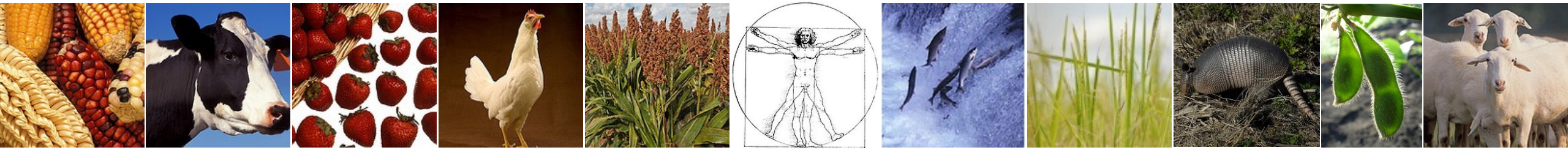

Genome Annotation of Arabidopsis thaliana T2T Col-CC By NCBI RefSeq

Terence D. Murphy – RefSeq Eukaryotes and CGR Project Team Lead

NIH National Library of Medicine
National Center for Biotechnology Information

Outline

- The CGR project and NCBI tools for Arabidopsis
- NCBI annotation of Col-CC
- What's Next

NCBI Genome Resources Workshop

Monday January 15, 2024, 4:00 – 6:10 pm, Palm 8

Time	Topic
4:00 – 4:20	SRA: Submission Improvements and Accelerating Discoveries through Lighter Data *Yuriy Skripchenko*
4:20 – 4:40	Lean, Mean, Genome Cleaning Machine: Removing Contamination with FCS-GX *Eric Tvedte*
4:40 – 5:00	GenBank Submission: How Do I Submit Diploid Genome Assemblies to NCBI? *Shelby Bidwell*
5:00 – 5:20	Advances in Eukaryotic Annotation at NCBI *Vamsi Kodali*
5:20 – 5:40	Using NCBI Datasets to Easily Gather Data from across NCBI Databases *Sally Chang*
5:40 – 6:00	The NIH Comparative Genomics Resource (CGR) *Terence Murphy*

