玉米矮化突变CT1基因的功能分析

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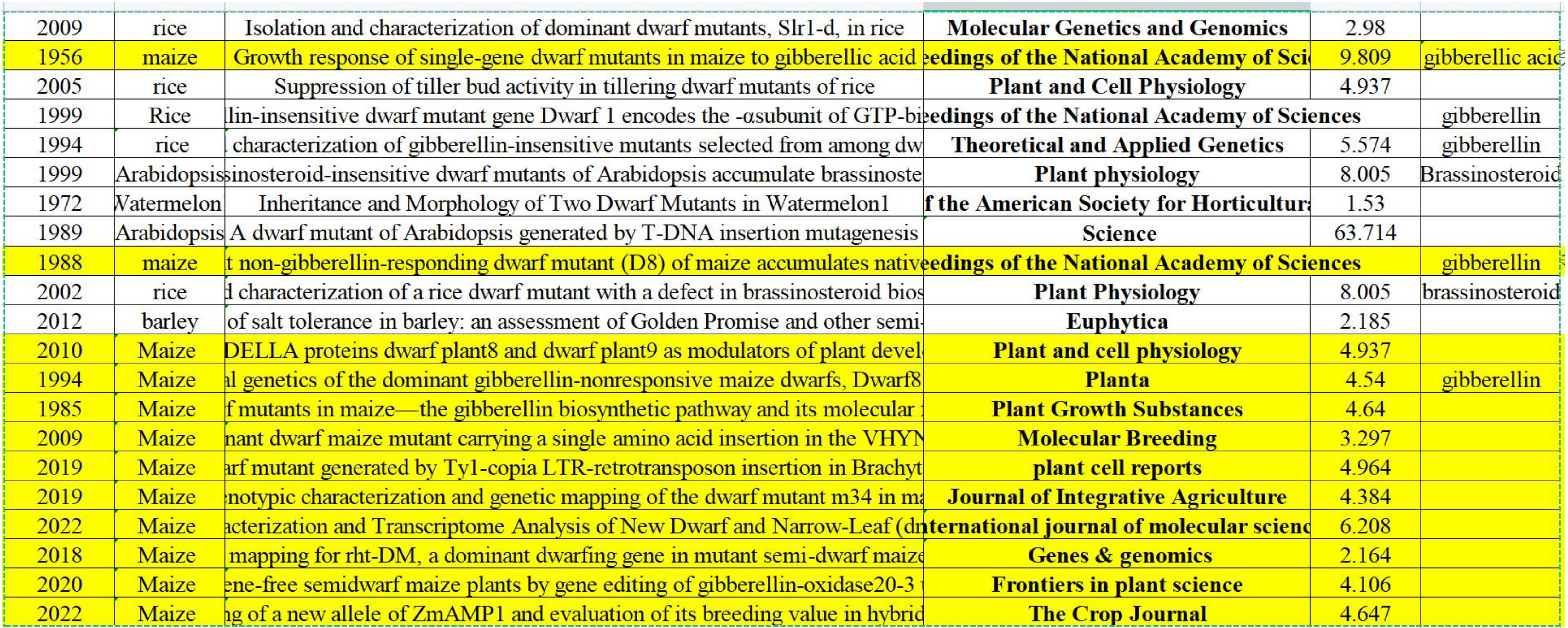
研究背景与意义

矮化性状重要性

- 农业绿色革命被认为是全球农业转型中最关键的点之一，对全球粮食生产、社会经济条件和环境可持续性产生了巨大的影响
- 这一革命是通过并行实施先进的**农艺实践、现代肥料和一种显著提高了抗倒伏性能的新植物理想型**来实现的。
  - 在小麦和水稻等小谷物作物中，化肥投入的增加和种植密度的增加促进了茎的快速伸长，使作物更容易倒伏，这导致了与产量、质量和收获效率降低等相关的重大经济损失
  - 为了解决上述问题，一种降低株高的理想植物型，显著提高了植物的整体坚固性，并保证了更高的种植密度
  - 最初利用矮化性状育种的作物包括小麦和水稻。其株高降低的分子机制主要涉及到内源植物激素赤霉素(GA)的生物合成和信号转导。

(Khush, 2001; Peng et al., 1999)

国内外研究进展



2009	rice	Isolation and characterization of dominant dwarf mutants, Slr1-d, in rice	**Molecular Genetics and Genomics**	2.98	
1956	maize	Growth response of single-gene dwarf mutants in maize to gibberellic acid	**eedings of the National Academy of Sci**	9.809	gibberellic aci
2005	rice	Suppression of tiller bud activity in tillering dwarf mutants of rice	**Plant and Cell Physiology**	4.937	
1999	Rice	lin-insensitive dwarf mutant gene Dwarf 1 encodes the -αsubunit of GTP-bi	**eedings of the National Academy of Sciences**		gibberellin
1994	rice	characterization of gibberellin-insensitive mutants selected from among dw	**Theoretical and Applied Genetics**	5.574	gibberellin
1999	Arabidopsis	sinosteroid-insensitive dwarf mutants of Arabidopsis accumulate brassinoste	**Plant physiology**	8.005	Brassinosteroid
1972	Vatermelon	Inheritance and Morphology of Two Dwarf Mutants in Watermelon1	**f the American Society for Horticultur**	1.53	
1989	Arabidopsis	A dwarf mutant of Arabidopsis generated by T-DNA insertion mutagenesis	**Science**	63.714	
1988	maize	t non-gibberellin-responding dwarf mutant (D8) of maize accumulates nativ	**eedings of the National Academy of Sciences**		gibberellin
2002	rice	d characterization of a rice dwarf mutant with a defect in brassinosteroid bios	**Plant Physiology**	8.005	brassinosteroid
2012	barley	of salt tolerance in barley: an assessment of Golden Promise and other semi-	**Euphytica**	2.185	
2010	Maize	DELLA proteins dwarf plant8 and dwarf plant9 as modulators of plant devel	**Plant and cell physiology**	4.937	
1994	Maize	al genetics of the dominant gibberellin-nonresponsive maize dwarfs, Dwarf8	**Planta**	4.54	gibberellin
1985	Maize	f mutants in maize—the gibberellin biosynthetic pathway and its molecular	**Plant Growth Substances**	4.64	
2009	Maize	nant dwarf maize mutant carrying a single amino acid insertion in the VHYN	**Molecular Breeding**	3.297	
2019	Maize	arf mutant generated by Ty1-copia LTR-retrotransposon insertion in Brachyt	**plant cell reports**	4.964	
2019	Maize	notypic characterization and genetic mapping of the dwarf mutant m34 in ma	**Journal of Integrative Agriculture**	4.384	
2022	Maize	acterization and Transcriptome Analysis of New Dwarf and Narrow-Leaf (d	**nternational journal of molecular scienc**	6.208	
2018	Maize	mapping for rht-DM, a dominant dwarfing gene in mutant semi-dwarf maize	**Genes & genomics**	2.164	
2020	Maize	ene-free semidwarf maize plants by gene editing of gibberellin-oxidase20-3	**Frontiers in plant science**	4.106	
2022	Maize	ng of a new allele of ZmAMP1 and evaluation of its breeding value in hybrid	**The Crop Journal**	4.647	

drawf mutant

(maize)

