**Methods:**

**Linear mixed model (LMM):**

A large-scale LMM is fitted to all the characteristics of genetic materials data. This is used to assess sources of variation in characteristics of genetic materials at an overall level, i.e. to estimate and interpret the variance components (https://academic.oup.com/bioinformatics/article/30/11/1555/284126). The main purpose of this model is to measure the variation of characteristics of genetic materials. To validate our results, we extended our model by including additional factors i.e. region, and temperature that may be associated with characteristics of genetic materials.

**Results:**

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | ORF | | | | | | N | | | | | |
|  | Ct Value | | Viral Load | | Copy Number | | Ct Value | | Viral Load | | Copy Number | |
|  | Estimate | P-value | Estimate | P-value | Estimate | P-value | Estimate | P-value | Estimate | P-value | Estimate | P-value |
| Without Drainage | 0.860 | 0.087 | -0.083 | 0.395 | -0.084 | 0.426 | 0.937 | 0.062 | -0.149 | 0.140 | -0.167 | 0.132 |
| With Drainage | Ref | - | Ref | - | Ref | - | Ref | - | Ref | - | Ref | - |
| Southern region | -0.006 | 0.993 | 0.035 | 0.806 | 0.016 | 0.916 | -0.988 | 0.180 | 0.110 | 0.456 | 0.089 | 0.583 |
| Central region | 0.997 | 0.159 | -0.179 | 0.195 | -0.200 | 0.180 | -0.147 | 0.836 | 0.003 | 0.981 | -0.007 | 0.960 |
| North-Eastern region | 0.556 | 0.441 | -0.205 | 0.145 | -0.217 | 0.154 | -0.175 | 0.808 | -0.004 | 0.979 | -0.037 | 0.815 |
| Northern region | Ref | - | Ref | - | Ref | - | Ref | - | - | - | Ref | - |
| Temperature | 0.010 | 0.404 | -0.002 | 0.384 | -0.002 | 0.474 | 0.001 | 0.966 | -0.001 | 0.881 | -0.001 | 0.955 |

Table X has given the relationship of Ct value, viral load, and copy number with the category of the drainage system and four regions. The LMM also explained the relationship characteristics of genetic materials with Temperature. Adding the covariates in the model as explanatory variables, all the variables reduced substantially the variability. Thus, this low variability shows that the effect of explanatory variables was not significant as a p-value greater than 0.05. However, for the Ct value of the both gene, according to estimate (coefficient), it showed a positive relation with drainage and non-drainage system. That means with the increase of without drainage 0.860 and 0.937 times increase Ct value by comparing to the drainage system for ORF and N Gene, respectively. In contrast, viral load and copy number of ORF and N gene estimate negative relation. The estimate of viral load was -0.083 and -0.149 in ORF and N gene, respectively and the estimate of copy number was -0.084 and -0.167 in ORF and N gene, respectively. According to our study, temperature was not shown any significant relationship with the characteristics of genetic materials.