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Research Journal of Agricultural Sciences
Volume 14; Issue 05 (Sep-Oct 2023); pp 1608–1614

ISSN: 0976-1675 (P)
ISSN: 2249-4538 (E)

Full Length Research Article

Analysis of Seed Dormancy Breaking in Buffel Grass (*Cenchrus ciliaris*) via Experimental Procedure

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Abstract

In this article, an experimental analysis of Buffel grass (*Cenchrus ciliaris*) was conducted. This experiment was conducted at the Central Laboratory, Seed Administration-Ministry of Agricultureand Forests-Khartoum-Sudan. The case study and experimental procedure are carried out on the different effects of dormancy-breaking methods on seeds of Buffel grass (*Cenchrus ciliaris*). In the experiment, three techniques were used for seed-breaking dormancy. The acid was added as an additive to increase the prevention and germination levels of seeds. These acids are sulfuric acid (H_2SO_4) with a concentration of 50, 70, and 90%, and Gibberellic acid with levels of 50, 100, 150, 200, 250, and 300 ppm respectively suggested in the experiment. Moreover, the hot water (H.W) was considered at 60 °C for 15, 30, 45, 60, and 75 minutes at different intervals. The experiment was carried out in a completely randomized design (CRD) with five replications. These experiments arebased on random samples to check and test the germination rate and other related parameters of the seeds and validate the quality of the seed. This thing improves the satisfaction of the customer as germination rate. Germination percentage and rate of germination were recorded for the seeds to study the effects of treatments. The results demonstrate that, in comparison to the control and other treatments, the Gibberellic acid treatment with a concentration of 300 ppm provided the highest germination percentage (71.86), germination rate (62.20), and mean germination percentage (43.24). Following that, the germination percentage was determined using sulfuric acid at a 90% concentration (63.52).

***Key words*:** *Cenchrus ciliaris*, Germination percentage, Gibberellic acid, Germination rate, Least square design (LSD), Mean germination percentage, Sulfuric acid

A grass known as Buffel grass, or *Cenchrus ciliaris*, is indigenous to southern Asia, as well as parts of Africa, Arabia, the Middle East, and India. It was brought to Australia in the late 19th century, and today it may be found all over tropical, subtropical, and warm temperate regions. In many locations, it is either produced or naturalized. It has been classified as poisonous in some places and invasive in dry and semi-arid habitats (northern America, Hawaii, Mexico, and northern Australia). In the majority of warm, dry, and semi-arid regions of the world, it has been cultivated for fodder and to manage erosion. It frequently eludes plantings, particularly in disturbed areas where it fosters a cycle of grass fires. Invaded ecosystems can change, changing ecosystem processes and endangering native plants and animals [1-4] due to increased fire frequency and intensity as well as thick Buffel grass growth.

Buffel grass is frequently associated with dispersed woody legumes like *Leucaena leucocephala*, *Acacia spp*., and *Prosopis spp*. It may co-occur with other drought-tolerant grasses such as *Urochloa maxima*, *Panicum maximum*, *Chloris spp*., and *Eragrostis spp*. Along seasonal dry riverbanks and drainage pathways in South Africa, a population of *Cenchrus ciliaris* and *Cyperusm arginatus* has been identified [5]. On sandy (fast-draining) soils with annual rainfall as high as 1200 mm, buffelgrass has been documented to persist [6]. Magnaporthe (*Pyricularia grisea*) is a dangerous fungus that has led to the dieback of Buffel grass in several nations and regions. The research [7] depicts a few more insects. Around the world, the most crucial factors for feeding on seed heads are seed quality and affordability [8-9].

Seed dormancy

Dormancy can be broken by most ideal growing conditions. A seed is necessary for a little plant whose life activities are at their most basic. A seed that is physiologically given enough water, and oxygen for proper aerobic metabolism, and an environment with a temperature that does not exceed physiological limitations but does not germinate is said to be dormant [10-11].

