

Horizontal gene transfer between the parasitic plant *Cynomorium songaricum* and its host *Nitraria tangutorum*

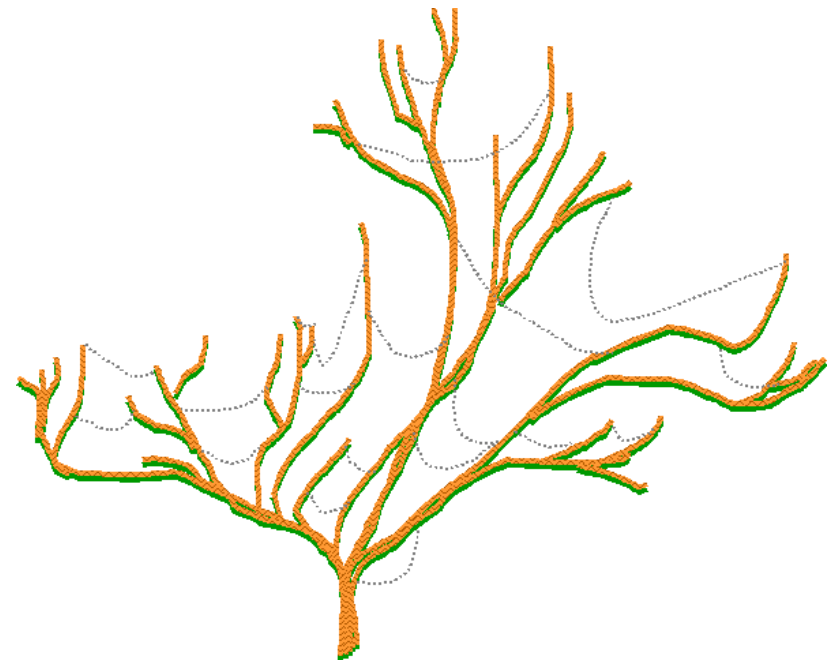


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Horizontal gene transfer (HGT) is the transfer of genes between non-mating species; parasitic relationship may facilitate HGT between flowering plants, but mechanistic explanations for HGTs have remained speculative. In this research, we report a discovery of *atp1* gene HGT from host *Nitraria tangutorum* to its obligate parasitic plant *Cynomorium songaricum*.





