Genetic meta-analysis and Mendelian randomisation

Investigating the association of fibrinogen with cardiovascular disease

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• “Classes make children sick”
• “Breast milk curbs asthma”
• “Fighting spirit not enough to beat cancer”

• “Your hormones can stop you going gaga as you get older, say researchers”
• “Frisky men and women who engage in risky sex could be suffering psychiatric problems…”
• “One footie in the grave – thrilling matches can kill, say docs”

Bartlett, Sterne and Egger, BMJ 2002
Beta-carotene and cardiovascular mortality

Cohorts
- Male health workers, USA
- Social insurance, men, Finland
- Social insurance, women, Finland
- Male chemical workers, Switzerland
- Hyperlipidaemic men, USA
- Nursing home residents, USA

Trials
- Male smokers, Finland
- Skin cancer patients, USA
- (Ex)-smokers, asbestos workers, USA
- Male physicians, USA

Relative risk (95% CI)

Egger et al *BMJ* 1998
Can genetic epidemiology help?

Mendelian randomisation to the rescue?
Mendelian randomisation

In a genetic association study the laws of Mendelian genetics imply that comparison of groups of individuals defined by genotype should only differ with respect to the locus under study (and closely related loci in linkage disequilibrium with the locus under study).

With control for population structure there should be little residual bias, or confounding by any behavioural, socioeconomic or physiological factors excepting those influenced by alleles at closely proximate loci.

Fibrinogen and CVD

• Fibrinogen is a well-established risk indicator for cardiovascular disease

• It is increased by smoking (and hence may mediate smoking-CHD associations or be confounded by smoking)

• Fibrinogen is an acute phase reactant (and hence an indicator of propensity to illness) and also may be increased by the presence of atherosclerosis
A meta-analyses of prospective observational studies

Mendelian randomisation?

- Several polymorphisms are associated with elevated fibrinogen
- A number of studies suggest that these polymorphisms are NOT related to CHD risk
- However – genetic association studies tend to be of low power and may be susceptible to reporting biases
  - What is the combined evidence?
Mendelian randomisation, fibrinogen and CVD

Genotype (β-fibrinogen)

1

Intermediate phenotype (plasma fibrinogen)

2

Disease (CVD risk)

3
Methods

• MEDLINE and EMBASE search

• Eligible studies were case-control or prospective cohort studies relating fibrinogen polymorphisms (ascertained via genotyping) to cardiovascular disease
  – Family studies excluded
Information extracted

• Associations between:
  – genotype and CVD
  – genotype and fibrinogen
  – Fibrinogen and CVD

• Details of study design
  – case definition, exclusion criteria, restriction to patients with a particular disease, control selection, confounding variables, blinding of those genotyping to participants’ disease status, geographic location, characteristics (e.g. gender, age, etc.) of cases and controls, and whether subgroup analyses had been performed, tests for Hardy-Weinberg equilibrium
Models for genetic inheritance

• Dominant inheritance model
  – Either one or two A alleles produces an increase in fibrinogen – compare GG with GA/AA

• Recessive inheritance model
  – Only two A alleles produces an increase in fibrinogen
    – compare GG/GA with AA

• Codominant inheritance model
  – Fibrinogen level: AA > GA > GG – use logistic regression to estimate the increase in the log odds per A allele
1. G – IP association (in controls)

- 10 studies give useable data on plasma fibrinogen level by genotype in controls.
- However 4 small studies combine GA and AA.
- Shall present GA and AA separately, so only 6 studies remain, with 8871 controls in total.
1. G – IP association in controls – Summary

- Pooled estimates of mean difference are:
  - GA - GG: 0.11 (0.08, 0.14) g/l
  - AA - GG: 0.22 (0.14, 0.30) g/l
- Good evidence that the gene is codominant.
- Suggests a per-allele model for the effect of gene on disease is reasonable.
- Using this gives a per-allele difference of 0.111 (0.084, 0.138) g/l.
2. G – D association

• 18 studies provide data on G-D
• However, one small study (van der Bom 1998) presents data for GA+AA combined only.
• Leaves 17 studies:
  10918 cases, 20252 controls.
2. G–D association: per-allele model

OR per A allele, random-effects model:

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<table>
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<td>All studies</td>
<td>0.97 (0.90, 1.05)</td>
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<td>1.00 (0.95, 1.04)</td>
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3. IP – D association: observed

- Meta-analysis of OR for a 1g/l increase in plasma fibrinogen.
- Taken from logistic regression where available in paper, else converted from mean difference in plasma fibrinogen between cases and controls, assuming normal distributions with common SD
  - Useful references:
    G. Chêne and S.G. Thompson (*American Journal of Epidemiology* 1996; 144: 610-621)

- Assuming a linear-logistic relationship between odds of disease and fibrinogen level:
  \[ \log \text{OR}_{PD} \approx \log \text{OR}_{GD} / \text{MD}_{GP} \]

where, for the per-allele model:
- \( \log \text{OR}_{GD} \) is the log-odds ratio per allele
- \( \text{MD}_{GP} \) is the difference in mean plasma fibrinogen per allele
- \( \log \text{OR}_{PD} \) is the estimated log-odds ratio per unit increase in fibrinogen.

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<th>Studies in G-D estimate</th>
<th>G-D OR per allele</th>
<th>OR for a 1 g/l increase</th>
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<td>All studies</td>
<td>0.97 (0.90, 1.05)</td>
<td>0.79 (0.38, 1.58)</td>
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<td>Excluding Yu (1996)</td>
<td>0.99 (0.94, 1.06)</td>
<td>0.95 (0.54, 1.67)</td>
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<tr>
<td>Excluding Ma (1999)</td>
<td>0.97 (0.90, 1.04)</td>
<td>0.75 (0.39, 1.39)</td>
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Summary

• Mendelian Randomisation can be used to look at the causal effect of fibrinogen on CHD.
• Suggests any causal effect is smaller than from observational evidence.
• However a small effect cannot be ruled out.