Article Title

Dennis Psaroudakis ¹, Ha Vu ¹, Colleen Yanarella ¹, Steven Cannon ¹, Darwin Campbell ¹, Parnal Joshi ¹, Iddo Friedberg ^{1,4}, Kokulapalan Wimalanathan ^{1,2}, Carolyn J. Lawrence-Dill ^{1,2,3*}

- ¹ Bioinformatics and Computational Biology, Iowa State University, Ames, IA, USA
- ² Department of Genetics, Development, and Cell Biology, Iowa State University, Ames, IA, USA
- ³ Department of Agronomy, Iowa State University, Ames, IA, USA
- ⁴ Department of Veterinary Microbiology, Iowa State University, Ames, IA, USA

Correspondence*:

Carolyn J. Lawrence-Dill

triffid@iastate.edu

2 ABSTRACT

- 3 Abstract length and content varies depending on article type. Refer to http://
- 4 www.frontiersin.org/about/AuthorGuidelines for abstract requirement and length
- 5 according to article type.

1 INTRODUCTION

7 Hello, how are we doing?

2 METHODS

8 **2.1 Clean up**

- 9 All functional annotation sets were cleaned up the following way (using definitions from the Gene
- 10 Ontology version 2019-07-01):
- 1. Any annotations where the GO accession was marked as obsolete were removed.
- 12 2. Some terms in the GO have 'alternative ids'. When naively removing duplicates, two entries will not
- be recognized as duplicates if they have different accessions pointing to the same GO term. Therefore,
- all GO accessions were changed to their respecitve 'main id' and the dataset was again scanned for
- duplicates.
- Table 1 provides information on the number of annotations that were removed this way from each dataset.
- 17 All further analyses were performed on the cleaned datasets since we assume the user will only be interested
- 18 in still valid and non-redundant functional annotations.

19 2.2 Choosing the right evaluation metric

- 20 A plethora of different metric to evaluate the quality of functional annotation predictions is available using
- 21 different approaches and there seems to be no clear standard yet. TODO: THIS IS WEAK Additionally,

Psaroudakis et al. **Short Title**

Table 1. Number of removed annotations during cleanup.

| Genome | Dataset | Obsolete Annotations | Duplicates |
|--------------------------|---------------|----------------------|------------|
| Arachis hypogaea | GOMAP | 0 | 0 |
| Glycine max | GOMAP | 203 | 0 |
| Hordeum vulgarum | GOMAP | 101 | 0 |
| Medicago truncatula A17 | GOMAP | 0 | 0 |
| Medicago truncatula R108 | GOMAP | 0 | 0 |
| | GOMAP | 111 | 2 |
| Oryza sativa | GoldStandard | 38 | 556 |
| | Gramene61-IEA | 10 | 14 |
| Phaseolus vulgaris | GOMAP | 0 | 0 |
| Triticum aestivum | GOMAP | 285 | 0 |
| Vigna unguiculata | GOMAP | 0 | 0 |
| | GOMAP | 1107 | 70 |
| 7. a. m. a. a. D.72 v.2 | GoldStandard | 1 | 0 |
| Zea mays B73.v3 | Gramene49 | 94 | 2 |
| | Phytozome | 54 | 0 |
| Zea mays B73.v4 | GOMAP | 752 | 83 |
| | GoldStandard | 1 | 23 |
| Zea mays Mo17 | GOMAP | 726 | 77 |
| Zea mays PH207 | GOMAP | 798 | 76 |
| Zea mays W22 | GOMAP | 754 | 82 |

each of the metrics has a different focus and lalala so choosing a metric for quality evaluation is not trivial. When we first published GOMAP (Wimalanathan et al., 2018), we used a modified version of the 23 hierarchical evaluation metrics originally introduced in (Verspoor et al., 2006) because they were simple, 24 clear, and part of an earlier attempt at unifying and standardizing GO annotation comparisons (Defoin-Platel et al., 2011). In the meantime, Plyusnin et al. (2018) have published an approach for evaluating different 26 metrics showing substantial differences within the robustness of different approaches. TODO DESCRIBE THEIR APPROACH We have applied their method on the Gold Standards available to us to determine which evaluation metric is the most appropriate in our case. The results of this analysis can be seen in TODO.

31 We then evaluated our predictions and the other annotation sets using the best performing metrics as well as the one we previously used. TODO 32

3 **RESULTS**

27

29

30

... a quantitative comparison of the datasets in Table.

3.1 **Quality Evaluation**

- TODO If it turns out that our predictions are good with hF but bad with more approriate metrics, 35
- explanation would be that score thresholds for the prediction tools used in the GOMAP pipeline have
- been chosen to maximize this hF value. It now seems reasonable to re-adjust these thresholds to maximize

Psaroudakis et al. Short Title

Table 2. Quantitative metrics of the cleaned functional annotation sets. CC, BF, MP, and A refer to the aspects of the GO: Cellular Component, Biological Function, Molecular Process, and Any/All.

