MOLECULAR MAPPING OF QUANTITATIVE GENES

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SUMMARY

Genetic maps based on DNA-level markers open the way for comprehensive mapping of quantitative trait loci in livestock. Marker-associated quantitative effects have been detected in a number of experiments and improved statistical designs and analyses are available. Marker-assisted-selection can contribute to within population improvement and to exploitation of between population genetic variation. Extensive synteny of livestock, mouse and human chromosomes will enable a flow of information between the human, mouse and livestock gene-mapping programs. "Microsatellites", a new class of DNA-level polymorphism, may well be the marker of choice for developing saturated gene maps of livestock species.

Methodologies for developing genetic maps based on DNA-level polymorphisms have been enthusiastically embraced by human geneticsts and plant breeders. As a result, large numbers of DNA-level polymorphisms (mostly RFLPs) have been mapped in man (Donis-Keller et al., 1987) and in many species of agricultural plants (Gebhardt et al., 1989; Havey et al., 1989; Helentjaris et al., 1986; Landry et al., 1987; McCouch et al., 1988). Progress in developing DNA-level maps for agricultural animals has been much slower (Fries et al., 1989). The most important reason for this relates to the perceived potential for application of genetic maps. In humans, the potential for using marker genes to track genetic diseases was immediately evident (Botstein et al., 1980). This was followed by map-based reverse genetics for cloning disease genes (Kerem et al., 1989; Rommens et al., 1989; Suthers et al., 1989). In plant breeding, since many domesticated plants are selfers, outside sources of genetic variation are often utilized for genetic improvement; but existing means for identifying and exploiting such genetic variation are limited (Soller and Beckmann, 1988). It was quickly realized that DNA-level genetic markers provided a powerful tool for exploring and manipulating genetic variation in resource populations (Beckmann and Soller, 1986; Soller and Beckmann 1983; Soller and Beckmann 1988).

In contrast, domestic animals are outcrossers, so that genetic variation for most traits of interest is already present within breeding populations. Animal breeders have therefore concentrated on developing methods for efficient utilization of within-population genetic variation. In particular, development of family selection schemes has enabled phenotype-based selection to be extended to sex-limited and carcass quality traits that are not amenable to mass selection. Areas of genetic improvement which could not be tackled by classical biometrical methods, such as improvement of crossbred populations, or utilization of genetic variation in resource populations have been for large part ignored. Thus, generation of genetic maps in livestock populations was not driven by an immediate realized pressing need for marker-assisted methods.

Nevertheles, in the past few years a number of studies have been carried out in animal populations, successfully uncovering marker-associated quantitative effects (Beever et al., 1990; Geldermann et al., 1985; Gonyon et al., 1989; Haenlein et al., 1987; Jung et al., 1989). Some of these studies are reviewed in this workship. For the most part these have involved classical blood group and protein polymorphisms, but experiments involving DNA-level polymorphisms have also been carried out (Jung et al., 1989).

Awareness of DNA-level markers has stimulated theoretical studies on marker-QTL mapping (Lander and Botstein, 1989; Lebowitz et al., 1987; Soller and Beckmann, 1990; Weller, 1986; Weller, 1987). Effective designs are now available for marker-QTL mapping within a single segregating population (Soller and Genizi, 1978; Weller et al., 1990), and for marker-QTL mapping in crosses between populations that differ wtih respect to a quantitative trait, but share marker locus polymorphisms (Beckmann and Soller 1988). Some advances in this area will also be reviewed in this workshop.

Theoretical studies of marker-assisted selection have pointed to marker-assisted-selection of young sires as a promising application for MAS in dairy cattle (Kashi et al., 1990a; Smith and Simpson, 1986; Soller, 1978; Soller and Beckmann, 1982, 1983). There is also a developing interest in marker analyses of exotic populations for novel genes not present in improved breeds. These include disease resistances, particularly the trypanotolerance of the N'Dama cattle of West Africa (Soller and Beckmann, 1987) and tick resistance in Zebu cattle from India. Marker-mapping can also find useful application in tracking single genes of economic importance, such as the double muscling gene in cattle, and the high fertility Booroola gene in sheep. Studies along these lines using DNA fingerprint techniques will also be reported here.

Theoretical studies of marker-QTL mapping and MAS, stress the importance of polyallelic markers (Beckmann and Soller, 1988; Kashi et al., 1990a). Considerable progress has been made toward a saturated map of the bovine genome based on such markers, as will also be reported in this workshop.

A major change in the perspective of mapping livestock genomes has come with the crystallization of the human genome mapping project. It may be possible to transfer human genome information to livestock genomes. Conversely, information gathered from mapping of livestock genomes can be used to further the structural and functional understanding of the human genome. Cross flow of information between human, mouse and livestock genomes will be greatly aided by studies of comparative (syntenic) mapping data in bovine, mouse and man carried out by J.E. Womack and his research group at Texas A&M University (Womack and Moll, 1986). To quote from a recent talk by Prof. Womack (Womack, 1990).

