

## A Review

# Genetic basis of Aroma gene badh1 and badh2 in rice

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### Abstract

An indigenous to the Indian sub-continent, basmati rices evolved with quality traits that are in highest demands of the rice cultivars. This ends up in its premium prices in domestic moreover as international market. Many studies are conducted to grasp the extent of diversity and unique characteristics of this special rice. One among the foremost unique trait of basmati rice is its pleasant aroma, which is liable for its high demands and premium prices. There are many naturally found traditional basmati and a few are evolved from these traditional basmati referred to as evolved basmati by many time organic process, many studies are conducted to grasp the genetics behind the aroma and that they concluded that BADH gene present on chromosome 8 and chromosome 4, most of the researchers assume that the fragrance is controlled by one factor (fgr), on the 8th chromosome of rice genome, that's one gene closely associated with the fragrance, and therefore the fgr gene has been separated and cloned at the moment. With the advancement in sequencing technology and functional genomics of rice, there has been a great progress to understand the aroma gene in rice, for screening of aroma gene and breeding new rice varieties with better-quality a series of functional markers has been developed. This review article mainly focuses on genetic basis of aroma in aromatic rice, gene function and regulation of aroma gene and application of functional markers present in the genetic improvement of aromatic rice.

**Keywords:** gene function, aromatic rice, fragrance, gene

### 1. Introduction

Rice (*Oryza sativa*L.) is the most staple food crop in the world, rice provides 35% of energy (Peng et al, 2014a; Li et al., 2015) to mostly south Asians and supplies 25% of energy to more than 3 billion people (Tian et al., 2009; Peng et al., 2017a). A varied association which has distinguished itself as conclusion of human and natural selection which has been accepted worldwide rice is known as “basmati”, Basmati has been in

cultivation since 1700 AD. Basmati accepted as the best aromatic due to its unique and special characteristics, fragrance counts as one of the most important eating quality traits in rice (Tang et al. 2009). This special trait of these varieties are responsible for their high market price and , as standard of living enhances demand for aromatic rice has been significantly increased in recent years (Shao et al., 2013; Shi et al., 2014; Peng et al, 2016a). The traditional rice varieties having poor resistance and lower yield are difficult to be introduced in large areas (Bai et al, 2009; Du et al, 2009) , hence aroma gene research and its implementation in genetic breeding has been reviewed by many geneticists and rice breeders.

With the enhancement of living standards and continuous growth in world population traditional rice breeding is not able to satisfy the strong demands for high quality and good fragrant rice (Peng et al, 2016b).new breeding methods like whole genome breeding , marker assisted selection , all these breeding strategies will cover the shortage of traditional rice breeding to have good rice yield and improvement of rice quality simultaneously. Great progress have been made in the genetics and breeding of aromatic rice , which also includes isolation and cloning of aroma genes and their functional analysis (shan et al., 2013, 2015; he and park et al., 2015; yan et al., 2015) .

2-acetyl-1-pyrroline (2-AP) is recognized as one of the main volatile substances out of 200 kinds of volatile substances present in rice out of which only (2-AP) is responsible for aroma in aromatic rice (Mahattanatawee and Rouseff, 2014; He and Park, 2015), there are number of ways to detect aroma in rice two of the main methods include KOH method and chewing method , used by traditional rice breeders this methods rely on human sense to determine the fragrance(Bradbury et al., 2005b; Liu et al, 2014; Yan et al, 2015) , which is not always accurate and creates difficulties for rice breeders. The genetic basis of aroma in aromatic rice is complex , researchers after many studies have concluded that the fragrance in aromatic rice is controlled by the gene named fgr gene present on the 8<sup>th</sup> chromosome of rice genome , fgr gene has been isolated and cloned at present (Bradbury et al., 2005a; Chen et al., 2006; Chen et al., 2008),this fgr gene encodes for betaine aldehyde dehydrogenase (Badh2) enzyme it catalyzes the oxidation of amino aldehydes to carboxylic acid and an inactive Badh2 also promotes fragrance in aromatic rice by inhibiting the expression of fgr gene or any mutation in fgr gene , which causes increase in 2-AP precursor substance which leads to accumulation of 2-AP precursor substance and enhances the fragrance in aromatic rice grain (Niu et al., 2008; Shan et al., 2013).

