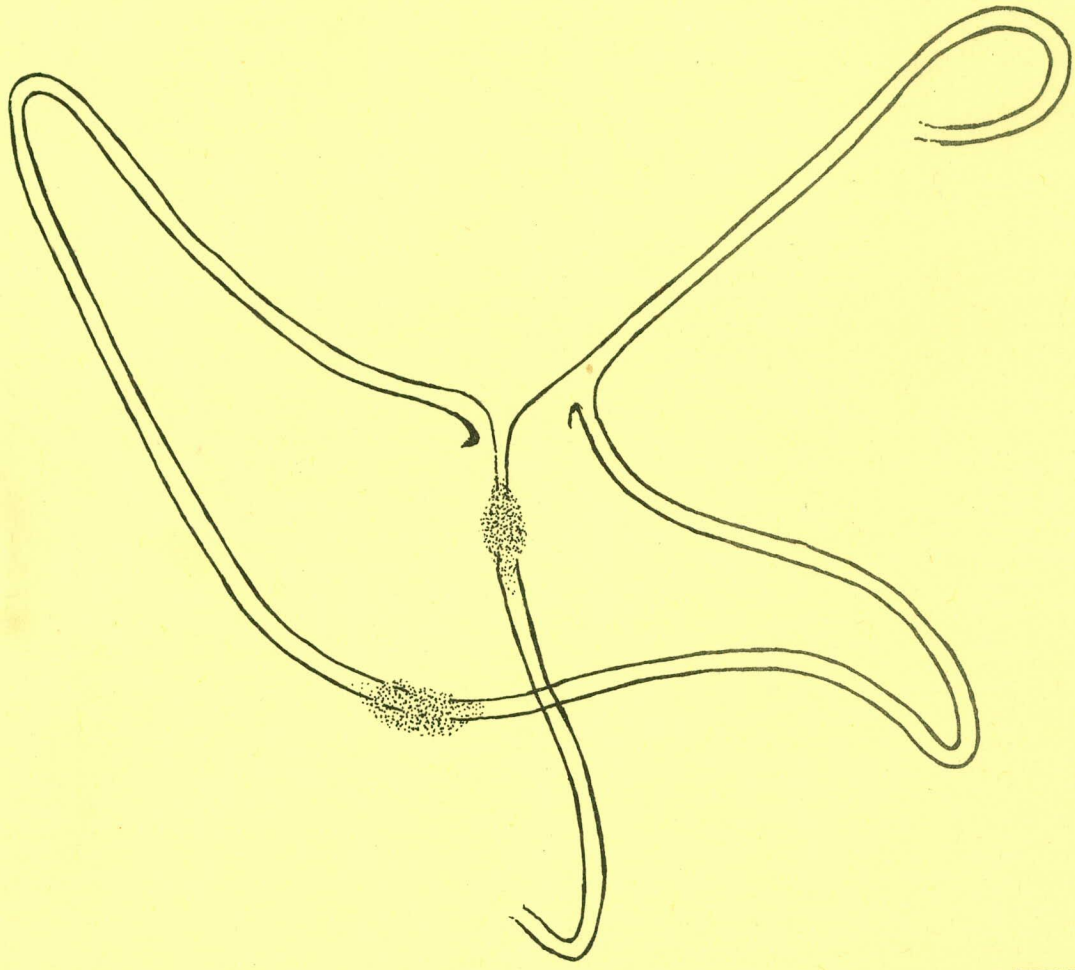
GENETICS SOCIETY OF AUSTRALIA

32nd ANNUAL CONFERENCE AUGUST, 1985



MACQUARIE UNIVERSITY

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COVER:

Above: The Allied Rock Wallaby (*Petrogale assimilis*)

Below: Synaptonemal complex of a hybrid between the nominate *assimilis* race (2n = 20) and the Mareeba race (2n = 18).

Drawings by Betty Thorn.

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PROGRAMME

TUESDAY 27TH AUGUST

7.00-10.00 pm Mixer and Registration in Building W5C, Seminar Room 220.

WEDNESDAY 28TH AUGUST

8.30- 9.00 am Registration in Building W5C, Seminar Room 220.

9.00-10.20 am **Session 1A (P.G. Price Theatre)**
Chairman - Dr. D.L. Hayman

9.00- 9.20 am J.D. Murray, C. Moran, M.P. Boland, C.D. Nancarrow and R.J. Scaramuzzi (CSIRO Animal Production, Prospect)

Chromosome analysis of sheep embryos and fetuses - the incidence of haploid and polyploid cells

9.20- 9.40 am A.L. Lavelle, R.C. Moore and R.F. Martin (Cancer Research Institute, Melbourne)

Radiation-induced chromosome damage in G_2 cells

9.40-10.00 am G.M. Clarke and G.G. Foster (CSIRO Entomology, Canberra)

Male recombination in *Lucilia cuprina*

10.00-10.20 am D. Bedo (CSIRO Entomology, Canberra)

Meiotic pairing of sex chromosomes in *Lucilia cuprina* males

10.20-10.50 am Morning Tea

9.00-10.20 am **Session 1B (Theatre W5A, T2)**
Chairman - Professor R.H. Crozier

9.00- 9.20 am J. Fegent, G. Weller and J.A. McKenzie (University of Melbourne)

Frequency-dependent selection in *Lucilia cuprina*

9.20-9.40 am M.S. Johnson (University of Western Australia)

An electrophoretic analysis of phylogeny and evolutionary rates of *Partula* from the Society Islands

P. Oldroyd and C. Moran (Plant Research Institute, Vic.)

Some effects of homozygosity on honeybee phenotypes

10.00-10.20 P.A. Davies, P.J. Larkin, M.A. Pallota and W.R. Scowcroft (CSIRO Plant Industry, Canberra)

Somaclonal mutants at the ADH-1 loci in wheat

10.20-10.50 am Morning Tea

CHROMOSOME ANALYSIS OF SHEEP EMBRYOS AND FOETUSES - THE INCIDENCE OF HAPLOID AND POLYPLOID CELLS.

J.D. Murray, C. Moran[1], M.P. Boland, C.D. Nancarrow and R.J. Scaramuzzi.

C.S.I.R.O. Division of Animal Production, Prospect, N.S.W. and [1] Department of Animal Husbandry, The University of Sydney.

The presence of chromosomal abnormalities has been correlated with early embryonic mortality in many mammals, including most of those of agricultural importance. Up to 11% of day 1 to 5 sheep and cattle embryos analysed have been chromosomally unbalanced. In addition the techniques of superovulation, which is extensively used in conjunction with embryo transfer, has been reported to increase the frequency of chromosome aberrations in early embryos (King, 1985).

In conjunction with experiments on early embryonic loss in androstenedione-immune Merino ewes we analysed the chromosome constitution of 108 day 1 to 3 embryos, 103 day 13-14 blastocysts and 116 day 24-32 foetuses. In addition the chromosome composition was assessed for 48 day 1 to 5 embryos collected from superovulated Merino ewes.

There were no significant differences between the level of chromosomal abnormalities in early stage embryos collected from untreated (12.8%), androstenedione-immune (11.6%) or superovulated (10.4%) ewes. Variation in the number of chromosome sets was more common than aneuploidy (15 : 3), with half of the euploid abnormalities being attributable to polyspermy. The frequency of chromosomally unbalanced embryos dropped to less than 2% by day 13 and was 0% in the 24-32 day old foetuses.

Polyploid cells, both 4N and 8N, were observed in half of the day 13-14 blastocysts analysed. This probably reflects the normal differentiation of the trophoblast, which is known to be polyploid. However, surprisingly 70% of the foetuses also contained polyploid cells, including 4N, 6N and 8N cells. The foetuses were dissected free of extra-embryonic membranes so these cells represent foetal tissue and presumably are important in differentiation. It should be noted that 6N cells can only be obtained via cell fusion.

We conclude that in Merinos approximately 10% of embryos are chromosomally unbalanced, largely due to polyspermy, and that the incidence of abnormalities is not increased by the superovulatory treatments used. Furthermore, there are polyploid cells in foetal tissue, some of which are derived via cell fusion, which presumably are part of the normal differentiation pathway of those tissues.

King, W.A. (1985). Theriogenelogy. 23: 161-174.

RADIATION-INDUCED CHROMOSOME DAMAGE IN G_2 CELLS

by

A. L. LAVELLE[+], R. C. Moore[*] and R. F. MARTIN[*]

[+]MONASH UNIVERSITY
DEPARTMENT OF GENETICS
CLAYTON, VICTORIA

[*]CANCER INSTITUTE
LITTLE LONSDALE STREET
MELBOURNE, VICTORIA

Abstract

Chromosome damage sustained by cells irradiated during G_2 has been examined in an asychronous population of marsupial (JU56) cells exposed to low dose rate gamma rays; X-rays or the ^{125}I-labelled DNA binding ligand (^{125}I)-iodohoechst. The frequency of different classes of chromosome aberrations seen at division following irradiation, was dependent upon linear energy transfer (LET) and the conditions under which irradiation took place.

Cells were gamma-irradiated over an extended period of 18 hours. Cell cycling during this period was prevented by irradiating at 4°C. Gamma-irradiated G_2 cells sustained more damage than cells X-irradiated with an equivalent acute dose at 37°C. The increase of chromosome aberrations after gamma irradiation appears to be due to the low dose rate and the reduced temperature.

High LET-type radiation like ^{125}I decay induces two-hit type chromosome aberrations more efficiently than low LET X or gamma radiation. It was found that exposing G_2 cells to (^{125}I)-iodohoechst produced more chromosome-type aberrations compared with chromatid-type aberrations than gamma irradiated cells. This result indicated that the replicated chromatids in G_2 cells are very closely apposed.

The amino acid cysteine is known to be a radioprotector, and therefore would be expected to reduce chromosome damage after irradiation. Cells exposed to X-rays at 37°C show reduced chromosome damage when cysteine was present at the time of irradiation. However, when X-irradiation took place at 4°C, chromosome damage was not reduced in the presence of cysteine. In addition, split dose experiments demonstrated that repair of radiation-induced lesions were inhibited when cells were irradiated at 4°C.

This study has revealed striking differences in the degree and type of chromosome damage after irradiation under different conditions. These differences can be explained in terms of dose rate, temperature and repair.

Male Recombination in Lucilia cuprina.

G.M. Clarke and G.G. Foster. CSIRO Division of Entomology.

Recombination in Lucilia cuprina has been previously thought to be restricted to the female sex. However recent observations have found recombination to be present in males at a frequency in the order of 0.1% and furthermore that this frequency may be influenced by the presence of chromosomal rearrangements.

A series of sex-linked and autosomal translocations in which the genetic background was controlled was constructed to examine more fully this phenomenon of male recombination. Examination of four chromosome 5 translocations yielded male recombination frequencies in the order of 1.4-2.1% some 10 to 15 times greater than the frequency observed in relevant controls.

Results are discussed with reference to the mass rearing and subsequent release of sex-linked translocations for the genetic control of Lucilia cuprina.

MEIOTIC PAIRING OF SEX CHROMOSOMES IN LUCILIA CUPRINA MALES

D. Bedo, Division of Entomology, CSIRO, Canberra.

Meiotic pairing relationships of X and Y chromosomes in male Lucilia cuprina was studied by cytological observation of normal, rearranged and deficient sex chromosomes in spermatogenesis. Only two X/Y pairing sites could be defined, one in each arm of the X and Y chromosomes. These pairing sites show specific recognition of their partners. Short arm pairing was localised to their ends, very close to or within the nucleolar organising secondary constriction. This pairing is very tight and not disrupted by chromosome rearrangement, deficiency for the Y chromosome long arm or supernumerary X chromosomes. Pairing of long arms could not be well localised but is likely to involve their distal ends in a much weaker association readily disrupted by chromosome rearrangement. Long arm pairing fails in flies deficient for the Y short arm. No significant pairing was observed between X chromosomes and the remainder of the Y chromosome. In males with an extra X chromosome, the ends of both X chromosomes pair to form multivalents with normal or rearranged Y chromosomes if the Y short arm is present, or an X bivalent if the Y short arm is deleted. The mechanism of pairing in Lucilia sex chromosomes seems to depend on specific loci of unique structure within heterochromatic regions. Sites of rDNA may have a strong influence in the function of the pairing sites.

FREQUENCY-DEPENDENT SELECTION IN LUCILIA CUPRINA

Jennifer Fegent, Gaye Weller and John McKenzie

Department of Genetics, University of Melbourne

Laboratory studies have shown egg to adult viability of susceptible (SS) genotypes of the R_1 diazinon resistance locus of the Australian Sheep Blowfly L. cuprina, to be facilitated by the presence of RS genotypes. SS individuals emerged from mixed (ISS:IRS) culture at diazinon concentrations that were lethal to pure SS cultures. RS viability was similar in pure and mixed cultures.

Egg to adult viability of SS and RS was compared over a range of genotypic frequencies and larval densities. At certain insecticide concentrations frequency-dependent selection was observed, the viability of SS genotypes being enhanced. The percentage of adults emerging declined with increasing larval density but similar frequency/viability associations were observed at each density.

Studies have commenced to attempt to define the mechanisms of genotypic interaction. The viability of SS was enhanced on insecticide medium if the medium had been previously conditioned by resistant genotypes.

M. S. Johnson
Department of Zoology, University of Western Australia

An Electrophoretic Analysis of Phylogeny and Evolutionary Rates of *Partula* from the Society Islands

An allozymic study of 30 species in the three genera of partulid snails was used to examine phylogeny and rates of evolution in the genus *Partula* from the Society Islands, French Polynesia. Genetic identities between congeneric species are generally high, with 42% of the *I* values above 0.85, and 24% above 0.90. Identities between species from the same island are particularly high.

