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A REVIEW :

MAGIC : A magical genetic resource for multiple trait enhancements in rice

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SUMMARY : MAGIC is the multi-parent advanced generation inter-cross. It is a simple extension of the advance inter cross. The MAGIC is an alternative resource for the genetic dissection of complex traits. The development of MAGIC population initiated by using the two major ecotypes: indica and japonica. Japonica rice grains are short, roundish, spikelet's are awnless to long awned and having 0-20 per cent amylose content in grain. Whereas counterpart indica rice grains are long to short selender grain, awnless spikelets and 23-31 per cent amylose content observed in grain. In rice, developed 4 multi-parent populations: indica MAGIC (8 indica parents); MAGIC plus (8 indica parents with two additional rounds of 8-way F_1 inter-crossing); japonica MAGIC (8 japonica parents); and Global MAGIC (16 parents – 8 indica and 8 japonica). The parents used in creating these populations are improved varieties with desirable traits for biotic and abiotic stress tolerance, yield and grain quality. The purpose is to fine map QTLs for multiple traits and to directly and indirectly use the highly recombined lines in breeding programmes.

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BACKGROUND AND OBJECTIVES

Rice is a major staple food feeding over 50 per cent of the global population. Cultivated rice was domesticated from the common ancestor of wild rice (*Oryza rufipogon*) under both natural and human selective pressures and displays large genetic diversity across thousands of varieties (Khush, 1997 and Huang *et al.*, 2012). Rice cultivars are mainly grouped into two subspecies Japonica and Indica with marked differences in plant architecture, agronomic and physiological features (e.g. stress resistance, cold tolerance, and seed quality) (Khush, 1997). Although Japonica and Indica cultivars exhibit clear variation in genome sequences as well as in the morphological and physiological features (Huang, 2010 and Huang, 2012).

MAGIC is the multi parent advanced generation inter-cross. The development of MAGIC in rice and discusses potential applications for mapping quantitative trait loci (QTLs) and for rice varietal development. Bandillo *et al.* (2013) developed 4 multi-parent populations: indica MAGIC (8 indica parents); MAGIC plus (8 indica parents with two additional rounds of 8-way F_1 inter-crossing); japonica MAGIC (8 japonica parents) and Global MAGIC (16 parents – 8 indica and 8 japonica). The parents used in creating these populations are improved varieties with desirable traits for biotic and abiotic stress tolerance, yield and grain quality. The purpose is to fine map QTLs for multiple traits and to directly and indirectly use the highly recombined lines in breeding programmes. These MAGIC populations provide a useful germplasm resource with diverse allelic combinations to be exploited by the rice community.

It is a simple extension of the advanced inter cross which seeks to recombine the genetic makeup of many different highly successful variety and allowing them reshuffle to see new combinations of gene which are previously not imagined. The MAGIC is an alternative resource for the genetic dissection of complex traits. In comparison to mapping in bi-parental crosses, the increased recombination and diversity of MAGIC gives greater precision in QTL location and greater opportunity to detect more QTL for multiple traits. MAGIC populations represent one of a new generation of crop genetic mapping resources combining high genetic recombination and diversity. The main objective of developing MAGIC population is to promote intercrossing and shuffling of the genome. Multi-parent populations (MPPs) have emerged as powerful nextgeneration mapping resources combining diverse genetic founder contributions with high levels of recombination (Mackay and Powell, 2007 and Ongom and Ejeta, 2018). MAGIC population fulfills the major limitations of existing mapping populations (Bandillo et al., 2013 and Huang et al., 2015).

Startup of MAGIC population concept:

The concept was first proposed and applied to mice as 'heterogeneous stock' by Mackay and Powell (2007) and in plants, MAGIC populations were first developed and described in Arabidopsis (Cavanagh *et al.*, 2008; Huang *et al.*, 2012 and Kover *et al.*, 2009). Cavanagh *et al.* (2008) discuss in detail the production of the MAGIC population, methods of mapping genes to traits and the relevance of such population to breeders. In Arabidopsisa MAGIC population was derived from inter-crossing of 19 accessions and mapping of quantitative traits detected QTLs contributing to low levels of phenotypic variation (Kover *et al.*, 2009). Arabidopsis multi-parent RIL (AMPRIL) populations were genotyped at the F_4 stage and phenotyped at F_5 and mixed-models were used for fine mapping (Huang et al., 2012). More recently, Huang et al. (2012) demonstrated the use of a large MAGIC wheat population for mapping QTLs underlying complex traits such as plant height and hectolitre weight. The increased recombination in MAGIC populations can lead to novel rearrangements of alleles and greater genotypic diversity. Given the potential benefits of MAGIC populations, IRRI scientist initiated the development of MAGIC populations using the two major rice ecotypes: indica and japonica aimed to produce at least 2,000 inbred lines to capture the broadest genotypic diversity through intercrossing of 8 indica parents for indica MAGIC and 8 japonica parents for the japonica MAGIC population. Subsequently, MAGIC populations have been developed in wheat (Cavanagh et al., 2008; Mackay et al., 2014; Huang et al., 2014 and Rebetzke et al., 2014), chickpea (Gaur et al., 2012), sorghum (Higgins et al., 2014), maize (Dell'Acqua et al., 2015), barley (Sannemann et al., 2015), tomato (Pascual et al., 2015), Strawberry (Wada et al., 2017) etc.

