Molecular Genetics of Migraine

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<th>IHBI Research Themes</th>
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<td>Health Determinants and Health Systems</td>
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<tr>
<td>Injury Prevention and Trauma Management</td>
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<td>Chronic Disease and Ageing</td>
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*Prevention  Intervention  Translation*

>1200 researchers
Research Focus

- Cancer research
  - Prostate, breast, ovarian, skin and lymphoma
- Cardiovascular disease and diabetes
- Mental health and neurological research
- Dementia and Ageing research
- Vision research
Migraine

• Migraine is a frequent, debilitating and painful disorder that affects a large proportion of the population

• No laboratory based diagnostics and current treatments exhibit variable efficacy

• Migraine affects
  – 4% of children,
  – 6% of men, and
  – 18% of women

Incidence of Migraine

Stewart et al. Am J Epidemiol (1993);34:1111
Migraine Prevalence Estimates

1-Year Prevalence (%)

- Males (~7.5%)
- Female (~19%)
- Overall (~12%)

Migraine Classification

- Recurrent attacks of headpain widely variable in intensity, frequency and duration
- Nausea, vomiting, photophobia & phonophobia

Migraine without Aura
- Recurrent headaches, 4-72 hours duration
- Usually unilateral and pulsating

Migraine with Aura
- Recurrent headaches, 4 - 72 hours duration
- Preceded or associated with neurological symptoms such as visual disorders, unilateral numbness, weakness, speech defects
Migraine Comorbidity

Migraineurs ~5 times more likely to be diagnosed with comorbid condition than non-migraineurs – shared genetic factors?
Migraine Genes

• shows strong familial aggregation

• RR (1.5-3.8), Twin heritability 40-65%

• complex disorder, environmental triggers

• MA and MO within same families and same individual

• Co-morbidity with several disorders eg epilepsy, CVD and depression – indicating potential shared genetic factors

• Number of genes unknown at present
Case/Control Samples

Population

Case Sample  Control Sample

Affected – Non-Affected
(Age, sex and ethnicity matched)
Migraine Family
Study Populations

• Cross-Sectional - case versus control
  • Affected 275 - Unaffected 275: ii) 300-300  iii) 500+/-
  • Age, sex and ethnicity matched
  • History, at least one affected relative
  • 68% female, 63% MA

• Pedigrees - family linkage approach
  • >100 pedigrees, 15 with > 8 affected ascertained
  • av 12 affected, range 8-21; 12-55 total individuals
  • 67% female, 68% MA

Diagnosis using IHS Criteria
Mutiny on the HMS Bounty
Pacific 1789

Mutiny on the Bounty
Clark Gable
Charles Laughton
Clark Gable

The Bounty
Mel Gibson
Anthony Hopkins

Petcairn Islands
Bounty
$3.50
Norfolk Island Project

• Geographically remote, isolated population with known founder effect and well-defined family groupings

• Strong family groupings and well documented family histories

• 12 generational pedigree involving ~6300 Individuals- going back to 1780’s

• Many of the current population can trace ancestry back to Pitcairn Island

• Limited number of original founders
  – 12 Maternal (Tahitian)
  – 9 Paternal (Bounty mutineers)
The Norfolk Island Pedigree

• Core Pedigree Reconstruction (n=2596)

~500 individuals have DNA and phenotype data - CVD risk traits, ocular phenotypes and migraine
## Migraine Demographics

<table>
<thead>
<tr>
<th>Sample</th>
<th>Characteristics</th>
<th>Migraine</th>
<th>Unaffected</th>
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<tbody>
<tr>
<td><strong>Entire Cohort</strong></td>
<td>Total</td>
<td>154</td>
<td>446</td>
</tr>
<tr>
<td></td>
<td>Female (%)</td>
<td>73.4</td>
<td>50.7</td>
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<tr>
<td></td>
<td>Mean age in years (SD)</td>
<td>49.0(16.3)</td>
<td>51.5(16.4)</td>
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<tr>
<td><strong>Pedigree</strong></td>
<td>Total</td>
<td>96</td>
<td>281</td>
</tr>
<tr>
<td></td>
<td>Female (%)</td>
<td>74.0</td>
<td>48.0</td>
</tr>
<tr>
<td></td>
<td>Mean age in years (SD)</td>
<td>46.4(16.5)</td>
<td>50.2(16.6)</td>
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**Migraine heritability analyses**

