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Genetic Characterisation of Anthocyanin Accumulation in Diverse Rice Germplasm

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ABSTRACT

Anthocyanins are natural pigments found in various crops, including rice, such as black rice. They enhance food quality and help fight oxidative stress, inflammation, and chronic diseases. New black rice genotypes have been introduced into Uganda, however, the genetic analysis of anthocyanin content among them is unknown, which complicates efforts to improve anthocyanin levels in Ugandan rice germplasm. Thus, this study aimed to determine the gene action, heritability and combining ability for anthocyanin content in black rice genotypes. The study was carried out on black rice genotypes (BR-11, BR-12, BR-14 and BR-15) and commercial white rice varieties (NR-1, NR-2, NR-3, NR-4 and OKILE) crossed using the North Carolina II mating design. The F₂ seeds were evaluated for anthocyanin content using the pH-differential method, and data were analysed using GenStat of the 23rd edition. The study revealed significant differences in general combining ability (GCA) and specific combining ability (SCA) mean squares, indicating the importance of additive and nonadditive gene action in controlling the trait under investigation. The GCA and SCA variance components revealed a very high broad-sense heritability (> 0.9), indicating the predominance of the genotype over the environment effect, a high narrow-sense heritability (> 0.5), and a Baker's ratio (> 0.5), indicating the predominance of additive inheritance among crosses evaluated in Namulonge at the National Crops Resources Research Institute (NaCRRI), thus anthocyanin content being controlled by an additive type of gene action. Maternal effects were suspected to be present in anthocyanin content, reflected by a higher female GCA variance than male GCA variance. Genotypes BR-11 and BR-12 were good combiners and could be used as donors in improving rice for anthocyanin content by plant breeders. Crosses NR-2/BR-11, NR-3/BR-11, OKILE/BR-11, NR-2/BR12, NR-3/BR-12, NR-4/BR-12, NR1/BR-14, NR1/BR-15 and NR-4/BR-15 were recorded as the best performers for anthocyanin content as they had a high positive SCA, a higher mean, and at least one of the parents had a good GCA for anthocyanin content. Therefore, advancing them and the selections made during early generations is necessary. This study recommends further evaluation of studied materials across different agroecologies for several seasons in Uganda to know the stability, adaptability and generate conclusive results on the type of gene action controlling anthocyanin content. It also recommends the need to evaluate reciprocal crosses to have conclusive results on the maternal inheritance of anthocyanin content.

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1 | Introduction

Rice, a widely cultivated crop belonging to the *Oryza* genus, serves as a crucial staple food, nourishing over half of the global population [1]. It is often hailed as the “grain of life” due to its exceptional nutritional and medicinal attributes, playing a pivotal role in ensuring food security [2]. Rich in bioactive compounds, particularly in landraces, rice demonstrates a significant therapeutic potential. Among these landraces, black rice stands out for its exceptional nutritional and therapeutic properties, containing high levels of beneficial compounds such as phenols, flavonoids and anthocyanins [3]. Black rice, distinguished by its heightened anthocyanin content compared to other varieties, is regarded as a superfood, offering a range of health benefits, including antioxidant, anti-inflammatory, antimutagenic, anticarcinogenic and antiangiogenesis properties [4]. These health-promoting properties help reduce the risk of noncommunicable diseases associated with oxidative stress, which are increasingly prevalent in Uganda and sub-Saharan Africa [5]. With the introduction of black rice lines in Uganda, there is a need for its conservation and detailed genetic evaluation of anthocyanin content. However, comprehensive studies on the inheritance of anthocyanins in African rice breeding materials remain scarce. Understanding the genetic control of anthocyanin accumulation will facilitate effective parent selection and marker-assisted breeding for nutrient-dense varieties, thus strengthening rice breeding efforts by incorporating nutritional quality traits into variety development, contributing to food security, value addition and improved public health [6].

