

# The Role of Muriate of Potash in Reducing Leaf Nicotine Levels

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**Abstract:** Nicotine is widely considered to be addictive, reducing consumer dependency on the constituent has been a focus of CORESTA regulatory groups in recent years. This study aimed to investigate the impact of NPK fertilizers, incorporating different potassium (K) sources, on nicotine content in K326 tobacco leaves. The research was conducted using a Randomized Complete Block Design (RCBD) at three sites (Chunya, Kahama, and Tabora) during the 2023–24 cropping season. Five fertilizer treatments were tested: control (T1), basal standard (T2: 30g N<sub>10</sub>P<sub>18</sub>K<sub>24</sub> with K sources from KCl and K<sub>2</sub>SO<sub>4</sub>, top-dressed with 8g CAN 27%), T3 (30g N<sub>11</sub>P<sub>22</sub>K<sub>21</sub> from KCl, top-dressed with 8g CAN 27%), T4 (two splits of 15g N<sub>11</sub>P<sub>22</sub>K<sub>21</sub> from KCl, top-dressed with 8g CAN 27%), and T5 (two splits of N<sub>11</sub>P<sub>22</sub>K<sub>21</sub> at 10g and 12.5g bi-weekly, without CAN 27%). Seedlings were transplanted into trial plots with 1.2m ridge spacing and 50cm intra-spacing. Basal applications were administered 14 days post-transplant for T2 and T3, and every two weeks for T4, with T5 receiving no CAN 27%. Results indicated that incorporating KCl and K<sub>2</sub>SO<sub>4</sub> with CAN 27% significantly increased nicotine levels (2.66%). In contrast, using KCl alone in NPK fertilizer reduced nicotine levels to 2.02%, but not tobacco leaf yields.

**Keywords:** nicotine; sulphate; muriet; potash; fertilizer

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## 1. Introduction

Cigarette smoking (*Nicotiana tabacum* L.) is widely known to cause addiction (Liu et al., 2023; Granata et al., 2024; Rao et al., 2024), and is a choice behavior for humans because of its related stimuli (aroma) attraction to smokers (Ferguson and Shiffman, 2009; Schröder et al., 2024). Despite the freedom to smoke cigarettes, the agronomic reduction of nicotine in tobacco leaves, could enhance the reduction of addiction to smokers. In addition, the World Health Organization (WHO) has recommended reducing cigarette filler nicotine levels to below 0.4 mg/g (Lewis, 2019).

Advancing tobacco addiction reduction through scientific collaboration has been a focus of regulatory groups in recent years under the CORESTA - Cooperation Centre for Scientific Research Relative to Tobacco (Lee et al. 2022; CORESTA, 2024). Therefore, the current focus is on lowering nicotine levels as an alternative strategy to reduce addiction among smokers. To reduce nicotine levels in cured tobacco leaves, agronomic practices are a crucial approach (Henry et al., 2019). Therefore, it is necessary to understand agronomic practices and identify factors that result in a reduction of nicotine in tobacco leaves.

Tobacco growth and development depend on N, P, and K nutrient availability in soils. Since soils are generally deficient in N, supplementation with fertilizers rich in a major inorganic N source ( $\text{NH}_4^+$ ) is essential (Coletto et al., 2023). The Tanzanian tobacco sector uses  $\text{N}_{10}\text{P}_{18}\text{K}_{24}$  with additional nutrients such as 7% S, 0.012% B, 3% CaO, and 0.5% MgO for basal application and CAN 27% N with 1.7% MgO, 3% CaO, and 3% S for top-dressing. In the 2022–23 crop season, a bag of  $\text{N}_{10}\text{P}_{18}\text{K}_{24}$  costed \$70.72, while a bag of CAN 27% costed \$59.99 (TTC, 2024). The compound  $\text{N}_{10}\text{P}_{18}\text{K}_{24}$  fertilizer's high cost is largely due to the tobacco's requirement for a specific potassium source, sulfate of potash (75%), and muriate of potash (25%). The muriate of potash contains more potassium than the sulfate of potash, and it is widely known to be a cheaper source of K than the sulfate of potash (Jan and ul Hussan, 2022). Thus, recommending a higher sulfate of potash by 75% as a source of K is associated with higher fertilizer costs. On the other hand, the two sources of K, KCl and  $\text{K}_2\text{SO}_4$ , differed from their anion-embedded counterparts, and both contain nutrients required by the tobacco crop. The KCl provides the micronutrient Cl, while  $\text{K}_2\text{SO}_4$  provides sulfur to the plant. However, KCl contains higher levels of K and solubility potential than  $\text{K}_2\text{SO}_4$  (Keeney, 2019). The disadvantage of  $\text{K}_2\text{SO}_4$  is that it is much more expensive than KCl, while the disadvantage of KCl is its higher salt potential than  $\text{K}_2\text{SO}_4$ . Therefore, based on this,  $\text{K}_2\text{SO}_4$  is mostly preferred for tobacco crops over KCl. The application of potassium sulfate fertilizer significantly had impact due to its minerals present. The effects results into enhancing tobacco biomass growth (Jiang et al. 2024). In connection to this, calcium sulfate is included in  $\text{N}_{10}\text{P}_{18}\text{K}_{24}$  fertilizer to improve soil quality and nutrient supply, which benefits tobacco plant growth and enhances cadmium (Cd) tolerance by suppressing Cd root uptake and lowering the Cd content in tobacco plants (Huang et al., 2022). Tobacco crops do not primarily use calcium from calcium chloride due to its influence on fungal biomass (Kortekamp, 2006).

