelong and incurable disorder, LQTS management imposes a potentially significant decline in quality of life. Severe exercise restriction is currently recommended in patients with LQTS to prevent symptoms from arising.² This sudden change in lifestyle can be especially devastating for children, as it can leave them feeling like outcasts (Figure 1). This article examines the impact of LQTS on the quality of life of patients and their families.

Long QT Syndrome

The QT interval, seen on the electrocardiogram, is a measure of ventricular depolarization and subsequent repolarization. LQTS is characterized by abnormally delayed repolarization of ventricular myocytes. It is caused by abnormalities of cardiac ion channels which result in abnormal amounts of sodium or potassium moving across the cell membrane, interfering with the normal action potential. At least 12 subtypes of congenital type long QT syndrome have been described resulting from mutations in any one of nearly a dozen genes controlling the expression, regulation, or assembly of cell membrane ion channels.³ Subtypes are classified based on the gene affected, with LQT1 (KVLQT1 mutation) accounting for approximately 50% of cases, LQT2 (HERG mutation) nearly 40%, and LQT3 (SCN5A) about 5%.³

Although most patients remain asymptomatic, the first manifestation of LQTS may be syncope, cardiac arrest or sudden cardiac death. Specific activities or triggers can precipitate cardiac events in LQTS patients – these include startling auditory stimuli such as an



Figure 1. Picture drawn by a young male diagnosed with LQTS. The patient experienced syncope during an ice hockey game which led to the subsequent diagnosis of LQTS. As a result of the diagnosis and resultant exercise restriction, the patient quit his ice hockey team and abandoned his dream of playing professional hockey. Patient identifiers have been removed from the drawing.

alarm clock or loud door bell, emotionally charged events, or even physical exertion as brief as running to catch the bus.⁴ Commonly prescribed medications including certain antibiotics, bronchodilators, hypertension medications, and antidepressants can cause further prolongation of the QT interval and increase the risk of cardiac arrhythmias in susceptible patients. While 50% of patients remain asymptomatic throughout their lifetime, the severity of the symptoms has created a focus on preventative medicine as an important aspect of management.

Quality of Life Issues – Impact on Patients and Families

The diagnosis of LQTS can be quite devastating to individuals. The implications that it has on longevity, mental state, and day to day life are immense. Farnsworth *et al.* (2006) conducted a series of interviews with parents with LQTS, and one patient's description was: "your whole life is shattered. You feel whammied off this earth."⁴ Another parent described the changes in her son after being diagnosed: "He is sports-oriented, social, and wants to fly for the Air Force. He can't play hockey now....He can't fly. He is on hold, hoping this will all go away."⁴

Sudden cardiac death is the first manifesting symptom in almost 12% of cases. Because of this, patients constantly live with the uncertainty of their own future as well as the

future of their loved ones in the context of a genetic disorder.⁴ Two thirds of parents interviewed by Farnsworth *et al.* (2006) expressed fear of their children dying. For example, one interviewee said, “When I walk into their rooms in the morning to wake them I am always aware in the back of my mind...will they be cold?”⁴

The management of unrelated medical conditions such as infections may also create anxiety because certain prescribed medications may prolong cardiac repolarisation as a side-effect. There are over 130 recognized drugs, ranging from anti-hypertensives, to antibiotics, heartburn aids, and antidepressants, that should be avoided by LQT patients as they have been shown to further prolong the QT interval, thus increasing the risk of a cardiac arrhythmia. Furthermore, cardiac events may be precipitated by dehydration and inadequate nutrition, which are commonly seen in many disease states. The fear of precipitating a cardiac event is always present: “Anytime she gets sick it is a big deal. It is a totally different ballgame. Medications are a big deal....We do flu shots every year. I am more worried, it is always in the back of your mind.”⁴

Quality of Life Issues – Impact of Treatment

Once diagnosed, a management strategy focusing on prevention of LQTS related cardiac events is implemented. Management is multimodal, combining lifestyle modification, medication or even surgical options. Lifelong beta blocker therapy is the mainstay of medical treatment for LQT1 and LQT2 subtypes.² Trigger avoidance is also important for successful prevention. The most common trigger is physical exertion. As a result, the American Heart Association guidelines recommend severe exercise restriction for LQT patients (Table 1). These restrictions may range from withdrawal from all competitive sports to students potentially sitting out of physical education class. Such broad exercise restriction may impair physical as well as psychological wellbeing.

Individuals that are physically active are less likely to suffer from mental health problems.

Physical activity improves self-esteem, can foster identification with peers, reduces stress, and is inversely related to adolescent depression.⁵ Exercise deprivation has been shown to lead to increased symptoms of pain, fatigue, cognitive problems, and negative mood.⁶ With children there is added danger because proper social development requires common identification with peers, and thus this targeted exclusion from team activities can have a significant negative psychological impact (Figure 1).⁷ The elderly are also at particular risk from exercise restriction as exercise has been shown to reduce CNS dysfunctions such as cognitive decline.⁸

With the rising prevalence of obesity, the physical health benefits of exercise have been given increased attention. Physical activity is unanimously recognized as an essential element in combating the obesity epidemic.⁹ Research has repeatedly shown regular exercise decreases lipid profiles, obesity, hypertension, and development of some cancers and cardiovascular diseases, and it increases bone mineral density. Physical activity is also used in the management of numerous chronic diseases.¹⁰

Physical activity rates are decreasing in Western populations. Numerous community interventions are being implemented to promote regular exercise to achieve the recognized benefits. The diagnosis of LQTS presents another road block in achieving a healthy lifestyle. Participation in competitive sports as a child, an activity shown to be predictive of physical activity levels in adulthood, is not permitted in patients with LQTS.⁵

Participation in sports is an enjoyable method of physical activity. Participants report enjoyment stemming from affiliation with peers, competitive excitement, gaining competency, and a source of psychosocial support.^{11,12} On the field people can learn sportsmanship, fair play, personal responsibility, and moral reasoning.¹³ Because LQTS patients are not permitted to participate in popular sports such as basketball, hockey, soccer and football, these key elements of personal development are potentially missed.

Table 1. Permissibility of common physical activities for LQTS patients²

Activity	Guidelines		
	Not Permitted	Maybe Permitted	Permitted
HIGH INTENSITY			
Basketball	√		
Body building	√		
Ice hockey	√		
Racquetball/squash		√	
Rock Climbing	√		
Running	√		
Skiing		√	
Soccer	√		
Tennis (singles)	√		
Touch football	√		
Windsurfing	√		
MODERATE INTENSITY			
Baseball/softball		√	
Biking			√
Motorcycling	√		
Jogging		√	
Sailing		√	
Surfing	√		
Swimming	√		
Treadmill			√
Weightlifting	√		
Hiking		√	
LOW INTENSITY			
Bowling			√
Golf			√
Horseback riding		√	
Scuba diving	√		
Skating			√
Snorkelling	√		
Brisk Walking			√

Conclusions

Long QT syndrome has a large impact on patients and their families on multiple levels. Physicians need to understand the psychological stress of parents facing the possibility of cardiac arrest or sudden cardiac death in their children. Physicians should educate patients and their families on the genotype-specific triggers and encourage them to take an active part in managing this disease. Patients also need to be aware that certain prescribed medications prolong the QT interval putting them at increased risk for cardiac events, and they should be instructed to screen all their future medications against a list of known QT-prolonging drugs (www.qtdrugs.org).

The American Heart Association (AHA) has published recommendations for the management of LQTS. They call for severe exercise restriction as a preventative measure. The premise is based on athletes not using proper judgement in extracting themselves immediately from the physical activity, even upon realization that medical attention may be needed. Due to the unique nature of sports, the AHA consensus statement asserts that cardiac symptoms such as palpitations, fatigue, and dizziness can incorrectly be attributed to the sensations which accompany intense exercise.²

The AHA recommendations are based on the experience and insights of a panel of experts and by their own admission, are not specifically from scientific evidence. With the negative psychological and physical health effects that exercise restriction can have on patients, it is important for future research to focus on this area, which might serve as a basis for validating or amending the current recommendations. Important areas of research include determining the effectiveness of exercise restriction on the prevention of cardiac events in each LQTS subtype, as well as classifying the degree of QT prolongation that occurs while participating in different sports. This will assist in developing more comprehensive and genotype specific recommendations which may limit the negative iatrogenic effects of LQTS management.

References

1. McAllister M, Davies L, Payne K, Nicholls S, Donnai D, MacLeod R. The emotional effects of genetic diseases: Implications for clinical genetics. *American Journal of Medical Genetics Part A*. 2007;143A(22):2651-2661.
2. Maron BJ, Chaitman BR, Ackerman MJ *et al*. Recommendations for physical activity and recreational sports participation for young patients with genetic cardiovascular diseases. *Circulation*. 2004;109(22):2807-2816.
3. Vohra J. The long QT syndrome. *Heart, Lung Circ*. 2007;16:S5-S12.
4. Farnsworth MM, Fosyth D, Haglund C, Ackerman MJ. When I go in to wake them ... I wonder: Parental perceptions about congenital long QT syndrome. *Journal of the American Academy of Nurse Practitioners*. 2006;18(6):284-290.
5. Hallal PC, Victora CG, Azevedo MR, Wells JCK. Adolescent physical activity and health - A systematic review. *Sports Medicine*. 2006;36(12):1019-1030.
6. Glass JM, Lyden AK, Byrne-Dugan CJ *et al*. Increased symptoms of pain, fatigue, cognitive problems and negative mood after exercise deprivation and sleep restriction are predicted by baseline autonomic and HPA function. *Arthritis and Rheumatism*. 2006;54(9):S828.
7. Ussher MH, Owen CG, Cook DG, Whincup PH. The relationship between physical activity, sedentary behaviour and psychological wellbeing among adolescents. *Social Psychiatry and Psychiatric Epidemiology*. 2007;42(10):851-856.
8. Adlard PA, Cotman CW. Voluntary exercise protects against stress-induced decreased in brain-derived neurotrophic factor protein expression. *Neuroscience*. 2004;124(4):985-992.
9. Duche P. Physical activity and infantile obesity: Tracking, prevention and treatment. *Science & Sports*. 2008;23(6):278-282.
10. Biddle SJH, Gorely T, Stensel DJ. Health-enhancing physical activity and sedentary behaviour in children and adolescents. *Journal of Sports Sciences*. 2004;22(8):679-701.
11. McCarthy PJ, Jones MV, Clark-Carter D. Understanding enjoyment in youth sport: A developmental perspective. *Psychology of Sport and Exercise*. 2008;9(2):142-156.
12. McCarthy PJ, Jones MV. A qualitative study of sport enjoyment in the sampling years. *Sport Psychologist*. 2007;21(4):400-416.
13. Bailey R. Physical education and sport in schools: A review of benefits and outcomes. *Journal of School Health*. 2006;76(8):397-401.

Genetic Susceptibility in Rheumatoid Arthritis

Kimberley Colangelo (Meds 2011) and Caleb Zelenietz (Meds 2011)

Faculty Reviewer: Dr. Janet Pope

Rheumatoid arthritis is a condition that causes chronic inflammatory arthritis. It has been linked to multiple etiologies; the cause is thought to be a combination of the environment, infection, hormones and a genetic component. The precise contribution of genetics to the development and course of rheumatoid arthritis is currently under investigation, and numerous gene candidates have been identified. The HLA-DR gene has been widely studied and numerous alleles have been identified that correlate to the development of rheumatoid arthritis. The role of genetics in rheumatoid arthritis is explored in this article.

Background

Rheumatoid arthritis (RA) is a chronic autoimmune disorder which causes inflammatory arthritis and occurs in approximately 1% of the population.¹ It frequently presents as peripheral polyarticular arthritis, is symmetrical, and often progresses with joint damage and loss of function. There may be periods of remission, although variations on this pattern occur. In addition, the extra-articular manifestations vary greatly among patients. Without treatment, it leads to significant deformities as cartilage and bone are destroyed, so called erosive disease.

The precise cause is unknown, but genetic predisposition, the environment, infections, and hormones (potential precipitators) are thought to all play a role. The genetic component has been the subject of much research in recent decades, as RA has been found to cluster in families and occurs more frequently in families with other autoimmune disorders. The heritability of RA is estimated to be approximately 60%, with a 12-15% concordance rate among twins.²

Several key genes have been targeted as they are implicated in the development of RA and the course of the disease. The group of human leukocyte antigen (HLA) genes on chromosome 6 has been at the forefront of genetic research in RA. Certain alleles have been associated with an increased risk of developing the disease and

predicting its severity, and others are involved in protection from RA. The HLA gene products are involved in immune function, particularly coding for antigens expressed on the surface of white blood cells. Many genes aside from the HLA group have been linked to RA.

Current Research

The HLA region in the human genome is one of the most heterogeneous and is frequently targeted for studies involving disease susceptibility. For RA, the HLA-DR family has been studied extensively. Several theories exist for how the HLA molecule could influence the immune system. HLA molecules bind peptides and present them to receptors on the CD4+ T lymphocytes, so the epitope associated with RA could determine which peptides bind successfully, the affinity with which they bind, the epitope itself could be antigenic, or the epitope could influence which T lymphocytes survive in the thymus (i.e. giving a propensity for autoreactive lymphocytes to survive).¹ In the majority of genetically-linked cases of RA, a shared epitope at amino acid positions 67-74 of HLA-DRB1 (the beta 1 subunit of the HLA-DR molecule) has been found. HLA-DR4, one of the most well studied, seems to be associated with both higher risk of disease and worse prognosis, particularly certain haplotypes within HLA-DR4 such as 0401.³

Interestingly, some HLA-DR alleles are associated with protection from RA, even counteracting the increased susceptibility when the sister allele was associated with an increased RA risk.⁴

These protective HLA-DR alleles have a different conserved sequence than the so-called shared epitope associated with an increased risk. A well known example is HLA-DRB1 with the conserved epitope DERA at positions 70-74 of the amino acid sequence. This allele was not only correlated with a lesser risk of developing RA, but was also linked to a less severe form in people with RA.⁵

The presence of the HLA alleles with the shared epitope has also been shown to have a high concurrence with the presence of anti-cyclic citrullinated peptides antibodies (anti-CCP), which are used in diagnosing RA. One theory is that the citrullinated peptide binds the HLA shared epitope resulting in activation of T lymphocytes. This may in turn lead to increased autoantibody production that results in the formation of anti-CCP antibodies.⁶ Anti-CCP is a stronger predictor of radiographic damage in early inflammatory arthritis than Rheumatoid Factor (RF) and is especially useful as a marker in those with negative RF who have RA. However, anti-CCP is not sufficiently sensitive to identify all those who will have radiological damage in early inflammatory arthritis.