In black rice, the presence of anthocyanin pigments is primarily determined by the expression of specific genes involved in their biosynthesis and regulation, and the understanding of the gene action for anthocyanin content in offspring provides insights into breeding strategies for developing rice varieties with improved anthocyanin content. A heritability study on the anthocyanin content will indicate the likelihood of the trait being passed on to subsequent generations and enhance the prospects of obtaining improved genotypes through selection. A higher heritability value will facilitate selection processes and accelerate breeding cycles [5]. The combining ability study will determine the pattern of genetic effects in the manifestation of anthocyanin content by identifying possibly superior parents and hybrids. General combining ability (GCA), which is the mean performance of a line when expressed as a deviation from the mean of all crosses, is attributed to additive gene effects and additive $\times$ additive epistasis and is theoretically fixable [7, 8]. Conversely, specific combining ability (SCA) pertains to the deviation of each cross from the expected value, attributable to nonadditive gene effects, potentially stemming from dominance or epistasis, or both, and is nonfixable.

North Carolina Design II (NCII), a factorial mating scheme used to evaluate inbred lines for both GCA, SCA and various types of gene actions [9, 10], is a rectangular mating design involving a single group of females mated to a single group of males in all possible combinations, and it has a provision for maternal effects. NCII is reported to have greater precision than NCI, and it is more applicable to self-pollinated crops [11]. So, this study aimed at identifying black rice genotypes with good GCA, determining the best-performing crosses between black rice and white rice for all the studied traits and knowing the mode of gene action and

inheritance of the studied traits using the NCII mating design. The outcome of this study will guide breeding programmes on how best to improve rice for anthocyanin content.

2 | Materials and Methods

2.1 | Experimental Materials

The black rice parents were introduced into Uganda from South Korea, while the white rice parents were chosen from Uganda's commercial rice varieties. Details concerning the materials are reflected in Table 1.

2.2 | Generation of Crosses

Nursery beds for parental materials were established in the screen house for 21 days. Seedlings of the genetic materials were then transplanted into 10 L capacity plastic buckets filled with loam soil at a seedling rate of 3 seedlings per bucket. The plants were crossed at flowering following the North Carolina II mating design of 4×5 , giving 20 F_1 offspring (Figure 1). This was done following the manual emasculation and hooking method described by Jennings et al. [12] to obtain the F_1 seed.

2.3 | North Carolina II Mating Design Scheme

2.3.1 | Experimental Location and Design

This study was conducted in Namulonge, at the National Crops Resource Research Institute (NaCRRI), Wakiso district, in Uganda, during the 2023/2024 cropping season. F_1 seeds were pregerminated in petri dishes in the seed laboratory for 7 days and later transferred onto soil on plastic plates for 14 days. The seedlings were later transplanted in the rice field using a randomised complete block design of 3 replicates. The experimental plot consisted of a single row at a spacing of 20×20 cm and a seeding rate of one seedling per hill. All recommended practices for good crop management were followed. The F_2 seed was further evaluated for anthocyanin content in the laboratory.

2.4 | Data Collection

Anthocyanin data were obtained in the laboratory using the pH differential method with slight modification as described by Sutharut and Sudarat [13] and Jaksomsak et al. [14]. This method quantifies monomeric anthocyanins based on their reversible structural and colour changes at different pH levels, coloured at pH 1.0 and colourless at pH 4.5.

Buffer solutions were prepared using 0.025 M potassium chloride (pH 1.0) and 0.4 M sodium acetate (pH 4.5), with pH adjusted using concentrated HCl. Rice grains (14% moisture) were dehulled, ground, sieved (2 mm) and anthocyanins extracted from 5 g of sample using 10 mL of acidified methanol. The mixture was shaken (500 rpm, 15 min) and centrifuged (6000 rpm, 10 min) to obtain the supernatant. Aliquots of the extract were mixed with each buffer and analysed in triplicate.

Absorbance was measured at 492 nm and 620 nm using a microplate reader, with distilled water as the blank. The corrected absorbance was calculated as $(A_{492} - A_{620})$ at pH 1.0 minus $(A_{492} - A_{620})$ at pH 4.5. Monomeric anthocyanin pigment (MAP) was calculated using the molecular weight (449.2 g mol^{-1}) and molar absorptivity ($\epsilon = 26,900 \text{ L mol}^{-1} \text{ cm}^{-1}$) of

TABLE 1 | Description of parental materials used in the study.

