CIM

Clinical and Investigative Medicine

Vol. 29 • No. 6 • December • 2006

In this issue

CIM

Clinical and Investigative Medicine

Official Journal of the
Canadian Society for Clinical Investigation

CIM is printed by University of Toronto Press, Toronto, ON. and published every two months (February, April, June, August, October, and December). Publication Mail Agreement no. 41136075; PAP registration no. 9845. Return undeliverable copies to the Canadian Society for Clinical Investigation, 774 Echo Drive, Ottawa, ON, Canada K1S 5N8.

The CSCI acknowledges the support of the Government of Canada, through the Publications Assistance Program (PAP), toward mailing costs.

"We acknowledge the finacial support of the Government of Canada through the Publication Assistance Program towards our mailing costs." Canada

Published by the Canadian

Society of Clinical Investigation.

Correspondence and inquiries concerning manuscripts should be sent to the Editor: David R. Bevan, University of Toronto, Rm 126, FitzGerald Building, 150 College St., Toronto, ON, M5S 3E2; TEL: 416-978-4306/7; FAX: 416-978-2408; e-mail: david.bevan@utoronto.ca.

The Canadian Society for Clinical Investigation
La Société Canadienne de Recherches Cliniques

CSCI SCRC

About CSCI

MISSION

To promote clinical and basic research in the field of human health throughout Canada, to lobby for adequate research funding at the federal, regional and local levels, and to support Canadian researchers in their endeavours and at all stages of their careers.

The Society still fulfills its original mandate today. It has evolved, however, to include the active promotion of clinical science and lobbying for support of basic and applied biomedical research from the federal and provincial governments. CSCI members represent researchers across Canada who are studying issues of disease and health care across the spectrum, from basic research to issues of health care delivery.

ORIGINS

The Canadian Society for Clinical Investigation (CSCI) was founded in 1951 and its original purpose was to provide a forum for the exchange of scientific information. It was envisaged as a "travel club for those interested in clinical investigation in Canada". As detailed by J.S.L. Browne, one of the four founding members of the CSCI, the idea was for it to be a very informal organization and not a society.

Its first meeting was attended by 44 people and was an outstanding success. Over the next several years, discussion continued as to the proposed nature, structure and organization of a society for Canadian clinical investigators. These discussions culminated in the formation of the CSCI in 1959. Its first meeting was held in Vancouver that year and the meetings have continued to grow in size and are now held conjointly with the annual meeting of the Royal College of Physicians and Surgeons of Canada.

ORGANIZATION

The CSCI is composed of individuals interested and active in clinical investigation from across the country. Membership is open to those who are interested and active in clinical research and who are sponsored by a member of the Society.

The Executive of the Society consists of 3 members: the President, President-Elect and Past-President. The Council consists of 13 members from medical schools across Canada.

Service Information

Subscription rates (2006)
Annual Subscription rates for libraries, research establishments and other multiple-reader institutions in Canada, Can$308; in the United States and other countries, US$365. Individuals, complete volume in Canada, Can$125 (single copies Can$45); the USA and other countries US$180 (single copy US$55). Replacement copies of older issues, where available, must be prepaid at the single-copy rate. Shipping and handling charges may apply. Canadian subscribers please add 7% GST or 15% HST (NS, NB, NL) as applicable. Payments should be made to the Canadian Society for Clinical Investigation (CSCI) in the funds specified, drawn on a Canadian or a US bank, respectively. VISA and MasterCard are also accepted. Orders and requests for information should be addressed to:

CSCI Head Office (CHO),
774 Echo Drive, Ottawa, ON, Canada
K1S 5N8
FAX (613)730-1116; TEL (613) 730-6240
E-mail: csci@rcpsc.edu
website: www.csci-scrc.org

Replacing Missing Issues
Claims for missing issues must be made to the CHO within three months of the date of publication, to be honoured and replaced (subject to availability)free of charge.

Change of Address
We require eight weeks notice to ensure uninterrupted service. Please send your current mailing label, your new address and the effective date of change to the CHO. Address changes should be directed to CSCI Head Office (CHO), 774 Echo Drive, Ottawa, ON , Canada K1S 5N8.

Reprints
Reprints of articles in Clinical and Investigative Medicine (CIM) are available in orders of at least 50. For information, please contact CSCI.

Permissions

Electronic form, abstracting and indexing
CIM is available from EBSCO Information Services (www.ebsco.com) and Micromedia (www.micromedia.on.ca). CIM appears in several indexing/abstracting services including BIOSIS, CAB Abstracts, Current Contents: Clinical Medicine, Current Contents: Life Sciences, Elsevier BIOBASE, EMBASE/Excerpta Medica, Global Health, the ISI Science Citation Index, MEDLINE/Index Medicus and Ulrich's International Periodicals

Joining CSCI

CRITERIA FOR ADMISSION TO GENERAL MEMBERSHIP

Individuals with an active interest in all medical and allied health research are welcome to apply for membership. Full membership is limited to those already holding degrees in disciplines relevant to medical health sciences.
Members will receive all official publications, including the peer-reviewed journal of the CSCI, Clinical and Investigative Medicine, which is referenced by Index Medicus. Members are eligible to vote and to hold office within CSCI. They are encouraged to participate in the Society's meetings and will receive prompt notification of such events.

CRITERIA FOR ADMISSION TO ASSOCIATE MEMBERSHIP

Any person who is in a training, or a graduate or fellowship program, with an active interest in medical and allied health research is welcome to apply for membership.
Associate Members will receive all official publications of the CSCI and the journal Clinical and Investigative Medicine. They are encouraged to participate in the meetings of the Society, but will not be eligible to vote or hold office.
Associate Members will be invited to become full members upon completion of their training.

Fees:

Full Member: $160.50
Associate Member: $50.00
Emeritus Member: No dues. $45.00
for a subscription to CIM

Contact:

CSCI Secretariat
CSCI Head Office
774 Echo Drive, Ottawa, ON
K1S 5N8
Tel: (613) 730-6240
Fax: (613) 730-1116
E-mail: csci@rcpsc.edu
www.csci-scrc.org

CIM
Clinical and Investigative Medicine
Official Journal of the
Canadian Society for Clinical Investigation
Vol. 29, No. 6 • December 2006

TABLE OF CONTENTS

The Canadian Society for Clinical Investigation
La Société Canadienne de Recherches Cliniques

CSCI SCRC

CSCI AWARDS

CALL FOR SUBMISSIONS

The Canadian Society for Clinical Investigation is pleased to request submissions for the following awards:

Deadline January 31, 2007: CSCI/RCPSC Henry Friesen Award
Canadian Research Awards for Specialty Residents
(Medicine and Surgery)

Deadline June 25, 2007: CSCI/CAPM Core Medical Residents Research Award

Deadline July 6, 2007: CSCI/CIHR Resident Research Prize

For more information, please visit the CSCI website at **www.csci-scrc.org**
or contact the CSCI Office at **csci@rcpsc.edu** / 613-730-6240

CSCI's New Website Address

It is time to update your bookmarks as the CSCI has changed its website address! Please visit **www.csci-scrc.org** to access CSCI online.

~~~~~~~~~

Il est temps de mettre à jour vos signets car le SCRC a changé son adresse de site Web ! Svp visiter **www.csci-scrc.org** afin d'accéder SCRC en ligne.

Canadian Society for Clinical Investigation
Société canadienne de recherches cliniques
774 Echo Drive, Ottawa ON K1S 5N8
Tel: 613-730-6240 Fax: 613-730-1116
csci@rcpsc.edu
www.csci-scrc.org
~~~~~~~~~

ORIGINAL ARTICLE

Cardiovascular manifestations of autosomal dominant polycystic kidney disease in young adults

S. Paul Handa, MD, FRCP, FACP

Department of Nephrology, Atlantic Health Sciences Corporation, Saint John, New Brunswick Canada

Manuscript Submitted: 10th July 2006
Manuscript Accepted: 7th August 2006

Clin Invest Med 2006; 29 (6): 339–346.

Abstract

Purpose: Autosomal Dominant Polycystic Kidney Disease (ADPKD) is the most common hereditary renal disorder. In addition to renal structure and function abnormalities, the cardiovascular changes (extra renal manifestations) are the frequent findings observed in these subjects. This pilot study describes the viscoelastic properties of the aorta, a predisposing factor for the genesis of hypertension and left ventricular hypertrophy in young adults diagnosed with ADPKD.

Methods: Twenty new patients with ADPKD, all the off springs of previously diagnosed patients with ADPKD were recruited to participate in the study. Each patient underwent the measurement of 24-hour creatinine clearance (Ccr), 24-hour Ambulatory Blood Pressure Monitoring (ABPM), and ultrasonographic determination of the aortic dimensions and left ventricular measurements by M-mode echocardiogram and 12-lead electrocardiogram (ECG) in the echo laboratory.

Results: The mean age of our ADPKD subjects was 29.9±6.5 yr. Five patients had impaired renal functions (Ccr < 1.48ml/sec). Fourteen patients had hypertension and nine were considered Non-dippers on ABPM. The median value of LVMI and AOD was 84.9g/m^2 and 28.5×10^{-3}/kPa respectively. Forty five percent of these subjects had non- dipping circadian pattern with smaller nocturnal BP reduction. No relationship between AOD, LVMI and 24-hr ABPM was observed; however, a positive trend towards Ccr and AOD was evident.

Conclusion: The majority of our ADPKD subjects have an unfavourable cardiovascular risk profile: hypertension and a non-dipping circadian BP rhythm. Subjects with evidence of renal function impairment had reduced aortic distensibility, placing them at an increased risk for cardiovascular morbidity and mortality.

Autosomal dominant polycystic kidney disease (ADPKD) ranks the most common genetic disorders occurring in 1 in 400 to 1000 people of European descent.[1, 2] In addition to renal function abnormality, extra renal manifestations have been observed in patients with ADPKD. Hypertension, cardiac involvement, valvular and vascular abnormalities are the frequent extra renal manifestations of ADPKD. Left ventricular hypertrophy (LVH) is a common finding in hypertensive autosomal dominant polycystic disease patients. Chapman et al.[3], Bardji et al.[4] and Zeier et al[5] have shown increased left ventricular mass (LVM) in early stages of the disease. In a recent study, Martinez-Vea et al.[6] confirmed that normotensive patients with ADPKD have greater LVM indices (LVMI) compared with healthy control subjects, and 25% of these patients showed LVH, defined as LVMI ≥ 131 g/m^2 in men and ≥ 110 g.m^2 in women.

Valvular disorders are common among patients with ADPDK. Hossack et al.reported that, in addi-

tion to frequent occurrence of mitral and tricuspid valve prolapse, aortic valve incompetence was eight times more common in subjects with ADPKD than in control subjects.[7] In the two prospective studies, aortic valvular incompetence was found in 9 and 19% of the ADPKD patients compared to 1 and 5% of the random controls.[7, 8]

The other major vascular abnormality in ADPKD is intracranial aneurysms; it appears that in ADPKD patients, rupture of cerebral aneurysms occurs at a young age and carries a greater morbidity and mortality than patients in the general population suffering from cerebral aneurysms.[3] Furthermore, ruptured intracranial aneurysms in young ADPKD patients seem to be associated with hypertension.[9] This vascular abnormality coupled with occasional occurrence of vertebral artery aneurysm does indeed suggest a diffuse systemic vascular disorder. This is probably secondary to abnormal vascular collagen matrix.[10]

The mechanisms responsible for elevation in blood pressure in ADPKD are not well understood. Hypertension is usually noted in association with enlarged palpable kidneys, produced by multiple increasing size cysts. There may be many factors to induce high BP but none seems to be consistent. Patients with impaired glomerular filtration rate (GFR) may retain salt and water leading to volume expansion and elevation in blood pressure. A relationship between activated systemic renin-angiotensin system (RAS) and hypertension has been postulated. There is no definite correlation exists between systemic RAS and hypertension; however, some evidence points towards the role of intrarenal RAS and the progression of polycystic renal disease [11]. Recognizing the potential role of intrarenal RAS, studies have tried the use of angiotensin converting enzyme inhibitors but failed to demonstrate their beneficial effect on the progression of disease. In addition, an open labeled randomized treatment study using atenolol or enalapril for hypertension showed moderately large decline in BP in patients on enalapril compared with the atenolol treated patients; the difference was not statistically significant. Furthermore, no difference in the rate of decline in glomerular filtration rate was noted between these two-treatment groups.[12]

It is likely that the generalized vascular disease process involving the aortic valve and aortic wall contribute to reduced aortic distensibility, which in turn, contributes to the development of hypertension and LVH in some patients with ADPKD. To date, there are no published data concerning the viscoelastic properties of the arterial tree, particularly the aorta. The present study describes the cardiovascular manifestations of ADPKD in young adults. The manifestations of interest are aortic distensibility, LVM and the 24-hour blood pressure profile.

Materials and methods

Subjects

Forty-three adults with ADPKD aged 25-70 yr were being followed in the Nephrology service at Atlantic Health Sciences Corporation in Saint John, New Brunswick. Some of these individuals were either being maintained on dialysis or had received renal transplant. The patient files identified 12 additional cases with ADPKD who had died for various reasons during their follow-up over the past 20-year, but the spouse might still be alive. If we assume that on average, each patient has 1.5 children, this would provide a population base of 82 off springs. If we further assume that at least 15 subjects are already aware of their having ADPKD, the remaining 67 subjects are needed to be screened. Supposing that 70% of these subjects participate, this will help in detecting 23 more cases. The combined numbers of 15 and 23 cases will give us a cohort of subjects with ADPKD for the investigation.

Children of these adults were recruited. Eligibility criteria were (1) a creatinine clearance between 1.19 - 2.13 ml/sec/1.73 m^2 and urinalysis showing no hematuria and proteinuria, (2) older than 16 years of age, and (3) diagnosed with ADPKD. The diagnosis of ADPKD was confirmed by ultrasonographic examination of the kidneys. ADPKD was defined by the presence of five or more cysts bilaterally on ultrasonography. The study protocol was in accordance with the ethical standards of the Helsinki declaration of 1983 and approved by the Hospital Research Ethics Board. All subjects gave written informed consent.

Study protocol

Study subjects were screened for renal function by urinalysis, measurement of serum creatinine and 24-hour creatinine clearance. Supine cuff BP was recorded by a clinic nurse using a mercury sphygmomanometer following 30-minute resting. Korotkoff phases 1 and 5 were taken as the systolic blood pressure (SBP) and diastolic blood pressure (DBP), respectively. Immediately after the cuff BP determination, all subjects underwent 24-h BP monitoring using a programmable BP monitor (SpaceLabs Model 90207 Ambulatory Blood Pressure Monitor, Redmond, WA). The monitor used the oscillometric method to

measure BP and was preset at 30-min intervals during daytime (0601 to 2200 h) and at hourly intervals during nighttime (2201 to 0600 h).

Ultrasonographic determination of the aortic dimensions and left ventricular measurements were made by M-mode echocardiogram (echo) and 12-lead electrocardiogram (ECG) in the echo laboratory. The aortic diameter was recorded by M-mode echo 3 cm above the aortic valve according to the methods recommended by the American Society of Echocardiography. The measurement of aortic diameters has been described elsewhere [13]. M-mode measurements of the left ventricle were obtained to estimate the LVM. An experienced echo technologist performed all echocardiograms under the supervision of a cardiologist experienced in echo. The records were coded and a cardiologist blinded to the data about the subjects independently made the measurements on five consecutive cardiac cycles. The aortic diameters in combination with BP measurements obtained during echo formed the basis for calculation of aortic distensibility (AOD) using the formula described by Stefandis et al.[14.]

AOD = 2 × (Δ aortic diameter)/(diastolic diameter) × (Δ aortic pressure) where Δ aortic diameter = systolic – diastolic diameter, and Δ aortic pressure = cuff SBP- cuff DBP (brachial artery pressure by sphygmomanometer obtained during echo). AOD is expressed in kPa.

The LVM was derived from measurements obtained from M-mode echo examination with the subjects in a partial left decubitus position. LVM was determined by the conventional method developed by Devereux and Reichek.[15] The following formula was used for calculation of the LVM:

LVM (g) = $[(\text{LVIDd} + 2\ \text{LVPWTd})^3 - (\text{LVIDd}^3)] \times 1.05$ where LVIDd is the left ventricular internal end-diastolic diameter and LVPWTd is the left ventricular posterior wall end-diastolic thickness. LVMI was determined by dividing LVM by the body surface area.

Subjects were classified as dippers and non-dippers according to their BP circadian variation. Pierdomenico et al.'s definition was used to classify subjects as dippers and non-dippers.[16] A non-dipping pattern was defined as a difference in the mean daytime and nighttime BP <10%, while a difference of >10% and <20% in mean daytime and nighttime BP was considered as a dipping pattern. Some patients, who were non-dippers for SBP and dippers for DBP, and others who were dippers for SBP and non-dippers for DBP, were classified according to SBP changes because nocturnal decrease in SBP has been shown to confer a reduced cardiovascular risk.[17]

Statistical analysis

In view of the small number of subjects enrolled in the study coupled with uneven number of subjects with and without evidence of renal impairment, nonparametric statistics were used to analyze the data. Results are presented as the median and quartiles. To test for differences in median values between groups (abnormal and normal renal function; dippers and non-dippers), nonparametric one-way analysis of variance statistics were used. The proportions of dippers and renal impairment were compared with the use of χ^2. Multiple regression analysis models were used to examine the relationship of renal function to LVM and AOD. Data were expressed as median and quartile. The data were processed using the software packages SAS/STAT (SAS Institute, Cary, North Carolina). Values of $p<0.05$ were considered statistically significant.

Results

Clinical characteristics of ADPKD subjects with and without impaired renal function.

Twenty patients (6 male and 14 female, mean age 29.9 ± 6.5 yr, range, 26 to 42 yr) with autosomal dominant polycystic kidney disease (ADPKD) were included in the study. None of our study subjects had any valvular abnormalities or symptoms to suggest intracranial aneurysm. Renal function was assessed by 24-h creatinine clearance. Using Levy et al.'s [18] criterions for classifying adequacy of renal function, subjects whose creatinine clearance > 1.5 ml/s/1.73 m^2 were classified as normal renal function, while those with creatinine clearance < 1.48 ml/s/1.73 m^2 were categorized as impaired renal function. These two groups did not differ in age, sex distribution, 24-h BP, heart rate. However, the normal renal function group had significantly greater nocturnal BP reduction (SBP and DBP) and aortic distensibility than the impaired renal function subjects (Table 1).

We defined hypertension as SBP of 130 mm Hg or greater or DBP of 80 mm Hg or greater on average 24-hour ABPM [19]. Seventy percent (n=14) of subjects had SBP or DBP > 130/80 mm Hg. The median value of LVMI and AOD for this subgroup was 84.9 g/m^2 (range 62.4 – 109.9 g/m^2) and 28.5×10^{-3}/kPa, (range 8 – 57×10^{-3}/kPa), respectively.

TABLE 1. Clinical characteristics of ADPKD subjects with normal and abnormal renal function. Results are expressed in median and quintiles.

Parameter	*Normal renal function n=15* *Median (Min.,Q1.Q3, Max)*	*Abnormal renal function n=5* *Median (Min.,Q1.Q3, Max)*
Age (yr)	29 (16, 27, 35, 37)	34 (26, 30, 37, 42)
Sex (%)		
• male	20	80
• female	100	0
24-h ABPM (mm Hg)		
• 24-h SBP	129 (104, 121, 138, 153)	131 (122, 127, 139, 148)
• 24-h DBP	88 (68, 75, 93, 102)	87 (73, 77, 88, 105)
• 24-h PP	44 (36, 41, 47, 51)	49 (43, 43, 50, 52)
• daytime SBP	135 (118, 127, 141, 160)	134 (126, 130, 140, 150)
• daytime DBP	94 (75, 84, 97, 109)	90 (76, 80, 93, 107)
• nighttime SBP	119 (100, 108, 131, 137)	124 (113, 119, 137, 141)
• nighttime DBP	77 (61, 63, 88, 96)	79 (63, 67, 81, 100)
• nocturnal BP reduction (mm Hg)		
SBP	17 (4, 12, 18, 30)	10 (3, 9, 11, 13)*
DBP	16 (1, 11, 21, 30)	13 (7, 8, 13, 14)*
24-h HR (b/min)	71 (55, 62, 77, 90)	73 (60, 61, 76, 96)
AOD (10^{-3}/kPa)	28.5 (8, 20, 39, 57)	16 (8, 12, 17, 36)*
LVMI (g/m^2)	88.9 (62.4, 79.8, 107.6, 119.9)	97.9(69.1, 87.2, 99.1, 100.9)
Creatinine (ml/s/m^2)	95 (52, 65, 106, 146)	155 (99, 113, 254, 325)

*P <0.05 versus renal impairment.

Min = minimum; Q1 = first quintile, Q3 = third quintile, Max = maximum; ABPM = ambulatory blood pressure monitoring; SBP = systolic blood pressure; DBP = diastolic blood pressure; HR = heart rate; AOD = aortic distensibility; LVMI = left ventricular mass index.

TABLE 2. Clinical characteristics of ADPKD dippers and non-dippers

Parameter	*Dipper (n=11)* *Median (Min.,Q1.Q3, Max)*	*Non-dipper (n=9)* *Median (Min.,Q1.Q3, Max)*
Age (yr)	31 (16, 27, 35, 37)	294 (19, 27, 372, 42)
Sex (%)		
• male	66.7	33.3
• female	50.0	50.0
24-h ABPM (mm Hg)		
• 24-h SBP	127 (104, 108, 136, 153)	135 (125, 129, 139, 148)
• 24-h DBP	88 (66, 73, 90, 102)	88 (77, 87, 93, 100)
• 24-h PP	46 (36, 41, 49, 51)	44 (41, 43, 50, 52)
• daytime SBP	135 (118, 122, 141, 160)	139 (127, 132, 140, 150)
• daytime DBP	92 (75, 78, 97, 109)	93 (80, 89, 97, 107)
• nighttime SBP	113 (100, 105, 124, 137)	135 (119, 120, 135, 141)*
• nighttime DBP	76 (61, 61, 80, 96)	81 (67, 77, 88, 100)*
• nocturnal BP reduction (mm Hg)		
SBP	18 (13, 16, 228, 30)	10 (3, 8, 11, 14)**
DBP	17 (1, 14, 21, 23)	11 (6, 7, 13, 16)**
24-h HR (b/min)	75.5 (55, 64, 78, 90)	66.5 (59, 60.5, 71.5, 96)
AOD (10^{-3}/kPa)	26.5 (8, 16, 39, 57)	23 (8, 17, 32, 42)
LVMI (g/m^2)	93.7 (62.4, 82.28, 101.4, 119.9)	87.2(69.1, 78.9, 99.1, 109.9)
Creatinine clearance(ml/s/m^2)	103 (57, 65, 126, 325)	93.0 (52, 82.5, 113, 183)*

*P <0.05 versus rénal impairment.

**P<0.005 versus non-dippers.

Min = minimum; Q1 = first quintile, Q3 = third quintile, Max = maximum; ABPM = ambulatory blood pressure monitoring; SBP = systolic blood pressure; DBP = diastolic blood pressure; HR = heart rate; AOD = aortic distensibility; LVMI = left ventricular mass index.

Circadian BP profile of ADPKD subjects

In the entire cohort of study subjects, 55% exhibited dipping circadian pattern while 45% did not. Non-dippers had significantly higher nighttime SBP and DBP, as well as smaller nocturnal BP reduction than dippers. Their creatinine clearance was lower than that of the dippers (Table 2).

Relationship of renal function to aortic distensibility, cardiac mass and 24-h BP:

Renal function assessed by creatinine clearance, did not correlate with AOD, LVMI and 24-h BP. In the model that examined the relationship between renal function and AOD, there was a positive trend towards a direct relationship between creatinine clearance and AOD, but this relationship did not reach statistical significance (r=0.2824, *P*=0.2930).

Discussion

The Clinical evaluation of our ADPKD patients recorded no valvular or symptomatic intracranial vascular abnormalities. Seventy five percent of our patients had normal and adequate renal function and almost the same percentage were found to have hypertension based on the criteria established by 24 h ABPM blood pressure. Circadian BP readings showed that 45% of our patients were non-dippers and each of them had higher systolic and diastolic blood pressure at night. Furthermore, nocturnal reduction in BP was much smaller than that in the dippers. We also observed that renal function impairment was an additional factor in causing smaller reduction in BP readings. There was no correlation of renal functions to the findings of AOD, LVMI and 24 h BP, however, positive trends were observed among these CV variables to suggest a relationship between glomerular filtration rate and AOD.

Blood pressure and circadian blood pressure profile.

Hypertension by office criteria (BP > 140/90) tends to occur early in patients with ADPKD before any substantial decrease in kidney function.[19] Kelleher et al. reported a highly significant occurrence of hypertension in young patients with ADPKD when compared with patients aged 20 to 34 yr in the U.S. population; hypertension in patients with ADPKD occurs in the absence of abnormal renal function.[20] According to Andersen and colleagues' ABPM values for identifying hypertension [21], 70% of the ADPKD subjects in our study were hypertensive at a mean age of only 30 yr. This agrees with Milutinovic et al. [22] who reported a 29% incidence of hypertension in ADPKD patients who were younger than 30 years of age. Similarly, Gabow et al. [23] found that the mean age of initial diagnosis of ADPKD patients in their study was 29 yr.

Studies of circadian blood pressure (BP) rhythm profile in ADPKD have yielded inconsistent and inconclusive results. Covic et al. [24] reported a widespread abnormality (loss of nocturnal BP reduction) in diurnal BP rhythm in ADPKD patients with renal impairment. Whereas Zeier et al [5] observed no difference in daytime and nighttime BP between children with ADPKD and controls, the daytime and nighttime BP in young adults with ADPKD was higher than that in controls. Seeman and colleagues [25] found no difference in nocturnal BP dip between ADPKD children and normal pediatric population. On the other hand, Li et al.[26] and Cerasola et al. [27] reported that the nocturnal reduction in BP in hypertensive patients with ADPKD was smaller than that in patients with essential hypertension. A blunted or absent nocturnal fall of ambulatory BP has been found to be associated with higher incidence of cardiovascular disease (CVD), a poorer long-term survival and profound autonomic dysfunction.[28] Mercuro et al [29] stated that persistent pressure overload expressed by a blunted nocturnal reduction in BP might be the possible mechanism of increased CV risk.

