



Effect of fruit age, pre-storage and seed fermentation durations on seed germination and seedling vigor in *Lagenaria siceraria*

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ABSTRACT

Objectives: the study was conducted to determine the optimum age of the fruit at harvest and to evaluate the effect of pre-storage and fermentation on the seeds' agronomic performance of oleaginous *Lagenaria siceraria*.

Methodology and results: Fruits of the “round-fruited” and “blocky-fruited”, two cultivars of the species, were harvested at 3 different ages (30, 40 and 50 days after anthesis (DAA)). These fruits were pre-stored during 0, 30 and 60 days after harvest (DAH), and then their seeds were fermented for 0, 5 and 10 days. Seed viability (germination percentage and germination speed index) and seedlings vigor (emergence percentage, emergence speed index, seedling shoot length and seedling dry weight) were significantly ($P < 0.001$) influenced by each of the three factors. Following the positive interaction of these factors, the best viability and the most vigorous seedlings were obtained with seeds from fruits tardily harvested (50 DAA), pre-stored for a long time (60 DAH) and longer fermented (10 days). The simultaneous variation of the fruit wet weight and the seed color, dry weight and moisture content with their viability and the seedlings vigor indicates that these four parameters can be used as indicators of both cultivars maturity.

Conclusions and application of findings: The results suggest that in oleaginous *L. siceraria*, fruits must be harvested 50 DAA, pre-stored up to 60 DPA. Good agronomic quality seeds must have yellow-orange color and be fermented up to 10 days.

Key-words: *Lagenaria siceraria*, fruit age, pre-storage, seed fermentation, maturity, viability, seedling vigor

INTRODUCTION

Lagenaria siceraria is a wild species comprising a very great morphological diversity of fruits (Morimoto *et al.*, 2005). Some of its representatives are the bottle-gourds used as containers or music instruments while the others, oleaginous ones, produce oilseeds consumed as soup in most tropical Africa and Asia areas (Ndabalishye, 1995; Zoro Bi *et al.*, 2003). Among the oleaginous cucurbits, *this* species shows the

best agronomic potentialities mainly due to its big fruits, and large seeds (Morimoto *et al.*, 2005). Furthermore, *L. siceraria* is reported to be rich in nutrients, namely protein ($36 \pm 2.17\%$) and fat ($45.89 \pm 4.73\%$) and is well adapted to extremely divergent agro-ecosystems and various cropping systems characterized by minimal inputs (Zoro Bi *et al.*, 2006). Thus it represents an important source of income for farmers, mainly for women, in

West and Central Africa (Zoro Bi *et al.*, 2003; Achigan *et al.*, 2006; Zoro Bi *et al.*, 2006). Despite of the economic, social and nutritional role played by this group of oleaginous cucurbits, they are classified as orphan crops (Chweya & Ezaguirre, 1999; IPGRI, 2002).

Rural farmers generally harvest fruits of this species after complete senescence of the plants. Because of the fruiting extent, all the fruits do not reach maturity before plant senescence, resulting in a mixing of mature and immature seeds at the harvest (Nerson & Paris, 1988). This can explain the poor seed yield and low germination rate widely reported in the species (Achigan *et al.*, 2006). The enhancement of the yield of any crop is a prerequisite for its popularization. Improved seed system development is one of main tasks allowing the sustainable production of the oleaginous cucurbits (Almekinders *et al.*, 1994). Indeed, seed agronomic quality, i.e. their aptitude to germinate and produce vigorous seedlings (Egli & Tekrony, 1995; Cantliffe, 1998; Nabi *et al.*, 2001), influences strongly the yield of the future plants (Embaye *et al.*, 2003; Al-Maskri *et al.*, 2004). Seed germinability depends mainly on its physiological maturity (McCaughey & Stephenson, 2000; Demir & Samit, 2001; Torres & Marcos-Fhilo, 2005). The production of high-quality seed in cucurbits

depends on the fruit age at harvest and the extraction procedure (Nerson & Paris, 1988) as well as fruit pre-storage duration. For the cultivated cucurbit species, fermentation is the main extraction procedure ensuring the separation of seeds from the fruit flesh in which they are strongly encrusted. This process is reported to considerably improve seed nutritive value and germinability (Edwards *et al.*, 1986; Nerson & Paris, 1988). However, the fermentation duration is deterministic for seed nutritive and agronomic qualities. Longer fermentation has decreased germination percentage and rate in cucumber (Nienhuis & Lower, 1981).

Several studies have been conducted to examine the effects of some fruit post-harvest treatments such as pre-storage and fermentation on seed germination and seedling vigor in the cucurbit species (Edwards *et al.*, 1986; Nerson & Paris, 1988). However, studies analyzing the combined effects of fruit maturity at harvest, pre-storage and fermentation durations in cucurbits are scant. To our knowledge, such studies have never been conducted on the oleaginous bottle gourd *L. siceraria*. The results obtained are a report from a study examining the effect of fruit age, pre-storage and fermentation durations on seed germination and seedling vigor in the oleaginous *L. siceraria*.

MATERIALS AND METHODS

Plant material: Two cultivars of oleaginous *Lagenaria siceraria*, the one blocky-fruited (BFC) producing seed without a cap (Figure 1 a and b) and the other round-fruited (RFC) producing seed with a cap on distal side (Figure 1 c and d) (Zoro Bi *et al.*, 2006), were used.

These seeds, recorded respectively under the introduction numbers NI304 and NI195, were selected in the collection of the University of Abobo-Adjamé (Abidjan, Côte d'Ivoire).



a



b



Figure 1: Fruit of “blocky-fruited cultivar” (a) and its seeds without cap (b); fruit of “round-fruited cultivar” (c) and its seeds with a cap on a distal side of the oleaginous *Lagenaria siceraria*

Obtaining various aged fruits: Each of the two cultivars (BFC and RFC) of *Lagenaria siceraria* was grown during two rainfall seasons (from March to June 2007 and from April to July 2008) in an isolated field at the experimental farm of University of Abobo-Adjamé (Abidjan, Côte d’Ivoire). To obtain vigorous seedlings, then good yield, both fields were pre-fertilized with pig manure. These fields were regularly maintained by three weeding and one insecticidal treatment (Cypercal 50 EC). Approximately 150 pistillate flowers of each cultivar were tagged at anthesis, and 15 fruits which developed from them were manually harvested by simple rupture of stalk at 30, 40 and 50 days after anthesis (DAA).

Fruits pre-storage and seed fermentation: For both cultivars (BFC and RFC), the fruits of each age (30, 40 and 50 DAA) were pre-stored during 30 and 60 days after harvest (DAH) on the experimental field in order to keep them under conditions close to those of the culture. A sample of non-pre-stored fruits (0 DAH), constituted the control treatment. At the end of each pre-storage duration, fruits were split using machete. A sample of non-fermented seeds was used as the control. The other split fruits were packed in a polyethylene bag and hidden to 30 cm depth in the ground for fermentation. At the end of each fermentation duration, 5 days and 10 days, the seeds were manually extracted, abundantly washed with tap water and then sun-dried at ambient air (22-32°C) until constant weight. Seed moisture content was determined by the high-temperature oven method (103°C, 17h) according to ISTA methods (ISTA, 1999). Finally 27 different treatments of seeds were performed: 3 fruit ages (30, 40, and 50 DAA) × 3 pre-storage durations (0, 30 and 60 DAH) × 3 fermentation durations (0, 5 and 10 days) by cultivar.

