Molecular evolution of *restorer-of-fertility*:

the case of sugar beet *Rf1*
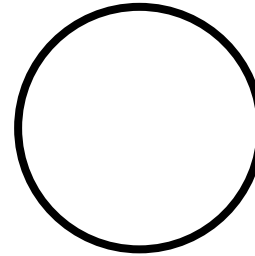
N mito DNA
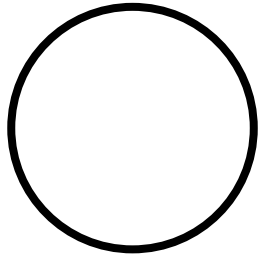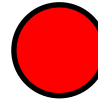
S mito DNA

Genome rearrangement

S-mito unique protein (preSATP6)

Sugar beet *Rf1*

Oligomer form
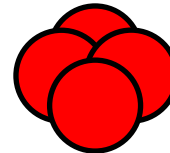
Male sterility

How has sugar beet *Rf1* evolved?

Sugar beet *Rf1* encodes an *Oma1*-like protein.

NH$_2$ — Mt signal — TM — TM — TM — COOH

Supplementary information of *Oma1*

Oma1 is a ubiquitous gene in eukaryotes. It plays multiple roles such as;

Quality control of mitochondrial inner membrane proteins (yeast).

Inhibiting mitochondrial fusion via OPA1 cleavage (mammal).

KO mutant is viable but exhibits several abnormalities (Arabidopsis).

Oma1 is single copy in most of eukaryotes.

Possible evolutionary relationship between sugar beet *Rf1* and *Oma1*

1. Sugar beet *Rf1* is the genuine *Oma1*.

Arabidopsis
Oma1
Orthologous
Sugar beet
Rf1
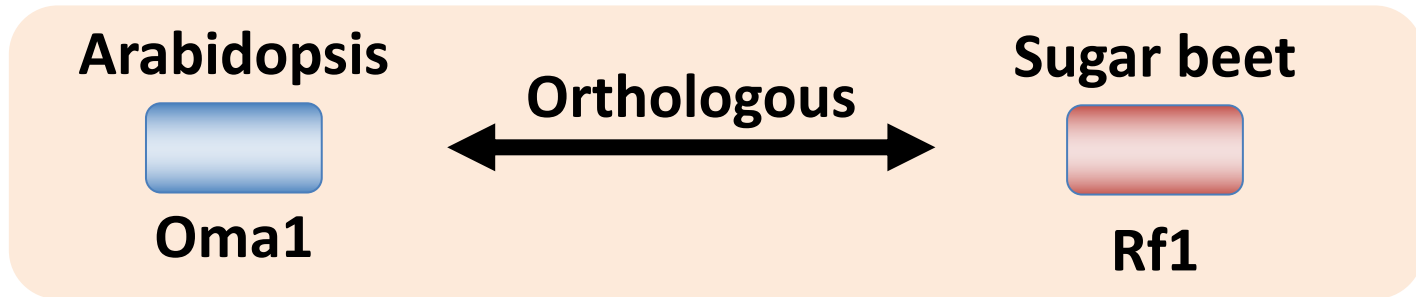
2. Sugar beet *Rf1* is a derivative of *Oma1*.

