

Variation of Plant Macronutrients in Sisal (*agavesisalana*) Leaves Biomass

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Abstract: - Sisal growing is one of the key agricultural activities in Kenya. Sisal is a high waste generating plant as 97% of its leaves are a waste. Disposal of the waste generated has remained a challenge and are burned to ashes or used as landfills. They are hardly used in promoting plant growth despite their potential. Discarding the biomass arises from the fact that they are acidic and when used would burn crops! When composted however acidity reduces and so is a potential fertilizer. The study set out to determine levels of N, P, K and Ca in relatively young, old, smooth and thorn edged sisal leaf biomass. Sisal leaf samples were collected from farms in Lugari, western Kenya and used to generate sample wastes that were used in the laboratory analysis. Digested samples were subjected to standard procedures and methods: Kjeldhal (N), Spectrophotometry (P) and Photometry (K and Ca) to establish the levels. It was established that sisal leaf wastes analyzed had 1719.00 ± 516.86 mg/100 g of nitrogen, 257.19 ± 131.41 mg/100 g of phosphorous, 2154.94 ± 471.45 mg/100 g potassium and 3419.20 ± 1234.39 mg/100 g calcium. It was further noted that apart from nitrogen all other macronutrients were significantly higher in relatively young leaves compared to correspondingly older leaves. Both potassium and calcium were also significantly high in thorn edged leaf varieties compared to the smooth edged ones. It is hoped that results of this study will influence the disposal mechanism shifting it from the present methods to composting or otherwise that would enable their utilization in promoting plant growth.

Key words: Sisal, waste, disposal, macronutrient levels, composting, fertilizer.

I. BACKGROUND INFORMATION

Of the 240 tons of sisal fibers produced globally, Kenya produces 27,600 tons [4]. Sisal occupies 6th place among fibre plants representing 2% of the world's production of the plant fibres [22]. It grows best in a hot climate and may be grown throughout humid and sub humid low land tropics [29]. In Kenya it is grown as plantations in Taita Taveta, Kilifi, Voi and Mwatate within the coastal region, Kibwezi in eastern region, Solai in Nakuru, Juja in Thika and as a demarcation to distinguish land ownership in other areas like western Kenya. As a cactus Agave plants survive and produce marketable product in fertile and arid regions which in many cases would otherwise be unproductive. It is a high waste generating industry with a current ratio of useful fibre to waste at 2.98% [19]. Recent studies on the anaerobic digestions of the waste to produce biogas have been found feasible [16]. Possible utilization in electrical production has equally been reported [23; 13]. The protein content is reported as 0.31g/100 g and mineral material 1.03 g/100 g [27]. The levels in sisal leaf waste provide the basis for the potential of using them as fertilizer. Attempts in the direct utilization of the wastes in the

cultivation and yield performance of *Coprinuscinereus* (schaeff) Gray established that the mixture of the wastes and cow dung gave the highest yield of mushrooms [21].

Utilization of sisal leaf wastes by communities growing sisal to promote growth of other plants has been limited in Kenya partly because the leaf extracts are acidic and direct use burns crops. When composted however the pH value rises. [25] Reported composting as a sustainable sisal waste management technique. Composting would therefore make sisal biomass use in farming viable. Though sisal plant leaf macronutrients composition has been reported their variation with leaf age or nature of the edge has not been reported.

II. MATERIALS AND METHODS

Three sisal leaf samples (young and old) were collected in duplicate from each sisal plant on farms in Lugari, western Kenya. Apart from relative age criteria samples collected were either smooth edged and thorn aged. The leaves were stripped before sisal fibers were removed to obtain sisal leaf wastes (SLW). The wastes were subjected to standard digestion procedures before instrumental analysis to establish levels of nitrogen, phosphorous, potassium and calcium.

Materials included concentrated sulphuric acid (18 M); standardized hydrochloric acid (0.01M) made by adding 2.207 ml of concentrated acid (35% purity) to one litre of distilled water before 10 ml of it was diluted to 250 ml; the salt and catalyst mixture; the sodium hydroxide (10 molar) made by dissolving 40 g NaOH in 100 cm³ distilled water; boric acid.

