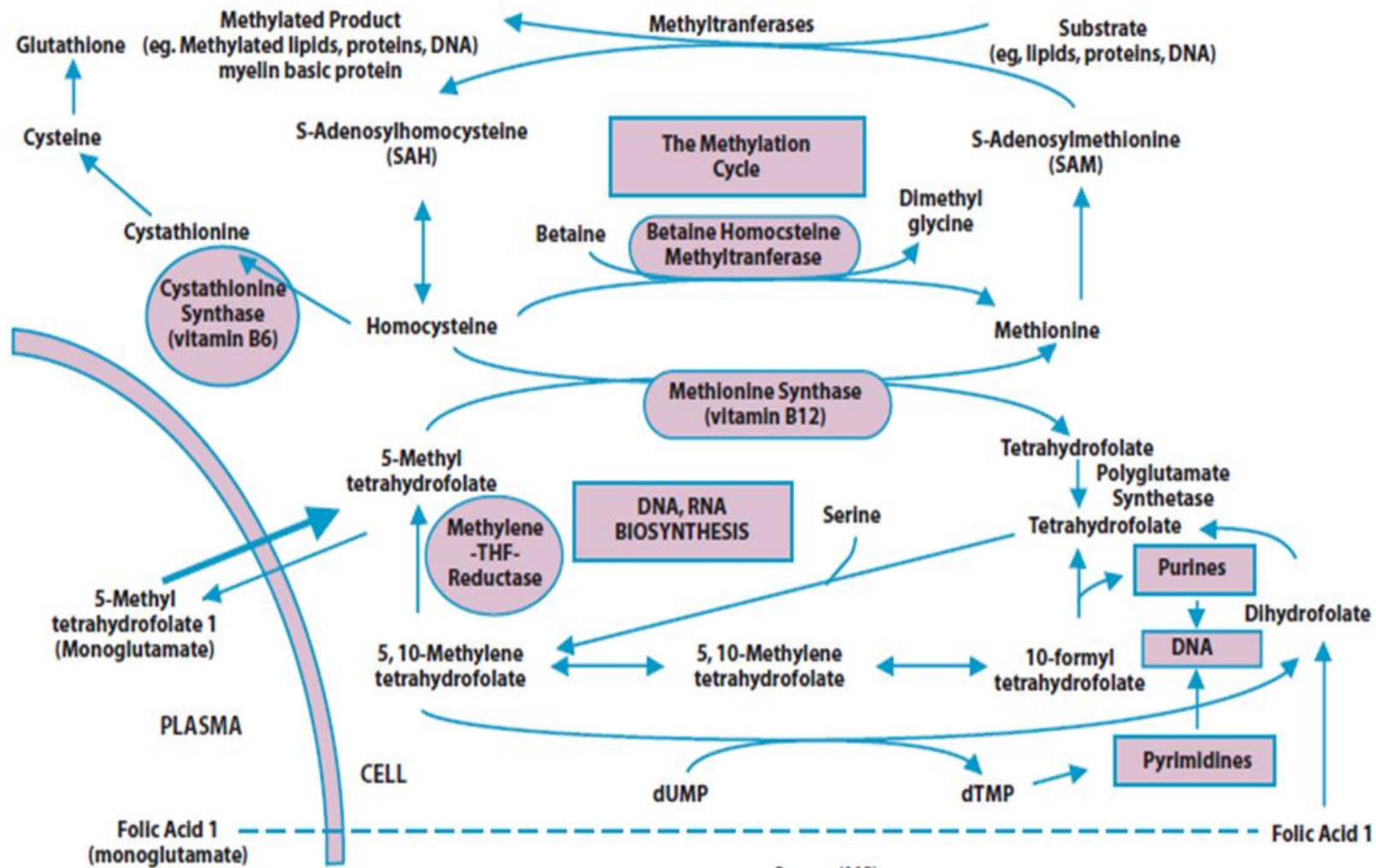


# Breeding for Nutritional Enhancement in Potato: Exploring Vitamin B9 Diversity in Wild and Cultivated Potatoes.

