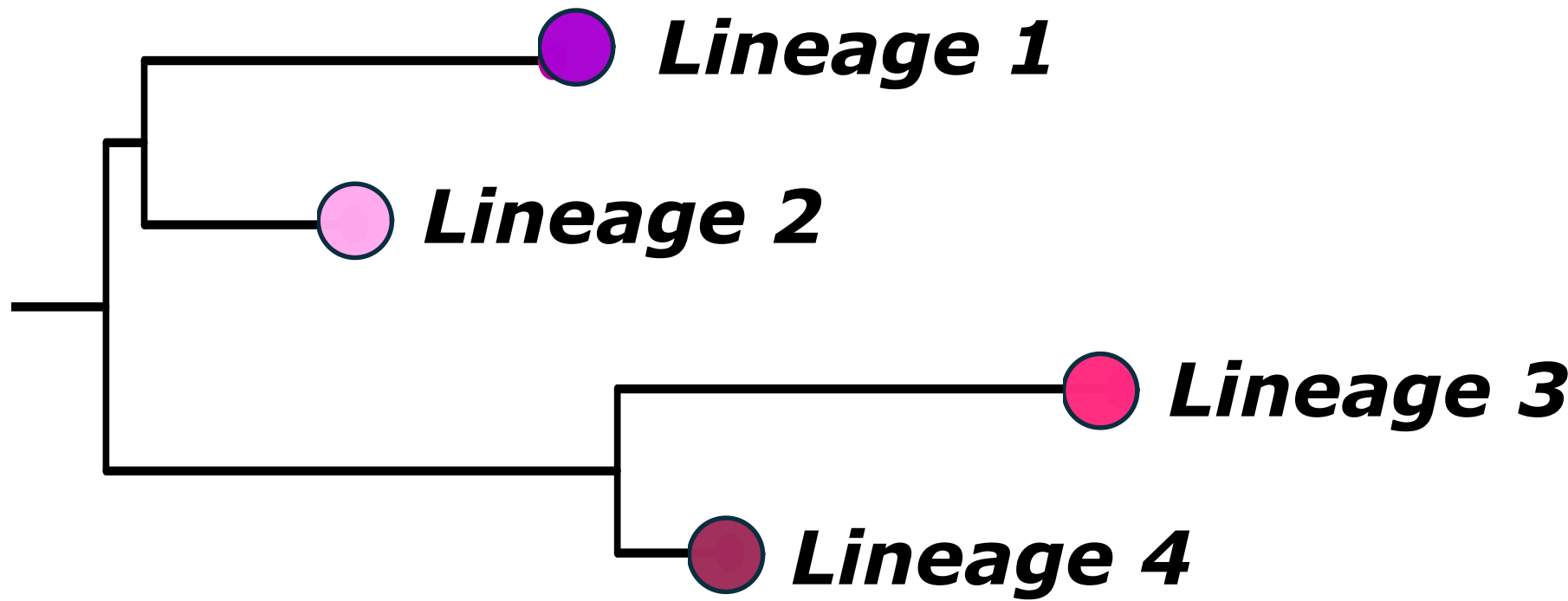
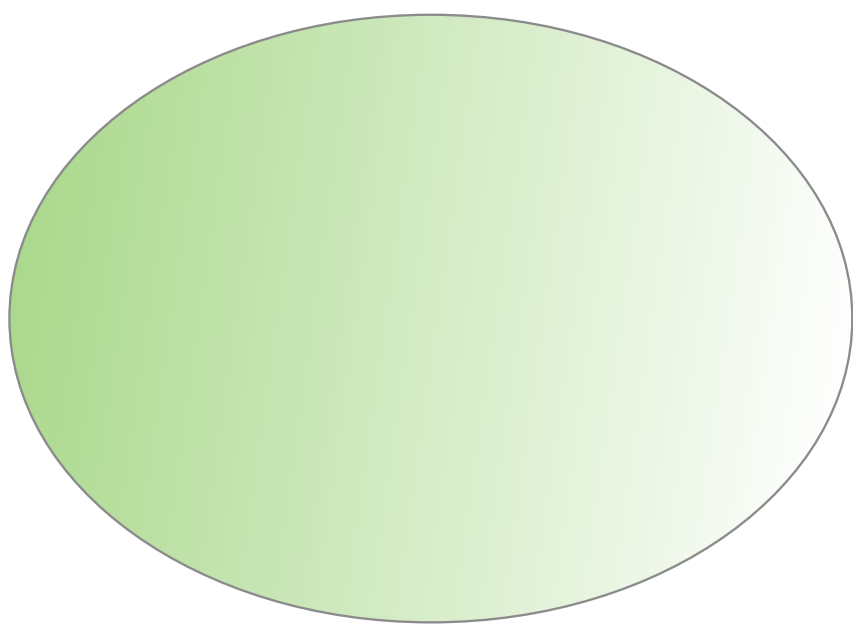


