

Mezinárodní vědecký časopis vydávaný z pověření Ministerstva zemědělství České republiky a pod gescí České akademie zemědělských věd

An international journal published under the authorization by the Ministry of Agriculture and under the direction of the Czech Academy of Agricultural Sciences

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**Periodicita:** Časopis vychází měsíčně (12x ročně), ročník 45 vychází v roce 1999.

**Přijímání rukopisů:** Rukopisy ve dvou vyhotoveních je třeba zaslat na adresu redakce: RNDr. Eva Stříbrná, vedoucí redaktorka, Ústav zemědělských a potravinářských informací, Slezská 7, 120 56 Praha 2, Česká republika, tel.: 02/24 25 79 39, fax: 02/24 25 39 38, e-mail: editor@login.cz. Den doručení rukopisu do redakce je publikován jako datum přijetí k publikaci.

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**Aims and scope:** Original scientific papers, results of research, review studies and analyses from the crop production sector, particularly care of crops, crop yield formation, quality of plant products, seed production, plant physiology, agrochemistry, soil science, microbiology and agri-ecology are published in this periodical.

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**Periodicity:** The journal is published monthly (12 issues per year), Volume 45 appearing in 1999.

**Acceptance of manuscripts:** Two copies of manuscript should be addressed to: RNDr. Eva Stříbrná, editor-in-chief, Institute of Agricultural and Food Information, Slezská 7, 120 56 Praha 2, Czech Republic, tel.: 02/24 25 79 39, fax: 02/24 25 39 38, e-mail: editor@login.cz. The day the manuscript reaches the editor for the first time is given upon publication as the date of reception.

**Subscription information:** Subscription orders can be entered only by calendar year (January–December) and should be sent to: Institute of Agricultural and Food Information, Slezská 7, 120 56 Praha 2. Subscription price for 1999 is 195 USD (Europe), 214 USD (overseas).

# QUANTITATIVE PARAMETERS OF SOME BURLEY TOBACCO TRAITS

## KVANTITATIVNÍ PARAMETRY NĚKTERÝCH ZNAKŮ TABÁKU TYPU BURLEY

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**ABSTRACT:** The purpose of this study was to determine manner of inheritance, heritability, heterosis and inbreeding in burley tobacco for days to flowering, topping height, number of leaves, yield and grade index. Four parental cultivars and their F<sub>1</sub>, F<sub>2</sub>, Bc<sub>1</sub> and Bc<sub>2</sub> progenies were studied in a four-year experiment carried out in the Northern Western Croatia. The experiment was organized as a RCBD with four replications. Larger nonadditive, than additive, variance was found for all the studied traits. Rather high values of  $h_n^2$  were determined for almost all the traits whereas  $h_n^2$  was very low. Positive heterosis in F<sub>1</sub> was found for topping height, number of leaves and yield, and negative for days to flowering and grade index. Also in F<sub>2</sub>, inbreeding was found for all the traits for which the positive heterosis in F<sub>1</sub> was found. Distribution of parental genotypes along the expected regression line showed genetic divergence of the parental cultivars. The highest number of dominant genes was found for the TN 86 and of recessive for the BL1 and Hy 71.

**Keywords:** *Nicotiana tabacum* L.; burley; cultivar; inheritance; heritability; heterosis; inbreeding; agronomic and morphological traits

**ABSTRAKT:** Předmětem studia byly dědičnost, dědivost, heteroze a inbridink u tabáku typu burley pro počet dní do kvetení, výšku rostlin, počet listů, výnos a index kvality. Ve čtyřletém pokusu, realizovaném v severozápadním Chorvatsku, jsme sledovali čtyři rodičovské odrůdy a jejich potomstva F<sub>1</sub>, F<sub>2</sub>, Bc<sub>1</sub> a Bc<sub>2</sub>. Pokus byl založen jako RCBD se čtyřmi opakováními. Pro všechny sledované vlastnosti byla větší neaditivní než aditivní variance a vyšší hodnoty  $h_n^2$ , zatímco hodnota  $h_n^2$  byla velmi nízká. Byla zaznamenána pozitivní heteroze v generaci F<sub>1</sub> pro výšku rostlin, počet listů a výnos a negativní heteroze pro počet dní do kvetení a index kvality. Také v generaci F<sub>2</sub> byl zjištěn inbridink u všech vlastností, které vykazovaly kladnou heterozu v generaci F<sub>1</sub>. Rozložení rodičovských genotypů podél modelové regresní přímky naznačilo genetickou divergenci rodičovských odrůd. Nejvyšší počet dominantních genů jsme stanovili pro TN 86 a recesivních genů pro BL1 a Hy 71.

**Clíčová slova:** *Nicotiana tabacum* L.; typ burley; odrůda; dědičnost; dědivost; heteroze; inbridink; agronomické a morfologické vlastnosti

### INTRODUCTION

Most economically important tobacco traits are of quantitative nature. Investigations were carried out on burley tobacco because of its importance for the production of blend type cigarettes, but also because quantitative inheritance of major tobacco properties has been little studied on domestic genetic material in Croatia. The object of this research was to determine on specific burley tobacco materials: the manner of inheritance of some morphological (topping height, number of leaves and days to flower) and agronomic traits (yield and grade index) by means of heritability, heterosis and inbreeding and regression analysis.

The manner of inheritance by means of heritability of major economic traits of tobacco (yield, plant height,

number of leaves, days to flower, etc.) is generally rather low (Legg, Collins, 1971a; Oinuma, 1971). According to Legg, Collins (1971b, 1975), Okamura, Nakahara (1983) progress in selection may be expected after hybridization on specific materials.

Commercial achievements of growing F<sub>1</sub> tobacco hybrids, in major economic traits, have been reported by many authors (Legg et al., 1970; Legg, Collins, 1971b; Matzinger et al., 1971; Gudoy et al., 1987; Wilkinson, Rufty, 1990). However, the obtained mean performance of F<sub>1</sub> hybrids is in most cases higher than the mid-parent value only, while very rarely higher than the better parent value.

Based on the so far research, inbreeding rendered varying results, depending on the trait studied. According to Legg et al. (1970), Legg, Collins (1971a, b),

Matzinger et al. (1971), this also holds for yield and plant height, but not for number of leaves and days to flower.

Diallel design is used as one of the basic procedures for investigating the problems of inheritance of quantitative traits. Some authors find the additive-dominant model after Hayman (1954), Jinks (1954), Mather, Jinks (1971) to be one of the best models for estimating heritability and the manner of inheritance. Some studies using the graphic method indicate that the additive variance plays a somewhat greater role in inheriting most of the morphological traits (Gopinath et al., 1966; Shamsuddin et al., 1980; Jung et al., 1982; Pandeya et al., 1983; Shoai Daylami, Honarnejad, 1996).

## MATERIAL AND METHODS

Four genotypes have been chosen for these investigations: the American cultivar TN 86 (Miller, 1987) and three burley lines, BL1, Hy 71 (Devčić, Bolsunov, 1975) and Poseydon (Devčić et al., 1984), developed at the Tobacco Institute Zagreb.

