Cowpea - MNPs Analysis

Original Data Extracted from VCF File

```
In [1]:
         from VCFtoTable import *
         from GTtable import *
         from GTplots import *
         from GTplot import *
         from BarPlots import *
         from CTbarPlots import *
         from variant_hist import*
         from stats import *
         from FilterVCF import *
         from GTfilter import*
In [2]:
         vcf cowpea = '/home/anibal/genome files/freebayes~bwa~GCF 004118075.1 ASM411807
In [3]:
         samples_all, vcf_df, chrom_len = VCFtoTable(vcf_cowpea)
In [4]:
         samples_all
Out[4]: array(['CBC1_P1', 'CBC5_A1'], dtype=object)
In [5]:
         progenitor = 'CBC1 P1'
         mutant = 'CBC5_A1'
         samples = [progenitor, mutant]
         samples
Out[5]: ['CBC1_P1', 'CBC5_A1']
In [6]:
         vcf df
                     CHROM
                                POS
                                                                                       REF
Out[6]:
              0 NC_018051.1
                                                                                          Τ
                               11786
              1 NC_018051.1
                               11801
                                                                                   TCTTCCT
              2 NC 018051.1
                               11813
                                                                                      AGCC
              3 NC_018051.1
                               11825
                                                                                GGTAGGTAAT AC
              4 NC 018051.1
                                                                                          G
                               18327
                                                                                         ...
         1968687 NC 040289.1 41659114
                                                                                          Т
         1968688 NC_040289.1 41659137
                                                                                          G
         1968689
                NC 040289.1 41667130
                                                                                    GTTTCA
         1968690 NC_040289.1 41667148
                                                                                          Т
```

CHROM POS REF

1968691 NC_040289.1 41668013 CAGGGTTTAGGGTTAGGGTTAGGGTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGGTTTAGGG

```
1968692 rows × 14 columns
```

```
In [7]:
          chrom len
                          LEN
Out[7]:
             CHROM
         NC_040279.1 42129361
         NC_040280.1 33908088
         NC_040281.1 65292630
         NC_040282.1 42731077
         NC_040283.1 48746289
         NC_040284.1 34463471
         NC_040285.1 40876636
         NC_040286.1 38363498
         NC_040287.1 43933251
         NC_040288.1 41327797
         NC_040289.1 41684185
```

PART 0: Raw

152415

NC_018051.1

Contingency Table - RAW - All Chromosomes - (No 0/0, 0/1, 1/1 Filtered)

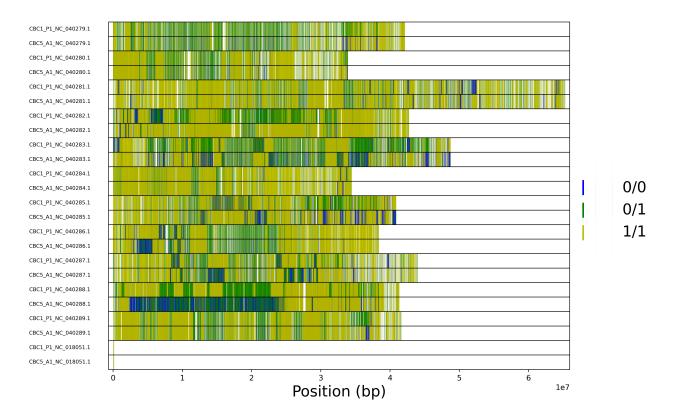
```
contingency_table_0 = contingency_table(samples, vcf_df, 'all')
Contingency Table - Chromosome all
```

```
CBC5_A1_GT
                         0/0
                                  0/1
                                          1/1
                                               other
CBC1_P1_GT 0/0
                           0
                                52496
                                       132507
                                               45048
                      287090
           0/1
                              273476
                                       178974
                                               45048
           1/1
                      211458
                                19728
                                       767915
                                               45048
                       45048
                                45048
                                        45048
                                               45048
           other
```

