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Research article

Polyaspartic acid increases potassium content and reduces the ratio of total sugar to nicotine in tobacco leaves

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A R T I C L E I N F O

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A B S T R A C T

Tobacco is an important cash crop in China, but the low potassium (K) content and high ratio of total sugar to nicotine in tobacco leaves have seriously affected the quality of tobacco leaves. As a fertilizer synergist, polyaspartic acid (PASP) can improve the K content in tobacco leaves, but it is unknown how it affects the K content in different parts of tobacco leaves, and how PASP affects the ratio of total sugar to nicotine in tobacco leaves has not been reported. Therefore, "Zhongyan 100" was selected for pot experiments with 5 different PASP addition levels: CK (0.0 %), P1 (0.1 %), P2 (0.2 %), P3 (0.4 %) and P4 (0.6 %), to reveal the effects of PASP on tobacco growth, K content, sugar content, nicotine content and the ratio of total sugar to nicotine in different tobacco parts, and determine the optimal PASP dosage for regulating the K content and the ratio of total sugar to nicotine in tobacco. The results showed that P1 (0.1 %) and P2 (0.2 %) only had slighter effects on tobacco growth and quality, while P3 (0.4 %) and P4 (0.6 %)treatments significantly promoted dry matter accumulation, increased K and nicotine content in leaves, decreased reducing sugar and total soluble sugar content in leaves, thereby reducing the ratio of total sugar to nicotine in tobacco leaves, especially in upper leaves. Considering the economic cost savings, 0.4% PASP was determined as the best application level to improve the growth and quality of tobacco. Thus, proper application of PASP is beneficial to improve tobacco leaf quality and reduce chemical K fertilizer application, thereby decreasing agricultural environmental risks of chemical fertilizer and alleviating the rapid depletion of potash in the world.

1. Introduction

The planting area and total output of tobacco in China rank first in the world [1]. As an important economic crop, tobacco has made great contributions to the development of the national economy. Tobacco is typical potassium (K)-loving crop, and K plays an important role in increasing leaf size, leaf yield, single leaf weight and improving leaf color [2,3]. At the same time, as an activator of more than 60 enzymes in plants, K affects the enzyme activity to participate in various physiological metabolic activities such as

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photosynthesis, carbohydrate transportation, etc., thereby regulates the accumulation of sugars, pigments, and aromatic substances that can directly affect the combustibility, aroma and other chemical qualities of flue-cured tobacco leaves [4,5]. The K level in tobacco leaves is one of the important indicators to evaluate the quality of tobacco. Usually, within a reasonable range of K content that would not induce K toxicity to tobacco, the higher the K content, the better the quality of tobacco leaves. The K content of high-quality flue-cured tobacco leaves should generally be higher than 2.5%. However, the average K content of Chinese tobacco leaves generally is 1%–2% and is significantly lower than the international level, which has become a major factor restricting the further improvement of tobacco leaf quality in China [6,7]. At present, there are some measures to improve tobacco leaf K content, such as K-efficient varieties breeding [8], scientific and rational fertilization [9], and chemical regulation with plant hormones or sprout inhibitors [10].

Reducing sugar, total sugar and nicotine content are also commonly indicators to evaluate the chemical composition of tobacco leaves. Sugar plays a key role in tobacco by participating in the synthesis of proteins, nucleic acids, lipids and other substances, to provide energy for the growth and development of tobacco and participate in the synthesis of aromatic substances. So, sugar directly affects the sensory quality of leaves, the quality of cut tobacco, the aroma and taste of smoking [3,4]. Suitable nicotine content is the goal pursued by the production of high-quality and low-harm tobacco leaves. The nicotine content of tobacco is generally in the range of 1.5–3.5%, preferably 2.5%. The ratio of total sugar to nicotine is an important indicator that can reflect the coordination between the taste and pungency of tobacco leaves. Too high ratio of total sugar to nicotine lowers the smoke strength and taste, while too low ratio of total sugar to nicotine will cause greater smoke irritation. In many tobacco-growing areas of China, low nicotine content and high sugar content in tobacco leaves resulted in high ratio of total sugar to nicotine and poor quality of tobacco leaves.