Investigations started in 1992 at the experimental field Božjakovina of the Tobacco Institute Zagreb and lasted four years. The trial was set up as a RCBD with four replications. In the first year, the trial involved ten genotypes, viz. the four above mentioned genotypes and their  $F_1$  hybrids, obtained a year earlier: Hy 71 x TN 86, Hy 71 x BL1, Hy 71 x Poseydon, Poseydon x BL1, Poseydon x TN 86 and TN 86 x BL1. In 1993, the trial was extended by additional six  $F_2$  genotypes obtained by self-pollination of the  $F_1$  progeny, and in 1994 and 1995 by further 12  $Bc_1$  and  $Bc_2$  progenies.

The studied agronomic traits included the yield of tobacco leaves and grade index, while the morphological traits involved topping height, number of leaves and days to flower. Tobacco was harvested when it reached technical maturity. Curing was done by the standard

procedure for burley tobacco. Grade index is based on a 1 to 100 scale, with 100 representing the highest quality. Height after topping was measured from plant base to topping height. Leaf number was determined at the end of the growing season. Days to flower were recorded when 25% of flowers opened. Data for the analysis of studied traits were taken on a sample of 80 plants per experimental plot.

Data for all the studied traits and for each year were statistically processed by the analysis of variance and LSD test was performed. The broad-sense heritability was estimated according to the formula by Mahmud, Kramer (1951),  $h_b^2 = VF_2 - \sqrt{VP_1 \cdot VP_2} / VF_2$ , in 1993, 1994 and 1995, and the narrow-sense heritability according to the formula by Mather (1949)  $h_n^2 = 2VF_2 - (VBC_1 + VBC_2) / VF_2$  in 1994 and 1995.

Heterosis was calculated as relative to the mean of the better parent value and mid-parent value in all four years using the following formulas:  $H_1(\%) = F_1 - BP \cdot 100 / BP$  and  $H_2(\%) = F_1 - MP \cdot 100 / MP$ . Inbreeding was calculated in 1993, 1994 and 1995 according to the formula I.D.  $F_2(\%) = F_1 - F_2 \cdot 100 / F_1$ .

Regression analysis was applied to the studied material for all four years according to the methods of Jinks (1954), Hayman (1954), Mather, Jinks (1971).

## RESULTS

Significant differences between parents and  $F_1$  hybrids were found in all traits measured and all years, except for grade index in 1993, 1994 and 1995 and days to flowering in 1993 (Tab. I). The average performance of all parents and  $F_1$  hybrids from 1992 to 1995 are presented in Tab. II. Generally, medium to low  $h^2$  values were obtained in most investigation years for studied traits, irrespective of the year (Tab. III). There are also large differences within each genotype and between estimated heritability in its broad- and

I. Analysis of variance for yield, grade index, topping height, leaf number and days to flowering from 1992 to 1995

Year	Source of variation	d.f.	F values				
			yield	grade index	topping height	leaf number	days to flowering
1992	crosses	9	8.41**	4.06**	4.46**	10.37**	4.86**
	error	27					
	total	39					
1993	crosses	9	7.11**	1.95 n.s.	3.12*	3.97**	1.87 n.s.
	error	27					
	total	39					
1994	crosses	9	14.46**	1.08 n.s.	2.88*	7.64**	6.93**
	error	27					
	total	39					
1995	crosses	9	14.79**	0.92 n.s.	2.65*	3.29**	3.68**
	error	27					
	total	39					

II. Means of parents and F<sub>1</sub> hybrids for yield, grade index, topping height, leaf number and days to flowering from 1992 to 1995

Trait	Year	Genotype										LSD	
		TN 86	BL1	Hy 71	Poseydon	Hy 71 x TN 86	Hy 71 x BL1	Hy 71 x Poseydon	Poseydon x BL1	Poseydon x TN 86	TN 86 x BL1	0.05	0.01
Yield (kg/ha)	1992	2 738	2 156	1 486	2 391	2 688	2 814	1 878	3 054	2 969	2 618	502	678
	1993	3 095	3 094	1 891	2 579	2 718	3 103	2 940	3 526	3 391	3 034	500	676
	1994	3 130	1 296	2 183	3 160	2 526	2 609	2 786	3 489	3 198	3 193	492	665
	1995	2 512	1 464	1 598	1 903	2 626	2 437	2 089	2 721	2 564	2 749	354	479
Grade index	1992	39.68	33.26	23.76	34.49	38.63	41.27	32.32	42.20	38.91	35.16	7.86	10.62
	1993	36.33	35.70	35.15	40.85	39.30	39.53	35.86	43.51	38.44	37.42	n.s.	n.s.
	1994	38.20	37.65	38.90	39.92	41.56	37.65	40.93	40.08	42.11	36.40	n.s.	n.s.
	1995	32.46	32.44	33.59	33.49	31.13	34.46	33.46	33.02	33.85	28.90	n.s.	n.s.
Topping height (cm)	1992	135	131	107	138	150	151	132	144	149	136	17.89	24.18
	1993	135	133	132	142	148	160	155	159	157	133	19.37	n.s.
	1994	140	119	120	147	124	137	134	130	145	147	18.26	n.s.
	1995	145	126	135	120	137	147	126	133	151	143	16.83	n.s.
Leaf number	1992	26	23	20	26	25	26	21	27	26	28	2.21	2.99
	1993	25	24	23	25	24	25	24	25	26	25	1.18	1.60
	1994	26	25	23	26	26	26	25	27	26	27	1.27	1.71
	1995	28	24	28	24	26	27	26	27	27	28	2.04	2.82
Days to flowering	1992	92	86	95	92	79	79	89	86	89	94	7.58	10.24
	1993	83	82	76	77	76	75	74	79	75	81	n.s.	n.s.
	1994	79	82	80	82	76	72	70	70	74	81	5.43	7.23
	1995	85	82	94	77	84	82	77	79	85	85	7.34	9.77

narrow-sense, depending on the investigation year. In contrast to other traits, broad-sense heritability for the number of leaves shows less variation. As a rule, narrow-sense heritability for studied traits is low in all genotypes, with the exception of the leaf yield in genotype Poseydon x TN 86, the plant height in genotype Hy 71 x BL1, number of leaves in genotype Hy 71 x Poseydon and days to flowering in genotype Poseydon x BL1. It also shows less variation than the broad-sense heritability.

It is noticeable that the F<sub>1</sub> hybrids gave higher yield and height than the mid-parent average, but also than better parent. The higher heterosis relative to the mid-parent value was estimated. The highest increase relative to the higher parent was recorded in the F<sub>1</sub> hybrids Poseydon x BL1 and Hy 71 x BL1. Negative heterosis for the number of leaves was recorded in most hybrids, or its absence relative to better parent values. Earlier flowering was recorded in our investigations in all F<sub>1</sub> hybrids relative to the mid-parent and better parent values.

In comparison with the F<sub>1</sub> hybrids, a yield and height decrease were recorded in most F<sub>2</sub> hybrids. The highest decrease was determined in the F<sub>2</sub> hybrids Poseydon x TN 86 and Hy 71 x BL1. Similar positive and negative values were determined for grade index and number of leaves. Occurrence of negative inbreeding was also recorded for days to flowering.

