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Effect of harvesting time on seed quality of two bell pepper cultivars (*Capsicum annuum*)

Efecto del momento de cosecha sobre la calidad de semilla en dos cultivares de pimiento (*Capsicum annuum*) tipo cuatro cascos

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Abstract

The maturation status of seeds is fundamental at the harvest time to get high rates of germination and vigor. The aim of this study was to determine the effect of harvest time on the quality of pepper seeds. The trial was conducted in two bell type cultivars: Fyuco INTA and Lungo INTA. Fruits were harvested at 4, 5, 6, 7, 8 and 9 weeks after anthesis. Starting from the eighth week the average seed weight stabilized, coincidently with the change in the fruit colour to red. Germination percentage increased linearly as it increased the age of the seed. The electrical conductivity of the leachate was also stabilized in seeds got from fruit harvested from 8 weeks after anthesis, showing that they had reached physiological maturity. The results demonstrate to harvest the fruits of pepper cv Fyuco INTA and Lungo INTA with over 8 weeks of age after anthesis to reach high quality seeds.

Keywords
physiologica maturity • germination • vigor

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Resumen

El nivel de maduración de la semilla es fundamental al momento de cosecha para obtener elevadas tasas de germinación y buen vigor. El objetivo de este trabajo fue determinar el efecto del momento de cosecha sobre la calidad de semillas de pimiento. El ensayo se realizó en dos cultivares tipo cuatro cascos: Fyuco INTA y Lungo INTA. Se cosecharon frutos a las 4, 5, 6, 7, 8 y 9 semanas después de la antesis. A partir de la octava semana el peso promedio de las semillas se estabilizó, coincidiendo con el cambio de coloración del fruto a rojo. La germinación se incrementó linealmente conforme aumentó la edad de la semilla. La conductividad eléctrica del sobrenadante se estabilizó también, en las semillas obtenidas de los frutos cosechados a partir de las 8 semanas después de la antesis, indicando que estas habían alcanzado la madurez fisiológica. Los resultados demuestran la importancia de cosechar los frutos de pimiento de estas cultivares con más de 8 semanas de edad para lograr semillas de calidad.

Palabras claves
madurez fisiológica • germinación • vigor

Introduction

The pepper is grown in all countries of the world in temperate and warm areas (22). Its production includes fresh and dried fruits. According to FAO (2016) pepper is the fifth global horticultural crop in terms of area and the eighth in terms of production volume. In Argentina, in 2008 it accounted for 2% of the production (weight) of fresh vegetables (16). Many authors have reported difficulty in achieving a fast and efficient emergency in pepper seeds (23, 27). The low quality of the seed can affect significantly the productivity because affects crop establishment (13).

One factor that can changed seed quality is the stage of fruit maturation at the time of the harvest. The physiological maturity of the fruit is essential to achieve high rates of germination and good vigor (21, 32). During seed development we can distinguish diferents stages in which the ripening of the seed is fundamental (18). During embryogenesis the seed accumulated proteins, carbohydrates and lipids (7), used by the embryo during germination process (3, 12).

The seed quality increases with the time after anthesis, reaching the maximum viability and vigour at the physiological maturity, to then begin a process of aging and deterioration (18, 25). Physiological maturity is related to reserve accumulation in optimal amounts (7, 26), but also with the acquisition of tolerance to desiccation (2). The time of harvest to get the best quality of seed depends on the species, cultivar and even the production system (13).

Previous studies on the effect of fruit maturity on pepper seed quality indicate that the highest percentages of germination was got in a over ripe mature fruits (6, 8). In *Capsicum frustescens*, Edwards & Sundström (1987) conclude similar results. However in these works, the authors do not related seed maturity with the age. The objective of the present study was to determine the effect of the age of bell pepper seed on quality to establish the optimum harvest time.

Materials and methods

Plant material and crop management

We used two bell pepper cultivars: Fyuco INTA and Lungo INTA. Both are open pollinated cultivars, resistant to *Phytophthora capsici* Leonian and used for fresh market or dehydrated industry (17). The plants were produced in trays of 416 cell polyurethane and transplanted into field 80 days after sowing (19/12/2014), with a of 33,000 plants per hectare (0.85 x 0,35 m) density, in the experimental field of INTA in San Juan, Argentina (latitude: 31°37' S and longitude 68°32' W, altitude: 618.23 meters above the sea level). Irrigation was carried out by a pressurized system using a tape of 3 droppers/meter with a flow rate of 3.3 l h^{-1} m^{-1} to 1 atm (Naan peace) and fertigated with 100 kg ha^{-1} of nitrogen during the entire crop cycle.