Experimental design for seed germination and seedling vigor evaluation: The experimental sowing

device was a completely randomized block design with 5 replications of 3 plots each one; hence 15 plots on the whole. There were spacing of 3 m width between replications and 0.5 m between plots. Each plot of 1 m × 0.5 m, representing one of the 3 pre-storage durations (0 DAH or 30 DAH or 60 DAH), was divided into two equal parts in its length. Each part, corresponding to one cultivar (BFC and RFC) comprised 9 treatments [3 fruits ages (30, 40 and 50 DAA) × 3 durations of fermentation (0, 5 and 10 days)]. Each treatment counted 20 seeds resulting from a same fruit. Sowings were carried out on plot well plowed with only one seed per seed hole. Seed holes, with 3 cm depth, were spaced each 7 cm. A total of 5400 seeds (5 replications × 2 cultivars × 27 treatments × 20 seeds) were sown by repetition. Two repetitions of sowing (the first, in July 2007 and the second, in August 2008) were carried out during this experiment.

Data collection: The fruit weight, the seed color and the dry weight of 100 seeds (DW_{100}) were used to test morphological maturity, and the seed moisture content (Smc) to test physiological maturity (Day, 2000; Gupta *et al.*, 2005). Seed germinability was evaluated using the seed germination percentage (GeP) and germination speed index (GSI). Seeds were considered as germinated when the cotyledons appeared above the ground level. The sown seeds were surveyed daily for 14 days (ISTA, 1996). The germination speed index (GSI) was calculated using the formula suggested by Maguire (1962) as follow:

$$GSI = \frac{G_1}{N_1} + \frac{G_2}{N_2} + \dots + \frac{G_n}{N_n}, \text{ where:}$$

G_1 , G_2 , and G_n represent the numbers of germinated seeds on the first count, the second count, and the last count; N_1 , N_2 , and N_n are the numbers of days elapsed of the first, second, and last count.

The seed vigor was examined through the seedling emergence percentage (EmP), emergence speed index (ESI), shoot length (SSL), and dry weight (DWS). A seedling was considered as emerged when its two cotyledonary leaves were completely opened (Koffi *et al.*, 2009). The seedling emergence speed index (ESI) was also calculated on the basis of the procedure used by Maguire (1962) according to the following equation:

$$ESI = \frac{E_1}{N_1} + \frac{E_2}{N_2} + \dots + \frac{E_n}{N_n}, \text{ where:}$$

E_1 , E_2 , and E_n represent the numbers of emerged seedlings on the first count, the second count, and the last count; N_1 , N_2 , and N_n are the numbers of days elapsed of the first, second, and last count.

Statistical analysis: Percentage data were arcsin-transformed before analysis (Little, 1985) but

untransformed data were used to calculate means to present the results. Multivariate analysis of variance (MANOVA) appropriate for five-way fixed model was performed using SAS statistical package (SAS, 2004) to check difference between the variable means for each factor tested (repetition, cultivar, fruit age, pre-storage duration and fermentation duration). This analysis allowed evaluating firstly, the individual effect of these 5 factors on each parameter, then their interactions. When the null hypothesis was rejected for each parameter, multiple comparisons using the Least Significant Difference (LSD) test were carried out to determine differences between fruits ages, pre-storage duration, fermentation duration and their interaction (Dagnelie, 1998). All LSD tests were performed at $\alpha = 0.05$ significance level.

RESULTS

Overall effect of analyzed factors: MANOVA revealed significant influence of repetitions, cultivar,

fruit age, pre-storage duration, and fermentation duration (**Table 1**).

Table 1: MANOVA results of repetitions (Rep), cultivars (Cult), fruit age at harvest (FrAg), pre-storage duration (PsDu), seed fermentation duration (FeDu) and their different interactions on seed germination and seedling vigor in oleaginous *Lagenaria siceraria*

Factors ¹	DF	F	P
Rep	1	38,957	<0,001
Cult	1	2,598	0,036
FrAg	2	94,153	<0,001
PsDu	2	53,377	<0,001
FeDu	2	43,821	<0,001
Rep×Cult	3	8,997	<0,001
Rep×FrAg	5	13,290	<0,001
Cult×FrAg	5	4,867	<0,001
Rep×PsDu	5	2,069	0,036
Cult×PsDu	5	1,530	0,142
FrAg×PsDu	8	1,960	0,012
Rep×FeDu	5	5,717	<0,001
Cult×FeDu	5	2,048	0,038
FrAg×FeDu	8	0,882	0,589
PsDu×FeDu	8	2,153	0,005
Rep×Cult×FrAg	11	2,006	0,043
Rep×Cult×PsDu	11	1,978	0,046
Rep×FrAg×PsDu	17	2,283	0,002
Cult×FrAg×PsDu	17	0,703	0,793
Rep×Cult×FeDu	11	2,915	0,003
Rep×FrAg×FeDu	17	2,033	0,009
Cult×FrAg×FeDu	17	1,177	0,279
Rep×PsDu×FeDu	17	1,571	0,069
Cult×PsDu×FeDu	17	2,145	0,005
FrAg×PsDu×FeDu	26	0,911	0,611
Rep×Cult×FrAg×PsDu	35	1,004	0,450
Rep×Cult×FrAg×FeDu	35	1,061	0,389

Rep×Cult×PsDu×FeDu	35	1,293	0,193
Rep×FrAg×PsDu×FeDu	53	1,103	0,317
Cult×FrAg×PsDu×FeDu	53	1,042	0,404
Rep×Cult×FrAg×PsDu×FeDu	107	1,381	0,077

¹Factors in bold type have significant effect ($P < 0.05$)

Consequently, most part of the double, the three, the four and the five interactions were significant. Nevertheless, for most of the eight traits examined, the trends of results related to single effects of fruit age, pre-storage duration and cultivar did not change through the two repetitions. The trend of the results related to the effects of fruit age, pre-storage and fermentation durations in first repetition (July 2007) were similar to those of the second repetition (August

2008) for all the measured parameters (data not presented) for each cultivar. Therefore, data for these three factors were pooled over repetitions and only the means are presented for each cultivar.

Fruit age: When fruit age at harvest is delayed from 30 to 50 DAA, their wet weight (**Figure 2**) and the dry weight of 100 seeds (DW_{100}) increased very significantly ($P < 0.001$) (**Table 2**).