The expected regression line for F<sub>1</sub> hybrids cuts the *Wr* axis below the origin, indicating overdominance in

inheritance of the studied traits, except for topping height in 1994 and for number of leaves in 1992 and 1993 when they were inherited partially dominantly (Figs 1 to 5). Since regression coefficient does not differ significantly from  $b = 1$ , no interallelic interaction is present, except for grade index in 1994 and 1995 and days to flowering in 1994. Distribution of parent genotypes on the scatter diagram along the expected regression line points to genetic divergence of the parents. Differences in terms of investigation years are also present. The highest number of dominant genes were found for most studied traits and most investigation years for the cultivar TN 86 and of recessive genes for the cultivars BL1 and Hy 71.

## DISCUSSION

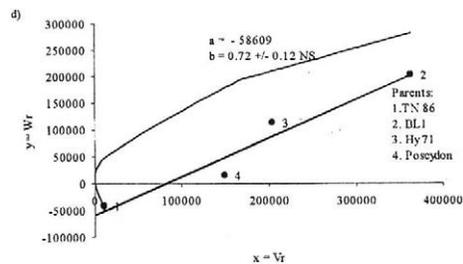
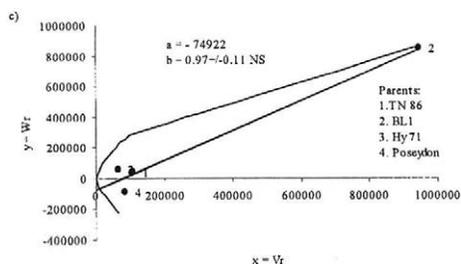
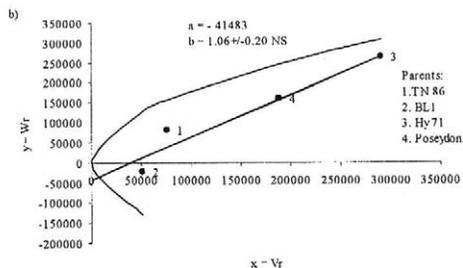
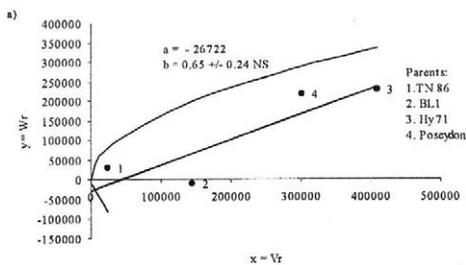
In recent years, selection programmes have been aimed at increasing the yield and grade index, but have been also aimed at decreasing the tobacco plant height without reducing the number of leaves. This should result in more lodging resistant stalks, and consequently in higher yields. Due to the generally small participation of additive variance in the yield, grade index and plant height inheritance, there is also a greater difference between the estimated broad- and narrow-sense heritability. Still, estimated heritability is

III. Values of the estimated parameters: heritability in broad- and narrow-sense, heterosis and inbreeding for yield, grade index, topping height, leaf number and days to flowering from 1992 to 1995

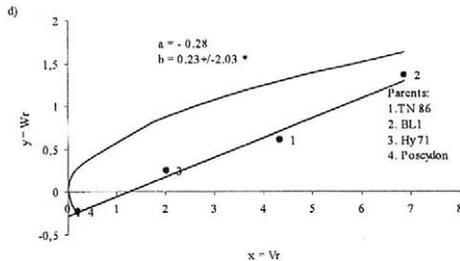
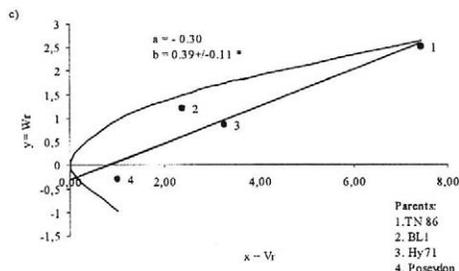
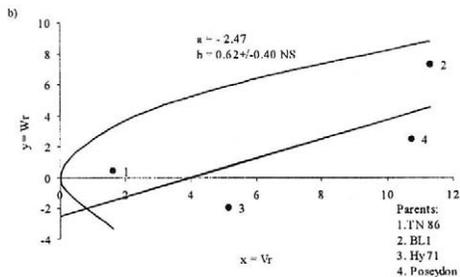
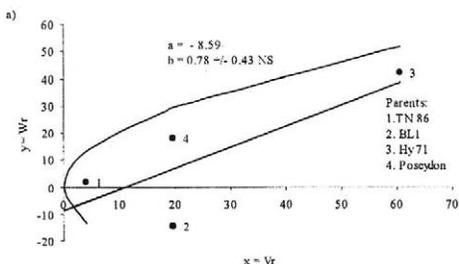
Genotype	Trait	Heritability (%)		Heterosis (%)		Inbreeding (%)
		in broad-sense	in narrow-sense	in comparison to the better parent	in comparison to the mid-parent	
Hy 71 x TN 86	yield	29.8–55.0	18.0–21.5	-15.71–4.53	9.02–45.17	-7.48–16.07
Hy 71 x BL1		24.5–39.5	10.6–14.8	-16.64–52.50	-1.80–59.17	9.72–23.68
Hy 71 x Poseydon		1.6–39.1	3.3–32.0	21.45–13.99	-3.14–31.54	3.59–18.98
Poseydon x BL1		23.7–46.0	10.1–16.3	10.41–42.98	24.28–61.57	-4.00–15.41
Poseydon x TN 86		42.6–68.4	38.0–39.5	1.20–9.56	1.68–19.52	-9.71–24.71
TN 86 x BL1		39.0–72.1	11.4–24.3	-4.38–9.43	-1.97–44.28	1.87–16.22
Hy 71 x TN 86		grade index	50.4–69.7	21.4–39.1	-26.40–6.83	-5.72–21.28
Hy 71 x BL1	29.2–60.1		19.0–23.5	-3.21–24.08	-1.62–44.75	-5.60–4.26
Hy 71 x Poseydon	14.2–30.1		12.5–14.2	-12.21–2.53	-0.23–10.98	-8.28–4.72
Poseydon x BL1	14.3–24.0		11.1–18.1	-1.40–22.35	0.18–24.59	-4.99–14.36
Poseydon x TN 86	40.4–52.8		11.1–40.9	-5.89–5.48	-0.38–2.66	3.06–3.91
TN 86 x BL1	27.7–40.0		5.5–20.0	-11.39–0.03	-10.93–3.91	-9.45–0.21
Hy 71 x TN 86	topping height		33.8–73.4	1.1–22.7	-11.42–11.11	-4.61–23.96
Hy 71 x BL1		49.2–79.8	30.2–42.3	8.88–20.30	12.21–26.89	6.57–14.38
Hy 71 x Poseydon		35.2–74.9	12.5–19.8	-8.84–9.15	-1.56–13.4	-4.76–20.64
Poseydon x BL1		31.0–84.6	13.0–20.3	-11.56–11.97	-2.25–15.21	-5.67–4.40
Poseydon x TN 86		37.9–46.2	1.7–26.0	-1.36–10.56	0.69–13.53	9.65–23.56
TN 86 x BL1		27.4–71.1	1.8–34.2	-1.48–5.00	-0.74–13.07	-1.39–12.93
Hy 71 x TN 86		leaf number	21.6–42.7	2.4–18.2	-7.14–0.00	-7.14–8.69
Hy 71 x BL1	14.6–48.3		3.6–13.7	-3.57–13.04	3.84–18.18	0.00–7.69
Hy 71 x Poseydon	43.6–65.3		37.0–42.5	-19.23–(-3.84)	-8.69–0.00	-3.84–4.00
Poseydon x BL1	50.8–57.7		12.8–13.4	0.00–12.50	0.00–12.50	-4.00–11.11
Poseydon x TN 86	49.3–62.0		23.7–27.1	-3.57–4.00	0.00–4.00	-3.70–7.69
TN 86 x BL1	51.0–59.5		3.2–10.7	0.00–7.69	0.00–12.00	0.00–11.11
Hy 71 x TN 86	days to flowering		48.5–65.4	12.7–13.0	-16.84–(-5.00)	-15.95–(-5.00)
Hy 71 x BL1		53.1–71.6	15.8–16.4	-16.82–(-8.53)	-13.18–(-5.06)	-5.33–(-1.38)
Hy 71 x Poseydon		48.4–55.5	22.6–45.6	-18.08–(-3.89)	-13.58–(-3.89)	-16.88–(-7.14)
Poseydon x BL1		50.8–80.4	40.0–54.5	-14.63–(-3.65)	-14.63–1.25	-10.28–0.00
Poseydon x TN 86		58.8–73.3	20.1–44.4	-9.75–0.00	-8.64–4.70	-21.62–(-10.58)
TN 86 x BL1		65.4–85.7	26.6–41.0	-2.40–2.17	-2.40–5.61	-7.05–7.40