Left ventricle mass index and aortic distensibility:

The evolution of the ADPKD includes the progressive development of renal cysts and renal insufficiency in some patients. As stated earlier, one of the common extra renal manifestations of ADPKD is CV involvement of which hypertension and LVH have been identified as factors associated with progression to end-stage renal disease (ESRD).[30] Our analysis indicated that the LVMI of ADPKD subjects with impaired renal function was higher than that of the normal renal function subjects, albeit statistically non- significant. This finding concurs with previous studies, which showed that LVH or at least increased LVMI is a common finding in hypertensive ADPKD patients.[6, 31]

Augmented LVMI has been found in children [5] and in asymptomatic subjects with ADPKD before the onset of hypertension or renal dysfunction.[3.] The only plausible explanation for the failure to detect a difference in LVMI between the two groups may be the results of inadequate statistical power.

The result of lower aortic distensibility in ADPKD subjects with evidence of renal function impairment implies a stiffer aorta. This finding is in concordance

TABLE 3. Primary Renal Disease of Registered Patients in Canada 2004 (Number*):

Causes	*Prevalence Number*	*Percent*	*Incidence Number*	*Percent*
Total	30,942	100.0	5,097	100.0
Diabetes	7,636	24.7	1,742	34.2
Glomerulonephritis	7,079	22.9	659	12.9
Vascular Disease	4,014	13.0	939	18.4
Pyelonephritis	2,143	6.9	229	4.5
Polycystic Kidney Disease	2,180	7.1	219	4.3
Drug Induced	373	1.2	62	1.2
Other	3,486	11.3	504	9.9
Unknown	4,031	13.0	743	14.6

*Included in the table hemodialysis , peritoneal and transplant data.

TABLE 4. Stages of chronic kidney disease- As outlined by US National Kidney Foundation.

Stage 1.	Kidney damage, with normal or elevated glomerular filtration rate (GFR) 90ml/min or more {1.50ml/sec per 1.73m^2}
Stage 2.	Kidney damage, with mild decrease in GFR (60-89ml/min) {1.00-1.48ml/sec per 1.73m^2}
Stage 3.	Kidney damage, with moderate decrease in GFR. (30-59ml/min) {0.50-0.98ml/sec per 1.73m^2}
Stage 4.	Kidney damage, with severe decrease in GFR (15-29ml/min) {0.25-0.48ml/sec per 1.73m^2}
Stage 5.	Kidney Failure with GFR less than 15ml/min {<0.25 per 1.73 m^2} Patient needs dialysis.

SI units are given in square brackets

with previous studies. Data from in vitro studies showed increased aortic wall thickness and accumulation of collagen in experimental rat models of moderate and renal insufficiency[32, 33] In a recent clinical study, Mourad et al. [34] reported that increased stiffness of central arteries is statistically associated with decreased creatinine clearance in subjects with mild-to-moderate renal insufficiency, independent of age, BP, and standard risk factors. The mechanisms responsible for arterial stiffness in patients with mild and moderate renal insufficiency are incompletely understood. Ibels et al. [35] and Amann et al. [32] reported that ESRD patients undergo vascular remodeling which is characterized by fibroelastic intimal thickening, calcification of elastic lamellae and ground substance deposition. Arterial remodeling has important impact on the cushioning functions; changes in cushioning function are direct consequence of increased arterial stiffness, resulting in augmented systolic and pulse pressure, LVH [36] and compromised coronary circulation.[37, 38]

Clinical implications

The ADPKD patients represent 5 to 7% of the population with ESRD in Canada (Table 3). The challenges of facing the consequences of impaired renal functions and the subsequent impact of ESRD is complex and in some instances formidable. In Canada, ESRD affected approximately 31,000 Canadians in 2004 whereas eight years earlier, there were 16,000 patients on renal replacement therapy. The Canadian Organ Replacement Register concludes that the incidence of ESRD is increasing at an annual rate of about 7%.[39] Furthermore, Stigant et al [40] has estimated between 600,000 to 1 million persons who may have moderate (stage 3) or advanced renal disease in Canada. These combined figures represent an enormous public health burden. Table 4 describes the stages of chronic kidney disease as classified by the US National Kidney Foundation[41]

Besides a need for early diagnosis of kidney disease and /or its associated hematological and biochemical abnormalities, it is essential that the caregivers or the healthcare teams institute the necessary life style modifications and therapeutic interventions.[40] Nephrology societies and related organizations have drafted guidelines on the follow up and the treatment of these patients, which are appropriate to the stage of chronic kidney disease. These steps are important to retard the progression of renal disease and to prevent serious complications of renal failure.

With respect to our pilot study in patients with ADPKD, we hope that the concept of vascular stiffness especially in the central blood vessels such as aorta should stimulate wider scientific interest. Furthermore, large, randomized controlled studies are needed to examine the effects of early renal function impairment on the vascular biology including arterial remodeling and other metabolic consequences.

Conclusions

This pilot study indicates that the majority of our ADPKD subjects have an unfavorable CV risk profile: hypertension and a non-dipping circadian BP

rhythm.

Moreover, subjects with evidence of renal impairment had reduced aortic distensibility, placing them at an increased risk for cardiovascular morbidity and mortality.

A methodological limitation is a lack of control for covariates such as duration of hypertension, use of antihypertensive agents, and history of chronic anemia, which are known risk factors for LVH. The lack of a control group precludes us from comparing the ambulatory BP, LVMI, and aortic distensibility of ADPKD young adults with their non-ADPKD counterparts. Furthermore, the small sample size seriously limits the statistical power to detect significant differences in variables under investigation. A larger observational study is needed to better characterize the cardiovascular manifestations of autosomal dominant polycystic kidney disease in young adults.

Acknowledgements

This work was supported by a grant-in-aid from the New Brunswick Research Fund, Canada. I thank the cardiologists at the Atlantic Health Sciences Corporation, Saint John, New Brunswick for providing expert technical advice for the ultrasound measurements.

References

1. Dalgaard OZ. Bilateral polycystic disease of the kidneys : a follow-up of two hundred and eighty-four patients and their families. Acta Med Scand 1957; 328 [Suppl]: 1-255.
2. Iglesias CG, Torres VE, Offord KP, Holley KE, Beard CM, Kurland LT. Eipdemiology of adult polycystic kidney disease, Olmsted County, Minnesota 1935-1980. Am J Kidney Dis 1983; 2: 630-9.
3. Chapman AB, Johnson AM, Rainguet S, Hossack K, Gabow P, Schrier RW. Left ventricular hypertrophy in autosomal dominant polycystic kidnet disease. J Am Soc Nephrol 1997; 8: 1292-7.
4. Bardaj A, Martinez-Vea A, Valero A. Cardiac involvement in autosomal-dominant polycystic kidney disease: A hypertensive heart disease. Clin Nephrol 2001; 56: 211-20.
5. Zeier M, Geberth S, Schmidt KG, Mandelbaum A, Ritz E. Elevated blood pressure profile and left ventricular mass in childreb and young adults with autosomal deominant polycystic kidney disease. J Am Soci Nephrol 1993; 3: 1451-7
6. Martinez-Vea A, Bardaj A, Gutierrez C, et al. Exercise blood pressure, cardiac structure, and diastolic function in young normotensive patients with polycystic kidney disease: A prehypertensive state. Am J Kidney Dis 2004; 442: 1-14.
7. Hossack KF, Leddy CL, Johnson AM, Schrier RW, Gabow PA: Echocardiographic findings in autosomal dominant polycystic kidney disease. N Engl J Med 1988; 319: 907-12.
8. Timio M, Monarca C, Pede S, Gentili S, Verdura C, Lolli S. The spectrum of cardiovascular abnormalities in autosomal dominant polycystic kidney disease: A 10-year follow-up in a five-generation kindred. Clin Nephrol 1992; 37: 245-51.
9. Chauveau D, Pirson Y, Verellen-Dumoulin C, Macnicol AM, Gonzalo A, Grunfeld JP. Intracranial aneurysms in autosomal polycystic kidney disease. Kidney International 1994; 45: 1140-6.
10. Bardaj A, Vea AM, Gutierrez C, Ridao C, Richart C, Oliver JA. Left ventricular mass and diastolic function in normotensive young adults with autosomal dominant polycystic kidney disease. Am J Kidney Dis 1998; 32: 970-5.
11. Mahmoud Loghman-Adham , Carlos E. Soto , Tadashi Inagami , Lisa Cassis. The Intrarenal rennin-angiotensin system in autosomal dominant polycystic kidney disease. AJP- Renal Physiology .2004;287:1775-88.
12. Marjan A, van Dijk , Martijn H. Breuning ., Rik Duiser , Leendert A ,van Es and Rudi G.J. Westendorp . No effect of enalapril on progression in autosomal dominant kidney disease. Nephrology Dialysis Transplantation 2003;18:2314-2320
13. Handa SP, Wong J, Wolf HK. The relationship of plasma norepinephrine levels and aortic distensiblity in elderly with isolated systolic hypertension. Am J Hypertens 1995; 8: 422-5.
14. Stefandis C, Stratos C, Bourdoulos H, Kourouklis C, Toutoyzas P. Distensibility of the ascending aorta: comparison of invasive and noninvasive techniques in health men and in men with coronary artery disease. Eur Heart J 1990; 11: 990-6.
15. Devereux RB, Reichek N. Left ventricular hypertrophy. Cardiovasc Res 1980; 1: 55-61.
16. Pierdomenico SD, Bucci A, Constantini F, et al. Circadian blood pressure changes and myocardial ischemia in hypertensive patients with coronary artery disease. J Am Coll Cardiol 1998; 32: 1627-34.
17. Verdecchia P, Schillaci G, Borgioni C, et al. Altered circadian blood pressure profile and prognosis. Blood Press Monit 1997; 2: 347-52.
18. Levey AS, Eckardt KU, Tsukamoto Y, et al. Definition and classification of chronic kidney disease: A position statement from Kidney Disease: Improving Global

Outcomes (KDIGO). Kidney Int 2005; 67:2089-100.
19. Schrier R, McFann K, Johnson A, et al. Cardiac and renal effects of standard versus rigorous blood pressure control in autosomal-dominant polycystic kidney disease: results of seven-year prospective randomized study. J Am Soc Nephrol 2002; 13: 1733-9.
20. Kelleher CL, McMann KK, Johnson AM, Schrier RW. Characteristics of hypertension in young adults with autosomal dominant polycystic kidney disease compared with the general U.S. population, Am J Hypertens 2004; 17: 1029-34.
21. Andersen MJ, Khawandi W, Agarwal R. Home blood pressure monitoring in CKD. Am J Kidney Dis 2005; 45: 994-1001.
22. Milutinovic J, Fialkon PJ, Agodoa LY, Phillips LA, Ruad TG, Bryant JL. Autosomal dominant polycystic kidney disease. Symptoms and clinical findings. QJ Med 1984; 53: 511-22.
23. Gabow PA, Chapman AB, Johnson AM, et al. Renal structure and hypertension in autosomal dominant polycystic kidney disease. Kidney Int 1990; 38; 1177-80.
24. Covic A, Mititiuc I, Gusbeth-Tatomir P, Goldsmith DJ. The reproducibility of the circadian BP rhythm in treated hypertensive patients with polycystic kidnet disease and mild chronic renal impairment – a prospective ABPM study. J Nephrol 2002; 15: 497-506.
25. Seeman T, Dusek J, Vendrichona H, Kyncl M, John U, Miselwitz J, Janda J. Ambulatory blood pressure correlates with renal volume and number of renal cysts in children with autosomal dominant polycystic kidney disease. Blood Pressure Monit 2003; 8: 107-10.
26. Li KWTC, Macnicol AM, Watson ML. Ambulatory blood pressure in hypertensive patients with autosomal dominant polycystic kidney disease. Nephrol Dial Transplant 1997; 12: 2075-80.
27. Cerasola G, Vecchi M, Mule G, et al. Sympathetic activity and blood pressure pattern in autosomal dominant polycystic kidney disease hypertensives. Am J Nephrology 1998; 18: 391-8.
28. Liu M, Takahashi, Morita Y, et al. Non-dipping is a potent predictor of cardiovascular mortality and is associated with autonomic dysfunction in hemodialysis patients. Nephrol Dial Transplant 2003; 18: 563-9.
29. Mercuro G , Zoncu S , Piano D , et al. Estradiol -17 beta reduces blood pressure and restores the normal amplitude of the circadian blood pressure rhythm in postmenopausal hypertension. Am J Hypertens 1998; 11 (8 Pt 1): 909-13.
30. Chapman AB, Shrier RW. Pathogenesis of hypertension in autosomal dominant Polycystic kidney disease. Semin Nephrol 1991;11: 653-60.
31. Saggar-Malik AK, Missouris CG, Gill JS, Singer DRJ, Markandu ND, MacGregor GA. Left ventricular mass in normotensive subjects with autosomal dominant polycystic kidney disease. BMJ 1994; 309: 1617-8.
32. Amann K, Neustib R, Ritz E, Irzyniec T, Weist G, Mall G. Changes of vascular architecture independent of blood pressure in experimental uremia. Am J Hypertens 1995; 8: 409-17.
33. Amann K, Wolf B, Nichols C, et al. Aortic changes in experimental renal failure: hyperplasia or hypertrophy of smooth muscle cells? Hypertension 1997; 29: 770-5.
34. Mourad JJ, Pannier C, Blacher J, et al. Creatinine clearance, pulse wave velocity, carotid compliance and essential hypertension. Kidney Int 2001; 59: 770-5.
35. Ibels LS, Alfrey AL, Huffer WE, Craswell PW, Anderson TJ, Weil R. Arterial calcifications and pathology in uremic patients undergoing dialysis. Am J Med. 1979; 66: 790-6.
36. London GM, Guerin AP, Pannier B, Safar ME, Day M, Metivier F. Cardiac and arterial interactions in end-stage renal disease. Kidney Int 1996; 50: 600-8.
37. Buckberg GD, Towers B, Paglia DE, Mulder DG, Maloney JV. Subendocardial ischemia after cardiopulmonary bypass. J Thorac Cardiovasc Surg 1972; 64: 669-87.
38. Wantanabe H, Phtsuka S, Kaihana M, Sugishita Y. Coronary circulation in dogs with an experimental decrease in aortic compliance. J Am Coll Cardiol 1993; 21: 1497-506.
39 Canadian Organ Replacement Register. Annual report 2004.Ottawa; Canadian institute for Health Information; 2004 (correspondence)
40. Stigant Caroline , Stevens Lesley , Levin Adeera. Nephrology:4. Strategies for the care of adults with chronic kidney disease. Can Med Assoc J. 2003; 168:1553-60
41. K/DOQI clinical practice guidelines for chronic kidney disease: evaluation , classification , and stratification. Kidney Disease Outcome Quality Initiative. Am J. Kidney Dis. 2002; 39 (suppl 2):S1-246

Correspondence:

S. Paul Handa, MD, FRCP, FACP
Department of Nephrology, Atlantic Health Sciences Corporation
Saint John, New Brunswick Canada
E2E 5R6

ORIGINAL ARTICLE

The incidence of nausea and vomiting of pregnancy (NVP): a comparison between depressed women treated with antidepressants and non-depressed women

Pina Bozzo BSc, Gideon Koren MD
Alejandro A. Nava-Ocampo MD
Adrienne Einarson RN

The Motherisk Program, The Hospital for Sick Children, Toronto, Ontario, Canada

Manuscript Submitted: 21st June 2006
Manuscript Accepted: 7th August 2006

Clin Invest Med 2006; 29 (6): 347–350.

Abstract

Background: Nausea and vomiting of pregnancy (NVP) affects up to 80% of pregnant women. In many cases NVP causes changes in family, social, or occupational functioning. Several studies have linked NVP with depression; however, whether depression preceded or resulted from NVP, has not been established.
Objective: To examine whether pregnant women, diagnosed with depression pre-conceptionally, treated with an antidepressant, reported a higher incidence of NVP when compared with pregnant women without depression.
Method*:* In this pilot study, two groups of pregnant women who called the Motherisk Program in Toronto, Canada, were compared. Group 1 was comprised of 179 pregnant women who reported taking an antidepressant for the treatment of depression prior to pregnancy and in the first trimester. Group 2 was comprised of 179 pregnant women with no history of depression. The incidence of NVP in both groups was recorded and compared.
Results: In the depressed group 109/179 (61%) women reported suffering from NVP vs. 121/179 (68%) in the non-depressed group (P=0.1)**.** The logistic regression analysis did not identify any independent variable as significantly explaining NVP.
Conclusion: Depression and treatment with antidepressants prior to and in early pregnancy, does not appear to affect the incidence of NVP.

Background

Nausea and vomiting of pregnancy (NVP) is the most common medical condition of pregnancy, affecting an estimated 60-80% of all pregnant women.[1-2] Depression is also a relatively common medical condition, estimated to affect up to 20% of pregnant women.[3] Several studies have linked NVP with depression.[4-8]

Mazzotta and coworkers examined, as one of their endpoints, the prevalence of psychosocial morbity among 3201 women with NVP. Nausea and vomiting were quantified as mild, moderate or severe. They found that severe NVP was more frequently associated with reported feelings of depression; however information of previous history of depression was not obtained.[4]

Kitamura and colleagues administered a set of questionnaires documenting severity of NVP and included the Zung's Self Rating Depression Score (SDS) to 1329 women who were attending a prenatal clinic of a general hospital. They were the first to conclude that women who were depressed had a significantly higher mean score of nausea and vomiting than the comparison group.[5]

Chou and colleagues examined the relationship between psychosocial factors and frequency of NVP among 113 women. A 20-item Center for Epidemiologic Studies Depression Scale (CES-D) was used to measure depressive symptoms and a checklist documenting the incidence of occasional, frequent or

absence of nausea and vomiting was used to establish NVP. They concluded that pregnant women without nausea and vomiting had significantly lower CES-D than women with frequent nausea and vomiting, suggesting that depressive symptoms had higher correlation with nausea and vomiting.[6]

Swallow and coworkers examined 273 women using the General Health Questionnaire (GHQ), and mood and illness perception visual analogue scales to measure psychiatric morbidity. These scores were compared with the scores of prevalence and severity of NVP, which had been measured using the NVP instrument. They concluded that high scores of GHQ were associated with more severe NVP. [7]

Finally, Andersson and colleagues examined the obstetric outcome of 1495 women diagnosed with antenatal depression and anxiety. They used the Primary Care Evaluation of Mental Disorders system for screening of depressive and anxiety disorders. They concluded that women with a psychiatric diagnosis suffered from more nausea and vomiting of pregnancy then women with no psychiatric diagnosis.[8] However, the main limitation of the above studies [4-8] was that it was not established if depression preceded or resulted from the symptoms of NVP.

The Motherisk Program is a counseling service that provides pregnant, planning and breastfeeding women, and health professionals, evidence-based information on the safety/risk of exposures of prescription and over-the-counter (OTC) medications, natural health products, chemicals, radiation, and infectious diseases. We also have a telephone helpline specifically for information and counselling regarding NVP. At the general Motherisk line, we receive a substantial number calls regarding antidepressant use in pregnancy. In contrast, we observed that few of the women who call our NVP Helpline for advice regarding NVP, have been diagnosed with depression or are currently taking an antidepressant.

Biologically, we could not hypothesise that there would be a reason that depression would play a role in the incidence of NVP. However, we did speculate that perhaps the serotonergic properties of certain antidepressants, may play a role in minimising the symptoms of NVP, as ondansetron, which is indicated for nausea and vomiting also shares some of these pharmacologic properties. In an experimental animal study of memory, however, ondansetron, acting as 5-HT3-receptor antagonist showed opposite effects on learning than other drugs acting as 5-HT1A and 5-HT2-receptor antagonists.[9] Whether these differences observed in the central nervous system effects in experimental animal models may also apply for the control of NVP has not yet been explored.

Our primary objective was to confirm results from other studies that depression diagnosed in pregnancy is probably related to the severity of symptoms. Subsequently, we decided to conduct a study assessing the incidence of NVP in women who have been diagnosed with depression, treated with an antidepressant prior to pregnancy and compare them to a similar group of women who were not diagnosed with depression. This study was approved by The Hospital for Sick Children's Research Ethics Board in Toronto, Ontario, Canada.

Methods

At the Motherisk general line an intake form documenting obstetrical and medical history, including conditions such as depression, medication use and details of NVP during pregnancy is completed for every woman who calls. We conducted a pilot study, where we used a convenience sample of women who had called our line for information on the safety of various drugs and other exposures in pregnancy. The study was comprised of 2 groups of women; a depressed group and a comparison group who did not have a diagnosis of depression. Both groups were obtained by searching our database for women who had contacted the Motherisk line in 2004 or 2005 and were currently pregnant at the time of call. We were only were able to go back 2 years, because this was when we added the NVP question to the intake form. Women with severe medical conditions, or who were on a teratogenic medication, or who called before five weeks of pregnancy were excluded from both groups. They had to be at least five weeks pregnant to be included in the study, as this is usually when NVP starts.

The depressed group was comprised of women with a diagnosis of depression, taking an antidepressant prior to pregnancy. Women with co-morbid psychiatric conditions were excluded. Our comparison group was comprised of women with no history of depression or any other psychiatric conditions. They were matched for maternal characteristics which included age, gestational age at time of call, smoking and alcohol use. The comparison group was selected by entering the next woman who called after the depressed woman, who was similar in every way with the exception that she had not been diagnosed with depression or currently taking an antidepressant medication.

TABLE 1. Rates of NVP in women with depression versus women without depression prior to pregnancy

	Depressed women (n = 179)	*Not depressed women (n = 179)*	*P*
Age (yr)	32.5 ± 4.2	32.4 ± 5.8	0.9
Gravidity (n)	2 (0 – 8)	2 (1 – 8)	0.01
Parity (n)	1 (0 – 5)	0 (0 – 5)	0.02
NVP	109 (61%)	121 (68%)	0.1

TABLE 2. Depression and NVP as outcomes (logistic regression analysis) N = 358

Outcome: NVP	*OR (95%CI)*	*P*
Independent variables		
Depression (yes/no)	0.70 (0.43,1.12	0.1
Gravidity (n)	1.20 (0.93,1.54)	0.1
Parity (n)	0.73 (0.51,1.05	0.09
Age (yr)	0.99 (0.96,1.01)	0.8

The primary endpoint, the incidence of NVP, was compared between the two groups by means of a Chi Square test. Patient demographics which included age, gravidity, and parity were compared between groups by using the Student *t* test or Mann-Whitney rank-sum test if the data were not normally distributed. A logistic regression analysis was performed between NVP as the study outcome, and depression, gravidity, parity, age, and gestational age at call as the independent variables. A *P* value of <0.05 was considered statistically significant.

Results

There were 179 women in each group who fulfilled the criteria for inclusion into the study. There were no differences in the maternal characteristics for which we did not match, with the exception of gravidity (*P*=0.01) and parity (*P*=0.02) between the two groups (Table 1).

The depressed group reported higher gravidity and parity than the comparison group.

There was no difference in the incidence of NVP among women with depression prior to pregnancy, compared to non-depressed women. In the depressed group 109/179 women (61%) suffered from NVP vs. 121/179 (68%) in the control group (p=0.1) (Table 1). The logistic regression analysis did not identify any independent variable as significantly explaining NVP (Table 2).

Discussion

To our knowledge, this is the first study to compare the incidence of NVP between women with depression treated with an antidepressant prior to pregnancy and women not diagnosed with depression.

Previous studies[4-8] indicated an association between depression and severe NVP, however whether NVP preceded depression or depression preceded NVP was not established. Our study revealed that women with depression diagnosed prior to pregnancy did not have a significantly different incidence of NVP. The incidence was slightly less in the depressed women and the lack of significant difference, may be the result of been an inadequate sample size. However, these results do suggest that it is likely that depression is associated with NVP, and is a direct result of the nausea and/or vomiting of pregnancy, as opposed to depression prior to pregnancy.

A study by Atanackovic and colleagues found women who suffered from NVP and also suffered from depression were more likely to be hospitalized than women suffering from NVP but not depression.[10] Another study by Mazzotta and colleagues reported that depression was one of the factors associated with a woman's consideration of termination of pregnancy due to NVP.[11] As well, another and extremely important concern is the increased risk for women with prenatal depression for postpartum depression, which can have tragic consequences. Several studies have demonstrated that prenatal depression is the highest predictor of postpartum depression.[12-14]

With these factors in mind, it becomes apparent that it is very important to ensure the appropriate treatment of NVP to decrease the incidence of depression women during pregnancy. Subsequently, if depression is a result of suffering from NVP, the appropriate treatment should not only decrease the incidence of depression but should also decrease hospitalization rates, termination of wanted pregnancies and the incidence of postpartum depression. This in turn, will have health benefits for the women as well as economic savings for the health care system.

The major limitation of our study was that we were not able to examine the severity of depression or the effectiveness of the treatment, as this may have had some effect on the outcomes. Another limitation may be related to the differences in parity and gravidity, as we did not match the women for these variables. Kallen and colleagues reported that NVP tended to be more severe with higher gravidity.[15] Interestingly, although the depressed group did have a higher gra-

vidity and parity than the non- depressed group, they still suffered less from NVP than the non-depressed group, although the results were not statistically significant: 109/179 (61%) compared with 121/179 (68%) (Table 1).

In previous studies regarding the mechanism of action of antidepressants, it was evidenced that trazodone acts as an antidepressant via antagonist action at serotonin 5-HT2/1C receptors, while fluoxetine likely acts as an antidepressant via inhibition of serotonin uptake.[16] Ondansetron, which is an effective drug used for treatment of nausea and vomiting in the general population, acts as a 5-HT3-receptor antagonist.[9] Our results suggest that the antidepressant treatment does not interfere with the serotoninergic mechanism of control of nausea and vomiting. Indeed, ondansetron and the 5-HT2-receptor antagonist ritanserin exhibited opposite results in an experimental animal model of memory.[9]

In conclusion, depression diagnosed prior to pregnancy treated with an antidepressant, does not appear to affect the course of nausea and vomiting of pregnancy. Severe NVP during pregnancy does appear to increase the incidence of depressive symptoms. It is therefore important to treat NVP effectively to minimise the symptoms.

