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Effect of Hormonal Application on Physio-biochemical Characters to Improve Seed Yield of Sewan Grass (*Lasiurus sindicus* Henrard)

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Abstract: The natural grassland lie in the desert area are in highly deteriorating stage with the productivity of only 300-400 kg ha^{-1} year^{-1}. The dominant perennial grass i.e., indigenous sewas grass is popularly known as the "King of the Desert grasses" belongs to the family Poaceae. Sewan grass is very important in arid environment because it coves soil especially at the top 15-30 cm that helps to protect soil transportation and erosion. The quality seed production of grasses is a challenging task for breeder and agronomist. Many problems are associated with grass seed production. Very low efforts have been made to develop the high yielding grass varieties. It has also been noticed that the high yielder fodder varieties are very poor seed yielder. Thus, the present research was carried out to assess the improvement of seed yield through application of different of hormones on different morpho-physiological and biochemical characters that affect seed set and seed yield. The improvement in physiological growth and yield characters viz., increasing in leaf area, per cent green leaves, number of tillers, number of spikelets, seeds per spike were observed with different levels of hormones. Maximum increase in leaf area, per cent green leaves, number of tillers, number of spikelets, spike length over control was observed in the plants treated with cycocel 100 ppm + pactobutrazol 200 ppm at seed filling stage. The canopy temperature at seed filling stage also showed significant positive relationship with seed set and seed yield. Hormone application significantly increased total soluble sugar, phenol and free proline content over the control in leaf, stem and showed positive relationship with seed yield. Among the treatments, cycocel 100 ppm + pactobutrazol 200 ppm showed maximum seed set and seed yield. The increase in leaf area, per cent green leaves and relative water content due to hormones application helps in lowering canopy temperature and stimulate metabolic activity that ultimately increase seed set, yield attributing characters and seed yield of *Lasiurus* grass.

Key words: *Lasiurus sindicus*, hormones, phenol, proline, relative water content, seed set.

Indian hot arid zone cover an area of 32 million hectare called as "Thar Desert". About 91% of the Indian desert fall in Rajasthan covering about 61% geographical area of the state. Western part of Rajasthan, the *Thar* desert, is the most thickly populated desert of the world, possessing more than 55 million population each of human and livestock. Aberrant weather conditions and poor soil fertility pose major threats to arable crops cultivation. Livestock rearing is directly dependent on fodder availability but due to harsh agro-climatic conditions there is severe fodder scarcity. The natural grassland lie in the desert area are in highly deteriorating stage with the productivity of only 300-400 kg ha^{-1} year^{-1}. Dichanthium-*Cenchrus*-*Lasiurus* type grassland are associated with sub-tropical, arid, and semi arid region comprising the northern part of Gujarat and the whole of the Rajasthan. *Lasiurus sindicus*, perennial grass grown in hot and warm grassland climate thrives well under moisture stress on sandy plains, low dunes and hummocks of this region, receiving annual rainfall below 200 mm. For decades, these grasslands in turn supported an ever-increasing livestock population in the Indian desert. Sewan grass has a higher calcium and lignin content than the other grasses such as marvel grass, buffel grass, birdwood grass, and bermuda grass. The component of cruide fiber are cellulose, lignin and hemi-cellulose. However, in case of other nutritional properties sewan grasses as lower value than other grasses but

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due to it's drought resistant properties, it can be grown in very low rainfall area and useful for small ruminant such as sheep and goat. The establishment of this grass is hampered by low seed availability because of its poor seed yield due to poor seed set. While sufficient literature is available on forage productivity of sewan grass, however, information on seed set and seed yield production is lacking.

