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Is suicide a gender issue? Annette L Beautrais. Canterbury Suicide Project and Christchurch Health and Development Study, Department of Psychological Medicine, Christchurch School of Medicine & Health Sciences, Christchurch.

Recent publicity about suicide depicts it as a male problem. This paper presents data from two Canterbury studies to illustrate the gender paradox in suicidal behaviour: females make more suicide attempts than males, but males more often die by suicide. The first study compared 202 suicides, 302 suicide attempters, and 1028 randomly selected controls to show that suicide attempts are twice as common in females, but males are 3-4 times more likely to die by suicide.

The second study followed 1265 babies born in Christchurch in 1977 for 25 years and examined gender differences in rates of suicidal ideation, attempt and suicide in this cohort. By age 18, 27.9% of females and 16.9% of males reported a lifetime history of suicidal ideation ($p < .05$). This difference persisted to age 25 (females, 37.5%; males 30.3%, $p < .05$) but from age 19 onwards male and female rates of ideation tended to converge.

Females also had higher rates of suicide attempt. By age 18, 7.1% of females and 3.2% of males had made attempts ($p < .05$). Higher female rates persisted to age 25 (females, 10.4%; males, 6.5%; $p < .05$). However females were less likely to die by suicide than males: By age 25, five males (1.1%, $p < .05$) but no females had died by suicide.

These findings suggest that suicidal behaviour is a problem of both males and females. Females have a propensity for suicidal ideation and attempt, while suicide is a male behaviour. Female choice of method for suicide attempt provides protection from death. Suicide prevention strategies should focus on reducing both suicide attempts and suicide and need to address suicidal behaviour in both males and females to achieve these goals.

Adrenomedullin increases cardiac sympathetic nerve activity in normal conscious sheep. Chris J Charles, David L Jardine, M Gary Nicholls, A Mark Richards. Christchurch Cardioendocrine Research Group, Department of Medicine, Christchurch School of Medicine and Health Sciences, Christchurch.

The sympathetic nervous system and adrenomedullin (AM) both participate in the regulation of cardiac and circulatory function but their interaction remains uncertain. Furthermore, the effect of AM on sympathetic traffic to the heart has not been previously reported. Accordingly, we have examined the effects of AM infusions (33 ng/kg/min for 2 h) on cardiac sympathetic nerve activity (CSNA), haemodynamics

and hormones in eight normal conscious sheep. Each sheep also received on separate days vehicle control and pressure-matched nitroprusside (NP) administration.

Compared with vehicle control, arterial pressure fell similarly (approximately 10 mmHg) with AM ($p=0.04$) and NP ($p<0.001$). Heart rate rose in response to both AM (30%, $p<0.001$) and NP (10%, $p=0.002$) but the rise with AM was significantly greater than that induced by NP ($p<0.001$). Cardiac output increased approximately 40% in response to AM compared with both control and NP (both $p<0.001$). CSNA burst frequency (bursts/min) were increased in response to both AM (50%, $p<0.001$) and NP (10-15%, $p=0.005$) with the rise in burst frequency being greater with AM compared with NP ($p<0.001$). CSNA burst area/min was also raised by both AM (60%, $p=0.03$) and NP (25-50%, $p=0.002$) with a trend for burst area being greater with AM than NP ($p=0.07$). CSNA burst incidence (bursts/100 beats) showed no significant differences between any treatment day.

In conclusion, we have demonstrated that AM is associated with a greater increase in CSNA and heart rate for a given change in arterial pressure than seen with the classical balanced vasodilator NP. These results provide further evidence that AM plays a role in pressure and volume homeostasis.

Predictors of maternal stress in the neonatal intensive care unit. Caron A. Clark,¹ Lianne Woodward,¹ Carole Spencer,² Janet Carter,² Jamie Edgin.¹
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Clear evidence shows that having an infant born prematurely and spending several months in the neonatal intensive care unit is a highly stressful experience for parents. The aims of the present study were to, 1) describe the sources of stress for parents of children born very preterm, 2) describe the factors that are associated with higher levels of stress in these mothers.

124 mothers of infants born very preterm and admitted to the level III Neonatal Intensive Care Unit (NICU) at Christchurch Women's Hospital were interviewed at term equivalent. The interview included the Parental Stressor Scale: Neonatal Intensive Care Unit (Miles et al., 1993), the Edinburgh 10-item Depression Scale (Cox et al., 1987), the Mother and Baby Scales (Brazelton & Nugent, 1995) and custom-written interview questions regarding family background, pregnancy and delivery, feeding experiences and recent life events.

Parents reported their loss of parental role as being the most stressful factor encountered, followed by the fragile appearance of the infant. An examination of infant and parent characteristics related to high levels of stress revealed that maternal state of mind was more important than infant medical risk. Specifically, infant and maternal characteristics such as birth weight, gestation, days on oxygen, days in the NICU unit, maternal age, marital status and socio-economic status did not correspond significantly with high levels of maternal stress. Rather, mothers who had experienced more stressful life events over the past year ($F = 6.048$; $p<0.05$), who were less confident in parenting their baby ($F = 6.787$; $p<0.002$) and who had higher levels of depression ($F=7.038$, $p<0.001$) were more likely to endorse higher levels of stress.

Regression analysis confirmed these findings, with only life event and depression scores contributing a significant amount of variance to reported stress levels.

These findings suggest a need to address maternal stress levels within the NICU. Focus should be given to empowering mothers and encouraging parenting confidence. Such approaches should complement existing care philosophies and help to optimise long-term child outcomes.

Using gene therapy to activate paracetamol for anti-vascular treatment of cancer. Gabi U Dachs,¹ Ally I Watson,¹ Margaret J Currie,¹ Sarah P Gunningham,¹ Joanna Tupper,² Bridget A Robinson,¹ 1:Angiogenesis Research Group, Christchurch School of Medicine and Health Sciences, Christchurch, New Zealand; 2:Royal Holloway, University of London, Surrey, UK.

A functional vascular network is essential for the survival and growth of solid tumours, making blood vessels a key target for therapeutic strategies. Gene transfer represents a targeted alternative to anti-vascular approaches as it can provide a high level of specificity. Gene therapy is a novel cancer treatment which is being tested in 715 clinical trials worldwide. ‘Suicide’ gene therapy consists of two components: delivery of an enzyme-encoding gene followed by the administration of an inactive prodrug which is converted to a toxin by the enzyme.

The peroxidase enzyme from horseradish (HRP) is able to convert benign agents into cytotoxins. Specifically, HRP can convert the well known analgesic agent, paracetamol, into N-acetyl-p-benzoquinoneimine (NABQ), a potent cytotoxin. We wish to employ the HRP/paracetamol combination for vascular targeting for the following reasons: delivery of genes and prodrugs is simplified when targeting the lining of blood vessels; these cells may be particularly sensitive to NABQ since damage to the endothelial cells *in vivo* is the first indication of paracetamol overdose; and even minor damage to the vasculature can amplify into tumour destruction.

We have shown, using clonogenic assays, that HRP-gene-modified cancer cells (FaDu head&neck) were effectively killed by paracetamol *in vitro*. Our pilot studies, using the BrdU proliferation assay, have indicated that human primary endothelial cells (HUVEC) may be inherently more sensitive to HRP/paracetamol treatment than human tumour cells (T24 bladder) and normal human fibroblasts.

