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Scientific Article

Cooking quality and protein content of black-grain cowpea genotypes before and after cooking

Qualidade de cocção e teor de proteínas de feijão-caupi da classe preto antes e pós-cozimento

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ABSTRACT - The consumption of cowpea in the population's diet is an option to complement the intake of various nutrients, mainly proteins, and its black grain has great potential in the preparation of feijoada, as a more economical alternative to common bean for the Northeastern consumer. The objective of this work was to evaluate the cooking quality and protein content in black class cowpea genotypes, before and after cooking. Fifteen genotypes were evaluated, being 12 lines and 3 commercial cultivars. Cooking quality was evaluated using the percentage of cooked grains (PCG) and the protein content in raw and cooked grains. Data were expressed as mean ± standard deviation, with means compared by Student's t-test and grouped by Scott-Knott test ($p<0.05$). With regard to the cooking quality, a variation of the PCG between 31% and 87% was observed, with an overall mean of 71.07%. Protein content ranged from 23.35 to 29.80 g 100 g^{-1} in raw grains and from 24.72 to 33.70 g 100 g^{-1} in cooked grains. The cowpea genotypes evaluated in the present study showed high culinary quality and high protein content in raw and cooked grains. Thermal processing increased the protein content in the grain. Among the evaluated genotypes, the lines MNC10-982B-3-7 and MNC10-998B-20-3 show better cooking quality and higher post-cooking protein content. These genotypes meet consumer needs in terms of practicality and economy in grain preparation, constituting excellent options for the black cowpea market.

Keywords: *Vigna unguiculata.* Nutrition quality. Thermal processing. Fast cooking.

RESUMO - O consumo do feijão-caupi na dieta da população é uma opção para complementar a ingestão de vários nutrientes, principalmente proteínas, e o seu grão preto apresenta grande potencial no preparo da feijoada, como alternativa mais econômica que do feijão-comum para o consumidor nordestino. O objetivo deste trabalho foi avaliar a qualidade de cocção e o teor de proteínas em genótipos de feijão-caupi da classe preto, antes e após o cozimento. Foram avaliados 15 genótipos, sendo 12 linhagens e 3 cultivares comerciais. Avaliou-se a qualidade de cozimento por meio da percentagem de grãos cozidos (PCG) e o teor de proteínas nos grãos crus e cozidos. Os dados foram expressos como média ± desvio-padrão, com médias comparados pelos testes t de *Student* e agrupadas pelo teste Scott-Knott ($p<0,05$). Com relação à qualidade de cocção, observou-se variação da PCG entre 31% e 87%, com média geral de 71,07%. O teor de proteínas variou de 23,35 a 29,80 g 100 g^{-1} nos grãos crus e de 24,72 a 33,70 g 100 g^{-1} nos grãos cozidos. Os genótipos de feijão-caupi avaliados no presente estudo apresentaram alta qualidade culinária e alto teor de proteína nos grãos crus e cozidos. O processamento térmico aumentou o teor de proteína no grão. Dentre os genótipos avaliados, as linhagens MNC10-982B-3-7 e MNC10-998B-20-3 apresentam melhor qualidade de cozimento e maior teor de proteína pós-cozimento. Esses genótipos atendem às necessidades do consumidor em termos de praticidade e economia no preparo do grão, constituindo-se em excelentes opções para o mercado de feijão-caupi do tipo preto.

Palavras-chave: *Vigna unguiculata*. Qualidade nutricional. Processamento térmico. Cozimento rápido.

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INTRODUCTION

Cowpea (*Vigna unguiculata* L. Walp.) is a legume of great socioeconomic importance in the Northeast region of Brazil, generating jobs and income and contributing to food security for thousands of people. Its grains are sources of protein, carbohydrates, vitamins, fiber and minerals and can be sold as dry grains (main market), pods, immature grains (green beans) and flour (GOMES et al., 2021).

The cultivation of cowpea in Brazil is predominant in the Northeast region, with a cultivated area of 1.5 million hectares and a production of 416.7 thousand tons in the 2022/2023 harvest (BRASIL, 2023). In these regions, production is carried out both by medium and large producers and by small family farmers who still use traditional practices. In the Central-West region, production comes from medium and large businessmen, who use high technology in cultivation, which contributes to high levels of grain yield (ROCHA; DAMASCENO-SILVA; MENEZES-JÚNIOR, 2017).