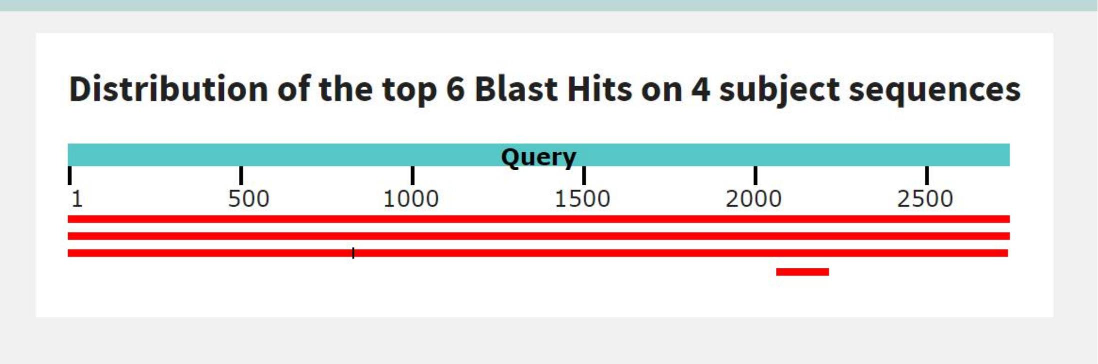
gibberellin 赤霉素

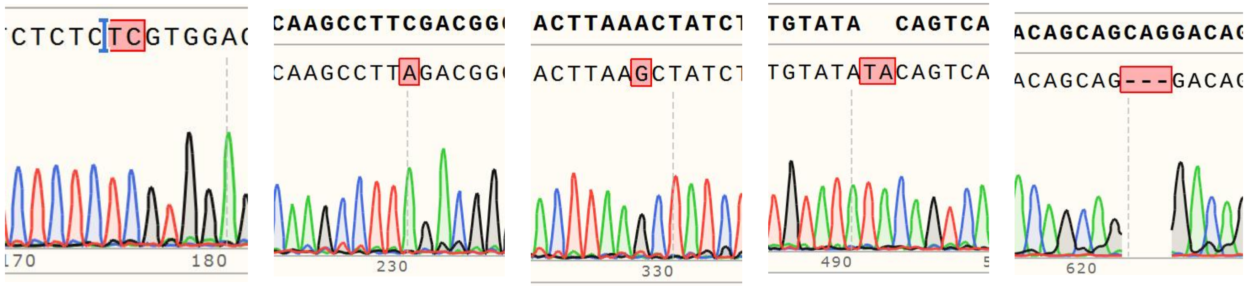
Brassinosteroid油菜素类固醇

玉米单基因矮化突变体对赤霉素的生长响应

显性赤霉素无反应植株

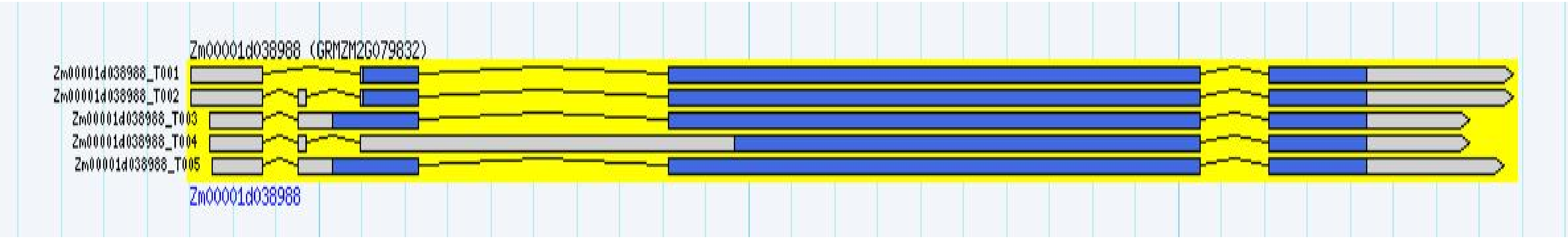
课题进展 基因测序与分析


Distribution of the top 6 Blast Hits on 4 subject sequences



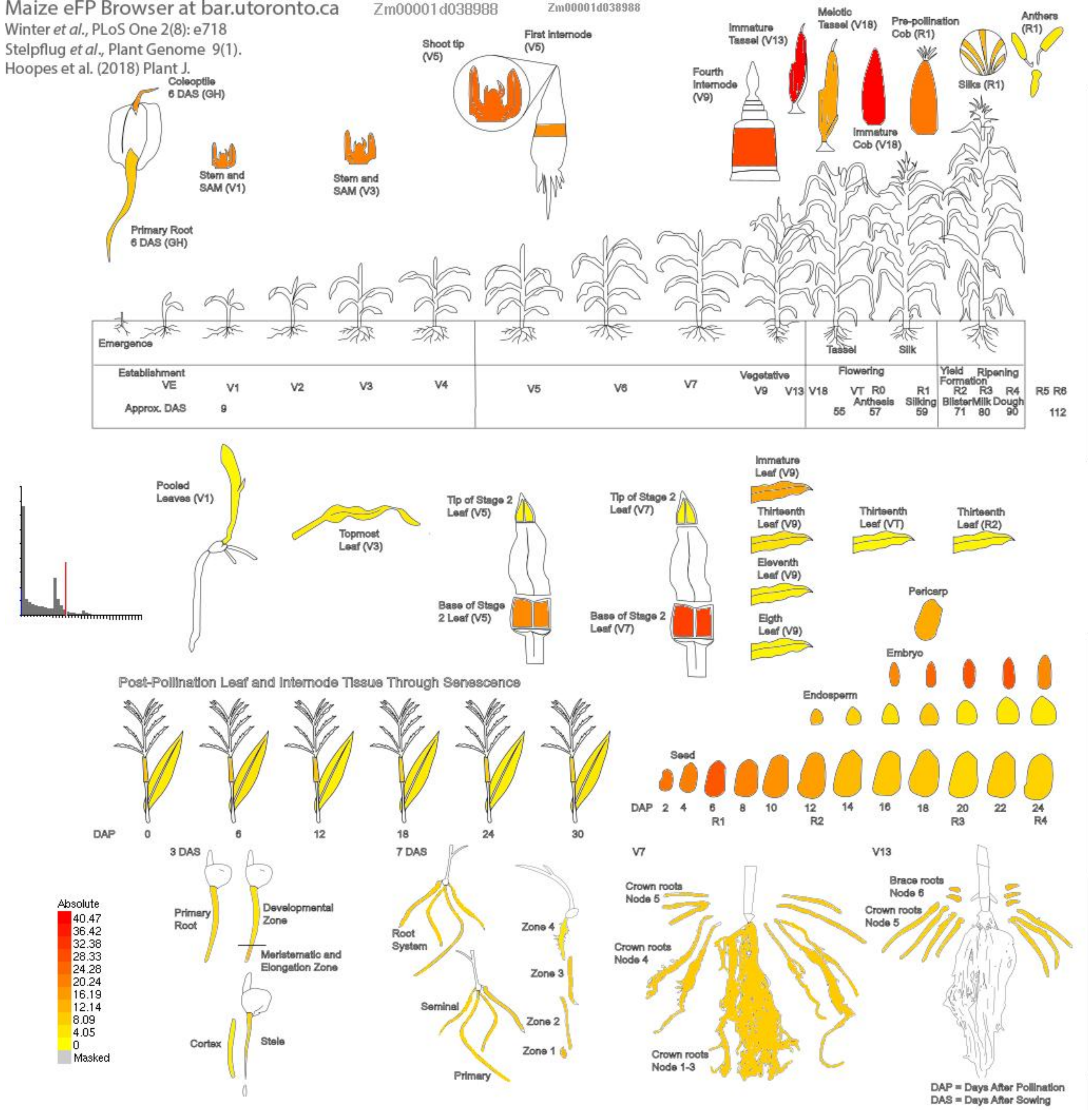
- ➢ Ct1 Zm00001d038988（mutant EMS4-091a53）
- ➢ 同源 Zm00001d009548 （mutant EMS4-0a700e）
- ➢ CT1基因位于chr6上，长度2996bp，物理位置，6:168296701..168299778，其中包含3个exon，自然突变是在exon1上，EMS突变在exon2上。
- ➢ 通过NCBI比对，同源染色体chr8，物理位置，8:69755639..69758896，与其有着极高的相似比（左图第3条），在这2996bp中，高达近90%的相似率。
- ➢ 自然突变ct1中，在第一个外显子60位点处含有TA插入，并在189位点处，缺失CAG三个碱基（与chr8序列相同），另外，还有三个内含子处的突变