Nitraria tangutorum



Cynomorium songaricum

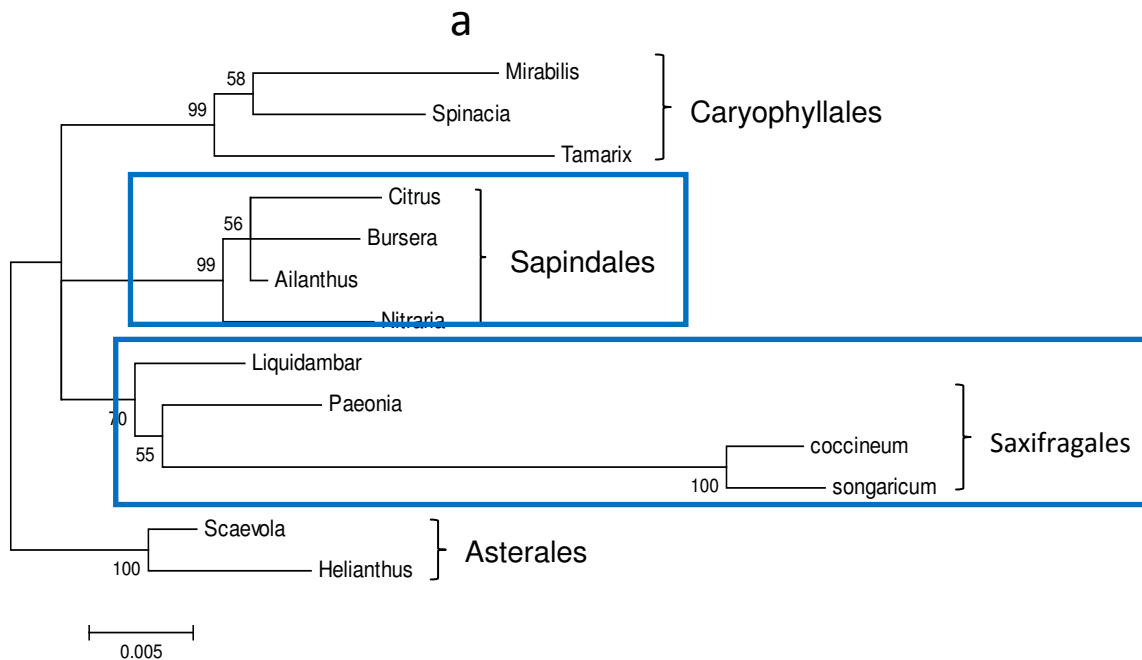
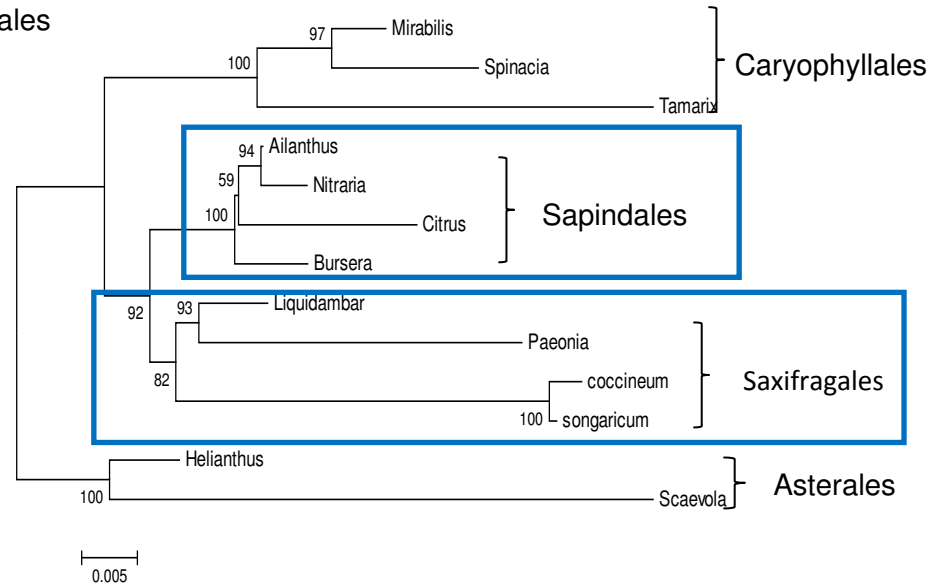
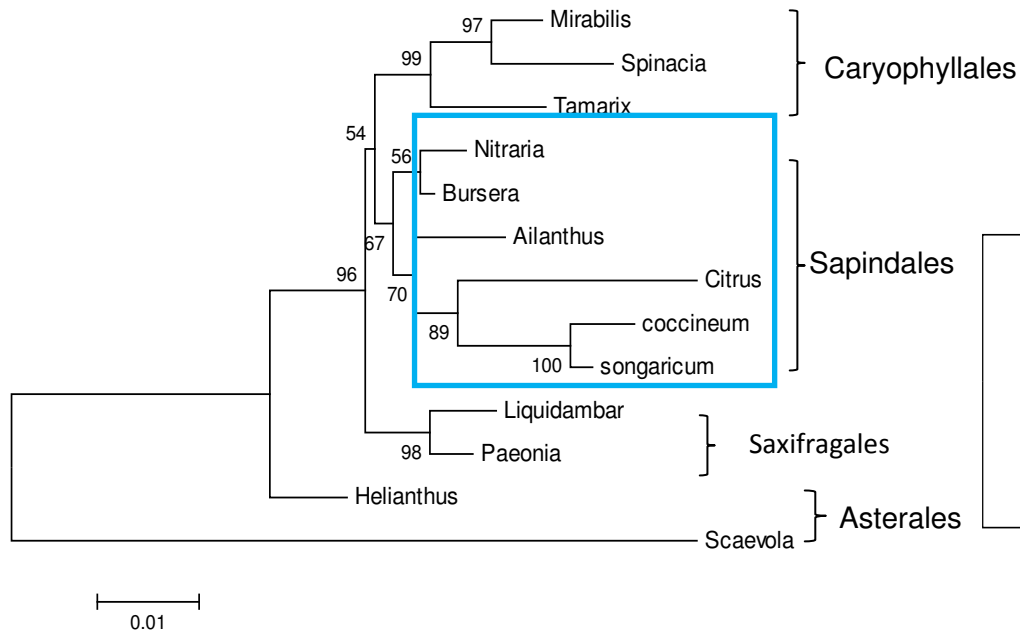


Cynomorium coccineum (Nickrent et al, 2005)

(Parasitic Plant Connection – Cynomoriaceae)

Cynomorium has two species, *Cynomorium coccineum* of northern Africa and the Mediterranean region and *Cynomorium songaricum* of western Asia, the latter species, known as "suo yang" in Chinese. *Cynomorium coccineum* belongs to Saxifragales (Nickrent et al, 2005). The hosts of *Cynomorium coccineum* mainly belong to Caryophyllales and Asterales and no host of the Sapindales has been discovered (Todd et al, 2007). The host of *Cynomorium songaricum* is *Nitraria tangutorum* in China, which belongs to Sapindales.

