

# Population Genomics of the Native and Invaded California Range of Palmer Amaranth (*Amaranthus palmeri*)

Josue Duque<sup>1</sup>, Alexander Lopez<sup>1</sup>, Romy Lum<sup>1</sup>, Chance Riggins<sup>2</sup>, Katherine Waselkov<sup>1</sup>



United States  
Department of  
Agriculture National Institute  
of Food and  
Agriculture

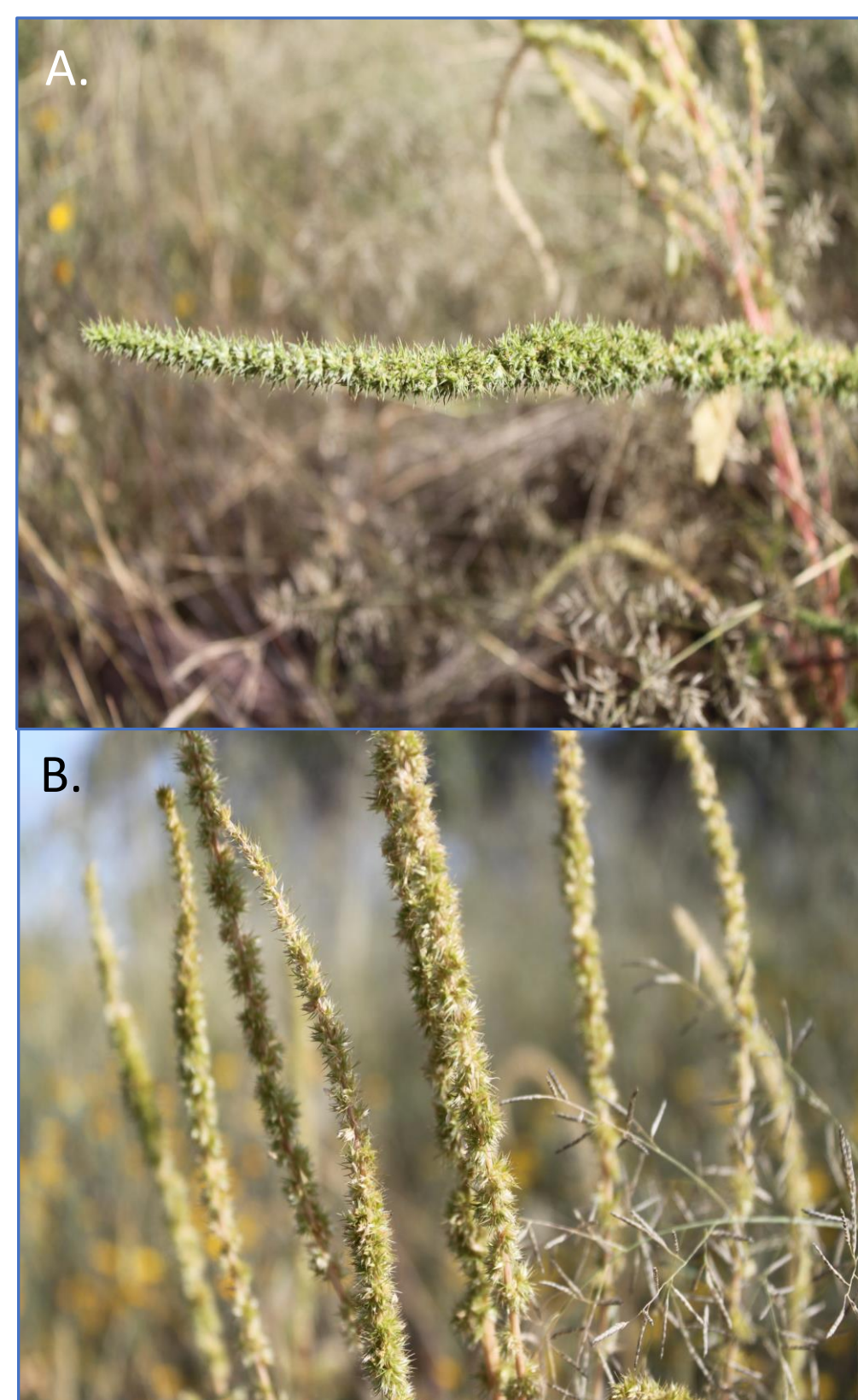
<sup>1</sup>Department of Biology, California State University Fresno, CA, USA;  
<sup>2</sup>Department of Crop Sciences, University of Illinois, Urbana, IL, USA