- 25.5% prevalence of migraine, Heritability of 0.53 (P=0.016)
**Serotonin Genes**
- Transporter (SERT)
- Receptor Genes (5HT2C, 5HT2A, 5HT1A)
- MAO-A, MAOB

**Dopamine Genes**
- Dopamine Beta-Hydroxylase (DBH)
- Dopamine Receptor Genes D 1,2,3,5

**19p13 Genes**
- CACNA1A
- INSR
- LDLR
- Notch3
- PRKCSH

**Nitric Oxide Synthases**
- inducible (iNOS)
- endogenous (eNOS)
- neuronal (nNOS)

**Vascular and Hormonal**
- MTHFR, ACE
- ESR, PgR, AR

**Ion Channel Genes**
- Calcium Channel Genes (CACNA1A, CACNA1S, CACNA1D)
- hKCa3 (Potassium Channel)
Migraine and Hormone Receptor Genes

- Tested variants in both the ESR and PR genes show replicated association with migraine

- Interaction analysis showed that those who possessed both the ESR and PR risk alleles were at increased risk

- Hence variants in these hormone receptor genes appear to play synergistic roles in migraine

- Implications to hormonal treatments and factors that affect hormonal levels in some migraineurs

- Commenced menstrual migraine genetics study with Dr Anne MacGregor and the London Migraine Clinic to determine the role of hormone receptor gene variants in this sub-type of migraine
MTHFR Gene

MTHFR gene

enzyme

5-10 MTH Folate

5-MTH Folate

homocysteine

methionine

circulatory folate

enzyme activity

folate levels

homocysteine levels

Vascular injury and dysfunction

C677T

migraine risk (MA 2.5 X)

stroke risk (1.5-2 times)

CVD risk
MTHFR C677T variant

- MTHFR TT genotype increased MA risk in Japanese (6-fold) (Kowa et al, 2001) (n=74 migraineurs, 22 with MA)

- MTHFR TT genotype increased MA risk in Turkish (10-fold) (Kara et al, 2003) (n=102 migraineurs, 23 with MA)

- MTHFR 677TT genotype increased MA risk (2.5 fold) in Australian cohort (n=268 migraineurs, 168 MA Lea et al, 2004)
  - also A1298C MTHFR polymorphism, gave 2.5 fold increased risk of MA (P= 0.015)

- Meta-analysis by Rubino et al, 2007, of 2961 migraineurs supports association of the TT C677T genotype with MA
Migraine Genes & Risk Variants

Genome-wide association study of migraine implicates a common susceptibility variant on 8q22.1

Verneri Anttila1,2,*, Hreinn Stefansson1, Mikko Kallela1, Unda Tödt1,6, Gisela M Terwindt7, M Stella Calafato1,8, Dale R Nyholt9, Antigone S Dimas1,10,11, Tobias Freilinger12,13, Bertram Müller-Myhsok14, Ville Artto4, Michael Inouye1,15, Kirsi Alakurtti1,2, Mari A Kaunisto1,16, Eija Hamalainen1,12, Bouke de Vries17, Anine H Stam7, Claudia M Weller15, Axel Heinze17, Katja Heinze-Kuhn17, Ingrid Goebel5,6, Guntram Borck5,6, Hartmut Göbel17, Stacy Steinberg1, Christiane Wolf14, Asgeir Björnsson3, Gretar Gudmundsson18, Malene Kirchmann19, Anne Hauge19, Thomas Werge20, Jean Schoenen21, Johan G Eriksson16,22,24, Knut Hagen25, Lars Stovner25, H-Erich Wichmann26,28, Thomas Meitinger29,30, Michael Alexander31,32, Susanne Moebus33, Stefan Schreiber34,35, Yurii S Aulchenko36, Monique M B Breteler36, Andre G Uitterlinden37, Albert Hofman36, Cornelia M van Duijn36, Paivi Tikka-Kleemola38, Salli Vespalainen3, Susanne Lucae14, Federica Tozzi19, Pierandrea Muglia39,40, Jeffrey Barrett1, Jaakko Kaprio24,24, Markus Färkkilä1, Leena Peltonen1,2,42,48, Kari Stefansson3, John-Anker Zwart25,43, Michel D Ferrari1, Jes Olesen19, Mark Daly12, Majia Wessman2,16, Arn M J M van den Maagdenberg7,5, Martin Dichgans12,13, Christiane Rune R Frants15 & Aarno Palotie12,42,46,47 for the International Headache Genetics Consortium.