Some important discoveries related to the HLA susceptibility region include⁷:

- Having two copies of a susceptibility allele makes a person even more likely to develop RA compared to having only one copy
- Having one or two copies of a susceptibility allele makes a person more likely to have severe disease
- The combination of alleles present alters the relative risk (i.e DR4/DR1 vs. DR4/X)
- Presence of the allele may also influence if a person will have extra-articular manifestations
- The timeline of progression of RA is influenced by the alleles present

The HLA-DR genes cannot be the only genetic contribution though, as not all RA patients have the implicated alleles and there is still a wide clinical heterogeneity amongst those that do. It has been estimated through sibship studies that HLA linked genes contribute about 30-40% of the genetic component in RA.⁸ Numerous other genes have been identified, and the more popular ones are summarized in Table 1.

The products of these genes are proteins or molecules that are involved in the immune response; as an intracellular signalling molecule, in the formation of antibodies, or in carrier molecules. A physiologic mechanism through which they contribute to the development of RA has been postulated for each one. Currently, screening of whole genomes of families where RA is prevalent is being undertaken to possibly discover other genes and better characterize the genes that have been discovered.

Table 1. Summary of genes that confer susceptibility to rheumatoid arthritis

Gene	Product
HLA-DR ⁹	Encodes cell surface antigens that present proteins to the T lymphocytes
STAT4 ¹⁰	Encodes a transcription factor for signals from certain cytokines
PAD14 ¹¹	Encodes an enzyme that catalyzes peptidyl arginine to peptidyl citrulline
N22/PTPN22 ¹¹	Encodes intracellular tyrosine phosphatase in T and B cells
SLC22A4 ¹²	Encodes for an organic cation transporter
FCRL3 ¹³	Encodes a B-lymphocyte specific membrane molecule
CTLA4/CD152 ¹	Encodes a protein produced by activated T cells

The presence and implications of these genes vary widely by ethnic population with respect to rheumatoid arthritis. The prevalence of RA itself differs by ethnic group, for example Native Americans have been reported to have a prevalence as high as 2% whereas Asian populations as low as 0.3%.¹⁴ In addition, the polymorphisms of the human genome sequence in the alleles that are related to RA vary by ethnic populations, with higher concordance amongst more closely related populations.¹⁵

Relevance

The key question regarding this research is whether it is limited to being an expensive research tool or if it will become relevant to patients and physicians. Given that many of these alleles are common in the general population, gene screening will not be useful as a screening tool to discover asymptomatic patients that will develop RA. However, since HLA alleles remain constant over a lifetime, typing may be useful as a predictor of the course of disease in patients already diagnosed with RA, it can be used to decide which patients will have more severe disease and who will progress quickly so that aggressive therapy can be initiated. Whether this will change outcomes needs to be studied. For now, its value seems limited to research where family members at risk for RA are being prospectively studied, particularly those who have already developed anti-CCP or RF.

Conclusions

It cannot yet be said what interaction and combination of alleles is necessary for the development of RA and for the specific phenotypes that are expressed. RF testing is done to determine who with early inflammatory arthritis will develop erosions, but combining RF and HLA may increase the sensitivity and specificity so that early aggressive treatment can be implemented. Since anti-CCP antibodies may have higher specificity for the development of RA, the combination of these two components could possibly increase the ability to predict the

development and course of progression of the disease.

References

1. Harney S, and Woodworth BP. Genetic epidemiology of rheumatoid arthritis. *Tissue Antigens*. 2002 Dec;60(6):465-73.
2. MacGregor AJ, *et al*. Characterizing the quantitative genetic contribution to rheumatoid arthritis using data from twins. *Arthritis Rheum*. 2000 Jan;43(1):30-7.
3. Taneja V, *et al*. Delineating the role of the HLA-DR4 "shared epitope" in susceptibility versus resistance to develop arthritis. 2008 Aug;181(4):2869-77.
4. van der Helm-van Mil AH, Huizinga TW, Schreuder GM, Breedveld FC, de Vries RR, Toes RE. An independent role of protective HLA class II alleles in rheumatoid arthritis severity and susceptibility. *Arthritis Rheum*. 2005 Sep;52(2):2637-44.
5. Carrier N, *et al*. The DERA HLA-DR alleles in patients with early polyarthritis. *Arthritis Rheum*. 2009 Mar;60(3):698-707.
6. Auger I, *et al*. Influence of HLA-DR genes on the production of rheumatoid arthritis-specific autoantibodies to citrullinated fibrinogen. *Arthritis Rheum*. 2005 Nov;52(11):3424-32.
7. Olsen NJ, *et al*. Associations of HLA-DR4 with rheumatoid factor and radiographic severity in rheumatoid arthritis. *Am J Med*. 1988 Feb;84(2):257-64.
8. Deighton CM, Kelly PJ, Walker DJ. Linkage of rheumatoid arthritis with HLA. *Ann Rheum Dis*. 1993 Sep;52(9):638-42.
9. Fries JF, *et al*. HLA-DRB1 genotype associations in 793 white patients from a rheumatoid arthritis inception cohort: frequency, severity, and treatment bias. *Arthritis Rheum*. 2002 Sep;46(9):2320-9.
10. Remmers EF, *et al*. Stat4 and the risk of rheumatoid arthritis and systemic lupus erythematosus. *N Engl J Med*. 2007 Sep;357(10):977-86.
11. Plenge RM, *et al*. Replication of putative candidate-gene associations with rheumatoid arthritis in >4000 samples from North America and Sweden: association of susceptibility with PTPN22, CTLA4, and PAD14. *Am J Hum Genet*. 2005 Dec;77(6):1044-60.
12. Tokuhira S, *et al*. An intronic SNP in a RUNX1 binding site of SLC22A4, encoding an organic

cation transporter, is associated with rheumatoid arthritis. Nat Genet. 2003 Dec;35(4):341-8.

13. Kochi Y, et al. A functional variant in FCRL3, encoding Fc receptor-like 3, is associated with rheumatoid arthritis and several autoimmunities. 2005 May;37(5):478-85.

14. Yamada R, Yamamoto K. Mechanisms of disease: genetics of rheumatoid arthritis – ethnic differences in disease-associated genes. Nat Clin Pract Rheumatol. 2007 Nov;3(11):644-50.
15. Mori M, et al. Ethnic differences in allele frequency of autoimmune-disease-associated SNPs. J Hum Genet. 2005 May;50(5):264-6.

ᐃᐅᑦᑲᑎᑦᑎᐅᑦᑲ



JOIN US IN NUNAVUT FAMILY PRACTITIONERS



This is your opportunity to begin your medical career in Nunavut in a collaborative family practice. Experience the rich Inuit culture, breathtaking scenery, and a change of pace

One of our regular physicians describes the practice like this: *"I chose to start my career as a family physician in Iqaluit after a rotation here as a resident. I was attracted to the opportunity to practice full service family medicine. I've expanded my clinical skills, learned about Inuit culture and worked with a supportive group of physicians from across the country".*

The Government of Nunavut offers competitive rates and travel expenses. Short or long term contract available

For more information,
Please contact Physician Services:

877.979.7343

email: medicine@gov.nu.ca



CARE CLOSER TO HOME

ᐃᐅᑦᑲᑎᑦᑎᐅᑦᑲ

www.nunavut-physicians.gov.nu.ca

Nuchal Translucency and Prenatal Diagnosis of Congenital Heart Disease: Progress and Future Directions

Xiangning Fan (*Meds 2010*)

Faculty Reviewer: Dr. Eva Welisch

Nuchal translucency thickness is an important component of prenatal screening programs, which exist to help quantify the risk of certain chromosomal and anatomical abnormalities in a fetus *in utero*. Abnormally increased nuchal translucency thickness has been used as a soft marker for Down's Syndrome and other aneuploidies for more than a decade. Recently, an association has been identified between increased nuchal translucency thickness and congenital heart disease, raising the possibility that nuchal translucency thickness may help identify affected fetuses as early as 11-14 weeks gestation. However, the pathophysiological mechanism which produces an abnormal nuchal translucency thickness in the fetus is not well understood and an abnormal measurement is not specifically associated with any one type of lesion. Therefore, nuchal translucency thickness cannot be used for diagnosis of congenital heart disease *in utero*, but may be very helpful in identifying fetuses at risk, which may help improve diagnostic accuracy and direct resource-efficient use of diagnostic methods such as fetal echocardiography.

Introduction

Although it has long been the holy grail of prenatal care to be able to assure every woman a healthy infant at term, 2-3% of infants are born with at least one anatomical deformity. The congenital heart diseases are collectively the most common congenital defects, with a combined incidence estimated at approximately 1/100 at term, and higher in the prenatal population.¹ Cardiac lesions may occur in isolation, or alternately as part of a genetic syndrome. Even in isolated cases, there appears to be a genetic, likely multifactorial, component to their etiology. They range from incidental findings of no great functional significance (e.g. small atrial septal defects) to lesions which, if left untreated, are immediately life-threatening to the neonate (e.g. transposition of the great arteries). Structural heart defects are thought to contribute to up to 20% of all neonatal deaths and up to 50% of all infant deaths.²

Recently, attention has focused on the utility of ultrasound measurement of the free fluid behind the neck of the fetus—termed the “nuchal

translucency” (NT)—in predicting congenital heart disease in the fetus. It has been suggested that the subset of fetuses with abnormal NT measurements is at increased risk for congenital heart defects; therefore the NT thickness may be of use in prenatal diagnosis of congenital heart disease.

What is Nuchal Translucency?

The NT measurement is a measurement of the thickness of the free fluid layer behind the fetal neck. This is measured by ultrasonography between 11 and 14 weeks gestation. There are strict criteria to establish an ultrasound image as adequate for the assessment of nuchal translucency and the normal limit to the measurement is dependent upon gestational age. The NT measurement was first suggested as a soft marker for fetal aneuploidy by Nicolaides and colleagues in 1994, and this association was later confirmed by other researchers.³⁻⁶ It is now known that the nuchal translucency measurement is 77% sensitive and 95% specific for Down's Syndrome/Trisomy 21, the most common of the

three autosomal aneuploidies that are compatible with life.⁴

However, the NT measurement is also associated with a host of other abnormalities in the fetus. Even in chromosomally normal fetuses, increasing NT is associated with adverse fetal outcomes.^{7,8} Bilardo and colleagues report that up to 20% of chromosomally normal fetuses with increased NT experienced an adverse outcome, defined as miscarriage, intrauterine death, parental choice to terminate the pregnancy, or one or more structural or genetic disorders in the fetus. Of the 86 adverse outcomes, 14 (16%) were pregnancy terminations. The likelihood of adverse outcome appeared to correlate with increasing degrees of NT abnormality.⁸

Nuchal Translucency and Congenital Heart Disease

Recently, in both euploid⁹⁻¹² and aneuploid¹³⁻¹⁶ fetuses, an abnormally elevated NT measurement has been described in association with congenital heart disease in the fetus. There is increasing risk of cardiac lesions as the NT becomes progressively more abnormal.¹² Ghi and colleagues report the overall incidence of major cardiac defects to be 4.5% in fetuses with NT above 2.5 mm, with 7% incidence in fetuses with nuchal translucency thickness equal to or greater than 3.5 mm.¹² They suggest that abnormal NT may be more of a risk factor for major congenital cardiac lesions than maternal diabetes, positive family history of congenital heart disease or exposure to teratogenic agents, and that therefore fetal echocardiography is indicated for all fetuses with elevated NT measurements. At a NT thickness at or above 3.5 mm (roughly the 99th percentile), major cardiac anomalies appear to be present in 5-10% of screen-positive fetuses.^{12,17-19} Hyett and colleagues¹¹ propose a cutoff of between the 95th-99th percentiles as the optimum point to initiate referral for fetal echocardiography. Hyett suggests the nuchal translucency measurement could contribute to the detection of approximately 30% of all cases of congenital heart defects.¹⁹

Previously, Hyett and colleagues¹³ have suggested that the nuchal translucency measurement may be related to failure of the fetal heart rather than to the direct effects of aneuploidy. As cardiac lesions are seen in many of the genetic syndromes, this is certainly a plausible explanation for the close association seen between increased nuchal translucency, aneuploid conditions and congenital heart disease. However, this interpretation is by no means unanimously accepted, and multiple authors^{20,21} report that the degree of NT abnormality is not specifically associated with any particular heart lesion. Makrydimas and colleagues²¹ report that there is considerable overlap between the average NT measurement for different cardiac lesions and the identity of the lesion; thus, the NT measurement is not helpful for the specific diagnosis of fetal congenital heart disease except to indicate that a cardiac lesion may be present. The underlying pathophysiology remains unclear, and alternate explanations for the elevated NT in cases of congenital heart disease include venous congestion in the head and neck of the fetus, abnormalities of lymphatic drainage, altered distribution of subcutaneous tissue, glycosaminoglycan and proteoglycan accumulation, fetal anemia and/or decreased fetal movement.^{22,23} Therefore, the NT may indeed identify fetuses who are at increased risk of congenital heart defects so that these can be referred on for more specialized diagnostic testing (e.g. fetal echocardiography). However, in isolation, it does not appear to be sensitive or specific enough to be a diagnostic test for congenital heart disease.¹⁷

Mol²⁴ has suggested that relying on nuchal translucency to identify fetuses with Down's Syndrome may in fact preferentially identify fetuses in whom there is an associated cardiac defect. Given the relative incidence of congenital heart disease (1/100) and Down's syndrome (1/800-1000 overall)^{25,26}, the NT measurement may in fact pick up more babies with congenital heart disease who incidentally have Down's syndrome than babies with Down's syndrome who have an associated cardiac defect.

The clinical implications of this finding are less clear. It could be argued that, even if increased NT is a marker for congenital heart disease in the fetus, it is not useful clinically as it is a standard of care to offer all pregnant women (regardless of NT measurement) a morphological ultrasound scan at 18-20 weeks during which structural cardiac lesions can be identified. The current recommendation is a four-chamber view of the heart and imaging of the ventricular outflow tracts in an attempt to identify major anomalies.²⁷ However, it has been previously noted in population studies of prenatal patients that morphological echocardiography is dependent upon the expertise of the ultrasonographer and consequently an inexperienced one may miss a significant proportion of major cardiac defects.^{28,29} In their study, Hyett and colleagues¹¹ report abnormal NT measurement in 55% of fetuses with major congenital heart disease, compared to an abnormal four chamber view on prenatal ultrasound in 26% of those same babies. The 26% detection rate in that study was consistent with prior estimates.²⁸ Wald and colleagues have suggested, based on a meta-analysis of studies of the utility of nuchal translucency measurements and congenital heart disease, a detection rate of 52% and a false-positive rate of 5% using a screening cutoff of 1.7 multiples of the median.³⁰ This is greater than that achieved using a four chamber view alone, and, therefore, the nuchal translucency shows great promise as a potential screening tool for congenital heart disease which can help direct appropriate and resource-efficient use of fetal echocardiography.