We amplified and sequenced *atp1*, *matR*, *18srDNA* genes of *Cynomorium songaricum* and its host *Nitraria tangutorum*, compared the sequences with the homologous genes of *Cynomorium coccineum* and other plants. Phylogenetic trees were estimated based on ML analysis. Phylogenetic analyses shows that *atp1* places *Cynomorium coccineum*, *Cynomorium songaricum* in Sapindales. However, according to the phylogenetic analyses of *matR* and *18srDNA*, *Cynomorium coccineum* and *Cynomorium songaricum* are placed in Saxifragales, which suggests HGTs might have happened between the common ancestor of *Cynomorium* and Sapindales.



b

Fig1.
 a. *atp1* phylogenetic tree based on MI analysis
 b. *matR* phylogenetic tree based on MI analysis
 c. *18s* phylogenetic tree based on MI analysis

c

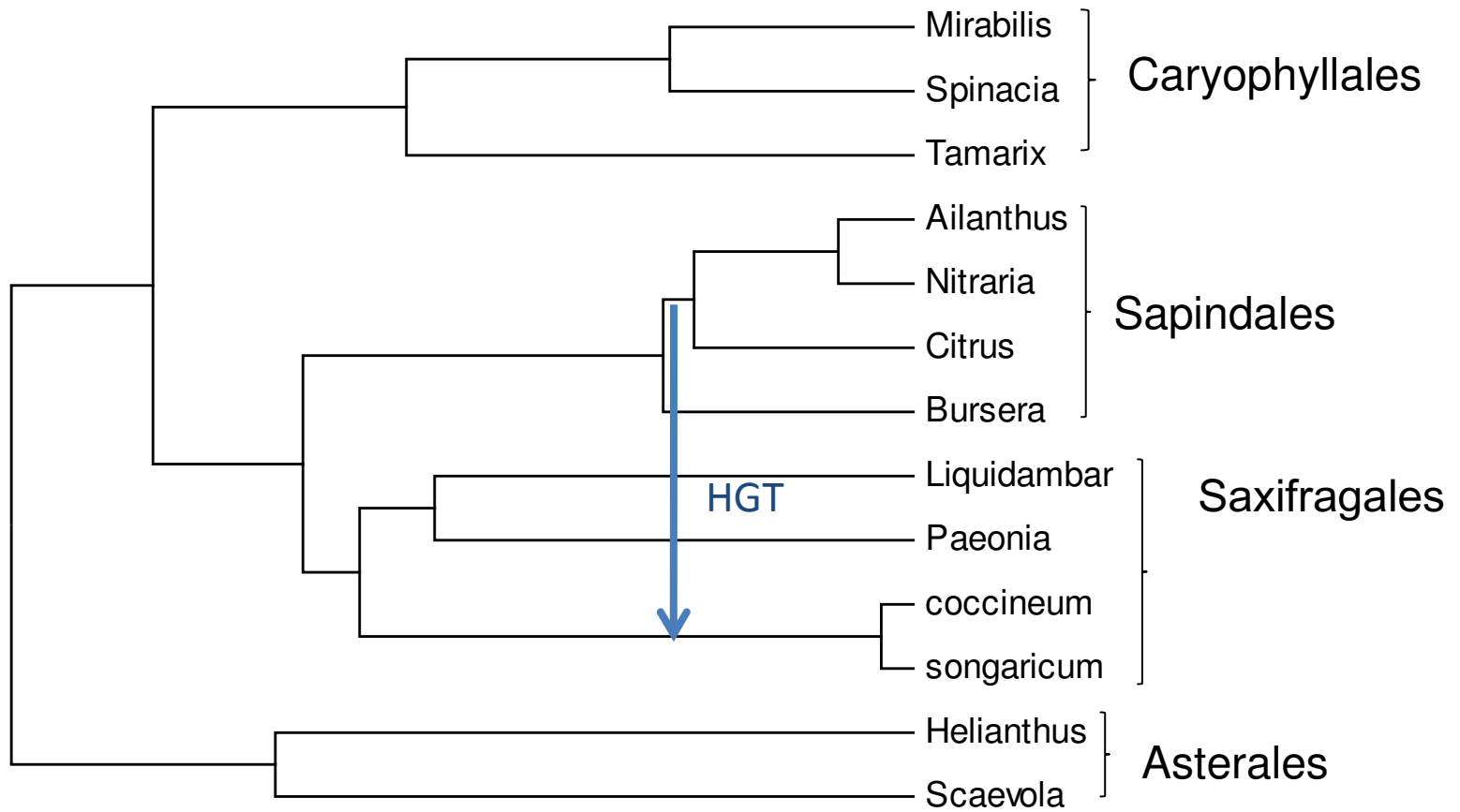
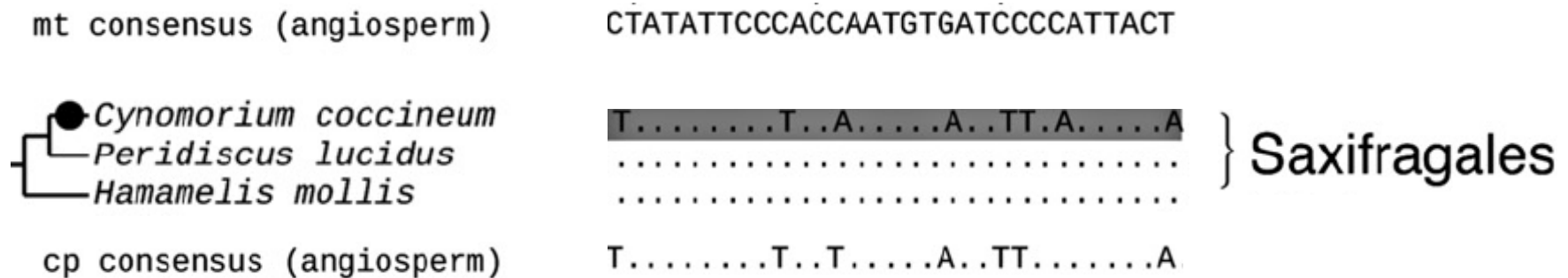


