Inheritance of Main Morphological Traits and Chemical Parameters' Analysis of Oriental Tobacco

Vesselina Masheva and Maria Kasheva

Breeding and Seed Production Department, Tobacco and Tobacco Products Institute, Plovdiv, Markovo 4108, Bulgaria

Abstract: This study was conducted to evaluate four different parental oriental varieties with their F_1 crosses to estimate inheritance of some morphological traits, such as plant height, number of leaves, total dry weight and chemical compounds, especially nicotine and sugars during sun-curing period. The oriental varieties Kozarsko 541, Plovdiv 50 and Krumovgrad 17 from Bulgaria and Basma Xanthi 101 from Greece and their respective crosses with Basma Xanthi 101 were studied. The study was set up in 2014 and 2015 in experimental field of the Tobacco and Tobacco Products Institute, Plovdiv, using randomized block design with four replications. Common agricultural practices were applied during the growing season. The result indicated that in hybrid combination with parental varieties Kozarsko 541 and Plovdiv 50, except total dry weight, additive gene effects are important for traits height of plant and number of leaves. Height of plant and number of leaves are inherited partially dominant to parent with higher value and total dry weight inheritance was over dominant. In hybrid combination Krumovgrad 17 $\times$ Basma Xanthi 101, inheritance of the plant height and leaves number were over dominant type. Total dry weight is inherited partially dominant. In a word, prevailing mode for inheritance of plant height and total dry weight was the over dominance and for number of leaves it was partial dominance. In sun-curing with yellowing stage, nicotine decreased and sugars increased in all tested varieties with a highest percentage in variety Kozarsko 541 (nicotine) and Krumovgrad 17 (sugars).

Key words: Oriental tobacco, inheritance character, morphological traits, chemical parameters.

1. Introduction

Tobacco plants and received tobacco raw material possess special characteristics, because, on the one hand, they are the result of phenotypic expression of a number of quantitative traits and response of the variety and environmental conditions, and on the other hand their use as a flavor product with specific consumer's qualities formed in the stages of curing and fermenting [1, 2].

Major signs determining the productivity of oriental tobacco are a number of leaves, plant height, size and absolute dry weight of the leaves, length of flowering period, etc.. Unlike other crops, tobacco varieties are exclusively related to the growing conditions, where a large part of chemical compounds and specific consumer qualities of the raw material are formed [3,

4]. In most studies [5-7], it was observed over dominant in Virginia and dominant inheritance of plant height in Burley tobacco. In Burley tobacco, it was found that the prevailing importance for the sign is the dominant gene effects, but in Virginia tobacco for height of the plants, the main type of inheritance is additive [8, 9]. In crosses of Bidi and Havana tobacco, the plant height is observed both additive and not additive (dominant and epistatic) gene effects [10, 11]. According to many studies in oriental tobacco, prevailing mode for inheritance of number of leaves is dominance and partial dominance [12, 13]. However, Radoukova et al. [14] and Butorac et al. [15] reported that in the mode of inheritance of Virginia and Burley tobacco, the most important are dominant and additive gene effects. The variability of quantitative traits of oriental tobacco in different environmental conditions is depending on the individual characteristics of genotype, and needs a specific study of inheritance of

Corresponding author: Vesselina Masheva, Ph.D., research fields: selection of new tobacco varieties, breeding and seed production.

morphological characters [16, 17]. The aim of this paper was to study the inheritance of plant height, number of leaves and dry weight, therefore to improve the knowledge of the mode of inheritance and gene type, and change of chemical parameters especially nicotine and sugars during sun-curing period.

2. Materials and Methods

Four oriental tobacco varieties Kozarsko 541, Plovdiv 50, Krumovgrad 17 from Bulgaria and Basma Xanthi 101 from Greece were tested. The genetic populations P_1 , P_2 and F_1 were grown for each of the crosses (Kozarsko 541 $\times$ Basma Xanthi 101, Plovdiv 50 $\times$ Basma Xanthi 101 and Krumovgrad 17 $\times$ Basma Xanthi 101) in a randomized block design with four replications under each cross at the experimental field of Tobacco and Tobacco Products Institute, Plovdiv during 2014-2015. All the cultural operations were done in accordance with the usual-recommended practices for oriental tobacco cultivation. The obtained results are from 20 plants selected at random per plot from the P_1 , P_2 and F_1 generation. Plant height (cm) was measured at maturity stage from base to first branch of the inflorescence. Numbers of leaves per plant were counted as a total number of leaves. Total dry weight was expressed in mg/100 cm².

The results of the height of plants, the number of leaves and total dry weight are expressed in the arithmetic mean ($\bar{X}$) $\pm$ the average error of the arithmetic mean ($S\bar{X}$), and the degree of dominance (h/d) was computed according to the formula of Mather [18, 19], as Eq. (1):

$$h/d = (F_1 - MP) / [\frac{1}{2}(P_1 - P_2)] \quad (1)$$

where, h is the difference between F_1 means and parent means (MP), and d is the half difference of two parents participators in a combination.