These early results support our hypothesis that tumour vascular endothelial cells may be a good target for gene therapy-activated paracetamol.

Ventricular gene expression changes during the development of cardiac hypertrophy in Npr-1 knockout mice. Leigh J Ellmers,¹ Nicola JA Scott,¹ Jarkko Piuhola,^{1,2} Nobuyo Maeda,³ Oliver Smithies,³ Chris M Frampton,¹ A Mark Richards,¹ and Vicky A Cameron.¹ 1:Christchurch Cardioendocrine Research Group, Department of Medicine, Christchurch School of Medicine and Health Sciences, PO Box 4345, Christchurch, New Zealand; 2:Department of Pharmacology and Toxicology, Biocenter Oulu, University of Oulu, Finland; 3:Department of Pathology and Laboratory Medicine, University of North Carolina, Chapel Hill, North Carolina 27599-7525, USA

Cardiac hypertrophy is initially an adaptive response to maintain normal cardiac function after injury or an increase in workload to the heart. Atrial natriuretic peptide (ANP) and brain natriuretic peptide (BNP) regulate cardiac remodelling by inhibiting both Miocene hypertrophy and cardiac fibrosis. To investigate signalling pathways involved during the development of cardiac hypertrophy and fibrosis we used cDNA microarray and quantitative real-time PCR to characterize gene pathways in 8-week and 6-month old male and female Npr1^{-/-} mice (n=6/group). Cardiac contractile responses to elevated ventricular stretch at these two ages were also studied in isolated hearts. The mean arterial pressure was significantly increased by a mean of 32mmHg (p<0.001) in all Npr1^{-/-} (KO) groups compared to wild-type (WT) and heart weight to body weight ratios were increased significantly (p<0.01) in all KO groups except in 6 month old males. cDNA microarray analysis identified 199 genes significantly changed (p<0.05) between 8-week male WT and KO, compared to 273 genes in 8-week females. In contrast, at 6 months 318 genes were significantly changed between WT and KO males, compared to 672 genes in 6-month female mice. Gene pathways identified included those involved in hypertrophy signalling, calcium signalling, structural proteins involved in muscle contraction, fibrosis and cardiomyocyte structure and cell ion channels. Real-time PCR analysis showed significant differences in gene expression of ANP, BNP, Hdac 7a, PKC ϵ , GATA 4, collagen 1, calmodulin 1, phospholamban, TGF- β 1 and GAPDH in KO mice compared to WT. Our results demonstrate the development and progression of cardiac hypertrophy is differentially regulated in Npr1 knockout mice.

The responses of luteinising hormone to interacting peptides: A mathematical model. T John Connolly, David JN Wall, and John J Evans. Laboratory for Cell and Protein Regulation, Centre for Neuroendocrinology, Department of O & G, Christchurch School of Medicine and Health Sciences, and Biomathematics Research Centre, University of Canterbury, Christchurch.

It is now recognised that many intracellular processes may participate in the luteinising hormone (LH) response to gonadotrophin-releasing hormone (GnRH). The complexity is such that it is impossible to understand the overall effect of a stimulatory pulse of GnRH with simple planar concepts. Thus any narrative which attempts to integrate all the processes will be deficient if we allow for the time-dependent nature of the response. For these reasons we have begun to construct a mathematical model of the processes.

To take account of the dynamic nature our model is based primarily on data from perfusion studies. Hemipituitaries from adult female rats at pro-oestrus were

collected, cut in two, and placed in a chamber of a perfusion apparatus. Peptide was delivered as required. The consecutive sampling of LH in the perfusate of the in vitro system allowed construction of a time course of LH release. To analyse the LH response a linear differential equation model was employed to model the profile of LH output.

Our model had characteristics that required a delayed effect, at least two pathways and an ability of one pathway to synergise with the other. We chose Ca²⁺-mediated processes as the first pathway and cyclic AMP-mediated processes as the second pathway. The model incorporates the phenomenon of GnRH self-priming, and synergistic responses to peptides such as oxytocin and neuropeptide Y in association with GnRH. The model describes the dynamic responses that occur in physiological environments.

The prevalence and genetic determinants of inflammatory bowel disease (IBD) in Canterbury. Richard B Gearty,^{1,2} Rebecca R Roberts,³ Ann Richardson,⁴ Christopher AM Frampton,² Martin A Kennedy,³ Murray L Barclay.^{1,2}

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IBD, comprising Crohn's disease (CD), ulcerative colitis (UC) and indeterminate colitis (IC), has increased exponentially in Westernised nations over the last 50 years. Previous NZ studies show a low prevalence of IBD. Mutations of the *CARD15* gene are associated with CD in hospital-based series. This study determined the prevalence and demographic characteristics of IBD in Canterbury, and the frequency of *CARD15* mutations in a population-based cohort of IBD patients for the first time.

IBD patients were recruited from Canterbury using multiple complimentary strategies. They gave informed consent, permission to have clinical notes reviewed, completed a questionnaire and were bled for DNA extraction. Cases were confirmed using standard criteria and demographic data were extracted. *CARD15* mutations were detected using a novel PCR assay. Data were analysed descriptively.

1454 IBD patients (712 CD, 680 UC and 62 IC) were recruited (>90% of Canterbury IBD patients). The prevalence of CD and UC was 153.3/100000, and 146.0/100000 respectively. CD patients were more likely to be female and younger than UC patients. IBD patients were predominantly Caucasian. Interim *CARD15* results (878 patients and 201 controls), show that mutations were significantly associated with CD ($p < 0.001$) in a 'dose-dependent' manner and non-significantly associated with UC ($p = 0.41$).

Contrary to previous NZ studies, IBD is at least as common in Canterbury as other western regions. CD appears slightly more common than UC, previously only seen in one other region. Characteristics of the IBD population are similar to other regions. *CARD15* mutations are less frequent in our population-based cohort than hospital-based cohorts.

Psychosocial and health profiles of pregnant women maintained on methadone: research issues and findings. Gray, A.C.,¹ Woodward, LJ,^{1,3} Spencer C,^{1,2} Quick ZL,¹ Wouldes TA,⁴ and Neha T.¹ 1:Canterbury Child Development Research Group, University of Canterbury; 2:Neonatal Services, Christchurch Women's Hospital; 3:Department of Psychological Medicine, Christchurch School of Medicine and Health Sciences, Christchurch; 4:Health Psychology, Faculty of Medical and Health Sciences, University of Auckland, Auckland.