Phylogenetic analysis of the allozymes indicates that the radiation of *Partula* has involved single colonizations of each island, followed by endemic speciation. The inferred sequence of colonization is generally consistent with the geological datings of the origins of these volcanic islands, which range from 1 My for Tahiti to 3 My for Bora Bora.

Based on the phylogeny and the ages of the islands, the average rate of allozymic divergence (measured in Nei's *D*) between species from different islands was 0.126/My. Genetic distances between species inhabiting the same island are consistent with this rate of divergence, confirming that recency of speciation is the reason for the generally high similarities between species.

The average rate of allozymic divergence for *Partula* is nearly twice that for amphibians and mammals. This discrepancy and a 3-fold variation in rates of change within *Partula* emphasize the potential errors in using genetic distance to estimate dates of divergence.

SOME EFFECTS OF HOMOZYGOSITY ON HONEYBEE (APIS MELLIFERA) PHENOTYPES

B.P. Oldroyd[1,2] and C. Moran[2]

[1]Plant Research Institute, Burnley Gardens, Swan Street, Richmond, VIC. 3121

[2]Department of Animal Husbandry, University of Sydney, Sydney, N.S.W. 2006

It has been shown in organisms other than Apis that individuals with a relatively greater level of homozygosity have increased morphological variance and asymmetry compared with similar but heterozygous individuals. The phenomenon of developmental homeostasis has been demonstrated in poikilotherms, but there are conflicting results for homeotherms. Honeybees are quasi-homeotherms, and it seemed interesting to investigate the phenomenon in this species.

Data were available for two characters of the honeybee: honey production, and hamuli number. These comprised measurements of a number of individuals of each of nine inbred lines and diallelic crosses made between the inbred lines.

Analysis of variance was used to estimate within-genotype variability for both the inbred and F1 groups. For honey production, within-genotype variation was greater for inbred colonies than for the F1's, indicating homeostasis. The results for hamuli number were conflicting. Where the bees were reared in a standard environment, developmental homeostasis was demonstrated. However, it could not be demonstrated where the bees were reared in their own hive.

P.A. Davies, P.J. Larkin, M.A. Pallotta and W.R. Scowcroft

CSIRO Division of Plant Industry, PO Box 1600, Canberra, ACT.

SOMACLONAL MUTANTS AT THE ADH-1 LOCI IN WHEAT

ADH-1 in wheat is constitutive in the endosperm and is coded by loci on each of the three group four homoeologous chromosomes. The functional enzyme is a dimer and with electrophoresis on cellulose acetate gels resolves into three bands which represent the six possible isozymes.

The progeny of 551 SC1 plants regenerated from tissue cultures of the wheat cultivar, Millewa, were screened by electrophoresis for aberrant zymograms. Sixteen aberrant SC1 families were identified. Cytogenetic analysis indicated that all were either aneuploid or involved chromosomal rearrangements.

The chromosomal rearrangements resulted in the duplication or deletion of an Adh-1 locus. One such variant, SV1, also involved a duplication of the Rht1 allele which had a distinctive effect on both seedling growth response to gibberellic acid and mature plant height.

Among the aneuploid variants some were a direct consequence of chromosome loss during culture, while others appeared to be due to chromosome non-disjunction at meiosis in SC1 plants, probably as a result of a translocation event during tissue culture.

The analysis provides definitive evidence that genomic rearrangements occur at an enhanced frequency during tissue culture. This has direct application to enhance introgression of alien genes from related species into bread wheat.

WEDNESDAY 28TH AUGUST (CONT'D)

10.50-12.30 pm — Session 2A (P.G. Price Theatre)
Chairman - Dr. D.S. Smyth

10.50-11.10 am — G. Peters (Queen Elizabeth Hospital, Adelaide)
The variable behaviour of univalents in first metaphase: an explanation

11.10-11.30 am — L.C. Shanahan (University of Adelaide)
Cytogenetics of Australian scorpions

11.30-11.50 am — R.I.S. Brettell, W.R. Scowcroft, E.S. Dennis and W.J. Peacock (CSIRO Plant Industry, Canberra)
Analysis of somaclonal mutants of maize at the alcohol dehydrogenase loci

11.50-12.10 pm — J. Wrigley (La Trobe University)
X-chromosome inactivation in monotremes?

12.10-12.30 pm — D. Hales (Macquarie University)
Sex determination and X-chromosome behaviour in aphids

12.30-2.00 pm — Lunch Break

10.50-12.30 pm — Session 2B (Theatre W5A, T2)
Chairman - Professor J.A. Pateman

10.50-11.10 am — M. Davis, C. Cobbett and M.J. Hynes (University of Melbourne)
amdS-lacZ fusions in *Aspergillus nidulans*

11.10-11.30 am — R.A. Sandemann and M.J. Hynes (University of Melbourne)
Cloning of glyoxylate bypass genes of *Aspergillus nidulans*

11.30-11.50 am — D. Rouch, J. Camakaris and B. Lee (University of Melbourne)
Multigenic copper resistance in *E. coli*

*11.50-12.10 pm — A.R. Walker, A.J. Howells and R.G. Tearle (Australian National University
The cloning and characterisation of the vermilion gene of *Drosophila melanogaster*

*12.10-12.30 pm — A.G. Tearle, D. Boyle and A.J. Howells (Australian National University)
Molecular biology of the scarlet and white genes of *D. melanogaster*

12.30-2.00 pm — Lunch Break

(*no abstract)

THE VARIABLE BEHAVIOUR OF UNIVALENTS IN FIRST METAPHASE: AN EXPLANATION.

G.B. Peters, Genetics Department, The Queen Elizabeth Hospital, Woodville, South Australia, 5011.

Abstract

When univalent chromosomes are present at first meiotic division, their behaviour is highly variable. In some cases, both sister kinetochores orient towards the same pole (syntely) while in others, the two kinetochores orient to opposite poles (amphitely). The former may be accompanied by recurrent pole-to-pole oscillation, although, as the present report suggests, this is not the only variation which can be found in conjunction with syntelic orientation. This study attempts to explain this diversity of orientation phenomena. A computer model was constructed which can generate all of the common univalent behaviours. Simulation experiments suggest that chromosome length is critical in determining whether amphitelic or syntelic orientation will predominate. Syntely should be common among large univalents, but oscillations will be rare or absent if the chromosome has any capacity for microtubule attachment at its non-centromeric end. It appears that gross attributes of the chromosome may influence the orientation of univalents during the first meiotic division, with consequent effects on the probability of transmission to the next generation.

CYTOGENETICS OF AUSTRALIAN SCORPIONS.

Catherine Shanahan

Department of Genetics, University of Adelaide,
South Australia 5001.

Australia has two major families of scorpions, Buthidae and Scorpionidae. General chromosome morphology and behaviour at mitosis and meiosis have been interpreted as evidence that species from both families possess non-localized centric activity. Males from both groups exhibit achiasmate meiosis and extensive chromosome polymorphism. Scorpionid species show inversion and fusion/fission polymorphism (2n=29 to 2n=64) manifested by trivalents and quadrivalents at meiosis. Buthid species (2n=14, 2n=16) exhibit ring formations involving from 4 to 12 chromosomes indicative of complex interchange polymorphism. Chromosomes from these two groups have been compared using X-ray induced chromosome breakage, C-banding, kinetochore staining, and transmission electron microscopy of synaptonemal complexes, kinetochores and regions of microtubule attachment. Results suggest there may be a basic difference in the chromosome organisation of the two families. Scorpionids possess features indicative of monocentric chromosomes while Buthids exhibit features indicative of holocentric chromosomes.

R.I.S. Brettell, W.R. Scowcroft, E.S. Dennis & W.J. Peacock

CSIRO Division of Plant Industry, P.O. Box 1600, Canberra City, ACT 2601

ANALYSIS OF SOMACLONAL MUTANTS OF MAIZE AT THE ALCOHOL DEHYDROGENASE LOCI

In many plant species, including maize, stable genetic variants have appeared after a cycle of tissue culture. The mechanisms responsible for this somaclonal variation have not been elucidated. One approach which may shed light on the phenomenon, is to examine mutants at defined loci which are amenable to molecular analysis. For this purpose, maize plants regenerated from tissue culture were screened for variant alcohol dehydrogenase (ADH) isozymes.

Shoots were regenerated from cultures that were initiated from immature embryos carrying both the Fast allele and the Slow allele of Adh1. These shoots developed roots upon transfer to medium lacking growth regulators. The roots had a good activity of both alcohol dehydrogenases, ADH1 and ADH2, without a specific induction treatment. Extracts from the roots were run on lithium borate starch gels which were then stained for ADH activity.

From 750 individual regenerant plants, representing 190 immature embryos, one ADH1 electrophoretic variant has been detected. The variant was not present among four other plants regenerated from the same immature embryo and is therefore presumed to have arisen as a consequence of the culture procedure. The variant has a slower electrophoretic mobility than the presumed progenitor allele, has full ADH1 activity, and appears to be stably transmitted to progeny. The Adh1 gene of the variant has now been cloned, and sequence data will be presented. Restriction endonuclease analysis reveals that the mutant is derived from the Slow allele of Adh1 and is not the result of a large insertion or deletion in the gene.

X-CHROMOSOME INACTIVATION IN MONOTREMES??

Jacki Wrigley
Department of Genetics and Human Variation
La Trobe University, Bundoora, 3083, Victoria.

In eutherian and metatherian mammals, dosage compensation is achieved in the females by X-chromosome inactivation. The inactive X is heterochromatic, late replicating and not transcribed.

Monotremes are prototherian (or non-therian) mammals which have evolved independently from the therian line for about 200 million years. Thus, it is of considerable interest to determine whether they, too, display this important regulatory mechanism.

It is not possible to study X-chromosome inactivation in monotremes using classical genetics or biochemical methods, as the animals don't breed in captivity and no genetic markers have been assigned to the X-chromosome. Consequently, the method of choice has been to determine whether or not one of the X chromosomes in female platypuses and echidnas is late replicating.

Diploid fibroblasts were cultured from female platypus and echidna, and were pulsed with ^{3}H-thymidine, or with BrdU, during early or late S phase. Chromosome spreads were examined after autoradiography or late replication banding, respectively. In those cells, the two X chromosomes replicate entirely synchronously; neither method revealed a late replicating X in any cell. This result contrasts with asynchrony reported previously (Murtagh 1977) for echidna lymphocytes, and raises the possibility that X chromosome inactivation is tissue-specific in monotremes.

Murtagh, C.E. (1977) Chromosoma *65*: 37--57.

SEX DETERMINATION AND X-CHROMOSOME BEHAVIOUR IN APHIDS

Dinah Hales,

School of Biological Sciences, Macquarie University, North Ryde, N.S.W. 2113.

Aphids in temperate climates produce males parthenogenetically in response to environmental cues signalling the arrival of unfavourable conditions. By treating aphids with precocene (cytotoxic to the corpus allatum) and subsequently with the juvenile hormone analogue kinoprene, it has been shown that male eggs are ovulated when the level of juvenile hormone falls below a certain threshold. Since male aphids have an XO sex chromosome constitution, this implies that juvenile hormone controls X-chromosome behaviour at the maturation division of parthenogenetically produced, diploid eggs in aphids. Some new observations on X-chromosome behaviour at this division are reported, with comments on their possible genetic implications.

amdS-lacZ fusions in *Aspergillus nidulans*

Meryl A. Davis, Chris Cobbett and Michael J. Hynes
Department of Genetics, University of Melbourne

ABSTRACT

The *amdS* (acetamidase) gene of *Aspergillus nidulans* is regulated by multiple independent controls. Expression of the gene is induced by acetate (*facB* and *amdA*), ω-amino acids (*amdR*) and benzamide/benzoate (?). The relief of carbon catabolite repression (*creA/B/C*) and/or nitrogen metabolite repression (*areA/tamA*) is also necessary for *amdS* expression.

The isolation of *cis*-acting mutations has allowed identification of the presumptive 5' sites of action of *facB* (*amdI9*), *amdA* (*amdI66*) and *amdR* (*amdI93*). To further characterise the 5' noncoding region of *amdS*, *amdS-lacZ* fusions have been constructed.

Using co-transformation with a selectable (*prn*$^{+}$) plasmid, the *lacZ* fusion plasmids were introduced into *A. nidulans*. Screening of the transformants on X-gal containing media showed that the *E. coli lacZ* can be expressed and the degree of expression is related to the gene copy number in the transformants. Plate tests and enzyme assays confirm that *lacZ* gene expression is regulated by the 5' *amdS* controlling region.

Construction of a *lacZ* fusion plasmid retaining only the most proximal 5' region of *amdS* is being used to further define the various *cis* regulatory regions and to explore nitrogen control in this organism.

CLONING OF GLYOXYLATE BYPASS GENES OF *ASPERGILLUS NIDULANS*

R. A. Sandeman and M. J. Hynes
Department of Genetics, University of Melbourne

The *amdS*, acetamidase, gene of *A. nidulans* is under the positive control of a number of independently acting regulatory genes. One of these genes, *facB*, also controls three enzymes involved in the glyoxylate bypass of the TCA cycle. This bypass allows the use of acetate as a sole carbon source. The genes, of the glyoxylate bypass, under *facB* control are *facA* - acetyl CoA synthase, *acuD* - isocitrate lyase and *acuE* - malate synthase.