PASP, a "green" fertilizer synergist made of amino acid polymerization, has the advantages of strong biocompatibility, biodegradability, and environmental friendliness. PASP has been gradually applied to agricultural production to strengthen the absorption of nutrients by plants, promote crop growth and improve crop stresses resistance [11–13]. PASP or PASP coated fertilizer application improved the ability to absorb nutrients, as well as the growth rate, biomass, yield and quality of corn, rice, mustard and other plants [14–16]. It has been reported that there was a significant positive correlation between PASP and K uptake by plants, for example, exogenous application of PASP promoted the content of K in tomato roots and stems [11]; Adding PASP to corn under the condition of sufficient nutrients, the K content in seedling was increased by 19% [17]; It also has been preliminarily found that PASP increased the available K content in the soil and the accumulation of K in tobacco leaves [18]. However, it is not yet known how PASP affects K transport in tobacco and K content in different parts of leaves. What is more, K has an extremely important influence on the internal chemical composition of flue-cured tobacco [19]. Previously study had pointed out K can affect the content of reducing sugar and total sugar in different parts of tobacco leaves by regulating the activity of sucrose invertase and amylase transformation among tobacco leaf carbohydrates [20]. The K content of tobacco leaves also can affect the content of nicotine in tobacco leaves [12] by controlling the expression of putrescine N-methyl transferase (PMT) genes and affecting nicotine synthesis [21]. However, the suitable PASP application amount to improve K content in tobacco leaves is not sure and it is unknown whether the increased K content by PASP will affect the sugar and nicotine content and thereby decreasing the ratio of total sugar to nicotine.

Therefore, in this study, to screen the appropriate application level of PASP to increase the K content and reduce the ratio of total sugar to nicotine in tobacco leaves, the K content, sugar content, nicotine content and the ratio of total sugar to nicotine in different parts of tobacco were analyzed under different PASP application levels. This study will provide scientific theoretical guidance for improving the quality of tobacco leaves and reducing chemical K fertilizer application to alleviates agricultural environment risks of chemical fertilizer.

2. Materials and methods

2.1. Experimental design

The soil pot experiment was carried out in a greenhouse at Qingdao Agricultural University, Qingdao, China. "Zhongyan 100" was tested as the experimental material and the tobacco seedlings were taken from the tobacco research institute of the Chinese academy of agricultural sciences. "Zhongyan 100", also known as "CF965" series, was selected and cultivated by the Qingzhou Tobacco Research Institute of China National Tobacco Corporation using a pedigree method. The field growth period of "Zhongyan 100" is about 116 days and 20–22 leaves can be harvested from one plant. The leaf shape is long and elliptical, with a waist length of 65.6 cm and a width of 30.1 cm, and with obvious yellowing and high disease resistance. The general yield is around 2550 kg/hm^2 and the average content of reducing sugar, nicotine, total nitrogen, the ratio of total sugar to nicotine and the ratio of nitrogen to nicotine in leaves is 20.37%, 2.41%, 1.78%, 8.45 and 0.74, respectively. The main chemical components are coordinated, with good aroma quality, sufficient aroma content, and excellent taste. It is suitable for planting in tobacco areas with good fertilizer and water conditions such as Southwest, Central, Southeast, and Northeast China. Uniform seedlings with four full leaves were transplanted into pots with 2 kg of the base soil and the soil pH, organic matter content, available phosphorus content and available potassium content are respectively 7.60, 17.90 g/kg, 12.53 and 143.60 mg/kg. This experiment set up 5 different PASP treatments: CK (0.0%, 0.00g/pot PASP), P1 (0.1%, 2.00g/pot PASP), P2 (0.2%, 4.00g/pot PASP), P3 (0.4%, 8.00g/pot PASP) and P4 (0.6%, 12.00g/pot PASP) and each pot was randomly arranged with 6 replicates. PASP used in the experiment was purchased from Hebei Wozi Environmental Protection Technology Co., Ltd, and existed as light yellow powder with a pH of 8.5 and unit quality of 134. In order to ensure that PASP and other nutrients were evenly applied to the soil, different PASP amount of corresponding treatment was dissolved in 500 ml nutrient solutions, and slowly poured into the pots. The Hoagland and Arnon nutrient solution was used for providing macronutrients and micronutrients: KNO_3 (5.0 mM), KH_2PO_4 (1.0 mM), $MgSO_4$ (2.0 mM), EDTA-Fe (0.05 mM), $ZnSO_4$ (0.8 mM), $CuSO_4$ (0.3 mM), Na_2MoO_4 (0.1 mM). During the cultivation period, 300 ml of deionized water was poured every 3 days to ensure the normal growth of tobacco, and 500 ml of nutrient solution was poured every 30 days to provide the nutrient requirements. After 3 months of cultivation, the tobaccos of different treatments were harvested to determine relevant growth indices.

2.2. Plant sampling and biomass determination

After appearing significant differences in growth status of the tobaccos of different PASP treatments, the seedlings were harvested and divided into roots, lower leaves and upper leaves. After the fresh simples of different tobacco parts were weighed with a balance, a certain amount of them were taken for the determination of sugar content and the rest samples were washed and placed in an oven dried at 75 °C to constant weight. After weighing and grounding, the dry samples of different tobacco parts were stored to measure K content and nicotine content.