GT Plot - RAW - All Chromosomes - (No 0/0, 1/1, 'Other' GTs Filtered)

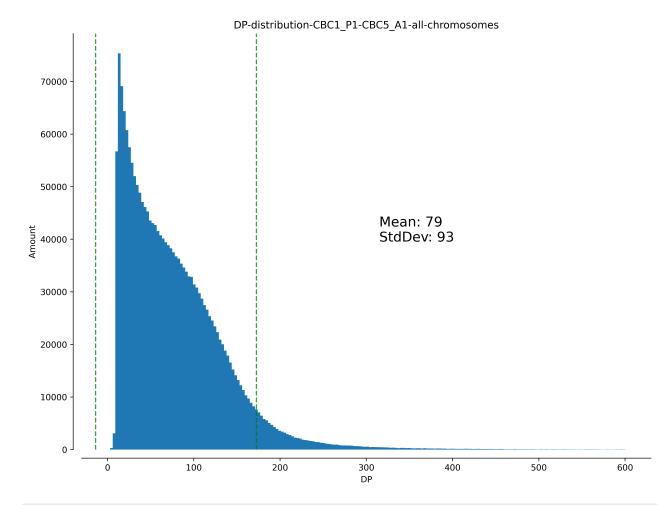
```
In [9]: plt.close('all')
GTplot(samples, vcf_df, chrom_len)
```

gt-plot-CBC1_P1-CBC5_A1

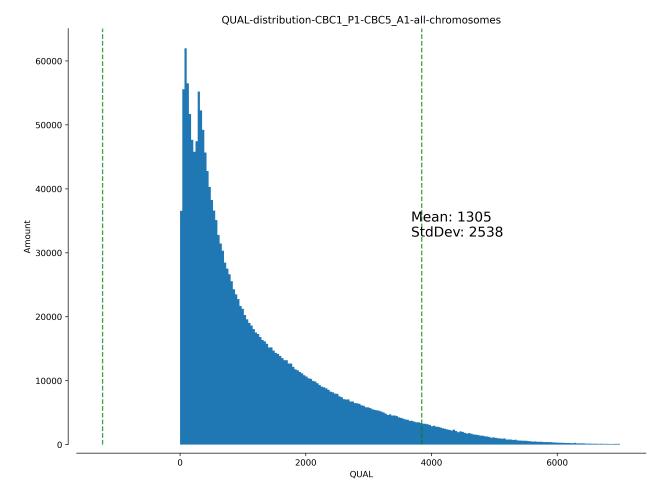


 $\operatorname{Histograms}$ - DP , QUAL , TYPE and GT Attributes - All Chromosomes - $\operatorname{Unfiltered}$

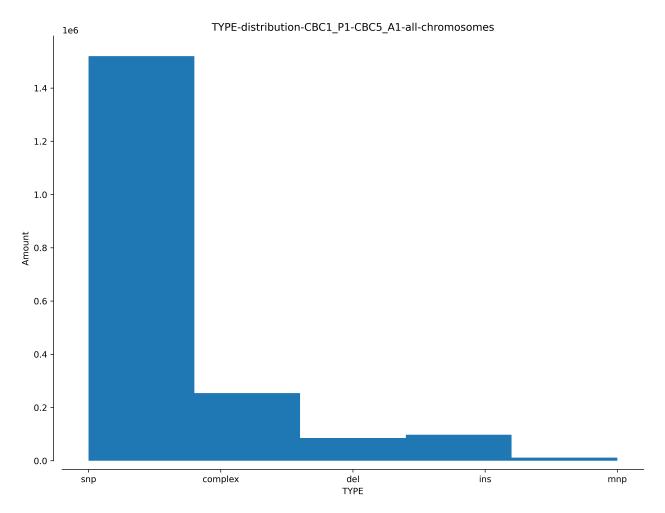
```
In [10]: plot_variant_hist(samples, vcf_df, 'all', 'DP', bins=200, MSTD=True, xmax=600)
```