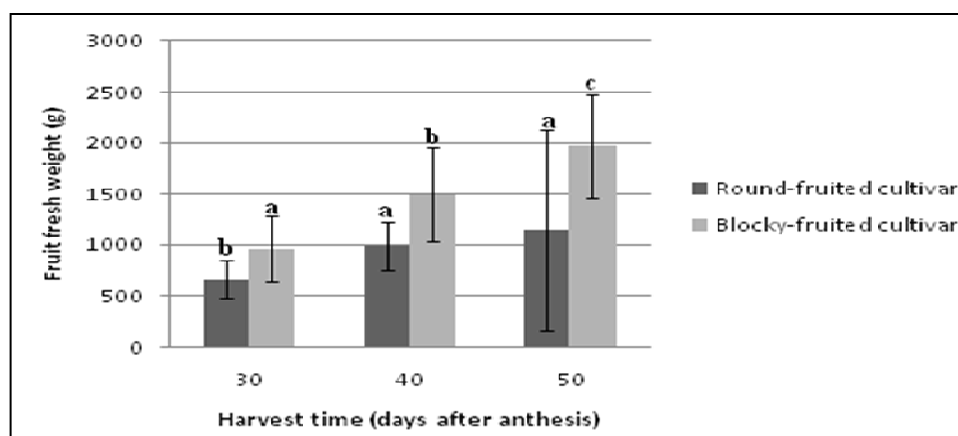


Figure 2: Histogram of fruit fresh weight variation according to their age at harvest (days after anthesis) for two cultivars of oleaginous *Lagenaria siceraria*. For each cultivar, vertical bars represent the standard deviation and the bands carrying the same letters are statistically equal

Table 2: Seed maturity, seed viability and seedling vigor response to fruit age in both cultivars of the oleaginous *Lagenaria siceraria*

Parameters ¹	Means according to fruit age at harvest ²			Statistics ³	
	30 DAA	40 DAA	50 DAA	F	P
DW_{100} (g) ²	10.74± 2.91 ^c	13.41± 2.54 ^b	16.09± 2.18 ^a	60.42	<0.001
Smc (%)	9.63± 1.16 ^a	8.65± 0.82 ^b	7.66 ± 0.96 ^c	32.19	<0.001
GnP(%)	42.77± 21.07 ^c	67.38± 19.07 ^b	78.45 ± 13.33 ^a	66.53	<0.001
EmP(%)	40.25± 21.03 ^c	65.58± 18.87 ^b	76.02± 12.90 ^a	72.59	0.003
GSI (Sd/d)	1.44± 0.98 ^c	2.33± 0.82 ^b	2.81 ± 0.76 ^a	38.63	<0.001
ESI (Sdl/d)	0.86 ± 0.49 ^c	1.42 ± 0.49 ^b	1.77 ± 0.45 ^a	40.58	<0.001
SSL (mm)	80.27± 15.32 ^c	88.50± 13.81 ^b	99.78± 11.51 ^a	401.99	<0.001
DWS (g)	0.27± 0.09 ^c	0.31± 0.08 ^b	0.34 ± 0.09 ^a	168.21	<0.001

¹ DW_{100} : dry weight of 100 seeds ; **Smc** : seed moisture content ; **GnP** : germination percentage, **GSI** : Germination speed index, **EmP** : emergence percentage, **ESI** : Emergence speed index, **DAH**: days after harvest, **SSL** : seedling shoot length, **DWS** : Dry weight of seedlings , **Sd/d** : seeds/day et **Sdl/d** : seedlings/day

²For each parameter, values with the same letters are statistically equal

³ In each row, the values with the same superscript letter are not significantly different from each other.

But for the same variations of fruits age, seeds moisture content (Smc) decreased very significantly (P

< 0.05) while their external color vary from white to yellow (Figure 3).

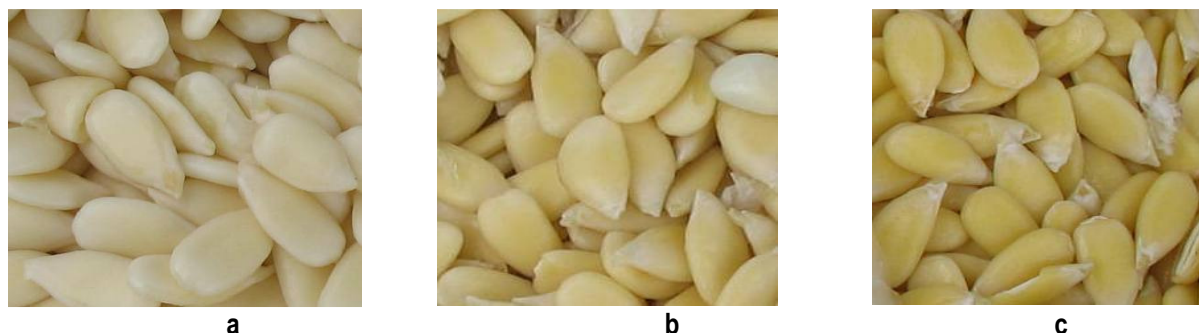


Figure 3: Variation of fresh seed external color from unfermented fruit harvested after 30 (a), 40 (b) and 50 (c) days after anthesis in oleaginous *Lagenaria siceraria*

The lowest germination percentage (GnP) and germination speed index (GSI) in both cultivars were observed with precocious seeds (30 DAA) and, their highest values with the oldest ones (50 DAA). Thus, the seed maturation increased with the age of fruits at harvest while their viability was improved. The emergence percentage (EmP), the emergence speed index (ESI), the seedling shot length (SSL) and the seedling dry weight (DWS) were very significantly influenced ($P < 100$) by the age of harvested fruits. The LSD tests showed that the lowest values of these vigor parameters were obtained with the seeds resulting from berries precociously harvested (30 DAA) while their highest values were observed with seeds from berries tardily harvested (50 DAA). The delayed-action of the berries harvest time, from 30 to 50 DAA, improved seedlings vigor resulting from their seeds.

Fruit pre-storage duration: Table 3 indicates that the lightest dry seeds (DW_{100}), the lowest germination percentage (GnP) and the lowest germination speed index (GSI) were obtained with the control fruits (non pre-stored: 0 DAH) while their highest values were found with the fruits pre-stored longer (60 DAH). But the moisture content of these seeds (Smc) decreased significantly with fruits pre-storage duration. Thus, pre-storage of fruits strongly improved the parameters of maturity and seed viability. The mean values of the seedlings vigor parameters (EmP, ESI, SSL and DWS) were very significantly ($P < 0.001$) low with the seeds of control berries (non pre-stored: 0 DAH), average with seeds of berries pre-stored during 30 DAH and, higher with seeds of berries pre-stored 60 DAH. The prolongation of fruits pre-storage duration improved seedling vigor.

