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Biometrics seeds, emergence and vigor of camu-camu seedlings depending on the seed coat coloring

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ABSTRACT

In the Brazilian Amazon is significant diversity of fruit species, the agronomic characteristics of which are still little studied. The objective of this study was to evaluate the influence of tegument coloration on germination and vigor of camu-camu seeds. The seeds were obtained from ripe and healthy fruits coming from population of the Urubu River in Roraima and were classified visually according to the tegument coloration of green, red and brown color. 200 seeds for each class of colors were utilized, evaluating the following biometric characters: length, breadth, thickness and individual mass, obtained with the aid of a digital caliper and precision balance. The experimental design utilized was the completely randomized with three treatments and 10 replications of 20 seeds. The highest means of mass and volume were obtained from brown and red seeds. The brown seeds produce higher content of root dry mass than both the green and red seeds. The physiological quality of camu-camu seeds of the Urubu River is associated with tegument coloration. Seeds with brown tegument have better development, greater growth of seedlings with greater accumulation of dry matter both in the roots and shoots.

Keywords

Myrciaria dubia; germination; seed thickness.

Academic Discipline And Sub-Disciplines

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INTRODUCTION

In the Brazilian Amazon is significant diversity of fruit species, relatively well known as far as the botanic aspects are concerned, but little studied concerning the agronomic characteristics. In that region, Cavalcante [8] catalogued 176 species with eatable fruits, a half of them represented by native fruit-fearing species. Nevertheless, the diversity is far greater, there being estimates, which point out that of the 50 Brazilian fruit-bearing species, about 44% have Amazonia as the center of origin [16]-[11]. The camu-camu (*Myrciaria dubia* (Kunth) McVaugh) is a fruit-bearing species of the Amazonian region, its being recently distributed to several Brazilian states for small commercial plantings [11], aiming at the exploration of the high natural source of vitamin C [9] and another componds [24] that its fruit presents.

The fruit is globose berry, with diameter between 10 mm and 30 mm and weight around 10g, coloration ranging from red to purple when ripe, thin, bright skin, with content of up to 5% of ascorbic acid, juicy, plentiful, acidic pulp and pinky in color, having from one to four seeds, de forma flattened or kidney-shaped, recalcitrant, covered with a small layer of fibers and length 8 mm to 15 mm in diameter [2].

The biometrics of fruits and seeds constitutes an important subside to the distinction of species of a same genus and varieties of a same species and also is related with the characteristics of spread, establishment and vigor of seedling, in addition to being utilized to distinguish pioneering and non-pioneering species in tropical forests ([3]- [10]- [1]. Seeds of the family Myrtaceae, Fabaceae, Euphorbiaceae differentiate as to tegument coloration and this heteromorphy, is associated with the physiological quality, the difference in the coloration of the seeds may be related with maturation stage and therefore, the visual morphological index a great deal of times points out information about the physiological maturity [5].

The tegument color of the seed has influence upon the physiological quality and is associated with the maturation degree, the light-colored seeds being more immature [21] similar to the seeds of *Pothomorpha umbellata* in which the black seeds present higher speed and percentage of germination [22].

In this aspect, in some species belonging to the family Myrtaceae were found in *M. dubia* seeds with three colorations (brown, mixed and green), the ones of brown coloration expressed less percentage of germination but they gave off greater number of leaves [29]. In *Quararibea grandiflora* Mart., the dark-brown seeds were viable and the light brown ones unviable [14], while in *Q. cordata* Spreng., the dark seeds possessed no embryo and the light-colored seeds were viable [17].

During the germination process, alterations in the chemical composition of seeds take place and the consumption of the store substances, such as carbohydrates, lipids and proteins, which supply energy for the embryo's development. The speed of utilizing the reserves during germination ranges according to the species, shape, color, precedence and with environment [4].

Before the exposed, the objective in this work was to determine the influence of the coloration of the seed tegument upon germination and vigor of the seedlings of camu-camu (*Myrciaria dubia*) coming from the State of Roraima.