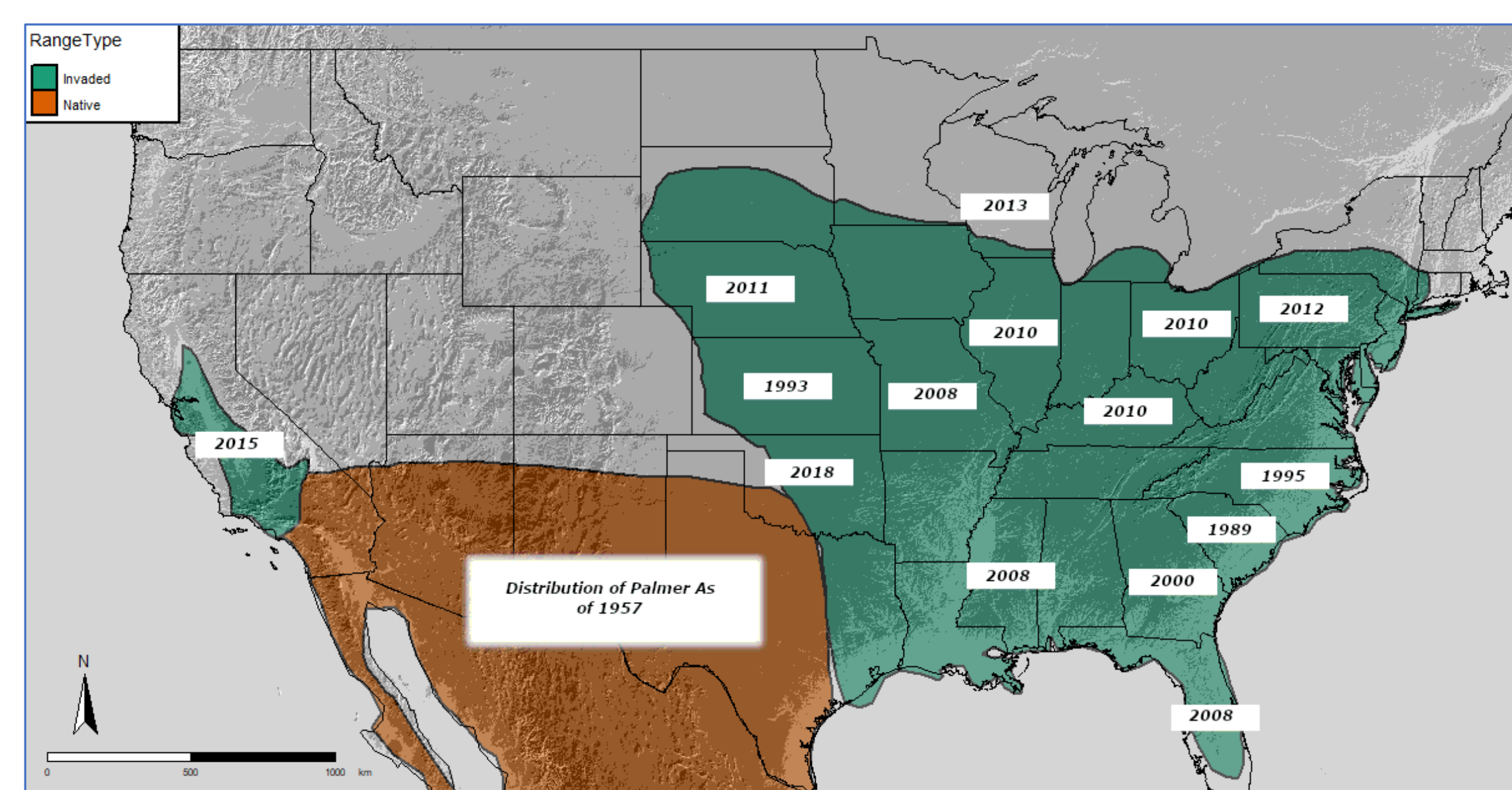
Discovery. Diversity. Distinction.

