



Unique Floral Morphology of Double Grained Heirloom Rice (*Oryza sativa* L.) Variety, Jugal, of West Bengal, India and the Need of its Conservation

Santi Ranjan Dey¹, Mitu De^{2*}

¹Department of Zoology, Rammohan College, Kolkata 70000, India

²Department of Botany, Gurudas College, Kolkata-700054, India

*Corresponding Author's Email: mitubotany@gmail.com & mitu.botany@gurudas.education

Abstract

Jugal, the unique heirloom rice folk or traditional variety from West Bengal, India, is a natural rice genotype which produces multiple fertile seeds. In this present investigation the floral development of double grained variety, Jugal is studied. It has been reported that homeotic mutation in a number of genetic loci causes an increased number of stamens and carpels. In rice (*Oryza sativa* L.) the unique structure of the inflorescence and flower has greatly diverged from those of model eudicots. In contrast to eudicot flowers which typically exhibit sepals and petals at their periphery, the flowers of grasses are distinguished by the presence of characteristic outer organs. The structural units of the grass flower are spikelets and florets. The spikelet is the primary unit of the grass inflorescence, and it comprises glumes (bract-like organs) and florets. The rice floret consists of one lemma, one palea, two lodicules, six stamens, and one central pistil that contain one pistal. Multiple-pistillate mutants are produced in rice to study the flower development genetics. Molecular studies of rice flowering require a number of mutant varieties of different abnormalities, which is produced by different techniques. Jugal, a natural double grained variety may be an important tool for the study of molecular biology of flowering in rice. Jugal needs to be conserved for use in molecular studies of floral development in rice. Documentation of Jugal must be undertaken before this heirloom rice variety fades away into oblivion.

Keywords: Floral Morphology; Double Grained; Heirloom Variety; Jugal; Molecular Biology of Flowering

Introduction

Jugal, a unique heirloom rice variety (*Oryza sativa* L.) from West Bengal, India, is renowned for its rare double-grain characteristic, where two grains develop within a single spikelet. Jugal is a folk variety or traditional rice cultivar which is distinguished by its unique floral morphology that results in multiple kernels per spikelet. Cultivated in specific regions of West Bengal and adjoining areas, Jugal holds cultural and agricultural significance due to its unique morphology, traditional value, and potential applications in breeding programs. This extraordinary feature not only enhances its cultural and historical significance but also offers immense potential for advancing rice genetics and breeding programs. However, with the increasing focus on commercial high-yield varieties (HYVs), Jugal is at risk of extinction, underscoring the urgent need for its conservation.

Heirloom Rice Varieties

These are folk or traditional varieties cultivated by indigenous farmers in rice terraces and passed on as heirlooms to the next generations. The unique floral characteristic of Jugal, the heirloom variety, is attributed to the development of multiple carpels within individual florets. The floral architecture of grass species is distinct from those of eudicots and other monocots. Specific to grass species, including rice, are the structural units of the inflorescence called the spikelet and floret, which comprise grass-specific peripheral organs and conserved sexual organs. The genetic framework of rice flower development is in part similar to that of model eudicots. The structural units of the grass flower are spikelets and florets. The spikelet, the basic unit of the grass inflorescence, consists of glumes and one to 40 florets that comprise the lemma and palea (possibly homologous to sepals), lodicules (homologous to petals),

Received on :7th December 2024; Revised version received on :15th January 2025; Accepted: 17th January 2025

stamens, pistils, and ovules (Bommert *et al.*, 2005; Itoh *et al.*, 2005; Malcomber *et al.*, 2006). The flower of rice diverged from those of model eudicot species such as *Arabidopsis*, *Antirrhinum*, or *Petunia*, and is thus of great interest in developmental and evolutionary biology.

