

Study of the Morphological and Agronomic Traits in Some Autochthonous and Commercial Tobacco Varieties in the Republic of Macedonia

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Abstract: Investigations were made on some morphological (height of the stalk with inflorescence, leaf number, length of the middle belt leaves) and agronomic trait dry mass yield in five autochthonous tobacco varieties of Prilep (P 10-3/2 and P 12-2/1), Djebel (Dj № 1) and Yaka tobaccos (YK 7-4/2 and KY) and five commercial oriental varieties of Prilep tobacco (P-23, P-84, NS-72, P-66-9/7 and P-79-94). The trial was set up in the Experimental field of Tobacco Institute – Prilep in 2012, 2013 and 2014 in randomized block design with three replications, using traditional cultural practices. The aim of investigations was to study the autochthonous and commercial varieties for the above quantitative traits, to evaluate their variability and to determine the importance of differences between varieties and years by the use of biometric analysis, in order to get a better knowledge of varietal stability, genetic homozygosity and the progress made in the selection of oriental tobacco in the Republic of Macedonia. Differences between the genotypes in the investigation period were highly significant, which is a sign of their mutual genotypic and phenotypic diversity. Standard error of the mean for all data is low, indicating stability and homozygosity of the genotypes. The highest values for plant height were achieved in KY, and the lowest in P 10-3/2 and P 12-2/1. The highest leaf number was recorded in variety P-66-9/7, i.e. 33 leaves more than YK 7-4/2 and Dj № 1. The highest value for leaf length was recorded in P-79-94 and it is 4.8 cm more than the lowest leaf length observed in YK 7-4/2. The highest yield was achieved in P-66-9/7, with 30,5% more than the lowest-yielding variety Dj № 1. The presented data point out to the successful breeding activity of the Tobacco Institute in the selection of oriental aromatic tobaccos. Through evaluation of the varietal stability, the breeder broadens his knowledge of their homozygosity and becomes more secure in the choice of parental pairs for implementing the selection programs.

Keywords: tobacco (*Nicotianatabacum*L.), autochthonous varieties, commercial varieties, quantitative traits, analysis of variance, variability.

I. Introduction

Tobacco Institute – Prilep dates from 1924 and its main activity is the selection of oriental, small-leaf, aromatic tobaccos. All breeding activities are directed to increasing its yield and quality.

Korubin – Aleksoska A. (2004) investigated some morphological and agronomic traits in Yaka tobacco (YK-23, YK-48, YK-87, YK-7-4/2, YV 125/3 and YK-68) and reported that YK-87 is ranked first in length, width and area of the middle belt leaves, green mass yield/stalk and green mass yield/decare; the highest plants were recorded in YK-68 and the highest leaf number in YK-48. The same author (2005) investigated some quantitative traits in Djebel tobacco (Dj-291, Dj-1, Pobeda 2, Dj-38, Orient 138 and line L-3) and reported that the best results for length, width and area of the middle belt leaves were found in Dj-291, which also had the highest green mass yield. The old standard high-quality aromatic variety Dj -1 was the smallest in size and had the lowest yield of green mass. The present standard Dj -38 has significantly higher yield than Dj -1, but compared to the other genotypes investigated, the difference in yield is not significant. Dimitrieski and Miceska (2006) investigated the qualitative and quantitative traits of some varieties and lines of oriental tobacco Prilep (P-12-2/1, P-7, P-84, P-23, P-76, NS-72, PV 156/1, PV 121/2, P-65/94, P.L.146-7/1 and P.L. 146-3/2) and concluded that all varieties, except for P-7, had higher yield than the standard P-12-2/1, but the lowest ecological variability was observed in the line P.L. 146-7/1. Mitreski (2011), in his two-year investigations of six varieties of Prilep tobacco, reported that their green and dry mass ratio ranged from 6.11:1 (Prilep 66-9/7) to 6.95:1 (P 12-2/1), which means that the highest productivity among varieties was observed in P 66-9/7 and the lowest in Prilep P 12-2/1. Dimitrieski and Miceska (2011), based on the results of comparative trial with some lines and varieties of Prilep tobacco, pointed out that P 66-9/7 should be especially emphasized for its productivity. Mitreski (2012), in his two-year investigations on height of the stalk with inflorescence in six Prilep tobacco varieties (Prilep P-23, P 12-2/1, NS-72, P 66-9 /7, P-79-94 and Prilep Basma 82), found that the average values ranged from 59,3 cm in Prilep P 12-2/1 to 148,1 cm in Prilep Basma 82. Compared to the check (P-23), stalk height is lower only in variety Prilep P 12-2/1 and in all other varieties it is higher. Korubin – Aleksoska A. (2015), in her four-year investigations on dry yield of some new Prilep varieties and old varieties of Prilep, Yaka and Djebel tobacco found that they were all stable, but the most stable one was P-66-9/7 and it also had the highest yield. The average yield of the new varieties was 173% higher than that of the old domestic varieties. This is the best confirmation of the successful work of Tobacco Institute - Prilep in the selection of oriental aromatic tobaccos.