Received: 08 Aug 2023; Revised accepted: 12 Oct 2023; Published online: 19 Oct 2023

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Citation: Ahmed M, Mohamed B. 2023. Analysis of seed dormancy breaking in buffel grass via experimental procedure. *Res. Jr. Agril. Sci.* 14(5): 1608-1614.

Breaking the seed dormancy

Scarification Bonner is a general term for any procedure that eliminates or lowers the impermeability of the seed coat. The degree and type of seed dormancy have a significant impact on the effectiveness of treatment. Making a tiny hole in the seed coat before sowing is one of the simplest and most direct physical approaches [12-14]. According to a study indicating that the hardest-coated seeds become accessible to water when the seed coat is damaged or pierced by mechanical abrasion or chemical treatment, the percentage of Acacia seeds that germinate increased from 7% in control to 19% in scarified seeds [15]. Buffel grass (*Cenchrus ciliaris* L.) is a preferred species for erosion management of railway banks in the semi-arid regions of Central Queensland, Australia, due to its wide distribution, rapid regeneration, and simplicity of maintenance. The species has a deeper and larger root system that can provide higher strength against erosion when compared to other grasses like Rhodes (*Chloris gayana* L.) and green panic (*Panicum maximum* var. tracheoles) [16].

By storing carbohydrates at the base of the swelling stem, the plant can withstand periods of drought, grow again after burning [17], and start to receive rain [18]. Buffel grass is a great option for dry communities and the colonization of disturbed lands due to its ability to regenerate after burning and resistance to drought [19].

If rapid vegetation establishment is achieved without delaying germination when batter slopes are constructed, buffelgrass can effectively reduce soil erosion on railway banks. However, because of the inherent dormancy of seeds, low seed germination when planted on railway ties prevents the development of a good grass cover within a constrained timeframe and may have a detrimental effect on soil erosion control [20].

To determine the most efficient method of reawakening four Adesmia species from dormancy, Tedesco conducted germination studies. The outcomes showed that mechanical scarification was a successful method for obtaining a significant number of germinated seeds, as opposed to immersion in hot water at 60°C and undamaged seeds as a control. By scarifying the *Sporades corilifolia* using sandpaper, intact seeds obtained 90% germination. Physical scarification, as opposed to chemical scarification with hydrochloric acid, was found to be more efficient at releasing *Rhynchosia minima* L. seeds from dormancy by Shaukat and Burhan [21-22]. *Cassia moschata* and *Entada polyarchy* had the best germination rate and seedling emergence when mechanical scarification was performed on the radishes on the opposite side.

According to Ghadiri and Torshiz [23], mechanical scarification improved *Glycyrrhiza glabra* L. seed germination to 49–98%. As per Girase *et al*. [25], mechanical scarification can remove the dormancy that exists in the seeds of *Acacia auriculifomis* and *A. torilis*. Histochemical analysis of the seed coat of *Leucaena glauca* L. revealed that pectin served as a water barrier and that the only method for removing the seed coat's dormancy was mechanical scarification. According to the research of Baskin and Baskin [26], mechanical scarification can totally end the dormancy of *Dalea foliose* seed, which is caused by the seed coat's water-impermeable nature.

Many aborigines' seeds have responded well to hot water treatment [27]. Typically, boiling water is used to soak the seeds before the water is quickly removed from the heat source and allowed to cool gradually. For 12 hours, the seeds are submerged in water [28]. Both the modification or softening of the hard seed coat and the leaching out of chemical inhibitors are effects of wet treatment. Levitt [29] discovered that seeds that don't expand while being boiled can endure being boiled in water for a number of hours. Using hot water to treat a variety of leguminous seeds has produced positive outcomes. The effectiveness of boiling water on the germination of three Acacia species was observed by Brown and Booysen [30]. With 3–5 minutes of soaking in hot water (80-88 °C).

A. *Gregii's germination percentage increased considerably*

In a similar manner, Clemens *et al*. [31] reported a substantial impact of hot water at three different temperatures for seven different time periods on five *Acacia species*. According to Wickens [32], the treatment of *Acacia albida* seeds with almost boiling water did not alter their characteristics from untreated seeds. The application of hot water therapy is very simple, safe, and successful.