## 2. Biosynthesis of 2AP Precursor Substance

There are several volatile and semi volatile compounds present in rice which are associated with unique fragrance, flavor and strength of aroma in fragrant rice. Over 300 volatile compounds are identified in rice varieties (Widjaja et al.1996) out of them 2-acetyl-1-pyrroline (2AP) precursor substance having popcorn-like aroma is reported the most important flavor compound having low odor threshold (Buttery et al., 1982) and a pleasant aroma. This popcorn-like aroma coming from basmati and jasmine rice varieties is the unique trait that attracts many consumers (Bergman et al., 2000) which is primarily stemming from its 2AP content.

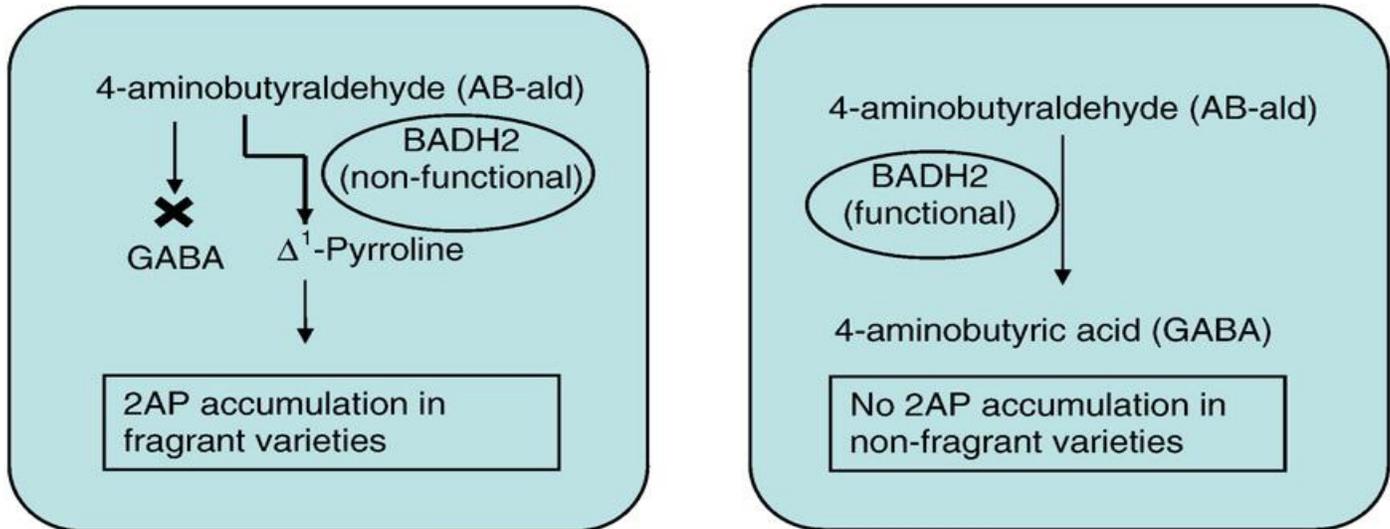
2 AP was observed to be present in all parts of rice plant, except for the roots (Sood and Siddiq, 1978), and also its concentration is 100 times lower, in non-fragrant rice varieties (reviewed by Grosch and Schieberle, 1997). The biochemical pathway for synthesis of 2AP compound is presently unknown (Niu et al., 2008; Fitzgerald et al., 2009) although many studies have revealed L-proline as a precursor of aroma in rice, Vanavichit et al. (2005) elucidated the biosynthetic pathways for 2AP suggested that 2AP is combined through the polyamine pathway. The primary precursor of 2AP was discovered to be 1-pyrroline (1P) which is formed from 4-aminobutyraldehyde (AB-ald; the direct precursor of 4-aminobutyric acid (GABA). With the discovery of gene for aroma, a more convincing proof for 2AP biosynthetic pathway was explained as of late (Fig. 1). As indicated by Chen et al. (2008), 4-aminobutyraldehyde (Abdominal muscle ald) is known to be kept up in an equimolar proportion with a direct 2AP precursor,  $\Delta^1$ -pyrroline, and the AB-ald levels give off an impression of being a significant factor controlling the pace of 2AP biosynthesis. They recommended that the functional badh2 catalyst (coded by the aroma gene *Fgr*) represses 2AP biosynthesis in non fragrant rice by changing over AB-ald, an assumed 2AP antecedent, to GABA while the non-functional badh2 (coded by *fgr*) bring about AB-ald aggregation leads to the development of 2AP in fragrant rice.