2.3. Determination of K content in tobacco

After the grounded root and leaf samples were pulverized through a 40-mesh sieve, an appropriate amount of samples were weighed for digestion: weigh 0.01g of sample powder into a triangular flask and add 2.0 mL of concentrated sulfuric acid and 10 drops of plant digestion accelerator in sequence. Then gently shake well and digest at low temperature on an electric furnace for 10 min finally, a certain amount of digestion solution was diluted and the total K content in different parts of tobacco was determined by a flame spectrophotometer (FP 640, Shanghai Precision & Scientific Instrument Co.).

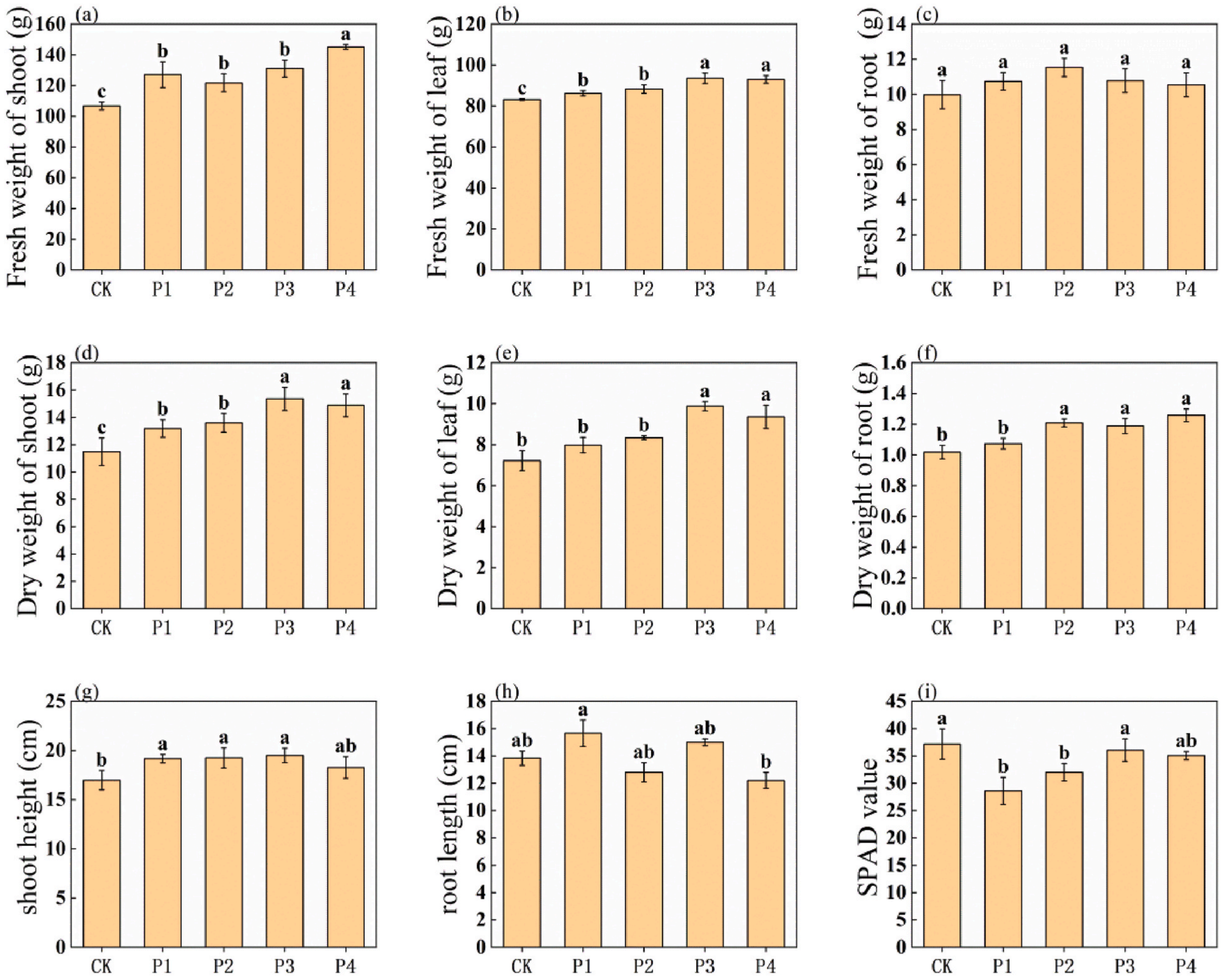

Fig. 1. Growth and dry matter accumulation of tobacco under different PASP dosage. (a–c) fresh weights of different part of tobacco; (d–f) dry weights of different part of tobacco; (g–i) shoot height, root length and SPAD value of tobacco. The data are represented as mean ± standard error, and different lower letters indicate significant differences between 5 treatments (P < 0.05).