基因结构


Zm00001d038988 (GRMZM2G079832)
Zm00001d038988_T001
Zm00001d038988_T002
Zm00001d038988_T003
Zm00001d038988_T004
Zm00001d038988_T005
Zm00001d038988

基因表达情况

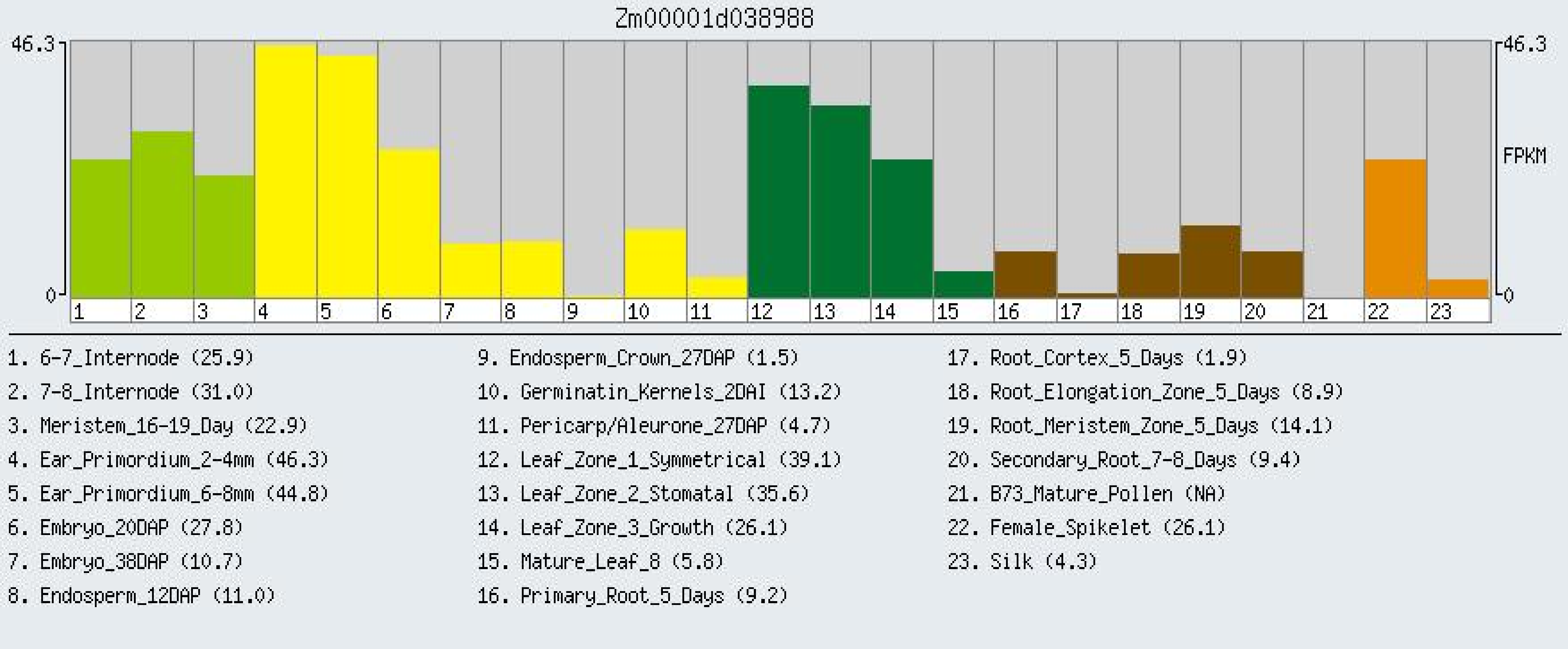
在MaizeGDB上可以看到在不同部位，不同生长时期的表达情况



Maize eFP Browser at bar.utoronto.ca
Winter *et al.*, PLoS One 2(8): e718
Stelpflug *et al.*, Plant Genome 9(1).
Hoopes et al. (2018) Plant J.

Adapters and low quality bases were removed using Cutadapt (v1.12) (Martin, 2011). All cleaned reads were aligned to the Z. mays inbred B73 AGPv4 genome assembly (Jiao et al., 2017) with Bowtie2 (v2.2.3) (Langmead and Salzberg, 2012) and TopHat2 (v2.0.14) (Kim et al., 2013). Fragments Per Kilobase of transcript per Million mapped reads (FPKM) gene expression values for Z. mays inbred B73 AGPv4 genes (Jiao et al., 2017) was quantified with Cufflinks (v2.2.1) (Trapnell et al., 2010).