In this case, the research trial's goal was to investigate if it would be possible to use NPK fertilizer, which gets its K from muriate of potash, to explore different agronomic techniques to lower leaf nicotine.

## 2. Materials and Methods

### 2.1. Site selection and location

The trial was conducted in low soil fertility; not been planted tobacco for the past ten years in three agro-ecological sites, Mtanila (Chunya, Mbeya), Tumbi (Tabora), and Ushetu (Kahama, Shinyanga). Soil samples were taken from all three sites to analyze the soil nutrient status, including soil pH. All sites had low levels of nitrogen (N), phosphorus (P), potassium (K), and boron (B). Mtanila-Chunya is located at latitude  $07^\circ 58' 59''$  South, longitude  $33^\circ 18' 59''$  East, at an altitude of 1439 meters above sea level (m.a.s.l), with an average annual rainfall of 750 mm. Tumbi-Tabora is at latitude  $05^\circ 03' 44.4''$  South, longitude  $32^\circ 40' 07.4''$  East, at an altitude of 1151 m a.s.l. with an average annual rainfall of 950 mm. Ushetu-Kahama is at latitude  $03^\circ 50' 15.2016''$  South, longitude  $32^\circ 35' 37.7808''$  East, at an altitude of 1156 m a.s.l. with an average annual rainfall of 990 mm. Mtanila and Tumbi sites have forest reserves and similar vegetative patterns, including long grass. These areas exhibit allelopathic effects on tobacco due to the proximity of the trial tobacco plots to the forest areas, whereas Ushetu boasts a well-developed land pattern with shrubs and seasonal rivers.

### 2.2. Experimental setup and treatments allocation

At all sites, the completely randomized block design (CRBD) was used with five treatments (Table 1) and three replications for flue-cured tobacco (K326) variety. Each treatment's plot size was 6 m in width and 6 m in length, making a total plot area of 36  $\text{m}^2$ . The plant spacing was 0.5 m, and ridge spacing was 1.20 m. The number of plants per ridge was 12, and the plant population per plot was 60. Tobacco seedlings in the control treatment (T1) were not fertilized. Tobacco seedlings in the standard  $\text{N}_{10}\text{P}_{18}\text{K}_{24}$  treatment (T2) were given 30 g  $\text{plant}^{-1}$  seven (7) days after transplanting (DAT), and then 8 g  $\text{plant}^{-1}$  of CAN 27% N on top of that at 21 DAT. Treatment T3 applied similar application

rates and times as the standard treatment, using  $N_{11}P_{22}K_{21}$  as the basal application and CAN 27% as the top-dressed fertilizer. The T4 applied  $N_{11}P_{22}K_{21}$  in two splits, the first at 14 DAT with 15 g plant<sup>-1</sup>, the second at 21 DAT with 15 g plant<sup>-1</sup> and CAN 27% N with 8 g plant<sup>-1</sup> at 35 DAT. The T5 also had an application of  $N_{11}P_{22}K_{21}$  in two splits: 10 g plant<sup>-1</sup> at 14 DAT and 12.5 g plant<sup>-1</sup> at 28 DAT without CAN 27% N.

Fertilizer  $N_{10}P_{18}K_{24}$  applied at a rate of 30 g per plant was equivalent to 50 kg N, 90 kg P, and 120 kg K ha<sup>-1</sup>, while fertilizer  $N_{11}P_{22}K_{21}$  applied at the rate of 30 g per plant was equivalent to 55 kg N, 110 kg P, and 105 kg K ha<sup>-1</sup>. The fertilizer  $N_{11}P_{22}K_{21}$  applied at the rate of 22.5 g per plant was equivalent to 37.5 kg N, 67.5 kg P, and 90 kg K ha<sup>-1</sup>. Fertilizer CAN 27% was applied at a rate of 8 g per plant, which was equivalent to 33.75 kg N and 5.4 kg Ca ha<sup>-1</sup>. Tobacco in field trials was monitored closely by weeding and earthing up the ridges. Plants were topped up 45 days after transplanting, and ripe plant leaves were harvested from the beginning of January to the end of April 2024.

