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Amino acid chelated zinc on growth and yield of cumbu napier hybrid grass-CO (BN) 5

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Abstract
About 40% of soils from all over India and 63.2% from Tamil Nadu were deficit in Zinc. The synthesized Zn lysinate complex AAS analysis revealed 20.6% of Zn content which is used as micronutrient supplement for cumbu napier hybrid grass-CO(BN)5. The field trail was conducted by comparing the Zn sulphate and the chelated Zn on different dosage along with the recommended dose of fertilizer (75: 50: 40 kg/ha). The experiment was laid out as randomized block design with five replications. The results showed that the chelated Zn @ 4 kg ha^{-1} with 0.5% foliar spray 20 days after cutting showed positive influence on green fodder (137.54 ton/ha) and dry fodder yield (26.13 ton/ha) which is 51.68% and 51.58% respectively increased over the control. The growth parameters such as plant height 40.96% (264.58 cm), leaf length 23.36% (148.21 cm), leaf breadth 31.35% (4.56cm) and tillers 40% (50 No.s) responds positively towards soil and foliar application over the control. Similarly the chlorophyll content (56.99) found maximum about 25.67% over the control. One of the important fodder quality parameter crude protein content was also shows positive effect on crude protein content 8.93% and the control having 7.43%. It is concluded that both soil and foliar application of chelated Zn shows positive effect on growth and productivity of the crop when compared to other source.

Keywords: Zn lysinate, cumbu napier hybrid grass, growth parameters and yield

Introduction
Cumbu Napier Hybrid CO (BN) 5 was released by the TNAU, Coimbatore is an interspecific hybrid between fodder Cumbu IP 20594 x Napier grass FD 437. Some of the unique characteristics are winter hardy, rapid regeneration potential, free from pests and diseases, superior ratooning rendering seven cuttings per year. Livestock plays a vital contribution in Indian economy as per the estimates of National Accounts Statistics (NAS) 2020 the Gross Value Added (GVA) contributed by livestock has increased from 24.32 per cent (2014-15) to 28.63 per cent (2018-19). Livestock sector contributed 4.19 per cent of total GVA in 2018-19. Currently, India is facing a net deficit of 35 percent green fodder and 11 percent dry fodder due to increasing population of livestock (Kumar *et al.*, 2020) [12]. Among the micronutrients, Zn is considered to be the most important nutrient next to the macronutrients *viz*., N, P, K and S throughout the world (Bharti *et al.*, 2013) [3]. Zn involves in major physiological functions in both plants and animals. The health and productivity of the plants will be adversely affected due to zinc deficiency. Plants generally uptake Zn as a free divalent cation (Zn^{2+}), but monovalent cation ($ZnOH^+$) prevails at high pH. On calcareous soils, adsorption and fixation reactions can substantially reduce the efficacy of micronutrients (Tahir *et al.*, 2009) [25]. Inorganic Zn sources such as $ZnSO_4$, ZnO and synthetic chelates such as EDTA or EDDHA are key players in balanced nutrition of cropping systems. Chelated fertilizers are more efficient than the non-chelated fertilizer salts (Shivay *et al.*, 2016) [19] in the chelated form, metal ions are less likely to react and immobilize (Tahir *et al.*, 2009) [25]. Nevertheless, due to synthetic chelates fate in agriculture as a potential contaminant in the ecosystems, their application is not regarded as a sustainable practice. Hence it is indispensable to give the solution to intricate problem in the production of chelated micronutrient without involving EDTA or any other harmful sources. Aminochelates are more natural and safer forms of chelating agents with higher use efficiency and without environmental side effects (Souri *et al.*, 2019) [21]. But when it comes to fodder crop it was rarely used and the main problem was despite of nutrient consumption rate their supplementation for consumed nutrients are not given to the soil, particularly for micronutrients. Hence this research mainly focuses on the production of chelated micronutrient to mitigate the micronutrient deficiency in fodder and to

increase the nutrient quality of the fodder thus helps in maintain animal health and soil health. However, information on extent of Zn enrichment in cumbu napier grass was lacking. Therefore, the objective of this experiment was to investigate the effects of soil and foliar application of chelated Zn on growth, herbage yield and quality of cumbu napier hybrid grass under field conditions.