Bradbury et al. (2008) likewise proposed that  $\gamma$ -aminobutyraldehyde (*gabald*) is a compelling substrate for badh2 and that accumulation furthermore, unconstrained cyclisation of *gabald* to form  $\delta^1$ -pyrroline because of a non-useful badh2 catalyst as the conceivable reason for 2ap accumulation in rice.

In another examination, increased expression of  $\delta^1$ - pyrroline-5-carboxylate synthetase in fragrant varieties thought about with non-fragrant varieties, just as corresponding raised concentrations of its product, Prompted the end that  $\delta^1$ -pyrroline-5- carboxylate, generally the quick precursor of proline combined

from glutamate, reacts directly with methylglyoxal to form 2AP. (huang et al., 2008), with no direct role proposed for badh2.

**(a) BADH2-dependant 2AP synthesis**



**(b) BADH2-independant 2AP synthesis**

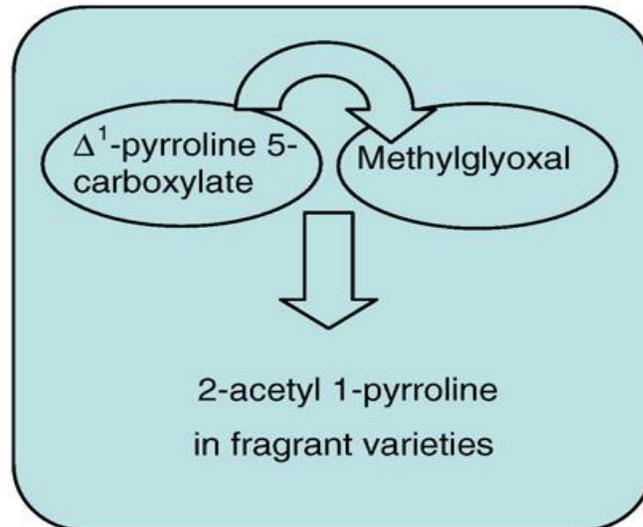


Figure 1 Pathway of 2AP biosynthesis in rice. (a) BADH2-dependant 2AP synthesis (Chen et al. (2008); Bradbury et al. (2008)), (b) BADH2-independant 2AP synthesis (Huang et al.,2008).

### 3. Genetic Basis of Rice Aroma Gene

Aroma is a very unique trait for aromatic rice varieties which is favored by many consumers worldwide (Kovach et al., 2009; Shan et al., 2015). The genetic basis of aroma in rice was discovered in 1992 by researchers and they found that a recessive gene that controls fragrance was present on chromosome 8 in aromatic rice variety. In 1992 researchers also discovered that the genetic distance between gene and molecular marker RG28 was 4.5 cm (Ahn et al., 1992).

Since then, many more research teams have detected the recessive gene controlling the rice fragrance by using different genetic populations and since then, a number of research teams also determined the gene controlling rice fragrance by using different genetic population and different types of molecular markers near the RG28 (Jin et al., 1995, 1996; Lorieux et al., 1996; Dong et al., 2001a, 2001b). Zhang Tao et al. (2008) used aromatic rice and *indica* non-fragrant rice in experiment, and determined the aroma gene in the region of approximately 252 kb on chromosome 8. Similarly, Wanchana et al. (2005) also determined the aroma gene *fgr* between the molecular marker RM223 and RM342, and concluded that the difference between aromatic and non-aromatic rice is due to the molecular markers present on the gene encoding betaine dehydrogenase.

