

Genetic Engineering of Local Cayenne Pepper (*Capsicum frutescens* L.) Through Breeding With Multigamma Irradiation Methods to Obtain Superior Offspring

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Abstract

This study purpose to genetically engineer local cayenne pepper through breeding with multigamma irradiation methods to obtain superior offspring that adapt to drought stress, extreme weather, pest tolerance, and high production. The method used consists of observation, sampling, irradiation, careful selection, purification, comparative, and interpretation. The brief procedure of the study included: observations for taking samples, inventorying the physical characteristics of local chili parent varieties, selecting research sites, cultivating the planting area, irradiating the sample at a dose of 2500 rads for 30 minutes, soaking the planting area, planting seeds, irrigation, observing the age and ability to grow seeds, doing embroidery, weeding and fertilizing, observing the condition of plants during growth, harvesting, weighing the mass of fruit per tree, analyzing several nutritional content, comparing the physical and chemical characteristics of the parent varieties and selected superior offspring, and interpreting. Result of research: Local cayenne pepper of superior selected offspring as a result of multigamma irradiation can adapt to drought conditions, extreme weather, tolerant of pests and diseases, and significantly increase production compared to the parent variety. The average production of the selected superior offspring was 11.11 t ha⁻¹, while the parent variety was 6.54 t ha⁻¹ with a percentage increase in the production of local cayenne pepper from the selected superior offspring was 41.13%.

Keywords: cayenne pepper, breeding, genetic, multigamma irradiation, superior offspring

1. Introduction

1.1 The Problem and Purpose of Research

The main problem investigated in this work was genetic engineering of local cayenne pepper through breeding to use multigamma irradiation. The purpose of this research is genetic engineering of local cayenne pepper through breeding with multigamma irradiation method to obtain superior offspring that can adapt to less water and extreme weather, tolerant of pests and diseases, and high production.

1.2 The Origin of Cayenne Pepper

Cayenne pepper belongs to the eggplant family (Solanaceae) originating from Peru, the American continent which then spread throughout America, Europe, Africa, and Asia. In the 16th century, Portuguese and Spanish traders introduced chili originating from South America to Asia and Africa through trade routes (Law et al., 2015; Assagaf, 2017). More than 100 chili species have been identified, of which 5 have been cultivated, namely *Casicum*, *Annuum*, *Chinense*, *Frutescens*, *Baccatum*, and *Pubescens* (Effendi et al., 2018).

1.3 The Importance of Cayenne Pepper Developed

Cayene Pepper is one of the important vegetable crops (horticultural commodities) because of its high nutritional content, especially cayenne pepper, has many benefits for consumers (Sofa et al., 2019), and has high economic value (Wulandari et al., 2018; Eliyatinationsih & Mayasari, 2019). The nutritional content or nutrients in 100 grams (g) of cayenne pepper are: 4.7 g protein, 4.2 g fat, 19.9 g carbohydrates, 45 mg calcium, 8 mg phosphorus, 11 mg vitamin A, 70 mg vitamin C, and 103 calories of energy (Assagaf, 2017; Hayatudin, 2021). Cayenne pepper contains resin, carotenoids, *capsantin* and *capsaicin* which give a spicy taste (Silvia et al., 2016;

Alimuddin et al., 2019), volatile alkaloids, volatile oil, vitamins A and C, tannins, phenols, flavonoids, anthraquinones, saponins, glucosides, limonoids, terpenoids, and carotenoids (Kusnadi et al., 2019), *dihydrocapsainin*, *capsainoid*, beta carotene (Sutomo et al., 2017), mineral salts such as calcium, iron, potassium, and phosphorus (Asmal, 2018), and in cayenne pepper seeds contained solanine, *solamargarine*, *solamidine*, *solasomine*, *solasodine*, and steroids. The special nutritional content in cayenne pepper includes vitamin A = 11.8836 IU/11 g, vitamin B = 763.52 ppm, protein = 5.83% (Mahyudi, 2015).