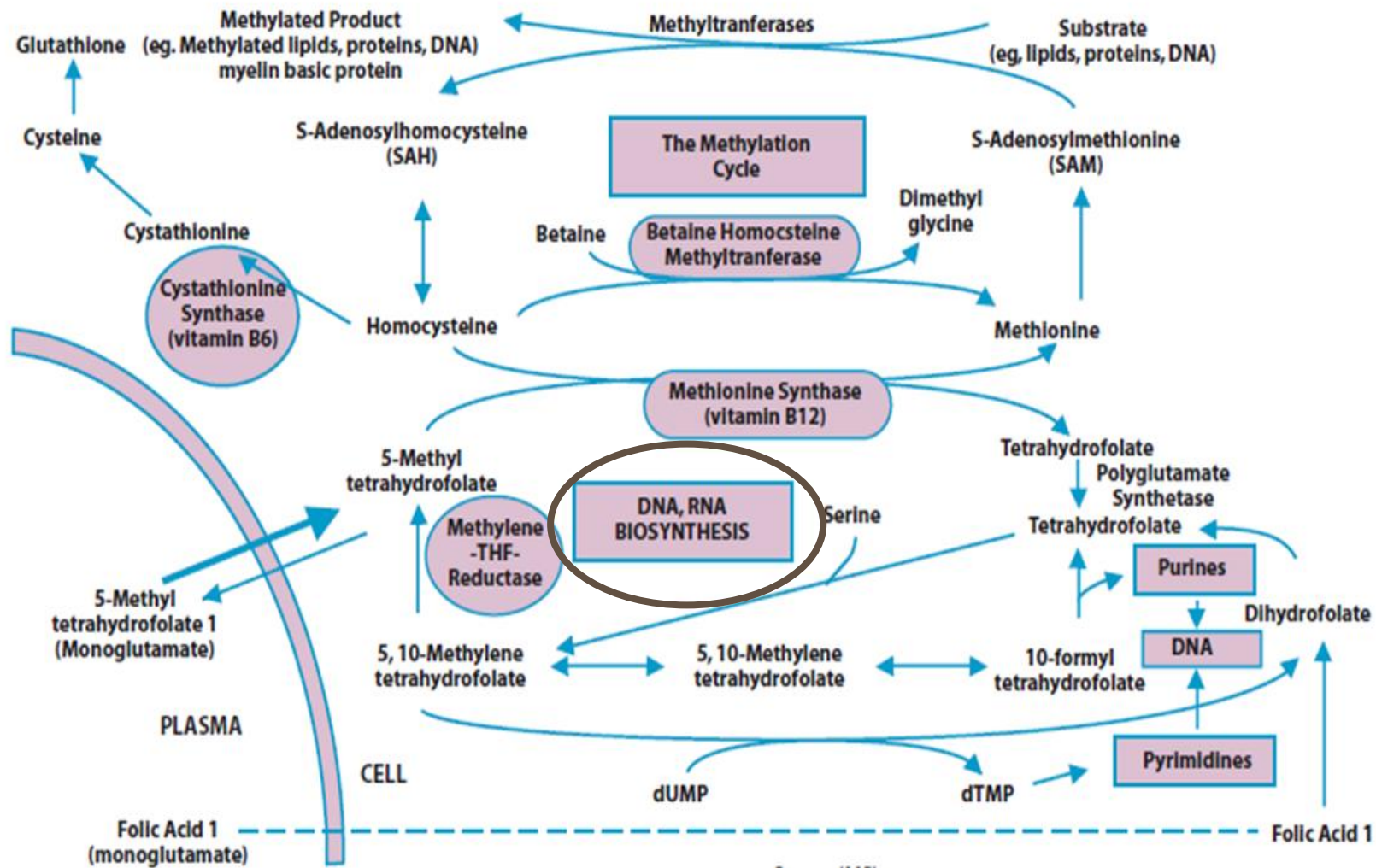
Bruce Reid Robinson II  
Department of Crop and Soil Sciences  
Hermiston Agricultural Research and Extension Center  
Oregon State University