In the determination of nitrogen, 0.5 g of samples (SLW) was weighed into digestion flasks alongside blanks. Spatula end full of the salt/catalyst mixture made by grinding 20 g Na₂SO₄ with 2 g copper in a mortar using a pestle was added to the samples, mixed well before 10 ml of concentrated sulphuric acid was added. The mixture was slowly heated to 200° C until frothing subsided before the temperature was adjusted to 350°-375° C and heating continued until a clear solution was obtained. The mixture was cooled to room temperature before topping up to 20 ml with deionised water. 50 cm³ of 8% boric acid (H₃BO₃) made by dissolving 40 g H₃BO₃ in 1000 cm³ of distilled water, warmed with stirring were measured into the 250 ml beaker. A funnel was connected to the Liebig condenser and immersed into boric acid solution to collect the ammonia liberated. About 0.6 g of Devard's alloy was added to the 20 ml digest before 30 ml of 10 molar NaOH was added with stirring. The tube to Kjeldahl distillation apparatus was connected to start the steam distillation that went on for 12 minutes. The NH₃ distilled into H₃BO₃ solution was then

titrated using standard 0.01MHCl. The end point 4.65 was monitored with a pH meter.

$$\text{Total mass (mg/100g), DM} = \frac{140,000 \times V_{axT}}{M_o \times M_s}$$

Where V_a = volume of the acid used

T_a = molarity of the acid used

M_o = % moisture M_s = mass of the sample used

In the determination of potassium, Calcium and Phosphorous, 0.5 g of the samples were weighed into separate digestion flasks. 10 ml of lanthornum solution made by dissolving 1.727 g of oxide in 8 ml concentrated hydrochloric acid, stirred and diluted to 1 litre with de-ionized water was added to the weighed sample. 10 ml of the concentrated nitric acid was then added to the mixture, heated on a hot plate, cooled, 10 ml more HNO_3 were added and heating continued. The mixture was cooled again before 2 ml of perchloric acid was added and the mixture heated until white fumes were seen. The resulting solution was made up to 100 ml. Portions were run in flame photometer at 766.5mm for potassium, 422.7 nm calcium and in spectrophotometer (Model Cecil-CE 2041 2000 series) at 660 nm phosphorous alongside standard solutions.

The standard solutions were prepared from stock solutions. The potassium stock solution (1000 ppm) was made by dissolving 0.1 g of KCl into 50 ml of de-ionised water. 2.5 ml of this stock solution was measured into 100 ml de-ionised water in a 250 ml volumetric flask. 10 ml of concentrated HCl was added before dilution to 250 ml to get the working solutions. Aliquotes of 4,6,10,12, and 15 ml of this working solution were separately diluted to 50 ml with deionised water to obtain the standard solutions with ranges 1.30 mg-10 mg potassium.

In the preparation of calcium standards 4.00 g of calcium carbonate were weighed into 50 ml volumetric flask. 2 ml of

1M HCl were added. After effervescence had subsided the mixture was topped up to the mark with de-ionised water giving rise to the stock solutions. 2.5 ml of the stock solution was then diluted to 250 ml to get the working solution. Aliquotes of 5, 10, 15, 20 and 25 ml of the working solutions were diluted to 50 ml to get standards.

Phosphorous standards involved dissolution of 0.23 g of KH_2PO_4 in 50 cm^3 to get the stock solution. 2.5 ml of this solution was then diluted to 250 ml. Aliquotes 10, 20, 40 and 60 ml of the working solutions were diluted to 100 ml to get the standards. Other reagents included acid molybdate stock solution which was made by dissolving 6.2 g of ammonium molybdate into 80 ml deionised water, heated to 60° C. The mixture was allowed to cool before adding about 0.7 g of antimonyl potassium tartrate. The flask holding the mixture was placed in an ice bath and slowly 70 ml of conc. H_2SO_4 added. Upon cooling it was diluted to 250 ml and stored in brown bottles at 4°C. The ascorbic acid stock solution made by dissolving 10.56 g of ascorbic acid was dissolved in 75 ml distilled water and diluted to 100 ml. The mixture was equally stored at 4°C. The working solutions were derived by adding 20 ml of the acid molybdate stock solution and 10 ml of the ascorbic acid stock solution to 800 ml of de-ionised water before diluting to 1 litre. In the colour development 1 cm^3 of either sample or standard solutions were added 100 ml of the working solutions and colour development allowed for 35 minutes. Absorbances by standard solutions were used to obtain regression equations used to calculate levels corresponding to absorbance by sisal leaf digest samples.

III. RESULTS AND DISCUSSION

The effects of different farms, plant sample, nature of the edges (normal smooth N or thorned T) and age (old-O or young-Y) on levels of macro elements were determined at 95% confidence. Table 1 shows results.