STEP 1: Identify comparative groups, focal trait of study, and environmental variables

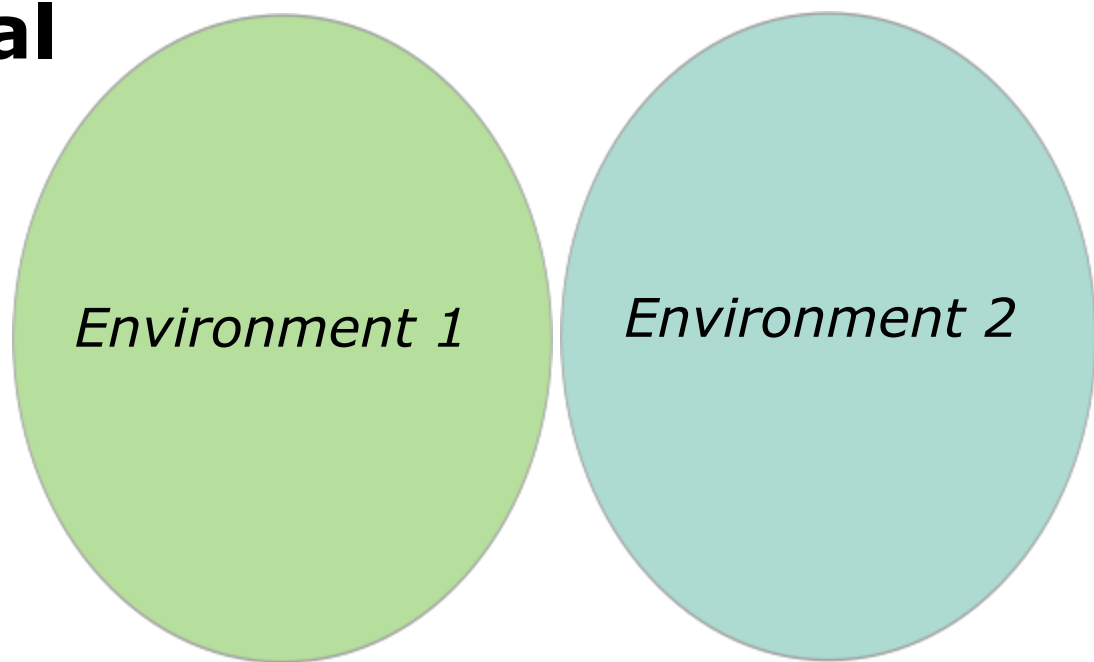
Identify genetic differentiation between selected lineages and choosing the right lineages