The aim of the study is to investigate and compare the height of the stalk, number and length of the leaves and dry mass yield per stalk, in some autochthonous varieties and new commercial varieties, in order to get a better knowledge and more successful selection of oriental tobacco in Macedonia.

II. Materials And Methods

Three-year investigations were made on three morphological traits: height of the stalk with inflorescence, number of leaves per stalk and length of the middle belt leaves, and dry mass yield per stalk as a agronomic trait, in five autochthonous varieties of the types: Prilep (P 10-3/2 and P 12-2/1), Djebel (Dj № 1) and Yaka (YK 7-4/2 and KY –

KishinskaYaka), and five new commercial varieties of the type Prilep: P-23, P-84, NS-72, P-66-9/7 and P-79-94. The morphological traits were measured during tobacco growth in the field, and the agronomic trait was measured at the manipulation of tobacco. Each amount represents the arithmetic average for the examined trait that is accompanied by error of the mean value, and also on variability of this trait by calculating the standard deviation and coefficient of variation (Najčevska, 2002).

During tobacco vegetation in field (May–September) in 2012, mean monthly temperature was 20.3⁰C, number of rainy days 26 and total precipitation 180 mm. In the same period in 2013 mean monthly temperature was 19.4⁰C, number of rainy days 34 and total precipitation 153 mm, and in 2014 mean monthly temperature was 18.3⁰C, number of rainy days 33 and total precipitation 223 mm.

General characteristics of the old domestic – autochthonous tobacco varieties

The cultivation of old tobacco varieties in this region began long ago, during the Ottoman Empire. The centuries–long presence led to their adaptation to the present agro–ecological conditions. Through successive natural selection they have acquired resistance to drought and diseases and can rightly be called autochthonous. Today, the old varieties make a valuable material for breeding activity in the Institute.

Prilep P 10-3/2 – characterized by cup–like habitus, average stalk height 50 cm, with 30–36 sessile leaves, dry mass yield averages 1200 kg/ha (Fig. 1).

Prilep P 12-2/1 – characterized by cup–like habitus, average stalk height 55 cm, with 34–38 sessile leaves, dry mass yield averages 1500 kg/ha (Fig. 2).

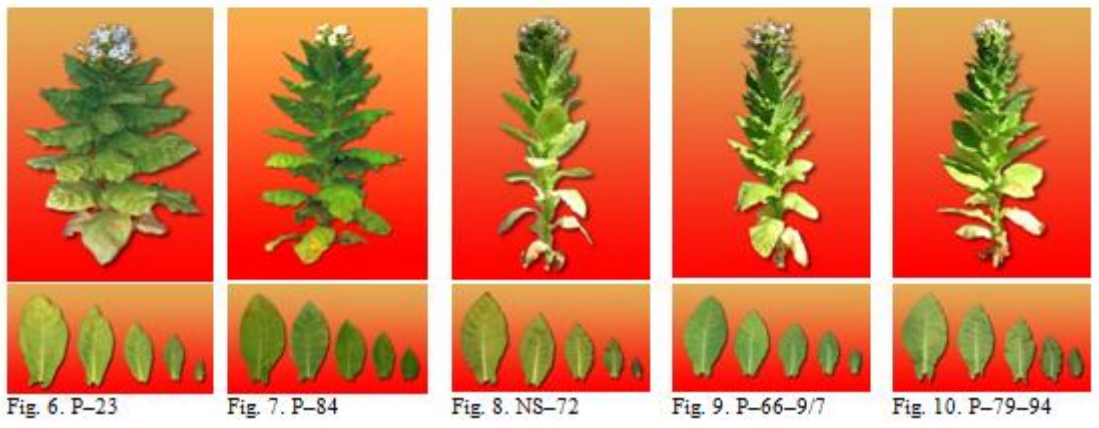
P 10-3/2 and P12-2/1 are put into production in the 30–ies of the last century; phenotypic and genotypic are very similar; originating from the local tobacco variety Djumaj–bale from GornaDjumaja–Bulgaria.