Based on the literature review, the method suggested for that analysis is the least different design method to establish the model based on the data. The data were analyzed according to the scale and rules.

The objective of this study is to find the different effects/methods for breaking dormancy in the Buffel grass seeds in this study, the experimental procedure about the Buffel grass seed breaking democracy with the help of the Ministry of Agriculture and Forests- Khartoum-Sudan was adopted. Furthermore, the germination percentage improvement and the period at which maximum germination rate is achieved.

This article is structured as follows: In Section 1, the literature review is discussed about Buffel grass and its production behavior. In Section 2, materials and methods are discussed. This section also describes the data collection methods and analysis methods. Section 3 covers the comprehensive procedure of results and a discussion of the proposed methods. In Section 4, the concluding remarks about the experimental analysis of germination are deliberated.

MATERIALS AND METHODS

A laboratory experiment was carried out to study the effect of different methods for breaking seed dormancy in Buffel grass (*Cenchrus ciliaris*). The study was conducted at the laboratory of the Seed Administration Ministry of Agriculture and Forestry, Khartoum, Sudan. This section is composed of two subsections. One is the materials about the research and the second is a method. The methodology is described to conduct the research.

Plant material

Buffel grass samples were taken from the fields such as Shabbat. The field locations were (latitude 15"40 N, longitude 32"32E, and 380m above sea level). The climate of the site is semi-desert and the soil is alkaline cracking clay with low permeability. The data collected from the fields, farmers, and surveys of the fields at the different locations, the nature of land and sand are different to check and validate the germination process and their related parameters.

Reagents

There are different reagents are used to boost the germination process and control the insects during the germination.

Sulfuric acid (H_2SO_4)

Sulfuric acid was used during the experiment and acid was obtained from the National Center for Research, Khartoum, Sudan.

Gibberellic acid (GA_3)

The second reagent is gibberellic acid (300 ml/L). It was provided by the Department of Horticulture University of Khartoum. In concentration, 0.3 gram per liter other concentrations were obtained by diluting from the stock solution. The different dilutions are obtained separately.

In this section, the research methodology is used. The different properties of seeds are evaluated.

Treatments: The process of treatment plays a key role in the seed quality, and germination rate and also improves the germination rate. This treatment improves the quality of the seed to prevent the different insects which damage the seed quality. The seeds of Buffel grass were subjected to the following treatments:

Control: No treatment was applied to the seeds

Hot water: A sub-sample of the seeds was soaked in hot water (60 °C) for 15, 30, 45, 60 and 75 minutes. The different timeframes were adopted to see the effects on the seeds.

Sulfuric acid (H_2SO_4): The different concentration of sulfuric acid was used. Seed samples were soaked in 50, 70, and 90% concentration of the acid. Thereafter the seeds were thoroughly rinsed several times in distilled water to remove any trace of the acid.

Gibberellic acid (GA_3): Six different concentrations of 50, 100,150, 200,250, and 300 PPM of GA_3.were prepared by dilution using distilled water from the stock (0.3g/L), and sub-samples of the sterilized seeds were soaked in each of the concentrations in the dark at room temperature for two hours.

Germination test

This experiment was carried out to determine how the treatments affected the Buffel grass seeds' ability to germinate. A completely randomized design with five replications was used to conduct the test. Each replication included 25 seeds in it. The seeds are prepared using various dormancy-breaking techniques, then stored in sterilized Petri dishes with a 9 cm diameter on double-layered filter paper that has been soaked with 8 cc of distilled water. After that, the Petri plates were put in an incubator set to 30 °C. For 55 days, germination seeds were counted and taken out every 24 hours. When the radical tip had separated from the seed coat, a seed was said to have germinated. The following parameters were then obtained using the collected data:

Germination percentage (%)

It is the ratio of germinated seeds to the total sprouted seeds. The number of germinated seeds was recorded daily and the germination percent was calculated as follows:

$$\text{Germination (\%)} = \frac{\text{No. of germinated seeds}}{\text{Total No. of seeds pe petri dish}}$$

Rate (speed) of germination (Gr)

The rate of germination was calculated according to Wiese and Binning [33] as follows:

$$\text{Germination rate (\%)} = \frac{1 + 2 + 3 + \ldots\ldots\ldots + N}{\text{Total No. of seeds}}$$

Where; 1, 2, 3, …. N are counting days from day 1 to N. speed of germination expresses the rate of germination in terms of the total number of seeds that germinate in a time interval. Higher values indicate greater and faster germination.