Continuous environmental variation Eg. precipitation or temperature

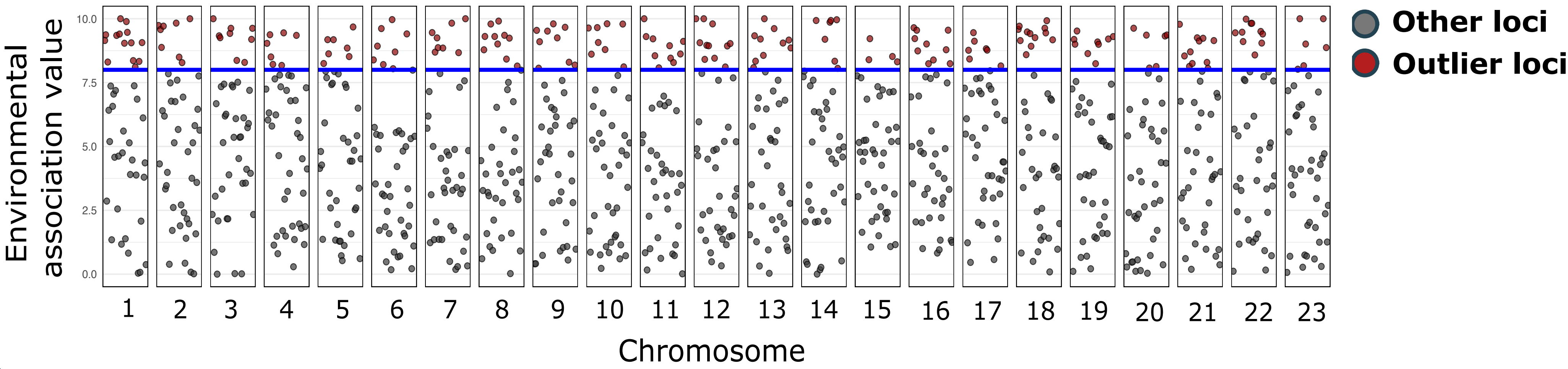


Discrete environmental variation Eg. novel host plant colonization



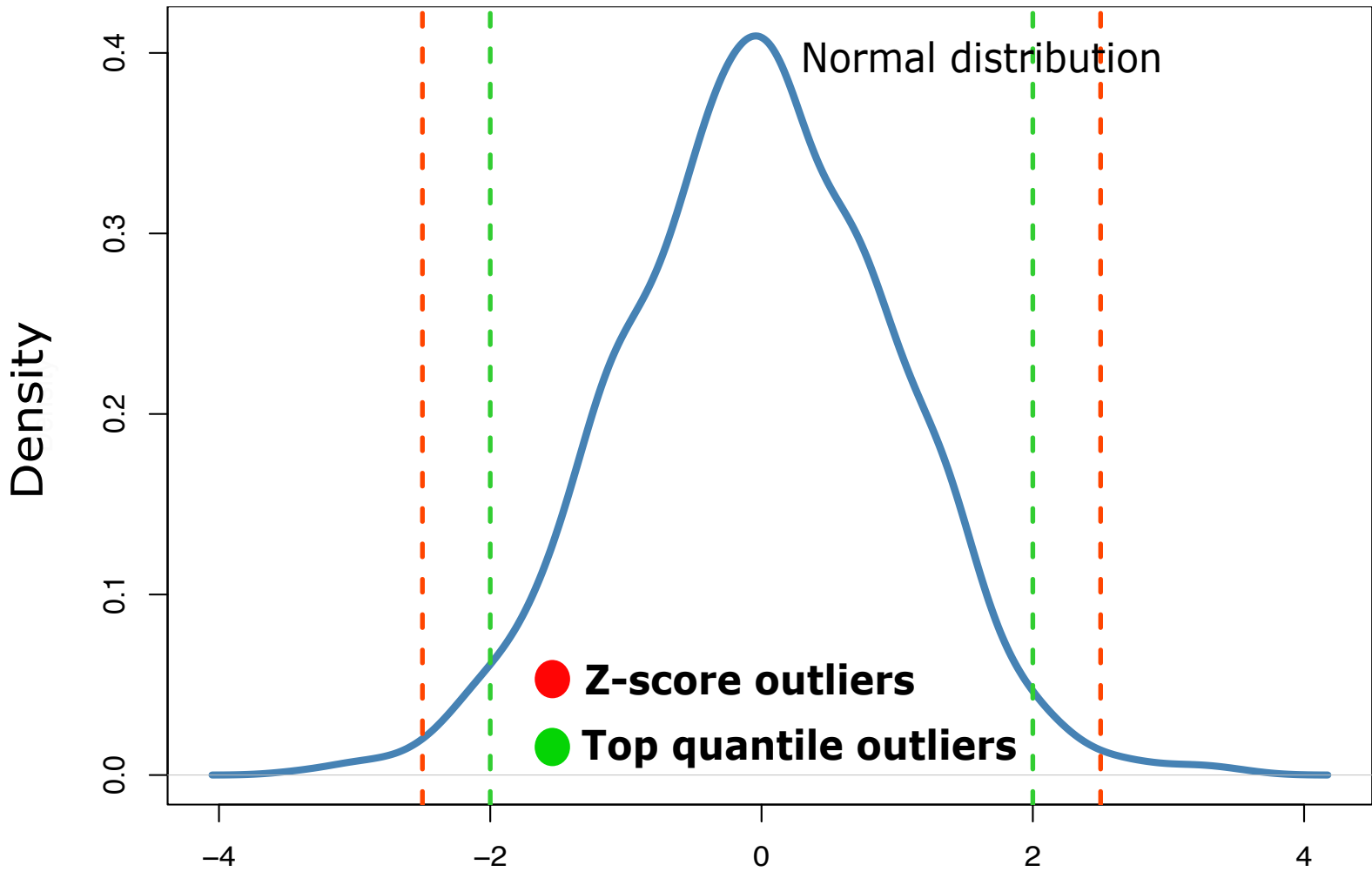
STEP 2: Identify loci showing exceptional association with trait/environment or outlier loci

Use GxE or Fst Outlier analysis to identify genome-wide candidates which show exceptional association with the identified environment or trait