largely a consequence of genetic differences between the parents. This is substantiated also by some other studies on inheritance involving several different tobaccos, as well as some inter-type crosses (Legg, Collins, 1971a, 1975; Oinuma, 1971; Pandeya et al., 1983; Povilaitis, 1966). Since the number of leaves is less influenced by external factors, according to most studies, estimated heritability is somewhat higher than in the other traits (Legg, Collins, 1971b). This is substantiated also by our investigation. Influenced by the extreme weather conditions during the investigation period in our investigations, the obtained broad-sense heritability values for days to flower were more variable. This is supported by the earlier investigations, as well as by large differences between estimated heritability in its broad- and narrow-sense (Legg, Collins, 1971a, 1975; Okamura, Nakahara, 1983).

The higher heterosis relative to the mid-parent value for yield and plant height in our investigations is in agreement with the investigations conducted by other authors (Legg et al., 1970; Legg, Collins, 1971a, b; Matzinger et al., 1971; Dean, 1974; Oupadisakoon, Wersman, 1976; Gudoy et al., 1987; Wilkinson, Ruffy, 1990). From the commercial point of view, positive heterosis is interesting only when the yield of a  $F_1$  hybrid exceeds the higher-yielding parent. Since genotypes TN 86 and Poseydon rendered highest yields in our investigations, while  $F_1$  hybrids Poseydon x BL1 and Poseydon x TN 86 achieved higher yields than the higher-yielding parents, these  $F_1$  hybrids might be chosen for the future commercial production. Negative heterosis for the number of leaves was recorded in most hybrids in our investigations, or its absence relative to better parent values. This is in agreement with the pre-



1.  $W_r V_r$  plots for yield (kg/ha) in 1992 (a), 1993 (b), 1994 (c) and 1995 (d)

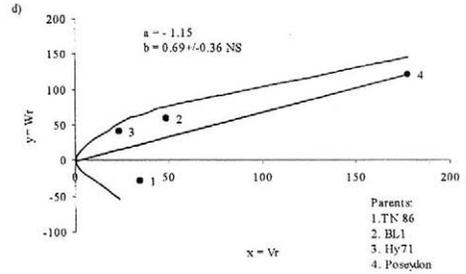
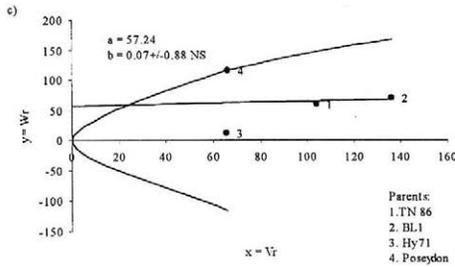
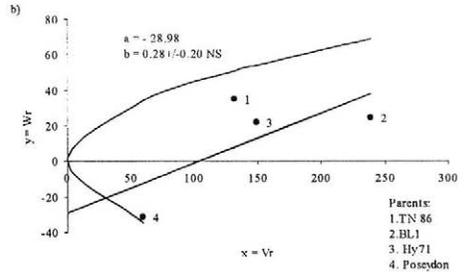
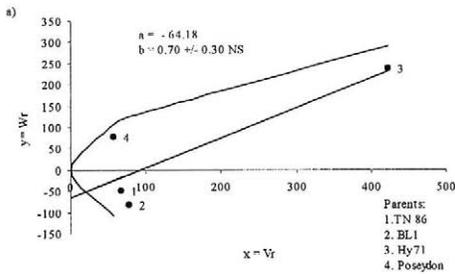


2.  $W_r V_r$  plots for grade index in 1992 (a), 1993 (b), 1994 (c) and 1995 (d)

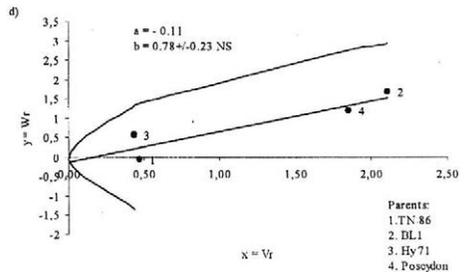
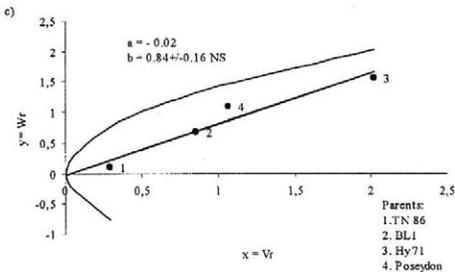
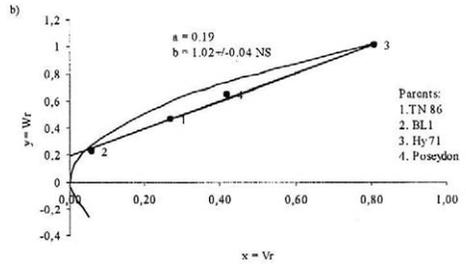
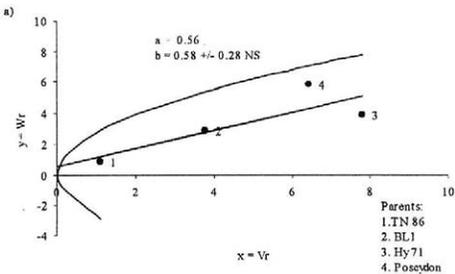
vious studies concerning this problem (Legg et al. 1970; Legg, Collins, 1971a, b; Gudoy et al., 1987). One of the advantages for which  $F_1$  hybrids are commercially used is their earlier flowering, which implies the presence of negative heterosis. This occurrence relative to the earlier maturing parent was mostly present in

other studies too. This indicates that early ripening genotypes may be created already in  $F_1$  generation by selection of adequate parents, which is also supported by our own results.