Fig 2. HGT from Sapindales to the common ancestor of *Cynomorium*.

The transferred *atp1* gene also contain a fragment that may involves mitochondrial and chloroplast conversion



Weilong Hao and Jeffrey D. Palmer. Fine-scale mergers of chloroplast and mitochondrial genes create functional, transcompartmentally chimeric mitochondrial genes. Proc Natl Acad Sci USA ,2009 106: 16728-16733.

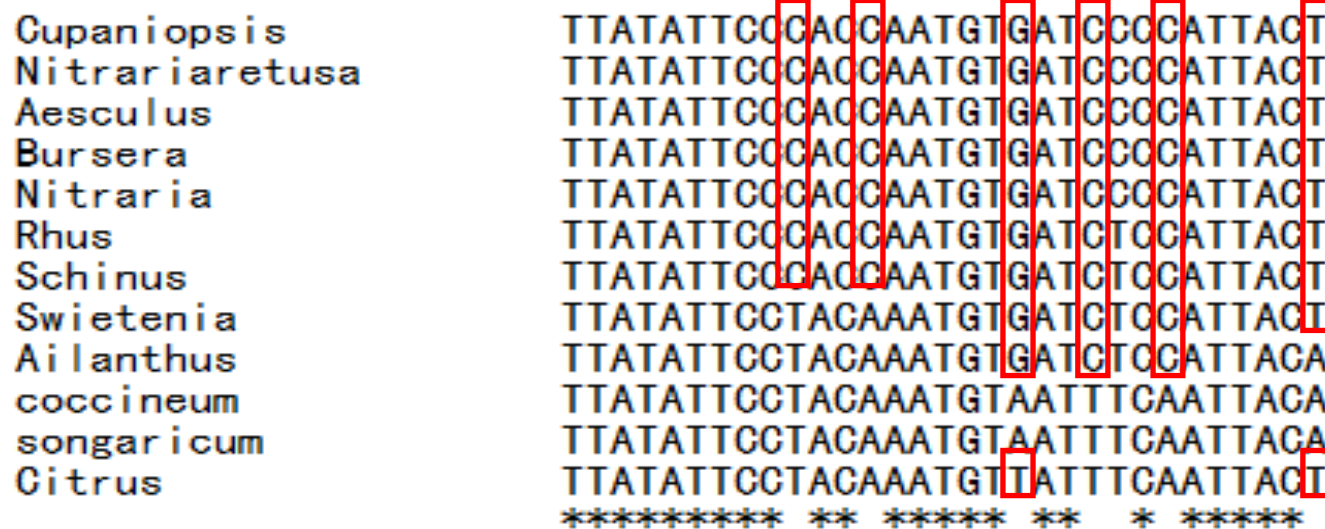


Fig 3 alignment of a fragment of *atp1* gene may involves mergers of chloroplast and mitochondrial genes