S/N	GEN	Code	Pedigree	Special attributes
1	Black Rice-11	BR-11	HR32054F1-1-28-1-3-2-3	Black, submergence tolerant, Blast and BLB
2	Black Rice-12	BR-12	BR-282-12	Black, tolerant to RYMV, submergence tolerant, Blast and BLB
3	Black Rice-14	BR-14	HR32054F1-1-28-1-1-3-1	Black, submergence tolerant, Blast and BLB
4	Black Rice-15	BR-15	HR32054F1-1-28-1-3-2-3	Black, submergence tolerant, Blast and BLB
5	NARO Rice-1	NR-1	PR 107	Aromatic, high milling recovery, and head rice recovery, early maturing, high yield, resistant to RYMV, Blast and BLS
6	NARO Rice-2	NR-2	ART34-88-1-2-B-1-Nam 2	High yield, slender grains, resistant to RYMV, Blast and BLS
7	NARO Rice-3	NR-3	ART35-272-1-2-B-P-1-UGA 3	Early maturing, heavy grains, resistant to RYMV, Blast and BLS
8	NARO Rice-4	NR-4	IR77454-22-B-20-20-2-2-B-TGRI-UGA1	High yield, heavy and big grains, resistant to RYMV, Blast and BLS
9	OKILE	OKILE	GSR 0057	High-yielding, late-maturing

Note: GEN; genotype. Abbreviations: BLB, bacterial leaf blight; BLS, bacterial leaf streak; RYMV, rice yellow mottle virus; S/N, serial number.

cyanidin-3-glucoside (C3G), which was used as the reference standard. Results were first expressed as mg C3G equivalents per litre (mg/L) and subsequently converted to mg C3G equivalents per 100 g dry weight sample, taking into account the dilution factor, extraction volume, sample weight, and a 1 cm path length. As shown in the following equation

$$MAP = \frac{A \times MW \times DF \times 10^3}{\epsilon \times 1} \tag{1}$$

Quality assurance was ensured by including a certified reference sample (PFSP 36 sweet potato) with known anthocyanin content in each analytical run.

2.5 | Statistical Analysis

Data were analysed using GenStat of the 23rd Edition. Means were separated using Fischer’s least significant difference test at the 5% significance level [3].

2.6 | Estimates of Combining Ability

The combining ability analysis was carried out as per the method suggested by Comstock and Robinson [15]. The variation between males and females and their interaction in the North Carolina II mating design was obtained following the statistical model.

$$Y_{ijk} = \mu + m_i + f_j + m \times f_{ij} + e_{ijk} \tag{2}$$

Here:

Y_{ijk} = K^{th} observation on ij^{th} progeny, μ = general mean. m_i = the effect of the i^{th} male, f_j = effect of the j^{th} female, $(m \times f)_{ij}$ = interaction effect and e_{ijk} = error associated with each observation.

The significances of GCA and SCA effects of the genotypes and crosses were obtained using a standard t -test, using standard errors of GCA and SCA effects.

2.7 | Estimates and Partitioning of Variance Components

Variance components were obtained from the expected mean squares. These were partitioned to obtain the general predictable ratio (GPR), Baker’s ratio (BR), and broad- and narrow-sense heritability estimates as described by Falconer [16].

$$\begin{aligned} \sigma_m^2 &= \frac{[MS_1 - MS_4]}{rf}, \\ \sigma_f^2 &= \frac{[MS_2 - MS_4]}{rm}, \\ \sigma_{mf}^2 &= \frac{[MS_3 - MS_4]}{r}, \\ \sigma_e^2 &= MS_4, \end{aligned} \tag{3}$$

where

MS_1 ; mean squares males, MS_2 ; mean squares females, MS_3 ; mean squares interaction, MS_4 ; mean square error σ_m^2 ; male GCA variance, σ_m^2 ; female GCA variance, σ_{mf}^2 ; SCA variance, σ_e^2 ; error variance.