2.4. Determination of nicotine content in tobacco

Firstly, adding methanol to an appropriate amount of sieved upper leaf and lower leaf samples and shaking in an air bath at 25 °C for 2h. After resting and layering, the supernatant was aspirated for chromatographic analysis with gas chromatography-mass spectrometry (GC-MS, 7890B/5977A, US, Agilent) [22].

2.5. Determination of total soluble sugar and reducing sugar content in tobacco

The total soluble sugar content was determined as the method described by [23]. The upper and lower leaves of fresh tobacco were extracted in a 65 °C water bath with ethanol, and then the supernatant was collected and concentrated. Finally, the total soluble sugar content was measured with a high performance liquid chromatography (HPLC, 1260, US, Agilent). The content of reducing sugar in tobacco leaves was determined using 3, 5 dinitro-salicylic acid method.

2.6. Statistical analysis

Data of different PASP treatments were subjected to one-way analysis of variance (ANOVA) using SPSS 25.0. Significant differences ($P < 0.05$) among the 5 treatments were determined by LSD test. Different lower case letters (a, b, c …) indicated the significant differences between the 5 treatments at the $P < 0.05$ level. Unless otherwise noted, results were presented as mean ± SE of 6 replicates.

3. Results

3.1. Effect of PASP on tobacco growth and dry matter accumulation

PASP promoted the growth and dry matter accumulation of tobacco leaves, while had no obvious effects on the fresh weight of roots and root length (Fig. 1a–h), indicating that the promotion of PASP on tobacco is mainly reflected in the growth and dry mass of leaves. What is more, comparing to CK, PASP application had a lower SPAD value of leaves (Fig. 1i). Our results also suggested an increasing of leaf growth and dry mass with the raising of PASP addition level, while P3 (0.4% PASP) and P4 (0.6% PASP) treatments had the most significantly effects for many indicators. Considering the economic input and benefits, 0.4% PASP was identified as the suitable PASP addition level for promoting tobacco growth and dry matter accumulation.

3.2. Effect of PASP on K content in different tobacco part

Compared with the CK treatment, the P1 (0.1% PASP) and P2 (0.2% PASP) treatments had no significant effect on K content in tobacco roots and leaves, while the P3 (0.4% PASP) and P4 (0.6% PASP) treatments obviously increased K content in roots, lower leaves and upper leaves (Fig. 2a–c). The results indicated that 0.4% and 0.6% PASP addition dosages increased K content in different parts of tobacco, and the effect of 0.4% PASP dosage on root K content was better than that of 0.6% PASP dosage (Fig. 2c).

3.3. Effect of PASP on the sugar content in tobacco leaves

Compared with the CK treatment, the P1 (0.1% PASP) treatment did not affect soluble sugar and total sugar contents in upper leaves, while the P2 (0.2% PASP), P3 (0.4% PASP) and P4 (0.6% PASP) treatments significantly decreased soluble sugar and total sugar contents in upper leaves (Fig. 3a). However, the decreasing effect on soluble sugar and total sugar contents in upper leaves did not rising with the increasing of PASP dosage. In addition, 0.1% and 0.2% PASP application had no effect on soluble sugar and total sugar contents but 0.4% and 0.6% PASP addition dosages decreased soluble sugar and total sugar content in lower leaves (Fig. 3b). The

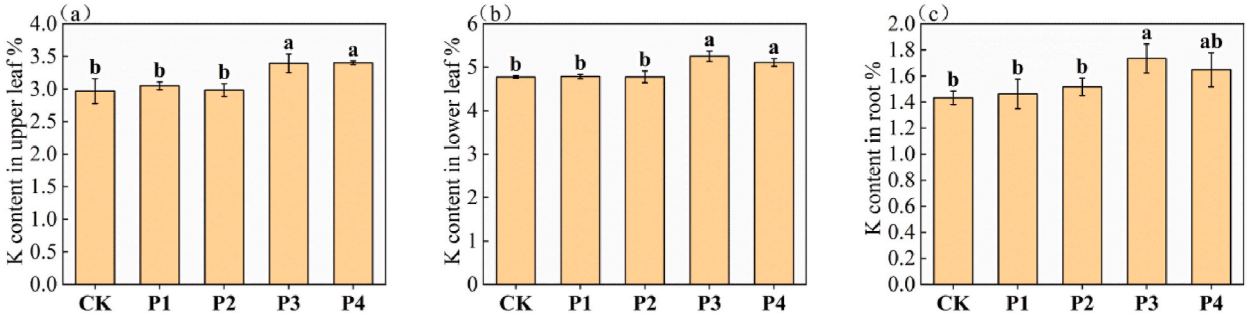

Fig. 2. Effects of different PASP dosages on K content in different parts of tobacco. (a) K content in upper leaves of tobacco; (b) K content in lower leaves of tobacco; (c) K content in roots of tobacco. The data are represented as mean ± standard error, and different lower letters indicate significant differences between 5 treatments ($P < 0.05$).