The rice (*Oryza sativa* L.) flower is composed of a, six stamens and two lodicules. The flower is enclosed by a pair of bract-like structure called the lemma and palea. The structure including lemma, palea and a flower is referred to as a floret (Kyojuka, 1999). Rice produces flower of typical grass species (Fig. 1). It has several advantages as a model species for studying molecular biology (Izawa & Shimamoto, 1996). A number of genes have been identified in rice which are responsible for the development of floral structure. These are named as RFL (Rice FLO/LFY), Rice APIA and Rice APIB etc. The rice floral biology is also important in maintaining varietal purity. Typically, short style and stigma, short anthers, limited pollen availability, rapid decline of pollen viability and a brief period (between 30 sec to 9 minutes) between opening of florets and release of pollen physically reduce cross pollination frequencies (Deb & Bhattacharya, 2005).

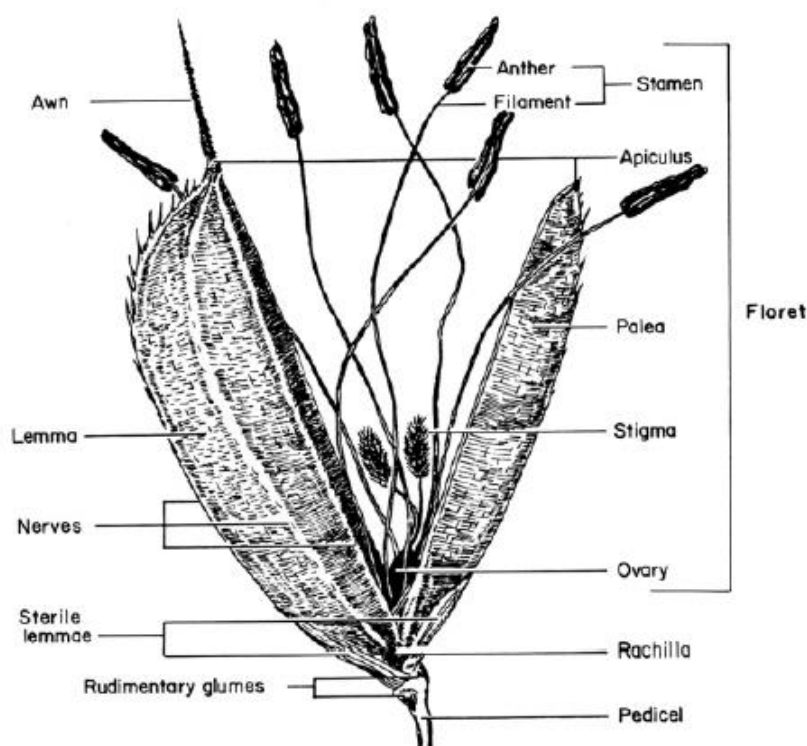


Figure 1: A typical rice floret

Multiple-pistillate Folk or Traditional Variety of Rice

In spite of the unrelenting advent of modernization involving industrialization and market-oriented land use policies, landraces survive in many indigenous societies (De, 2014; Deb & Malhotra, 2001). Jugal, an indica variety from West Bengal, India is a natural Multiple-pistillate landrace of rice which produces multiple fertile seeds. Multiple-pistillate mutants are produced in rice to study the flower development genetics. These are mostly of a bi- to tetra pistillate in nature are reported to be controlled by two one or two recessive genes (Suh, Heu & Khush, 1983). It has been reported that homeotic mutation in a number of genetic loci causes an increased number of stamens and carpels. Complete sterile natural mutant TDC 72 was reported from India (Li RongBai & Pandey, 2002).

Material and Methods

1. Multiple-pistillate rice folk variety, Jugal was collected from Midnapore district. It was propagated in the Bose Institute Experimental Farm in Madhyamgram.
2. 30 individual plants of the Jugal was used for the study of flower development.
3. The flowers were collected in different stage, starting from the booting stage to the grain filling stage.

4. The collected flowers were fixed in 70% ethyl alcohol.
5. These flowers were dissected, studied under microscope and a camera lucida drawing was prepared.
6. Photographs of different stages are taken (Fig 2a and 2b).
7. The dual seeded rice was germinated to examine whether both the seeds were fertile or not.

Results

When florets of Jugal were dissected it was seen that it had two carpels in most cases. In some rare incidences three carpels were also found. Photographs were taken at various stages. The dual seeded rice was germinated. It was seen that 2 seedlings emerged from the seed of Jugal germinated (Fig. 3).



