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EFFECT OF MONKEYS' DIGESTIVE TRACT AND SOME FACTORS ON *OPUNTIA TUNA* (L.) MILL. SEED GERMINATION IN THE SPECIAL RESERVE OF GUEUMBEUL (SENEGAL)

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ABSTRACT

Opuntia tuna is a resilient plant, used toprotect against wind and water erosion for the restoration of degraded soils. Its introduction, as a fence hedge, in the Special Reserve of Gueumbeul (Senegal), has led to a proliferation of the species in this area. This proliferation has drastically reduced the areas of predilection of the animals. This is aggravated by the lack of initiative in the processing of fruits and *cactus* snowshoes. In order to contribute to a better knowledge of the biology and good management of this plant, we proposed to evaluate the effect of transit in the digestive tract of monkeys and different treatments on the germination of *cactus* seeds. The results obtained showed that seeds transiting through the digestive tract of monkeys (SD), gave the highest germination rate (58.3%) compared to the germination rate (48.6%) of seeds directly extracted from fruits (SF). Soaking SD seeds in water for 24 hours greatly improved the germination rate to 68%. Scarification with sulfuric acid for 5 and 15 min and soaking in boiling water did not improve this rate.The direct seeding tests revealed that the sand substrate gives a better germination rate than the soil substrate for seeds kept at the ambient temperature.

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INTRODUCTION

The *cactus*, *Opuntia tuna*, also called prickly pear, is a plant native to the arid and semi-arid regions of Mexico. It has a great capacity to adapt to global warming and increasing risks of drought (FAO, 2017). The snowshoe, fruit and essential oils are widely used in food and cosmetics. Its introduction as a fence hedge in the Special Fauna Reserve of Gueumbeul has led to the proliferation of the species in this area. The fruits, which have a good nutritional value, are often consumed by animals, mainly the red monkey (*Erythrocebus patas*). These animals, considered as vectors, effectively contribute to the dissemination of *cactus* seeds. Among these vectors, the red monkey occupies the first place based on preliminary results (Diop 2011). Khardiata (2020) confirmed the important role of monkeys in the dissemination of *cactus* seeds. Indeed, by consuming these fruits, red monkeys disseminate the seeds present in their droppings in sites more or less favorable to germination. However, the seeds of *cactus* fruits are characterized by the hardness of their integument. Many authors such as Treca & Tamba (1997) and Gautier-Hion (1984) have reported the positive effect of passage through the digestive tract of animals on the lifting of integumentary dormancy. Therefore, in order to evaluate the effect of transit through the digestive tract of monkeys on the germination of *cactus* seeds, this work has been initiated. Other experiments such as scarification with sulfuric acid, soaking of seeds in water at ambient temperature or boiling were carried out. The effect of storage temperature on the viability and germination rate of *Opuntia tuna* seeds was also studied.

MATERIAL AND METHODS

Plant material: The plant material consists of a collection of types of seeds collected in 2019:

- blackish seeds extracted from the droppings of red monkeys(a, photo 1);
- light brown seeds extracted from the *cactus*fruits (b, photo 1).

For each type of seeds, a batch was preserved at ambient temperature and another batch at 4°C.

Tests of *Opuntia tuna* seeds germination

Effect of transit through the digestive tract on seed germination: Seeds extracted from *cactus* fruits (SF) and seeds extracted from monkey droppings (SD) stored at ambient temperature, were sown without prior treatment.

Effect of soaking seeds in water on germination: The seeds extracted from the fruits and those from the droppings were put in jars containing 150 ml of water for 24, 48 and 72 hours before sowing.

Effect of soaking in hot water on germination: SD and SF seeds were soaked in hot water (100°C) for 10 minutes.

Influence of sulfuric acid scarification on seed germination: The seeds from the fruits and those extracted from the droppings were soaked in 98% sulphuric acid for different duration (5, 15, 25 and 30 min); after they were rinsed three times in sterilized water before sowing.

Influence of storage temperature on seed germination: Seeds that were stored at room temperature (SF and SD) and those that were stored at 4°C (SFC and SDC) were used. These seeds were sown in a sandy substrate at room temperature.

