

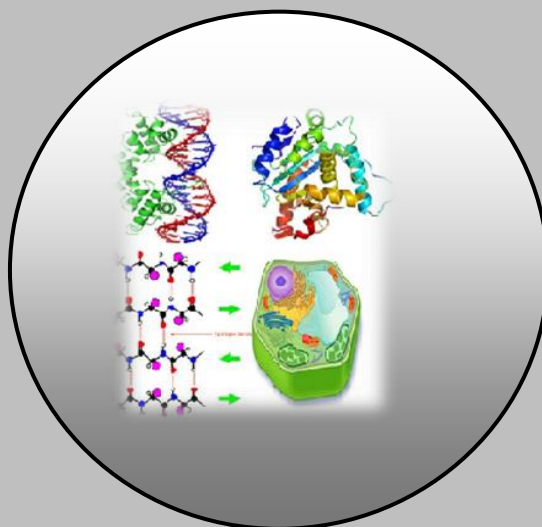
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***Grindelia camporum* Greene, A resinous Petrocrop
for sulphur-excess Wastelands**

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ABSTRACT

For generating information, hitherto not much available, on the nutritional, including S, aspects of petrocrop productivity, a pot-culture response study was made of graded soil S amendment (upto 100/ ug g⁻¹) as CaSO₄ .2H₂O on the biomass and biocrude productivity of a resinous petrocrop, Grindelia camporum Greene of family Asteraceae. The S-deficient, mildly calcareous Gomti up land alluvial soil (Entisol) of Mahibullapur, Lucknow, India was made adequate in all the other limiting nutrients. Ca increment through CaSO₄ was equated through CaCl₂ amendment. Plant at nil and 10/ug g⁻¹ applied s upto 12/ug g⁻¹ CaCl₂ and 23/ ug g⁻¹. Morgan's exhibited visible effects of S-deficiency. Plants at 100/ ug g⁻¹ S produced maximum top-biomass, most markedly in grindelane diterpenoid rich fruits (achenial aggregates), and biocrude and had highest (0.39%) concentration of tissue S. under S-deficiency, requirement of S for biocrude appeared being met preferentially over biomass. This along with S-utilization quotient ratio values, lower for biomass and slightly higher for biocrude with increasing rates of applied S, indicated biosynthetic role of S for the secondary metabolites of the biocrude. The critical levels of plant and soil S and the S-dose needed for optimal biomass and biocrude productivity indicated a high S requirement of the petrocrop. Suitability of this petrocrop for wastelands having low N, P, K, and Ca and high Fe and S was, thus, indicated.

Key words: Biomass & Biocrude; calcareous Entisol; Critical-S; grindelia camporum; resinous Petrocrop; S-excess Tolerance; S-response

INTRODUCTION

S-deficiency as well as it's excess is quite frequent in tropical soils (Dudal, 1976. and Mehrotra, 1991), the former in sandy, coarse textured and highly weathered Ultisols, Alfisols and Inceptisols of arid and semi-arid tropics (Mehrotra, 1990) and the latter in old, degraded and poorly drained humid tropical rice soils of the east including Indian acid sulphate 'kari',

some gypsiferous saline soils and vertisols, and S-rich-coal mine spoils (Mehrotra, 1991 and Eaton, 1965) Both arid to semi-arid and humid to sub-humid tropical regions in India, as also elsewhere, abound in waste and underutilised lands where large scale energy plantations, including those of petrocrops is being aimed at to meet the global energy deficit (Calvin, 1987. and Usmani, 1987). Little information is available, except that generated in the author's laboratory under a Govt. of India D.N.E.S. Research Project, on the nutritional, including S, aspects of biomass and biocrude productivity of petrocrops. Hence, with that aim in view, the present soil-pot culture study on the response of applied S to *Grindelia camporum* Greene, a resinous petrocrop in a calcareous S-deficient alluvial soil (Entisol) was undertaken. The petrocrop, with a high potentiality under Indian conditions (Srivastava, 1986) is, according to (Hoffman, 1983), capable of producing 11 tons biomass 12.7 bbl biocrude $\text{ha}^{-1} \text{yr}^{-1}$. Grindelane diterpenoids from the plant also possess aphid feeding deterrent activity (Rose et al. 1981).