The *facA* and *acuE* genes were cloned using the following procedure. Double stranded cDNA's were made from acetate induced mRNA, digested with Sau3A and inserted into M13mp9. These clones were then screened with uninduced and acetate induced cDNA probes. The positive clones obtained were then used to screen lambda gene banks. Their identity has been confirmed by transformation into suitable *facA*$^-$ and *acuE*$^-$ strains of *A. nidulans*. Northern analysis has shown that *facA* produces a message of 2.6 kb and *acuE* a message of 1.8 kb. The direction of transcription of these genes has been determined. Sequencing of the 5' regions of these genes is underway and will be compared to the 5' sequence of the *amdS* gene.

acuD and *facA* are located less than one map unit apart on chromosome V. At present a cosmid library is being screened with *facA* in order to clone the *acuD* gene.

MULTIGENIC COPPER RESISTANCE IN *E. COLI*

Duncan Rouch, J. Camakaris and Barry Lee

Department of Genetics, University of Melbourne

The conjugative plasmid pRJ1004 confers inducible copper resistance in *E. coli*. Two plasmid coded determinants contribute to the resistance and the function of at least one chromosomal gene is required for full expression of resistance.

The major copper resistance determinant *pco* has been cloned. Transposition ($\gamma\delta$) mutagenesis and complementation studies show that *pco* is a 5.8 kb segment which contains at least four genes which do not function as an operon. Functions have been assigned to two of the genes in this *pco* cluster.

WEDNESDAY 28TH AUGUST (CONT'D)

2.00- 2.10 pm Welcoming address by Emeritus Professor G.E. Roberts, Deputy Vice-Chancellor, Macquarie University

2.10- 4.10 pm Guest lecture and invited papers in P.G. Price Theatre
Chairman - Dr. W.J. Peacock

2.10- 3.10 pm Dr. S.J. O'Brien (National Cancer Institute, Maryland)
On the evolution of genomic organisation in mammals

3.10- 3.40 pm J.A.M. Graves, G.W. Dawson and A. Dobrovic (La Trobe University)
Gene Mapping in marsupials: the marsupial X chromosome

3.40- 4.10 pm J.H. Bennett, D.L. Hayman and R.M. Hope (University of Adelaide)
Genetic studies in the marsupial *Sminthopsis crassicaudata*

4.10- 4.30 pm Afternoon Tea

4.30- 5.30 pm Guest lecture in P.G. Price Theatre
Chairman - Dr. J.A. Sved

Professor J. Maynard-Smith (University of Sussex)
Evolution of Recombination

5.30- 6.30 pm Poster session in Seminar Room W5C, 220

GENE MAPPING IN MARSUPIALS; THE MARSUPIAL X CHROMOSOME

Jennifer A. Marshall Graves, Garey W. Dawson, Alex Dobrovic
Department of Genetics and Human Variation
La Trobe University, Bundoora, 3083, Victoria.

Many interesting questions regarding the evolution of the mammalian genome, and the functional significance of mammalian gene arrangements may be approached by comparing gene arrangements in closely- and distantly-related species. Marsupials diverged from placental mammals 130-150 million years ago; it is therefore of special interest to compare gene maps of closely and distantly related marsupial species, and to compare marsupial gene maps with information on placental mammals. The X chromosome is particularly interesting because information about the location and expression of genes is required for an understanding of the evolution of mammalian sex chromosomes and the X chromosome inactivation mechanism.

Classic genetic analysis of marsupials has been difficult. We have chosen the somatic cell genetic approach, which relies on fusion between cells from marsupial and placental species, segregation of marsupial chromosomes from cell hybrids, and correlations between segregation of genes (synteny testing) and between genes and particular chromosomes (chromosome assignment). We have obtained many hybrids from fusions with cells from 3 macropodid and 3 dasyurid species. The hybrids are unstable, and show extreme loss, and fragmentation of marsupial chromosomes, making mapping difficult (but also making it possible to determine gene order). We have determined that in all these species, genes which are X linked in placental mammals (*Hpt Pgk Gpd* and *Gla*) are syntenic, and can be assigned to the marsupial X chromosome; however, *Sts* is not present even in hybrids retaining an intact X. Another 20 markers can be excluded from the X. The four markers assigned to the X are also X linked, and subject to X chromosome inactivation in placental mammals; Sts is X linked, but not inactivated in humans, and is located on the XY pairing region in the mouse. This region appears to be absent from the marsupial X.

Gene order on the X has been determined for several species, and is consistent with the hypothesis of a chromosome-wide control of X chromosome inactivation, with tissue-specific spreading from a single inactivation centre.

1. J.H. Bennett, D.L. Hayman and R.M. Hope

2. Department of Genetics, University of Adelaide, Adelaide, South Australia 5001.

3. Title

Genetic studies in the marsupial *Sminthopsis crassicaudata.*

4. Abstract

A laboratory colony of the fat-tailed marsupial insectivore *Sminthopsis crassicaudata* was set up in Adelaide almost 20 years ago. The aim was to develop a small laboratory-bred marsupial suitable for intensive genetic studies as well as wider biological use (e.g. in the study of early development or for toxicological work). This colony, which has been closed for at least 10 years, has undergone a big increase in numbers in the last few years. Data on reproduction and breeding structure taking account of computerized records of ancestry and inbreeding levels will be reviewed.

Genetic variants for a number of characters are segregating in the colony. Analysis of blood samples using electrophoresis and isoelectric focussing has provided extensive family data and some population data for protein variants involving 7 gene loci. In addition, variation in dorsal pellage colour has been quantified using reflectance measurements, and heritability estimates, as well as population data on reflectance have been obtained. For the protein coding loci, family data are in close accord with Mendelian expectations. Differences between populations in gene frequency and pellage reflectance will be described and discussed.

Linkage has been established for a number of the autosomal gene markers. These linkage data, the first to become available for a marsupial species, show very unusual sex differences. These data will be reviewed and possible explanations for the sex differences will be discussed.

THURSDAY 29TH AUGUST

9.00-10.20 am — Session 3A (P.G. Price Theatre)
Chairman - Professor K.L. Williams

9.00- 9.20 am — J.L. Joseph, J.W. Sentry and D.R. Smyth (Monash University)
Comparative organisation of the *del* family of dispersed repeated sequences in *Lilium longiflorum and L. henryi*

*9.20- 9.40 am — P. Roberts and A. Lohe (CSIRO Entomology, Canberra)
Evolution in satellite DNAs in chromosomes of *Drosophila* species

9.40-10.00 am — W.L. Gerlach, J.M. Buzayan and G.E. Bruening (CSIRO Plant Industry, Canberra)
Biological activity of cloned transcripts of satellite tobacco ringspot virus RNA

*10.00-10.20 am — A.J. Prior and G. Lawrence (CSIRO Plant Industry, Canberra)
Inheritance of ds RNAs in *Melamspora lini*

10.20-10.50 am — Morning Tea

9.00-10.20 am — Session 3B (Theatre W5A, T2)
Chairman - Dr. C. Moran

9.00- 9.20 am — D. Haig (Macquarie University)
Kin conflict in the evolution of seed plants

9.20- 9.40 am — Y.L. Fripp and A.R. Griffin (La Trobe University)
Temporal variation of gene frequencies in a population of *Eucalyptus regnans*

9.40-10.00 am — R.M. Harding (La Trobe University)
Microevolution in Tasmanian registration districts, 1838-1950

10.00-10.20 am — R.J. Mitchell and M. Kosten (La Trobe University)
Coefficient of relationship in the white population of Tasmania - How homogeneous were the earliest immigrants?

10.20-10.50 am — Morning Tea

(*no abstract)

Comparative Organization of the *del* Family of Dispersed Repeated Sequences in *Lilium longiflorum* and *L. henryi*

J. L. Joseph, J. W. Sentry and D. R. Smyth

Department of Genetics, Monash University

The genomes of *Lilium* species are very large, falling in the range of 30 and 40 million kbp. The most highly repeated sequence component of two species has been studied. This was identified as DNA which reanneals by a C_0t of 1 M sec. This is greatly enriched for sequences which are repeated 10,000 times or more per genome.

In *Lilium henryi* 2% of DNA reanneals by this C_0t. About half of this is attributable to a family of dispersed sequences, *del*. Originally isolated in two parts from a BamHI digest of genomic DNA, the full element is about 8 kbp long and is scattered throughout *L. henryi* chromosomes. When a partial library of 15 kbp fragments of *L. henryi* DNA are screened with *del* sequences, 4% show homology. This indicates that about 50,000 *del* repeats occur per genome.

The *del* family is also present in a distantly related species, *L. longiflorum*. About 10% of the *L. longiflorum* genome reanneals by a C_0t of 1 M sec, and a major portion of this is comprised of *del* repeats. A *L. longiflorum* library yields almost one in three clones homologous to *del* probes, implying about 400,000 copies per genome. Although the *del* repeat is highly conserved between *L. henryi* and *L. longiflorum*, distinct differences occur in length and sites of restriction.

Within the 8 kbp *del* sequences a direct repeat of 1 kbp occurs at the termini. This, together with their scattered distribution and differing copy number between species, strongly indicates the *del* sequences are mobile.

BIOLOGICAL ACTIVITY OF CLONED TRANSCRIPTS OF SATELLITE TOBACCO RINGSPOT VIRUS RNA

W.L. Gerlach, J.M. Buzayan*, and G.E. Bruening*

CSIRO Division of Plant Industry, Canberra

*Department of Plant Pathology, University of California, Davis, U.S.A.

The satellite RNA of tobacco ringspot virus (STobRV RNA) increases to a detectable level in plant hosts only when co-inoculated with any of several TobRV strains that serve as supporting virus. STobRV RNA becomes encapsidated in TobRV coat protein and greatly reduces the severity of the symptoms that would have been observed with TobRV alone as the inoculum. STobRV RNA therefore has potential as the agent in a biological control system. We have cloned full-length, permuted monomeric and multimeric cDNA sequences of the ca.360 nucleotide STobRV RNA. As well as various cDNA sequences, RNA transcripts of both polarities have been prepared for testing. Double stranded cDNA and "+" strand RNA have biological activity when inoculated onto plants.

KIN CONFLICT IN THE EVOLUTION OF SEED PLANTS

David Haig
School of Biological Sciences, Macquarie University

A scenario for the evolution of double fertilization in angiosperms will be presented. Polyzygotic polyembrony is common among gymnosperms. In some species, a pollen grain will father more than one embryo within an ovule. It is suggested that the endosperm originated as an "identical twin" of the angiosperm embryo. This "twin" became specialized as a nutritive tissue for the benefit of the embryo. The theory of kin selection will be used to suggest explanations for some other features of seed development.

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TEMPORAL VARIATION OF GENE FREQUENCIES IN A POPULATION OF EUCALYPTUS REGNANS

Individual members of any potentially interbreeding group of plants may differ in the times at which flowering starts, peaks and finishes. The temporal heterogeneity that such variation may lead to in the gene frequencies of the pollen pool has been examined in a small population of *Eucalyptus regnans* using polymorphic allozyme loci.

Dates of first, last and peak flowering and number of flowers produced in the 1982 flowering season were estimated for each tree. The progress of pollen release and receptivity of stigmas during its flowering period was estimated for each tree by fitting a beta distribution to its observed flowering dates with the mode of the distribution at the date of peak flowering. Using these distributions, the estimated number of flowers for each tree and the known genotype of each tree, gene frequencies in the gamete pool were estimated as a function of time for the total population, and also for the outcrossing pollen pool of individual trees. These gene frequencies were then compared, for their ability to predict genotypic frequencies in the progeny of individual trees, with these derived following standard estimation procedures where gene frequencies are assumed to be constant over a flowering season.

The results obtained will be presented, and some implications of such temporal heterogeneity of gene frequencies in the pollen pool will be discussed.

Microevolution in Tasmanian registration districts, 1838-1950

Rosalind M. Harding
Department of Genetics and Human Variation,
La Trobe University,
Bundoora, Victoria 3083.

The historical demography and population structure has been investigated for the south-east coast of Tasmania, Australia using the civil registers of births and marriages for the districts of Glamorgan, Spring Bay and Sorell. Tasmania is an ideal choice for such studies because penal settlement and bureaucracy established procedures for excellent historical documentation of the population from its founding.

Means and variances in sibship sizes were calculated from aggregated births and reconstituted family data to determine effective population sizes and Crow's index of 'the maximum opportunity for natural selection'. Surnames, occupations and birthplaces were used to examine patterns of inbreeding, endogamy and marital mobility.

The potential for genetic drift and natural selection and the degree of population constancy and structure were comparable to levels reported for other rural mainstream populations in England and the United States for the same time period. However, all the analyses indicate that the most important evolutionary process underlying the genetic structure of these districts in Tasmania has been gene flow.

R. J. Mitchell and M. Kosten.

La Trobe University.

COEFFICIENT OF RELATIONSHIP IN THE WHITE POPULATION OF TASMANIA - HOW HOMOGENEOUS WERE THE EARLIEST IMMIGRANTS?

The surnames listed in the 1856 electoral roll for the House of Assembly in Tasmania are used to investigate two questions: (1) how representative were the early Tasmanian settlers of the populations they emigrated from, and (2) how genetically similar were these early immigrants across regions of the State?

The limited franchise of the 1856 roll (males over 21 who met specific qualifications) restricted the number of electors to 13.2% of the total population in 1857 and 37.6% of adult males. Despite this limitation the data are most informative.

Comparison of the 50 most frequent surnames on the roll with the 'top 50' in England and Wales in 1853 reveals a high correlation ($r = 0.55$). The correlation between the 'top 10' on both lists is even higher ($r = 0.76$). In fact, only 10 out of the 50 names are not common to both lists.

