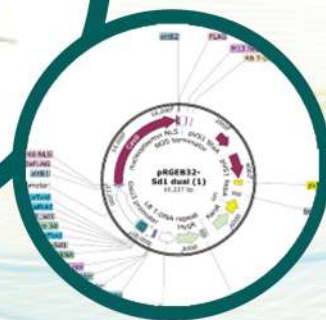
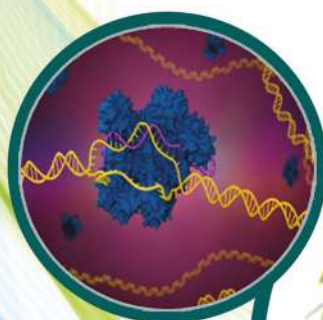


Catalogue of Genome Editing Vectors for Rice Breeding: CRRRI

S Samantaray, MJ Baig, KA Molla, Parameswaran C,
Devanna BN, AK Nayak



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Catalogue of Genome Editing Vectors for Rice Breeding: CRRI

**S Samantaray, MJ Baig, KA Molla, Parameswaran C,
Devanna BN, AK Nayak**

A comprehensive catalog of 30 genome editing vectors constructed at ICAR-CRRI, designed to address multiple challenges in rice cultivation. These vectors target a wide range of traits, including nitrogen use efficiency, lodging resistance, yield enhancement, abiotic and biotic stress tolerance, grain number increase, and more.



ICAR-Central Rice Research Institute
Cuttack 753 006, Odisha





Citation

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Preface

Rice, being the staple food for more than half of the global population, demands continuous improvement to sustain productivity and resilience against various biotic and abiotic challenges, especially in a changing climate. In this regard, genome editing has emerged as a revolutionary tool, enabling precise modifications in plant genomes to enhance key agronomic traits. The ICAR-Central Rice Research Institute (ICAR-CRRI) has been at the forefront of leveraging this cutting-edge technology to develop improved rice varieties with superior traits tailored to the needs of farmers and consumers.

This technical bulletin presents a comprehensive catalog of 30 genome editing vectors constructed at ICAR-CRRI, designed to address multiple challenges in rice cultivation. These vectors target a wide range of traits, including nitrogen use efficiency, lodging resistance, yield enhancement, abiotic and biotic stress tolerance, grain number increase, and more. The vectors have been designed to introduce precise genetic modifications for trait improvement. The vectors were based on versatile cutting-edge technologies, including Cas9, Cas12a, base editors, and prime editors. Notable, it also includes our indigenous miniature TnpB vectors. Each vector has been developed with a clear scientific rationale and holds the potential to contribute significantly to future rice breeding programs.

The development of these genome editing vectors is a testament to ICAR-CRRI's commitment to harnessing modern biotechnological advancements for sustainable rice production. I extend my appreciation to the Head, Crop improvement division, and Head, Crop Physiology and biochemistry and dedicated team of scientists whose efforts have made this endeavor possible. This bulletin will serve as a valuable resource for researchers in rice improvement programs, fostering collaborations and advancements in genome editing applications.

I hope this technical bulletin will inspire further innovations and pave the way for the deployment of genome-edited rice varieties in India.

Dr. AK Nayak
Director
ICAR-CRRI

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Introduction

Genome editing in rice, primarily using tools like CRISPR-Cas9, has shown significant potential in improving rice varieties for various purposes, including increasing yield, enhancing disease resistance, improving nutritional quality, and adapting to environmental stress. **CRISPR-Cas** is a precise and targeted genome editing tool that allows scientists to alter specific genes within the rice genome. It works by introducing a break in the DNA at a particular location, where the cell's repair mechanisms either introduce mutations or can be directed to insert new sequences. For example, editing genes involved in rice's growth and development can lead to varieties with higher productivity, bacterial blight, rice blast, and other emerging diseases. Similarly, abiotic stress tolerance, nutritional quality, and herbicide tolerant varieties can also be developed using the genome editing technology.

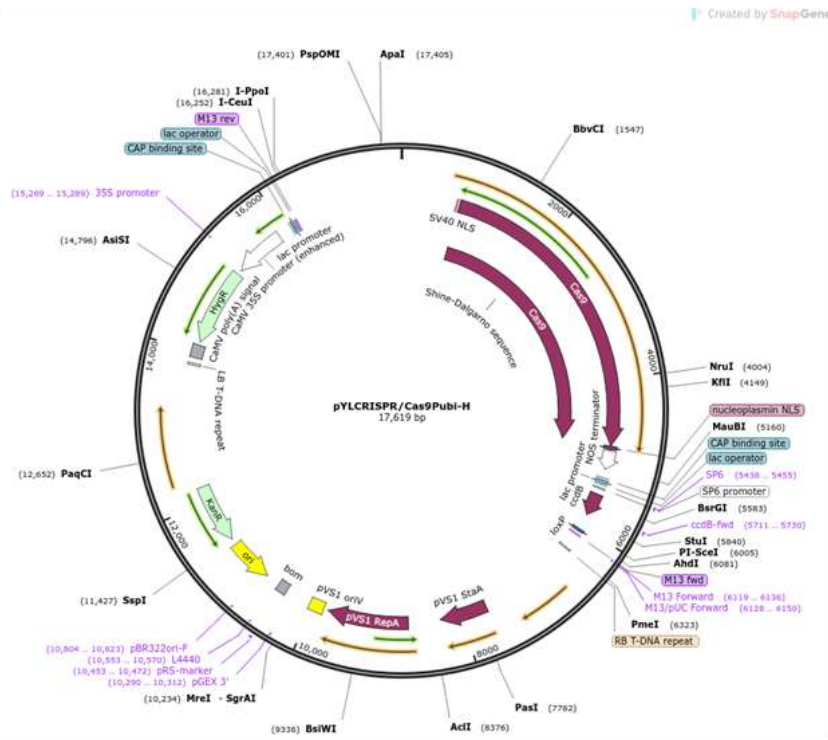
Genome editing work at CRRI

ICAR-CRRI, Cuttack initiated the genome editing work four years back with the objective of development of superior cultivars suited to the needs of our nation. The efforts of our institute resulted in the development of genome edited lines for *Ideal Plant Architecture 1* gene in Swarna variety which showed 20-30% increase in the number of spikelets per panicle. Recently, IBSC-NRRI has cleared the Swarna-*IPA1* for field evaluation as per the SoPs developed by DBT, Government of India. Besides, T₂ plants have been developed for two-line breeding system targeting *TMS5* gene in MTU1010 and Lalat, plant height reduction (*Sd1*) gene in Gobindobhog and Nuakalajeera varieties. Gn1a edited lines of Swarna and Nuakalajeera are also developed through using genome editing technology. The work is also in progress for the enhancement of nitrogen use efficiency through editing of *GRF4* gene in rice. Further, genotype independent transformation protocol has been established in five different cultivars and same protocol is now being evaluated in 20 different cultivars including wild rice for transformation efficiency. Apart from gene knockouts, efforts are in progress in vector engineering for genome editing, especially on the development of *SpCas9* orthologs, TnpB editing system, efficient base editing and also the development of novel fusion protein with nCas9 for prime editing. We are now validating the editing efficiency of those novel vectors. Besides, at least 21 genes has been targeted for different traits using knock out, multiplex editing, base and prime editing strategies in our institute. Moreover, five projects are currently continuing in our institute related to genome editing and one project has been approved by technical committee of DBT for funding. ICAR-CRRI, Cuttack has established expertise, infrastructure, and manpower for execution of genome editing work in accordance with the rapid development of the genome editing technology and to the needs of our nation.

Vector 1

Title of the work:	Editing of miR156 bonding site in IPA1 gene for yield improvement
Target gene: Name with full form	<i>IPA1</i> (Ideal Plant Architecture 1)
Target variety:	Swarna
Brief Description: Hypothesis, expected outcome	<i>IPA1</i> gene regulates the number of panicle branches and yield. This gene is negatively regulated by miR156. The editing of binding site would result in increased panicle branches and yield
Vector details: Background and guide details	pYLCRISPR-Cas9Pubi-H; guide RNA expressed by U6a promoter.