References

1. Emelianova S, Mazzotta P, Einarson A, Koren G. Prevalence and severity of nausea and vomiting of pregnancy and the effect of vitamin supplementation. Clin Invest Med. 1999; 22: 106-10.
2. Gadsby R, Barnie-Adshead AM, Jagger C. A prospective study of nausea and vomiting during pregnancy. Br J Gen Pract. 1993; 43: 245-8, 325.
3. Marcus SM, Flynn HA, Blow FC, Barry KL. Depressive symptoms among pregnant women screened in obstetric settings. J Women's Health (Larchmt). 2003; 12: 373-80.
4. Mazzotta P, Stewart D, Atanackovic G, Koren G, Magee LA. Psychosocial morbidity among women with nausea and vomiting of pregnancy: prevalence and association with anti-emetic therapy. J Psychosom Obstet Gynaecol. 2000; 21: 129-36.
5. Kitamura T, Sugawara M, Sugawara K, Toda MA, Shima S. Psychosocial study of depression in early pregnancy. Br J Psychiatry. 1996; 168: 732-8.
6. Chou FH, Lin LL, Cooney AT, Walker LO, Riggs MW. Psychosocial factors related to nausea, vomiting, and fatigue in early pregnancy. J Nurs Scholarsh. 2003; 35: 119-25.
7. Swallow BL, Lindow SW, Masson EA, Hay DM. Psychological health in early pregnancy: relationship with nausea and vomiting. J Obstet Gynaecol. 2004; 24: 28-32.
8. Andersson L, Sundstrom-Poromaa I, Wulff M, Astrom M, Bixo M. Implications of antenatal depression and anxiety for obstetric outcome. Obstet Gynecol. 2004; 104: 467-76.
9. Petkov VD, Kehayov R. Effects of agonists and antagonists of some serotonin-receptor subtypes on memory and their modulation by the 5-HT-uptake inhibitor fluoxetine. Acta Physiol Pharmacol Bulg 1994; 20: 83-90.
10. Atanackovic G, Wolpin J, Koren G. Determinants of the need for hospital care among women with nausea and vomiting of pregnancy. Clin Invest Med. 2001; 24: 90-3.
11. Mazzotta P, Stewart DE, Koren G, Magee LA. Factors associated with elective termination of pregnancy among Canadian and American women with nausea and vomiting of pregnancy. J Psychosom Obstet Gynaecol. 2001; 22: 7-12.
12. O'Hara MW, Swain AM. Rates and risk of postpartum depression: a meta-analysis. Int Rev Psychiatry. 1996; 8:37-54.
13. Beck CT. A meta-analysis of predictors of postpartum depression. Nurs Res. 1996; 45: 297-303.
14. Beck CT. Predictors of postpartum depression: an update. Nurs Res. 2001; 50: 275-85.
15. Kallen B, Lundberg G, Aberg A. Relationship between vitamin use, smoking and nausea and vomiting of pregnancy. Acta Obstet Gynecol Scand 2003; 82: 916-20.
16. Marek GJ, McDougle CJ, Price LH, Seiden LS. A comparison of trazodone and fluoxetine: implications for a serotonergic mechanism of antidepressant action. Psychopharmacology (Berl) 1992; 109: 2-11.

Correspondence:

Adrienne Einarson RN
The Motherisk Program,
Division of Clinical Pharmacology/Toxicology
The Hospital for Sick Children
555 University Avenue
Toronto, Ontario, M5G 1X8
Canada
E Mail: einarson@sickkids.ca

ORIGINAL ARTICLE

Multidisciplinarity, interdisciplinarity and transdisciplinarity in health research, services, education and policy: 1. Definitions, objectives, and evidence of effectiveness

Bernard C.K. Choi, PhD, MSc[1]
Anita W.P. Pak, PhD, MA, MEd[2]

[1]Senior Research Scientist, Centre for Chronic Disease Prevention and Control, Public Health Agency of Canada (PHAC), Ottawa, Ontario; Associate Professor, Department of Public Health Sciences, University of Toronto, Toronto, Ontario; Adjunct Professor, Department of Epidemiology and Community Medicine, University of Ottawa, Ottawa, Ontario, Canada. [2] Pak Consulting, Ottawa, Ontario, Canada.

Manuscript Submitted: 4th May 2006
Manuscript Accepted: 7th August 2006

Clin Invest Med 2006; 29 (6): 351–364.

Note: Opinions expressed in this paper are solely those of the authors and do not necessarily represent the views of any agencies, organizations or universities.

Abstract

Background/Purpose. Teamwork involving multiple disciplines is increasingly emphasized in health research, services, education and policy. The terms multidisciplinary, interdisciplinary and transdisciplinary are increasingly used in the literature, but are ambiguously defined and interchangeably used. This paper is the first of two in a series. It discusses the definitions, objectives, and evidence of effectiveness of such teamwork.
Methods. The paper is a literature review based on dictionaries, and Google and MEDLINE (1982-2006) searches.
Results. Multidisciplinarity draws on knowledge from different disciplines but stays within their boundaries. Interdisciplinarity analyzes, synthesizes and harmonizes links between disciplines into a coordinated and coherent whole. Transdisciplinarity integrates the natural, social and health sciences in a humanities context, and transcends their traditional boundaries.
The objectives of multiple disciplinary approaches are to resolve real world or complex problems, to provide different perspectives on problems, to create comprehensive research questions, to develop concensus clinical definitions and guidelines, and to provide comprehensive health services. Multiple disciplinary teamwork has both benefits and drawbacks.
Conclusion. The three terms refer to the involvement of multiple disciplines to varying degrees on the same continuum. The common words for multidisciplinary, interdisciplinary and transdisciplinary are additive, interactive, and holistic, respectively. With their own specific meanings, these terms should not be used interchangeably. The more general term "multiple disciplinary" is suggested for when the nature of involvement of multiple disciplines is unknown or unspecified. While multiple disciplinary teamwork is appropriate for complex problems, it is not always necessary in every single project.

Introduction

There is an increasing emphasis in teamwork that involves multiple disciplines.[1-8] It is generally assumed that efforts to involve more than one discipline are valuable and beneficial.[9-12] Multiple disciplinary approach is emphasized in health research,[13,14] health care services, [15-17] health education,[18-20] and health policy.[21,22] Funding agencies often call for research that involves multiple disciplines.[11,12,23] Hospitals establish multiple disciplinary teams to provide health care. [1,24] Universities establish multiple disciplinary departments and teaching programs.[13] Health policies and programs put their stress on building multiple disciplinary capacity.[21,22]

Terms like multidisciplinary, interdisciplinary, and transdisciplinary have been used to denote efforts that involve several disciplines.[18,25] However, these terms are ambiguously defined[5,9] and often used interchangeably [26] – a situation that Leathard refers to as a "terminological quagmire".[2] Do these terms mean the same or different things? Is there more than one method to bring together people from different disciplines? Are efforts to involve several disciplines really useful? Must we involve multiple disciplines in every project? What are the difficulties in carrying out these efforts? How can this approach be enhanced? However, there has been no previous attempt to comprehensively resolve the confusion.

This paper is a first step to examine the confusing topic. It reviews the definitions of the three terms of interest and then discusses why and under what circumstances multiple disciplinary efforts are useful, with health examples. A second paper will discuss the promotors and barriers, and propose a framework to look for and nurture multiple disciplinary efforts, by locating the place of health sciences in the knowledge universe.

Methods

"Discipline" and the three terms "multidisciplinary", "interdisciplinary", and "transdisciplinary" were looked up in early hard-copy dictionaries from the UK (1944 and 1974),[27,28] US (1975)[29] and Canada (1978),[30] as well as online dictionaries based on "OneLook Dictionary Search"[31] on the Internet.

In addition, Google searches[32], each using one of the three terms of interest plus "definition" as key words, were performed to identify the pertinent online literature on the definitions of the terms, and additional dictionary definitions that "OneLook Dictionary Search" did not locate. Another Google search using all three terms as key words was conducted to find the online literature that include all three terms. Finally, searches of MEDLINE [33] from 1982 to mid-2006 were conducted using a similar strategy to identify relevant publications in the medical and scientific literature.

Results

1. Definitions in the literature

"Discipline" is defined in hard-copy dictionaries as a branch of knowledge,[27,28,30] instruction,[27-30] or learning.[29] Examples are economics and history.[29] "OneLook Dictionary Search" found 19 online dictionaries with English definitions that include the word "discipline". They define "discipline" as a branch of knowledge (10 dictionaries), instruction (5), learning (3), teaching (3) or education (2); or a field of study (3) or activity (1). Examples of a discipline include anthropology, architecture, biology, economics, engineering, history, science, and theology.

The three terms of interest were not found in the 2-volume Oxford Dictionary of 1944,[27] indicating they had probably not been coined at the time (Table 1A). The term "multidisciplinary" was found in the US dictionary of 1975 only, indicating this term probably originated in the US. "Interdisciplinary" was found in all three dictionaries of the 1970's. "Transdisciplinary" was not found in any of the hard copy dictionaries, indicating it is a relatively new term. Dictionaries give the following meanings for the prefixes: Multi - many; more than one. Inter - among; between; mutual, mutually. Trans - across; over; beyond; on the far side of; through.[27-30]

"OneLook Dictionary Search" and additional Google search found 14 online dictionaries with English definitions that include the word "multidisciplinary", 23 dictionaries with the word "interdisciplinary", but only 2 dictionaries with the word "transdisciplinary". Multidisciplinary is defined as "involving" (4 dictionaries) or "of" (4) several disciplines. Interdisciplinary is defined as "involving" (7), "drawing from" (5), "combining" (3), "of" (2), "relating to" (2) or "characterized by" (2) two or more disciplines. The 2 dictionaries, Webster and Merriam-Webster, that defined transdisciplinary, both defined it as interdisciplinary (Table 1A). Thus the 3 terms are rather poorly differentiated in the dictionaries.

Our Internet search found other online documents, mostly glossaries, which define the terms. There is, however, little consistency in definitions of

the three terms, as many of the definitions are virtually interchangeable. Some of the more useful definitions are listed in Table 1B. The earliest online document that defined all three terms was a thesis by Gossman in 1979.[25]

MEDLINE search using the keywords "multidisciplinary interdisciplinary transdisciplinary" found the earliest scientific publication on this topic, written by Thomlinson in 1983, which used the three terms interchangeably.[18] Some definitions in peer-reviewed journals are listed in Table 1C.

TABLE 1. Definitions of multidisciplinary, interdisciplinary and transdisciplinary in the literature.

	A. Dictionary definitions
Multidisciplinary	[Not found] (UK, Oxford, 1944)[27]
	[Not found] (UK, Oxford, 1974)[28]
	Composed of or made up of several specialized branches of learning, as for achieving a common aim (US, Random House, 1975)[29]
	[Not found] (Canada, Funk & Wagnalls, 1978)[30]
	Composed of or combining several usually separate branches of learning or fields of expertise (US, Random House, 1997)[34]
	Involving several academic disciplines or professional specializations (UK, Oxford, 2005)[35]
Interdisciplinary	[Not found] (UK, Oxford, 1944)[27]
	Of more than one branch of learning, e.g. interdisciplinary studies/degrees (UK, Oxford, 1974)[28]
	Combining or involving two or more academic disciplines (US, Random House, 1975)[29]
	Pertaining to or involving two or more branches of knowledge (Canada, Funk & Wagnalls, 1978)[30]
	Combining or involving two or more academic disciplines or fields of study; or two or more professions, technologies, departments, or the like, as in business or industry (US, Random House, 1997)[34]
	Relating to more than one branch of knowledge (UK, Oxford, 2005)[35]
Transdisciplinary	[Not found] (UK, 1944)[27]
	[Not found] (UK, 1974)[28]
	[Not found] (US, 1975)[29]
	[Not found] (Canada, 1978)[30]
	Pertaining to or involving more than one discipline; interdisciplinary (US, Webster, 2006)[36]
	Interdisciplinary (US, Merriam-Webster Online, 2006)[37]

	B. Definitions in the online literature
Multidisciplinary	Group research whereby individuals from different disciplines work together on a common problem, but with limited interaction (Grossman, 1979)[25]
	Relating to or involving several disciplines, such as university, industry, and government (Alberta Inventors and Inventions, 2003)[38]
	Draws on knowledge from different disciplines but stays within the boundaries of those fields (NSERC, 2004)[11]
	A combination of many disciplines in an assignment, not necessarily working in an integrated or coordinated manner (International Rice Research Institute, 2005)[39]
	Involving or combining more than one discipline (University of Southampton, 2005)[40]
	Several branches of medicine, science, or other professions working together toward common goals (Cheshire Medical Center, 2006)[41]
	Involving several areas of medical science and practice (St. Louis Children's Hospital, 2006)[42]
	Of, pertaining to, or arising through the action of many disciplines or professions (Iowa Association of Cardiopulmonary Rehabilitation, 2006)[43]
Interdisciplinary	Joint, coordinated, and continuously integrated research done by experts with different disciplinary backgrounds, working together and producing joint reports, papers, recommendations, and/or plans, which are so tightly and thoroughly interwoven that the specific contributions of each researcher tend to be obscured by the joint product (Grossman, 1979)[25]

	Involves the interaction among two or more different disciplines and occurs at the inter face between disciplines. This may range from the sharing of ideas to full integration of concepts, methodology, procedures, theory, terminology, data, organization of research and training (NSERC, 2004)[11]
	The ability to analyze, synthesize and harmonize links between disciplines into a coordinated and coherent whole (CIHR, 2005)[12]
	Integrates knowledge and modes of thinking from two or more disciplines - such work embraces the goal of advancing understanding in ways that would have not been possible through single disciplinary means (Mansilla and Gardner, 2005)[44]
	A group of professional specialists with expertise in different resources that collaborate to develop and evaluate management alternatives (US Department of Agriculture, 2005)[45]
	At the interface between more than one discipline (University of Southampton, 2005)[40]
	Work which integrates concepts across different disciplines. New disciplines have arisen as a result of such syntheses (Wikipedia, 2006)[46]
	Comprehensive, concerning the cooperation of several disciplines, e.g. physicists with medical practitioners and others (RayMaster International, 2006)[47]
	A course or instructional program involving concepts, knowledge, or faculty from several disciplines (Oregon State University, 2006)[48]
Transdisciplinary	Group research whereby individuals from different disciplines work as a team within a mutually accepted systems organization with an overall set of systems goals (Grossman, 1979)[25]
	An approach that occasions the emergence of new data and new interactions from out of the encounter between disciplines. It offers us a new vision of nature and reality. Trandisciplinarity does not strive for mastery of several disciplines but aims to open all disciplines to that which they share and to that which lies beyond them (Charter of Transdisciplinarity, 1994)[49]
	A specific form of interdisciplinarity in which boundaries between and beyond disciplines are transcended and knowledge and perspectives from different scientific disciplines as well as non-scientific sources are integrated (Vrije University Amsterdam, 2005)[50]
	Of relevance to more than one discipline (University of Southampton, 2005)[40]
	Investigators with different disciplinary backgrounds know enough about other perspectives (conceptually, methodologically, statistically, substantively) to be able to work as a team to study a problem using shared perspectives informed by a range of disciplines (Centre for Addiction and Mental Health, 2006)[51]

C. Definitions in peer-reviewed publications

Multidisciplinary	Multidisciplinary projects are those in which researchers representing different fields contribute methods and ideas from their respective disciplines toward the analysis of a particular research question (Rosenfield, 1992)[52]
	In a multidisciplinary team, health care providers tend to treat patients independently and to share information with each other, while the patient may be a mere recipient of care (Bernard-Bonnin et al, 1995)[53]
	In multidisciplinary research, a variety of disciplines collaborate in one research program without integration of concepts, epistemologies, or methodologies. The degree of integration between disciplines is restricted to the linking of research results (Flinterman et al, 2001)[54]
Interdisciplinary	Interdisciplinary projects involve closer and more frequent collaborative exchanges among researchers drawn from different fields who are working together on a common problem (Rosenfield, 1992)[52]
	An interdisciplinary team aspires to a more profound level of collaboration (than a multi disciplinary team), in which constituents of different backgrounds combining their knowledge mutually complete different levels of planned care (Bernard-Bonnin et al, 1995)[53]
	Interdisciplinary research is a collaboration of several disciplines, but in this case concepts, methodologies, or epistemologies are explicitly exchanged and integrated, resulting in a mutual enrichment (Flinterman et al, 2001)[54]
Transdisciplinary	Transdisciplinary projects are those in which researchers from different fields not only work closely together on a common problem over an extended period but also create a

shared conceptual model of the problem that integrates and transcends each of their sepa rate disciplinary perspectives (Rosenfield, 1992)[52]

Transdisciplinary approaches to human health are defined as approaches that integrate the natural, social and health sciences in a humanities context, and in so doing transcend each of their traditional boundaries (Soskolne, 2000)[55]

Transdisciplinarity is a specific form of inter disciplinarity in which boundaries between and beyond disciplines are transcended and knowledge and perspectives from different scientific disciplines as well as non-scientific sources are integrated (Flinterman et al, 2001)[54]

Multidisciplinarity, Interdisciplinarity, and Transdisciplinarity

"Multidisciplinarity", according to Klein, is a process for providing a juxtaposition of disciplines that is additive, not integrative; the disciplinary perspectives are not changed, only contrasted.[56] An example is physics and history, biology and architecture.[104] A painting by Giotto can be studied not only within art history but also within history of religions, European history, and geometry.[57] Team-taught courses in which faculty provide serial lectures are often multidisciplinary.[58] In a multidisciplinary team dealing with pediatric undernutrition, members function as independent specialists rather than interactive team members. The child or the family is assessed individually by several professionals (such as nursing, social work, psychiatry, nutrition, education, etc) but generally at the discretion of the team leader, usually a physician in medical settings.[24]

"Interdisciplinarity" is a synthesis of two or more disciplines, establishing a new level of discourse and integration of knowledge.[56] For example, when nuclear physics is combined with medicine it leads to new treatments for cancer. When methods from mathematics were transferred to physics, mathematical physics was born, and when they were transferred to meteorological phenomena or stock market processes, they gave rise to chaos theory; transferring methods from particle physics to astrophysics produced quantum cosmology; and from the transfer of computer methods to art, computer art was generated.[57] Interdisciplinary efforts can create new disciplines.[46] For instance, quantum information processing amalgamates elements of quantum physics and computer science; bioinformatics combines molecular biology with computer science.[46] Other examples are biochemistry, ecophilosophy and astrophysics;[59] and psychoimmuno-neuroendocrinology.[60] In an interdisciplinary pediatric undernutrition team, members come together as a whole to discuss their individual assessments and develop a joint service plan for the child.[24]

"Transdisciplinarity" provides holistic schemes that subordinate disciplines, looking at the dynamics of whole systems.[56] Examples are structuralism and Marxism.[58] A transdisciplinary approach on an issue such as pollution or hunger both within and beyond disciplinary boundaries can lead to new perspectives.[59] Ecological economics, defined as the study of the relationship between human housekeeping (economics) and nature's housekeeping (ecology), is transdisciplinary.[61] It is more than drawing on the disciplines of economics and ecology, and requires a common perspective that "transcends" those that are standard in the two disciplines. The traditional perspective of economics needs to be modified to take on board the fact that humans are a species of animal; and the traditional perspective of ecology needs to recognise the role of humanity as a species in the functioning of all ecosystems.[61] In a transdisciplinary pediatric undernutrition team, members share roles as each specialist helps other members to acquire skills related to the specialist's area of expertise; this requires both role release (accepting that others can do what the specialist was trained specifically to do) and role expansion (allowing that one's job can include more than what one was specifically trained to do)[24].

Several authors contrast the three terms (Table 2). According to Rosenfield,[52] multidisciplinary teams work in parallel or sequentially from their specific disciplinary base to address a common problem. Interdisciplinary teams work jointly but still from a discipline-specific base to address a common problem. Transdisciplinary teams work using a shared conceptual framework, drawing together discipline-specific theories, concepts, and approaches to address a common problem. Young[66] suggested that in multidisciplinary teams the different professions work to individually set goals and meet to discuss their progress. In interdisciplinary teams goals are first agreed by the team, whose members then coordinate their input to the common project plan. In transdisciplinary teams, not only goals but skills are shared.

Pain describes the process of evolving approaches to involve several disciplines.[72] Multidisciplinarity is

TABLE 2. Comparison of multidisciplinary, interdisciplinary and transdisciplinary

Multidisciplinary	*Interdisciplinary*	*Transdisciplinary*
Working with several disciplines[9]	Working between several disciplines[9]	Working across[9,57] and beyond[57] several disciplines
Involves more than two disciplines[62,63]	Involves two disciplines[62,63] (i.e. focuses on reciprocal action of disciplines[64])	Involves scientists from relevant disciplines, as well as stakeholders,[61] nonscientists,[54] and non-academic participants[6]
Members from different disciplines working independently on different aspects of a project,[65] working in parallel or sequentially[52]	Members from different disciplines working together on the same project;[65] working jointly[52]	Members from different disciplines working together using a shared conceptual framework[52]
Individual goals in different professions[66]	Shared goals[66]	Shared goals and shared skills[66]
Participants have separate but inter-related roles[5]	Participants have common roles[67]	Participants have role release and role expansion[24]
Participants maintain own disciplinary roles[56,67]	Participants surrender some aspects of their own disciplinary role;[3,5,68] but still maintains a discipline-specific base[52]	Participants develop a shared conceptual framework, drawing together discipline-specific bases[52]
Does not challenge disciplinary boundaries[67]	Blurring of disciplinary boundaries[3]	Transcend the disciplinary boundaries[52,54,55,61]
Summation[5] and juxtaposition[56,69,70] of disciplines	Integration[5,71] and synthesis[56,70] of disciplines	Integration, amalgamation, assimilation, incorporation, unification and harmony of disciplines, views and approaches[60]
Additive,[56] Integrative,[54] Collaborative[54]	Interactive,[11] Integrative,[5,6,11,25,44,46,54,56,60,71] Collaborative[45,52-54]	Holistic,[56,72,73] Transcendental,[50,52,54,55] Integrative,[6,50,52,54,55,60] Collaborative[14]
Graphically analogous to two totally separate circles[59]	Graphically analogous to two partially overlapping circles[59]	Graphically analogous to a third circle that covers two partially overlapping circles[59]
External coherence (i.e. motivated by a desire to focus on clients' needs[5])	Internal coherence[9] (i.e. motivated by a desire to focus on team needs)	
Participants learn about each other[5]	Participants learn about and from each other[5]	
Separate methodologies[54,67,74]	Common methodologies[3,68]	
Instrumental[9]; use of complementary knowledge or perspectives to address a question[11]	Epistemological;[9,68] creation of new knowledge or perspective, even new disciplines[11,46]	
The outcome is the sum of the individual parts[5,75]	The outcome is more than the sum of the individual parts[5,75]	
Graphically analogous to a horizontal series of compartments, each linked by a vertical unidirectional arrow to a higher "control" compartment above[25]	Graphically analogous to a horizontal series of compartments, each linked by a vertical unidirectional arrow to a higher "control" compartment above; and also with horizontal bidirectional arrows between pairs of horizontal compartments[25]	

an approach when experts from different fields work together on a common subject within the boundaries of their own disciplines. However, if they stick to those boundaries they may reach a point where the project cannot progress any further. They will then have to bring themselves to the fringes of their own fields to form new concepts and ideas, and create a whole new, interdisciplinary field. A transdisciplinary team is an interdisciplinary team whose members have developed sufficient trust and mutual confidence to transcend disciplinary boundaries and adopt a more holistic approach.[72]

The Holistic Education Network presents four graphical schemes. "Disciplinary" is visualized as a single circle; "multidisciplinary" is represented by two totally separate circles; "interdisciplinary" is two partially overlapping circles; and "transdisciplinary" is a third circle that covers two partially overlapping circles.[59]

Multidisciplinarity and Interdisciplinarity

According to Pirrie et al[68] the distinction between interdisciplinary and multidisciplinary is based upon three dimensions: numerical, territorial, and epistemological. For the numerical dimension, interdisciplinary involves only two disciplines, and becomes multidisciplinary if more than two disciplines are involved[62,63] (Table 2). Although this may on surface seem like a numbers game[68], interdisciplinary actually focuses on the reciprocal action of two disciplines.[64] For the territorial (or disciplinary boundary) dimension, multidisciplinary often involves little interaction or collaboration across disciplines, like "one sees different facets of a crystal by turning it".[67] Interdisciplinary, on the other hand, is like "you are crossing into another space",[68] and there is blurring of the professional boundaries.[3] For the epistemological dimension, interdisciplinary involves the creation of a new way of working.[68]

Transdisciplinary

Kerne summarized the process to achieve transdisciplinarity.[76] "Trans-" means "across, to or on the farther side of, beyond, over".[77] To go across, beyond, and over disciplinary boundaries, there is a process to assemble disciplines and recombine information. Juxtaposition is a starting point for integration.[69] Juxtaposition and recontextualization draw the mind to puzzle about potential connection between information elements. The next step is recombinant information.[76] Recombination is "the process of taking existing coded compositions, breaking them down into constituent elements, and recombining those elements to form new codings",[76] or new knowledge. When information elements are recombined, if a combination makes sense immediately, the cognitive process is not likely to go anywhere. But, if there are potential relationships that are not immediately clear, the mind tends to work on making sense of them, to find new connections. Sometimes, this process does not lead anywhere. On other occasions, one experiences "Ah-ha!". This is emergence of new ideas and knowledge. Therefore, ambiguous and incongruous juxtaposition of heterogeneous information elements that are related through the operation of a transdisciplinary interface is likely to stimulate the emergence of new knowledge.[76]

A Team

According to Lorimer and Manion, "a team is a small number of consistent people committed to a relevant shared purpose, with common performance goals, complementary and overlapping skills, and a common approach to their work".[78] According to Wiecha and Pollard, an interdisciplinary team is a consistent grouping of people from relevant disciplines, whose interactions are guided by specific team functions and processes to achieve team-defined favourable outcomes.[79] Conventional teams are those whose members interact through traditional meetings and consultations. With modern technology, however, the Internet rapidly becomes a logical platform for supporting interdisciplinary teamwork.[79] Electronic teams are those whose members interact through new communications processes augmented by advances in electronic technology, such as the Internet, Web-based tools, multifunctional software applications, digital audio and video access, lists, forums and websites, that enables teamwork to occur anywhere, at any time.[79,80]

2. Why Pursue Multiple Disciplinarity?

There are several reasons that teamwork involving multiple disciplines are desirable.