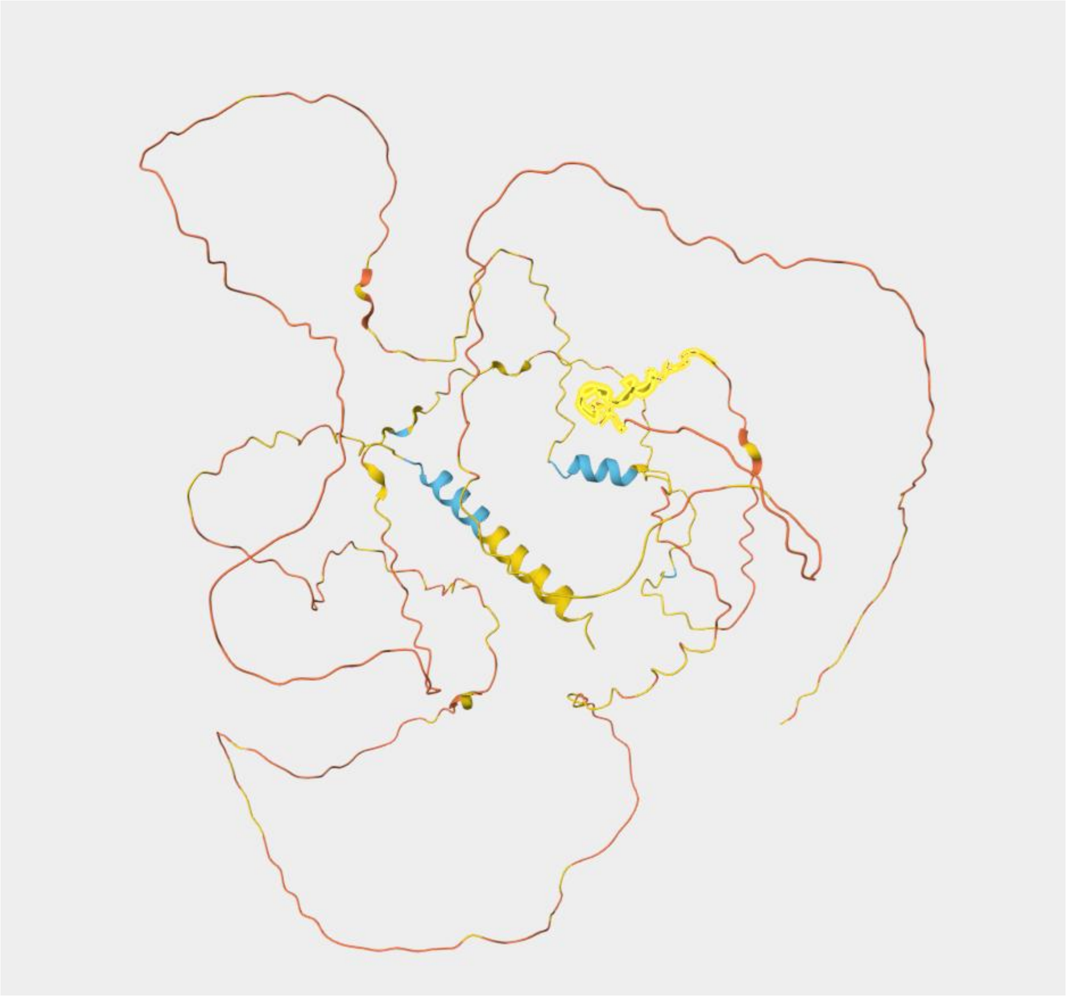
RNA-seq expression



1. 6-7_Internode (25.9)
2. 7-8_Internode (31.0)
3. Meristem_16-19_Day (22.9)
4. Ear_Primordium_2-4mm (46.3)
5. Ear_Primordium_6-8mm (44.8)
6. Embryo_20DAP (27.8)
7. Embryo_38DAP (10.7)
8. Endosperm_12DAP (11.0)
9. Endosperm_Crown_27DAP (1.5)
10. Germinatin_Kernels_2DAI (13.2)
11. Pericarp/Aleurone_27DAP (4.7)
12. Leaf_Zone_1_Symmetrical (39.1)
13. Leaf_Zone_2_Stomatal (35.6)
14. Leaf_Zone_3_Growth (26.1)
15. Mature_Leaf_8 (5.8)
16. Primary_Root_5_Days (9.2)
17. Root_Cortex_5_Days (1.9)
18. Root_Elongation_Zone_5_Days (8.9)
19. Root_Meristem_Zone_5_Days (14.1)
20. Secondary_Root_7-8_Days (9.4)
21. B73_Mature_Pollen (NA)
22. Female_Spikelet (26.1)
23. Silk (4.3)

Walley JW, Sartor RC, Shen Z, Schmitz RJ, Wu KJ, Urich MA, et al. 2016. Integration of omic networks in a developmental atlas of maize. Science.