According to the literature data for inbreeding for studied traits, different values were obtained (Legg et



3.  $W_r V_r$  plots for height after topping (cm) in 1992 (a), 1993 (b), 1994 (c) and 1995 (d)

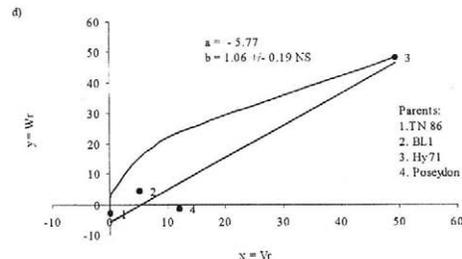
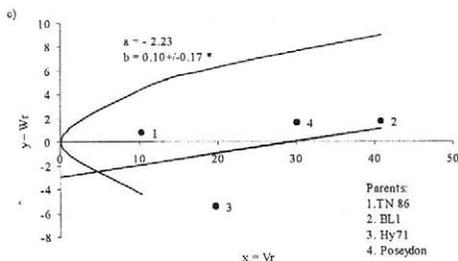
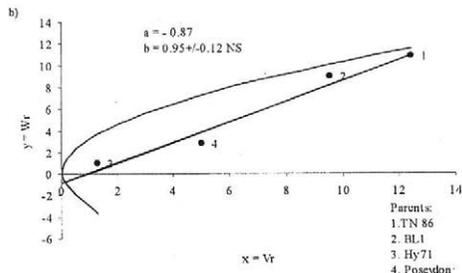
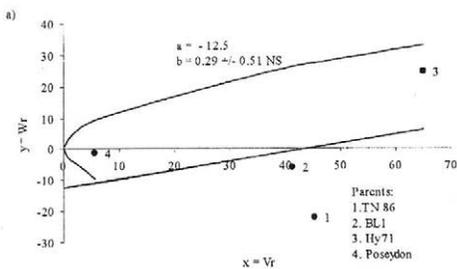


4.  $W_r V_r$  plots for number of leaves in 1992 (a), 1993 (b), 1994 (c) and 1995 (d)

al., 1970; Legg, Collins, 1971a, b; Dean, 1974; Oupadissakoon, Wersman, 1976). Significant inbreeding for yield was determined in our investigations, but its absence was recorded as well. No inbreeding for number of leaves has been recorded in most  $F_2$  hybrids. Occur-

rence of negative inbreeding for days to flower was also recorded in our investigations. It is mainly present in burley tobacco (Legg et al., 1970).

Regression analysis was applied in order to get a better insight into the quantitative traits studied. If the



5.  $Wt Vr$  plots for days to flowering in 1992 (a), 1993 (b), 1994 (c) and 1995 (d)

results obtained for the traits under study in our investigations are taken as a whole, it can be said that no interallelic interaction was present for all traits in most investigation years. The expected regression line cuts the  $Vr$  axis mostly below the origin which indicates to the prevalence of overdominance. Parent scatter in the coordinate system varies, however TN 86 has more dominant genes in most traits and years. The other three parents have mainly the same proportion of dominant and recessive genes, however with a trend towards recessive genes. In some earlier investigations similar results were obtained with different tobacco types by Shoai Daylami, Honarnejad (1996) for plant height, Jung et al. (1982) for yield and Pandeya et al. (1983) for days to flower. However, the overall literature results obtained to date suggest a somewhat greater role of additive variance in inheritance of these traits, but the role of non-additive variance cannot be fully excluded either.

## REFERENCES

- Dean C. E. (1974): Heterosis, inbreeding depression and combining ability in diallel crosses of cigar-wrapper tobacco. *Crop Sci.*, 14: 482-484.
- Devčić K., Bolsunov I. (1975): Čulinec, nova sorta burleya stvorena u Duhanskom institutu Zagreb. 9. Simp. duhanskih stručnjaka Jugoslavije: 10-12.
- Devčić K., Triplat J., Benković F. (1984): Sorte burleya selekcionirane u Duhanskom institutu Zagreb. *Tutun/Tobacco*, 1-2: 1-10.
- Gopinath D. M., Ramanarao V. V., Subrahmanyam M., Narayana C. L. (1966): A study of diallel crosses between

varieties of *Nicotiana tabacum* L. for yield components. *Euphytica*, 15: 171-178.

- Gudoy L. B., Ventura E. B., Rivera R. L. (1987): Diallel cross and combining ability in burley tobacco. *J. Tob. Sci. Techn.*, 1: 240-245.
- Hayman B. I. (1954): The theory and analysis of diallel crosses. *Genetics*, 39: 789-809.
- Jinks J. L. (1954): The analysis of continuous variation in a diallel cross of *Nicotiana rustica* varieties. *Genetics*, 39: 767-788.
- Jung S. H., Hwang J. K., Son S. H. (1982): The analysis of inheritance of quantitative characters with Oriental tobacco varieties (*Nicotiana tabacum* L.) in diallel cross. II. Gene distribution and analysis of variance for each character in  $F_1$  generation. *J. Korean Soc. Tob. Sci.*, 4: 15-20.
- Legg P. D., Collins G. B. (1971a): Genetic parameters in burley populations of *Nicotiana tabacum* L. I. Ky 10 x Burley 21. *Crop Sci.*, 11: 365-367.
- Legg P. D., Collins G. B. (1971b): Genetic parameters in burley populations of *Nicotiana tabacum* L. II. Virginia B-29 x Ky 12. *Tob. Ab.*, 10: 940.
- Legg P. D., Collins G. B. (1975): Genetic parameters in Ky 14 x Ky Ex 42 burley population of *Nicotiana tabacum* L. *Theor. Appl. Genet.*, 45: 264-267.
- Legg P. D., Collins G. B., Litton C. C. (1970): Heterosis and combining ability in diallel crosses of burley tobacco, *Nicotiana tabacum* L. *Crop Sci.*, 10: 705-707.
- Mahmud I., Kramer H. H. (1951): Segregation for yield, height and maturity following a soybean cross. *Agron. J.*, 43: 307-311.
- Mather K. (eds) (1949): *Biometrical genetics*. London, Methnen.
- Mather K., Jinks J. L. (eds) (1971): *Biometrical genetics*. London, Chapman and Hall.