Statistical analysis

According to the procedure outlined by Gomez and Gomez [34] for a fully randomized design, the gathered data were subjected to analysis of variance. After that, mean separation was carried out utilizing the least significant difference (LSD) technique. Stat analysis was used to perform the computations.

RESULTS AND DISCUSSION

A: *Germination percentage of Buffel grass seeds*

There are different treatment cases done for germination rate. These steps improve the quality of seeds and germination rate and also boost the optimization level of the germination. The obtained results can be summarized as follows:

Case 1: At 10 days

Buffel grass germination percentages in the various treatments varied significantly ($P \leq 0.01$) after ten days, according to an analysis of variance. The treatments were divided into three groups in (Table 1) based on the mean germination. The GA3 300ppm condition yielded the highest germination rate of 14.85.

Table 1 Mean germination percentage of Buffel grass seeds during the period 10 to 22 day after germination

Treatments	Days 10	Days 13	Days 16	Days 19	Days 22
Control	1.81	1.81	1.81	1.81	1.81
H_2SO_4 50	11.54	12.31	12.31	20.93	23.51
H_2SO_4 70	12.31	13.09	13.09	22.25	24.76
H_2SO_4 90	16.62	18.62	18.92	28.32	29.74
GA_3 50 PPM	13.09	15.62	15.62	21.59	25.92
GA_3 100 PPM	12.31	14.84	14.84	22.85	27.66
GA_3 150 PPM	16.39	18.33	18.33	27.07	30.90
GA_3 200 PPM	17.36	18.33	18.33	25.3	29.3
GA_3 250 PPM	16.39	17.36	17.36	28.69	30.80
GA_3 300 PPM	18.87	25.81	20.91	29.77	34.31
Hot water 60 °C / 15min	1.81	1.81	1.81	3.66	4.15
Hot water 60 °C / 30min	1.81	1.81	1.81	4.22	5.99
Hot water 60 °C / 45min	6.48	7.14	8.37	7.65	9.25
Hot water 60 °C / 60min	1.81	1.81	1.81	3.81	3.96
Hot water 60 °C / 75min	1.81	1.81	1.81	1.81	3.79
Mean	9.96	10.28	11.21	20.21	22.42
5% LSD	3.38	3.41	3.54	3.02	3.16

This germanium rate is not significantly greater than GA3 250ppm, GA3 200ppm, GA3150ppm, and H2SO4 (70%), but was significantly higher than all other treatments as well as the control are shown in (Table 1, Fig 1-3). On the other hand, the lowest value which is 1.81 was recorded in all hot water treatments and the control.

Case 2: At 16 days

After sixteen days of treatment, an analysis of the data showed that there were major variations (P≤0.01) between the various treatments in the proportion of Buffel grass that germinated. The GA_3 300ppm and GA_3 250ppm concentrations yielded the highest germination percentages 25.85 and 25.81, respectively. They were not significantly greater than GA_3 200 ppm, GA_3 150 ppm, and H_2SO_4 (90%), but were significantly higher than all other treatments as well as the control are shown in (Table 1, Fig 1-3). On the other hand, the low values (3.14) and (3.05) were recorded in H.W 60 °C/30min and 45min, the lowest values (1.81) were recorded in the remaining hot water treatments, and the control is also shown in (Table 1, Fig 1-3).

Case 3: At 31 days

Analysis of variance demonstrated highly significant deviations (P≤0.01) in the germination amount throughout the various treatments after thirty-one (31) day sessions. In GA_3 300ppm, the highest germination percentage (44.65) was discovered.