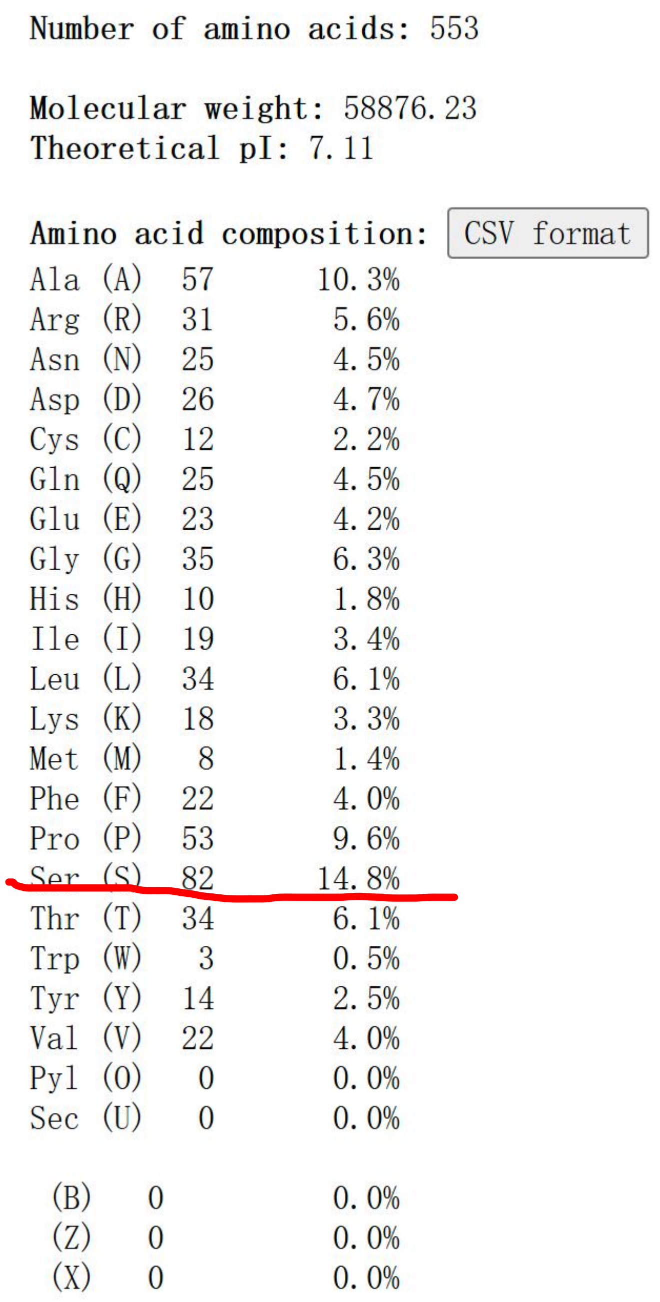
羟脯氨酸富含糖蛋白家族蛋白（预测）



90-139 185-207 266-288
disorder

96-116
polar

117-139
low complexity

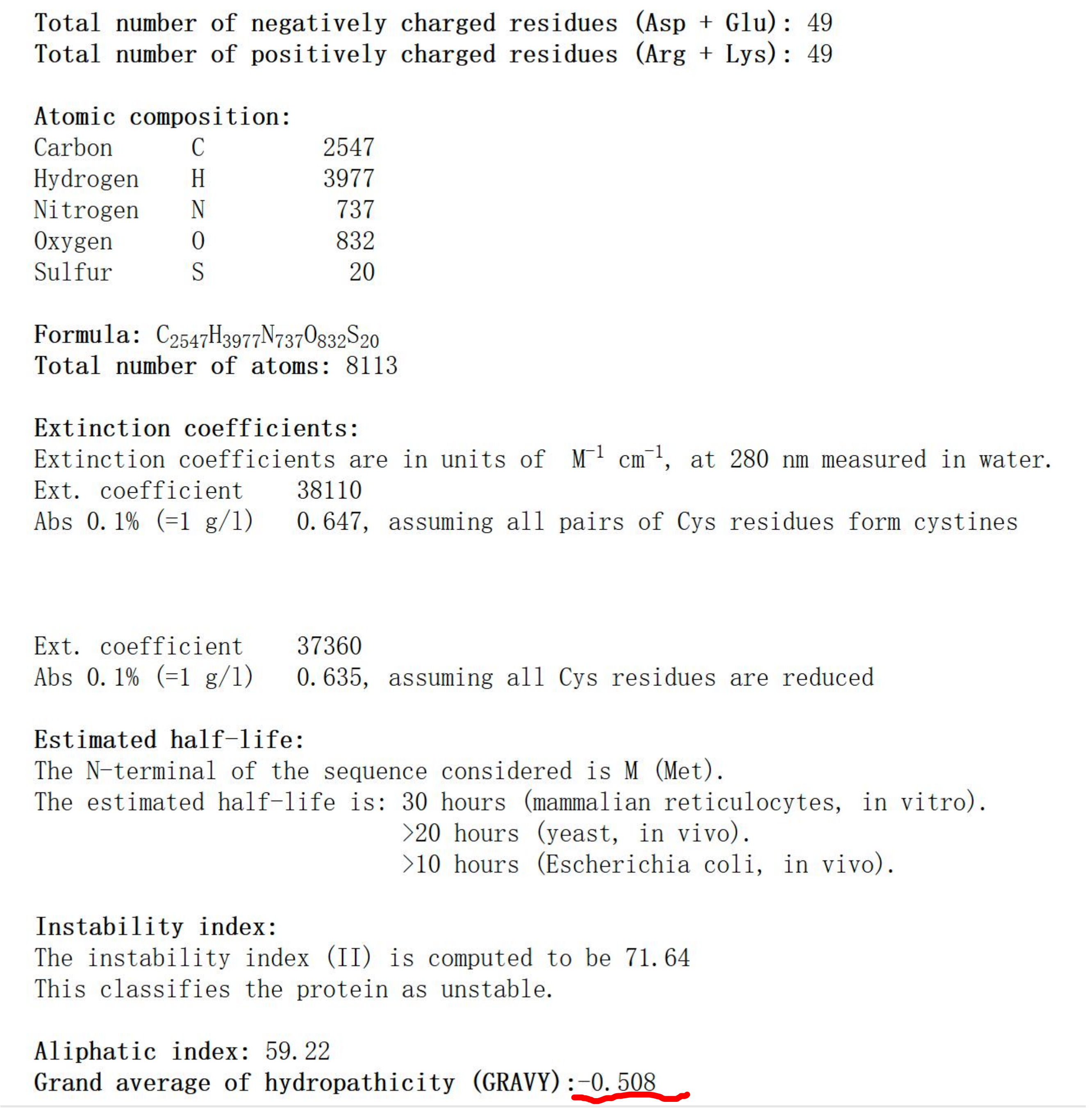


```
Number of amino acids: 553

Molecular weight: 58876.23
Theoretical pI: 7.11

Amino acid composition: CSV format
Ala (A)  57      10.3%
Arg (R)  31       5.6%
Asn (N)  25       4.5%
Asp (D)  26       4.7%
Cys (C)  12       2.2%
Gln (Q)  25       4.5%
Glu (E)  23       4.2%
Gly (G)  35       6.3%
His (H)  10       1.8%
Ile (I)  19       3.4%
Leu (L)  34       6.1%
Lys (K)  18       3.3%
Met (M)   8       1.4%
Phe (F)  22       4.0%
Pro (P)  53       9.6%
Ser (S)  82      14.8%
Thr (T)  34       6.1%
Trp (W)   3       0.5%
Tyr (Y)  14       2.5%
Val (V)  22       4.0%
Pyl (O)   0       0.0%
Sec (U)   0       0.0%

 (B)   0          0.0%
 (Z)   0          0.0%
 (X)   0          0.0%
```



```
Total number of negatively charged residues (Asp + Glu): 49
Total number of positively charged residues (Arg + Lys): 49

Atomic composition:
Carbon      C        2547
Hydrogen    H        3977
Nitrogen    N         737
Oxygen      O         832
Sulfur      S          20

Formula: C2547H3977N737O832S20
Total number of atoms: 8113

Extinction coefficients:
Extinction coefficients are in units of  M-1 cm-1, at 280 nm measured in water.
Ext. coefficient    38110
Abs 0.1% (=1 g/l)   0.647, assuming all pairs of Cys residues form cystines



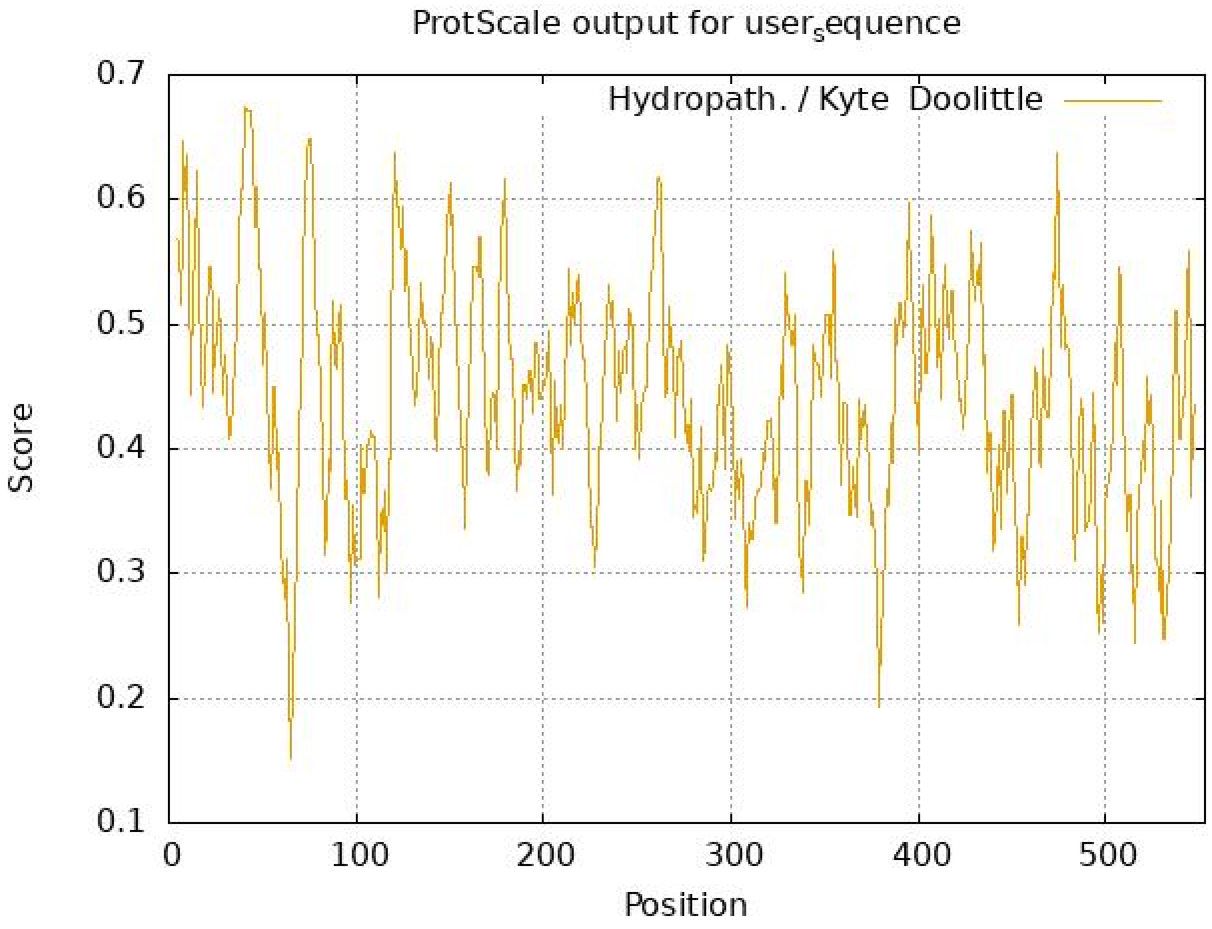
Ext. coefficient    37360
Abs 0.1% (=1 g/l)   0.635, assuming all Cys residues are reduced

Estimated half-life:
The N-terminal of the sequence considered is M (Met).
The estimated half-life is: 30 hours (mammalian reticulocytes, in vitro).
                            >20 hours (yeast, in vivo).
                            >10 hours (Escherichia coli, in vivo).

Instability index:
The instability index (II) is computed to be 71.64
This classifies the protein as unstable.

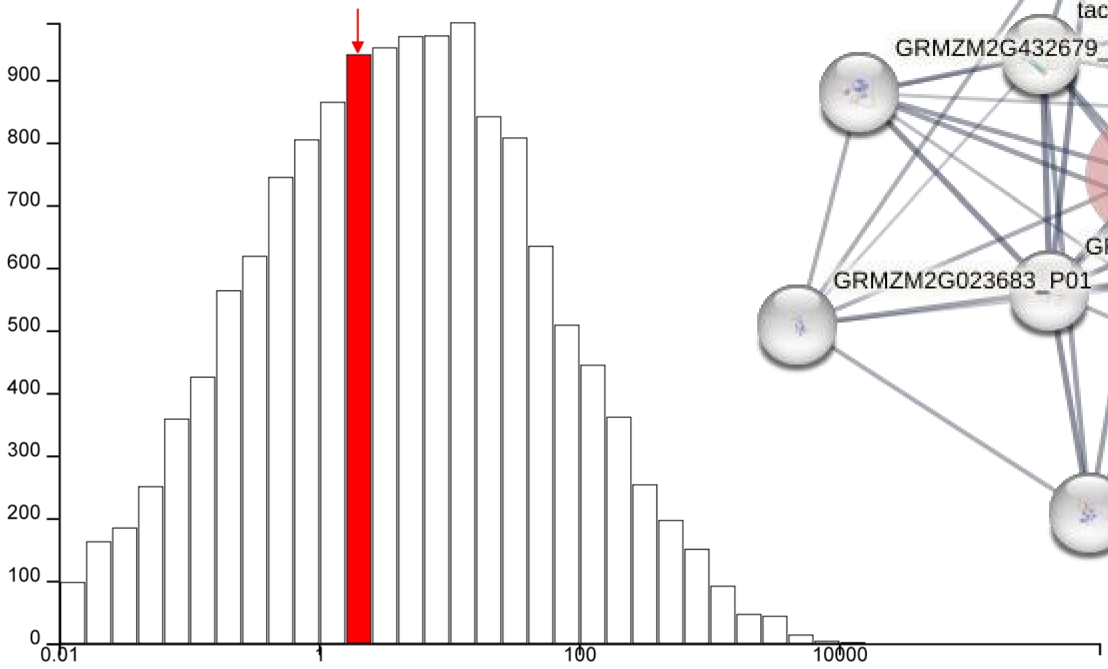
Aliphatic index: 59.22
Grand average of hydropathicity (GRAVY):-0.508
```