Table 1: The mean \pm Std Dev. levels of N, P, K and Ca in sisal leaf wastes

Sample	Mean macronutrient levels \pm Standard deviation (mg/100g, DM)			
	Total Nitrogen	Phosphorous	Potassium	Calcium
F ₃ N ₁ Y	1310.80 \pm 9.504	361.27 \pm 3.050	2618.00 \pm 4.57	2800.8 \pm 174.10
F ₃ N ₂ Y	1322.20 \pm 15.124	417.75 \pm 5.41	1961.10 \pm 4.88	3180.8 \pm 3.46
F ₂ N ₁ Y	1264.40 \pm 24.364	459.41 \pm 12.12	2308.00 \pm 9.57	2643.4 \pm 57.14
F ₂ N ₂ Y	1040.10 \pm 23.473	419.25 \pm 21.79	1663.70 \pm 6.18	1945.2 \pm 14.582
F ₁ N ₁ O	2459.60 \pm 86.483	244.85 \pm 18.78	2708.00 \pm 5.57	3898.1 \pm 0.864
F ₁ N ₂ O	2154.40 \pm 147.394	176.50 \pm 11.72	2063.10 \pm 6.88	3959.0 \pm 7.029
F ₄ N ₂ O	2044.90 \pm 34.573	110.01 \pm 0.71	1763.10 \pm 9.18	3827.2 \pm 94.69
F ₉ T ₁ O	2155.20 \pm 31.573	206.87 \pm 11.04	2537.60 \pm 0.88	6847.8 \pm 174.10
F ₆ T ₁ Y	1550.80 \pm 12.504	437.86 \pm 1.58	2708.00 \pm 5.57	5180.8 \pm 5.46
F ₉ N ₁ Y	1602.20 \pm 35.124	307.82 \pm 0.86	1863.10 \pm 6.88	2243.4 \pm 50.14
F ₇ N ₁ O	2004.40 \pm 27.164	147.43 \pm 0.83	2062.10 \pm 0.40	3945.2 \pm 114.582
F ₅ T ₂ O	1246.70 \pm 13.073	405.21 \pm 20.57	2158.00 \pm 8.71	2858.1 \pm 0.864
F ₄ T ₁ Y	1499.60 \pm 82.183	286.58 \pm 0.69	1908.60 \pm 6.13	2959.0 \pm 5.009
F ₇ N ₂ O	2351.40 \pm 67.394	165.18 \pm 3.81	2190.30 \pm 16.27	3627.2 \pm 204.69
F ₉ T ₂ O	1944.90 \pm 24.513	156.11 \pm 0.05	3023.00 \pm 156.46	5885.2 \pm 1.350
F ₈ N ₂ O	2415.30 \pm 35.503	96.28 \pm 2.77	1579.10 \pm 1.39	3703.1 \pm 1.501
Overall mean	1772.93 \pm 469.57	274.90 \pm 127.595	2194.68 \pm 420.51	3719.02 \pm 1309.17
p-value(Farm)	0.027	0.018	0.173	0.022
p-value(edge)	0.821	0.698	0.017	0.004
p-value(age)	0.000	0.000	0.676	0.017

Key: F- Farm, T- Thorn edged, N-Smooth edged, O-Old, Y-Young

Nitrogen

The mean levels of total nitrogen were found to average 1772.930 ± 469.574 mg/100g giving a range of 1040.10 – 2459.60 mg/100 g. The value found is slightly higher than 1.46 ± 0.02 % (or 1460 ± 200 mg/100 g) established in a separate study [15]. A one way analysis of variance (ANOVA) to establish variations in the levels of total nitrogen was done. It was found that at 95% confidence level there was a significant difference ($P=0.027$) when data values from different farms disregarding age and nature of the edge were compared. The plant soil environment [6], inputs including fertilizer application [3] determine mineral levels in plant parts.

An ANOVA with nature of edge as variable showed no significant ($p = 0.821$) variation. The nature of edge whether smooth or with thorns did not affect levels of nitrogen. However when the age of the sisal leaves was a factor, a significant difference ($p = 0.000$) was noted. Relatively older leaves exhibited higher levels of total nitrogen compared with younger leaves from the same plant. This compares well with the results obtained in the integrated analysis of nitrogen status in cucumber plants that found nitrate content in petiole sap was much higher in older leaves than in younger leaves when planted in full strength of coopers nutrients' solution [8]. While studying environmental factors and cultural measures affecting the nitrate content in spinach it was found that NO_3^- contents were highest in petioles and older leaves [2]. The same study noted that complete or partial replacement of NO_3^- nitrogen by NH_4^- nitrogen caused NO_3^- content in spinach to decrease meaning the levels of nitrogen are dependent on supply within the crop environment. This however is dependent on effectiveness of nitrification inhibitors, the soil type and the amount of available nitrogen.