Figure 2a: Dissected floret of the landrace, Jugal showing two carpels



Figure 2b: Two carpels from one floret of Jugal, each with bifid feathery stigma



Figure 3: Germination of two seedlings from a dual seed of Jugal

Discussion

Understanding of grass inflorescence morphogenesis has expanded significantly over the last two decades. Extensive studies in model plants have demonstrated common genetic factors regulating eudicot and grass floral development including MADS-box and non-MADS-box genes and epigenetic regulators (Ali *et al*, 2019). The spikelet which is a unique flower structure of grass inflorescence, contains highly specialized non-reproductive organs (Yuan, Persson & Zhang, 2020). OsMADS32, an orphan protein in monocotyledonous plants, regulates floral context by interacting with other floral homeotic proteins (Hu *et al*, 2021).

Jugal, a naturally occurring folk or traditional variety, may be an important tool for the study of molecular biology of flowering in rice. From the study of the development of gynoecium in Jugal, it is evident that the regulatory mechanism in flower development has some abnormalities. These inherent abnormalities itself may provide a clue in studying the developmental genetics of the rice floral development. The unique floral morphology of Jugal, characterized by its multi-ovary and multi-kernel traits, offers valuable insights for rice breeding programs aiming to enhance grain yield. Understanding the genetic and developmental mechanisms underlying these traits could contribute to the development of new rice varieties with improved productivity.

Research on Jugal shows that at first the floral meristems became enlarged and the enlarged floral meristem specifically produces increased number of carpels (Das, Deb & Dey, 2018). Recent research suggests that rice lodicule is a bracteopetal organ derived from a modified leaf, rather than an andropetal stamen-derived organ. In this case, duplication of rice B-class genes may have contributed to the diversification of petal-like organs in grasses, just like in eudicot (Wang *et al*, 2024).

From field visits and talks with the farmers it became apparent that most farmers do not want to cultivate Jugal in their rice fields. According to them Jugal does not fetch a good market price. As there are two seeds none of them attain good grain dimensions which is one reason for a low market price. However recent molecular studies of rice flowering requires a number of mutant varieties of different abnormalities, which is produced by different techniques. Widespread adoption of high-yielding rice varieties (HYVs) has led to the biological poverty of rice germplasms, as local folk rice varieties/landraces or rare heirloom rice varieties are abandoned for modern varieties (Gao, 2003).

Conclusion

Fabio Fornara, Professor at the University of Milan, Italy states that for 'guiding better breeding, driving selection with molecular rather than phenotypic data, and quickly tailoring new varieties to specific cultivation environments, possibly also with the use of gene-editing technologies will be the most daunting task, requiring close and constructive interactions between scientists and breeders' (Vicentini *et al.*, 2023). So for any molecular studies it is imperative to conserve these unique varieties which possibly holds the key to many unanswered questions of floral morphology and its genetics.

Jugal needs to be conserved for use in molecular studies of floral development in rice. Documentation of Jugal must be undertaken before this heirloom rice (*Oryza sativa* L.) variety fades away into oblivion. The conservation of Jugal is crucial for preserving its unique genetic traits, which have significant implications for understanding floral genetics and enhancing rice production. By integrating traditional knowledge with modern science, we can safeguard this heirloom variety, ensuring its availability for future generations and its potential contribution to global food security.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this work.

Acknowledgment

The author gives thanks to the Director, Bose Institute, Kolkata and Dr. Tapas Kumar Ghose of Bose Institute for providing the laboratory and experimental garden infrastructure.