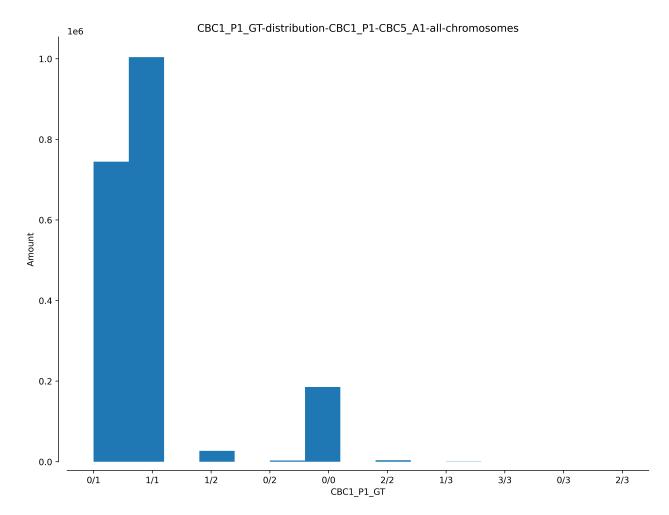
In [11]: plot_variant_hist(samples, vcf_df, 'all', 'QUAL', bins=200, MSTD=True, xmax=706



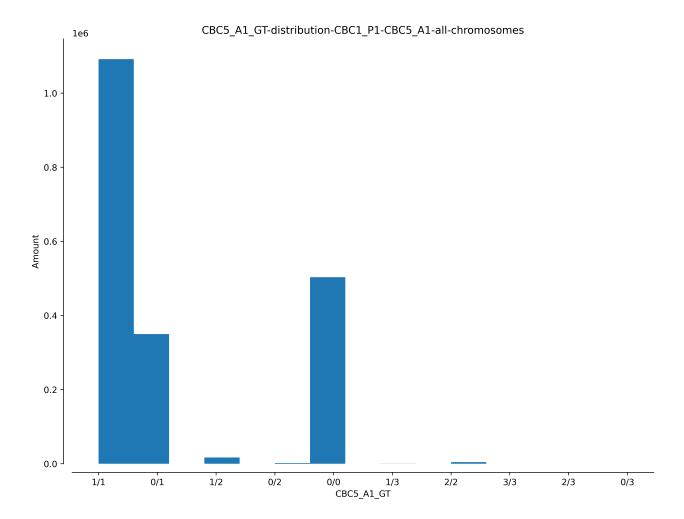
In [12]: plot_variant_hist(samples, vcf_df, 'all', 'TYPE', bins=5)



In [14]: plot_variant_hist(samples, vcf_df, 'all', 'CBC1_P1_GT', bins=15)



In [15]: plot_variant_hist(samples, vcf_df, 'all', 'CBC5_A1_GT', bins=15)



Stacked Bar Plots - RAW

PART 1: Filter Out Mitochondria and Chloroplast Chromosomes

Drop Mitochrondria and Chloroplast Chromosomes from vcf_df and chrom len

```
In [ ]: drop_mito_chloro = "CHROM != NC_018051.1"
    vcf_df_00 = filter_vcf(vcf_df, drop_mito_chloro)
    vcf_df_00
```

```
In [ ]:
         mito chloro = ['NC 018051.1']
         chrom_len_00 = chrom_len.drop(mito chloro)
         chrom len 00
        Create Mitochrondria and Chloroplast Variants and Chomosome Length
        Dataframes
In [ ]:
         drop chrom = "CHROM!=NC 040279.1, CHROM!=NC 040280.1, CHROM!=NC 040281.1, CHROM!
         vcf_df_mito_chloro = filter_vcf(vcf_df, drop_chrom)
         vcf df mito chloro
In [ ]:
         mito chloro len = chrom len.loc[mito chloro]
         mito chloro len
       Contingency Table - No Mitochondria/Chloroplast
In [ ]:
         contingency table 1 = contingency table(samples, vcf df 00, 'all')
       GT Plot - No Mitochondria/Chloroplast
In [ ]:
         # plt.close('all')
         # GTplot(samples, vcf df 00, chrom len 00)
       Histograms - DP, QUAL, TYPE and GT Attributes - No
       Mitochondria/Chloroplast
In [ ]:
         plot variant hist(samples, vcf df 00, 'all', 'DP', bins=200, MSTD=True, xmax=600
In [ ]:
         plot variant hist(samples, vcf df 00, 'all', 'QUAL', bins=200, MSTD=True, xmax=
In [ ]:
         plot_variant_hist(samples, vcf_df_00, 'all', 'TYPE', bins=5)
In [ ]:
         plot variant hist(samples, vcf df 00, 'all', '%s GT' % progenitor, bins=15)
In [ ]:
         plot variant hist(samples, vcf df 00, 'all', '%s GT' % mutant, bins=15)
```