The high protein and carbohydrate content, the relatively low lipid content, and the amino acid profile complementary to common cereal grains make cowpea an important nutritional food in the human diet (JAYATHILAKE et al., 2018). The thermal processing of the grain is necessary for consumption, and several studies have been carried out to identify genotypes with high cooking quality, as well as forms of thermal processing that cause less post-cooking loss of nutrients (PEREIRA et al., 2014; BARROS; ROCHA; MOREIRA-ARAÚJO, 2019;

VIEIRA; BEZERRA; SANTOS, 2021).

The cooking of bean grains is a hydrothermal process that results in starch gelatinization, protein denaturation and solubilization of part of the polysaccharides present in the cell wall of the grains (SHIGA; CORDENUNSI; LAJOLO, 2009). Studies have shown that the cooking process significantly reduces antinutritional factors and, in some cases, completely eliminate them, with wet heat treatment being the most effective. The effects of heat treatment on these compounds in cowpea grain may vary according to cultivar, antinutrient content, type of processing and heat exposure time (CARVALHO et al., 2023).

Cowpea cooking time is positively linked to the amount of water absorbed before the cooking process itself. The complexes formed by polysaccharides, phenolic compounds, and proteins are altered during the maceration step, favoring the entry of water and consequently interfering with the cooking time (SHIGA; CORDENUNSI; LAJOLO, 2009).

Black beans, in general, are an important food in Brazilian cuisine, being used mainly in the preparation of feijoada, which is a widespread dish in all regions of the country. However, the bean of this commercial class that has been most used for this purpose by the Northeast has been of the *P. vulgaris* species, produced in the South and Southeast regions, reaching the Northeast at higher prices. In the Brazilian market, only two black cowpea cultivars were registered in the RNC/MAPA and released, BRS Tapaihum (SANTOS, 2011) and BRS Guirá (FREIRE FILHO et al., 2023). However, both cultivars have a restricted recommendation, the first to the Juazeiro/Petrolina region (BA, PE, and PI) and the second to the state of Pará.

The black commercial class of cowpea is part of the special grain market, but there is a lack of cultivars and poor characterization of the grain in terms of nutritional quality and cooking. Therefore, this work aimed to evaluate the cooking quality and determine the protein content of cowpea genotypes before and after cooking.

MATERIAL AND METHODS

Step I - Obtaining the samples

Black grain samples of cowpea genotypes were obtained from the Work Collection of the Cowpea Genetic Breeding Program of Embrapa Meio-Norte, in Teresina, Piauí, Brazil, from a cultivation carried out under field conditions and under irrigation in the year 2021. Fifteen genotypes were evaluated, comprising 12 elite lines and three commercial cultivars (Pretinho, BRS Tapahium and BRS Guirá), as shown in Table 1.

Table 1. Name, genealogy and commercial subclass of the black commercial class cowpea genotypes studied.

Name/Code	Genealogy	Commercial subclass
MNC08-937C-6-1	(Pretinho x TE97-309G-9) x Pretinho	Opaque black
MNC09-981B-1	{[BRS Guariba x (Pretinho x TE97-309G-9)] x BRS Guariba	Slightly shiny black
MNC09-981B-3	{[BRS Guariba x (Pretinho x TE97-309G-9)] x BRS Guariba	Slightly shiny black
MNC09-981B-6	{[BRS Guariba x (Pretinho x TE97-309G-9)] x Guariba} x BRS Guariba	Slightly shiny black
MNC09-981B-9	{[BRS Guariba x (Pretinho x TE97-309G-9)] x Guariba} x BRS Guariba	Slightly shiny black
MNC09-988B-3	IT85F-1045 x MNC01-689F-11	Opaque black
MNC09-988-1B-3-20	IT85F-1045 x MNC01-689F-11	Opaque black
MNC09-988B-20	IT85F-1045 x MNC01-689F-11	Opaque black
MNC10-982B-3-7	{[BRS Guariba x (Pretinho x TE97-309G-9)] x Pretinho} x Pretinho	Slightly shiny black
MNC10-982B-11-1	{[BRS Guariba x (Pretinho x TE97-309G-9)] x Pretinho} x Pretinho	Slightly shiny black
MNC10-998B-8-1	F_3RC_1[Pretinho x TE97-309G-9) x Pretinho] x Pretinho	Slightly shiny black
MNC10-998B-20-3	F_3RC_1[Pretinho x TE97-309G-9) x Pretinho] x Pretinho	Slightly shiny black
Pretinho	Pretinho - Pará	Shiny black
BRS Tapahium	Epace 11 x 293588	Shiny black
BRS Guirá	{[BRS Guariba x (Pretinho x TE97-309G-9)] x Guariba} x BRS Guariba	Slightly shiny black