Age and seed extraction

To determine the age of the seed, during anthesis, the flowers were marked with different paper labels. In this way it was possible to harvest fruits with 4, 5, 6, 7, 8 and 9 weeks of age after anthesis. The seeds were extracted manually and dried in a closed container with silica gel at 30°C and 0% HR during 2 days. They were subsequently preserved in containers sealed at 20°C and 20% of HR for 30 days (year 0) for the evaluation of leachate electrical conductivity and germination. After a year (year 1), we repeated the tests to evaluate the effect of the time on the quality of the seed in them different treatments.

Dry weight

The extracted seeds were dehydrated in an oven at 70° C for 48 hours and weiged in a precision scale. Four replicates of 100 seeds were used for each cultivar and harvest age.

Leachate electrical conductivity

A hundred seeds (distributed in four replicates of 25 seeds) of both cultivars and harvest age were soaked in 30 ml of distilled water at 25°C, The conductivity was measured after four hours with a conductivity meter (Oacton Instruments, Vernon Hillls, Ill) and expressed as μS cm^{-1} g^{-1} of seed.

Germination tests

Four replicates of twenty-five seeds for each varieties and harvest age were sown in Petri dishes with germination paper (Munktell filter 1700, Sweden) moistened with 4 ml of distilled water (20) and were placed in a germination chamber scheduled at 25/15°C (16/8 h) for 14 days. The seed was considered germinated when the radicle emerged 1 mm. Germination was controlled every 24 h and calculated germination percentage and emergency rate index (ERI) (28).

Statistical analysis

All evaluations were conducted as a randomized design, with treatments replicated four times. Percentage data were converted and analyzed as the square root arc sine transformation. Means were compared with a least significant difference test (LSD) at P=0.05. Linear regression analyses were performed between age of the seeds (WAA) with seed weight, percentage of germination, ERI and leachate electrical conductivity. The SAS statistical programme (SAS Institute, Cary, NC) was used for the analysis of the data.

Results

The increase in seed weight was similar in both cultivars in response to a linear trend (figure 1). The average seed weight was stabilized from the eighth week after anthesis, coincidently with the change of the fruit color from green to red (figure 2, page 71). The seeds of Cv Lungo showed a significantly less average dry weight than Cv Fyuco eight weeks after flowering (p=0.0021) .

The leachate electrical conductivity in both cultivars showed a significative negative linear regression with the age of the seed (figure 3, page 72). The highest conductivity values were obtained from seeds of 4 weeks age. From the 8th week after anthesis we observed a marked decrease in the loss of electrolytes in both cultivars, coinciding with the stabilization of the seed weight and the change of the fruit color.

The time of harvest (seed age) and cultivar significantly modified the germination percentage, also in the fresh seeds (Year 0) there was a significant interaction between Cv and seed age (table 1, page 72). The Cv Fyuco showed percentages of germination significantly above the Lungo INTA. After one year (year 1) only seed age significantly modified the germination percentage, reached the highest value (73%) 9 weeks after anthesis.

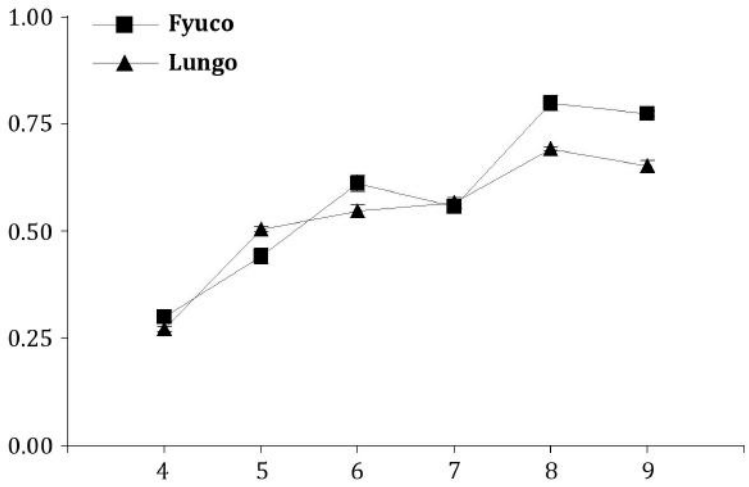

Figure 1. Relationship between seed age (WAA: week after anthesis) and the dry weight of the seeds in the two pepper cultivars evaluated. Lungo (y= 0.079+ 0.071(sdf) R^2= 0.79**) and Fyuco (y= -0.049+0.097(sdf) R^2= 0.87**).