Lasker's coefficent of relationship by isonymy (Ri) was used to measure genetic similarities across selected regions of Tasmania as at 1856. The matrix of Ri values will be presented and discussed in the context of the physiography and settlement pattern of the various regions of the State. The limitations of the methods of analysis will be discussed.

THURSDAY 29TH AUGUST (CONT'D)

10.50-12.30 pm — Session 4A (P.G. Price Theatre)
Chairman - Dr. M.S. Johnson

10.50-11.10 am — S. Burgin (Macquarie University)
Electrophoretic variation in *Lampropholis* skinks

11.10-11.30 am — A.L. Freeth and J.B. Gibson (Australian National University)
Alcohol dehydrogenase null activity alleles from natural populations of *Drosophila melanogaster*

11.30-11.50 am — D.J. Schafer and S.W. McKechnie (University of New England)
The *Adh* polymorphism of *Drosophila melanogaster*: Longevity of adults on ethanol at various temperatures

11.50-12.10 pm — B.W. Geer and S.W. McKechnie (Monash University)
Effects of dietary ethanol and *Adh* on lipids of *Drosophila melanogaster* larvae

12.10-12.30 pm — A. Hoffmann, G. Simmons and M. Turelli (University of California, Davis)
Unidirectional incompatibility between populations of *Drosophila simulans*

12.30- 2.00 pm — Lunch Break

10.50-12.30 pm — Session 4B (Theatre W5A, T2)
Chairman - Professor J.A. Thomson

10.50-11.10 am — N.G. Ehiobu and M.E. Goddard (Vic. Department of Agriculture)
Heterosis and genetic distance among populations of *D. melanogaster*

11.10-11.30 am — R. Appels, C.L. McIntyre, B.C. Clarke and C.E. May (CSIRO Plant Industry, Canberra)
Alien chromatin in wheat: ribosomal DNA spacer regions as specific probes for *nor* containing chromosome segments

11.30-11.50 am — R. Appels and L.B. Moran (CSIRO Plant Industry, Canberra)
Rye heterochromatin: studies on clusters of the major repeating sequence and the identification of a new dispersed repetitive sequence element

11.50-12.10 am — C.L. McIntyre (Australian National University)
Alien chromatin in wheat: studies on a dispersed repetitive DNA sequence from the E genome

12.10-12.30 pm — J.P. Gustafson and A.J. Lukaszewski (University of Missouri)
The manipulation of alien chromosome translocations in wheat utilizing wheat-rye hybrids

12.30- 2.00 pm — Lunch Break

SOME RELATIONSHIPS OF LAMPROPHOLIS SKINKS

Shelley Burgin

School of Biological Sciences

Macquarie University

Due to the effects of co-evolution, reptiles are often difficult to distinguish on morphological grounds alone. Relationships both among members of the Scincid genus Lampropholis and between the genus and its closest relatives are no exception. The use of techniques such as electrophoresis and micro-complement fixation have proved useful in investigating these relationships. The present study investigates Lampropholis relations using these techniques; some preliminary findings are discussed.

Alcohol dehydrogenase null activity alleles from natural populations of *Drosophila melanogaster*

A.L. FREETH and J.B. GIBSON

Department of Population Biology
Research School of Biological Sciences
Australian National University
Canberra, Australia

Alcohol dehydrogenase (*Adh*) null activity alleles have been detected in a number of Australian populations of *Drosophila melanogaster* at frequencies up to 3.9%, but with an average of 1.3% in 1983 and 0.7% in 1984. These values compare with a previously reported frequency of 0.09% for *Adh* (Langley and co-workers) in a North Carolina (USA) and a London (GB) population. Of the twelve extracted second chromosomes bearing *Adh* null alleles, four were homozygous lethal, but all were viable in combination with *Df(2L)64j*.

Six *Adh* null activity alleles isolated from the 1983 collections have been shown to be incapable of forming active heterodimers when heterozygous with either Adh^F or Adh^S. The levels of ADH activity and ADH protein in these heterozygotes were in the range expected for individuals with a single copy of the *Adh* gene. No interallelic complementation has been detected in crosses between these nulls. Southern Blot analysis has confirmed the presence of the *Adh* gene in the nulls.

Darren J. Schafer and Stephen W. McKechnie*

University of New England and Monash University*

THE ADH POLYMORPHISM OF *DROSOPHILA MELANOGASTER*: LONGEVITY OF ADULTS ON ETHANOL AT VARIOUS TEMPERATURES

Variation in two adult *Adh* fitness components, each measured at more than one temperature, was examined on flies extracted from a large and relatively outbred laboratory population recently established from the Tahbilk winery cellar population.

The first test measured the number of progeny produced by females after feeding, maturing and mating over wine seepage. Expectedly, higher numbers of progeny were produced at 20°C than at 15°C. However no significant differences in production occurred among genotypes at either temperature.

In the second test, carried out at 4 temperatures, adults were sealed in vials over vapour from (a) water, (b) 3% ethanol and (c) 6% ethanol. After 50% mortality genotypes of surviving adults were compared to those of adults from the same samples but not vapour exposed. Temperature dependent fitness variation occurred. At 20°C, but not at lower temperatures, *Adh*-SS survival was relatively poor, even in the absence of ethanol vapour. This latter result may help explain the observed association in the cellar of higher Adh^F frequencies with temperatures around 20°-22°C, near the high end of the cellar temperature range.

Billy W. Geer and Stephen W. McKechnie*

Department of Biology, Knox College, Galesburg, Illinois 61401 and Department of Genetics, Monash University*, Clayton, Victoria 3168

EFFECTS OF DIETARY ETHANOL AND ADH ON LIPIDS OF *DROSOPHILA MELANOGASTER* LARVAE

When cultured on a defined diet, ethanol was an efficient substrate for lipid synthesis in wild type *D. melanogaster* larvae. At certain dietary levels both ethanol and sucrose could displace the other as a lipid substrate. Comparison of wild type with *Adh*-null larvae indicated that more than 90% of the flux from ethanol to lipid was via the alcohol dehydrogenase (ADH) system. The activities of the lipogenic enzymes, sn-glycerol-3-phosphate dehydrogenase (GPDH), fatty acid synthetase (FAS), and ADH, together with the triacylglycerol (TG) content of wild type larvae increased in proportion to the dietary ethanol concentration, up to 4.5% (v/v).

Dietary ethanol (at 2.5%, v/v) reduced the chain length of total fatty acids (FA) and increased the extent of desaturation of short chain FA in larvae with a functional ADH. In these larvae dietary ethanol also stimulated an increase in free fatty acid levels and an increase in the relative amounts of phosphatidylethanolamine in comparison to phosphatidylcholine. Although the ethanol-stimulated reduction in saturation levels of short chain FA's occurred in ADH-null larvae, ethanol promoted in these mutants an *increase* in total FA chain length, a *decrease* in FAS activity, and a *decrease* in total TG content. In wild type larvae these ethanol-effected changes in lipid composition may alter important physiological and biochemical properties of the membranes.

Unidirectional Incompatiblity Between Populations of *Drosophila simulans*

Ary Hoffmann, Gail Simmons and Michael Turelli

Department of Genetics
University of California at Davis

Drosophila simulans females from a strain collected at Watsonville, California produce no offspring when mated with males from a strain collected at Riverside 480 km away. This incompatibility does not exist between Riverside females and Watsonville males. In the incompatible cross, mating and oviposition are normal, but eggs fail to hatch. A survey of other California populations indicates that unidirectional incompatibility is widespread. The incompatibility is maternally inherited and is partially overcome when old Riverside males are used. Rearing the strains at 28 C suppresses incompatibility. Culturing the strains on medium with tetracycline restores compatibility, suggesting the involvement of a microorganism.

N. G. Ehiobu[1] and M. E. Goddard[1,2]

1. Graduate School of Tropical Veterinary Science, James Cook University, Qld.

2. Department of Agriculture and Rural Affairs, Victoria.

Heterosis and Genetic Distance among Populations of D. melanogaster.

The amount of heterosis which occurs in crosses between two populations depends on the genetic divergence between the populations. Genetic distance can also be measured from gene frequencies at allozyme loci and expressed as an inbreeding coefficient (F) since the populations diverged. We have compared these two measures of genetic distance using 9 populations of D. melanogaster collected from Australia, PNG, Fiji and England and inbred lines. They were typed for 10 polymorphic loci and heterosis or inbreeding depression for larval survival, fecundity and cold stress mortality estimated.

Heterosis occurred in crosses between populations separated by as little as 300 km but it did not increase as the geographic distance between the populations increased. F showed a tendency to increase as the geographic distance and the climatic dis-similarly increased between the locations from which the populations were collected.

The rate of inbreeding was found to affect the inbreeding depression observed at a constant level of the inbreeding co-efficient. Slow inbreeding caused less depression than fast inbreeding for larval viability, slightly less for fecundity but no less for cold stress mortality.

By combining the estimate of genetic distance (F) between geographic populations with a knowledge of inbreeding depression per % F we attempted to predict the amount of heterosis that would occur in crosses. The average heterosis observed for fecundity agreed with the prediction but the observed heterosis for larval survival was less than predicted and for cold stress mortality it was more than predicted.

These results suggest the following hypothesis. Loci controlling larval mortality are subject to high selection pressures which are relatively uniform all over the world. Consequently genetic divergence between populations is limited even for widely separately populations. On the other hand loci controlling cold stress mortality are subject to different selection pressures in different places so that populations diverge greatly and consequently yield a large amount of heterosis when crossed. Some loci controlling fecundity show uniform selection across populations and others divergent selection.

R. Appels, C.L. McIntyre, B.C. Clarke, C.E. May
Division of Plant Industry, CSIRO, Canberra, ACT.

Alien Chromatin in Wheat: Ribosomal DNA Spacer Regions as Specific Probes for *Nor* containing Chromosome Segments.

The rDNA spacer region from grasses representing the R, S, P, E, N and J_1 J_2 genomes (defined by Dewey, 1984), as well as barley, have been analyzed using cloned DNA fragments. Specific sections of the spacer region were selected which allowed the respective *Nor* loci to be identified in a wheat background. In grasses such as *Secale cereale* (R genome), *Hordeum vulgare* (barley) and *Psathyrostachys junceum* (N genome) the region bounded by *Taq*1 restriction endonuclease sites provided a suitable probe. In all the grasses this *Taq*1 fragment comprises 60 - 80% of the spacer region. The S, P, E and J_1 J_2 genome grasses, being more closely related to wheat, required a smaller section of the spacer, containing only the internally repetitious sequences, to be isolated before a suitable probe was available.

The availability of the probes allows unambiguous identification of chromosome segments, from the grasses characterized, carrying the *Nor* locus as these are manipulated in wheat breeding programmes. Some specific examples are illustrated using wheat lines containing rye and *Thinopyrum* (E genome) chromosome segments.

R. Appels and L.B. Moran
CSIRO Division of Plant Industry, Canberra, ACT

Rye Heterochromatin : Studies on Clusters of the Major Repeating Sequence and the Identification of a New Dispersed Repetitive Sequence Element.

The structure of approximately 10 kb of DNA originating from rye heterochromatin is described and the implications of these findings to the origin and maintenance of this sequence family is discussed. The analysis of a molecule which contained a junction between the heterochromatic "350" DNA sequence family and apparently non - heterochromatic DNA is also presented. This non - heterochromatic DNA contains a dispersed repetitive type of sequence ("5.3") which is found adjacent to a wide range of sequences as judged from genomic analyses and independent isolates of the sequence. The value of this type of sequence for further analysis of the rye genome is discussed.

C.L. McIntyre, Australian National University, Canberra, ACT.

Alien Chromatin in Wheat: Studies on a Dispersed Repetitive DNA Sequence from the E Genome.

The E genome is one of the genomes found among the perennial members of the tribe Triticeae, and is present in the diploid grass Thinopyrum elongatum (Host) Dewey (= Agropyron elongatum (Host) Beauvois). T. elongatum and other E genome species are of interest to wheat breeders as they contain many agronomically important characteristics such as disease resistance genes. Genome-specific DNA sequences can be used as probes to detect successful introductions of alien chromatin into wheat. In addition, such sequences can provide information on evolutionary relationships among the related genomes of the tribe Triticeae.

The DNA sequence designated 145.3 is a 1.4kb fragment isolated from a clone containing part of a tandem array of repeated sequences in T. elongatum. 145.3 assays a unique restriction enzyme band in T. elongatum that is not present in wheat, and is dispersed on all the chromosome arms of T. elongatum. Sequencing studies reveal that it may have arisen from a duplication event, as well as allowing smaller more E-genome specific sequences to be isolated. A very similar sequence, designated 143.3, was isolated independently from a second clone, indicating that they form part of a major family of repeated sequences in T. elongatum. 143.3 contained only 6% base pair changes with respect to 145.3. 145.3 has also been shown to hybridize strongly to the N genome (Psathyrostachys juncea), a distantly related perennial genome, but not to a major N-genome specific dispersed repeat.

J.P. Gustafson and A.J. Lukaszewski
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The Manipulation of Alien Chromosome Translocations in Wheat Utilizing Wheat - Rye Hybrids.

Geneticists have always looked at related species as a vast pool of genes from disease resistance, improved quality, improvement under adverse conditions, etc. When wheat (Triticum aestivum L. em Thell.) and rye (Secale cereale L.) are studied these alien transfers can be accomplished in several ways : by creating amphiploids, by single chromosome additions and substitutions, and by inducing translocations. Each of these methods has had limited success and each has serious drawbacks. The use of translocations has been generally thought to be the most promising. However, this method was thought to require prohibitive amounts ofcytology and has never really been exploited.