**Table 1.** Treatments used for flue cured tobacco (K326)

T1 = Absolute control (no fertilizer)
T2 = 30g of $N_{10}P_{18}K_{24}$ at 7 DAT + 8g of CAN 27% at 21 DAT
T3 = 30g $N_{11}P_{22}K_{21}$ at 7 DAT + 8g of CAN 27% at 21 DAT
T4 = 15g $N_{11}P_{22}K_{21}$ at 14 DAT + 15g $N_{11}P_{22}K_{11}$ at 28 DAT + 8g of CAN 27% at 35 DAT
T5 = 10g $N_{11}P_{22}K_{21}$ at 14 DAT + 12.5 g $N_{11}P_{22}K_{11}$ at 28DAT

Key: DAT = Days After Transplanting

### 2.3. Plant leaf sampling for nutrient, nicotine, leaf area analysis and reaping tobacco leaf

Plant leaf sampling excluded the border rows and the first three plants at the edges of the innermost rows. Therefore, a mature middle leaf in each tobacco plant was sampled. The leaves were sampled from six plants in each treatment plot for each replication and at each site. These leaf samples were dried in the oven at 65°C at a constant weight, chopped, and sieved with a 0.5 mm wire mesh. The plant leaf analysis for the concentration of N, P, K, Mg and Ca was done using a dry ash and wet digestion method as given by Moberg (2000). For the plant leaf Zn and B, the diethylene triamine pentaacetic acid (DTPA) extraction method was deployed (Moberg, 2000). Nicotine was determined by a spectrophotometric method using a UV-visible single beam fixed at 602 nm (Figueiredo et al., 2009).

All plots were managed using agronomic good agricultural practices for field plot management, weeding, applying fertilizer, topping, and reaping tobacco leaves. The plant leaf length and width were determined using a measuring tape for the calculation of leaf area by multiplying leaf width and length using a correction factor of 0.6345 to get the leaf area (Equation 1). Ripened plant leaves were harvested weekly from each experimental plot and weighed using a digital scale. Harvested tobacco leaves were tied to a stick and loaded in a curing barn for about 7 days to dry. Dried leaves were offloaded from the curing barn, weighed, graded, and assigned a price through a registered Classifier from TTB. The grade index was calculated based on the weight of sold tobacco multiplied by the price allocation and divided by the overall tobacco weight. The trial was conducted in low soil fertility; not been planted tobacco for the past ten years in three agro-ecological sites, Mtanila (Chunya, Mbeya), Tumbi (Tabora) and Ushetu (Kahama).

$$A_s = \sum_{i=1}^n (L_i \times W_i \times 0.6345) \quad (1)$$

Where,  $A_s$  = flue cured tobacco leaf area per plant

$n$  = flue cured tobacco leaf number

$L_i$  = largest leaf length

$W_i$  = maximum leaf width

## 2.4. Statistical analysis

Statistical analysis of the two factors sites (Chunya; Tabora, Kahama) and fertilization were done using STATISTICA 8th Edition, StatSoft, Inc., Tulsa, OK, USA and analysis of variance (ANOVA). The significant means were compared using Fisher's least significance difference at  $p = 0.05$

## 3. Results and Discussion

### 3.1. Physical and Chemical Properties of the Experimental Soils

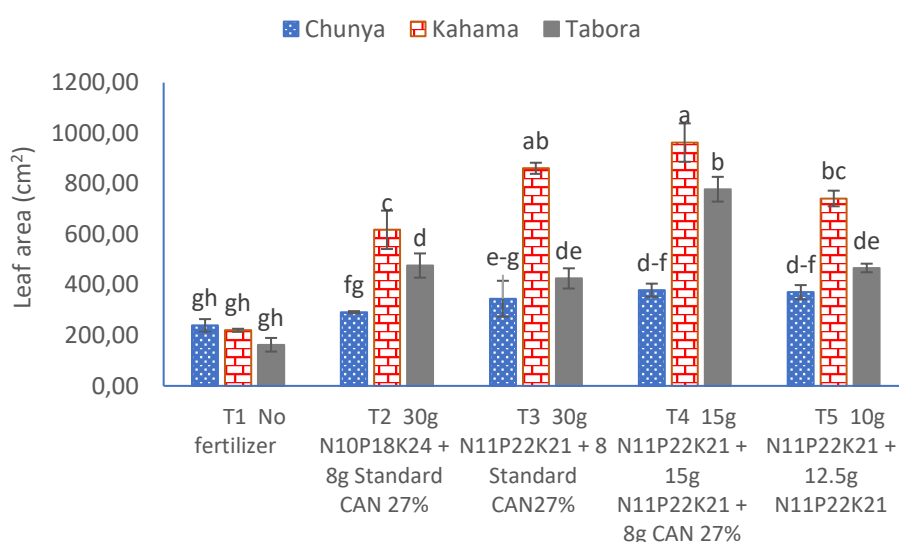
The physical and chemical properties of the Chunya (Mtanila), Tabora (Tumbi), and Kahama (Ushetu) sites (Table 2) were determined at TARI Serian, Arusha. The soil pH for Tabora soils (5.11), Chunya (5.09), and Kahama (5.17) were low moderately slightly acidic. Tabora and Chunya classified their soils as sandy, while Kahama exhibited a loamy sandy texture. Kahama had organic carbon (OC) of 2.91%, Chunya 2.40%, and Tabora 2.37%. Total N was ranked as very low across the sites, ranging from 0.13% for Chunya, 0.15% for Tabora, and 0.16% for Kahama. Available Sulphur (S), ranging from 48.37 (Chunya) to 71.92 mg kg<sup>-1</sup> (Tabora) and ranked as high. While across the sites, soil levels for P ranged from 3.23 (Tabora) to 8.03 mg kg<sup>-1</sup> (Chunya), had low levels of P, whereby levels of K ranged from 0.035 (Tabora) to 0.05 cmol (+) kg<sup>-1</sup> (Chunya), rated as very low. Calcium (Ca) levels across the sites were low, ranging from 0.98 (Kahama) to 3.83 cmol (+) kg<sup>-1</sup> in Tabora. Magnesium (Mg) across the site was very low (0.09-0.35 cmol (+) kg<sup>-1</sup>). Levels of zinc (Zn) were medium across the site, ranging from 0.70 to 0.74 mg kg<sup>-1</sup>, and boron (B) was very low across the sites with a narrow range from 0.01 to 0.02 mg kg<sup>-1</sup>. Therefore, the soil chemical properties of all three sites were slightly acidic with low levels of N, P, K, Ca, Mg, and B, making the trial suitable for soil fertility studies.

