RESEARCH ARTICLE Agricultural Science Digest, Volume 43 Issue 5: 604-609 (October 2023)

Seed Physiological Quality Testing Gambir (*Uncaria gambir roxb.*) at Various Ages of Harvest

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10.18805/ag.DF-538

ABSTRACT

Background: Gambier has a problem with low production levels which is caused by the use of inferior quality plants that come from inferior seeds. This study aims to determine the optimal harvesting age for producing gambir seeds with optimal physiological quality. The maturity phase of the fruit became the basis for determining the timescalation of the harvest that count from the day after anthesis. The research was carried out from June to December of 2021. This research was conducted at the Seed Technology Laboratory, Faculty of Agriculture, Andalas University, Padang. The plant material utilized is the seed of the Mancik Riau gambier.

Methods: The research employed a completely randomized design (CRD) with five treatments based on days after anthesis (DAA) because at this phase the plants pollinated and the fruit maturity phase begin and it used three replication : seeds harvested at 110-112 DAA, 107-109 DAA, 104-106 DAA, 101-103 DAA and 98-100 DAA. Quantitative data are analyzed at a 5% significance level using Analysis of Variance; if the effect is significant, the Duncan's multiple range test (MRT) is performed; and qualitative data are presented descriptively.

Result: The results revealed that the harvest age of 98-100 DAA provided the highest viability and vigor, as measured by germinated power testing at 80.50%, seed growth potential testing at 81.83%, T50 test, index value testing at 11.04 and soil emergence testing at 84.33%, indicating that the plant had reached physiological maturity.

Key words: Characteristics, Gambir, Seeds, Sprouts, Transport.

INTRODUCTION

Gambir is one of the most widely cultivated plantation crops in Indonesia. It has a high economic value due to its numerous industrial applications. However, its production rate remains low due to fewer seedlings of high quality. The high-quality seeds also originate from good seeds. One way to obtain seeds of high quality is to harvest Gambir at the optimal time, when physiological maturation occurs. It is necessary to harvest at the optimal time to prevent the collection of immature seeds or the loss of crop yields due to broken fruit (Brenna *et al*., 2019).

To comprehend the timing of seed harvesting, knowledge of the duration of flower and fruit development is essential. According to research conducted by Jamsari *et al*. (2007) on Riau type gambir, there are five phase in flower and fruit development, namely flower initiation, small bud cale, large bud scale, anthesis/flower opening and fruit development. Fruit development began with the fall of the flower crown (final stage / open flower stage). Large bud scale and anthesis phase each took place in 5 days and fruit development stage complete in 53 days. It is estimated that the fruit of the Anthesis phase or open flower (F3) matures in 58 days, given that the initiation to anthesis lasts approximately 52 days. The average duration of fruit maturation is 112 days, ranging from 107 to 119 days. According to Udarno and Setiyono, (2013), the duration of udang and cubadak is 116 days. This study aims to determine the optimal harvesting age for producing gambir seeds with optimal physiological quality. The objective of this study is to determine the optimal harvesting age for producing gambir seeds with optimal physiological quality.

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How to cite this article: Zainal, A., Monaliatrisna, Kasim, M., Gustian, Warnita and Yunita, R. (2023). Seed Physiological Quality Testing Gambir (*Uncaria gambir roxb.*) at Various Ages of Harvest. Agricultural Science Digest. 43(5): 604-609. doi: 10.18805/ag.DF-538.

Submitted: 06-02-2023 **Accepted:** 08-07-2023 **Online:** 08-08-2023

MATERIALS AND METHODS

From June to December 2021, this research was conducted at the Seed Technology Laboratory, Faculty of Agriculture, Andalas University in Padang, Indonesia. The plant material utilized is the Riau mancik gambir seed that collected from the tree. The seedlings were grown on Whatman qualitative filter paper No. 1 in sterile ultisol soil (pore size: 11 m; material: cellulose).

