# Bioinformatic Analysis of Strawberry PTOX Gene

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**Keywords:** PTOX; Malus x domestica closest relative; the plastid terminal oxidase.

**Abstract.** PTOX has always been known as a key factor of defense response in plant testa. Recent studies show that The plastid terminal oxidase (plastid terminal oxidase, PTOX) is a nuclear encoded, plastid located plastid quinoxone oxidoreductase (plastid quinone oxidase). PTOX is the terminal oxidase of the plant chloroplast respiration. In this research we chosen PTOX gene of 9 kinds species, downloaded their nucleotide and protein sequence from NCBI as the research object, found strawberry PTOX gene via bioinformation analyses, constructed phylogenetic tree. At the same time, we analyzed the strawberry PTOX gene of physical and chemical properties and its protein structure and so on. The phylogenetic tree showed that strawberry and Malus x domestica closest relative. By the protein prediction, we found that the protein owed one proper signal peptide without obvious transmembrane regions.

## 1. Introduction

PTOX has always been known as a key factor of defense response in plant testa. Recent studies show that The plastid terminal oxidase (plastid terminal oxidase, PTOX) is a nuclear encoded, plastid located plastid quinoxone oxidoreductase (plastid quinone oxidase). PTOX is the terminal oxidase of the plant chloroplast respiration. In this research we chosen PTOX gene of 9 kinds species, downloaded their nucleotide and protein sequence from NCBI as the research object, found strawberry PTOX gene via bioinformation analyses, constructed phylogenetic tree. At the same time, we analyzed the strawberry PTOX gene of physical and chemical properties and its protein structure and so on. The phylogenetic tree showed that strawberry and Malus x domestica closest relative. By the protein prediction, we found that the protein owed one proper signal peptide without obvious transmembrane regions.

## 2. Result and Analysis

### 2.1 Strawberry PTOX Find and Phylogenetic Tree Construction of PTOX

Run BLAST in the Fragaria ×ananassa database of GDR and Strawberry GARDEN respectively using Arabidopsis thaliana PTOX as a sample. Select the sequence with ideal S and C value, we find strawberry PTOX.

We have chosen PTOX-like gene of 9 kinds species (Table 1), downloaded their nucleotide and protein sequence from NCBI as the research object, analyzed sequences via MEGA6.0, constructed phylogenetic tree on the genetic distance base (Figure 1). The relationship between strawberry and petunia is closer than others, due to they all contain a lot of anthocyanins probably.

Table 1. Basic Information of Ptox Gene in Different Species

| Species | mRNA Accession Number | Protein Accession Number | Amino Acid Number |
|---|---|---|---|
| Fragaria vesca | XM_004293288.2 | 764550170 | 1,499 |
| Pyrus x bretschneideri | XM_009359240.2 | 1079217804 | 1,646 |
| Malus x domestica | XM_008386041.2 | 1039831879 | 1,247 |
| Prunus mume | XM_008226514.2 | 1027099556 | 1,795 |
| Prunus avium | XM_021967505.1 | 1220073398 | 1,754 |
| Ziziphus jujuba | XM_016022792.1 | 1009107174 | 1,636 |
| Juglans regia | XM_018953165.1 | 1098805633 | 1,424 |
| Jatropha curcas | XM_012235882.2 | 1173853084 | 1,352 |
| Vitis vinifera | XM_002271042.4 | 1104509441 | 1,312 |

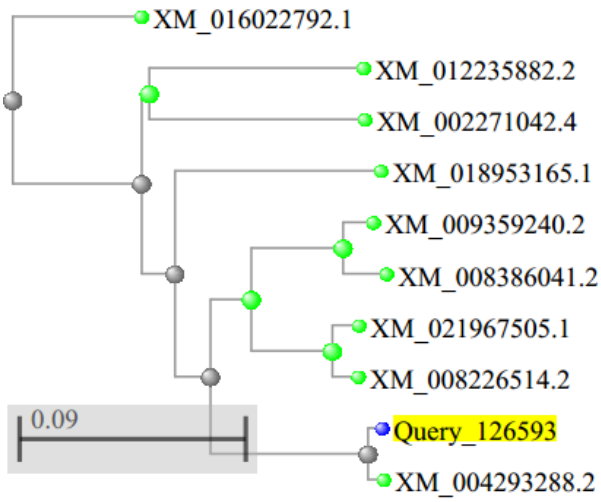


Figure 1. Phylogenetic Tree of Strawberry PTOX.

## 2.2 Strawberry Ptox Protein Physical Analysis

Analyze strawberry PTOX protein physical feature using ProtPram. Result shows as following: the protein is composed by 1107bp, molecular weight is 92443.58, theoretical pI is 5.07, electric neutrality, molecular formula is C3410H5715N1107O1450S219. Because the N-terminal of the sequence considered is A (Ala), the estimated half-life is 4.4 hours in mammalian reticulocytes (in vitro), 20 hours in yeast (in vitro) and 10 hours in Escherichia coli (in vitro). The instability index (II) is computed to be 45.15, so the protein is unstable. The aliphatic index of the protein is 26.47. Grand average of hydropathicity (GRAVY): 0.663.