# Folate – Water Soluble Vitamin B9



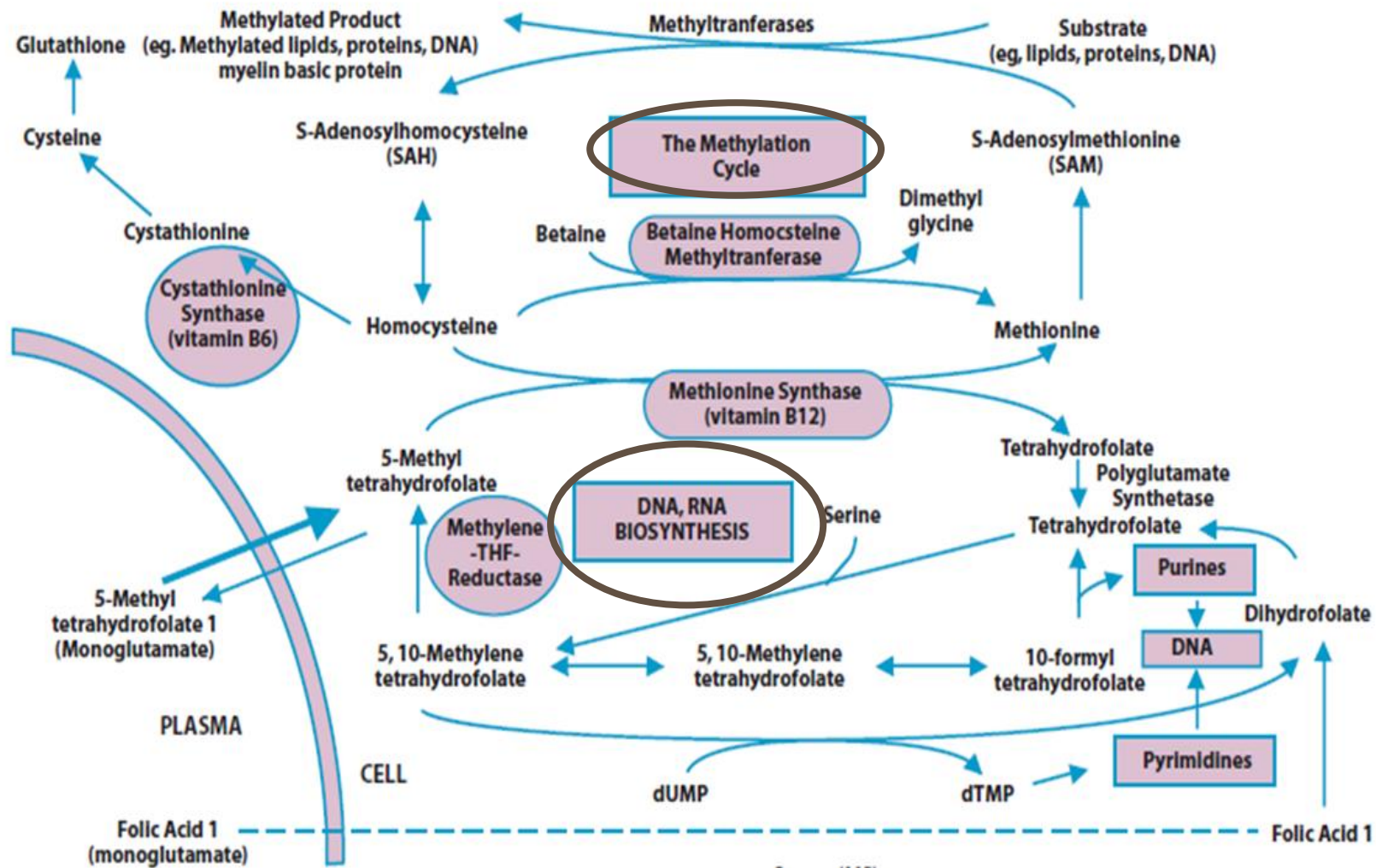
Source: Scott & Weir 1998

# Folate – Water Soluble Vitamin B9



Source: Scott & Weir 1998

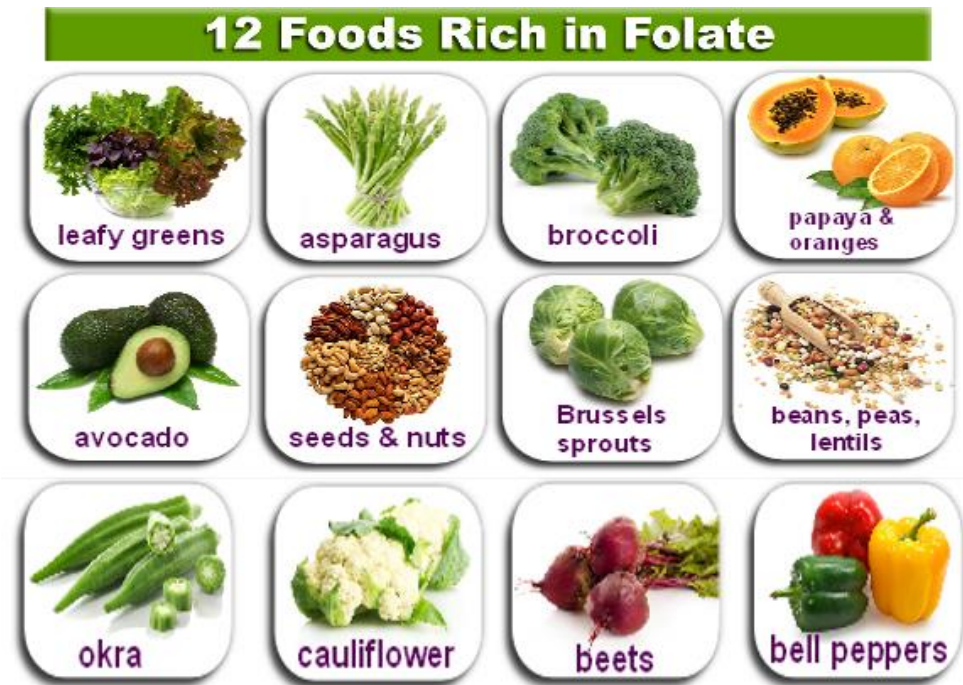
# Folate – Water Soluble Vitamin B9



Source: Scott & Weir 1998

# Folate Sources and Deficiency

- Folate deficiency has been linked to:
  - Neural Tube Defects (NTDs) such as spina bifida and anencephaly
  - Cardiovascular diseases
  - Stroke
  - Anemia
  - Development of certain types of cancers
  - Impaired cognitive performance



# Biofortification Through Breeding

- Has additional advantages compared to industrial fortification alone:
  - Cost-effective
  - Sustainable
  - Can impact areas that lack the political will, infrastructure, and money to utilize current fortification practices
- Requires that the target of the biofortification is a staple crop
- Requires that this crop demonstrates natural variation, stability, and heritability for the trait you are breeding for