In sun-curing period, the samples were divided into two groups. The first group was cured in a conventional method (control group). The tobacco leaves are manually removed from the plants whilst the plants are in the field, so as to leave the stalks

behind, and the leaves are then threaded onto a string to provide approximately 200-500 leaves/m of string material. The string (control group) was then hung allowing the tobacco leaves to be sun-cured for 15 d to 30 d. Another group of strings was placed in a controlled environmental chamber at 28 °C for 48 h (for yellowing stage) and after this period was cured.

The chemical compounds nicotine was determined according to ISO 5152-2003 and reducing sugars was determined according to ISO 5154-2003 in Tobacco and Tobacco Products Institute (TTPI) laboratory. Percentage change of nicotine and sugars was calculated according to Eq. (2):

$$\% \text{ up to control} = \frac{b - a}{a} \times 100 \quad (2)$$

where, b is variant with yellowing stage and a is control variant.

3. Results and Discussion

3.1 Inheritance of Plant Height

The highest stem among parental genotypes was observed in Krumovgrad 17 (133.20 cm), followed by Plovdiv 50 (127.30 cm), and the lowest height was on variety Basma Xanthi 101 (92.37 cm). The highest stem among hybrids are those where one of the parents are Krumovgrad 17 and Plovdiv 50. From the analysis of data in Table 1, it indicated that the nature of inheritance for plant height character was over dominance in hybrid Krumovgrad 17 $\times$ Basma Xanthi 101 ($h/d = 1.18$) and partial dominance in the other crosses ($h/d = 0.84$ and 0.51 , respectively) with positive direction towards the parent with higher stem. Genetic analysis in Fig. 1 showed that the additive component (d) in F_1 progenies is higher than the dominant one (h) in years of research (Kozarsko 541 $\times$ Basma Xanthi 101 $d = 11.62 > h = 9.76$; Plovdiv 50 $\times$ Basma Xanthi 101 $d = 17.47 > h = 8.91$). The trait expression is determined by additive gene effects. In cross Krumovgrad 17 $\times$ Basma Xanthi 101, $d = 20.42 < h = 24.09$ inheritance was over dominance, which indicates a stronger effect of dominant genes in

inheritance. Similar results were reported in studies of tobacco ecotypes Ustina, Dupnitsa and Djebel Basma [20, 21]. The values for environment effects ($e = -1.25, -6.82$ and -9.82 , respectively) indicate that the environment has impact on this property.

3.2 Inheritance of Number of Leaves

The largest number of leaves in the parental varieties was established in Kozarsko 541 (34.35) and the lowest in Basma Xanthi 101 (26.58). The analysis of data shown in Table 1 indicated that the nature of inheritance for number of leaves character was partial dominance with positive direction towards parent Kozarsko 541 with more number of leaves. Additive component (d) was significant for number of leaves as presented in Fig. 2. The values for d are significantly

higher than those for h , with parent Kozarsko 541 ($d = 3.89 > h = 2.26$), Plovdiv 50 ($d = 1.99 > h = 0.98$), which indicates that a major part of the genetic variance in the inheritance of number of leaves belongs to the additive component. The interaction in the investigated generations has a positive value, indicating dominance of the genes from parents with more number of leaves.

The cross with parental variety Krumovgrad 17 was observed over dominance in inheritance of this traits ($d = 2.4 < h = 3.57$). The low values for environment effects ($e = -0.23$ in cross 1, $e = -1.01$ in cross 2 and $e = -1.23$ in cross 3) indicate that the environment has negative impact on this characteristic, especially in F_1 Plovdiv 50 $\times$ Basma Xanthi 101 and Krumovgrad 17 $\times$ Basma Xanthi 101.

Fig. 1 Genetic parameters of plant height.

1: Kozarsko 541 $\times$ Basma Xanthi 101; 2: Plovdiv 50 $\times$ Basma Xanthi 101; 3: Krumovgrad 17 $\times$ Basma Xanthi 101.

Fig. 2 Genetic parameters of number of leaves.

1: Kozarsko 541 $\times$ Basma Xanthi 101; 2: Plovdiv 50 $\times$ Basma Xanthi 101; 3: Krumovgrad 17 $\times$ Basma Xanthi 101.

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Fig. 3 Genetic parameters of total dry weight.

1: Kozarsko 541 × Basma Xanthi 101; 2: Plovdiv 50 × Basma Xanthi 101; 3: Krumovgrad 17 × Basma Xanthi 101.

3.3 Inheritance of Total Dry Weight

Total dry weight is an indicator of the materiality of tobacco leaves and is in direct contact with both yield and qualitative indicators of raw materials [22].

