Vitamin Variation In *Capsicum* Spp. Provides Opportunities To Improve Nutritional Value Of Human Diets

Michael B. Kantar[1], Justin E. Anderson[1], Sarah A. Lucht[2], Kristin Mercer[3], Vivian Bernau[3], Kyle A. Case[4], Nina C. Le[4], Matthew K. Frederiksen[4], Haley C. DeKeyser[4], Zen-Zi Wong[4], Jennifer C. Hastings[4], and David J. Baumler[4,5,6]

[1] Department of Tropical Plant & Soil Sciences, University of Hawaii at Manoa, Honolulu, HI, USA, [2] Department of Epidemiology, Harvard T.H. Chan School of Public Health, Harvard University, Boston, MA, USA, [3] Department of Horticulture and Crop Science, The Ohio State University, Columbus, OH, USA, [4] Department of Food Science and Nutrition, [5]Microbial and Plant Genome Institute, and [6]Biotechnology Institute, University of Minnesota-Twin Cities, St. Paul, MN, USA

INTRODUCTION

Chile peppers, native to the Americas, have spread around the world and have been integrated into the diets of many cultures. Much like their heat content, nutritional content can vary dramatically between different pepper types. In this study, a diverse set of chile pepper types were examined for nutrient content. Some pepper types were found to have high levels of vitamin A, vitamin C, or folate.

Peppers are consumed raw, cooked, and as a spice [1]. The secondary metabolites commonly connected with peppers are capsaicinoids, the compounds that produce their "heat". Peppers are also a good source of vitamin C, vitamin A, vitamin E, and folate [2,3,4]. A number of factors can affect their nutritional content including agronomics [5], harvest time [6], storage and preparation technique [7,8,9], and cultivar type [10,11,12].

Peppers, with their high nutritional content and global consumption, may have a role to play in reducing nutrient deficiencies. In this study, a diverse collection of diverse peppers was evaluated for vitamin A, vitamin C, folate, and capsaicin content. Relationships were explored among nutrient levels, geographic origin, species, and breeding status (heirloom/landrace or modern cultivar). Pepper types were identified with high nutrient content and a range of Scoville heat levels, suggesting that subsequent breeding could develop nutrient-packed mild or hot peppers.

Correlations between nutrient content, species, cultivation status, or geographic region were limited. Varietal selection or plant breeding offer tools to augment nutrient content in peppers. Integration of nutrient rich pepper types into diets that already include peppers could help combat nutrient deficiencies by providing a significant portion of recommended daily nutrients.

MATERIALS AND METHODS

Assembly and propagation of diverse germplasm

- Pepper types were sourced from various heirloom seed producers across North America to explore their phenotypic diversity. Plants were grown in a completely random design with two replications in Madison, WI.

Vitamin concentration ascertainment

- Vitamin A concentrations using a Vitamin A Food Enzyme-Linked Immunosorbent Assay (ELISA) technique (Crystal Chem Inc., IL) and a microplate reader (450 nm) (Epoch 2, Biotek)
- Vitamin C concentrations were estimated using an EnzyChrom™ Ascorbic Acid Assay Kit (BioAssay Systems, Hayward, CA) and a microplate reader (570 nm)
- Folate concentrations were estimated using a Folic Acid ELISA kit (Eagle Biosciences, Nashua, NH) and using a microplate reader (450 nm)
- Capsaicin concentration was estimated using a Capsaicin HS Plate Kit (Beacon Analytical Systems Inc., Saco, ME) following manufacturer's instructions and a microplate reader at 450 nm.

Statistical analysis

- Each univariate nutritional compound data set was analyzed using an Analysis of Variance with cultivar as a fixed factor on complete univariate data sets. Cultivar means for each compound were separated using a Fishers Least Significant Difference (LSD, α=0.01) with a Bonferroni correction in the R statistical language and programming environment [13].

Exploring Public Data on Nutrition

- Data from the USDA National Nutrient Database for Standard Reference Release 28 was downloaded to compare concentration of vitamin A, vitamin C, and Folate in the examined pepper types to other foods known to be high in the nutrients and the effect preparation techniques have on nutrient content [14].