"Of the 160 genes on the current bovine syntenic map, 141 are homologous to mapped human genes. One hundred twenty-four are mapped in both humans and mice. Alignment of these 124 bovine and murine genes with their respective homologues on the human map demonstrates extensive genomic conservation. Thirty-two rearrangements are necessary to account for differences in this abbreviated human and mouse comparative map, while only 21 rearrangements account for the human and cow differences. Nadeau and Taylor (1984) estimated that as few as 150-200 rearrangements distinguish the entirety of the genomes of mouse and man. Our bovine syntenic mapping demonstrates approximately 50% more conservation between human and cattle than between humans and mice. Consequently, as few as 100 chromosomal rearrangements may differentiate the bovine and human genomes....With reasonable continued effort in physical mapping of bovine genes, the boundaries of evolutionary conservation will be defined."

Synteny of the bovine, mouse and human genomes means that as genes affecting quantitative traits are mapped to specific bovine chromosomal regions, it will be possible to turn to the human map for candidate genes having the identified effects. The human genes can then be used as heterologous probes to clone the corresponding bovine genes. These, in turn, could be used to screen for direct or linked quantitative effects of polymorphisms or haplotypes at the putative bovine genes on the quantitative trait of interest. Conversely, identification of specific bovine genes having quantitative effects, will contribute to assigning functions to the homologous human genes, and in this way expand functional understanding of the human gene map.

The past year has also seen a major advance in DNA-level marker methodologies. It was found that the the eukaryote genome is densely interspersed with simple tandemly-repeated motifs, termed "microsatellites. Remarkably, these appear to exhibit site-specific length variation, similar to that shown by minisatellite sequences (Nakamura et al., 1987). Poly(TG) is by far the most frequent microsatellite, appearing as $5\text{-}10 \times 10^4$ individual islets in the mammalian genome (Hamada et al., 1982) but many other microsatellites have been reported (Kirschoff, 1988; Tautz, 1989; Tautz and Renz, 1984; Zischler et al., 1989; Vergnaud, 1989).

Specific microsatellite islets contained within a stretch of sequenced unique DNA, can be individually amplified by means of the polymerase chain reaction (PCR, Saiki et al. 1988; Vosberg, 1989; White et al., 1989), using a pair of flanking unique oligonucleotides. When amplified sequences from a series of individuals have been examined, the microsatellite motifs are almost invariably found to exhibit highly polymorphic length variation (Litt and Luty, 1989; Smeets et al., 1989; Weber and May, 1989). Thus, each microsatellite islet may represent a highly variable, polyallelic locus of high information content. Following Olson et al., (1989) a microsatellite locus defined in this way will be termed a "sequence tagged microsatellite site" STMS (Beckmann and Soller, 1990). Such STMSs, each defined by its flanking unique oligonucleotides, can thus be used to prepare saturated genetic maps of any eukaryote genome. Screening of a bovine genomic library with a (TG)10 oligonucleotide probe in our laboratory showed that almost one-third of all clones gave hybridization signals (Kashi et al., 1990b). Thus, sequencing of random poly(TG) containing clones should rapidly provide large numbers of microsatellite loci. Even if polymorphic islets are spaced only every 100-300 kb, a complete map for a typical mammal based solely on STMSs could include as many as 10,000 reference points.

An STMS map will consist of a simple published list of oligonucleotide sequences. Utilization of the map will require only ad hoc synthesis of specific oligonucleotides corresponding to the desired STMSs, eliminating storage or shipment of cloned probes. As in the original Olson et al. (1989) proposal, the STMS map can be built up gradually, by the cooperative efforts of many laboratories; each sequenced fragment found to contain a microsatellite motif, would add to the map. As illustrated by Smeets et al. (1989) a microsatellite islet may well be located within short walking distance of any gene of interest. Thus, it should be possible to walk from any single-copy sequence until such a motif is met, and add it to the map. This could serve to build a highly informative STML map based on known genes or other unique sequences that have been physically mapped, but which are themselves monomorphic, or associated with diallelic RFLPs only. In this way physical and genetic maps could be united. Furthermore, because of the minute quantitites of DNA required for the PCR reaction, it may be possible to score individual embryos for microsatellite markers (Li et al., 1989). This would allow MAS to be carried out the embryo level, increasing the effectiveness of MOET schemes. The great degree of polymorphism anticipated for microsatellites will allow reference populations for linkage studies to be chosen that will be genetically most informative with respect to traits of biological or agricultural interest. In this way, as the genetic map is developed, a map of loci affecting these traits will be developed simultaneously. As these considerations show, the multiple advantages of an STMS map seem potentially so great as to make this by far the method of choice in mapping agriculturally important species.

