

# **GENETICS OF BIOLOGICAL NITROGEN FIXATION EFFICIENCY IN COWPEA**

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## **Abstract**

Cowpea (*Vigna unguiculata* [L.] Walp) is a critical grain legume in Sub-Saharan Africa, valued for its high protein content, adaptability to marginal environments, and its capacity to improve soil fertility through Biological Nitrogen Fixation (BNF). Despite its importance, the genetic mechanisms underlying BNF efficiency in cowpea remain insufficiently characterized, limiting the potential for targeted breeding of nitrogen-efficient varieties. This study was designed to elucidate the genetic and environmental determinants of BNF efficiency in cowpea and to identify superior genotypes and molecular markers for use in breeding programs.

The research was conducted in three major phases. The first phase involved the phenotypic evaluation of 252 cowpea genotypes under controlled screen house conditions to assess key BNF-related traits, namely the number of nodules (NN), active nodules (AN), nodule dry weight (NDW), nitrogen content in shoots (NC) and plant vigor. Significant genetic variability was observed across these traits. Genotypes such as TVu-14971, TVu-1477, and TVu-14691 exhibited superior nodulation and plant vigor performance. Multivariate analyses, including principal component analysis and hierarchical clustering, confirmed the existence of distinct groups within the evaluated material, highlighting genotypes with exceptional BNF efficiency.

The second phase evaluated 35 selected genotypes across seven contrasting environments in Uganda to investigate genotype  $\times$  environment (G $\times$ E) interactions. Using Additive Main Effects and Multiplicative Interaction (AMMI) and Genotype and Genotype  $\times$  Environment (GGE) biplot models, the study revealed that environmental variation had a substantial effect on both nodulation and seed yield. Genotypes TVu-1477 and TVu-14691 showed high and stable performance across all environments, confirming their broad adaptability and suitability for breeding programs targeting multiple regions.

In the third phase, a Genome-Wide Association Study (GWAS) was conducted on 232 cowpea genotypes using 40,246 SNP markers and the mrMLM.GUI software. Seventeen significant SNPs associated with BNF traits were identified across ten chromosomes. Candidate genes such as Vigun06g121800, Vigun01g160600 and Vigun07g221500 linked to nodule formation, and

Vigun09g246400 associated with nitrogen metabolism, were pinpointed. TVu-1477 emerged as a multi-trait superior genotype, possessing favorable alleles for all major BNF traits. The identified SNPs are proposed for conversion into Kompetitive Allele Specific-PCR (KASP) markers to facilitate marker-assisted selection in future cowpea breeding.

This study provides critical insights into the phenotypic and genotypic variation governing BNF efficiency in cowpea. It delivers a framework for the development of cowpea varieties that are not only high-yielding but also efficient in nitrogen fixation, thereby reducing reliance on inorganic fertilizers for enhancing soil fertility. These findings promise to improve smallholder farmer productivity and support sustainable agricultural systems in SSA.

The outcomes of this research directly contribute to multiple United Nations Sustainable Development Goals (SDGs). Specifically, the work supports SDG 2 (Zero Hunger) by enhancing food security through increased cowpea productivity and resilience. It contributes to SDG 13 (Climate Action) by promoting climate-smart agricultural practices that reduce dependency on nitrogen fertilizers, thereby mitigating greenhouse gas emissions. Additionally, it aligns with SDG 12 (Responsible Consumption and Production) by enhancing natural soil fertility and promoting sustainable land use.