EXPERIENCES WITH MULTIPLE-CROSS HYBRIDS IN FLUE-CURED TOBACCO

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ABSTRACT

Multiple-cross hybrids are routinely used in several outcrossing crops mainly to bypass difficulties in commercial hybrid seed production. An advantage of using a multiple-cross hybrid in tobacco, a self-pollinating crop, may lie in adding some variation to a cultivar thus making it more flexible in its response to environmental variability – e.g. soil- or weather-related. Even though self-fertile cultivars may show some residual variation left deliberately by the breeder, single-cross hybrids, now prevalent, are most often based on homozygous lines rather than cultivars and lack any inherent genetical variability. Another possible benefit may consist in more flexibility in constructing useful genotypes from available parental inbred lines or varieties. The main issue is the choice of the original inbred lines so that the introduced variation does not compromise the homogeneity of the agronomic traits required of a cultivar. Several three line hybrids of flue cured tobacco were produced, tested and compared against corresponding single-cross (F_1) hybrids based on the same true-breeding lines. One of these three line hybrids, dubbed VRG 5 TL, was found to outyield two F_1 hybrid combinations which involved the true-breeding parent and either of the homozygous lines used to produce the F_1 parent. VRG 5TL has been recently successfully released to tobacco growers in Poland. It shows acceptable agronomic performance which is in many ways superior to that of formerly released and widely grown F_1 hybrids.

OBJECTIVES

- Test the agronomic potential of three-line hybrids as flue-cured tobacco cultivars against their single-cross counterparts
- Test the degree of plant-to-plant variability exhibited by three-line hybrids of flue-cured tobacco.

MATERIALS & METHODS

For the F1 parent: two closely related lines WAC 120/3 and WAC 121 D7 representing a product of breeding resistance to black root root into cv. Wislica were chosen.

For the inbred parent three flue-cured genotypes distinct from Wislica were chosen: cv. Wisla, VP 06 and VB 08.

In the F1 parent WAC 120/3 was used as a CMS analogue and cms analogues of WAC 120/3 and WAC 121 D7 were used to produce single cross hybrids.

The following hybrid combinations were produced for the study:

- Six single-hybrid hybrid combinations which involved WAC 120/3 cms and WAC 121 D7 cms as female parents and Wisla, VP 06 and VB 08 as male parents
- Three three-line hybrid combinations which involved the F1 hybrid WAC 120/3 cms x WAC 121 D7 as the female parent and Wisla, VP 06 and VB 08 as male parents.

Hybrids were tested in a replicated field trial run in two years (2011 and 2012) and at two locations in eastern and south-eastern Poland. The soil was medium heavy silty loam at site 1 and sandy loam at site 2. For both locations plot size was 45 m^2 , number of plants per plot – 120. Planting distance was 70-90 cm x 45 cm for site 1 and 90 x 40 cm for site 2.

Standard agronomic practices for flue-cured tobacco were used which included transplant production in overhead watered plug trays, cultivation between rows, topping followed by application of contact suckericide, harvesting in six reapings of 2-3 leaves each and curing in a semi-automated bulk-curer.

The following yield parameters were measured: yield per unit area, percentage of the combined grades 1 and 2 in the total yield, quality index of cured crop ((grade 1 x 1.0 + grade 2 x 0.8+grade 3 x 0.5 + grade 4 x 0.2 + grade 5 x 0.1 + grade 6 x 0.0)/total yield), gross revenue in PLN per 1 ha

Morphological measurements included: plant height at full flowering, number of leaves length of internode (plant height divided by leaf number), leaf length, leaf width, width to length ratio, leaf area estimated as leaf length x leaf width x 0.65. The results were used to calculate plant to plant variation of morphological characteristics within an entry based on the measurements of 80 plants (4 plots x 20 plants on a plot). Variation was measured by using positional coefficient of variation based on the median and range between the 1st and 3rd quartile.

The experiment was set up as a randomized block design with four replications. Plot values of yield parameters and morphological measurements were subjected to ANOVA. The differences between means were examined for significance using Tukey's HSD at 0.05.

RESULTS

Yields. The yields were generally higher at site 1 than at site 2. The multiple hybrid with Wisla gave the highest yields at both sites, although the difference between it and the next entry was higher at site 2 (Tab. 1). It was also the highest yielder (Table when the average from two sites was calculated (Tab. 2). On aggregate across sites and years, the multiple with Wisła also gave the highest yields followed by the multiple with VB 08 (Tab. 3).