DjebelDj № 1– released in the first half of the last century; originated from the local variety XanthianYaka grown in the Djebel tobacco producing region in Bulgaria; characterized by a cylindrical habitus, average stalk height 80 cm, 26–30 sessile leaves erected toward the stalk, dry mass yield averages 1000 kg/ha (Fig. 3).

Yaka YK 7-4/2 – released in mass production in 1932. Originated from XanthianYaka originating from Xanthi–Greece;aplant with narrow, spindle shaped–elliptic habitus; average stalk height 100 cm, with 26–32 sessile leaves, dry mass yield averages 1000 kg/ha (Fig. 4).

These four domestic varieties were created in the Tobacco Institute–Prilep by Rudolf Gornik (Gornik, 1973), by individual selection(Borojević, 1981).

KY (KishinskaYaka) – According to some unconfirmed reports, this variety originates from village Kishino in the region of Veles, Republic of Macedonia. According to other surces it was transmitted in the past from Moldova (Uzunoski, 1985). Environmental conditions had a great influence on the morphology and chemistry of this genotype and with multi–decades selection a uniform and stable variety was formed. It is characterized by elongated–elliptic habitus with about 40 sessile leaves (Fig. 5).



General characteristics of the new commercial tobacco varieties

The new commercial oriental sun-cured varieties created in the Scientific Tobacco Institute-Prilep present a high quality raw material for the domestic and world market. Due to their pleasant aroma and harmonious chemical composition they enter in the mixtures of the highest-quality cigarette brands.

Prilep P-23– created by KostadinNikoloski and Milan Mitreski, through hybridization and selection in Tobacco Institute-Prilep; recognized by the Ministry of Agriculture, Forestry and Water Management of the Republic of Macedonia in 1995 (Korubin – Aleksoska,2004). It has elliptical-conical habitus, average stalk height 65 cm, with about 55 densely arranged leaves, dry mass yield 2000–2500 kg/ha (Fig. 6).

Prilep P-84 – created by Kiril Naumovski and Ana Korubin – Aleksoska, through hybridization and selection; recognized in 1988 in former Yugoslavia, as one of the first varieties of the type Prilep. Characterized by cylindrical-elliptical habitus, average stalk height 65 cm, with approximately 40–42 sessile leaves, elliptical in shape, dry mass yield 2500–3200 kg/ha (Fig. 7).

Prilep NS-72– created by Dushko Boceski and Simeon Karayankov; recognized in 1984 in former Yugoslavia as one of the first varieties of the type Prilep obtained by crossing (Korubin – Aleksoska et al.,2012); characterized by cylindrical-elliptical habitus, average stalk height 75 cm, with approximately 50 sessile leaves, elliptical in shape, dry mass yield 2800–3300 kg/ha (Fig. 8).

Prilep P-66-9/7– created in Tobacco Institute-Prilep by Miroslav Dimitrieski, Gordana Miceska and Aco Siskoski; recognized by the Ministry of Agriculture, Forestry and Water Management of R. Macedonia in 2004 (Korubin – Aleksoska et al., 2012); characterized by elliptical-conical habitus, with 54–60 ovate leaves, sessile and evenly distributed on the stem, dry mass yield 3000–3600 kg/ha. It has been the most represented tobacco variety in our country in recent years (Fig. 9).

Prilep P-79-94 – created in Tobacco Institute-Prilep by Milan Bogdanceski; recognized by the Ministry of Agriculture, Forestry and Water Management of R. Macedonia in 2001 (Korubin – Aleksoska, 2004); characterized by cylindrical-elliptical habitus, average stalk height 75 cm, with about 55 sessile leaves densely distributed, especially in the upper part of stem, dry mass yield 2500–3000 kg/ha (Fig. 10).