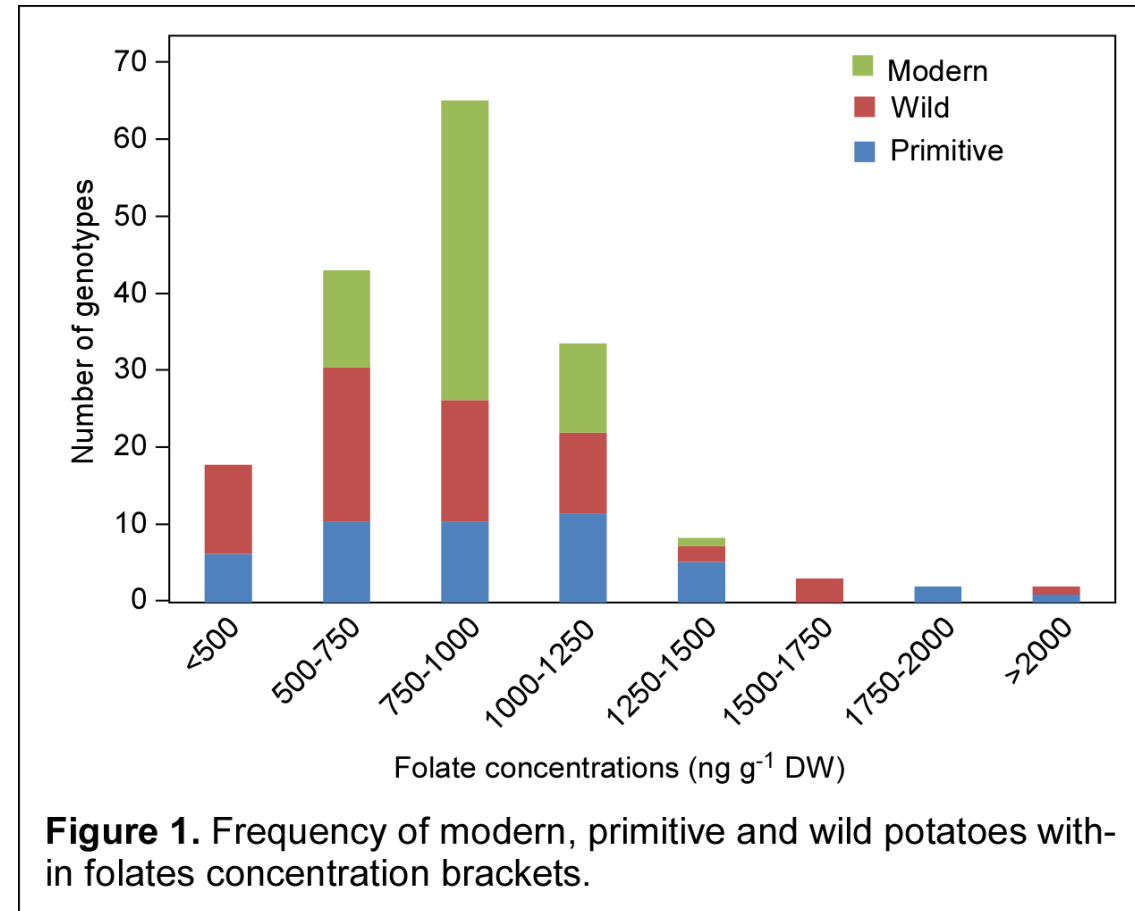
## Why Potatoes?

- Currently a 148g serving of potato (a medium sized potato) only provides about 6% of the 400 $\mu$ g RDA of folate
- There are approximately 200 tuber bearing *Solanum* species representing enormous genetic diversity
- Exploiting this variation between species is the paradigm for modern crop improvement, yet potatoes have not been a major focus of biofortification studies until now



# Folate Content Variability in Potatoes

- Wild type and primitive cultivated species show the greatest range of folate content
- Some demonstrate significantly higher levels of folate over modern cultivars





# Potato Materials – Wild and Primitive Cultivated Species

- 285 individual plants from 95 accessions representing 10 species evaluated with Russet Burbank as control
- Accessions were obtained from the U.S. Potato Genebank



# Potato Materials – Wild and Primitive Cultivated Species

## Harvested Selections:

1. *S. acuale* (3 accessions)
2. *S. boliviense* (25 accessions)
3. *S. candolleanum* (3 accessions)
4. *S. chacoense* (2 accessions)
5. *S. circaefolium* (3 accessions)
6. *S. demissum* (3 accessions)
7. *S. microdontum* (3 accessions)
8. *S. okadae* (3 accessions)
9. *S. tuberosum subsp. andigenum* (9 accessions)
10. *S. vernei* (23 accessions)