Materials and Methods

The present study was conducted to assess the improvement of seed yield through application of different levels of hormones on different morpho-physiological and biochemical characters that affect seed set and seed yield. The soil of the experimental site at Chandan, CAZRI, RRS, Jaisalmer, Rajasthan, India having elevation of 196 meters (latitude 26° 59′ 31.32 N and longitude 71°20′29.59E) was sandy loam and alkaline (pH 8.43) with <0.1% organic carbon, electrical conductivity- 0.113 dS m^{-1}, available nitrogen - <70 kg ha^{-1}, available phosphorus - <10 kg ha^{-1}, available potash - 221 kg ha^{-1}. The study was conducted during 2014 at Experimental Area, Chandan, CAZRI-RRS Jaisalmer in 20 x 6 m plots, where row-to-row and plant to plant spacing was kept 1m. Different levels of plant hormones [T_2- Cycocel 100 ppm, T_3- Cycocel 200 ppm, T_4- Paclobutrazol 200 ppm, T_5- Paclobutrazol 400 ppm, T_6- Cycocel 100 ppm + Paclobutrazol 200 ppm, T_7- Salicylic acid 100 ppm and T_8- Salicylic acid 200 ppm] were sprayed at pre-flowering and anthesis stage on the sewan grass planted in RBD design with three replications. Water sprayed plants (T_1) served as control. The meteorological data, namely average maximum and minimum temperature, humidity, photoperiod, wind speed, wind direction and rainfall were noted during the field experiment. Ten plants were randomly selected and growth data were recorded at weekly interval from all sets. Three cuts of grasses were taken and the growth data included number of raceme, number of tillers, tussock diameter, plant height, above ground biomass (fresh and dry) and below ground biomass (fresh and dry) weight per plant were taken at weekly intervals.

Leaf relative water content (RWC) was estimated by recoding the turgid weight of 0.5 g fresh leaf samples by keeping in water for 4 h, followed by drying in hot air oven till constant weight using the following relationships (Gao, 2000).

$$RWC\ (\%) = [(Fresh\ weight - Dry\ weight) / (Turgid\ weight - Dry\ weight)] \times 100$$

The total phenols were determined by Folin – Ciocalteau reagent method described by Malik and Singh, 1980. The dilute extracts of different concentrations were taken in 10 ml. glass tubes and total volume made to 3 ml. with distilled water these are then mixed with 0.5 ml Folin - Ciocalteau reagent (1:1 with water) and 2 ml Na_2CO_3 (20%). A blue coloured complex, molybdenum blue developed in each tube, as the phenols undergo a complex redox reaction with phosphomolibdic acid in Folin – Ciocalteau reagent in alkaline medium. The tubes containing the blue solutions were warmed for 1 min., cooled and absorbance was measured at 650 nm against the reagent blank. The standard curve was prepared using known concentrations of catechol at 650 nm. The total phenol content in the test samples was calculated from the standard curve and expressed as mg. catechol equivalent of phenol/ g. sample.

The sugar was estimated with the method of Dubois *et al*. (1951). The 100 mg leaves was ground with 50% ethyl alcohol (10 ml).The homogenate was centrifuged at 4000 rpm for 15 minutes and the supernatant was used for estimation of sugar. Sugar was estimated with anthrone reagent. Free proline was measured spectrophotometrically according to the method of Bates *et al*. (1973). The 200 mg fresh leaves were homogenized in 5 ml of 3% aqueous sulphosalicylic acid and centrifuged at 4000 rpm for 20 minutes. The residue was reextracted with 5 ml of 3% aqueous sulphosalicylic acid and make final volume to 10 ml. The 2 ml of this aliquote was used for proline estimation by ninhydrin reagent.

Results and Discussion

The meteorological data recorded during the period of experiment showed that the minimum atmospheric temperature varied from 21.0-27.0°C while the maximum varied from 35.0-42.0°C. The 18.3 to 28.4 mm variation in vapour pressure and 29 to 81% variation in relative humidity were also recorded during the period (Table 1). The total rainfall received during the period was 164.5 mm, out of which

Table 1. Effect of different levels of hormones on shoot: root ratio, leaf area, %green leaves RWC, number of tillers, number of spikelet, dry fodder yield, seed set and seed yield of Lasiurus sindicus L.