It was not significantly greater than GA_3 250ppm GA_3 200ppm, GA_3 150ppm, and H_2SO_4 90%, but was significantly higher than all other treatments as well as the control are shown in (Table 2, Fig 1-3).

Moreover, all hot water treatments were not significantly different from the control and recorded lower germination percentage values in (Table 2).

Table 2 Mean germination percentage of Buffel grass during the period 25 to 37 days after germination

Treatments	Days 25	Days 28	Days 31	Days 34	Days 37
Control	1.81	1.81	1.81	1.81	1.81
H_2SO_4 50	25.92	28.74	30.87	32.42	35.87
H_2SO_4 70	27.07	29.32	31.92	32.75	35.89
H_2SO_4 90	35.92	38.48	41.28	43.21	46.84
GA_3 50 PPM	28.74	31.40	32.91	33.41	38.28
GA_3 100 PPM	29.85	33.41	35.40	36.35	40.15
GA_3 150 PPM	34.45	35.41	38.28	38.28	42.93
GA_3 200 PPM	32.95	36.85	39.56	40.50	44.08
GA_3 250 PPM	34.85	38.23	40.95	42.34	45.34
GA_3 300 PPM	36.32	39.88	43.26	44.65	49.31
Hot water 60 °C / 15min	3.22	3.33	4.52	4.55	5.25
Hot water 60 °C / 30min	6.08	6.25	6.96	7.16	7.82
Hot water 60 °C / 45min	10.25	11.58	13.58	14.56	15.23
Hot water 60 °C / 60min	3.97	3.99	4.16	4.89	5.16
Hot water 60 °C / 75min	3.11	3.19	3.35	3.66	4.12
Mean	24.89	27.41	29.82	30.32	34.19
5% LSD	3.42	3.74	3.88	3.67	3.74

Case 4: At 49 days

The analysis of variance revealed highly significant variations (P≤0.01) in the percentage of grass germination across the various treatments at day forty-nine. The treatment GA_3 300ppm always gave the highest germination percentage 62.2 which is shown in (Table 3, Fig 1-3). All GA_3 250ppm, GA_3 200ppm, GA_3 150ppm, and GA_3 100ppm continued to give higher germination percentages, whereas all hot water treatments gave lower germination values than other treatments.

Table 3 Mean germination percentage of Buffel grass during the period 40 to 55 days after germination

Treatments	Days 40	Days 43	Days 46	Days 49	Days 52	Days 55
Control	1.81	1.81	1.81	1.81	1.81	1.81
H_2SO_4 50	39.09	42.35	44.88	50.67	48.54	52.67
H_2SO_4 70	40.06	42.93	45.92	48.91	52.18	56.05
H_2SO_4 90	48.45	51.25	54.26	57.38	59.93	63.52
GA_3 50 PPM	40.15	41.89	43.85	44.31	45.46	51.24
GA_3 100 PPM	44.07	45.69	48.45	49.39	50.77	53.05
GA_3 150 PPM	46.49	47.88	50.31	51.71	53.61	58.71
GA_3 200 PPM	47.53	50.32	52.61	54.11	55.36	61.95
GA_3 250 PPM	48.33	50.44	53.86	55.33	58.37	62.94
GA_3 300 PPM	53.31	55.83	58.98	62.23	63.51	71.86
Hot water 60 °C / 15min	9.22	10.35	11.11	12.03	15.44	15.81
Hot water 60 °C / 30min	9.98	10.66	11.95	12.48	15.78	16.79
Hot water 60 °C / 45min	16.23	17.58	17.91	18.24	18.65	19.39
Hot water 60 °C / 60min	7.66	8.96	9.45	9.81	10.87	12.37
Hot water 60 °C / 75min	6.22	7.33	8.24	9.29	10.41	11.47
Mean	37.24	37.79	38.64	38.64	41.84	43.24
5% LSD	3.88	3.85	3.75	3.757	3.68	4.13

Case 5: At 55 days:

The treatment GA_3 300ppm continued to provide the best germination percentage at 55 days, which is 71.86. The highest germination values were provided by GA_3 250 ppm (H_2SO_4 90%), GA_3 200 ppm, GA_3 150 ppm, and GA_3 100 ppm, which were significantly greater than the control and other treatments. However, the mean separation was observed to rise in all treatments except for these four.