**Table 2.** Some Physical and Chemical Characteristics of Experimental Soils

Soil Constituents	Sites		
	Tabora	Chunya	Kahama
pH	5.11	5.09	5.17
Sand	88.39	85.71	84.99
Silt	4.59	4.80	2.92
Clay	6.95	9.25	11.45
Texture Class	Sandy	Sandy	Sandy loam
Organic carbon (OC) %	2.37	2.40	2.91
Nitrogen (N) %	0.15	0.13	0.16
Phosphorous (P) mg kg <sup>-1</sup>	3.23	8.03	4.79
Sulphur (S) mg kg <sup>-1</sup>	71.91	48.37	71.70
Potassium (K) cmol (+) kg <sup>-1</sup>	0.27	0.34	0.25
Calcium (Ca) cmol (+) kg <sup>-1</sup>	3.82	1.55	0.97
Magnesium (Mg) cmol (+) kg <sup>-1</sup>	0.34	0.16	0.08
Boron (B) mg kg <sup>-1</sup>	0.02	0.01	0.01
Zinc (Zn) mg kg <sup>-1</sup>	0.71	0.74	0.70

### 3.2. Effects of Fertilizer Treatments on Leaf Area, Dry Leaf Yield and Grade Index

The leaf area had a correlation with the dry leaf yield, of which the Kahama site, which had a significantly higher leaf area, yielded  $2052.59 \pm 310.12 \text{ kg ha}^{-1}$  and did not differ significantly from the Tabora site dry leaf yield ( $1813.42 \pm 251.57 \text{ kg ha}^{-1}$ ). The lowest significant ( $P \leq 0.001$ ) leaf area was recorded in the unfertilized tobacco plot (T1), which had  $207.33 \pm 15.94 \text{ cm}^2$ , followed by the standard treatment (T2), which had a leaf area of  $461.82 \pm 53.94 \text{ cm}^2$ . The treatment (T3), which had  $\text{N}_{11}\text{P}_{22}\text{K}_{21}$ , had a significantly higher leaf area ( $543.72 \pm 83.90 \text{ cm}^2$ ) than the standard  $\text{N}_{10}\text{P}_{18}\text{K}_{24}$  (T2) and did not differ significantly from the treatment T5, which had two splits of  $\text{N}_{11}\text{P}_{22}\text{K}_{21}$  without being top-dressed with CAN 27% N. The highest and most significant leaf area ( $706.56 \pm 90.45 \text{ cm}^2$ ) was obtained in T4, which had two splits of  $\text{N}_{11}\text{P}_{22}\text{K}_{21}$  and was top-dressed with CAN 27% N after 35 DAT (Figure 1). Splitting fertilizer showed a significant impact for increasing leaf area and yields due to the effective use of fertilizers following root growth with the ability to absorb more nutrients.



**Figure 1.** Influence of fertilizer application on tobacco leaf area

Table 3 displays the data on the effects of fertilizer treatments on green, dry tobacco leaf yields and grade index. The Kahama site had a significantly ( $P \leq 0.001$ ) higher leaf area ( $680.58 \pm 71.55 \text{ cm}^2$ ), followed by Tabora ( $461.80 \pm 54.30 \text{ cm}^2$ ), and Chunya had the lowest leaf area ( $325.07 \pm 20.19 \text{ cm}^2$ ). The standard  $\text{N}_{10}\text{P}_{18}\text{K}_{24}$  top-dressed with standard CAN 27% N fertilizer (T2; Picture 2) showed no significant difference in green leaf yields between T3 and T5 (Picture 1). However, the lowest significant green leaf yield was obtained in T1, recorded at  $2586.46 \pm 511.31 \text{ kg ha}^{-1}$ . The significantly higher green leaf yield recorded at T4 was  $21522.06 \pm 2614.80 \text{ kg ha}^{-1}$ . The results for the green leaf yields followed a similar trend to the dry leaf yield, of which the application of 15 g of  $\text{N}_{11}\text{P}_{22}\text{K}_{21}$  per plant at 14 DAT and a second split of 15 g  $\text{N}_{11}\text{P}_{22}\text{K}_{21}$  at 28 DAT, followed by top-dressing of 8 g of CAN 27% N at 35 DAT, had significant dry leaf yields of  $2784.30 \pm 251.56 \text{ kg ha}^{-1}$ . The inherently very low soil levels of N, P, K and the sandy loam soil texture for Kahama could have impacted the response of applied fertilizers for higher dry leaf yield (Table 3; Figure 2).



