Snip-SNP mapping of *eq4*, a genetic suppressor of the *gtl-2* mutant phenotype in *Caenorhabditis elegans*

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Magnesium homeostasis

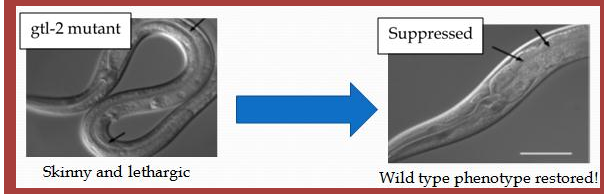
- Important cation involved in metabolic and enzymatic reactions.
- Abnormal level can lead to cardiac and neurologic disorders.
- Homeostasis maintained by TRPM6 and TRPM7 channels of Transient Receptor Potential (TRP) family.

- *C. elegans* possess 23 TRP channels, most of which have human homologs. To date, three TRPM orthologs have been identified:

1. *gon-2* and *gtl-1*: involved in Mg^{2+} uptake.
2. *gtl-2* (less studied): seems to be involved in excretion of Mg^{2+}.

- *gtl-2* mutants are lethargic, skinny and develop slowly. The mutant phenotype can be rescued by growth on a media lacking Mg^{2+}, indicating that *gtl-2* is involved in excretion of Mg^{2+}.

- Chen Lab identified three suppressors of *gtl-2* mutant phenotype. These suppressors are second site mutations that help restore wild-type phenotype in mutants.



Objective

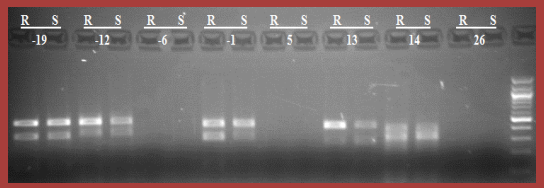
To map *eq4*, which is a genetic suppressor for *gtl-2* mutant phenotype by using Snip-SNP mapping. Single nucleotide polymorphisms between *C. elegans* Bristol strain and Hawaiin strain are used as genetic markers.

Methods

1. Hawaiian SNPs were crossed into *gtl-2; sup (eq4)* Bristol mutants.
2. Wild type progeny were picked and allowed to produce self-progeny.
3. Suppressed and non-suppressed progeny were isolated.
4. The two sets of progeny were lyzed for Snip-SNP mapping.
5. The chromosome containing the suppressor was identified:
   - PCR-> Dra1 digest-> Genome-wide scanning to locate linkage of *eq4.*

Results

(a) Chromosome I



(b) Chromosome V

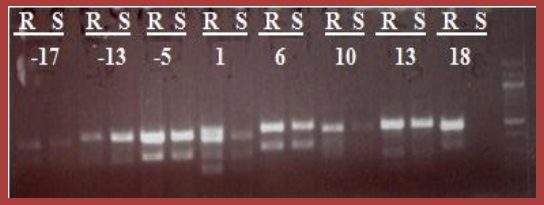


Figure 1. **Snip-*SNP mapping eq4* .** *Chromosome mapping.* Each pair of lanes shows results from SNPs at the indicated genetic map position of chromosome I and V, using either suppressed (R) or unsuppressed (S) DNA template.

Discussions

eq4 is on chromosome I of *C. elegans* genome. Linkage is visible as an increase in proportion of Bristol DNA in suppressed lanes (R) of chromosome I from -12 to 13 (Figure 1a). No linkage is visible on other chromosomes (Figure 1b).

eq4 is not a second site mutation in *gon-2* (also located on chromosome I) because of the absence of *gon-2* mutant phenotype (improper gonad development) in suppressed worms.

Future work

- Finely map and clone *eq4* to understand the maintenance of Mg^{2+} homeostasis in C. *elegans.*

- If *eq4* is conserved in humans, apply the knowledge to develop treatments of various human diseases related to magnesium level imbalance.

Literature

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