Summary

Nuchal translucency is typically measured at 11-14 weeks as part of prenatal screening programs in order to help estimate maternal risk of bearing a child with aneuploidy or neural tube defects. In recent years, an association has been demonstrated between abnormally elevated nuchal translucency and a group of structural, genetic and chromosomal abnormalities in the developing fetus, including the congenital heart diseases, as well as poorer pregnancy outcomes for the fetus, and this is seen even in fetuses with

normal chromosomes. Routine measurement of the nuchal translucency shows great promise in helping to identify fetuses at increased risk of congenital heart disease, and therefore may help direct resource-efficient use of fetal echocardiography. However, by itself, increased nuchal translucency does not predict the diagnosis of a specific type of cardiac defect in the fetus.

References

1. Hoffman JJ. Incidence of congenital heart disease: II. Prenatal incidence. *Pediatr Cardiol.* 1995;16:155-165.
2. Yogel S, Cohen SM, Achiron R. Examination of the fetal heart by five short-axis views: a proposed screening method for comprehensive cardiac evaluation. *Ultrasound Obstet Gynecol.* 2001;17:367-369.
3. Nicolaides KH, Brizot ML, Snijders RJM. Fetal nuchal translucency: ultrasound screening for fetal trisomy in the first trimester of pregnancy. *Br J Obstet Gynaecol.* 1994;101:782-786.
4. Pandya PP, Snijders RJM, Johnson SP, De Lourdes Brizot M, Nicolaides KH. Screening for fetal trisomies by maternal age and fetal nuchal translucency thickness at 10-14 weeks of gestation. *Br J Obstet Gynaecol.* 1995;102:957-962.
5. Hackshaw AK, Wald NJ, Haddow JE. Down's syndrome screening with nuchal translucency. *Lancet.* 1996;348:1740.
6. Snijders RJM, Noble P, Sebire N, Souka A, Nicolaides KH. UK multicentre project on the assessment of risk of trisomy 21 by maternal age and fetal nuchal-translucency thickness at 10-14 weeks of gestation. *Lancet.* 1998;351:343-346.
7. Souka AP, Krampfl E, Bakalis S, Heath V, Nicolaides KH. Outcome of pregnancy in chromosomally normal fetuses with increased nuchal translucency in the first trimester. *Ultrasound Obstet Gynecol.* 2001;18:9-17.
8. Bilardo CM, Muller MA, Pajkrt E, Clur SA, van Zalen MM, Bijlsma EK. Increased nuchal translucency thickness and normal karyotype: time for parental reassurance. *Ultrasound Obstet Gynecol.* 2007;30:11-18.
9. Hyett JA, Moscoso G, Papapanagiotou G, Perdu M, Nicolaides KH. Abnormalities of the heart and great arteries in chromosomally normal fetuses with increased nuchal translucency thickness at 11-13 weeks of gestation. *Ultrasound Obstet Gynecol.* 1996;7:245-250.

10. Hafner E, Schuchter K, Liebhart E, Philipp K. Results of routine fetal nuchal translucency measurement at weeks 10-13 in 4233 unselected pregnant women. *Prenat Diagn.* 1998;18:29-34.
11. Hyett J, Perdu M, Sharland G, Snijders R, Nicolaides KH. Using fetal nuchal translucency to screen for major congenital cardiac defects at 10-14 weeks of gestation: a population based cohort study. *BMJ.* 1999;318:81-85.
12. Ghi T, Huggon IC, Zosmer N, Nicolaides KH. Incidence of major structural cardiac defects associated with increased nuchal translucency but normal karyotype. *Ultrasound Obstet Gynecol.* 2002;18:610-614.
13. Hyett J, Moscoso G, Nicolaides KH. Increased nuchal translucency in trisomy 21 fetuses: relationship to narrowing of the aortic isthmus. *Hum Reprod.* 1995;10:3049-3051.
14. Hyett JA, Moscoso G, Nicolaides KH. Cardiac defects in 1st-trimester fetuses with trisomy 18. *Fetal Diagn Ther.* 1995;10:381-386.
15. Hyett JA, Moscoso G, Nicolaides KH. First trimester nuchal translucency and cardiac defects in fetuses with trisomy 21. *Am J Obstet Gynecol.* 1995;172:1411-1413.
16. Hyett J, Moscoso G, Nicolaides KH. Abnormalities of the heart and great arteries in first trimester chromosomally abnormal fetuses. *Am J Med Genet.* 1997;172:1411-1413.
17. Macrides E, Cobian-Sanchez F, Tekay A, Moscoso G, Campbell S, Thilaganathan B, Carvalho JS. Limitations of using first-trimester nuchal translucency measurements in routine screening for major congenital heart defects. *Ultrasound Obstet Gynecol.* 2001;17:106-110.
18. Galindo A, Comas C, Martinez JM, Gutierrez-Larraya F, Carrera JM, Puerto B, Borrell A, Mortera C, de la Fuente P. Cardiac defects in chromosomally normal fetuses with increased nuchal translucency at 10-14 weeks of gestation. *J Matern Fetal Neonatal Med.* 2003;13:163-170.
19. Hyett JA. Does nuchal translucency have a role in fetal cardiac screening? *Prenat Diagn.* 2004;24:1130-1135.
20. Simpson JM, Sharland GK. Nuchal translucency and congenital heart disease: heart failure or not? *Ultrasound Obstet Gynecol.* 2000;16:30-26.
21. Makrydimas G, Sotiriadis A, Huggon IC, Simpson J, Sharland G, Carvalho JS, Daubeney PE, Ioannidis JPA. Nuchal translucency and fetal cardiac defects: a pooled analysis of major fetal echocardiography centers. *Am J Obstet Gynecol.* 2005;192:89-95.
22. Nicolaides KH, Heath V, Cicero S. Increased fetal nuchal translucency at 11-14 weeks. *Prenat Diagn.* 2002;22:308-15.
23. von Kaisenberg CS, Prols F, Nicolaides KH, Maass N, Meinhold-Heerlein I, Brand-Saberi B. Glycosaminoglycans and proteoglycans in the skin of aneuploid fetuses with increased nuchal translucency. *Human Reprod.* 2003;18:2544-2561.
24. Mol BWJ. Down's syndrome, cardiac anomalies and nuchal translucency. *BMJ.* 1999;318:70-71.
25. Weijerman ME, van Furth AM, Vonk Noordegraaf A, van Wouwe JP, Broers CJ, Gemke RJ. Prevalence, neonatal characteristics and first-year mortality of Down syndrome: a national study. *J Pediatr.* 2008;152:15-19.
26. Collins VR, Muggli EE, Riley M, Palma S, Halliday JL. Is Down syndrome a disappearing birth defect? *J Pediatr.* 2008;152:20-24.
27. ACOG Practice Bulletin No. 98. Ultrasonography in pregnancy. *Obstet Gynecol.* 2008;112:951-961.
28. Tegnander E, Eik-Ness SH, Johansen OJ, Linker DT. Prenatal detection of heart defects at the routine fetal examination at 18 weeks in a non-selected population. *Ultrasound Obstet Gynecol.* 1995;5:372-380.
29. Chew C, Halliday JL, Riley MM, Penny DJ. Population-based study of antenatal detection of congenital heart disease by ultrasound examination. *Ultrasound Obstet Gynecol.* 2007;29:619-624.
30. Wald NJ, Morris JK, Walker K, Simpson JM. Prenatal screening for serious congenital heart defects using nuchal translucency: a meta-analysis. *Prenat Diagn.* 2008;28:1094-1104.

The Legacy of a Life Lived: Trans-Generational Epigenetics and Cancer Development

Mark Kirchhof (Meds 2009)

Faculty Reviewer: Department of Pathology, UWO

Your grandfather smoked everyday, your mother was stressed while pregnant, your father worked in the petrochemical industry, should any of this matter to you? Evidence is emerging that the lives we live affect us far beyond the boundaries of our own body and can reach well into the lives of future generations. These lingering traces of lives lived manifest themselves in changes to the DNA of our cells, commonly referred to as epigenetics. Epigenetics is defined as inheritable changes in gene expression that don't alter the actual DNA sequence itself, most commonly mediated by DNA methylation and/or histone methylation or acetylation. In this paper, we will discuss some of the epigenetic changes that have been associated with the development of disease with specific focus on the development of cancer. Moreover, we will look at environmental exposures in current generations that may affect cancer development in future generations. We may merely be guardians of our DNA for future generations, and perhaps the impact of the way we treat our bodies should now be examined in this new light.

My parents are children of World War II. They grew up amongst the bombings, death and destruction. Their lives were forever changed by these experiences at a level far deeper than one would expect. Beyond the psychological, emotional or physical impact of those experiences, those moments would forever change their DNA. Such are the findings of Dr. Yehuda, who found that traumatic events can permanently alter the expression of the stress hormone, cortisol.¹ Yehuda found that children of mothers who suffered psychological trauma from the events of WWII had children who had lower levels of cortisol production.² This change in the offspring related to maternal behaviour or experiences has been linked to changes within the DNA of the children. These changes are referred to as epigenetics, which is defined as inheritable changes in gene expression that don't alter the actual DNA sequence itself.³ In this article, we will explore the relationship between the environment, epigenetics and cancer.

There are two major modalities by which DNA can be altered to change the expression of a particular gene: the methylation of DNA and

modification of histones. The methylation of DNA involves the addition of methyl groups to the cytosine bases in the CpG domains.^{4,5} The modification of histones is the other major epigenetic regulatory mechanism. The more tightly the DNA is wrapped around the histone complex, the lower the expression of the genes coded on that piece of DNA. Histone modification involves the methylation or acetylation of amino acids such as lysine, arginine and serine. Histone acetylation generally results in the loosening of the association between the DNA and the histone, allowing for increased gene expression.^{6,7} The effects of histone methylation are more variable and can result in the increase or decrease in gene expression depending on which residues are modified and where within the histone protein the modification takes place.^{6,7}

Epigenetic changes have long been associated with cancer. The first major finding was that the DNA of cancerous cells was globally hypomethylated.⁸ In addition, the degree of hypomethylation of the DNA seemed to correlate with the aggressiveness and prognosis of a particular cancer.^{4,9} There is some debate as to the

effects of DNA hypomethylation, but several theories have garnered attention. These include the idea that hypomethylated DNA is more unstable and thus leads to deletions and translocations, potentially creating truncated genes or genes that have different promoter elements.¹⁰ Other theories are based on the observation that imprinted DNA, that is selective DNA methylation to inactivate the expression of either a maternal or paternal allele, is disrupted in cancer cells.^{3,11} Therefore, hypomethylated DNA may correspond to the disruption of genomic imprinting. As an example, it is known that the loss of imprinting of the insulin-like growth factor gene is a risk factor for colorectal cancer.¹²⁻¹⁴

The other major epigenetic mechanism that has been found to be prevalent in cancer cells is the inactivation of tumour-suppressor genes.⁶ Tumour-suppressor genes prevent the progression of cancerous cells and some of their functions have been related to the inhibition of cell division, the initiation of apoptosis and the mediation of cell adhesion.⁴⁻⁶ Hypermethylation in the promoter regions of tumour-suppressor genes is one of the major steps required for the transformation from benign to malignant. Two well known genes, the retinoblastoma tumour-suppressor gene (Rb) and the breast cancer susceptibility gene 1 (BRCA1) are examples of genes that have hypermethylated promoter regions in cancerous cells.^{15,16} The inactivation of tumour-suppressor genes by hypoacetylation and hypermethylation of histones have been associated with colon cancer, prostate cancer, liver and breast cancer.^{6,12,17}

The question now concerns *why* these changes arise and how they result in cancer. We are all familiar with the concepts of nature and nurture intersecting to result in the genetic changes required to initiate the transformation into cancerous cells. For example, lung cancer requires a genetic predisposition on top of an environmental factor, commonly smoking. Research has shown that exposure to cigarette smoke can induce the hypermethylation of BRCA1/BRCA2 and XRCC, all tumour-suppressor genes, as well as the hypomethylation of the pro-metastatic oncogene, SNCG.^{18,19} Other

examples of environmental agents that induce changes to the epigenome resulting in increased incidence of cancer include aniline dyes and bladder cancer, solar UV radiation and skin cancer, and aflatoxins found in the soil and on plants and liver cancer.²⁰⁻²² We are all aware of this direct link between an environmental factor and the development of cancer. But what of the offspring of a person exposed to these environmental agents, are they ultimately at risk as well?

Exposure to environmental toxins has been shown to increase the transgenerational risk of diseases, including cancer. One of the most widely studied toxins is vinclozolin, which is an anti-androgenic compound used as a fungicide in the fruit industry.²³ Transient exposure of gestating rats to vinclozolin leads to a variety of diseases in offspring up to four generations later, including tumour development, prostate disease, kidney disease, immune abnormalities and defects in sperm formation.^{23,24} Bisphenol A is perhaps the most well known endocrine disruptor believed to change the epigenome. Bisphenol A is an organic compound that, until recently, could be found in many baby bottles, baby and children's toys, and in the epoxy resins that coat almost every food and beverage can.^{20,22,24} Neonatal exposure to bisphenol A has been shown to alter the methylation status of a variety of genes and result in cellular transformations that promote the development of prostate cancer.²⁵ Prenatal exposure to bisphenol A has been linked with an increased risk of breast cancer via hypomethylation.²⁶ While the temporal effects of bisphenol A have been shown in the first generation, further studies are required to determine if changes are transmitted to subsequent generations. Other links have emerged between environmental toxins and cancer including the transgenerational effects of alcohol leading to colon cancer and benzene leading to leukemia.^{27,28} As the research into epigenetics grows, we will undoubtedly continue to see an expansion of the environmental agents that can alter the epigenome.