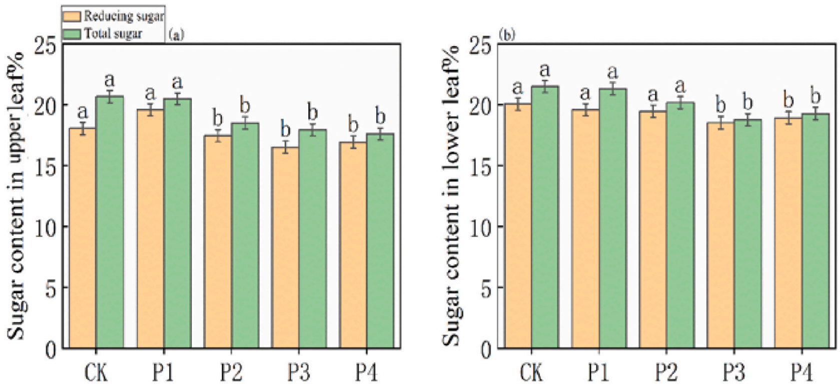

Fig. 3. Effects of different PASP dosages on sugar content in leaves of tobacco. (a) soluble sugar and total sugar contents in upper leaves; (b) soluble sugar and total sugar contents in lower leaves. The data are represented as mean ± standard error, and different lower letters indicate significant differences between 5 treatments (P < 0.05).

results showed that P3 (0.4% PASP) and P4 (0.6% PASP) treatments had the same effect on soluble sugar and total sugar contents both in upper and lower leaves and 4% PASP was identified as the most suitable PASP dosage for decreasing sugar content in tobacco leaves.

3.4. Effect of PASP on nicotine content in tobacco leaves

Comparing to the nicotine content (1.8%) in the upper leaves of the CK treatment, the nicotine content of P2 (0.2% PASP), P3 (0.4% PASP) and P4 (0.6% PASP) treatments were significantly increased by 22.7%, 46.8% and 49.1% (Fig. 4a). While PASP had no obviously effect on the nicotine content in lower leaves of tobacco (Fig. 4b).

3.5. Effect of PASP on the ratio of sugar-nicotine in tobacco leaves

The ratio of sugar-nicotine in lower leaves is higher than that in upper leaves, and PASP application significantly reduced the ratio of sugar-nicotine in tobacco leaves, especially in the upper leaves. P2 (0.2% PASP), P3 (0.4% PASP) and P4 (0.6% PASP) treatments decreased the ratio of sugar-nicotine in upper leaves from 13.8 to 10.0, 8.1 and 7.8 (a suitable range of sugar-nicotine ratio is 8–12) (Fig. 5a). In the meantime, P3 (0.4% PASP) and P4 (0.6% PASP) treatments decreased the ratio of sugar-nicotine in lower leaves from 15.6 to 12.5, and 12.9 (Fig. 5b). Although PASP did not reduce the sugar-nicotine ratio of lower leaves to an appropriate level, suitable PASP dosage decreased the ratio of sugar-nicotine in tobacco leaves, to a great extent. The results indicated that P3 (0.4% PASP) treatment was better to maintain a more suitable ratio of sugar-nicotine in tobacco leaves.

4. Discussions

The improvement of flue-cured tobacco quality and yield is related to the development of China's national economy. As a "green" fertilizer synergist, PASP had significant effects on crop growth, nutrient absorption, and yield and quality improvement [24]. PASP increased K content in tobacco [25], but it is not yet known how it affects other quality indicators such as sugar content, nicotine content, and sugar-nicotine ratio. Our study suggested that suitable PASP application dosage significantly increased the tobacco leaf biomass and improved leaf quality with increased K content and decreased sugar-nicotine ratio. What is more, 0.4% PASP was

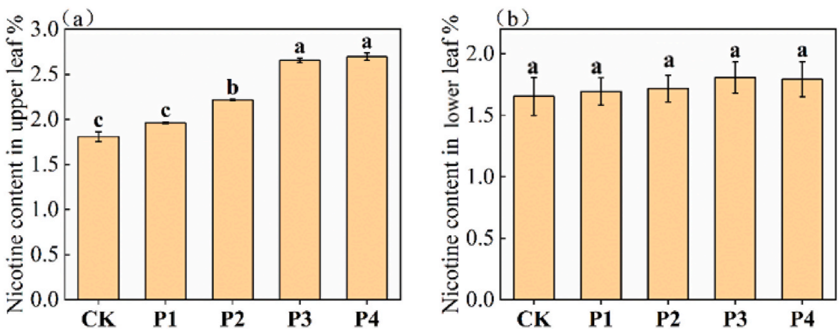

Fig. 4. Effects of different PASP dosages on nicotine content in leaves of tobacco. (a) nicotine content in upper leaves; (b) nicotine content in lower leaves. The data are represented as mean ± standard error, and different lower letters indicate significant differences between 5 treatments (P < 0.05).

