Single nucleotide polymorphism(SNP) mapping of mutant 2.1 in *Caenorhabditis elegans*

Shivsha Rajkumar, Cristina Slatculescu and Dr. Antonio Colavita

Department of Cellular and Molecular Medicine
University of Ottawa

Introduction

Caenorhabditis elegans is a powerful model organism that can be used for neurobiological research. Study of the DD motor neurons of the nematode has displayed that mutation 2.1(prk1-1 mutation of prickle protein) in *C.elegans* causes the migration of the motor neurons closer to the anterior than the wildtype worm and a reduced distance between motor neurons DD1 and DD2. Single nucleotide polymorphism(SNP) markers can easily be utilized to pinpoint the specific chromosomal location of mutant 2.1 as they are extremely dense and usually have no associated phenotype. The study of altered neuronal migration such as those resulting from mutant 2.1 can help one develop new treatments to establish neural repair for many neurodegenerative diseases.

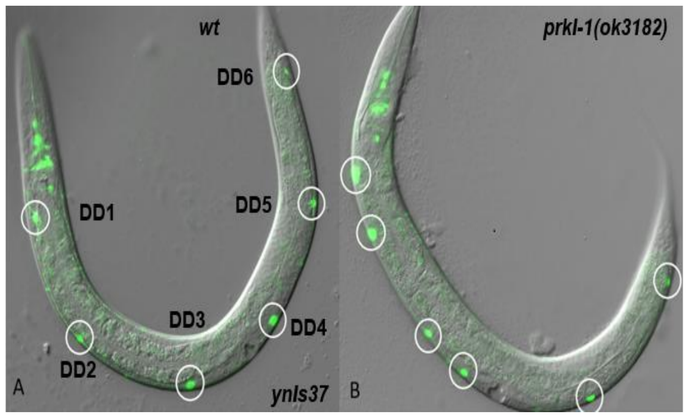

Figure 1: Image of *C.elegans* mutant 2.1 with DD motor neurons translocated to a more anterior region (image obtained from graduate student of Dr. Antonio Colavita).

Hypothesis

1) Mutant 2.1(prk1-1 mutation) is located on Chromosome 2 (LG 11) between SNPs F39E9 to F15D4 corresponding to -7.8 and 13.40 mbp respectively.

2) The closer the mutant 2.1 lies to the given SNPs being tested, the less likely the homozygous mutant will contain a CB4856 loci of that polymorphism. Thus, an over representation of the N2 homozygous allele among mutant worms will be observed due to the decreased likelihood of recombination.

Objective

Determine the chromosomal location of mutant 2.1 (mutation of prickle protein) responsible for the translocation of motor DD neurons of *C. elegans* via SNP-mapping.

Methods

Chromosomal mapping was performed to identify mutant 2.1 to a broad general region on chromosome 2(LG11). A technique known as interval mapping was utilized to narrow the known interval to a smaller region via PCR and restriction enzyme digest.

Recombinant worms were genotyped via Polymerase Chain Reaction(PCR) for SNPs(F39E9, F54D10 and F15D4) located in the region in which mutant 2.1 was mapped.

Restriction Enzyme Dra1 was utilized to digest DNA. Gel electrophoresis was performed using the digested DNA products.

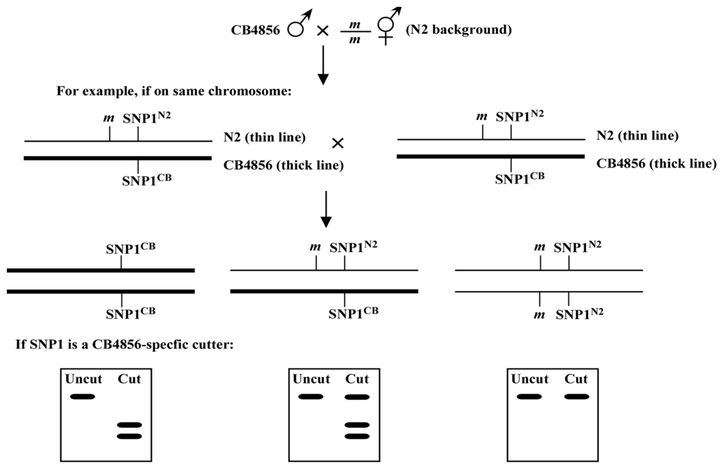

Figure 2: Basic outline for two-point SNP mapping. Hawaiian and English strains or isolates are represented as CB4856 and N2 respectively.