The relationship between nutrition, epigenetics and the disease processes warrants

further consideration. One of first multi-generational epigenetic studies was done in Sweden, where parish registries and harvest records were used to link disease in generations after feasts or famines.^{29,30} These studies found that if a paternal grandfather experienced a surplus of food before the onset of puberty, the grandchild had a four-fold increased chance of dying from complications of diabetes.^{29,30} Interestingly, the children of mothers who experienced abundance of food before the onset of puberty had children who were protected from diseases related to diabetes.^{29,30} In terms of cancer research, evidence is emerging that links nutrition, epigenome status and cancer development. Excessive maternal weight gain during pregnancy leads to increased risks of breast cancer in the child.³¹ In addition, the nutritional content of the diet has been shown to be important in developing and maintaining the epigenome. Specifically, the availability of methyl donors, including folate, choline and methionine seem to be important in preventing disease.^{32,33} In rat models, decreased availability of methyl donors has been shown to induce the development of liver cancer, even in the absence of any known carcinogens.³³ In the agouti mouse models, dietary supplementation *in utero* with folic acid, vitamin B12, choline, betaine and zinc was associated with a lower risk of cancer, diabetes and obesity and an increased life expectancy.^{34,35} Low protein diets in utero also seem to affect DNA methylation and disease in offspring, leading to increased susceptibility to diabetes, hypertension and cancer.^{31,36}

Since epigenetic changes to the DNA are reversible, it provides the perfect target for cancer therapies. Already, we are seeing the emergence of therapies that are based on altering DNA methylation and histone acetylation.³⁷ In particular, *in vitro* evidence using DNA demethylating drugs and the ability to re-express tumour suppressor genes has led to the development of clinical therapies. Two DNA demethylating drugs, 5-azacytidine (Vidaza) and 5-aza-2'-deoxycytidine (Decitabine), have been approved for the treatment of myelodysplastic syndrome and leukemia.³⁸⁻⁴⁰ The other major class of drugs developed on the principles of

epigenetics is the histone deacetylase (HDAC) inhibitors. These drugs have been found to induce cell-cycle arrest and apoptosis *in vitro*.^{4,37,41,42} Suberoylanilide hydroxamic acid (Vorinostat) is the first HDAC inhibitor approved for the treatment of cancer, specifically cutaneous T-cell lymphoma.^{41,42} While these drugs have been approved, they lack specificity for cell type which may lead to unintended effects, including the expression of previously silenced oncogenes. As with any cancer therapeutic, the eventual goal will be to develop agents that target only cancerous cells, sparing normal tissue and cells.

If the last century belonged to DNA and the genome, this century will surely belong to epigenetics and the epigenome. We are slowly discovering that the epigenome is an important player in the development of disease. What has come as a surprise is the degree to which the epigenome is influenced by the environment, by everything from drugs, toxins to vitamins and caloric intake. Even more astonishing is the emerging data indicating that what we experience in our lives will ultimately influence the lives of our children and perhaps even their children. In effect, we are the guardians of our genes and need to watch over them so that we may pass them on in the best condition we can.

References

1. Yehuda R, Bierer LM. Transgenerational transmission of cortisol and PTSD risk. *Prog Brain Res.* 2008;167:121-35.
2. Yehuda R, Bell A, Bierer LM, Schmeidler J. Maternal, not paternal, PTSD is related to increased risk for PTSD in offspring of Holocaust survivors. *J.Psychiatr.Res.* 2008.
3. Jiang YH, Bressler J, Beaudet AL. Epigenetics and human disease. *Annu Rev Genomics Hum Genet.* 2004;5:479-510.
4. Esteller M. Epigenetics in cancer. *N Engl J Med.* 2008;358:1148-59.
5. Esteller M. CpG island hypermethylation and tumor suppressor genes: a booming present, a brighter future. *Oncogene.* 2002;21:5427-40.
6. Bachman KE, Park BH, Rhee I, Rajagopalan H, Herman JG, Baylin SB *et al.* Histone modifications and silencing prior to DNA

- methylation of a tumor suppressor gene. *Cancer Cell*. 2003;3:89-95.
7. Kouzarides T. Chromatin modifications and their function. *Cell*. 2007;128:693-705.
 8. Feinberg AP, Vogelstein B. Hypomethylation distinguishes genes of some human cancers from their normal counterparts. *Nature* 1983;301:89-92.
 9. Herranz M, Esteller M. DNA methylation and histone modifications in patients with cancer: potential prognostic and therapeutic targets. *Methods Mol Biol*. 2007;361:25-62.
 10. Eden A, Gaudet F, Waghmare A, Jaenisch R. Chromosomal instability and tumors promoted by DNA hypomethylation. *Science* 2003;300:455.
 11. Feinberg AP, Tycko B. The history of cancer epigenetics. *Nat Rev Cancer*. 2004;4:143-53.
 12. Cui H, Cruz-Correa M, Giardiello FM, Hutcheon DF, Kafonek DR, Brandenburg S et al. Loss of IGF2 imprinting: a potential marker of colorectal cancer risk. *Science*. 2003;299:1753-5.
 13. Kaneda A, Feinberg AP. Loss of imprinting of IGF2: a common epigenetic modifier of intestinal tumor risk. *Cancer Res*. 2005;65:11236-40.
 14. Cruz-Correa M, Cui H, Giardiello FM, Powe NR, Hyland L, Robinson A et al. Loss of imprinting of insulin growth factor II gene: a potential heritable biomarker for colon neoplasia predisposition. *Gastroenterology*. 2004;126:964-70.
 15. Esteller M, Silva JM, Dominguez G, Bonilla F, Matias-Guiu X, Lerma E et al. Promoter hypermethylation and BRCA1 inactivation in sporadic breast and ovarian tumors. *J.Natl.Cancer Inst*. 2000;92:564-9.
 16. Sakai T, Toguchida J, Ohtani N, Yandell DW, Rapaport JM, Dryja TP. Allele-specific hypermethylation of the retinoblastoma tumor-suppressor gene. *Am.J.Hum.Genet*. 1991;48:880-8.
 17. Greger V, Passarge E, Hopping W, Messmer E, Horsthemke B. Epigenetic changes may contribute to the formation and spontaneous regression of retinoblastoma. *Hum.Genet*. 1989;83:155-8.
 18. Lee MN, Tseng RC, Hsu HS, Chen JY, Tzao C, Ho WL et al. Epigenetic inactivation of the chromosomal stability control genes BRCA1, BRCA2, and XRCC5 in non-small cell lung cancer. *Clin.Cancer Res*. 2007;13:832-8.
 19. Liu H, Zhou Y, Boggs SE, Belinsky SA, Liu J. Cigarette smoke induces demethylation of prometastatic oncogene synuclein-gamma in lung cancer cells by downregulation of DNMT3B. *Oncogene*. 2007;26:5900-10.
 20. Heindel JJ. The fetal basis of adult disease: Role of environmental exposures--introduction. *Birth Defects Res A Clin Mol Teratol*. 2005;73:131-2.
 21. Herceg Z. Epigenetics and cancer: towards an evaluation of the impact of environmental and dietary factors. *Mutagenesis*. 2007;22:91-103.
 22. Weidman JR, Dolinoy DC, Murphy SK, Jirtle RL. Cancer susceptibility: epigenetic manifestation of environmental exposures. *Cancer J*. 2007;13:9-16.
 23. Anway MD, Leathers C, Skinner MK. Endocrine disruptor vinclozolin induced epigenetic transgenerational adult-onset disease. *Endocrinology*. 2006;147:5515-23.
 24. Anway MD, Skinner MK. Epigenetic transgenerational actions of endocrine disruptors. *Endocrinology* 2006;147:S43-S49.
 25. Ho SM, Tang WY, Belmonte dF, Prins GS. Developmental exposure to estradiol and bisphenol A increases susceptibility to prostate carcinogenesis and epigenetically regulates phosphodiesterase type 4 variant 4. *Cancer Res*. 2006;66:5624-32.
 26. Dolinoy DC, Huang D, Jirtle RL. Maternal nutrient supplementation counteracts bisphenol A-induced DNA hypomethylation in early development. *Proc Natl Acad Sci U.S.A*. 2007;104:13056-61.
 27. Bollati V, Baccarelli A, Hou L, Bonzini M, Fustinoni S, Cavallo D et al. Changes in DNA methylation patterns in subjects exposed to low-dose benzene. *Cancer Res*. 2007;67:876-80.
 28. Ross SA. Diet and DNA methylation interactions in cancer prevention. *Ann N.Y. Acad Sci*. 2003;983:197-207.
 29. Kaati G, Bygren LO, Edvinsson S. Cardiovascular and diabetes mortality determined by nutrition during parents' and grandparents' slow growth period. *Eur J Hum Genet*. 2002;10:682-8.
 30. Kaati G, Bygren LO, Pembrey M, Sjostrom M. Transgenerational response to nutrition, early life circumstances and longevity. *Eur J Hum Genet*. 2007;15:784-90.
 31. Bertram C, Trowern AR, Copin N, Jackson AA, Whorwood CB. The maternal diet during pregnancy programs altered expression of the glucocorticoid receptor and type 2 11beta-hydroxysteroid dehydrogenase: potential molecular mechanisms underlying the programming of hypertension in utero. *Endocrinology*. 2001;142:2841-53.
 32. Cooney CA, Dave AA, Wolff GL. Maternal methyl supplements in mice affect epigenetic

- variation and DNA methylation of offspring. *J.Nutr.* 2002;132:2393S-400S.
33. Davis CD, Uthus EO. DNA methylation, cancer susceptibility, and nutrient interactions. *Exp Biol Med (Maywood)*. 2004;229:988-95.
 34. Wolff GL, Kodell RL, Moore SR, Cooney CA. Maternal epigenetics and methyl supplements affect agouti gene expression in Avy/a mice. *FASEB J.* 1998;12:949-57.
 35. Dolinoy DC, Weidman JR, Waterland RA, Jirtle RL. Maternal genistein alters coat color and protects Avy mouse offspring from obesity by modifying the fetal epigenome. *Environ Health Perspect.* 2006;114:567-72.
 36. Rees WD, Hay SM, Brown DS, Antipatis C, Palmer RM. Maternal protein deficiency causes hypermethylation of DNA in the livers of rat fetuses. *J Nutr.* 2000;130:1821-6.
 37. Mack GS. Epigenetic cancer therapy makes headway. *J.Natl.Cancer Inst.* 2006;98:1443-4.
 38. Oki Y, Aoki E, Issa JP. Decitabine--bedside to bench. *Crit Rev.Oncol.Hematol.* 2007;61:140-52.
 39. Oki Y, Jelinek J, Shen L, Kantarjian HM, Issa JP. Induction of hypomethylation and molecular response after decitabine therapy in patients with chronic myelomonocytic leukemia. *Blood* 2008;111:2382-4.
 40. Kantarjian H, Oki Y, Garcia-Manero G, Huang X, O'brien S, Cortes J et al. Results of a randomized study of 3 schedules of low-dose decitabine in higher-risk myelodysplastic syndrome and chronic myelomonocytic leukemia. *Blood* 2007;109:52-7.
 41. Marks PA, Breslow R. Dimethyl sulfoxide to vorinostat: development of this histone deacetylase inhibitor as an anticancer drug. *Nat.Biotechnol.* 2007;25:84-90.
 42. Rosato RR, Grant S. Histone deacetylase inhibitors in cancer therapy. *Cancer Biol.Ther.* 2003;2:30-7.

Amish, Mennonite and Hutterite Genetic Disorder Database

Michael Payne (Meds 2010)

Faculty Reviewers: Dr. C. Anthony Rupar and Dr. Victoria Mok Siu

Clinical Example

A newborn baby girl presented with bowel obstruction. The possibility of cystic fibrosis was raised, particularly since there was a positive family history of cystic fibrosis in a deceased relative. CFTR mutation analysis was ordered using a panel that included $\Delta F508$ and 28 other common mutations which account for 85% of all CF mutations. The results were negative for all mutations in the screening panel.

Since the parents were members of the Old Order Amish community, the Amish, Mennonite and Hutterite Genetic Disorder Database was accessed via the Internet and it was determined that a specific mutation (3905 ins T) had been described in the Old Order Amish. Further DNA testing confirmed that the baby was homozygous for the 3905 ins T mutation. Although the sweat chloride test is the gold standard for cystic fibrosis testing, it can be difficult to obtain sufficient sweat volume collection in the newborn period.¹

Correct diagnosis of newborns with CF through mutation analysis is important as this will help direct therapies and improve overall outcomes.² This case illustrates the usefulness of the Amish, Mennonite and Hutterite Genetic Disorder Database as it allows medical professionals to identify genetic disorders and specific mutations which otherwise may be rare in the North American general population.

Introduction

The Amish, Mennonite, and Hutterite Genetic Disorder Database was created in

response to a paucity of resources for Canadian medical practitioners who treat "Plain People." The database focuses on genetic diseases which are found with higher prevalence in these groups due to their unique cultural history. The database can be viewed at:

<http://www.biochemgenetics.ca/plainpeople/>

Common Background

The Amish, Mennonite and Hutterite populations share some similarities. They all arose in Europe during the Protestant Reformation in the 16th century.³ In contrast to other Protestants and Catholics, they are Anabaptists, believing that children should not be baptized at birth, but instead at an age when they can make a conscious decision to join the church. They also believe in the separation of church and state. These views were seen as heretical and led to their persecution in Europe by the Protestant and Catholic majority. The Anabaptists are also pacifists who do not engage in war and believe in segregating themselves from the general population.³ "Plain People" is a term which refers to their plain dress and restricted use of technology.⁴

History and Migrations

In order to understand present day genetic issues surrounding the Plain People it is important to understand where they originated and how they came to Canada and the USA. They can broadly be divided into 4 groups: the Hutterites, Old Colony Mennonites, Old Order Mennonites and Amish.

The Hutterites were followers of Jakob Hutter in Austria, and their colony was founded in 1528.⁵ They were persecuted for most of their history and were forced to migrate between settlements in Moravia, Romania, Hungary and, Ukraine. From 1874 to 1879, 443 individuals immigrated to the United States.⁵ They settled on the prairies and prospered until WWI when they refused to accept conscription. Canada offered them land and exemption from military service. Many Hutterites immigrated to Canada during this period and settled in Alberta, Saskatchewan and Manitoba. There are also a few colonies in British Columbia. Their current population in the US and Canada numbers over 40,000.⁵ The Hutterites believe in communal ownership, with minimal personal possessions. They live in communal colonies, use modern farming and manufacturing techniques, and utilize modern health care.⁶

Mennonites are named for their founding leader, Menno Simons. The Mennonites can be divided into two groups: Dutch-North German and Swiss-South German. The Dutch-North German (Old Colony) Mennonites originated in the low countries of Europe in the 16th century.³ As a result of persecution they were forced to move to Prussia and lived there in relative peace until the 18th century. With continued growth, land shortages forced emigration from Prussia to Russia. In 1870, 7000 Mennonites immigrated to Manitoba, while another 10,000 immigrated to the American Midwest.³ Another wave of Russian Mennonites came to Canada in 1920 following the Russian revolution.³

In the 1920s, 7000 of the Canadian Old Colony Mennonites emigrated to Mexico, Belize and South America. Recently, many Old Colony Mennonites have returned to Canada from Mexico and South America, settling mostly in Southwestern Ontario, Alberta and Manitoba.³ Today, Old Colony Mennonites in Canada number over 27,000. They speak Low German at home, and High German for more formal occasions, such as church.³

The Swiss-South German Mennonites were founded in the 16th century in Switzerland.