Table 3: Evolution of seed maturity, seed viability and seedling vigor parameters according to fruit pre-storage duration in the oleaginous *Lagenaria siceraria*

Parameters ¹	Pre-storage duration ²			Statistics ³	
	0 DAH (control)	30 DAH	60 DAH	F	P
DW_{100} (g)	10.50 ± 2.93 ^c	13.47 ± 2.54 ^b	16.74 ± 2.18 ^a	61.43	<0.001
Smc (%)	9.49 ± 1.36 ^a	8.68 ± 0.62 ^b	7.96 ± 0.98 ^c	24.78	<0.001
GnP(%)	52.02 ± 22.58 ^c	63.48 ± 19.98 ^b	72.90 ± 20.16 ^b	20.42	<0.001
EmP(%)	49.30 ± 23.49 ^b	62.07 ± 21.19 ^a	70.30 ± 19.52 ^a	19.80	<0.001
GSI (Sd/d)	1.55 ± 0.83 ^c	2.46 ± 0.86 ^b	2.72 ± 0.83 ^a	44.49	<0.001
ESI (Sdl/d)	0.99 ± 0.52 ^b	1.42 ± 0.52 ^a	1.62 ± 0.52 ^a	32.67	<0.001
SSL (mm)	67.83 ± 12.44 ^c	74.27 ± 12.96 ^b	84.96 ± 1.07 ^a	116.73	<0.001
DWS (g)	0.27 ± 0.09 ^c	0.31 ± 0.08 ^b	0.33 ± 0.09 ^a	58.21	<0.001

¹ DW_{100} : dry weight of 100 seeds ; **Smc** : seed moisture content ; **GnP** : germination percentage, **GSI** : Germination speed index, **EmP** : emergence percentage, **ESI** : Emergence speed index, **DAH**: days after harvest, **SSL** : seedling shoot length, **DWS** : Dry weight of seedlings , **Sd/d** : seeds/day et **Sdl/d** : seedlings/day

²For each parameter, values with the same letters are statistically equal

³ In each row, the values with the same superscript letter are not significantly different from each other

Seeds fermentation duration: Seeds fermented for 5 days and 10 days had a dry weight (DW_{100}) statistically equal ($F= 1.04$; $F=0.35$) to that of control seeds (not fermented). However, their moisture content (Smc)

decreased significantly ($P < 0.001$) from $9.43 \pm 1.26\%$ to $7.95 \pm 0.91\%$ during the 10 days of fermentation (0 to 10 days) (Table 4).

Table 4: Evolution of seed maturity, seed viability and seedling vigor parameters according to seed fermentation duration in the oleaginous *Lagenaria siceraria*

Parameters ¹	Fermentation duration			Statistics ²	
	0 day (control)	5 days	10 days	F	P
DW ₁₀₀ (g)	13.91 ± 4.10	13.72 ± 2.69	13.02 ± 2.63	1.04	0.350
Smc (%)	9.43 ± 1.26 ^a	8.78 ± 0.97 ^b	7.95 ± 0.91 ^c	21.82	<0.001
GnP(%)	49.80 ± 20.30 ^c	64.85 ± 21.51 ^b	72.95 ± 19.52 ^a	27.21	<0.001
EmP(%)	48.88 ± 20.40 ^c	62.56 ± 20.97 ^b	70.43 ± 20.93 ^a	24.70	<0.001
GSI (Sd/d)	1.78 ± 0.81 ^b	2.29 ± 0.98 ^a	2.49 ± 0.91 ^a	17.01	<0.001
ESI (Sdl/d)	1.13 ± 0.53 ^b	1.37 ± 0.61 ^a	1.54 ± 0.59 ^a	8.82	0.003
SSL (mm)	85.00 ± 14.37 ^c	92.04 ± 15.05 ^b	94.97 ± 15.16 ^a	72.35	<0.001
DWS (g)	0.28 ± 0.10 ^c	0.31 ± 0.09 ^b	0.33 ± 0.09 ^a	27.28	<0.001

¹ DW₁₀₀ : dry weight of 100 seeds ; Smc : seed moisture content ; GnP : germination percentage, GSI : Germination speed index, EmP : emergence percentage, ESI : Emergence speed index, SSL : seedling shoot length, DWS : Dry weight of seedlings , Sd/d : seeds/day et Sdl/d : seedlings/day

² In each row, the values with the same superscript letter are not significantly different from each other

The lowest germination percentage (GnP) and germination speed index (GSI) were observed with non-fermented seeds (control) and their highest values, with the longer fermented seeds (10 days). Fermentation did not affect seeds dry weight but its duration significantly improved their viability. Extending seeds fermentation duration from 0 day (control) to 10 days, significantly ($P < 0.001$) increased the emergence percentage (EmP), the emergence speed index (ESI), the seedlings shoot length (SSL) and the dry matter of the seedlings. Extending the fermentation duration improved seedlings of oleaginous *L. siceraria*.

Interaction between fruit age, pre-storage and fermentation: In both cultivars RFC and BFC, for each age of fruit (30, 40 and 50 DAA), seed dry weight (DW_{100}) increased significantly ($P < 0.001$) following pre-storage duration (from 0 to 60 DAH) while their moisture content (Smc) showed a very strong decrease ($F = 273.19$; $P < 0.001$). However, except the control treatment 30 DAA × 0 DAH, the dry weight of seeds resulting from all treatments decreased with fermentation duration (from 0 to 10 days). This decrease of seed weight was also followed by a decrease of their moisture content (Table 5). From 30th to 50th day after pollination, the seeds of both cultivars RFC and BFC of the oleaginous *Lagenaria siceraria* accumulated dry matter and dehydrated gradually. It

increased their dry weight (of 100 seeds) respectively from 6.25 ± 2.36 to 17.58 ± 1.44 g and from 9.98 ± 3.34 to 21.61 ± 0.91 g. This accumulation continued for immature seeds in the pre-stored fruits. During fermentation, the seeds are dehydrated considerably; what decreases their dry weight. The germination percentage (GnP) and the germination speed index (GSI) increased with the age of fruits at harvest, the duration of storage and the duration of fermentation. Seed viability begun during the fruit formation and continued during pre-storage and fermentation. For each fruit age at harvest (30, 40 and 50 DAA), seedlings resulting from their seeds presented an emergence percentage (EmP), an emergence speed index (ESI), a mean seedlings shoot length (SSL) and a dry weight of seedlings (DWS) increasing significantly ($P < 0.001$) with storage duration in both cultivars RFC and BFC (Table 6). Moreover, for all the duration of pre-storage (0, 30 and 60 DAH), these vigor parameters of the seedlings (EmP, ESI, SSL and DWS), resulting from each harvest age (30, 40 and 50 DAA), increased significantly with the duration of seed fermentation (0 to 10 days) from which they resulted. Thus, the delayed-action of harvest, the pre-storage of fruits and the duration of seeds fermentation improved seedlings vigor.