Picture 1: Tobacco plants for T 5 (Tabora) Picture 2: Tobacco plants for T2 (Tabora)

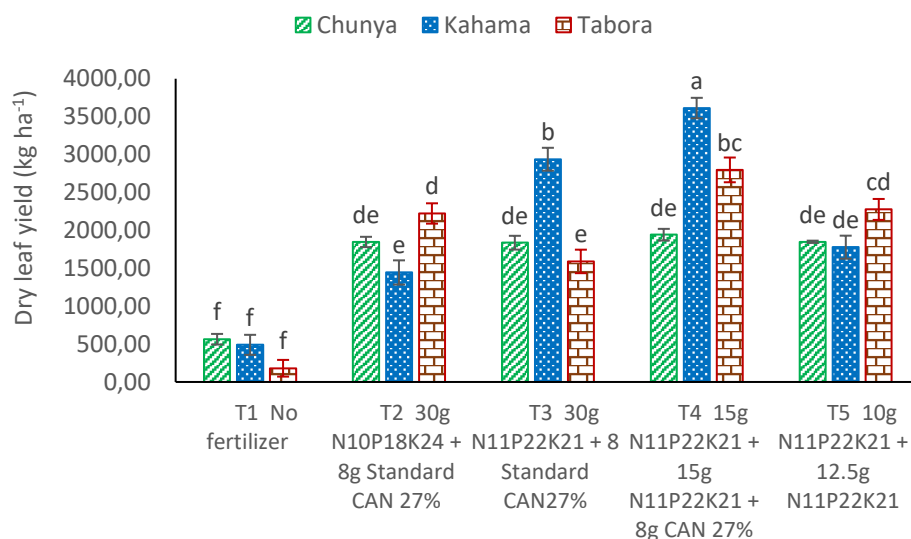
**Table 3.** Green Leaf, Dry Leaf Yields and Grade Index Produced Using different NPK Fertilizers

Sites	Green leaf yield (kg ha <sup>-1</sup> )	Dry leaf yield (kg ha <sup>-1</sup> )	Grade Index
Chunya	9220.44 ± 881.44 c	1607.84 ± 145.96 b	0.08 ± 0.01 c
Kahama	19684.63 ± 2401.65 a	2052.59 ± 310.12 a	0.29 ± 0.04 a
Tabora	16362.14 ± 2285.35 b	1813.42 ± 251.57 ab	0.16 ± 0.02 b
Treatments			
T1 No fertilizer	2586.46 ± 511.31 c	411.57 ± 83.75 c	0.03 ± 0.01 b
T2 30g N <sub>10</sub> P <sub>18</sub> K <sub>24</sub> + 8g CAN 27%	16321.22 ± 1710.75 b	1837.89 ± 129.10 b	0.22 ± 0.04 a
T3 30g N <sub>11</sub> P <sub>22</sub> K <sub>21</sub> + 8 CAN 27%	17811.98 ± 2322.85 b	2121.72 ± 266.69 b	0.23 ± 0.05 a
T4 15g N <sub>11</sub> P <sub>22</sub> K <sub>21</sub> + 15g N <sub>11</sub> P <sub>22</sub> K <sub>21</sub> + 8g CAN27%	21522.06 ± 2614.80 a	2784.30 ± 251.56 a	0.24 ± 0.04 a
T5 10g N <sub>11</sub> P <sub>22</sub> K <sub>21</sub> + 12.5g N <sub>11</sub> P <sub>22</sub> K <sub>21</sub>	17203.64 ± 2103.88 b	1967.60 ± 107.17 b	0.19 ± 0.03 a
2-WAY ANOVA F-statistics			
Site (S)	50.03***	6.67***	72.86***
Treatment (T)	55.42***	61.12***	27.52***
S x T	4.58***	8.28***	5.61***

Values presented are means ± SE (Standard Error); \*\*\* significant at  $P < 0.001$ ; ns= non-significant; Means in the same category of evaluated interface sharing similar letter(s) do not differ significantly based on their respective Least Significance Difference (LSD) value at 5% error rate

Furthermore, the Kahama soil exhibited a high level of organic carbon (T2), indicating a higher level of organic matter in the soil that effectively retains applied nutrients and facilitates their uptake by tobacco plants.