Influence of the substrate on the seeds germination: Seeds stored at ambient temperature (SF and SD) were used. These were sown in two different substrates (sand and compost) at ambient temperature.

Experimental conditions: For experiments on the effects of transit through the digestive tract, soaking in water at ambient temperature or at 100°C and scarification, seeds are sown on sterile cotton placed in plastic small jars. For all these experiments, the germination temperature is 27°C in the dark room. For the all experiences, a total of 24 seeds were tested with a repetition number of 03.

Germination parameters: The germination record was done daily during two (02) months. Two parameters weremeasured (germination time and germination rate). We consider that a seed has germinated when there is emergence of the radicle.

-germination rate (%) $= \frac{total\ number\ of\ germinated\ seeds}{total\ number\ of\ sown\ seeds} \times 100$

- germination time : duration between the sowing date and the beginning of germination

Acclimatization: After germination, the seedlings were transferred to the plastic pots containing compost for hardening. The seedlings were kept under green house with proper watering for 02 months. The number of acclimated plants was 50.

Statistic treatments: The data analysis was performed with xlstat (version 2016). For the data that did not meet the criteria of normality and variance homogeneity, a non-parametric Kruskal-Wallis test at the 5% threshold was performed. Dunn's test (1964) was performed in order to make pair-wise comparisons and to classify the different types of groups according to the variable studied.

RESULTS

Effect of transit through the digestive tract on seed germination: Figure 1 shows the effect of seed transit through the digestive tract on seed germination. Analysis of the figure shows that the germination of the seeds extracted from the droppings, begins on the eleventh day after sowing. For the control seeds extracted from *cactus* fruits, germination started after 21 days.The germination rates of both types of seeds increasedwith time. The higher germination rate (58.3%) was noted in seeds that had undergone the effect of the digestive tract compared to the germination rate (50%) of seeds extracted from *cactus* fruits. However, there is no significant difference according to the Kruskal-Walllis test at the 5% threshold.

Effect of water Soaking Seeds on Germination: Figure 2 shows the influence of the soaking time on the seeds germination. In SD seeds, the highest germination rate (68%) was obtained with 24 hours soaking time after two months of sowing. However, for SF seeds, it was found that the germination rate (45%) was higher in control seeds. From these results, it is clear that water soaking combined with the effect of digestive transit, increases the germination rate of SD seeds. Also, the Kruskal-Wallis non-parametric test shows that there is a significant difference between the rate of germination of the two types seeds ($P<0,05$).

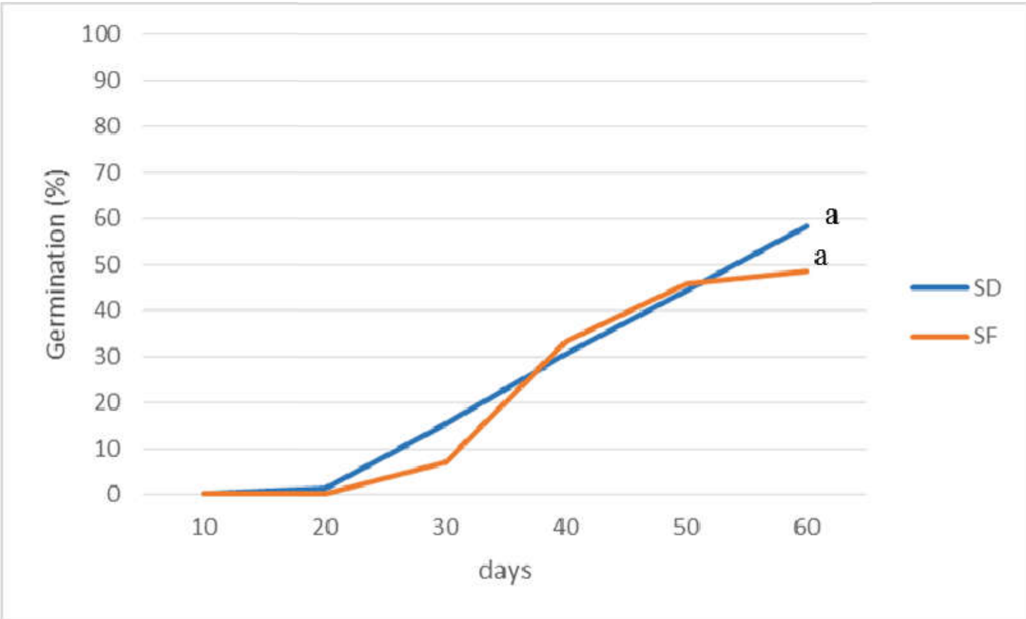
Effect of soaking in hot water on seed germination: The effect of soaking seeds in hot water on the germination of both seed types (SD and SF) has been illustrated in Figure 3. Analysis of the figure shows that the germination rate is higher in the SD control seeds. The hot water treatment has a negative effect on the germination time, which is 10 days in the SD seeds untreated and 40 days in the treated seeds. This treatment also significantly reduces the germination rate, which is almost zero with treated SD seeds. The same observations are also noted with SF seeds.