蛋白质亲水性（using expacy ProtScale）

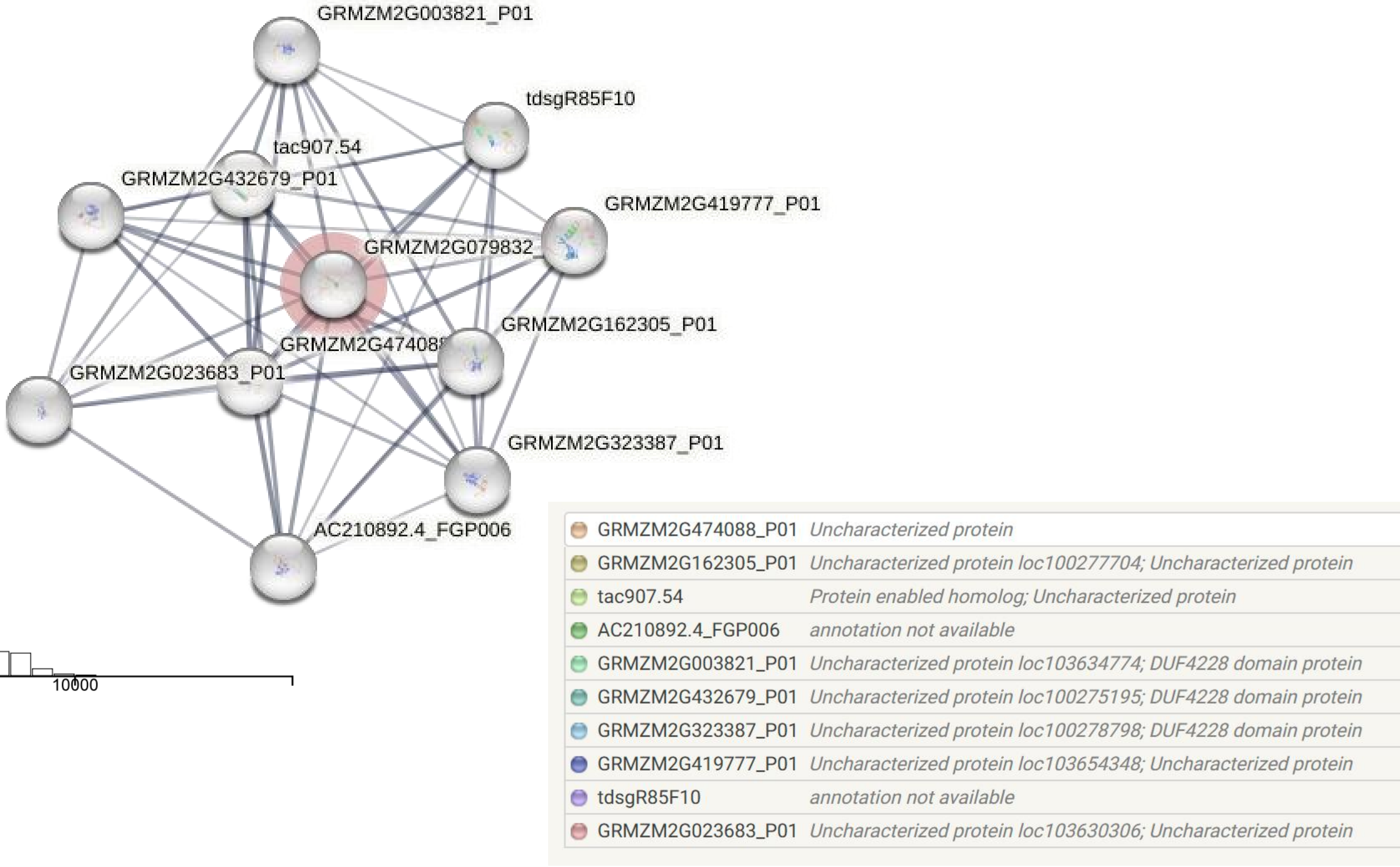


蛋白质等电点pH7.88

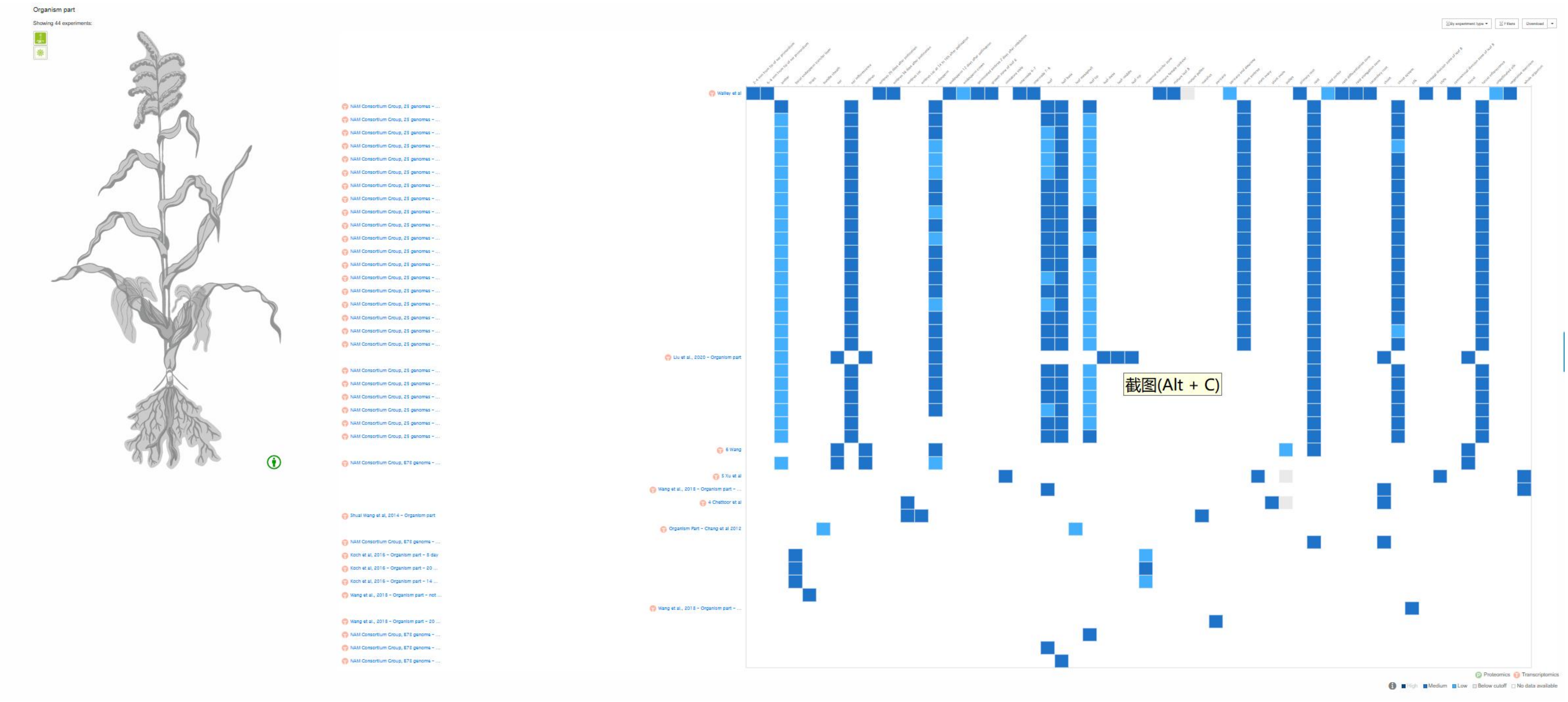
蛋白质丰度



互作蛋白



●	GRMZM2G474088_P01	*Uncharacterized protein*
●	GRMZM2G162305_P01	*Uncharacterized protein loc100277704; Uncharacterized protein*
●	tac907.54	*Protein enabled homolog; Uncharacterized protein*
●	AC210892.4_FGP006	*annotation not available*
●	GRMZM2G003821_P01	*Uncharacterized protein loc103634774; DUF4228 domain protein*
●	GRMZM2G432679_P01	*Uncharacterized protein loc100275195; DUF4228 domain protein*
●	GRMZM2G323387_P01	*Uncharacterized protein loc100278798; DUF4228 domain protein*
●	GRMZM2G419777_P01	*Uncharacterized protein loc103654348; Uncharacterized protein*
●	tdsgR85F10	*annotation not available*
●	GRMZM2G023683_P01	*Uncharacterized protein loc103630306; Uncharacterized protein*



Organism part

Showing 44 experiments:

Walley et al
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
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NAM Consortium Group, 25 genomes – ...
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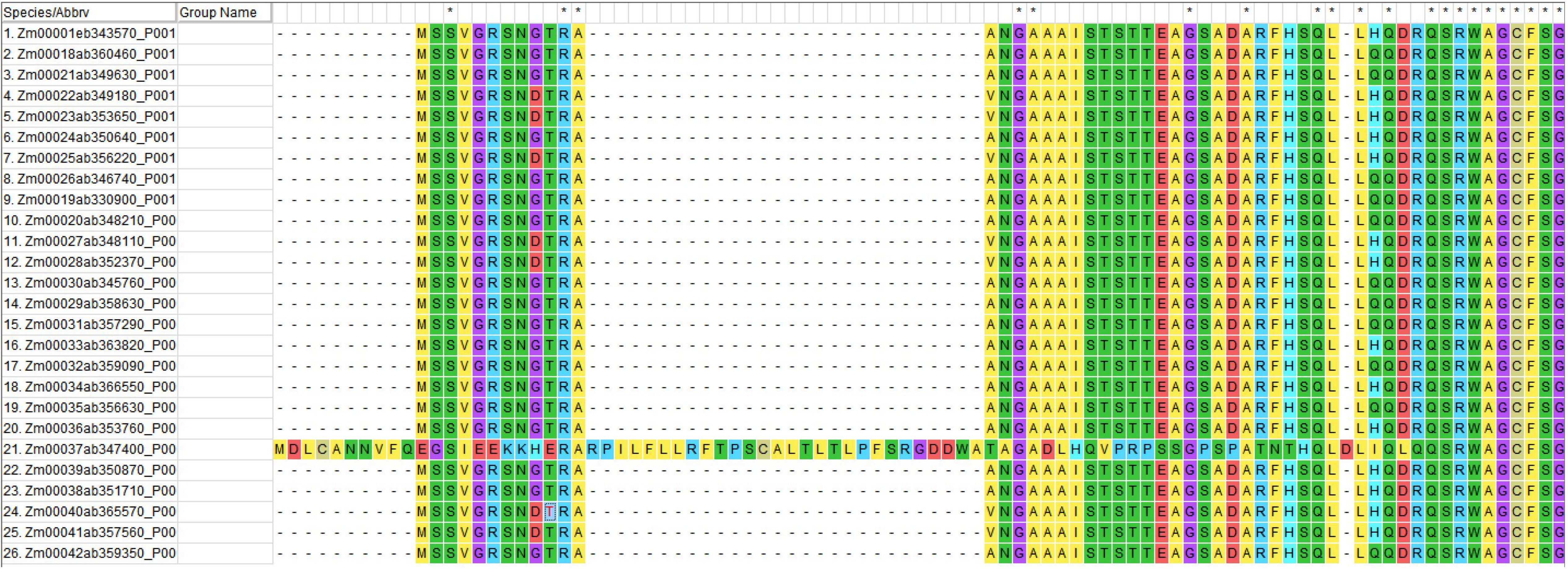
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
Liu et al., 2020 – Organism part
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
NAM Consortium Group, 25 genomes – ...
6 Wang
NAM Consortium Group, B73 genome – ...
5 Xu et al
Wang et al., 2018 – Organism part – ...
4 Chettoor et al
Shuai Wang et al, 2014 – Organism part
Organism Part – Chang et al 2012
NAM Consortium Group, B73 genome – ...
Koch et al, 2016 – Organism part – 8 day
Koch et al, 2016 – Organism part – 20 ...
Koch et al, 2016 – Organism part – 14 ...
Wang et al., 2018 – Organism part – not ...
Wang et al., 2018 – Organism part – ...
Wang et al., 2018 – Organism part – 20 ...
NAM Consortium Group, B73 genome – ...
NAM Consortium Group, B73 genome – ...
NAM Consortium Group, B73 genome – ...