Research suggests that children of drug-dependent parents are at high risk of a range of adverse outcomes; including behaviour problems, learning difficulties, mental health and substance abuse problems, and juvenile offending. However, relatively little is known about the personal characteristics and care-giving practices of drug-using mothers, which may in part contribute to these later adverse child outcomes. This study draws on prospective longitudinal data from a regional cohort of 41 women on methadone maintenance during their pregnancy and a comparison sample of 32 randomly-identified non-methadone maintained mothers. In the third trimester of pregnancy, all women were interviewed extensively about their pregnancy history, physical and mental health, as well as licit and illicit drug use during pregnancy. This information was cross checked and supplemented with information from women's obstetric and health records. Results showed that pregnant women enrolled in methadone maintenance represent a high risk group, characterised by high rates of educational under-achievement and welfare dependence. In addition to methadone use, participants also reported high rates of licit and illicit drug use, including tobacco, marijuana, benzodiazepines, stimulants and opiates. High rates of mental health and physical health problems were also evident. These findings raise significant concerns about the capacity of these women to provide an ideal caregiving environment for their infants, as well as highlighting the potential need for longer term follow-up and support of these mothers and their infants. Implications for their children's development and future study plans will be discussed.

Vasopressin in Meniere's Disease. Jeremy Hornibrook,^{1,3} Peter George.² 1:Department of Otolaryngology, Christchurch Hospital; 2:Department of Clinical Biochemistry, Christchurch Hospital; 3:Department of Communication Disorders, University of Canterbury, Christchurch.

Meniere's disease is an inner ear disorder characterised by attacks of vertigo, usually accompanied by fluctuating hearing, tinnitus and aural pressure. The fundamental histological feature is an excess of endolymph fluid (endolymphatic hydrops). Japanese investigators have induced endolymphatic hydrops in guinea pigs with vasopressin infusions. They also claim that vasopressin concentrations are elevated in Meniere's patients at the time of the attack, thereby supporting a long-held notion that stress may be a trigger. The objective of this study was to measure vasopressin levels in subjects with a certain (symptoms and electrophysiological proof of hydrops) diagnosis of Meniere's disease at a range of times after their last vertigo attack. The Endolab community normal for vasopressin concentration is < 5 pmol/L. Time of the last vertigo attack was noted when the sample was taken. In 47 subjects (time of last attack: 1 day–years) the mean vasopressin concentration was 2.2 (SD 1.9) pmol/L; in 12 subjects whose last attack was within a week or less the mean concentration was 2.3(1.6) pmol/L, i.e. no significant difference.

A neurorehabilitation tool for off-road assessment of driving ability in subjects with brain disorders. Carrie RH Innes,^{1,2} Richard D Jones,^{1,2,3} Tim J Anderson,^{1,2,4} John C Dalrymple-Alford,^{1,5} Sarah Hayes,⁶ Sue Hollobon,^{1,6} Julie Severinsen,^{1,6} Gwyneth Smith,⁶ and Angela Nicholls.⁶ 1:Van der Veer Institute for Parkinson's Disease and Brain Research; 2:Department of Medicine, Christchurch School of Medicine and Health Sciences, University of Otago; 3:Department of Medical Physics & Bioengineering, Christchurch Hospital; 4:Department of Neurology, Christchurch Hospital; 5:Department of Psychology, University of Canterbury; 6:Department of Occupational Therapy, Burwood Hospital, Christchurch.

Due to physical or cognitive deficits, brain disorders can lead to a decreased ability to drive safely. A battery of 17 computerized sensory-motor and cognitive tests (*SMCTests*TM) has been developed and used in a study to determine the predictive value of *SMCTests* for driving in people with brain disorders.

SMCTests and an independent on-road driving assessment were applied to 50 people with brain disorders referred to the Driving and Vehicle Assessment Service at Burwood Hospital, Christchurch (36 males, 14 females; age 43-85 years, mean age 71.3 years; 35 stroke, 4 traumatic brain injury, 4 Alzheimer's disease, 7 other diagnoses).

Binary logistic regression and nonlinear causal resource analysis (NCRA) were used to build model equations for prediction of on-road driving ability based on *SMCTests* performance. Logistic regression correctly classified 47 of 50 referrals as on-road pass or fail, while NCRA correctly classified 45 of 50 referrals. Leave-one-out cross-validation analysis estimated that logistic regression would correctly predict 86% of an independent referral group as on-road pass or fail, while NCRA would correctly predict 76%.

Accurate estimation of driving ability can minimise on-road assessment of patients who will inevitably fail, thus decreasing an unnecessary risk of accidents. Reducing the proportion of referrals needing an on-road assessment will increase throughput of referrals, shorten waiting lists, and return safe drivers to driving as quickly as possible. *SMCTests* can also identify sensory-motor and cognitive deficits underlying an inability to drive safely so that rehabilitation can, where possible, be optimally applied to reduce such deficits and lead to safe driving.

Cyclosporin monitoring in kidney transplantation: is C₂ really superior? John Irvine, Kelvin Lynn, David McGregor, Martin Searle, Nickolas Cross, Richard Robson. Department of Nephrology, Christchurch Hospital, Christchurch.

Cyclosporin (Neoral®) is an immunosuppressant drug used in kidney transplantation. The aim of this study was to compare the cyclosporin dose, determined by two methods of drug monitoring, with the incidence of kidney rejection or toxicity. C₂ (serum concentration 2 hours after dose) is considered the most accurate marker of area under the curve for the first four hours (AUC₀₋₄) following drug administration. AUC₀₋₄ is a highly sensitive predictor of acute rejection. C₀ is the serum concentration 12 hours after dose.

C₂ monitoring began at Christchurch Hospital in March 2001. Twenty-nine patients with complete records for one-year post transplant used C₂. The results were compared with the same number of patients who had used C₀ immediately prior to March 2001. Serum creatinine, cyclosporin dose and serum concentration were observed at 1, 2, 4, 8 and 52 weeks after transplantation. Rejection or toxicity was determined by clinical judgement and/or biopsy.

The mean creatinine at 52 weeks was 0.14 mmol/L for the C₂ group, compared with 0.12 mmol/L for the C₀ group. The mean dose of cyclosporin was 6.11 mg/kg in the C₂ group and 4.78 mg/kg in the C₀ group (P<0.05). The number of rejections in the C₂ group was 16 (55%) compare with 7 (24%) in the C₀ group (p=0.02). Eleven patients (38%) had cyclosporin toxicity in the C₂ group and 2 (7%) in the C₀ (p=0.005).

Since we introduced C₂ monitoring, the mean cyclosporin dose used and the number of toxic events have increased. There is a trend towards an increased number of rejections in the C₂ group.

Effects of ageing on liver sinusoid caveolin-1 expression. HA Jamieson,¹ VC Cogger,¹ S Hilmer,¹ DG Le Couteur,¹ and R Fraser.² 1:Centre for Education and Research on Ageing and ANZAC Medical Research Institute, University of Sydney, Sydney, Australia; 2:Department of Pathology, Christchurch School of Medicine, Christchurch, New Zealand.

Liver sinusoidal endothelial cells are perforated by 100nm fenestrations. We have previously shown that ageing is associated with a reduction in the frequency of fenestrations (known as defenestration). These changes have implications for a wide range of age-related diseases.¹ One protein that is associated with fenestration formation is caveolin-1. Caveolin-1 is found in vesicular invaginations on the plasma membrane and is particularly abundant in endothelial cells. It is known to play a key role in endothelial vesicular trafficking, signal transduction and vascular permeability.²

In this study we have explored the relationship between liver sinusoidal endothelial cell expression of caveolin-1 and age. Understanding the underlying mechanisms of defenestration could provide means to pharmacologically modify fenestrae and novel ways to treat several age-related diseases

In this study human tissue from our tissue bank of cadaveric liver specimens was processed for immunohistochemistry. Microwave antigen retrieval techniques were employed.³ The primary antibody was caveolin-1 (1:200, Santa Cruz) was and the secondary antibody was anti-rabbit IgG (1:800).