- Matzinger D. F., Wersman E. A., Ross H. F. (1971): Diallel crosses among burley varieties of *Nicotiana tabacum* L. in the F<sub>1</sub> and F<sub>2</sub> generations. *Crop Sci.*, 11: 275-279.
- Miller R. D. (1987): Registration of TN 86 burley tobacco. *Crop Sci.*, 27: 365-366.
- Oinuma T. (1971): Inheritance and selection of quantitative characters in burley tobacco. I. Heritability and genotypic correlation in F<sub>2</sub> generation of three varietal crosses. *Tob. Ab.*, 2: 202.
- Okamura T., Nakahara K. (1983): Heritabilities and genotypic correlations of morphological traits in F<sub>2</sub> generation of the cross, Awa (Chisa) x N 303. *Bull. Okayama Tob. Exp. Stn.*, 42: 67-73.
- Oupadissakoon S., Wersman E. A. (1976): Agronomic performance and nature of gene effects in progenitor species-derived genotypes of tobacco. *Crop Sci.*, 17: 843-847.
- Pandeya R. S., Dirks V. A., Poushinsky G. (1983): Quantitative genetic studies in flue-cured tobacco (*Nicotiana tabacum*). I. Agronomic characters. *Can. J. Genet. Cytol.*, 25: 336-345.
- Povilaitis B. (1966): Diallel cross analysis of quantitative characters in tobacco. *Can. J. Genet. Cytol.*, 8: 336-346.
- Shamsuddin A. K. M., Newaz M. A., Razzaque C. A. (1980): Genetic analysis of leaf yield and component characters in tobacco (*Nicotiana tabacum* L.). *Z. Pfl.-Zücht.*, 84: 139-147.
- Shoai Daylami M., Honarnejad R. (1996): Gene effects and combining ability of some quantitative characteristics of tobacco varieties (*Nicotiana tabacum*). *Bull. Spec. Cores-ta*: 182.
- Wilkinson C. A., Rufty R. C. (1990): Diallel analysis of crosses among United States and European burley tobacco cultivars. *Tob. Sci.*, 34: 15-18.

Received on March 17, 1998

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# GENETIC ANALYSIS OF SOME CHEMICAL TRAITS IN BURLEY TOBACCO

## GENETICKÁ ANALÝZA NĚKTERÝCH CHEMICKÝCH VLASTNOSTÍ TABÁKU TYPU BURLEY

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**ABSTRACT:** Four parent burley tobacco cultivars (TN 86, BL1, Hy 71 and Poseydon), and their six F<sub>1</sub> hybrids (Hy 71 x TN 86, Hy 71 x BL1, Hy 71 x Poseydon, Poseydon x BL1, Poseydon x TN 86 and TN 86 x BL1) were separated for the needs of this study. Selected materials from a four-year trial, set up according to the RCBD in four replications, were used to study the manner of inheritance of nicotine, proteins, total nitrogen and ash content by means of the components of genetic variance and regression analysis. According to the components of genetic variance, the significant additive variance was estimated for nicotine, proteins and ash content, while the significant additive and nonadditive variance were estimated for total nitrogen content. Other results point to the presence of overdominant inheritance for all the traits studied, but not for all investigation years, as well as to the preponderance of dominant genes over recessive genes, however not of their symmetric distribution.

**Keywords:** *Nicotiana tabacum* L.; burley; components of genetic variance; regression analysis; chemical traits

**ABSTRAKT:** Vybrali jsme čtyři rodičovské odrůdy tabáku typu burley (TN 86, BL1, Hy 71 a Poseydon) a jejich šest hybridů F<sub>1</sub> generace (Hy 71 x TN 86, Hy 71 x BL1, Hy 71 x Poseydon, Poseydon x BL1, Poseydon x TN 86 a TN 86 x BL1). Na tomto materiálu jsme ve čtyřletém pokusu, založeném podle RCBD ve čtyřech opakováních, studovali dědičnost pro obsah nikotinu, proteinů, celkového dusíku a popela. Výsledky jsme vyhodnotili pomocí genetické variance a regresní analýzy. Na základě komponent genetické variance jsme u obsahu nikotinu, proteinů a popela zjistili průkaznou aditivní varianci, zatímco u obsahu celkového dusíku jsme zaznamenali průkaznou aditivní i neaditivní varianci. Výsledky naznačují existenci superdominantní dědičnosti u všech sledovaných vlastností, nikoliv však ve všech pokusných letech, a dále převahu dominantních genů nad recesivními, které však nejsou symetricky rozloženy.

**Klíčová slova:** *Nicotiana tabacum* L.; typ burley; komponenty genetické variance; regresní analýza; chemické vlastnosti

### INTRODUCTION

Quantitative traits are characterized by continuous distribution and include the effects of genes and the effects resulting from the genotype x environment interaction. Discrete effects of genes, however, cannot be measured directly, but the nature of their action may be detected by different statistical methods (Vasilij, 1973).

Most traits of breeding interest are polygenic controlled and are influenced by external factors. Chemical traits of tobacco, which are the subject of this paper, are also mainly quantitatively inherited. Thus, for instance, Legg et al. (1969) and Legg, Collins (1971) determined two loci with major genes and a few minor genes for nicotine content, the most important component of the tobacco chemical complex. This means that nicotine content is inherited qualitatively-quantitatively. Nicotine is affected by a number of external factors (fertilization, stalk position and ripeness) but also the cultivar itself (Beljo, 1992).

According to the studies conducted by Povelaitis (1966), significant additive and dominant variance was estimated for nicotine content on specific flue-cured tobacco materials using the components of genetic variance, while only significant additive variance was estimated by Matzinger, Wersman (1971) and Chiang, Chen (1985). Using the graphic method, Pandeya et al. (1984) estimated partial dominance in inheritance of total alkaloids, total nitrogen and ash content. No inter-allele interaction was estimated, however genetic divergence of the studied parent genotypes was present. Beljo (1980) estimated intermediary and partially dominant nicotine content inheritance in F<sub>1</sub> progeny of the crosses between flue-cured, burley, semioriental and oriental tobaccos. Beljo (1984) came to a similar conclusion in investigations following crossing of semioriental, burley and flue-cured tobaccos, and so did Chang, Shyu (1980). According to Beljo (1980), only dominant variance was significant in protein content inheritance.

The goal of these investigations was to estimate on specific materials: the manner of inheritance of nicotine, proteins, total nitrogen and ash content using the components of genetic variance and regression analysis.

## MATERIAL AND METHODS

Four-year investigations of selected burley tobacco materials were carried out according to the RCBD in four replications at the experimental field of the Tobacco Institute Zagreb in Božjakovina from 1992 to 1995. The trial involved four parent genotypes: American line cultivar TN 86 (Miller, 1987) and three burley lines BL1, Hy 71 (Devčić, Bolsunov, 1975) and Poseydon (Devčić et al., 1984) developed in the Tobacco Institute Zagreb. In all four investigation years, apart from the mentioned parent genotypes, the trial included also their six  $F_1$  hybrids, viz. Hy 71 x TN 86, Hy 71 x BL1, Hy 71 x Poseydon, Poseydon x BL1, Poseydon x TN 86 and TN 86 x BL1.