Phosphorous

The overall mean \pm standard deviation levels of phosphorous found in this study was 274.90 ± 127.595 mg/100 g disregarding location, or age or edge characteristic of the sisal leaf sampled. This gave a range of 96.28-459.41 mg/100 g. This compares well with the 250 mg/100 g reported in the audit and characterization of sisal post-harvest wastes as a bio resource [15]. According to Alberta Agriculture and rural development guide on phosphorous application in crop production [11] phosphorous occurrence in plants ranges 0.1-0.4% i.e. 100-400 mg/100g. The two values are well within this range.

The quantity of phosphorous in the soil solution, even when relatively high is only in the range of 0.136 to 1.36 kg per acre [11]. From this study it means that sisal wastes can supplement 2.48% on the basis that though crops may differ in phosphorous needs but an average of 13.154 to 20.876 kg per acre of sisal leaf wastes would support the soil by this percentage. Considering the wastes generated annually of 611,875 tons in Kenya [19] and 1,222,000 tons in Tanzania [14] their utilization then would go a long way to improve

crop production. They are discarded otherwise as wastes causing environmental pollution.

A one way analysis of variance was done on the data generated. Individual farm as a factor showed significant variation ($P = 0.018$) in the levels of phosphorous. Samples from different plants but within the same environment differed. This is likely because mineral levels in plant parts are affected by factors such as variety, time of harvest, climate and soil conditions including fertilizer application [3]. The moisture and mineral composition of leaves furthermore show large variations in mineral composition due to cultivar, plant age, ecological conditions and cultural practices [26] and [7].

Sisal leaf wastes analyzed were grouped as young and old. A significant difference ($P=0.000$) was recorded when the relative age of the sisal leaves was considered as a factor. The mean values varied from 96.28 mg/100 g to 459.41mg/100 g in the samples investigated for phosphorous. It is noted that old leaves had lower levels of phosphorous than younger leaf wastes. The outcome that young leaves had higher levels of phosphorous than relatively old leaves could be because phosphorous readily trans-locates within plants moving from old leaves to young tissues as the plant forms cells and develops root, stems and leaves. Young actively growing plant tissues have more abundant phosphorous than in older tissues [11]. The nature of the sisal leaf edge (whether with thorns or smooth) did not affect the levels of phosphorous in sisal wastes.

Potassium

The study sought to establish the levels of potassium in sisal wastes generated from sisal leaf samples. On average sisal wastes collected and analyzed showed potassium levels of 2154.90 ± 420.51 mg/100 g. This was in the range 1579.10-2708.00 mg/100 g. While studying the chemical characteristics of sisal bole wastes (SBW) and remnant leaf stubs, [15] found the levels of potassium as 1340 mg/100 g and 5260 mg/100 g respectively. In the auditing the characteristics of sisal wastes as bio resource for value additions [18] reported levels of potassium in assorted sisal wastes- sisal leaf decortification residues as (5.56 mg/100 g), sisal leaf decortification (waste water as (18 mg/l), sisal leaf decortification waste (fresh) as 4.28 mg/l and sisal leaf decortification waste dry (9.96 mg/l). Typical values of potassium in plants are quoted to range 3-4 % [24] with young leaves having 3-5 % range [20]. Potassium has no structural purpose but it is the most common cation in the plant biochemical process that functions in the protein synthesis, photosynthesis and transport of sugar from leaves to the fruits. The action of potassium on protein synthesis enhances conversion of nitrate into the protein which contributes to enhanced efficiency of supplied nitrogen fertilizers [24].

Sisal leaves can either have smooth edges or be with thorns. When this factor was considered a significant difference ($p = 0.017$) was noted at 95% confidence level in the levels of potassium. The difference was brought about by tendency of

those with thorny edges having higher levels of potassium in their leaf wastes compared with those with smooth edges. Samples F₆T₂ (2158.00), F₅T₁ (2537.00), F₆T₁ (2708.60) and F₅T₂ (3023.00) clustered higher than smooth edged samples F₄N₂ (1519.00), F₈N₁ (1863.10), F₇N₁ (2062.10) in mg/100 g. Young leaves too had relatively higher levels compared with older ones from the same plant as observed on samples F₄N₂ (1763.00 mg/100 g), F₄N₁ (1908.60), F₈N₂ (1579.10 mg/100 g) and F₈N₁ (1863.10 mg/100 g), F₆T₂ (2158.00 mg/100 g) and F₆T₁ (2708.60), F₅T₁ (2537.60 mg/100 g) and F₅T₂ (3023.00 mg/100 g). The nutrient content of a plant varies not only among its various parts but changes with age and stage of development [20]. There are also varietal differences that affect the nutrient content of various plant parts [3].