Map:

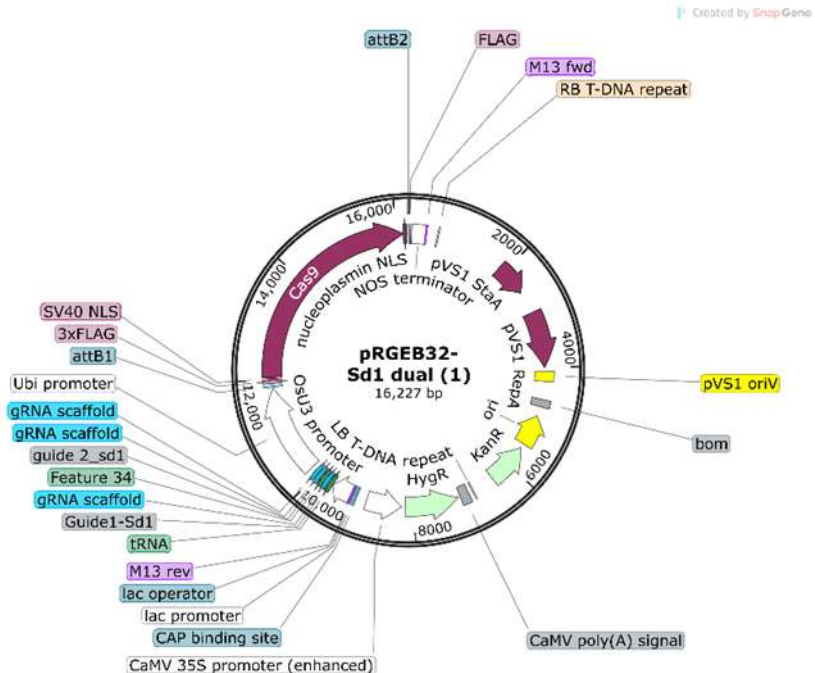


Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray
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Vector 2

Title of the work:	Development of semidwarf rice lines through CRISPR-Cas9 mediated genome editing
Target gene: Name with full form	Semidwarf-1 (<i>SD1</i>)
Target variety:	Nuakalajeera and Gobhindobhog (<i>Oryza sativa</i> . indica cultivar)
Brief Description: Hypothesis, expected outcome	Genome editing is a rapidly advancing tool for crop improvement, but challenges include recalcitrant varieties and limited CRISPR validation protocols. We hypothesize that a standardized in vitro regeneration protocol will enhance edited plant recovery, while protoplast-based guide RNA validation will confirm mutagenesis efficiency. Editing <i>OsSD1</i> in Nuakalajeera rice may produce semi-dwarf, lodging-resistant lines without yield loss.
Vector details: Background and guide details	pRGEB32 Two guides targeting exon 1 and exon 2

Map:

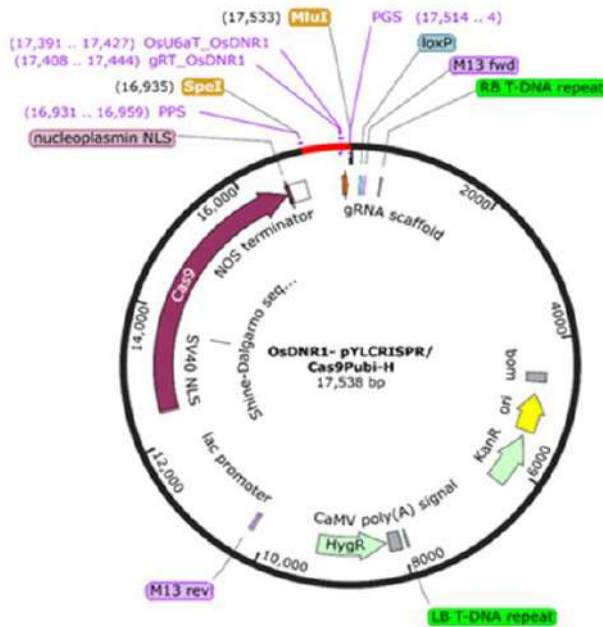


Name of the scientist(s)	Dr. KA Molla and Dr. MJ Baig
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Vector 3

Title of the work:	Editing of <i>Dull nitrogen Response 1 (DNRI)</i> nitrogen use efficiency.
Target gene: Name with full form	Dull nitrogen Response 1 (<i>DNRI</i>)
Target variety:	Swarna
Brief Description: Hypothesis, expected outcome	<i>DNRI</i> gene is involved in the regulation of auxin homeostasis. Modulation of the levels of auxin in response to the availability of nitrogen affects root architecture and growth, hence affecting the ability of the plant to absorb nutrients. Reduced expression was associated with increased rates of nitrogen uptake; this implies that lower expression of <i>DNRI</i> may enhance the ability of the plant to use available nitrogen more effectively.
Vector details: Background and guide details	pYLCRISPR-Cas9Pubi-H; guide RNA expressed by U6a promoter.

Map:

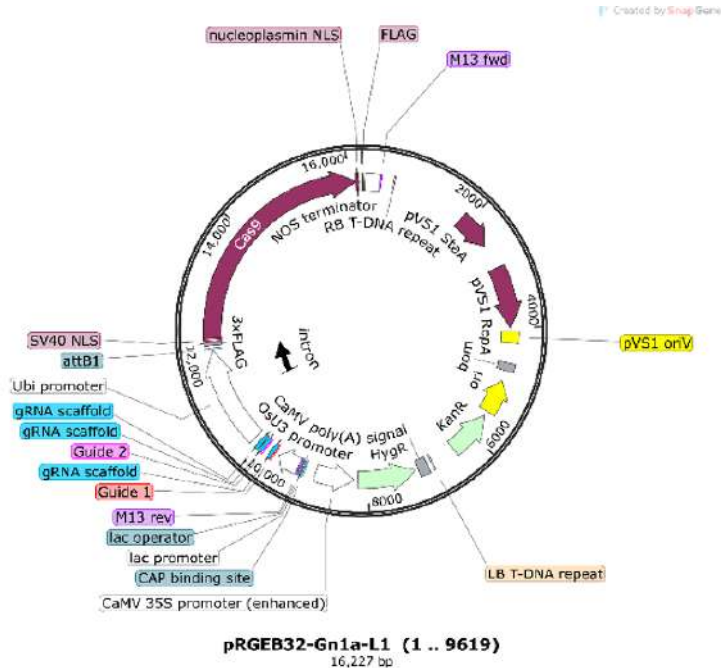


Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray
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Vector 4

Title of the work:	Development of <i>Gn1a</i> mutant rice to enhance tiller number and yield using CRISPR-Cas9-mediated genome editing.
Target gene: Name with full form	<i>Gn1a</i> (Cytokinin oxidase/dehydrogenase 2)
Target variety:	Nuakalajeera (<i>Oryza sativa</i> . indica cultivar)
Brief Description: Hypothesis, expected outcome	Genome editing is a powerful tool for crop improvement, but challenges such as low regeneration efficiency and the need for precise validation of edits limit its effectiveness. We hypothesize that optimizing an in vitro regeneration system will enhance the recovery of edited plants. Targeted editing of <i>Gn1a</i> in rice has the potential to increase tiller number, leading to improved yield without negatively impacting plant architecture.
Vector details: Background and guide details	pRGEB32 Two guides targeting exon 3

Map:

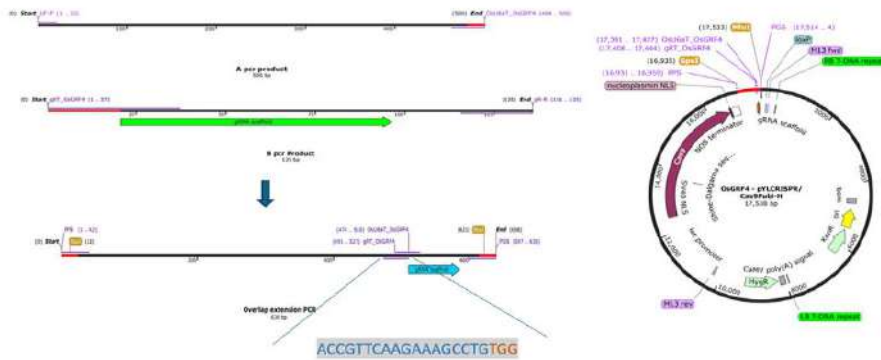


Name of the scientist(s)	Dr. KA Molla
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Vector 5

Title of the work:	Editing of <i>miR396</i> bonding site in <i>GRF4</i> gene for yield nitrogen use efficiency.
Target gene: Name with full form	Growth Regulation Factor 4 (<i>GRF4</i>)
Target variety:	Swarna
Brief Description: Hypothesis, expected outcome	The <i>GRF4-GIF-miR396</i> module plays a crucial role in regulating grain size and overall yield. This module is characterized by the interaction between microRNA396 (<i>miR396</i>), growth-regulating factors (<i>GRFs</i>), <i>miR396</i> is known to post-transcriptionally repress GRF expression (negative regulator), thereby modulating their activity in developmental pathways. Disruption of <i>miR396</i> binding sites in <i>GRF4</i> can lead to enhanced nitrogen uptake, grain size and yield. Hence, enhance overall plant performance.
Vector details: Background and guide details	pYLCRISPR-Cas9Pubi-H; guide RNA expressed by U6a promoter.