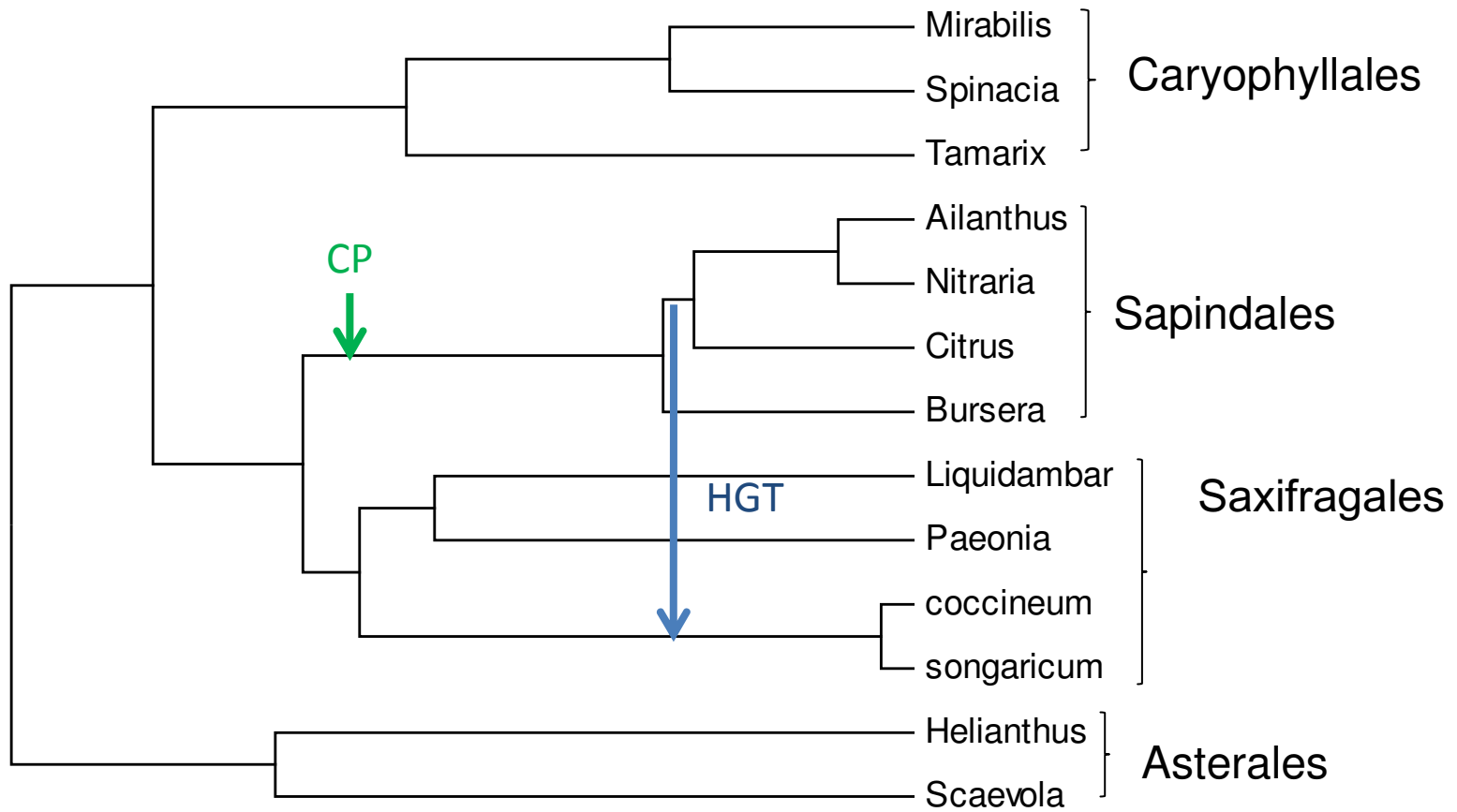


Fig 4. There is mitochondrial and chloroplast conversion in the common ancestor of Sapindales . Then HGT from Sapindales to the common ancestor of Cynomorium.

The similarity between *atp1* gene of *Cynomorium songaricum* and *Nitraria tangutorum* is higher than *Cynomorium coccineum* and *Nitraria tangutorum*

We use [Comp3Seq](http://www.indiana.edu/~orgconv/) (To detect conversion in three sequences <http://www.indiana.edu/~orgconv/>) to further analysis the transferred *atp1* gene

(Hao et. BMC Bioinformatics 2010, 11:114)