Arabidopsis
Oma1
Paralogous
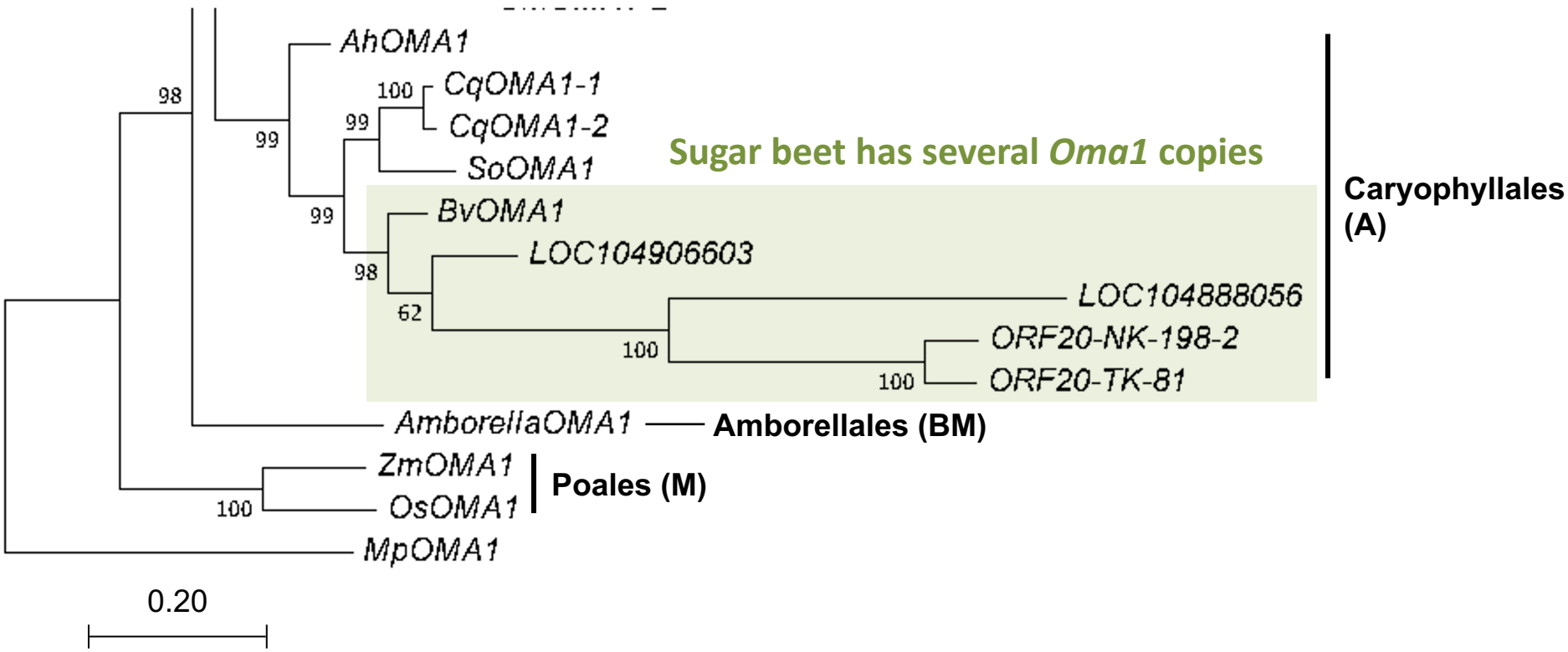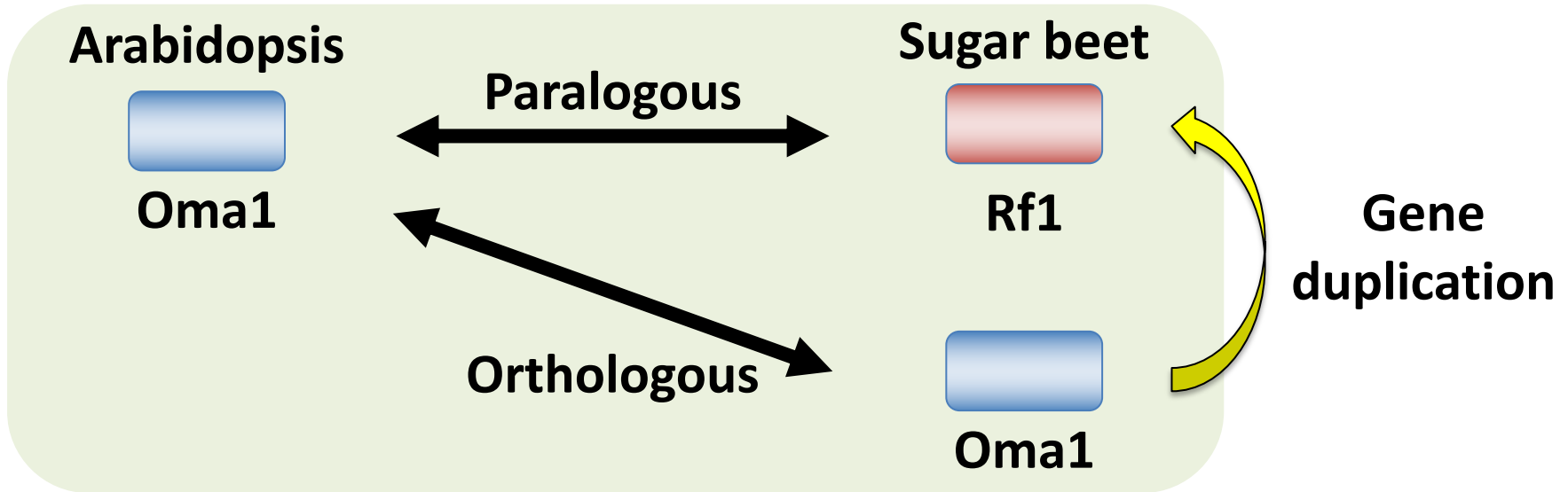Sugar beet