MATERIALS AND METHODS

The study was conducted at Embrapa Roraima, in the period of May to September of 2013 in the facilities of the Seed Analysis Laboratory and in ten seedling nursery of the Fruit culture sector localized on BR 174, Km 8, Industrial District, under geographical coordinates of reference 02º45'28"N and 60º43'54"W, 90m of altitude. Boa Vista is located in the Tropical Climate Zone, there not being either extremely dry season or average monthly temperature lower than 20 °C, according to Köppen its climate is tropical wet of the Aw type: rainy tropical climate, hot and wet, with rainy season in the summertime; the driest month presents rainfall inferior to 60 mm. The average rainfall is of 1,750 mm yearly, air temperature of 26.7 °C and relative humidity of air, 79% [20].

The seeds of camu-camu were obtained from completely ripe with tegument of the fruit skin purple in color, health of good quality, coming from the banks of the Urubu River in the state of Roraima. The fruits were pulped by hand; the seeds were washed in running water and placed to dry in the shade at room temperature for 24h. Next, the seeds were classified visually according to the tegument coloration, for in the camu-camu population of the Urubu river there was variation in the tegument coloration (green, red and brown). From these, 200 seeds were utilized for each class of colors, with further measure of the biometric characters: length (C), breadth (L) and thickness (E) of the seeds obtained with the aid of digital caliper (0.01 mm). The volume of the seed was determined by multiplying the dimensions: C x L x E as well as the individual mass of the seeds was quantified on a precision balance of 0.001g, with correction for moisture determined. The experimental design utilized was the completely randomized with three treatments and 10 replications of 20 seeds per plot. Next, the seeds were sown singly in polyethylene tubes containing as substrate ORG: Organoamazon® (Table 1).

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Table 1. Chemical analysis, micronutrients and particle size of the substrate utilized for the germination and early development of the camu-camu seedlings.

		Sorptive complex[2]											
Sub[1]	pH	Ca^{2+}	Mg^{2+}	K^{+}	Al^{3+}	H+Al	S	t	T	V	M	P	MO
		---------- $cmol_c dm^{-3}$ ----------								-- % ---		$mg\ dm^{-3}$	$g\ kg^{-1}$
ORG	5.8	10.5	7.9	1.6	-	2.08	20	20	22	90.6	-	176.77	69.2

	Micronutrients[3]						Particle size		
Sub	Zn	Fe	Mn	Cu	B	S	Clay	Silt	Sand
	---------- $mg\ dm^{-3}$ ----------						--------$g\ kg^{-1}$----------		
ORG	19.5	27.08	124	0.3	0.33	19	170	290	540

[1] Substrate: ORG: Organoamazon®. [2]pH in water (1:2.5); Ca^{2+}. Mg^{2+} e Al^{3+}: KCl extractor 1 mol L^{-1}; K^{+} and P: mehlich-1extractor; H+Al: extractor SMP; M.O.: organic matter – oxidation $Na_2Cr_2O_7$ 4N + H_2SO_4 10N; S: sumo f exchangeable base; t: effective cation exchangeable capacity (CTC); T: CTC at pH 7.0; V: base saturation index; m: Aluminum saturation index. [3]Zn, Fe, Mn and Cu mehlich-1: extractor; B: hot water extractor; S: monocalcium phosphate extractor in acetic acid.

Irrigation was conducted by scheduled sprinkling in the interior of greenhouse covered with plastic and screened sides, every five hours during the day, each irrigation having the duration of 5 minutes. So, at 20 days after sowing, the count of the germinated seeds was started daily till 40 days. With these data, the average time to obtain 50% of the total germination and the time for start of germination was determined, taking into account the first seedling to emerge. At the end of the experiment at 90 days, after the development of the seedlings, the following items were evaluated: height of the shoot taken from the collar to the plant tip (millimeter ruler) and the stem diameter, measured at 1 cm from soil (caliper), quantified the number of leaves (NF), measured the length of the root system (CSR), the same ones being put separately into identified paper bags, placed into oven at the temperature of 60 ºC for 72 hours, next weighted in semi-analytical balance to obtain the dry matter of the shoot (MSPA) and root dry mass of the root system (MSR).

