# Promising rice mutants with high yields, good grain quality, and disease resistance due to irradiation technology

**Duy Duong Tran[1], Manh Ha Nguyen[2], Duy Quy Tran[3*], Thi Dung Quan[4], Hoang Dung Tran[5]**

[1]*Institute of Agricultural Genetics (IAG)*
[2]*Technical Economic Vocational College To Hieu*
[3]*Institute for Asian - Pacific science technology research cooperation*
[4]*Tan Trao University*
[5]*Nguyen Tat Thanh University*

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***Abstract:***

**Three rice genotypes of NPT4, NPT5, and QP-5 were successfully selected through mutation breeding using $Co^{60}$ gamma rays with 250-300 Gy. Of the three lines, two genotypes, NPT4 and NPT5, obtained high yields of 8.9-9.2 tonnes/ha, had growth duration of 103-110 days in the season of autumn and 130-135 days in the spring season. They exhibited dark green leaves, which were found to be adapted to intensification. They were resistant to major pests and diseases, and tolerant to major abiotic stress due to unfavourable conditions, as compared to the popular rice hybrids of Nhi Uu 868, Thien Nguyen Uu 9, and HYT100. The grain quality properties of NPT4 and NPT5 genotypes were also found to be good, identified as such due to their light aroma and amylose content of 15-16%. The genotype QP-5 exhibited a high grain quality and high yield of 6.5-7.0 tonnes/ha, with a growth duration of 105-110 days, and 125-135 days in autumn and spring seasons, respectively. The aromatic rice genotypes offered 10-12% amylose content and became more delicious as compared to the Bac Thom 7 variety.**

***Keywords:*** ***amylose contents, gamma ray, irradiation technology, mutation.***

***Classification number:*** ***3.1***

## Introduction

Ever since the epoch-making discoveries made by Muller and Stadler eighty years ago, the application of mutation techniques using different agents of physical and chemical nature has generated a vast amount of genetic variability and has played a significant role in modern plant breeding and genetic studies. The use of induced mutations over the past five decades has played a major role in the development of smart crop varieties all over the world. The widespread use of induced mutants in plant breeding programmes across the globe has led to the official release of 3,222 plant mutant varieties from more than 170 different plant species in more than 60 countries throughout the world; the developed varieties increase biodiversity and provide breeding material for conventional plant breeding, thus directly contributing to the conservation and use of plant genetic resource [1].

Before and up through 2016, Vietnam had developed 78 mutant rice varieties, according to the IAEA database. Vietnam is ranked 8$^{th}$ for mutant breeding in the world [2, 3]. The list includes 32 mutant rice varieties accordingly identified as: DT10, DT11, DT13, DT33, A20, DT21, DV2, DCM1, Khang Dan Dot Bien, DT37, DT39, VND-95-20, VND-99-3, Tai Nguyen Dot Bien, Tam Thom Dot Bien, P6DB, ST3DB, DB5, and DT39 [4]. Three rice genotypes NPT4, NPT5, and QP-5 were created and selected from leading rice cultivars as ST19 and DH18 using $Co^{60}$ gamma rays.

According to the Department of Phytosanitary VIII across several regions and ranging from 1/10/2015 to 2/12/2015, department procedures completed nearly 70 batches of imported hybrid rice seeds from China in spring 2014, with the total amount of seeds being 5,616 tonnes, including 4,496 tonnes of Nhi Uu 868 hybrid seeds; 376 tonnes of Nhi Uu 63; 350 tonnes of Nhi Uu 7; 271 tonnes of San Uu 63, and some varieties including: Nghi Huong 305, Xuyen Huong 178, Kinh So Uu 1588, Thinh Du 4, Thinh Du 11, and Hoa Uu 2. Although dominant rice hybrids usually produce high yields, in fact, no pest resistant rice hybrids is as pure as rice with *Xanthomonas* bacteria, and as far as pests of rice hybrids, they were

*Corresponding author: Email: duyquyvaas@gmail.com

great, especially when combined with China's rice hybrids. Furthermore, the average yield of rice hybrids cultivated in Vietnam is higher than the amount cultivated of pure rice by only about 10-15%. So if Vietnam does not actively study the creation of rice hybrids and pure rice varieties that produce high yields of high-quality rice, we will always spend foreign currency to import rice hybrids and continue our dependence on China and other countries [5].