## Background

- Palmer amaranth (Figure 1), *Amaranthus palmeri*, is an herbaceous annual plant native to the arid regions of northeastern Mexico (Sonora) and the southwestern United States (southern California, Arizona, New Mexico, and Texas (Sauer 1957; Figure 2).
- Palmer amaranth is an aggressive weed that has become a pervasive issue in the eastern United States, where herbicide and multi-resistant biotypes have emerged.
- The plant has found recent success in agricultural fields in the Californian Central Valley, as chemical herbicides began to fail in the region in 2015 (Rios et al. 2016; Figure 2).
- This ongoing range expansion gives us an opportunity to examine the genetic diversity and structure of emerging central California demes of this weedy species.
- The source populations of these demes remains unknown. Their genetic origins could be in the ancestral range (orange shading in Fig 2), or they could be the result of gene flow from the previously invaded regions in the Eastern U.S.
- Conducting a population genetic analysis on these new demes in comparison to the individuals in the ancestral range is a critical first step to understanding local (southwestern United States) population structure and trends in genetic variation.



**Figure 1.** Close up images of Palmer amaranth inflorescences. Palmer is a dioecies species; both female (A) and (B) male individual plants are depicted above.



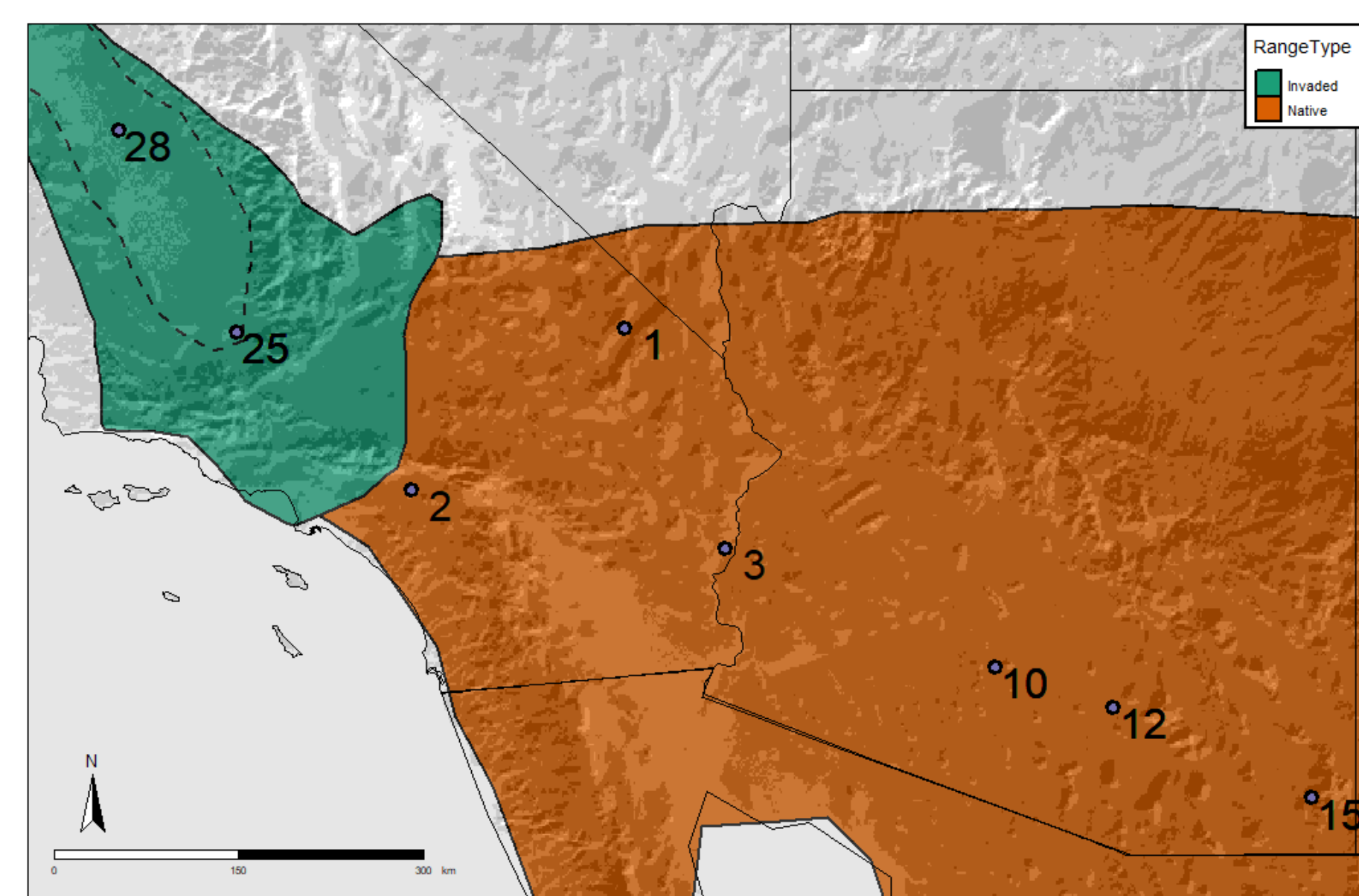
**Figure 2.** The current range of Palmer amaranth has expanded from its original arid range (orange shading) well into the northeastern and Midwestern U.S., and most recently into central California. States in the invade regions (green shading) are labeled with the year that the first herbicide resistant biotype was detected (Sauer 1957; Heap 2020).