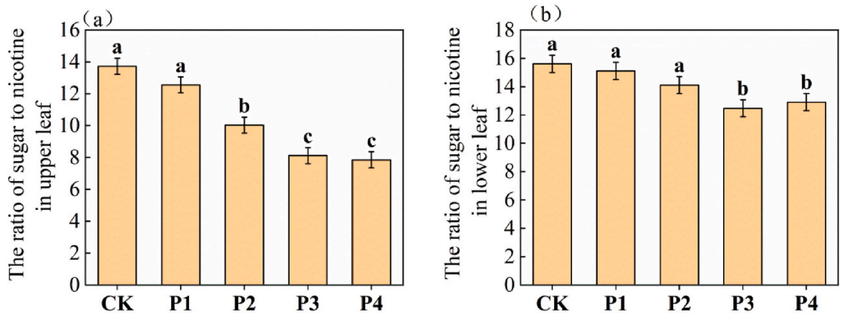

Fig. 5. Effects of different PASP dosages on the ratio of sugar to nicotine in tobacco leaves. (a) the ratio of sugar to nicotine in upper leaves; (b) the ratio of sugar to nicotine in lower leaves. The data are represented as mean ± standard error, and different lower letters indicate significant differences between 5 treatments ($P < 0.05$).

determined to be the optimal addition dosage for increasing K content and maintaining optimal sugar-nicotine content in tobacco leaves.

4.1. PASP promoted K absorption of tobacco

The application of PASP increased K content in tobacco, which was consistent with the researches on wheat seedling and tobacco [26]. Previous study indicated that PASP can increase K content by promoting root growth and K absorption by roots [18], while in our experiment, different PASP dosages had no obvious effect on root growth, and the improvement of PASP on the absorption of K by plants may be due to the K chelation and adsorption by free carboxyl and amide groups in PASP [11,27]. In this study, 0.4% PASP and 0.6% PASP addition dosages improved the K content in roots, upper leaves and lower leaves (Fig. 2), and considering the input cost and increase effect, 0.4% PASP was considered more suitable for combined supplying with fertilizers to tobacco.

4.2. PASP decreased the ratio of sugar to nicotine in tobacco leaves

Total sugar and reducing sugar are important indicators for evaluating tobacco smoke quality [28]. It has been pointed out that K was mainly involved in carbohydrates synthesis, transportation, and distribution in plants [29]. In present study, the increased K content induced by PASP supplying may promote the transportation of sugar from leaves to other parts, thereby decreasing sugar content in lower and upper leaves. It is generally believed that the optimal total sugar content for high-quality cigarettes is 18%–22%, and the reducing sugar content is 16%–18%. Without PASP application, there were higher total sugar and reducing sugar contents in tobacco leaves, while PASP treatments (P2, P3 and P4) significantly decreased total sugar and reducing sugar contents in upper leaves, and P3 (0.4% PASP) and P4 (0.6% PASP) treatments kept optimal reducing sugar content in lower leaves. K also affects nicotine content by affecting the biological chemical process of tobacco [21], but different studies have yielded opposite results. It has been indicated that K increased nicotine accumulation in tobacco [30], while other studies suggested that K decreased nicotine content by affecting putrescine formation and expression of genes related to nicotine synthesis [21,31]. In present study, PASP treatments, especially P3 (0.4% PASP) and P4 (0.6% PASP) improved nicotine content in upper leaves, and the effects on nicotine was consistent with the changes in K content. The synthesis site of nicotine is in the plant roots, and root activity affects the synthesis intensity of nicotine. It has been reported that there was no significant correlation between root activity and nicotine content in middle and lower tobacco leaves, but a significant positive correlation with nicotine content in upper tobacco leaves [32]. Thus, the increased nicotine content in upper leaves may be related to improved root activity and promoted nicotine biosynthesis induced by suitable PASP application [33].