Amino acid composition result is settings as following (Table 2): the peptide chain consists many Ala as 26.5%.

Table 2. Strawberry Ptox Gene Amino Acid Composition

| Amino Acid | Count | Percentage |
|---|---|---|
| Ala (A) | 293 | 26.5% |
| Gly (G) | 253 | 22.9% |
| Thr (T) | 342 | 30.9% |

## 2.3 Strawberry Ptox Protein Signal Peptide Forecast

Forecast strawberry PTOX protein signal peptide using SignalP 4.1. Result shows as following (Figure 2). There is no signal peptide in the PTOX.

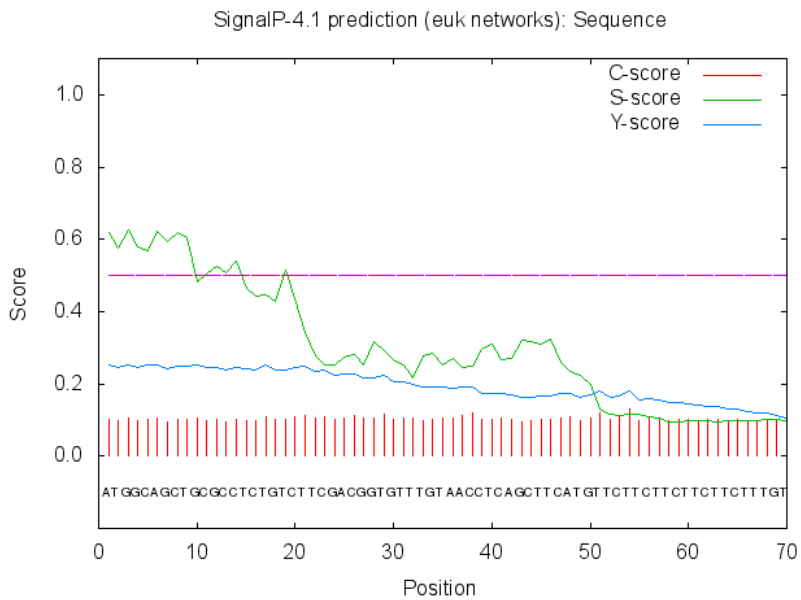


Figure 2. Signal peptide forecast of strawberry PTOX.

### 2.4 Strawberry Ptox Protein Transmembrane Domain Forecast

Forecast strawberry PTOX protein transmembrane domain using TMHMM Server. Result shows as following (Figure 3). There isn't transmembrane domain in the strawberry PTOX, thus the protein is not cytomembrane receptor or located on cytomembrane.

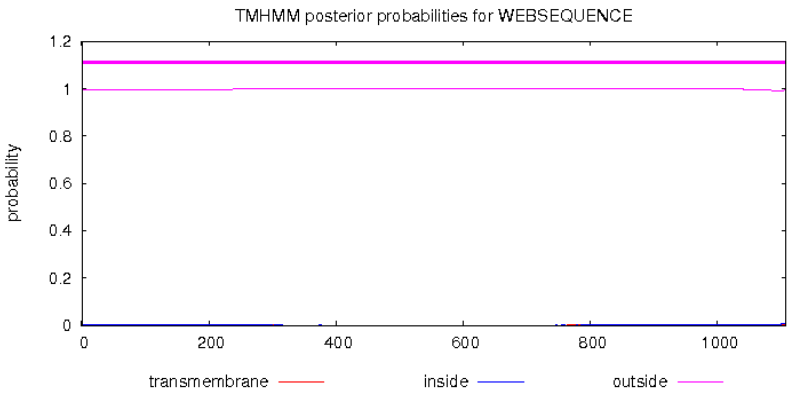


Figure 3. Transmembrane domain forecast of strawberry PTOX.

## 3. Materials and Methods

Sequences access from NCBI (https://www.ncbi.nlm.nih.gov/). BLAST run in GDR (https://www.rosaceae.org/), and Strawberry GARDEN(http://strawberry-garden.kazusa.or.jp/). Phylogenetic tree is constructed via MEGA6.0, forecast strawberry PTOX protein signal peptide using SignalP 4.1 (http://www.cbs.dtu.dk/services/SignalP/), forecast strawberry PTOX protein transmembrane domain using TMHMM Server (http://www.cbs.dtu.dk/services/TMHMM/).

## 4. Conclusion

PTOX is a double ferric oxidase, and there are two iron atoms in its active center and two His and 4 Glu residues chelating with them [8]. PDS is a key enzyme in carotenoid biosynthesis, the activity of PDS will lead to the loss of carotenoid synthesis blocked PTOX is a major force in maintaining the proplastids redox PQ library, in the light under the stress of proplastids due to lack of PTOX, PDS are not functioning properly; there is no synthesis of carotenoids, raw the body cannot be the normal development of chloroplast [9]. The exploration on strawberry TT12, which is forward gene to strawberry PTOX, have been attempt in our lab, we would focus on the interaction between strawberry TT12 and PTOX so as to provide reference for further study and utilization.

## Acknowledgments

I thank my teachers: Haoru Tang, for the directions, Jiangang He for the discussion. This work was financially supported by National Innovation Experiment Program for University Students (201510626005) fund.

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