**Hierfstat software**

To get a summary statistic of the data loaded, heirfstat r package was used. As expected, the overall observed heterozygosity was 0 since *M. Oryzae* has a haploid genome.

> summary(df\_2) # summarizes a genind object

// Number of individuals: 226

// Group sizes: 56 37 58 56 9 10

// Number of alleles per locus

// Number of alleles per group: 331909 336362 342627 284122 256010 315506

// Percentage of missing data: 1.62 %

// Observed heterozygosity: 0

Calculating the Fst

> wc(df\_3, diploid = F) # Fst

$FST

[1] 0.3375974

# Pairwise Fst

> wc84 <- genet.dist(df\_3, method = "WC84", diploid = F)

> wc84

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | Ethiopia | Kenya | Tanzania | Uganda | Historic\_Uganda |
| Kenya | 0.386813120 |  |  |  |  |
| Tanzania | 0.164666561 | 0.272751250 |  |  |  |
| Uganda | 0.522569906 | 0.055839201 | 0.432473888 |  |  |
| Historic\_Uganda | 0.481448629 | 0.050712401 | 0.376374068 | 0.126959750 |  |
| Historic\_Kenya | 0.394441152 | 0.004639372 | 0.283999165 | 0.127215827 | 0.048087892 |

> nei87 <- genet.dist(df\_3, method = "Nei87", diploid = F)

> nei87

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | Ethiopia | Kenya | Tanzania | Uganda | Historic\_Uganda |
| Kenya | 0.3851 |  |  |  |  |
| Tanzania | 0.1647 | 0.2746 |  |  |  |
| Uganda | 0.5222 | 0.0527 | 0.4327 |  |  |
| Historic\_Uganda | 0.5091 | 0.0628 | 0.4151 | 0.1248 |  |
| Historic\_Kenya | 0.3907 | 0.0050 | 0.2923 | 0.1056 | 0.0494 |

> stats\_hier <- basic.stats(df\_3, diploid = F) # Ho and He analyses (nei87)