Crop quality indicators. The percentage of the 1st and 2nd grade in the total crop of cured leaves showed little variation from entry to entry but varied substantially depending on the year of the experiment. Generally, the studied hybrids scored much higher for the percentage of top grades in 2011 than in 2012 regardless of site (Tab. 2). On average, the highest percentage of top grades was shown by the single hybrid WAC 120/3 x Wisla (Tab. 3). Crop quality index also showed little variation from entry to entry and also generally showed much higher values in 2011 than in 2012 (Tab. 2). Overall, the highest crop quality index was shown by WAC 120/3 x Wisla. and the lowest by the multiple hybrid with Wisla (Tab. 3).

Gross money returns from 1 ha. Averaged across years, the highest revenue regardless of experiment year was obtained from the multiple with VB 08 at site 1 and from the multiple hybrid with Wisla at site 2 (Tab. 1). The multiple with Wisla performed best in both years, as an average of two sites (Tab. 2). Overall, the multiple hybrid with Wisla was also the highest producer of gross revenue per 1 ha when aggregated results from sites and years were considered (Tab. 3).

Generally, both in terms of yields per 1 ha and the money returns per 1 ha two multiple hybrids (WAC 120/3 x WAC 121 D7) x Wisla and WAC 120/3 x WAC 121 D7) x VB 08) were superior to their single-cross counterparts. The third multiple gave yields and money returns only slightly worse than the better single-cross counterpart. Of the single-cross hybrids WAC 121 D7 x Wisla was the best yielder and the best provider of gross revenue, ranking third after the two best performing multiples: (WAC 120/3 x WAC 121 D7) x Wisla and WAC 120/3 x WAC 121 D7) x VB 08 (Tab. 1, 2, 3).

Table 1. Selected parameters of the yield of cured leaves of three multiple (three-line) hybrids of flue-cured tobacco and of their single-cross counterparts at two locations averaged across two years of the experiment.

No.	Hybrid formula	yield t·ha^{-1}		% top grades (1+2)	
		site 1	site 2	site 1	site 2
1	WAC 120/3 x Wisla (single)	3.44	2.65	26.3	22.4
2	WAC 121 D7 x Wisla (single)	3.56	2.87	28.4	29.3
3	(WAC 120/3 x WAC 121 D7) x Wisla (multiple)	3.97	3.13	25.1	27.0
4	WAC 120/3 x VP 06 (single)	3.38	2.50	29.0	27.5
5	WAC 121 D7 x VP 06 (single)	3.29	2.64	21.8	25.9
6	(WAC 120/3 x WAC 121 D7) x VP 06 (multiple)	3.20	2.61	27.3	31.9
7	WAC 120/3 x VB 08 (single)	3.52	2.74	26.1	26.4
8	WAC 121 D7 x VB 08 (single)	3.54	2.70	26.6	22.8
9	(WAC 120/3 x WAC 121 D7) x VB 08 (multiple)	3.78	2.84	34.5	19.5
	HSD	0.572	0.553		
		crop quality index		crop value PLN·ha^{-1}	
		site 1	site 2	site 1	site 2
1	WAC 120/3 x Wisla (single)	0.67	0.59	31875	23545
2	WAC 121 D7 x Wisla (single)	0.62	0.60	33641	26233
3	(WAC 120/3 x WAC 121 D7) x Wisla (multiple)	0.63	0.54	37816	27393
4	WAC 120/3 x VP 06 (single)	0.62	0.58	30286	23852
5	WAC 121 D7 x VP 06 (single)	0.60	0.57	30571	25994
6	(WAC 120/3 x WAC 121 D7) x VP 06 (multiple)	0.63	0.57	31369	24974
7	WAC 120/3 x VB 08 (single)	0.67	0.55	33233	24911
8	WAC 121 D7 x VB 08 (single)	0.64	0.55	32603	25346
9	(WAC 120/3 x WAC 121 D7) x VB 08 (multiple)	0.65	0.53	35206	25976
	HSD			4790	

Table 2. Selected parameters of the yield of cured leaves of three multiple (three-line) hybrids of flue-cured tobacco and of their single-cross counterparts at two locations averaged across two years of the experiment.