In 1693, followers of Jakob Amman split from the Swiss-South Mennonites to form the Amish sect, a more conservative group.⁷ Both the Swiss-South German Mennonites and the Amish suffered persecution in Europe. Between 1683 and 1880, approximately 8000 Swiss-German Mennonites immigrated to the United States.⁷ With the onset of the revolutionary war in the USA, members of the Mennonite and Amish communities travelled to Canada, settling in the Kitchener-Waterloo and Aylmer-St. Thomas areas.

In 1824, Amish families began immigrating directly to the Waterloo and Perth County areas of Ontario from Europe.⁸ In the late 19th century, Old Order Mennonites (OOM) split from the other Swiss-Mennonite groups in the USA and Canada due differing opinions over Sunday school and higher education.⁷ OOM and the Amish are both quite conservative and can be seen driving buggies and wearing conservative clothes.⁷

Suitability for Genetic Study

The unique history of Plain People groups makes them especially suitable for genetic research. They are socially isolated with little genetic inflow. They keep extensive genealogical records, maintained by local ministers. Their initial founder populations are well known. They have experienced many genetic bottleneck events caused by successive migrations over their history. Large families are common, with low rates of non-paternity. They have a relatively high standard of medical care.⁹

The founder effect is quite pronounced in Plain People populations. For example, most of the over 40,000 Hutterites in North America can trace their ancestry back to a group of 89 founding members.⁵ The Amish and Mennonite populations of North America are similar with small founding populations and isolation resulting in genetic homogeneity.⁷ This can highly skew the prevalence of certain diseases in the isolated populations, particularly autosomal recessive conditions. For example, nephropathic cystinosis is a rare autosomal recessive lysosomal storage disorder affecting about 1 in 100,000 children in

the general population with a carrier frequency of about 1 in 150.¹⁰ In the Old Order Amish population of Ontario, the cystinosis carrier frequency is 1 in 4.5, predicting an extraordinarily high incidence of 1 in 78.¹¹ The increased incidence of these otherwise rare conditions allow for linkage analysis and identification of causative genes.¹²

These genetic studies are not only useful for Plain People, but also for the general population. Since 10-20% of each generation of Old Order Amish and Mennonite children have chosen to leave their communities, many alleles have entered the general population.¹³ Also, many of the mutations causing disease in the Plain People can be found in Europe, where the founding populations originated.¹³ Genetic linkage studies in the Mennonites has led to the discovery of the genes for many conditions, such as hypophosphatasia and X-linked congenital stationary night blindness.³ The Clinic For Special Children in Pennsylvania specializes in care for Plain People and have been successful in isolating over 50 different mutations causing disease in the Mennonite and Amish communities over the past 5 years.¹⁴

The Database

The Amish, Mennonite, and Hutterite Genetic Disorder Database is intended to be used as a resource to assist in research and diagnosis of genetic conditions for patients of Plain People ancestry. It has been compiled by researching published genetic conditions for Plain People. Literature searches were performed on PubMed. Some disorders and mutations were entered based on personal communication with other genetic researchers.

For each disorder, the group affected is recorded along with the geographic location (e.g. Amish, Ontario). This is important because disease prevalence can vary between locations, even within a specific group (e.g. Hutterite), as a result of differences in the founding populations.¹² The specific gene and mutation(s) are also listed, if known. The journal articles detailing the illness with specific mutation(s)

found in Plain People are listed along with the OMIM reference number. Finally, the clinical symptoms of each condition are listed, assisting healthcare providers in recognition and diagnosis.

In order to navigate the database, users search by disorder, mutation or clinical signs and symptoms. When searching by disorder, the user is able to input either the OMIM number or the name and is directed to the corresponding page. Alternatively, the user can input a specific gene and identify any disorders which are caused by mutations in that gene. Finally, the user can search the database by clinical manifestations of the genetic conditions. This is done using a drop-down menu divided by different body systems. The clinical search may be limited to one specific Plain People group and a geographic location. Finally, users are provided a link in order to communicate with the site administrator if they have questions or new information to add to the database.

Conclusion

Plain People have unique healthcare needs as a result of small founding populations and cultural isolation. It is our hope that The Amish, Mennonite, and Hutterite Genetic Disorder Database will improve the healthcare delivery to Plain People in Canada. We view it as an ongoing project, with continuing updates to the database with future discoveries.

References

- 1 Farrel, P., Rosenstein, B., White, T., Accurso, F., *et al.* Guidelines for Diagnosis of Cystic Fibrosis in Newborns through Older Adults: Cystic Fibrosis Foundation Consensus Report. *The Journal of Pediatrics.* 2008; 153:2.
- 2 Sharp, J. and Rock, M. Newborn Screening for Cystic Fibrosis. *Clinic Rev Allerg Immunol.* 2008; 35:107-115.
- 3 Orton, N, Innes, M, Chudley, A. and Torben Bech-Hansen, B. Unique Disease Heritage of the Dutch-German Mennonite Population. *Am J Med Genet Part A.* 2008; 146A: 1072-1087.
- 4 Scott, Stephen E. (1989). "Plain People." *Global Anabaptist Mennonite Encyclopedia Online.* Retrieved 20 March 2009. Available from:

- <http://www.gameo.org/encyclopedia/contents/P59ME.html>
- 5 Boycott K, Parboosingh J, Chodirker B, Lowry B, *et al.* Clinical Genetics and Hutterite Population: A Review of Mendelian Disorders. *American Journal of Medical Genetics Part A.* 2008; 146A: 1088-1098.
 - 6 Derek Suderman. 1998. Mennonite Historical Society of Canada. Retrieved March 20, 2009. Available from: <http://www.mhsc.ca/index.asp?content=http://www.mhsc.ca/mennos/tamish.html>
 - 7 Puffenberger EG. Genetic Heritage of the Old Order Mennonites of Southeastern Pennsylvania. *American Journal of Medical Genetics Part C (Semin. Med. Genet.)*. 2003; 121C:18-31.
 - 8 Nolt SM. *A History of the Amish* (2003). Good Books, Intercourse, PA. p125.
 - 9 Francomano C, McKusick VA, Biesecker L. Medical Genetic Studies in the Amish: Historical Perspective. *American Journal of Medical Genetics Part C (Semin. Med. Genet.)*. 2003; 121C: 1-4.
 - 10 Shotelersuk V, Larson D, Anikster Y, McDowell G, Lemons R, Bernardini I, Guo J, Thoene J, Gahl WA. CTNS mutations in an American-based population of cystinosis patients. *Am J Hum Genet.* 1998;63(5):1352-62.
 - 11 Siu V, Kuepfer S, Dewar R, Rupa CA. High carrier rate for cystinosis in the Ontario Old Order Amish. Poster presentation at the American Society of Human Genetics, Toronto, Ontario, October 26-30, 2004. A1804.
 - 12 Arcos-Burgos M, Muenke M. Genetics of Population Isolates. *Clin Genet.* 2002: 61:233-247.
 - 13 Morton H, Morton C, Strauss K, Robinson D, *et al.* Pediatric Medicine and the Genetic Disorders of the Amish and Mennonite People of Pennsylvania. *American Journal of Medical Genetics Part C (Semin. Med. Genet.)*. 2003; 121C: 5-17.
 - 14 Clinic For Special Children. Retrieved March 20, 2009. Available from: http://www.clinicforspecialchildren.org/CSC/Brochure_files/brochure.pdf.

Hemochromatosis – Screening for a Common Condition

Nick Sunderland (Meds 2010)

Faculty Reviewer: Dr. Kamilia Rizkalla

Hemochromatosis is a common genetic disorder in the Caucasian population. It can go unrecognized in many patients for years before manifesting itself as multisystem end-organ damage. There is debate as to whether this condition should be screened for on a routine basis; as such this article serves to explore the basics of the disease and to review current guidelines for screening at the primary care level.

Case

A 26 year old female fashion student getting lab work for allergy testing was incidentally found to have significantly increased serum transferrin and ferritin levels. Subsequent genetic testing revealed homozygous mutation for the gene encoding the HFE protein. Retrospectively, the mother reports a history of improvement in chronic fatigue and joint pains following elective phlebotomies at the local blood bank. Her testing revealed a compound (double) heterozygous mutation in the HFE gene, but no abnormality in her liver function tests or iron levels. The father and 24 year old brother have normal ferritin and liver enzymes; the 28 year old brother, however, has markedly increased iron stores.

Introduction

Hereditary hemochromatosis (HH) is an autosomal recessive inherited disorder in which mutations in specific genes related to iron transport proteins lead to increased iron absorption from the diet. This eventually leads to deposition of iron in the parenchyma of the liver, heart, pancreas and pituitary glands if left untreated, and can cause significant long term morbidity.¹

This disorder was originally described as “bronze diabetes” by Armand Trousseau in 1865, when he observed an association of skin

pigmentation with diabetic patients.² The first report of deposition of iron in tissue leading to dysfunction was by Von Recklinghausen in 1890. The sequencing of the human genome has allowed identification of the specific mutations associated with the HFE protein, the commonly affected protein in HH. Mutations in this protein are the focus of this article. Note that there are other, less common heritable defects in iron metabolism.

Genetics

The most common genetic defect is in the HFE protein, which interacts with the transferrin receptor and other iron regulating proteins to regulate absorption of iron from the GI tract.

There are two common mutations in HFE, C282Y and H63D. The mutation is most prevalent in Caucasians of European descent, with a homozygote prevalence (C282Y) of about 1 in 200. The prevalence is about 1 in 250 in the general population.

In this same population, 12% are carriers of the C282Y and 25% the H63D mutation. Most HH patients are homozygous for the C282Y mutation. The penetrance of the H63D mutation is considerably lower, thus compound heterozygotes (C282Y/H63D) and H63D homozygotes constitute 10% of HH patients.³⁻⁵

Pathogenesis

The HFE protein as mentioned is important in regulating the uptake of iron from the diet in the GI tract. Iron can only be lost through sweat, skin and the GI tract, menses in females, and during times of growth such as pregnancy and adolescence. Males lose approximately 1mg/day and females proportionally more (1.5-2mg/day). In HH, the ability of the body to sense overall iron stores is lost, leading to an extra absorption of 1-4mg of iron daily, which averages over a year approximately 1 gram. The slow accumulation of iron is generally asymptomatic until the total body iron stores are greater than 20 grams.

Excessive iron is toxic to tissues by free radical reactions, stimulation of collagen formation, and interaction with DNA. Most of these changes are reversible if the iron is removed. Deposition of hemosiderin, an intracellular iron storage complex, in the cells of the liver, pancreas, myocardium, pituitary, adrenal, thyroid and parathyroid glands, joints and skin lead to discolouration, fibrosis, and dysfunction. Interaction with DNA has led to increased risk for hepatocellular carcinoma (HCC) in advanced cases.^{1,7} HCC and cirrhosis are the leading causes of mortality due to HH.

Presentation and Differential

Most cases of HH are discovered through screening, as either an incidental finding when bloodwork is done for another reason or because of a relative with a diagnosis. The manifestations of HH usually occur after age 40 in males (50 in females), after iron stores are greater than 20 grams. The most frequent symptoms include hepatomegaly, abdominal pain, skin pigmentation (especially on sun exposed areas), altered glucose metabolism or diabetes, cardiac arrhythmias, and an atypical arthritis. Hypogonadism resulting in amenorrhea in females and a drop in libido in males is also a common initial presentation.

Each of the symptoms is due to hemosiderin deposition in organ parenchyma and

the ensuing fibrotic reaction from chronic damage. Cirrhosis, pancreatic insufficiency secondary to islet cell destruction and skin pigmentation are all fairly late findings, but are considered the classic "triad". Hypogonadism is related to alteration of the hypothalamic-pituitary axis.¹

The differential for HH includes other causes for elevated iron stores. Most alternative diagnoses are secondary causes of iron overload, most commonly chronic transfusions, either alone or in association with ineffective erythropoiesis. The latter condition results from RBC being destroyed before they can leave the bone marrow.⁶

Screening

The understanding of the underlying pathophysiology of this disorder has come a long way. At a population level, it has been debated whether routine screening (via a fasting serum transferrin saturation) should be implemented, due to the high prevalence of the mutation, ability for early diagnosis and treatment, and ease of genetic testing. However, at this time widespread screening has not been recommended, due to lack of predictability about the expression of the mutations and lack of information on risk-stratifying patients.^{3, 6-8} Labelling of individuals who test positive through genetic testing but are asymptomatic is also another concern and could lead to difficulties with obtaining insurance, for example.⁷

The variable expression in this condition has been a focus of some relatively large screening studies, including a study by Beutler *et al.* (2002) which involved 41,000 US individuals tested for the C282Y and H63D mutations. They found no difference in the age distribution or frequency of signs and symptoms of HH between the cases and controls. However, a more recent study of 31,000 Australian patients of northern European decent (followed for 12 years), showed iron-overload related disease in 28% of male and 1.2% of female homozygotes.⁵ Other observational studies have identified a substantial percentage of first degree relatives of

homozygotes with subclinical conditions related to HH.¹⁰

Following from this, a reasonable strategy for screening would include targeting high risk individuals, such as adult men > 25 years old of northern European decent and first degree relatives of patients with known hereditary hemochromatosis. The screening test of choice would be a fasting serum transferrin saturation (men >52% and women >50%) or a fasting serum unsaturated iron binding capacity. Follow-up to this would be genetic testing.^{3,4,8,10}

Diagnosis

The diagnosis of HH rests on a combination of the clinical features mentioned above, and an elevated serum ferritin and fasting transferrin saturation. In the past, a liver biopsy was used in confirmation of the disease; however, genetic testing has largely replaced this invasive test except in the case of documented liver impairment and need to determine extent of cirrhosis. The serum ferritin value needs to be analyzed with caution as it is an acute phase reactant. Excluding the secondary causes of iron overload is important (Table 1).

Treatment

The definitive treatment of HH and the associated iron-overload is phlebotomy, 500ccs weekly. This is continued until iron-deficiency erythropoiesis is induced as evidenced by a reduced MCV and decreased Hb value, and/or

serum ferritin levels and transferrin saturation markers are within the target range, which are below 50% and 50ng/mL, respectively. Patients will usually require a phlebotomy every 2-3 months as maintenance therapy, which in many cases can be accomplished by regular donations to the local blood bank. Patients testing positive for HFE mutation but without laboratory evidence of iron overload can be followed annually for evidence of disease, but do not require active treatment.