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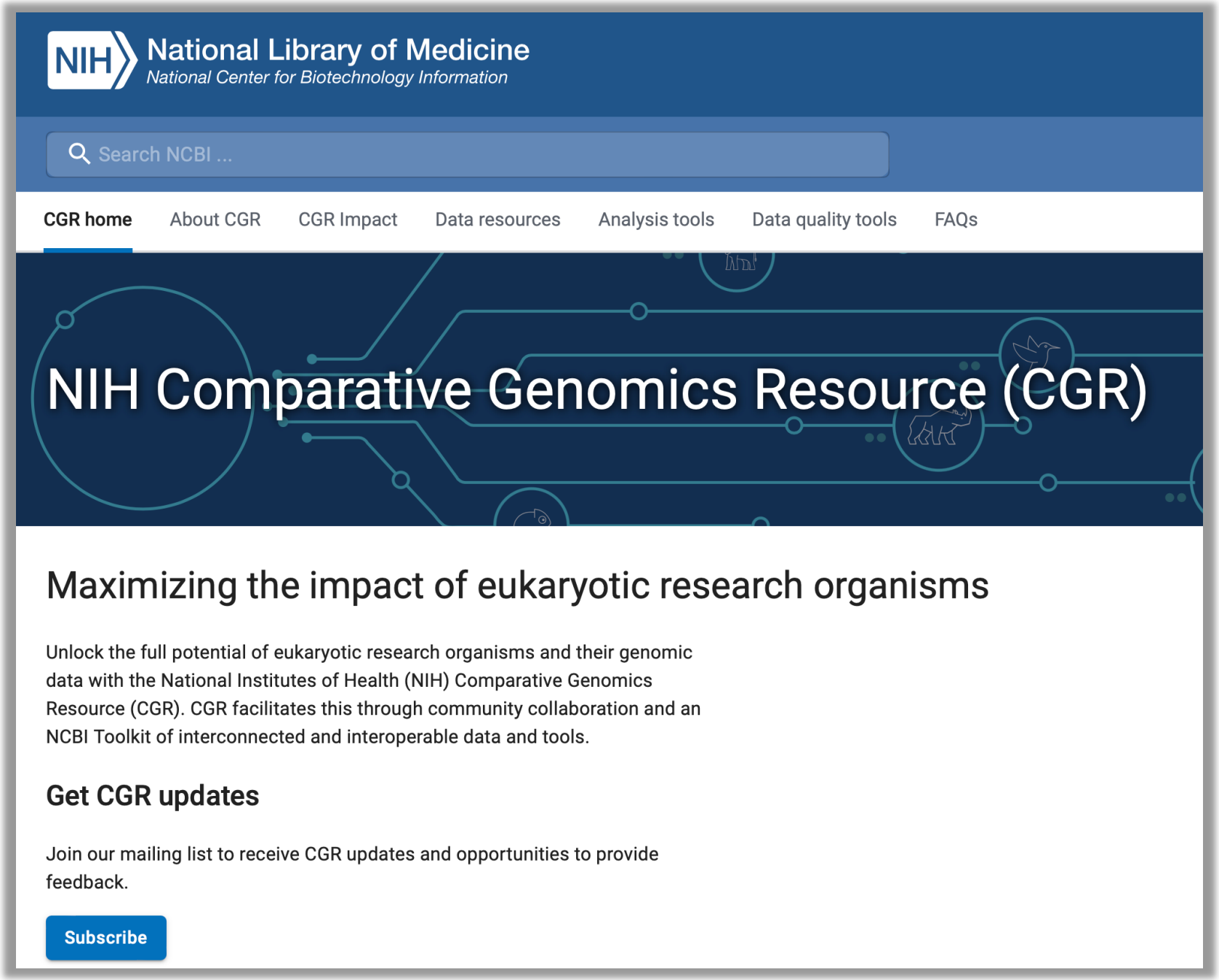
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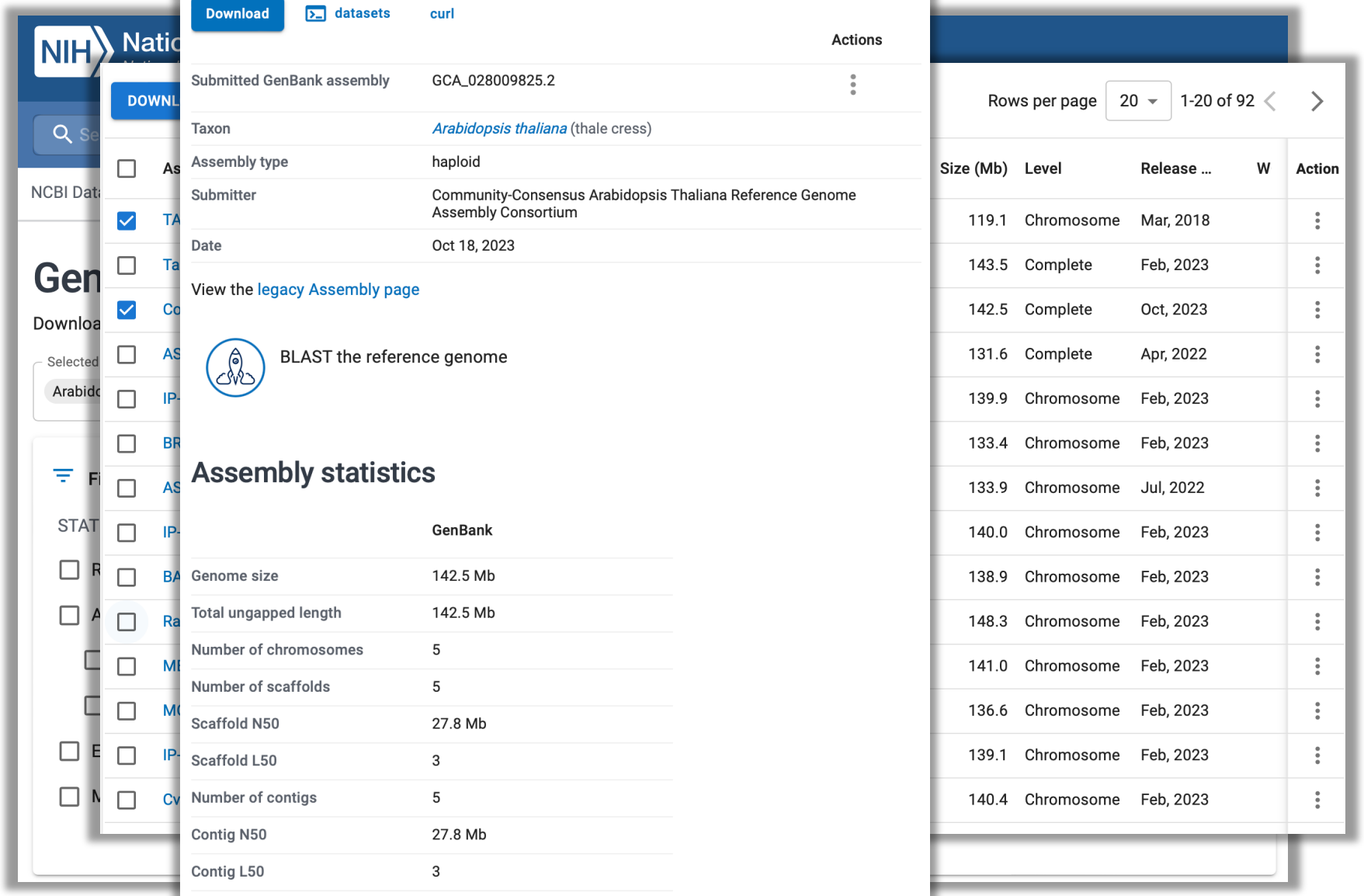
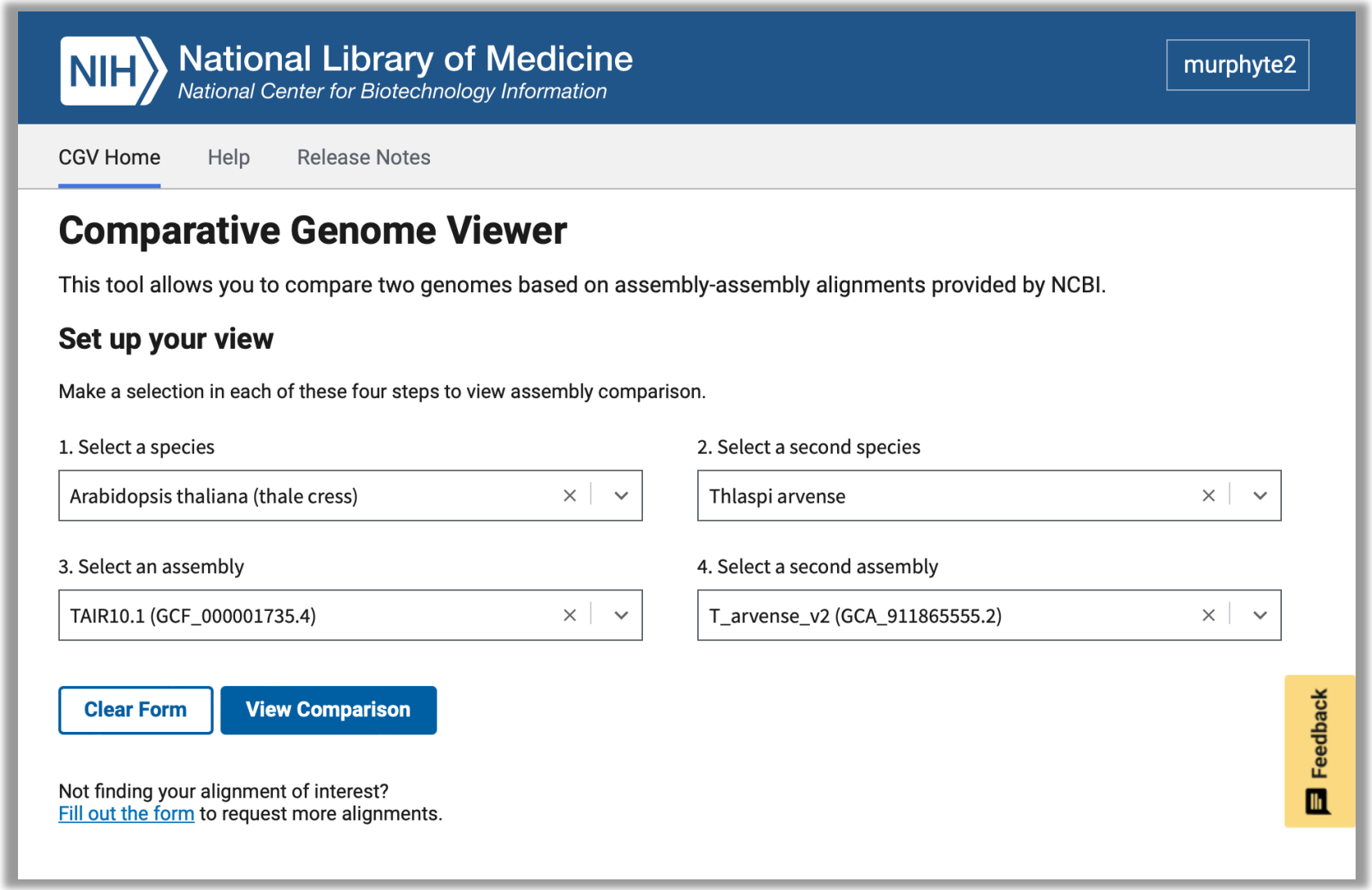


Comparative Genome Viewer (CGV)