To resolve a real world problem

Life is multiple disciplinary. Disciplines are the result of artificial fragmentation of knowledge. Real world problems are rarely confined to the artificial boundaries of academic disciplines.[81] Multiple disciplinary research evolves to meet the demands of many societal, environmental, industrial, scientific and engineer-

ing problems that cannot be adequately addressed by single disciplines alone.[11] There are real world problems and issues that are broader than any single discipline, and can be fruitfully examined in a multiple disciplinary framework.[82]

To resolve a complex problem

In the olden days, problems were relatively simple. A person could build a horse drawn cart or a sail boat by knowing the six simple machines (lever, pulley, wheel and axle, inclined plane, wedge, and screw).[83] With modern technology, however, such thorough knowledge is no longer possible or necessary. Modern automobiles and ocean liners must be built by teams of experts from different disciplines, who themselves can understand and contribute to only a small part of the complex problems.[83] The requirement for multiple disciplinarity is emerging at a time when pace and complexity of science and technology is accelerating, such as in the fields of bioinformatics, hydrogen fuel cells, and broadband infrastructure.[84] Multiple disciplinary teams, with people trained in different fields, are common in complex environments such as research,[11,12,44,80,85] health care,[46,79] teaching,[44,81,82,86] and public health.[9,21]

To provide different perspectives on a problem

Experts from different disciplines read things differently. Quoting from Bernd, "We observe and react to data within the structure of our subject. We read books as 'Englishers' because that is what we are. Quite obviously an historian would not read books in the same manner. In certain sense, he would not be reading the same books".[86]

To create a comprehensive prospective theory-based hypothesis for research

It is necessary to develop "the right question" to lead research.[87] Explicit use of biomedical theory (e.g. sex, physiology) and inexplicit use of social theory (e.g. gender, social capital) to generate hypotheses for test by public health research requires a comprehensive multiple disciplinary approach.[88] After data have been collected and analysed, use of post-hoc theories to explain findings also requires a similar approach. Furthermore, individual disciplines can get "tired", become predictable, and then a crisis of ideas can ensue, after which, progress is difficult.[89] Multiple disciplinary approaches can give the research a "look in" from many different stances at issues in research and reduce "one dimensional" evaluation.[10]

To develop consensus clinical definitions and guidelines for complex diseases and conditions

Consensus of multidisciplinary clinicians is often required to develop definitions for complex diseases such as idiopathic interstitial pneumonias,[90,91] and complex conditions such as multiple chemical sensitivity.[92] For example, the joint American Thoracic Society and European Respiratory Society international consensus definition for idiopathic interstitial pneumonias was worked out by an international multidisciplinary core panel of some 20 pulmonologists, pulmonary pathologists, and thoracic radiologists, and was then approved by an extended review panel of some 65 experts from over 20 countries.[90,91]

To provide comprehensive services such as health care and health education

Multiple disciplinary teamwork is required in health and social care, such as primary health care,[1] education in health care,[4,5,68] and training of medical, dental and nursing students.[19,62,64] Such teamwork can offer a coordinated range of skills, expertise and clinical experience in a setting of interprofessional support.[66] Multidisciplinarity is in some cases required by law[93,94] or guidelines.[95] For example, the US federal Public Law 101-630, the Indian Child Protection and Family Violence Prevention Act, states that "each multidisciplinary team established under this section shall include, but is not limited to, personnel with a background in 1) law enforcement, 2) child protective services, 3) juvenile counselling and adolescent mental health, and 4) domestic violence".[94] In Australia, the multidisciplinary care team for women with breast cancer must minimally include surgery, oncology, pathology, radiology and supportive care.[95] A multidisciplinary team, consisting of an ophthalmologist and an educator, is recommended for comprehensive services to myopic children in developing countries.[96]

3. Teamwork Effectiveness

Some evidence has emerged demonstrating teamwork benefit.[7,97-112] The purported benefits of teamwork include increased learning and development of people and organizations, better utilization of resources and planning for the future, minimization of unnecessary costs, and improving job performance and work quality.[101] Benefits also include discussions among participants, networking, teamwork, gaining new insights and skills, publications, merit points, and a positive effect on careers.[6] In a study of the effect of coordination dimensions (including communication, shared

goals, shared knowledge, problem solving and mutual respect) in orthopedic surgical care, it was found that the more coordination the team demonstrated, the better the patients' postoperative functioning and the shorter the hospital stays.[102] Teamwork for collaborative care and shared care has been shown to improve patient outcomes.[104] In psychiatric disorders management, it has been shown that multidisciplinary care models that include patient education, psychiatric and primary care co-management improve patient outcomes.[105] Patients with depression rated the quality of their care more highly,[106,107] were more adherent to medications,[105,108] had fewer symptomatic days,[109,110] and had decreased depression scores[107,111] when treated collaboratively. Similar results have been reported with panic disorder.[112] How these collaborative models improve outcomes, however, is not clear.

4. Teamwork Ineffectiveness

Other studies provide conflicting results on the effectiveness of teamwork. For example, a literature review reported weaknesses in research rigor, with great inconsistency in terminology and little empirical evidence for the effectiveness of interdisciplinary teams.[113] Another literature review rated the state of interdisciplinary teamwork as poor.[114]

Proposed Definitions

Based on a review of Table 1, we recommend the following definitions:

Multidisciplinarity draws on knowledge from different disciplines but stays within the boundaries of those fields (NSERC, 2004).[11]

Interdisciplinarity analyzes, synthesizes and harmonizes links between disciplines into a coordinated and coherent whole (CIHR, 2005).[12]

Transdisciplinarity integrates the natural, social and health sciences in a humanities context, and in so doing transcends each of their traditional boundaries (Soskolne, 2000).[55]

Based on a review of Table 2, we propose that the terms "multidisciplinary", "interdisciplinary" and "transdisciplinary" are to be used to describe multiple disciplinary approaches to varying degrees on the same continuum.

We further propose that when the exact nature of a multiple disciplinary effort is not known, the specific terms "multidisciplinary", "interdisciplinary" and "transdisciplinary" should be avoided, and the general term "multiple disciplinary" used instead.

Discussion

This paper, the first of two in a series, serves to clarify the terms "multidisciplinary", "interdisciplinary" and "transdisciplinary". In our literature review, these terms are found to be relatively new, poorly differentiated even in dictionaries, confusing and often used interchangeably among many authors, but starting to converge or "gel" towards certain specific meanings.

We conclude that the three terms are used by many authors to refer to the involvement of multiple disciplines to varying degrees on the same continuum. Multidisciplinary, being the most basic level of involvement, refers to different (hence "multi") disciplines that are working on a problem in parallel or sequentially, and without challenging their disciplinary boundaries. Interdisciplinary brings about the reciprocal interaction between (hence "inter") disciplines, necessitating a blurring of disciplinary boundaries, in order to generate new common methodologies, perspectives, knowledge, or even new disciplines. Transdisciplinary involves scientists from different disciplines as well as nonscientists and other stakeholders and, through role release and role expansion, transcends (hence "trans") the disciplinary boundaries to look at the dynamics of whole systems in a holistic way.

Through this review, we believe that common everyday words that can be used to describe the nature of multidisciplinary, interdisciplinary, and transdisciplinary are additive (serving or tending to increase[30]), interactive (producing action on each other[30]), and holistic (producing a material object that has a reality other and greater than the sum of its constituent parts[30]), respectively (Table 3). Mathematically, multidisciplinary is analogous to 2+2=4[115] (additive[116], as in linear combination); interdisciplinary is analogous to 2+2=5[117] (deviation from linear combination, thus requiring an interaction term in a linear model[116,118]); and transdisciplinary is analogous to 2+2=yellow[119] (where the outcome is of a different kind). To these, we add our everyday food examples: multidisciplinary is like a salad bowl (such as a vegetable platter or

TABLE 3. The authors' views on multidisciplinary, interdisciplinary and transdisciplinary

	Multidisciplinary	*Interdisciplinary*	*Transdisciplinary*
Keyword	Additive	Interactive	Holistic
Mathematical example	2+2=4	2+2=5	2+2=yellow
Food example	a salad bowl	a melting pot	a cake

mixed salad, in which the ingredients remain intact and clearly distinguishable); interdisciplinary is like a melting pot (such as a fondue or stew, in which the ingredients are only partially distinguishable); transdisciplinary is like a cake (in which the ingredients are no longer distinguishable, and the final product is of a different kind from the initial ingredients).

Because multidisciplinary, interdisciplinary, and transdisciplinary are starting to gain specific meanings in the literature, we suggest that the use of these terms should be restricted to describe specific approaches with a known or specified level/nature of involvement of multiple disciplines. For the more general situation, the term "multiple disciplinary" should be used when the level/nature of involvement of multiple disciplines is unknown or unspecified.

There is little documented evidence in the literature about the effectiveness of multiple disciplinary teamwork. The limited available evidence indicates conflicting results. In theory, multiple disciplinary approaches are necessary to resolve real world, complex problems. Multiple disciplinary teamwork can provide different perspectives on a problem by generating comprehensive prospective hypotheses before a study, and providing comprehensive post-hoc theories to explain study results. Multiple disciplinary teams have been found with success in situations such as consensus clinical definitions for complex diseases, and comprehensive health care services and health education.

Multiple disciplinary teamwork does not always work, nor does it always deliver what it promises to deliver. Obviously it is not necessary to involve multiple disciplines in every single project. Some projects are so simple and straightforward that they are best performed by one person, or experts from one discipline. Other projects may be more complex and require multiple disciplines, but the expertise may not be available, or even exist. During the project, team conflicts, discipline conflicts and other factors can lead to failure.

A second paper will examine the promotors and barriers for multiple disciplinary teamwork, under what conditions and by what criteria multiple disciplinary efforts are called for, and ways to look for and nurture multiple disciplinary efforts.

References

1. Poulton BC, West MA. Effective multidisciplinary teamwork in primary health care. *J Advanced Nursing* 1993; **18**: 918-25.
2. Leathard A. *Going Inter-Professional: Working together for Health and Welfare.* London: Routledge, 1994.
3. Nolan M. Towards an ethos of interdisciplinary practice. *Br Med J* 1995; **312**: 305-7.
4. Barr H, Hammick M, Koppel I et al. Evaluating interprofessional education: two systematic reviews for health and social care. *Br Educ Res J* 1999; **25**: 533-44.
5. Wilson V, Pirrie A. *Multidisciplinary Teamworking: Beyond the Barriers? A Review of the Issues.* Edinburgh: The Scottish Council for Research in Education, 2000.
6. Tress B, Tress G, Fry G. Researchers' experiences, positive and negative, in integrative landscape projects. *Environ Manage* 2005; **36**: 792-807.
7. Fauchald SK, Smith D. Transdisciplinary research partnership: making research happen! *Nurs Econ* 2005; **23**: 131-5.
8. Mitrany M. Gauging the transdisciplinary qualities and outcomes of doctoral training programs. *J Plan Educ Res* 2005; **24**: 437-49.
9. Whitfield K, Reid C. Assumptions, ambiguities, and possibilities in interdisciplinary population health research. *Can J Public Health* 2004; **95**: 434-6.
10. Peterson CL, Martin C. A new paradigm in general practice research - towards transdisciplinary approaches. The utilisation of multiple research methodologies in general practice research. URL: http://www.priory.com/fam/paradigm.htm. (Accessed on December 17, 2005).
11. Natural Sciences and Engineering Research Council of Canada (NSERC). *Guidelines for the Preparation and Review of Applications in Interdisciplinary Research.* Ottawa: NSERC, 2004. Updated on August 3, 2004. URL: http://www.nserc.ca/professors_e.asp?nav=profnav&lbi=intre (Accessed on April 29, 2006).
12. Canadian Institutes of Health Research (CIHR). *Training Program Grant Guide: Strategic Training Initiative in Health Research.* Ottawa: CIHR, 2005. Updated on May 19, 2005. URL: http://www.cihr-irsc.gc.ca/e/17948.html. (Accessed on April 29, 2006).
13. Slatin C, Galizzi M, Devereaux Melillo K, et al. Conducting interdisciplinary research to promote healthy and safe employment in health care: promises and pitfalls. *Public Health Rep* 2004; **119**: 60-72.
14. Stokols D, Harvey R, Gress J, et al. In vivo studies of transdisciplinary scientific collaboration: lessons learned and implications for active living research.

Am J Prev Med 2005; **28** (2 Suppl 2): 202-13.
15. Carson M, Williams T, Everett A, et al. The nurse's role in the multidisciplinary team. *Eur J Palliat Care* 1997; **4**: 96-8.
16. Nicholson D, Artz S, Armitage A, Fagan J. Working relationships and outcomes in multidisciplinary collaborative practice settings. *Child Youth Care Forum* 2000; **29**: 39-73.
17. Reilly C. Transdisciplinary approach: an atypical strategy for improving outcomes in rehabilitative and long-term acute care settings. *Rehabil Nurs* 2001; **26**: 216-20.
18. Thomlinson R. Interdisciplinarity in the teaching of demography in North America. *Janasamkhya* 1983; **1**: 91-7.
19. Dyer JA. Multidisciplinary, interdisciplinary, and transdisciplinary education models and nursing education. *Nurs Educ Perspect* 2003; **24**: 186-8.
20. Ewing KM, Jones TW. An educational rationale for deaf students with multiple disabilities. *Am Ann Deaf* 2003; **148**: 267-71.
21. Taub A. Transdisciplinary approaches to building the capacity of the public health workforce. *Ethn Dis* 2003; **13** (2 Suppl 2): S45-7.
22. Kassel JD, Ross H. Global tobacco control issues: the role of training in global tobacco control research. *Am J Public Health* 2005; **95**: 946-9.
23. National Institute of Environmental Health Sciences. Health disparities: linking biological and behavioral mechanisms with social and physical environments. Request for application ES-00-00004; 1999 Nov 4. URL: http://www.niams.nih.gov/rtac/funding/grants/rfa/es_00_004.pdf. (Accessed on December 24, 2005).
24. Kessler D. Transdisciplinary approach to pediatric undernutrition. *Developmental Behavioral News* 1999; **8**: online URL: http://www.dbpeds.org/section/fall_1999/kessler.html (Accessed on January 8, 2006).
25. Grossman DG. *A quantitative system for the assessment of initial organizational needs in transdisciplinary research.* MSc thesis. East Lansing, Michigan: Michigan State University, 1979. URL: http://gossman.org/daveg/thchapt1.htm. (Accessed on December 11, 2005).
26. Schoonen M. Multidisciplinary working groups. URL: http://www.research.sunysb.edu/research/workgroups.html. (Accessed on December 17, 2005).
27. *The Shorter Oxford English Dictionary.* Volumes 1 and 2. London: Oxford University Press, 1944.
28. *Oxford Advanced Learner's Dictionary of Current English.* Oxford: Oxford University Press, 1974.
29. *The Random House College Dictionary.* New York: Random House, Inc, 1975.
30. *Funk & Wagnalls Standard College Dictionary.* Canadian Edition. Toronto: FitzHenry & Whiteside Limited, 1978.
31. OneLook Dictionary Search. URL: http://www.onelook.com/ (Accessed on December 11, 2005).
32. Google. URL: http://www.google.com (Accessed on December 9, 2005).
33. MEDLINE. Entrez PubMed. A service of the National Library of Medicine and the National Institutes of Health. URL: http://www.ncbi.nlm.nih.gov/entrez/query.fcgi (Accessed on December 22, 2005)
34. *Random House Unabridged Dictionary.* Copyright Random House, Inc, 1997, on Infoplease. URL: http://www.infoplease.com/dictionary/ (Accessed on December 11, 2005).
35. *Compact Oxford English Dictionary.* Copyright Oxford University Press, 2005. URL: http://www.askoxford.com/ (Accessed on December 11, 2005).
36. *Webster's New Millenium Dictionary of English. Preview Edition (v 0.9.6).* URL: http://dictionary.reference.com/ (Accessed on January 7, 2006).
37. Merriam-Webster OnLine. URL: http://www.m-w.com/ (Accessed on January 7, 2006).
38. Alberta Inventors and Inventions, 2003. URL: www.abheritage.ca/abinvents/glossary.htm (Accessed on January 7, 2006).
39. International Rice Research Institute. Acronyms and Glossary of Rice Terminology, 2005. URL: http://www.knowledgebank.irri.org/glossary/Glossary/M.htm (Accessed on January 7, 2006).
40. University of Southampton, School of Electronics and Computer Science, 2005. URL: http://www.ecs.soton.ac.uk/~sgb/istdocs/ISTglossary.html (Accessed on January 7, 2006).
41. Cheshire Medical Center. Glossary of Oncology Terms. URL: http://www.cheshire-med.com/programs/kingsbur/terms.html (Accessed on January 7, 2006).
42. St. Louis Children's Hospital, Brachial Plexus Palsy Center. URL: http://brachialplexus.wustl.edu/glossary.html (Accessed on January 7, 2006).
43. Iowa Association of Cardiopulmonary Rehabilitation. URL: http://www.iacpr.net/services/glossary_terms.php (Accessed on January 7, 2006).
44. Mansilla VB, Gardner H. Assessing interdisciplinary work at the frontier: an empirical exploration

of "symptoms of quality". Interdisciplines, 2005. URL: http://www.interdiscipline.org/interdisciplinary/papers/6/3/1. (Accessed on December 11, 2005).
45. US Department of Agriculture, 2005. URL: http://www.fs.fed.us/r2/psicc/publications/environmental_impact_statements/eis_uspwp/Glossary.htm (Accessed on January 7, 2006).
46. Wikipedia. URL: http://en.wikipedia.org/wiki/. (Accessed on January 7, 2006).
47. RayMaster International. URL: http://www.raymaster.at/index.htm (Accessed on January 7, 2006).
48. Oregon State University. URL: http://oregonstate.edu/ap/curriculum/terms.html (Accessed on January 7, 2006).
49. Charter of Transdisciplinarity. Translated from French. Adpoted at the First World Congress of Transdisciplinarity, Convento da Arrabida, Portugal, November 2-6, 1994. URL: http://nicol.club.fr/ciret/english/charten.htm. (Accessed on December 10, 2005).
50. Vrije University Amsterdam. URL: http://www.bio.vu.nl/vakgroepen/bens/HTML/transdiciplinair.html (Accessed on December 11, 2005).
51. Centre for Addiction and Mental Health, University of Toronto, Canada. URL: http://www.camh.net/education/strattraintusp_transdisciplinary.html. (Accessed on January 7, 2006).
52. Rosenfield PL. The potential of transdisciplinary research for sustaining and extending linkages between the health and social sciences. *Soc Sci Med* 1992; **35**: 1343-57.
53. Bernard-Bonnin AC, Stachenko S, Bonin D, et al. Self-management teaching programs and morbidity of pediatric asthma: a meta-analysis. *J Allergy Clin Immunol* 1995; **95**: 34-41.
54. Flinterman JF, Teclemariam-Mesbah R, Broerse JEW et al. Transdisciplinary: the new challenge for biomedical research. *Bull Sci* 2001; **21**: 253-66.
55. Soskolne C. Transdisciplinary approaches for public health. *Epidemiology* 2000; **11**: S122.
56. Klein JT. *Interdisciplinarity: History, Theory, and Practice*. Detroit, Mich: Wayne State University Press, 1990.
57. Nicolescu B. The transdisciplinary evolution of learning. Centre International de Recherches et d'Etudes Transdisciplinaires (CIRET), Paris. URL: http://perso.club-internet.fr/nicol/ciret. (Accessed on December 17, 2005).
58. DeZure D. Interdisciplinary teaching and learning. Eastern Michigan University, Ypsilanti, MI, USA. URL: http://www.unm.edu/~castl/Castl_Docs/Packet1/Interdisciplinary%20Teaching%20&%20Learning.html (Accessed on January 8, 2006).
59. Holistic Education Network. Transdisciplinary inquiry incorporating holistic principles. URL: http://www.hent.org/transdisciplinary.htm. (Accessed on December 17, 2005).
60. Fedor-Freybergh PG. Psychoimmuno-neuroendocrinology: an integrative approach to modern philosophy in medicine and psychology. *Neuro Endocrinol Lett* 1999; **20**: 205-13.
61. Common M, Stagl S. *Ecological Economics: An Introduction*. Cambridge: Cambridge University Press, 2005.
62. Carpenter J. Interprofessional education for medical and nursing students: evaluation of a programme. *Med Educ* 1995; **29**: 265-72.
63. Rawson D. Models of interprofessional work: likely theories and possibilities. In: Leathard A, ed. *Going Interprofessional. Working Together for Health and Welfare*. London: Routledge, 1994.
64. Carpenter J. Doctors and nurses: stereotypes and stereotype change in interprofessional education. *J Interprofessional Care* 1995; **9**: 151-61.
65. Mallon WT, Burnton S. The functions of centers and institutes in academic biomedical research. *Analysis in Brief* 2005; **5**(1), June 2005, Association of American Medical Colleges (AAME) URL: http://www.ammc.org/data/aib/aibissues/aibvol5_no1.pdf. (Accessed on December 11, 2005).
66. Young CA. Building a care and research team. *J Neurol Sci* 1998; **160** Suppl 1: S137-40.
67. Clarke PG. A typology of multidisciplinary education in gerontology and geriatrics: are we really doing what we say we are? *J Interprofessional Care* 1993; **7**: 217-27.
68. Pirrie A, Wilson V, Elsegood J et al. *Evaluating Multidisciplinary Education in Health Care*. Edinburgh: The Scottish Council for Research in Education, 1998.
69. Nowotny H. The potential of transdisciplinarity. In: Rethinking Interdisciplinarity, January, 2004. URL: http://www.interdisciplines.org/interdisciplinarity/papers/5. (Accessed on December 17, 2005).
70. Klein JT. *Crossing Boundaries: Knowledge, Disciplinarities, and Interdisciplinarities*. Charlottesville: University of Virginia Press, 1996.
71. Wilmot S. Professional values and interprofessional dialogue. *J Interprofessional Care* 1995; **9**: 257-65.
72. Pain E. Multidisciplinary research: Today's hottest buzzword? January 3, 2003. URL: http://science-

careers.sciencemag.org/career_development/issue/articles/2100/multidisciplinary_research_today_s_hottest_buzzword (Accessed on January 8, 2006).
73. Anderson GW, Monsen RB, Rorty MV. Nursing and genetics: a feminist critique moves us towards transdisciplinary teams. *Nurs Ethics* 2000; **7**: 191-204.
74. Klein JT, Grossenbacher-Mansuy W, Haberli R, et al, eds. *Transdisciplinary - Joint Problem-solving among Science, Technology and Society: An Effective Way of Managing Complexity.* Basel: Birkhauser Verlag, 2001.
75. Parker GM. *Cross-functional teams: working with allies, enemies and other strangers.* San Francisco: Jossey-Bass Publishers, 1994.
76. Kerne A. Doing interface ecology: the practice of metadisciplinary. Proceedings of the ACM SIGGRAPH 05 electronic art and animation catalog, August 1-4, 2005, Los Angeles, California, USA. URL: portal.acm.org/citation.cfm?id=1086057.1086144 (Accessed on January 8, 2006).
77. *Oxford English Dictionary on Compact Disk.* Second Edition. Oxford: Oxford University Press, 1992.
78. Lorimer W, Manion J. Team-based organizations: leading the essential transformation. *PFCA Rev* 1996; Spring: 15-9.
79. Wiecha J, Pollard T. The interdisciplinary eHealth Team: chronic care for the future. *J Med Internet Res* 2004; **6**: Article e22. URL: http://www.jmir.org/2004/3/e22. (accessed on December 11, 2005).
80. Heintz C, Gloria Origgi. Rethinking interdisciplinary: emergent issues. Interdisciplines URL: http://www.interdisciplines.org/interdisciplinarity/papers/11. (Accessed on December 17, 2005).
81. Krahn G. Interdisciplinary culture - a result not a goal. *Proceedings of the Interdisciplinary Workshop on Core Mathematics: Considering Changes in the First Two Years of Undergraduate Mathematics.* November 4-7, 1999, West Point, NY, USA. URL: http://www.dean.usma.edu/math/activities/ilap/workshops/1999/files/krahn.pdf (Accessed on January 8, 2006).
82. Seipel M. Interdisciplinary: An Introduction. Truman State University, Kirksville, Missouri. URL: http://www2.truman.edu/~mseipel/Interdisciplinarity.pdf (Accessed on January 8, 2006).
83. Choi BCK. Future challenges for diagnostic research: striking a balance between simplicity and complexity. *J Epidemiol Community Health* 2002; **56**: 334-5.
84. Mandel M, Hamm S. Industry and academia weigh in. *Business Week.* October 11, 2004, page 1. URL: http://www.businessweek.com/magazine/content/04_41/b3903487.htm. (Accessed on January 8, 2006).
85. Sillitoe P. Interdisciplinary experiences: working with indigenous knowledge in development. *Interdisciplinary Sci Rev* 2004; **29**: 6-23.
86. Bernd D. Prolegomenon to a definition of interdisciplinary studies: the experience at Governors State University. *ADE Bulletin*, 1971; **031**: 8-14.
87. Syme G. Integration initiatives at CSIRO: reflections of an insider. *J Res Practice* 2005; **1**: Article M3. URL: http://jrp.icaap.org/content/v1.2/syme.html. (Accessed on December 17, 2005).
88. Frohlich KL, Mykhalovskiy E, Miller F, et al. Advancing the population health agenda. *Can J Public Health* 2004; **95**: 392-5.
89. Nissani M. Ten cheers for interdisciplinarity: the case for interdisciplinary knowledge and research. *Soc Sci J* 1997; **34**: 201-16.
90. Travis WD, King TE, Bateman ED, et al. ATS/ERS international multidisciplinary consensus classification of the idiopathic interstitial pneumonias: general principles and recommendations. *Am J Respir Crit Care Med* 2002; **165**: 277-304.
91. Demedts M, Costabel U. ATS/ERS international multidisciplinary consensus classification of the idiopathic interstitial pneumonias. *Eur Respir J* 2002; **19**: 794-6.
92. Environmental Law Center. Multiple Chemical Sensitivity: A 1999 Consensus. URL: http://www.elc.org.uk/pages/healthmcsdefinition.htm. (Accessed on December 11, 2005).
93. *California Welfare and Institutions Code.* Division 4.5. Services for the Developmentally Disabled. Chapter 1. General Provisions. URL: http://www.dds.ca.gov/statutes/WICSectionView.cfm?Section=4500-4519.7.htm (Accessed on December 11, 2005).
94. US federal Public Law 101-630. The Indian Child Protection and Family Violence Prevention Act. Title 25. Indians. Chapter 34. Indian Child Protection and Family Violence Prevention. Sec. 3209. Indian Child Resource and Family Services Centers. URL: http://uscode.house.gov/download/pls/25C34.txt (Accessed on December 11, 2005).
95. Zorbas H, Barraclough B, Rainbird K et al. Multidisciplinary care for women with early breast cancer in the Australian context: what does it mean? *Med J Aust* 2003; **179**: 528-31.