**Figure 2.** Influence of fertilizer application on tobacco dry leaf yields

### 3.3. Effects of fertilizer application on plant leaf nicotine

Table 4 presents the content of leaf nicotine and reducing sugars. Reducing sugars in tobacco leaves differed significantly across the sites, with Kahama having the lowest reducing sugars and the highest nicotine levels ( $2.42 \pm 0.16\%$ ), which did not differ from Tabora's ( $2.41 \pm 0.16\%$ ) (Figure. 3). The unfertilized tobacco leaves (T1) had significantly higher reducing sugar ( $26.85 \pm 0.18\%$ ) and the lowest nicotine ( $1.21 \pm 0.04\%$ ) in tobacco leaves (Table 4). The treatment (5), which received  $N_{11}P_{22}K_{21}$  (K from KCl) in two splits on 14 DAT and 28 DAT without CAN 27&N, had the least amount of nicotine in the leaves ( $2.02 \pm 0.11\%$ ), next to T1. The leaves of treatments (T2–T4) fertilized with  $N_{10}P_{18}K_{24}$ , which contained 25% K from KCl and 75%  $K_2SO_4$  (T2), and with  $N_{11}P_{22}K_{21}$ , which contained K from KCl alone (T3, T4) and top-dressed with CAN 27%N, showed significantly higher leaf levels of nicotine. This indicates that  $Cl^-$  could also be associated with improving nitrogen (N) use efficiency from applied CAN 27% N and hence increasing leaf nicotine levels. Our findings are similar to the results obtained by Peinado-Torrubia et al. (2023), who demonstrated that  $Cl^-$  improves nitrogen (N) use efficiency (NUE).

**Table 4.** Effect of NPK fertilizer with different sources of K on leaf nicotine

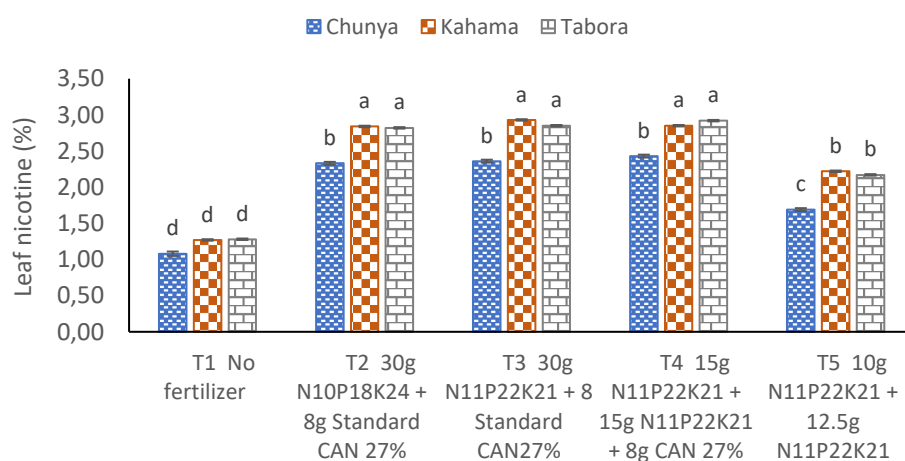
Sites	Reducing sugars (%)	Leaf nicotine (%)
Chunya	$24.54 \pm 0.43$ a	$1.98 \pm 0.16$ b
Kahama	$23.45 \pm 0.44$ c	$2.42 \pm 0.16$ a
Tabora	$24.38 \pm 0.35$ b	$2.41 \pm 0.16$ a
Treatments		
T1 No fertilizer	$26.85 \pm 0.18$ a	$1.21 \pm 0.04$ c
T2 30g $N_{10}P_{18}K_{24}$ + 8g CAN 27%	$23.06 \pm 0.18$ c	$2.66 \pm 0.11$ a
T3 30g $N_{11}P_{22}K_{21}$ + 8g CAN27%	$22.98 \pm 0.18$ c	$2.71 \pm 0.11$ a
T4 15g $N_{11}P_{22}K_{21}$ + 15g $N_{11}P_{22}K_{21}$ + 8g CAN 27%	$22.96 \pm 0.19$ c	$2.74 \pm 0.10$ a
T5 10g $N_{11}P_{22}K_{21}$ + 12.5g $N_{11}P_{22}K_{21}$	$24.77 \pm 0.17$ b	$2.02 \pm 0.11$ b
2-WAY ANOVA F-statistics		
Site (S)	299***	19.90***
Treatment (T)	1506***	82.82***
S x T	10***	0.45ns

Values presented are means  $\pm$  SE (Standard Error); \*\*\* significant at  $P < 0.001$ ; ns= non-significant; Means in the same category of evaluated interface sharing similar letter(s) do not differ significantly

based on their respective Least Significance Difference (LSD) value at 5% error rate

The results further indicate that applying  $N_{10}P_{18}K_{24}$  fertilizer with K sources derived from  $K_2SO_4$  (75%) and KCl (25%), is more likely to encourage the production of tobacco leaves with a higher nicotine content. The higher percentage of  $K_2SO_4$  also implies a higher S source, which enhances plant N uptake and encourages more nicotine production. Adding CAN 27%N as a top-dressing fertilizer increased N and favored the production of nicotine. On the other hand, our study showed that using  $N_{11}P_{22}K_{21}$  fertilizer, which gets its K from the muriate of potash (KCl), without adding CAN 27%N fertilizer, lowers the amount of nicotine in the leaves. This indicates that since N levels in soil were significantly lower, the  $Cl^-$  could not effectively improve N use efficiency, and hence reduced total N uptake, resulting in leaves with a significantly low nicotine content ( $2.02 \pm 0.11\%$ ). However, this did not result in a decrease in the yields of tobacco leaves (Table 3: Picture 1). Our study showed that the nicotine levels are much lower than in other flue-cured tobacco leaves, ranging from 4.9% to 3.3% on average (Tassew and Chandravanshi 2015; Tayoub et al., 2015; Henry et al., 2019).