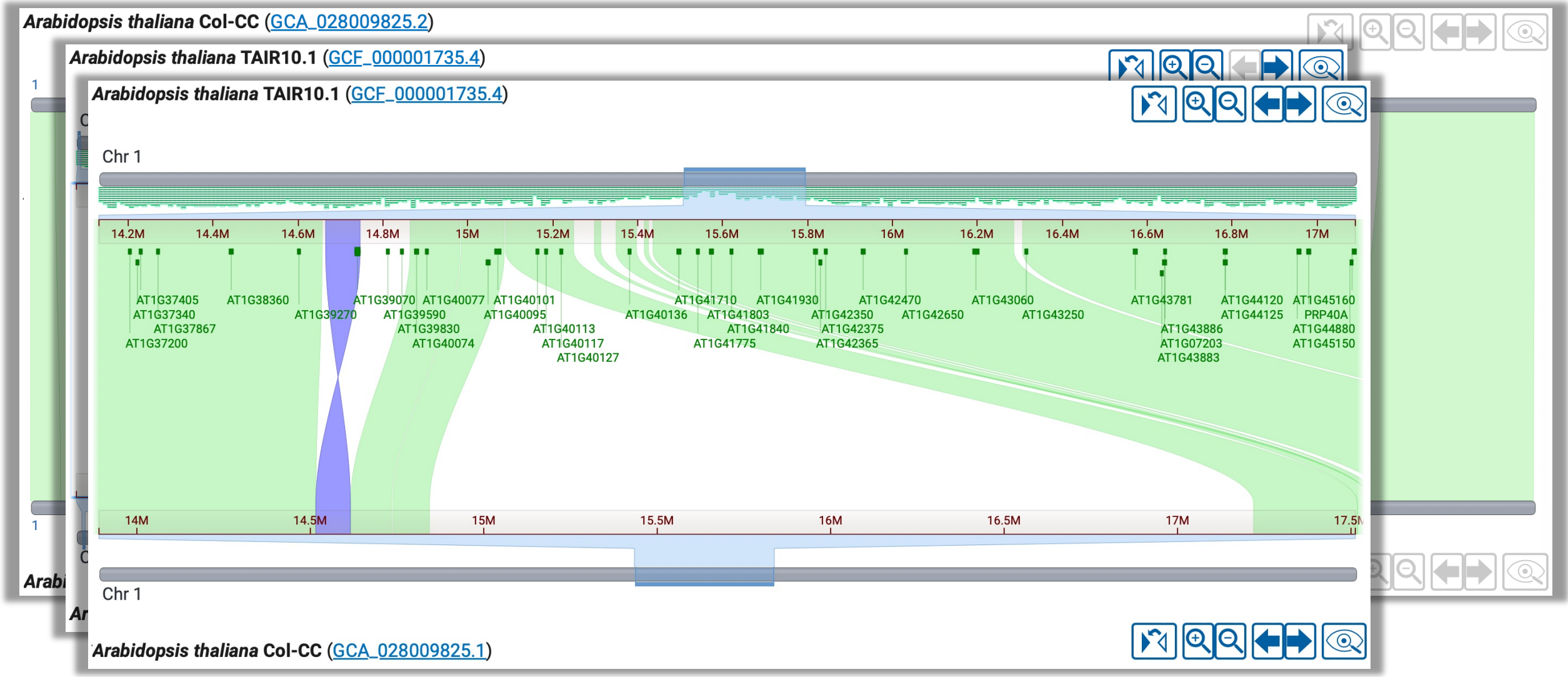
- Compare genomes through assembly alignments
- Over 700 alignments for over 300 species!
- Preprint: PMC10705539

https://www.ncbi.nlm.nih.gov/genome/cgv/



CGV: TAIR10 vs Col-CC

First Pass	Total
Col-CC (Current) Coverage: 88.74%	Col-CC (Current) Coverage: 89.58%
TAIR10.1 (Previous) Coverage: 99.03%	TAIR10.1 (Previous) Coverage: 99.32%
Percent Identity: 98.64%	Percent Identity: 98.61%



Outline

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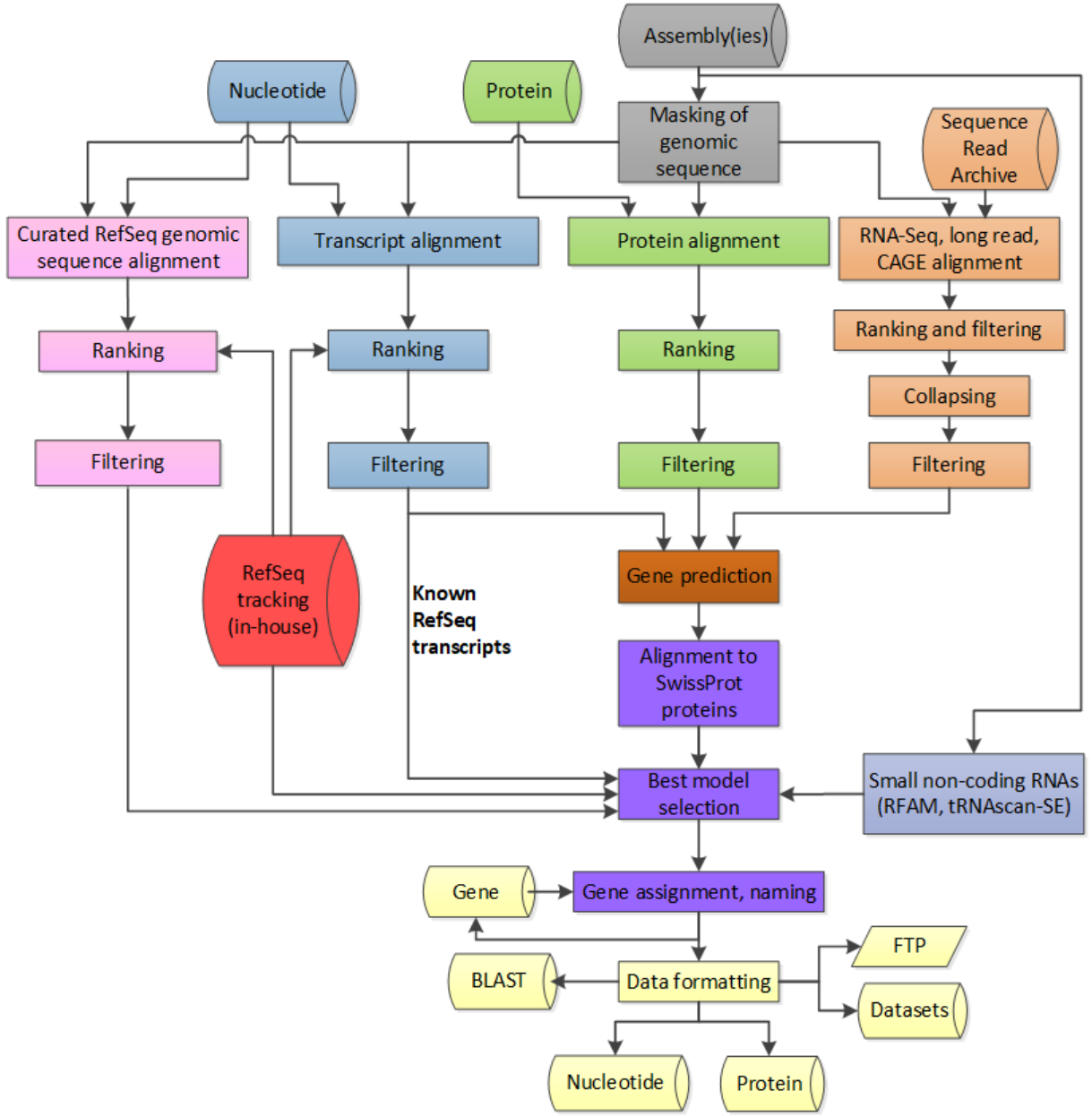
EGAP: Eukaryotic Genome Annotation Pipeline