Influence of sulfuric acid scarification on seed germination: Figure 4 illustrates the effect of sulfuric acid scarification time on SD and SF seed germination. The analysis of the figure shows that the germination time is shorter in the control SD seeds than in the seeds treated with sulfuric acid during 5 and 15 min. The same observation is noted in SF seeds. For both categories of seeds, scarification has a depressing effect on the germination rate. However, for seeds extracted from fruits, we note a positive effect of scarification with a duration of 25 min. With SD seeds, the germination rate (60%) of the of the seeds untreated is significantly higher than the rates obtained with different scarification durations. For SF seeds, seeds untreated and seeds treated with sulfuricacid for 25 min, have relatively same germination rates (50%).

Influence of storage temperature on *Opuntia tuna* seed germination: The influence of storage temperature on germination of SD and SF seeds has been shown in Figure 5.

a b

Photo 01. Seeds extracted from the droppings of singes (a), seeds extracted from cactus fruits (b)



SD: Seeds extracted from the droppings of monkeys
SF: Seeds extracted from *cactus* fruits

Figure 1. Influence of digestive tract on *Opuntia tuna* seeds germination

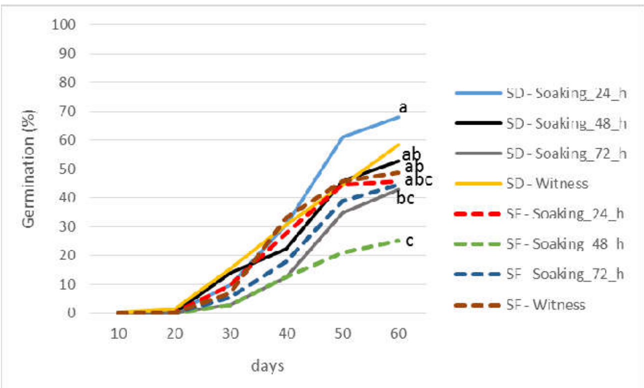


Figure 2. Effect of the soaking time on *Opuntia tuna* seeds germination

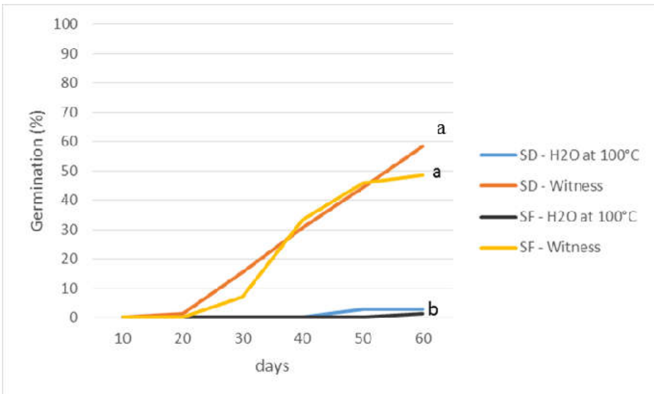


Figure 3. Effect of hot water on *Opuntia tuna* seed germination

The analysis of the results on the germination rates of the SD and SDC seeds, shows a clear difference on the delay and the germination rate. The SD seeds have a better germination rate (62.5%) after 2 months of sowing.For SF seeds, we have not noticedany difference in germination time. However, the germination rate (30%) is slightly higher in seeds that have been stored at room temperature.