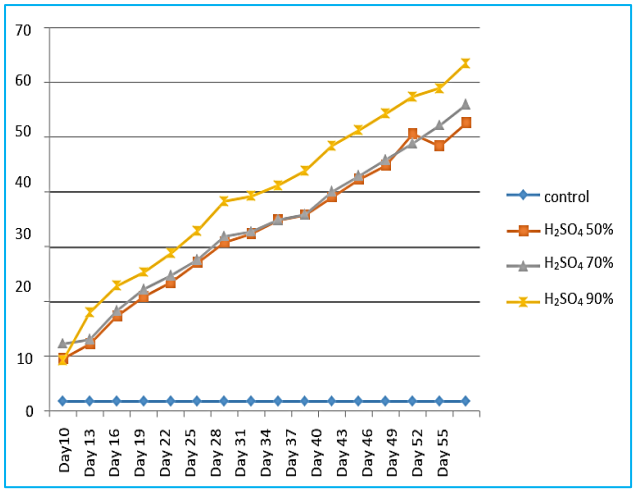

Fig 1 H_2SO_4 effects on pre-treatments on germination (10-55 days after treatments)

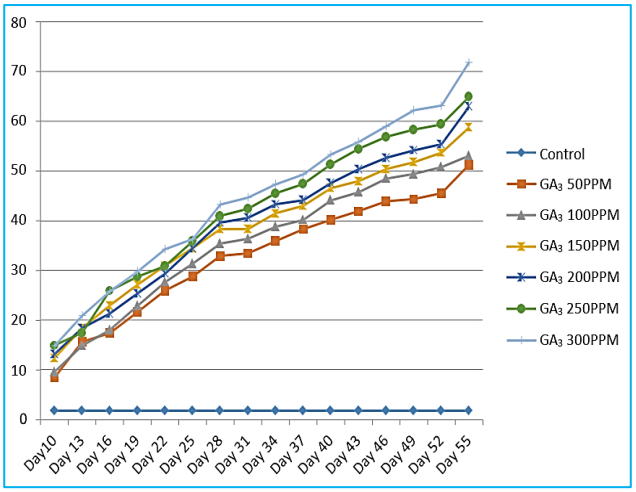

Fig 2 GA_3 effects on pre-treatments on germination (10-55 days after treatments)

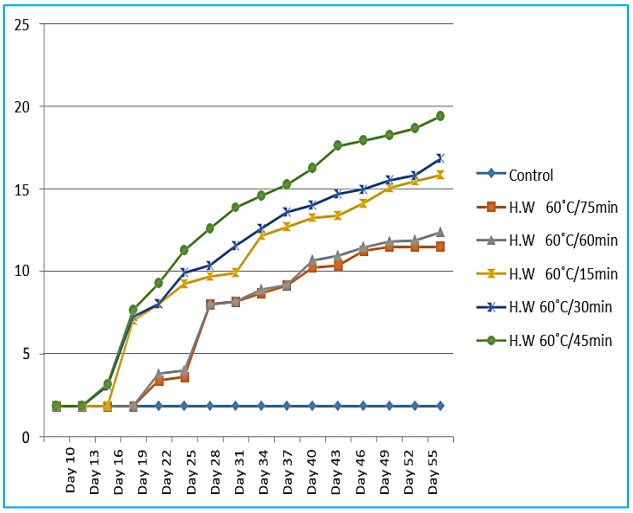

Fig 3 Hot water effects on pre-treatments of germination (10-55 days after treatments)