EBI



```
Species/Abbrv           Group Name             *       **                              **          *   *    ** * *  **********
1. Zm00001eb343570_P001            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LHQDRQSRWAGCFSG
2. Zm00018ab360460_P001            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LQQDRQSRWAGCFSG
3. Zm00021ab349630_P001            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LQQDRQSRWAGCFSG
4. Zm00022ab349180_P001            ----------MSSVGRSNDTRA----------------------------VNGAAAISTSTTEAGSADARFHSQL-LHQDRQSRWAGCFSG
5. Zm00023ab353650_P001            ----------MSSVGRSNDTRA----------------------------VNGAAAISTSTTEAGSADARFHSQL-LHQDRQSRWAGCFSG
6. Zm00024ab350640_P001            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LHQDRQSRWAGCFSG
7. Zm00025ab356220_P001            ----------MSSVGRSNDTRA----------------------------VNGAAAISTSTTEAGSADARFHSQL-LHQDRQSRWAGCFSG
8. Zm00026ab346740_P001            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LQQDRQSRWAGCFSG
9. Zm00019ab330900_P001            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LQQDRQSRWAGCFSG
10. Zm00020ab348210_P00            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LQQDRQSRWAGCFSG
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15. Zm00031ab357290_P00            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LQQDRQSRWAGCFSG
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18. Zm00034ab366550_P00            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LHQDRQSRWAGCFSG
19. Zm00035ab356630_P00            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LQQDRQSRWAGCFSG
20. Zm00036ab353760_P00            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LHQDRQSRWAGCFSG
21. Zm00037ab347400_P00            MDLCANNVFQEGSIEEKKHERARPILFLLRFTPSCALTLTLPFSRGDDWATAGADLHQVPRPSSGPSPATNTHQLDLIQLQQSRWAGCFSG
22. Zm00039ab350870_P00            ----------MSSVGRSNGTRA----------------------------ANGAAAISTSTTEAGSADARFHSQL-LHQDRQSRWAGCFSG
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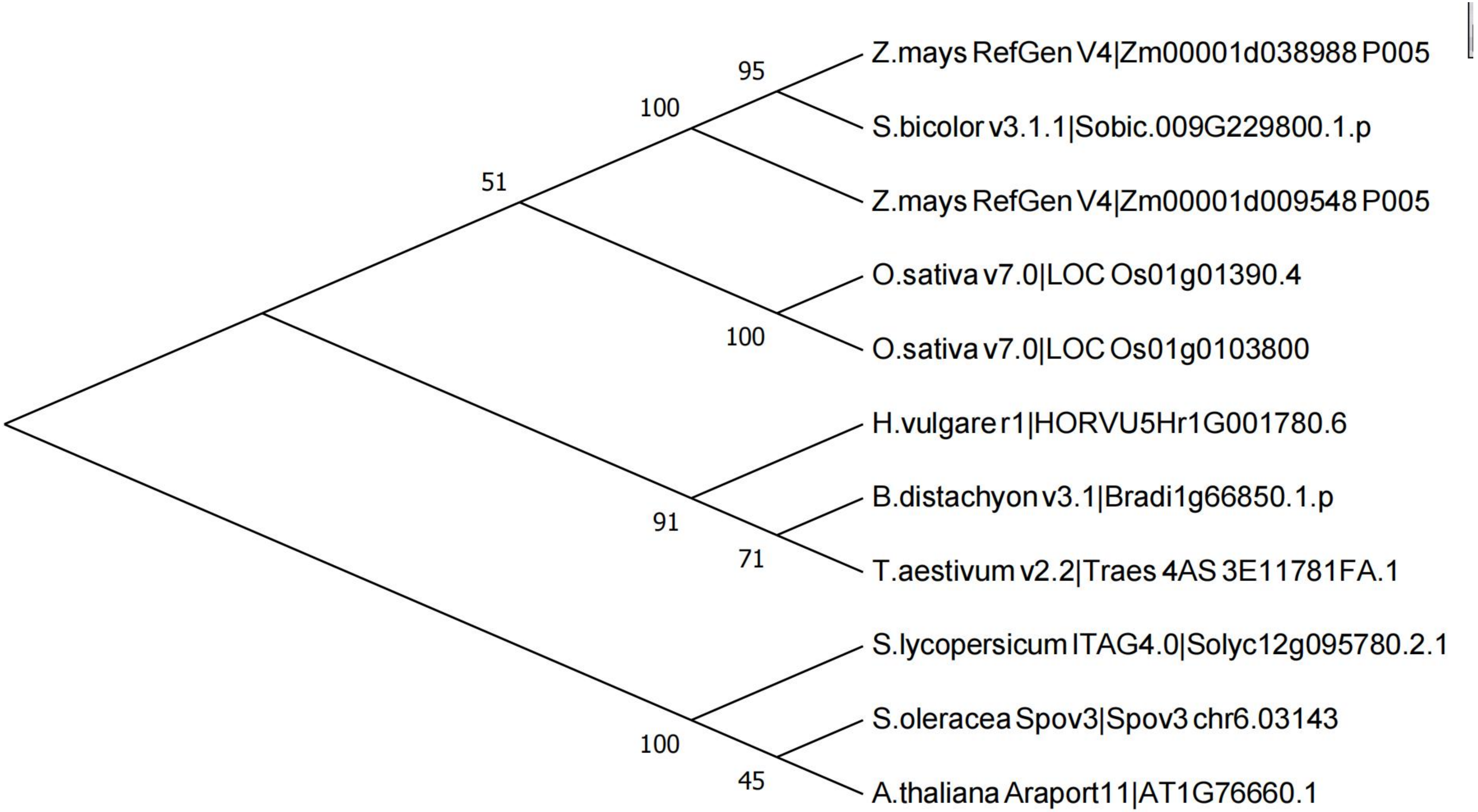
95
100
51
100
91
71
100
45
Z.mays RefGen V4|Zm00001d038988 P005
S.bicolor v3.1.1|Sobic.009G229800.1.p
Z.mays RefGen V4|Zm00001d009548 P005
O.sativa v7.0|LOC Os01g01390.4
O.sativa v7.0|LOC Os01g0103800
H.vulgare r1|HORVU5Hr1G001780.6
B.distachyon v3.1|Bradi1g66850.1.p
T.aestivum v2.2|Traes 4AS 3E11781FA.1
S.lycopersicum ITAG4.0|Solyc12g095780.2.1
S.oleracea Spov3|Spov3 chr6.03143
A.thaliana Araport11|AT1G76660.1

THANK YOU