> stats\_hier

$perloc

Ho Hs Ht Dst Htp Dstp Fst Fstp Fis Dest

V3 NA 0.2023 0.4115 0.2092 0.4534 0.2511 0.5084 0.5538 NA 0.3147

V4 NA 0.2023 0.4115 0.2092 0.4534 0.2511 0.5084 0.5538 NA 0.3147

V5 NA 0.2013 0.4110 0.2097 0.4529 0.2516 0.5102 0.5555 NA 0.3150

V6 NA 0.2013 0.4110 0.2097 0.4529 0.2516 0.5102 0.5555 NA 0.3150

V7 NA 0.1935 0.4093 0.2158 0.4525 0.2590 0.5272 0.5723 NA 0.3211

V8 NA 0.1935 0.4093 0.2158 0.4525 0.2590 0.5272 0.5723 NA 0.3211

V9 NA 0.1935 0.4092 0.2157 0.4524 0.2589 0.5272 0.5723 NA 0.3210

V10 NA 0.1951 0.4088 0.2137 0.4516 0.2565 0.5227 0.5679 NA 0.3186

V11 NA 0.1944 0.4097 0.2153 0.4528 0.2584 0.5255 0.5706 NA 0.3207

V12 NA 0.1944 0.4097 0.2153 0.4528 0.2584 0.5255 0.5706 NA 0.3207

V13 NA 0.1983 0.4144 0.2162 0.4577 0.2594 0.5216 0.5668 NA 0.3235

V14 NA 0.2006 0.4119 0.2113 0.4541 0.2536 0.5130 0.5583 NA 0.3172

V15 NA 0.1871 0.4046 0.2175 0.4481 0.2610 0.5376 0.5824 NA 0.3211

V16 NA 0.1856 0.4065 0.2209 0.4506 0.2650 0.5434 0.5881 NA 0.3254

V17 NA 0.1950 0.4075 0.2125 0.4500 0.2550 0.5215 0.5667 NA 0.3168

V18 NA 0.1998 0.4128 0.2129 0.4553 0.2555 0.5159 0.5612 NA 0.3193

V19 NA 0.1942 0.4084 0.2141 0.4512 0.2570 0.5243 0.5695 NA 0.3189

V20 NA 0.1990 0.4136 0.2146 0.4565 0.2575 0.5188 0.5640 NA 0.3215

V21 NA 0.1990 0.4136 0.2146 0.4565 0.2575 0.5188 0.5640 NA 0.3215

V22 NA 0.1998 0.4131 0.2133 0.4558 0.2560 0.5163 0.5616 NA 0.3199

V23 NA 0.1998 0.4131 0.2133 0.4558 0.2560 0.5163 0.5616 NA 0.3199

V24 NA 0.1991 0.4140 0.2149 0.4569 0.2579 0.5191 0.5644 NA 0.3220

V25 NA 0.1991 0.4140 0.2149 0.4569 0.2579 0.5191 0.5644 NA 0.3220

V26 NA 0.2013 0.4114 0.2100 0.4534 0.2520 0.5106 0.5559 NA 0.3156

V27 NA 0.2084 0.4167 0.2083 0.4584 0.2500 0.5000 0.5454 NA 0.3158

V28 NA 0.2008 0.4132 0.2124 0.4557 0.2549 0.5141 0.5594 NA 0.3190

V29 NA 0.1942 0.4084 0.2141 0.4512 0.2570 0.5244 0.5695 NA 0.3189

V30 NA 0.2006 0.4119 0.2113 0.4541 0.2536 0.5130 0.5583 NA 0.3172

V31 NA 0.2006 0.4119 0.2113 0.4541 0.2536 0.5130 0.5583 NA 0.3172

V32 NA 0.1982 0.4146 0.2164 0.4579 0.2597 0.5220 0.5672 NA 0.3239

V33 NA 0.2013 0.4110 0.2097 0.4529 0.2516 0.5102 0.5556 NA 0.3150

V34 NA 0.2013 0.4110 0.2097 0.4529 0.2516 0.5102 0.5556 NA 0.3150

V35 NA 0.1886 0.4032 0.2146 0.4461 0.2575 0.5322 0.5772 NA 0.3174

V36 NA 0.1944 0.4098 0.2154 0.4528 0.2585 0.5256 0.5707 NA 0.3208

V37 NA 0.1952 0.4093 0.2141 0.4521 0.2569 0.5231 0.5683 NA 0.3192

V38 NA 0.1974 0.4065 0.2091 0.4484 0.2510 0.5145 0.5598 NA 0.3127

V39 NA 0.1970 0.4155 0.2185 0.4592 0.2622 0.5258 0.5710 NA 0.3265

V40 NA 0.1998 0.4128 0.2129 0.4553 0.2555 0.5159 0.5612 NA 0.3193

V41 NA 0.2013 0.4110 0.2097 0.4529 0.2516 0.5102 0.5556 NA 0.3150

V42 NA 0.1935 0.4098 0.2164 0.4531 0.2596 0.5279 0.5730 NA 0.3219

V43 NA 0.2028 0.4096 0.2068 0.4509 0.2481 0.5048 0.5502 NA 0.3112

V44 NA 0.2021 0.4105 0.2084 0.4522 0.2501 0.5077 0.5531 NA 0.3134

V45 NA 0.1996 0.4166 0.2170 0.4600 0.2605 0.5210 0.5662 NA 0.3254

V46 NA 0.1981 0.4150 0.2168 0.4583 0.2602 0.5225 0.5677 NA 0.3245

V47 NA 0.1958 0.4070 0.2112 0.4492 0.2535 0.5190 0.5643 NA 0.3152

V48 NA 0.1958 0.4070 0.2112 0.4492 0.2535 0.5190 0.5643 NA 0.3152

V49 NA 0.1908 0.4126 0.2218 0.4570 0.2662 0.5376 0.5825 NA 0.3290

V50 NA 0.1935 0.4093 0.2158 0.4525 0.2590 0.5272 0.5723 NA 0.3211

V51 NA 0.1927 0.4102 0.2174 0.4537 0.2609 0.5301 0.5752 NA 0.3232