In the present study four wheat - rye hybrids (triticales: X Triticosecale Wittmack) were crossed as females to four different wheats with the resulting generations being analysed plant by plant. The results indicate : 1) the number of wheat - rye translocations identified were several times higher than observed in any previous study and that each arm of every rye chromosome was involved at least once ; 2) the number of translocations per plant ranged from 1 to 3 and one in every five plants had at least one translocation ; 3) since four different triticale x wheat populations were involved and translocations were present in all of them, then the results were not restricted to one single genotype of parent ; and 4) the maximum retention of alien variation in a population using the methodology discussed occurs when the populations are not subjected to selection before the F generation.

THURSDAY 29TH AUGUST (CONT'D)

2.00- 3.00 pm — The M.J.D. White Presidential Address (P.G. Price Theatre)
Professor J.A. Pateman (Australian National University)
Evolution: some thoughts on the future

3.10- 4.10 pm — Session 5A (P.G. Price Theatre)
Chairman - Professor M.J. Hynes

3.10- 3.30 pm — K.L. O'Hoy and V. Krishnapillai (Monash University)
Genetic analysis of *Pseudomonas aeruginosa* using transposons and a suicide plasmid vector

3.30- 3.50 pm — R. Hirst, D. Strom, J. Petering and A. Morgan (Monash University)
Transposon mutagenesis in *Pseudomonas putida*

3.50- 4.10 pm — S. Davies, V. Obeyesekere and V. Krishnapillai (Monash University)
Physical and genetic characterization of the transfer regions of the *Pseudomonas* plasmid R91-5

4.10- 4.30 pm — Afternoon Tea

3.10- 4.10 pm — Session 5B (Theatre W5A, T2)
Chairman - Professor R. Frankham

3.10- 3.30 pm — R.J. Mitchell (La Trobe University)
The use of vital data and other historical records in examining the genetic demography of human populations

3.30- 3.50 pm — J.C. Daly (CSIRO Entomology, Canberra)
Evolution of insecticide resistance in the moth, *Heliothis armiger*

3.50- 4.10 pm — R. Poulter (University of Otago)
Parasexual genetics of *Candida albicans*

4.10- 4.30 pm — Afternoon Tea

GENETIC ANALYSIS OF *PSEUDOMONAS AERUGINOSA* USING TRANSPOSONS AND A SUICIDE PLASMID VECTOR

K. L. O'Hoy and V. Krishnapillai

Department of Genetics, Monash University, Clayton, Victoria 3168

In order to extend the genetic map of *P. aeruginosa* strain PAO, transposon mutagenesis of the chromosome has been achieved using 2 transposons: the *E. coli* Tn*5*, and the native *P. aeruginosa* transposon Tn*2521*. These transposons were loaded onto a temperature sensitive, replication defective derivative of the IncP-1 plasmid, R68. At the non-permissive (43°C) temperature, auxotrophic mutations were scored.

Tn*5* mutagenesis generated 30 stable mutants with a wide variety of amino acid requirements. By genetic tests the inserts were shown to be dispersed on the PAO chromosome. From the 30 mutants, 6 new auxotrophic mutations were identified. A variety of auxotrophs were also found using Tn*2521* including two new auxotrophic mutations. The majority of these mutants retained the plasmid stably integrated into the chromosome which led to transfer of the bacterial chromosome in a polarized manner. These Hfr donors have been used in nalidixic acid-interrupted mating crosses to determine the origins of chromosome transfer and the precise location of a range of markers by time of entry.

R.Hirst, D. Strom, J. Petering and A. Morgan

Department of Genetics, Monash University, Clayton, Victoria 3168

TRANSPOSON MUTAGENESIS IN PSEUDOMONAS PUTIDA

The inability of the IncP-10 plasmid R91-5 to maintain itself in Pseudomonas putida PPN allows its use as a natural "suicide" plasmid in this strain. When R91-5 loaded with the transposon Tn5 (pMO75) was conjugally transferred from P. aeruginosa PAO to P. putida PPN, kanamycin resistant exconjugates were obtained at a frequency of 10^{-5} per donor cell, and 0.3% of these were auxotrophic. Identification of the nutritional deficiency, growth response to pathway intermediates, interspecific complementation by R primes and conjugal mapping of the mutations have shown that they are widely distributed around the PPN chromosome, and at least three new loci have been identified and mapped. Reversion studies suggest that the majority of auxotrophic mutations contain a single chromosomal Tn5 insert.

The reintroduction of pMO75 into P. putida chromosome::Tn5 derivatives results in two classes of Hfr donor strains. Each class mobilises the chromosome in a polarised manner from the original site of Tn5 insertion, but in opposite directions. As proximal markers are transferred at 10^{-1} per donor cell, these Hfr strains have been used as donors in interrupted matings. The time-of-entry of markers of known map position allows the accurate mapping of the site of the original Tn5 mutation.

PHYSICAL AND GENETIC CHARACTERIZATION OF THE TRANSFER REGIONS OF THE *PSEUDOMONAS* PLASMID R91-5

S. Davies, V. Obeyesekere and V. Krishnapillai

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R91-5 is a narrow host range conjugative plasmid which replicates and maintains itself stably only in *P. aeruginosa*. This plasmid does however have a conjugational wide host range as shown by its promotion of conjugation between *P. aeruginosa* and a number of other *Pseudomonas* species, as well as between *P. aeruginosa* and *E. coli*. Two unlinked regions control conjugation in R91-5, TraI which is associated with conjugal DNA metabolism, and Tra2 which is associated with sex pili synthesis and functionality. In this study two classes of TraI mutants were isolated, *Tn7* insertion mutants and hydroxylamine induced point mutants. The mapping of a large number of these insertion mutants has allowed the precise physical limits of TraI to be determined, while complementation tests between point mutants has identified three transfer cistrons. These cistrons have been shown to map in a region which overlaps the 2.2 kb found to encompass TraI. Complementation tests are currently being undertaken to confirm the position of these cistrons within TraI and to deduce the transcriptional organization of this region.

R.J. Mitchell

La Trobe University
Department of Genetics, Bundoora, 3083.

THE USE OF VITAL DATA AND OTHER HISTORICAL RECORDS IN EXAMINING THE GENETIC DEMOGRAPHY OF HUMAN POPULATIONS

Human population geneticists often wish to know as much as possible regarding the historical population structure and relevant demographic characteristics of a population. In particular, they wish to measure kinship levels, both within and between populations, in a particular region.

These issues can be most appropriately investigated in humans because of the existence of written records. Civil registration of vital events commenced earliest in Tasmania (1838) and the first census was in 1841. Tasmania, therefore, presents the greatest time-depth for human micro-evolutionary studies in white Australia. The implications of these data for explaining genetic variation in the contemporary Tasmanian population as revealed through a study of blood polymorphisms will be discussed.

Evolution of Insecticide Resistance in the moth, *Heliothis armiger*

Joanne C. Daly
CSIRO, Division of Entomology, Canberra, A.C.T.

Evolutionary events are normally very slow, relative to the generation time of population geneticists. One exception is the evolution of resistance to insecticides which occurs very rapidly in many insect pest species. Populations of the noctuid moth, *Heliothis armiger*, are segregating for an allele(s) conferring resistance to the synthetic pyrethroid insecticides. Studies have begun to elucidate both the genetic basis of this resistance and those factors which are determine frequency of the resistance alleles in the population.

Results from preliminary crosses in the laboratory suggest that resistance is conferred by one semi-dominant allele, although more than one locus may be involved. Populations of *H. armiger* were observed this past season in the irrigation area of Emerald, Queensland. Individuals were sampled from fields before commercial applications of pyrethroids to screen for resistance. Even though resistance was as high as 20 percent, estimates of mortality were 100%. This suggests that the allele for resistance is recessive under standard commercial conditions. Resistant individuals were uniformly distributed over a wide area. The evolution of resistance is discussed in light of both these results.

DR RUSSELL POULTER

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Parasexual genetics of *Candida albicans*.

The imperfect diploid yeast *Candida albicans* is the most common and one of the most serious opportunistic fungal pathogens of man. Recent research[1] has enabled a start to be made on the genetic analysis of this yeast. In the absence of a sexual cycle two approaches have been used: parasexual genetics and molecular genetics. The parasexual systems involve the following procedures: protoplast fusion between complementing auxotrophs on selective media; UV irradiation induced mitotic crossing-over; heat-shock induced chromosome loss. These procedures enable one to perform complementation and recombination analyses. The molecular systems involve the following procedures: construction of plasmids carrying *C. albicans* prototrophic genes, the most useful plasmids are those capable of growing in both *E. coli* and *S. cerevisiae*; the transformation of auxotrophic *C. albicans* with such plasmids, so far only integrative transformation has been achieved; the analysis of *C. albicans* transformants by Southern blotting. Both genetical approaches have made extensive use of red, adenine requiring auxotrophic, strains of *C. albicans* analagous to the ade1 and ade2 mutants of *S. cerevisiae*.

[1] Shepherd, M.G., Poulter, R.T.M. and Sullivan, P.A.
Ann. Rev. Microbiol. (1985). 39, 579-614.

THURSDAY 29TH AUGUST (CONT'D)

4.30-5.30 pm Invited Lecture (P.G. Price Theatre)

Chairman - Dr. D.A. Briscoe

R.L. Close and G.B. Sharman (Macquarie University)

Chromosomal speciation in rock wallabies - stasipatric speciation?

5.30-6.30 pm Annual General Meeting - P.G. Price Theatre

7.00 pm for 7.30 pm - Society Dinner

Macquarie University Union,
Function Room, Level 3.

CHROMOSOMAL SPECIATION IN ROCK WALLABIES - STASIPATRIC SPECIATION?

R. L. Close and G. B. Sharman

School of Biological Sciences, Macquarie University.

The essential features of the stasipatric model of speciation (White, et al. 1967) are the origin and establishment of a chromosomal rearrangement which reduces fecundity when heterozygous and which spreads geographically through a part of the area occupied by a species and may act as an incipient isolating mechanism between the population homozygous for it and the original population (White 1978a). It is also an essential part of the model that it should be possible to recognise that derived taxa occupy geographically interior areas and ancestral taxa peripheral ones (White 1978b).

The rock wallabies (Petrogale: Macropodoidea) are widely distributed in continental Australia and some offshore islands. Between 1841 and 1982 twenty two taxa were described mostly as full biological species. Karyology of valid described and undescribed taxa has revealed the occurrence of nineteen different "chromosome races". In the lateralis - penicillata group of thirteen chromosome races the basic macropodoid marsupial karyotype (Hayman and Martin 1974) was found in the lateralis race of peripheral s.w. Western Australia distribution. The remaining races, of Australian Shield Arid Zone and Eastern Uplands distribution, were distinguished by one or more chromosome rearrangements interpreted as centromere transpositions and inversions (resulting in changes in chromosome shape) and centric and tandem fusions, resulting in changes in chromosome number (Briscoe, et. al. 1982). These arrangements appear to have spread through various parts of the overall range of the lateralis - penicillata group and the derived taxa occupy geographically interior areas as required by the stasipatric model. In the Eastern Uplands, where rock wallabies are virtually continuously distributed from north to south, the chromosome races are of parapatric distribution.

Two narrow hybrid zones were found in the field and hybridization between various chromosome races was studied in captivity. F1 and backcross hybrids exhibited various degrees of reproductive inviability roughly correlated with degree of karyotypic difference between parental races.

The role of chromosome rearrangements in speciation in rock wallabies in relation to stasipatric and classical allopatric models of speciation will be discussed.

References:-

Briscoe, D.A., Calaby, J.H., Close,R.L., Maynes, G.M. Murtagh, C.E. and Sharman, G.B. (1982). In R.H. Groves and W.D.L. Ride (eds.) "Species at Risk: Research in Australia", 73-87 (Australian Academy of Science, Canberra).
Hayman, D.L. and Martin, P.G. (1974). In B. John (ed.) "Animal Cytogenetics 4, Mammalia I: Monotremata and Marsupialia" (Gebrüder Borntraeger, Berlin).
White, M.J.D. (1978a). "Modes of Speciation" (W.H. Freeman and Company,San Francisco).
White, M.J.D. (1978b). Systematic Zoology 27, 285.
White, M.J.D. Blackith, R.E., Blackith, R.M. and Cheney, J. (1967) Australian Journal of Zoology 15, 263.

FRIDAY 30TH AUGUST

9.10-10.30 am — Session 6A (P.G. Price Theatre)
Chairman - Dr. J.A. McKenzie

9.10- 9.30 am — J.L. Harry and D.A. Briscoe (Macquarie University)

Anomalous expression of the Pgk locus in Loggerhead *(Caretta caretta)* turtle embryos

9.30- 9.50 am — M. Mahony (Macquarie University)

Polyploidy and natural hybridisation in Australian ground frogs *(Neobatrachus)*

9.50-10.10 am — E.S. Robinson (Macquarie University)

X-chromosome replication and condensation patterns in marsupials

10.10-10.30 am — P.J. Sharp and D.L. Hayman (University of Adelaide)

The role of chiasmata in the genetic systems of marsupials

10.30-11.00 am — Morning Tea

9.10-10.30 am — Session 6B (Theatre W5A, T2)
Chairman - Dr. R. Appels

9.10- 9.30 am — M.R. Gillings, R. Frankham, J. Speirs and J.M. Whalley (Macquarie University)

Coevolution of X and Y rDNA arrays in *D. melanogaster*

9.30- 9.50 am — A.G. Mackinlay, C.F. Hawkins, J.R. Ovenden and R.H. Crozier (University of New South Wales)

Organization of an avian mitochondrial genome

9.50-10.10 am — D.J. Ayre and J.M. Resing (University of Wollongong)

Coral chimeras

10.10-10.30 am — D.G. Colgan (Australian National University)

Hormonal effects on the developmental changes in the glycolytic enzymes of *Caledia captiva* and other acridid grasshoppers

10.30-11.00 am — Morning Tea

Anomolous expression of the Pgk locus in Loggerhead (Caretta caretta) turtle embryos

J.L. HARRY and D.A. BRISCOE

School of Biological Sciences, Macquarie University, North Ryde, 2113.