## Objectives

- Curate a high-quality panel of single nucleotide polymorphisms (SNPs) and Indels from genomic data generated from Palmer amaranth (*Amaranthus palmeri*) sampled from individuals across the native and invaded Central California ranges. These sampling locations will encompass the two distinct geographic regions.
- Using hierarchical clustering algorithms to see if we can detect population structure within our sampled populations using the genetic variation among samples.
- Use other population genetics statistics (estimates of genetic distance and diversity) to supplement and corroborate our findings in the clustering analysis.

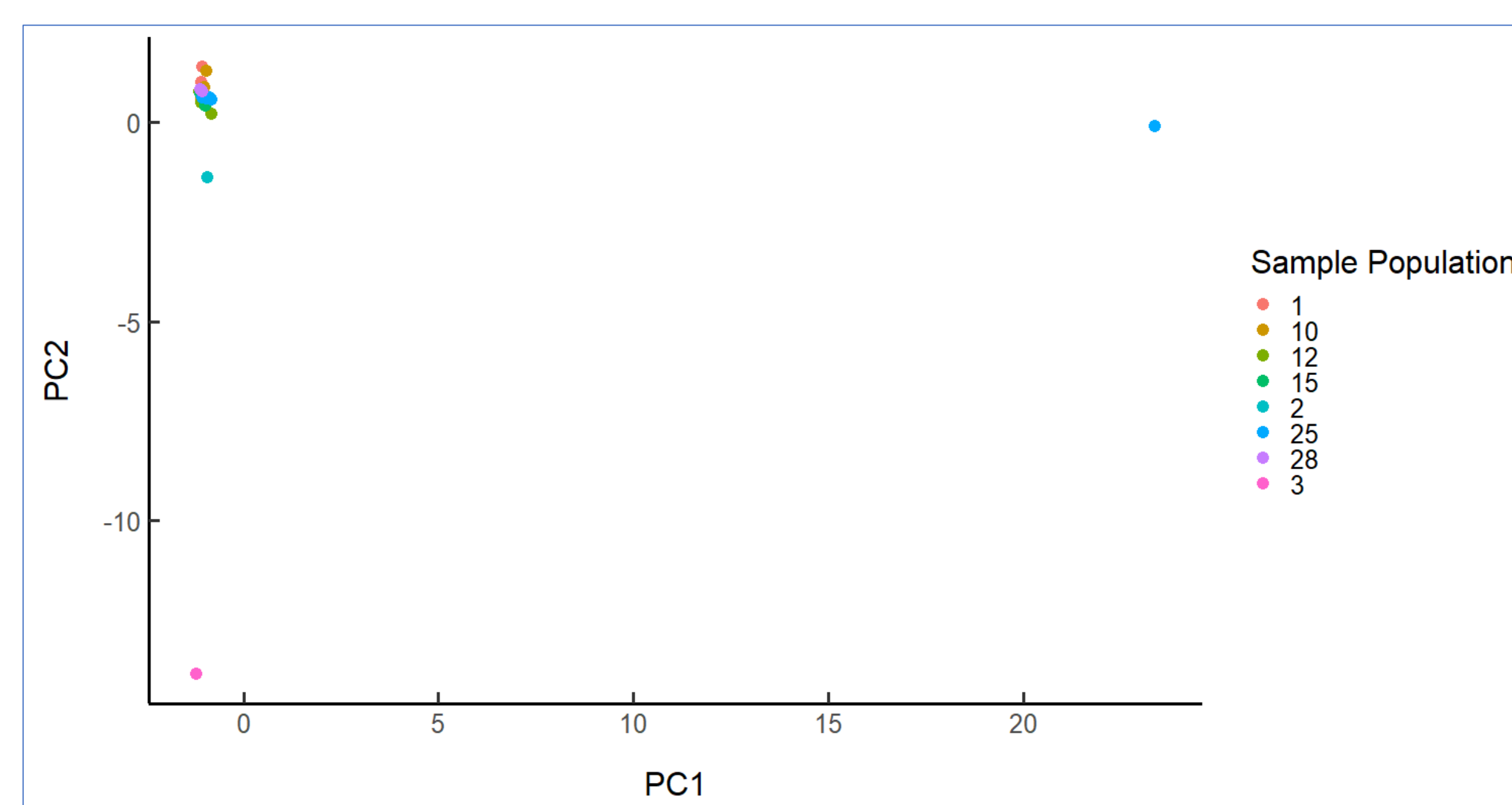
## Approach and Results

- Our preliminary dataset is based on sequence data from 24 individuals from locations in Palmer amaranth's native range and the Central California invaded range. Sampling locations 25 and 28 represent locations that are well outside of Palmer's native range.
- Libraries were prepared using a RipTide library construction kit (iGenomX), quantitated with qPCR, and sequenced on a single lane in an Illumina NovaSeq 6000.
- We used the Genome Analysis Toolkit (GATK) for variant (SNP/Indel) calling (McKenna et al. 2010). In this process, we used the genome of closely related species, *Amaranthus tuberculatus* (Moq.) Sauer (waterhemp), as our reference genome (Kreiner et al. 2019).