The highest total dry weight among parental genotypes was observed in Basma Xanthi 101 (394 mg/100 cm²), followed by Kozarsko 541 (390 mg/100 cm²) and the lowest in variety Krumovgrad 17 (372 mg/100 cm²).

Results in this investigation revealed the prevailing of over dominance in inheritance of this trait (Table 1) The analysis of data showed in Fig. 3 indicated that the dominant component (h) in F₁ is higher than the additive one (d) in research crosses Kozarsko 541 × Basma Xanthi 101 ($h = 6 > d = 2$) and Plovdiv 50 × Basma Xanthi 101 ($h = 23.5 > d = 7.5$), which indicates a stronger effect of dominant genes in the inheritance of total dry weight. In cross Krumovgrad 17 × Basma 101, inheritance was partial dominance, the value of dominant parameter $h = 4$ is lower than the additive one $d = 11$, which indicates a stronger effect of recessive genes in the inheritance of total dry weight. The degree of dominance (h/d) in F₁ generation has a positive value, indicating the dominance of genes from higher total dry weight parents.

3.4 Change of Nicotine and Sugars in Sun-Curing Period

For the essential for tobacco products, except

typical morphological characteristics of each oriental tobacco ecotype, are also important their chemical components forming both in the growing period and the next stages of raw tobacco passed through curing and fermentation.

Tobacco is a complex plant material in a chemical composition standpoint. Yet even today, the quality of tobacco is judged largely by empirical experience and subjective sensorial evaluation. It is not possible to replicate the smoking properties of the tobacco leaf synthetically [23].

Nicotine is the chemical component which is the most typical alkaloid in the tobacco plant. It determines physiological strength of smoking that occurs only when content of nicotine is in certain range. During the sun-curing, nicotine levels decreased by an average of 10%-30% as a result of oxidative decomposition. This degradation occurs mainly in autolysis in the second phase of drying [24].

The nicotine is a varietal characteristic, but it varies considerably from environmental conditions and the curing. The data in Table 2 show that in all tested varieties after yellowing stage, a significant reduction in nicotine was observed, with the highest percentage of 22.77% in variety Kozarsko 541. According to Refs. [23, 25, 26], the accumulated evidence in tobacco curing suggested that nicotine and total alkaloid contents decreased with the curing process.

Table 1 Parameters of parental varieties and F₁ hybrids.

Crosses	P1 ($\bar{X} \pm S \bar{X}$)	P2	F1	h/d
Plant height (cm)				
Kozarsko 541 × Basma Xanthi 101	115.60 ± 2.74	92.37 ± 2.25	113.75 ± 1.96	0.84
Plovdiv 50 × Basma Xanthi 101	127.30 ± 3.71	92.37 ± 2.15	118.75 ± 2.43	0.51
Krumovgrad 17 × Basma Xanthi 101	133.20 ± 1.93	92.37 ± 2.15	136.88 ± 1.58	1.18
Number of leaves				
Kozarsko 541 × Basma Xanthi 101	34.35 ± 0.92	26.58 ± 0.53	32.73 ± 0.68	0.58
Plovdiv 50 × Basma Xanthi 101	30.56 ± 0.64	26.58 ± 0.53	29.55 ± 0.92	0.49
Krumovgrad 17 × Basma Xanthi 101	31.38 ± 0.79	26.58 ± 0.53	32.55 ± 0.72	1.49
Total dry weight (mg/100 cm ²)				
Kozarsko 541 × Basma Xanthi 101	390.00	394.00	398.00	3.00
Plovdiv 50 × Basma Xanthi 101	379.00	394.00	419.00	3.13
Krumovgrad 17 × Basma Xanthi 101	372.00	394.00	379.00	0.36

Table 2 Chance of chemical components during sun-curing process.

Variety	Sun-curing without yellowing stage (control)	Sun-curing with yellowing stage	% to control
Nicotine (%)			
Kozarsko 541	1.01	0.78	-22.77
Plovdiv 50	1.17	1.08	-7.70
Krumovgrad 17	1.62	1.38	-14.82
Reducing sugars (%)			
Kozarsko 541	17.60	18.90	7.39
Plovdiv 50	14.90	16.80	12.75
Krumovgrad 17	12.45	14.36	15.34

From the data presented in Table 2, it can be seen that in all the varieties, the amounts of reducing sugars in the curing after yellowing stage were relatively higher to the control, with the increase being at a high in Krumovgrad 17 to 15.34%. The results were similar to those reported in Refs. [27-29].

4. Conclusions

In this study, both additive and non-additive gene action play a significant role in the inheritance of all characters. Prevailing mode for inheritance of plant height and total dry weight was the over dominance, and for number of leaves, it was partial dominance. In sun-curing with yellowing stage, nicotine decreased and sugars increase in all tested varieties with a highest percentage in variety Kozarsko 541 (nicotine) and Krumovgrad 17 (sugars).

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