Liver samples from 21 adult (aged 15-64 years) and 27 old (aged over 65 years) were studied. There was an increase in hepatic caveolin-1 expression (odds ratio 3.6, p<0.04) which appeared to be secondary to both increased hepatocellular and liver sinusoidal endothelium staining.

These results suggest that there is increased caveolin-1 expression in the liver sinusoid with age. Increased expression of liver sinusoid endothelial caveolin-1 has also been reported with liver disease where defenestration also occurs.⁴ These results suggest that caveolin-1 expression may be a marker of defenestration. Understanding the link

between defenestration and caveolin-1 expression may provide further insight into the pathogenesis of age-related defenestration.

References:

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Coordinating respiration and nutritive swallowing in the first year of life. Bronwen N Kelly,^{1,4} Maggie Lee Huckabee,^{1,4} Richard D Jones,^{2,4} Christopher M Frampton.^{3,4} 1:Department of Communication Disorders, University of Canterbury; 2:Department of Medical Physics and Bioengineering, Christchurch Hospital; 3:Department of Medicine, Christchurch School of Medicine and Health Sciences, Christchurch; 4:Van der Veer Institute for Parkinson's and Brain Research, Christchurch.

This study monitored the maturation of respiratory and swallowing coordination during feeding in healthy term human infants through the first year of life which has never before been documented. Over 15,000 swallows were obtained from ten term infants monitored during their first year of life. Assessments were made during breast or bottle feeds within the first 48 hours of life and again at one, two, three and four weeks, two, three, six, nine and 12 months of age. Swallows were categorised into five respiratory phase categories and expressed as percentage frequency of occurrence of all swallows (for each infant for every age) prior to statistical analysis. For the group of infants, an average of 64.34% of swallows were followed by expiration (irrespective of age): an adult-like characteristic. Pre- and post-swallow expiration (mid-expiratory swallows) was the dominant pattern of breathing-swallowing coordination within the first 48 hours (mean of 44.11%). Mid-expiratory swallows declined over time such that only an average of 23.93% of swallows occurred in this category and 50.44% at the inspiratory-expiratory cusp by 12 months. These data suggest that while post-swallow expiration is a robust feature of breathing-swallowing coordination during feeding in healthy infants from birth, a shift in the pattern of breathing-swallowing coordination occurs after one week of postnatal feeding experience.

Gene and protein expression changes induced by chronic exposure to the antidepressant paroxetine. MA Kennedy,¹ PC McHugh,¹ GR Rogers,¹ DM Glubb,¹ M Allington,¹ PR Joyce.² 1:Dept Pathology, Christchurch School of Medicine & Health Sciences, University of Otago; 2:Dept of Psychological Medicine, Christchurch School of Medicine & Health Sciences, University of Otago, Christchurch

Our current understanding of the mechanisms underlying patient responses to, and therapeutic actions of, antidepressants is poor. In order to better understand these aspects of drug action, and to determine whether inter-individual genetic differences may contribute to the observed variability in responses, we are exploring the molecular effects of antidepressants in neurons and brain tissue. For this purpose we

have exposed cultured neurons (derived from mouse embryonic stem cells) and laboratory rats to the selective serotonin reuptake inhibitor paroxetine. Microarray and proteomic methods were applied to identify genes and proteins undergoing differential expression as a result of chronic (12 day) exposure to paroxetine. Several proteins and genes with synaptic or brain-specific functions were identified in this screen, and the observed expression changes are currently being validated by real-time quantitative PCR and immunoblotting analysis. Expression changes identified in this screening approach may be relevant to antidepressant function and individual drug responses or they may be unrelated bystander effects, and it is necessary to further evaluate the specificity and relevance of the observed expression changes. However, it is possible that a subset of the novel candidate genes and proteins identified here may contribute to the therapeutic effects of paroxetine or provide pharmacogenetic insights into treatment of depression and related illnesses.

Pitch and loudness effect on the electroglottographic measures in voice patients.

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Electroglottography (EGG), which monitors changes of glottal contact during phonation, has been found useful in detecting abnormal laryngeal behaviours. Since dysphonic patients tend to exhibit limited dynamic range across frequencies and their laryngeal stability may differ from that of normal speakers, examining how pitch and loudness affect EGG measures, such as open quotient (OQ) and speed quotient (SQ), in voice patients will help identify the source of dysphonia. Forty-five subjects (28 females and 17 males), randomly selected from patients seen in a voice clinic over a one-year period, were asked to sustain vowels in four different conditions: habitual loudness at comfortable, high, and low pitch and maximum loudness at habitual pitch. Results of one-way Repeated Measures Analysis of Variances on the male data revealed a significant condition effect for both SQ [$F(3, 44) = 2.899, p = 0.046$] and OQ [$F(3, 44) = 4.001, p = 0.013$]. Post-hoc tests revealed that the high-pitch condition had a significantly lower average SQ than both normal-pitch and maximum loudness conditions while the maximum loudness condition had a significantly lower average OQ than both normal-pitch and high-pitch conditions. No significant condition effect was found in females. Findings from further analysis of data obtained from patients who were classified as one of the three diagnostic groups, namely, “mass lesion”, “hyperfunctional voice with no detectable lesion”, and “glottal incompetence with no sign of lesion or vocal hyperfunction”, suggest that voice patients with lesions or vocal hyperfunction tend to vary laryngeal vibratory patterns by pitch and loudness.

Modelling stochastic insulin sensitivity variability in critical care. J Geoffrey Chase,¹ Jessica Lin,² Dominic Lee,³ Geoffrey M. Shaw,⁴ Thomas Lotz,² Christopher E Hann,⁵ Jason Wong.² 1: Sr. Lecturer, Dept of Mechanical Engineering, Centre for Bio-Engineering, University of Canterbury, Private Bag 4800, Christchurch, and University of Otago, Christchurch School of Medicine and Health Sciences, Christchurch; 2: Research Assistant, Dept of Mechanical Engineering, Centre for Bio-Engineering, University of Canterbury, Private Bag 4800, Christchurch; 3: Senior Lecturer, Dept of Mathematics and Statistics, University of Canterbury, Private Bag 4800, Christchurch. 4: Consultant and Sr. Lecturer, University of Otago, Christchurch School of Medicine and Health Sciences, Christchurch, and Canterbury District Health Board, Dept of Intensive Care Medicine, Christchurch Hospital, Christchurch; 5: Post-Doctoral Fellow, Dept of Mechanical Engineering, Centre for Bio-Engineering, University of Canterbury, Private Bag 4800, Christchurch.

Hyperglycaemia is prevalent in critical care, and tight control can reduce mortality by 29–45%. Targeted control of glucose levels in critical care patients relies on frequent fitting and prediction of a patient's modelled insulin sensitivity, S_I . This parameter varies significantly in the critically ill due to the evolution of their condition and drug therapy. This research presents a stochastic model of the S_I variation that enables better prediction and control of glucose levels.