Materials and Methods

Zn lysinate synthesis

The $ZnCl_2$ hydrolyzed using excess alkali which precipitate into $Zn(OH)_2$ The Zinc lysinate complex was obtained by refluxing the $Zn(OH)_2$ to get fine white color powder. Then the Zn content was analysed using AAS model used here is ICE 3000 Series and physio-chemical properties such as colour, texture, moisture percentage, solubility, pH, EC of chelated Zn was analysed based on testing methods of fertilizer guidelines (2016) by FAMIC, Japan..

Field experiment

The field trail was conducted in the farmer's field during April-June of 2021 at Valaypatti village, Madurai district, Tamil Nadu with the GPS coordination of 9º82' N latitude and 78 º08' E longitude and the field soil properties have shown in table.1. The experiment was laid out as Randomized Block Design with ten treatments of five replication. Plot size of 5 x 4m (20m^2) was adopted to grow the cumbu napier hybrid grass- CO (BN)-5 with the treatment *viz.,* T1: Recommended dose of NPK (75:50:40) as standard control, T2: RDF + 12.5 Kg ha^{-1} $ZnSO_4$ soil application, T3: RDF + 25 Kg ha^{-1} $ZnSO_4$ soil application, T4: RDF + 4 Kg ha^{-1} Zn lysinate soil application, T5: RDF + 2 Kg ha^{-1} Zn lysinate soil application, T6: RDF + 1 Kg ha^{-1} Zn lysinate soil application, T7: RDF + 0.5% Zn lysinate foliar application @ 20 days after cutting, T8: RDF + 4 Kg ha^{-1} Zn lysinate in soil + 0.5% Zn lysinate foliar application @ 20 DAC, T9: RDF + 2 Kg ha^{-1} Zn lysinate in soil + 0.5% Zn lysinate foliar application @ 20 DAC and T10: RDF + 1 Kg ha^{-1} Zn lysinate in soil + 0.5% Zn lysinate foliar application @ 20 DAC. The growth parameters such as plant height, leaf length, leaf breadth, no. of tillers and chlorophyll value were recorded at the three different duration *viz.,* 15, 30 and at harvest of the crop (45th DAC). The chlorophyll value was recorded by using SPAD-502 meter. The green fodder yield and dry fodder yield were recorded on hectare basis. The collected data's were tabulated and statistically analysed using AGRES software.

Results and Discussions

Zn lysinate

A fine white colour powder was obtained by refluxing $Zn(OH)_2$ and lysinate (1:2). The synthesis of Zn lysinate was two step processes. First, preparation of $Zn(OH)_2$ precipitates by excess alkali hydrolysis method which yielded 72.6% of $Zn(OH)_2$ and the second, refluxing $Zn(OH)_2$ with Lysine which yielded 84.3% of Zn lysinate. The physio-chemical properties of Zn lysinate were analyzed and tabulated in the table.2.

Effect of Zn lysinate on Cumbu napier hybrid grass

The growth parameters observed in the field such as plant height, number of tillers, leaf length, leaf breadth and chlorophyll content are positively influenced by the Zn lysinate application.

Plant Height

The maximum plant height was recorded in the treatment applying chelated Zn @ 4 Kg ha^{-1} with the 0.5% foliar spray at 20 days after cutting (T8) of about 112.04cm, 159.35cm and 264.58cm and the least plant height of 102.04cm, 141.38cm and 156.16cm was observed in treatment T1(RDF alone- 75:50:40 Kg ha^{-1}) at 15, 30 and 45 days respectively (Table.3). This positive effect of Zn fertilization on plant height is similar to Salwinder *et al*. 2020 and Khanda *et al.*, 2013. They reported that plant height improves on application of Zn which may be due to its inter relationship with auxin production, which is an important growth promoter regulating all enlargement and stem elongation. The effectiveness of chelated Zn treatments over the $ZnSO_4$ treatments (T2&T3) were in close conformity with Khalid *et al.*, 2013 [9] who revealed that chelated Zn application have high positive effect on increase in plant height significantly compared to Zn applied through $ZnSO_4$. The maximum plant height recorded might be due the increase in Zn content of rhizosphere as well as quick utilization of Zn through the foliar spray these results were similar to findings of Sajid *et al.,*2009, Ghasemi *et al.,*2013 [7] and Rizwan *et al.*, 2017 [16].