Table 5: Interaction of fruit age, fruit pre-storage duration and fermentation duration on seed maturity, seed viability of two cultivars of *Lagenaria siceraria*

Fruit age at harvest	Fruit pre-storage duration	Seed fermentation duration	Round fruited cultivar				Blocky fruited cultivar			
			DW ₁₀₀ (g)	Smc (%)	GnP (%)	GSI (Sd/d)	DW ₁₀₀ (g)	Smc (%)	GnP (%)	GSI (Sd/d)
30 DAA	0 DAH	0 day	6,25 ± 2,36 ^m	11,97 ± 0,32 ^a	18,33 ± 12,24 ^l	0,45 ± 0,17 ^k	9,98 ± 3,34 ⁱ	12,13 ± 0,19 ^a	15,55 ± 8,81 ^j	0,39 ± 0,20 ^m
		5 days	9,01 ± 2,18 ^l	10,47 ± 0,12 ^b	31,11 ± 9,93 ^k	0,94 ± 0,27 ^{jk}	11,43 ± 2,86 ^{ghi}	11,39 ± 0,25 ^b	23,88 ± 15,36 ^j	0,64 ± 0,18 ^m
		10 days	10,85 ± 3,26 ^{ijkl}	9,56 ± 0,44 ^c	43,88 ± 17,81 ^{hij}	1,19 ± 0,57 ^{ij}	11,39 ± 1,38 ^{ghi}	10,52 ± 0,41 ^c	38,33 ± 23,97 ⁱ	0,71 ± 0,17 ^m
	30 DAH	0 day	11,56 ± 1,06 ^{hij}	9,35 ± 0,08 ^d	37,22 ± 18,21 ^{jk}	1,39 ± 0,63 ^{hij}	12,65 ± 1,65 ^{fgh}	10,17 ± 0,23 ^d	24,44 ± 11,30 ^j	0,90 ± 0,34 ^{klm}
		5 days	10,68 ± 1,04 ^{kl}	8,91 ± 0,11 ^{fg}	54,44 ± 20,83 ^{gh}	1,94 ± 1,18 ^{efgh}	11,14 ± 1,28 ^{ghi}	9,19 ± 0,12 ^g	44,44 ± 23,64 ^{hi}	1,28 ± 0,46 ^k
		10 days	11,17 ± 2,29 ^{ijk}	8,39 ± 0,24 ^{kl}	63,33 ± 18,37 ^{ef}	2,55 ± 0,87 ^{cd}	10,62 ± 1,82 ^{hi}	8,61 ± 0,14 ⁱ	52,77 ± 25,87 ^{efgh}	1,70 ± 0,65 ^{ij}
	60 DAH	0 day	12,34 ± 2,27 ^{ghij}	9,24 ± 0,06 ^{1d}	42,22 ± 8,70 ^{jk}	1,61 ± 0,56 ^{hi}	14,47 ± 0,30 ^{cdef}	9,60 ± 0,15 ^e	46,11 ± 19,00 ^{hi}	1,24 ± 0,78 ^{jk}
		5 days	11,43 ± 0,39 ⁱ	8,49 ± 0,17 ^{jk}	59,44 ± 13,09 ^{efg}	2,21 ± 0,79 ^{defg}	13,52 ± 1,26 ^{efgh}	9,08 ± 0,10 ^{gh}	48,33 ± 12,74 ^{ghi}	1,91 ± 0,62 ^{hi}
		10 days	11,49 ± 0,61 ^{ij}	8,16 ± 0,03 ^{mn}	64,44 ± 17,40 ^{ef}	2,49 ± 1,07 ^{cdef}	13,34 ± 0,48 ^{efgh}	8,16 ± 0,04 ^{kl}	61,66 ± 20,61 ^{def}	2,33 ± 0,68 ^{gh}
40 DAA	0 DAH	0 day	12,73 ± 2,75 ^{efghij}	10,58 ± 0,16 ^b	41,11 ± 11,66 ^{jk}	1,13 ± 0,41 ⁱ	14,88 ± 2,15 ^{cdef}	9,53 ± 0,15 ^e	43,33 ± 20,91 ^{hi}	1,18 ± 0,38 ^{kl}
		5 days	12,54 ± 2,32 ^{ghij}	9,18 ± 0,11 ^{de}	53,88 ± 12,69 ^{ghi}	1,57 ± 0,67 ^{hi}	14,12 ± 3,66 ^{cdef}	8,57 ± 0,19 ^j	60,55 ± 10,44 ^{defg}	1,78 ± 0,33 ^j
		10 days	9,04 ± 2,54 ^{kl}	8,03 ± 0,09 ^{no}	65,55 ± 15,29 ^{ef}	1,93 ± 0,55 ^{gh}	14,03 ± 3,53 ^{def}	8,18 ± 0,11 ^{kl}	69,44 ± 12,61 ^{cd}	1,89 ± 0,47 ^{hi}
	30 DAH	0 day	13,32 ± 1,05 ^{efg}	9,18 ± 0,08 ^{de}	54,44 ± 14,24 ^{fgh}	1,85 ± 0,59 ^{gh}	15,26 ± 0,94 ^{cde}	9,42 ± 0,11 ^e	60,55 ± 13,33 ^{defg}	2,06 ± 0,67 ^{ghi}