The study used a complete randomized design (CRD) with five treatments as follow:
A: Seeds harvested at the age 110-112 DAA.
B: Seeds harvested at the age 107-109 DAA.
C: Seeds harvested at the age 104-106 DAA.
D: Seeds harvested at the age 101-103 DAA.
E: Seeds harvested at the age 98-100 DAA.

Seeds were sown on a medium of filter paper and ultisol soil, 200 seeds each/ repeats. Each treatment has three repeats so that the number of seeds for the filter paper medium was and the ultisol soil was 3000 seeds each. So, the total of all seeds used were 6000 seeds.

Per experimental unit, 200 seeds were planted on filter paper and soil in laboratory The F test was used to analyze the data statistically; if there is a significant difference, the Duncan test at a significance level of 5% is performed, followed by analysis using the STAR application. The labeling of interest rates commences with those of progressively older ages, 112 DAA for the oldest treatment and 98 DAA for the youngest. The fruit is harvested 112 days after the first flower has been marked. The fruit stalks are gathered by severing them and placing them in seeding containers. The diameter of the fruit, the length of the capsule, the color and shape of the fruit were measured as parameters. Then, place it in an envelope and expose it to two hours of direct sunlight before storing it in a dry location. Additionally, the seeds were planted in seeding containers. The seeds were arranged in a plant medium in humid conditions. he incubation temperatures ranged between 25 and 27°C. Observations were performed every day for thirty days. Seed germination vigor, abnormal seeds, dead seeds, seed germination potential, first count test, T50 test, value index test and soil emergence test were examined, in addition to fruit physical characteristics, seeds and seed germination.

RESULTS AND DISCUSSION

Seed sprout power, abnormal seeds and dead seed

the statistical analysis of test parameters for seed germination and dead seeds at multiple harvest ages produced results that deviated significantly from the norm, but not for seed parameters (Table 1). There is an effect of harvest age on the quality of the seeds produced, with the highest germination percentage (80.50%) occurring at harvest age 98-100 DAA, which meets the ISTA criteria for good seed quality (2018). This indicated the seed has reached physiological maturity. 101-103 DAA, 104-106 DAA, 107-109 DAA and 110-112 DAA did not differ significantly in terms of harvesting age. The typical germination rate falls below 34.17 per cent. This indicated that the quality of the seed diminished as a result of physiological maturation. This setback will have a negative effect on seed viability, vigor, plant development and yield. At this stage, seeds were vulnerable to environmental influences. According to Marcos (2015), seeds are physiologically mature when they are no longer genetically related to their parents.

Table 1 depicts the effect of harvest age on the percentage of dead seeds, with the lowest percentage occurring between 98 and 100 DAA. This result indicated that in addition to physiological maturity, the seeds obtained at an age of harvest between 98 and 100 DAA were optimal condition. Due to the condition of the young seeds, the proportion of dead seeds is greatest at harvest ages 101-103 DAA. Internal factors, such as a small embryo size, influence the development of abnormal sprouts. According to Pereira *et al*. (2014), embryos vary in size at each maturity stage. Even though they were growing in good reservoirs, these abnormal sprouts cannot develop into normal plants (Fadhilah, 2020). Some seeds may fail to develop into the optimal embryo for germination, whereas seeds harvested at 110-112 DAA were physiologically mature.

Fresh seeds, seed growing potential and soil emergence test

The statistical analysis of fresh seeds, seed growth potential and soil germination tests conducted at various harvest ages yielded significantly distinct results (Table 2). According to ISTA, fresh seeds did not grow and did not change color until the end of the experiment, but they were not classified as hard seeds. Some seeds in the tetrazolium test turned red (red circle) while others did not change color (black circle). Conditions after soaking in 0.5% tetrazolium can be seen in Fig 1.

Tests for tetrazolium should be conducted on seeds that have been cut and soaked in a solution of TZ (Fig 2). However, based on phase I, the absorbed tetrazolium solution does not necessarily indicate that the seed has

Table 1: Percentage of seed sprout strength, percentage of abnormal seeds and percentage of seeds dead at various harvest ages.