96. Siddiqui AP, Backman O, Awan HR. Multidisciplinary approach in the development of comprehensive low-vision services in developing countries. *Eastern Mediterranean Health J* 1997; **3**: 149-53.
97. Mcghee SM, Mcinnes GT, Hedley AJ, et al. Coordinating and standardizing long-term care: evaluation of the west of Scotland shared-care scheme for hypertension. *Br J Gen Pract* 1994; **44**: 441-5.
98. Shortell SM, Zimmerman JE, Rousseau DM, et al. The performance of intensive care units: does good management make a difference? *Med Care* 1994; **32**: 508-25.
99. Young GJ, Charns MP, Desai K, et al. Patterns of coordination and clinical outcomes: a study of surgical services. *Health Serv Res* 1998; **33**: 1211-36.
100. Risser DT, Rice MM, Salisbury ML, et al. The potential for improved teamwork to reduce medical errors in the emergency department. The MedTeams Research Consortium. *Ann Emerg Med* 1999; **34**: 373-83.
101. Ingram H, Desombre T. Teamwork in health care: lessons from the literature and from good practice around the world. *J Manag Med* 1999; **13**: 51-8.
102. Gittell JH, Fairfield KM, Bierbaum B, et al. Impact of relational coordination on quality of care, post-operative pain and functioning, and length of stay: a nine-hospital study of surgical patients. *Med Care* 2000; **38**: 807-19.
103. Heinemann GD, Zeiss AM. *Team Performance in Health Care: Assessment and Development.* New York: Plenum, 2002.
104. Katon W, Russo J, Von Korff M, et al. Long-term effects of a collaborative care intervention in persistently depressed primary care patients. *J Gen Intern Med* 2002; **17**: 741-8.
105. Katon W, Von Korff M, Lin E, et al. Collaborative management of achieve depression treatment guidelines. *J Clin Psychiatry* 1997; **58** Suppl 1: 20-3.
106. Katon W, Robinson P, Von Korff M, et al. A multifaceted intervention to improve treatment of depression in primary care. *Arch Gen Psychiatry* 1996; **53**: 924-32.
107. Unutzer J, Katon W, Callahan CM, et al. Collaborative care management of late-life depression in the primary care setting: a randomized controlled trial. *JAMA* 2002; **288**: 2836-45.
108. Liu C, Hedrick SC, Chaney EF, et al. Cost-effectiveness of collaborative care for depression in a primary care veteran population. *Psychiatr Serv* 2003; **54**: 698-704.
109. Katon W, Von Korff M, Lin E, et al. Stepped collaborative care for primary care patients with persistent symptoms of depression: a randomized trial. *Arch Gen Psychiatry* 1999; **56**: 1109-15.
110. Schoenbaum M, Unutzer J, Sherbourne C, et al. Cost-effectiveness of practice-initiated quality improvement for depression: results of a randomized controlled trial. *JAMA* 2001; **286**: 1325-30.
111. Katon W, Von Korff M, Lin E, et al. Collaborative management to achieve treatment guidelines: impact on depression in primary care. *JAMA* 1995; **273**: 1026-31.
112. Katon W, Roy-byme P, Russo J, et al. Cost-effectiveness and cost offset ofa collaborative care intervention for primary care patients with panic disorder. *Arch Gen Psychiatry* 2002; **59**: 1098-104
113. Schofield RF, Amodeo M. Interdisciplinary teams in health care and human services settings: are they effective? *Health Soc Work* 1999; **24**: 210-9.
114. Mccallin A. Interdisciplinary practice - a matter of teamwork: an integrated literature review. *J Clin Nurs* 2001; **10**: 419-28.
115. Golden Gate University. University Library. 2+2=4. Intellectual Freedom Page. URL: http://www.buraphalinux.org/~twatchai/OLDWEB/hotel/index-4.html (Accessed on January 19, 2006).
116. Last JM. *A Dictionary of Epidemiology.* New York: Oxford University Press, 1995.
117. Sharp A. Making two plus two equal five. *Evaluation J Australasia* 2001; **1**: 41-5.
118. Rothman KJ, Greenland S. *Modern Epidemiology.* Philadelphia, PA: Lippincott, Raven Publishers, 1998.
119. Dixon MJ, Smilek D, Cudahy C, Merikle PM. Five plus two equals yellow. *Nature* 2000; **406**: 365

Correspondence:

Dr. Bernard C.K. Choi,
Centre for Chronic Disease Prevention and Control, Public Health Agency of Canada, Government of Canada, P.L. 6701A, 120 Colonnade Road, Ottawa, Ontario, Canada K1A 1B4
E-mail: Bernard_Choi@phac-aspc.gc.ca.

ORIGINAL ARTICLE

Effects of Losartan on Urinary Secretion of Extracellular Matrix and Their Modulators in Type 2 Diabetes Mellitus Patients with Microalbuminuria

Vincent Woo MD, Lian-Song Ni MD
Dixie Hak RN, Lori Berard RN
Fuqin Zhu BSc, Shagufta Khan MD
Guoping M. Ma MD, Brian Penner MD
Garry X. Shen MD PhD

Department of Internal Medicine, University of Manitoba, Winnipeg, Canada

Manuscript Submitted: 18th July 2006
Manuscript Accepted: 3rd September 2006

Clin Invest Med 2006; 29 (6): 365–372.

Abstract:

Purpose: Angiotensin II receptor Type 1 antagonists postpone the development of nephropathy in type 2 diabetes mellitus (DM). We hypothesize that Losartan may ameliorate renal function in diabetic patients through the regulation on the generation of transforming growth factor (TGF)-β and fibrinolytic regulators.

Methods: Twenty-two type 2 DM patients with microalbuminuria were treated with 50-100 mg/day of Losartan for 6 months. Urinary secretion of TGF- , plasminogen activator inhibitor-1 (PAI-1), tissue and urokinase plasminogen activators (tPA and uPA) fibronectin, collagen IV and plasma levels of TGF- β, PAI-1, tPA and uPA of the patients before and after the treatment were analyzed using enzyme-linked immunoabosorbance assay.

Results: Losartan effectively reduced arterial blood pressure and urinary albumin excretion. The levels of TGF-β in urine, but not in plasma, were reduced after 2, 4 and 6 months of the treatment (-32% to -48%, $P<0.05$ or 0.01). Urinary or plasma levels of PAI-1, tPA or uPA, and urinary secretion of fibronectin or collagen IV were not significantly altered by Losartan treatment. Urinary levels of collagen IV positively correlated with uPA, and that of fibronectin negatively correlated with PAI-1 in the patients ($P<0.01$). Urinary TGF-β negatively correlated uPA in urine of the patients ($P<0.01$).

Conclusion: Losartan reduced urinary excretion of TGF-β and albumin in type 2 DM patients with microalbuminuria. Fibrinolytic regulators and TGF-β are implicated in the regulation of ECM turnover in kidneys of the patients with diabetic nephropathy.

Introduction

Diabetes mellitus (DM) has become epidemic in most developed and many developing countries. Diabetes is the single largest cause of end-stage renal disease (ESRD) requiring dialysis or kidney transplantation in North America.[1] The lifespan of patients with ESRD is 20%-30% shorter than that in general population.[2] Microalbuminuria and hypertension occur from 5 to 15 yr after the onset of diabetes, and are considered as clinical markers for the onset of diabetic nephropathy.[3] The majority of DM or diabetic nephropathy is type 2 DM. The pathology of diabetic nephropathy is characterized by glomerular membrane thickening and mesangial expansion. Excess accumulation of extracellular matrix (ECM) and mesangial expansion are major pathological findings in the kidneys of diabetic patients.[7]

Transforming growth factor (TGF)-β is a fibrogenic growth factor and regulates the generation of multiple types of ECM proteins.[8] Fibrinolytic regulators, urokinase plasminogen activator (uPA),

tissue plasminogen activator (tPA) and their major physiological regulator, plasminogen activator inhibitor-1 (PAI-1), are abundant in kidneys. The fibrinolytic regulators are involved in the regulation of ECM turnover in kidneys. Renin-angiotensin system plays a crucial role in the pathophysiology of diabetic nephropathy. The results from several large-scale randomized controlled trials indicated that angiotensin II receptor type 1 (AT1) antagonists, including Losartan, reduced urinary albumin excretion and the requirements for dialysis or kidney transplantation in type 2 DM patients (4-6). Previous results on the effects of Losartan on urinary secretion of TGF-β or ECM in diabetic patients were inconsistent.[9-12] High concentrations of glucose altered the production of uPA, tPA and PAI-1 in cultured human mesangial cells.[13] AT1 antagonists prevented angiotensin II-induced expression of PAI-1 in rat kidneys.[14] The impact of AT1 antagonists on urinary secretion of fibrinolytic regulators and the relationship between fibrinolytic regulators and ECM in diabetic patients remains unknown.

The present study examined urinary secretion of TGF-β, ECM and fibrinolytic regulators in type 2 DM patients with microalbuminuria before and after Losartan treatment. The relationships between ECMs and their regulators in the patients were further analyzed.

Methods

Subjects

Patients were recruited from the Diabetes Clinics or Diabetes Research Group at the Health Sciences Centre and the Rossmere Medical Clinic in Winnipeg. Sixty-three patients were screened for eligibility to the study and were instructed to collect 3-times of 24 h urine for measuring urine albumin. The inclusion criteria of the study included a confirmed diagnosis of type 2 DM based on the classification of World Health Organization[15] and microalbuminuria (20-200 μg/min) from at least two out of the three 24-h urine collections. Patients who had type 1 diabetes, macroalbuminuria, serious medical conditions, massive obesity or pregnancy were excluded from the study. Twenty-two patients (16 male and 6 female, 40-72 yr) meeting the criteria were enrolled in the study. All participants signed an informed consent approved by the Research Ethics Board of the University of Manitoba. Among the participants, 14 of them had a diagnosis of hypertension (≥140/90 mmHg), 6 with retinopathy, 5 with neuropathy, and 4 with coronary artery disease (Table 1). Eighteen of the participants were on metformin, glyburide or both, and 4 of others were receiving insulin injections. Five patients received calcium channel antagonists or β-blockers. One patient was on dietary intervention alone without any medication. Nine patients were receiving one type of angiotensin I converting enzyme (ACE) inhibitors at the enrollment. Four others were on AT1 antagonists. For the patients who were taking ACE inhibitors or AT1 antagonists, one month of washout period was applied before the start of the study medication.

TABLE 1 Baseline characteristics of subjects

Parameters	*Subjects*
Number	22
Sex (M/F)	16/6
Age (yr)	57.1 ± 8.4
During of Diabetes (yr)	13.7 ± 9.8
Body mass index (kg/M^2)	32.5 ± 5.6
Microalbuminuria	22
Hypertension	14
Hyperlipidemia	14
Retinopathy	6
Neuropathy	5
Coronary artery disease	4

Values present as mean ± SD or the number of cases.

Regimen

The patients received Losartan (Merck Frosst Canada) from research nurses at an initial dose of 50 mg/day. The dosage of Losartan was titrated to 100 mg/day after two weeks if no sign of intolerance to Losartan appeared. The later dosage was maintained until the end of the study. No placebo or randomized control group was involved in the study. Hypoglycemic treatment or other anti-hypertensives beside renin-angiotensin antagonists were not interrupted in the patients during the study.

Follow-up visits, sample collection and general biochemical analysis

Follow-up visits of the participants were arranged at the enrollment, titration, 1, 2, 4 and 6 months after the start of the Losartan treatment. Data on their blood pressure and general health were collected during each visit. Blood and urine samples were collected

at enrollment, 2, 4 and 6 months after the start of the treatment. Urinary albumin, blood HbA1c, fasting blood glucose, serum lipid profile, liver enzymes, serum and urine creatinine at entry and 6 months after the start of the treatment were analyzed in the Department of Clinical Chemistry in Health Sciences Centre in Winnipeg.

Enzyme-linked immunosorbant assay (ELISA)

Plasma and urinary TGF-β, PAI-1, uPA, tPA, urine fibronectin and collagen IV at enrollment, 2, 4 and 6 months after the start of the treatment were analyzed in research laboratory using ELISA. Blood samples were collected in EDTA containing vacuum tubes for measuring the fibriolytic regulators in plasma, and in heparin-tubes for measuring plasma TGF-β. Blood was centrifuged at 1000xg for 10 min at 4°C within 1 hour. Plasma was collected and stored at –70°C. Urine samples were freshly frozen and stored at –70°C. Plasma and urine PAI-1, uPA and tPA were analyzed using IMUBIND ELISA kits for PAI-1, uPA or tPA from America Diagnostica Inc. as previously described.[16] Plasma and urine TGF-β were analyzed using the immunoassay kits from Biosource International Inc. Urine fibronectin was measured using QuantiMatrix™ Human Fibronectin ELISA kits from Chemicon International. Urine collagen IV was measured with Biotrin Urinary Collagen IV enzymatic immunoassay kits from Daiichi Fine Chemical Co. Ltd., Japan.

Statistical analysis

Data are presented as means ± standard deviation (SD). Differences between two groups were analyzed using paired *t*-test. Correlations were analyzed using Pearson coefficient and regression analysis (two-tailed). Significant level was preset at *P* value <0.05. SPSS11.0 software was used for the statistical analysis.

Results

Changes in arterial blood pressure

The levels of arterial systolic and diastolic blood pressure (DBP) were reduced by 7.4% and 16.4%, respectively, in the type 2 DM patients after 1 month of Losartan treatment ($P<0.05$ or 0.01). The blood pressure lowering effect of Losartan was continued to 6 months after the start of the treatment (Fig. 1). The levels of blood pressure were normalized to ≤130/80 mmHg in 41% of the patients at end of their treatment.

TABLE 2 Clinical chemical variables in patients with type 2 diabetes mellitus before and after Losartan treatment

Variables	*Before treatment*	*After treatment*
24h urine albumin (μg/min)	76.0 ± 73.6	49.7 ± 93.0*
HbA1c (%)	7.7 ± 1.9	8.2 ± 2.3
Triglycerides (mmol/L)	2.3 ± 1.4	2.2 ± 1.9
Total cholesterol (mmol/L)	4.4 ± 0.9	4.3 ± 1.4
HDL-cholesterol (mmol/L)	1.3 ± 0.5	1.3 ± 0.5
LDL-cholesterol (mmol/L)	2.0 ± 0.9	1.9 ± 0.9
Serum potassium (mmol//L)	4.2 ± 0.5	4.2 ± 0.5
Serum Creatinine (mmol/L)	80.7 ± 40.0	83.7 ± 43.6
Alanine transaminase (U/L)	29.8 ± 14.1	30.5 ± 16.9
Aspartate transaminase (U/L)	24.7 ± 9.8	23.8 ± 9.4

Values presented in mean ± SD (n = 22). *: $P<0.05$ versus before treatment.

Effect of Losartan on albuminuria and general biochemical variables

After 6-months of Losartan treatment, the average urinary albumin excretion rate from the 24 h urine collections was reduced by 30% in the diabetic patients compared with baseline ($P<0.05$). The urinary albumin excretion in 27% of the patients was normalized to the levels of <20 μg/min after 6 months of the Losartan treatment. The levels of serum creatinine, HbA1c, total cholesterol, triglycerides, HDL-cholesterol, LDL-cholesterol, serum potassium, serum creatinine and liver enzyme activities were not significantly altered during or after the Losartan treatment (Table 2).

TGF-β and fibrinolytic regulators

Plasma levels of TGF-β in diabetic patients were not altered by the Losartan treatment. However, the levels of TGF-β in urine during and after the Losartan treatment were lower than that before the treatment ($p<0.05$ or 0.01). After 2 months of the treatment, the levels of urine TGF-β were 35% lower than that prior to the Losartan treatment, which were further decreased after 4 or 6 months of the treatment (-48% and -42% versus basal level). The levels of plasma or urinary uPA, tPA and PAI-1 in patients before and after the Losartan treatment were not significantly different (Table 3).

Urinary secretion of fibronectin and collagen IV

TGF-β is one of the most important physiological modulators for the production of ECM in human body.[8] Collagen IV and fibronectin have been detected in the ECM of glomerular mesangial complex.[17] We

TABLE 3 Plasma and urinary transforming growth factor (TGF-β), plasminogen activator inhibitor-1 (PAI-1), urokinase plasminogen activator (uPA), tissue plasminogen activator (tPA), urinary fibronectin and collagen IV in patients with type 2 diabetes mellitus before and after Losartan treatment

Variables	*Before*	*After Losartan treatment*		
		2 months	*4 months*	*6 month*
Plasma TGF-β (pg/ml)	2289 ± 1093	2019 ± 1097	2511 ± 1285	2356 ± 807
Urine TGF-β (pg/ml)	256 ± 326	173 ± 308**	141 ± 192**	191 ± 308*
Plasma PAI-1 (ng/ml)	127 ± 89	125 ± 70	132 ± 84	162 ± 98
Urine PAI-1 (ng/ml)	142 ± 129	140 ± 80	157 ± 107	161 ± 85
Plasma tPA (ng/ml)	10.5 ± 3.8	10.8 ± 5.1	11.1 ± 4.7	11.2 ± 4.7
Urine tPA (ng/ml)	416 ± 612	565 ± 796	495 ± 572	515 ± 612
Plasma uPA (ng/ml)	0.84 ± 0.28	0.85 ± 0.28	0.89 ± 0.33	1.50 ± 2.91
Urine uPA (ng/ml)	17.9 ± 15.6	16.8 ± 21.1	21.2 ± 28.6	22.3 ± 28.1
Urine fibronectin (ng/ml)	162 ± 112	187 ± 188	225 ± 299	158 ± 107
Urine collagen IV (µg/L)	6.1 ± 4.5	5.8 ± 4.5	4.7 ± 3.6	4.7 ± 4.9

Values presented in mean ± SD (n = 22 for plasma, n = 20 for urine analysis due to 2 patients failed to submit complete urine samples on time). *: $P = 0.05$ versus before; **: $P = 0.01$ versus before.

TABLE 4 Correlations between biochemical and clinical variables

Variables	*HbA1c*	*U-TGF*	*P-TGF*	*U-PAI*	*P-PAI*	*U-uPA*	*P-uPA*	*U-tPA*	*P-tPA*	*U-fibron*	*U-CIV*	*U-album*	*SBP*
U-TGF	0.312*												
P-TGF	0.293*	NS											
U-PAI	0.416**	NS	NS										
P-PAI	0.363**	0.296*	NS	NS									
U-uPA	NS	-0.302**	NS	NS	NS								
P-uPA	NS	NS	NS	NS	NS	0.324**							
U-tPA	NS	NS	NS	NS	NS	0.441**	NS						
P-tPA	NS	-0.247*	NS	NS	NS	NS	NS	0.506**					
U-fibron	NS	NS	NS	-0.358**	0.249*	NS	NS	-0.233*	NS				
U-CIV	NS	NS	NS	NS	NS	0.418**	0.386**	NS	NS	0.454**			
U-album	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS		
SBP	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	
DBP	NS	NS	0.235*	0.250*	NS	NS	NS	NS	NS	NS	NS	NS	0.536**

HbA1c: hemoglobulin A1c; U-: urine; P-: plasma; TGF: transforming growth factor-β; PAI: plasminogen activator inhibitor-1; uPA: urokinase plasminogen activator; tPA: tissue plasminogen activator; fibron: fibronectin; CIV: collagen IV; album: albumin; SBP: systolic blood pressure; DBP: diastolic blood pressure; NS: not significant. *, **: $P<0.05$ or 0.01 for r values (n = 88 for analyses with plasma variables and SBP or DBP, n = 80 for analyses with U-TGF, U-PAI, U-uPA, U-tPA, U-fibron and U-CIV due to two patients failed to submit complete urine specimens; n = 44 for analyses with HbA1c and U-albumin, which were only collected at entry and 6 months after the treatment).

measured the levels of collagen IV and fibronectin in urine of the diabetic patients before and after the Losartan treatment. The levels of urinary collagen IV and fibronectin in the diabetic patients after Losartan treatment were not significantly different from the basal level, although there was a trend of decrease in urinary collagen IV levels after more than 4 months of the Losartan treatment (Table 3).

Correlations between clinical and biochemical variables

The levels of HbA1c positively correlated with plasma and urinary TGF-β and PAI-1 (p<0.05 or 0.01), but did not correlate with the levels of tPA, uPA, fibronectin or collagen IV in the diabetic patients during or after the Losartan treatment (Table 4). In addition, urinary TGF-β positively correlated with plasma PAI-1, and negatively correlated with urinary uPA and plasma tPA in the patients (P<0.05). The levels of collagen IV positively correlated with the levels of urinary and plasma uPA (P<0.01). The levels of urinary fibronectin negatively correlated with urinary PAI-1 and tPA, and positively correlated with plasma PAI-1 and urinary collagen IV (P<0.05 or 0.01). Urinary uPA positively correlated with plasma uPA and urinary tPA (P<0.01). Positively correlation was detected between urinary and plasma tPA (P<0.01). Plasma TGF-β and urinary PAI-1 positively correlated with DBP (P<0.05). No correlation was detected between urinary albumin and the tested ECM or their regulators in the present study (Table 4).

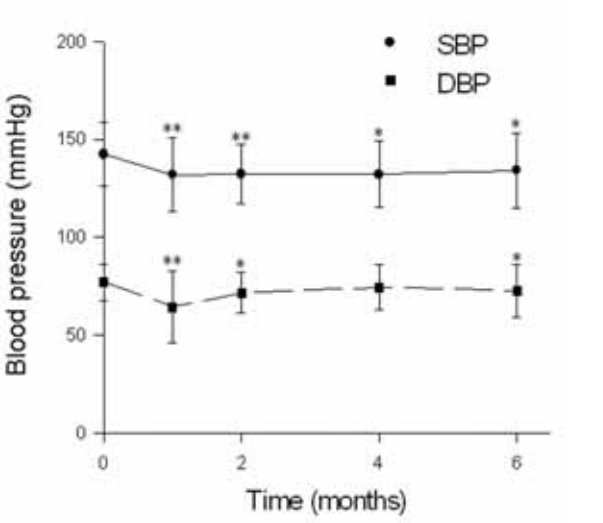


FIGURE Changes of arterial blood pressure in patients with type 2 diabetes mellitus with microalbuminuria receiving Losartan. Twenty-two diabetic patients received 50-100 mg/day of Losartan for 6 months. Blood pressure was measured at sitting position between 8-10 am before and after 1, 2, 4 and 6 months of the Losartan treatment. Values were expressed in mean ± SD (n = 22). *, **: P<0.05 or 0.01 versus baseline measurements.

Discussion

Microalbuminuria is an early biochemical marker of renal functional impairment in incipient diabetic renal disease. Early management of diabetic patients at the stage of microalbuminuria may improve the prognosis of the patients. ACE inhibitors postponed the progression of nephropathy in patients with type 1 DM or non-diabetic renal diseases in comparison to calcium channel blockers, β-blockers or diuretics.[18] Some studies did not show better efficacy of ACE inhibitors compared to other antihypertensives in terms of delaying the onset of ESRD in type 2 DM patients.[19-21] Several large clinical trials demonstrated that treatment of AT1 antagonists provides renal protection for type 2 DM patients.[4-6] The results of the present study on the effect of Losartan on blood pressure and urinary albumin in type 2 DM patients were consistent to previous reports [4-6] which helps to validate the treatment of Losartan in the present study.

Previous studies suggest that a blood pressure-independent mechanism possibly contributes to the renal protective effect of renin-angiotensin antagonists in type 2 DM patients.[6,22,23] Multiple lines of evidence suggest that TGF-β is implicated in the pathogenesis of tubulointerstitial fibrosis of diabetic nephropathy.[24] Angiotensin II stimulates the expression of TGF-β in the kidney via AT1 receptor.[25] Losartan inhibits the expression of TGF-β in mesangial cells through down-regulating phosphatidylinositol-3 kinase and mitogen activating protein kinase activity.[26] Results from limited available studies regarding to the effects of Losartan on the levels of TGF-β in type 2 DM patients are incoherent. Esmatjes et al. [9] reported that Losartan treatment reduced plasma levels of TGF-β1 and urinary albumin excretion in 14 type 2 DM patients with hypertension and microalbuminuria. Houlihan et al.[10] found that Losartan decreased urinary TGF-βexcretion in 11 type 2 DM patients compared with placebo group. The results of the present study demonstrated that Losartan administration reduced urinary, but not plasma, levels of TGF-β in diabetic patients with microalbuminuria, which supports the findings of Houlihan et al.[10] The results imply that

Losartan may have selective effect on the metabolism of TGF-β in kidneys.

TGF-β regulates the generation of multiple ECM presenting in the glomerular mesangial complex. High levels of tubulointerstitial TGF-β and glomerular fibronectin mRNA was associated with deterioration of renal function.[27] Murayama et al.[12] found that candesartan did not reduce urinary secretion of collagen IV in diabetic patients with microalbuminuria. The results of the present study suggest that Losartan does not significantly alter urine secretion of collagen IV or fibronectin in diabetic patients, which is consistent to the findings by Murayama et al.[12] In addition, the results of the present study do not exclude the possibility that Losartan may affect other ECM proteins in kidney, including TGF-β-inducible gene h3 (βig-h3).[28,29] in diabetic patients. Analysis assay kit for βig-h3 is not commercially available at the time of the study.

Previous studies in hypertensive patients, AT1 antagonist administration reduced the levels of PAI-1 in plasma.[30,31] Fogari et al.[32] reported that ACE inhibitor, but not Losartan, decreased plasma levels of PAI-1 in type 2 DM patients. In vitro study found that ACE inhibitor, but not Losartan, inhibited the expression of PAI-1 in proximal tubular epithelial cells.[33] These findings suggest that PAI-1 expression in peripheral vascular or renal cells may be regulated through an AT1-independent mechanism in kidney. The effects of Losartan on urinary secretion of fibrinolytic regulators in diabetic patients have not been documented to our knowledge. The present study demonstrated that the levels of PAI-1, tPA or uPA were readily detectable in urine of type 2 DM patients with microalbuminuria. Losartan did not significantly alter the levels of the fibrinolytic regulators in peripheral circulation or urine in type 2 DM patients with microalbuminuria. One of potential explanation is that the secretion of uPA or tPA may not be regulated by AT1-dependent pathway as that of PAI-1 in EC.[34] Further studies to observe the changes in urinary section of uPA and tPA in diabetic patients using ACE inhibitors will help to examine this hypothesis.