Map:

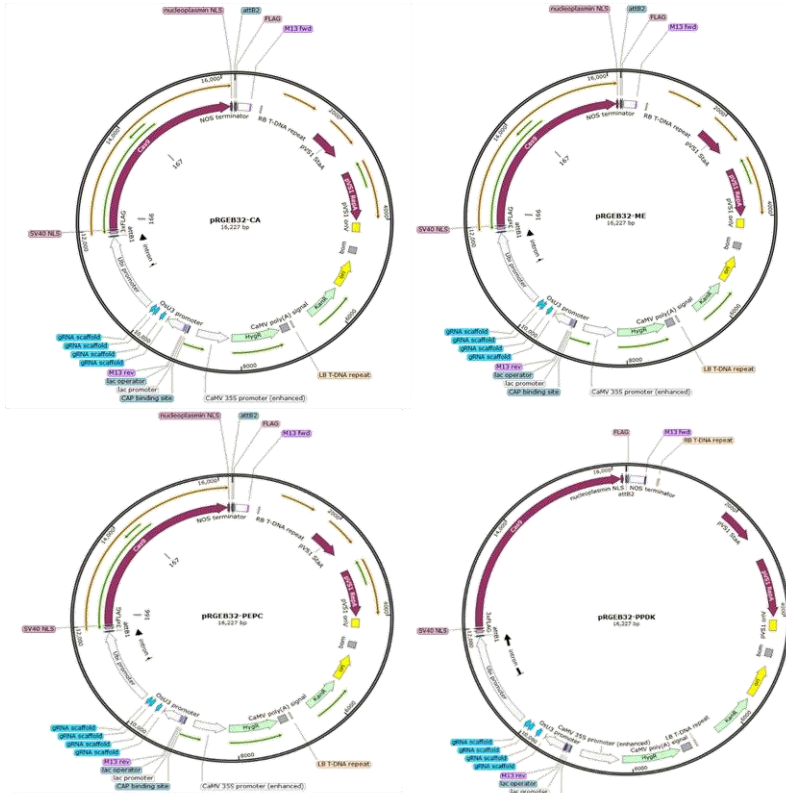


Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray
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Vector 6

Title of the work:	Deciphering the role of C4 photosynthetic pathway genes through CRISPR mediated knock-out
Target gene: Name with full form	<i>OsCA</i> (Carbonic Anhydrase), <i>OsPEPC</i> (Phospho Enol Pyruvate Carboxylase), <i>OsME</i> (Malic Enzyme), <i>OsPPDK</i> (Pyruvate orthophosphate dikinase)
Target variety:	CR DHAN 317, Kitaake
Brief Description: Hypothesis, expected outcome	This study focuses to identify the role of endogenous <i>PEPC</i> , <i>CA</i> , <i>PPDK</i> and <i>ME</i> in rice by CRISPR mediated knockout of these genes.
Vector details: Background and guide details	Two guides were selected to target each gene. The final vector backbone is pRGEB32

Map:

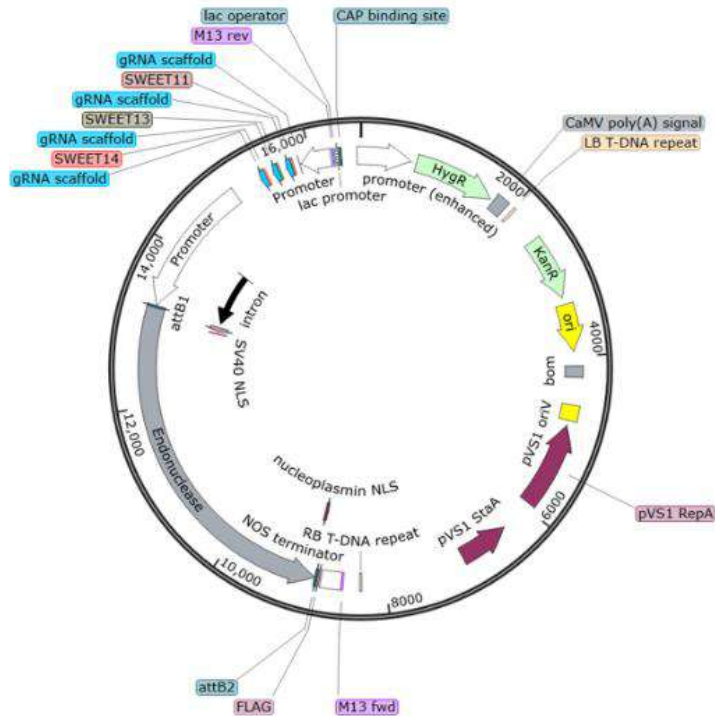


Name of the scientist(s)	Dr. M J Baig and Dr. K A Molla
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Vector 7

Title of the work:	Developing Sheath blight and Bacterial blight resistance in rice
Target gene: Name with full form	<i>OsSWEET11/13/14</i> (Sugars Will Eventually be Exported) or any one of them
Target variety:	Naveen, MTU1010, CR DHAN 807, CR DHAN 805
Brief Description: Hypothesis, expected outcome	The hypothesis is to inhibit the binding of <i>Xoo</i> TALEs and ultimately reduce <i>Xoo</i> pathogenicity by altering the EBE sequences of all three genes or any one gene using Cas9 and Cas12a.
Vector details: Background and guide details	One guide was selected to target each gene. The final vector is pRGEB32

Map:

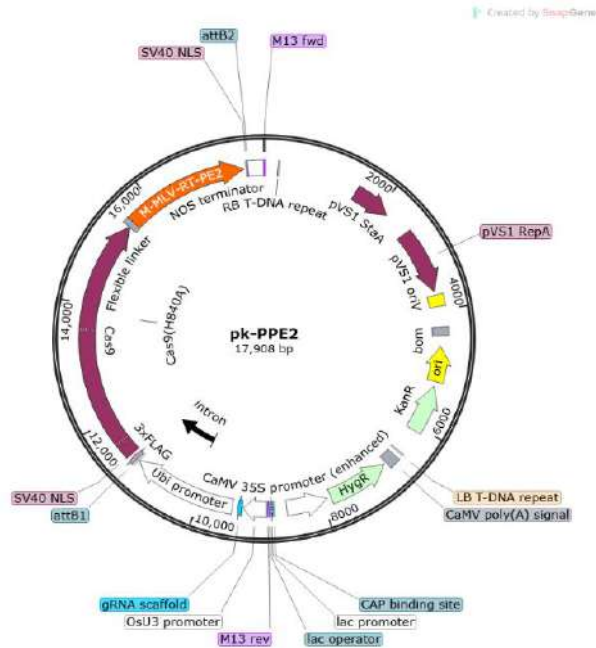


Name of the scientist(s)	Dr. M J Baig and Dr. K A Molla
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Vector 8

Title of the work:	Increasing the catalytic efficiency of <i>OsPEPC</i> for photosynthesis
Target gene: Name with full form	<i>OsPEPC</i> (Phosphoenol pyruvate carboxylase)
Target variety:	Kitaake (Japonica)
Brief Description: Hypothesis, expected outcome	Rice utilizes the C3 photosynthetic pathway, which is less efficient under elevated temperatures and water-limited conditions. Attempts to introduce the C4 photosynthetic pathway into C3 rice have met with limited success. In this study we are trying to edit the endogenous PEPC gene in rice via prime editing to achieve precise genomic modifications at two specific loci to convert its function from C3-like to C4-like that could improve enzyme functionality.
Vector details: Background and guide details	pK-PE2, PEPC prime editing guide RNA (pegRNA-Protospacer, scaffold, RT template and PBS)

Map:

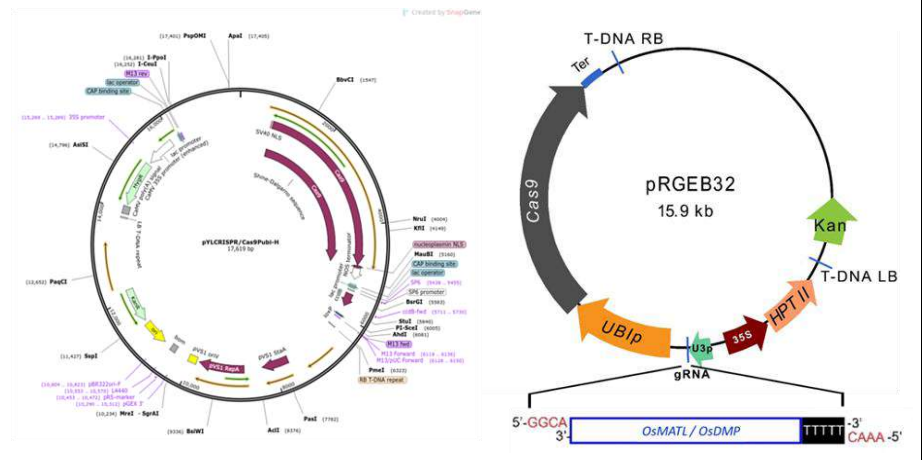


Name of the scientist(s)	Dr. M J Baig and Dr. K A Molla
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Vector 9

Title of the work:	Development of Haploid Inducer rice lines using CRISPR Cas9 gene editing system for high induction frequency
Target gene: Name with full form	<i>CENH3</i> (Centromere specific histone H3) <i>DMP</i> (Domain Membrane Protein) <i>MATL</i> (Matrilineal gene)
Target variety:	Kasalath, Japonica (Ac 41023), Manipuri Black Rice
Brief Description: Hypothesis, expected outcome	<i>MATL</i> , is Phospholipase specific to sperm cell cytoplasm and any disruption causes the inactivation of sperm cell. <i>DMP</i> is a membrane protein associated with the pollen wall. It helps in maturation of pollen wall. <i>CENH3</i> , controls the formation of kinetochore for spindle fibre attachment to chromosome during cell division. Any alteration to these genes may result in the inactivation of pollen cells and cause haploid induction.
Vector details: Background and guide details	pGEB32; guide RNA expressed by U3. pYLCRISPR-Cas9Pubi-H; guide RNA expressed by U6a, U6b and U3a promoter.