Harvest age (DAA)	Sprout strength (%)	Abnormal seeds (%)	Dead seed (%)
110-112	9.83b	2.50	9.83b
107-109	27.50b	2.00	6.17bc
104-106	34.17b	2.83	5.1c
101-103	11.00b	2.83	12.17a
98-100	80.50a	1.33	3.83c
	CV= 31.19	CV= 21.66	CV= 15.59

Description: The numbers in the same column followed by different lowercase letters are significant differences according to MRT at a level of 5%. CV is obtained in the transformation result = ASIN [SQRT (%DATA/100)]*180(22/7) for sprout strength, the result of transformation =√x for abnormal seeds and dead seeds.

germinated. Instead, it indicates that the seed is viable and has the potential to germinate. For instance, the tetrazolium test is useful not only for verifying other indicators of seed vigor, but also for evaluating the presence of damage, which may be the cause of low seed vigor (Franca-Neto and Krzyzanowski, 2018).

The results are presented in Table 2, which shows that the potential of growing seeds at harvest age 98-100 DAA is 81.83 percent, the highest of the entire harvest life, indicating that the seeds have likely reached physiological maturity at this point. Mello *et al*. (2010) state that physiologically mature seeds have perfectly formed embryos and sufficient endosperm to support seed metabolic activity and the development of its essential structures, such as roots, hypocotyls, epicotyls and plumules, so that they sprout normally.

Age of harvest between 98 and 100 DAA yields the best results for SET (Table 1). This is due to the physiological maturity of the seeds and subsequent sun-drying after harvest. Consequently, the seeds are vigorous. In accordance with Fauza's (2011) research, post-harvest gambir seeds are obtained from fruit picked when it has reached physiological maturity, but is not rotten and then dried in the sun for three to four days, or until the fruit breaks. According to Prasad *et al*. (2016), certain seeds can germinate within a few days of fertilization, with maximum germination occurring later in life.

The day it takes for seeds to germinate 50% (T50) first count test and percentage index value test gambir at various harvest ages

One factor affecting the germination rate of seeds is technical handling during seedlings, such as the medium used. The filter produces 50% more germinated seeds than ultisol soil media in paper media (Table 3). However, filter paper media can only be used to see the percentage of germination, then the growth of plants becomes slow because nutrients are not available to be absorbed by the roots and media conditions since the decreasing of water resistance occurs.

The results of the first counting test or germination of the first count on gambir seeds at various harvest ages indicate that there is no significant difference between the marked results. The results of the first count test (FCT) are shown in Table 3. Growing seeds fail because they are still

Table 2: Percentage of fresh seeds, seed growing potential and soil emergence test gambir at various harvest ages.

Harvest age (DAA)	Fresh seeds (%)	Seed growing potential (%)	Soil emergence test (%)
110-112	77.83a	12.33b	5.50c
107-109	64.33a	29.50b	31.67b
104106	57.83a	37.00b	12.67c
101-103	74.00a	13.83b	6.50c
98-100	14.33b	81.8a	84.33a
	CV= 21.73	CV= 30.59	CV= 20.42

Note: The numbers in the same column followed by different lowercase letters are significant differences according to MRT at a level of 5%. CV is obtained in the transformation result = ASIN [SQRT (%DATA/100)]*180(22/7).

Table 3: First count test and percentage of first counting test gambir at various harvest ages.

Harvest age (DAA)	Fiterpapar media	Ultisol landmedia	First count test (%)	Index value test	
	T50			Filter paper media	Ultisol land media
110-12	26	-	1.83	1.33b	0.87c
107-109	29	-	2.67	3.80b	5.21b
104-106	17	15	2.50	4.53b	1.87c
101-103	16	13	0.67	1.31b	0.82c
98-00	14	13	4.00	11.04a	12.83a
CV			36.29%	24.05%	16.12%

Note: The numbers in the same column are not significant according to the F test at a significance level of 5%. CV obtained in the transformation result=√x.