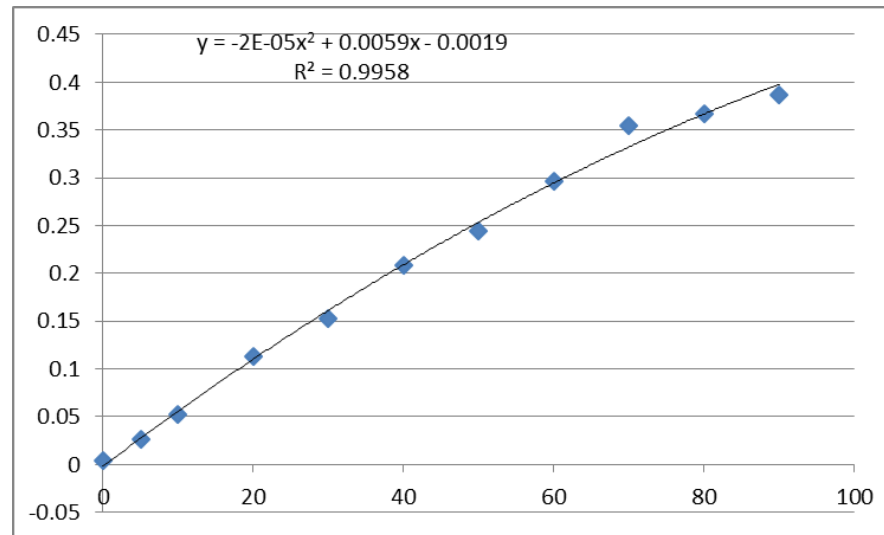
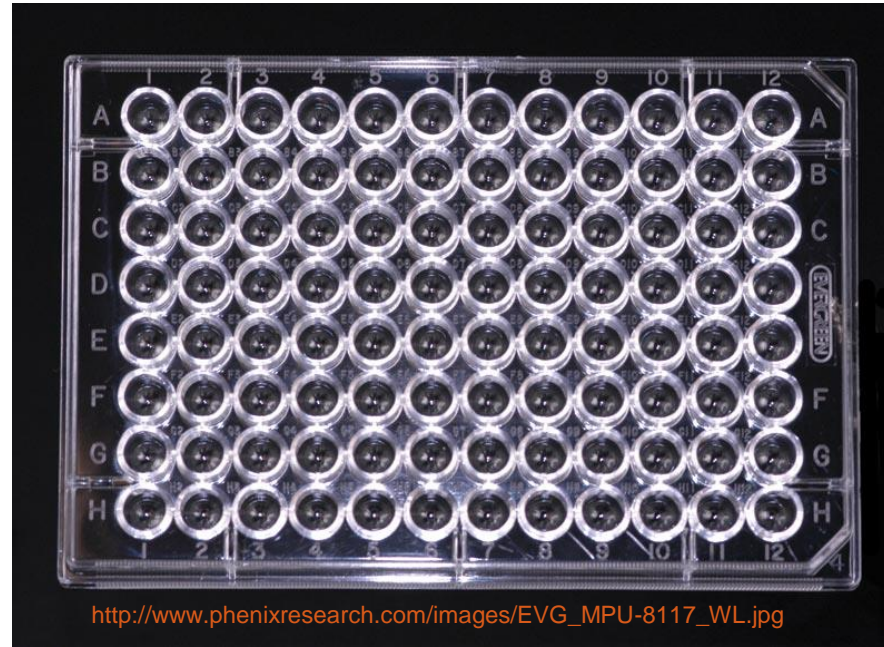
# Tri-Enzyme Extraction Method

- General Principle: Folate species must be released from food matrices and processed without degrading the sample so determination can be performed
- HEPES/CHES buffer, protease,  $\alpha$ -amylase, and conjugase allow for this with reasonable throughput

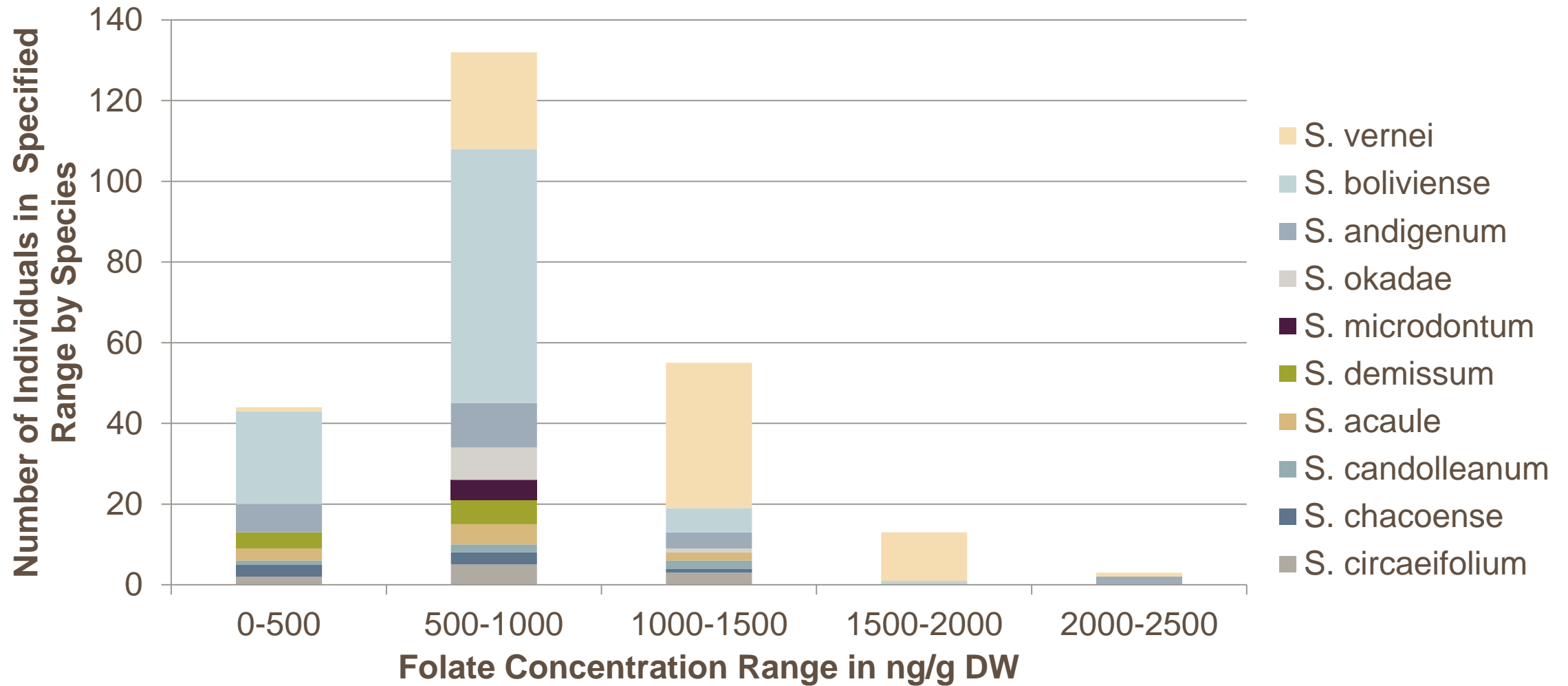
Freeze-dried Tuber Sample  
Homogenize in HEPES/CHES Buffer  
Heat (10min at 100° C)  
Ice Bath  
Incubate with Protease (2hrs at 37° C)  
Heat (5 min at 100° C)  
Ice Bath  
Incubate with  $\alpha$ -amylase and conjugase  
(2-3hrs at 37° C)  
Heat (10min at 100° C)  
Ice Bath  
Centrifuge  
Storage at -80° C