Map:

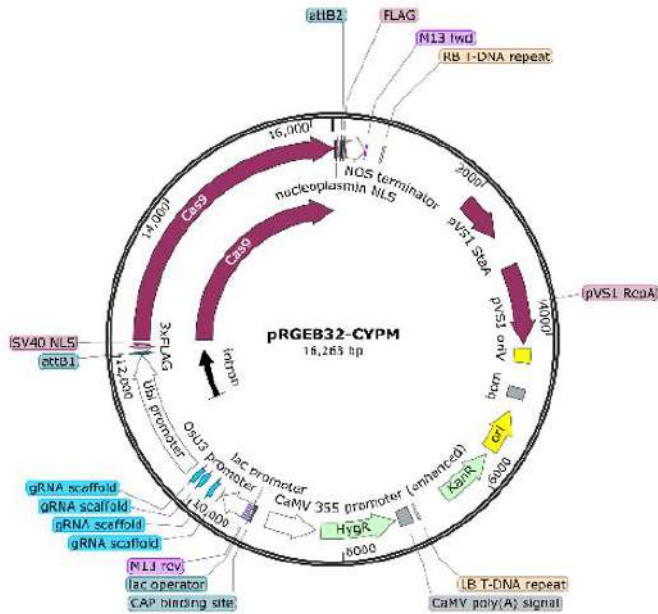


Name of the scientist(s)	Dr. Sanghamitra Samantaray
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Vector 10

Title of the work:	Increasing Apigenin secretion by rice roots to promote diazotroph population
Target gene: Name with full form	CYP75 B3/B4 (Cytochrome P450 75B3/B4)
Target variety:	Naveen
Brief Description: Hypothesis, expected outcome	Apigenin is a rice root secreted flavonoid that is likely to promote diazotroph population, in turn promoting biological nitrogen fixation. CYP75B3/B4 are involved in apigenin degradation, so knocking out CYP75B3/B4 can promote apigenin concentration in root exudates.
Vector details: Background and guide details	Vector: pRGEB32 Guide: 2 guides, both targeting the genes CYP75B3/B4

Map:



Name of the scientist(s)	Dr. K A Molla
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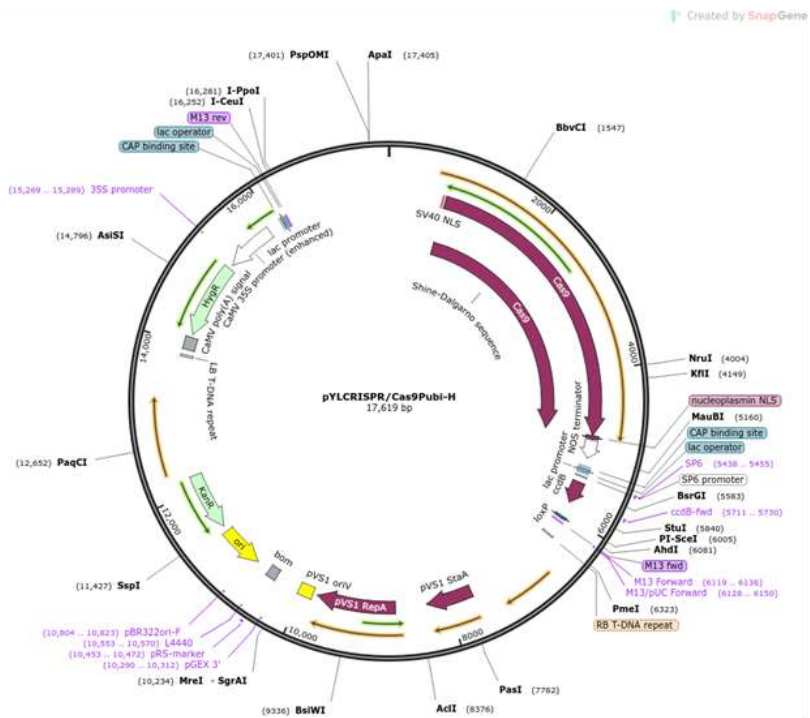
Vector 11

Title of the work:	Enhancing yield by altering <i>OsKRN2</i> gene in rice
Target gene: Name with full form	<i>OsKRN2</i> (Karnel row number 2)
Target variety:	Nuakalajeera
Brief Description: Hypothesis, expected outcome	The convergent selection of <i>KRN2</i> / <i>OsKRN2</i> in maize and rice has played a significant role in increasing grain number, and its complete loss-of-function can enhance grain yield without negatively impacting other agronomic traits. So, we targeted <i>OsKRN2</i> using CRISPR/Cas9 aiming that it would increase the grain number in the targeted variety.
Vector details: Background and guide details	Two guides were selected to target <i>OsKRN2</i> . The final vector backbone is pRGEB32
<p>Map:</p>	
Name of the scientist(s)	Dr. K A Molla and Dr. M J Baig

Vector 12

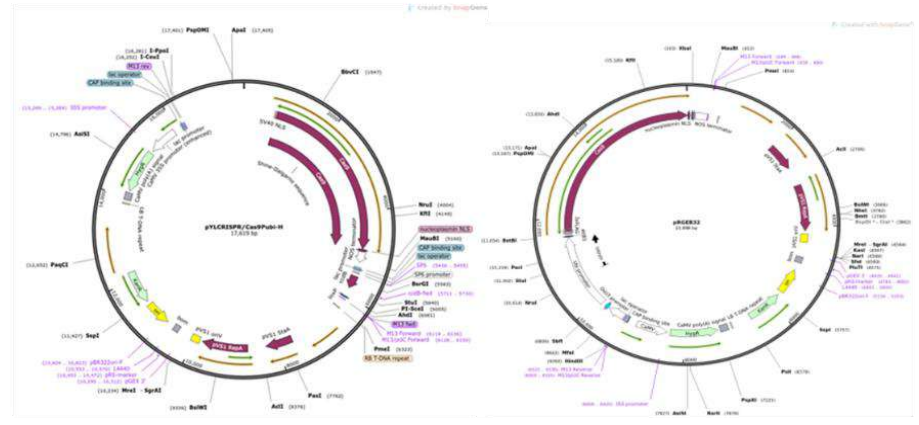
Title of the work:	Editing the Dense Erect Panicle 1 gene for enhancing Nitrogen Use Efficiency (NUE).
Target gene: Name with full form	<i>DEP1</i> (Dense Erect Panicle1)
Target variety:	Swarna
Brief Description: Hypothesis, expected outcome	<i>DEP1</i> knock down will reduce panicle length and enhance seed number in panicle in addition to Nitrogen uptake.
Vector details: Background and guide details	pYLCRISPR-Cas9Pubi-H; 2 guide RNA expressed by U6a, U6b, U6c and U3 promoter.

Map: Source: (Vector map: <https://www.addgene.org/>)



Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray
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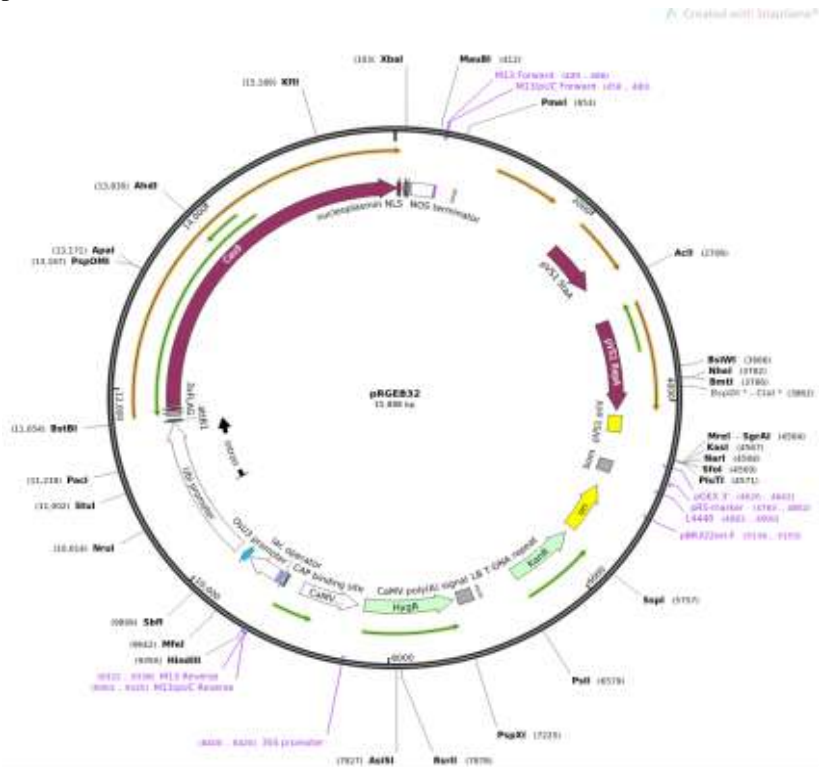
Vector 13