Table 4: Observation of fruit diameter, seed capsule size and seed number per capsule at different harvest ages.

Harvest age (DAA)	Fruit diameter (cm)	Capsule size (cm)	Number of seeds per capsule
110-112	3.82	2.62	7
107-109	3.73	2.45	137
104-106	4.47	3.01	153
101-103	4.48	2.72	341
98-100	5.64	3.05	95
Average	4.5	2.77	105

respirating. The slow growth rate indicates weak seed vigor. However, the harvest age between 98 and 100 is the best indicator of the number of seeds that germinate on the first day. The first day of germination, according to FCT bservations in this study, occurs on day 10. Overall, the calculated FCT value is relatively low. The germination potential and vigor of seeds characterize their physiological quality (Silva *et al*., 2016). According to Melia (2019), pre-germination treatment and seed quality influence FCT. Temperature and humidity will also impact germination rate. The condition of ultisol soil media rapidly dries out, disrupting the hydration process and enzymes and other components that promote germination.

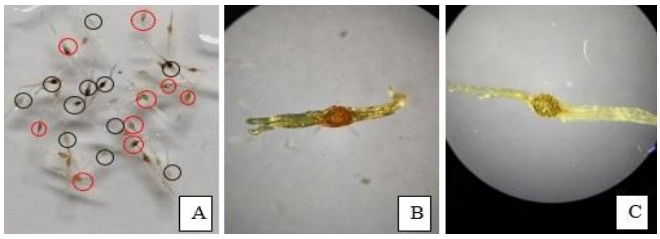
The results of observations of index value test (IVT) or the speed of seed growth at various ages of harvesting after analysis show different results in filter paper media and soil ultisol media. The observations of IVT can be seen in Table 3. Seeds at harvest age 98-100 DAA have a better-growing speed than the entire harvest age. The longer the day needed, the lower the index value. The index value obtained at the harvest age >101 DAA in Table 3 is relatively low because the number of days for the seed to germinate is relatively long. The expression of seed vigor can result from pre-harvest treatments such as drying at too high a temperature or a warm environment, the chemical composition of the seeds and hard seeds (Elias *et al*., 1997).

Observation of physical characteristics

Observations of physical characteristics are also made of the size of the fruit and the number of seeds produced. Observational data are presented in Table 4. Gambir flower has a diameter ranging from 3.82-5.64 cm with an average of 4.5 cm. The size of one seed capsule ranges from 2.45-3.05 cm with an average of 2.77 cm. This size may vary depending on the type. The number of seeds in each capsule is 7-195 seeds, with an average of 105 seeds. Based on Fauza's Research (2011), the number of each fruit's capsules ranges from 25 to 113, with an average of 62 capsules. Therefore, in one flower's cluster has the potential to produce 22,035 seeds. However, it can differ depending on the

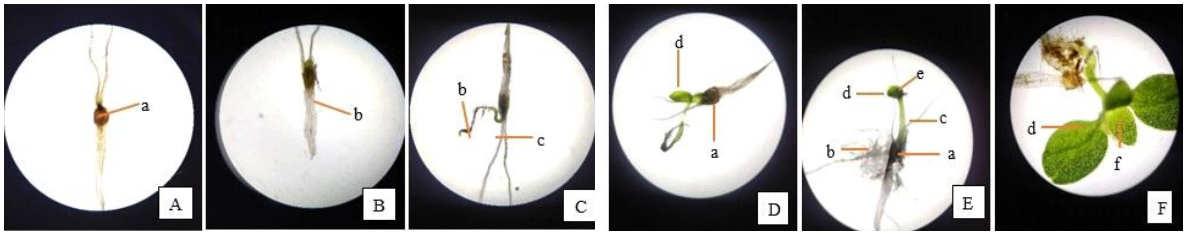
Harvest Age (HSA)	Color and shape of fruits		
	Repeat 1	Repeat 2	Repeat 3
110-112			
	Blackish brown-very broken	Brown-very broken	Brown-very broken
107-109			
	Brown-broken	Brown-cracked	Partially brown-broken
104-106			
	Light-cracked Brown	Light-cracked Brown some	Brown-cracked
101-103			
	Brown-very Broken	Light-cracked Brown	Brown-very broken

Fig 1: Observation of the color and shape of gambir fruit of various harvest ages.