III. Results And Discussion

The highest stalk with inflorescence among the genotypes was found in KY ($\bar{x} = 121.7$ cm), and the lowest in P 10-3/2 ($\bar{x} = 51.7$ cm), (Table 1). Analysis of variance for height of the stalk showed highly significant differences among the varieties (only in the combinations Dj № 1 – P-66-9/7 and P-23 – P-84 the difference was not significant).

Table 1 - Height of the stalk with inflorescence in autochthonous and new commercial tobacco varieties.Prilep, May 2015

Tobacco varieties	Height of the stalk with inflorescence (cm)								
	2012			2013			2014		
	$\bar{x} \pm s_x$	δ	V (%)	$\bar{x} \pm s_x$	δ	V (%)	$\bar{x} \pm s_x$	δ	V (%)
Autochthonoustobacco varieties									
1. P 10-3/2	50 ± 0.21	5.74	9.78	51 ± 0.20	5.44	9.07	54 ± 0.18	5.03	8.45
2. P 12-2/1	50 ± 0.22	5.81	9.92	52 ± 0.22	5.53	9.43	56 ± 0.29	5.79	10.28
3. Dj № 1	75 ± 0.27	5.99	7.90	76 ± 0.25	5.82	7.73	80 ± 0.21	5.52	6.73
4. YK 7-4/2	98 ± 0.35	7.28	6.84	100 ± 0.33	7.26	6.01	100 ± 0.32	6.94	5.36
5. KY	120 ± 0.41	7.47	5.98	120 ± 0.36	7.47	5.98	125 ± 0.33	7.44	5.96
Commercial tobacco varieties									
6. P-23	65 ± 0.24	6.17	5.62	66 ± 0.24	6.08	5.03	70 ± 0.24	6.12	5.02
7. P-84	65 ± 0.27	5.45	5.37	67 ± 0.27	5.34	5.28	72 ± 0.27	5.03	5.01
8. NS-72	70 ± 0.25	6.69	10.38	72 ± 0.25	6.47	11.53	75 ± 0.25	6.29	10.05
9. P-66-9/7	73 ± 0.31	6.48	5.92	75 ± 0.31	6.55	5.83	79 ± 0.31	6.37	5.79
10. P-79-94	70 ± 0.29	6.73	5.97	73 ± 0.29	6.46	5.40	78 ± 0.29	5.92	5.02

LSD_{0,05} = 1.36

LSD_{0,01} = 2.44

The highest leaf number per stalk among the autochthonous varieties was found in Kishinska Yaka-KY ($\bar{x} = 37$) and the lowest in YK 7-4/2 and Dj № 1 ($\bar{x} = 27$). Among the commercial varieties, the highest leaf number was found in P-66-9/7 ($\bar{x} = 59$) and the lowest in P-84 ($\bar{x} = 40$). The variety P-66-9/7 has 32 leaves more than YK 7-4/2 and Dj № 1 (Table 2). Analysis of variance for the number of leaves per stalk showed highly significant differences among the varieties (only in the combination Dj № 1 – YK 7-4/2 the difference was not significant). This is an indication of different genotypes. The

Study of the Morphological and Agronomic Traits in Some Autochthonous and Commercial Tobacco..

differences between varieties in the number of leaves between 2012 and 2013 are not significant, while between 2012 and 2014, and between 2013 and 2014 the differences are small but significant, due to the fact that in 2014 there was more rain.

Table 2 - Number of leaves per stalk in autochthonous and new commercial tobacco varieties. Prilep, May 2015