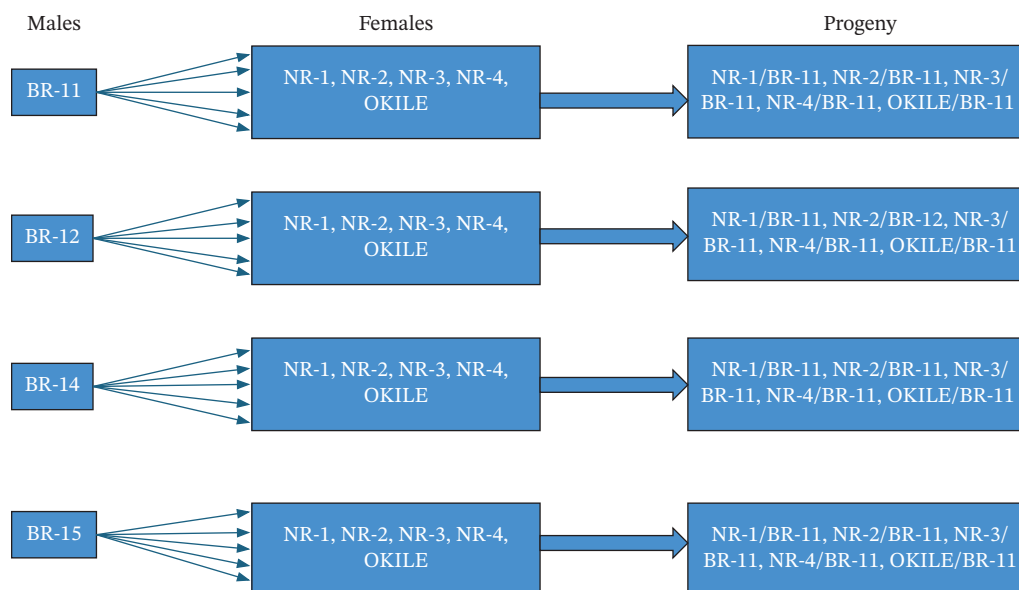


FIGURE 1 | Mating design as used in the experiment.

According to Baker [17], the ratio of combining ability variance components (GPR) is used to determine the gene action involved in the expression of a trait, hence guiding the strategic planning and execution of breeding programmes aimed at enhancing desirable traits in plant species. The GPR and BR are obtained as shown in the following [17]:

$$\text{GPR} = \frac{[\sigma_m^2 + \sigma_f^2]}{[\sigma_{mf}^2]}, \quad (4)$$

$$\text{BR} = \frac{[\sigma_m^2 + \sigma_f^2]}{[\sigma_m^2 + \sigma_f^2 + \sigma_{mf}^2]}.$$

The broad-sense heritability (H) was obtained using the total genetic variance in the following:

$$H = \frac{[\sigma_m^2 + \sigma_f^2 + \sigma_{mf}^2]}{[\sigma_m^2 + \sigma_f^2 + \sigma_{mf}^2 + \sigma_e^2]}. \quad (5)$$

The narrow-sense heritability (h) was obtained as follows:

$$h^2 = \frac{[\sigma_m^2 + \sigma_f^2]}{[\sigma_m^2 + \sigma_f^2 + \sigma_{mf}^2 + \sigma_e^2]}. \quad (6)$$

3 | Results

3.1 | Combining Ability and Heritability for Anthocyanin Content

The analysis of variance for anthocyanin content indicated highly significant ($p < 0.001$) differences in the various sources of variation (GCA male, GCA female and SCA). The female GCA values were larger than the male GCA values, while the SCA values were lower than the GCA of both males and females (Table 2). The female GCA variance was higher than the male GCA variance and SCA variance. The GPR and BR were greater than 0.5 (0.58), while the broad-sense heritability (H) was close to 1 (0.999), and the narrow-sense heritability (h^2) was greater than

0.5 (0.58) (Table 2). The mean anthocyanin content for the crosses averaged 120.65 mg/100 g (ranging from 48.73 to 195.37 mg/100 g), with OKILE/BR-11 having the highest and NR-3/BR-15 having the lowest anthocyanin content. The black rice parents ranged from 243 to 308.9 mg/100 g (Table 3).