The objectives of this study aim at: (1) Releasing new rice genotypes with short growth durations of 105-110 days in the summer season, and 120-135 days in the spring season, (2) and enhancing the highest yields by 7-10 tons/ha with resistance to blast, sheath blight, brown plant hopper, and being widely adaptable. These new genotypes could partly replace the proportion of cultivable area used for rice hybrid varieties, including in Khang Dan 18 and Q5 introduced from China.

## Materials and methods

### *Materials*

Rice seeds of two varieties: DH18 and ST19 were collected and stored in IAP.

### *Methods*

Dry seeds of two varieties DH18 and ST19 with 13% grain moisture were irradiated by gamma ray $Co^{60}$ at 25-30 Krad in the summer season of 2009.

After treatment, the normal seeds were collected, germinated, and planted in the IAP experimental field; this happened through individual selections of M2, M3, M4, and M5 from 2010 to 2012 during two different seasons.

The selection criteria emphasised the growth duration, plant height, panicle length, number of filled grains per panicle, grain shape, 1,000-grain weight, and grain yield. Biotic stress evaluation was done through IRRI-SES score with 9 levels.

- Fertiliser (NPK/ha) was applied at a rate of 100N: 90P: 80K kg/ha.

- Data is processed using IRRISTAT and Excel.

- DUS and VCU were conducted at the National Center for Yield Trial and Seed Certification - MARD.

- Quality test: TCVN 1643:1992.

- VCU test: Area of at least 1,000 $m^2$ per each site was required with local control varieties.

## Results and discussions

### *Breeding schedule of new mutants NPT4, NPT5, and QP-5*

Dry seeds of two varieties, DH18 and ST19, had 13% moisture and were irradiated with gamma ray $C0^{60}$ at 250-300 Gy during the 2009 autumn season. Individual selection was done from the M2 up to M7 (Fig. 1).

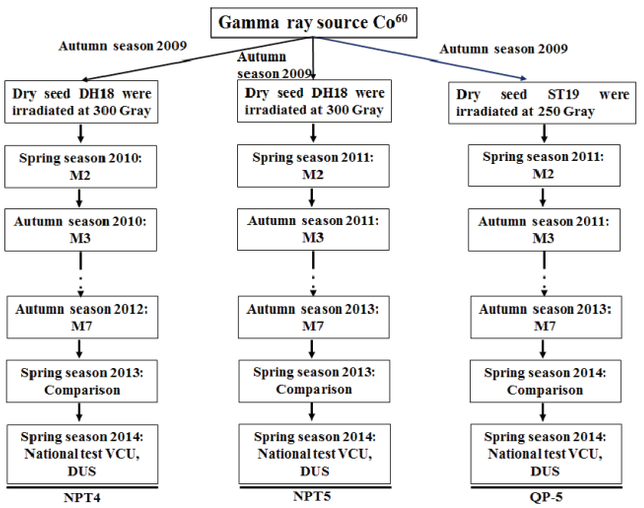


**Fig. 1. Breeding schedule of three rice mutants viz. NPT4, NPT5, and QP-5.**

### *Agronomical traits of NPT4, NPT5, and QP-5 mutants*

The growth duration of the NPT4 genotype occurred for between 130 to 135 days in spring, and between 103 to 110 days in autumn as well as Khang Dan and also 5-7 days earlier than DH18. The NPT4 genotype exhibited as a compact plant type with pale green leaves, and it adapted to intensive practices performed to obtain high yields. Its grain amount per panicle offered 500-600 long, slender grains at a 1,000-grain weight of 20-22 g. Its grain quality property addressed a desirable amylose content of 15.8% to fit the consumer preference; with its gray husk colour as well as Bac Thom 7 (Table 1, Fig. 1).