Number of tillers

The number of tillers per plant found to be significant at different growing duration of cumbu napier hybrid grass (Table.3). However at 15 DAC the more number of tillers per plant 34.4 on T8 treatment which is statistical at par with treatment T4 having 33.26 numbers was recorded. On the other hand the lowest number of tillers per plant was recorded in treatment T1 (RDF only). At 30 DAC the number of tillers more in treatment T8 having 42.17 tillers followed by the treatments T9 and T4 which are statistical at par with 33.81 and 33.65 tillers respectively. At 45 DAC maximum number of tillers per plant was recorded in the treatment T8 having 50.37 tillers followed by the treatment T4 having 47.14 tillers which is statistically at par with treatment T9 having 45.83 tillers. From the obtained result the number of tillers recorded on the treatment T4 and T9 are at par confirms the effect done by RDF+4 Kg ha^{-1} Zn chelate was similar to RDF+2 Kg ha^{-1} Zn chelate + 0.5% foliar at 20DAC. The maximum number of tillers was recorded due to both soil and foliar application of Zn, which might be resulted in high concentration of Zn in soil rhizosphere and better utilization of foliar Zn fertilization this resulted in increase in plant height, leaf area result in high photosynthetic rate which in turn helped in formation of tillers. Above results were in thorough agreement with the findings of Dhaliwal *et al.,* 2020 and Sandhu *et al.,* 2020.

Leaf length and leaf breadth

At 15 DAC the leaf length and breadth of the crop found to be maximum in the treatment T8 recorded 77.64 cm and 3.05cm respectively (Table.4). Which is at par with the treatment T4 having 76.17 cm length and 3.07 cm breadth and the minimum one was in the treatment T1 recorded 63.11cm length and 2.38cm breadth. At 30 DAC the maximum effect was on the treatment T8 recorded 98.89cm length and 3.38cm breadth followed by treatment T4 and T9 those treatments are at par with each other recorded values of 95.04 cm length, 3.43 cm breadth (T4) and 94.97cm length, 2.93cm breadth (T9). At 45DAC the maximum effect was on the treatment T8 recorded 148.21cm length and 4.56 cm breadth followed by T9 recorded 140.43 cm length and 4.09 cm breadth. The minimum leaf length and breadth was recorded in the treatment T1 of about 113.58 cm length and 3.13 cm breadth. From the recorded values we confirmed that the combined application of both soil and foliar fertilization of chelated Zn shows maximum effect on crops. Here significant effect on Zn chelate on leaf area was observed, Bakyt and Sade (2002), Tabir *et al.,* 2009 [25] reported that flag leaf blade area was increased by Zn application in barley. An increase in the leaf area and other growth parameters due to the application of chelate Zn shows improvement in plant metabolism responsible for cell division and cell enlargement due to the optimum supply of nutrients (Dhanalakshmi *et al.,* 2019, Krishnaraj *et al.,* 2020, Kothaipriya *et al.,* 2020) [6, 11]

Chlorophyll

Throughout the entire duration the chlorophyll content of the crop shows increasing trend (Table.4). At 15 DAC the chlorophyll content was found to be maximum in T4 having 46.94 followed by T3 and T8 having value of 45.83 and 45.16. From the result we can tell that the Zn absorption for the treatments T3 and T8 are at par showing $ZnSO_4$ and chelated Zn source shares a similar effect within 15 days. At 30 DAC the maximum chlorophyll content was observed in the treatment T8 recorded value of 51.40 which is followed by the treatment T9 and T4 recorded value of 48.20 and 47.55 respectively. The sudden increase in chlorophyll on T8 was due to the effect of foliar application of chelated Zn at 20 DAC. At 45 DAC the maximum chlorophyll content was observed in the treatment T8 having 56.99 followed by the treatment T9 and T4 having value of and 53.47 and 52.07 and the minimum chlorophyll content was recorded in the treatment T1 during the entire crop growing period. After the foliar application there is a steady trend in increase in chlorophyll content than the other treatments this result was closely related to the findings of Krishnaraj *et al.,* 2020 [11] who revealed that foliar application of amino chelated sources increases the yield and growth attributes in maize. The present study suggest that the integrated effect of aminoacid with macro and micro nutrient improves the growth and photosynthetic activities in pearl millet (Mostafa *et al.,*2014) [14] and it was confirmed by the research findings given by Sadah *et al.,* 2015 [18] and Souri (2016).