		5 days	12,41 ± 2,01 ^{ghj}	8,55 ± 0,18 ^{jk}	70,55 ± 8,07 ^d	2,77 ± 0,51 ^{bcd}	14,06 ± 0,42 ^{cdef}	8,59 ± 0,16 ⁱ	69,44 ± 7,26 ^{cd}	2,50 ± 0,54 ^{efg}
		10 days	12,12 ± 2,61 ^{ghij}	8,06 ± 0,14 ^{mn}	81,11 ± 9,27 ^{cd}	2,84 ± 0,34 ^{bc}	13,54 ± 0,86 ^{efgh}	7,95 ± 0,10 ^{lm}	77,77 ± 10,03 ^c	2,59 ± 0,50 ^{def}
	60 DAH	0 day	15,15 ± 2,48 ^{bode}	8,78 ± 0,27 ^{ghi}	64,44 ± 18,44 ^{ef}	2,49 ± 0,31 ^{cdef}	15,56 ± 0,82 ^{cde}	9,16 ± 0,08 ^g	67,77 ± 13,48 ^{cd}	2,62 ± 0,87 ^{def}
		5 days	12,58 ± 2,38 ^{ghij}	8,26 ± 0,06 ^{lm}	83,33 ± 16,00 ^{abc}	3,07 ± 0,37 ^{ab}	15,03 ± 0,05 ^{cdef}	8,28 ± 0,06 ^{jk}	77,22 ± 11,21 ^c	3,01 ± 0,45 ^{bcd}
		10 days	11,04 ± 1,95 ^{ijl}	7,84 ± 0,11 ^o	86,66 ± 7,07 ^{ab}	3,24 ± 0,33 ^{ab}	14,77 ± 0,31 ^{cdef}	7,53 ± 0,20 ^o	85,55 ± 12,61 ^a	3,30 ± 0,69 ^{abc}
50 DAA	0 DAH	0 day	15,81 ± 2,27 ^{ab}	9,09 ± 0,09 ^{ef}	61,66 ± 5,59 ^{ef}	1,95 ± 0,88 ^{efgh}	18,49 ± 2,98 ^{ab}	9,17 ± 0,09 ^g	49,44 ± 6,82 ^{fghi}	2,01 ± 0,57 ^{hi}
		5 days	14,19 ± 0,69 ^{cdef}	8,68 ± 0,09 ^{hij}	73,33 ± 7,50 ^{cd}	2,53 ± 0,44 ^{cd}	17,06 ± 3,31 ^{bc}	8,46 ± 0,14 ^{ij}	70,00 ± 10,30 ^{cd}	2,30 ± 0,61 ^{fgh}
		10 days	12,98 ± 1,48 ^{efg}	7,29 ± 0,06 ^p	81,11 ± 12,69 ^{bcd}	2,31 ± 0,63 ^{defg}	16,00 ± 3,45 ^{cd}	8,09 ± 0,13 ^{kl}	89,44 ± 6,34 ^a	2,98 ± 0,25 ^{bode}
	30 DAH	0 day	15,78 ± 1,03 ^{abcd}	8,59 ± 0,08 ^{jk}	66,66 ± 7,50 ^e	2,51 ± 0,60 ^{cde}	17,05 ± 2,52 ^{bc}	8,88 ± 0,15 ^h	63,33 ± 15,41 ^{de}	2,57 ± 0,50 ^{def}
		5 days	13,95 ± 1,63 ^{defg}	8,05 ± 0,05 ^{mno}	85,00 ± 6,12 ^{abc}	3,02 ± 0,49 ^{abc}	16,14 ± 3,03 ^{cd}	8,28 ± 0,07 ^{jk}	80,00 ± 9,35 ^c	2,85 ± 0,55 ^{cde}
		10 days	12,92 ± 1,45 ^{efghij}	7,24 ± 0,06 ^p	85,55 ± 8,07 ^{ab}	3,25 ± 0,44 ^{ab}	15,94 ± 2,99 ^{cd}	7,71 ± 0,26 ^{no}	88,88 ± 6,00 ^a	3,18 ± 0,33 ^{abc}
	60 DAH	0 day	17,58 ± 1,44 ^a	7,21 ± 0,10 ^q	71,11 ± 10,54 ^d	2,69 ± 0,60 ^{bcd}	21,61 ± 0,91 ^a	8,25 ± 0,07 ^{jk}	82,77 ± 10,63 ^b	2,86 ± 0,48 ^{cde}
		5 days	17,10 ± 1,49 ^{ab}	6,65 ± 0,19 ^r	90,55 ± 10,73 ^{ab}	3,50 ± 0,52 ^a	19,83 ± 0,19 ^{ab}	7,83 ± 0,06 ^{mn}	86,66 ± 11,45 ^a	3,37 ± 0,55 ^{ab}
		10 days	15,30 ± 1,81 ^{bcd}	6,11 ± 0,13 ^s	93,88 ± 6,00 ^a	3,54 ± 0,39 ^a	19,78 ± 0,39 ^{ab}	6,98 ± 0,07 ^p	92,77 ± 7,12 ^a	3,55 ± 0,44 ^a
Statistics		F	13,68	273,19	20,64	15,71	7,93	238,57	20,15	26,32
		P	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001