Tobacco varieties	Number of leaves per stalk								
	2012			2013			2014		
	$\bar{x} \pm s_x$	δ	V (%)	$\bar{x} \pm s_x$	δ	V (%)	$\bar{x} \pm s_x$	δ	V (%)
----- Autochthonous tobacco varieties -----									
1. P 10-3/2	31 ± 0.09	1.80	5.43	32 ± 0.09	1.70	4.79	34 ± 0.08	1.72	4.88
2. P 12-2/1	32 ± 0.10	1.90	5.94	34 ± 0.10	1.90	5.57	36 ± 0.11	1.83	5.31
3. Dj № 1	25 ± 0.17	1.70	5.29	26 ± 0.16	1.50	5.13	29 ± 0.14	1.51	5.04
4. YK 7-4/2	26 ± 0.09	2.98	5.18	26 ± 0.17	2.28	4.85	28 ± 0.15	2.59	4.27
5. KY	37 ± 0.09	3.11	7.55	36 ± 0.15	3.06	7.16	38 ± 0.15	2.74	6.25
----- Commercial tobacco varieties -----									
6. P-23	45 ± 0.09	3.84	6.22	45 ± 0.20	3.51	6.73	48 ± 0.18	3.39	5.41
7. P-84	38 ± 0.10	3.05	5.71	40 ± 0.14	2.84	5.94	42 ± 0.15	2.59	5.07
8. NS-72	43 ± 0.17	5.73	5.93	43 ± 0.21	4.92	5.32	45 ± 0.17	4.26	5.39
9. P-66-9/7	56 ± 0.09	6.04	6.20	58 ± 0.15	5.87	6.52	62 ± 0.15	5.27	6.18
10. P-79-94	51 ± 0.09	6.38	8.45	52 ± 0.14	5.78	7.83	54 ± 0.15	5.82	7.24

LSD_{0,05} = 1.02

LSD_{0,01} = 1.84

The highest leaf length among the investigated varieties were measured in P-79-94 (\bar{x} = 23.2 cm) and the lowest in YK 7-4/2 (\bar{x} = 18.3 cm). The difference in length of the middle belt leaves between the two varieties is 4.9 cm (Table 3).

The comparison of the middle belt leaf length of the semi-oriental variety Otlia O 9-18/2 - \bar{x} = 32.75 cm (Fig. 11), with that of the variety P-79-94 shows that leaves of O 9-18/2 are 9.55 cm longer compared to P-79-94 and 14.45 cm longer compared to YK 7-4/2. The analysis of variance for the length of the middle belt leaf shows highly significant differences among varieties in 62.22% and 0.05 significance in 17.8%. No significance was observed in 20% of the combinations. The significance of differences in about 80% of the combinations indicates that they are different genotypes.

Table 3 -Length of the middle belt leaves in autochthonous and new commercial tobacco varieties. Prilep, May 2015

Tobacco varieties	Length of the middle belt leaves (cm)								
	2012			2013			2014		
	$\bar{x} \pm s_x$	δ	V (%)	$\bar{x} \pm s_x$	δ	V (%)	$\bar{x} \pm s_x$	δ	V (%)
----- Autochthonous tobacco varieties -----									
1. P 10-3/2	21.5 ± 0.12	2.24	8.33	22.1 ± 0.10	2.23	7.87	22.4 ± 0.11	2.05	7.93
2. P 12-2/1	23.09 ± 0.17	2.26	8.39	22.5 ± 0.12	2.22	8.23	23.3 ± 0.11	2.18	8.04
3. Dj № 1	20 ± 0.11	2.25	7.54	19.4 ± 0.11	2.20	7.51	20.2 ± 0.10	2.22	7.46
4. YK 7-4/2	18 ± 0.09	2.57	9.55	18.2 ± 0.08	2.46	9.55	18.7 ± 0.09	2.47	9.35
5. KY	21.82 ± 0.13	2.56	11.73	21.8 ± 0.12	2.56	10.90	22.1 ± 0.11	2.36	11.15
----- Commercial tobacco varieties -----									
6. P-23	20 ± 0.18	1.98	7.86	20.2 ± 0.13	2.15	7.99	22.4 ± 0.12	1.87	7.58
7. P-84	20 ± 0.15	2.03	7.92	20.4 ± 0.10	1.78	7.75	21.3 ± 0.09	1.95	7.83
8. NS-72	22.12 ± 0.14	2.04	8.69	21.8 ± 0.17	2.13	8.54	22.9 ± 0.15	1.99	8.44
9. P-66-9/7	22.03 ± 0.12	2.27	9.41	20.9 ± 0.12	2.16	9.15	21.5 ± 0.14	2.12	9.29
10. P-79-94	23 ± 0.11	2.41	10.33	23.2 ± 0.12	2.37	10.42	23.4 ± 0.11	2.18	9.95

LSD_{0,05} = 0.62

LSD_{0,01} = 1.12

The highest dry mass yield per stalk among the investigated tobacco genotypes was found in P-66-9/7. The lowest yield among the commercial varieties was recorded in P-23. The yield of the autochthonous varieties was 17.5% lower compared to the newly-created varieties. The highest yield among them was recorded in Kishinska Yaka and the lowest in Dj