Used by NCBI to annotate ~1100 species

Evidence used for gene prediction:

- ✓ ESTs
- ✓ cDNAs
- ✓ Same and cross-species proteins
- ✓ RNA-Seq
- ✓ PacBio IsoSeq, ONT transcriptomes
- ✓ CAGE
- ✓ PhyloCSF (for targeted use)



NCBI EGAP annotation

- Software: EGAP v10.1
- Annotation Name: GCA_028009825.1–RS_2023_03 (internal release)

Input type	amount	notes
Oxford Nanopore	41 runs, 61M reads	~90% identity/coverage
PacBio	33 runs, 17M reads	99+% identity/coverage
Illumina	332 runs, 9.3B reads	Mostly unpaired, unstranded
CAGE	52 runs, 3.4M reads	63% aligned
Transcripts	195k cDNA + 1.5M ESTs	99+% identity/coverage
Proteins	• TAIR A. thaliana annotation • Subset of Brassicaceae proteins	

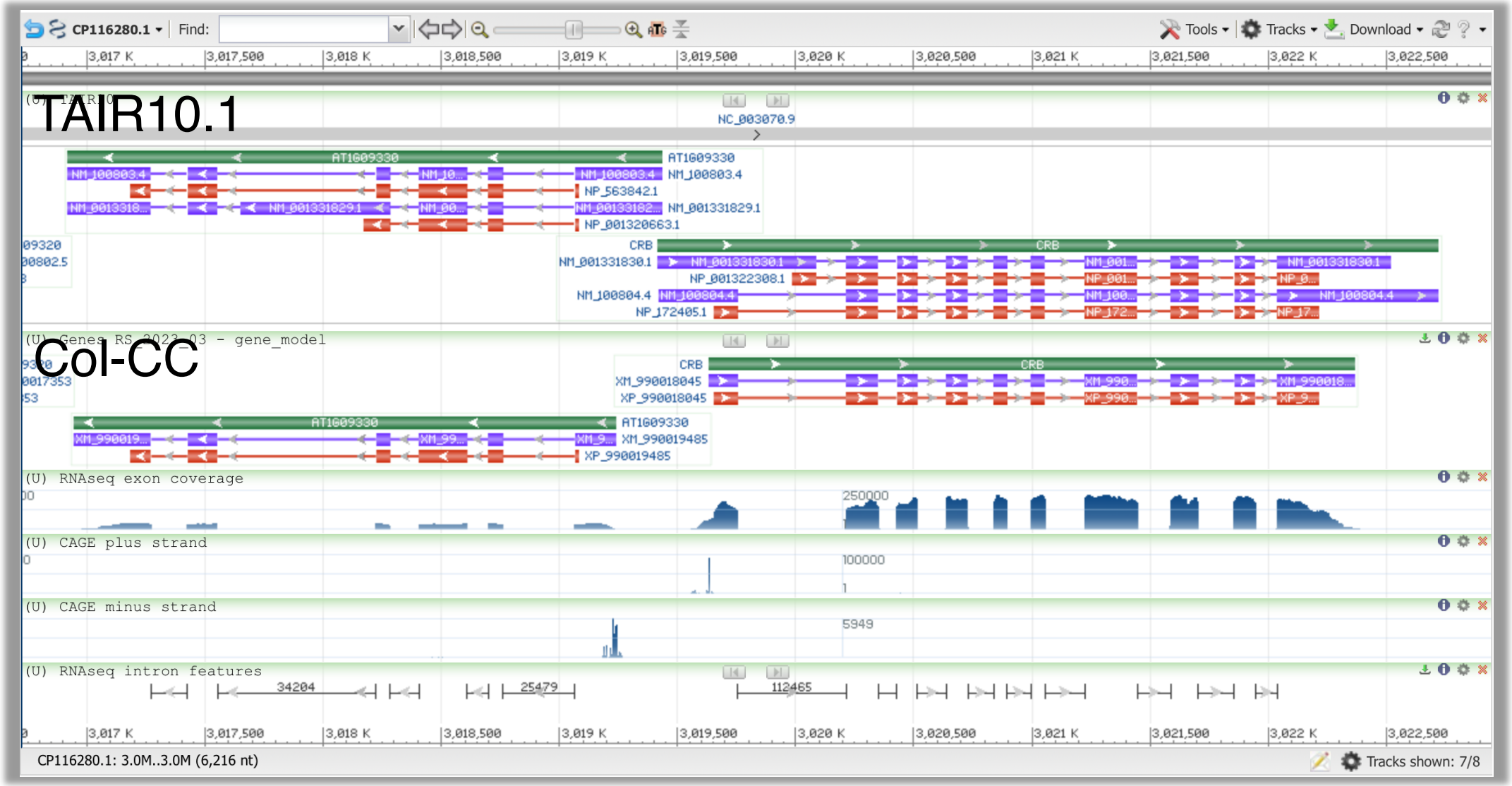
Gene counts

Gene biotype	source	Col-CC	TAIR10.1
Protein-coding	Gnomon	25,565	27,444
with alt transcripts		5,574	10,701
with major corrections		570	
Pseudogene	Gnomon	2,337	4,842
lncRNA	Gnomon	833	3,480
rRNA (v1 assembly)	Rfam	3441	4
snRNA	Rfam	62	82
snoRNA	Rfam	578	287
tRNA	tRNA-scan	545	625
miRNA	previous	325	325
Antisense	previous	79	79