Cooking and protein content analyses were carried out at the Laboratory of Bromatology at Embrapa Meio-Norte in the year 2022.

Step II - Preparation of samples for cooking quality

Cooking quality was evaluated using the methodology proposed by Carvalho et al. (2017), with adaptations for cowpea grains made by Barros (2019) and Freitas et al. (2022). Two samples with 50 grains of each genotype without mechanical damage were placed in organza bags and identified. Two bags per genotype were used, so the analysis was performed in duplicate. The bags were individually placed in a beaker with distilled water for 60 minutes for the soaking step.

For cooking, the bags were placed at the bottom of an electric pressure cooker (Electrolux®). The water level was checked in such a way that the bags were completely soaked (2/5 of the container’s capacity). The samples were cooked one at a time, in order to avoid interaction between the genotypes, for 30 minutes, being removed immediately for cooling after the completion of the process. The soaking and cooking time were predetermined in preliminary tests carried out by Barros (2019).

The evaluation of the percentage of cooked grains was carried out using a Mattson cooker, after the cooking stage. A total of 25 grains were used per sample genotype, randomly chosen. The plungers were placed over the grains for immediate perforation verification. The perforated grains were counted and expressed as a percentage

Step III - Preparation of samples for determination of the protein content

The raw grain samples were washed with distilled water to remove dirt, placed in paper bags and taken to a drying oven with air circulation at 60 ºC for 48 hours. The dried grains were crushed in a zirconium ball mill to obtain flours. The flours were placed in polypropylene test tubes duly closed with a lid, identified, and kept under refrigeration temperature at 4 ºC.

Before the cooking process, the grains remained in the soaking step in deionized water for 1 hour. The cooking of samples of cowpea genotypes was carried out in an electric pressure cooker (Electrolux®) for 30 minutes, using the capacity of 2/5 of the cooker for the amount of water used in the process, considering the soaking water that was used. After completion, the samples were transferred to an air circulation drying oven at 60 °C for 48 hours. After drying, the samples were transformed into flour with the aid of a zirconium ball mill and placed in properly closed test tubes and stored under refrigeration temperature for further analysis of the protein content.

The determination of proteins was carried out by the macro Kjeldahl method, based on three steps: digestion, distillation, and titration, where the organic matter is decomposed, and the existing nitrogen is transformed into ammonia. The 6.25 factor was used to convert the total nitrogen content into proteins (AOAC, 2012).

About 0.2 g of the sample was weighed on parchment paper and then transferred to digestion tubes. 2 g of catalytic mixture (96% potassium sulfate and 4% copper sulfate) and 5 mL of concentrated sulfuric acid were added, then heated in a digester block at a temperature of 430 ºC for 1h40, until the solution became greenish clear, free of undigested material. After cooling the tubes, 10 mL of distilled water was added. The tube was attached to the distiller. In an Erlenmeyer® flask, 10 mL of boric acid was added as an indicator and inserted into the equipment to collect the distillate.

In the equipment, 15 mL of 50% sodium hydroxide solution was added to the tube with the sample until a slight excess of base was guaranteed. After boiling, distillation was performed until obtaining 100 mL of the distillate. After this step, the distillate was titrated with 0.02 N hydrochloric acid. The protein content was calculated by the formula:

$$\text{Protein content} = V \times 0.14 \times F/P$$

where V is the volume of sulfuric acid used minus the volume of sodium hydroxide used in the titration, F is the conversion factor for vegetable protein (6.25), and P is the sample weight.