Rf1
Orthologous
Oma1
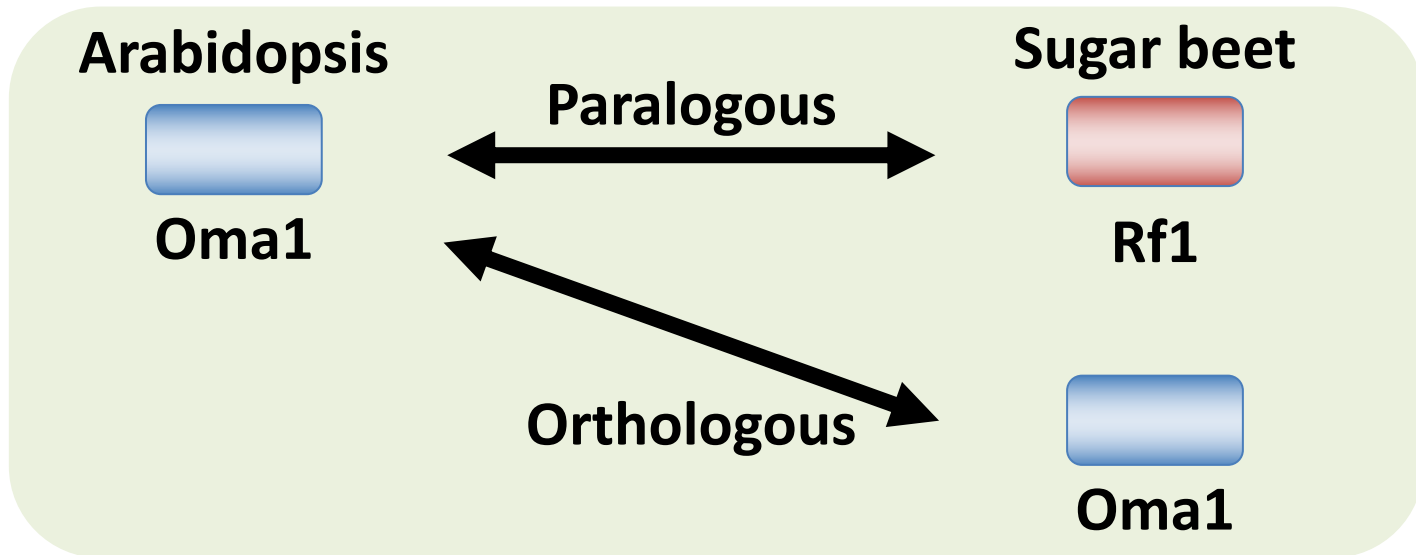
Sugar beet *Rf1* is a paralogue of *Oma1*