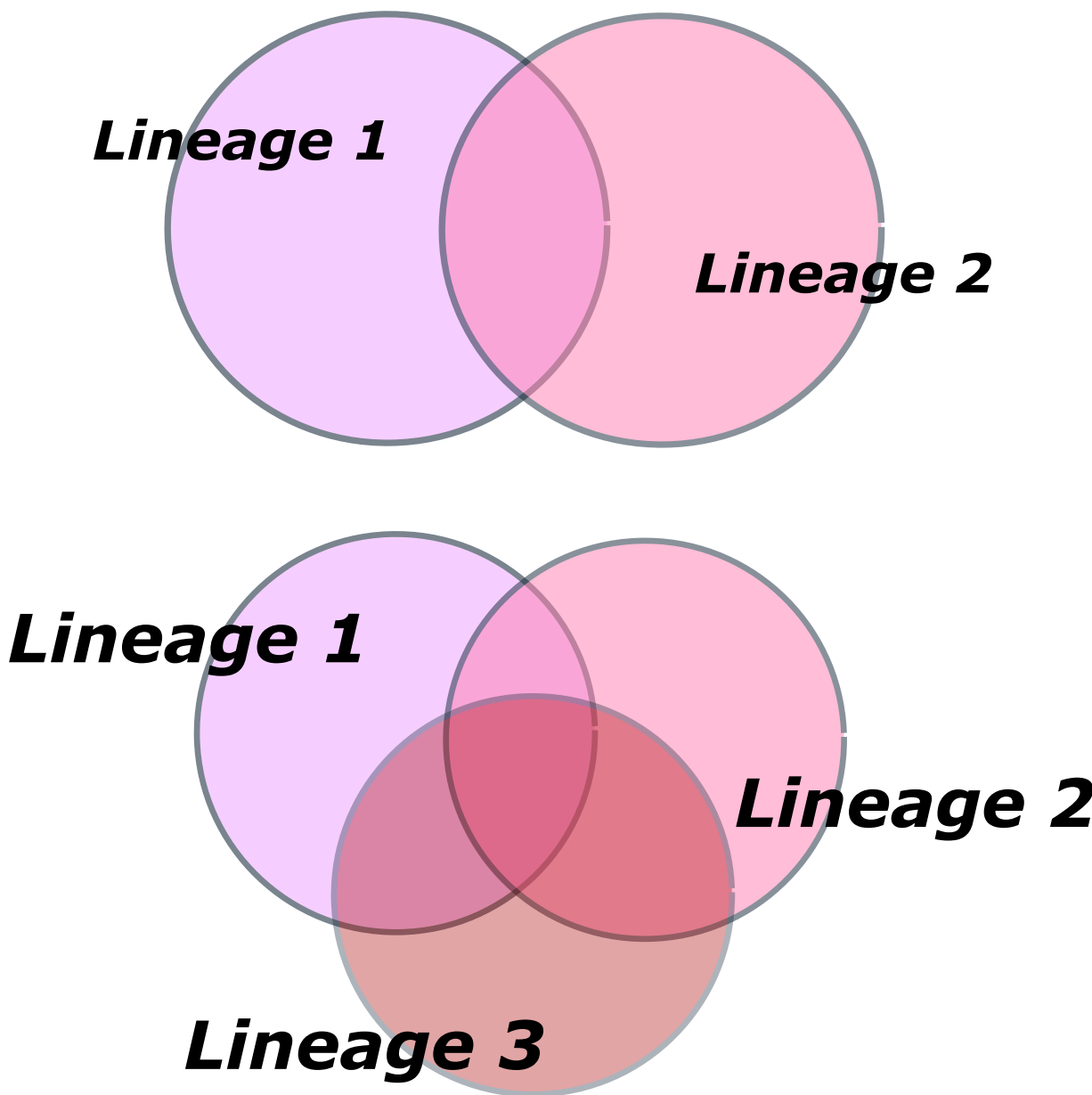


STEP 3: Identify loci in top quantiles or candidate loci and quantify overlap of loci