Crude protein

Crude protein an important fodder quality parameter was analysed. Among the treatments T8 recorded 8.93% shows maximum followed by the treatment T9 recorded 8.58%. The minimum crude protein content was recorded in the treatment T1 having 7.43% (Table.5). Significant difference noticed between the treatments with respect to crude protein content because there is significant difference in nitrogen content among the treatments this result was coinciding with the findings of Soni *et al.,* 2016 [22]. This may be due to the fact that the nitrogen plays a vital role in synthesis of plant proteins. Sufficient amount micronutrients such as Zn and Cu in rhizosphere region induce the intake of nitrogen and results in protein synthesis (Raveena *et al*., 2020)

Yield

The green fodder and dry fodder yield was found to be maximum in the treatment T8 recorded 137.54 ton ha^{-1} and 26.13 ton ha^{-1} respectively followed by the treatments T4 recorded 121.39 ton ha^{-1} of green fodder and 23.06 ton ha^{-1} of dry fodder yield and T9 recorded 113.54 ton ha^{-1} of green fodder and 21.53 ton ha^{-1} of dry fodder yield. The minimum yield was recorded in treatment T1 having 66.45 ton ha^{-1} of green fodder and 12.62 ton ha^{-1} of dry fodder yield (Table.5). These findings are in close conformity to those of Bama *et al*., 2015 [2]. This might be due to the combined effect of soil and foliar fertilization of chelated Zn which was similar to Rizwan *et al.,* 2017 [16] who noted that Zn amino chelate has higher efficiency in plants which favours the positive effect on growth and yield of the crop than the $ZnSO_4$ source. The higher biomass production with application of Zn lysinate might be due to accompanying effect of both Zn and lysine on plants. The maximum fodder yield might be due to the role of Zn in various growth parameters like photosynthesis; N metabolism; protein synthesis; hormone production and regulation of auxin in plants, these results in taller plant with maximum leaf area which interms reflects in green fodder yield (Kumar *et al.,* 2012 and Sulthana *et al.,* 2015) [13,23] the obtained results also coincide with Trehan and Sharma (2000) [26] who assess the wheat, maize and sunflower results a significant response to Zn in terms of maximum dry matter yield.

Table 1: Soil Physio-chemical properties

A. Physical Properties		Methodology	Reference
Soil Texture	Sandy clay loam	International pipette method	Piper (1966)
Bulk density	1.24 g/cc	Core sampler method	Gupta and Dakshinamoorthy (1981)
Particle density	2.22 g/cc	Core sampler method	Gupta and Dakshinamoorthy (1981)
Pore space	44.7%	Core sampler method	Gupta and Dakshinamoorthy (1981)

B. Chemical Properties		Methodology	Reference
Ph	8.27	Potentiometry (Soil-water suspension of 1:2 ratio)	Jackson (1973)
EC	0.25 dSm^{-1}	Conductometry (Soil water suspension of 1:2 ratio)	Jackson (1973)
Available N	234 kg ha^{-1}	Alkaline Permanganate method	Subhiah and Asija (1956)
Available P	11.5 kg ha^{-1}	Olsen method (0.5M $NaHCO_3$ extractable P)	Olsen (1954)
Available K	125 kg ha^{-1}	Neutral normal NH_4OAc method	Stanford and English (1949)
DTPA Zn	1.12 mg kg^{-1}	DTPA extract- AAS	Lindsay and Norvell (1978)

Table 2: Physio-chemical properties of Zn lysinate

A. Physical properties	
Colour	Creamy White
Texture	Fine powder
Moisture percentage	3-4%
Water Solubility	Highly soluble on water
Organic solvent solubility	Insoluble in organic solvents