## Sugar beet genome is examined to find *Oma1* homologues.

In contrast to Arabidopsis, sugar beet has several copies of duplicated *Oma1* as lineage specific paralogues.

Evolutionary model of sugar beet *Rf1*

Considering Arabidopsis *Oma1* retains ancestral features, are Arabidopsis *Oma1*, sugar beet *Rf1* and sugar beet *Oma1* functionally equivalent to each other?

Neither Arabidopsis OMA1 or sugar beet OMA1 binds to preSATP6 protein.
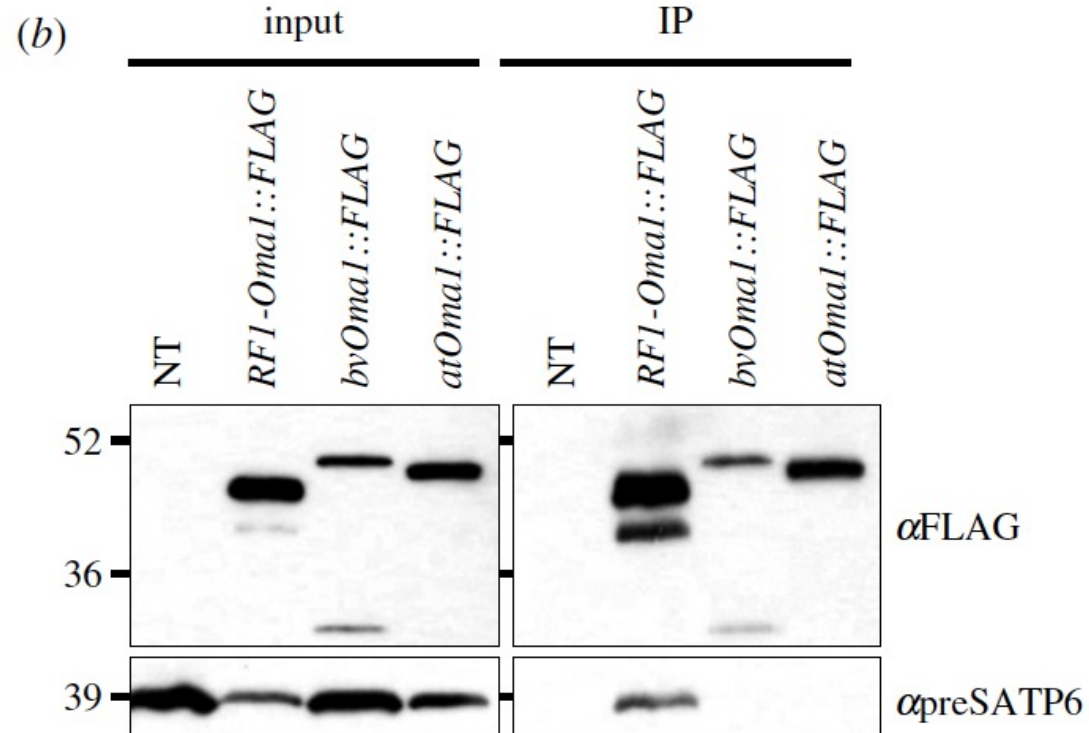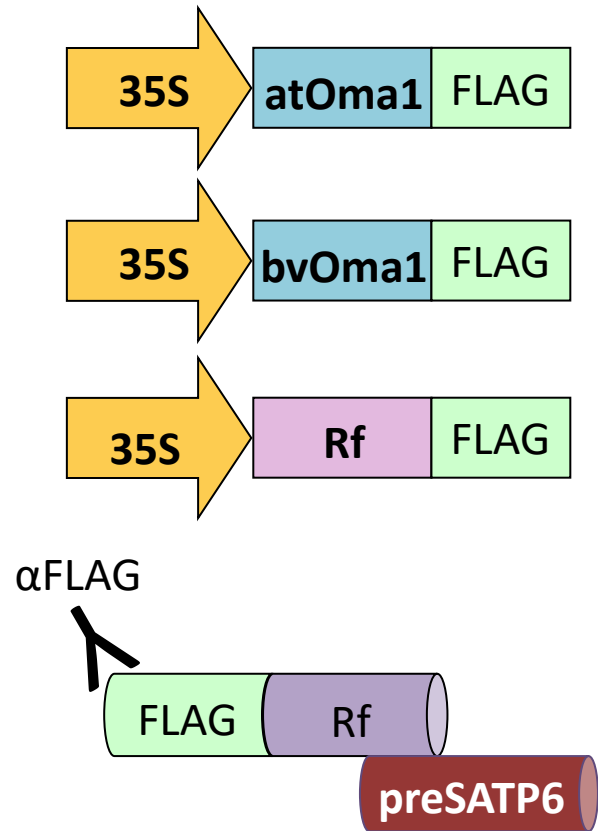
OMA1 is inherently unable to bind to preSATP6 protein.