¹ For each parameter, values with the same letters are statistically equal

Table 6: Combined effects of fruit age, fruit pre-storage duration and fermentation duration on seedling vigor in both cultivars of *Lagenaria siceraria*

Fruit age at harvest	Fruit pre-storage duration	Seed fermentation duration	Round fruited cultivar				Blocky fruited cultivar			
			EmP	ESI (Sd/d)	SSL (mm)	DWS (g)	EmP	ESI (Sd/d)	SSL (mm)	DWS (g)
30 DAA	0 DAH	0 day	16,11 ± 10,54 ^g	0,32 ± 0,10 ^m	59,39 ± 14,25 ^p	0,19 ± 0,07 ⁱ	13,33 ± 9,01 ^j	0,23 ± 0,08 ^j	64,01 ± 8,48 ^o	0,19 ± 0,07 ⁱ
		5 days	28,88 ± 8,20 ^f	0,63 ± 0,18 ^m	65,35 ± 11,09 ^o	0,24 ± 0,09 ^{hij}	21,66 ± 14,57 ^j	0,37 ± 0,16 ^{kl}	71,92 ± 9,16 ⁿ	0,24 ± 0,08 ^{jk}
		10 days	40,55 ± 16,09 ^f	0,68 ± 0,47 ^m	69,07 ± 15,20 ^{no}	0,26 ± 0,08 ^{ghi}	35,55 ± 23,10 ⁱ	0,45 ± 0,10 ^{kl}	73,11 ± 14,95 ⁿ	0,26 ± 0,07 ^{ji}
	30 DAH	0 day	35,55 ± 19,59 ^f	1,10 ± 0,42 ^{ghijk}	74,45 ± 11,03 ^m	0,22 ± 0,08 ^{kl}	22,22 ± 10,34 ⁱ	0,56 ± 0,21 ^{jk}	71,42 ± 11,90 ⁿ	0,22 ± 0,07 ^k
		5 days	52,77 ± 22,09 ^e	1,07 ± 0,76 ^{hijk}	71,62 ± 16,41 ^{mn}	0,23 ± 0,09 ^{ij}	42,22 ± 22,09 ^{hi}	0,78 ± 0,34 ^{hi}	84,58 ± 10,41 ^{kl}	0,27 ± 0,06 ^{ij}
		10 days	54,44 ± 26,15 ^e	1,35 ± 0,62 ^{efghi}	77,33 ± 12,94 ^{kl}	0,27 ± 0,09 ^{gh}	50,55 ± 24,16 ⁱ	1,03 ± 0,44 ^{gh}	81,65 ± 9,65 ^m	0,30 ± 0,05 ^{fg}
	60 DAH	0 day	40,00 ± 9,35 ^f	0,98 ± 0,37 ^{kl}	85,85 ± 11,56 ^{gh}	0,20 ± 0,09 ^{kl}	44,44 ± 9,27 ^{hi}	0,76 ± 0,49 ^{hij}	87,55 ± 4,94 ^{ij}	0,22 ± 0,08 ^k
		5 days	56,66 ± 11,18 ^e	1,31 ± 0,49 ^{fghij}	88,89 ± 7,88 ^{fg}	0,27 ± 0,09 ^{gh}	47,77 ± 13,01 ^{fghi}	1,14 ± 0,38 ^g	92,12 ± 4,54 ^h	0,27 ± 0,05 ^{ij}
		10 days	61,66 ± 15,41 ^e	1,41 ± 0,70 ^{efgh}	90,17 ± 6,17 ^{ef}	0,29 ± 0,08 ^{efg}	60,00 ± 19,36 ^{ef}	1,32 ± 0,44 ^{efg}	91,49 ± 4,48 ^h	0,29 ± 0,06 ^{ghi}
40 DAA	0 DAH	0 day	39,44 ± 11,02 ^f	0,75 ± 0,28 ^{kl}	68,27 ± 9,49 ^o	0,23 ± 0,06 ^{ij}	41,11 ± 20,58 ^{hi}	0,77 ± 0,26 ^{hi}	74,10 ± 13,40 ⁿ	0,26 ± 0,08 ^{ij}
		5 days	52,22 ± 11,21 ^e	0,94 ± 0,51 ^{kl}	79,47 ± 14,23 ^k	0,25 ± 0,08 ^{hij}	58,88 ± 11,66 ^{efg}	1,16 ± 0,21 ^{fg}	85,62 ± 6,03 ^{jk}	0,27 ± 0,07 ^{ij}
		10 days	63,88 ± 15,96 ^e	1,15 ± 0,39 ^{ghij}	83,41 ± 10,46 ^{hi}	0,28 ± 0,08 ^{fgh}	67,22 ± 12,27 ^{de}	1,21 ± 0,28 ^{efg}	80,94 ± 14,07 ^m	0,31 ± 0,08 ^{efg}
	30 DAH	0 day	53,88 ± 14,95 ^e	1,32 ± 0,39 ^{fghij}	77,43 ± 13,21 ^{kl}	0,29 ± 0,09 ^{ef}	60,00 ± 12,74 ^{ef}	1,31 ± 0,45 ^{efg}	84,33 ± 8,98 ^j	0,29 ± 0,05 ^{ghi}
		5 days	68,33 ± 7,50 ^{cd}	1,32 ± 0,47 ^{fghij}	83,07 ± 13,75 ^{hi}	0,28 ± 0,08 ^{fgh}	68,33 ± 7,07 ^{cd}	1,59 ± 0,39 ^{def}	90,42 ± 4,74 ^h	0,31 ± 0,06 ^{efg}
		10 days	78,88 ± 8,57 ^{bc}	1,72 ± 0,16	85,75 ± 12,89 ^{gh}	0,32 ± 0,08 ^{cde}	75,55 ± 8,45 ^{bcd}	1,64 ± 0,30 ^{cd}	91,99 ± 7,47 ^h	0,35 ± 0,07 ^{cd}
	60 DAH	0 day	61,11 ± 19,00 ^e	1,47 ± 0,19 ^{cdefg}	92,02 ± 5,56 ^{de}	0,31 ± 0,07 ^{de}	66,66 ± 13,91 ^{de}	1,61 ± 0,52 ^{de}	94,36 ± 5,47 ^g	0,31 ± 0,06 ^{efg}
		5 days	80,55 ± 15,50 ^{ab}	1,82 ± 0,25 ^{abcd}	94,38 ± 5,28 ^d	0,30 ± 0,07 ^{ef}	76,11 ± 9,93 ^{bcd}	1,79 ± 0,26 ^{bcd}	97,99 ± 5,54 ^f	0,32 ± 0,07 ^{ef}
		10 days	85,55 ± 6,34 ^{ab}	1,80 ± 0,41 ^{bode}	99,39 ± 8,41 ^c	0,33 ± 0,06 ^{bcd}	82,77 ± 12,52 ^{ab}	2,04 ± 0,40 ^{ab}	100,06 ± 6,23 ^e	0,36 ± 0,07 ^{cd}
50 DAA	0 DAH	0 day	60,00 ± 6,12 ^e	1,32 ± 0,58 ^{fghij}	82,43 ± 7,42 ^{ij}	0,29 ± 0,10 ^{ef}	46,66 ± 7,07 ^{ghi}	1,31 ± 0,36 ^{efg}	88,93 ± 4,02 ^j	0,29 ± 0,10 ^{ghi}
		5 days	72,22 ± 7,12 ^c	1,44 ± 0,36 ^{defgh}	88,70 ± 7,84 ^{fg}	0,27 ± 0,08 ^{gh}	66,66 ± 9,35 ^{de}	1,55 ± 0,44 ^{def}	97,39 ± 5,94 ^f	0,30 ± 0,07 ^{fg}
		10 days	79,44 ± 11,84 ^{bc}	1,58 ± 0,24 ^{cdef}	89,52 ± 5,63 ^{ef}	0,32 ± 0,10 ^{cde}	83,88 ± 8,20 ^{ab}	1,94 ± 0,12 ^{abc}	96,07 ± 4,41 ^{fg}	0,37 ± 0,08 ^{ab}
	30 DAH	0 day	65,55 ± 7,68 ^d	1,61 ± 0,34 ^{cdef}	89,22 ± 7,65 ^{ef}	0,32 ± 0,09 ^{cde}	60,55 ± 16,09 ^{ef}	1,63 ± 0,29 ^{cd}	96,04 ± 5,63 ^{fg}	0,34 ± 0,09 ^{de}
		5 days	82,77 ± 5,06 ^{ab}	1,85 ± 0,29 ^{abc}	94,03 ± 8,64 ^d	0,34 ± 0,08 ^{bc}	77,22 ± 7,54 ^{bcd}	1,81 ± 0,34 ^{bcd}	102,64 ± 4,69 ^d	0,39 ± 0,06 ^a
		10 days	84,44 ± 7,26 ^{ab}	2,01 ± 0,26 ^{ab}	97,86 ± 5,72 ^c	0,35 ± 0,08 ^b	86,11 ± 6,97 ^{ab}	2,01 ± 0,15 ^{ab}	104,92 ± 6,35 ^c	0,36 ± 0,08 ^{cd}
	60 DAH	0 day	68,33 ± 10,30 ^{cd}	1,56 ± 0,43 ^{cdef}	98,68 ± 7,78 ^c	0,34 ± 0,07 ^{bc}	80,55 ± 10,73 ^{abc}	1,8 ± 0,30 ^{bcd}	104,00 ± 7,22 ^c	0,37 ± 0,09 ^{ab}
		5 days	86,66 ± 10,00 ^{ab}	2,06 ± 0,39 ^{ab}	105,35 ± 8,18 ^b	0,38 ± 0,07 ^a	84,44 ± 11,02 ^{ab}	2,03 ± 0,28 ^{ab}	110,57 ± 6,47 ^b	0,38 ± 0,11 ^{ab}
		10 days	92,22 ± 6,18 ^a	2,18 ± 0,23 ^a	110,43 ± 9,24 ^a	0,37 ± 0,08 ^a	90,55 ± 7,26 ^a	2,16 ± 0,20 ^a	113,90 ± 9,93 ^a	0,36 ± 0,10 ^{cd}
Statistics		F	20,19	13,68	133,84	24,23	20,99	11,00	203,20	24,23
		P	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001	<0,001