Results

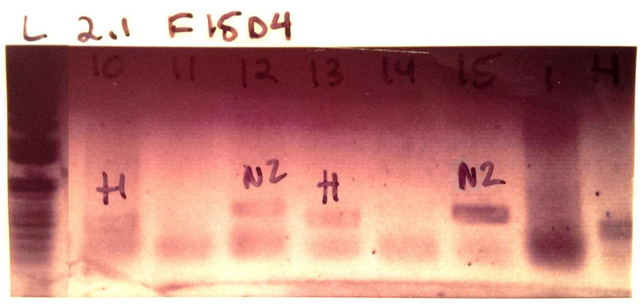

Figure 3: Gel Electrophoresis results illustrating recombinant DNA products using SNP F15D4 marker (CB4856 and homozygous N2 loci observed). 1kb DNA ladder and controls were utilised.

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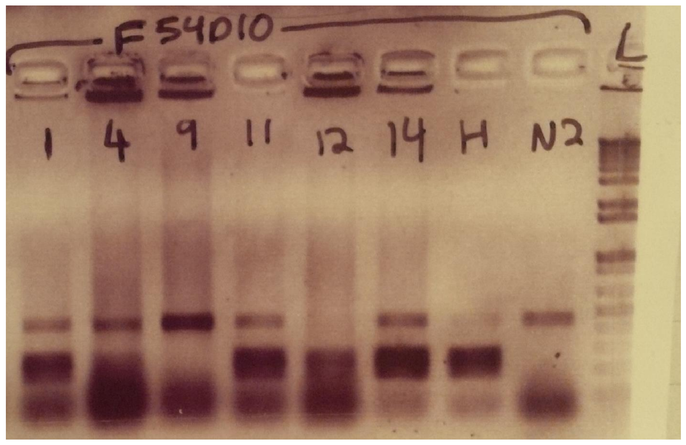

Figure 4: Gel Electrophoresis results illustrating recombinant DNA products using SNP F54D10 marker (CB4856 , homozygous N2 and CB4856/N2 loci observed). DNA 1kb ladder and controls were utilised.

	2.1	F39E9	F54D10		F15D4	SNP
		3.3	3.8		13.2	position on physical map
	LGII	-7.8	-5.27		13.40	position on genetic map
	recombinant					
	2	N			N	
	4	H	H		N	
	5	N	N		H	
	6	H	HH		N	
	7	N	N		H	
	8	H	N		N	
	9	N	N		HH	
	12	HH	HH		N	
	14	HH	H		N	
	15	N			N	

Figure 5: Final results depicting that mutant 2.1 is located between SNPs F54D10 and F15D4 corresponding to -5.27mbp and 13.40mbp of the genetic map respectively. If the mutant lies closer to the SNP being studied one will observe an overexpression of the N2 homozygous loci. N, H, and HH correspond to the expression of homozygous N2 , CB4856/N2 and CB4856 loci of the recombinant s(2-15) respectively.

Conclusion

Mutant 2.1 was located between SNPs F54D10 and F15D4 corresponding to -5.27mbp and 13.40mbp of the genetic map respectively. One could further pinpoint the exact location of the mutant using SNPs between F54D10 and F15D4. Future work includes identifying and characterizing the particular gene that is disrupted in the 2.1 mutant strain using molecular cloning techniques.

References

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3) Wicks, Stephen R., et al. "Rapid gene mapping in Caenorhabditis elegans using a high density polymorphism map." Nature genetics 28.2 (2001): 160-164.