ABSTRACT

Electrophoretic analysis of 806 Caretta caretta embryos revealed aberrant genotypic ratios within clutches at the polymorphic Pgk locus. Eight of the nine clutches where 3 phenotypes were observed (N, N/S and S), exhibited a significant deficit (50%) of the presumed heterozygote (N/S) phenotype. It is proposed that this deficit results from only one copy of the Pgk locus being expressed in one half of the embryos. Under this hypothesis Pgk is expressed as a normal autosomal locus in one half of the embryos. In the remaining embryos, random inactivation of one gene copy results in heterozygotes being phenotypically indistinguishable from one or other homozygote. This hypothesis is derived from 3 aspects of the data; (i) the observed ratios of phenotypes within clutches, (ii) the observed heterozygote frequency 0.253, and (iii) the population gene frequencies, are entirely consistent with those predicted under the model. Alternative hypotheses to explain the phenomenon are not compatible with the data. Further, it is proposed that the inactivation of one copy of the Pgk locus, in one half of the embryos, is associated with the inactivation of one set of genetic loci involved in sexual differentiation.

MICHAEL MAHONY

SCHOOL OF BIOLOGICAL SCIENCES, MACQUARIE UNIVERSITY, NORTH RYDE, 2113.

Polyploidy and natural hybridisation in Australian ground frogs (Neobatrachus)

Four of the nine species of Neobatrachus are bisexual tetraploids. Chromosomal studies indicate that the tetraploid species are closely related and probably represent the results of speciation from a tetraploid ancestor rather than independent occurrences of polyploidy. Comparisons of chromosome morphology (relative lengths and centromere positions) and heterochromatin location reveal uniformity, but variation in the location and form of the nucleolar organiser region indicates that structural changes to chromosomes have occurred. There is no indication of diploidization from either mitotic or meiotic chromosomes.

Natural hybridization between diploid and tetraploid species has been detected at three localities. Triploid male and female hybrids are viable and reach maturity, but show much reduced fecundity, although a small number of gametes are produced. Identification of one pentaploid individual, most likely the result of fertilization of an unreduced triploid gamete with a diploid gamete from a tetraploid, indicates that backcrossing does occur. The possible role of hybridization in the origin of polyploidy is discussed.

X CHROMOSOME REPLICATION AND CONDENSATION PATTERNS IN MARSUPIALS

E. S. Robinson

School of Biological Sciences

Macquarie University

Monodelphis domestica, a Brazilian opossum, has rapidly become an important species for laboratory-based marsupial research. It is now being used in North America and Europe for a variety of investigations from general morphogenesis and immunological tolerance to DNA repair mechanisms and genetic control of serum cholesterol concentration and it is widely preferred to Didelphis virginiana as an experimental didelphid model. The limited earlier reports on X chromosome behaviour in D. virginiana include some unusual or contradictory results. The X chromosomes of M. domestica are easily identified because they are the smallest of the (2n=18) female complement and possess a nucleolus organizer region (NOR). X chromosome replication was examined in cells from blastocysts using a 5-bromodeoxyuridine (BrdU)-acridine orange fluorescence technique. In female spreads with satisfactory BrdU incorporation, one of the two Xs was late replicating, except for the NOR, in both the embryonic and extra-embryonic regions. X-linked markers are not available in M. domestica so the type of X inactivation has yet to be determined. Evidence of an interphase condensed X was found in female blastocyst cells and in female juvenile and adult cerebellar neurones. These results are compared with earlier observations on X chromosome behaviour in D. virginiana and with recent work on two Australian marsupials (Macropus robustus and Antechinus stuartii).

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2. Genetics Department, University of Adelaide, South Australia 5001

* Present address: Plant Breeding Institute, Trumpington, Cambridge, England.

3. Title of paper

The role of chiasmata in the genetic system of marsupials.

4. Abstract

Variation in chiasma frequency in the male has been studied in thirty three species of marsupial. The relationship between these data and aspects of the biology of the species studied has been examined to see what patterns may be present. The results of these studies will be described.

Coevolution of X and Y rDNA Arrays in D. melanogaster

M. R. Gillings[1], R. Frankham[1], J. Speirs[2] and J. M. Whalley[1].

[1] Macquarie University, [2] CSIRO Plant Physiology Group

The nucleolus organizers on the X and Y chromosomes of D. melanogaster are the sites of 200-250 tandemly repeated genes for ribosomal RNA. The genes (rDNA) exhibit considerable heterogeneity both within and between locations. This heterogeneity is caused by polymorphism for the length of the spacer regions and by interruption of the 28S coding region by insertion sequences. Despite this heterogeneity, the rRNA transcribed by individual genes on the X and Y is very similar, if not identical.

Molecular, genetic and cytological analyses of a series of X chromosome rDNA deletions (bb alleles) showed that these deletions arose by unequal exchange through the nucleolus orrganizers of the X and Y chromosomes. The exchange events generated compound $X{\cdot}Y^L$ chromosomes carrying mainly Y specific rDNA, although in reduced amounts from that on wild type chromosomes.

Some sublines founded from these homozygous $X{\cdot}Y^L$bb stocks showed spontaneous loss of part or all of the appended Y^L and phenotypic reversion of bb. Some sublines therefore carry Y rDNA in the X nucleolus organizer, yet no longer show gross cytological or phenotypic consequences of the original exchange event. Thus all the criteria for the Y to periodically donate rDNA repeats to the X have been demonstrated. Events such as these are sufficient to account for the coevolution of X and Y rDNA arrays.

A. G. MacKinlay[1]
C. F. Hawkins[1]
J. R. Ovenden[2,3]
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ORGANIZATION OF AN AVIAN MITOCHONDRIAL GENOME

We have cloned approximately four out of five Hind III restriction fragments from the mitochondrial genome of Platycercus elegans elegans, representing 6400 base pairs, from a total of 16,900 (±150). Sequence comparisons show that the gene organization of this section of the Crimson Rosella mtDNA is the same as that of bovine mtDNA from the cytochrome oxidase I gene through to URF 5. Sequence studies on parts of the genes for $tRNA^{ARG}$, cytochrome oxidases I & III, and ATPase 6 show nucleotide homologies as high as 80% with homologies for the amino acid sequences as high as 93%.

Delta values from 26 samples of the various rosella species show heterogeneity in amount of divergence per unit time (although, in view of the time-depth problem, this is not evidence of variation in evolutionary rate), and illustrate the differences in result between tree-building dendrograms assuming constant evolutionary rates and those which do not. Sequence data from the genome being intensively studied also enable determination of the phylogenetic position of rosellas relative to other genetically-significant animals.

CORAL CHIMERAS

BY
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ABSTRACT

Tissue-grafting bioassays have previously been used to assess the genotypic diversity of populations of scleratinian corals. The application of this technique is dependent on the validity of the hypothesis that corals possess a precise system of "self-recognition", such that tissue fusion will occur only between grafted clonemates. In this study, comparisons with multi-locus electrophoretic data show that results from parabiotic tissue grafts are inadequate indicators of clonal identity and population structure.

For each of four species, both tissue-grafting bioassays and electrophoresis were used in an attempt to determine the number of clones present within samples of 16 colonies from areas <800 m^2. Colonies which were electrophoretically distinct were considered to be non-clonemates. The efficiency of the tissue-grafting bioassay as an indicator of clonal identity (relative to electrophoresis) varied between areas and species but was estimated to be 60, 70, 80 and 100% for *Porites cylindrica*, *Seriatopora hystrix*, *Porites nigrescens* and *Stylophora pistillata*, respectively. The self-recognition hypothesis was rejected for *P. cylindrica*, *P. nigrescens* and *S.hystrix*, since pairs of non-clonemates were observed to fuse. This phenomenon was most pronounced within a sample of *S. hystrix*, where 41% of scorable allografts fused and all fusions of different colour morphs). Electrophoresis was therefore considered to be the more reliable indicator of clonal identity, although in one area its usefulness was restricted by our failure to detect more than one variable enzyme-encoding locus. For all species, except *P. nigrescens*, allograft rejections proved valuable adjuncts to electrophoresis in discriminating between colonies which were electrophoretically indistinguishable. We suggest that the best available estimates of genotypic diversity and population structure will combine the results of electrophoretic and allograft rejections.

HORMONAL EFFECTS ON THE DEVELOPMENTAL CHANGES IN THE GLYCOLYTIC ENZYMES OF *CALEDIA CAPTIVA* AND OTHER ACRIDID GRASSHOPPERS

Don Colgan

Department of Population Biology, R.S.B.S., A.N.U.

More than half of the glycolytic enzymes of *Caledia captiva* differ in electrophoretic phenotypes between embryos and adults. The changes between the patterns, which appear to be mostly due to differences in the regulation of individual members of gene families, are coincident in time. They occur near hatching from the egg for all enzymes.

This talk focusses on two questions relating to these observations. The first is the determination of the phylogenetic extent of similar patterns of changes. They have now been found in each of the six studied species of acridids. They are not found in the grasshoppers examined from other families or in insects in other orders than the Orthoptera. Acridid species may also show the startling phenotypic plasticity of phase polymorphism. Comparisons of solitary and gregarious *Locusta migratoria* were made to ascertain whether there is any relation between this polymorphism and the changes in glycolytic enzymes.

The second question addressed by the talk is whether the temporal coincidence of the changes reflects a common underlying cause such as hormonal induction. To test this *C. captiva* embryos and adults were variously treated with juvenile hormone I, ecdysterone and homogenates of first instar hatchlings. Six enzymes and one general protein were studied in these experiments. Significant effects on the enzyme phenotypes were obtained in seven instances involving five different systems and all three types of treatment. There was no evidence that any treatment exerted a concerted effect on all enzymes suggesting that there may not be a single effector of the changes in electrophoretic patterns between stages of the life-cycle.

FRIDAY 30TH AUGUST (CONT'D)

11.00-12.40 pm — Session 7A (P.G. Price Theatre)
Chairman - Professor D.W. Cooper

11.00-12.00 am — F. Nicholas and J.M. Nicholas (University of Sydney)
Charles Darwin in Australia

12.00-12.20 pm — S. Ryan (CSIRO Plant Industry, Canberra)
Novel B-amylase isozymes revealed by tissue culture of wheat

12.20-12.40 pm — J. Landsmann, T.J.V. Higgins, C.A. Appleby, A. Kortt, E.S. Dennis and W.J. Peacock (CSIRO Plant Industry, Canberra)
Evolution of plant hemoglobins

11.00-12.40 pm — Session 7B (Theatre W5A, T2)
Chairman - Dr. D.E.A. Catcheside

11.00-11.20 am — L. Ritchie (Macquarie University)
Frameshift mutagenesis in *S. typhimurium*

*11.20-11.40 am — R. Hall (CSIRO Molecular Biology, Sydney)
Effects of plasmid PKM101 on growth and viability of *E. coli* WP2

11.40-12.00 am — M. Nayudu and B.G. Rolfe (Australian National University)
Identification of host specificity DNA regions determining the broad host range of *Rhizobium* strain NGR234

12.00-12.20 pm — B. Sherwin (University of Melbourne)
The detection of selection in wild populations

12.20-12.40 pm — C.B. Gillies (University of Sydney)
Meiotic chromosome pairing at zygotene in rye

(*no abstract)

CHARLES DARWIN IN AUSTRALIA

F.W. Nicholas and J.M. Nicholas

Department of Animal Husbandry,
University of Sydney

On 12th January, 1836, H.M.S. *Beagle*, with Charles Darwin on board, entered Sydney Harbour on the final stages of its voyage round the world. This was the beginning of Darwin's one and only visit to Australia. During this visit, Darwin spent some time in Hobart and Albany as well as in Sydney, and he also took a trip inland from Sydney to Bathurst. With the 150th anniversary of Darwin's visit only a few months away, attention will be focussed on Darwin's impressions of Australia as recorded in his unpublished Australian notebook, and in his diary, journal, and letters. In addition, background information will be given in relation to some of the places and people visited by Darwin during his stay in Australia.

By a fortunate coincidence, Darwin's shipmates on the Beagle during an earlier stage of the voyage included the artists Augustus Earle and Conrad Martens. Both artists spent time in Australia, and both have left behind paintings and drawings depicting what they saw. Many of these sketches and paintings provide attractive and contemporary illustrations for Darwin's Australian travelogue.