Statistical analyses

The data obtained were entered into Excel spreadsheets and then statistical analyses were performed. Analyses of variance were performed for all characteristics, and means were compared by Student's t test ($p<0.05$) to verify the existence of differences between two means (comparison between samples of raw and cooked grains by genotype) and grouped by the Scott-Knott test ($p<0.05$) to verify the existence of differences between three or more samples (groups of genotypes), with the aid of the GENES program (CRUZ, 2016).

RESULTS AND DISCUSSION

Cooking quality

The percentage of cooked grains (PCG) of 15 black commercial class cowpea genotypes in descending order is shown in Table 2. There was a significant difference ($p<0.05$) between the genotypes evaluated for PCG. The Scott-Knott test grouped the means of the genotypes into six groups (a, b, c, d, e, f), with group “a” concentrating the genotypes with the highest PCG, with emphasis on MNC10-982B-3-7, Pretinho and MNC09-988B-20, with 87%, 86% and 85%, respectively. Among the fifteen evaluated genotypes, fourteen of them showed good cooking quality, with PCG above 52% of grain perforation, standing out as promising genotypes for the development and commercialization of cultivars with good attributes related to consumption. According to Mattson (1946), when 13 of the 25 (52%) plungers pierce the grains, the samples are considered cooked.

The results found for the cultivars Pretinho (86%) and BRS Tapaihum (73%) were superior to those found by Barros (2019), who, evaluating the cooking quality of these two cultivars, reported PCG of 59 and 39%, respectively, using the same cooking time. These differences observed for the cooking quality between these cultivars in the different studies may be due to differences in the post-harvest time of the grains, the quality of storage before analysis and the positioning of the samples in the electric pressure cooker.

On the other hand, the MNC08-937C-6-1 genotype exhibited the lowest PCG (31%), thus showing poor cooking quality. This indicates that this genotype will spend more time and energy (gas, firewood or electricity) for cooking, which is undesirable for the consumer. Addy et al. (2020), when studying the genetic heritage of cooking time of cowpea in Africa, stated that long periods of cooking for cowpea lead to loss of nutrients, loss of useful time and increased emission of greenhouse gases by the increase in wood burning.

When 13 of the 25 Mattson cooker plungers, i.e., 52%, pierce the grains, the sample is considered cooked. Thus, the procedure used in this study allowed evaluating the cooking quality of a satisfactory number of genotypes in an optimized time. Carvalho et al. (2017) evaluated 252 common bean genotypes of the carioca type and observed a mean percentage of cooked grains of 36.71%, below the mean obtained in the present work. This striking difference between the results may be due to the species, the genotype, the time of immersion in water before cooking, the average cooking time and the capacity of the electric pressure cooker used.

The overall mean of the PCG of the cowpea genotypes evaluated in this study was 71.07%, above the mean observed by Freitas et al. (2022), who, using the same methodology of this study, evaluated 100 cowpea genotypes of the commercial color class and observed a mean of 68.7% of PCG, lower than that found in this study, but still higher than the reference percentage for the classification of cooked grain

(52%) according to the Mattson method, indicating that, in general, the black commercial class cowpea genotypes studied had good cooking quality. The overall mean of PCG observed in the present study was more similar to that obtained by Oliveira et al. (2023), when evaluating three cowpea cultivars, observed an overall mean of 75.98% for PCG.

The soaking step was essential for cooking the grains, as immersion in water facilitates starch reactions, helping to soften the grains, reducing cooking time and, consequently, energy costs during cowpea grain cooking.

Silva et al. (2017), when studying 24 cowpea genotypes, observed that the cooking time of the grain depends on the genotype. The genetic variability of this characteristic has its cause evidenced in the various physical and chemical factors that differ between the various types of seeds, such as thickness and composition of the seed coat, as well as the composition of the cotyledon (WAINAINA et al., 2021).

The thickness and appearance of the cowpea seed coat can influence the water absorption rate, which is closely associated with cooking time, a fact that may partly explain the low cooking quality of the genotype MNC08-937C-6-1. According to Smýkal et al. (2014), the complexity of water penetration in grains with darker coats is due to the thicker coating as a consequence of the deposition of polyphenols that supposedly play a role in the permeability of the seed coat.

Hamid et al. (2016) compared red and black seed coat cowpea grains and observed that black grains had lower water absorption rate and cooking time when compared to red seed coat grains. The differential behavior in relation to the cooking quality shown by the black grain genotypes evaluated in the present study indicates that the physical characteristics of the seed coat (thickness, appearance, porosity, water absorption capacity and others) may vary even within a same business class.