A significant difference is present in the sequence of *Badh2* gene between the aromatic rice and non-aromatic rice after sequencing the *fgr* region, a mutation is present in the 7<sup>th</sup> exon region of *badh* gene, which leads to loss of function of *badh2* protein. A *badh2* gene. Therefore, *Badh2* gene is most likely linked to *fgr* gene, which control the rice fragrance (Bradbury et al., 2005a).

The complete length of *badh2* gene is 1509 bp, which includes 15 exons and 14 introns, which encodes further 503 amino acids (Chen et al., 2008; Shan et al., 2015). When aromatic rice and non-aromatic rice varieties are compared various different mutations were observed in *badh2* gene among different rice varieties. In aromatic rice varieties, 8 bp deletion and 3 single nucleotide polymorphism sites (SNPs) in the 7<sup>th</sup> exon of *badh2* gene (Bradbury et al., 2005a) are common in all aromatic rice varieties. In different varieties of aromatic rice, there is a 7 bp deletion in exon 2 of the *Badh2* gene and a 803 bp deletion between exon 4 and exon 5 (Shi et al., 2008; Kovach et al., 2009; Shao et al., 2011), variations are present in exons site 1, 10, 13 and 14 (Shi et al., 2008; Amarawathi et al., 2008; Kovach et al., 2009). There are different mutation and splice sites are present on the first exons and the first introns of the *badh2* gene, the promoter region and the 5' UTR region also include insertion, deletion, or single nucleotide mutation sites (Shao et al., 2011, 2013; Shi et al., 2014; Ootsuka et al.,

2014). The different aroma in different rice varieties is due to the allelic variation of badh2 gene . hence , badh2 gene gets mutated in coding region or regulatory region , which produces betaine dehydrogenase which performs no biological activity , but is responsible for fragrance in rice .

To further analyze the genetic basis of aroma gene, the whole genome analysis was performed and results concluded that there are 2 quantitative trait loci (QTL) which were also responsible to control the fragrance in rice grain with badh2 gene present on eighth chromosome. The two loci were present on the third and fourth chromosomes respectively of the rice grain (Amarawathi et al., 2008). The badh1 gene present on chromosome 4 as located by QTL could be responsible for controlling aroma in rice grains , as badh1 and badh2 belongs to the homologous gene , badh1 gene in rice is also highly homologous to badh1gene in sorghum and the barley genome (Bradbury et al.2005a, 2008). Recent studies have found that badh1gene has a positive role in presenting the salt tolerance at the germination stage (He et al., 2015). The mutation in badh1gene is also linked with the aroma in rice grain , but the effect is much less than the badh2 gene (Amarawathi et al., 2008). Hence , relation between aroma and badh1gene need further study.

#### **4. Perspective**

The desirability of aroma has brought about strong human inclination and choice for this quality during development and taming. Despite the fact that sensory examination has distinguished over a dozen distinct smells and flavors in rice (Champagne, 2008), an interesting aroma explicit to specific varieties may be expected to the differential mix of at least one significant mixes of one or more major compounds in definite proportions. Though, there is almost no connections between various unpredictable different compound combinations and the smell/flavor. The altered aroma or the unique smell in relation to the popcorn-like (or on the other hand 2AP-related) smell may be because of (i) the presence of QTLs for significant mixes in various blends and their connection (ii) the impact of modifier qualities, (iii) the diverse hereditary foundation, and their cooperation with the climate in creating aroma. As badh2 appears to clarify just the 2AP accumulation in most (yet not every) fragrant variety, and as there is an absence of data with respect to the hereditary relationship for different other flavor/off-flavor compounds, it would be profoundly imperative to portray the coordinated connection between these compounds and fundamental hereditary impact . Further, identification and portrayal of other gene(s) in aroma advancement and their encoding gene product(s) in uncommon varieties , hereditary complementation experiments coupled with the investigation of the biochemical responses catalyzed by those

proteins may give further bits of knowledge into the atomic premise of aroma in rice.

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