There is no need for patients to avoid red meats and other dietary sources rich in iron; a well balanced diet is acceptable. Patients can be at higher risk for certain infections such as *Vibrio vulnificus*, and as such eating raw seafood is discouraged.

Case Discussion

The case represents a common scenario, in this case a Caucasian family with a compound heterozygote mother (C282Y/H63D) and the daughter homozygous for the C282Y mutation. This case illustrates the need for awareness on the part of family doctors of at-risk populations to determine the need for genetic testing. Had the fashion student (homozygote) not had her blood tested she may have gone years without any knowledge of her condition, until presentation with signs of end organ damage. While a number of incidental cases of iron-overload are inevitable until better evidence is available for specifically identifying those at risk, targeting the at-risk sub-groups should minimize this occurrence.

Table 1: Causes of iron overload⁶ (*focus of article)

Hereditary hemochromatosis	Exogenous iron overload
Related to <i>HFE</i> gene*	Chronic iron supplementation (in absence of blood loss)
African (Bantu) hemochromatosis	Transfusion
Juvenile hemochromatosis	Iron dextran injection
Neonatal hemochromatosis	
Chronic anemias	Chronic liver diseases
Thalassemia major	Viral hepatitis
Sideroblastic anemia	Alcoholic liver disease
Congenital dyserythropoietic anemia	Nonalcoholic steatohepatitis
Congenital atransferrinemia	Porphyria cutanea tarda

References

1. Kumar, Cotran and Robbins. Basic Pathology. Philadelphia: Saunders; 2003. p 615.
2. Trousseau A. "Glycosurie, diabete sucre". Clinique medicale de l'Hotel-Dieu de Paris. 1865. 2: 663-98.
3. Phatak PD, Bonkovsky HL, and Kowdley KV. Hereditary hemochromatosis: time for targeted screening. *Ann Intern Med.* 2008; 149: 270-272.
4. Bacon BR, Britton RS. Editorial: clinical penetrance of hereditary hemochromatosis. *New Engl J Med.* 2008; 358: 291.
5. Allen KJ, Gurrin LC, Constantine CC, *et al.* Iron-overload-related disease in HFE hereditary hemochromatosis. *New Engl J Med.* 2008; 358:221-30.
6. Brandhagen DJ, Fairbanks VF, Baldus W. American Family Physician: Recognition and Management of Hereditary Hemochromatosis [internet]. March 1, 2002. Available at: <http://www.aafp.org/afp/20020301/853.html>
7. Rakel R. Textbook of Family Medicine, 7th edition. Philadelphia: Saunders; 2007. Chp 56, iron overload and hemachromatosis. Online at MD Consult.
8. Qaseem A, Aranson M, Fitterman N, Snow V, Weiss K., Owens D, *et al.* Clinical efficacy assessment subcommittee of the ACP. Screening for hereditary hemochromatosis: a clinical practice guideline from the ACP. *Ann Intern Med.* 2005; 143: 517-21.
9. Beutler E, Felitti V, Koziol JA, Ho NJ, Gelbart T. Penetrance of 845G->A (C282Y) HFE hereditary hemochromatosis mutation in the USA. *Lancet.* 2002; 359: 211-8.
10. Bulaj ZJ, Ajioka RS, Phillips JD, LaSalle BA, Jorde LB, Griffen LM, Edwards CQ, Kushner JP. Disease-related conditions in relatives of patients with hemochromatosis. *N Engl J Med.* 2000; 343(21): 1529-35.

Amniocentesis: Safety, Reliability and Alternatives

Caleb Zelenietz (Meds 2011), Kimberley Colangelo (Meds 2011), and Ishvinder Chattha (Meds 2011)

Faculty Reviewer: Dr. Robert Gratton

Amniocentesis is the most commonly used technique for prenatal genetic diagnosis. It is an effective procedure with a high diagnostic success rate, but carries the risk of severe complications including miscarriage. Despite the risks, amniocentesis performed in the second trimester remains the safest means of prenatal genetic diagnosis. With the introduction of biochemical and first trimester ultrasound screening for fetal aneuploidy, amniocentesis usage rates have declined.

Background of Prenatal Genetic Screening

Pregnancy can be a time of worry, when concerns over fetal health and well-being are at the forefront of many women's minds. Modern clinicians have a wide array of tools at their disposal to allay parents' fears. Prenatal screening for genetic anomalies has become a cornerstone of care for expectant mothers.

In Ontario, Integrated Prenatal Screening (IPS), Serum Integrated Prenatal Screening (SIPS), and First Trimester Screen (FTS) are offered to all women presenting before 14 weeks gestation.¹ The testing uses serum markers and nuchal translucency (in the case of IPS and FTS) to assess the risk of trisomies 18 and 21 along with open neural tube defects. Testing is undertaken in both the first and second trimester, with results becoming available after the second set of testing. Many screening options are possible but IPS and SIPS (if nuchal translucency is not available) prove most accurate and safe.^{2,3} These tests give the mother the risk of her fetus having one of these conditions, but are not diagnostic.

The costs to the health care system of screening have been estimated to be around \$15,000 US per quality adjusted life year gained.⁴ The screening tests are non-invasive and do not pose a risk to the fetus in and of themselves, but are associated with an increased risk of pregnancy

loss due to follow-up diagnostic tests. After a woman has screened positive as having increased risk of one of the fetal anomalies, she must be offered further testing; screening should therefore only be performed when follow-up diagnostic testing is available. When used in the absence of screening, based on maternal age, amniocentesis and chorionic villus sampling cost about \$100,000 US per abnormal birth averted, giving substantial weight to the use of screening programs as a first step in assessing the risk of genetic abnormalities in a fetus.⁵ From a safety perspective, however, the use of amniocentesis with the sole risk factor of advanced maternal age is justified as the risk of aneuploidy is comparable to the risk of miscarriage.⁶

Positive Screen: What Comes Next?

Several options are available after a woman has screened positive: she can choose to continue the pregnancy without further testing, she can have chorionic villus sampling (CVS) in the case of FTS, or she can have amniocentesis. Both CVS and amniocentesis are invasive procedures that pose risks to the fetus, but are also highly effective diagnostic tools with close to 100% diagnostic success rates.^{7,8}

Amniocentesis involves the withdrawal of amniotic fluid from the uterine cavity. Typically, a 20 or 22 gauge needle is inserted into the amniotic sac transabdominally under ultrasound

guidance. It can be done both for obtaining cells to culture for genetic testing, fetal blood typing, and diagnosing fetal infection.⁹ When used for genetic diagnosis, amniocentesis is typically performed in the second trimester. It is most effective at obtaining a useful sample after 15 weeks gestation and before 24 weeks.¹⁰ Cell cultures typically take 7 days to grow before they can be tested. For more rapid results, interphase fluorescence in situ hybridization (FISH) can detect aneuploidy of chromosomes 13, 18, 21, X, Y in one to two days. An alternate procedure for obtaining genetic material from the fetus is chorionic villus sampling (CVS). CVS involves sampling the placenta either transabdominally or transcervically. It is usually performed in the first trimester, after 10 weeks gestation. The rate of both amniocentesis and CVS use has decreased with the introduction of IPS, FTS, and SIPS.^{11,12}

Safety of Early Versus Late Amniocentesis and CVS

The safety of early versus late (first rather than second trimester) amniocentesis and CVS has been studied extensively in the literature. Earlier testing provides a definitive diagnosis much quicker after a positive test, but is not without added risk of complication. A Cochrane review of 16 randomized controlled trials found second trimester amniocentesis to be the safest method of prenatal genetic diagnosis.⁸ Amniocentesis performed before the second trimester is associated with a higher rate of pregnancy loss (7.6% versus 5.9% relative risk of 1.29 95% CI 1.09 to 1.81) than second trimester testing, thus it is not as safe an alternative as later amniocentesis. CVS is the safest method of first trimester sampling, but has a higher loss rate than second trimester amniocentesis with a higher total pregnancy loss (relative risk of 1.40; 95% CI 1.09 to 1.81) and spontaneous miscarriage (9.4%; RR 1.50; 95% CI 1.07 to 2.11).⁸

Risks of Amniocentesis

The maternal risks of amniocentesis are low; of much greater concern are potential fetal complications. The only randomized controlled trial comparing second trimester amniocentesis to

placebo, a study of 4606 low risk women aged 25-34 years, found the rate of spontaneous abortion to be 1.7% in the second trimester amniocentesis group versus 0.7% in the control ultrasound group (relative risk of 2.3).¹³ Current consensus from various trials estimates the loss rate from second trimester amniocentesis to be between 0.6% to 1.0%.¹⁴ Fluid leakage is more common after amniocentesis than in controls, but is not associated with negative pregnancy outcomes.¹³

With ultrasound guidance, direct fetal injury is extremely rare¹³, although it has been reported.^{15,16} Indirect fetal injury is more common, with respiratory distress syndrome and pneumonia being more common in newborns after second trimester amniocentesis.^{13,17} A large cohort study of women 35-49 years old (with 71,586 participants) found an increased incidence of musculoskeletal deformities and respiratory distress in newborns whose mothers underwent 2nd trimester amniocentesis. There was no increase in limb reduction defects, infant and fetal mortality, prematurity, low birth weight, or fetal distress with amniocentesis.¹⁸ There is no long term increased risk for disabilities in children after amniocentesis.¹⁹

Vertical transmission of infections such as cytomegalovirus, hepatitis C Virus, and human immunodeficiency virus has been linked to amniocentesis performed in infected women.^{20,21} Like all invasive procedures in chronically infected women, amniocentesis should be used only when absolutely necessary.

Advantages of Early Versus Late Diagnosis

Although second trimester amniocentesis has been conclusively shown to be the safest method of prenatal genetic diagnosis, it is not without disadvantages. Because it is performed in the second trimester, there is a long lag time between a screen positive test from IPS or SIPS and definitive diagnosis. CVS is available in the first trimester, thus cutting the lag time considerably. Several studies of women's subjective well being have shown shorter time to maternal fetal bonding, earlier anxiety reduction,

and improved health related quality of life in women having a positive screening test followed by a negative CVS as compared to a negative second trimester amniocentesis.²²⁻²⁴ The magnitude of the reduction of anxiety post test is the same for the CVS and amniocentesis groups, but occurs sooner because the CVS is done earlier.²³ This benefit in anxiety reduction exists despite the increased risk for miscarriage with CVS.²³ The same is true for the maternal-fetal bonding improvements, the bonding improves markedly after a negative test result, equally for CVS and amniocentesis, but the CVS is performed earlier.²² Whether these subjective improvements outweigh the very real increased risk of miscarriage should be discussed with each patient.

Aftermath of a Positive Test

After a positive diagnosis of a chromosomal abnormality, the patient faces the difficult decision of whether or not to terminate the pregnancy. The attitudes and knowledge of the health care provider providing the diagnosis affects the patient's choice.^{25,26} The patient's level of education and socio-economic status also plays a role in her decision making.²⁵ Detection rates do not correlate with termination rates; older women are more likely to continue the pregnancy.^{27,28} Pregnancies with sex chromosome abnormalities (SCA) causing infertility and abnormal sexual development were more likely to be terminated than SCA's that did not affect fertility and sexual development.²⁸⁻³¹ The lowest termination rates for SCA are for the 47 XYY and 47 XXX karyotypes.^{28,30,31} Abnormalities visible on ultrasound make the pregnancy less likely to be continued.^{28,30-32} Overall detection rates are increasing for SCA while termination rates have steadily declined.^{29,33,34}

Conclusions

Second trimester amniocentesis is an effective tool for prenatal genetic diagnosis. Although it is not without risks of serious complications such as miscarriage, it remains the safest technique for prenatal genetic diagnosis. The greatest limitation of second trimester

amniocentesis remains the late availability of results, which gives parents little time to make decisions based on the results of the testing. As detection rates of genetic disorders by amniocentesis have increased, termination rates have decreased. Amniocentesis remains a valuable tool for prenatal genetic diagnosis.

References

1. Carroll J. Reference Guide for Health Care Providers Prenatal Screening Tests for the Detection of: Down Syndrome, Trisomy 18 and Open Neural Tube Defects. London Health Sciences Center; 2007 [cited 2009 Feb 20]. Available from: <http://www.lhsc.on.ca/programs/rmgc/mss/provider.pdf>
2. Wald NJ, Rodeck C, Hackshaw AK, Walters J, Chitty L, Mackinson. First and second trimester antenatal screening for Down's syndrome: the results of the Serum, Urine and Ultrasound Screening Study (SURUSS). *Am J Med Screen* 2003; 10:56-104.
3. Wald, NJ, Rodeck, C, Hackshaw, AK *et al.* First and second trimester antenatal screening for Down's syndrome: the results of the Serum, Urine and Ultrasound Screening Study (SURUSS). *Health Technol Assess* 2003; 7:1.
4. Harris RA, Washington AE, Nease RF Jr, Kuppermann M. Cost utility of prenatal diagnosis and the risk-based threshold. *Lancet*. 2004 Jan 24;363(9405):276-82.
5. Heckerling PS, Verp MS. A cost-effectiveness analysis of amniocentesis and chorionic villus sampling for prenatal genetic testing. *Med Care*. 1994 Aug;32(8):863-80.
6. Bornstein E, Lenchner E, Donnenfeld A, Barnhard Y, Seubert D, Divon MY. Advanced maternal age as a sole indication for genetic amniocentesis; risk-benefit analysis based on a large database reflecting the current common practice. *J Perinat Med*. 2009;37(2):99-102.
7. Borrell A, Fortuny A, Lazaro L, Costa D, Seres A, Pappa S, Soler A. First-trimester transcervical chorionic villus sampling by biopsy forceps versus mid-trimester amniocentesis: a randomized controlled trial project. *Prenat Diagn*. 1999 Dec;19(12):1138-42.
8. Alfirevic Z, Mujezinovic F, Sundberg K. Amniocentesis and chorionic villus sampling for prenatal diagnosis. *Cochrane Database of Systematic Reviews* 2003, Issue 3. Art. No.: CD003252. DOI: 10.1002/14651858.CD003252.
9. Ghidini A. Amniocentesis: Technique and complications. *Uptodate*; 1 October 2008. [updated