A three-dimensional stochastic model of S_I variability is constructed using retrospective data from 18 long term critical care patients. The stochastic behaviour of the hourly variation in fitted S_I is a function of its magnitude over the physiological range. The three-dimensional model permits the stochastic parameter behaviour to match the retrospective data without having a known distribution forced upon it. The stochastic model defines the distribution of blood glucose levels one hour following a known insulin and/or nutrition intervention, and thus enables more knowledgeable and accurate prediction for glycaemic control.

Across the 18 patients, 454 out of 460 measurements (98.7%) were within the 0.95 probability interval the stochastic model produces. The stochastic model is a tool to assist clinical glycaemic control intervention using currently developed glucose-insulin models and control protocols. In particular, the probability interval generated in predicting patient response to insulin further prevents hypoglycaemic episodes and provides tighter predictive control. The model also enables “virtual patients” to be generated for Monte Carlo simulation of new control protocols, overcoming the need to use limited numbers of clinical trials available for study.

Model-based assessment of insulin resistance in broad populations. J Geoffrey Chase,¹ Thomas F Lotz,¹ Kirsten A McAuley,² Geoffrey M Shaw,³ Chris E Hann,¹ Jessica Lin,¹ Jim I Mann.² 1: Centre for Bioengineering, Department of Mechanical Engineering, University of Canterbury, Christchurch; 2: Edgar National Centre for Diabetes Research, Dunedin; 3: Department of Intensive Medicine, Christchurch Hospital, Christchurch

Insulin resistance is a major risk factor for type-2 diabetes and cardiovascular disease and its early diagnosis can significantly reduce further complications. Hence, there is a great need for simple, accurate assessment of this marker in broad populations to

enable early intervention and regular quantification of changes due to clinical intervention.

A physiological three compartment model of the glucose/insulin metabolism is fit to euglycaemic-hyperinsulinaemic clamp data (N=195) to assess the metabolic information it can provide during steady and transient states. The insulin model consists of two differential equations modelling the main transports and losses of insulin in plasma and interstitium. The glucose model describes the pharmacokinetics of plasma glucose, dependent and independent of insulin.

Based on the results of this study, a new dynamic model-based insulin sensitivity test is proposed. The objectives for the test are: short duration, simple protocol, physiological dosing and low cost, with high accuracy. Pancreatic insulin secretion is estimated by plasma C-Peptide concentrations. Simulations on clamp trial data are used to assess the performance of this method.

Clamp data validation yielded mean errors of 7%. The model-based insulin sensitivity parameter, S_I , correlated $r^2=0.96$ in steady state and $r^2=0.88$ at a transient state. Simulations of the proposed insulin sensitivity test resulted in correlations with clamp assessed ISI of $r^2 > 0.80$. These results are significantly better than similar attempts in the literature.

The model is able to effectively capture all the dynamics of the euglycaemic-hyperinsulinaemic clamp. The model-based insulin sensitivity parameter highly correlates to the equivalent ISI from the clamp. This result led to the development of a new simple test to assess insulin resistance, applicable in clinical settings and broad population studies. First simulations show promising, highly correlated results.

The Christchurch Tissue Bank: a biorepository for cancer research. Helen R Morrin,¹ Sarah P Gunningham,¹ Margaret J Currie,¹ Gabi U Dachs,¹ Stephen B Fox,² and Bridget A Robinson,³ 1:Department of Pathology, Christchurch School of Medicine & Health Sciences, Christchurch; 2:Department of Clinical Laboratory Sciences, University of Oxford, UK; 3:Department of Oncology, Christchurch Hospital, Christchurch, New Zealand.

Breakthroughs in the understanding of cancer biology, and the development of novel treatments are increasingly dependent on accessing human cancer tissues with their associated clinicopathological data. The Christchurch Tissue Bank (CTB) has been established to meet this need, by developing a central repository of consented cancer tissues for genomic and proteomic studies. It is a collaboration involving staff from the Christchurch School of Medicine and Health Sciences and the Canterbury District Health Board. The CTB operates to international biorepository standards but must also comply with New Zealand's ethical, legal and cultural requirements.

The development of tissue banking is reported, including the number and type of samples collected. Samples have been banked from more than 2000 donors. Our consent form has evolved over time with new ethical guidelines and cultural consultation. Donor options have been added to allow some choice in how the gifted tissue may be used. Most donors (99.6%) consented to allow access to medical records, 98.3% to their tissue being sent overseas to research collaborators, and 97.4% to their tissue being used in research with commercial collaborators. Since this option

became available and regardless of ethnicity, 35.6% of donors, requested sample disposal with a *karakia*, at the end of a study. A secure relational database of all tissue and associated clinicopathological data within the bank is maintained to protect patient privacy, facilitate the optimal matching of tissue to research projects and provide an auditable trail from donation to allocation.

The CTB is successfully providing quality tissue samples for cancer research whilst appropriately addressing ethical, legal and cultural aspects of their collection.

Analysis of cells by atomic force microscopy. James J Muys,¹ Maan M Alkaisi,¹ Junko Nagase,² and John J. Evans.²; 1:MacDiarmid Institute for Advanced Materials & Nanotechnology, Electrical and Computer Engineering Dept, University of Canterbury, Christchurch; 2:Laboratory for Cell and Protein Regulation, Centre for Neuroendocrinology, Christchurch School of Medicine and Health Sciences, University of Otago, Christchurch.

Currently, optical microscopy techniques are the primary method for cell visualization and microscale features have traditionally been used for diagnosis and classification. However, because the differences in characteristics can be subtle, accurate investigation can be challenging and ambiguous. Resolution limits due to the wavelength of light used in optical microscopy cause finer details to be neglected, which if utilised could provide more accurate characterisation.

The atomic force microscope (AFM) is a probing-based instrument enabling surface imaging of living biological systems and their components at high resolution and in real time. Using the nanoscale capabilities of the AFM, a detailed understanding of cell topography can be resolved. Furthermore, when incorporated with a Biochip platform designed to position cells at known locations for single-cell studies using microelectrodes, a rapid and organised process for identification and characterisation is introduced.

Pituitary cells were prepared as a single cell suspension in culture medium by standard methods. A biochip platform for the precise trapping of cells using microelectrodes was developed. The biological cells were transferred to the platform and imaged by AFM. Particular focus of this study was utilizing the AFM to image the fusion pore at the cell membrane, through which hormones are released from the cell interior to the peripheral circulation and subsequent transport to the target organ. Pores in the cell membrane were observed in characteristic clusters. The increased information on cell behaviour will yield a better understanding of the secretion of hormones from the pituitary gonadotroph cell.