1.4 Literatures Study

Chili which is one of the leading national horticultural commodities experiences fluctuations in production from year to year (Nurjanni, 2016) so that it is not comparable to the demand for chili needs among the people which continues to increase (Septiadi et al., 2020; Prabowo et al., 2018; Wachdijono, 2020; Salampessy et al., 2019). The results of the BPS survey (2017) reported that the consumption of cayenne pepper in 2013 was 316.57 thousand tons per year, and in 2014 it increased to 318.21 thousand tons per year. The relatively high demand for cayenne pepper is caused by the content of nutrients that are needed for health (Fitri et al., 2020). Some areas producing cayenne pepper in Indonesia experienced a decline in production, including: (1) Cayenne pepper production in Aceh in 2015 was 58,918 tons (t) per year, decreased to 46,405 t in 2016, and in 2018 cayenne pepper production in Aceh decreased to 62,167 t to 61,887 t in 2019; (2) Cayenne pepper production in South Sumatra in 2017 was 15,826 t per year, decreased to 13,451 t in 2018, and 11,014 t in 2019; (3) Lampung area: cayenne pepper production in 2017 was 14,705 t decreased to 14,648 t in 2018, 12,796 t in 2019; (4) In 2017 the production of cayenne pepper in West Java amounted to 13,910 t, decreased to 131,418 t in 2018, and continued to decline in 2019 to 128,494 t; (5) Cayenne pepper production in West Kalimantan in 2016 was 4,753 t, down to 4,719 t in 2017, and production continued to decline in 2018 to 4,165 t; (6) In 2017, the production of cayenne pepper in Maluku was 4,039 t, down to 3,732 t in 2018, and continued to decline in 2019 to 3,440 t; (7) Cayenne pepper-producing areas that have decreased production for 3 consecutive years are West Papua, namely production of 1,304 t in 2016, 889 t in 2017, and 625 t in 2018 (BPS, 2019). In 2012-2015, the production of cayenne pepper in North Central Timor Regency decreased successively, namely 66.6 t (in 2012), 63.5 t (in 2013), 50.8 t (in 2014) and 35.5 t (in 2015) (Bete & Taena, 2018).

The low productivity of chili is caused by several factors, including: (1) the use of superior varieties of seeds is still relatively lacking (Azwir et al., 2018); (2) the use of types of fertilizers with inappropriate doses (Adetya et al., 2018; Ernita et al., 2020; Hutubessy, 2017; Ilyasa et al., 2018; Karim et al., 2019); (3) pest control has not maximum; (4) inadequate planting of land; (5) the presence of toxic elements in the soil in chili plants (Edy et al., 2017); (6) inadequate irrigation; (7) post-harvest processing adequate (Edowai et al., 2016; Putri et al., 2020).

1.5 Problem Solution

To increase the productivity of cayenne pepper, the first step that must be taken is the procurement of superior variety seeds through breeding using modern technology. One of the effective and efficient methods that requires a relatively short time is the breeding of cayenne pepper with Nuclear Techniques (multigamma irradiation).

2. Method

2.1 Research Sites

Samples of local cayenne pepper developed in this study were taken at the center of cayenne pepper farmers' plantations in Fukdale Kupang Timur, Kefamenanu, and Malaka. The irradiation of local cayenne pepper samples was carried out at the Mini Nuclear Bioscience Laboratory, Nusa Cendana University. Planting locations are carried out in farmers' gardens in four provinces, namely East Penfui, East Nusa Tenggara Province, Mamuju, West Sulawesi Province, Tinoring, South Sulawesi Province, and Palu, Central Sulawesi Province.

2.2 Research Method

The methods used in this study include observation: carry out activities for sampling local cayenne pepper and determining planting locations (research locations), sampling: Sampling of local cayenne pepper at farmer's plantation centers in Menado North Sulawesi and Palu Central Sulawesi, irradiation: irradiation of local cayenne pepper seed samples at a dose of 2500 rads for 30 minutes, careful selection: selecting cayenne pepper plants individually starting at the age of 30 days after planting until harvesting is complete, analysis: calculate the percentage of seed growth, average production, and the percentage increase in cayenne pepper production, comparative: Comparing the characteristics of superior offspring varieties with their parent varieties, and interpretation: interpret the results that have been achieved.