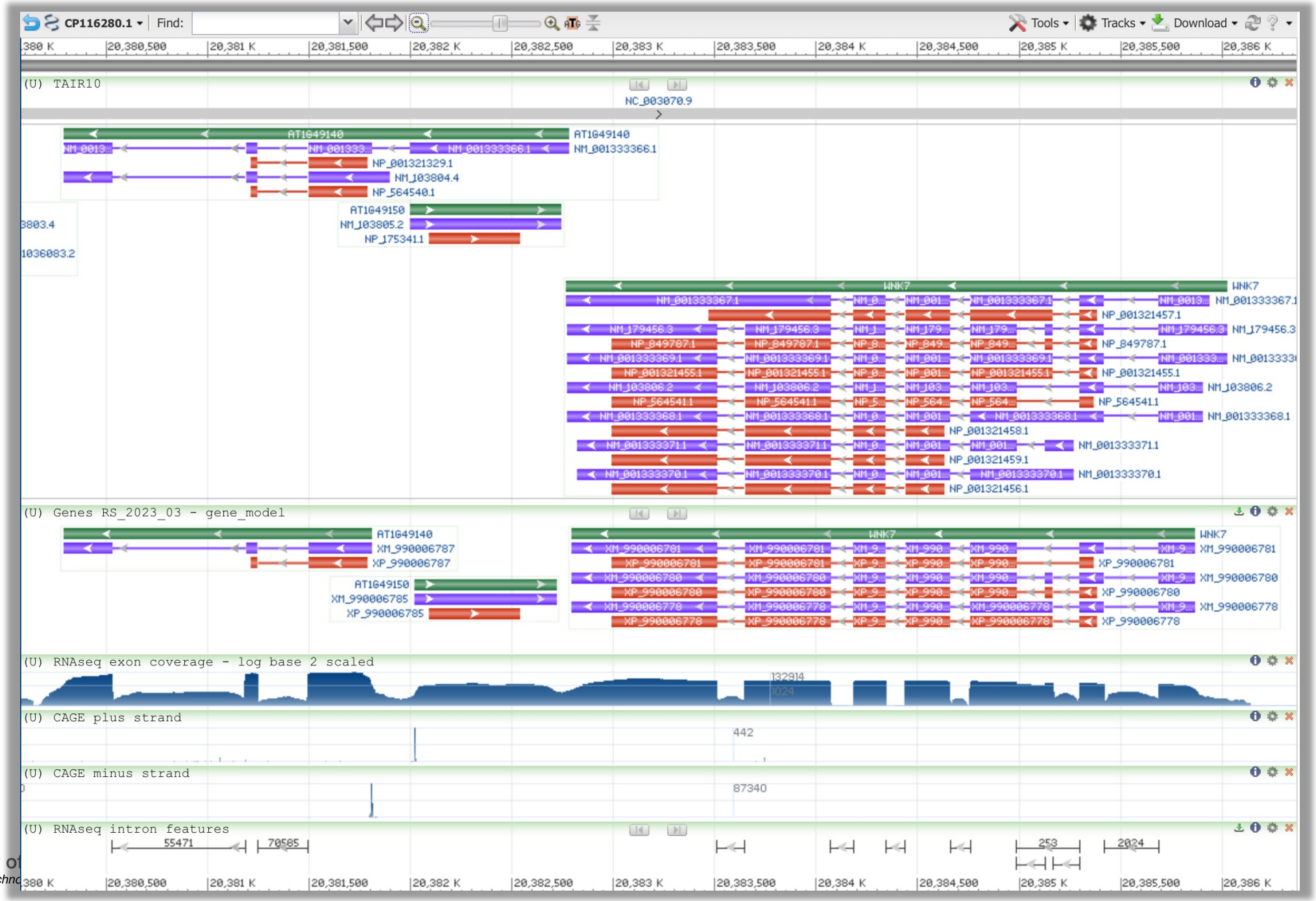
BUSCO:

- TAIR10.1: C:99.8%(S:98.7%,D:1.1%),F:0%,M:0.2%
- RS_2023_03: C:98.8%(S:97.7%,D:1.2%),F:0%,**M1.1%**

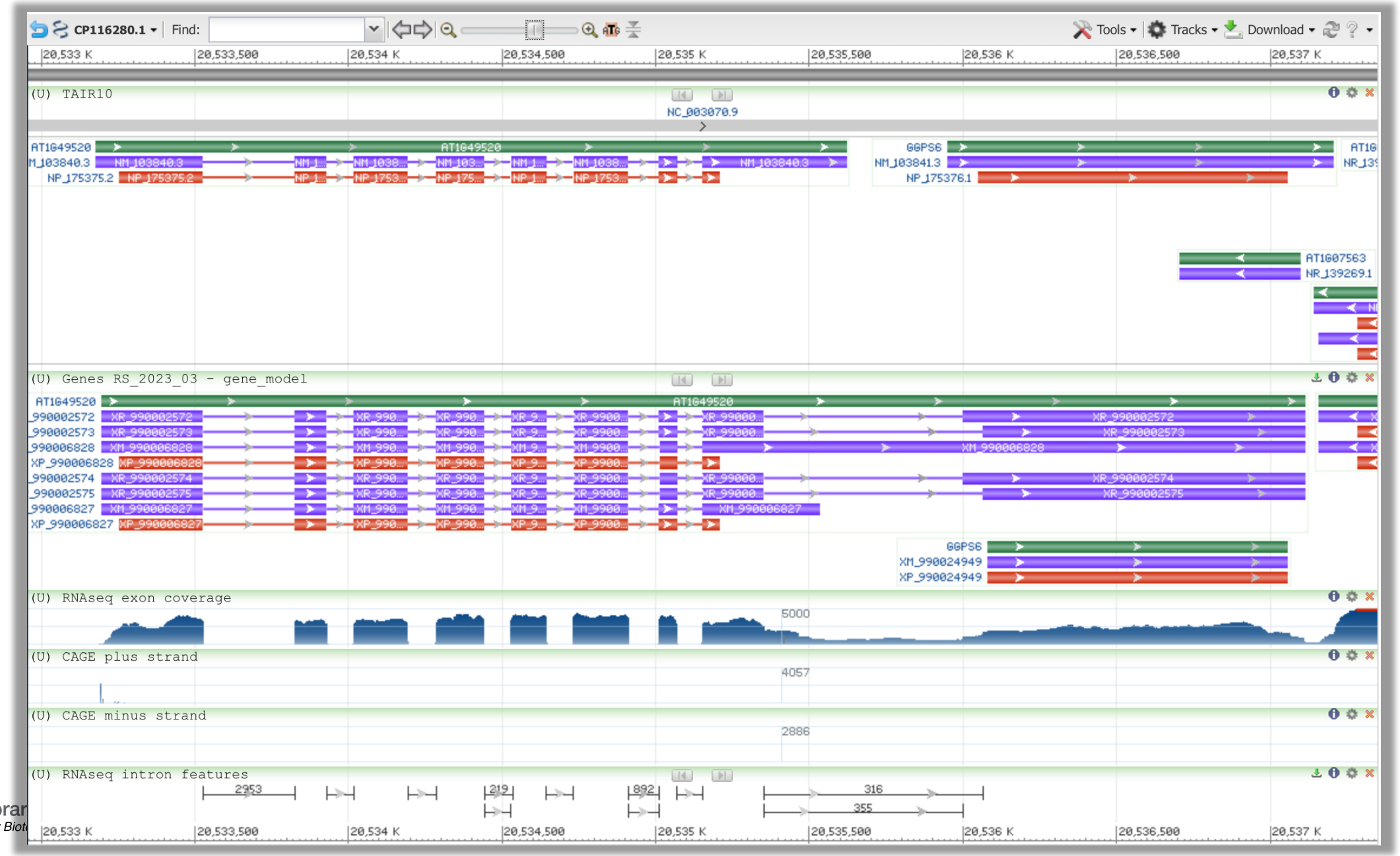
Chr1: AT1G09330 and CRB



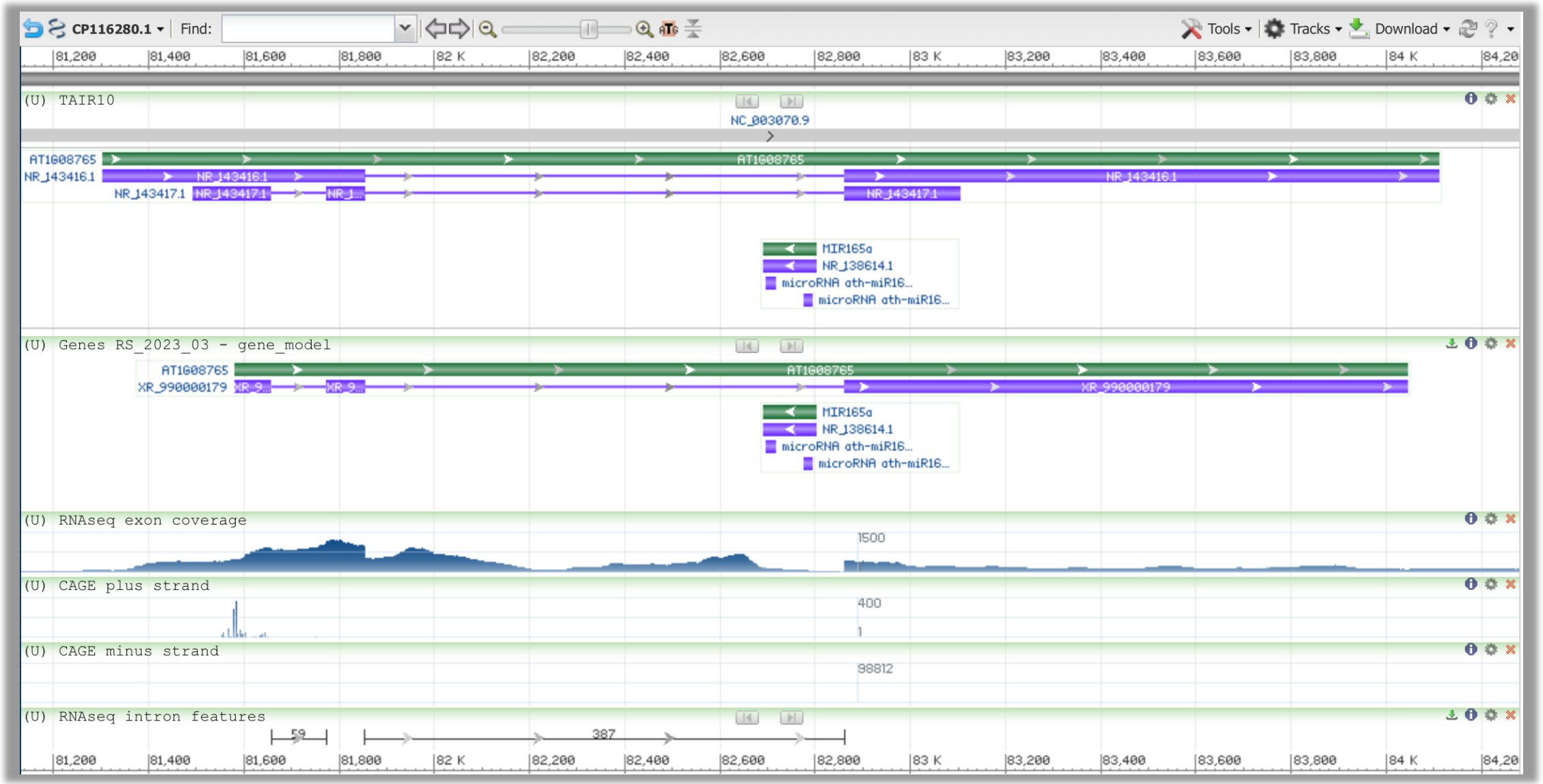
WNK7



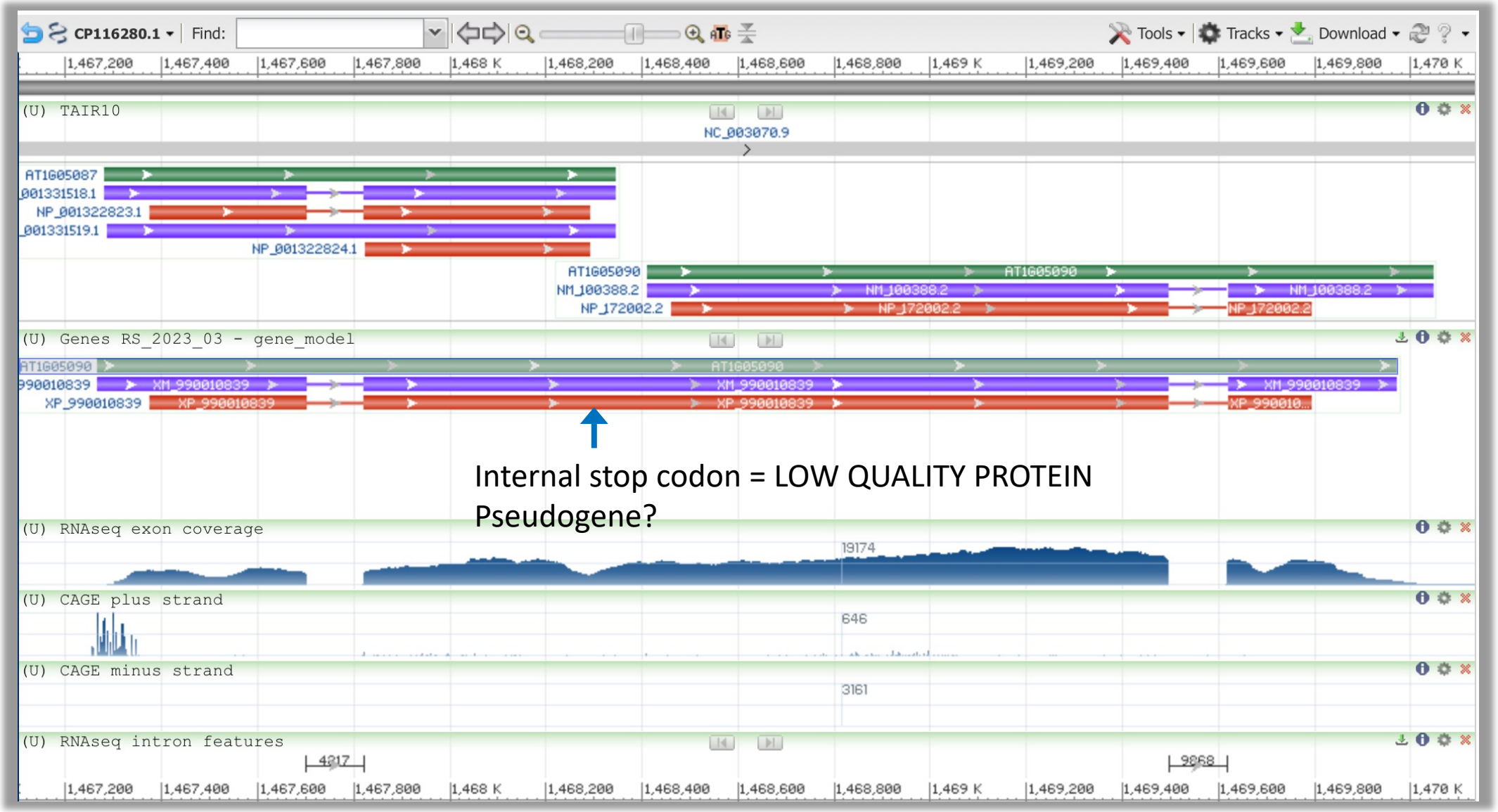
AT1G49520 and GGPS6



AT1G08765 and MIR165a



AT1G05087 and AT1G05090



What's Next

- Review and refinement
  - Major differences vs TAIR10.1
  - Low quality proteins
- Merge other annotations and shift to adjust for added NORs
- Validation and GenBank submission
- Adoption:
  - Incorporate into NCBI RefSeq
  - Switch to Col-CC in TAIR (TAIR11?)
  - Add additional data (expression, variation, etc) to the new reference