Genetic polymorphisms as predictors of clinical outcome after acute myocardial infarction. Barry R Palmer, Teresa E Baird, Richard P Collins, Anna P Pilbrow, Lorraine Skelton, Chris M Frampton, Tim G Yandle, A Mark Richards, Vicky A Cameron. Christchurch Cardioendocrine Research Group, Department of Medicine, Christchurch School of Medicine & Health Sciences, University of Otago, Christchurch

Genetic association studies have shown potential for defining at-risk patient groups and may allow risk stratification to occur well before the onset of serious heart disease

symptoms. Studies aimed at defining a selection of genetic polymorphisms with predictive value for clinical outcome after myocardial infarction (MI) using DNA samples from 985 post-MI (PMI) patients are in progress. The PMI cohort was recruited between November 1994 and June 2001, 78.4% of patients were male and the mean age of patients of 62.2 years. DNA samples from the PMI cohort have been genotyped for polymorphisms from 7 candidate genes, implicated with a role in heart disease. Polymorphisms in the angiotensin converting-enzyme (ACE intron 16 I/D), aldosterone synthase (CYP11B2 C-344T) and AMP deaminase I (AMPD1 C34T) genes have significant univariate associations with survival in the PMI cohort. PMI patients with at least one copy of the ACE gene D allele had twice greater covariate-adjusted mortality after MI than those with II genotype ($p=0.047$). PMI patients homozygous for the CYP11B2 -344T allele had 3 times lower covariate-adjusted mortality after MI than those with CC or CT genotype ($p=0.005$). PMI patients with the AMP deaminase I T34 allele and a previous history of MI were at greater risk of death ($p<0.001$). Hazard ratios for these genetic predictors compare favourably with established predictors such as age, plasma N-terminal brain natriuretic peptide levels and left ventricular ejection fraction. This suggests genetic profiling may ultimately assist in improved targeting of treatment to individuals.

Glial activation precedes neurodegeneration in ovine Batten disease, begins during prenatal brain development, and spreads from specific foci associated with later symptoms. DN Palmer,¹ MJ Oswald,¹ GW Kay,¹ P Rezaie,² JD Cooper.³ **1:Agriculture and Life Sciences Division, Lincoln University; 2:Department of Biological Sciences, The Open University, Milton Keynes, UK; 3:Department of Neuroscience, Institute of Psychiatry, King's College London, UK.**

Batten disease (neuronal ceroid lipofuscinoses NCLs) are fatal inherited neurodegenerative diseases of children characterised by brain atrophy, and the accumulation of lysosomal storage bodies containing mainly a single protein. Little is known of the pathogenesis and traditional ideas are based on observations in humans, made after the development of clinical signs.

Animal forms of Batten disease include a flock of affected sheep with pathology closely resembling that seen in humans. The development of preclinical CNS pathology was studied in these sheep, by comparing a series of 12 age-matched affected and control brains, 6 prenatal and 6 after birth.

Differences noted in affected brains were: 1. Perivascular macrophages were more activated 130 days after conception. 2. Astrocytes were activated in affected grey and white matter at this age, microglia were activated in affected grey matter at birth, and MHC II activated glia detected only 12 days after birth. 3. Focal clusters of activated microglia were evident in outer layers of affected occipital and somatosensory cortical regions at only 12 days. Glial activation was not seen in control brains.

Glial activation was progressive, regionally defined, and preceded the degeneration of different cortical layers and brain areas, foremost affected being regions associated with clinical symptoms. Widespread generalized neurodegeneration followed. The peri-natal onset of glial activation suggests that glia may be central to NCL pathogenesis, which begins during brain development. Storage body accumulation

was more evenly spread across regions at all ages, suggesting that neurodegeneration and storage body accumulation are independent manifestations of disease.

Vitamin D analogues for the treatment of bone disease outcomes in chronic kidney disease: a systematic review. Suetonia C Palmer,¹ David O McGregor,¹ Jonathan C Craig,² Giovanni FM Strippoli.² 1:Department of Nephrology, Christchurch Hospital, Christchurch, New Zealand; 2:Cochrane Renal Group, NHMRC Centre for Clinical Research Excellence in Renal Medicine, The Children's Hospital at Westmead, Westmead, Australia.

Bone disease is a universal complication of chronic kidney disease (CKD) and reduces quality of life. However, treatment with vitamin D to ameliorate musculoskeletal morbidity causes hypercalcaemia, and tissue calcification. We used systematic review methods to examine the benefits and harms of vit D treatment in CKD.

A comprehensive literature search was conducted for all randomised controlled trials (RCTs) of vit D in CKD. Data were extracted for mortality, bone-related outcomes, hypercalcaemia, hyperphosphataemia, and parathyroid hormone (PTH). Treatment effects were summarised as a relative risk (risk ratio, RR) or standardised mean difference (SMD) with 95% confidence interval (CI).

Sixty-five of 1,272 articles identified were eligible. Eighteen compared vit D with placebo, and 11 compared newer vit D with placebo or another vit D. No beneficial effect of treatment on mortality, fracture, or bone pain for any comparison was found. Any vit D reduced the end of treatment PTH concentration (9 RCTs, 350 patients, SMD -0.53; 95% CI, -0.77 to -0.29) at the expense of increased episodes of hypercalcaemia (9 RCTs, 540 patients; RR 2.55; 95% CI 1.36 to 4.81) and hyperphosphataemia (2 RCTs, 82 patients; RR 2.84; 95% CI 1.13 to 7.13). The newer vit D analogues also suppressed PTH secretion (3 RCTs, 163 patients; SMD -0.52; 95% CI -0.84 to -0.21) but without increasing the risk of hypercalcaemia (3 RCTs, 163 patients; RR 5.13; 95% CI 0.57 to 46.29).

A cautious recommendation for the use of the newer vit D sterols to reduce treatment related toxicity is made until further, larger trials are completed.

Association of angiotensinogen M235T and T174M gene polymorphisms with mortality in heart failure. Anna P Pilbrow, Barry R Palmer, Chris M Frampton, Tim G Yandle, Richard W Troughton, Elizabeth Campbell, Lorraine Skelton, John G Lainchbury, A Mark Richards, and Vicky A Cameron. Christchurch Cardioendocrine Research Group, Department of Medicine, Christchurch School of Medicine and Health Sciences, University of Otago, Christchurch

The renin-angiotensin system contributes to adverse ventricular remodelling in heart failure (HF) patients. Two polymorphisms of the angiotensinogen gene (AGT M235T and T174M) have been individually associated with elevated levels of plasma AGT, hypertension or left ventricular hypertrophy. The aim of this study was to investigate association of these polymorphisms, separately and in combination, on clinical outcome in HF patients admitted to Christchurch Hospital.

In HF patients genotyped for AGT M235T (n=451) and T174M (n=448) polymorphisms, interactions between genotype, hormonal prognostic markers, echocardiography measures and clinical outcome were investigated. Mortality was recorded over a median 4.2 years follow-up. Patients carrying the 235TT genotype (n=84) were 3 years younger at admission (age±SEM: MM, 76.0±0.8; MT, 74.1±0.8; TT, 71.6±1.3 p=0.011) and, in those with documented hypertension, diagnosis was made 8 years earlier than other patients (age±SEM: MM, 54.2±1.9; MT, 55.1±1.9; TT, 46.0±3.6 p=0.038). Patients carrying one or more 174M alleles (n=93) were more likely to have a previous history of HF (p=0.044) and had higher mortality (p=0.060) compared with 174TT homozygotes (n=355), despite having significantly better cardiac function as indicated by echocardiography. Patients with 'high-risk' haplotypes (AGT 235TT combined with either 174TM or MM) had a 1.8-fold (95%CI: 1.25-2.64) increased risk of dying during the follow-up period. These haplotypes predicted mortality independently of established risk factors, including neurohormonal status. The percentage of deaths that could be attributed to 'high-risk' AGT haplotypes in this HF cohort was 21%. This study indicates an association between the AGT polymorphisms, M235T and T174M, and increased mortality in HF.