No.	Hybrid formula	yield t·ha^{-1}		% top grades (1+2)	
		year 2011	year 2012	year 2011	year 2012
1	WAC 120/3 x Wisla (single)	3.89	3.04	59.4	24.4
2	WAC 121 D7 x Wisla (single)	4.45	3.22	48.7	28.8
3	(WAC 120/3 x WAC 121 D7) x Wisla (multiple)	4.90	3.55	45.6	26.1
4	WAC 120/3 x VP 06 (single)	4.00	2.94	48.5	28.2
5	WAC 121 D7 x VP 06 (single)	4.38	2.96	46.8	23.8
6	(WAC 120/3 x WAC 121 D7) x VP 06 (multiple)	4.36	2.90	46.0	29.6
7	WAC 120/3 x VB 08 (single)	4.23	3.13	52.9	26.3
8	WAC 121 D7 x VB 08 (single)	4.31	3.12	46.4	24.7
9	(WAC 120/3 x WAC 121 D7) x VB 08 (multiple)	4.60	3.31	50.6	27.0
	HSD	0.637	0.537		
		crop quality index		crop value PLN·ha^{-1}	
		year 2011	year 2012	year 2011	year 2012
1	WAC 120/3 x Wisla (single)	0.69	0.57	32874	22547
2	WAC 121 D7 x Wisla (single)	0.62	0.59	35534	24339
3	(WAC 120/3 x WAC 121 D7) x Wisla (multiple)	0.61	0.56	39336	25873
4	WAC 120/3 x VP 06 (single)	0.62	0.59	32177	21961
5	WAC 121 D7 x VP 06 (single)	0.62	0.56	35165	21400
6	(WAC 120/3 x WAC 121 D7) x VP 06 (multiple)	0.61	0.59	34915	21429
7	WAC 120/3 x VB 08 (single)	0.65	0.57	34982	23162
8	WAC 121 D7 x VB 08 (single)	0.61	0.58	34935	23014
9	(WAC 120/3 x WAC 121 D7) x VB 08 (multiple)	0.63	0.55	37246	23936
	HSD			6019	3089

Table 3. Selected parameters of the yield of cured leaves of three multiple (three-line) hybrids of flue-cured tobacco and of their single-cross counterparts at two locations averaged across two years of the experiment.

No.	Hybrid formula	yield t·ha^{-1}	% top grades (1+2)	crop quality index	crop value PLN·ha^{-1}
1	WAC 120/3 x Wisla (single)	3.47	41.9	0.631	27710
2	WAC 121 D7 x Wisla (single)	3.84	38.7	0.608	29937
3	(WAC 120/3 x WAC 121 D7) x Wisla (multiple)	4.23	35.8	0.584	32604
4	WAC 120/3 x VP 06 (single)	3.47	38.3	0.604	27069
5	WAC 121 D7 x VP 06 (single)	3.67	35.3	0.589	28282
6	(WAC 120/3 x WAC 121 D7) x VP 06 (multiple)	3.63	37.8	0.601	28172
7	WAC 120/3 x VB 08 (single)	3.68	39.6	0.611	29072
8	WAC 121 D7 x VB 08 (single)	3.71	35.5	0.596	28974
9	(WAC 120/3 x WAC 121 D7) x VB 08 (multiple)	3.95	38.8	0.588	30591
	HSD	0.58			4677

Variability of morphological parameters. Relatively large cQ for leaf length was recorded for the multiple hybrid with Wisla at site 2 and for the multiple with VP 06 at site 1. The two multiples also showed the highest variation for leaf length as a two-site average. Leaf width showed little site-to-site repeatability of cQ. The highest value was recorded at site 2 for the multiple with Wisla. When averaged across sites, the multiple hybrid ranked third after the single hybrid WAC 120/3 x VP 06 and its multiple counterpart. Leaf to length ratio was generally smaller in hybrids with Wisla than in those in the remaining entries. The highest values were recorded for the multiple hybrid with VB 08 and for the single-cross WAC 120/3 x VP 06. Leaf area was the most variable parameter of those studied. Across the sites, the highest cQ was recorded for multiple the multiple hybrid WAC 120/3 x WAC 121 D7) x VP 06. (Tab. 4).

Plant height and number of leaves showed little variability. As the average from two sites, the highest cQ value for height was recorded in the multiple with VP 06 but the highest single value was recorded for two singles at site 2: WAC 120 x VP06 and WAC 121 D7 x VB 08. Variation for leaf number as site average was the highest for the single cross WAC 121 D7 x VB 8.

Generally, except those for leaf area at site 2 cQ values did not exceed 10% regardless of hybrid formula . (Tab. 4).

Table 4. Variation for the dimensions of the 8th leaf, plant height and leaf number measured as the positional variation coefficient of three multiple (three-line) hybrids of flue-cured tobacco and of their single-cross counterparts at two locations of the experiment in 2012