Title of the work:	Targeting serotonin and senescence pathways for enhancing brown plant hopper resistance and yield in rice (<i>Oryza sativa</i> L.) using genome editing approaches
Target gene: Name with full form	<i>CYP71A1</i> (Cytochrome p450 71a1), <i>OsNAP</i> (NAC-like activated by apetala3/pistillata)
Target variety:	IR64, MTU1010
Brief Description: Hypothesis, expected outcome	Knock out of <i>CYP71A1</i> and <i>OsNAP</i> gene enhances Bph tolerance, delay in senescence and increase yield in rice.
Vector details: Background and guide details	pYLCRISPR-Cas9Pubi-H; guide RNA expressed by U6a promoter. pRGEB32; guide RNA expressed by U3a promoter.
Map: 	
Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray

Vector 14

Title of the work:	Genome editing of <i>AAP3</i> and <i>ARF6</i> genes associated with tiller number and angle in <i>indica</i> rice for yield improvement
Target gene: Name with full form	<i>AAP3</i> (Amino acid permease), <i>ARF6</i> (Auxin response factor)
Target variety:	IR64, MTU1010
Brief Description: Hypothesis, expected outcome	Knock out of <i>AAP3</i> and <i>ARF6</i> will increase tiller number and decrease angle in rice.
Vector details: Background and guide details	pRGEB32; guide RNA expressed by U3a promoter.

Map:

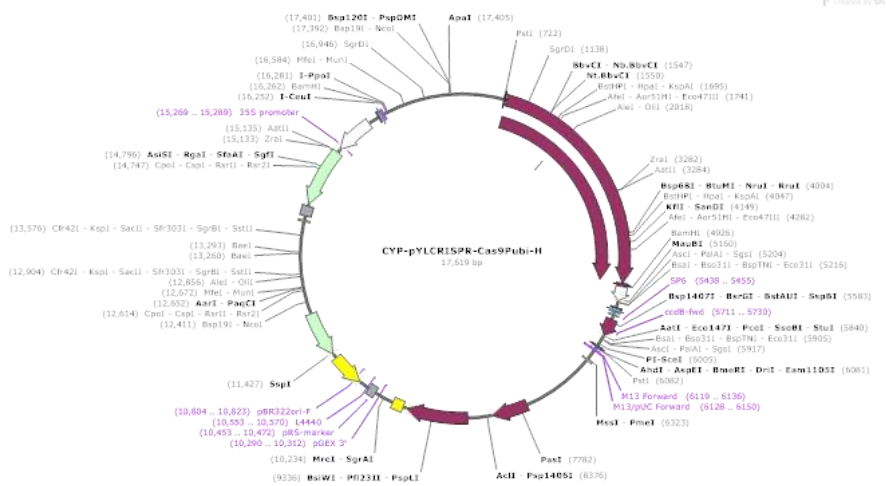


Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray
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Vector 15

Title of the work:	Deciphering and deploying low phosphorus tolerance and nitrogen use efficiency in rice using targeted genomics approach
Target gene: Name with full form	<i>CYP</i> (Cytochrome P450)
Target variety:	MTU1010
Brief Description: Hypothesis, expected outcome	<i>CYP</i> gene regulates the number of tiller number and yield. <i>CYP</i> genes are linked to nitrogen metabolism, affecting how plants utilize nitrogen for growth. CRISPR-mediated editing of <i>CYP</i> genes can enhance nitrate absorption by regulating lateral root growth while indirectly controlling phosphate transporter expression for improved phosphorus uptake. CRISPR-edited <i>CYP</i> genes will enhance NUE and PUE, improving nutrient uptake, optimizing growth, and increasing crop yield sustainably.
Vector details: Background and guide details	pYLCRISPR-Cas9Pubi-H; guide RNA expressed by U6a promoter.

Map:

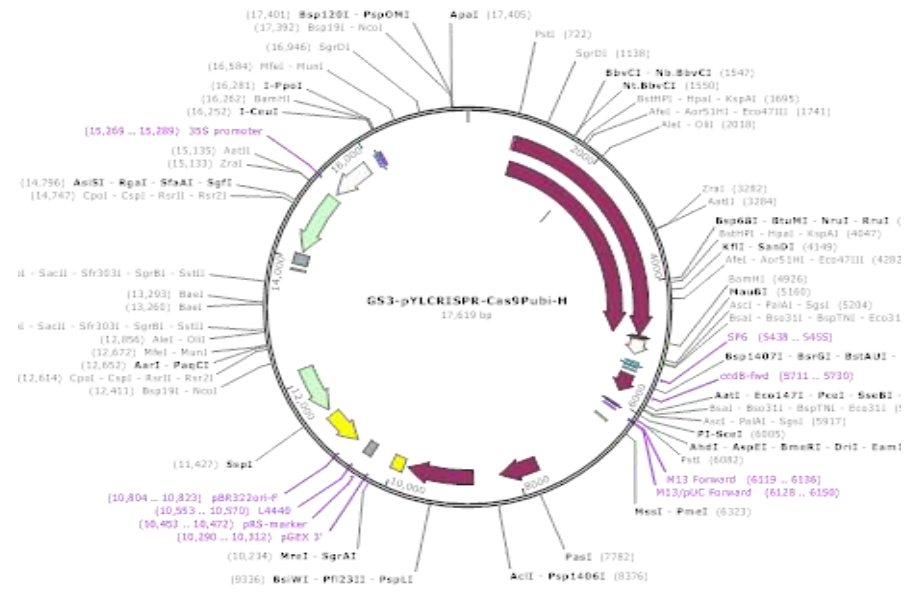


Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray
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Vector 16

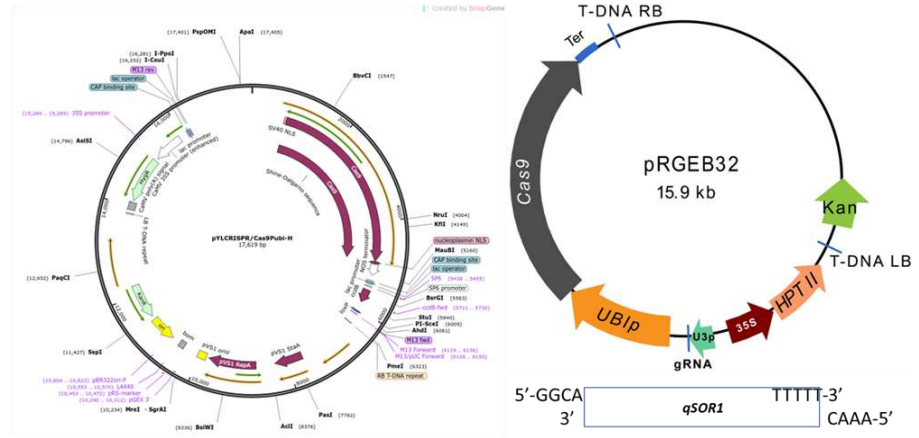
Title of the work:	Genome editing of Grain Size 3 (GS3) gene in popular indica varieties for enhancement of yield and brown plant hopper resistance in rice (<i>Oryza sativa</i> L.)
Target gene: Name with full form	GS3 (Grain Size 3)
Target variety:	Swarna
Brief Description: Hypothesis, expected outcome	GS3 encodes a protein with a G-protein gamma subunit-like domain, associated in the negative regulation of grain size. Loss- of-function of GS3 gene will enhance grain size and BPH tolerance through gene editing. This will contribute to the development of high-yield, pest-resistant crop varieties, ultimately supporting global food security and sustainable agriculture.
Vector details: Background and guide details	pYLCRISPR-Cas9Pubi-H; guide RNA expressed by U6a promoter.