Amino-terminal proCNP: a putative marker of growthplate activity in post natal growth. Timothy CR Prickett,¹ Adrienne M Lynn,² Martin Wellby,³ Graham K Barrell,³ Brian A Darlow,^{1,2} Eric A Espiner,¹ A Mark Richards,¹ Timothy G Yandle.¹ 1:Department of Medicine, Christchurch School of Medicine & Health Sciences; 2:Department of Pediatrics, Christchurch Hospital; 3:Agricultural and Life Sciences Division, Lincoln University.

C-type natriuretic peptide (CNP), acting via its specific receptor NPR-B, is an important regulator of endochondral bone growth. We have recently identified a stable product of proCNP, amino-terminal proCNP (NT-proCNP), which unlike CNP is readily measurable in human and ovine plasma. Hypothesizing that plasma NT-proCNP concentrations reflect in part CNP synthesis within growth plates of rapidly growing cartilage, we studied levels of CNP forms in both children and lambs and related these to age, growth velocity and biochemical markers of bone turnover. Plasma NT-proCNP levels were elevated at birth and fell progressively with age. Significant associations between plasma NT-proCNP and height velocity ($r^2=0.32$, $p=0.005$), plasma alkaline phosphatase activity (ALP, $r^2=0.30$, $p<0.001$), and Type 1 collagen C telopeptide ($r^2=0.11$, $p=0.013$) were identified in children (n=60) aged 5 to 18 years. In longitudinal animal studies, elevated plasma concentration of NT-proCNP in 1 week old lambs (61.7 + 1.9 pmol/L, n = 24) fell progressively to mature adult levels (26.7 + 0.9 pmol/L, $p<0.001$) at age 27 weeks. Plasma NT-proCNP showed a highly significant association with ALP ($r^2=0.89$, $p<0.001$) and metacarpal growth velocity ($r^2=0.31$, $p<0.001$). Glucocorticoid administration (dexamethasone 0.25mg/kg/day for 15 days), a treatment known to inhibit cartilage proliferation, reduced metacarpal growth elongation ($p<0.001$) in 4-week-old lambs (n=8) and markedly lowered circulating NT-proCNP levels ($p<0.001$) during the treatment period. In summary, NT-proCNP levels in blood show a strong association with growth velocity and markers of bone formation and may well serve as a useful marker of growth plate activity in humans and other mammals.

Clinical and neurobehavioural outcomes of infants exposed to methadone during pregnancy. Woodward LJ,^{1,2} Quick Z,¹ Spencer C,^{1,3} Woules TA.⁴

1:Canterbury Child Development Research Group, University of Canterbury, 2:Department of Psychological Medicine, Christchurch School of Medicine and Health Sciences; 3:Christchurch Women's Hospital, Christchurch; 4:Health Psychology, Faculty of Medical and Health Sciences, University of Auckland, Auckland.

Methadone maintenance is the most widely used treatment for opiate addiction in pregnant women. However, despite a large number of proven benefits associated with methadone maintenance, the teratological effects of this drug on the developing foetus are poorly understood. To examine the effects of prenatal methadone exposure on infant health and neurobehavioural development and in particular, whether a relationship exists between maternal methadone dose and infant outcomes, a regional cohort of 73 mother-infant dyads were recruited from the Canterbury region. This cohort consisted of two groups; 41 of these dyads were recruited consecutively from the CADS methadone maintenance programme, and 32 were randomly identified non-methadone exposed controls. Prior to birth, all pregnant women completed an extensive structured interview covering areas such as pregnancy history, physical and mental health and licit and illicit drug use. Following birth, clinical data and hospital information on each child was collected. At around 42 + 1 week gestation infants underwent a comprehensive, neurobehavioural evaluation using the NICU Network Neurobehavioural Scale (NNNS). Results revealed significant ($p < 0.05$) differences between the two groups with methadone exposed infants tending to be lighter, shorter, have a smaller head circumference at birth and a longer duration of hospital stay. Exposed infants also had a lower mean gestational age at delivery, as opposed to their control counterparts. On the neurobehavioural (NNNS) assessment methadone exposure was significantly ($p < 0.05$) related to increased infant dysregulation, poorer sustained alertness, higher excitability, increased arousal and a tendency to habituate less well. More refined analyses also provided support for the presence of a dose-response relationship with several neurobehavioral component measures showing significance ($p < 0.05$). These findings support the presence of high levels of behavioural disorganisation amongst infants prenatally exposed to methadone and offers evidence that a dose-response relationship between methadone exposure and adverse neurobehavioural outcome does exist.

Changes in gene expression and the role of the natriuretic peptides in cardiac development. Nicola Scott,¹ Leigh Ellmers,¹ John Lainchbury,¹ Nobuyo Maeda,² Oliver Smithies,² and Vicky Cameron.¹ 1:Department of Medicine, Christchurch School of Medicine and Health Sciences, PO Box 4345, Christchurch, New Zealand; 2:Department of Pathology and Laboratory Medicine, University of North Carolina, Chapel Hill, NC 27599-7525, USA.

Atrial (ANP) and Brain (BNP) natriuretic peptides are hormones that protect against the adverse changes in heart structure and function known as cardiac remodelling. The Npr-1 receptor mediates both ANP and BNP bioactivity, and mice that lack the Npr-1 gene exhibit cardiac remodelling. We have observed that Npr-1 knockout (KO) mice have decreased survival during gestation and the neonatal period, proposing a

previously unrecognized role of the natriuretic peptides in fetal cardiac development. Cardiac anatomy and gene expression profiles were compared in Npr-1 KO and wild-type (WT) hearts at three key time points in cardiac development, 12.5 and 15.5 days post coitum (dpc) and neonatal day one, in male and female mice (n=6 per group). Npr-1 KO mice had significantly larger hearts from 15.5 dpc ($p<0.05$). Microarray analysis on 22k oligo arrays indicated the altered gene expression of at least 3,000 genes ($p<0.05$), including genes involved in cardiac structure, developmental axis formation, regulation of transcription, cell proliferation and hypertrophy. The extent of altered expression of selected cardiac and developmental genes was further investigated through real-time PCR analysis. ANP expression was seen to be significantly increased in Npr-1 KO mice from 12.5 dpc ($p<0.001$), similar trends were observed for BNP expression. Interestingly, the cardiogenic transcription factors investigated (Mef2A, Mef2C, GATA-4 and GATA-6), were all down regulated at 12.5 dpc, with relative over-expression observed at 15.5 dpc. In summary, in addition to their well-characterised cardioprotective effects, the natriuretic peptide family appears to interact extensively with several signalling pathways regulating cardiac development.

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Outcomes of Christchurch Early Intervention for Psychosis Service. Mark A Turner. Totara House, Mental Health Division, Canterbury District Health Board, Christchurch.