Treatments	Shoot : Root ratio	Leaf area (cm^2)	RWC (%)	Green leaves (%)	Tillers/ tussock	Spikelet/ raceme	DFY (g $plant^{-1}$)	Seed set (%)	Seed yield (g $plant^{-1}$)
Control	1.20	4.06	35.50	11.20	91.67	16.58	720.30	21.60	2.06
CCC 100 ppm	0.90	4.98	37.00	12.20	110.67	15.50	944.30	34.10	2.38
CCC 200 ppm	0.90	6.14	41.20	13.90	140.50	27.67	876.00	32.60	2.86
PBZ 200 ppm	0.80	5.59	40.30	11.70	109.50	25.33	696.00	34.80	2.64
PBZ 400 ppm	0.80	7.23	45.00	16.10	129.83	38.67	826.30	36.60	3.05
CCC 100+PBZ 200 ppm	0.90	8.25	46.90	19.30	147.33	44.50	1097.00	38.90	3.41
SA 100 ppm	0.90	5.21	38.30	13.60	102.67	21.92	1050.70	35.50	2.21
SA 200 ppm	0.90	5.75	39.90	16.70	122.67	25.50	1244.30	37.50	2.49
Min	0.80	4.10	35.50	11.20	91.70	13.50	696.00	21.60	2.10
Max	1.20	8.20	46.90	19.30	147.30	44.50	1244.30	38.90	3.40
Mean	0.90	5.90	40.50	14.30	119.40	27.00	931.90	33.90	2.60
S.D.	0.07	0.98	2.89	2.28	7.39	4.06	152.22	3.43	0.35
Variance	0.01	1.73	14.93	7.93	14.37	34.92	36262.00	28.91	0.20
Variance/N	0.002	0.217	1.867	0.992	1.796	4.365	4533.00	3.613	0.025
SEm	0.04	0.47	1.370	1.00	1.34	2.09	67.33	1.90	0.16
C.V.	8.00	16.60	7.10	15.90	6.20	15.10	16.30	10.10	13.40

127.5 mm received during July and August. Sunshine hours recorded during observation period varied from 2-10 hours.

Hormonal application has significant effects on leaf area, perc ent green leaves, RWC and others yield attributing characters (Table 1). However, the effect on shoot: root ratio was non-significant. Hormone application increase leaf area of sewan grass and maximum increase in leaf area was measured with the treatment

Table 2. Effect of different levels of hormones on total sugar phenol content and free proline content in leaves and stem of Lasiurus sindicus L.

Treatments	Total sugar [(mg g^{-1}) Fr. wt]		Total phenol [(mg g^{-1}) Fr. wt]		Free proline [(mg g^{-1}) Fr. wt]	
	Leaf	Stem	Leaf	Stem	Leaf	Stem
Control	9.19	18.00	3.67	3.12	2.81	3.45
CCC 100 ppm	12.29	21.00	4.31	5.24	3.07	4.34
CCC 200 ppm	13.94	23.10	5.03	5.12	3.51	5.76
PBZ 200 ppm	11.81	17.50	5.10	4.61	3.21	4.89
PBZ 400 ppm	16.14	28.90	5.89	5.59	5.35	7.56
CCC 100+PBZ 200 ppm	20.22	34.90	6.34	5.79	5.84	8.20
SA 100 ppm	10.35	16.60	4.18	3.41	3.22	5.34
SA 200 ppm	15.72	28.90	4.85	4.58	4.16	6.86
Min	9.20	16.60	3.70	3.12	2.80	3.50
Max	20.20	34.90	6.34	5.79	5.84	8.20
Mean	13.70	23.60	4.90	4.68	3.90	5.80
S.D.	2.80	2.40	0.67	0.37	0.92	1.31
Variance	12.81	22.50	0.78	16.44	1.27	2.67
Variance/N	1.602	2.807	0.098	2.055	0.159	0.334
SEm	1.27	1.68	0.31	1.43	0.40	0.58
C.V.	20.40	10.30	13.60	7.90	23.50	22.50

Table 3. Range, Mean and C.V. values of different characters of Lasiurus sindicus at different intervals

Characters	10th September			17th September			24th September		
	Range	Mean ± S.Em	C.V.	Range	Mean ± S.Em	C.V.	Range	Mean ± S.Em	C.V.
Root:Shoot length	0.33-0.5	0.4±0.02	10.07	0.34-0.49	0.41±0.02	9.03	0.39-.50	0.44±0.01	6.72
No. of raceme	55-120	88.9±8.13	21.52	59-125	95.0±8.04	19.7	63-132	100±8.22	18.9
Spikelets/raceme	12.0-53.7	28.4±5.63	45.80	13.5-56.6	31.5±5.70	42.3	15.6-58.7	34.5±5.68	38.6
Spike length	9.63-14.8	11.0±0.65	12.50	9.22-14.9	11.05±0.67	12.7	10.3-16.2	12.6±0.76	13.8
Tussock diameter	21.9-35.8	28.2±1.74	14.60	22.2-38.9	30.0±2.06	15.8	23.3-40.3	31.3±2.09	15
Tillers/tussock	89.7-146.7	119.0±7.51	15.10	94.6-152.3	123.4±7.46	13.9	96.8-155.4	125.8±7.3	13.4
Green fodder yield	0.78-1.7	1.1±0.12	25.90	0.82-1.79	1.18±0.12	24.6	0.85-1.80	1.26±0.13	24.7
Dry fodder yield	0.25-0.80	0.5±0.07	33.50	0.27-0.79	0.47±0.07	32.9	0.27-0.90	0.53±0.08	33.5
Av. canopy temp.	40.0-45.1	42.5±0.60	3.14	38.9- 41.2	41.8±0.62	3.23	38.3- 43.7	41.2±0.66	3.49
Phenol content in leaf	-	-	-	1.47- 4.57	2.71±0.37	30.6	1.74- 4.70	3.16±0.40	31.2
Phenol content in stem	-	-	-	5.83- 7.60	6.83±0.21	7.27	4.62- 9.00	7.19±0.57	16.5
Sugar content in leaf	-	-	-	4.13- 6.55	5.36±0.37	16.6	3.78-9.16	6.09±0.68	24.1
Sugar content in stem	-	-	-	6.74- 21.4	13.2±1.65	25.9	10.43-21.0	15.7±1.29	18.5