Previous studies demonstrated that the expression of PAI-1 was increased in kidneys of humans and animals with diabetic nephropathy and was associated with ECM accumulation in mesangial complex.[35] Genetic PAI-1-deficiency retarded the development of diabetic nephropathy in mice through a TGF-β-dependent mechanism. The effect of PAI-1-deficiency was neutralized by uPA in the animal model.[36] The results of the present study demonstrated that the levels of uPA positively correlated with the levels of collagen IV in urine. Urinary PAI-1 levels negatively correlated with the levels of fibronectin in urine. Urinary secretion of TGF-β negatively correlated with the levels of uPA in urine, which probably acts as a negative feedback for ECM turnover. These findings suggest that both fibrinolytic system and TGF-β are implicated in ECM turnover in kidneys of diabetic patients with microalbuminuria. TGF-β may modulate the metabolism or the effects of fibrinolytic regulators on ECM turnover in kidneys. Experimental studies using animals with genetic deficiency of fibrinolytic regulators or TGF-β may provide additional insight to the role of the ECM modulators in the development of diabetic nephropathy.

In conclusion, Losartan administration effectively reduced urinary albumin excretion and arterial blood pressure in type 2 DM patients. A reduction in urinary TGF-β was detected in the diabetic patients receiving Losartan treatment. Urinary levels of TGF-β correlate with fibrinolytic regulators in urine or plasma of type 2 DM patients. Crosstalk between TGF-β and fibrinolytic regulators may modulate ECM turnover in kidneys of type 2 DM patients with latent renal dysfunction.

Acknowledgments

The authors thank for the assistance from the staff in Diabetes Research Group, and Dr. Harvey Lee (Rossmere Medical Clinic) on the recruitment or sample handling, the helps from Ms. Lori-Jean Manness, Ms. Jill Spiegel and Mr. Tom McDougall for the start of the project, research laboratory space provided by the Health Sciences Centre Foundation, and the grant supports from Merck Medical School Grant, Canadian Institute of Health Research Industrial Partnership Program (G.X.S., W.V., and B.P.) and Canadian Diabetes Association (G.X.S). Dr. Lian-Song Ni was a Visiting Scientist supported by the First Affiliated Hospital in the Wenzhou Medical College, Wenzhou, China.

References

1. Amos AF, McCarty DJ, Zimmet P. The rising global burden of diabetes and its complications: estimates and projections to the year 2010. Diabet Med. 1997;14 Suppl 5:S1-85
2. US Renal Data System: USRDS 2002 Annual Data Report. Atlas of end-stage renal disease in the United States. Besthesda MD, National Institutes of Health,

National Institute of Diabetes and Digestive and Kidney Disease. www.usrds.org/adr_2002.htm
3. Ismail N, Becker B, Strzelczyk P, Ritz E. Renal disease and hypertension in non-insulin-dependent diabetes mellitus. Kidney Int 1999;55:1-28
4. Brenner BM, Cooper ME, de Zeeuw D, et al. Effect of losartan on renal and cardiovascular outcomes in patients with type 2 diabetes and nephropathy. N Engl J Med 2001;345:861-9
5. Parving HH, Lehnert H, Brochner-Mortensen J, Gomis R, Andersen S, Arner P. Irbesartan in Patients with Type 2 Diabetes and Microalbuminuria Study Group. The effect of irbesartan on the development of diabetic nephropathy in patients with type 2 diabetes. N Engl J Med 2001;345:870-8
6. Lewis EJ, Hunsicker LG, Clarke WR, et al. Renoprotective effect of the angiotensin-receptor antagonist irbesartan in patients with nephropathy due to type 2 diabetes. N Engl J Med 2001;345:851-60
7. Wogensen L, Krag S, Chai Q, Ledet T. The use of transgenic animals in the study of diabetic kidney disease. Horm Metab Res 2005;37 Suppl 1:17-25
8. Mauviel A. Transforming growth factor-beta: a key mediator of fibrosis. Methods Mol Med 2005;117:69-80
9. Esmatjes E, Flores L, Inigo P, Lario S, Ruilope LM, Campistol JM. Effect of losartan on TGF-beta1 and urinary albumin excretion in patients with type 2 diabetes mellitus and microalbuminuria. Nephrol Dial Transplant 2001;16 Suppl 1:90-3
10. Houlihan CA, Akdeniz A, Tsalamandris C, Cooper ME, Jerums G, Gilbert RE. Urinary transforming growth factor-beta excretion in patients with hypertension, type 2 diabetes, and elevated albumin excretion rate: effects of angiotensin receptor blockade and sodium restriction. Diabetes Care 2002;25:1072-7
11. Nishimura M, Sasaki T, Ohishi A, et al. Angiotensin-converting enzyme inhibitors and probucol suppress the time-dependent increase in urinary Type IV collagen excretion of Type II diabetes mellitus patients with early diabetic nephropathy. Clin Nephrol 2001;56:96-103
12. Murayama S, Hirano T, Sakaue T, Okada K, Ikejiri R, Adachi M. Low-dose candesartan cilexetil prevents early kidney damage in type 2 diabetic patients with mildly elevated blood pressure. Hypertens Res 2003;26:453-8
13. Tada H, Tsukamoto M, Ishii H, Isogai S. A high concentration of glucose alters the production of tPA, uPA and PAI-1 antigens from human mesangial cells. Diabetes Res Clin Pract 1994;24:33-9
14. Nakamura S, Nakamura I, Ma L, Vaughan DE, Fogo AB. Plasminogen activator inhibitor-1 expression is regulated by the angiotensin type 1 receptor in vivo. Kidney Int 2000;58:251-9
15. WHO Expert Committee on Diabetes Mellitus. Second report. World Health Organ Tech Rep Ser 1980;646:1-80
16. Zhang J, Ren S, Sun D, Shen GX. Influence of glycation on LDL-induced generation of fibrinolytic regulators in vascular endothelial cells. Arterioscler Thromb Vasc Biol 1998;18:1140-8
17. Jones CL, Buch S, Post M, McCulloch L, Liu E, Eddy AA. Renal extracellular matrix accumulation in acute puromycin aminonucleoside nephrosis in rats. Am J Pathol 1192;141:1381-96
18. Kshirsagar AV, Joy MS, Hogan SL, Falk RJ, Colindres RE. Effect of ACE inhibitors in diabetic and nondiabetic chronic renal disease: a systematic overview of randomized placebo-controlled trials. Am J Kidney Dis 2000;35:695-707
19. Efficacy of atenolol and captopril in reducing risk of macrovascular and microvascular complications in type 2 diabetes. UKPDS 39. UK Prospective Diabetes Study Group. BMJ 1998;317(7160):713-20
20. Fogari R, Zoppi A, Corradi L, et al. Long-term effects of ramipril and nitrendipine on albuminuria in hypertensive patients with type II diabetes and impaired renal function. J Hum Hypertens 1999;13:47-53
21. Nielsen FS, Rossing P, Gall MA, Skott P, Smidt UM, Parving HH. Long-term effect of lisinopril and atenolol on kidney function in hypertensive NIDDM subjects with diabetic nephropathy. Diabetes 1997;46:1182-8
22. Viberti G, Wheeldon NM. MicroAlbuminuria Reduction With VALsartan (MARVAL) Study Investigators. Microalbuminuria reduction with valsartan in patients with type 2 diabetes mellitus: a blood pressure-independent effect. Circulation 2002;106:672-8
23. Davis BJ, Cao Z, de Gasparo M, Kawachi H, Cooper ME, Allen TJ. Disparate effects of angiotensin II antagonists and calcium channel blockers on albuminuria in experimental diabetes and hypertension: potential role of nephrin. J Hypertens 2003;21:209-16
24. Wang SN, LaPage J, Hirschberg R. Role of glomerular ultrafiltration of growth factors in progressive interstitial fibrosis in diabetic nephropathy. Kidney Int 2000;57:1002-14
25. Wolf G. Link between angiotensin II and TGF-beta in

the kidney. Miner Electrolyte Metab 1998;24:174-80
26. Perlman A, Lawsin LM, Kolachana P, Saji M, Moore J Jr, Ringel MD. Angiotensin II regulation of TGF-beta in murine mesangial cells involves both PI3 kinase and MAP kinase. Ann Clin Lab Sci 2004;34:277-86
27. Eikmans M, Baelde HJ, Hagen EC, et al. Renal mRNA levels as prognostic tools in kidney diseases. J Am Soc Nephrol 2003;14:899-907
28. Cha DR, Kim IS, Kang YS, et al. Urinary concentration of transforming growth factor-beta-inducible gene-h3(beta ig-h3) in patients with Type 2 diabetes mellitus. Diabet Med 2005;22:14-20
29. Ha SW, Kim HJ, Bae JS,et al. Concentration of urinary betaig-h3, transforming growth factor-beta-induced protein in patients with type 2 diabetes and nephropathy. Diabetes Res Clin Pract 2004;65:167-73
30. Erdem Y, Usalan C, Haznedaroglu IC,et al. Effects of angiotensin converting enzyme and angiotensin II receptor inhibition on impaired fibrinolysis in systemic hypertension. Am J Hypertens 1999;12:1071-6
31. Brown NJ, Agirbasli M, Vaughan DE. Comparative effect of angiotensin-converting enzyme inhibition and angiotensin II type 1 receptor antagonism on plasma fibrinolytic balance in humans. Hypertension 1999;34:285-90
32. Fogari R, Mugellini A, Zoppi A, et al. Losartan and perindopril effects on plasma plasminogen activator inhibitor-1 and fibrinogen in hypertensive type 2 diabetic patients. Am J Hypertens 2002;15:316-20
33. Gesualdo L, Ranieri E, Monno R, et al. Angiotensin IV stimulates plasminogen activator inhibitor-1 expression in proximal tubular epithelial cells. Kidney Int 1999;56:461-70
34. Kerins DM, Hao Q, Vaughan DE. Comparative effect of angiotensin-converting enzyme inhibition and angiotensin II type 1 receptor antagonism on plasma fibrinolytic balance in humans. Hypertension 1999;34:285-90
35. Rerolle JP, Hertig A, Nguyen G, Sraer JD, Rondeau EP. Plasminogen activator inhibitor type 1 is a potential target in renal fibrogenesis. Kidney Int 2000;58:1841-50
36. Nicholas SB, Aguiniga E, Ren Y, et al. Plasminogen activator inhibitor-1 deficiency retards diabetic nephropathy. Kidney Int 2005;67:1297-307

Correspondience

Garry X. Shen MD PhD
Diabetes Research Group
University of Manitoba
835-715 McDermot Ave
Winnipeg, Manitoba R3E 3P4
Canada
e-mail: gshen@ms.umanitoba.ca

HISTORY OF MEDICINE

Control of Coagulation: a Gift of Canadian Agriculture

Vivian McAlister

Department of Surgery, University of Western Ontario, Canada

Manuscript Submitted: 28th June 2006
Manuscript Accepted: 7th August 2006

Clin Invest Med 2006; 29 (6): 373–377.

Summary

Vitamin K, heparin and their antagonists remain the basis of coagulation therapies today, more than half a century after their discovery. Failure of blood clotting in chicks that were fed a fat-depleted diet was observed by William McFarlane, William Graham Jr. and Frederick Richardson of the Ontario Agricultural College; it led to the search that yielded vitamin K. Investigation of hemorrhagic disease in cattle by Francis Schofield of the Ontario Veterinary College found an anti-thrombin substance in spoiled clover which was later characterized as dicoumarol, a vitamin K antagonist, and led to the development of warfarin. In Toronto, a systematic approach lead by Charles Best resulted in the world's first plentiful supply of purified heparin. Clinical usefulness of heparin in thrombosis, embolism, cardiovascular surgery, dialysis and transplantation was demonstrated first by Gordon Murray and Louis Jaques. The roles and the careers of Canadian coagulation research pioneers are briefly presented in this review, which shows how clinical medicine benefited by the systematic development of agricultural science in Guelph, Ontario.

Introduction

The lives of Toronto scientists, Charles Best (1899 – 1973) and Gordon Murray (1894 – 1976) including their crucial roles in making heparin available for clinical use are well documented if not generally known.[1,2] The role of the scientists from Guelph, Ontario in allowing control of vitamin K-dependant coagulation is even less well known. In his 1943 Nobel Prize lecture, Henrik Dam (Copenhagen, 1895 – 1976) credited 'McFarlane and co-workers of the Ontario Agricultural College' (OAC) with the initial observation that led to the discovery of vitamin K.[3] Contemporary newspapers failed to mention this honour and it is not clear if the scientists themselves were aware of it. The discovery of vitamin K was actually preceded by the description of a factor which antagonized it, causing haemorrhage in cattle. This observation, which was made in 1922 by Francis William Schofield (1889 – 1970), a pathologist from the Ontario Veterinary College (OVC), led to the discovery of the coumarin-derived anti-caogulants.[4] The Ontario agricultural scientists' contribution, if considered at all in history, is portrayed as serendipitous. More careful examination reveals an elaborate development in Guelph of agricultural and related sciences, from which human medicine benefited on more than one occasion. The purpose of this review is to outline the roles of the participants in this story and to briefly describe their careers.

Background: a plentiful supply of heparin

Toronto's role in bringing heparin into clinical use is as grand and as important as its contribution to the development of insulin.[5] Although Charles Best was disappointed not to be included in the Nobel Prize of 1923 for his role in the discovery of insulin, it did not go unrewarded. In 1928 at the age of 33 years, he became the youngest head of department in the University of Toronto. He succeeded Banting's

Nobel co-laureate, John MacLeod, as Professor of Physiology, a job he added to his role as Associate Director of Connaught Laboratories. He immediately assembled a team dedicated to the inexpensive and plentiful production of purified heparin and to its introduction into clinical practice.

Twelve years earlier Jay McLean (1851 – 1957), a Johns Hopkins medical student, had discovered an anti-thrombin factor in liver. His supervisor, William Howell was able by 1918 to extract the substance which he called heparin but only in small amounts. Best recruited Arthur F. Charles (1905-1972) to join David A. Scott (1892-1971), a veteran of insulin production, at Connaught; their job was to find a cheap source of heparin. They switched from McLean's dog liver to beef liver and when that became too expensive they found a plentiful source in beef intestine. As soon as reliable production of heparin seemed possible, Best approached Gordon Murray to join the team as an experimental surgeon.[2] Louis Jaques, a Toronto chemistry graduate who had registered with Best for post-graduate studies, was assigned to Murray as an assistant.[6] Murray and Jaques demonstrated the usefulness of heparin in the treatment of thrombosis and embolism. Murray realized that safe local circulatory arrest might permit vascular surgery. He pioneered the fields of peripheral vascular and complex cardiac surgery.[2] Murray's discovery that heparin prevented blood clotting in extra-corporeal circuits allowed him to pioneer the development of renal dialysis. Combining both vascular surgery and renal failure therapy, Murray used heparin to introduce the modern era of renal transplantation.[7] Louis Jaques continued heparin research as Head of Physiology at the University of Saskatchewan becoming the world's leading expert on heparin for the next 60 years.

The Ontario Agricultural College and the discovery of vitamin K.

In 1930, William McFarlane, William Graham Jr. and Frederick Richardson from OAC submitted a report to the Journal of Biochemistry regarding a series of experiments on the fat soluble components of chick feed.[8] Vitamins A and D were known to be essential for growth in the chick. The authors noted that previous researchers had failed to account for small amounts of fat and fat soluble products in their specialized diets. They decided to use ether to remove all the fat from the basic meal and to restore various elements in different comparative groups. In the control group of chicks on the fat-free diet they noted persistent bleeding from the small wounds where identification tags were applied and they found that the chick blood would not clot. After determining that the phenomenon was not due to the lack of known vitamins or due to the disturbance of calcium - inorganic phosphorous homeostasis, they returned to the principal focus of their experiment, which was to compare the effect of fish meal and meat meal dietary supplements on chick growth.

Holst and Halbrook discovered that cabbage could restore coagulation in fat-depleted-chow fed chicks but mistakenly identified vitamin C as the missing ingredient.[9] Dam found anti-haemorrhagic factor in a variety of other foods and named the vitamin 'K' for 'koagulation' although it is also said that vitamin discovery was so rapid in those years that he was uncertain whether the lower alphabetical titles were already 'taken'.[3] In 1939, Edward Doisey (Illinois, 1893 – 1986) extracted, characterized and synthesized vitamin K, using alfalfa as the source.[10] Vitamin K was soon available to treat cholestatic patients who were bleeding or who needed surgery. Dam and Doisey shared the 1943 Nobel Prize in Physiology or Medicine.

McFarlane, Graham Jr. and Richardson

The OAC researchers who started it all have faded from view before we got to know them. William Douglas McFarlane was born in Glasgow on June 26th, 1900. He studied dairy science, receiving the National Diploma in Agriculture of Great Britain. He came to Canada in 1923 to work as a butter-maker for the Walkerton Egg and Dairy Company and to study at the OAC.[11] He transferred from Chemistry to the Poultry Department under Professor Dick Graham, where he stayed until he completed his Ph.D. in 1932. Also in the department was William R Graham Jr. (born 1906, Guelph), the professor's son.[11] Graham Jr. collaborated with McFarlane on the chick feed study. The third member of the team was Frederick Richardson (born 1900, Minneapolis) who was an associate student.[11] It would seem that McFarlane was the principal investigator as his M.A. thesis dealt with a similar topic[12] whereas Graham Jr. studied the nutritional role of the thyroid in cattle for his 1933 Ph.D.[13] McFarlane completed his Ph.D. in 1932 studying nutritional anaemia.[14] He was recruited by the University of Edmonton as an Assistant Professor of Chemistry. In 1941, McFarlane published a survey of farm product research in Canada.[15] He returned to Guelph as Professor of Chemistry at MacDonald

College and in 1947 he became Director of Research for Canadian Breweries in Toronto.

Graham Jr. went to Kansas where he worked in the research department of Cerophyl Laboratories. There he studied the nutritional value of grasses such as alfalfa, ironically Dam's source of vitamin K.[16] In 1949 he became Director of Research for Quaker Oats in Chicago. He was an early enthusiast for the use of soybeans. In 1952, he published in *Science* the mechanism of absorption of nitrogen in rats fed soybeans.[17] He continued this research by looking at the effect of adding methionine and tetracycline to his model.[18,19]

Richardson remained in Canada, working at the Dominion Experimental Station in Nappan, Nova Scotia before he too joined the brewing industry in Toronto at the Canada Malting Company.[11] These incomplete biographies were gleaned from the records of the OAC that are now kept by the University of Guelph, from records of post-graduate theses at the University of Toronto and from a scan of medical literature for their names as authors. Unfortunately no photographs were located. Should this article stimulate the memories of some of its readers, the author would appreciate more information.

Schofield and vitamin K inhibition.

What was not known to Dam or other vitamin researchers was that vitamin K antagonists had been discovered almost a decade earlier. Again the discovery derived from an observation in Agriculture and this time the scientist who reported it was from OVC. In the early part of the 20th century it was known that cattle could sporadically die of spontaneous haemorrhage. In 1922, Francis W Schofield (1889-1970) was able to track the cause down to sweet clover that had been spoiled by mould.[4] He had been sent animals affected by the problem from two herds, in Markham and in St. Mary's Ontario, both of which had been fed clover silage 'of poor quality'. To prove his hypothesis he conducted trials at OVC comparing diets of mouldy sweet clover to both sweet clover and to grass. The syndrome appeared similar to septicemia but no infectious agent was found. He eliminated low calcium as a cause and suggested it was due to low prothrombin. Schofield then focused on advice for the treatment of affected cattle.[4] Karl Paul Link and Harold Campbell of the University of Wisconsin found that mould oxidized coumarin in clover and coupled it to formaldehyde, forming a potent anti-coagulant, dicoumarol.[20] Subsequent derivatives included warfarin, named after Link's funding agency, Wisconsin Alumni Research Fund. Warfarin was originally used solely as a rodent pesticide because it was considered too powerful for human use. Recovery of attempted suicide victims suggested that this was not true. The use of dicoumarol and subsequently of warfarin was popularized when President Dwight Eisenhower was treated with it after a heart attack in 1955.

Frank Schofield was well known in his day not only for his prolific publication in veterinary medicine but also for his unusual career. Schofield emigrated from England to Canada at the age of 16, one year before he entered OVC in 1907.[21] Five years after graduation he was invited by Dr. Oliver R. Avison (1860 – 1956), a former Professor of Medicine in Toronto, to teach Bacteriology and Sanitation at the Severance Medical School in Seoul. He became active in the defense of the Korean people against Japanese occupation. He documented and photographed several atrocities. He hid student leaders while he negotiated with Japanese officials on their behalf. Eventually it was considered too dangerous for him to remain in Korea. He returned to OVM in 1921 where he worked for 33 years. He published 148 papers despite limited funding.[22] On retirement he returned to Korea to work for the College of Veterinary Medicine in Seoul. The status he achieved in Korea, which has been compared to that of Norman Bethune in China, is evident by his inclusion as the only foreigner in the Patriots' Corner of Korea's National Cemetery.[23] It contrasts with his status in Canada where he, like the other pioneers of coagulation research, has been allowed to fade from memory.

Agricultural science research at Guelph.

The systematic research which led to the discovery of vitamin K can be easily traced to William Richard (Dick) Graham (1875 - 1958), Professor of Poultry Science at the OAC (Figure). Dick Graham was born in Hastings, Ontario in 1875.[24] He graduated from OAC in 1894 and returned to run the family farm. In 1899 he was recruited back to OAC to head its poultry department. He introduced scientific methods to study the breeding, growth and egg production of poultry. Professor Graham arranged with the University of Toronto for OAC students to continue postgraduate studies within his department. In the process he seeded the food and agriculture industry of North America with scientist-leaders. He collaborated with the Nutritional Research Laboratory at the Hospital for Sick Children on the development of the

FIGURE: Controlled trials by Professor Dick Graham (shown here) were used in best practice guidelines for poultry management, published by the Ontario Department of Agriculture in 1906.
Photograph courtesy of University of Guelph

infant formula, *Pablum,* which was derived from his chick starter-feed. The experimental techniques used by Fred Tisdall, Theo Drake and Alan Brown in their animal studies of nutritional supplements were similar to those used by McFarlane and co-workers.[25,26] Professor Graham was recognized by the University of Toronto with a Doctor of Science (honoris causa) in 1938. At his retirement his close friend Frederick Banting gave the eulogy. [24]

At the turn of the last century, the Ontario Department of Agriculture published a series of evidenced-based reports written by Dick Graham regarding the breeding and rearing of poultry.[27,28,29] The evidence was based on controlled experiments performed by Graham. Graham's technique was passed on to his students who observed coagulopathy in chicks on fat-free diets. The antecedent role of his interests and methods in the work of Tisdall, Drake and Brown is clear, if not well recognized. In a similar fashion Schofield back up his hunch regarding spoiled clover with a controlled trial. Although Graham's reports for the Ontario Department of Agriculture are now rare documents (microfiche copies are available from the University of Toronto Library), they were originally published and circulated for general use in a concerted effort to improve outcomes. This evidence-based campaign to promote best practice which started one hundred years ago preceded its equivalent in human medicine by most of the intervening century. While the systematic approach to agricultural science promoted by Dick Graham, Frank Schofield and their colleagues in Guelph permitted the observations that unlocked the mysteries of coagulation, perhaps it was the approach itself that was Canadian agriculture's gift to human medicine.

Acknowledgement
I am particularly grateful to Ms. Darlene Wiltsie, Librarian, Archival & Special Collections, McLaughlin Library, University of Guelph, for access to files and for assistance searching.

References.

1. Best HBM. Margaret and Charley: The Personal Story of Dr. Charles Best, the Co-Discoverer of Insulin. Dundurn Press. Toronto 2003
2. McKellar S. Surgical Limits: The Life of Gordon Murray University of Toronto Press. Toronto 2003
3. Dam H. The discovery of vitamin K, its biological functions and therpeutical application. Nobel Lecture, December 12, 1946. http://nobelprize.org/medicine/laureates/1943/dam-lecture.pdf (accessed October 14, 2006).
4. Schofield FW. A brief account of a disease in cattle simulating haemorrhagic septicaemia due to feeding sweet clover. Can Vet Record 1922; 3: 74-8
5. Baird RJ. "Give us the tools...". The story of heparin--as told by sketches from the lives of William Howell, Jay McLean, Charles Best, and Gordon Murray. J Vasc Surg. 1990 Jan;11(1):4-18.
6. Dr. Louis Jaques. The Banting Research Institute Website http://www.utoronto.ca/bantresf/HallofFame/Jaques.html (accessed October 14, 2006).
7. McAlister VC. A 50th anniversary review of the first series of clinical kidney transplantation. Am J Surg 2005; 190: 485-8
8. McFarlane WD, Graham Jr. WR, Richardson F. The fat-soluble vitamin requirements of the chick. 1. The

vitamin A and vitamin D content of fish meal and meat meal. Biochem J 1931; 25: 358-66
9. Holst WF. Halbrook ER. A 'scurvy-like' disease in chicks. Science 1933; 77: 354
10. Edward A. Doisey – Biography. *Nobel Lectures, Physiology or Medicine 1942-1962*, Elsevier Publishing Company, Amsterdam, 1964 http://nobelprize.org/medicine/laureates/1943/doisy-bio.html (accessed October 14, 2006).
11. Bureau of Records of the Ontario Agricultural College. Archival and Special Collections, University of Guelph Library.
12. McFarlane WD. *Study of the comparative nutritional value of certain commercial protein supplements in poultry nutrition I by feeding trial; II analytically. 1929* Thesis (M.A.) University of Toronto.
13. Graham WR. *The effect of thyroidectomy and thyroid feeding on the mild fat formation and milk secretion of cows. 1933* Thesis (Ph.D.) University of Toronto.
14. McFarlane WD. *Studies in nutritional anaemia. 1932* Thesis (Ph.D.) University of Toronto
15. McFarlane WD. **A survey of Canadian research on the utilization of farm products** 1941 Canadian Chamber of Commerce. Montreal
16. Kohler GO, Graham Jr. WR. A chick growth factor found in leafy green vegetation. Poultry Sci 1951; 30: 484
17. Carroll RW, Hensley GW, Graham WR. The site of nitrogen absorption in rats fed raw and heat-treated soybean meals. Science. 1952; 115: 36-9.
18. Carroll RW, Hensley GW, Sittler CL, Wilcox EL, Graham WR. Absorption of nitrogen and amino acids from soybean meal as affected by heat treatment or supplementation with aureomycin and methionine. Arch Biochem Biophys. 1953; 45: 260-9.
19. Carroll RW, Hensley GW, Wilcox EL, Graham WR. The effects of aureomycin and methionine supplements fed to rats receiving soybean meals. Arch Biochem Biophys. 1953; 45: 270-4.
20. Link PK. The discovery of dicumarol and its sequels. Circulation.1959; 19: 97-90.
21. Citation for Honorary degree awarded to Francis W Schofield (Archival and Special Collections, University of Guelph Library)
22. Murphy DA, Hulland TJ. Frank W. Schofield – Bibliography (notes held by Archival and Special Collections, University of Guelph Library)
23. Smith DL. Francis William Schofield 1889-1970. Vet Pathol 1971; 8: 282-8.
24. Citation for Dr. William Richard Graham. Canadian Agricultural Hall of Fame 1975. http://www.cahfa.com/ag/Graham.html (accessed October 14, 2006).
25. Tisdall FF, Drake TG, Summerfeldt P, Brown A. A new whole wheat irradiated biscuit, containing vitamins and mineral elements. CMAJ 1930; 22: 166-70.
26. Tisdall FF, Drake TG, Brown A. The incorporation of vitamins in bread. CMAJ 1931; 24: 210-3.
27. Graham WR **Farm poultry: with the results of some experiments in poultry houses and fattening chickens. Ontario Department of Agriculture. Toronto, Canada. 1906**
28. Graham WR **Experiments in hatching and rearing chickens. Ontario Department of Agriculture. Toronto, Canada. 1911**
29. Graham WR **Egg production in cities, towns and villages. Ontario Department of Agriculture. Toronto, Canada. 1917**

Correspondence:

Dr. Vivian McAlister
University Hospital,
University of Western Ontario
London, Ontario
Canada, N6A 5A5
vmcalist@uwo.ca

CSCI AWARDS 2006

CSCI/RCPSC Henry Friesen Lecture: Cell therapy for Duchenne Muscular Dystrophy.