Use multi parent population:

Breeders and molecular geneticists have routinely used populations derived from bi-parental crosses for variety development and mapping quantitative trait loci (QTLs) for traits of interest. Most of the varieties developed by breeders in self-pollinated crops like rice are based on single crosses between two parents. Breeders have also attempted to make multiple crosses (e.g., three-way involving three parents, or double crosses involving four parents) to increase the genetic variation in breeding populations however, extensive use of these multiplecrosses may be restricted by technical limitations (e.g., intensive labour for crossing and large population sizes required for recovering recombinants with all the desira traits). More complex crossing schemes involving 6- way, 8-way crosses or diallel selective mating for use in breeding of self - pollinated crops were proposed many years ago (Allard, 1960 and Jensen, 1970) but have seldom been used in plant breeding programmes.

More targeted traits from each of the parents can be analyzed based on the selection of parents used to make the multi-parent crosses.Increased precision and resolution with which QTLs can be detected due to the increased level of recombination.In bi-parental population only two alleles can be analyze and that genetic recombination in this population is limited.

Production of MAGIC populations:

Bandillo et al. (2013), worked with two initial MAGIC populations. They developed it by inter-crossing eight elite lines from the Asia indica pool (indica MAGIC) and the japonica group (japonica MAGIC). Each population is comprised of 8 founder lines that include elite and modern varieties known to exhibit high yield potential, good grain quality and tolerance to a range of biotic and abiotic stresses. The indica and japonica MAGIC populations followed the same scheme of development. The first stage followed a half-diallele mating system by inter-mating the eight founder lines and 28 bi-parental crosses were made. The resulting 28 F₁'s were inter-crossed to derive 4-way crosses for which 70 such 4-way crosses (out of 210 possible crosses) were made. The 70 crosses were selected in such a manner that no parent was represented more than once in the 4-way cross. Also only one of the possible 4way combinations was selected (e.g., one of ABCD or ACBD or BCAD etc.). The last stage involved intercrossing of the 70 4-way crosses to derive 8-way crosses. Only 35 out of 105 such possible 8-way crosses were made keeping in mind that no parent was represented more than once. From each of the 35 8way crosses ~60 seeds were advanced by selfing. Single plant selections were made to advance to the next generation. Thereby the population size targeted was 35 \times 60 = 2100 lines. A pedigree for a line in the MAGIC indica population would for example be written as A/C// E/H///B/F//D/G.In the case of the MAGIC plus population the 8-way crosses derived during the development of the MAGIC population underwent two extra-rounds of inter-crossing prior to selfing. A set of 175 multiple crosses were made for each of the inter-crossing rounds.

The pedigree for a line derived from the MAGIC plus population would for example be written as:

(H/E//A/F///C/D//B/E/4/A/E//F/C///B/H//D/G/5/C/ H//E/F///A/B//D/G/4/H/E//G/A///D/C//B/F).

The Global MAGIC population was developed by crossing the 8-way crosses derived during the development of the indica MAGIC population to the 8way crosses derived during the development of the japonica MAGIC population. A total of 150 such multiple crosses were made. Therefore, the Global MAGIC population is representative of 16 parents (8 indica type and 8 japonica type). The 16-way crosses were then advanced by selfing. The indica MAGIC population consists of 2000 lines advanced by single seed descent (SSD) of which currently 1328 are in the S7 generation and the remaining lines are following as they are late maturing.

The japonica MAGIC population is smaller relative to the indica MAGIC population, consisting of 500 lines at the S5 stage. This population will be suitable to temperate environments, and these lines will initially be tested for their performance in Korea. The MAGIC plus population is currently at the S4 stage and is potentially more valuable due to the added round of inter-crossing. This population is expected to further extend the amount of recombination when compared to the indica MAGIC population. We hope to address whether the extra two rounds of inter-crossing increased the levels of recombination enough to enable direct fine-mapping of QTLs using the MAGIC population.

The Global MAGIC is an attempt to combine traits from both indica and japonica gene pools that have been adapted to different environments. In addition to being a source of potential novel variation, the Global MAGIC population provides useful materials for studying the relative contributions from the two rice ecotypes and the level of recombination among 16 genomes constituting this population. The crossing schemes for the different MAGIC populations are shown in Fig. 1a and 1b.



Fig. 1a : Development of MAGIC population for Indica





Fig. 1b : Development of MAGIC- plus and MAGIC global populations

Advantages:

- Shuffling the genes across different parents enable accurately ordering the genes.

- Increased recombination - novel rearrangements of alleles and greater genetic diversity.

- Best combinations of genes for important traits development.

- 1000 Magic individuals.

– Fine mapping.

- Epistatic and G X E interactions.

- Facilitate the discovery, identification and manipulation of new forms of allelic variability.

Disadvantages:

- Extensive segregation.

– More time consuming approach.

- Large scale phenotyping.

Future opportunities of MAGIC rice development:

- Requirement of food security and self-sufficiency in rice

– Rice export opportunity

– Progress in technology innovations

- Increased trends in public-private partnership (PPP)

– Increased support to IPR – plant breeder right Future.

Conclusion:

The MAGIC populations can also be explored to study Inter-allelic interactions. This may be more challenging as the background is a complex mix of eight parents. For instance a QTL may not be effective in a single background (one of the eight parents), but maybe be effective in the MAGIC populations (a mixed background of the eight parents). The MAGIC populations also present opportunities for studying the interactions of genome introgressions and chromosomal recombination.

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