PART 2: Cutting Off by Mean ± 2 StdDev Histograms of DP Attribute

```
cutoff_left = vcf_df_00.DP.mean() - (2 * vcf_df_00.DP.std())
cutoff_right = vcf_df_00.DP.mean() + (2 * vcf_df_00.DP.std())
filter_dp = "DP >= %i, DP <= %i" % (cutoff_left, cutoff_right)</pre>
```

```
print(filter dp)
         vcf_df_01 = filter_vcf(vcf_df_00, filter_dp)
         vcf df 01
       Verify DP Histogram Cutoff Off by Mean±StdDev
In [ ]:
         plot_variant_hist(samples, vcf_df_01, 'all', 'DP', bins=200, xmax=200)
In [ ]:
         plot_variant_hist(samples, vcf_df_01, 'all', 'QUAL', bins=100, MSTD=True)
       Contingency Table After DP Cutoff by Mean±StdDev
In [ ]:
         contingency_table_2 = contingency_table(samples, vcf_df_01, 'all')
       GT Plot After DP Cutoff by Mean±StdDev
In [ ]:
         # plt.close('all')
         # GTplot(samples, vcf_df_01, chrom_len_00)
       Histogram 'GT' Attribute after DP Cutoff
In [ ]:
         plot variant hist(samples, vcf_df_01, 'all', '%s_GT' % progenitor, bins=15)
In [ ]:
         plot_variant_hist(samples, vcf_df_01, 'all', '%s_GT' % mutant, bins=15)
       PART 3: Extract mnp from TYPE Attribute
       Extract mnp TYPE Attribute
In [ ]:
         extract type = "TYPE != snp, TYPE != complex, TYPE != ins, TYPE != del"
         vcf_df_02 = filter_vcf(vcf_df_01, extract_type)
         vcf df 02
       Examples of mnp mutation type
In [ ]:
         vcf df 02[ ['REF', 'ALT'] ].head()
In [ ]:
         vcf df 02[ ['REF', 'ALT'] ].tail()
       TYPE mnp Histogram Verification
In [ ]:
         plot_variant_hist(samples, vcf_df_02, 'all', 'TYPE', bins=5)
```

Contingency Table - mnp TYPE only

```
In [ ]: contingency_table_3 = contingency_table(samples, vcf_df_02, 'all')

GT Plot - mnp TYPE only
```

In []: # plt.close('all') # GTplot(samples, vcf_df_02, chrom_len_00)

Histograms - DP, QUAL, and GT Attributes after TYPE Filtering

```
In [ ]:    plot_variant_hist(samples, vcf_df_02, 'all', 'DP', bins=200, MSTD=True, xmax=600
In [ ]:    plot_variant_hist(samples, vcf_df_02, 'all', 'QUAL', bins=200, MSTD=True, xmax=
In [ ]:    plot_variant_hist(samples, vcf_df_02, 'all', '%s_GT' % progenitor, bins=15)
In [ ]:    plot_variant_hist(samples, vcf_df_02, 'all', '%s_GT' % mutant, bins=15)
```