A dominant-negative mutation in the TRESK potassium channel is linked to familial migraine with aura

Ronald G Lafrenière1,2,3, M Zameel Cader2,3,4,13, Jean-François Poulin2, Isabelle Andres-Enguix5, Maryse Simonneau2, Namrata Gupta2, Karine Boisver2, François Lafrenière2, Shannon McLaughlan2, Marie-Pierre Dubé8, Martin M Marcinikiewicz7, SreeRam Ramagopal9, Olaf Ansorge9, Bernard Brais1, Jorge Sequeiros18, Jose Maria Pereira-Monteiro11, Lyn R Griffiths12, Stephen J Tucker3, George Ebers8 & Guy A Rouleau1,2

Migraine with aura is a common, debilitating, recurrent headache disorder associated with transient and reversible focal neurological symptoms. A role has been suggested for the two-pore domain (K2P) potassium channel, TWIK-related spinal cord potassium channel (TRESK), encoded by KCNK18, in pain pathways and general anaesthesia. We therefore examined whether TRESK is involved in migraine by screening the KCNK18 gene in subjects diagnosed with migraine. Here we report a frameshift mutation, F139WfsX24, which segregates of the trigeminal ganglia afferents and may lead to central sensitization. Considerable insights into the pathogenesis of migraine have come from the investigation of the rare autosomal dominant subtype of migraine with aura, familial hemiplegic migraine. Three susceptibility genes (CACNA1A, ATP1A2 and SCN1A), which encode either ion channels or ion transport proteins, have so far been identified, and it is likely that mutations in these genes reduce the threshold for CSF5.10 However, such mutations are not found in typical migraine with aura, suggesting that other ion channels or transport proteins may be involved.
**Figure.** Genomic regions reporting significant evidence of linkage to migraine phenotypes (red) and FHM (blue).
FHM Implicated Loci

• FHM- severe, rare autosomal dominant form of migraine

• FHM1 mapped to C19 with mutations in a PQ calcium channel gene implicated - *Ophoff et al 1996, Cell*

• FHM2 mapped to 1q23 with mutations in an ATPase gene implicated - *De Fusco et al 2003, Nature Genetics*

• FHM3 mapped to 2q24, voltage gated sodium channel gene, SCN1A - *Dichigans et al 2005, Lancet*

• NATA accredited diagnostic testing since 1999
FHM Types 1, 2 and 3

- Familial Hemiplegic Migraine Types 1, 2 and 3

- FHM is considered a subtype of migraine with aura

- In addition to aura (sensory disturbances) FHM manifests with several more severe symptoms

  - These include hemiparesis (one sided paralysis), deafness, nystagmus (involuntary eye movement), retinal degeneration and coma.

- Linked to mutations in CACNA1A, ATP1A2 and SCN1A
Migraine Genetic Studies

- therapeutics
MTHFR Gene

C677T

- ↓ enzyme activity
- ↓ folate levels
- ↑ homocysteine levels
- ↑ migraine risk (MA 2.5 X)
- ↑ stroke risk (1.5-2 times)
- ↑ CVD risk

Vascular injury and dysfunction

5-10 MTH Folate

5-MTH Folate

homocysteine

methionine

circulatory folate

enzyme

MTHFR gene

MTHFR enzyme
Migraine Pharmacogenetic Trial

• MTHFR C677T mutation is associated with enzyme function

  – TT genotype exhibits ~50% reduction in enzyme activity and results in higher homocysteine levels