MATERIAL AND METHODS

A mild calcareous Gomti upland soil (CaCO_3 1.25%, pH 1:2 CaCl_2) (0.1N) 6.98, pH 1:2 water 7.53, $\text{Ec}_2 \text{ mS cm}^{-1}$ at 25°C 6.43), deficient in available S (CaCl_2 extr. 6.7 ppm), was collected in bulk from Mahibullapur, Lucknow, U.P., India. The soil was adequately fertilized through basal applications of N, P_2O_5 , K_2O , Cu, Zn, Mo, Mn, Fe and B at 25, 22.2, 40.5, 2.5, 0.5, 10, 10 and 0.5/ ug g^{-1} (w/w), respectively. The amended soil was divided into 5 lots. Each lot was separately amended with A.R. grade $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$ at 10, 25, 50 and 100 / ug g^{-1} S (w/w), with no S amended serving as control. The soil was filled in 25 cm clay flower pots whose inside surfaces were painted with bitumen and lined with clean alkathene. There were 4 pots for each treatment arranged in two blocks in a randomized fashion. Five *Grindella camporum* Greene seedlings (4 weeks old), initially raised through seeds in purified sand, were transplanted in each pot. Plants were irrigated with de-ionised water, according to need, in a manner that they did not wilt and excess water did not drain out. To meet the N requirement, additional 12.5 ppm N was top-dressed at 21 weeks after transplantation (wat). Plants of all the treatments were given 8 bi-weekly sprays of 56 ppm Fe^{+3} -EDTA (prepared from FeCl_3) solution, starting from 9 wat.

A periodical record was made of the growth (height, branching, foliation, leaf area) and visual effects. The plants were harvested at 26 wat by cutting at the ground level, chopped, sun-dried and estimated for dry biomass. A portion of the sun-dried material was oven-dried at 70°C for 48-72 h for the estimate of dry matter yield. The dry biomass from each of the different S treatments was separately ground in a Wiley mill and utilized for the estimation of biocrude through sequential soxhlet extraction in ethyl acetate and methanol (Timmermann et al. 1983). For nutrient analysis, known portions of the fresh plant material were cleaned against surface contamination by successive washings in a detergent, 0.01N HCL and deionised water before oven-drying. Tissue S was estimated in the nitric perchloric wet-digests (1:15:1) of the oven-dried plant material (Piper, 1942) turbidimetrically (Williams, 1959).

After harvest, composite soil samples were drawn and estimated for available S. Available soil S was extracted by three extractions: 0.15% CaCl_2 sulphur (Williams and Steinberg, 1959), 0.5M NHCO_3 Ph 0.5 (Kilmer and Nearing, 1960) and Morgan's reagent (Harward et al., 1962). In the soil extracts, S was estimated as for plants.

All the data were statistically analysed and tested for significance at $p=0.05$ (Panse and Sukhatme, 1961). Critical S values, both for available and tissue S, indicative of optimal, threshold of deficiency for the biomass as well as biocrude yield were worked out through extrapolation by the method described by (Agarwala and Sharma, 1979).

Available soils

Soil S amendment, range $10\text{-}100/\text{ug g}^{-1}$, increased all the three forms of available S. However, a good part of the applied S as SO_4^{2-} became non-extractable (fixed) whose magnitude increased with increasing levels of applied S. The magnitude of such S-fixation was least in the case of NaHCO_3 extractable S (Table 1). The values of available soil S, CaCl_2 as well as Morgan's for soils of control (S_0) and up to $10/\text{ug g}^{-1}$ applied S (S_1), were indicative of its deficiency ($<10/\text{ug g}^{-1}$ for CaCl_2 - Williams and Steinberg¹⁵, and $<20/\text{ug g}^{-1}$ for Morgan's – (Harward et al., 1962).

Visible effects

Within 6 watt, the plants at S_0 and S_1 levels exhibited overall growth depression. Later S_0 (control) plants developed visible effects of S-deficiency i.e., chlorosis, by 9watt, the size of young leaves got markedly reduced. The apices of young chlorotic leaves turned necrotic and the markedly reduced sized middle leaves developed irregular necrotic patches on their lamina. By 14 watt, plants developed visible effects of S-excess i.e., the mature leaves, with reduced size, developed apical and marginal necrosis, and incurring of margins leading to cupping and ultimate drying up of the affected leaves.