Sequence	Donor	start	end	Pvalue(L/N)	Pvalue(L-N)
>songaricum	>Ailanthus	75	699	1.90e-03	1.84e-02
Sequence	Donor	start	end	Pvalue(L/N)	Pvalue(L-N)
>songaricum	>Aesculus	96	699	5.19e-04	4.95e-03
Sequence	Donor	start	end	Pvalue(L/N)	Pvalue(L-N)
>songaricum	>Bursera	96	699	5.32e-03	4.67e-02
Sequence	Donor	start	end	Pvalue(L/N)	Pvalue(L-N)
>songaricum	>Rhus	96	699	1.29e-03	1.24e-02
Sequence	Donor	start	end	Pvalue(L/N)	Pvalue(L-N)
>songaricum	>Nitraria	96	699	2.08e-07	1.78e-06
Sequence	Donor	start	end	Pvalue(L/N)	Pvalue(L-N)
>songaricum	>Citrus	96	699	8.56e-04	1.13e-02

Fig 5 detect conversion in three sequences using *atp1* gene *Cynomorium songaricum*, *Cynomorium coccineum* and Sapindales

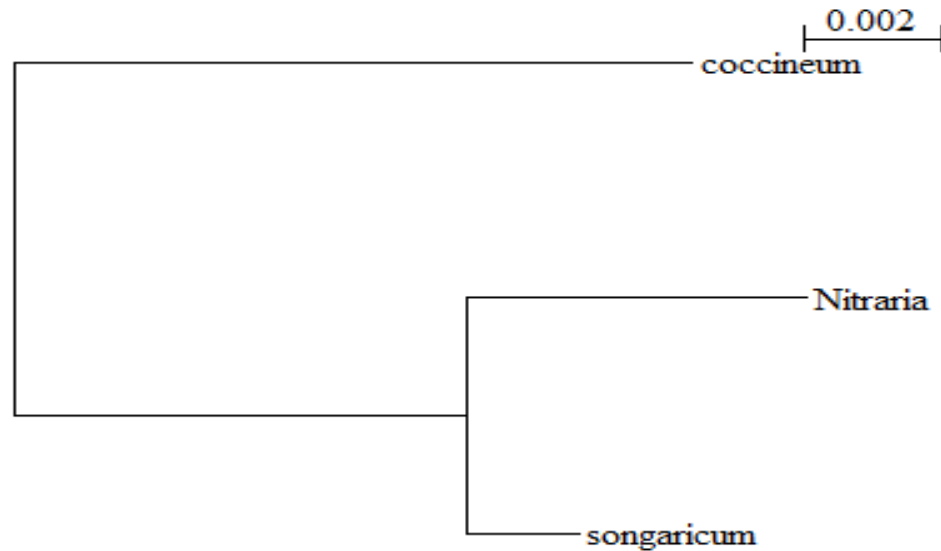


Fig 6 Cluster of the *atp1* fragment of *Cynomorium songaricum* , *Nitraria tangutorum* and *Cynomorium coccineum*.

atp1 of *Cynomorium songaricum* shows the character of chimerism , a fragment of the *atp1* gene of *Cynomorium songaricum* shows high similarity to *Nitraria tangutorum*, even higher than *Cynomorium coccineum*. This suggests that after the divergence of *Cynomorium coccineum* and *Cynomorium songaricum*, a part of the *atp1* gene was horizontally transferred from *Nitraria tangutorum* to *Cynomorium songaricum*.

There are 4 bases different between *Cynomorium songaricum* and *Cynomorium coccineum*. But no amino acid different. There are 6 bases different between *Cynomorium songaricum* and *Cynomorium coccineum*, but there is only 1 amino acid different, and it's an Arginine to Lysine substitution. These two amino acids have very similar properties, they are both basic amino acids and hydrophilic amino acids. We also did not find RNA editing sites in *Cynomorium songaricum atp1* gene. This suggests that the gene fragment of *atp1* horizontally transferred may not affect the function of the protein.

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songaricum      ENENVGIVVFGSDTAIKEGDLVKRTGSIVDVPAGKAMETLGRVVDGLGVPIDGRRGALS DH
Nitrarria      ENENVGIVVFGSDTAIKEGDLVKRTGSIVDVPAGKAMETLGRVVDGLGVPIDGRRGALS DH
coccineum      ENENVGIVVFGSDTAIKEGDLVKRTGSIVDVPAGKAMETLGRVVDGLGVPIDGKRGALS DH
*****

songaricum      ERRRVEVKAPGIIERKSVHEPMETQTGLKAVDSLVP IGRGQREL IIGDRQTGKTAIAIDT
Nitrarria      ERRRVEVKAPGIIERKSVHEPMETQTGLKAVDSLVP IGRGQREL IIGDRQTGKTAIAIDT
coccineum      ERRRVEVKAPGIIERKSVHEPMETQTGLKAVDSLVP IGRGQREL IIGDRQTGKTAIAIDT
*****