V52 NA 0.1871 0.4046 0.2175 0.4481 0.2610 0.5376 0.5825 NA 0.3211

V53 NA 0.1841 0.4081 0.2241 0.4529 0.2689 0.5490 0.5936 NA 0.3295

V54 NA 0.1823 0.4100 0.2276 0.4555 0.2732 0.5553 0.5998 NA 0.3341

V55 NA 0.1935 0.4092 0.2157 0.4524 0.2589 0.5272 0.5723 NA 0.3210

V56 NA 0.1883 0.4147 0.2264 0.4600 0.2716 0.5459 0.5906 NA 0.3347

V57 NA 0.2028 0.4096 0.2068 0.4509 0.2481 0.5048 0.5502 NA 0.3112

V58 NA 0.1982 0.4150 0.2168 0.4583 0.2602 0.5225 0.5676 NA 0.3245

V59 NA 0.2021 0.4105 0.2084 0.4522 0.2501 0.5077 0.5531 NA 0.3134

V60 NA 0.2014 0.4114 0.2100 0.4534 0.2520 0.5105 0.5559 NA 0.3156

V61 NA 0.1998 0.4128 0.2129 0.4553 0.2555 0.5159 0.5611 NA 0.3193

V62 NA 0.2006 0.4123 0.2117 0.4546 0.2540 0.5134 0.5587 NA 0.3177

V63 NA 0.1955 0.4173 0.2218 0.4616 0.2661 0.5315 0.5765 NA 0.3308

V64 NA 0.1935 0.4092 0.2157 0.4524 0.2589 0.5272 0.5723 NA 0.3210

V65 NA 0.1864 0.4055 0.2191 0.4493 0.2630 0.5404 0.5852 NA 0.3232

V66 NA 0.1879 0.4050 0.2171 0.4484 0.2606 0.5361 0.5811 NA 0.3208

V67 NA 0.1841 0.4073 0.2233 0.4520 0.2679 0.5481 0.5928 NA 0.3284

V68 NA 0.1872 0.4072 0.2201 0.4513 0.2641 0.5404 0.5853 NA 0.3249

V69 NA 0.1871 0.4199 0.2327 0.4664 0.2793 0.5543 0.5988 NA 0.3436

V70 NA 0.1849 0.4175 0.2325 0.4640 0.2790 0.5570 0.6014 NA 0.3424

V71 NA 0.1944 0.4097 0.2153 0.4528 0.2584 0.5255 0.5707 NA 0.3207

V72 NA 0.1851 0.4173 0.2322 0.4637 0.2786 0.5564 0.6008 NA 0.3419

V73 NA 0.1946 0.4175 0.2228 0.4621 0.2674 0.5338 0.5787 NA 0.3320

V74 NA 0.2015 0.4124 0.2109 0.4546 0.2530 0.5113 0.5566 NA 0.3169

V75 NA 0.2015 0.4124 0.2109 0.4546 0.2530 0.5113 0.5566 NA 0.3169

V76 NA 0.2023 0.4115 0.2092 0.4534 0.2511 0.5085 0.5538 NA 0.3148

V77 NA 0.1993 0.4150 0.2157 0.4581 0.2588 0.5198 0.5650 NA 0.3233

V78 NA 0.1951 0.4088 0.2137 0.4516 0.2565 0.5227 0.5679 NA 0.3186

V79 NA 0.2015 0.4124 0.2109 0.4546 0.2530 0.5113 0.5566 NA 0.3169

V80 NA 0.1906 0.4217 0.2311 0.4679 0.2773 0.5479 0.5926 NA 0.3426

V81 NA 0.2000 0.4141 0.2141 0.4570 0.2569 0.5170 0.5623 NA 0.3212

V82 NA 0.1932 0.4192 0.2260 0.4644 0.2712 0.5392 0.5841 NA 0.3362

V83 NA 0.1965 0.4178 0.2213 0.4621 0.2656 0.5298 0.5748 NA 0.3305

V84 NA 0.1919 0.4125 0.2205 0.4566 0.2646 0.5347 0.5796 NA 0.3275

V85 NA 0.1944 0.4098 0.2154 0.4528 0.2585 0.5256 0.5707 NA 0.3208

V86 NA 0.1944 0.4098 0.2154 0.4528 0.2585 0.5256 0.5707 NA 0.3208

V87 NA 0.1954 0.2315 0.0361 0.2387 0.0433 0.1560 0.1815 NA 0.0539

V88 NA 0.1950 0.4079 0.2129 0.4505 0.2554 0.5219 0.5671 NA 0.3173

V89 NA 0.1942 0.4084 0.2141 0.4512 0.2570 0.5244 0.5695 NA 0.3189

V90 NA 0.1892 0.4139 0.2247 0.4589 0.2697 0.5430 0.5878 NA 0.3326

V91 NA 0.1942 0.4084 0.2141 0.4512 0.2570 0.5244 0.5695 NA 0.3189

V92 NA 0.1998 0.4127 0.2129 0.4553 0.2554 0.5158 0.5611 NA 0.3192

V93 NA 0.2018 0.4138 0.2120 0.4562 0.2544 0.5122 0.5575 NA 0.3187

V94 NA 0.1967 0.4175 0.2208 0.4616 0.2649 0.5288 0.5738 NA 0.3298

V95 NA 0.2008 0.4132 0.2124 0.4557 0.2549 0.5140 0.5593 NA 0.3190

V96 NA 0.1998 0.4127 0.2129 0.4553 0.2554 0.5158 0.5611 NA 0.3192

V97 NA 0.1998 0.4127 0.2129 0.4553 0.2554 0.5158 0.5611 NA 0.3192

V98 NA 0.1998 0.4127 0.2129 0.4553 0.2554 0.5158 0.5611 NA 0.3192

V99 NA 0.1998 0.4127 0.2129 0.4553 0.2554 0.5158 0.5611 NA 0.3192

V100 NA 0.1998 0.4131 0.2133 0.4558 0.2560 0.5163 0.5616 NA 0.3199

V101 NA 0.1998 0.4127 0.2129 0.4553 0.2554 0.5158 0.5611 NA 0.3192