# Folate Determination

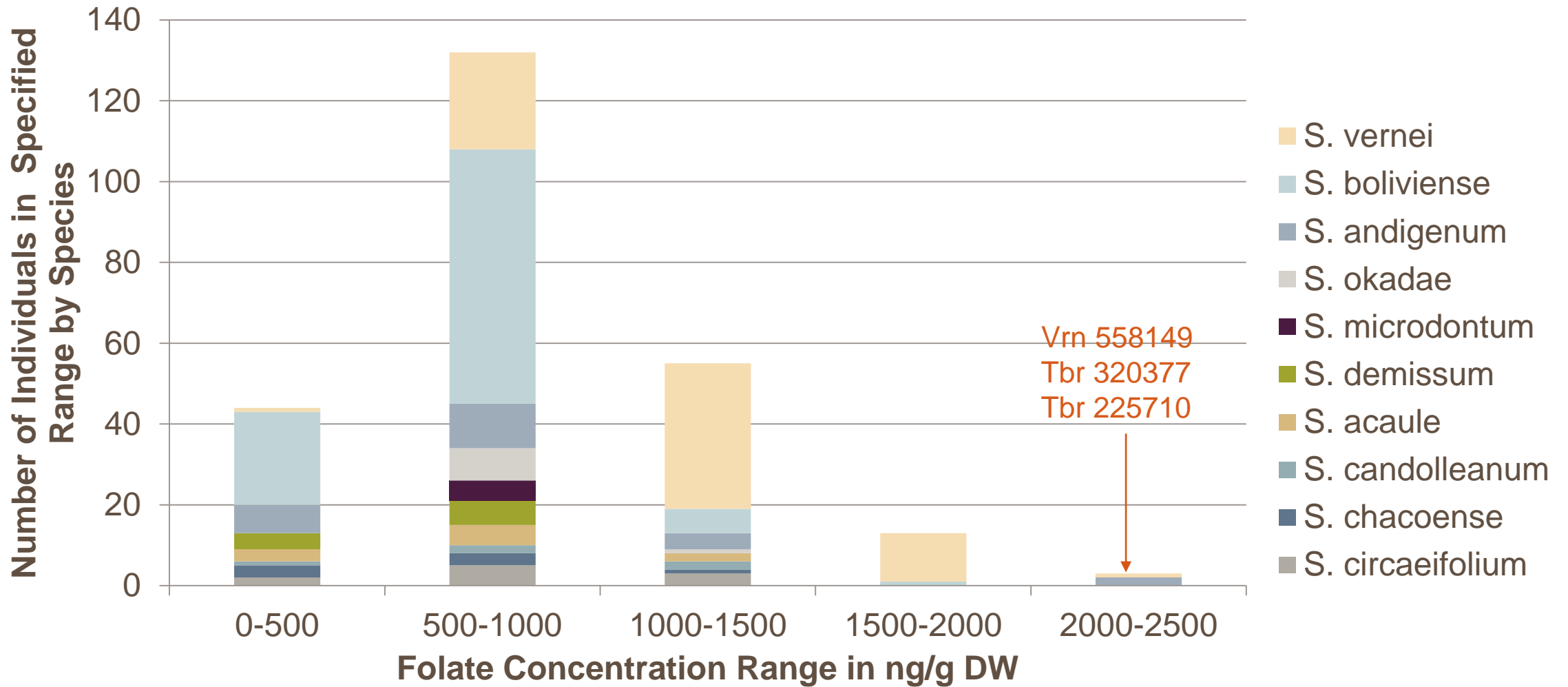
- Microbiological Assay using *L. Rhamnosus*
- Wells loaded with Folic Acid Medium, standards, or samples
- Incubated for 18-24 hours
- Read with microplate reader
- Folate values calculated from standard curve