Map:



Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray
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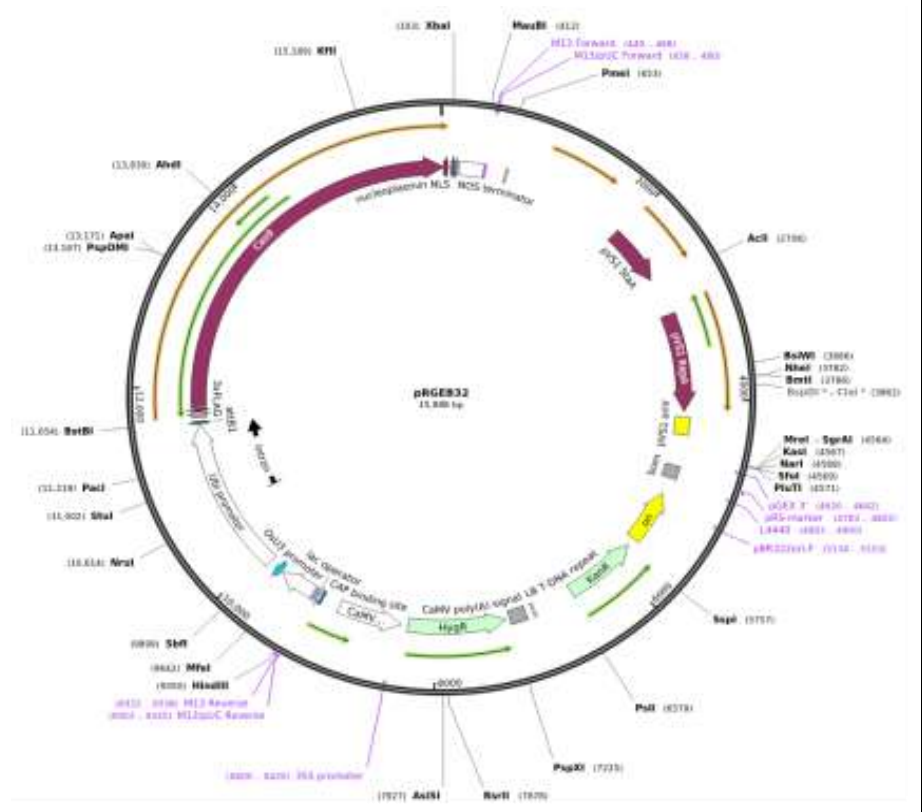
Vector 17

Title of the work:	Editing of <i>qSOR1</i> gene for reproductive stage salinity tolerance and yield improvement in rice.
Target gene: Name with full form	<i>qSOR1</i> (QTL for SOIL SURFACE ROOTING 1)
Target variety:	Luna Shankhi, Pooja
Brief Description: Hypothesis, expected outcome	Loss of function mutation in exon 3 of <i>qSOR1</i> gene has been observed to result in SOR (soil surface roots) in rice which helps the plants to avoid reducing stress of saline soil, resulting in increased yield compared to the parental plants without SOR. Editing of <i>qSOR1</i> gene would provide reproductive stage salinity tolerance and improved yield in saline condition.
Vector details: Background and guide details	pYLCRISPR-Cas9Pubi-H; guide RNA expressed by U6a promoter. pRGEB32; guide RNA expressed by U3 promoter.
Map: 	
Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray

Vector 18

Title of the work:	Genetic diversity and CRISPR based editing of <i>Trehalose-6-phosphate phosphatase 7 (OsTPP7)</i> gene in rice (<i>Oryza sativa</i> L.) for regulating germination under submergence
Target gene: Name with full form	<i>OsTPP7 (Trehalose-6-phosphate phosphatase 7)</i>
Target variety:	Bhalum 2
Brief Description: Hypothesis, expected outcome	Knockout of the <i>OsTPP7</i> gene will reduce germination and survival under submergence conditions in rice.
Vector details: Background and guide details	pRGEB32; guide RNA expressed by U3a promoter.

Map:

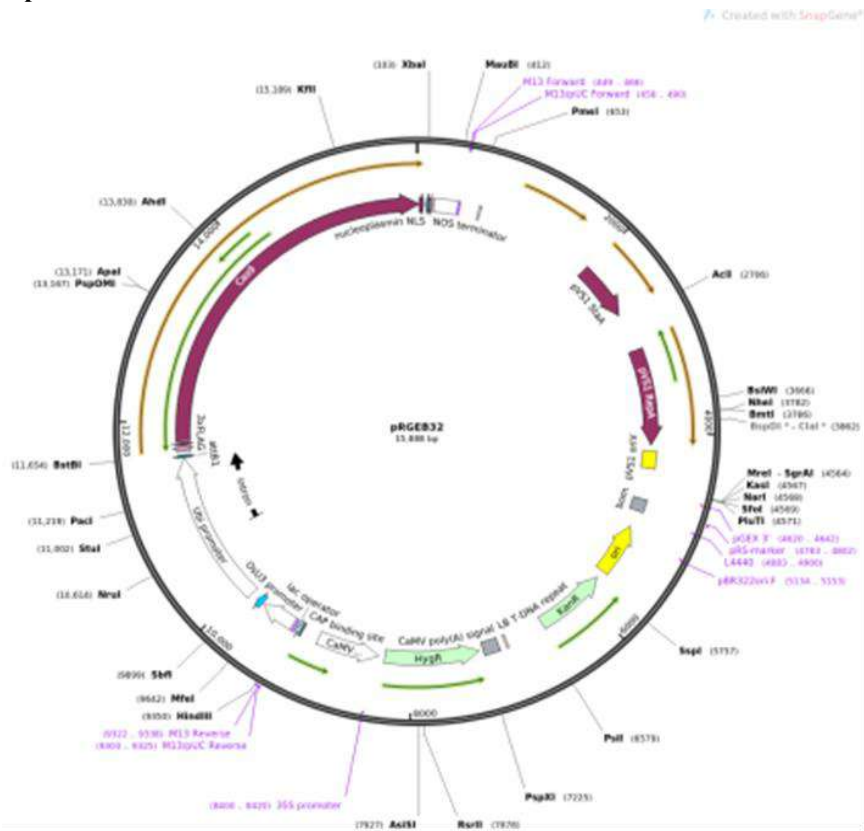


Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray
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Vector 19

Title of the work:	Breaking yield barriers in black rice using CRISPR-cas9 genome editing
Target gene: Name with full form	<i>SD1</i> (Semidwarf 1)
Target variety:	Manipuri black rice (chakhao)
Brief Description: Hypothesis, expected outcome	Knockout of <i>SD1</i> gene will reduce plant height, preventing loss of yield due to lodging in rice.
Vector details: Background and guide details	pRGEB32; guide RNA expressed by U3a promoter.

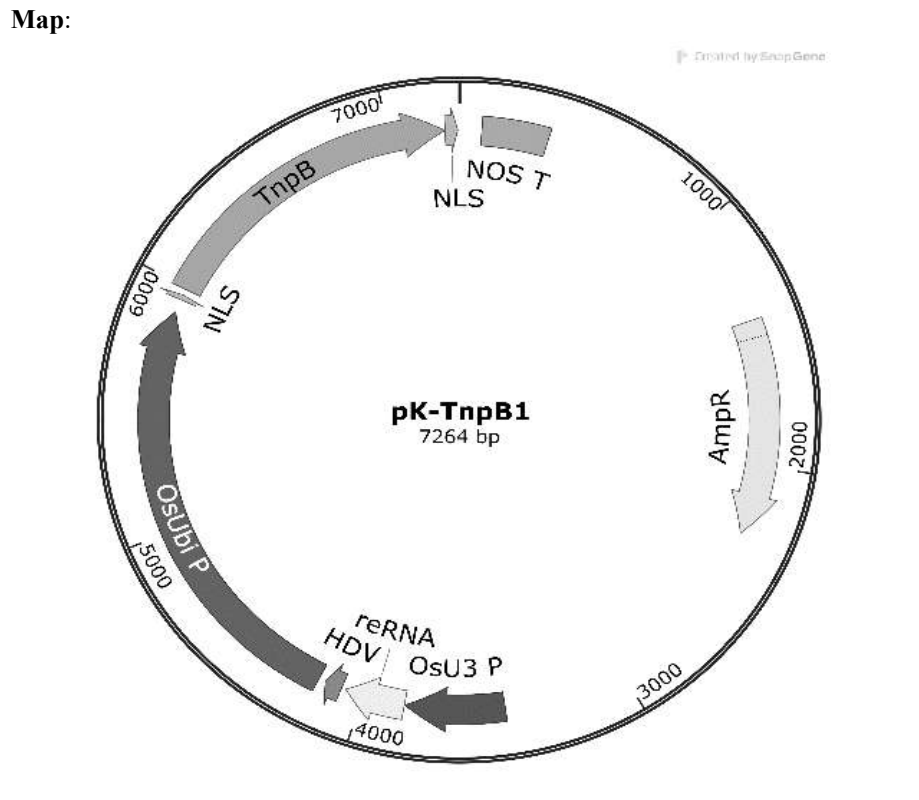
Map:



Name of the scientist(s)	Dr. Parameswaran C and Dr. Sanghamitra Samantaray
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Vector 20

Title of the work:	Developing Novel Genome editing vectors
Target gene: Name with full form	<i>HMBPP2</i> (4-hydroxy-3-methylbut-2-enyl diphosphate reductase), <i>Sla4G2</i> (Seedling-Lethal-Albino), <i>Pi21</i>
Target variety:	Naveen
Brief Description: Hypothesis, expected outcome	The vector is based on TnpB nuclease, which is way smaller than Cas9 an dCas12a. It could be used as an alternative to Cas9/Cas12a and facilitate easy delivery. Guide RNA is driven by Pol-III promoter.
Vector details: Background and guide details	pK-TnpB1