References

- Ali, Z., Raza, Q., Atif, R. M., Aslam, U., Ajmal, M., & Chung, G. (2019). Genetic and molecular control of floral organ identity in cereals. *International Journal of Molecular Sciences*, 20(11), 2743. <https://doi.org/10.3390/ijms20112743>
- Bommert, P., Satoh-Nagasawa, N., Jackson, D., & Hirano, H. Y. (2005). Genetics and evolution of inflorescence and flower development in grasses. *Plant and Cell Physiology*, 46(1), 69-78. <https://doi.org/10.1093/pcp/pci504>
- Das, S. P., Deb, D., & Dey, N. (2018). Micromorphic and molecular studies of floral organs of a multiple seeded rice (*Oryza sativa* L.). *Plant Molecular Biology Reporter*, 36, 764-775. <https://doi.org/10.1007/s11105-018-1116-9>
- De, M. (2014). Biocultural Diversity of Folk Rice Variety Heritage of West Bengal. *Journal of Environment and Sociobiology*, 95-100.
- Deb, D., & Bhattacharya, D. (2005). *Seeds of tradition, seeds of future: Folk rice varieties of Eastern India*. Research Foundation for Science, Technology, and Ecology. Retrieved from: <https://www.scirp.org/reference/referencespapers?referenceid=1547975>
- Deb, D., & Malhotra, K. C. (2001). Conservation ethos in local traditions: the West Bengal heritage. *Society & Natural Resources*, 14(8), 711-724. <http://dx.doi.org/10.1080/08941920152524909>
- Gao, L. Z. (2003). The conservation of Chinese rice biodiversity: genetic erosion, ethnobotany and prospects. *Genetic Resources and Crop Evolution*, 50, 17-32. <https://doi.org/10.1023/A:1022933230689>
- Hu, Y., Wang, L., Jia, R., Liang, W., Zhang, X., Xu, J., ... & Yuan, Z. (2021). Rice transcription factor MADS32 regulates floral patterning through interactions with multiple floral homeotic genes. *Journal of Experimental Botany*, 72(7), 2434-2449. <https://doi.org/10.1093/jxb/eraa588>
- Itoh, J. I., Nonomura, K. I., Ikeda, K., Yamaki, S., Inukai, Y., Yamagishi, H., ... & Nagato, Y. (2005). Rice plant development: from zygote to spikelet. *Plant and Cell Physiology*, 46(1), 23-47. <https://doi.org/10.1093/pcp/pci501>
- Izawa, T., & Shimamoto, K. (1996). Becoming a model plant: the importance of rice to plant science. *Trends in Plant Science*, 1(3), 95-99. [https://doi.org/10.1016/S1360-1385\(96\)80041-0](https://doi.org/10.1016/S1360-1385(96)80041-0)
- Kyozuka, J. (1999). Flower development of rice. *Molecular Biology of Rice*, 43-58. <https://doi.org/10.1093/jxb/err272>
- Li RongBai, L. R., & Pandey, M. P. (2002). The spt1 locus for sept-pistillate spikelet mutant in rice. *International Rice Research Notes*, 27(1), 18-19. Retrieved from: <https://www.cabidigitallibrary.org/doi/full/10.5555/20023156875>
- Malcomber, S. T., Preston, J. C., Reinheimer, R., Kossuth, J., & Kellogg, E. A. (2006). Developmental gene evolution and the origin of grass inflorescence diversity. *Advances in Botanical Research*, 44, 425-481. [https://doi.org/10.1016/S0065-2296\(06\)44011-8](https://doi.org/10.1016/S0065-2296(06)44011-8)
- Suh, H. S., Heu, M. H., & Khush, G. S. (1983). Inheritance of polycaryopsis and breeding of polycaryoptic male-sterile rice. *International Rice Research Newsletter*, 8(3): 6-7. Retrieved from: <https://www.cabidigitallibrary.org/doi/full/10.5555/19831624045>
- Vicentini, G., Biancucci, M., Mineri, L., Chirivì, D., Giaume, F., Miao, Y., ... & Fornara, F. (2023). Environmental control of rice flowering time. *Plant Communications*, 4(5). <https://doi.org/10.1016/j.xplc.2023.100610>
- Wang, L., Li, Q. L., Hu, J. P., & Yuan, Z. (2024). Neofunctionalization of B-class genes in regulating rice flower development. *Seed Biology*, 3(1). <https://doi.org/10.48130/seedbio-0024-0012>
- Yuan, Z., Persson, S., & Zhang, D. (2020). Molecular and genetic pathways for optimizing spikelet development and grain yield. *Abiotech*, 1(4), 276-292. <https://doi.org/10.1007/s42994-020-00026-x>