The ratio of sugar to nicotine is also a quality indicator of flue-cured tobacco, and it depends on the total sugar content and nicotine content. Optimal ratio of sugar to nicotine (8–12) is conductive to improve the coordination between the taste and irritation of tobacco leaves [34]. P3 (0.4% PASP) and P4 (0.6% PASP) treatments obviously decreased the ratio of sugar to nicotine of upper leaves from 13.8 to 8.1 and 7.8, and decreased the ratio of sugar to nicotine of lower leaves from 15.6 to 12.5 and 12.9. Thus, P3 (0.4% PASP) treatment was conductive to maintain a more suitable ratio of sugar to nicotine especially in tobacco upper leaves by affecting sugar and nicotine contents.

5. Conclusions

This study investigated the effect of PASP application on tobacco K content, sugar content, nicotine content and the ratio of sugar and nicotine in upper and lower leaves of tobacco and the results indicated that 0.4% and 0.6% PASP dosage significantly improved the quality of tobacco leaves, especially in upper leaves. Considering the economic investment of PASP, 0.4% PASP is screened as the most suitable dosage of PASP for increasing K content and reducing ratio of sugar to nicotine in tobacco leaves.The results provided a theoretical basis for PASP to promote the growth and development of flue-cured tobacco, improve nutrient absorption rate and quality, reduce the use of K fertilizer, and protect the ecological environment.

CRediT authorship contribution statement

Zetao Zhang: Writing – original draft, Data curation. **Hui Tian:** Methodology, Data curation. **Jinsheng Li:** Writing – original draft, Data curation. **Dian Wang:** Writing – review & editing, Writing – original draft. **Xiuwen Wu:** Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