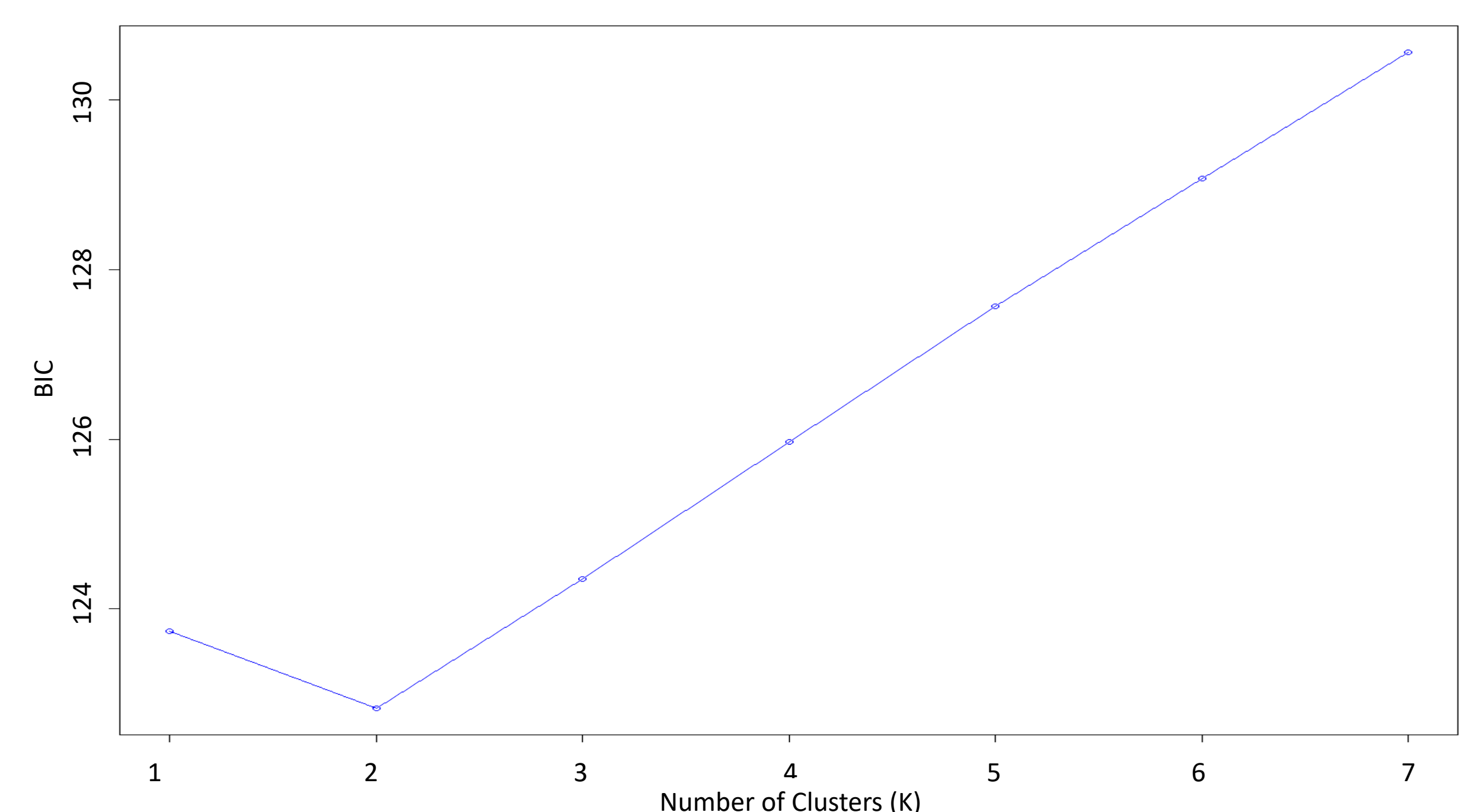


**Figure 3.** Map of sample sites from pilot analysis. Note that 7 of our 24 initial samples were from the invaded region (green shading) and 17 were from the native range (orange shading).

- Clustering analysis: in order to identify clusters (K) of closely related individuals based on shared genetic variance, we used the R package *adegenet* v2.1.3 (Jombart 2008).
- Principal components analysis (PCA) (Figure 4)
- Discriminant analysis of principal components (DAPC), a method that uses K-means and model selection (Jombart et al. 2010; Figure 5) to identify the most likely value for K, and then can be used to assign individual samples to those K clusters.



**Figure 4.** In the scatter plot of the first two principal components, one out of five sampled members of population 25 cluster further away from the remaining individuals examined in this PCA.