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BIBLIOGRAPHY

BECKMANN JS, SOLLER M, 1986. Oxford Surveys of Plant Molecular and Cell Biology. Oxford Press, Oxford. Volume 3: 196-250.

BECKMANN JS, SOLLER M, 1988. Theor. appl. Genetics 76:228-236.

BECKMANN JS, SOLLER M, 1990. (submitted).

BEEVER JE, GEORGE PD, FERNANDO RL, STORMONT CJ, LEWIN HA, 1990. Animal Science (in press).

BOTSTEIN D, et al. 1980. Am. J. Hum. Genet. 32: 314-331.

DONIS-KELLER et al., 1987. Cell 51:319-337.

FRIES T, et al. 1989. Animal Genetics 20:3-29.

GEBHARDT D, et al. 1989. Theor. appl. Genet. 78:65-75.

GELDERMANN H, et al. 1985. Theor. appl. Genetics 70: 138-146.

GONYON DS, et al. 1987. J. Dairy Sci. 70:2585-2598.

HAENLEIN GFW, et al. 1987. J. Dairy Sci. 70:2599-2609.

HAMADA H, et al. 1982. PNAS (USA) 79:6465-6469.

HAVEY JJ, MUEHLBAUER FJ, 1989. Theor. appl. Genet. 77:395-401.

HELENTJARIS T, et al. 1986. Theor. appl. Genet. 72:7561-769.

JUNG YC, et al. 1989. Animal Genetics 20:79-81.

KASHI Y, HALLERMAN EM, SOLLER M, 1990a. Anim. Prod. (in press).

KASHI Y, IRAQI F, TICKOSCHINSKY Y, RUDINSKY B, NAVE A, BECKMANN, JS, FRIEDMANN A, SOLLER M, GRUENBAUM Y. 1990b. Genomics (in press).

KEREM B, et al. 1989. Genetic analysis. Science 245:1073-1080

KIRSCHOFF C, 1988. Chromosoma (Berl.) 96:107-111.

LANDER ES, BOTSTEIN D, 1989. Genetics 121:185-199.

LANDRY BS, et al. 1987. Genetics 116:331-337.

LEBOWITZ RJ, et al. 1987. Theor. appl. Genet. 73:556-562.

LI H, et al. 1988. Nature 335:414-417.

LITT M, LUTY JA, 1989. Am J. Hum Genetics 44:397-401.

McCOUCH SR, et al. 1988. Theor. appl. Genet. 76:815-829.

NADEAU JH, TAYLOR BA, 1984. PNAS (USA) 81:814-818.

OLSON M, et al. 1989. Science 245:1434-1435.

ROMMENS, JM, et al. 1989. Science 245:1059-1065

SAIKI RK, et al. 1988. Science 239:487-491.

SMEETS HJM, et al. 1989. Human Genet. 83:245.

SMITH C, SIMPSON, SP, 1986. J. Anim. Breeding Genet. 103:205-217.

SOLLER M, 1978. Animal Prod. 27:133-139.

SOLLER M, BECKMANN JS, 1982. Proc. 2nd World Congress Genetics Applied to Livestock Production 6:396-404.

SOLLER M, BECKMANN JS, 1983. Theor. appl. Genet. 67: 25-33.

SOLLER M, BECKMANN, JS. 1987. Toward an Understanding of the Genetic Basis of Trypanotolerance in the N'Dama Cattle of West Africa. Consultation Report submitted to FAO, Rome, March 1987.

SOLLER M, BECKMANN JS, 1988. In: (Eds. BS Weir, EJ Eisen, MM Goodman and G Namkoong) Proc. 2nd Int. Conf. Quant. Genetics. Sinauer Assoc. Inc. Publ. Sunderland, Mass. p:161-188.

SOLLER M, BECKMANN JS, 1990. Theor. appl Genetics (in press).

SOLLER M, GENIZI A, 1978. Biometrics 34:47-55.

SUTHERS GK, et al., 1989. Science 246:1298-1300.

TAUTZ D, RENZ M, 1984. Nucl. Ac. Res. 12: 4127-4138.

TAUTZ D, 1989. Nucl. Ac. Res. 17: 6463

VERGNAUD G, 1989. Nucl. Ac. Research 17:7623-7630.

VOSBERG HP, 1989. Hum. Genet. 83:1-15.

WEBER JL, May PE, 1989. Am. J. Hum. Genetics 44:388-396.

WELLER JI, 1986. Biometrics 42: 627-640.

WELLER JI, 1987. Heredity 59: 413-421.

WELLER JI, KASHI Y, SOLLER M, 1990. J. Dairy Science (in press).

WHITE TJ, et al. 1989. Trends in Genetics 5:185-189.

WOMACK JE, 1990. In: Mapping Agricultural Animals, Banbury Conf. Feb., 1990.

WOMACK JE, MOLL YD, 1986. J. Heredity 77:2-7.

ZISCHLER H, et al. 1989. Nucleic Acids Research 17:4411.