(A) seed immersion in TZ solution, (B) portions of the embryo turn red to indicate viability, (C) absence of seed discoloration indicates nonviability.

Fig 2: Tetrazolium test.



(A) the seed begins to appear swelling on 5th day after the censure marking the imbibe, (B) radicle begins to grow on 6th day after being debauched, (C) the hypocotyl extends on the 7th day, (D) the cotyledon begins to grow on the 9th day, (E) the epicotyl begins to appear and the seed coat detaches from the sprouts on the 10th day, (F) the first leaves begin to grow on the 20th day; (a) seed coat, (b) radicle, (c) hypocotyl, (d) cotyledon, (e) epicotyl, (f) true leaves.

Fig 3: The germination phases of gambir.

climate because the fruit is very easy to fall caused by the extreme weather such as heavy rains and strong winds, which can reduce crop yields.

According to Ningsih (2012), the physiological maturation process of fruits and seeds usually coincides with the maturing time of the seeds. Physiological maturing of the fruit is also an increase in the production of sugar and water so that there is a change in color in the flesh of the fruit; discoloration occurs due to decreasing levels of chlorophyll and increasing levels of carotenoid and anthocyanin. Observations on the physical condition of the fruit are presented in Fig 3, showing the condition of the fruit at the harvest age of 110-112 DAA is brown and blackish-brown with very broken capsules. Therefore, lesser seeds were harvested.

The color of the fruit at the harvest age of 107-109 DAA is brown and partially broken. It is not much different from the harvest age fruit 104-106 DAA with light brown fruit color. The condition of the fruit at the harvest age of 101-103 DAA is light brown, but some capsules are in a very broken condition. The condition of the fruit at the harvest age of 98-100 DAA is yellowish-green to bright brown, with the condition of the capsule intact until partially broken.

Gambir seeds have an epigeal germination type with mount rooting. The phases of germination of gambir can be seen in Fig 3. Gambir seeds size about 1-2 mm. The structure of the gambir seed consists of wings and embryos. The wings only serve as seed protectors and aid to spread. The enlarged part of the seed embryo contains tissues and living cells that will develop into epicotyls,radicle ,plumules in the germination process.

The germination process (Fig 3) begins with the phase of imbibing, which is characterized by the start of swelling of the embryo in the seed coat (6A) to the appearance of radicle (6B), the growth of radicle followed by hypocotyl lengthening (6C), then the cotyledon comes out of the seed coat and lifts upwards followed by epicotyl growth (6D) and produces intact sprouts that have formed roots, hypocotyl, cotyledon, epicotyl (6E), The end of the seed germination process produces one plant that is intact and continues to grow with multiple leaf growths (6F).

Research on the maturity level of color has been conducted by Rohaeni (2019). She studies the maturity level of coffee fruit based on color showing the red color yield has a 100% seed germination rate. This color grouping needs to be further investigated because each type of gambir result may be different.

CONCLUSION

The viability and vigor of gambir seeds harvested between 98 and 100 DAA indicate that the seed has reached physiological maturity. The best viability and vigor resulted from a germinated power test of 80.50 per cent, a seed growth potential test of 81.83 per cent, a T50 test, an index value test of 11.04 and a soil emergence test of 84.33 per cent, indicating that the plant had reached its physiological maturity.

ACKNOWLEDGEMENT

We thank the Directorate General of Higher Education, the Ministry of Research, Technology and Higher Education of the Republic of Indonesia and LPPM Andalas University for Reputable Publication Research with contract No.T/2/ UN.16.17/PT.01.03/ food-RPB/2022, date 11 April 2022. SK Rektor No. 360/KPT/R/PTN-BH/UNAND/2022, date 8 April 2022.

Conflict of interest: None.

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