- Get back to the real science!

Thank you

This research was supported by the Intramural Research Program of the NIH, National Library of Medicine.

RefSeq/Gene

Shashi Pujar
Tamara Goldfarb
Diana Haddad
John Jackson
Vinita Joardar
Kelly McGarvey
Michael Murphy
Barbara Robbertse
Brian Smith-White
Pooja Strope
Anjana Vatsan
David Webb

EGAP

Francoise Thibaud-Nissen
Wratko Hlavina
Avi Kimchi
Jinna Hoffman
Vamsi Kodali
Patrick Masterson
Eyal Mozes
Robert Smith
Alexandre Souvorov
Olga Ermolaeva
Craig Wallin

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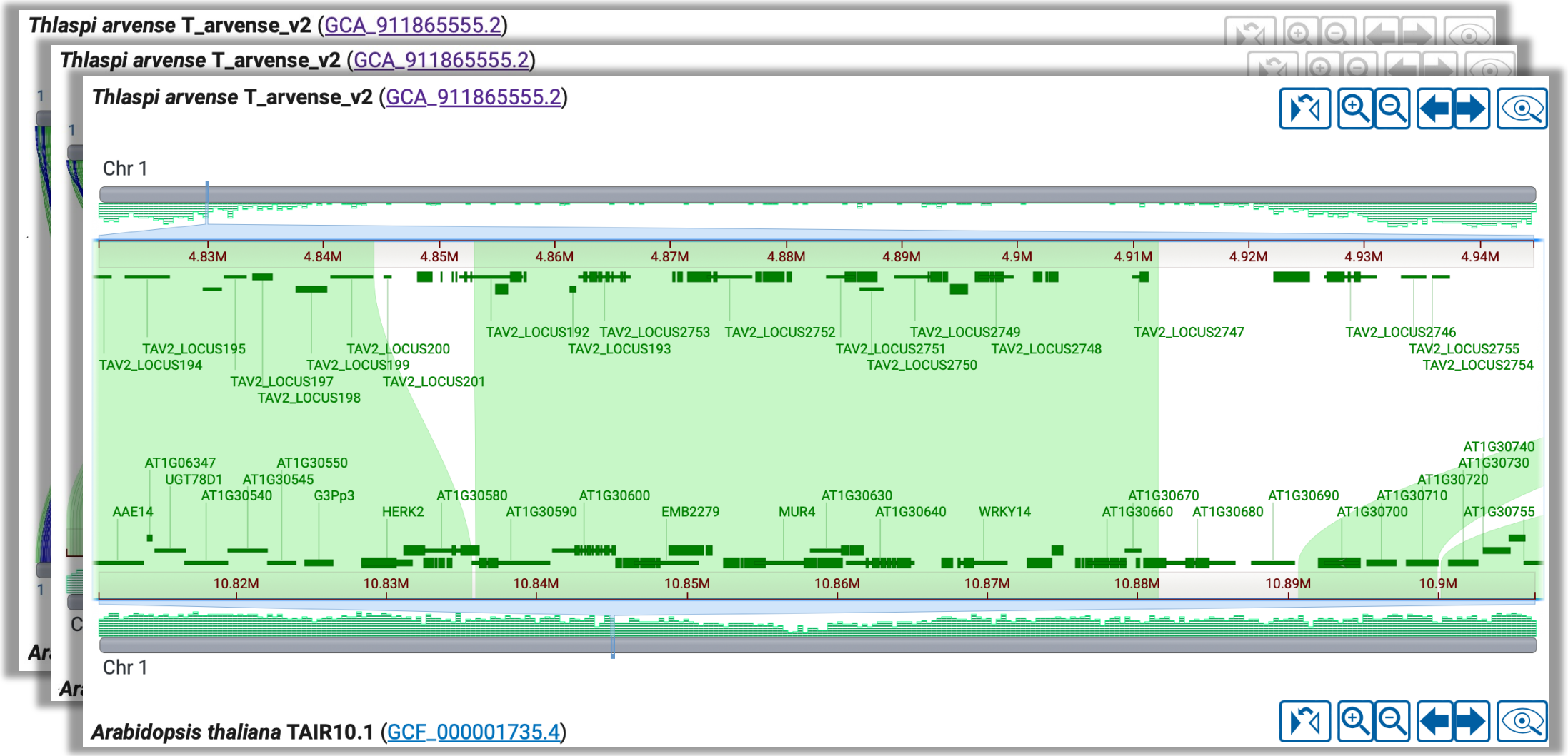
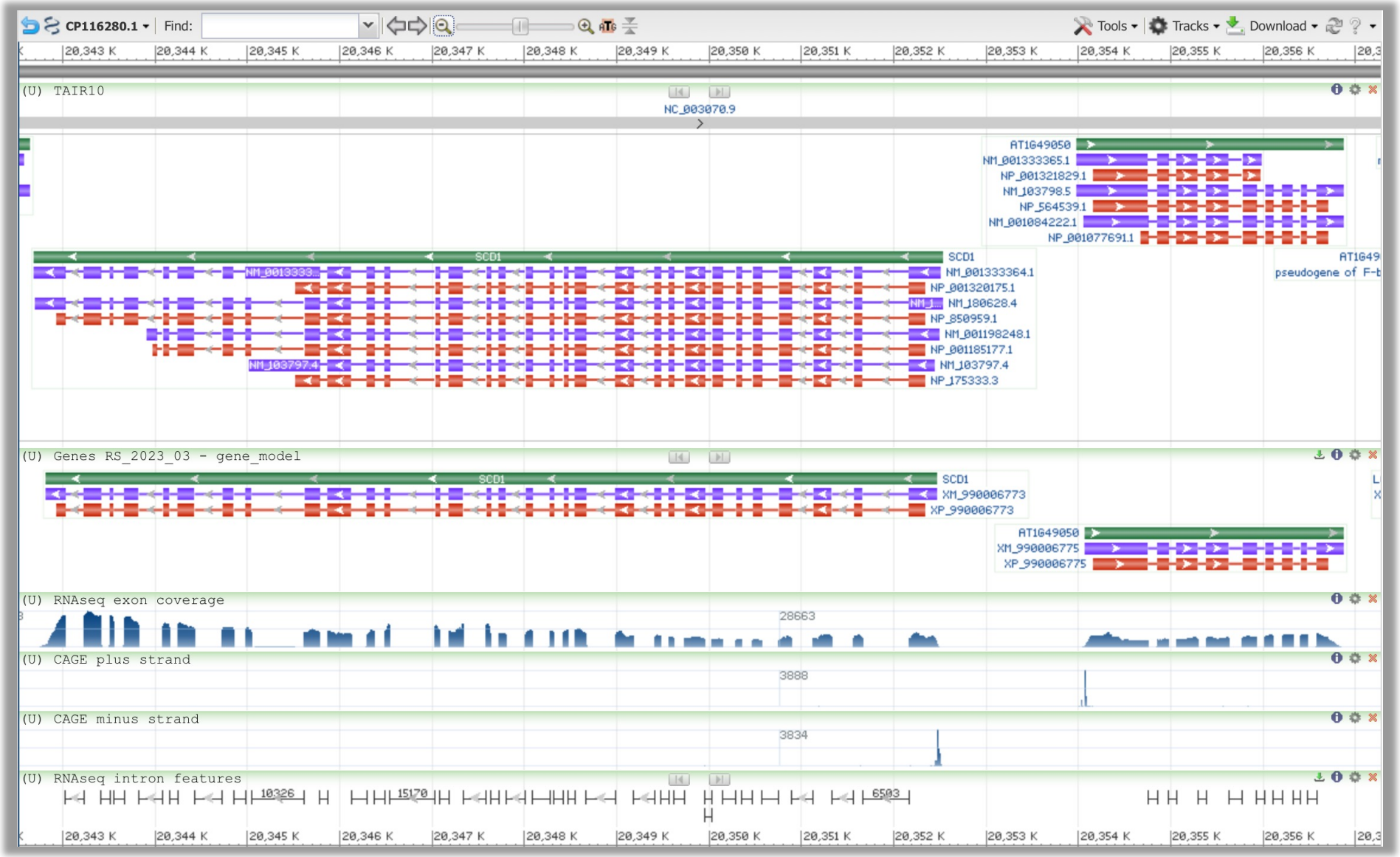
CGV: Arabidopsis vs Thlaspi