Figura 1. Relación entre la edad (WAA: semanas después de antesis) y el peso seco de las semillas en las dos Cultivares de pimiento evaluados. Lungo (y= 0,079+ 0,071(sdf) R^2= 0,79**) y Fyuco (y= -0,049+0,097(sdf) R^2= 0,87**).

Error bars represent standard deviations / Las barras de error representan desviaciones estándar.

Figure 2. Fruits of Cvs Fyuco INTA and Lungo INTA 6 and 8 weeks after anthesis (WAA).
Figura 2. Evolución del color de frutos de los Cvs Fyuco INTA y Lungo INTA 6 y 8 semanas después de antesis (WAA).

In Cv Fyuco a significant linear response was observed when we relate the germination percentage with the seed age (figure 4, page 73) in new seeds (Year 0) and old seed (Year 1) (figure 4, page 73). Seeds extracted 4, 5, 6 and 7 weeks after the anthesis showed germination percentages lower than 40% while the seeds harvested 8 and 9 weeks after anthesis the percentages ranged between the 50 and 80%. Seeds from of one year age, in all harvest time, showed lower germination percentages, but with the same trend as in the new seeds. In Cv Lungo the similar response was observed, however the germination percentages were lower compared to Cv Fyuco (figure 5, page 73).

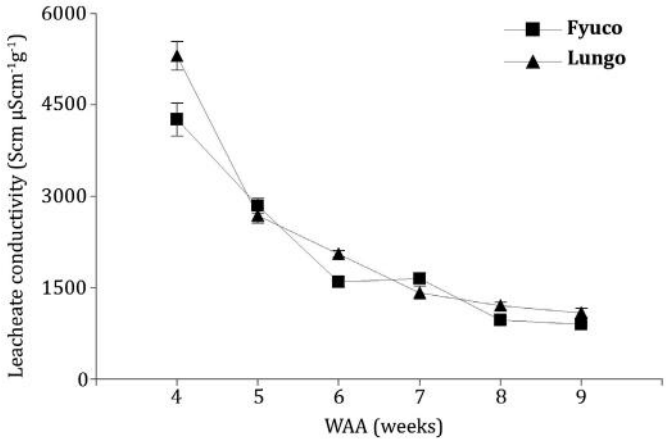

Figure 3. Relationship between seed age (WAA) and leachate electrical conductivity ($\mu Scm^{-1}g^{-1}$) of bell pepper cultivars evaluated. Lungo (y = 7152.38 - 747.33 (WAA) R^2 = 0.75 **); Fyuco (y = 6193.42 - 639.15 (WAA) R^2 = 0.82 **).

Figura 3. Relación entre la edad de la semilla (WAA) y la conductividad del sobrenadante en las cultivares de pimiento evaluados. Lungo (y= 7152,38 - 747,33 (WAA) R^2= 0,75**); Fyuco (y= 6193,42 - 639,15 (WAA) R^2= 0,82**).

Error bars represent standard deviations. / Las barras de error representan desviaciones estándar.

Table 1. Effect of cultivar (Cv) and seed age (WAA) on germination percentage and germination rate (ERI) in two bell pepper (Cv Lungo INTA and Fyuco INTA) after seed extraction (Year 0) and one year later (Year 1).

Tabla 1. Efecto del cultivar y edad de la semilla sobre el porcentaje de germinación y velocidad de germinación (ERI) en pimiento Cvs. Fyuco INTA y Lungo INTA luego de la extracción de la semilla (Año 0) y un año después (Año 1).

	Year 0		**Year 1**	
	Germination (%)	**ERI**	**Germination (%)**	**ERI**
Fuente de variación	Significancy		Significancy	
Cultivar (Cv)	< 0.0001	< 0.0001	0.4988	0.0106
Seed Age (WAA)	< 0.0001	< 0.0001	< 0.0001	< 0.0001
Cv x WAA	0.0332	< 0.0001	0.0772	0.0098
Cv	Means			
Fyuco INTA	38.33	4.25	24.60	2.77
Lungo INTA	27.33	2.11	22.60	1.87
LSD	5.06	0.51	5.90	0.67
SDF	Means			
4 Weeks	0.00	0.00	0.00[a]	0.00
5 Weeks	0.00	0.43	0.00[a]	0.00
6 Weeks	20.00	1.96	5.50[a]	0.54
7 Weeks	36.50	3.12	25.50[b]	2.27
8 Weeks	59.00	5.72	38.00[c]	4.00
9 Weeks	76.00	7.83	73.00[d]	7.13

Column means not followed by the same letter differ significantly LSD test (5%).

Medias de las columnas no seguidas por la misma letra difieren significativamente LSD (5%).