songaricum      ILNQKQLNSRATSESETLYCVYVAIGQKRSTVAQLVQILSEANALEYSILVAATASDPAP
Nitrarria      ILNQKQLNSRATSESETLYCVYVAIGQKRSTVAQLVQILSEANALEYSILVAATASDPAP
coccineum      ILNQKQLNSRATSESETLYCVYVAIGQKRSTVAQLVQILSEANALEYSILVAATASDPAP
*****

songaricum      LQFLAPYSGCAME TGEYFRDNGMETHAL I
Nitrarria      LQFLAPYSGCAME TGEYFRDNGMETHAL I
coccineum      LQFLAPYSGCAME TGEYFRDNGMETHAL I
*****

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Fig7 amino acid alignment of the transferred fragment of *atp1* gene

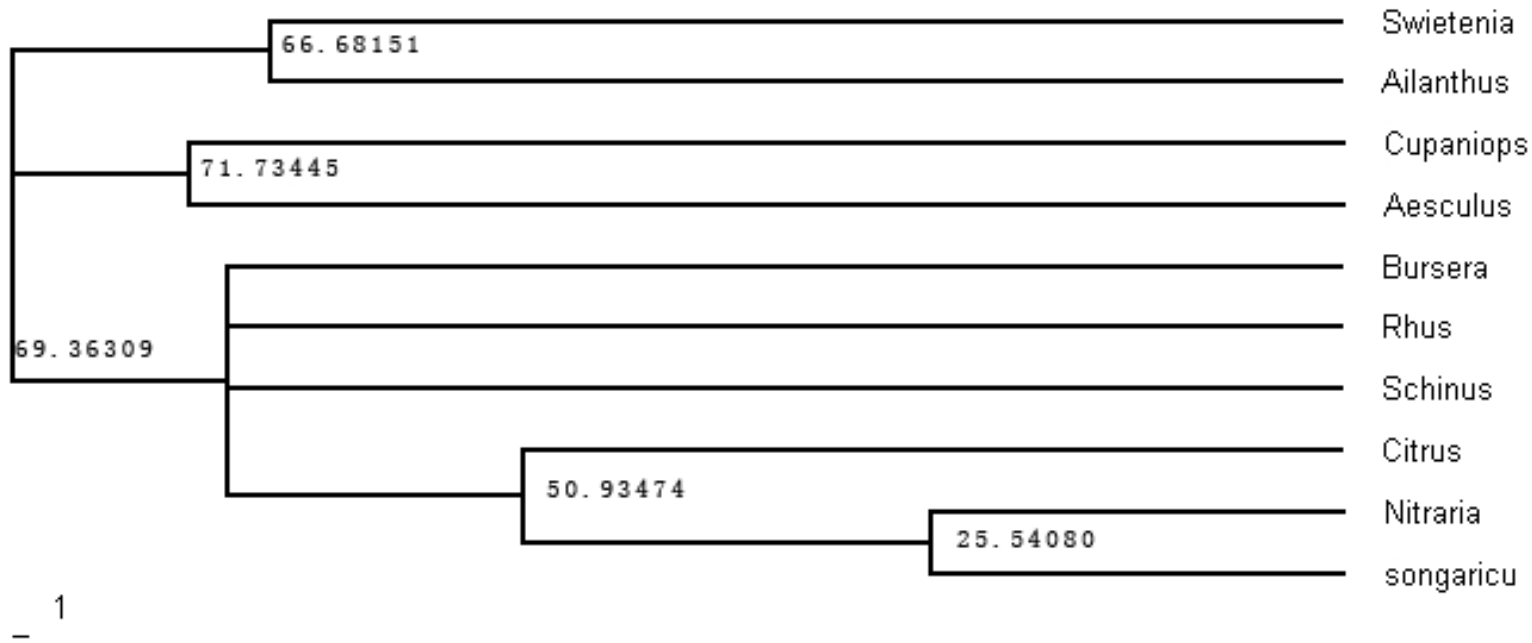


Fig 8 Molecular clock analysis of transferred gene fragment. Fossil data reference :Muellner et al .
Plant System Evolution.2007, 266: 233~252)

According to the Molecular clock analysis , HGT happened about 25 Ma year ago, and *Nitraria tangutorum* has already spread on west China since the end of Eocene Epoch according to the fossil record (about 45Ma)(Miao et al, SCIENCE IN CHINA SERIES D EARTH SCIENCES, 2008). HGT may happened in this time range.