- 24 Mar 2008, cited 20 Feb 2009]. Available from www.uptodate.com
10. Gosden, CM. Amniotic fluid cell types and culture. *Br Med Bull* 1983; 39:348.
 11. Benn PA, Egan JF, Fang M, Smith-Bindman R. Changes in the utilization of prenatal diagnosis. *Obstet Gynecol* 2004 Jun;103(6):1255-60.
 12. Chasen ST, McCullough LB, Chervenak FA. Is nuchal translucency screening associated with different rates of invasive testing in an older obstetric population? *Am J Obstet Gynecol* 2004 Mar;190(3):769-74.
 13. Philip J, Madsen M, Bang J, Obel EB, Nørgaard-Pedersen B. Randomised controlled trial of genetic amniocentesis in 4606 low-risk women. *Lancet* 1986 Jun 7;1(8493):1287-93.
 14. Wilson RD, Langlois S, Johnson J. Mid-Trimester Amniocentesis Fetal Loss Rate. *J Obstet Gynaecol Can* 2007;29(7):586-590.
 15. Isenberg SJ, Heckenlively JR. Traumatized eye with retinal damage from amniocentesis. *Pediatr Ophthalmol Strabismus*. 1985; Mar-Apr;22(2):65-7.
 16. Admoni MM, BenEzra D. Ocular trauma following amniocentesis as the cause of leukocoria. *J Pediatr Ophthalmol Strabismus*. 1988; Jul-Aug;25(4):196-7.
 17. Sant-Cassia LJ, MacPherson MB, Tyack AJ. Midtrimester amniocentesis: is it safe? A single centre controlled prospective study of 517 consecutive amniocenteses. *Br J Obstet Gynaecol* 1984 Aug;91(8):736-44.
 18. Cederholm M, Haglund B, Axelsson O. Infant morbidity following amniocentesis and chorionic villus sampling for prenatal karyotyping. *BJOG* 2005 Apr;112(4):394-402.
 19. Baird PA, Yee IM, Sadovnick AD. Population-based study of long-term outcomes after amniocentesis. *Lancet*. 1994 Oct 22; 344(8930):1134-6.
 20. Minola, E, Maccabruni, A, Pacati, I, Martinetti, M. Amniocentesis as a possible risk factor for mother-to-infant transmission of hepatitis C virus. *Hepatology*. 2001; 33:1341.
 21. Mandelbrot L, Mayaux MJ, Bongain A, Berrebi A, Moudoub-Jeanpetit Y, Benifla JL, Ciraru-Vigneron N, Le Chenadec J, Blanche S, Delfraissy JF. Obstetric factors and mother-to-child transmission of human immunodeficiency virus type 1: the French perinatal cohorts. SEROGEST French Pediatric HIV Infection Study Group. *Am J Obstet Gynecol*. 1996 Sep; 175(3 Pt 1):661-7.
 22. Caccia N, Johnson JM, Robinson GE, Barna T. Impact of prenatal testing on maternal-fetal bonding: chorionic villus sampling versus amniocentesis. *Am J Obstet Gynecol*. 1991 Oct;165(4 Pt 1):1122-5.
 23. Robinson GE, Garner DM, Olmsted MP, Shime J, Hutton EM, Crawford BM. Anxiety reduction after chorionic villus sampling and genetic amniocentesis. *Am J Obstet Gynecol*. 1988 Oct;159(4):953-6.
 24. Feeny D, Townsend M, Furlong W, Tomkins DJ, Robinson GE, Torrance GW, Mohide PT, Wang Q. Health-related quality-of-life assessment of prenatal diagnosis: chorionic villi sampling and amniocentesis. *Genet Test*. 2002 Spring;6(1):39-46.
 25. Yilmaz Z, Sahin FI, Bulakbasi T, Yüregir OO, Tarim E, Yanik F. Ethical considerations regarding parental decisions for termination following prenatal diagnosis of sex chromosome abnormalities. *Genet Couns*. 2008;19(3):345-52.
 26. Robinson A, Bender BG, Linden MG. Decisions following the intrauterine diagnosis of sex chromosome aneuploidy. *Am J Med Genet*. 1989 Dec;34(4):552-4.
 27. Forrester MB, Merz RD. Pregnancy outcome and prenatal diagnosis of sex chromosome abnormalities in Hawaii, 1986-1999. *Am J Med Genet A*. 2003 Jun 15;119A(3):305-10.
 28. Holmes-Siedle M, Ryyanen M, Lindenbaum RH. Parental decisions regarding termination of pregnancy following prenatal detection of sex chromosome abnormality. *Prenat Diagn*. 1987 May;7(4):239-44.
 29. Brun JL, Gangbo F, Wen ZQ, Galant K, Taine L, Maugey-Laulom B, Roux D, Mangione R, Horovitz J, Saura R. Prenatal diagnosis and management of sex chromosome aneuploidy: a report on 98 cases. *Prenat Diagn*. 2004 Mar;24(3):213-8.
 30. Mezei G, Papp C, Tóth-Pál E, Beke A, Papp Z. Factors influencing parental decision making in prenatal diagnosis of sex chromosome aneuploidy. *Obstet Gynecol*. 2004 Jul;104(1):94-101.
 31. Christian SM, Koehn D, Pillay R, MacDougall A, Wilson RD. Parental decisions following prenatal diagnosis of sex chromosome aneuploidy: a trend over time. *Prenat Diagn*. 2000 Jan;20(1):37-40.
 32. Hamamy HA, Dahoun S. Parental decisions following the prenatal diagnosis of sex chromosome abnormalities. *Eur J Obstet Gynecol Reprod Biol*. 2004 Sep 10;116(1):58-62.
 33. Shaw SW, Chueh HY, Chang SD, Cheng PJ, Hsieh TT, Soong YK. Parental decisions regarding prenatally detected fetal sex chromosomal abnormality and the impact of genetic counselling: an analysis of 57 cases in Taiwan. *Aust N Z J Obstet Gynaecol*. 2008 Apr;48(2):155-9.
 34. Vestergaard CH, Lidegaard O, Tabor A. Invasive prenatal diagnostic practice in Denmark 1996 to 2006. *Acta Obstet Gynecol Scand*. 2009 Jan 26:1-4. [Epub ahead of print]

Ductal Carcinoma in situ in a 25-Year-Old Male with Unilateral Gynecomastia

Christopher J. Coroneos (Meds 2010)

Faculty Reviewer: Dr. Caroline Hamm

DCIS in a young male is rarely reported. Our patient is a 25 year old male who presented with symptomatic unilateral gynecomastia. He presented with a strong family history of cancer on both maternal and paternal sides of his family including breast, lung (maternal) and melanoma, colon and pancreatic (paternal). His mother tested negative for BRCA1 and BRCA2. There is no information on the paternal genetic testing. He was treated with left subcutaneous mastectomy. Upon histologic review of the sample, concurrent gynecomastia and ductal carcinoma in situ was discovered. To date, only four cases of gynecomastia and DCIS have been described in younger male patients. Since only 30 – 50% of patients with DCIS eventually develop invasive cancer in the subsequent 10 – 20 years, this figure in the general population may be higher. This case underscores the importance of family history in any patient presenting with a breast mass. Patients must be made aware of the risk, however small it may be, and physicians must remain cautious of malignancy in young males with gynecomastia.

Introduction

By definition, gynecomastia is a benign condition affecting males characterized on histology by glandular tissue proliferation.¹ It must be distinguished first from pseudogynecomastia, the deposition of fat with absence of glandular tissue proliferation seen in obese males and second, from breast carcinoma. The increasing size of the breast is due to parenchyma and/or fatty tissue. Gynecomastia is common and most prevalent in the neonatal, pubertal and elderly periods.² However, it is also present in 33-41% of adult males aged 25-45.² Long-standing cases that are resistant to medical management or aesthetically displeasing are treated with surgical excision with or without liposuction.³

In contrast, cases of male breast cancer (MBC) are uncommon, occurring in approximately 1 in 100 000 men⁴ and leading to under 0.5% of male cancer deaths annually.⁵ The age of incidence is typically 65-67 years.⁶⁻⁹ Risk factors include testicular abnormality, estrogenic/androgenic imbalance, Klinefelter

syndrome, BRCA mutation, positive family history, obesity, radiation exposure and liver disease.⁸ While gynecomastia has been hypothesized to be associated with MBC, research indicates its incidence is not higher among MBC patients when compared to the general population.^{4, 10, 11} The rare coexistence of the two conditions has been identified in the literature.^{12, 13} Still more rare are cases of ductal carcinoma in situ (DCIS) with gynecomastia, especially in the young adult population. To date, only four cases of DCIS in the setting of gynecomastia have been described in patients that are 25 year old or less¹⁴⁻¹⁷ and only one of these describes unilateral gynecomastia as our patient does.¹⁶

Case Report

A 25 year old male presented in 2007 for cosmetic mastectomy. He reported a history of new onset localized discomfort in the periareolar region of the left breast and a growth in the left breast for one year, increasing in size. On presentation, both the size of the mass and the associated pain had decreased in magnitude.

Physical exam noted slight swelling deep to the left nipple areolar complex with no axillary lymphadenopathy. The right breast had no discernable changes. Family history was significant for malignancy including the maternal grandmother (metastatic lung cancer to bone and bilateral breast cancer at ages 45 and 46 respectively, death at 49), maternal great-aunt (breast cancer, death at 65), second maternal aunt (breast carcinoma at age 60, relapse and bone metastases, currently undergoing treatment), maternal grandfather (pancreatic cancer), paternal grandfather (colon cancer), paternal great-uncle and two paternal uncles (melanoma). He reported no other health concerns, and history was notable only for surgical hernia repair at age three. The remainder of the physical exam was unremarkable.

Ultrasound reported a small amount (1.5cm) of mixed echogenic tissue in the retroareolar region, consistent with benign changes and gynecomastia. Left subcutaneous mastectomy was performed. The suspected gynecomastia was completely excised, measuring approximately 6.5x6.0x2.5cm, weighing 48 grams. On microscopic examination, pathology reported gynecomastia having abundant fibrous stroma separating ducts having columnar cells with inbudding. In addition, a cribriform pattern of cellular change was present. Pathology was consistent with nuclear grade I/III DCIS, largest focus 7mm. The pathology report mentioned the relative rarity of intraductal carcinoma in the demographic group of our patient.

Further investigation included MR mammogram at three months post-operative, reporting increased signal intensity in the left retroareolar region consistent with post-surgical changes and no abnormal enhancement. CA 19-9 was normal at 13 kU/L. Ultrasound of the abdomen and bone scan demonstrated no abnormalities. Ultrasound of the right (unaffected) breast was unremarkable. The patient's mother has tested BRCA1/2 negative. The patient underwent follow-up with medical and radiation oncologists and received genetic counselling. Given the negative surgical margins described on pathology, low grade malignancy,

absence of axillary findings and potential toxicities of adjuvant treatment in this young age group, no further intervention was recommended.

Discussion

Breast cancer in males is far less common than in females. On average, the typical age of onset is up to ten years older than in females.⁶ Similar to female cases, incidence of MBC has increased from 0.86 per 100 000 in 1973 to 1.08 in 1998.¹⁸ HER2 overexpression, a negative prognostic factor in women, is found less often in males.¹⁹ Male cases are more frequently ER and PR positive,⁸ perhaps indicating increased proliferative activity.²⁰

DCIS accounts for approximately 7%²¹ of MBC and is far less common than invasive ductal (90% of cases), though lobular malignancy is still more rare with only 1.5% of cases.¹⁸ Median age of incidence is 65 years.²² On histology, the papillary form is most common though all subtypes present in females can potentially occur in males.²² Pure DCIS occurs only in 5% of cases as the pathology is often present in association with infiltrating malignancy elsewhere.²² DCIS is more common in females, representing 20% of cases²¹. Only 30 – 50% of all male and female patients with DCIS eventually develop invasive cancer in the subsequent 10 – 20 years, so actual prevalence in the general population may be higher.²³⁻²⁵ Compared to males, fewer intraductal papillary cases with higher grade and younger age are seen in females.^{22, 26} The etiology of DCIS is unknown for males since they lack the terminal duct lobular unit (TDLU) where the malignancy has frequently been found to originate in females.¹³ However, not all cases are associated with the TDLU and it is hypothesized cases in males originate from epithelium of ducts.²² Only a limited number of large studies pertaining to male DCIS are available.^{22, 27, 28}

Breast cancer of any form in young men is exceptionally rare. At the time of our patient's presentation, only six cases had been described in males under age 25,^{14, 15, 29-32} and only four cases of males of any age with breast cancer revealed on pathological examination following surgical

intervention for gynecomastia.^{14,15,29,33} To our knowledge, among males 25 years of age or younger, a case of unilateral gynecomastia and DCIS has only been described once in the literature with Chang's account of a 16 year old male.¹⁶

Our patient originally presented with unilateral gynecomastia, though bilateral cases are more common.³⁴ There is no evidence that either unilateral or bilateral gynecomastia increase risk for MBC.³⁵ Obesity is an independent risk factor, and may confound the association with gynecomastia.¹⁵ Gynecomastia in adults is most often due to persistence from puberty, drugs, cirrhosis, hypogonadism, testicular tumor, hyperthyroidism and idiopathic.³⁶ Beyond his breast, our patient had an otherwise normal physical exam and investigations. Of note however, is the comment in the surgical pathology report of putative anabolic steroid use. Use of anabolic steroids is not mentioned anywhere else in the patient's medical record. Some state that cases of gynecomastia should be biopsied if presentation includes query Klinefelter's syndrome, bloody discharge, firm, irregular or unilateral mass.^{37,38} Others endorse mammography to recognize mass since it distinguishes between glandular tissue and fat,^{39,40} though it is not universally supported since male breasts are small and dense.^{41,42} Mammography for MBC has been reported as successful as 92% sensitive, 91% specific.⁴³ Our patient received an ultrasound which demonstrated findings consistent with gynecomastia but made no mention of possible malignancy. It did not report microcalcifications typical of DCIS.¹⁷ The patient was seen again in follow-up with no improvement and surgical intervention of gynecomastia for cosmesis was planned. The possible increased risk of squamous cell carcinoma and testicular cancer later in life⁴⁴ is considered prior to surgery in a younger patient and annual screening is suggested for cases of gynecomastia.¹⁶

DCIS will most often present with bloody nipple discharge and a mass²¹, though mild symptoms, concurrent gynecomastia and the scarcity of its incidence result in frequent mis-

diagnoses.¹⁴ Our patient had malignancy diagnosed on routine post-operative pathological investigation. Thus he did not benefit from pre-operative diagnostic evaluation and staging. Fine needle aspiration cytology is accurate in men when sufficient tissue is obtained, though up to a quarter of cases have insufficient samples.⁴⁵ ER, PR and HER2 profiles are obtained. The remainder of diagnostic routine mirrors cancer in women and investigation includes lab work, bone scan, CT pelvis/abdomen and plain films as clinically relevant.⁴⁶

Research pertaining to the prognosis of DCIS is conflicting. Studies in the past concluded a poorer prognosis in MBC when compared to females, though the older age and delay in diagnosis are thought to have been factors.¹⁸ It is currently believed that prognosis MBC is equal to that in females,⁶ though specific outcomes for DCIS unavailable. Breast cancer specific survival for treated DCIS is as high as 99% in females and 85% in cases of invasive recurrence.⁴⁷ Cutuli *et al.* observed four recurrences among their 27 DCIS patients in follow-up following lumpectomy who were then treated with radical salvage surgery.²⁷

Treatment for DCIS in males does not have an established benchmark and no definitive trial has been published. Current treatment has been guided by experiences reported in the literature.¹⁴ For any MBC, surgeons have the option of mastectomy, local excision or lumpectomy and adding adjuvant therapy as well as axillary dissection. Total mastectomy is suggested²¹ and a recurrence following this procedure has yet to be described.^{42, 49} Research pertaining to lumpectomy and adjuvant radiation therapy are sparse and it is not endorsed.²¹ Though it bears the potential to improve cosmesis,⁵⁰ the potential long-term toxicities of radiation treatment in our patient's young age group outweighs benefit. Nipple excision is often necessitated⁴² since the male breast most often leads malignancy to a subareolar presentation.¹⁴ There is no evidence for axillary lymph node dissection, tamoxifen and adjuvant radiation or chemotherapy.²¹ Since our patient received treatment for what was thought to be

gynecomastia, subcutaneous mastectomy was performed. The patient was well and no recurrence was detected on follow-up at three months.

