SELECTIVE GENOTYPING FOR DETERMINATION OF A MAJOR GENE ASSOCIATED WITH CRANIAL CRUCIATE LIGAMENT DISEASE IN THE NEWFOUNDLAND DOG

P.E. Macrossan[1], B.P. Kinghorn[1,2], V.L. Wilke[3] & M.F. Rothschild[3]
[1]Department of Animal Science, University of New England, Armidale
[2]Sygen Chair of Genetic Information Systems
[3]Department of Animal Science and Center for Integrated Animal Genomics, Iowa State University

SUMMARY
Cranial cruciate ligament disease (CCLD) is the leading cause of lameness in the dog. The objective of this study was to determine the most informative subset of animals on which to carry out selective genotyping for the identification of any major gene affecting CCLD. Two groups of animals were chosen for genotyping from candidate groups of animals having high probabilities (calculated from segregation analysis) for carrying zero and two copies, respectively, of the putative causative allele. A stochastic search algorithm was employed to search the solution space for the 'best' two groups of individuals for genotyping based on two alternating fitness functions. The first fitness function was designed to minimize the genetic relationship amongst dogs within groups for each group individually. The second fitness function was designed to maximize the genetic relationship amongst dogs between groups. This approach, and the ramifications of its use, are discussed.
Keywords: Cranial cruciate ligament disease (CCLD), selective genotyping, segregation analysis.

INTRODUCTION
Cranial cruciate ligament disease (CCLD) is the leading cause of lameness in the dog (Johnson *et al.* 1994). Particular breeds of dogs are predisposed to CCLD, leading to the hypothesis that the disease has a heritable component. Experience has shown that significant morphological defects having all-or-none expression, such as CCLD, are often associated with single loci of major effect (termed Quantitative Trait Loci or QTL) (Falconer and Mackay 1996). Since whole genome scanning is an expensive procedure, the objective of this study was to determine the most informative subset of animals on which to carry out genotyping to identify the major gene responsible for CCLD.

Selective genotyping or DNA pooling has been used to choose individuals for genotyping on the basis of phenotype, or some other criteria of merit (Lander and Botstein 1989; Darvasi and Soller 1992). It can be used to reduce costs in QTL detection, by ensuring strong contrast in merit among individuals with genetic marker information. Selective genotyping can be seen as a special case of the approach to group genotyping formulated in the simulation study of Macrossan and Kinghorn (2005). In the work presented here, pre-selection on probabilities of carrying opposing disease genotypes was first carried out. Final selection was made in a manner that:

1. Reduced the relationships among animals within each selected group, to increase the effective number of dogs within each group, and

2. Increased the relationship between animals in different groups, to reduce the confounding effect of polygenes in the between-group contrast.