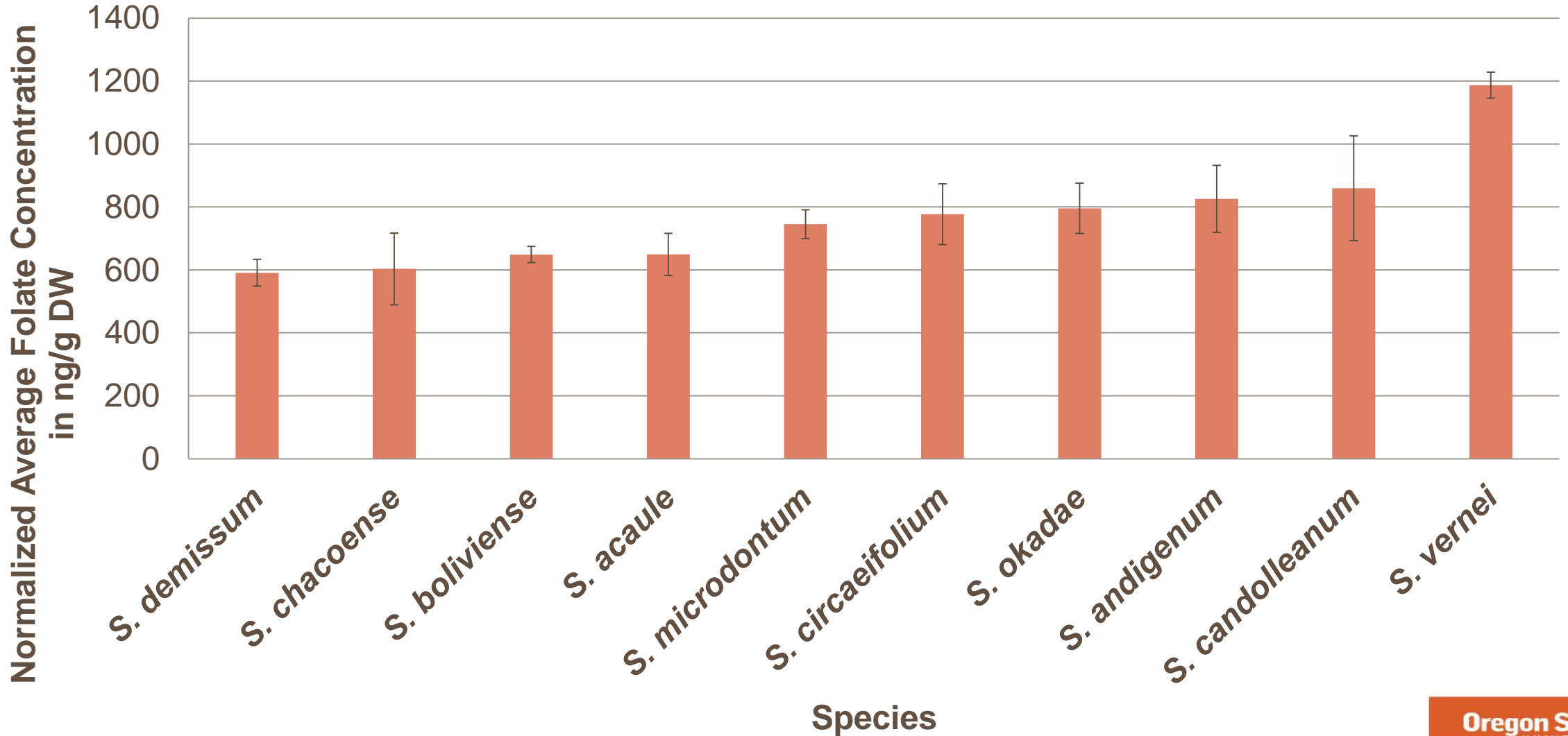
## Wild and Primitive Cultivated Species Folate Distribution