V102 NA 0.2006 0.4119 0.2113 0.4541 0.2536 0.5130 0.5583 NA 0.3172

[ reached 'max' / getOption("max.print") -- omitted 174200 rows ]

$overall

Ho Hs Ht Dst Htp Dstp Fst Fstp Fis Dest

NA 0.2564 0.3456 0.0893 0.3635 0.1071 0.2583 0.2947 NA 0.1440

**AMOVA analysis using Poppr software**

> amova\_results <- poppr.amova(df\_2\_genclone\_strat, pop, cutoff=0.95)#, clonecorrect = TRUE)

> amova\_results

$call

ade4::amova(samples = xtab, distances = xdist, structures = xstruct)

$results

|  |  |  |  |
| --- | --- | --- | --- |
|  | Df | Sum Sq | Mean Sq |
| Between samples | 5 | 4293769 | 858753.77 |
| Within samples | 220 | 10001020 | 45459.18 |
| Total | 225 | 14294789 | 63532.39 |

$componentsofcovariance

|  |  |  |
| --- | --- | --- |
|  | Sigma | % |
| Variations Between samples | 23038.93 | 33.6344 |
| Variations Within samples | 45459.18 | 66.3656 |
| Total variations | 68498.11 | 100.0000 |

$statphi

|  |  |
| --- | --- |
|  | Phi |
| Phi-samples-total | 0.336344 |

># test for significance

> amova\_test <- randtest(amova\_results, nrepet = 1000)

> amova\_test

Monte-Carlo test

Call: as.randtest(sim = res, obs = sigma[1])

Observation: 23038.93

Based on 1000 replicates

Simulated p-value: 0.000999001

Alternative hypothesis: greater

|  |  |  |
| --- | --- | --- |
| Std.Obs | Expectation | Variance |
| 48.487552 | -6.188493 | 225890.172076 |

The high genetic similarity between the Tanzanian and Ethiopian isolates can be explained by factors such as the variety of finger millet grown. This could be of importance to investigate since the varieties grown in the different regions are adopted based on the planting seasons along with other biotic and geographical considerations which could influence the adaptations of the pathogen (Chadha et al., 2005).

Considering the latitudes of these four countries, Tanzania and Ethiopia are more or less equidistant from the equator while Kenya and Uganda are positioned along the equator. Our results suggest that climatic conditions could be the main factor contributing to the population structure observed since the different methods employed revealed a consistent clustering pattern with regard to the latitude. The latitude is an important factor that determine the climate of a place as it affects the amount of sunlight received and various atmospheric circles (prevailing winds) that affect the distribution of heat and pressure on the surface of the earth (Huang et al., 2014). This is subjective to the presence of other geographical features like elevation and proximity to rivers, lakes and oceans. Environmental conditions have also been shown to play a big role in fungal adaptation and infection of plants (Bidartondo et al., 2018). Despite this, our results show evidence of climatic conditions affecting the diversity of the blast fungus observed across the four countries. The isolates that could be observed overlapping within the two groups could be evidence of seed traveling. Possibilities of horizontal gene transfer between the host and the pathogen have been reported between rice blast and rice (Chadha et al., 2005; Kim et al., 2000). This could be a possible explanation for the clustering patterns observed assuming that the finger millet varieties grown are also influenced similarly by climatic conditions.