Jacques P. Tremblay, Ph.D.

Human Genetic Unit, CRCHUL and Laval University
Quebec, P.Q. Canada G0A 4P0

Presented: 28th September 2006

Clin Invest Med 2006; 29 (6): 378–382.

Description of Duchenne Muscular Dystrophy (DMD):

DMD is a fatal hereditary disease for which there is currently no treatment. Progressive muscle weakness starts at age 5, leads to wheelchair confinement by age 10-12, and death from respiratory complications or secondary heart disease between 17 and 30 years. DMD is caused by an X-linked genetic defect leading to the absence of dystrophin.[1] Dystrophin is normally located under the muscle membrane, and it links the cytoskeleton via a group of proteins called (the dystrophin complex) located in the membrane with the extracellular matrix.[2] The absence increases the vulnerability of muscle fibers to damage during normal activity.

Muscle fibers are cellular syncitia. Fibers damaged during normal activity are repaired by proliferation and fusion of stem cells, called satellite cells, located close to the muscle fibers under the basal lamina. When proliferating, these satellite cells are called myoblasts. Unfortunately, in the case of DMD, the repaired muscle fibers still lack dystrophin and are thus still vulnerable to subsequent activity. This leads to frequent repair of the muscle fibers, progressive myoblast senescence and loss of regenerative potential by 4 to 5 years.[3] Damaged muscle fibers are no longer adequately repaired and are gradually replaced by fat and connective tissue leading to progressive muscle weakness.

Dystrophin is a very large gene (2.6 million base pairs) containing 79 exons. This produces a final messager RNA containing 14,000 bases coding for a large protein (427 kDa). Mutations in this gene can result in Duchenne or in Becker Muscular Dystrophy (BMD), a less severe disease, the patient being confined to a wheel chair later or sometimes not at all.[1] The mutations leading to these dystrophies can be either deletions, duplications or in about 30% of the cases point mutations. In the case of DMD the deletion of one or several exons causes a shift in the reading frame of the messager RNA. The end of the dystrophin protein is thus missing and the protein can not form a complex with the glycoproteins forming the dystrophin complex. This complex is thus totally absent from the membrane. In BMD, a multiple of 3 nucleotides is deleted so that there is no change of the reading frame of the messager RNA and thus the beginning and the end of the dystrophin protein are present and the protein can incorporate in the dystrophin complex. There is thus in BMD the presence of a dystrophin complex in the muscle fiber membrane only a part of dystrophin being absent. Depending on the portion of dystrophin which is absent, the protein is more or less functional and thus the symptoms of the BMD patients are more or less severe.

Possible therapies of DMD

Several approaches for treating DMD are currently being pursued. *Gene therapy* aims to introduce the dystrophin gene directly in the patient muscle fibers. The main problem facing gene therapy for DMD is that because of the large size of the dystrophin cDNA there is no adequate vector to integrate this gene in the muscle fiber genome. However, widespread expression of a truncated version (missing the central part) of the dystrophin gene have been recently obtained in dystrophic mice with an Adeno Associated Virus (AAV).[4] However, as

in other forms of gene therapy, prolonged gene expression, avoiding a host immune response to vector proteins and producing large amount of the vector in GMP (Good Manufacturing Practice) conditions remain problematic for human application. A new approach to gene therapy based on exon skipping permit to restore a correct reading frame for the dystrophin mRNA or to bypass a nonsense mutation (stop codon) and allows expression of truncated Dys which is likely to result in a milder disease phenotype (such as that seen in Becker forms of dystrophy).[5] Such exon skipping may be obtained with small systemic oligonucleotides which have been chemically modified to increase their stability for systemic delivery or with AAV vectors coding for such an antisense.[6] Moreover, since the Dys expressed is identical to that endogenously expressed by rare revertant fibers, immunogenicity may be eliminated. Nonetheless, an immune response remains conceivable, since in lupus models, evidence suggests that higher antigen dose can augment the immune response and precipitate autoimmunity when counter-regulatory mechanisms are overwhelmed.

The principle of cell therapy

The damaged muscle fibers are repaired by the proliferation of mononucleated cells located on the muscle fibers under the basal lamina. Following muscle fiber damage, these satellite cells divide and form myoblasts which further proliferate and fuse with the damaged muscle fiber to repair it. The myoblasts present in DMD patients contain the mutated dystrophin gene and thus the repaired muscle fibers still lack the expression of dystrophin and are thus still vulnerable. However, if myoblasts obtained from a healthy donor are transplanted in the patient muscle they contain the normal dystrophin gene and their fusion with damaged DMD muscle fibers introduces the normal dystrophin gene and protects them from further damage. Importantly, some of the transplanted cells will not fuse immediately and will remain as quiescent satellite cells, which will be able to repair subsequent damage induced by normal muscle activity in neighbouring dystrophin negative muscle fibers. Indeed, transplantation of wild-type syngeneic myoblasts can restore dystrophin expression in mdx mice, which lack this protein and thus serve as a murine model for DMD.[7] Importantly, our research group has shown that dystrophin expression following myoblast transplantation can protect muscles in mdx mice from myonecrosis normally induced by eccentric exercise.[8] Thus, restoration of Dys expression in muscle actually improves disease phenotype.

Early clinical trials

The absence of strength increase in most of the early clinical trials of myoblast transplantation in the 1990 decade.[9-13] led to the conclusion that myoblast transplantation was not effective in human patients. Controversy also surrounded the clinical trial conducted by one research group.[14] However, our research group had reported some improvements in a few patients.[15-19] Experiments conducted by our research group in animal models following these clinical trials identified some of the problems that limited the success of these early clinical trials. An important problem was the choice of an inadequate immunosuppressive drug, e.g., cyclophosphamide that either killed the myoblasts early after their transplantation[10] or cyclosporine, which induced apoptosis of the differentiating myoblasts.[20] We also demonstrated that myoblasts transplanted in a muscle did not migrate more than 100 µm away from the injection trajectory while in the clinical trial myoblast injections were done 1 cm apart.[21] Finally, 75 to 80% of the myoblasts died in the first 3 days following transplantation, a phenomenon which also contributed to reduce the number of dystrophin positive fibers formed by the transplantation.[22]

Our recent Phase 1 clinical trial

Our research group recently completed a new phase 1 clinical trial on 9 DMD patients. We transplanted 30 million myoblasts in only a cm^3 of the Tibialis anterior muscle in patients immunosuppressed for one month with Tacrolimus. A muscle biopsy of the site injected with the myoblasts and of the controlateral muscle injected with saline was made. The expression of the normal dystrophin mRNA was detected in all 9 patients using primers which did not amplify the patient mutated dystrophin mRNA. We also observed the presence of dystrophin positive muscle fibers in 8 of these patients in the biopsies of the muscle injected with myoblasts. In most of these patients, the monoclonal antibody used to detect dystrophin was specific for a dystrophin exon deleted in the patient genome. Thus the dystrophin detected was due to the fusion of the donor myoblasts with the patient existing muscle fibers. In one of the patients, 29% dystrophin positive fibers were present while only a few revertant fibers were present in the muscle injected with saline (figure 1).[23,24] Smaller dystrophin positive muscle fibers

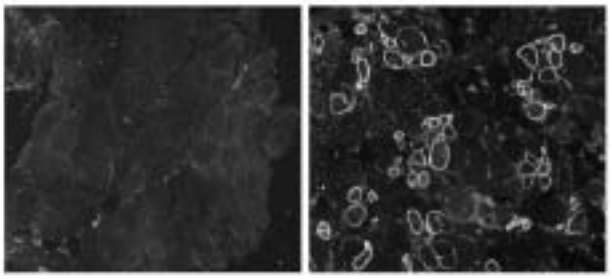


FIGURE: Cryostat section of muscle biopsies obtained one month after the transplantation of myoblasts grown from a normal donor (left side) and of the controlateral muscle injected only with saline (right side). Dystrophin was detected by immunohistochemistry using a mAb specific for dystrophin which was revealed using a rabbit anti-mouse coupled with biotin and Streptavidin coupled with Cy3. Abundant dystrophin positive muscle fibers are present only in the muscle injected with myoblasts and not in the control muscle injected with saline. These dystrophin positive muscle fibers are formed by the fusion of the donor myoblasts with the patient muscle fibers. This forms hybrid muscle fibers containing mostly the patient nuclei with the mutated dystrophin gene and a few donor nuclei containing a normal dystrophin gene. These normal nuclei express normal dystrophin mRNA which codes for a normal dystrophin protein, which incorporates over roughly 100 µm of the muscle fiber membrane. Thus a single normal nucleus can produce dystrophin for a part of the muscle fiber which contains several hundreds nuclei with a mutated gene.

were also present in biopsies of the myoblast injected muscles. These smaller fibers are probably the result of the fusion of the donor myoblasts with each others. We do not know the long term fate of these smaller fibers, but if they survive and increase in size, they may eventually contribute to increase the patient strength.

These better results obtained in our recent clinical trial were due to the following three improvements in the transplantation protocol:

1) the use of a better immunosuppressive drug (Tacrolimus),

2) more closely spaced injection trajectories to compensate for the absence of migration of the transplanted myoblasts and

3) injection of a higher amount of cells (30 million in a cm^3 of muscle. A second clinical trial is currently under way to answer the important question of whether myoblast transplantation can increase the strength of a muscle. In this clinical trial, donor myoblasts will be injected throughout one muscle (Brachio radialis) while the contralateral muscle will receive only saline injections. The patients and the physiotherapists determining the strength of the muscle will not know which side has been injected with cells. The patients will be immunosuppressed for 6 months and evaluated at 3 and 6 months.

A look at the future

Genetic modification of the patients own cells

The main problem associated with our current clinical trial is the potential adverse effects associated with a sustained immunosuppression (increase risk of cancer, nephrotoxicity, pancreatic toxicity, neurotoxicity). These adverse effects could be avoided by transplanting to the patients their own genetically modified myoblasts. Our research group has obtained recently very good results by transplanting myoblasts in mice and in monkeys genetically modified with a lentiviral vector containing a truncated dystrophin gene (unpublished results) or the full length dystrophin cDNA introduced by a combinaison of nucleofection and the PhiC31 integrase.[25] We have also been able to transplant myoblasts genetically modified with a lentiviral vector to skip a dystrophin exon to restore the normal reading frame of the dystrophin mRNA (unpublished results).

Development of specific immunological tolerance

Another strategy to avoid the need for a sustained immunosuppression is the development of immunological tolerance. Specific tolerance to donor myoblasts has recently obtained by our research group by a central mechanism involving the death of donor reactive lymphocytes in the thymus following the development of mixed chimerism by pre-transplanting donor hematopoietic stem cells.[26,27] The development of peripheral immunological tolerance by developing specific regulatory T cells (Treg) for donor myoblasts is also currently under investigation in my laboratory.

Reducing the myoblast death

As mentioned before, one of the problems reducing the success of myoblast transplantation is the rapid death of up to 80% of the transplanted cells in first few days after the transplantation.[22] Our laboratory has identified several mechanisms responsible for this problem. Part of this death is due to the inflammatory reaction which rapidly follows the intramuscular injection of cells.[28] The death is also due to an apoptotic

process called anoikis, due to the lack of contact of the transplanted cells with the extracellular matrix (unpublished results). Anoxia and pressure may also be responsible for some of the death. Several of these mechanisms may be controlled by reducing apoptosis by blocking the expression of pro-apoptotic genes with specific siRNA.

Improving the graft success with IGF-1 administration or by myostatin blockade

The success of myoblast transplantation may also be increased by interfering with growth factors involved in myogenesis. One of these factors is IGF-1, which increases the proliferation and the fusion of myoblasts and thus induces muscle hypertrophy. We have recently shown that the administration of this factor increases the percentage of dystrophin positive fibers obtained in a dystrophic mouse muscle after the transplantation of normal myoblasts (unpublished results). Myostatin is on the contrary a growth factor which reduces the proliferation and the fusion of myoblasts. Blocking the myostatin signal by various strategies not only increased the number of dystrophin positive fibers formed following the transplantation of normal myoblasts but also induced a hypertrophy of the dystrophin positive muscle fibers.[29]

Deriving myogenic cells from embryonic stem cells (ES cells)

Currently our research group is making primary myoblast culture from a specific donor for each dystrophic patient. This is a labour intensive process since every donor has to be tested for infectious diseases and every cell culture has to be done one at the time in a clean room facility. We have recently been able to derive myoblasts from mouse ES cells, a pluripotent cell capable of differentiating into every cell type of the body. In collaboration with the Stem Cell Network of Canada, we are currently trying to derive not only myoblasts from human ES cells but also hematopoietic stem cells (HSCs). This would eventually permit to develop mixed chimerism and thus central immunological tolerance towards the myoblasts derived from the same ES cells used to produce the HSCs. With this approach, the same human ES cell line could be used to treat several hundred dystrophic patients. ES cells may also be a source of other types of cells able to fuse with the existing muscle fibers, such as the mesoangioblasts a stem cells able to migrate from the blood to the muscles and thus a cell, which could be delivered systemically.

References

1. Hoffman, E.P., Kunkel, L.M. Dystrophin abnormalities in Duchenne/Becker muscular dystrophy. *Neuron* 1989;2:1019-29.
2. Campbell, K.P. Three muscular dystrophies: loss of cytoskeleton-extracellular matrix linkage. *Cell* 1995.:80:675-9.
3. Petrof, B.J. Molecular pathophysiology of myofiber injury in deficiencies of the dystrophin-glycoprotein complex. *Am J Phys Med Rehabil* 2002:81:S162-174.
4. Liu, M., Yue, Y., Harper, S.Q., Grange, R.W., Chamberlain, J.S., Duan, D. Adeno-associated virus-mediated microdystrophin expression protects young mdx muscle from contraction-induced injury. *Mol Ther* 2005:11:245-56.
5. Aartsma-Rus, A., Janson, A.A., Kaman, W.E., et al. Antisense-induced multiexon skipping for Duchenne muscular dystrophy makes more sense. *Am J Hum Genet* 2004:74:83-92.
6. Goyenvalle, A., Vulin, A., Fougerousse, F., et al. Rescue of dystrophic muscle through U7 snRNA-mediated exon skipping. *Science* 306: 2004:1796-9.
7. Partridge, T.A., Morgan, J.E., Coulton, G.R., Hoffman, E.P., Kunkel, L.M. Conversion of mdx myofibres from dystrophin-negative to -positive by injection of normal myoblasts. *Nature* 1989:337:176-9.
8. Brussee, V., Merly, F., Tardif, F., Tremblay, J.P. Normal myoblast implantation in MDX mice prevents muscle damage by exercise. *Biochem Biophys Res Commun* 1998:250:321-7.
9. Gussoni, E., Pavlath, G.K., Lanctot, et al. Normal dystrophin transcripts detected in Duchenne muscular dystrophy patients after myoblast transplantation. *Nature* 1992:356:435-8.
10. Karpati, G., Ajdukovic, D., Arnold, D., et al. Myoblast transfer in Duchenne muscular dystrophy. *Ann Neurol* 1993:34:8-17.
11. Mendell, J.R., Kissel, J.T., Amato, A.A., et al. Myoblast transfer in the treatment of Duchenne's muscular dystrophy [see comments]. *N Engl J Med* 1995:333:832-8.
12. Miller, R.G., Sharma, K.R., Pavlath, G.K., et al. Myoblast implantation in Duchenne muscular dystrophy: the San Francisco study. *Muscle Nerve* 1997:20:469-78.
13. Neumeyer, A.M., Cros, D., McKenna-Yasek, D., et al. Pilot study of myoblast transfer in the treatment of Becker muscular dystrophy. *Neurology* 1998:51:589-92.

14. Thompson, L. Cell-transplant results under fire. *Science* 1992:257:472-4.
15. Huard, J., Bouchard, J.P., Roy, R., et al. Myoblast transplantation produced dystrophin-positive muscle fibres in a 16-year-old patient with Duchenne muscular dystrophy [letter]. *Clin Sci (Colch)* 1991:81:287-8.
16. Huard, J., Bouchard, J.P., Roy, R., et al. Human myoblast transplantation: preliminary results of 4 cases. *Muscle Nerve* 1992:15:550-60.
17. Huard, J., Roy, R., Bouchard, J.P., Malouin, F., Richards, C.L., Tremblay, J.P. Human myoblast transplantation between immunohistoincompatible donors and recipients produces immune reactions. *Transplant Proc* 1992:24:3049-51.
18. Tremblay, J.P., Bouchard, J.P., Malouin, F., et al. Myoblast transplantation between monozygotic twin girl carriers of Duchenne muscular dystrophy. *Neuromuscul Disord* 1993:3:583-92.
19. Tremblay, J.P., Malouin, F., Roy, R., et al. Results of a triple blind clinical study of myoblast transplantations without immunosuppressive treatment in young boys with Duchenne muscular dystrophy. *Cell Transplant* 1993:2:99-112.
20. Hong, F., Lee, J., Song, J.W., et al. Cyclosporin A blocks muscle differentiation by inducing oxidative stress and inhibiting the peptidyl-prolyl-cis-trans isomerase activity of cyclophilin A: cyclophilin A protects myoblasts from cyclosporin A- induced cytotoxicity. *Faseb J* 2002:16:1633-5.
21. Lafreniere, J.F., Mills, P., Tremblay, J.P., Fahime, E.E. Growth factors improve the in vivo migration of human skeletal myoblasts by modulating their endogenous proteolytic activity. *Transplantation* 2004:77:1741-7.
22. Skuk, D., Caron, N.J., Goulet, M., Roy, B., Tremblay, J.P. Resetting the problem of cell death following muscle-derived cell transplantation: detection, dynamics and mechanisms. *J Neuropathol Exp Neurol* 2003:62:951-67.
23. Skuk, D., Roy, B., Goulet, M., Chapdelaine, P., et al. Dystrophin expression in myofibers of Duchenne muscular dystrophy patients following intramuscular injections of normal myogenic cells. *Molecular Therapy* 2004:9:475-82.
24. Skuk, D., Goulet, M., Roy, B., et al. Dystrophin expression in muscles of duchenne muscular dystrophy patients after high-density injections of normal myogenic cells. *J Neuropathol Exp Neurol* 2006:65:371-86.
25. Quenneville, S.P., Chapdelaine, P., Rousseau, J., et al. Nucleofection of muscle-derived stem cells and myoblasts with phiC31 integrase: stable expression of a full-length-dystrophin fusion gene by human myoblasts. *Mol Ther* 2004:10:679-87.
26. Camirand, G., Rousseau, J., Ducharme, M.E., Rothstein, D.M., Tremblay, J.P. Novel Duchenne Muscular Dystrophy Treatment Through Myoblast Transplantation Tolerance with Anti-CD45RB, Anti-CD154 and Mixed Chimerism. *Am J Transplant* 2004;4:1255-65.
27. Stephan, L., Pichavant, C., Bouchentouf, M., et al.. In press. Induction of tolerance across fully mismatched barriers by a non myeloablative treatment excluding antibodies or irradiation use. *J. Cell Transplantation.*
28. Guerette, B., Asselin, I., Skuk, D., Entman, M., Tremblay, J.P. Control of inflammatory damage by anti-LFA-1: increase success of myoblast transplantation. *Cell Transplant* 1997;6:101-7.
29. Benabdallah, B.F., Bouchentouf, M., Tremblay, J.P. Improved success of myoblast transplantation in mdx mice by blocking the myostatin signal. *Transplantation* 2005;79:1696-702.

Address correspondence to

Jacques P. Tremblay, Ph.D.
Human Genetic Unit,
CRCHUL and Laval University,
Quebec, P.Q.,
Canada G0A4P0
jacques-p.tremblay@crchul.ulaval.ca

CSCI AWARDS 2006

CSCI Joe Doupe Lecture: End Stage Renal Disease among Aboriginal People.

Brenda R Hemmelgarn PhD, MD

Department of Medicine, Division of Nephrology, University of Calgary, Calgary, Alberta, Canada
Department of Community Health Sciences, University of Calgary, Calgary, Alberta, Canada

Presented: 28th September 2006

Clin Invest Med 2006; 29 (6): 383–387.

Aboriginal people in Canada bear a disproportionate burden of illness, and are at increased risk for a variety of chronic conditions including diabetes and cardiovascular disease. One condition in particular, end-stage renal disease (ESRD), appears to be increasing at epidemic proportions among Aboriginal people. The reasons for this increase are not entirely clear, but are believed to be related to the increasing prevalence of diabetes mellitus and vascular disease in this population. The outcome for patients with ESRD who start dialysis is dismal. Cardiovascular mortality rates, the most common cause of death among dialysis patients, are 10 to 20 times those of the general population, and dialysis patients also have a significant reduction in their quality of life.

While inequities in health outcomes for Aboriginal people have been demonstrated in other conditions such as cardiovascular disease, trauma and musculoskeletal syndromes, little research has been conducted regarding Aboriginal people with ESRD. It is unclear as to whether or not renal replacement therapy modality, survival, and quality of care post initiation of dialysis varies for Aboriginal and non-Aboriginal people with ESRD. In addition the likelihood of receiving a kidney transplant and survival post kidney transplant for Aboriginals with ESRD has not been studied in the past. The purpose of this review is to discuss these issues as they relate to Aboriginal people with ESRD.

Health of Aboriginal People in Canada

Mortality from infectious diseases among Aboriginal people in Canada has declined over the past century. However despite this decline life expectancy still remains lower for this population compared to non-Aboriginal Canadians [1]. Aboriginal Canadians have nearly a 4-fold greater risk of severe trauma compared to non-Aboriginals, which may be contributing to their higher mortality rates [2]. However the prevalence of chronic conditions and their associated morbidity and mortality is also increasing. Compared to non-Aboriginal people, Aboriginal people in Canada have a two-fold higher prevalence of cardiovascular disease (CVD) [3]. In addition, their rates of hospitalization for CVD related disease conditions have doubled in the past 20 years [4]. This is in contrast to that of society as a whole, where the prevalence of CVD has been decreasing, as have hospitalization rates for CVD related conditions [4].

The prevalence of Type 2 diabetes mellitus has also been increasing in the Aboriginal population, at near epidemic proportions [5]. The prevalence of diabetes among Aboriginal people is reported to be 6.4% [6], with rates among Aboriginal people three to five times higher than the general population, across all age categories and for both males and females. Not only are the rates higher, but cases of Type 2 diabetes are being diagnosed in Aboriginal people at a younger age, including children as young as 5 to 8 years of age [7].

Prevalence and Causes of End-Stage Renal Disease Among Aboriginal People

In the United States the number of Aboriginal people with ESRD has tripled over the past 15 years, with prevalence rates 3.5 times greater than that of

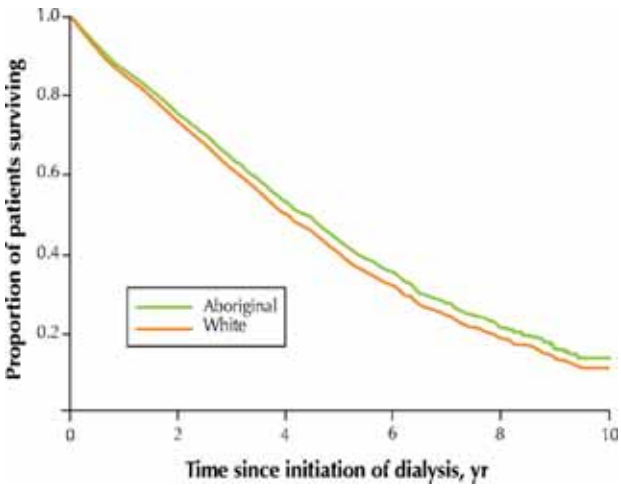


FIGURE 1: Adjusted survival of patients, by race. Adjusted for age, sex, cause of end-stage renal disease, diabetes mellitus, other comorbidity, mode of dialysis, dialysis centre, era effect, socioeconomic status and location of residence. Reprinted from, CMAJ 14-Sept-04; 171(6), Page(s) 577-582 by permission of the publisher. © 2004 Canadian Medical Association.