The binding activity may have evolved after the gene duplication.

Arakawa et al. R Soc Open Sci 2019

	Meiosis	Tetrad	Micro spore
Arabidopsis Oma1			
Sugar beet Oma1			
Rf1			
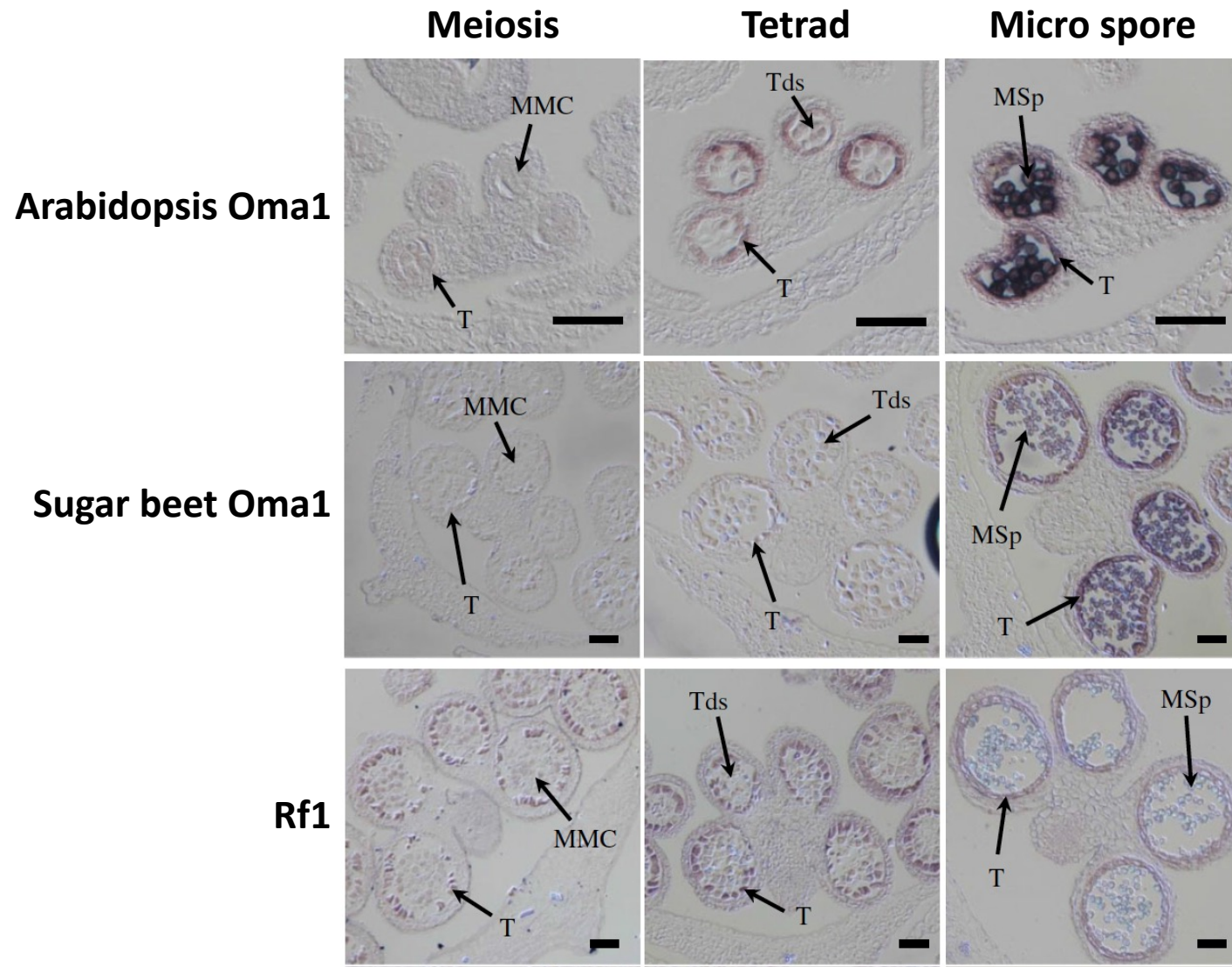
Gene expression pattern is different among Arabidopsis Oma1, sugar beet Oma1 and Rf1.