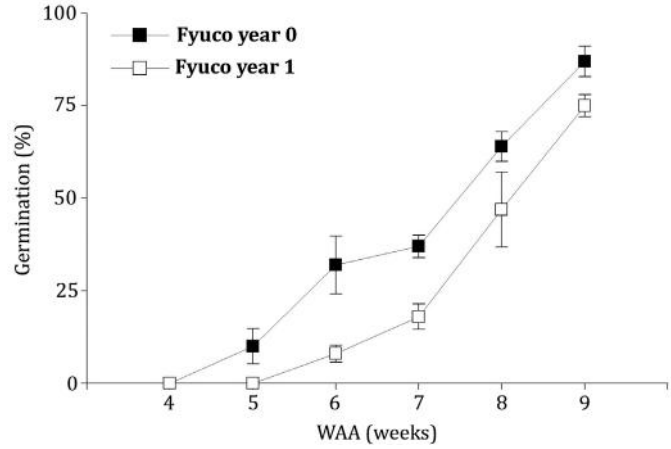

Figure 4. Relationship between seed age (WAA) and germination percentage in fresh seeds (Year 0) of Cv Fyuco INTA in (y = -73.47 + 17.2 (WAA) R^2 = 0.90 **) and one year old seeds (Year 1) (y = -73, 02 +15.2 (WAA) R^2 = 0.79 **).

Figura 4. Relación entre la edad de la semilla (WAA)y el porcentaje de germinación en la Cv Fyuco INTA en semillas frescas (y= -73,47+17,2 (WAA) R^2= 0,90**) y semillas de un año (y=-73,02 +15,2 (sdf) R^2= 0,79**).

Error bars represent standard deviations. / Las barras de error representan desviaciones estándar.

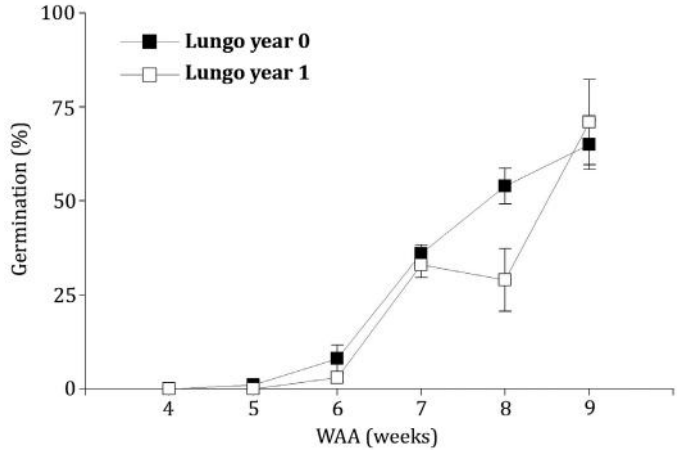

Figure 5. Relationship between seed age (WAA) and germination percentage in fresh seeds (Year 0) of Cv Lungo INTA, (y = -67.75+ 14.63 (WAA) R^2 = 0.88 **) and one year old seeds (Year 1) (-64.99 + 13.48 (WAA) R^2 = 0.70 **).

Figura 5. Relación entre la edad de la semilla (WAA) y el porcentaje de germinación en la Cv Lungo INTA, en semillas frescas (y= -67,75+ 14,63 (WAA) R^2= 0,88**) y semillas de un año (-64,99+13,48 (WAA) R^2= 0,70**).

Error bars represent standard deviations. / Las barras de error representan desviaciones estándar.

As in the analysis of germination percentage, the germination speed, expressed as ERI, showed a significant differences between cultivars, seed age and its interaction (table 1, page 72). The maximum values of ERI corresponded to the Cv Fyuco INTA (figure 6, page 75). The seeds harvested 9 weeks after anthesis germinated significantly more quickly than the seeds harvested 4, 5, 6, 7and 8 weeks after anthesis.

Discussion

The results observed in this study confirm that the age of the fruits, at the time of seed extraction is essential to obtain good quality seed (8, 11, 23, 29).

The seeds of both bell pepper cultivars (Fyuco INTA and Lungo INTA) extracted from fruits of 7 or less weeks after anthesis, do not accumulate enough metabolic reserve and also do not develop physiological mechanisms that allow seeds to tolerate dessication.

High values of electrical conductivity in seeds harvested between 4 and 7 weeks are related to the inability of the cell membranes to control the loss of metabolits (2, 30). Cell membranes undergo changes in their molecular organization during the process of physiological seed ripeness, allowing to tolerate dessecation. Quickly recover of membranes integrity allows selective permeability of ions and solutes from the cell (2).

In pepper the electrical conductivity of the supernatant test has proven to be the most efficient method to determine physiological quality of seeds (1) since it correlates strongly with the germination percentages.