- and is associated with increased risk of MA

• Plasma homocysteine levels can be lowered by vitamin supplementation with folic acid, vitamin B12 and vitamin B6

• Double blind, placebo controlled trial of 52 MA patients over 6 month period, seen at baseline, 3 mths and 6 mths

• Vitamin tablets (containing 2mg of folic acid, 25mg vitamin B6 and 400μg of Vitamin B12) or placebo - one tablet daily for 6 months
Change in plasma folate and vitamin B12 concentration in vitamin and placebo groups

A. Change in plasma folate

B. Change in plasma vitamin B\textsubscript{12}

Values are mean ± SEM.
Change in plasma B6 and homocysteine concentration in vitamin and placebo groups

C. Change in plasma vitamin B6

D. Change in plasma homocysteine

Values are mean ± SEM.
Change in secondary clinical outcomes ie. 6-month migraine frequency (A) and average pain score (B) over the treatment period for vitamin and placebo groups. Values are medians. Quartiles not shown but reductions are statistically significant (P<0.05).
Change in primary clinical outcomes ie. frequency of high level migraine disability (MIDAS >11) over the treatment period in vitamin and placebo groups

- frequency of disease disability decreased 2-fold following 6 months of daily supplementation (61% to 30%, P= 0.01)
- reduction in placebo group not statistically significant (53% to 46%, P=0.3).
Migraine Clinical Trial

- Disease disability decreased 2-fold following 6 months of daily supplementation (61% to 30%, P=0.01)

- Decrease in headache frequency, from 4 attacks to 1 per month (P=0.04),

- Decrease in pain severity, from an average score of 6 to 4.5 (P=0.002)
  - placebo group, no change either headache variable (P>0.1)

- Effect of vitamin therapy on migraine disability was dependent on MTHFR C677T genotype - reduction in disability occurring predominantly in patients carrying the CC/CT genotype (P=0.002).

- Results suggest that MTHFR genotype plays a role in migraine response to vitamin supplementation, with a significant reduction in migraine frequency, severity and disability

Lea et al 2009 *Pharmacogenetics and Genomics*
Recent RCT of Vitamin Supplementation

• 245 MA patients, female Caucasians

• No statistically significant differences between the vitamin and placebo groups for the test variables at baseline.

• 206 patients completed- treatment well tolerated, no adverse reactions

• At 6 mths, folate, B6 and B12 marked increases, compared to baseline and placebo (P<0.001), homocysteine marked decrease compared to placebo
Change in primary clinical outcome - frequency of high-level migraine disability (Migraine Assessment Score > 11) over the treatment period in vitamin and placebo groups.

Menon et al 2012  *Pharmacogenetics and Genomics*
Trial Conclusions

- Promising evidence that homocysteine-lowering via vitamin B/folate supplementation reduces migraine disability in a subgroup of migraineurs.

- Vitamin supplementation appears to be a safe, inexpensive and effective preventative treatment for increasing the quality of life of migraineurs.

- There is also a need to determine whether such treatment and treatment dosage should be based on MTHFR genotype.

- There is thus some evidence that vitamin supplementation can affect migraine disability.

- The identification of the genes that play a role in migraine may lead to other novel migraine treatments.

Menon et al 2012  *Pharmacogenetics and Genomics*
Conclusions

- DNA studies can aid in defining genes involved in disease and this can have important diagnostic and treatment implications.

- DNA diagnostics - effective way to differentiate the genes involved in familial hemiplegic migraine, episodic ataxia, hereditary stroke and pediatric epilepsy - and hence define personalised treatments.

- Not all genes for these disorders have yet been identified and we are undertaking studies to identify new migraine genes.

- Concussion genetics study – to investigate ion channel genes in relation to susceptibility.

- Also undertaking trials to develop new treatments for migraine.
MIGRAINE HEADACHES

• Are you 18-65 years old?
• Do you have more than 1 migraine headache a month?
• Would you like to prevent another migraine?

Would you like to participate in a medical research study?

Contact Emily on 1800 397 371
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• Saras Menon, Sharon Quinlan
  - clinical collections, trials, phenotype analyses

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