Difference in gene expression pattern between three *Oma1*-homologues

		Meiosis	Tetrad	Microspore
atOma1	Tapetum		▓	▓
atOma1	Meiocyte		▓	▓
bvOma1	Tapetum			▓
bvOma1	Meiocyte			▓
Rf1	Tapetum	▓	▓	
Rf1	Meiocyte	▓	░	

1. Rf1 acquired meiosis expression.

2. bvOma1 and Rf1 alternately expresses to complement atOma1 expression pattern.

⬇

Evolution of expression pattern after gene duplication is suggested.

Kubo et al. Plants 2020

Evolution of sugar beet *Rf1* may be very complex but all have happened recently.

Ancestral *Oma1* may resemble Arabidopsis*Oma1*

Regulatory element — Gene coding region
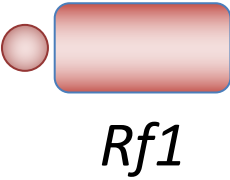
1. Gene duplication

2. Alteration of regulatory element
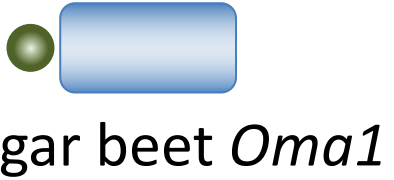
3. Alteration of regulatory element
4. Alteration of coding region