Tiecher et al. (2023) found in their research studies that the application of Cl rates ranging from 0 to  $112 \text{ kg ha}^{-1}$  did indeed reduce the tobacco yield and quality. In our study we used a Cl rate of  $105 \text{ kg ha}^{-1}$ , which was lower than the rate used by Tiecher et al. (2023). Therefore, the current study recommends the application of  $N_{11}P_{22}K_{21}$  with its K sourced from KCl in two splits for higher yield results and to avoid the possibility of Cl levels being toxic to plants. This can be achieved by either applying with CAN 27% N or without CAN 27% N to reduce nicotine levels in leaves. Other studies by Tabaxi et al. (2020), using organic NPK fertilizer, showed that fertilization did not significantly affect the nicotine and sugar on Virginia tobacco. However, studies by de Marchi Soares et al. (2020), Bozhinova (2021), Ya'nan et al. (2021), and Wang et al. (2024) have linked the use of inorganic NPK fertilizer with K sourced from  $K_2SO_4$  to an increase in leaf nicotine.



**Figure 3.** Effect of NPK (source of K from KCL and  $K_2SO_4$ ) on leaf nicotine content

### 3.4. Effects of fertilizer application on plant nutrients

Table 5 presents the tobacco macronutrient leaf concentrations for N, P, and K. With the exception of leaf K, leaf N and P differed significantly ( $p < 0.001$ ) across the sites. Leaf K was significantly ( $p < 0.001$ ) higher in Chunya. The data in Table 5 shows that the differences between N, P, and K in the leaves of fertilized and unfertilized tobacco were significant. In fertilized tobacco plants, the leaf concentrations of N, P, and K increased significantly compared with the quantities obtained in unfertilized plants (Table 5). The application of NPK and CAN fertilizers, along with the high nutrient requirements of tobacco, is responsible for the significant increase in these nutrients in the leaves of fertilized plants. Table 2 reveals that the initial P and N concentrations in the soils across



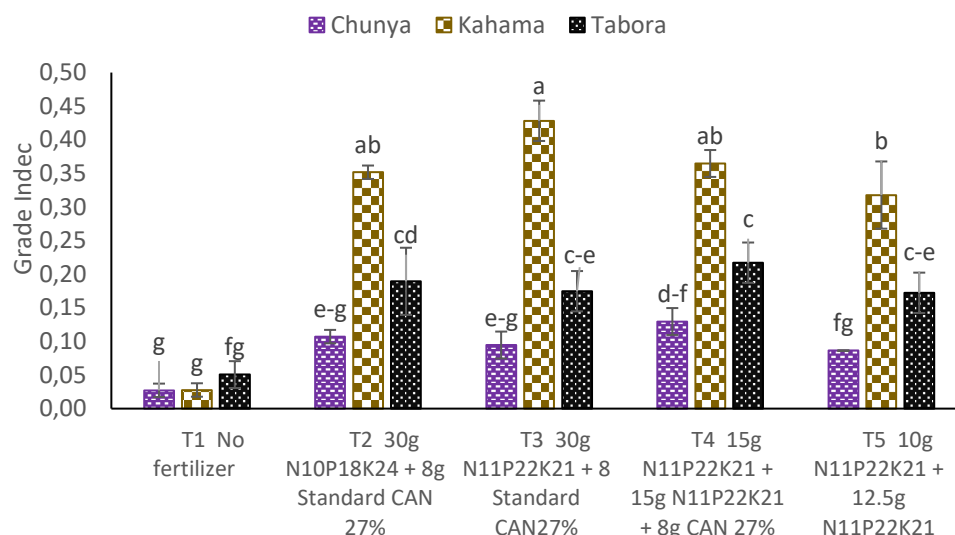
the sites were extremely low, likely contributing to the observed increase in leaf N and P concentrations under the  $N_{11}P_{22}K_{21}$  treatment. As a result of fertilization, the tobacco leaf concentrations for N, P, and K were above the critical levels of 1.5% N given by Haghighi et al. (2011), 0.1-1.0% P, and 1.6-4.1% K given by Bryson and Mills (2014). The source of K is crucial for tobacco quality and treatment. T3,  $N_{11}P_{22}K_{21}$  which sourced its K from KCl, exhibited a higher K content than the other treatments. Its leaf K content was  $2.39 \pm 0.20\%$ , which was much higher than the other treatments. This signifies that KCl has higher K levels compared to  $K_2SO_4$  (Jan and ul Hussan, 2022).