Effect of the substrate on the *Opuntia tuna* seeds germination: The influence of the substrate on the germination of SD and SF seeds is shown in Figure 6. The results show that significantly higher germination rates were obtained with the soil substrate for both categories of seeds. We noteda germination rate of 60% for SD seeds and 30% for SF seeds after two months of sowing.

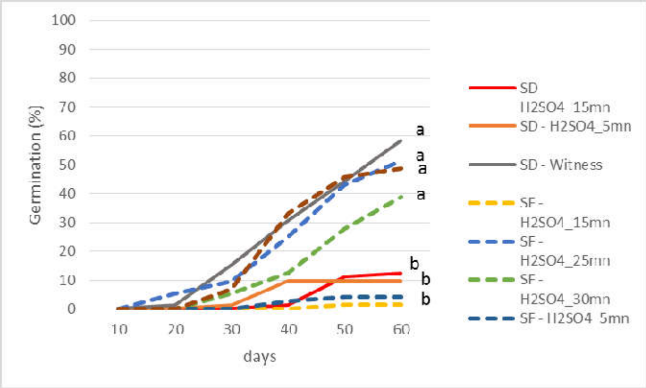
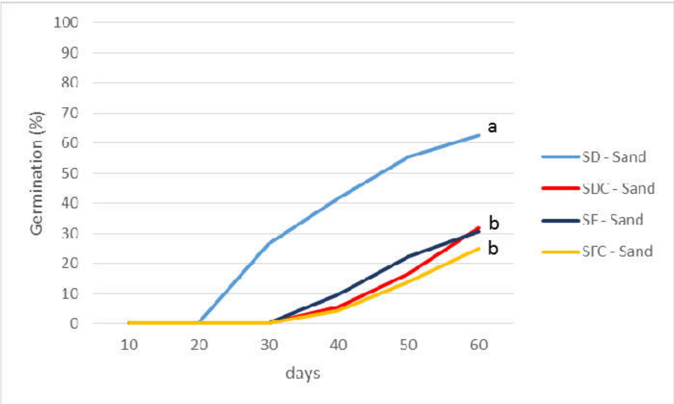


Figure 4. Effect of sulfuric acid scarification on *Opuntia tuna* seed germination



SD : Seeds extracted from the droppings of singes and stored at ambient temperature
SDC : Seeds extracted from the droppings of singes and stored at 4°C
SF : Seeds extracted from the *Opuntia tuna* fruits and stored at ambient temperature
SFC : Seeds extracted from the *Opuntia tuna* fruits and stored at4°C

Figure 5. Effect of storage temperature on seed germination

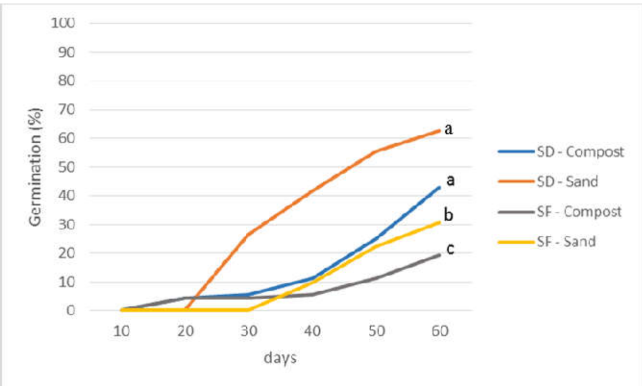


Figure 6. Influence of substrate on the *Opuntia tuna* seeds germination

Photo 2. Acclimatized *Opuntia tuna* plants

Acclimatization: After following the process of hardening and acclimatization, the seedlings transplanted in plastic pots containing the compost showed a successful acclimatization rate of 34%.