2.3 Research Procedure

The brief procedure of this research includes: (1) observations for sampling local cayenne pepper, selection of research sites, and collecting characteristics of parent varieties; (2) irradiation of samples; (3) cultivation of planting land; (4) planting; (5) observation of early seed growth; (6) weeding, fertilizing, and watering; (7) individual plant selection; (8) observation during growth (pest and disease resistance, adaptation to water shortages, and extreme weather); (9) harvesting, drying, and storage; (10) analysis of several nutrients.

2.4 Mathematical Equations for Data Analysis

The mathematical formulation required for data analysis consists of (Pasangka and Rfli, 2016; Pasangka and Irvandi, 2021).

2.4.1 Calculation of the Percentage of Growing Local Cayenne Pepper Seeds on Superior Offspring and Parent Variety

Treatment sample:

$$PG_{TS} = \left(\frac{T_{AS} - A_{TS}}{T_{AS}} \right) \times 100\% \quad (1)$$

Where, PG_{TS} : percentage of growth on treatment (%), T_{AS} : the total number of seeds planted, A_{TS} : number of seeds that did not grow on treatment sample.

Control sample:

$$PG_{CS} = \left(\frac{T_{AS} - A_{CS}}{T_{AS}} \right) \times 100\% \quad (2)$$

Where, PG_{CS} : percentage of growth on control sample (%), A_{CS} : the number of seeds that did not grow on control sample (%).

2.4.2 Calculation of the Average Production of Selected Superior Offspring at the Four Planting Locations

$$A_{PTS} = \frac{P_{L1} + P_{L2} + P_{L3} + P_{L4}}{4} \quad (3)$$

Where, A_{PTS} : the average production of selected superior offspring (t/ha), P_{L1} , P_{L2} , P_{L3} : production of superior offspring and control samples in four planting locations.

2.4.3 Calculation of the Average Production in the Control Sample at the Four Planting Locations

$$A_{PCS} = \frac{P_{L1} + P_{L2} + P_{L3} + P_{L4}}{4} \quad (4)$$

Where, A_{PCS} : the average production of control sample.

2.4.4 Mathematical Formulation to Calculate the Average Percentage Increase in Production

$$I_{PAP} = \left(\frac{A_{PTS} - A_{PCS}}{A_{PTS}} \right) \times 100\% \quad (5)$$

Where, I_{PAP} : percentage increase in average production (%).

3. Results

3.1 Observation, Measurement, and Calculation Results

The results obtained based on observation, measurement, and calculation data show the physical characteristics of local cayenne pepper varieties of parent and offspring developed through breeding using multigamma (nuclear) irradiation methods and careful selection to obtain superior offspring that can adapt to drought conditions (lack of land water), extreme climate, pest-disease tolerance, and high production. The real results obtained are shown in Figures 1a, 1b, 2a, and 2b, Tables 1-3 as follows.



Figure 1a. Show an example of local cayenne pepper fruit of selected superior offspring at the age of 48 days after planting (d a p)



Figure 1b. Show an example of local cayenne pepper fruit of selected superior offspring at the age 80 d a p



Figure 2a. Show an example of local cayenne pepper fruit of parent variety at the age of 80 d a p



Figure 2b. Show an example of local cayenne pepper fruit of parent variety at the age of 100 d a p

3.2 Data to Calculate the Percentage of Growing Seeds

To calculate the percentage of growing local cayenne pepper seeds in the control sample and the treatment sample, five (5) groups of samples were taken randomly with the number of seeds observed as many as 30 seeds in each group. The treatment sample consisting of five (5) groups was taken with five (5) variations of observations which are clearly visible in Table 1.

Table 1. The results of the observation of the number of cayenne pepper seeds that did not grow in the control and the treatment samples

GS	Number of seeds in each group	CS	Number of seeds that do not grow				
			Selected Superior Offspring				
			1	2	3	4	5
I	30	8	4	5	3	5	4
II	30	10	6	5	7	4	5
III	30	9	5	3	4	7	5
IV	30	9	4	6	6	5	7
V	30	11	7	6	7	3	5
Average = 30		9.40	5.20	5.00	5.40	4.80	5.20
The total average in the selected superior offspring					5.12		
Percentage of Growth in control and treatment samples		68.67%	82.67%	83.33%	82.00%	84.00%	82.67%
Average total of growth percentage		68.67%			82.93%		

Note. GS: Group Sample; CS: Control Sample.