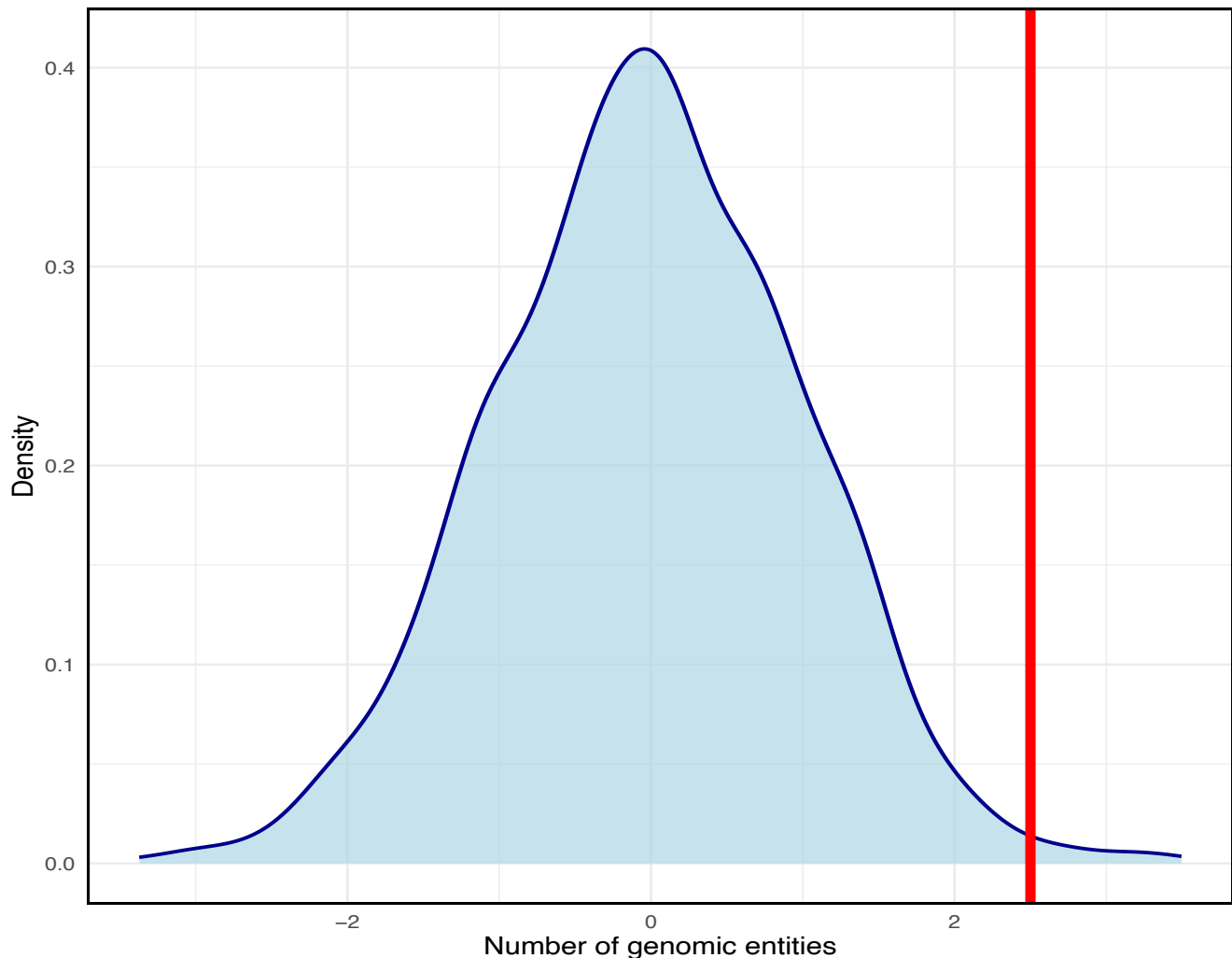
Use outlier analyses such as top quantiles or Z-score tests to identify top candidate genomic entities to test for repeated adaptation



Identify genomic entities associated with repeated adaptation in two or more lineages



Use randomization tests to validate if the observed number of genomic entities associated with repeated adaptation are more than expected under chance



Examples of cross-validation indices which quantify the observed amount of repeatability relative to expectations under a null hypothesis

Metric	Description	Test data	Author
SuperExactTest	Calculates probabilities of multi-set intersections based on combinatorial theory	Simulation based	Wang et al. (2015)
Rank–Rank Hypergeometric Overlap (RRHO)	Ranks gene lists by differential expression and calculates statistical significance of overlapping genes	Differential gene expression data from cancer cells	Plaisier et al. (2010)
Pairwise Hypergeometric Test	SNP or gene overlap among lineages; functional responses to environmental changes	Gene sets	Cheng et al. (2021)
Null-W Method	Strength of association (r^2) for SNPs in candidate orthologs; null distribution from non-candidate orthologs	Single nucleotide polymorphisms in orthologous genes	Yeaman et al. (2016)
C-score and C_{hyper} Index	Probability of observed repeatability; adaptation-effective loci; genomic constraints	Orthologous genes	Yeaman et al. (2018)
X-Fold Enrichment Metric	SNP overlap associated with host plant use and climatic adaptation	Single nucleotide polymorphism data	Chaturvedi et al. (2018, 2023)
PicMin	Significance of repeated molecular evolution at individual genes; genome scan results	Gene sets	Booker et al. (2022)