DISCUSSION

Germination tests carried out on *cactus* seeds revealed a high germination rate and a shorter germination time for seeds those had passed through the digestive tract. The positive effect on germination of seedspassing through the digestive tract of animals has been reported in many species (Hladik & Hladik, 2011; Tréca & Tamba, 1997; Gautier-Hion, 1984; Charles-Dominique, 1995). In contrast, Danthu (1996) reported no improvement of hard seed germination in many plant species (A. *niloticaadansonii*, *A. raddiana*, *A. senegal*, *A. seyal*, *B. rufescens*, *F. alb in ida*, *P. juliflora*),evenafter their passage through the digestive tract of ruminants. The germination process in SD seeds was improved by soaking them in water. Soaking, seeds ingested by red monkeys and the seeds extracted from *cactus* fruits, for 24, 48 and 72 hours in water, improved the germination rate. But the positive effect is much more noted in seeds extracted from the droppings; this could be related to a weakening of the integuments by the digestive juices, which would facilitate the entry of water and oxygen. Soaking would also eliminate certain chemical inhibitors of germination. Bourou & Braconnier (2010), in two varieties of Niébé (*Vigna unguiculata*), showed that pre-soaking in water improves the germination rate. Bamba *et al* (2018), on germination of *Pterocarpus erinaceus* seeds, revealed that soaking in water had a remarkable positive effect in improving germination rate. The low germination rate observed in seeds soaked in hot water (100°C), could be related to the heat sensitivity of the seeds. This negative heat effect on germination of some seeds was noted by Bamba *et al* (2018) on *Pterocarpus erinaceus* seeds. However, good results were obtained on *Prosopis africana* seeds (Ahaton *et al*., 2009).

Germination rates noted with sulfuric acid scarification of SD and SF seeds during 5 and 15 min are low. For SD seeds, this low rate would be related to a lethal action of the sulfuric acid for seeds whose seed coats have already been weakened by digestive juices; however, these same durations are not sufficient to weaken the SF seed embryo. Similar results were revealed by the work of Ahaton *et al* (2009) on *Prosopis africana* seeds. Berka & Harfouche (2001) were unable to improve germination rates of Argan seeds by sulfuric acid scarification. The results obtained on the effect of storage temperature on germination show that temperature influences seed germination physiology. They revealed that seeds stored at ambient temperatures, show the best germination rates (60% and 30%) than seeds stored at 4°C. The work of Berka & Harfouche (2001) showed that the ambient temperature is an important factor in the germination of Argan seeds. However, the germination rate is more important in SD seeds. The sand substrate records the highest germination rates; but the germination time is shorter with the compost substrate. Authors like Benmahioul *et al* (2010) and Ahaton *et al* (2009) have shown in their work that the germination rate is higher in the sand substrate. The Nguema Ndoutoumou *et al* (2013) showed that the mixture of sand and humus soilgave better results with respect to the germination of *Gambeya lacourtiana* seeds. In our experiences, the seedlings transplanted in plastic pots containing the compost showed a successful acclimatization rate of 34%. According to Ndiaye (2006, 2014), the *Oxytenanthera abyssinica* plants raised through seeds, showed 75% survival after acclimatization. In summary, the results showing the high germination rates in SD seeds, are linked to the positive effect of the digestive tract of monkeys on the germination process. Nevertheless, this germination rate is moreimportant with the combined effect of the seeds soaking in water for 24 hours. The positive effects of seeds SFscarificationsulfuric acid, confirm the integumentary inhibitor effect of *Opuntia tuna* plant.

Conclusion

The germination rate (58.3%) in seeds extracted from monkey droppings is higher than the germination rate observed in seeds from fruits 48.6%. These results confirm the positive effect of the digestive tract on the germination of *cactus* seeds. However, pre-soaking SD and SF seeds in water these results increases the percentage to 68%. Increasing the SF seeds scarification time to 25 min gave a good germination rate. The results also showed the positive effect of the sand substrate on seed germination.

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