| | | | Annotations ^a Annotated Genes [| | d Genes [% | 6] ^b | Med | Median Ann. per G.c | | | | | | |
|--------------------------|-------|---------------|--|--------|------------|-----------------|-------|---------------------|--------|--------|----|----|----|----|
| Genome | Genes | Dataset | CC | BF | MP | A | CC | BF | MP | A | CC | BF | MP | A |
| Arachis hypogaea | | GOMAP | 153433 | 132944 | 493799 | 780176 | 57667 | 56855 | 67123 | 67124 | 2 | 2 | 6 | 10 |
| Glycine max | | GOMAP | 129215 | 113827 | 417555 | 660597 | 46020 | 47034 | 52871 | 52872 | 2 | 2 | 6 | 11 |
| Hordeum vulgarum | | GOMAP | 88130 | 80282 | 272823 | 441235 | 35237 | 36470 | 39733 | 39734 | 2 | 2 | 5 | 10 |
| Medicago truncatula A17 | | GOMAP | 107362 | 99719 | 364065 | 571146 | 42325 | 43736 | 50443 | 50444 | 2 | 2 | 6 | 10 |
| Medicago truncatula R108 | | GOMAP | 112343 | 108031 | 382322 | 602696 | 40332 | 50220 | 55706 | 55706 | 1 | 2 | 5 | 9 |
| _ | | GOMAP | 72780 | 64685 | 248700 | 386165 | 28619 | 29853 | 35824 | 35825 | 2 | 2 | 6 | 9 |
| Oryza sativa | | GoldStandard | 7730 | 11060 | 19378 | 38176 | 5725 | 7383 | 9031 | 11387 | 1 | 1 | 1 | 3 |
| · | | Gramene61-IEA | 14633 | 32787 | 39105 | 86529 | 10771 | 15537 | 16705 | 21446 | 1 | 1 | 1 | 3 |
| Phaseolus vulgaris | | GOMAP | 72005 | 64583 | 229630 | 366218 | 25934 | 25539 | 27432 | 27433 | 2 | 2 | 6 | 11 |
| Triticum aestivum | 100 | GOMAP | 267741 | 218623 | 785960 | 1272324 | 95604 | 98187 | 107890 | 107891 | 2 | 2 | 6 | 10 |
| Vigna unguiculata | 100 | GOMAP | 75867 | 68313 | 243278 | 387458 | 27173 | 27124 | 29772 | 29773 | 2 | 2 | 6 | 11 |
| 0 0 | | GOMAP | 135211 | 87420 | 291251 | 513882 | 34866 | 38073 | 39468 | 39469 | 3 | 2 | 6 | 11 |
| 7 | | GoldStandard | 1565 | 65 | 299 | 1929 | 1548 | 60 | 151 | 1634 | 1 | 0 | 0 | 1 |
| Zea mays B73.v3 | | Gramene49 | 20072 | 31056 | 30089 | 81217 | 11834 | 17991 | 15800 | 21926 | 1 | 1 | 1 | 3 |
| | | Phytozome | 4787 | 19044 | 13100 | 36931 | 4524 | 13728 | 11365 | 16132 | 0 | 1 | 1 | 2 |
| 7 D72 4 | | GÓMAP | 88827 | 82251 | 278719 | 449797 | 36717 | 37337 | 39323 | 39324 | 2 | 2 | 6 | 10 |
| Zea mays B73.v4 | | GoldStandard | 1371 | 63 | 286 | 1720 | 1349 | 58 | 144 | 1428 | 1 | 0 | 0 | 1 |
| Zea mays Mo17 | | GOMAP | 87567 | 79214 | 277787 | 444568 | 33618 | 35105 | 38619 | 38620 | 2 | 2 | 6 | 10 |
| Zea mays PH207 | | GOMAP | 90617 | 85500 | 288677 | 464794 | 35170 | 36762 | 40556 | 40557 | 2 | 2 | 6 | 10 |
| Zea mays W22 | | GOMAP | 95390 | 85039 | 289780 | 470209 | 36987 | 37685 | 40689 | 40690 | 2 | 2 | 6 | 10 |

 $^{^{}a}$ How many annotations in the CC, BF, and MP aspect does this dataset contain? A = How many in total? A = CC + BF + MP

Table 3. Quality evaluation of the used GO annotation sets.

| Genome | Dataset | SimGIC2 score |
|-----------------|---------------------------------|----------------------------------|
| Oryza sativa | GOMAP Gramene61-IEA | 0.253680 0.330437 |
| Zea mays B73.v3 | GOMAP Gramene49 Phytozome | 0.052182 0.091475 0.028721 |
| Zea mays B73.v4 | GOMAP | 0.050760 |

a different metric which will likely result in a drop in hF score but increase in other metrics. Again emphasizes the importance of choosing the right evaluation metric.

REFERENCES

- Defoin-Platel, M., Hindle, M. M., Lysenko, A., Powers, S. J., Habash, D. Z., Rawlings, C. J., et al. (2011). AIGO: Towards a unified framework for the Analysis and the Inter-comparison of GO functional
- 42 annotations. *BMC Bioinformatics* doi:10.1186/1471-2105-12-431
- Plyusnin, I., Holm, L., and Töoröonen, P. (2018). Novel Comparison of Evaluation Metrics for Gene
 Ontology Classifiers Reveals Drastic Performance Differences. *bioRxiv*, 427096doi:10.1101/427096
- Verspoor, K., Cohn, J., Mniszewski, S., and Joslyn, C. (2006). A categorization approach to automated
 ontological function annotation. *Protein Science* doi:10.1110/ps.062184006
- 47 Wimalanathan, K., Friedberg, I., Andorf, C. M., and Lawrence-Dill, C. J. (2018). Maize GO Annotation-
- 48 Methods, Evaluation, and Review (maize-GAMER). Plant Direct 2, e00052. doi:10.1002/pld3.

49 52

Frontiers 3

b How many genes in the genome have at least one GO term from the CC, BF, MP aspect annotated to them? A = How many at least one from any aspect? $(A = CC \cup BF \cup MP)$

^c Take a typical gene that is present in the annotation set. How many annotations does it have in each aspect? A = How many in total? Ask your favorite statistician why $A \neq CC + BF + MP$