3.2 | GCA for Parental Lines Among Studied Crosses for Anthocyanin Content

The GCA was utilised to pinpoint superior parents for direct integration into breeding programmes. Both male and female genotypes showed significant differences in their GCA for anthocyanin content (Table 3). Amongst the males, BR-11 and BR-12 exhibited significant positive GCA effects, with BR-11 having the highest effects (19.37). BR-14 exhibited significant negative GCA effects, and BR-15 exhibited nonsignificant GCA effects. Amongst the females, NR-1, NR-4 and OKILE exhibited significant positive GCA effects, with NR-4 having the highest effects (35.9). NR-2 and NR-3 exhibited significant negative GCA effects (Table 3).

3.3 | SCA Among Studied Crosses for Anthocyanin Content

Crosses NR-2/BR-11, NR-3/BR-11, OKILE/BR-11, NR-2/BR-12, NR-3/BR-12, NR-4/BR-12, NR1/BR-14, NR-3/BR-14, NR1/BR-15 and NR-4/BR-15 had significant positive SCA effects, with OKILE/BR-11 having the highest positive SCA effect (51.81***) while NR1/BR11, NR-4/BR-11, NR1/BR-12, OKILE/BR-12, NR-2/BR14, NR-4/BR-14, OKILE/BR-14, NR-2/BR15, NR-3/BR-15 and OKILE/BR-15 had significant negative SCA effects for anthocyanin content (Table 3).

4 | Discussion

Anthocyanins are natural edible pigments with high nutritional value and pharmacological effects such as improving vision, promoting antioxidant activity, capturing free radicals, and treating cancer in humans [18]. They are becoming increasingly

TABLE 2 | Mean squares, combining abilities, general predictable ratio, Baker's ratio and heritability for anthocyanin content in F₂ rice progenies evaluated at the National Crops Resources Research Institute.

SOV	Df	AN (mg/100 g)
Rep	2	2.05
GCA male	3	5715***
GCA female	4	13,560***
SCA(<i>m</i> * <i>f</i>)	12	3222***
Residuals	38	0.6078
Variance components		
σ_f^2		1130
σ_m^2		381
σ_{mf}^2		1074
GPR		1.4
BR		0.58
<i>H</i>		0.999
<i>h</i> ²		0.58

Note: SOV; source of variation, Df; degrees of freedom, AN; anthocyanin content, σ_m^2 ; male variance, σ_f^2 ; female variance, σ_{mf}^2 ; male * female variance, *H*; broad-sense heritability, *h*²; narrow-sense heritability.

Abbreviations: BR, Baker's ratio; GPR; general predictable ratio.

****p* ≤ 0.01.

popular in food and medicine and are highly concentrated in black rice. Several studies [2, 5, 19], involving crosses between black and white rice, have been carried out to understand the genetics of desired traits. This hybridisation involving the incorporation of desired genes in rice has been reported to be one of the most important techniques to broaden the genetic base of the crop [20] and understand the genetics of the trait of interest. Therefore, this study aimed to determine the mode of gene action of anthocyanin in rice germplasm. This information will help in the selection of suitable breeding methods for improving anthocyanin content in rice.

4.1 | Estimates of Genetic Components for Anthocyanin Content

The ANOVA revealed significant variation for GCA of a trait only if the female and/or male parents had a statistically significant effect, while SCA was considered significant if the parents had a statistically significant female × male interaction for the trait [21]. In this study, all the GCA males, GCA females and SCA crosses for anthocyanin content revealed statistically significant differences, suggesting the presence of both additive and non-additive gene effects in the expression of the trait. This suggestion is in support of several findings [22, 23]. The significance of the SCA mean squares called for the need to know how important the interactions were in determining the performance of single-cross progeny, as suggested by Baker [17]. According to Griffing [24], the best way to determine progeny performance was to use variance components other than the mean squares of the SCA and GCA.

The variance components revealed a larger female GCA variance than the male GCA variance, indicating the possible presence of maternal effects in anthocyanin content, and Khamphasan et al. [4] reported similar findings in their study materials.

TABLE 3 | Means of anthocyanin content, general combining ability effects and specific combining ability effects of parents and crosses generated at the National Crops Resource Research Institute.