B. Chemical properties	
pH	7.5
EC (dS m^{-1})	0.42
MW	323.75 g/mole
Zn content	20.6%
C content	26.84%

Table 3: Effect of Zn lysinate on plant height and no. of tillers

Treatments	Plant height (cm)			No. of tillers		
	15DAC	30DAC	45DAC	15DAC	30DAC	45DAC
T1	102.5	141.4	156.2	21.11	25.42	29.98
T2	107.9	156.5	186.2	23.62	26.58	33.81
T3	115.8	174.0	194.8	29.52	36.05	43.76
T4	110.5	156.3	214.0	33.26	39.65	47.14
T5	107.3	150.9	207.9	31.29	37.11	42.85
T6	103.2	145.4	188.9	27.62	31.97	34.46
T7	102.7	144.8	178.5	21.48	25.05	31.73
T8	112.0	159.3	264.6	34.40	42.17	50.37
T9	107.2	154.3	229.0	32.30	39.81	45.83
T10	105.0	146.4	218.7	29.26	33.97	41.31
SEd	2.0	1.7	4.5	1.86	1.07	1.42
CD (P= 0.05)	4.1	3.5	9.2	3.76	2.16	2.89

T1- RDF (75:50:40 Kg ha^{-1})
T2- T1 + $ZnSO_4$ @ 12.5 Kg ha^{-1}
T3- T1 + $ZnSO_4$ @ 25 Kg ha^{-1}
T4- T1 + Zn lysinate @ 4Kg ha^{-1}
T5- T1 + Zn lysinate @ 2Kg ha^{-1}
T6- T1 + Zn lysinate @ 1Kg ha^{-1}
T7- T1 + Zn lysinate 0.5% foliar at 20DAC
T8- T1 + T4 + Zn lysinate 0.5% foliar at 20DAC
T9- T1 + T5 + Zn lysinate 0.5% foliar at 20DAC
T10- T1 + T6 + Zn lysinate 0.5% foliar at 20DAC

Table 4: Effect of Zn lysinate on leaf length, leaf breadth and chlorophyll content at harvest

Trt.	15 DAC		30 DAC		45 DAC		Chlorophyll		
	Leaf length	Leaf breadth	Leaf length	Leaf breadth	Leaf length	Leaf breadth	15 DAC	30 DAC	45 DAC
T1	63.11	2.38	82.36	2.49	113.58	3.13	37.53	39.93	42.36
T2	67.25	2.81	86.85	3.09	118.74	3.52	43.46	43.96	47.55
T3	75.65	3.22	94.18	3.43	134.47	3.70	45.83	46.08	48.55
T4	76.17	3.07	95.04	3.39	128.13	3.73	46.94	47.55	52.07
T5	71.36	2.78	91.66	2.94	125.92	3.61	42.06	44.86	46.98
T6	66.73	2.58	88.20	2.78	125.40	3.37	38.18	39.62	43.49
T7	63.99	2.30	83.54	2.50	116.13	3.17	39.60	45.00	48.91
T8	77.64	3.05	98.89	3.38	148.21	4.56	45.16	51.40	56.99
T9	70.79	2.85	94.97	2.93	140.43	4.09	44.09	48.20	53.47
T10	65.20	2.75	90.92	2.82	133.85	3.54	40.92	43.30	48.70
SEd	1.45	0.16	1.44	0.13	1.89	0.16	2.08	3.01	1.04
CD (P= 0.05)	2.94	0.33	2.92	0.27	3.82	0.33	4.22	6.11	2.11

Table 5: Effect of Zn lysinate on green fodder, dry fodder yield and crude protein content

Treatment	GFY (ton ha^{-1})	DFY (ton ha^{-1})	Crude Protein (%)
T1	66.45	12.62	7.43
T2	88.41	16.80	7.54
T3	92.05	17.49	7.74
T4	121.39	23.06	7.85
T5	104.66	19.89	7.78
T6	91.58	17.40	7.50
T7	81.05	15.40	7.65
T8	137.54	26.13	8.93
T9	113.29	21.53	8.58
T10	106.89	20.31	8.26
SEd	6.06	1.15	0.10
CD (P= 0.05)	12.29	2.34	0.21

Conclusion
Usage of organic synthesized chelated micronutrients along with the NPK recommended fertilizers improves the growth, yield and nutrient status of fodder crops. The green fodder and dry fodder yield increase about 51.68% and 51.58% over the control and the crop growth parameters also shows positive effect throughout the research. So, chelated Zn can be recommended to use as a cheap source of amino acids and solve the problems of micronutrients thus improves soil and crop quality.

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