Fig 1. All Peppers explored for nutritional analysis. 1: Bhut Jolokia, 2: Trinidad 7 Pot, 3: Trinidad Butch T Scorpion, 4: Trinidad Douglah, 5: Trinidad Moruga Scorpion, 6: Yellow Bhut Jolokia (Ghost Pepper), 7: Aji Crystal, 8: Jalamundo, 9: Manzano, 10: Corno Di Toro, 11: Banana Sweet, 12: Lilac Bell, 13: Succette de Provence, 14: Thai Red, 15: Ancho, 16: Joe Parker, 17: Big Jim, 18: Sandia, 19: Feher Ozon Paprika, 20: Aconcagua, 21: Santa Fe Grande, 22: 5 Color Marble, 23: Twilight, 24: Naga Dorset, 25: Pasilla, 26: Gold Nugget, 27: Sangria, 28: Peach Habanero, 29: Korean, 30: Chilly Chili, 31: Scotch Bonnet, 32: Espanola, 33: Haiti Cluster, 34: Guajillo, 35: Atomic Starfish, 36: Nepal, 37: Spanish Cayenne, 38: Chinese Giant Sweet, 39: Amish Chicken, 40: Bulgarian, 41: Rocoto Yellow, 42: Shishito, 43: Aji Limon, 44: Sunrise, 45: Mayan Cobanero, 46: Szegedi Giant, 47: Marseilles Sweet Yellow, 48: White Habanero, 49: Tepin, 50: Apple Pimento, 51: Dulcetta Orange, 52: Japones, 53: Joe's Long Cayenne, 54: Tequila Sunrise, 55: Uba Tuba, 56: White Bhut Jolokia, 57: Lady Bug Cherry Bomb, 58: Brown Bhut Jolokia, 59: Orange Trinidad Moruga, 60: Chili De Arbol, 61: Big Bertha, 62: Mustard Habanero, 63: Bahama Fish, 64: Barancio Paprika, 65: India Byadagi Mirchi , 66: Gundo Mirchi, 67: Naga Morich, 68: Yellow Moruga, 69: Ancient Sweet, 70: Bever Dam, 71: Aji Amarillo, 72: Orange Suave, 73: Chinese Ching Choo, 74: Pakistan, 75: Paprika De Cayenne, 76: Sweet Chocolate Bell, 77: Orange Thai, 78: Buran, 79: Zavory Habanero, 80: Pinata, 81: Red Suave, 82: Piquillo, 83: Mulato, 84: Chocolate Habanero, 85: Marconi Gold Sweet Bell, 86: Red Habanero Hot, 87: Red Rocoto, 88: Laotian, 89: Jamaican Yellow, 90: Orange Habanero Hot, 91: Malaysian Goronong, 92: Cajamarca, 93: California Mild, 94: Antilles, 95: Peter Pepper Red, 96: Congo Red, 97: Pumpkin Habanero, 98: Golden Habanero, 99: Assam, 100: Trinidad Perfume, 101: Black Pearl.