PART 4: Cutting Off by Mean±StdDev Histograms of QUAL Attribute

```
In []: # cutoff_left = vcf_df_02.QUAL.mean() - vcf_df_02.QUAL.std()
# cutoff_right = vcf_df_02.QUAL.mean() + vcf_df_02.QUAL.std()

# filter_qual = "QUAL >= %i, QUAL <= %i" % (cutoff_left, cutoff_right)
# print(filter_qual)

# vcf_df_03 = filter_vcf(vcf_df_02, filter_qual)
# vcf_df_03</pre>
```

Verify DP and QUAL Histograms after QUAL Cutoff Off by Mean±StdDev

```
In [ ]: # plot_variant_hist(samples, vcf_df_03, 'all', 'DP', bins=200, xmax=200)
In [ ]: # plot_variant_hist(samples, vcf_df_03, 'all', 'QUAL', bins=100, xmax=3500)
```

Contingency Table After QUAL Cutoff by Mean±StdDev

```
In [ ]:  # contingency_table_4 = contingency_table(samples, vcf_df_03, 'all')
# contingency_table_4
```

GT Plot After QUAL Cutoff by Mean±StdDev

```
In [ ]: # plt.close('all')
# GTplot(samples, vcf_df_03, chrom_len_00)
```

Histograms after QUAL Cutoff by Mean±StdDev

```
In [ ]: # plot_variant_hist(samples, vcf_df_03, 'all', 'PAHAT_1_GT', bins=9)
In [ ]: # plot_variant_hist(samples, vcf_df_03, 'all', 'GHP-2-2_GT', bins=9)
```

PART 5: Filtering GTs 0/0, 1/1, 'Other'

Filter out where samples GTs are the same (0/0, 1/1) and have 'Other'

```
In []:
    progenitor_gts_filter = "CBC1_P1_GT != ./., CBC1_P1_GT != 0/2, CBC1_P1_GT != 1/2
    vcf_df_04 = filter_vcf(vcf_df_02, progenitor_gts_filter)

mutant_gts_filter = "CBC5_A1_GT != ./., CBC5_A1_GT != 0/2, CBC5_A1_GT != 1/2, CE
    vcf_df_04 = filter_vcf(vcf_df_04, mutant_gts_filter)

genotypes = ['0/0', '1/1']
    for genotype in genotypes:
        vcf_df_04 = filter_similar_gt(samples, vcf_df_04, genotype)

vcf_df_04
```

Contingency Table after GT Filtering

```
In [ ]: contingency_table_5 = contingency_table(samples, vcf_df_04, 'all')
```

GT Plot after GT Filtering

```
In [ ]: plt.close('all')
GTplot(samples, vcf_df_04, chrom_len_00)
```

Histograms GT after GT Filtering

```
In [ ]: plot_variant_hist(samples, vcf_df_04, 'all', '%s_GT' % progenitor, bins=9)
In [ ]: plot_variant_hist(samples, vcf_df_04, 'all', '%s_GT' % mutant, bins=9)
```

PART 6: Stacked Bar Plots

```
In [ ]: ct_guide()
In [ ]:    plt.close('all')
    window_size = 1000000
```

```
CTbarPlots(samples, vcf_df_04, chrom_len_00, window_size)
```

PART 7: Bar Plots per Chromosome

```
In []:  # suppress all the warnings from the inverted tickes of bar plots
import warnings
warnings.filterwarnings('ignore')

plt.close('all')
GTbarPlots(samples, vcf_df_04, chrom_len_00, window_size)
```

PART 8: GT Plots per Chromosome

```
plt.close('all')
GTplots(samples, vcf_df_04, chrom_len_00)
```

PART 9: Contingency Table per Chromosome

```
import dataframe_image as dfi
for chromosome in chrom_len_00.index:
    chromosome_df = vcf_df_04[ vcf_df_04.CHROM == chromosome ]

# reset chromosome_df indexes for contingency table
    chromosome_df.reset_index(inplace=True, drop=True)
    chromosome_ct = contingency_table(samples, chromosome_df, chromosome)
```