Sarah Ryan

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NOVEL B-AMYLASE ISOZYMES REVEALED BY TISSUE CULTURE OF WHEAT

The isolation and analysis of somaclonal variants at specific loci will exemplify the types of variation which may be expected at other loci, and will help elucidate the genetic mechanisms involved in this phenomenon. Amongst the progeny of ninety wheat plants regenerated from culture, one family was found which was segregating for at least two novel B-amylase bands of mature endosperm identified by isoelectric focussing. The new bands are inherited concurrently and segregate 9 parental : 26 variant in the SC3 generation thus appearing to represent a dominant or co-dominant mutation. The other seven regenerants from the same culture did not contain the variant bands, indicating that the variant did arise in culture. No parental bands appear to be missing in the variant, and both new bands occur in positions not represented in any of 90 diverse cultivars examined for allelic variation. The new bands do not appear in the immature seed of the parent and thus do not represent an altered expression of existing immature isozymes. Mitotic and meiotic chromosome counts and N-banding demonstrate a normal karyotype for this variant. This suggests a fine structural alteration is involved. However a simple alteration of an existing strutural gene does not seem to be implicated, since additional and multiple bands have appeared.

Jorg Landsmann, T.J.V. Higgins, C.A. Appleby, A. Kortt, E.S. Dennis & W.J. Peacock

Evolution of Plant Hemoglobins

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The symbiosis between certain plants and nitrogen-fixing bacteria is of great evolutionary and agricultural interest, but is still poorly understood in molecular terms. An important component in the symbiosis is hemoglobin.

Leghemoglobin (Lb) is detected only in bacterially-induced root nodules of legumes where it can make up 30% of the nodule protein. The apoprotein is synthesized by the plant while the heme moiety is contributed by the bacteria. Lb has a high affinity for oxygen, allowing it to facilitate oxygen flux, bacterial respiration and oxidative phosphorylation at a free oxygen concentration too low to damage the bacterial nitrogenase enzymes.

Plant hemoglobins were thought to be unique to the legume-Rhizobium symbiosis. Recently, similar hemoglobins (Hb) have been isolated from non-leguminous plants such as Parasponia and Casuarina. The nodule-inducing bacteria are not restricted to the genus Rhizobium (although Rhizobium associates with legumes and Parasponia) but can also include the Actinomycetes (as in the case of Casuarina).

There is over 85% DNA and protein homology between the Hbs of two legume species, soybean and kidney bean.

Comparison of the primary structure of leghemoglobins and Hbs from the non-legume Parasponia shows an overall homology of about 40% and significantly higher levels in selected regions of the protein such as in the oxygen and heme-binding regions. This level of homology between legumes and Parasponia Hbs is interpreted as evidence for a common ancestral origin of the plant hemoglobins.

So far only the genes for Lbs have been analysed in any detail. Lbs are encoded by a small family of nuclear plant genes. At least eight genes have been identified in the soybean genome but four of these are pseudogenes. Four Lb genes have been shown to be present in kidney bean. The genes are arranged in two unequal clusters in soybean but there is only one cluster in kidney bean. The complete Lb genes are all interrupted by three intervening sequences (IVS) at identical positions. The positions of IVS-1 and IVS-3 correspond to the positions of the two IVS found in mammalian α, β-and myo-globin genes. These data support the view that all globin genes are derived from a common ancestor. The central exon of the mammalian globin genes may have evolved from a fusion of two ancestral exons.

Comparison of Hb genes from non-legume plants with both Lbs and animal globins will shed more light on the origin and evolution of these globin families.

We have isolated cDNA and genomic clones for Hb from Parasponia, a tree belonging to the Ulmaceae family. Antibodies prepared against soybean Lb show little or no reaction with Parasponia or Casuarina Hb in Western blots. Furthermore, soybean Lb cDNA does not hybridize to Parasponia nodule mRNA or DNA. Therefore synthetic oligo-nucleotides, derived from the Parasponia Hb protein sequence, were used to identify Parasponia Hb specific cDNA clones. Two cDNA clones were sequenced. One of them contains the entire protein coding region for Parasponia Hb1. The second cDNA clone differs by 4 nucleotides. Parasponia Hb is 157 amino acid long, contains 5-6 Met residues and a Cys, the first one found in a plant hemoglobin. The cDNA sequence predicts 4 additional amino acid residues at the NH_2-terminal end, which could not be found in the mature protein. Therefore a preprotein has to be predicted. The cDNA sequence of Parasponia Hb1 is slightly more than 50% homologous to soybean Lbs in the coding region whereas the 5' and 3' noncoding regions exhibit only 25% homology to those from the Lb genes.

Southern analyses with a Parasponia cDNA clone shows a single hyridizing band with various restriction enzymes, indicating only one Hb-gene locus in the Parasponia genome. This might reflect a more primitive gene organization.

We have isolated two Parasponia genomic clones from a 12 kb BamH1 fraction. These differ slightly in their restriction pattern. We are currently analysing these two Parasponia Hb genes. Comparison of the structure of the non-legume Hb genes with Lb genes, especially the position of introns, will lead to a better understanding of the evolution of hemoglobin genes.

Frameshift mutagenesis in S.typhimurium.

by

Lyndal Ritchie.

Macquarie University/CSIRO Division of Molecular Biology.

Abstract.

Frameshift mutations in particular strains of S.typhimurium induced by 9-aminoacridine (9AA) are thought to occur via a single, novel repair pathway (1). In an attempt to identify genes in this pathway S.typhimurium hisC3076 uvr-304 was mutagenised with nitrosoguanidine and single colonies were screened selecting for mutants with zero and low mutagenesis in the presence of 9AA. Mutants detected in the initial screen were separated into several groups on the basis of responses in well tests. Besides zero and low responders to 9AA a high proportion of mutants were found to be sensitive to 9AA.

Representatives from the different classes of mutants have been characterised using the plate mutation assay in combination with survival assays. Further characterisation of the mutants has been included by looking at their mutagenic response to other chemicals and their sensitivity to methylmethanesulfonate (MMS) and 2-aminopurine (2AP). Since it is known that mutants deficient in DNA polymerase I are sensitive to MMS, while mutants deficient in DNA methylation are sensitive to 2AP, these tests may give further clues to the nature of the mutations. Mutants from the selected group were found to be sensitive to one, both and neither of these mutagens.

Assays are underway to confirm whether any of the mutants sensitive to MMS have mutations which affect DNA polymerase while mutants sensitive to 2AP will have methylation patterns examined using restriction enzyme analysis and methylation assays.

Reference.

1. D. M. Podger, G. W. Grigg and D. G. MacPhee. Mutation Research, 119 (1983) 113-120.

Identification of host specificity DNA regions determining the broad host range of Rhizobium strain NGR234. MURALI NAYUDU and BARRY G. ROLFE, Australian National University, Canberra, ACT 2601, Australia.

An in vivo genetic engineering technique using an "R68.45" like plasmid has been used to construct over 100 hybrid plasmids (R-prime plasmids) containing various segments of the NGR234 Sym plasmid. These R-primes carry approximately 180 kb to 330 kb of Sym plasmid DNA. Extensive analysis of one of these R-prime plasmids carrying approximately 330 kb of the 470 kb NGR234 Sym plasmid has shown that it contains all the genetic information required for the broad host range nodulation of the tropical legumes nodulated by the parent strain NGR234, including soybean. Furthermore, this construct is able to nodulate the non-legume Parasponia andersonii more efficiently than the parent NGR234. Analysis of these R-primes in Escherichia coli K-12 using Southern hybridization techniques has established linkage (spanning a maximum of 180 kb) between two copies of the structural genes for the nitrogenase enzyme complex present in this strain, a site involved in the fixation process and a region containing nodD already identified to be essential for nodulation. The differences in the nodulating ability of the different sized R-primes in the Sym plasmid deficient Rhizobium strain ANU265 has indicated that distinctive host specific regions (hsn) involved in Soybean and Parasponia symbiosis reside on the NGR234 Sym plasmid. In addition the R-primes retain the wide host range transfer property of their parent R plasmid so the expression of the NGR234 nodulation genes has been shown in other bacterial species.

Bill Sherwin, Department of Zoology, University of Melbourne, Parkville, VIC., 3052. present address: Arthur Rylah Institute for Environmental Research, 123 Brown Street, Heidelberg, VIC., 3084.

The detection of selection in wild populations

The detection of selection in wild populations poses considerable problems. Where incomplete familial data are not available, it may be possible to obtain samples of juveniles which have survived a stage of considerable mortality, and a sample of the adults which produced the juveniles. Under these circumstances, several authors have used the allelic frequencies in the adult sample to calculate expected genotypic frequencies in juveniles, for comparison with observed frequencies by Chi-square. However, this test does not involve the size of the adult sample; that is, it assumes that the sample was infinitely large, so the allelic frequencies have no variance. It is shown that the alpha errors in this testing procedure are significantly higher than expected; that is, selection is detected much more often than it should be. An alternative test is presented, which gives a frequency of alpha errors much closer to the significance level chosen for the test.

C.B. GILLIES

School of Biological Sciences
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University of Sydney, N.S.W. 2006

Meiotic chromosome pairing at zygotene in rye

A spreading technique was used to allow electron microscopic analysis of synaptonemal complex formation in whole nuclei of rye (Secale cereale) at zygotene stages of meiosis. The following features were characteristic of rye chromosome pairing:
(1) lateral elements decreased in length by about one third from the beginning to the end of zygotene;
(2) there is a bouquet at early zygotene, and pairing appears to be initiated near the clustered telomeres of bivalents;
(3) multiple sites of pairing initiation subsequently occur interstitially in each bivalent;
(4) initiation of new pairing sites and extension of existing synaptonemal complexes occur simultaneously;
(5) interlockings of lateral elements and synaptonemal complexes are common in early zygotene, where they may delay the completion of pairing, but such interlocks ultimately appear to be resolved before pachytene is reached.

POSTERS

P.W. Atkinson and M.J. Hynes (University of Melbourne)

Comparison of the 5' regulatory regions of two co-regulated genes in *Aspergillus nidulans*

J. Bonsing, A.F. Stewart and A.G. Mackinlay (University of New South Wales)

Intragenic domain duplication in an α-casein

* C. Collis and G.W. Grigg (CSIRO Molecular Biology, Sydney)

A radiation-sensitive mutant of *E. coli* is phleomycin-, bleomycin- and heat resistant

C.M. Corrick, A.P. Twomey and M.J. Hynes (University of Melbourne)

The physical structure of the acetamidase gene of *Aspergillus nidulans*

N. Dear (University of Adelaide)

The mitochondrial genome of *Sminthopsis crassicaudata*

A. Delves, A. Mathews, D.D. Day, B.J. Carroll and P.M. Gresshoff (Australian National University)

Root and shoot factors affecting nodulation in soybean mutants

* G.G. Foster, R.J. Mahon and T.L. Woodburn (CSIRO Entomology, Canberra)

Genetic control of *Lucilia cuprina* - report on the Shoalhaven Field Trial

* A.A. Hoffman and P.A. Parsons (La Trobe University)

Habitat marking: parallel genetic divergence in two *Drosophila* species

M. Jenkin and K.W. Shepherd (CSIRO Plant Industry, Canberra)

Aconitate hydratase isozymes in wheat-cultivar variation and use as a genetic marker

T.G. Littlejohn, M.A. Davis, J.M. Kelly, L.M. Corrick and M.J. Hynes (University of Melbourne)

Regulatory elements of the *amdS* gene of *Aspergillus nidulans*

* J. Patton, J. Camakaris and D.M. Danks (University of Melbourne)

Studies on copper-resistant Chinese Hamster ovary (CHO) cells

* C. Roberts, J. Brasch and M.H.N. Tattersall (Ludwig Institute for Cancer Research, Sydney)

Amplication of RNA genes: a selective advantage in tissue culture

J.W. Sentry and D.R. Smyth (Monash University

Characterization of the *del* sequence family dispersed through the genome of *Lilium henryi*

(*no abstract)

(continued over)

POSTERS (CONT'D)

B. Sherwin and P.R. Brown (Arthur Rylah Institute of Environmental Research, Heidelberg)

Problems in the estimation of the effective size of an endangered population of bandicoots, *Perameles gunni*

M. Westerman, C. Randell and R.C. Moore (La Trobe University)

DNA α-polymerase activity and aphidicolin inhibition as a function of cysteine concentration

S. Yuguang and J.S.F. Barker (University of New England)

Effects of a 2nd chromosome MR element on selection responses in *Drosophila melanogaster*

P.A. Zelesco and J.A.M. Graves (La Trobe University)

Investigations of the mitotic apparatus in mammalian hybrid cells

COMPARISON OF THE 5' REGULATORY REGIONS OF TWO CO-REGULATED GENES IN ASPERGILLUS NIDULANS

Peter Atkinson and Michael Hynes

Department of Genetics, University of Melbourne,
Parkville, Victoria, 3052

The amdS and aciA genes are co-regulated by the amdA regulatory gene of Aspergillus nidulans. A cis-dominant mutation amdI66 which maps to the 5' non-coding region of amdS increases the amdA-controlled expression of this gene. Analysis of the 5' regulatory regions of aciA, amdS and amdI66 revealed the presence of a purine-rich sequence located immediately upstream from the TATA box in each of them. The significance of this region of homology is discussed.

J. Bonsing, A.F. Stewart and A.G. Mackinlay.

School of Biochemistry, University of N.S.W., Kensington, N.S.W. 2033

INTRAGENIC DOMAIN DUPLICATION IN AN α-CASEIN.

The caseins comprise the major protein fraction of milks. They are unusual proteins, in that while being biologically important, there have been relatively few constraints imposed upon their structures. As a result, a large repertoire of functional homologues have evolved.

We have determined the complete nucleotide sequence of the bovine α_{s2}-casein mRNA, the structure of which is compared to its homologues in the guinea pig, mouse and rat. The organisation of these casein mRNA sequences is such that discrete blocks of sequence have been the subject of various rearrangements and duplications. The simplest explanation for this observation is that the blocks represent either existing or ancestral exons.