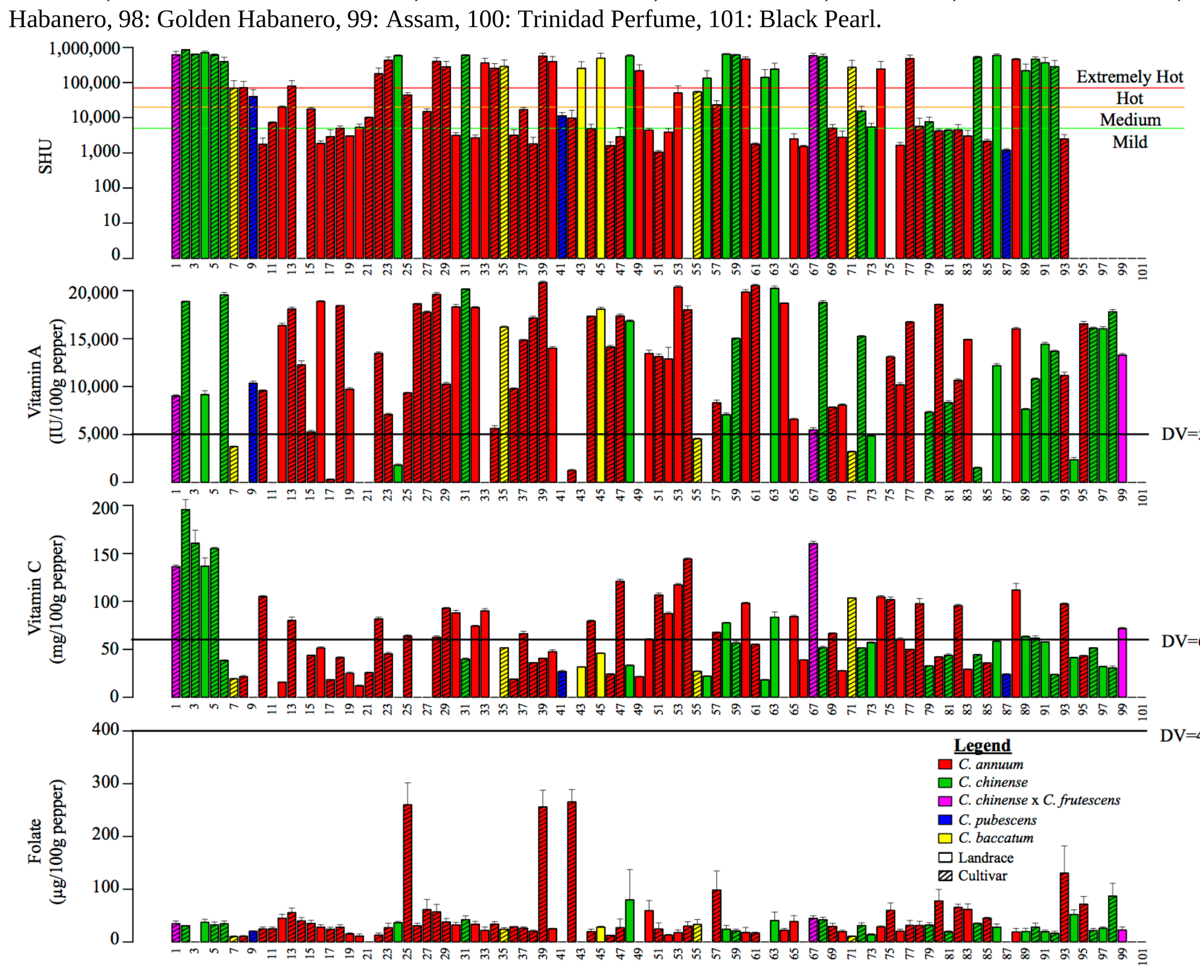

Fig 2. Levels of capsaicin (SHU), vitamin A, vitamin C, and folate detected in pepper types. In the plot, each bar represents a single pepper type in order of the numbering in Figure 1. Each bar represents the average of two biological replicates and the error bars correspond to standard error. The bars are colored based on species and shaded according to status as landrace or cultivar according to the legend. The top plot of capsaicin content is on a log base 10 scale with horizontal lines at 70,000 SHU (red), 20,000 SHU (orange), and 5,000 SHU (green) separating labeled levels on pungency. Recommended daily values (DV) for adults consuming 2,000 calories are plotted and labeled for the three lower plots of pepper nutritional content (FDA, 2011). LSD = 262963 SHU, 1624 vitamin A, 18.7 vitamin C, 94.3 folate.

