Progress Report 2012 - 14
NIAA Anaesthesia/AAGBI small project award (A.P.Morley)

Molecular mechanisms of action of propofol in clinical use: a gene association study

Clinical data collection and DNA sampling:

Clinical data collection and DNA sampling was completed in July 2009. 515 Caucasian patients were enrolled – less than the 550 stated on the grant application, which included a 50 patient genotyping failure contingency. The laboratory conducting DNA extraction and genotyping deemed 515 sufficiently in excess of the 500 subjects necessary for effective statistical analysis, genotyping failure rates being only 1-2%. A further 254 non-Caucasian patients were recruited over the data collection period.

DNA extraction and genotyping:

Because of the large study size, nearly four years elapsed from the initial literature search on the molecular basis of propofol’s effects, and the genotyping. This being the case, it was necessary to revise completely the selection of CNS receptor gene polymorphisms to take account of new literature relating to gene association studies in general, and our selected genes in particular.

Hypnotic polymorphisms: Having reviewed the evidence, we restricted our genotyping choice to four genes for central nervous system receptors, rather than the seven genes originally proposed. These four were GLRA1 (glycine receptor alpha 1 subunit), GRIN2A (N-methyl D-aspartate receptor 2A subunit 2) and GABRB2 and GABRB3 (gamma-aminobutyric acid A β2 and β3 receptor subunits, respectively). These receptor subunits were deemed of potential relevance to propofol’s hypnotic effect, largely on the basis of knock-out/knock-in mutation studies in mice.

We elected to genotype two tagging sets of single nucleotide polymorphisms (SNPs) to cover common variation in two genetic regions. Using HapMap (www.hapmap.org), the first set of 15 SNPs was generated for the smallest of the genes, GLRA1, and its 10 kb margins. A second set of 6 SNPs was generated for a 21 kb region including the 5’ end of the GABRB3 gene. This region has been previously shown to be associated with transcriptional effects. We chose seven further polymorphisms across the four selected genes, on the basis of predicted functional relevance or clinical association, making a total of 28 hypnotic polymorphisms in all.

Hypotensive polymorphisms

Our choice of polymorphisms for genotyping with respect to propofol's hypotensive effect remained unchanged from our grant application.
Results

Maximum percentage decrease in mean arterial blood pressure from baseline in the first ten minutes of propofol infusion. No statistically significant association was detected between this endpoint and genotype at the following five polymorphisms (genes) - rs4961 (α adducin, ADD1); rs699 and rs5051 (angiotensin, AGT); rs5186 (angiotensin II receptor type1, AGTR1), In/Del (ACE). A manuscript containing detailed results, and those relating to the other five cardiovascular polymorphisms, is in preparation.

Propofol dose requirement for loss of verbal response. No statistically significant association was detected between this endpoint and any of the SNPs tested in the GLRA1 gene. A manuscript containing detailed results, and those relating to the hypnotic polymorphisms we tested in other genes, is in preparation.

Finance: The full sum of the grant, £14,820, has now been claimed to fund the genotyping.

Update July 2014: The experiments and analyses reported in the papers and abstracts above have now been written up as a doctoral thesis (Doctor of Medicine, University of Cambridge) which will be submitted in the next few months. The title is: 'Genetic susceptibility to the effects of the intravenous anaesthetic, propofol.'

Publications from this project: (a summary sentence appears after each reference)