Table 2. Important physical and chemical characteristics from observations, measurements, and calculations on local cayenne pepper parent and selected superior offspring

No.	Description	Parent Variety	Selected Superior Offspring
1	Growing time	11 days after planting (d a p)	7 days after planting (d a p)
2	Growth percentage	68.67%	82.93%
3	Flowering age range	(78-97) d a p	(43-55) d a p
4	Average flowering age	83 d a p	48 d a p
5	Plant height range	(82.5-108.5) cm	(92.3-130.0) cm
6	Average plant height	98.5 cm	115.5 cm
7	Adapt to dry land and extreme weather	Less adaptive	Adaptive
8	Pests and diseases tolerant	Less tolerant	Tolerant
9	Harvest age range	(114-125) d a p	(70-95) d a p
10	Average harvest age	118 d a p	88 d a p
11	Fruit length range	(1.20-2.16) cm	(2.31-3.50) cm
12	Average length of fruit	1.97 cm	2.85 cm
13	Fruit diameter range	(1.25-1.95) cm	(2.20-3.25) cm
14	Average fruit diameter	1.65 cm	2.86 cm
15	Range of fruit per tree	96-155	127-215
16	Average number of fruits per tree	115	185
17	Fruit mass range per tree	(300-400) g	(550-750) g
18	Average fruit mass per tree	365 g	650 g
19	Young fruit color	Green	Green
20	Ripe fruit color	Dark red	Light red
21	Production range	(5.83-7.15) t ha ⁻¹	(10.78-11.29) t ha ⁻¹
22	Average production	6.54 t ha ⁻¹	11.11 t ha ⁻¹
23	Maximum production potential	7.15 t ha ⁻¹	11.29 t ha ⁻¹
24	Percentage of production increase	-	41.13%
25	Protein	4.2 g	4.9 g
26	Calcium	43.5 g	51.3 mg
27	Phosphor	7.8 mg	9.6 mg
28	Vitamin A	10.5 mg	11.85 mg
29	Vitamin C	68.2 mg	76.3 mg
30	Carbohydrate	19.7 g	16.8 g

Table 3. Production levels at the four planting sites in the control (parent variety) and the treatment samples (selected superior offspring)

No.	Planting Site	Parent Variety	Selected Superior Offspring
1	East Penfui Province of East Nusa Tenggara Timur	7.15 t ha ⁻¹	11.29 t ha ⁻¹
2	Mamuju Province of West Sulawesi	5.83 t ha ⁻¹	11.26 t ha ⁻¹
3	Tinoring Province of South Sulawesi	6.45 t ha ⁻¹	10.78 t ha ⁻¹
4	Palu Province of central Sulawesi	6.72 t ha ⁻¹	11.12 t ha ⁻¹
Average production		6.54 t ha ⁻¹	11.11 t ha ⁻¹
Percentage increase in production			41.13%

3.3 Statistical Analysis

The growth percentage of seed in the control and treatment samples calculated by using Equations (1) and (2), and data in Table 1 as follows.

The number of observed seeds that were randomly selected was 30 seeds, the average seed that did not grow in the control sample was 9.40 seeds. The average of seeds that did not grow in the selected superior offspring was 5.12 seeds.

Percentage of growth in the control sample:

$$PG = \left(\frac{T_{AS} - A_{SG}}{T_{AS}} \right) \times 100\% = \left(\frac{30 - 9.40}{30} \right) \times 100\% = 768.67\%$$

Percentage of growth in the treatment sample:

$$PG = \left(\frac{T_{AS} - A_{SG}}{T_{AS}} \right) \times 100\% = \left(\frac{30 - 5.12}{30} \right) \times 100\% = 82.93\%$$

Average production on control and treatment samples of cayenne pepper calculated by Equation (3) dan (4) and percentage of increase in production of selected superior offspring of local cayenne pepper as a result of multigamma irradiation calculated by Equation (5) as follows.