**Table 1. Agronomical traits of NPT4 rice genotype growing in IAP, Binh Xuyen District, Vinh Phuc Province, 2015.**

| Trait | NPT4 | DH18 | KD18 | Nhi Uu 86b | HYT100 | Thien Nguyen Uu 9 |
|---|---|---|---|---|---|---|
| Growth duration (days) | | | | | | |
| +Spring season | 130-135 | 135-140 | 130-135 | 135-145 | 130-135 | 130-135 |
| +Autumn season | 103-105 | 110-115 | 100-105 | 110-120 | 105-110 | 110-115 |
| Plant height (cm) | 121-125 | 115-125 | 105-110 | 115-120 | 105-112 | 110-115 |
| Plant type | V | V compact | V compact | V compact | V compact | V compact |
| Leaf angle | Horizontal | Horizontal | Straight | Straight | Straight | Straight |
| Grain no./ pan. | 500-600 | 200-450 | 180-200 | 130-160 | 190-220 | 200-250 |
| Unfilled grain (%) | 15-20 | 20-25 | 12-14 | 10-12 | 12-15 | 12-15 |
| 1,000-gr.wgt. (g) | 20-22 | 16-17 | 19-20 | 27-28 | 23-24 | 27-29 |
| Yield (100 kg) | 90-100 | 75-85 | 50-65 | 70-75 | 80-85 | 80-85 |
| Amylose (%) | 15.8 | 16.5 | 26-27 | 16-17 | 20-21 | 16-17 |
| Alkaline digestion | 4.7 | 4.8 | 4.9 | 4.2 | 3.9 | 4.3 |
| Brown rice (%) | 78.5 | 75.5 | 79.8 | 78.2 | 79.6 | 72.0 |
| Milled rice (%) | 70.5 | 69.5 | 70.2 | 70.4 | 70.8 | 65.6 |
| Grain length (mm) | 6.5 | 6.1 | 5.7 | 6.8 | 7.0 | 6.4 |
| L : l ratio | 2.90 | 2.69 | 2.70 | 2.85 | 3.05 | 2.30 |

The growth duration of NPT5 occurred at between 130-135 days in spring and between 105-110 days in autumn (105-110 days) as well as Khang Dan and 3-5 days earlier than DH18. The NPT5 exhibited a compact plant type with pale green leaves and adapted to intensive practices utilised to obtain high yields. Its grain number per panicle offered 400-550 long, slender grains and had a 1,000-grain weight of 20-23 g. Its grain quality addressed the desirable amylose content of 15.9%, which is enough to fit with consumer preference for a bright yellow husk (Table 2, Fig. 2).

**Table 2. Agronomical traits of NPT5 rice genotype growing in IAP, Binh Xuyen District, Vinh Phuc Province, 2015.**

| Trait | NPT5 | DH18 | KD18 | Nhi Uu 86b | HYT100 | Thien Nguyen Uu 9 |
|---|---|---|---|---|---|---|
| Growth duration (days) | | | | | | |
| + Spring season | 130-135 | 135-140 | 130- 135 | 135-145 | 130-135 | 130-135 |
| + Autumn season | 105-110 | 110-115 | 100-105 | 110-120 | 105-110 | 110-115 |
| Plant height (cm) | 120-128 | 115-125 | 105-110 | 115-120 | 105-112 | 110-115 |
| Plant type | V | V compact | V compact | V compact | V compact | V compact |
| Leaf angle | Straight | Horizontal | Straight | Straight | Straight | Straight |
| Grain no./ pan. | 400-550 | 200-450 | 180-200 | 130-160 | 190-220 | 200-230 |
| Unfilled grain (%) | 13-15 | 20-25 | 12- 14 | 10-12 | 12-15 | 12-15 |
| 1,000-gr.wgt. (g) | 20-23 | 16-17 | 19-20 | 27-28 | 23- 24 | 27-29 |
| Yield (100 kg) | 95-100 | 75-85 | 50-65 | 70-75 | 80-85 | 80-85 |
| Amylose (%) | 15.9 | 16.5 | 26-27 | 16-17 | 20-21 | 16-17 |
| Alkaline digestion | 4.8 | 4.8 | 4.9 | 4.2 | 3.9 | 4.3 |
| Brown rice (%) | 79.7 | 75.5 | 79.8 | 78.2 | 79.6 | 72.0 |
| Milled rice (%) | 72.4 | 69.5 | 70.2 | 70.4 | 70.8 | 65.6 |
| Grain length (mm) | 6.7 | 6.1 | 5.7 | 6.8 | 7.0 | 6.4 |
| L: l ratio | 2.85 | 2.69 | 2.70 | 2.85 | 3.05 | 2.30 |