[1] Rebs, 2019-2025 China's Flue-Cured Cigarette Industry Market Panorama Assessment and Development Trend Research Forecast Report, Huajing Industry Research Institute Beijing, 2018.
[2] E.C. Schwamberger, J.L. Sims, Effects of soil-pH, nitrogen-source, phosphorus, and molybdenum on early growth and mineral-nutrition of burley tobacco, Commun. Soil Sci. Plant Anal. 22 (1991) 641–657.
[3] W. Hu, J. Wei, Q. Di, T. Tao, J. Zhang, et al., Flue-cured tobacco (*Nicotiana tabacum* L.) leaf quality can be improved by grafting with potassium-efficient rootstock, Field Crops Res. 274 (2021) 108305.
[4] K. Miyamoto, A. Takahashi, J. Ueda, Stimulation of hypocotyl elongation in etiolated lettuce seedlings by sodium chloride and potassium chloride at low concentrations: relevance to osmoregulation and cell wall mechanical properties, Plant Cell Physiol. 44 (2003) S37.
[5] X.Y. Dai, Y.R. Su, W.X. Wei, et al., Effects of top excision on the potassium accumulation and expression of potassium channel genes in tobacco, J. Exp. Bot. 60 (2009) 279–289.
[6] Q. Li, Study on Characteristic of Tobacco Quality and Effect of Main Ecological Factors on Tobacco Quality in Qujing, Hunan Agricultural University, 2011.
[7] G. Huo, T. Wen, et al., Research progress on the way to increase potassium content in flue-cured tobacco leaves, Hunan Agric. Sci. 413 (2) (2020) 112–114.
[8] T.Z. Yang, J.H. Fan, The roots charateristics of different genotype flue-cured tobacco varieties in absorption potassium, Acta Agriculturae Boreali-Occidentalis Sinica (3) (2006) 41–44.
[9] L.F. Lin, Effect of K accumulation and distribution of flue-cured tobacco under k fertilizer topdressing time retroposition, Journal of Southwest Agricultural University 29 (7) (2016) 1660–1665.
[10] W. Long, F.L. Chen, X.Y. Wang, et al., Effects of applying suckering agent and growth regulator on potassium and nicotine content in flue-cured tobacco leaves, Acta Tabacaria Sinica 16 (4) (2010) 51–57.
[11] M.M. Hu, Q.H. Dou, X.M. Cui, et al., Polyaspartic acid mediates the absorption and translocation of mineral elements in tomato seedlings under combined copper and cadmium stress, J. Integr. Agric. 18 (5) (2019) 1130–1137.
[12] X.L. Wang, S.X. Tu, J.h. Kang, X.h. Tang, W.j. Wei, M.X. Peng, G. Guan, Effect of phosphorus and potassium nutrition on nicotine and nutrient accumulation during topping stage of Nicotiana tabacum L, J. Food Agric. Environ. 10 (2012) 732–740.
[13] Y. Liu, L. Yuan, et al., Effects of polyaspartic acid with different molecular weights on root growth and nutrient uptake of wheat, Sci. Agric. Sin. 55 (13) (2022) 2526–2537.
[14] L.P. Koskan, A.R.Y. Meah, J.L. Sanders, R.J. Ross, Method and Composition for Enhanced Plant Productivity Comprising Fertilizer and Cross-Linked Poly Amino Acid, United States Patent. Patent, 1999 5861356.
[15] W.E. King, R.P. Fister, S.J. Norris, Slow-release Fertilizer and Method of Making and Using Same, United States Patent, 2009 20090298688.
[16] F. Deng, L. Wang, W.J. Ren, X.F. Mei, Enhancing nitrogen utilization and soil nitrogen balance in paddy fields by optimizing nitrogen management and using polyaspartic acid urea, Field Crops Res. 169 (2014) 30–38.
[17] W. Jiang, D.B. Zhou, H.S. Zhang, et al., The effect of polyaspartic acid on maize growth at seedling stage under different fertilizer applied condition, Journal of Maize Sciences 15 (5) (2007) 121–124.
[18] B.F. Cao, Y.G. Lu, L. Liu, et al., Effects of polyaspartic acid on physiological characteristics and fate of nitrogen fertilizer in flue-cured tobacco with nitrogen fertilizer reduction, J. Soil Water Conserv. 33 (5) (2019) 223–229.
[19] X.L. Pu, J.H. An, G.X. Nan, Z.X. Qu, et al., Research progress and direction of potassium(K) content in tobacco leaf, Jiangsu Agric. Sci. (5) (2009) 115–117.
[20] L. Wang, L. Liu, W.Y. Yang, W.H. Liu, et al., Effects of plant population and k application level on the quality of leaves of flue-cured tobacco, Journal of North China Agriculture S1 (2007) 106–110.
[21] J.Y. Li, Y. Jin, J. Wang, G.X. Su, Study on key genes in potassium nutrition regulating nicotine biosynthesis, Jiangsu Agric. Sci. 47 (16) (2019) 106–109.
[22] L. Tang, H.W. Yang, L. He, et al., Direct analysis of free-base nicotine in tobacco leaf by headspace solid phase micro extraction combined with gas chromatography/mass spectrometry, Accred Qual. Assur. 24 (5) (2019) 341–349.
[23] X. Li, J. Li, Determination of the content of soluble sugar in sweet corn with optimized anthrone colorimetric method, Storage Process 13 (04) (2013) 24–27.
[24] M. Dai, H.B. Guo, Y.D. Wei, Effects of polyaspartic urea application methods on nitrogen balance and rice yield in cold region of China, Rice Farming in China 28 (4) (2022) 74–78.
[25] B.F. Cao, Y. Gui, Q.X. Zu, et al., Effects of polyaspartic acid on growth, yield and nutrient absorption of flue-cured tobacco with reduced fertilization, Chin. Tobac. Sci. 39 (5) (2018) 57–63.
[26] W.Y. Li, X.H. Du, et al., Effects of potassium fertilizer reduction combined with supplement of polyaspartic acid on soil physicochemical properties, potassium absorption and utilization of flue-cured tobacco, Tob. Sci. 56 (2) (2023) 1–10.
[27] Y.W. Shen, H.T. Lin, W.S. Gao, et al., The effects of humic acid urea and polyaspartic acid urea on reducing nitrogen loss compared with urea, J. Sci. Food Agric. 100 (12) (2020) 4425–4432.
[28] X.H. Deng, S.Y. Zhao, et al., Total sugar content of cutter leaf in flue-cured tobacco from Hunan province and its correlation with smoke quality, Acta Tabacaria Sinica 15 (5) (2009) 43–47.
[29] C.M. Ramage, Mineral uptake in tobacco leaf discs during different developmental stages of shoot organogenesis, Plant Cell Rep. 21 (11) (2003) 1047–1053.
[30] G.S. Hu, Z.B. Yang, et al., Characteristics of nicotine accumulation and effect of some nutrients on nicotine content of flue cured tobacco, Henan Agricultural Sciences (1) (1999) 10–14.
[31] F.J. Richards, R.G. Coleman, Occurrence of putrescine in potassium deficient barley, Nature 170 (4324) (1952) 460.
[32] F. Ding, Regulating Effect of Nicotine Content of Flue-Cured Tobacco with Exogenous Regulators, Nanjing Agricultural University, 2006.
[33] X. Xu, X. Zhang, C. Liu, H. Qin, F. Sun, J. Liu, Appropriate increasing potassium supply alleviates the inhibition of high nitrogen on root growth by regulating antioxidant system, hormone balance, carbon assimilation and transportation in apple, Sci. Hortic. 311 (2023) 111828.
[34] J.N. Gao, D. Zhang, et al., Effects of combined application of nitrogen, phosphorus and potassium on biomass and quality components content of tobacco leaves, Southwest China J. Agric. Sci. 34 (6) (2021) 1269–1276.