Parents	Mean AN (mg/100 g)	GCA	Rank
<i>Male genotype</i>			
BR-11	243	19.37***	1
BR-12	301.8	7.21***	2
BR-15	273.9	0.18	3
BR-14	308.9	−26.75***	4
<i>Female genotype</i>			
NR-4	—	35.9***	1
NR-1	—	28.51***	2
OKILE	—	3.55***	3
NR-2	—	−26.97***	4
NR-3	—	−40.98***	
Crosses		SCA	Rank
OKILE/BR-11	195.37	51.81***	1
NR-4/BR-15	186.13	29.38***	2
NR-1/BR-15	185.87	36.52***	3
NR-4/BR-12	177.33	13.58***	4
NR-2/BR-12	127.97	27.11***	5
NR-1/BR-14	127.07	4.67***	6
NR-2/BR-11	124.83	11.77***	7
NR-3/BR-11	103.63	4.58***	8
NR-3/BR-12	103.3	16.4***	9
NR-1/BR-11	140.77	−27.76***	10
NR-1/BR-12	142.93	−13.43***	11
NR-2/BR-14	59.37	−7.57***	12
NR-2/BR-15	62.53	−31.32***	13
NR-3/BR-14	63.03	10.12***	14
NR-3/BR-15	48.73	−31.09***	15
NR-4/BR-11	135.53	−40.41***	16
NR-4/BR-14	127.2	−2.55***	17
OKILE/BR-12	87.73	−43.66***	18
OKILE/BR-14	92.73	−4.69***	19
OKILE/BR-15	120.9	−3.48***	20
Means	120.65		
LSD	1.27		
CV	0.64		

Note: LSD; least significant difference at 5%, —; did not contain anthocyanin.

Abbreviations: AN, anthocyanin content; CV, coefficient of variation; GCA, general combining ability; SCA, specific combining ability.

Anthocyanin content had a lower SCA variance than the GCA variance, and its GPR was more than unity, consistent with a substantial role for additive gene action in anthocyanin content. Therefore, genes controlling anthocyanin content are additive and fixable, making it possible for them to be carried on to the next generation and improve for anthocyanin content using pure line selection or pedigree breeding. This study disagreed with the findings of Jaksomsak et al. [14], who reported a nonadditive type of gene action for anthocyanin content among their materials.

This discrepancy could be attributed to differences in germplasm genetic backgrounds, population diversity and experimental conditions between the two studies.

Regarding heritability estimates in the broad sense, the findings indicated a very high heritability (> 90%), suggesting that the environment minimally influenced anthocyanin content. Therefore, direct phenotypic selection for anthocyanin content proves highly effective and accelerates the breeding cycle. These broad-sense heritability results align with the classification by Singh [25], who considered heritability values above 80% to be very high. In a narrow-sense heritability exceeded 50%, indicating high heritability, consistent with Khamphasan et al. [4] and Singh's [25] classification of narrow-sense heritability values greater than 50% as high. A high narrow-sense heritability and a BR (> 0.5) suggested the additive inheritance among crosses for anthocyanin content under the Namulonge environment, implying that progeny performance could be predicted from that of parents and confirming potential for selection in early generations [27]. Because the experiment was conducted at a single location, the estimates are considered environment specific.

4.2 | Evaluation of Parents Based on GCA Effects

Combining ability defines the pattern of gene effects in the expression of quantitative traits by identifying potentially superior parents and crosses and developing appropriate breeding processes for further enhancement of various traits [5, 22]. GCA was defined as the average performance of a genotype in a series of hybrid combinations by Sprague and Tatum [28] and is attributed to additive gene effects and additive $\times$ additive epistasis and is theoretically fixable [5]. Parents with higher positive significant GCA effects were considered better combiners, while those with negative significant GCA effects were generally considered poor combiners, as reported by Jaksomsak et al. [14].