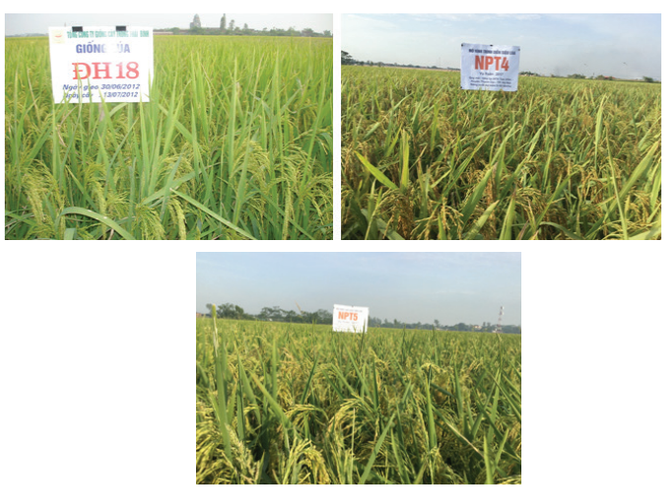


**Fig. 2. NPT4, NPT5 varieties compared with DH18 varieties.**

QP-5 rice mutants exhibited shorter growth duration than ST19 (5-8 days), which was used as a control because of its plant height of 95-100 cm, bright yellow grain husks, and 1,000-grain weigh of 24.8-25.4 g. Its grain quality addressed the desirable amylose content of 10.4-12% as compared to the control of ST19 at 28.7% with a slight aroma (Table 3).

**Table 3. Agronomical traits of the Q5 rice genotype grown in IAP, Cam Giang district, Hai Duong province, 2015.**

| Trait | QP-5 | ST19 |
|---|---|---|
| Growth duration (day) | 105-135 | 115-145 |
| Plant height (cm) | 95-100 | 105-108 |
| Trunk diameter (mm) | 0.4-0.5 | 0.4-0.5 |
| Husk color | Bright yellow | Gold - yellow |
| 1,000-grain weight (g) | 24.8 -25.4 | 22.5-23.8 |
| Amylose content (%) | 10.4-12.0 | 28.7 |
| Aroma (SES score from 0 to 2) | 1 | 1 |
| Grain length (mm) [L] | 6.46 | 5.8 |
| L : l ratio | 3.01 | 2.87 |

***Yield trial of NPT4, NPT5, and QP-5 mutant rice varieties***

The yield trials were conducted over three years continuously at the IAP (2013-2015). The results indicate that NPT4 and NPT5 offered higher yields than the checks of DH18, Khang Dan 18, and the leading hybrid rice varieties as Nhi Uu 86b, Thien Nguyen Uu, and HYT100 (Table 4).

**Table 4. Yield trials of NPT4 and NPT5 as compared to checks at IAP (tonnes/ha).**

| Designation | 2013 Spring | 2013 Autumn | 2014 Spring | 2014 Autumn | 2015 Spring | Average yield |
|---|---|---|---|---|---|---|
| NPT4 | 9.65 | 8.27 | 9.88 | 8.72 | 9.75 | 9.25 |
| NPT5 | 9.05 | 8.14 | 9.56 | 8.54 | 9.57 | 8.97 |
| DH18 | 8.41 | 6.78 | 8.54 | 7.28 | 8.63 | 79.2 |
| Khang Dan 18 (C) | 6.85 | 5.85 | 6.65 | 5.67 | 6.52 | 6.30 |
| Nhi Uu 86b | 8.15 | 7.25 | 8.03 | 7.14 | 8.15 | 7.75 |
| HYT100 | 8.26 | 7.35 | 8.34 | 7.38 | 8.52 | 7.95 |
| Thien Nguyen Uu 9 | 8.44 | 7.46 | 8.47 | 7.46 | 8.5 | 8.07 |
| CV (%) | 4.90 | 6.60 | 7.50 | 5.80 | 7.40 | |
| LSD 0.05 | 2.30 | 2.80 | 1.90 | 2.30 | 2.40 | |

Table 4 shows that the average yield of NPT4 and NPT5 mutants offered higher yields than hybrid rice varieties. NPT4 average yield obtained 9.25 tonnes/ha, which is more than NTP5, at 8.97 tonnes/ha, which was higher than the hybrid rice varieties *viz.* Nhi Uu 86b, HYT100, and Thien Nguyen Uu 9 at 7.75, 7.95, and 8.07 tonnes/ha, respectively.