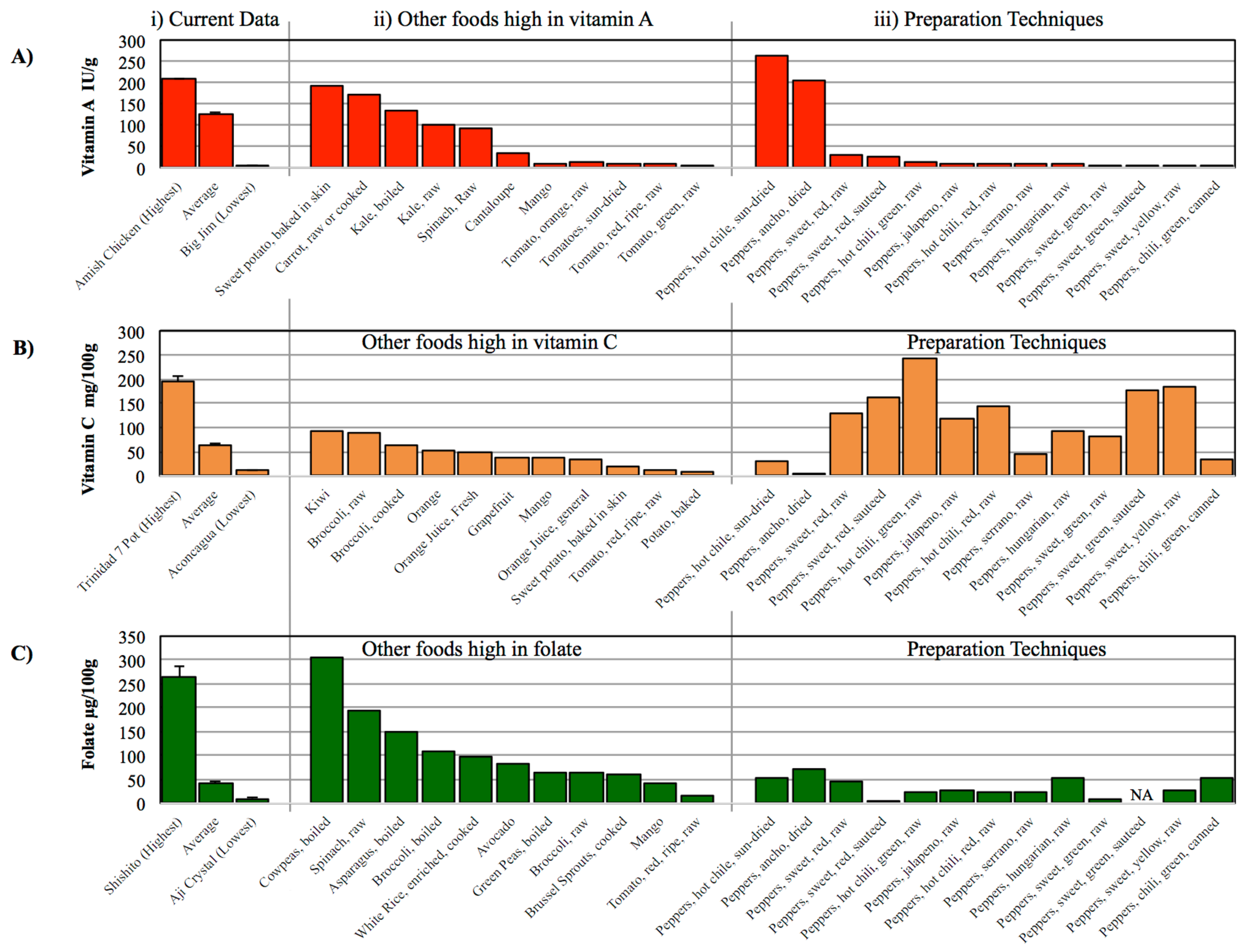

Fig 3. Comparison of our pepper types to other commonly consumed foods and preparation techniques. Plotted are the vitamin A (A), vitamin C (B), and folate (C) content for the current data (i), other foods high in each nutrient (ii), and preparation techniques from the USDA National Nutrient Database for Standard Reference Release 28. Error bars in the current data correspond to standard error.

Capsaicin Content		Vitamin A Content		Vitamin C Content		Folate Content	
Ten Lowest	Ten Highest	Ten Lowest	Ten Highest	Ten Lowest	Ten Highest	Ten Lowest	Ten Highest
Dulcetta Orange	Trinidad 7 Pot	Big Jim-	Amish Chicken+	Aconcagua-	Trinidad 7 Pot+	Aji Crystal-	Shishito
Red Rocoto	Trinidad Douglah	Shishito	Big Bertha+	Lilac Bell	Trinidad Butch T Scorpion	Jalamundo-	Pasilla
Gundo Mirchi	Brown Bhut Jolokia	Chocolate Habanero	Joe's Long Cayenne+	Mustard Habanero	Naga Morich	Aji Amarillo-	Amish Chicken+
Szegedi Giant	Trinidad Butch T Scorpion	Naga Dorset-	Bahama Fish	Big Jim-	Trinidad Moruga Scorpion	Aconcagua-	California Mild
Sweet Chocolate Bell	Bhut Jolokia	Antilles	Scotch Bonnet	Nepal	Tequila Sunrise	Szegado	Lady Bug Cherry Bomb
Corno De Toro	Orange Trinidad Moruga	Aji Amarillo-	Chili De Arbol	Aji Crystal-	Trinidad Douglah	Japones	Golden Habanero
Big Bertha+	Trinidad Moruga Scorpion	Aji Crystal-	Peach Habanero	Jalamundo-	Bhut Jolokia	5-color marble	White Habanero
Chinese Giant Sweet	Scotch Bonnet	Uba Tuba	Yellow Ghost Pepper	Tepin	Marseilles Sweet Yellow Bell	Ching Choo	Pinata
Joe Parker+	Red Habanero Hot	Chinese Ching Choo	Joe Parker+	White Bhut Jolokia	Joe's Long Cayenne+	Feher Ozon	Peter Pepper Red
Marconi Gold	Naga Dorset-	Ancho	Trinidad 7 Pot+	Cajamarca	Laotian	Cajumarca	Piquillo

+ Pepper types that show up twice in this table for ten highest nutrient content or low capsaicin content.

- Pepper types that show up twice in this table ten lowest nutrient content or high capsaicin content.