Of chemical traits, nicotine, proteins, total nitrogen and ash were estimated in an average third harvest sample on 80 plants for each genotype. Coresta methods were used for nicotine estimation, and AOAC methods for proteins, total nitrogen and ash.

Data for all the studied traits and for each year were statistically processed by the analysis of variance and LSD test was performed. Genetic variability was analyzed on the tested material in all four investigation years, components of genetic variance were calculated ( $D$ ,  $H_1$ ,  $H_2$ ,  $F$ ,  $E$ ,  $\sqrt{H_1/D}$ ,  $H_2/4H_1$  and  $Kd/Kr$ ), and regression analysis was done according to the methods of Jinks (1954), Hayman (1954) and Mather, Jinks (1971).

## RESULTS

Significant differences between parents and  $F_1$  hybrids were found in all traits measured and all years,

except for total nitrogen content in 1992 and 1993, and ash content in 1993 (Tab. I). The average performance of all parents and  $F_1$  hybrids for all studied traits from 1992 to 1995 are presented in Tab. II. The statistics representing additive effects of genes were significant for all four traits and all four years except for total nitrogen content in 1992 and 1993 and ash content in 1993 (Tab. III). Also, the statistics representing dominance effect were significant for all traits but only in 1994 for nicotine content, in 1992 and 1995 for protein content, and in 1994 and 1995 for total nitrogen and ash content. This, it may be taken that additive and nonadditive variance participated to nearly the same extent in inheritance of total nitrogen content. However, nicotine, proteins and ash content were additively inherited. The sign of the component  $F$  indicates the relative frequencies of dominant and recessive alleles. Positive values of  $F$  indicate an excess of dominant alleles and negative values an excess of recessive alleles.  $F$  for the studied traits is mainly of a positive sign and mostly marks dominant action, but only the  $F$  values for the ash content in 1995 were significant. A further ratio,  $Kd/Kr$  provides the estimate of the ratio between the total number of dominant to recessive genes in all the parents.  $Kd/Kr$  ratios are in line with the interpretation of values obtained for  $F$  values. The proportion  $H_2/4H_1$ , which provides an estimate of the average frequency ( $uv$ ) of positive ( $u$ ) versus negative ( $v$ ) alleles in the parents, where  $u + v = 1$  attains a maximum value of 0.25 when these frequencies are equal ( $uv = 0.50 \cdot 0.50 = 0.25$ ). The estimated values for all four traits deviated from 0.25, except for nicotine in 1993 and for ash content in 1992. For the proteins content in 1992, 1994 and 1995 and total nitrogen and ash content in 1993, however  $H_1 < H_2$  and therefore the ratio has no particular meaning. Due to negative values  $D$  obtained for ash content in 1993, and values  $H_1$  and  $H_2$  for nicotine content in 1995, it was not possible to calculate the values for  $\sqrt{H_1/D}$  and  $Kd/Kr$ . According to the degree of dominance ( $\sqrt{H_1/D}$ ), over

I. Analysis of variance for nicotine, proteins, total nitrogen and ash content from 1992 to 1995

Year	Source of variation	d.f.	F values			
			nicotine	proteins	total nitrogen	ash
1992	crosses	9	3.94**	3.43**	1.15 n.s.	2.35*
	error	27				
	total	39				
1993	crosses	9	6.12**	2.49*	1.06 n.s.	1.26 n.s.
	error	27				
	total	39				
1994	crosses	9	8.26**	2.43*	4.99**	4.74**
	error	27				
	total	39				
1995	crosses	9	3.26**	3.06*	3.30**	3.16**
	error	27				
	total	39				

II. Means of parents and F<sub>1</sub> hybrids for nicotine, proteins, total nitrogen and ash content from 1992 to 1995

Trait	Year	Genotype										LSD	
		TN 86	BL1	Hy 71	Poseydon	Hy 71 x TN 86	Hy 71 x BL1	Hy 71 x Poseydon	Poseydon x BL1	Poseydon x TN 86	TN 86 x BL1	0.05	0.01
Nicotine (%)	1992	2.16	2.35	1.92	1.55	1.57	1.67	1.75	2.01	1.78	2.01	0.38	0.52
	1993	2.40	2.31	1.84	1.64	2.28	1.99	1.82	2.12	1.61	2.41	0.35	0.48
	1994	3.02	3.59	2.54	2.59	2.50	2.62	2.73	2.38	2.46	2.94	0.37	0.50
	1995	1.76	1.97	1.36	1.06	1.62	1.72	1.25	1.35	1.32	1.80	0.45	0.60
Proteins (%)	1992	10.64	11.03	12.67	11.83	10.54	10.79	11.69	10.64	10.18	9.98	1.27	1.71
	1993	8.32	8.31	9.26	8.35	8.10	8.03	8.77	8.32	8.18	7.69	0.78	n.s.
	1994	7.99	8.05	8.76	7.70	8.16	7.94	8.46	7.53	7.69	7.75	0.69	n.s.
	1995	7.25	7.64	7.55	8.51	7.17	7.12	7.75	7.70	7.25	7.42	0.68	n.s.
Total nitrogen (%)	1992	4.29	3.95	3.52	4.76	4.07	4.68	5.27	3.92	4.27	4.21	n.s.	n.s.
	1993	3.22	3.18	3.35	2.95	3.16	3.11	3.16	3.24	2.88	3.11	n.s.	n.s.
	1994	3.13	3.27	3.46	2.98	3.14	3.05	3.34	2.80	3.00	2.87	0.27	0.36
	1995	2.23	2.54	2.06	2.19	2.23	2.25	2.06	2.02	1.96	2.39	0.29	0.39
Ash (%)	1992	24.29	24.43	22.64	22.58	23.05	23.95	23.98	23.46	22.73	25.01	1.60	n.s.
	1993	25.56	25.57	25.07	24.79	25.39	24.96	24.20	25.68	25.45	25.25	n.s.	n.s.
	1994	24.99	23.17	23.19	22.62	23.01	23.70	22.90	24.33	23.59	25.20	1.19	1.60
	1995	34.62	34.41	38.43	32.32	33.94	32.38	33.41	32.12	36.04	33.30	3.17	4.29

dominance in inheritance was estimated for all traits in most investigation years, except in 1993 for nicotine content, and for proteins content in 1994 and 1995. In these years, the mentioned traits were inherited partly dominantly.

*Wr Vr* graphs for nicotine, proteins, total nitrogen and ash in all four investigation years are presented in Figs 1 to 4. The expected regression line for total nitrogen and ash cuts the *Wr* axis below the origin in all four years, which points to overdominance in inheritance of these traits. It may be said that nicotine and proteins are also inherited overdominantly, however only in two investigation years, while there was partial dominance in the remaining two years. Regression coefficient does not differ significantly from  $b = 1$  for any of the studied traits and hence interallelic interaction could not be evaluated. In all four investigation years, there were noticeable differences in carriers of a larger number of dominant or recessive genes. Thus, for example, for nicotine the largest number of dominant genes in three investigation years was estimated in genotype Poseydon, and the largest number of recessive genes in genotype BL1. In contrast to nicotine, genotypes TN 86 and BL1 had the largest number of dominant genes for proteins, and genotypes Hy 71 and Poseydon had the largest number of recessive genes equally in two investigation years. The largest number of dominant genes for total nitrogen was also estimated in genotypes TN 86 and BL1, though only in two years, while genotype Poseydon had the largest number of recessive genes. Finally, TN 86 was furthest from the origin for ash in two investigation years, thus having the largest number of recessive genes. Occurrence of

scatter diagram points in the third quadrant for all the studied traits points to the presence of overdominance.