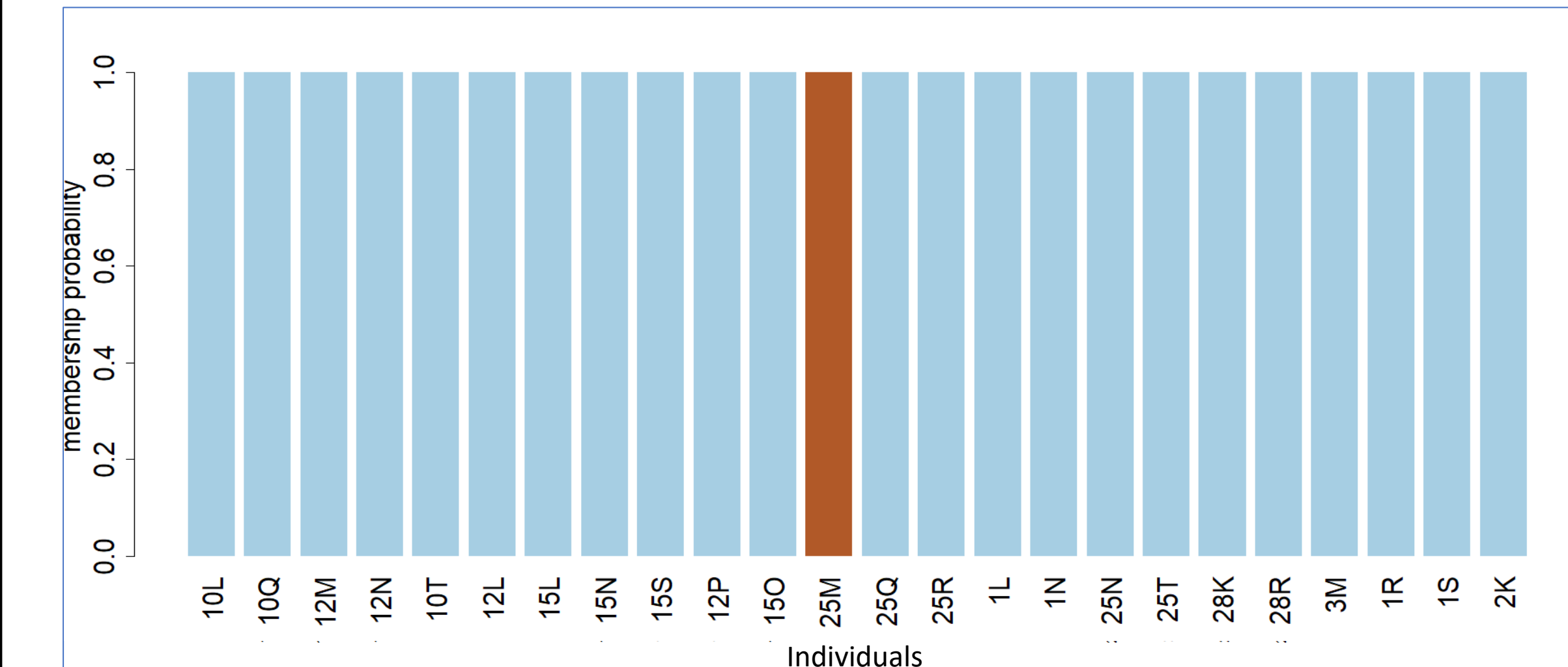


**Figure 5.** Using the R package *adegenet*, we examined the likelihood of the existence of discrete clusters K, from K = 1 to 7. The plot shows the value of the Bayesian information criterion (BIC) versus the number of clusters, and indicates that the scenario K = 2 is most likely given the data.

## References

- Heap, I. 2020. International survey of herbicide resistant weeds. Website [www.weedscience.org](http://www.weedscience.org).
- Jombart, T., et al. 2008. *adegenet*: a R package for the multivariate analysis of genetic markers. *Bioinformatics* 24: 1403-1405.
- Jombart, T., et al. 2010. Discriminant analysis of principal components: a new method for the analysis of genetically structured populations. *BMC Genetics* 11: 94.
- Kreiner, J., et al. 2019. Multiple modes of convergent adaptation in the spread of glyphosate-resistant *Amaranthus tuberculatus*. *PNAS* 116: 21076-21084.
- McKenna, A., et al. 2010. The Genome Analysis Toolkit: a MapReduce framework for analyzing next-generation DNA sequencing data. *Genome Research* 20: 1297-1303.
- Rios, S., et al. 2016. Tolerance of *Amaranthus palmeri* populations from California to postemergence herbicides at various growth stages. *Crop Protection* 87: 6-12.
- Sauer, J. 1957. Recent migration and evolution of the dioecious amaranths. *Evolution* 11: 11-31.

## Results



**Figure 6.** Each bar in this DAPC plot represents an individual. The color shows the probability of assignment of individuals to the different clusters (1 = blue and 2 = orange), based on the proportion of genetic marker variation from that samples showing an affiliation with each cluster.

- Initial PCA analysis indicates that there is some evidence for genetic distance between individuals in the new Central Valley demes and individuals in the ancestral range (Figure 4).
- DAPC analysis indicates that there is strong evidence for the existence of two genetically distinct clusters in the SNP data (K = 2: Figure 5); however, only one individual from population 25 is included in Cluster 2 (Figure 6).

## Ongoing and Future WorkOur

- Work is currently underway to generate more sequence data from 96 additional individuals. This new data set will include additional individuals from the same populations in both the invaded range and the native range, to lend statistical strength to our analyses.
- We will be analyzing the combined current and new sequence data with a recently released high quality reference genome assembly for *A. palmeri*. This will improve the accuracy of the variant calling step, as well as increase the confidence in our results.
- We will also carry out STRUCTURE analysis and calculate standard within- and among-population genetic diversity statistics to corroborate the conclusions for K reached in our DAPC analysis, and further compare the genetics of native and invaded range populations.

## Acknowledgments

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## For More Information

- To learn more about our lab and our research visit us at: [waselkovlab.com](http://waselkovlab.com)
- To contact us directly, email us at:
  - J. Duque
    - Email: [duqu9804@mail.fresnostate.edu](mailto:duqu9804@mail.fresnostate.edu)
  - K. Waselkov
    - Email: [kwaselkov@fresnostate.edu](mailto:kwaselkov@fresnostate.edu)
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