Average production in the control sample (parent variety) at the four planting sites:

$$A_{PCS} = \frac{P_{L1} + P_{L2} + P_{L3} + P_{L4}}{4} = \left(\frac{7.15 + 5.83 + 6.45 + 6.72}{4} \right) = 6.54 \text{ t ha}^{-1}$$

The average production of treatment sample or selected superior offspring of cayenne pepper from multigamma irradiation at the four planting sites:

$$A_{PTS} = \frac{P_{L1} + P_{L2} + P_{L3} + P_{L4}}{4} = \left(\frac{11.29 + 11.26 + 10.78 + 11.12}{4} \right) = 11.11 \text{ t ha}^{-1}$$

Percentage of increase in production in selected superior offspring of local cayenne pepper as a result of multigamma irradiation:

$$I_{PAP} = \left(\frac{A_{PTS} - A_{PCS}}{A_{PTS}} \right) \times 100\% = \left(\frac{11.11 - 6.54}{11.11} \right) \times 100\% = 41.13\%$$

4. Discussion

4.1 Growing Time and Percentage of Seed Growth, Adaptation to Drought and Extreme Weather Conditions, Tolerance to Pests and Diseases

The growing time for the parent variety of local cayenne pepper was 11 days after planting (da p), while the high-yielding cultivars were selected 7 days after planting (d a p) with growth percentages of 68.67% (parent variety) and 82.93% (selected superior offspring). Figure 1 shows the growth of selected local cayenne peppers as a result of multigamma irradiation with thriving leaves, large and smooth fruits, while Figure 2 shows the growth of local cayenne peppers of parent varieties with rapidly yellowing leaves and small fruits. These data indicate that local chilies of superior breeds grow faster, adapt to drought and extreme weather conditions, and are tolerant of pests and diseases compared to their parent varieties.

4.2 Flowering and Harvesting Age

The selected superior local cayenne pepper has a flowering age range (43-55) dap with an average flowering age of 48 dap, while the range and average flowering age of the parent variety are (78-97) dap and 83 dap, respectively. The harvest age range of the selected superior offspring was (70-95) dap with an average harvest age of 88 dap, while the harvest age range for the parent variety (114-125) dap with an average harvest age of 118 dap. This proves that the selected superior local cayenne peppers flower and harvest faster than their parent varieties.

4.3 Production Rate and Percentage of Production Increase

The number of fruits per tree in selected local cayenne peppers ranged from (127-215) fruit with an average fruit per tree of 185 fruit, while the range for the number of fruits in the parent variety was (96-155) fruit with an average number of fruits per tree 115 fruit. The mass for selected superior cayenne peppers per tree ranged between (550-750) g with an average mass per tree of 650 g, while the mass range per tree of the parent variety was (300-400) g with an average mass of 365 g. The production range of selected superior cayenne pepper was (10.78-11.29) t ha⁻¹, with an average production of 11.11 t ha⁻¹, while the production range of the parent variety was (5.83-7.15) t ha⁻¹, with an average production of 6.54 t ha⁻¹. This shows that the superior cayenne pepper selected as a result of multigamma irradiation has more fruit, higher mass per tree, and higher production than the parent variety with a percentage increase of 41.13% in production.

4.4 Nutritional Content

The content of several nutrients in selected local cayenne peppers includes protein 4.9 g, calcium 51.5 mg, phosphorus 9.6 mg, vitamin A 11.85 mg, vitamin C 76.3 mg, and carbohydrates 16.8 g, while the parent variety: protein 4.2 g, calcium 43.5 g, phosphorus 7.8 mg, vitamin A 10.5 mg, vitamin C 68.2 mg, and carbohydrates 19.7 g. These data indicate that the nutritional content of selected high-yielding cayenne peppers is higher than the nutritional content of the parent varieties, except for carbohydrates.

5. Conclusion

The selected superior offspring of local cayenne pepper variety as a result of multigamma irradiation can adapt to drought conditions, extreme weather, tolerant to pest-disease, high nutrition content, and significantly increase production compared to the parent variety. The average production of the selected superior offspring was 11.11 t ha⁻¹ while the parent variety was 6.54 t ha⁻¹ with a percentage increase in the production of local cayenne pepper from the selected superior offspring was 41.13%.

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