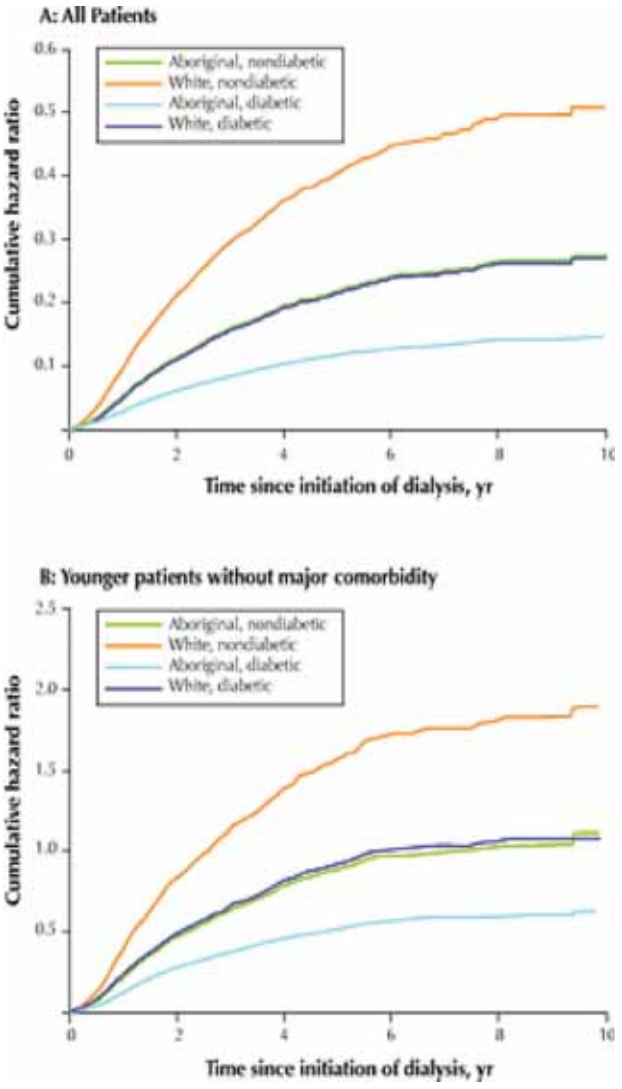


FIGURE 2: Likelihood of renal transplantation by race and diabetic status. Adjusted for age, sex, cause of end-stage renal disease, diabetes mellitus, mode of dialysis, dialysis centre, era effect, socioeconomic status and location of residence. A: All patients commencing dialysis treatment; these data are also adjusted for comorbidity. B: Younger patients without major comorbidity. In both analyses, Aboriginal patients were significantly less likely to receive renal transplants ($p < 0.001$). Reprinted from, CMAJ 14-Sept-04; 171(6), Page(s) 577-582 by permission of the publisher. © 2004 Canadian Medical Association.

Caucasians [8]. Aboriginal people in Canada have experienced disproportionately higher rates of ESRD, with an 8-fold increase in the number of prevalent dialysis patients between 1980 and 2000 [9].

Diabetes is the most common cause of ESRD among Aboriginal people. In the United States in 1999, 68% of all Aboriginals initiating ESRD treatment developed kidney failure due to diabetes, a rate three times higher for Aboriginals than Caucasians [8]. We recently reported similar results based on data from Manitoba, Saskatchewan and Alberta, where diabetic nephropathy was the cause of ESRD in 56.5% of all Aboriginal patients initiating dialysis between the years 1990 and 2000, compared to 26.8% of white patients [10].

Survival and Quality of Care for Aboriginal Patients with End-Stage Renal Disease

While recent studies have highlighted racial inequalities in morbidity and mortality among patients with ESRD, most of these studies have focused on black or Hispanic populations [11]. We conducted a study of all Aboriginal and white adult patients who initiated dialysis in Alberta, Saskatchewan or Manitoba between 1990 and 2000, and followed them until death, transplantation, loss to follow-up or end of the

study (December 31 2001)[10]. A total of 4840 patients commenced dialysis, of whom 685 (15.8%) were Aboriginal. As shown in Figure 1, after adjusting for patient demographics and comorbidities survival post initiation of dialysis was similar for Aboriginal and white patients (Hazard ratio [HR] 0.89; 95% confidence interval [CI] 0.77 – 1.02).

Racial differences in hemodialysis adequacy and outcomes in the North American ESRD population have also been reported, although once again most studies have focused on the African American, Hispanic and Asian ethnic groups and have not included the Aboriginal population[12-15]. We conducted a study of all adults who were established on hemodialysis in Alberta to compare the quality of hemodialysis care between Aboriginal and non-Aboriginal patients [16]. Quality of hemodialysis care was assessed by small solute clearance, blood pressure (BP) control, mineral metabolism, and anemia management. Of the 835 patients, 95 (11.4%) were Aboriginal. Quality of care was similar for Aboriginal compared with non-Aboriginal hemodialysis patients, except for differences in predialysis systolic BP and mineral metabolism, with Aboriginal patients less likely to achieve target levels. The reasons for these differences are unclear, but may be influenced by individual and cultural factors. Explanations for these differences and their impact on long-term morbidity and mortality warrant further investigation.

The choice of dialysis modality, either hemodialysis or peritoneal dialysis, may also vary by patient race. Aboriginal patients with ESRD in Canada are more likely to live in rural locations with a greater distance to travel to a hemodialysis unit than other dialysis patients [17]. Peritoneal dialysis, which would enable the patients to continue to reside in their local community without the need to relocate for dialysis, would seem like the most likely choice of dialysis modality for this population. We found, however, that compared to non-Aboriginals, Aboriginal patients with ESRD in Western Canada were significantly less likely to initiate therapy on peritoneal dialysis compared with white patients (Odds Ratio 0.51; 95% CI 0.40 – 0.65) [18]. Importantly, however, survival among Aboriginal peritoneal dialysis patients seemed similar to both white peritoneal dialysis patients and Aboriginal patients who were treated with hemodialysis, after adjusting for comorbidity. Given the similar survival outcomes, these results suggest that barriers to increase uptake of peritoneal dialysis in the Aboriginal population should be determined and addressed.

Kidney Transplantation among Aboriginal Patients with End-stage Renal Disease

Kidney transplantation is associated with better health outcomes and lower costs, and is the preferred therapy for individuals with ESRD. Racial disparities in access to kidney transplantation have been documented, although most studies have limited detail regarding the Aboriginal population [19]. Using data from the Canadian Organ Replacement Registry, and including all Aboriginal and white patients who commenced dialysis in Western Canada between 1990 and 2000, we found that Aboriginal patients were over 50% less likely to receive a kidney transplant than white patients (Figure 2, Panel A) [10]. This disparity was consistent regardless of living or cadaveric donor source, and persisted even when the analysis was limited to a subgroup of younger patients without major comorbidity (Figure 2, Panel B).

Because renal transplantation is associated with lower rates of death, higher quality of life and lower health care costs than dialysis treatment, reasons for lower transplantation rates among the Aboriginal population are important to determine. In an attempt to better understand there reasons, we also investigated potential barriers to kidney transplantation for Aboriginal patients in Alberta. In this study of all adult hemodialysis patients in Alberta we found that the likelihood of referral for transplantation was similar for Aboriginal and non-Aboriginal people, however Aboriginal people were half as likely to be successfully activated to the transplant waiting list, and that delays early in the course of evaluation were occurring [20]. These results suggest that barriers to transplantation for Aboriginal patients occur after referral, but early in the course of the work-up for eligibility.

Given their often remote location of residence, it is conceivable that distance between residence location and the transplant center may explain the lower rate of kidney transplantation in the Aboriginal population. Using data from a national dialysis registry, we were able to confirm that the majority of Aboriginal patients on dialysis (57%) lived more that 150 kilometers from the nearest transplant center, compared to only 26% of non-Aboriginal patients [17]. Somewhat surprisingly however, the likelihood of transplantation was lower for Aboriginal patients compared to non-Aboriginal patients regardless of the distance they lived from the transplant center, suggesting that remote location of residence does not explain the lower rate of kidney transplantation among Aboriginal people with ESRD in Canada. Further exploration of potential barriers to

kidney transplantation for Aboriginal ESRD patients is the focus of ongoing research.

Summary
Aboriginal people in Canada have an increased prevalence of ESRD, are more likely to initiate renal-replacement therapy on hemodialysis, and are much less likely to receive a kidney transplant compared to non-Aboriginals. Once established on dialysis quality of care and survival is similar for Aboriginal and non-Aboriginal people with ESRD. Understanding issues central to the health and outcomes of Aboriginal patients with ESRD, and documenting potential inequities in care, is essential to guide strategies to develop a new model for delivery of services for management of ESRD among Aboriginal people. Two areas in particular require further investigation for Aboriginal people with ESRD, namely the need to further investigate potential barriers to kidney transplantation, and reasons for lower rates of peritoneal dialysis as a form of renal replacement therapy. Research in this area will enable targeted interventions to be developed in partnership with the Aboriginal communities, with a goal to increase the health outcomes and quality of life for Aboriginal people with ESRD.

Acknowledgements
This work was conducted by members of the Alberta Kidney Disease Network (AKDN), a collaborative group of researchers at the Universities of Alberta and Calgary. The contributions of the investigators of the AKDN are gratefully acknowledged. These studies were supported by operating grants from the Canadian Institutes for Health Research, the Alberta Heritage Foundation for Medical Research and the Kidney Foundation of Canada. Dr Hemmelgarn is supported by a Population Health Investigator Award from Alberta Heritage Foundation for Medical Research and by a New Investigator Award from the Canadian Institutes for Health Research.

References

1. Martens PJ, Sanderson D, Jebamani LS: Mortality comparisons of First Nations to all other Manitobans: a provincial population-based look at health inequalities by region and gender. Can J Public Health 2005;96: Suppl 1:S33-S38,
2. Karmali S, Laupland K, Harrop AR, Findlay CF, Kirkpatrick AW, Winston B, Winston BF, Kortbeek JF, Crowshoe LF, Hameed M: Epidemiology of severe trauma among status Aboriginal Canadians: a population-based study. CMAJ 2005;172:1007-11.
3. Anand SS, Yusuf S, Jacobs R, Davis AD, Yi Q, Gerstein H, Montague PA, Lonn E: Risk factors, atherosclerosis, and cardiovascular disease among Aboriginal people in Canada: the Study of Health Assessment and Risk Evaluation in Aboriginal Peoples (SHARE-AP).Lancet 2001;358:1147-53.
4. Shah BR, Hux JE, Zinman B: Increasing rates of ischemic heart disease in the native population of Ontario, Canada. Arch Intern Med 2000;160:1862-6.
5. Young TK, Reading J, Elias B, O'Neil JD: Type 2 diabetes mellitus in Canada's first nations: status of an epidemic in progress. CMAJ 2000;163:561-6.
6. Statistics Canada. 1991 Aboriginal peoples survey: language, tradition, health, lifestyle and social issues. 1993. Ottawa, Statistics Canada.
7. Dean HJ, Young TK, Flett B, Wood-Steiman P: Screening for type-2 diabetes in aboriginal children in northern Canada. Lancet 1998;352:1523-4.
8. United States Renal Data System. USRDS 2003 Annual Data Report: Atlas of End-Stage Renal Disease in the United States. Bethesda, MD, United States Renal Data System Coordinating Center.
9. Dyck RF: Mechanisms of renal disease in indigenous populations: influences at work in Canadian indigenous peoples. Nephrology 2001;6:3-7.
10. Tonelli M, Hemmelgarn B, Manns B, etal: Death and renal transplantation among Aboriginal people undergoing dialysis. CMAJ 2004;171:577-82.
11. Powe NR: To have and have not: Health and health care disparities in chronic kidney disease. Kidney Int 2003;64:763-72.
12. Frankenfield DL, Ramirez SP, McClellan WM, Frederick PR, Rocco MV: Differences in intermediate outcomes for Asian and non-Asian adult hemodialysis patients in the United States. Kidney Int 2003;64:623-31.
13. Frankenfield DL, Rocco MV, Roman SH, McClellan WM: Survival advantage for adult Hispanic hemodialysis patients? Findings from the end-stage renal disease clinical performance measures project. J Am Soc Nephrol 2003;14:180-6.
14. Wong JS, Port FK, Hulbert-Shearon TE, et al: Survival advantage in Asian American end-stage renal disease patients. Kidney Int 1999;55:2515-23.
15. Pei YP, Greenwood CM, Chery AL, Wu GG: Racial differences in survival of patients on dialysis. Kidney Int 2000;58:1293-9.
16. Chou SH, Tonelli M, Bradley J, Gourishanker S, Hemmelgarn BR: Quality of Care among Aboriginal Hemodialysis Patients. Clin J Am Soc Nephrol 2006;1:58-63.

17. Tonelli M, Hemmelgarn B, Kim AK, et al: Association between residence location and likelihood of kidney transplantation in Aboriginal patients treated with dialysis in Canada. Kidney Int 2006;70:924-30.
18. Tonelli M, Hemmelgarn B, Manns B, Davison S, Bohm C, Gourishankar S, Pylypchuk G, Yeates K, Gill JS: Use and outcomes of peritoneal dialysis among Aboriginal people in Canada. J Am Soc Nephrol 2005;16:482-88.
19. Yeates KE, Schaubel DE, Cass A, Sequist TD, Ayanian JZ: Access to renal transplantation for minority patients with ESRD in Canada. American Journal of Kidney Diseases 2004;44:1083-9.
20. Tonelli M, Chou S, Gourishankar S, Jhangri GS, Bradley J, Hemmelgarn B: Wait-listing for kidney transplantation among Aboriginal hemodialysis patients. Am J Kidney Dis 2005;46:1117-23.

Address for Correspondence:

Dr Brenda Hemmelgarn
Division of Nephrology, Foothills Medical Center
1403 29th St NW
Calgary, Alberta CANADA
T2N 2T9
Phone: (403) 944-2745
Fax: (403) 944-2876
Email: brenda.hemmelgarn@calgaryhealthregion.ca

POSTER AWARDS

Poster Winners 2006

Clin Invest Med 2006; 29 (6): 388–393.

The 2005 Core Medical Residents Research Awards were presented to Dr. Michelle Robinson from the University of Calgary, Dr. Brian Clarke, Dalhousie University, and Dr. Alexandre Boudreault, Université Laval at the Annual Meeting of the Canadian Society for Clinical Investigation. These awards, co-sponsored by the CSCI and the Canadian Association of Professors of Medicine are to recognize outstanding research by core medicine residents and to highlight the importance of research participation as a component of the core medical training experience. Here are the abstracts of their award winning research.

MICHELLE ROBINSON

STANDARDIZED PATIENTS ARE LESS LIKELY TO FAIL STUDENTS ON THE INTERNAL MEDICINE OSCE.

Michelle Robinson, Deirdre Jenkins, Sylvain Coderre, and Kevin McLaughlin. University of Calgary.

The objective structured clinical examination (OSCE) is a commonly used evaluation method. Because it can be difficult to recruit physician evaluators (PE), trained standardized patient evaluators (SPE) have been used. The benefits are that it is cost effective, and alleviates the pressure on physicians. However, there are concerns that SPE's are not qualified to evaluate clinical skill, and grade differently than PE's. The first objective of this study was to assess the correlation between SPE and PE scores for OSCE stations. The second objective was to compare the mean score and pass/fail rate for SPE and PE. On seven stations in the internal medicine clerkship OSCE, students were evaluated simultaneously by a PE and by a trained SPE using the same scoring form. The overall rating of student performance by PE by SPE was collected for all stations. Correlation between scores for PE and SPE was evaluated using Pearson's correlation coefficient. Mean scores for PE and SPE were compared using a paired t-test. The pass/fail rate for PE and SPE was compared using McNemar's discordant pair analysis. Of 178 stations, overall ratings for PE and SPE were available for 123 stations. There was a significant correlation between PE and SPE scores ($r = 0.52$, $p<0.0001$). The mean SPE score was higher than PE score (74.9% vs. 71.2%, $p<0.0001$). On discordant pair analysis the number of students failed by PE/passed by SPE was greater than the

BRIAN CLARKE

number of students failed by SPE/passed by PE (22 vs. 3, p=0.0001). Despite a significant correlation between SPE and PE, we found a significant difference between rating of student performance for SPE and PE. SPE rated student performance higher and were less likely to score performance unsatisfactory. Compared to PE scores the distribution of SPE scores suggests a leniency bias, which may reduce validity of SPE ratings on the OSCE.

A RISK SCORE PREDICTING NON SUDDEN CARDIAC DEATH IN CONGESTIVE HEART FAILURE.

Clarke B, Howlett J, Parkash R. Dalhousie University/ Queen Elizabeth II Health Science Center Halifax, Nova Scotia

Sudden cardiac death (SCD) or non SCD is responsible for the majority of deaths in heart failure patients. The relative risks and predictors of these two modalities of death have not been well defined. A retrospective analysis of 830 patients followed in a tertiary care heart failure clinic from 1998-2004 was performed. Modality of death was reviewed independently by two physicians using predefined clinical criteria in each case. A risk score was constructed using a Cox proportional hazards model to predict non SCD. Mean age was 64.1 $\pm$ 14.7 years, 68.6% were male, mean EF was 32.8$\pm$13.5%, 60.6% were NYHA III/IV, 59.4% had coronary artery disease, and 34.4% had a Charlson Comorbidity Index (CCI) <3. The following variables were used to construct the risk score: age $\geq$ 75 ($p<0.0001$), CCI $\geq$ 3 ($p<0.0001$), NYHA III/IV ($p<0.0001$), creatinine >180mmol/L ($p<0.0001$), a history of ischemic heart disease ($p=0.0083$) and peripheral vascular disease ($p=0.0011$). A risk score $\leq$ 2 predicted a 3.6% risk of non SCD, whereas a score of $\geq$5 predicted a 45.6% risk of non SCD. The risk of SCD remained constant at 5% irrespective of the risk score. A simple clinical risk score can identify patients at a higher relative risk of non SCD compared to SCD. These findings may have implications in optimal selection of heart failure patients for implantable cardioverter defibrillator (ICD) therapy.

SCREENING FOR BK VIRUS ASSOCIATED NEPHROPATHY IN RENAL TRANSPLANTATION WITH BLOOD VIRAL LOAD MEASUREMENT.

Alexandre Boudreault, Chantal Courtemanche, Isabelle Houde, Isabelle Côté, Claude Delage, Eva Latulippe and Louise Deschênes. Hôtel-Dieu de Québec Hospital, Université Laval, Quebec City.

In the past decade, introduction of more potent immunosuppressive agents has led to the emergency of BK virus associated nephropathy (BVAN) in renal transplant recipients. This disease affects 8 % of patients and is now an important cause of graft failure in renal transplant population. Screening for BVAN would reduce graft loss by an early reduction in immunosuppression therapy. Screening is done using urine cytology which has excellent sensitivity but poor specificity. Recently, others have reported promising results of BVAN screening with plasma viral load measurement by quantitative PCR. The goal of this study was to design a quantitative real-time PCR assay for BK viral load measurement and to establish usefulness of this test in BVAN screening in our renal transplant popu-

ALEXANDRE BOUDREAULT

SANDY KISILEVSKY

lation. Since January 2005, a screening protocol for BKVAN including blood BK viral load measurement was used for the graft renal recipient population in the Hôtel-Dieu de Québec hospital. During the study period, 33 patients had a renal biopsy and blood BK viral load measurement. Eight of these patients had a diagnosis of BK virus associated nephropathy after histological examination. All 8 patients with BVAN had a blood BK viral load above 3 X 10^3 copies/ml and 24 of the 27 patients without BVAN had a blood BK viral load under 3 X 10^3 copies/ml. With the real time PCR developed in our institution a cut-off value of 3 X 10^3 copies/ml yield a 100 % sensitivity and a 88,9% specificity for BVAN screening. Our study demonstrates usefulness of blood BK viral load measurement in BK virus associated nephropathy screening.

2006 CSCI Young Investigators' Forum Award Recipients

The Annual Meeting of the Canadian Society for Clinical Investigation once again featured the Young Investigators Forum, an opportunity for students from Canadian MD/PhD and Clinician Investigator Programs to present their research in poster and oral format. Here are this year's award recipients.

Sandy Kisilevsky, a student in the MD Plus program at the University of Calgary is in her 4th year of her PhD in neuroscience at the University of Calgary. Her research is being conducted under the supervision of Dr. Gerald Zamponi, a world leader in the calcium channel field. Using electrophysiology, protein biochemistry, molecular biology and confocal microscopy, she investigates how neuronal calcium channel activity is modulated by different dopamine receptor isoforms. This work has therapeutic implications for neuropathological conditions which result from disturbances in dopaminergic signaling (e.g. schizophrenia and Parkinson's Disease) and is presently supported by funding from the Alberta Heritage Foundation for Medical Research (AHFMR) and the Heart & Stroke Foundation of Canada. After completing the dual degree program, she interested in pursuing post-graduate medical training in neuropa-

KIRSTEN NILES

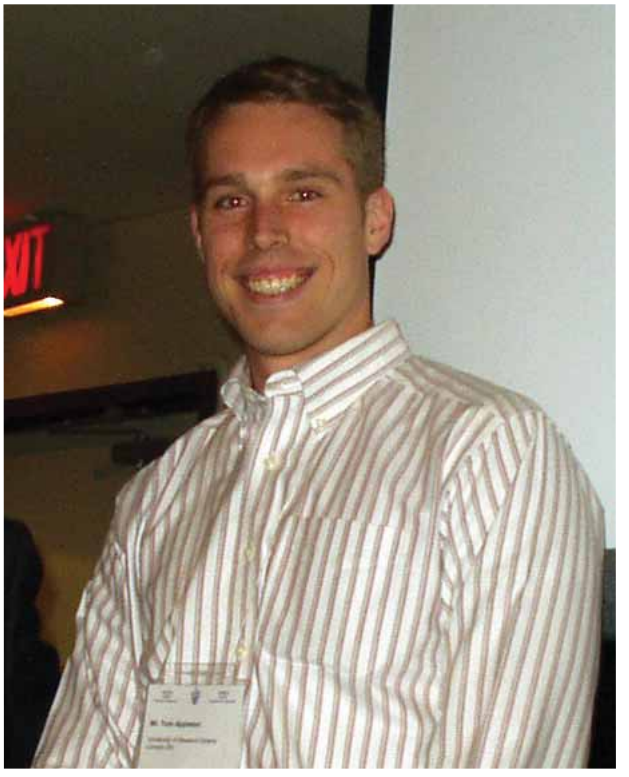
JAMES KENNEDY

thology or neurology. Sandy is an outdoor enthusiast and enjoys long-distance running, triathlon, hiking and snowboarding.

Kirsten Niles is currently a third year MD/PhD student at McGill University. She has just commenced her research in the lab of Dr. Jacquetta Trasler examining epigenetic changes in the male germ line, specifically DNA methylation. Her project involves investigating the global methylation patterns in normal prenatal germ cells and how these patterns change when the system is perturbed by the absence of one or a combination of the methylating enzymes. Her work will move towards attaining a better understanding of the mechanisms of DNA methylation during development of male germ cells. In the future, she hopes to combine her medical degree and PhD in human genetics to focus in pediatric genetics. Outside of academics, Kirsten plays ultimate Frisbee for the McGill Martlets and is a ski instructor at Sugarbush Mountain in Vermont. She also enjoys playing violin and viola. She currently serves as an editor for the McGill Journal of Medicine and as one of two student representatives for the McGill University MD/PhD program.

James Kennedy is an MD/PhD student at the University of Toronto, currently in the final year of graduate studies under the supervision of Dr. John Dick at the University Health Network. His research has focused upon the molecular mechanisms of human leukemogenesis, studying the impact of oncogene expression upon the developmental program of normal human hematopoietic stem and progenitor cells. James has focused particularly upon using this approach to develop in vivo models of human hematologic malignancies in immunodeficient mice. In line with this field, he plans to pursue a residency in hematology/oncology following medical school and hopes to eventually have a career which combines his clinical and research interests.

CLAIRE HESLOP



TOM APPLETON

Claire Heslop is in her third year of the MD/PhD program at the University of British Columbia. In the Atherosclerosis Specialty Laboratory at St. Paul's Hospital's Healthy Heart Lipid Clinic, she is researching the value of biochemical and genetic markers of oxidative stress for the prediction of adverse events in patients with coronary artery disease. Her supervisor is Dr. John S. Hill from the Department of Pathology and Laboratory Medicine at the University of British Columbia, and an iCAPTURE Investigator. At the completion of her program at UBC, Claire plans to apply for a research residency program, and continue working to address the needs of under-served populations.

Tom Appleton is a second year MD/PhD student at the University of Western Ontario's Schulich School of Medicine & Dentistry. Currently, Tom works with Dr. Frank Beier in the field of Osteoarthritis (OA), one of the New Emerging Team (NET) initiatives in Canada co-funded by the Canadian Arthritis Network and CIHR. Tom's project focuses on the identification of signaling pathways controlling articular cartilage degradation due to OA. Integrating histopathological, pharmacological, biochemical, and molecular biology techniques, his project is designed to gain further understanding of the processes involved in the early stages of OA development. This project complements Tom's clinical interests in the OA field, as well as other degenerative diseases.

Arezoo Astanehe is a second year MD/PhD student at the University of British Columbia under the supervision of Dr. Nelly Auersperg and Dr. Sandra Dunn. Her research is focused in understanding the mechanisms involved in ovarian carcinogenesis. Ovarian cancer is the fifth most common cause of death from cancer among Canadian women, and the leading cause of death from gynaecological malignancies.

AREZOO ASTANEHE

MICHAEL WARD

Most cases of ovarian cancer are detected in advanced stage, because symptoms generally appear after the cancers have spread, and screening tests are available only for the detection of advanced cases. Furthermore, the poor success rate in the treatment of women with advanced, recurrent or persistent ovarian cancer has remained largely unchanged for the past few decades. In order to develop effective therapies and to detect disease at an early stage, improvements in our understanding of the processes leading to the initiation and progression of ovarian cancer are required. Arezoo is particularly interested in understanding the contribution of *PIK3CA* oncogene to ovarian carcinogenesis. So far she have shown that p53 tumour suppressor protein negatively regulates *PIK3CA* transcription through direct binding to its promoter, and suggested a mechanism by which loss of p53 function in cancer may lead to increased *PIK3CA* signaling. Her ultimate goal is to develop targeted therapies to treat and improve survival outcomes of women affected by ovarian cancer.

Michael Ward is a 4th year MD/PhD student at the University of Toronto who has been working for two and half years under the supervision of Dr. Duncan Stewart and Dr. Michael Kutryk. His research contributes to the field of cell therapy for the treatment of acute myocardial infarction. Specifically, he studies the effect of coronary artery disease (CAD) risk factors on the capacity of endothelial progenitor cells (EPCs) to stimulate neovascularization and improve myocardial function following injury. Michael has been able to show that patients with multiple CAD risk factors have compromised EPC function, which may reduce the potential benefit of autologous cell therapy for these patients. Using gene therapy approaches, he is currently working on improving the regenerative properties of human EPCs derived from these patients and rescuing their dysfunction. With his supervisors, he hopes that by improving EPC function, they can improve the ability of EPCs to regenerate the myocardium following myocardial injury and translate their work into exciting clinical trials.

INDEX TO VOLUME 29

The title for each article is followed by a code that designates the article's category. Categories used in this volume are:

(Brief) – Brief Report
(S) – Snap Shot
(CC) – Current Comments
(CR) – Case Report
(CSCI/RCPSC) – CSCI/RCPSC Symposium
(E) – Editor's Page
(G) – Guideline
(H) - History
(MP) – Meeting Presentation
(NN) –News and Newsmakers
(OP) - Opinion
(OR) – Original Research
(R) - Review

Clin Invest Med 2006; 29 (6): 394–404.

AUTHORS

Note Jt.Auth=joint author

Z