The average results obtained in the variables under study were submitted to the normality test and test for homogeneity and next they were submitted to the variance analysis (ANOVA) and comparison of the means by the Tukey test at 5% of probability, by utilizing the statistic program Sisvar [15].

RESULTS

The greatest means of biometric measures for mass and volume were obtained with the seeds classified in the brown and red coloration. While the seeds of green coloration presented smaller biometric measures, except for the variable breadth (Table 2), where the average values presented no significant differences for the seeds with teguments of green, red and brown coloration. However, the greatest length was verified in the seeds with tegument of brown coloration in relation to the green ones.

Table 2. Average values of mass (g), length (cm), breadth (cm), thickness (mm) and volume (cm^3) of camu-camu seeds (*Myrciaria dubia*) collected in Roraima for different classes of tegument colors

Coloration	Mass	Length	Breadth	Thickness	Volume
Green	0.91 b	16.74 b	12.55 a	5.63 b	1192.02 b
Red	1.03 a	17.16 ab	12.73 a	6.08 a	1358.13 a
Brown	1.05 a	17.26 a	12.87 a	6.22 a	1365.35 a
CV%	18.71	5.35	7.13	8.13	16.27

Means followed by the same letter in the column do not differ from one another by the Tukey test (p>0.05).

It was found that the maximum emergence percentage of camu-camu seedlings took place in the seeds of brown coloration, but not differing significantly from the red-colored seeds, but superior to those of green coloration. The same was found for the start of emergence, emergence speed ad average time of emergence of the seedlings in which there was a significant superiority of the brown-colored seeds in relation to the others (Table 3).

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Table 3. Average values for early emergence (IEMG, days), seedling emergence (EMG, %), emergence velocity (VE, index) and average emergence time (TME, days) according to the coloration of the tegument of the camu-camu seeds (*Myrciaria dubia*) collected on the Urubu river in Roraima obtained at 50 days after sowing

Coloration	IEMG (days)	EMG (%)	VE (index)	TME (days)
Green	36 b	63.0 b	0.14 b	42 b
Red	36 b	75.0 a	0.13 b	42 b
Brown	28 a	79.0 a	0.16 a	37 a
CV%	10.06	8.53	9.13	5.87

Means followed by the same letter in the column do not differ from one another by the Tukey test (p>0.05).

In this work, it was found that camu-camu seedlings originated from brown-colored seeds are also more vigorous than those obtained from red and green seeds, according to what can be verified in the results of height (cm), diameter (mm) and number of leaves (Table 4).

Table 4. Average values of height (ALT), stem diameter (DC) and number of leaves (NF) obtained from camu-camu seedlings (*Myrciaria dubia*) at 90 days according to the coloration of the tegument of seeds collected on the Urubu river in Roraima

Coloration	ALT (cm)	DC (mm)	NF
Green	10.7 b	1.2 b	19.8 b
Red	9.7 b	1.2 b	18.8 b
Brown	15.8 a	1.6 a	27.1 a
CV%	10.01	6.10	7.27

Means followed by the same letter in the column do not differ from one another by the Tukey test (p>0.05).

The greatest content of dry mater of the shoot, rots and length of the root system in seedlings of *M. dubia* was obtained in the present study when they were originated from seeds with brown tegument and the smallest values occurred in seedlings originated from seeds green and red in color (Table 5).

Table 5. Average values of the root system length (CSR, cm), dry mass of the shoot (MSPA, g) and dry mass of the root system (MSSR, g) obtained from camu-camu seeedling (*Myrciaria dubia*) according to the coloration of the tegument of seeds collected on the Urubu river at 90 days after sowing

Coloration	CSR(cm)	MSPA(g)	MSSR(g)
Green	12.3 b	0.17 b	0.10 b
Red	12.1 b	0.14 b	0.07 b
Brown	12.9 a	0.37 a	0.15 a
CV%	4.90	9.26	17.80

Means followed by the same letter in the column do not differ from one another by the Tukey test (p>0.05).