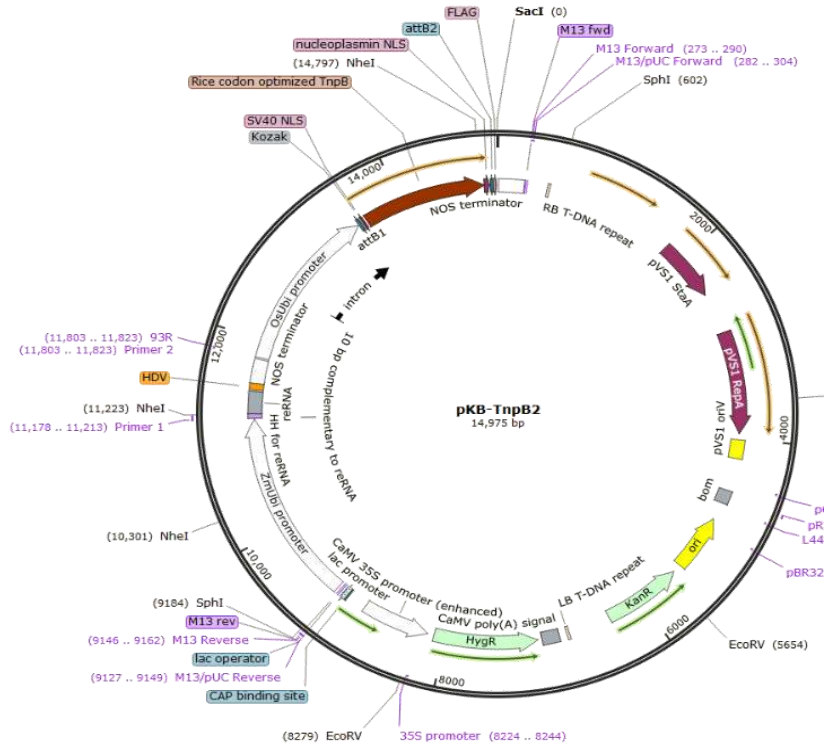


Name of the scientist(s)	Dr. K A Molla and Dr. M J Baig
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Vector 21

Title of the work:	Developing Novel Genome editing vectors
Target gene: Name with full form	<i>HMBPP2</i> (4-hydroxy-3-methylbut-2-enyl diphosphate reductase), <i>Sla4G2</i> (Seedling-Lethal-Albino), <i>CKX2</i> (cytokinin oxidase 2), <i>Waxy</i> , <i>SD1</i> (Semi Dwarf1)
Target variety:	Naveen
Brief Description: Hypothesis, expected outcome	The vector is based on TnpB nuclease, which is way smaller than Cas9 or dCas12a. It could be used as an alternative to Cas9/Cas12a and facilitate easy delivery. Guide RNA is driven by Pol-II promoter
Vector details: Background and guide details	pK-TnpB2

Map:

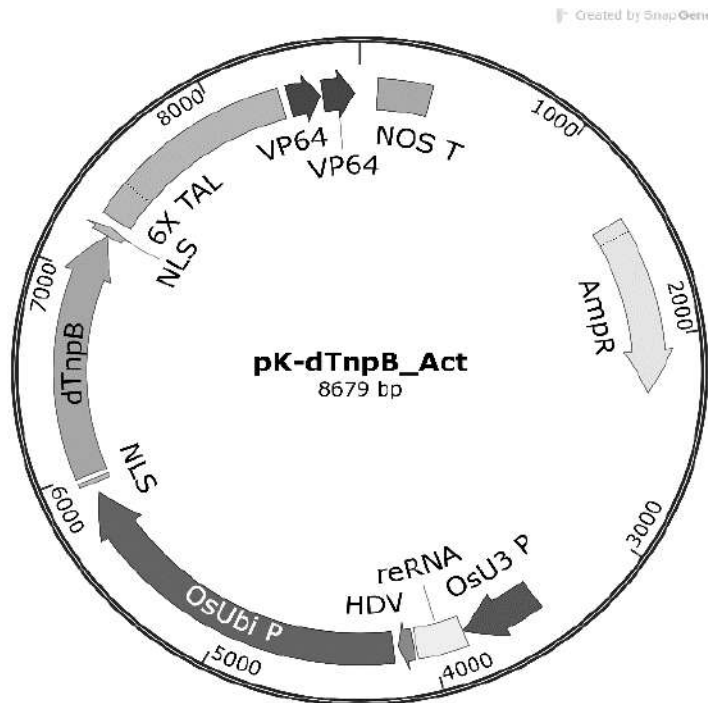


Name of the scientist(s)	Dr. K A Molla and Dr. M J Baig
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Vector 22

Title of the work:	Developing Novel Genome editing vectors
Target gene: Name with full form	<i>CHS</i> (chalconesynthase), <i>DSX</i> (1-deoxy-D-xylulose-5-phosphatesynthase), <i>PDS</i> (phytoene desaturase)
Target variety:	Naveen
Brief Description: Hypothesis, expected outcome	The vector is based on dead TnpB nuclease. It is useful for transcriptional activation of any gene.
Vector details: Background and guide details	pK-dTnpB-Act

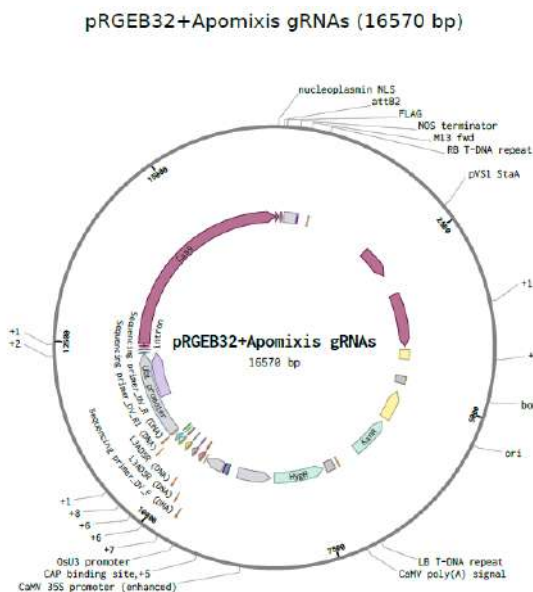
Map:



Name of the scientist(s)	Dr. K A Molla and Dr. M J Baig
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Title of the work:	Inducing quadruple mutations in key rice genes for development of apomictic hybrid rice lines
Target gene: Name with full form	<i>MTL: MATRILINEAL, OSD1: Omission of second division 1, PAIR1: Homologous pairing aberration in rice meiosis 1, REC8: RECombination 8</i>
Target variety:	Ajay and Rajalaxmi
Brief Description: Hypothesis, expected outcome	Mitosis instead of Meiosis (MiMe) is achieved by artificially inducing mutations in the meiotic genes REC8, PAIR1, and OSD1 to produce clonal diploid gametes, and mutating MTL to induce haploid seed formation. The quadruple mutant rice hybrid lines would have the potential of clonal propagation without losing the hybrid vigor.
Vector details: Background and guide details	The multiplex gRNA cloning vector was developed in the background of pRGE32, a binary vector for rice transformation having Kanamycin and Hygromycin markers for bacterial and plant selection, respectively. The fragment consisting of guide-RNAs for along with scaffold RNA was inserted at BsaI sites in pRGE32 plasmid.

Map:

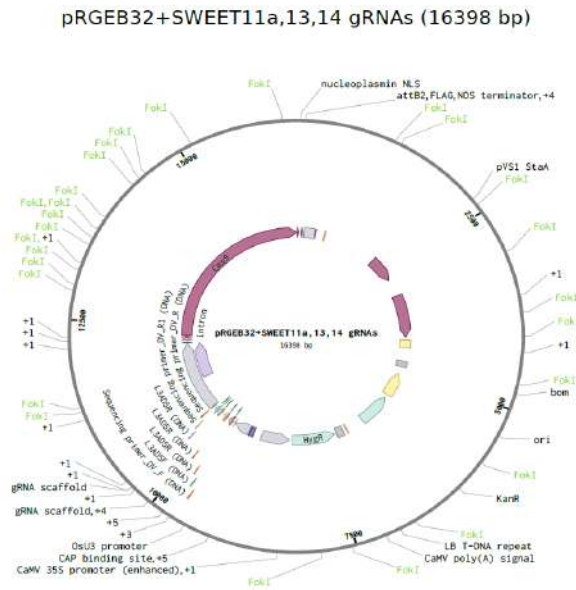


Name of the scientist(s)	Dr. Devanna, Parameswaran C, and Dr. Sanghamitra Samantaray
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Vector 24