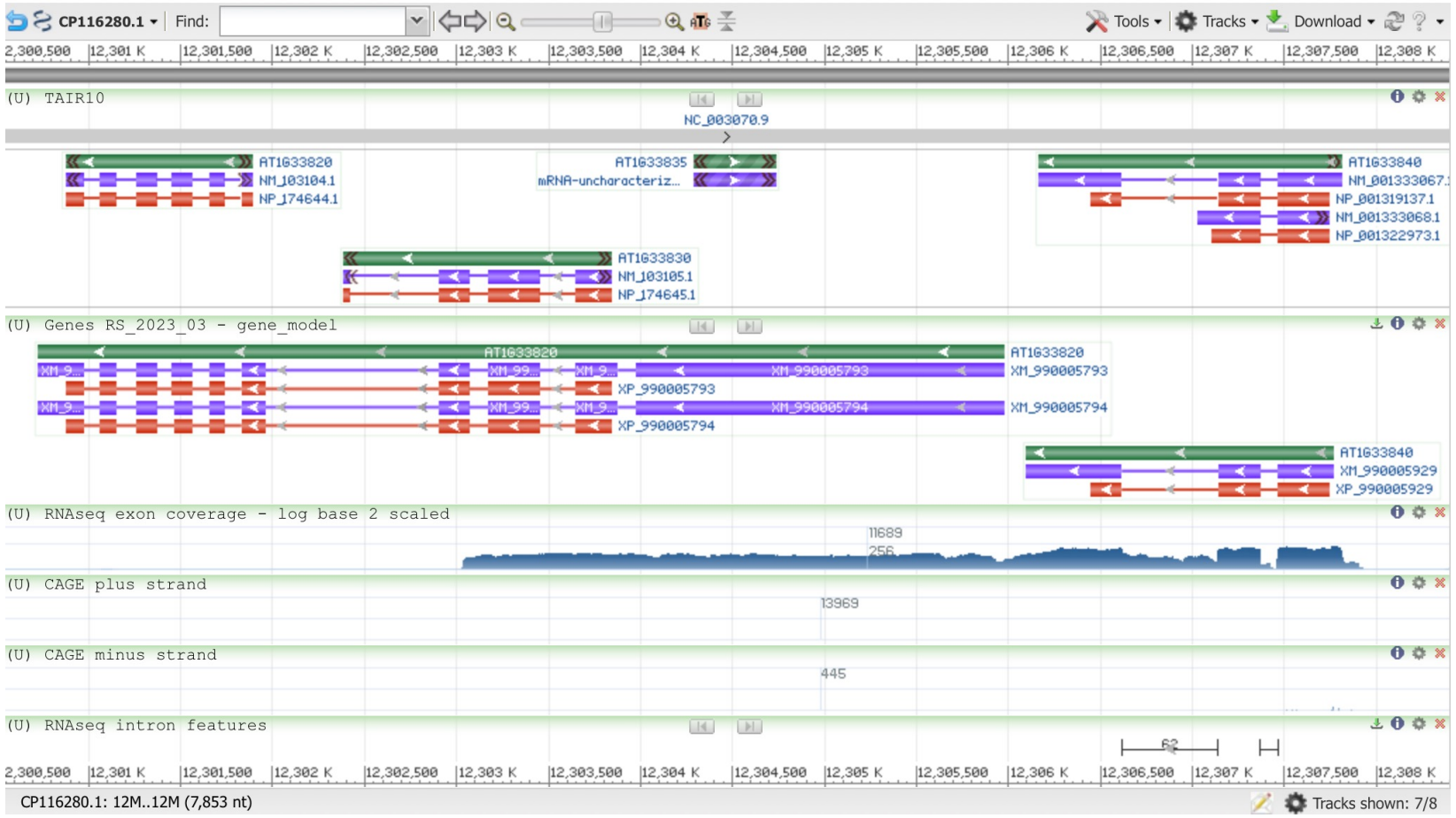


Chr1: AT1G48870

Chr1: SCD1 and AT1G49050



AT1G33820 and AT1G33840



AT1G36920 and AT1G36925