Early intervention for psychosis (EIP) is a relatively new approach to managing and ameliorating the potentially devastating impact of psychosis in bipolar disorder, schizophrenia spectrum disorders and other psychoses. Internationally, little research has been published on the effectiveness of EIP Services. The present study examines the outcomes of 180 patients with first episode psychosis consecutively accepted to Totara House EIP service. Patients are assessed on a range of psychometric instruments at baseline, twelve months and discharge.

Overall, clients who remain in the service for a minimum of twelve months had a significant improvement in: i) symptoms, as assessed by the Positive and Negative Syndrome Scale (PANSS) $t(99) = 8.21$; $p<0.001$ ii) quality of life, as assessed by the Heinrichs Quality of Life Scale (QLS) $t(95) = -3.88$; $p<0.001$ iii) functioning as assessed by the Health of the Nation Outcome Scale (HoNOS) $t(96) = 8.42$; $p<0.001$ and Global Assessment of Functioning (GAF) $t(100) = -10.07$; $p<0.001$. In addition, those people with a long duration of untreated psychosis at baseline had significantly poorer outcomes at discharge, while those with a diagnosis of schizophrenia spectrum disorder did not.

These results suggest that although improvements can be made to the level of symptoms, quality of life and general functioning of those with first episode psychosis while in treatment, those with a long duration of untreated psychosis generally have a poorer outcome. Efforts to decrease duration of untreated psychosis may lead to greater improvements for this challenging population.

Vitamin C is essential for neutrophil apoptosis. Implications for the resolution of inflammation and death by scurvy. Margret CM Vissers. Free Radical Research Group, Pathology Dept., Christchurch School of Medicine and Health Sciences, Christchurch.

Vitamin C (L-ascorbic acid) is a versatile antioxidant essential for life. Humans require dietary intake to maintain good health, and deficiency leads to the development of scurvy. The devastating symptoms of this disease are thought to result from decreased collagen hydroxylation by the Fe-containing hydroxylases that require ascorbate as a co-factor, although this remains unproven. Tissue levels of ascorbate vary, and high intracellular concentrations indicate an important function. Neutrophils, the primary defence against micro-organisms, maintain high ascorbate concentrations, but its role in these cells has never been determined.

In this study I show that ascorbate profoundly affects neutrophil apoptosis. Using *Gulo* ^{-/-} mice, that are unable to synthesise ascorbate, I found that apoptosis was prevented in ascorbate-deficient neutrophils. Both spontaneous apoptosis and phosphatidylserine (PS) exposure in stimulated neutrophils were inhibited. Initially there was an increase in cell survival, but within 24h cell death occurred by necrosis. Neutrophils lacking ascorbate had elevated levels of the transcription factor hypoxia-inducible factor (HIF)-1 α , the primary oxygen sensor that also inhibits neutrophil apoptosis under hypoxic conditions. This factor is regulated by Fe²⁺-dependent hydroxylases requiring ascorbate for optimal activity.

My results indicate that up-regulation of HIF-1 α in ascorbate-deficient neutrophils blocks apoptosis under normoxic conditions. Protection of neutrophil apoptosis therefore represents a critical role for vitamin C. Widespread neutrophil necrosis in ascorbate deficiency provides a new explanation for the devastating symptoms of scurvy, suggesting that this, rather than lack of collagen hydroxylation causes the breakdown of extracellular matrix collagen.

Role of NADPH oxidase – derived oxidants in neutrophil apoptosis. Rachel Wilkie, Margret Vissers, Mark Hampton. Department of Pathology, Free Radical Research Group, Christchurch School of Medicine & Health Sciences, University of Otago, Christchurch.

When neutrophils ingest bacteria by phagocytosis the active NADPH oxidase complex is assembled, generating large amounts of reactive oxygen species to facilitate bacterial killing. After the infection is cleared neutrophils undergo apoptosis (programmed cell death), whereby macrophages ingest the neutrophils before they release their cytotoxic contents. The pathways leading to apoptosis following phagocytosis are currently unclear. It is proposed that oxidants derived from the NADPH oxidase have a fundamental role, but there are conflicting reports. The objective of our study was to use diphenyleneiodonium (DPI), an inhibitor of the NADPH oxidase, and neutrophils isolated from an X-linked gp91phox knockout mice with a non-functional NADPH oxidase to determine exactly what role NADPH derived oxidants have in the execution and resolution of apoptosis. We show a 3.5-fold increased uptake of phagocytic neutrophils by human macrophages does not occur when the NADPH oxidase is not functional. Oxidants derived from the NADPH oxidase trigger exposure of a 4-fold increase in expression of surface marker

phosphatidylserine, which is critical for the clearance of apoptotic cells. The phosphatidylserine exposure and macrophage uptake is not caspase-mediated, indeed, we show that NADPH-derived oxidants actually reduces caspase activity 2-fold as assessed by enzyme assays and immunofluorescence. These results may help to explain the formation of granuloma in chronic granulomatous disease (CGD), a human condition wherein patients have a defect in their NADPH oxidase. Their inability to generate oxidants may result in impaired neutrophil apoptosis and clearance leading to a dysfunctional inflammatory response.

Clinical trials of active and adaptive insulin and nutrition control to control hyperglycaemia in critically ill patients. XW Wong,³ JG Chase,¹ GM Shaw,² J Lin,³ T Lotz,³ and CE Hann.⁴ 1:Department of Mechanical Engineering, University of Canterbury, Christchurch School of Medicine and Health Science, University of Otago, Christchurch; 2:Department of Intensive Care, Christchurch Hospital, Christchurch School of Medicine and Health Science, University of Otago, Christchurch; 3:Department of Mechanical Engineering, University of Canterbury, Christchurch; 4:Department of Mechanical Engineering, University of Canterbury, Christchurch.

Stress-induced hyperglycaemia caused by increased hepatic glucose production and insulin resistance is prevalent in intensive care, impairing the immune response. Nutritional support regimes with high glucose content further exacerbate the problem. Tight glucose control can reduce mortality by up to 45% if levels are kept below 6.1mmol/L. This research develops an adaptive control algorithm with variable insulin and nutritional inputs for targeted glucose control of critically ill patients. Clinical trials are performed for verification.

Proof-of-concept clinical trials were conducted on intubated, insulin-dependent Christchurch ICU patients (n=7). A target 10-15% reduction in glucose level per hour for a desired glucose level of 4-6mmol/L was set. 43% and 91% of glucose targets were achieved within $\pm 5\%$ and $\pm 20\%$ respectively for a one-hour prediction window. The mean error was 8.9% (0.5mmol/L), with an absolute range [0, 2.9] mmol/L.

All large target errors were attained at low glucose levels and are attributable to sudden changes in patient physiology, rather than systemic model deficiencies. Target errors are consistent with and explainable by the published sensor error distributions. End glucose levels were 40% lower than their starting values. The results show that intensive glucose management insulin therapy and nutrition control not only reduced absolute glucose levels, but also the severity of fluctuation in glucose values even with significant inter-patient variability and time-varying physiological condition.

Trials spanning longer periods of time are in development to verify the long-term trial simulations performed and to test the adaptability of the controller. Clinically, these results indicate potential in clinical use to reduce ICU mortality as well as reduce risk of severe complications.