Surveying the germination results, one can predict that the seeds of brown coloration had best development, which possibly allow them to accumulate increased amount of reserves, which were transferred to the growth of the seedling de forma a accumulate greater amount of dry mass of the shoot and dry mass of the rot system and length of the roots in relation to those obtained with the other seeds.

DISCUSSION

The coloration of the camu-camu seeds can be correlated with their physiological maturity, although works as that of Santos *et al.* [27] in studying the maturity of fruits and germination of seeds of six species of Myrtaceae in Southern Brazil, showed that that there is variability in the morphology of ripe seeds as to their coloration, shape and size.

Research results which relate the biometric studies and coloration of teguments of camu-camu seeds are scarce. Several authors verified the scarcity of biometric studies in native species, which could give support in programs for recovery of degraded area and genetic improvement due to the great variability in seed size, coloration of seed tegument and others

[10]- [12]- [19]-[13]- [28]. Thus, the results of biometric studies of fruits and seeds represent important subsides to the establishment of techniques of eatable fruits [26].

According to Marcos Filho [23], seeds of determined species, manifest the capacity of germinating even before the physiological maturity to be reached; but, in general, they are less vigorous for not having reached the maximum accumulation of reserve , like that found by the green and red seeds of this work.

The germination of the seeds of *M. dubia* is of the hypogeal type [30] and occurs between 30 and 120 days and reaches on average 60% of germination, in the present study means ranging from 63.0 to 79% of emergence of seedlings being observed (Table 3). Still, according to the same authors, the seeds of camu-camu green in color have greatest rate of germination as compared with seeds brown in color, divergent results were obtained in the present study, which can be explained by the genetic differences of the material of precedence, in addition to the differences in the conditions of conservation of the seeds, culminating in the smallest or greatest capacity of germination of the immature seeds. Carvalho and Nakagawa [7] justified that non-completely ripe seeds can germinate and further result into seedlings so vigorous as the ones which reached physiological maturation.

It was verified by Alves *et al.* [1] which brown-colored seeds of *Clitoria fairchildiana* (R.A. Howard) possess greater emergence rate of seedling compared with those of light colors, similar results were obtained in the seeds of the present study.

The coloration of the seeds of *Clitoria fairchildiana* influenced in a significant way the vigor of the seedlings, the greatest values of which were obtained with brow-colored seeds [1], which are corroborated by the results of the present study. Those results suggest that the brown-colored seeds of this population are at the physiological maturity point. So, the conduction of seed maturation studies is important in order to define with greatest precision how the relation between seed coloration and the characteristics of ripe fruits, dehiscence and the chronological age of the seeds of each coloration occurs.

According to Carneiro [6], increased values for the dry mass of the root are indicators of greatest percentage of survival in the field, since the presence of fibrous roots allows greatest capacity of them to keep growing and forming new roots, more active, enabling greatest tolerance under extreme conditions.

The dry mass of the shoot according to Gomes and Paiva [18] should be considered since it indicates the seedling's hardiness, the larger the harder it will be. Taking as basis this statement, it is possible for the seedlings coming from brown-colored seeds to be harder than the other seedlings produced. For C*litoria fairchildiana* brown-colored seeds, Alves *et al*. [1] obtained greater values for the variables dry mass of the shoot and dry mass of the root system, similar results were obtained in the present study.

Species of Myrtaceae as *Eugenia pyriformis* Cambess and *Eugenia involucrata* D.C. do not express in the seeds characteristics of the physiological maturation, but they can have as an indicative the epicarp coloration of fruits [25]. Though, it is believed that there is a relation between camu-camu seed coloration of the studied population with the development, being able to influence the germination index, further studies of the seeds of *Myrciaria dubia*, at different maturation stages of the fruit in order to identify the ideal harvest time for the utilization in works with improvement are suggested.

CONCLUSIONS

The physiological quality of camu-camu seeds of the Urubu river population is associated with the tegument coloration. Seeds with brown tegument have best early development, greatest growth of the seedlings with greatest accumulation of dry mass both in the roots and in the shoot.

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