Growth

By 14 watt, maximum growth of plants in terms of height, branching, leaf number and average area of leaves was found at S_3 ($50/\text{ug g}^{-1}$ S) (Table 2). This S treatment had $13.3/\text{ug g}^{-1}$ CaCl_2 and $30/\text{ug g}^{-1}$ Morgan's S (Table 1). At this growth stage, plants at S_4 ($100/\text{ug g}^{-1}$ S) had reduced growth in terms of all the parameters. However, at maturity (26 watt), maximum growth of plants in terms of all the growth parameters were found at S_4 ($100/\text{ug g}^{-1}$ S) which had $16.7/\text{ug g}^{-1}$ CaCl_2 and $32/\text{ug g}^{-1}$ Morgan's S. the S_4 plants also had the maximum productivity of capitula, both in terms of number and 100 capitula weight. No capitula were formed at S_0 (control). A higher requirement of S at the post-flowering stage/reproductive phase of plant growth was, thus, indicated.

Biomass

Maximum vegetative as well as total above-ground plant biomass was found at S_4 ($100/\text{ug g}^{-1}$ S) which had $16.7/\text{ug g}^{-1}$ CaCl_2 and $32/\text{ug g}^{-1}$ Morgan's S. S amendment, in the range of 25 to $100/\text{ug g}^{-1}$, also increased tissue dry matter percentage or decreased level of tissue hydration in the above ground plant parts (Table 2).

Biocrude

Maximum biocrude yield, both of total extractives as well as its ethyl acetate and methanol extractive components, particularly of the former, was found at S₄ (100/ug g⁻¹ S) having 16.7/ug g⁻¹ CaCl₂ and 32/ug g⁻¹ Morgan's S (Table 3).

Tissue S-concentration and uptake

Soil S amendment, in the entire range (10 to 100/ug g⁻¹), markedly increased both concentration and total content of S in the above ground plant parts. While the increase at S₄ (100/ug g⁻¹ S) *vis a vis* control, was only 3 folds for tissue concentration, it was nearly 6 folds for the S uptake (table 3). S amendment markedly decreased the S-utilization quotient ratio for biomass but slightly increased it for the biocrude (Table 3).

Critical soil available and tissue S

The critical values indicative of S-excess, threshold or severe could not be determined in the range of applied S (up to 100/ug g⁻¹) of the present study. The critical S values indicative of optimum and threshold of deficiency were found to be almost similar for biomass and biocrude (Table 4). However the values indicative of severe deficiency were lower for the biomass than for the biocrude. Such available S value indicative of severe S- deficiency for biomass and biocrude productivity, respectively were 10.3 and 13-12.4/ug g⁻¹ for NaHCO₃ extractable soil S, and 0.20 and 0.25-0.27% for tissue S.

DISCUSSION

Higher extractability of S by NaHCO₃ pH 8.5 extractant could be attributed to the dissolution of CaCO₃ by HCO₃⁻¹ and the consequent occlusion of CaCO₃—chemie-adsorbed SO₄⁻² in solution (Virmani, 1971). Though anionic form of soil applied S is generally not considered fixed (Kardos, 1964), the present findings regarding non-extractability of bulk of the applied S by all the different soil extractants, being least with NaHCO₃ pH 8.5, however, suggests otherwise. This could possibly be due to some inherent edaphic property like calcareous.

In the above ground plant parts, S amendment related increase in the tissue dry matter percentage could be attributed to the role of S in increasing photosynthesizing ability of plants (Mengal and Kirkby, 1982). The improvement in biomass by applied S was most marked in the reproductive (fruit) yield than in the vegetative biomass. A greater requirement of S during the reproductive phase of plant growth, especially for the productivity of gridelane diterpenoid-rich fruits and seeds (Timmermann, et al., 1985) was, thus, suggested. This was further confir,ed by the observation of visible phytotoxicity of S during vegetative growth phase at a level of applied S (100/ug g⁻¹) where maximum biomass was found when the the plants turned reproductive and attained maturity (Table 3). In controlled sand culture study with a laticiferous petrocrop,

Pedilanthus tithymalocides also, the S requirement found to increase with plant's maturity (Mehrotra et al., 1991).

Table 1. Effect of graded S amendment (as CaSO₄ .2H₂O) to a Gomti upland alluvial soil of Mahibullapur, Lucknow, on available soil sulfur: Data after harvest (26 watt) of *Grindelia camporum* Greene plants grown in pot – culture.