The most prominent sequence duplication occurs in both the bovine and cavine sequences, and comprises a large, tandem intragenic repeat of approximately 90 codons. The origin of this and other smaller duplications is probably unequal sister chromatid exchange.

THE PHYSICAL STRUCTURE OF THE ACETAMIDASE GENE OF *ASPERGILLUS NIDULANS*

C.M. Corrick, A.P. Twomey and M.J. Hynes

Department of Genetics, University of Melbourne

The acetamidase of *Aspergillus nidulans* enables the utilization of acetamide as a sole nitrogen or carbon source. The *amdS* gene is subject to complex multiple control mechanisms which are independent of one another. These regulatory gene products operate at a control region adjacent to the *amdS* structural gene.

The extent of the control region has been described by genetic and physical mapping of various strains containing control region mutations.

Transcriptional mapping has shown the *amdS* gene to have two small introns. Northern blot analysis shows an mRNA size of 1.6 - 1.7 kb.

The DNA sequence of the *amdS* genomic clone has been determined and the putative promoter and intron splice junctions located. The control regions and first exons of several mutants have been sequenced, including *cis*-acting regulatory mutants.

1. Neil Dear

2. Department of Genetics, University of Adelaide, Adelaide, South Australia 5001.

3. Title

The mitochondrial genome of *Sminthopsis crassicaudata*.

4. Abstract

A laboratory colony of the dasyurid marsupial *Sminthopsis crassicaudata*, maintained by the Genetics Department, University of Adelaide, has facilitated a study of the structure and variability of the mitochondrial genome of this organism.

The mitochondrial genome is a circular molecule of 16.7 ± 1 kb in length. Restriction endonuclease analysis of mitochondrial DNA revealed a very low level of individual variability in the colony, much lower than has been reported for natural populations of other organisms. This may be due, in part, to the small number of wild-caught animals from which the colony has descended.

In the past, mitochondrial DNA has been assumed to be homoplasmic within an individual and solely maternally inherited. Preliminary evidence suggests that this may not be strictly true in *S. crassicaudata*.

ABSTRACT

Root and Shoot Factors Affecting Nodulation in Soybean Mutants.

Angela C. Delves, Anne Mathews, David D. Day, Bernard J. Carroll, Peter M. Gresshoff.

Botany Department, Australian National University, Canberra. A.C.T., 2601, Australia.

A number of mutants which supernodulate and are tolerant to nitrate (nitrate tolerant symbiosis - nts) have been isolated from soybean cultivar Bragg (1). Two nts lines, 382 and 1116 (a marginal supernodulating mutant) have been used to investigate the regulation of nodulation.

In soybeans, a non-nodulating condition controlled by a single recessive gene (rj_1,rj_1) was reported by Williams and Lynch (2). Three non-nodulating mutants (nod49, nod139 and nod772) were isolated from mutagenized soybean populations in the presence of Rhizobium japonicum strain CB 1809 (=USDA 136) (3). The non-nodulating mutant, nod49 has been characterized in more detail.

Experiments in which shoots from nts382 and 1116 were grafted onto Bragg and Williams root stocks, and vice versa, have shown that both supernodulation and nitrate tolerance are determined by shoot factor(s). Bragg roots grafted with nts shoots had many more nodules than wildtype controls and these occurred even in the presence of applied nitrate. Nts roots grafted with Bragg shoots had fewer nodules, which were restricted to the upper portion of the root, than nts controls. The same results occurred to a lesser extent with nts1116, confirming its marginal nature. Grafts using the cultivar Williams confirmed that the supernodulating character was not cultivar specific as results did not differ significantly from those using Bragg.

Tanner and Anderson (4) observed that non-nodulation is controlled in the roots of non-nodulating (rj_1,rj_1) plants of soybeans. Grafting experiments using the parent cultivar Bragg and the non-nodulating mutant nod49 indicated that the non-nodulation trait was controlled by the genotype of the root tissue.

References

(1) Carroll, B.J., McNeil, D.L., and Gresshoff, P.M. (1985). Plant Physiol. 78: 34-40.

(2) Williams, L.J., and Lynch, D.L. (1954). Agron. J., 46: 28-29.

(3) Carroll, B.J., McNeil, D.L., and Gresshoff, P.M. (1985) Submitted to Plant Science.

(4) Tanner, J.W., and Anderson, I.C., (1963). Can. J. Plant Sci., 43: 542-546.

ACONITATE HYDRATASE ISOZYMES IN WHEAT-CULTIVAR VATIATION AND USE AS A GENETIC MARKER

(Poster)

M. JENKIN* AND K.W. SHEPHERD#

(*CSIRO, Division of Plant Industry, BLACK MOUNTAIN, ACT and #Waite Agricultural Research Institute, GLEN OSMOND, SA)

Hart (1983) has shown that aconitate hydratase isozymes are monomeric isozymes controlled by group 6 homoeologues of Chinese Spring wheat and some related species. In a survey of 77 world wheats we identified 9 different zymogram patterns involving 7 different band positions. Thus this character shows some scope for cultivar identification. Most variation was observed in the two slower, more cathodal bands. We have shown that an *Agropyron* aconitate hydratase band is present in wheats containing the stem rust gene *SR26* from *Agropyron elongatum*. This isozyme band has proved to be a valuable marker characteristic for identifying the presence of this resistance gene in some wheat backgrounds and it has been used in genetic studies aimed at recombining the *Agropyron* segment with wheat chromosomes.

REF: Hart, G.E. Hexaploid wheat (*Triticum aestivum* L. em Thell). (p. 35-54) in "Isozymes in Plant Genetics and Breeding" Tanksley/Orton eds., Elsevier Scientific (Amsterdam) 1983.

"REGULATORY ELEMENTS OF THE *amd*S GENE OF ASPERGILLUS NIDULANS"

T.G. Littlejohn, M.A. Davis, J.M. Kelly, L.M. Corrick and M.J. Hynes
(University of Melbourne)

Aspergillus nidulans is a filamentous fungus which can utilise acetamide as a sole carbon and nitrogen source through the acetamidase enzyme encoded by the *amd*S gene. At least five independently acting regulatory pathways control the level of *amd*S expression via known effectors and regulatory genes. *amd*S has been cloned and sequencing of the controlling (5') region of the gene and *cis* regulatory mutants has exposed the targets at which regulatory gene products exert their effect on *amd*S expression. These mutants are, however, very rare and so constitute a biased representation of sequences required for regulation of *amd*S. Further, they give little indication of the mode of action of regulatory gene product - DNA interactions.

To determine the precise DNA sequences required for regulation, and the manner in which they operate, two approaches will be presented. They are, the use of DNA mediated transformation and *in vitro* mutagenenis to :-

(i) identify the 5' sequences which cause, when found in multi-copy form, titration of regulatory gene products (*fac*B and *amd*R) and hence represent their most probable site of action.

(ii) manipulate 5' sequences in their orientation, distance from the startpoint of transcription and their sequence composition and to their assay *in vivo* the effects of such mutations on regulation.

Characterization of the *del* Sequence Family Dispersed Through the Genome of *Lilium henryi*

John W. Sentry and D. R. Smyth

Department of Genetics, Monash University

The true lilies have genomes among the largest known. For example, an unreplicated haploid nucleus of *Lilium henryi* contains 32 million kbp of DNA, ten times that usually found in mammals. To see what types of DNA may be present in this huge genome, we have begun characterizing a large (8 kbp), dispersed, and abundantly repeated sequence -- the *del* family.

The sequence usually has three *Bam* HI sites, and we have cloned a sample of the two internal *Bam* subregions into pBR322. Twelve clones (pLh200 series) corresponding to the 2 kbp *Bam* HI subregion and ten clones (pLh500 series) of the 5 kbp *Bam* HI subregion have been characterized. *In situ* hybridization showed the repeats to be dispersed throughout lily chromosomes. Restriction mapping of the cloned subregions shows family members to be somewhat variable, with consensus sites present on average on 80-90% of occasions.

To investigate the genomic organization of the *del* family we prepared a library of *Lilium henryi* DNA. The lily DNA was partially digested with *Mbo* I into 15-20 kbp fragments and cloned into the lambda phage replacement vector EMBL3. Around 4% of plaques were positive when screened with previously cloned *Bam* HI subregions, reflecting an abundance of about 50,000 copies per genome. The 8 kbp *del* elements are bound by 1 kbp direct repeats suggesting transposability. The terminal direct repeat is occasionally found in isolation, and one clone contains tandem copies of the *del* element.

Bill Sherwin and Peter R. Brown, Arthur Rylah Intsitute for Environmental Research, 123 Brown St., Heidelberg, VIC., 3084.

Problems in the estimation of the effective size of an endangered population of bandicoots, Perameles gunnii.

Since the 1940s, the mainland population of P.gunnii has been declining. It is now restricted to the city of Hamilton and its outskirts,in Western Victoria. The census size of the population is 1135 or lower. Dispersal of more than 1 km is known to occur, but detailed dispersal data are not available. On the assumption that the whole population represents a single neighbourhood, available demographic data have been used to calculate effective population size, which appears to be 200 or lower. The Hamilton population may have been at this size for ten years or longer. It is calculated that a detectable loss of genetic variability may have ocurred in this period. Electrophoretic investigation of variability has commenced, using the numerous Tasmanian population as a reference. This work will also address other conservation genetic questions: subdivision of the Hamilton population, and taxonomic relationship to the Tasmanian population. Genetic methods for estimating Ne are not suitable for endangered populations. Radio-telemetry studies will commence soon.

M. Westerman*, Chris Randell+ and Ruth C. Moore+

*La Trobe University, +Cancer Institute, Melbourne

DNA α-polymerase activity and aphidicolin inhibition as a function of cysteine concentration.

DNA α-polmerase in the cell line JU56 has been found to be an -SH containing, -SH dependent enzyme, as it is in other cell types. The activity of the enzyme and the inhibitory effect of aphidicolin, as a function of -SH concentration, is described.

Effects of a 2nd Chromosome MR Element on Selection Responses in *Drosophilia Melanogaster*

Shi Yuguang and **J. S. F. Barker**
Department of Animal Science, The University of New England

MR elements of *Drosophilia melanogaster* are responsible for male recombination as well as increasing mutation. The current experiments using an MR-h12 stock (MR on chromosome 2 between *Tft*,53.2 and *pr*,54.5) examined whether the anticipated extra mutation could alter selection responses for high and low numbers of sternopleural bristles. The mutagenic activity depends on patroclinous inheritance of MR and on the female parent cytotype, this allowing control over mutation by manipulating parent matings.

A first experiment compared seven generations of selection for high and low bristle number in two replicated lines. In both lines MR was active for two generations before beginning selection; the activity of MR was then continued through selection in one (patroclinous) line but discontinued in the other (maternal) line. The realized heritabilities for bristle number were slightly (but not statistically significantly) higher in the patroclinous line.

A second experiment examined high and low selection over five generations in three replicated lines two of which were similar to experiment 1 and a third line where MR was inactive prior, as well as during, selection. Despite the same trends as in experiment 1, the differences in realized heritabilities were not statistically significant.

Possible reasons why MR activity fails to increase selection responses will be discussed.

INVESTIGATIONS OF THE MITOTIC APPARATUS IN MAMMALIAN HYBRID CELLS.

P.A. Zelesco and J.A.M. Graves
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La Trobe University
Bundoora, Victoria

Abstract

Gene mapping studies rely on interspecific somatic cell hybrids preferentially losing chromosomes of one parent. The mechanism of chromosome segregation remains unknown. One suggested theory is that segregant chromosomes fail to engage in functional attachments with the mitotic apparatus. Microtubules are the major protein components of mitotic and meiotic spindles. They are composed principally of dimeric subunits of one α- and one β - tubulin polypeptide.

We hypothesized that tubulin gene repression in hybrid cells determines the direction of chromosome segregation. Two predictions that follow from this are:

1. Tubulin genes of one species will be repressed in hybrid cells;

2. Tubulin gene repression will be concordant with the direction of chromosome segregation in hybrids.

A colcemid-resistant Chinese hamster cell line (CHO, Strain 10193) with a mutant β-tubulin marker was fused with a wild-type mouse line (LTA). We found all hybrids to be colcemid-sensitive. This reflects an absence of mutant hamster tubulin. Since loss of colcemid resistance is independent of loss of particular hamster chromsome(s), we conclude that hamster genes for the mutant β-tubulin are repressed in hamster-mouse hybrids.

Some of these hybrids lost predominantly mouse and some predominantly hamster chromosomes. Therefore, we conclude that the direction of chromosome segregation is not concordant with the repression of hamster mutant β-tubulin genes.

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POSITIONS FOR GENETICISTS:

Department of Genetics,

University of Melbourne

Summer Research Studentship - available for work on a defined project in the Department between November and February. Applications from interstate final year students are welcomed. Enquiries to:

Professor Michael Hynes,
Chairman,
Department of Genetics,
University of Melbourne,
Grattan Street,
PARKVILLE VIC. 3052

Telephone: 3446246

School of Biological Sciences

Macquarie University

Research (Postdoctoral) Fellow

The project concerns the genetic control of hypertension in pregnant women. The appointee will be required to use DNA probes as part of an attempt to detect linkage between marker genes and genes for pregnancy hypertension. A PhD and experience in molecular biology is essential, preferably with recombinant DNA techniques. Some background in general genetics would also be an advantage. Further information from:

Professor D. W. Cooper,

(02) 88 9483.