Screening of specific genes on U chromosome of *Saccharina japonicab* by comparative transcriptomics

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Abstract

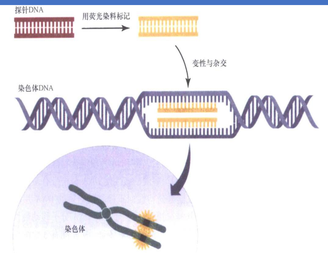
Saccharina japonica is a large perennial edible economic algae whose life cycle alternates between diploid and haploid stages. In organisms with haploid stage sex-determination systems, the chromosomes containing sex-determination region (SDR) are known as U and V sex chromosomes. In this experiment, we used U chromosome combined genome, transcriptome, bioinformatics analysis and other techniques to screen sex-specific genes located on the U chromosome of *S. japonica* , and then verified the final results according to the female sex-determining gene (SJ-13_000170) that had been screened. It lays a foundation for the characterization of the assumed UV sex chromosome SDR region of *S. japonica*, and also provides a certain basis for the drawing of pseudoautosomal region (PAR) of *S. japonica* with Chorus2 software.

Background

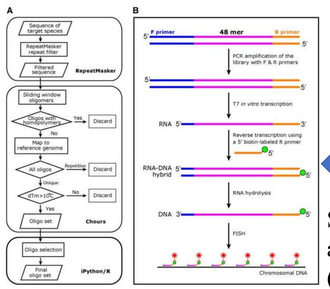
So far, most studies have focused on the diploid sex determination system (classical XY or ZW), while few studies have focused on the haploid sex determination system (Bull J J., 1984). SDR is a region closely linked with gender. The SDR region of the model organism long sac water cloud in brown algae has been preliminarily defined (Ahmed *et al*., 2014), but the SDR of *S. japonica* remains to be further explored. Recently, Du (Du *et al*., 2022) developed a female-linked (SJ-f_000170) molecular marker based on the sex-determining region (SDR) of the genus *Ectocarpus* sp., which can be used to validate our sex-specific genes. Therefore, transcriptomics was used to screen sex-specific genes to provide a basis for the characterization of SDR region in *S. japonica*. Meanwhile, the Chorus2 software developed by Han (Han *et al*., 2015) also provided a basis for the painting of U chromosome PAR in *S. japonica*.

Materials and methods

Culture male, female gametophyte and sporophyte of *S. japonica*.

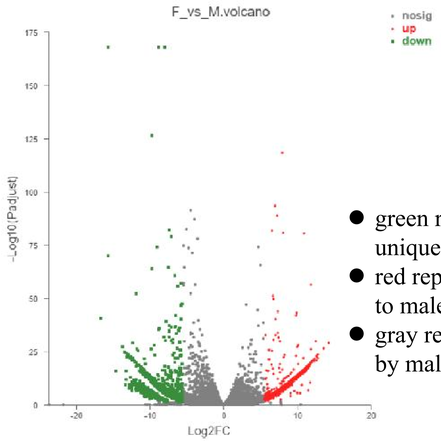


Use FISH technique to distinguish the putative U and V chromosomes of *S. japonica* (Jocelyn 《基因X》)



Specific oligo probe design (A) and amplification and labeling diagram (B)(Han et al., 2015)

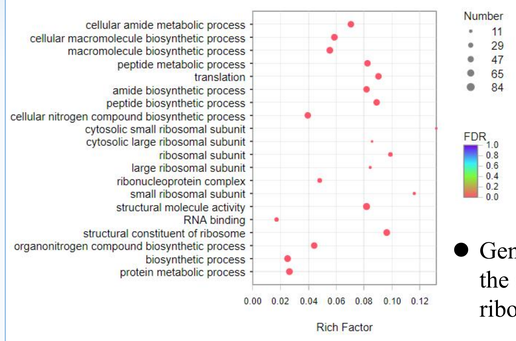
Results and discussions



Volcanic map of transcriptome expression differences

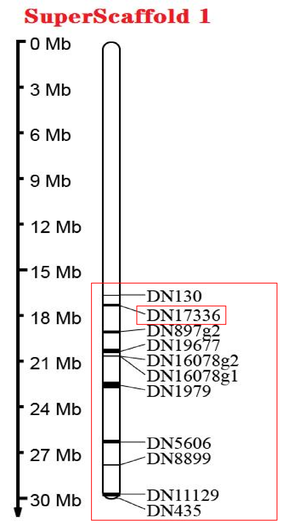
- green represents the transcriptome unique to female gametophytes
- red represents the transcriptome unique to male gametophytes
- gray represents the transcriptome shared by male and female gametophytes

GO enrichment analysis(F_specific)



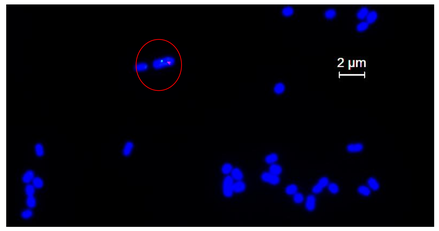
Proposed GO cluster analysis of female sex-specific genes

- Gene function is enriched in the structure and function of ribosomes



- There were 11 female sex-specific genes, and one specific gene DN17336 was verified by FISH

Distribution of female sex-specific genes in the genome



Location of female linkage marker SJ-13_000170 (red) and DN17336 (green) probe on metaphase chromosome of female gametophyte in *S. japonica*.

Conclusions

In summary, the SDR region accounted for roughly 15 million bp of SuperScaffold1 and the 18 million bp of SuperScaffold1 is likely to be PAR territory. After comparison between SuperScaffold1 on U chromosome and chr2 on V chromosome, it was obtained that the approximate PAR area of chr2 was about 8 million bp. Probe using the Chorus2 software (https://github.com/zhangtaolab/Chorus2), end up with 29000 oligos. The determination of PAR regions of U and V chromosomes in *S. japonica* needs further study.