¹ For each parameter, values with the same letters are statistically equal

DISCUSSION

The agricultural productivity depends mainly on the quality of sown seeds (Al-Maskri *et al.*, 2004). The performance of *Lagenaria siceraria* seeds, resulting from the age of fruits, the durations of pre-storage and fermentation, was evaluated in this study. Seed aptitude to germinate and produce vigorous seedlings was tested. The results showed that these three factors considered individually or simultaneously, significantly influenced the seed viability and seedling vigor in this oleaginous species. Precocious seeds (30 DAA) presented, at least, viability. This relatively high viability (approximately 50%) indicates that physiological maturity of the first seeds began before 30 DAA. Moreover, delaying harvest time from 30 to 50 DAA, lead to significant improvement of seed viability through the increase of germination percentage from 42.77 ± 21.07 to $78.45 \pm 13.33\%$. Nerson and Paris (1988) reported some viable seeds in two cucurbits crops through the germination percentage of seeds harvested at 28 DAA which was 35% for melon (*Cucumis melo* L.) and 75% for watermelon (*Citrullus lanatus* (T.) Matsum. and Nakai). These researchers observed an improvement of seed viability through the increase of the germination percentage from 35 to 75% for melon and from 75 to 100% for watermelon when the age of harvest, was delayed from 28 to 49 DAA. Seed viability improvement following fruit growing time (corresponding to its age at harvest) could be explained by a seed continuation to accumulate dry matter while being dehydrated (Demir & Mavi, 2004). Seed dry matter accumulation can be justified by the increase in seed dry weight that we observed. In addition, the increase in fruit wet weight can support this matter accumulation of seeds. About seed dehydration during their formation, Miklic *et al.* (2006) observed in sunflower a very high decrease in seed moisture content (from 80 to 20 %) with an improvement of their germinability (from 0 to 99 %) when the fruit harvest time is delayed from 10th to 35th DAA. According to Nerson (2002), the moisture content of cucurbits' seeds declines considerably during their development. Moreover, we observed changes on seed external color with delayed harvest time. Similar results were obtained by Sangakkara (1995) studying papaya (*Carica papaya*, L.) seed ripeness. Seed viability was very significantly improved during the pre-storage of fruits through the germination percentage and the germination speed index. Seed viability improvement, highly related to a profit of weight and dehydration, could be explained by a completion of physiological

maturation of some immature seeds in the harvested fruits. Indeed, during pre-storage, the remaining immature seeds would accumulate nutrients temporarily stored in the flesh of harvested fruits. Thus, the number of physiologically mature seeds would be higher in more pre-stored fruits (60 DAH) than those of less pre-stored ones (30 DAH). These mature seeds number in pre-stored fruits would be higher than that of non pre-stored fruits (0 DAH: control). A beneficial effect of fruits pre-storage on the improvement of seeds germinability in watermelon and melon had already been mentioned by Nerson *et al.* (1985), Nerson (1991). According to Giovannoni (2001), as cucurbits are climacteric species, their ripening can continue during post-harvest treatment (pre-storage) through increased respiration and ethylene biosynthesis. The occurrence of postharvest maturity in *L. siceraria* is phenomenon widely observed in cultivated cucurbits (Zong *et al.*, 1995; Beaulieu *et al.*, 2004; Olson *et al.*, 2009).

Seed fermentation allowed an increase in their germination percentage and germination rate in *Lagenaria siceraria*. This improvement of seed viability is related to the decrease of their moisture content. Fermentation is a process which allows the physiological maturation of immature seeds, at harvest, to complete (Nerson & Paris, 1988; Demir & Mavi, 2004). This completion increased the rate and the percentage of germination (Edwards *et al.*, 1986). However, seeds dry weight constancy during fermentation would be related to a suspension of nutrients accumulation, probably due to the deterioration of flesh by the micro-organisms. Viability increase, following fermentation duration, could have two explanations: a completion of physiological maturation or a strong lifting of seed germination inhibitor. About first hypothesis, the micro-organisms responsible for fermentation would have sufficient time (from 0 to 10 days) to proliferate and enhance seed germinability. Zheng and Shetty (2000) observed for pea (*Pisum sativum*) an increase of germination rate when the seeds were mixed with fungi (*Trichoderma spp.*). This increase of microbial activity following fermentation duration would have allowed some remaining immature seeds to complete their maturation (Nerson, 2007). Kermode and Bewley (1985) observed the synthesis of some germinative proteins during the dehydration of seed (as it is the case during fermentation). These proteins (L-leucyl- β -naphthylamidase and isocitrate lyase) are implied in the

production of enzymes responsible for the seed reserves hydrolysis during germination of *Ricinus communis* L. Their production would be more significant in fermented seeds (5 and 10 days) than in unfermented ones (F0). About the second hypothesis, Nerson and Paris (1988) suggested the existence of a water-soluble inhibitor in immature seeds of watermelon, which effect would be neutralized by fermentation. The neutralization of this inhibitor, cucurbitacine (Nerson, 1991), would be more significant in seeds fermented longer (10 days: F10).

Whatever was the fruits age at harvest (30, 40 and 50 DAA), pre-storage improved their seed viability. This improvement could mean that physiological maturity of all the seeds would not be completed even at 50 DAA. According to Nerson (1991), the embryo of immature seed continues its development in the harvested fruit during pre-storage; what explains the improvement of germinability. Moreover, for all pre-stored fruits (30 and 60 DAH), fermented seeds (especially 10 days) showed the highest viability compared to unfermented seeds (F0). Seed viability improvement by fermentation was already reported (Nerson, 2007). It could mean that even in pre-stored fruits, some seeds still remain immature. Completion of physiological maturation of these seeds would have allowed improvement of viability parameters. Finally, the combination of fruit age, fruit pre-storage duration and seed fermentation provided the best results of seed germination and seedling vigor. The positive effect resulting from this interaction was also tested by Edwards *et al.* in cucurbits (1986).

Usually, seeds viability and seedling vigor are correlated (Cantliffe, 1998). In both cultivars RFC and BFC, all the seedling vigor parameters (emergence

percentage, emergence speed index, seed shoot length and seedling dry weight), showed the same variations as those of viability. Indeed, these parameters showed the highest values for the seedlings resulting from seeds tardily harvested (50 DAA), longer pre-stored (60 DAH) and fermented during 10 days (F10). The most vigorous seedlings are produced by the most viable seeds. The fact that the first germinated seedlings are also the first to emerge confirms the correlation between seed viability and seedlings vigor. According to Hamman *et al.* (2002), poor emergence is generally not due to a decrease of viability but rather to a decrease of seedlings growth: what prevents them from emergence on the ground surface.

In conclusion fruits harvested tardily provide seeds with the highest viability and the most vigorous seedlings than those harvested precociously. Pre-storage of fruits allows the immature seeds, at harvest, to complete their physiological maturation. This improves their germinability and seedling emergence. All the seeds fermented longer show the best germinability. It indicates that fermentation, a part from facilitating seed extraction, improves their viability. Following the positive interaction between fruit age at harvest, durations of pre-storage and fermentation, and the similar reaction of both cultivars RFC and BFC of *Lagenaria siceraria*, fruits must be harvested tardily (50 DAA), longer pre-stored (60 DAH) and longer fermented (10 days). Variations of seed viability and seedling vigor analyzed in this study were closely associated to those of fruit wet weight, seed color, seed dry weight and moisture content. Fruit weight and seed color, dry weight and moisture content can be used as indicators of maturity in both cultivars of *Lagenaria siceraria*.

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