There is no indication in the literature that samples from gynecomastia procedures should be routinely sent for pathological examination. Given the unilateral nature in our patient, samples were sent for examination to rule out the rare occurrence of malignancy. Liao *et al.* recognize the routine histological examination of samples in female cases of reduction mammoplasty.¹⁵ It is further suggested that the same procedure be following for gynecomastia samples when possible, noting suction lipectomy may make it difficult because of large volume and destruction of cells.¹⁵ Perhaps surgical mastectomy should be applied to more cases, allowing for better review of not only the sample itself, but also the surgical margins.

Our patient had a significant family history of breast cancer, though his mother had tested negative for BRCA 1 and 2. Fifteen to twenty percent of all MBC patients have a family history of breast cancer. Just as our patient was, all MBC cases are routinely offered genetic counselling. Beyond BRCA, MBC has been associated with PTEN mutation in Cowden's syndrome⁵¹ as well as MLH1, a mismatch repair gene related to hereditary nonpolyposis colorectal cancer syndrome.⁵² Our patient did not undergo genetic testing since his mother had tested negative.

References

1. Braunstein G. Pubertal gynecomastia. In: Lifshitz F, ed. *Pediatric Endocrinology*. New York: Marcel Dekker; 1996:197.
2. Niewoehner C, Schorer AB. Gynaecomastia and breast cancer in men. *BMJ* 2008;336:709-13.
3. Gikas P, Mokbel K. Management of gynaecomastia: an update. *International journal of clinical practice* 2007;61:1209-15.
4. Fentiman I, Fourquet A, Hortobagyi GN. Male breast cancer. *Lancet* 2006;367:595-604.
5. Jemal A, Siegel R, Ward E, Hao Y, Xu J, Murray T, Thun MJ. Cancer statistics, 2008. *CA: a cancer journal for clinicians* 2008;58:71-96.
6. Anderson W, Althius MD, Brinton LA, Devesa SS. Is male breast cancer similar or different than female breast cancer? *Breast cancer research and treatment* 2004;83:77-86.
7. Thomas D. Breast cancer in men. *Epidemiologic reviews* 1993;15:220-31.
8. Giordano S, Buzdar AU, Hortobagyi GN. Breast cancer in men. *Annals of Internal Medicine* 2002;137:678-87.
9. Cutuli B, Lacroze M, Dilhuydy JM, Velten M, DeLaFontan B, Marchal C, Resbeut M, Graic Y, Campana F, Moncho-Bernier V, DeGislain C, Tortochaux J, Cuillere JC, Reme-Saumon M, N'Guyen TD, Lesaunier F, LeSimple T, Gamelin E, Hery M, Berlie J. Male Breast Cancer: Results of the Treatments and Prognostic Factors in 397 Cases. *European Journal of Cancer* 1995;31:1960-5.
10. Carlsson G, Hafstrom L, Jonsson P. Male breast cancer. *Clinical Oncology* 1981;7:149-55.
11. Sasco A, Lowenfels AB, Pasker-de Jong J. Review article: epidemiology of male breast cancer. A meta-analysis of published case-control studies and discussion of selected aetiological factors. *International Journal of Cancer* 1993;53:538-49.
12. Crichlow R. Breast cancer in males. *Breast* 1976;2:12.
13. Crichlow R. Carcinoma of the male breast. *Surgery, gynecology & obstetrics* 1972;134:1011-9.
14. Wadie G, Banever GT, Moriarty KP, Courtney RA, Boyd T. Ductal carcinoma in situ in a 16-year-old adolescent boy with gynecomastia: a case report. *Journal of pediatric surgery* 2005;40:1349-53.
15. Liao E, Kish JB, Hertl MC. Incidental discovery of bilateral breast cancer in a 24-year-old man presenting with gynecomastia. *Annals of Plastic Surgery* 2007;58:673-6.
16. Chang H, Kish JB, Smith BL, Goldstein AM. A 16-year-old male with gynecomastia and ductal carcinoma in situ. *Pediatric surgery international* 2008;24:1251-3.
17. Corroppo M, Erculiani E, Zampieri N, Gobbato M, Camoglio FS, Giacomello L. Ductal carcinoma in situ in a 15-year-old boy with gynaecomastia: a case report. *Pediatric surgery international* 2008;24:943-5.
18. Giordano S, Cohen DS, Buzdar AU, Perkins G, Hortobagyi GN. Breast carcinoma in men: a population-based study. *Cancer* 2004;101:51-7.
19. Bloom KJ GH, Gattuso P, Reddy V, Francescatti D. Status of HER-2 in male and female breast

- carcinoma. *American Journal of Surgery* 2001;182:389-92.
20. Muñoz de Toro MM MM, Kass L, Luque EH. Proliferative activity and steroid hormone receptor status in male breast carcinoma. *The Journal of steroid biochemistry and molecular biology* 1998;67:333-9.
 21. Camus M, Joshi MG, Mackarem G, Lee AK, Rossi RL, Munson JL, Buyske J, Barbarisi LJ, Sanders LE, Hughes KS. Ductal carcinoma in situ of the male breast. *Cancer* 1994;74:1289-93.
 22. Hittmair A, Lininger RA, Tavassoli FA. Ductal carcinoma in situ (DCIS) in the male breast: a morphologic study of 84 cases of pure DCIS and 30 cases of DCIS associated with invasive carcinoma--a preliminary report. *Cancer* 1998;83:2139-49.
 23. Page D, Dupont WD, Rogers LW, Landenberger M. Intraductal carcinoma of the breast: follow-up after biopsy only. *Cancer* 1982;49:751-8.
 24. Betsill Jr W, Rosen PP, Lieberman PH, Robbins GF. Intraductal carcinoma. Long-term follow-up after treatment by biopsy alone. *JAMA* 1978;239:1863-7.
 25. Collins LC TR, Baer HJ, Connolly JL, Colditz GA, Schnitt SJ. Outcome of patients with ductal carcinoma in situ untreated after diagnostic biopsy: results from the Nurses' Health Study. *Cancer*;103:1778-84.
 26. Anderson W, Devesa SS. In situ male breast carcinoma in the Surveillance, Epidemiology, and End Results database of the National Cancer Institute. *Cancer* 2005;104:1733-41.
 27. Cutuli B, Dilhuydy JM, De Lafontan B, Berlie J, Lacroze M, Lesaunier F, Graic Y, Tortochaux J, Resbeut M, Lesimple T, Gamelin E, Campana F, Reme-Saumon M, Moncho-Bernier V, Cuilliere JC, Marchal C, De Gislain G, N'Guyen TD, Teissier E, Velten M. Ductal carcinoma in situ of the male breast. Analysis of 31 cases. *European Journal of Cancer* 1997;33:35-8.
 28. Cutuli B, Florentz P, Lacroze M, Dilhuydy JM, Allavena C, De Lafontan B, Resbeut M, Campana F, Graic Y, Tortochaux J. Breast cancer in males: a study of 15 cases of pure ductal carcinoma in situ. *Bulletin du cancer* 1992;79:1045-53.
 29. Fodor P. Breast cancer in a patient with gynecomastia. *Plastic and reconstructive surgery* 1989;84:976-9.
 30. Hartman A, Magrish P. Carcinoma of breast in children; case report: six-year-old boy with adenocarcinoma. *Annals of Surgery* 1955;141:792-8.
 31. Simpson J, Barson AJ. Breast tumours in infants and children: a 40-year review of cases at a children's hospital. *Canadian Medical Association journal* 1969;101:100-2.
 32. de Bree E, Tsagakatakis T, Kafousi M, Tsiftsis DD. Breast enlargement in young men not always gynecomastia: breast cancer in a 22-year-old man. *ANZ journal of surgery* 2005;75:914-6.
 33. Brenner P, Berger A, Schneider W, Axmann HD. Male reduction mammoplasty in serious gynecomastias. *Aesthetic plastic surgery* 1992;16:325-30.
 34. Wiesman I, Lehman JA Jr, Parker MG, Tantri MD, Wagner DS, Pedersen JC. Gynecomastia: an outcome analysis. *Annals of plastic surgery* 2004;53:97-101.
 35. Al-Qattan M, Hassanain J, Mahmoud S, El-Shayeb A, Tashkandi M, Al-Kattan WM. On the neglected entity of unilateral gynecomastia. *Annals of Plastic Surgery* 2005;55:255-7.
 36. Braunstein G. Gynecomastia. *New England Journal of Medicine* 1993;328:490-5.
 37. Bland K, Graves TA, Page DL. Gynecomastia. In: Bland K, Copeland III EM, ed. *The breast: comprehensive management of benign and malignant diseases*. Philadelphia: WB Saunders; 1998:153-89.
 38. Sullivan J, Magee HR, Donald KJ. Secretary (juvenile) carcinoma of the breast. *Pathology* 1977;9:341-6.
 39. Haagensen C. *Diseases of the breast*. 2 ed. Philadelphia: WB Saunders; 1971.
 40. Cooper R, Gunter BA, Ramamurthy L. Mammography in men. *Radiology* 1994;191:651-6.
 41. Carlson H. Gynecomastia. *New England Journal of Medicine* 1980;303:795-9.
 42. Wilhelm M, Langenburg SE, Wanebo HJ. Cancer of the male breast. In: Bland K, Copeland III EM, ed. *The breast: comprehensive management of benign and malignant diseases*. 2 ed. Philadelphia: WB Saunders; 1998:1416-20.
 43. Evans G, Anthony T, Turnage RH, Schumpert TD, Levy KR, Amirkhan RH, Campbell TJ, Lopez J, Appelbaum AH. The diagnostic accuracy of mammography in the evaluation of male breast disease. *American Journal of Surgery* 2001;181:96-100.
 44. Olsson H, Bladstrom A, Alm P. Male gynecomastia and risk for malignant tumours--a cohort study. *BMC Cancer* 2002;16:26.
 45. Joshi A, Kapila K, Verma K. Fine needle aspiration cytology in the management of male

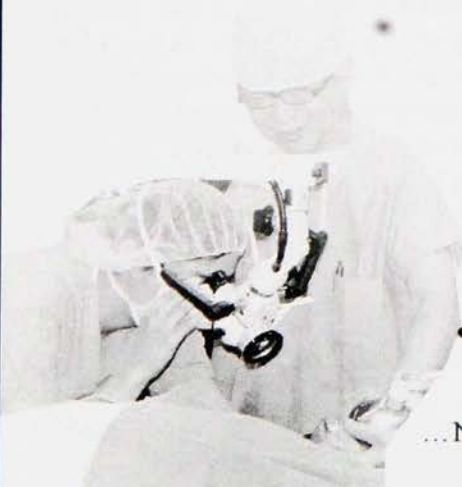
breast masses. Nineteen years of experience. *Acta cytologica* 1999;43:334-8.

46. American Joint Committee on Cancer: AJCC Cancer Staging Manual. 6 ed. New York: Springer-Verlag; 2002.
47. Romero L, Klein L, Ye W, Holmes D, Soni R, Silberman H, Lagios MD, Silverstein MJ. Outcome after invasive recurrence in patients with ductal carcinoma in situ of the breast. *American Journal of Surgery* 2004;188:371-6.
48. Auvinen A, Curtis RE, Ron E. Risk of subsequent cancer following breast cancer in men. *Journal of the National Cancer Institute* 2002;94:1330-2.
49. Frykberg E, Masood S, Copeland EM 3rd, Bland KI. Ductal carcinoma in situ of the breast.

Surgery, gynecology & obstetrics 1993;177:425-40.

50. Deutsch M, Altomare FJ Jr, Mastrian AS, Chervenak JP. Carcinoma of the male breast following thymic irradiation. *Radiology* 1975;116:413-4.
51. Fackenthal J, Marsh DJ, Richardson AL, Cummings SA, Eng C, Robinson BG, Olopade OI. Male breast cancer in Cowden syndrome patients with germline PTEN mutations. *Journal of medical genetics* 2001;38:159-64.
52. Boyd J, Rhei E, Federici MG, Borgen PI, Watson P, Franklin B, Karr B, Lynch J, Lemon SJ, Lynch HT. Male breast cancer in the hereditary nonpolyposis colorectal cancer syndrome. *Breast cancer research and treatment* 1999;53:87-91.

Discover what the Rural Ontario Medical Program can do for you....



- Core & elective rotations in family medicine & select specialties
- Accommodation & travel funding for clerks & residents
- Faculty development
- Preceptor funding

...New preceptors always welcome!

Providing clinical experiences to medical trainees for twenty years.



Rural Ontario Medical Program

Toll Free: 1-877-445-7667 ~ www.romponline.com ~ romp@romponline.com

SE

www.sehealth.mb.ca

South Eastman Health/Santé Sud-Est Inc.

Fields of Opportunities



South Eastman Health/Santé Sud-Est Inc.

is a Regional Health Authority located in the fastest growing region of Manitoba where vibrant, progressive and multi-cultural communities offer a pace to suit your lifestyle. Minutes from Winnipeg, home to beautiful golf courses, lakes and private beaches. Come home to a good life.

FAMILY PHYSICIANS

Our Recruitment Officer would be pleased to provide further information about our Rural Physician opportunities.

For more information, please contact: South Eastman Health/
Santé Sud-Est Inc. • Human Resources Department • Box 470
La Broquerie MB R0A 0W0 • Fax: (204) 424-5888 •
Email: hr@sehealth.mb.ca