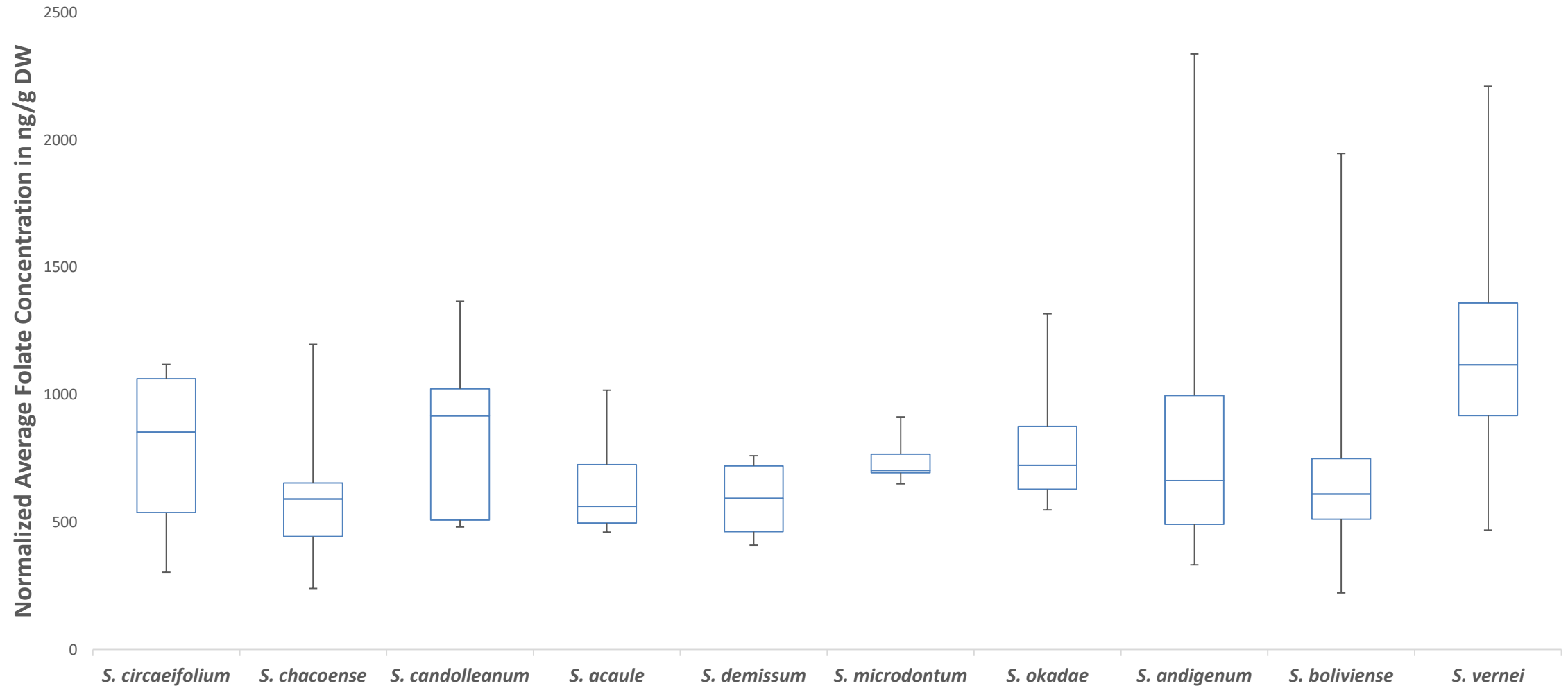
## Wild and Primitive Cultivated Species Folate Distribution



## Wild and Primitive Cultivated Species Average Folate Concentration



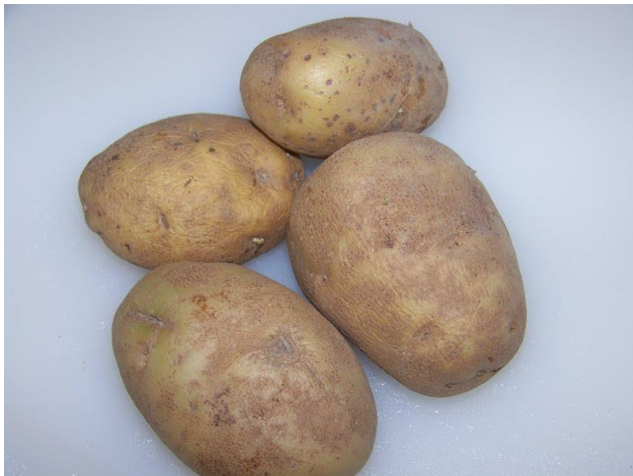
## Normalized Average Folate Concentration by Species



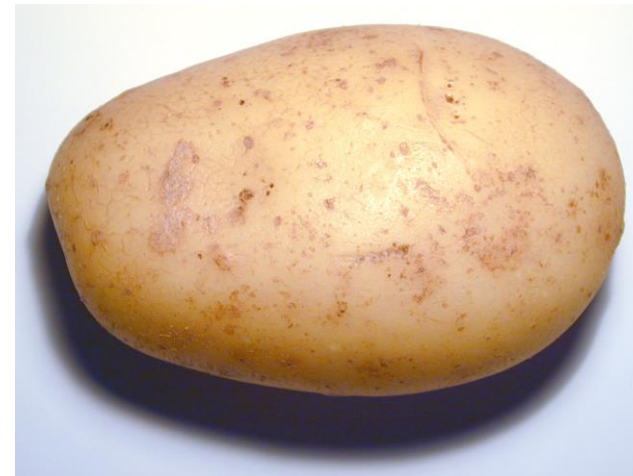
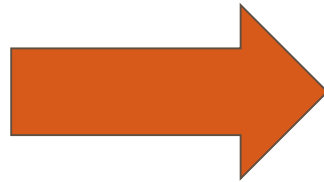


## Results

- Wild and primitive cultivated species demonstrated an averaged range of 220 – 2200 ng/g folate based on dry weight
- Highest measured individuals were in *S. vernei* and *S. tuberosum subsp. andigenum*
- If modern cultivars' average folate concentration could be increased to 2000 ng/g dry weight or more this would represent a 4-5X increase



[https://the3amigoz.files.wordpress.com/2014/01/6761e-100\\_9485.jpg](https://the3amigoz.files.wordpress.com/2014/01/6761e-100_9485.jpg)



<https://hauntingthelibrary.files.wordpress.com/2013/03/potato.jpg>

# Conclusions

- Research demonstrates that there is genetic material with significantly higher folate concentration available for breeding purposes
- Hybridization and evaluation of folate content in these materials will be necessary to determine:
  - Heritability of high folate traits
  - Which species and accessions are the most useful for this process
- Further research is currently underway to try and establish molecular markers associated with high folate phenotypes

# Acknowledgements

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