Soil applied S (/ug g ⁻¹)					L.S.D. p0.05
Nil	10	25	50	100	
(S ₀)	(S ₁)	(S ₂)	(S ₃)	(S ₄)	
Available soil S (/ug g⁻¹) : 0.45% CaCl₂ extractable					
6.7	9.2(75)	11.7 (80)	13.3 (87)	16.7(90)	4.4
: Morgan's reagent extractable					
15.8	20.8(58)	22.5(73)	30.0 (72)	31.7 (85)	12.5
: 0.5 M NaHCO₃, pH 8.5 extractable					
26.7	35.5(17)	40.0 (47)	41.7(70)	45.0(82)	4.6

Figures in parentheses denote the percentage of applied S rendered non-extracting (fixed in the soil) after plants harvest.

Table 2. Growth response of *Grindelia camporum* Greene plants to graded S amendment in a Gomti upland alluvial soil of Mahibullapur, Lucknow in pot-culture.

Growth parameter	Growth Stage (wat)	Soil applied S (/ug g ⁻¹)					L.S.D. p0.05
		Nil	10	25	50	100	
		(S ₀)	(S ₁)	(S ₂)	(S ₃)	(S ₄)	
Height-tops	14	9.5	14.5	13.3	25	19	4.5
(cm)	26	33.3	39	45.5	48.3	66.8	24.4
Secondary branches (No. plant)	26	0	0	0.25	0.75	4	1.61
Leaves	14	8	14	16.5	20	18.5	5.8
(No. plant)	26	24	26	30	32	62	11.1
Mean average	14	5.16	5.25	6.22	7.7	6.54	0.79
leaf area (cm)	26	6.53	9.56	10.37	10.56	12.56	4.37
Capitula	14	0	0	0.5	0.5	0	
(No. plant)	26	0	2.5	4	3.5	8.5	
100 capitula weight (g)	26	0	6.4	9.6	10.3	13	

Wat = weeks after transplantation.

Table 3. Biomass, biocrude and tissue –S response of *Crindelia camporum* plants to graded amendment in Gomti uplant alluvial soil of Mahibullapur, Lucknow.

Plant attribute	Soil applied S (/ug g ⁻¹)					L.S.D. p0.05
	Nil	10	25	50	100	
	(S ₀)	(S ₁)	(S ₂)	(S ₃)	(S ₄)	
Dry matter yield (g plant⁻¹)						
- capitula	0	0.06	0.13	0.21	0.9	0.18
- total tops	2.76	2.85	3.36	3.57	4.68	1.54
Dry matter(%)- tops	26	26	29.2	28.2	31.3	8.9
Top-biomass (g g⁻¹ tissue)	769	564	423	399	259	
Tissue S-tops (% D.M.)	0.13	0.18	0.23	0.27	0.39	0.03
Tissue S-tops (mg plant⁻¹)	3.6	5.1	8	8.9	18.1	2.4
Biocrude - tops (% D.M.)						
Et ₂ OAC extractable	2.7	-	4.8	7.8	8.7	1.2
MeOH extractable	2.6	-	4.5	6.8	7.6	0.9
Total (Et₂ Oac+OeOH)	5.3	-	9.3	14.6	16.3	2
Biocrude yield tops (g plant⁻¹)						
Et ₂ OAC extractable	0.07	-	0.16	0.29	0.41	0.03
MeOH extractable	0.07	-	0.15	0.24	0.36	0.01
Total (Et₂ OAC+MeOH)	0.14	-	0.32	0.52	0.76	0.3
biocrude (g g tissue S)	39	-	40	58	42	

-= Not estimated, Et₂ OAC = Ethyl acetate, MeOH = Methanol.

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Table 4. Critical S values available soil and tissue S for *Grindelia camporum* Greene plants grown in Gomti upland alluvial soil of Mahibullapur, Lucknow in pot-culture.

Productivity (g pant)	Critical S value*		
	Optimal	Deficiency	
		Thershold	Severe
	<u>Available soil S (/ug g⁻¹): 0.45% CaCL₂ extractable</u>		
Biomass - tops	16.7	15.5	10.3
Total Biocrude - tops Et ₂ OAC extractable	16.7	15.3	13.1
	<u>: Morgan's extractable</u>		
Biomass - tops	31.7	31.1	20.8
Total Biocrude - tops Et ₂ OAC extractable	31.7	31.3	27.9
	<u>: 0.5 M NaCHO₃, pH 8.5 extractable</u>		
Biomass - tops	45	43.8	37.5
Total Biocrude - tops Et ₂ OAC extractable	45	43.7	41.2
	<u>Tissue (top) - S (% dry matter)</u>		
Biomass - tops	0.39	0.34	0.2
Total Biocrude - tops Et ₂ OAC extractable	0.39	0.35	0.25

* Excess value could not be determined in the range of S amendment used.

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