Characters	1st October			8th October		
	Range	Mean±S.Em	C.V.	Range	Mean±S.Em	C.V.
Root:Shoot length	0.40-0.51	0.45±0.01	6.44	0.40-0.50	0.43±0.01	6.27
No. of raceme	66-135	105±8.19	17.60	58-130	98±8.5	19.10
Spikelets/raceme	15.6-62.8	37.9±6.01	36.60	18.5-55.2	30.4±5.9	45.60
Spike length	10.7-18.2	13.9±0.97	16.50	9.6-15.4	11.8±0.76	14.80
Tussock diameter	24.2-42.2	33.6±2.21	15.20	21.3-39.3	30.4±2.21	16.40
Tillers/tussock	101.5-157.4	128.1±7.02	12.40	93.4-153.2	122.7±7.7	14.50
Green fodder yield	0.87-1.86	1.32±0.13	24.20	0.80-1.80	1.20±0.13	25.00
Dry fodder yield	0.29-0.92	0.54±0.08	32.70	0.30-0.90	0.50±0.07	35.20
Av. canopy temp.	37.7- 43.4	40.7±0.70	3.86	39.1- 43.9	41.6±0.60	3.20

CCC 100 + PBZ 200 ppm followed by PBZ 400 ppm and CCC 200 ppm. Per cent green leaves were also increases by hormonal application and 4.5 to 72% increase in greenness was observed with different treatment (Table 1). The maximum greenness, 72% more over control was recorded with CCC 100 + PBZ 200 ppm followed by PBZ 400 ppm (Table 1). The control plants showed 35.5% leaf relative water content (RWC) that increases with hormonal application. The increase was 4.2 to 32.1% over the control plants. The maximum increase in RWC was measured with the treatment CCC 100 + PBZ 200 ppm. The finding of Nasrabadi *et al*. (2016) also showed positive and significant effect of paclobutrazol and salicylic acid on RWC (Table 1). The tillers per tussock was also significantly improved by different hormone treatment. The increase in tillers per tussock over control was varied from 12% to 60.63%. The maximum increase in tillers number was measured with CCC 100 + PBZ 200 ppm, followed by 200 ppm CCC (Table 1). While Salicylic acid 100 ppm showed minimum 12% increase in tiller numbers. The similar trend was also recorded in spikelet numbers. The dry fodder yield measured for per plant also significantly influenced by hormone application (Joaquin *et al*., 2007) and maximum increase in dry fodder yield over control was recorded with CCC 100 + PBZ 200 ppm treatment (Table 1 and 3). It showed 52% increase in fodder yield over control plants. The natural seed set in *Lasiurus sindicus* is very low as the control plant showed 21.6% seed set. The application of hormones improved seed set and maximum 38.9% seed set was observed with treatment CCC 100 + PBZ 200 ppm followed by paclobutrazole 400 ppm with 36.6% seed set (Table 1). Thus the best treatment CCC 100 + PBZ 200 ppm showed 80% more seed set than the control. The control sewan plant showed 2.06 g plant^{-1} seed yield, while the hormone spray increases seed yield significantly. The maximum 65.5% increase