**Table 5.** Effect of Fertilizer Treatments on Tobacco Leaf N, P and K Concentrations

Sites	N (%)	P (%)	K (%)
Chunya	$1.50 \pm 0.06$ c	$0.21 \pm 0.01$ a	$2.77 \pm 0.08$ a
Kahama	$1.84 \pm 0.09$ a	$0.14 \pm 0.01$ c	$1.75 \pm 0.07$ b
Tabora	$1.56 \pm 0.09$ b	$0.17 \pm 0.01$ b	$1.69 \pm 0.14$ b
Treatments			
T1 No fertilizer	$1.25 \pm 0.08$ d	$0.16 \pm 0.01$ b	$2.00 \pm 0.13$ b
T2 30g $N_{10}P_{18}K_{24}$ + 8g Standard CAN 27%	$1.72 \pm 0.10$ b	$0.14 \pm 0.01$ c	$1.91 \pm 0.19$ b
T3 30g $N_{11}P_{22}K_{21}$ + 8 Standard CAN27%	$1.92 \pm 0.08$ a	$0.19 \pm 0.02$ a	$2.39 \pm 0.20$ a
T4 15g $N_{11}P_{22}K_{21}$ + 15g $N_{11}P_{22}K_{21}$ + 8g CAN 27%	$1.91 \pm 0.05$ a	$0.18 \pm 0.02$ a	$2.01 \pm 0.25$ b
T5 10g $N_{11}P_{22}K_{21}$ + 12.5g $N_{11}P_{22}K_{21}$	$1.38 \pm 0.05$ c	$0.19 \pm 0.02$ a	$2.04 \pm 0.26$ b
2-WAY ANOVA F-statistics			
Site (S)	83.35***	202.57***	52.05***
Treatment (T)	140.71***	68.39***	2.83*
S x T	22.79***	71.01***	2.93*

Values presented are means  $\pm$  SE (Standard Error); \*, \*\*\* significant at  $P < 0.05$  and  $P < 0.001$  respectively; ns= non-significant; Means in the same category of evaluated interface sharing similar letter(s) do not differ significantly based on their respective Least Significance Difference (LSD) value at 5% error rate

Since the concentrations of N, P, and K in the leaf are above the critical level, P vigorously promoted root development to enhance the uptake of more nutrients. This allowed N to influence the growth, quality, taste, and aroma of flue-cured tobacco, while K influenced the leaf yield and quality of the tobacco. As a result, the tobacco grade index significantly improved (Figure 4). Consequently, there was no significant difference in the grade index between the standard  $N_{10}P_{18}K_{24}$  and evaluated  $N_{11}P_{22}K_{21}$  across all treatments except for the unfertilized treatment (Table 3). Therefore, the two fertilizers have a similar impact on the quality of tobacco. However, the new  $N_{11}P_{22}K_{21}$  fertilizer product benefits from a subsidy, with a bag costing \$ 30.4. Without this subsidy, the cost per 50 kg is approximately \$ 55.6, compared to \$ 68.5 for the standard  $N_{10}P_{18}K_{24}$  fertilizer.



**Figure 4.** Effect of plant nutrients uptake on the quality of leaf grade index

The study's findings indicate that when using  $N_{11}P_{22}K_{21}$ , with a K source derived from KCl, in conjunction with CAN 27% N as a top-dressing fertilizer on tobacco plants, the CI tends to improve N efficiency. However, application of  $N_{11}P_{22}K_{21}$  without CAN 27% N automatically causes the soil pool's total N levels to drop, and the Cl<sup>-</sup> levels hinder total N uptake. Therefore, further research is required to observe the mechanism of Cl uptake by the plants in association with other nutrients, focusing on N nutrient uptake.

#### 4. Conclusions

The evaluated  $N_{11}P_{22}K_{21}$  fertilizer (T4) that was applied in two equal splits and then top-dressed with CAN 27% N produced a significant dry leaf yield ( $2784.30 \pm 251.56 \text{ kg ha}^{-1}$ ) compared to the standard  $N_{10}P_{18}K_{24}$  (T2) that was applied once and then top-dressed with CAN 27% N, which had a dry leaf yield of  $1837.89 \pm 129.10 \text{ kg ha}^{-1}$ . The application of  $N_{11}P_{22}K_{21}$  fertilizer (T5) in two splits, with a lower rate of 10 g per plant and a higher rate of 12.5 g per plant, and it did not contain CAN 27% N. It produced  $1967.60 \pm 107.17 \text{ kg ha}^{-1}$  of tobacco leaves, which did not differ significantly from the standard treatment (T2)  $N_{10}P_{18}K_{24}$  on yields ( $1837.89 \pm 129.10 \text{ kg ha}^{-1}$ ) when CAN 27%N was added, but the leaf nicotine content was lower ( $2.02 \pm 0.11\%$ ) than the standard treatment ( $2.66 \pm 0.11\%$ ). Therefore, application of  $N_{11}P_{22}K_{21}$  in two splits with a K source derived from KCl only and without CAN 27%N could lower the leaf nicotine content but not the tobacco leaf yields. The application  $N_{11}P_{22}K_{21}$  with a K source derived from KCl must be applied in two splits to bring good results with no toxicity of Cl levels. Thus, further research is necessary to understand how tobacco plants absorb Cl in relation to other nutrients, with a specific focus on N nutrition uptake.

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Please turn to the CRediT taxonomy for the term explanation. Authorship must be limited to those who have contributed substantially to the work presented.

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**Data availability statement:** We hereby declare the availability of our data and datasets in case needed.

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