Table 5 indicates that QP-5 yielded higher than the control (BT7) with an average yield of 6.94 tonnes/ha as compared to BT7, which yielded 5.92 tonnes/ha.

**Table 5. Yield trial of QP-5 as compared to checks at IAP (tonnes/ha).**

| Designation | 2013 Spring | 2013 Autumn | 2014 Spring | 2014 Autumn | 2015 Spring | 2015 Autumn | Average yield |
|---|---|---|---|---|---|---|---|
| QP-5 | 7.02 | 6.58 | 7.38 | 6.76 | 7.25 | 6.67 | 6.94 |
| BT7 (C) | 6.55 | 5.27 | 6.43 | 5.37 | 6.52 | 5.35 | 5.92 |
| CV (%) | 4.80 | 6.60 | 6.30 | 6.20 | 7.10 | | |
| LSD 0.05 | 3.10 | 3.77 | 2.90 | 2.40 | 2.50 | | |

***Disease response by NPT4, NPT5, and QP-5 mutants***

Under field conditions, NPT4 and NPT5 mutants exhibited a resistance to blast, brown planthopper, and sheath blight; even though it was lightly infected by bacterial leaf blight (NPT4).

**Table 6. Reaction to major stresses from NPT4 and NPT5 mutants in two autumn seasons (2013-2014).**

| Designation | Scirpophaga incertulas | Plant hopper | Sheath blight | Bacterial leaf blight | Lodging |
|---|---|---|---|---|---|
| NPT4 | 1-3 | 1-3 | 1-3 | 3-5 | 1-3 |
| NPT5 | 1-3 | 1-3 | 1-3 | 1-3 | 1-3 |
| DH18 | 1-3 | 3-5 | 3-5 | 3-5 | 3-5 |
| Nhi Uu 86b | 1-3 | 1-3 | 1-3 | 3-5 | 1-3 |
| Thien Nguyen Uu 9 | 1-3 | 1-3 | 1-3 | 3-5 | 1-3 |
| HYT100 | 1-3 | 1-3 | 1-3 | 3-5 | 1-3 |
| QP-5 | 1-2 | 1-3 | 1-3 | 1-3 | 1-3 |
| Bao Thai Hong | 1-3 | 3-5 | 3-5 | 1-3 | 3-5 |
| Khang Dan 18 | 3-5 | 3 | 3 | 3-5 | 3-5 |
| BT7 | 3-5 | 3 | 3 | 5-7 | 3-5 |

Sources: IAP, Plant protection research Institute.

Table 6 shows that NPT4, NPT5, and QP-5 mutants highly responded to major stresses as compared to wild rice types and Khang Dan 18 (a leading variety). They also expressed the better reaction as compared to some hybrid rice cultivars in the north.

***Grain quality properties of NPT4, NPT5, and QP-5 mutants***

Table 7 shows that NPT4 and NPT5 offer a higher milled rice percentage than hybrid rice varieties offer. Its amylose content was lower (15-17%) than with BC15, Khang Dan 18, Thien Nguyen Uu 9, and Nhi Uu 86b. Their productivity and quality exhibited better than some rice hybrids. In particular, the QP-5 grain quality was as good as Bac Thom 7 to meet the demand of consumers, with its soft, sticky, and transparent milled rice (Table 8, Fig. 3).

**Table 7. Grain appearance quality of NPT4, NPT5, and QP-5 mutants in Cam Giang district, Hai Duong province in the 2015 autumn season.**

| Designation | Brown rice (%) | White rice (%) | Grain length (mm) | L : l ratio | Amylose contents (%) |
|---|---|---|---|---|---|
| Khang Dan 18 | 79.8 | 70.2 | 5.68 | 2.72 | 26-27 |
| BC15 | 83.3 | 54.1 | 5.70 | 2.9 | 18-20 |
| BT7 | 78.18 | 69.06 | 5.56 | 2.7 | 11.2-12.4 |
| NPT4 | 78.9 | 70.5 | 6.12 | 2.90 | 15-16.2 |
| NPT5 | 79.7 | 72.4 | 6.75 | 2.85 | 15-17 |
| QP-5 | 80.2 | 69.6 | 6.46 | 3.01 | 11.5-12.5 |
| Nhi Uu 86b | 79.1 | 67.5 | 6.20 | 2.3 | 23-25 |
| HYT100 | 80.0 | 65.5 | 6.70 | 2.9 | 18-20 |
| Thien Nguyen Uu 9 | 79.5 | 64.1 | 6.90 | 3.0 | 20-22 |

Sources: IAP Institute.