№ 1. In 2014, dry mass yield in P-66-9/7 was 24.47 g / stalk, which is 28% higher than the yield of Dj № 1 – 6,93 g / stalk in 2012 (Table 4). The analysis of variance for dry mass yield per stalk showed highly significant differences in 42 combinations, and only in three combinations 5% significant difference was observed (P 10-3/2 – YK 7-4/2, P-23 – P-79-94 and P-84 – NS-72). These values indicate that the tested varieties are different genotypes. According to the above results, somewhat higher values were obtained in 2014, due to the higher precipitation during the growing season.



Fig. 11. O 9-18/2

Table 4 - Dry mass yield per stalk in autochthonous and new commercial tobacco varieties. Prilep, May 2015

Tobacco varieties	Dry mass yield per stalk (g)								
	2012			2013			2014		
	$\bar{x} \pm s\bar{x}$	δ	V (%)	$\bar{x} \pm s\bar{x}$	δ	V (%)	$\bar{x} \pm s\bar{x}$	δ	V (%)
----- Autochthonoustobacco varieties -----									
1. P 10-3/2	10.72 ± 0.13	0.59	5.81	10.54 ± 0.12	0.55	5.71	11.48 ± 0.12	0.38	5.21
2. P 12-2/1	13.12 ± 0.17	0.66	5.06	12.97 ± 0.11	0.46	5.16	13.65 ± 0.14	0.26	4.86
3. Dj № 1	6.93 ± 0.11	0.47	6.93	7.39 ± 0.07	0.44	6.03	8.02 ± 0.09	0.17	5.78
4. YK 7-4/2	10.18 ± 0.17	0.51	4.96	9.55 ± 0.12	0.56	4.36	10.83 ± 0.13	0.21	4.25
5. KY	15.25 ± 0.12	0.52	3.26	15.01 ± 0.10	0.50	3.79	15.91 ± 0.13	0.45	3.11
----- Commercial tobacco varieties -----									
6. P-23	17.15 ± 0.39	1.76	5.15	17.11 ± 0.16	0.72	6.37	17.61 ± 0.23	0.51	5.07
7. P-84	19.73 ± 0.30	1.35	3.64	19.23 ± 0.17	0.75	6.12	19.86 ± 0.21	0.63	5.18
8. NS-72	19.75 ± 0.43	1.91	6.38	19.34 ± 0.12	0.54	6.92	21.88 ± 0.97	0.24	5.79
9. P-66-9/7	24.18 ± 0.38	1.71	5.99	24.37 ± 0.12	0.54	6.23	24.47 ± 0.55	0.14	5.23
10. P-79-94	17.58 ± 0.42	1.87	4.45	17.99 ± 0.12	0.52	3.53	18.48 ± 0.81	0.31	4.55

LSD_{0,05} = 0.48

LSD_{0,01} = 0.87

The three-year biometric investigations of the autochthonous and commercial tobacco varieties for the investigated quantitative traits show low standard deviation and low degree of variability, which indicates stability and uniformity as a result of their homozygotness.

IV. Conclusion

- The paper includes three-years studies of some old domestic – autochthonous oriental varieties (P 10-3/2, P 12-2/1, Djebel Dj № 1, Yaka YK 7-4/2 and KY–Kishinska Yaka) and new commercial varieties (P-23, P-84, NS-72, P-66-9/7 and P-79-94).
- The highest among the investigated varieties is KY, and the lowest P 10-3/2. With the highest number of leaves is characterized P-66-9/7, and with the lowest number YK 7-4/2 and Dj № 1. The longest leaves has P-79-94, and shortest YK 7-4/2. The highest dry mass yield per stalk has P-66-9/7, and the lowest Dj № 1.
- Compared to the semi-oriental variety Otlia O 9-18/2 it can be concluded that differences in leaf length among the ten genotypes is within allowed limits for oriental aromatic tobaccos.
- Small error of the mean value and low variability for the traits is an indication of correct setting of the experiment, high genetic stability of the traits and homozygosity of the varieties.
- Analysis of variance for tested traits gives highly significant differences between the varieties, and it indicates that they are different genotypes.

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