Table 4. Correlation study of different characters

Character	SDW	RDW	NTPT	NSPP	PGL	LA	RWC	PSSPS	SYPT	Ta	Tc	WS
Shoot dry wt (SDW)												
Root dry wt (RDW)	0.943**											
No. tillers (NTPP)	0.538*	0.693**										
No. spikelets (NSPP)	0.820**	0.857**										
% green leaves (PGL)	0.635**	0.802**	0.89**	0.65**								
Leaf area (LA)	0.673**	0.848**	0.682**	0.805**	0.8**							
RWC (RWC)	0.628**	0.826**	0.901**	0.542*	0.885**	-0.802						
% seed set per spike (PSSPS)	0.482*	0.718**	0.642**	0.57*	0.762**	0.844**	0.863**					
Seed yield/ Tussock (SYPT)	0.474*	0.691**	0.89**	0.573*	0.963**	0.807**	0.867**	0.791**				
Atm temp (Ta)	0.111	-0.074	0.181	-0.219	-0.117	-0.198	-0.179	-0.539	-0.158			
Canopy temp (Tc)	0.073	-0.141	-0.710	0.117	-0.607	-0.168	-0.607	-0.395	-0.667	0.101		
Wind speed (WS)	0.040	0.028	0.219	-0.167	0.327	-0.233	0.148	0.028	0.192	-0.097	-0.529	
Per cent seed shed	-0.508	-0.68	-0.68	-0.511	-0.785	-0.526	-0.837	-0.715	-0.739	0.436	0.676**	-0.409

in seed yield over control was observed with treatment CCC 100 + PBZ 200 ppm (Table 1 and 3). The increase in seed yield was mainly because of improvement in seed set, increase in tillers and spikelet number. The increase in tiller numbers, spikelet and seed yield due to paclobutrazole was also reported by Tesfahun and Manzir (2018). Application of fertilizer increased the fresh and dry fodder yield, dry crown weight, dry root weight and fresh root volume per tussock by 85.93, 125.63, 75.51, 67.36 and 63.02% more than the control, respectively (Kumawat *et al*., 2017)

The phenol content varied from 3.7 to 6.3 (mg g^{-1} fresh weight) in leaves and 3.12 to 5.79 (mg g^{-1} fresh weight) in stem (Table 2). The control plant shows minimum phenol content while the application of hormones increases its level significantly over the control. The maximum 72% increase in leaf phenol and 85% increase in stem phenol over control were estimated with CCC 100 ppm + PBZ 200 ppm followed by PBZ 400 ppm. Similar to phenol, maximum 119% increases in sugar content were also observed in leaf and 94% increase in stem with CCC 100 ppm + PBZ 200 ppm treated plant as compared to control (Table 2 and 3). Hormonal application also increases the free proline content in leaves and stem of sewan grass and CCC 100 ppm + PBZ 200 ppm treated plant showed maximum proline accumulation in leaves and stem (Table 2 and 3). Enhancement of metabolic activity by hormones application is the possible cause of increase in phenol, sugar and free proline content of leaves and stem. Thus, our study revealed that seed setting and yield of sewan grass could be significantly increased (21.7% and 30.0% over control) by foliar spray of combination of CCC 100 ppm and PBZ 200 ppm. The soil and ambient temperature coupled with plant water status (RWC) seem to play important role in seed yield as also reported earlier by Ramírez and Hacker, 1993; Mathiassen *et al*., 2007.

Correlation Studies

The correlation study conducted for different characters showed that the seed yield has positive and significate correlation with number of tillers (0.89**), per cent green leaves (0.963**), leaf area (0.807**), RWC (0.867**), seed set per spike (0.791**) while it showed negative relationship with canopy temperature (-0.667) and per cent seed shed (-0.739**).

The per cent seed set also showed positive significant relationship with RWC (0.863**), leaf area (0.844**). Per cent green leaf (0.762**) and seed yield (0.791**) (Table 4).

Conclusion

The hormones application enhanced metabolic activity resulted in increased phenol, sugar and free proline content in leaves and stem. It showed positive relationship with seed yield. Among the treatments CCC 100 cppm + PBZ 200 ppm showed maximum seed set and seed yield. The increase in leaf area, per cent green leaves and relative water content due to hormones application helps in lowering canopy temperature. Thus, the study revealed that seed setting and yield of sewan grass could be significantly increased by foliar spray of combination of CCC 100 ppm and PBZ 200 ppm. The soil and ambient temperature coupled with plant water status (RWC) seem to play important role in seed yield.

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