**Table 8. Grain cooking quality of rice genotypes.**

| Season | Designation | Aroma | Softness | Stickiness | Whiteness | Gloss | Exquisite |
|---|---|---|---|---|---|---|---|
| 2014 Spring | Huong Thom 1 | 2 | 4 | 4 | 5 | 4 | 3 |
| | Bac Thom 7 | 2 | 4 | 4 | 5 | 3 | 3 |
| | Nang Tien | 2 | 4 | 4 | 5 | 3 | 2 |
| | QP-5 | 1 | 4 | 4 | 4 | 3 | 3 |
| | AGPPS 151 | 3 | 4 | 4 | 5 | 3 | 3 |
| | HQT6 (MB68) | 1 | 4 | 4 | 5 | 3 | 3 |
| | KTH08 | 1 | 4 | 4 | 5 | 4 | 3 |
| 2014 Autumn | Huong Thom 1 | 2 | 4 | 4 | 5 | 4 | 3 |
| | Bac Thom 7 | 3 | 4 | 4 | 5 | 4 | 4 |
| | Nang Tien | 2 | 4 | 4 | 5 | 3 | 4 |
| | QP-5 | 2 | 4 | 4 | 5 | 4 | 3 |
| | HQT6 (MB68) | 2 | 4 | 4 | 5 | 3 | 2 |
| | ADI30 | 1 | 4 | 4 | 5 | 3 | 2 |

(Assessing by 10TCN-590-2004 Standard; Source: Data from Center for national testing 2014).

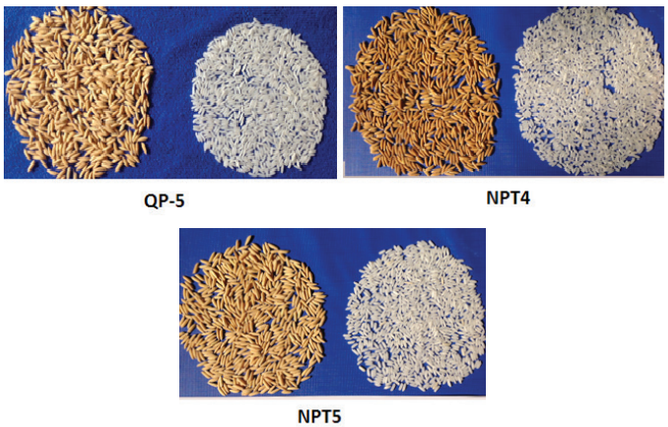


**Fig. 3. Paddy and milled rice of QP-5, NPT4, and NPT5 varieties.**

## Conclusions

Three mutant rice genotypes NPT4, NPT5, and QP-5 were successfully selected and released through $Co^{60}$ induction at 250-300 Gy.

NPT4 and NPT5 yielded an impressive form from between 8.9 to 9.2 tonnes/ha, with shorter growth durations (103-110 days in autumn and 130-135 day in spring), and good phenotypical acceptability as dark green leaves that are compact plant types. This is suitable for intensive practice as compared to Nhi Uu 86b, Thien Nguyen Uu 9, and HYT100, which are the leading cultivars.

NPT4 and NPT5 exhibited their good grain quality properties as lightly scented grain, with an amylose content of 15-16%, and able to meet consumer demand in Northern Vietnam.

The QP-5 rice mutant exhibited high quality and yielded 6.5-7.0 tonnes/ha over a short growth duration of 105-110 days in autumn, and 125-135 days in spring. QP-5 is considered as an aromatic rice genotype with an amylose content of 10-12% to be recommended to commercialise as Bac Thom 7 cultivar.

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