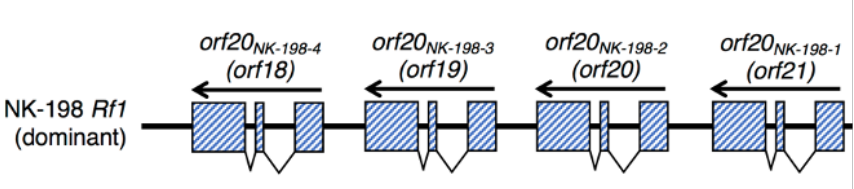Sugar beet *Oma1*   *Rf1*

Rf1 evolution may be ongoing
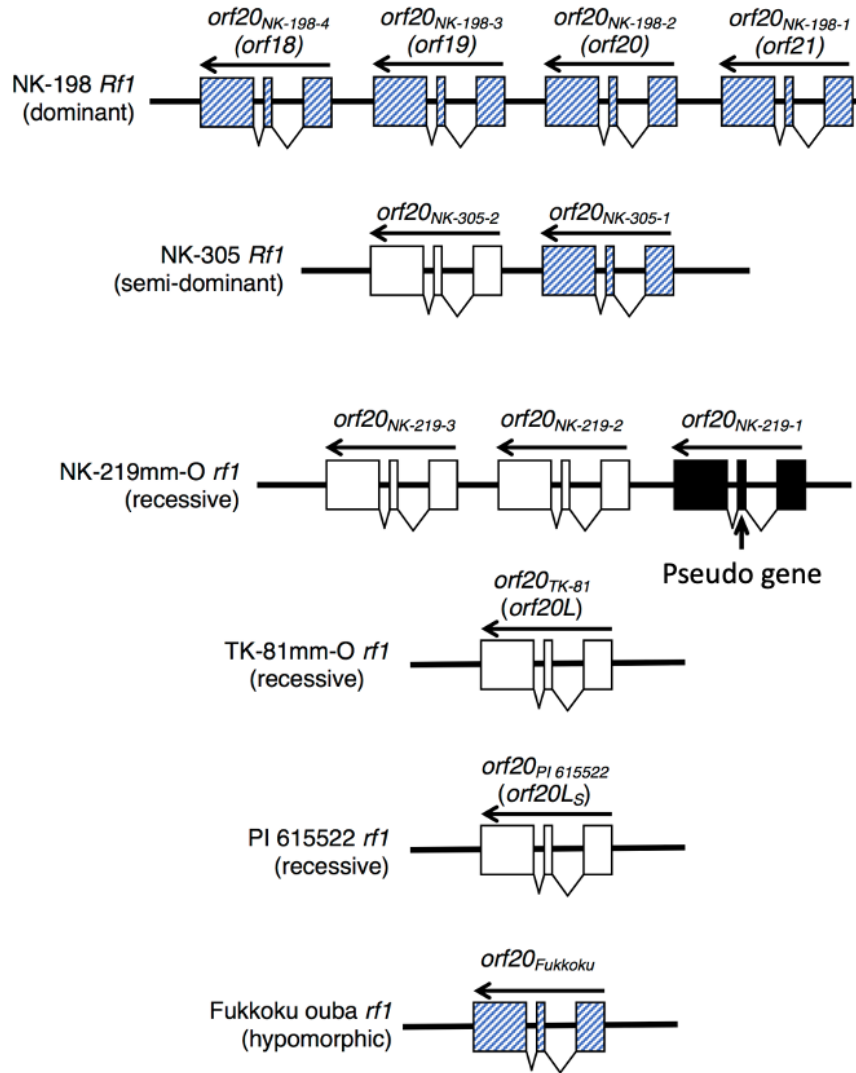
Copy number variation (CNV) of *Rf1* locus in cultivated beet.
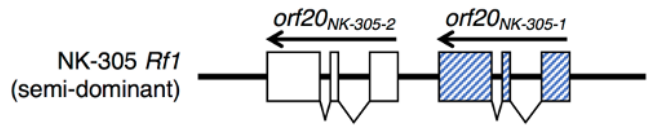
Rf1 evolution may be ongoing

Genetic action in field

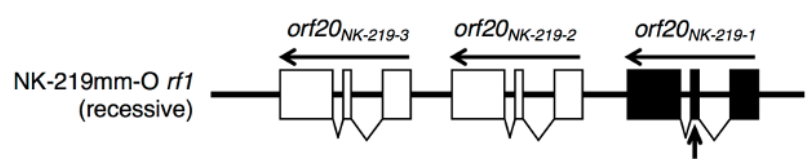All copies have potential to bind preSATP6

Strong restoration

Single copy has potential to bind preSATP6

Weak

No copy has potential to bind preSATP6

No

Rf1 is a complex locus whose genetic action is determined by its constituent copies.

1. Sugar beet *Rf1* is an evolutionarily young gene.

2. Evolution of sugar beet *Rf1* is a complex process that includes many steps such as gene duplication, alterations of coding- and regularory elements, and alteration(s) in the original copy.

3. Sugar beet *Rf1* likely continues its evolution to form functionally different alleles (haplotypes).

4. Evolution of *Rf1* locus and resistance (*R*) gene locus shares some features, such as CNV and positive selection.

References

Arakawa T, et al. How did a duplicated gene copy evolve into a restorer-of-fertility gene in a plant? The case of *Oma1*, **Royal Society Open Science**, 6: 190853, 2019.

Arakawa T, et al. A lineage-specific paralogue of *Oma1* evolved into a gene family from which a suppressor of male sterility-inducing mitochondria emerged in plants. **Genome Biology and Evolution**, 12:2314-2327, 2020.

Arakawa T, et al. The molecular basis for allelic differences suggests *Restorer-of-fertility 1* is a complex locus in sugar beet (*Beta vulgaris* L.). **BMC Plant Biology**, 20:503, 2020.