CONCLUSIONS

Direct consumption of the more nutritious peppers assayed here as well as future consumption of nutritionally enhanced varieties could be used in international efforts to address vitamin deficiency. Thus, though not a silver bullet, peppers could constitute an important part of an integrated strategy, including nutrient supplementation and food fortification, for combatting vitamin deficiency. While lower quantities of pepper are consumed daily compared to staples, such as maize or wheat, highly nutritious or nutritionally improved peppers can contribute to a diverse and healthy diet.

REFERENCES

1. Sherman PW, Billing J. Darwinian Gastronomy: Why We Use Spices Spices taste good because they are good for us. BioScience. 1999;49: 453-463.
2. Phillips KM, Ruggio DM, Ashraf-Khorassani M, Haytowitz DB. Difference in folate content of green and red sweet peppers (*Capsicum annuum*) determined by liquid chromatography-mass spectrometry. J. Agric. Food Chem. 2006;54: 9998–10002. doi: 10.1021/jf062327a.
3. Wahyuni Y, Ballester AR, Sudarmonowati E, Bino RJ, Bovy AG. Secondary metabolites of Capsicum species and their importance in the human diet. J. Nat. Prod. 2013;76: 783-793.
4. Wahyuni Y, Ballester AR, Sudarmonowati E, Bino RJ, Bovy AG. Metabolite biodiversity in pepper (Capsicum) fruits of thirty-two diverse accessions: variation in health-related compounds and implications for breeding. Phytochemistry. 2011;72: 1358–70. doi: 10.1016/j.phytochem.2011.03.016.
5. Pérez-López AJ, López-Nicolas JM, Núñez-Delicado E, Amor FMD, Carbonell-Barrachina ÁA. Effects of agricultural practices on color, carotenoids composition, and minerals contents of sweet peppers, cv. Almuden. J. Agric. Food Chem. 2007;55: 8158-8164.
6. Deepa N, Kaur C, George B, Singh B, Kapoor HC. Antioxidant constituents in some sweet pepper (*Capsicum annuum* L.) genotypes during maturity. LWT - Food Sci. Technol. 2007;40: 121–129. doi: 10.1016/j.lwt.2005.09.016.
7. Martínez S, López M, González-Raurich M, Bernardo Alvarez A. The effects of ripening stage and processing systems on vitamin C content in sweet peppers (*Capsicum annuum* L.). Int. J. Food Sci. Nutr. 2005;56: 45–51. doi: 10.1080/09637480500081936.
8. Pugliese A, Loizzo MR, Tundis R, O'Callaghan Y, Galvin K, Menichini F, O'Brien N. The effect of domestic processing on the content and bioaccessibility of carotenoids from chili peppers (Capsicum species). Food Chem. 2013;141: 2606-2613.
9. US Department of Agriculture, Agricultural Research Service, Nutrient Data Laboratory. Nutrient Data : USDA National Nutrient Database for Standard Reference, Release 28; 2015. Available at: http://www.ars.usda.gov/nea/bhnrc/ndl [Accessed April 14, 2016].
10. Guil-Guerrero JL, Martínez-Guirado C, del Mar Rebolloso-Fuentes M, Carrique-Pérez A. Nutrient composition and antioxidant activity of 10 pepper (*Capsicum annuun*) varieties. Eur. Food Res. Technol. 2006;224: 1-9.
11. Topuz A, Ozdemir F. Assessment of carotenoids, capsaicinoids and ascorbic acid composition of some selected pepper cultivars (*Capsicum annuum* L.) grown in Turkey. J. Food Compost. Anal. 2007;20: 596-602.
12. Perla V, Nimmakayala P, Nadimi M, Alaparthi S, Hankins GR, Ebert AW, et al. Vitamin C and reducing sugars in the world collection of Capsicum baccatum L. genotypes. Food Chem. 2016;202: 189–98. doi: 10.1016/j.foodchem.2016.01.135.
13. R Core Team. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria; 2015. Available at: http://www.R-project.org/.
14. FDA. Center for Food Safety and Applied Nutrition:Labeling & Nutrition - Guidance for Industry: A Food Labeling Guide (14. Appendix F: Calculate the Percent Daily Value for the Appropriate Nutrients). 2013. Available at: http://www.fda.gov/Food/GuidanceRegulation/GuidanceDocumentsRegulatoryInformation/LabelingNutrition/ucm064928.htm [Accessed April 14, 2016].

ACKNOWLEDGEMENTS

The authors would like to thank Dr. Joseph Delaney and Dr. "Pepper" Steve Marier for insightful discussions and guidance of this study.