There are several protocols to evaluate the cooking of cowpea beans and there are no specific guidelines for a given commercial class or variety. According to Wainaina et al. (2021), in the absence of a validated cooking procedure, the determinant for choosing an appropriate preparation method is accessibility and convenience linked to time and energy costs.

Table 2. Means and standard deviation (SD) for the percentage of cooked grains (PCG) of 15 cowpea genotypes of black commercial class.

Genotype	PCG (%) Mean ± SD
MNC10-982B-3-7	87 ± 1.41a
Pretinho	86 ± 2.82a
MNC09-988B-20	85 ± 4.24a
MNC09-988B-3	82 ± 2.82b
MNC09-981B-9	81 ± 4.24b
MNC09-988-1B-3-20	78 ± 2.82b
MNC09-981B-3	77 ± 1.41b
BRS Tapahium	73 ± 4.24c
MNC10-998B-20-3	71 ± 1.41c
BRS Guirá	69 ± 1.41c
MNC09-981B-6	65 ± 4.24d
MNC10-982B-11-1	65 ± 1.41d
MNC10-998B-8-1	63 ± 1.41d
MNC09-981B-1	53 ± 1.41e
MNC08-937C-6-1	31 ± 1.41f
Overall mean	71.07

Means with equal letters in the same column do not differ significantly by the Scott-Knott test ($p<0.05$).

Protein content

The means and respective standard deviations of protein contents in raw and cooked grains of commercial black cowpea genotypes are shown in Table 3.

In raw grains, protein content ranged from 23.35 (BRS Tapahium) to 29.80 g 100 g^{-1} (MNC09-988-1B-3-20), with an overall mean of 27.42 g 100 g^{-1} (Table 3). In the cooked grains, the protein content varied from 24.72 (BRS Tapahium) to 33.70 g 100 g^{-1} (MNC10-998B-20-3), with an overall mean of 29.15 g 100 g^{-1}. The lines MNC09-988-1B-3-20 and MNC10-981B-20-3 had the highest protein contents in the raw and cooked grains, respectively. Protein contents lower than those of the present study were observed in evaluations of cowpea cultivars carried out by Rios et al. (2018), who found variation of 21.73 to 25.77 g 100 g^{-1}.

The protein contents found for the cultivars Pretinho (27.51 g 100 g^{-1}), BRS Tapaihum (23.35 g 100 g^{-1}) and BRS Guirá (25.35 g 100 g^{-1}) were lower than those found by Barros (2019), who evaluated the cooking quality of the cultivars Pretinho and BRS Tapaihum, and Freire Filho et al. (2023), who evaluated the cultivar BRS Guirá and reported protein contents of 29.83, 25.12 and 29.43 g 100 g^{-1}, respectively. These differences in protein content observed in different studies may be due to differences in the field management, post-harvest time of grains and quality of storage of the grain before analysis.

Table 3. Means and standard deviation (SD) for protein contents in raw and cooked grains of 15 cowpea genotypes of black commercial class, analyzed on a dry basis.

Genotype	Protein content (g 100 g^{-1})	
	Raw grain Mean ± SD	Cooked grain Mean ± SD
MNC08-937C-6-1	26.58 ± 0.32b E	28.48 ± 0.23a E
MNC09-981B-1	26.29 ± 0.82b E	28.32 ± 0.20a E
MNC09-981B-3	27.70 ± 0.17b D	28.50 ± 0.45a E
MNC09-981B-6	28.32 ± 0.42b C	29.10 ± 0.16a D
MNC09-981B-9	28.35 ± 0.08b C	29.53 ± 0.28a D
MNC09-988B-3	29.27 ± 0.13a B	29.46 ± 0.11a D
MNC09-988-1B-3-20	29.80 ± 0.13b A	31.66 ± 0.27a B
MNC09-988B-20	28.81 ± 0.27b B	30.34 ± 0.31a C
MNC10-982B-3-7	23.98 ± 0.26b G	27.15 ± 0.39a G
MNC10-982B-11-1	29.34 ± 0.19b B	31.34 ± 0.21a B
MNC10-998B-8-1	26.38 ± 0.12b E	28.06 ± 0.31a E
MNC10-998B-20-3	29.75 ± 0.20b A	33.70 ± 0.01a A
Pretinho	27.51 ± 0.32b D	29.23 ± 0.31a D
BRS Tapahium	23.35 ± 0.33b H	24.72 ± 0.41a H
BRS Guirá	25.83 ± 0.30b F	27.72 ± 0.20a F
Overall mean	27.42 b	29.15 a

Data are presented as mean of three replicates ± standard deviation. Means with equal lowercase letters in the same row and equal capital letters in the same column do not differ significantly by Student's *t* and Scott-Knott tests, respectively ($p<0.05$).