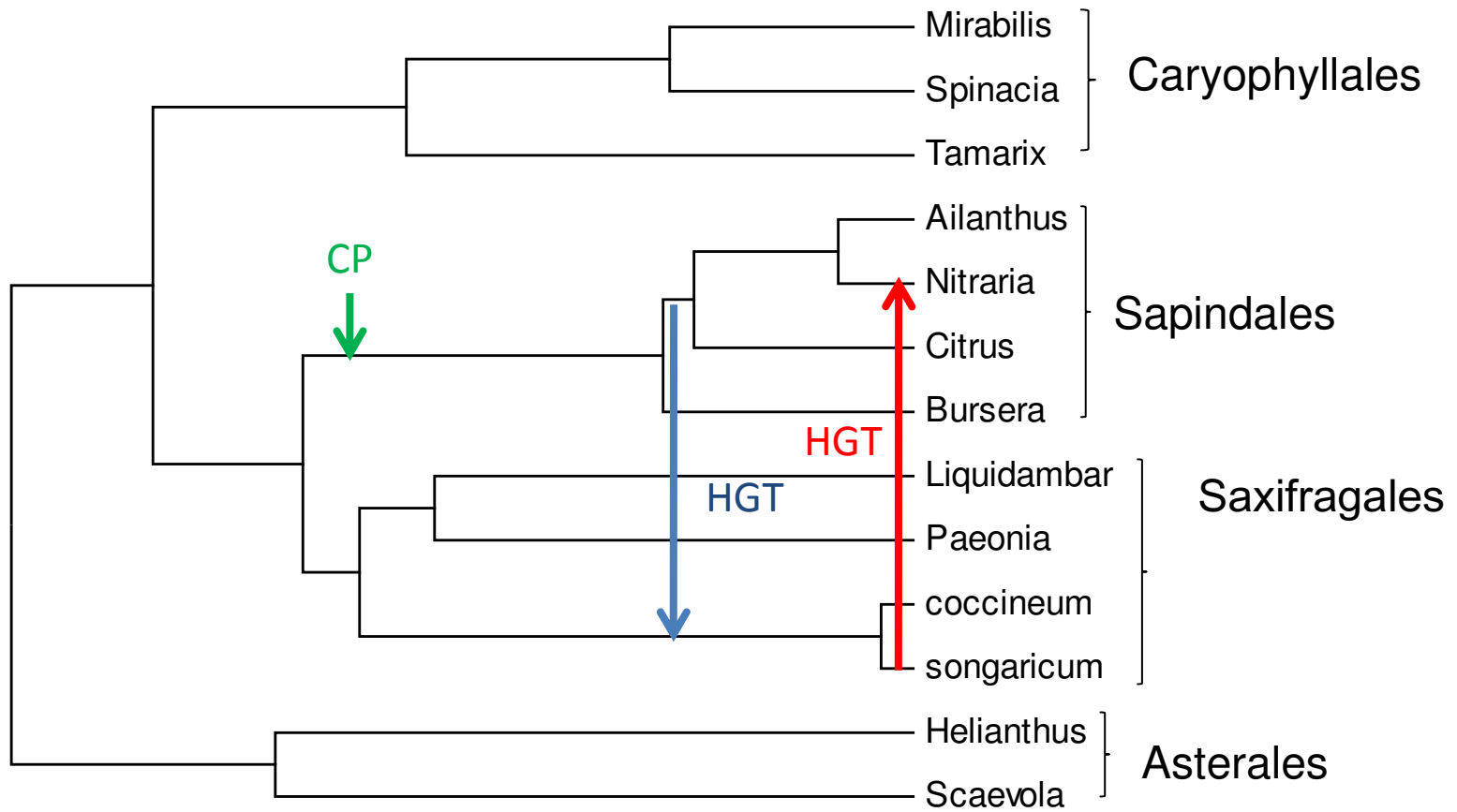
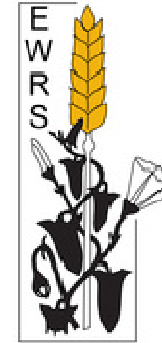


Fig 9. HGT still happened from *Nitraria tangutorum* to *Cynomorium songaricum* after the divergence of *Cynomorium coccineum* and *Cynomorium songaricum*

The results of this research suggest there were HGT from Sapindales to the common ancestor of *Cynomorium*. After the divergence of *Cynomorium coccineum* and *Cynomorium songaricum*, HGT still happened from *Nitraria tangutorum* to *Cynomorium songaricum*. Although *Cynomorium songaricum* and *Cynomorium coccineum* belong to the same genus, some genes will co-evolve with the different hosts they chose.

The results also shows HGT analysis may reveal the history of some parasitic plants.

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Thanks For Your Attention!