No.	Hybrid formula	8th leaf length			8th leaf width		
		site 1	site 2	avg 2 sites	site 1	site 2	avg 2 sites
1	WAC 120/3 x Wisla (single)	4.9	5.8	5.3	6.1	5.9	6.0
2	WAC 121 D7 x Wisla (single)	3.8	3.7	3.7	8.8	3.8	6.3
3	(WAC 120/3 x WAC 121 D7) x Wisla (multiple)	4.2	8.4	6.3	4.2	9.9	7.1
4	WAC 120/3 x VP 06 (single)	5.5	4.0	4.8	7.6	9.4	8.5
5	WAC 121 D7 x VP 06 (single)	6.1	5.4	5.8	4.5	4.5	4.5
6	(WAC 120/3 x WAC 121 D7) x VP 06 (multiple)	8.6	4.8	6.7	7.9	8.5	8.2
7	WAC 120/3 x VB 08 (single)	5.9	3.7	4.8	5.6	6.1	5.8
8	WAC 121 D7 x VB 08 (single)	5.5	4.8	5.1	5.4	7.4	6.4
9	(WAC 120/3 x WAC 121 D7) x VB 08 (multiple)	6.1	5.5	5.8	7.1	6.3	6.7
		8th leaf width/length ratio			8 th leaf area		
		site 1	site 2	avg 2 sites	site 1	site 2	avg 2 sites
1	WAC 120/3 x Wisla (single)	2.7	4.7	3.7	9.6	9.6	9.6
2	WAC 121 D7 x Wisla (single)	4.0	3.7	3.8	11.7	6.4	9.0
3	(WAC 120/3 x WAC 121 D7) x Wisla (multiple)	2.6	2.3	2.5	6.5	16.9	11.7
4	WAC 120/3 x VP 06 (single)	4.6	6.0	5.3	9.2	13.4	11.3
5	WAC 121 D7 x VP 06 (single)	3.9	5.3	4.6	11.0	10.0	10.5
6	(WAC 120/3 x WAC 121 D7) x VP 06 (multiple)	4.5	3.6	4.0	14.1	13.4	13.7
7	WAC 120/3 x VB 08 (single)	5.0	5.0	5.0	9.8	11.8	10.8
8	WAC 121 D7 x VB 08 (single)	5.0	4.8	4.9	8.3	11.0	9.7
9	(WAC 120/3 x WAC 121 D7) x VB 08 (multiple)	3.5	6.9	5.2	10.8	12.3	11.5
		plant height			leaf number		
		site 1	site 2	avg 2 sites	site 1	site 2	avg 2 sites
1	WAC 120/3 x Wisla (single)	2.9	4.2	3.6	4.5	6.8	5.7
2	WAC 121 D7 x Wisla (single)	2.6	2.5	2.5	4.3	6.0	5.2
3	(WAC 120/3 x WAC 121 D7) x Wisla (multiple)	3.2	2.1	2.7	4.5	4.4	4.5
4	WAC 120/3 x VP 06 (single)	3.6	5.4	4.5	3.8	6.8	5.3
5	WAC 121 D7 x VP 06 (single)	3.8	3.1	3.4	4.3	4.3	4.3
6	(WAC 120/3 x WAC 121 D7) x VP 06 (multiple)	4.6	4.7	4.7	4.3	6.5	5.4
7	WAC 120/3 x VB 08 (single)	3.9	3.6	3.8	6.3	7.8	7.0
8	WAC 121 D7 x VB 08 (single)	3.6	5.4	4.5	6.0	8.3	7.2
9	(WAC 120/3 x WAC 121 D7) x VB 08 (multiple)	5.0	3.6	4.3	4.0	4.2	4.1

APPENDIX

In a field trial run on a farmer's field at the village of Dorbozy, southeastern Poland, in 2012, the three-line flue-cured tobacco hybrid VRG 5TL ((WAC 120/3 x WAC 121 D7) x Wisla) was compared to commercially grown black-root rot resistant single-cross hybrids and to the standard inbred cultivar Wislica. VRG 5 TL ranked the highest in terms of yield and second highest in terms of gross money returns to the grower (Tab. A). However, it scored the lowest with regard to crop quality indicators (percentage of top grades and crop quality index calculated based on contribution of grades 1 to 6 in the total crop. The low performance on crop quality of VRG 5TL was partly due to the genetic makeup of the multiple hybrid which was different from that of the other cultivars resulting in a different leaf ripening pattern and curing requirements not fully compatible with those of Wiślica and Wiślica-based single-cross hybrids.

Table A. Agronomic performance of standard inbred cv. Wiślica, three single-cross hybrid varieties currently under cultivation (VRG 1, VRG 2, VRG 4), a new single cross hybrid variety VRG 7 and a three-line hybrid variety VRG 5TL (WAC 120/3 cms x WAC 121 D7) x Wisła grown in a variety trial established on a tobacco farmer's field at the village of Dorbozy, south-eastern Poland in 2012.

No.	Cultivar	yield t·h^{-1}	crop quality index	percentage of grades 1 +2	gross money revenue PLN·ha^{-1}
1	VRG 1	3.33	0.73	54	29275
2	VRG 2	3.28	0.70	48	28661
3	VRG 4	3.09	0.71	48	27072
4	VRG 7	3.47	0.70	47	30156
5	VRG 5TL	3.49	0.65	38	29431
6	Wiślica	3.05	0.73	56	26803

VRG 5TL is now commercially produced along with single-cross hybrid varieties by many tobacco growers across Poland. Photo shows a field of VRG 5TL at the village of Łukowa, south-estern Poland

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