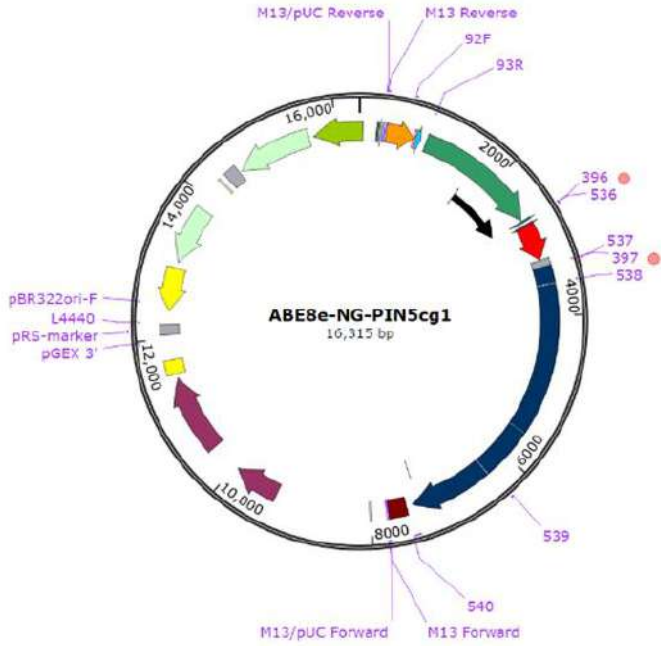
Title of the work:	Inducing triple mutations in key rice genes for imparting bacterial blight resistance
Target gene: Name with full form	<i>SWEET11a</i> : Sugars Will Eventually be Exported 11a, <i>SWEET13</i> : Sugars Will Eventually be Exported 13, <i>SWEET14</i> : Sugars Will Eventually be Exported 14
Target variety:	Naveen, Swarna
Brief Description: Hypothesis, expected outcome	SWEET genes have positive a role in sugar transportation in plants, including rice. Expression of these genes is enhanced by bacterial pathogen <i>Xanthomonas oryzae</i> pv. <i>oryzae</i> (Xoo) through its TALE effectors which bind at the promoter regions SWEET genes. Hence, modification of those nucleotides which are target sites for TALE will abolish their binding. This would deprive bacteria of essential nutrients and restrict the growth.
Vector details: Background and guide details	The multiplex gRNA cloning vector was developed in the background of pRGE32, a binary vector for rice transformation having Kanamycin and Hygromycin markers for bacterial and plant selection, respectively. The fragment consisting of guide-RNAs for along with scaffold RNA was inserted at BsaI sites in pRGE32 plasmid.

Map:



Name of the scientist(s)

Dr. Devanna and Dr. Sanghamitra Samantaray

Title of the work:	Adenine Base Editing for crop improvement
Target gene: Name with full form	<i>PIN5c</i> (PIN-FORMED 5c)
Target variety:	Kitaake
Brief Description: Hypothesis, expected outcome	A highly efficient ABE was developed for precise editing to enhance C4-like vein density in rice. <i>OsPIN5c</i> overexpression induces C4-like venation, establishing a key regulatory link for C4 trait introduction in C3 crops. The base editing is likely
Vector details: Background and guide details	RGEB32 One guide (guide 1) for <i>PIN5c</i> target
<p>Map:</p>  <p style="text-align: center;">ABE8e-NG-PIN5cg1 16,315 bp</p>	
Name of the scientist(s)	Dr. M J Baig and Dr. K A Molla

Vector 26

Title of the work:	Adenine Base Editing for crop improvement
Target gene: Name with full form	<i>PIN5c</i> (PIN-FORMED 5c)
Target variety:	Kitaake
Brief Description: Hypothesis, expected outcome	A highly efficient ABE was developed for precise editing to enhance C4-like vein density in rice. The SHR/SCR pathway regulates vein density, with OsSHR1 and OsSHR2 interacting with OsIDD12 and OsIDD13 to influence auxin efflux via OsPIN5c. OsPIN5c overexpression induces C4-like venation, while SHR represses OsPIN5c through IDD binding, establishing a key regulatory link for C4 trait introduction in C3 crops.
Vector details: Background and guide details	pRGEB32 One guide (guide 2) for PIN5c target
Map:	
Name of the scientist(s)	Dr. M J Baig and Dr. K A Molla

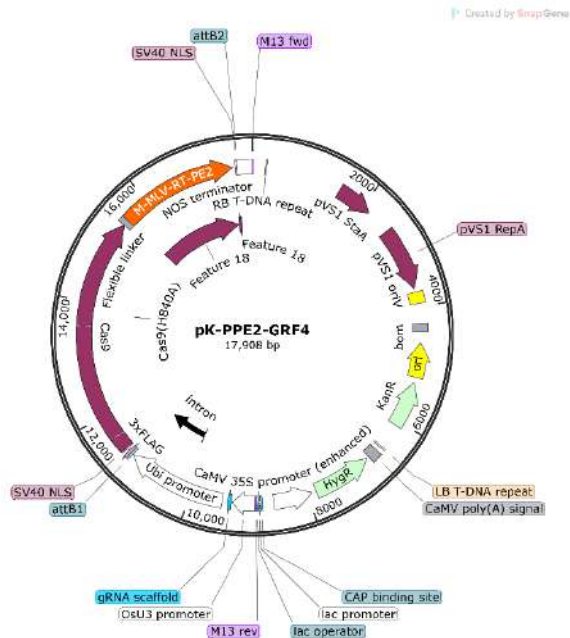
Vector 27

Title of the work:	Adenine Base Editing for crop improvement
Target gene: Name with full form	<i>ACC</i> (Aminocyclopropane-1-Carboxylic Acid), <i>GS1</i> (Glutamine Synthetase 1)
Target variety:	MTU1010
Brief Description: Hypothesis, expected outcome	Development of a highly efficient ABE for precise editing and improvement of rice traits such as multiple herbicide tolerance.
Vector details: Background and guide details	RGE32 1 guide each for ACC and GS1 target
Map:	<p style="text-align: center;">ABE-NG-ACC1+GS1 16,753 bp</p>
Name of the scientist(s)	Dr. M J Baig and Dr. K A Molla

Vector 28

Title of the work:	Improving nitrogen use efficiency in Rice
Target gene: Name with full form	<i>OsGRF4</i> (Growth Regulating Factor 4)
Target variety:	MTU1010 (Indica)
Brief Description: Hypothesis, expected outcome	The GROWTH REGULATING FACTOR 4 (<i>GRF4</i>) transcription factor in rice enhances nitrogen assimilation, carbon fixation, and growth. In the NM-73 variety, a natural TC-to-AA base change disrupts the miRNA binding site in <i>GRF4</i> , improving Nitrogen Use Efficiency (NUE). This study aims to mimic this mutation in the MTU1010 variety using prime editing technology to enhance NUE.
Vector details: Background and guide details	pK-PE2-GRF4, <i>GRF4</i> prime editing guide RNA (pegRNA- Protospacer, scaffold, RT template and PBS)

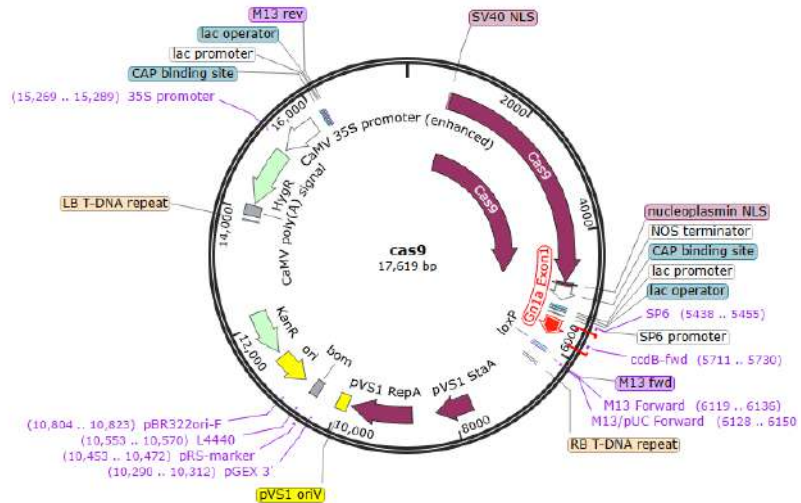
Map:



Name of the scientist(s)	Dr. M J Baig and Dr. K A Molla
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Title of the work:	Downregulation of OsGn1a gene to improve yield in indica rice
Target gene: Name with full form	Grain number 1 a (<i>Gn1a</i>)
Target variety:	Swarna
Brief Description: Hypothesis, expected outcome	The Gn1a gene encodes the OsCKX2 (Cytokinin Oxidase 2) enzyme, which plays a key role in the degradation or metabolism of active cytokinins in the inflorescence meristem of rice. Downregulating the OsGn1a gene leads to a reduction in CKX2 enzyme levels in the inflorescence meristem, resulting in elevated cytokinin levels. This increase in cytokinin promotes spikelet formation and grain number, ultimately enhancing rice yield.
Vector details: Background and guide details	pYLCRISPR-Cas9Pubi-H; guide RNA expressed by U6a promoter.

Map:



Name of the scientist(s)	Dr. Sanghamitra Samantaray
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Vector 30

Title of the work:	Editing of OsXND1 gene for development of drought tolerance rice
Target gene: Name with full form	Xylem NAC-domain 1 (<i>XND1</i>)
Target variety:	Swarna
Brief Description: Hypothesis, expected outcome	AtXND1 acts as a negative regulator of xylem formation and drought tolerance in Arabidopsis. Identifying the homologous gene of AtXND1 in rice and downregulating its expression could enhance drought tolerance in rice.
Vector details: Background and guide details	pYLCRISPR-Cas9Pubi-H; guide RNA expressed by U6a promoter.
Map: Source: Addgene	
Name of the scientist(s)	Dr. Sanghamitra Samantaray

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