The potassium requirement of plants varies widely depending on the plant species. The potassium level in a plant can change quickly as potassium is quite mobile and moves readily within the plant. Because it is mobile in the plants, potassium deficiency symptoms appear in older plant tissues first. Thus potassium concentration in sisal leaves' tissues decrease with age [20] in the roles of nitrogen and potassium in plant nutrition. [1]while studying the uptake and distribution of potassium in tomato plant parts, reported that concentration of potassium in the stems, petiole and luminal tissues increased from the base to the apex of the plant irrespective of the potassium concentration in the nutrient feed. The same study also reported a gradient of decreasing potassium concentration along the leaves from the proximal to distal lamina. The concentration of potassium in all parts increased with supply.

Calcium

The mean levels \pm standard deviation of calcium was found to be 3719.02 \pm 1309.17 mg/100 g of the sisal leaf wastes. This is in close proximity to the findings of earlier studies. In a study on the integration of livestock systems into sisal production levels of calcium in sisal leaves are reported as 5720 mg/100 g [9]. In the analysis of maize ear leaf during silking and soy bean during the first trifoliolate at the time of early flowering found the parts analyzed to have 310-600 mg/100 g and 1100-2000 mg/100g as sufficient amounts respectively. Plants growing in adequate calcium in their natural habitat usually have shoot calcium concentrations of 0.1 and 5.0 % dry weight [10]. These values reflect both calcium available in the environment and contrasting calcium requirements of different plant species. Given that soils exchangeable calcium ranges from 300-500 mg/100 g [20] and that different plants require different amounts, utilization of sisal leaf wastes in low base saturation or high levels of acidic deposition can be a reliable supplement of this element in crop production [12]

A one way analysis of variance (ANOVA) was done with a view to establish any variations that may have existed between the sisal leaf wastes. This was considered with farms, age (young and old) and nature of the edges (with thorns or smooth) as the variables. At 95 % confidence limits the different individual sisal plant environment significantly

(P=0.022) affected the levels calcium in the samples. This was consistent with observations made in other studies. The moisture and mineral composition of leaves furthermore show large variations in mineral composition due to cultivar, plant age, ecological conditions and cultural practices [7, 26]).

A significant variation (P=0.004) between samples from thorn and smooth edged leaves was noted. The samples with thorn edges (F₅T₂O, F₅T₁O, and F₆T₁Y) showed higher levels of calcium in their leaf wastes than samples with smooth edges like F₈N₂O, F₄N₂O, F₇N₂O and F₇N₁O. It was also noted that young leaves were inferior in the levels of calcium than the corresponding older leaf waste of the same plant. Sample F₈N₂O (old leaves) from farm 8 had 3703.10 mg/100 g while younger leaves from the same plant and location F₈N₁Y showed 2243.40 mg/100 g. Equally 2643.4 mg/100 g in F₂N₁Y (younger leaves) were lower compared with 3959.0 mg/100 g (F₂N₁O). The range of calcium and magnesium in plant tissues varies considerably within a given crop and among crops [20]. Generally variation in the same plant species occurred because varieties differ in the amounts of nutrients they take up [20]. Old leaves contain more calcium and less potassium and magnesium than young ones [5]. The calcium content of leaves changes downward in the shoot and crown and from the crown peripheries to its center. Calcium uptake is passive and does not require energy input and therefore calcium uptake is directly related to the plant transpiration rate. Younger leaves show deficiency earlier because they have low transpiration rate. Calcium is immobile and cannot be redistributed, therefore only relies on immediate supply [28].

IV. CONCLUSION

Sisal (*Agavesisalana*) leaf wastes have significant amount of plant macronutrients nitrogen, phosphorous, potassium and calcium that can be concentrated to support plant growth. This would in turn solve disposal challenges of the biomass within sisal growing zones. As observed old sisal leaves had significantly higher values of nitrogen and calcium while relatively younger leaves had more of phosphorous and potassium. This means any formulation targeting planting which requires phosphorous relatively young leaves be involved while older ones being rich in nitrogen and calcium are involved in making side dressing formulations. It is recommended that further studies be done to not only establish methods of concentrating these macronutrients but also hasten accessibility by plants.

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