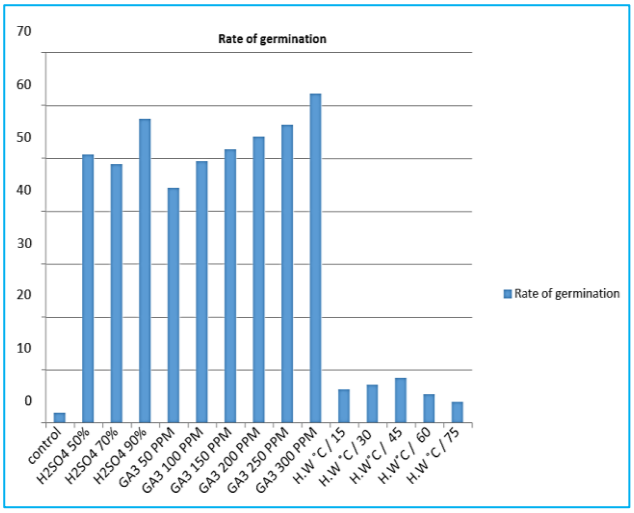

Fig 4 Germination rate effects after 55 days experiment

B: *Germination rate*

In this section, the germination rate of the seed is calculated through the least squared difference. The scale is set by the variance to estimate the level of germination rate. The germination rate of Buffel grass during the lab experiment of 55 days is shown in (Fig 4). This behavior shows a highly significant difference with respect to the scale $P \leq 0.01$. The germination treatments are shown in (Table 4). This data and facts of results show that the germination rate follows the linear rate of germination. The results show that the highest germination rate was recorded for GA_3 300ppm, which is highly significant from all remaining treatments. In the proceeding of the results, the higher the concentration (level of sulfuric acid) is, the germination rate is higher than the previous dilution rate such as H_2SO_4 (90%) is a higher germination rate than H_2SO_4 (70%). but gibberellic acid (GA_3) effects are also considerable on the germination rate. The higher level of gibberellic acid (GA_3) increases the germination rate such as GA_3 (250) Higher germination was recorded than other dilutions. All hot water treatments recorded the lowest germination rate value, which is shown in (Table 4).

Table 4 Rate of germination in Buffel grass seeds after germination treatments for breaking dormancy under laboratory conditions, for 55 days

Treatments	Rate of germination
Control	1.81
H_2SO_4 50	50.67
H_2SO_4 70	48.91
H_2SO_4 90	57.38
GA_3 50 PPM	44.31
GA_3 100 PPM	49.39
GA_3 150 PPM	51.71
GA_3 200 PPM	54.11
GA_3 250 PPM	56.33
GA_3 300 PPM	62.20
Hot water 60 °C / 15min	6.26
Hot water 60 °C / 30min	7.13
Hot water 60 °C / 45min	8.42
Hot water 60 °C / 60min	5.34
Hot water 60 °C / 75min	3.89

The results of this study showed that gibberellic acid (GA_3) in different concentrations gave a significantly higher germination percentage than the other treatments. This demonstrated that GA_3 was effective in removing these seeds' dormancy. These outcomes indicate that GA_3 might be involved in the control of both protein and nucleic acid metabolism during dormancy. These results suggest that the enhancement of germination might occur due to the role played by GA_3 in breaking endogenous dormancy. These results agree with those reporting that the promotion of germination may be due to the fact that, GA_3 at increased concentration may act as a signal for amylase production, which plays an essential role in seed germination in rice seed, and in grass *Leymus arenarius*. The results indicated that seeds of *Cenchrus ciliaris* did not exhibit physical dormancy (no seed coat dormancy was found in the seeds), and the type of seed dormancy was endogenous, physiological, or morphophysiological.

CONCLUSION

The concluding remarks are written as:

1. The optimization is achieved at 300ppm because the breaking seed dormancy in Buffel grass seed is the application of GA_3. This is the best and most convenient method to achieve the level of optimization.
2. The reliable achievements are obtained by the experiment by using sulfuric acid. The different dilutions of acid are used to achieve the maximum breaking seed dormancy in Buffel grass. The optimum concentration is 90%.

The future recommendation based on the experiment is to emphasize on using seeds of different ages treated with GA_3 in different concentrations and H_2SO_4, to determine the appropriate concentration of GA_3 to be used for breaking seed dormancy in Buffel grass seeds.

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