A significant difference ($p<0.05$) was observed between genotypes for protein contents in both raw and cooked grains (Table 3), showing that the evaluated cowpea genotypes, although belonging to the same commercial class and many sharing the same genealogy (Table 1), show genetic divergence. According to Haider et al. (2018) and Oliveira et al. (2023), in addition to the genotype, the chemical composition of the grains depends on environmental conditions, soil properties and cultivation practices, which may also explain the variation between the protein contents obtained in the present study.

Protein contents showed a significant difference ($p<0.05$) between raw and cooked grains. This statistical difference was also verified by Melo et al. (2017), who analyzed the influence of cooking on the centesimal composition of immature grains in four cowpea cultivars and observed that the BRS Tumucumaque cowpea cultivar obtained protein content of 10.05 g 100 g^{-1} in the raw grains, while after cooking this content increased to 11.45 g 100 g^{-1}.

On the other hand, some studies have reported a decrease in protein content after thermal processing. Bezerra et al. (2019), evaluating the chemical composition of eight cowpea cultivars, observed mean protein contents in raw and cooked grains of 32.29 and 9.19 g 100 g^{-1}, respectively. Affrifah, Phillips and Saalia (2021), reviewing the nutritional profile, processing methods and products of cowpea, observed a mean of 23.52 g 100 g^{-1} in raw grains and 7.73 g 100 g^{-1} in cooked grains.

Just as increases and losses in protein content after cooking were observed in the studies above in the total group of evaluated cowpea genotypes, the literature also reports cases in which there were increases, losses and similarities in the same group of genotypes in the contents of this nutrient after cooking. Silva et al. (2017), when evaluating 24 cowpea genotypes, observed that five genotypes showed similar protein contents in the grains, ten genotypes showed losses, and nine genotypes showed increases after cooking.

Thermal processes such as cooking applied to cowpea can cause physicochemical modifications in the proteins, starch, and other components of the grains and, therefore, affect the nutritive value. The increase in protein content after cooking the grains of the black-grain cowpea genotypes evaluated in this study can be attributed to thermal processing, because of humid heat that causes the denaturation of antinutritional factors of protein nature and, at the same time, prevents the exacerbated degradation of essential amino acids (RAMÍREZ-CÁRDENAS; LEONEL; COSTA, 2008).

The Normative Instruction nº 75, of October 8, 2020 by Anvisa, which establishes the technical requirements for declaring nutritional labeling on packaged foods (BRASIL, 2020), determines that the Recommended Daily Value (RDV) of protein is 50 grams per day^{-1}. Considering that the average protein contents in this study were 27.42 g 100 g^{-1} in raw grains and 29.15 g 100 g^{-1} in cooked grains, grains can be classified as "high protein content" because they have contents greater than 20% of the RDV (10 g). Therefore, the means correspond to 54.84% of the RDV in raw grains and 58.30% of the RDV in cooked grains. Freitas et al. (2022) and Oliveira et al. (2023), evaluating raw grains of cowpea genotypes, observed RDV percentages of 48.30% and 51.44%, respectively, which are lower than those found in the present study.

CONCLUSIONS

The black commercial cowpea genotypes evaluated in present study, in general, showed high cooking quality and high protein content in raw and cooked grains. Thermal processing increases protein content. Among the evaluated genotypes, the elite lines MNC10-982B-3-7 and MNC10-998B-20-3 show better cooking quality and higher post-cooking protein content. These genotypes, because they already have excellent agronomic characteristics, can be used as parents in breeding crosses or directly recommended as commercial cultivars, meeting consumer needs in terms of practicality and economy in grain preparation, constituting excellent options for the special grain cowpea market of the black type.

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