Panayotov & Aladhzadhzian (1999) reported that the largest increases in pepper seed dry weight were produced between 20 and 45 days after flowering and the physiological maturity 70 days after anthesis, where the leachatte conductivity levels are stabilize.

The results obtained in this experiment showed that the seeds dry weight, in both cultivars, reached the highest value 8 weeks after anthesis however the highest germination percentage was observed 9 weeks after anthesis. In several species such as pepper (23), eggplant (10), tomato (9) and beans (5) the maximum dry matter acumulation and seed quality are not coincident.

The germination percentages and ERI responded linearly regarding the age of seeds. These results are coincident with those obtained by other authors (15, 23) where the highest germination percentages was observed in fruits harvest of 8 to 10 weeks of age.

The Cv Fyuco presented higher and faster germination than Cv Lungo. These differences between cultivars have been informed by other authors and usually are associated to different optimal temperatures for germination (8).

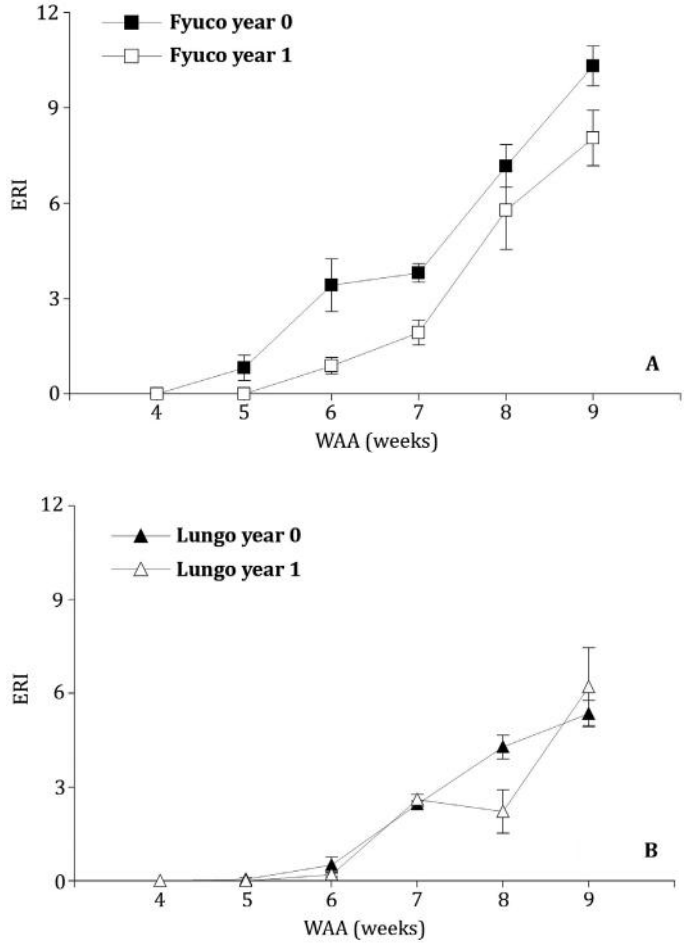

Figure 6. Relationship between seed age (WAA) and ERI (Emergency Rate Index) in both bell pepper cultivars. A) Fresh seeds in Fyuco INTA (Year 0) (y = -8.93 + 2.03 (WAA) R^2 = 0.88 **) and one year old seed (Year 1) (y = -8.12 + 1.67 (WAA) R^2 = 0.76 **). B) Lungo INTA (y = -5.57+ 1.18 (WAA) R^2 = 0.88 **); and one year old seed (Year 1) (y = -5.57 + 1.15 (WAA) R^2 = 0.65 **).

Figura 6. Relación entre la edad de la semilla (WAA) y ERI (índice de velocidad de emergencia) en las dos cultivares de pimiento evaluados. A) en semillas frescas de Fyuco INTA (y= -8,93+2,03 (WAA)R^2= 0,88**) y en semillas de un año (y= -8.12+1.67 (WAA) R^2= 0,76**). B) Lungo INTA (y= -5,57+ 1,18 (WAA) R^2= 0,88**); y en semillas de un año (y= -5,57+1,15 (WAA) R^2= 0,65**).

Error bars represent standard deviations / Barras verticales representan error estándar.

Conclusions

The optimal timing of fruit harvest for seed production in both cultivars is coincident with the change of color of the fruit to red, wich is consistent with reported by other authors for other varieties of peppers (8, 11, 15, 27, 29).

The results demonstrate that the fruits should be harvest over 9 weeks after anthesis to get high seed quality in Cvs Fyuco INTA and Lungo INTA.

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