Genotypes BR-11, BR-12, NR-1, NR-4 and OKILE were good combiners and therefore considered desirable, while BR-14, NR-2 and NR-3 combined poorly for anthocyanin content and were undesirable. This study supported the findings of Jaksomsak et al. [14], who reported both good and poor combiners among their study materials for anthocyanin content. Several researchers [2, 5, 27] also reported a variation in the GCA effects among their studied materials for anthocyanin content and other traits.

4.3 | Evaluation of Parents Based on SCA Effects

SCA effects are used in plant breeding to describe the performance of the crosses relative to the averaged performance of crosses in an experiment [28]. SCA was used in identifying the best crosses for traits of interest and is related to nonadditive gene effects such as dominance and epistasis [5]. The crosses that showed high and significant SCA effects may be valuable in a breeding programme, and, in rice, hybridisation has not yet been explored on a large scale, but with advancements in technology, rice hybridisation might become necessary. However, Khamphasan [4] showed that SCA effects should be used together with mean performance and GCA of parents to identify good-performing crosses. During selections, at least one of the parents with a high GCA should be considered for favourable allele combinations. According to Baker [17], high and positive SCA effects can arise from different GCA combinations, but their

genetic implications differ. Crosses involving one parent with high GCA and another with low GCA suggest that additive effects from the good general combiner can compensate for or overcome the poorer combining ability of the other parent, resulting in favourable SCA. On the other hand, crosses where both parents possess high GCA indicate strong additive $\times$ additive interactions, which are heritable and fixable through selection. While low $\times$ low GCA combinations typically reflect dominance or nonadditive gene action, which is less amenable to fixation. Therefore, prioritising parents with high GCA increases the chances of capturing favourable additive variation while still benefiting from positive SCA in certain crosses. Several researchers have also reported this [2, 29]. This study revealed good-performing crosses (NR-2/BR-11, NR-3/BR-11, OKILE/BR-11, NR-2/BR12, NR-3/BR-12, NR-4/BR-12, NR1/BR-14, NR1/BR-15 and NR-4/BR-15) based on SCA, mean performance and at least one of the parents to have high GCA effects for anthocyanin content, and they can therefore be advanced and selection can start at an early generation. This suggestion is in support of Khamphasan [4], which states that if one of the parents has a high significant positive GCA for the desired trait, with a highly significant positive SCA, selection can be earlier as the trait is easily fixed.

4.4 | Limitation of the study

The present study was conducted at a single location, which does not provide conclusive results on the genetic action of anthocyanin content, and therefore, results from this study can be considered environment-specific. The study also did not include reciprocal crosses, which limited information on possible maternal contributions.

5 | Conclusion

For healthier populations, there is a need to improve foods for their therapeutic value to boost the population's immunity through daily intakes. BR-11 and BR-12 could be exploited via hybridisation programmes to improve therapeutic value in white rice, as they had a highly significant GCA effect. Crosses NR-2/BR-11, NR-3/BR-11, OKILE/BR-11, NR-2/BR12, NR-3/BR-12, NR-4/BR-12, NR1/BR-14, NR1/BR-15 and NR-4/BR-15 were the best performers for anthocyanin content based on their SCA effect, mean performance and GCA effects of the parents and can therefore be advanced to later generations for inbred development. The study revealed that anthocyanin content is controlled by an additive type of gene action under the Namulonge environment, making it possible for genes controlling the trait to be carried onto the next generation. Breeders will use this information to select the appropriate breeding strategies for anthocyanin content improvement in rice germplasm within Uganda. There is a need for future work to evaluate the promising parents and crosses across multiple environments and seasons to validate additive components controlling anthocyanin content in rice germplasm and also to perform reciprocal crosses on a representative subset to test for maternal effects.

Author Contributions

Conceptualization, Nashim Nassanga, Tonny Obua and Emmanuel Amponsah Adjei; methodology, Nashim Nassanga; formal analysis,

Nashim Nassanga; investigation, Arnold Katungisa; laboratory analysis, Nashim Nassanga; data curation, Nashim Nassanga; writing—original draft preparation, Nashim Nassanga; writing—review and editing, Tonny Obua, Jimmy Lamo, Emmanuel Amponsah Adjei; supervision, Emmanuel Amponsah Adjei, Tonny Obua and Jimmy Lamo.

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Disclosure

All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

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