## DISCUSSION

There is no doubt that tobacco plays an important role in the development of genetics and breeding of all cultivated plants. Due to the still insufficiently known genetic constitution of quantitative traits (most traits of interest to breeders), extensive research into the problem of their inheritance is continuing in the world, while tobacco, as the test crop, is still now subject of a great deal of fundamental and applied research.

In addition to the yield, tobacco quality is the most important goal of breeding, in which the quality of dried tobacco leaf should be regarded as a complex trait (Kozumplik et al., 1991). One of the criteria for tobacco quality is the chemical composition of the leaf. It is well known that each tobacco leaf contains a large number of organic and inorganic substances, and that uncountable new compounds are formed during smoking, which finally define the smoking characteristics of cigarettes. Chemical traits can be of a decisive importance for the evaluation of cigarettes, as well as of cultivars involved in their production. Chemical traits are changed under the influence of the intensity and constellation of ecological and vegetational factors, agrotechnical practices applied, and also the cultivars. Accordingly, as already said, chemical traits are polygenically controlled. Knowledge of the manner of inheritance of particular chemical traits is of vital impor-

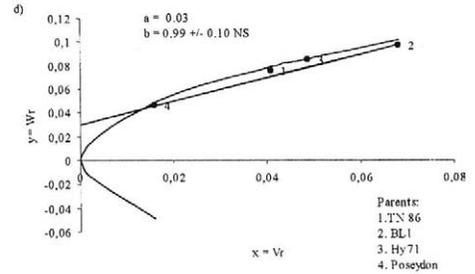
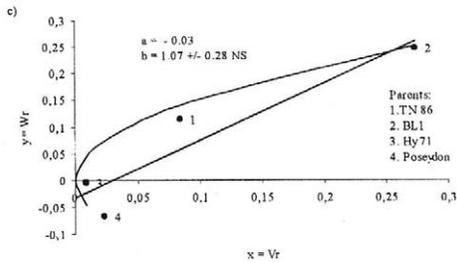
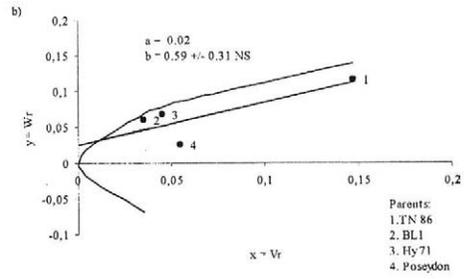
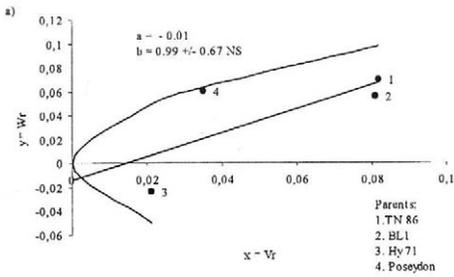
Year	Components of genetic variance	Trait			
		nicotine	proteins	total nitrogen	ash
1992	<i>D</i>	9.16+/-0.23*	0.40+/-4.28*	3.92+/-0.15 n.s.	0.70+/-0.22*
	<i>H</i> <sub>1</sub>	0.11+/-6.89 n.s.	0.63+/-0.12*	0.58+/-0.43 n.s.	0.99+/-0.65 n.s.
	<i>H</i> <sub>2</sub>	8.90+/-6.36 n.s.	0.70+/-0.11*	0.46+/-0.40 n.s.	0.99+/-0.60 n.s.
	<i>F</i>	4.70+/-6.09 n.s.	-0.37+/-0.11 n.s.	0.16+/-0.38 n.s.	5.64+/-0.57 n.s.
	<i>E</i>	2.50+/-1.06*	0.22+/-1.19*	0.24+/-6.75*	0.33+/-0.10*
	<i>H</i> <sub>2</sub> / <i>4H</i> <sub>1</sub>	0.20	0.28	0.20	0.25
	$\sqrt{H_1/D}$	1.11	1.24	3.84	1.19
	<i>Kd/Kr</i>	1.60	0.46	3.42	1.07
1993	<i>D</i>	0.11+/-0.24*	0.15+/-5.93*	7.77+/-6.98 n.s.	-7.68+/-7.59 n.s.
	<i>H</i> <sub>1</sub>	0.11+/-0.71 n.s.	0.28+/-0.17 n.s.	8.96+/-2.03 n.s.	0.25+/-0.22 n.s.
	<i>H</i> <sub>2</sub>	0.11+/-6.55 n.s.	0.21+/-0.15 n.s.	1.03+/-1.87 n.s.	0.26+/-0.20 n.s.
	<i>F</i>	-1.83+/-6.27 n.s.	2.76+/-0.15 n.s.	1.28+/-1.79 n.s.	-0.19+/-0.19 n.s.
	<i>E</i>	1.64+/-1.09 n.s.	6.89+/-0.26*	2.05+/-3.12*	0.15+/-3.39*
	<i>H</i> <sub>2</sub> / <i>4H</i> <sub>1</sub>	0.25	0.19	0.29	0.26
	$\sqrt{H_1/D}$	0.95	1.38	1.07	-
	<i>Kd/Kr</i>	0.85	1.15	1.17	-
1994	<i>D</i>	0.21+/-0.39*	0.13+/-2.46*	3.00+/-6.10*	0.88+/-0.14*
	<i>H</i> <sub>1</sub>	0.29+/-0.11*	1.48+/-7.17 n.s.	9.30+/-1.77*	1.79+/-0.42*
	<i>H</i> <sub>2</sub>	0.26+/-0.10*	2.45+/-6.21 n.s.	7.35+/-1.63*	1.21+/-0.39*
	<i>F</i>	0.16+/-0.10 n.s.	-7.72+/-6.33 n.s.	2.89+/-1.56 n.s.	0.53+/-0.37 n.s.
	<i>E</i>	2.00+/-1.75 n.s.	0.08+/-1.10*	1.08+/-2.73*	0.18+/-0.65*
	<i>H</i> <sub>2</sub> / <i>4H</i> <sub>1</sub>	0.22	0.41	0.20	0.17
	$\sqrt{H_1/D}$	1.16	0.33	1.77	1.42
	<i>Kd/Kr</i>	1.96	7.07	1.06	1.53
1995	<i>D</i>	0.13+/-2.45*	0.23+/-1.53*	3.16+/-0.01*	5.29+/-0.74*
	<i>H</i> <sub>1</sub>	-3.67+/-7.12 n.s.	0.11+/-4.46*	5.25+/-2.37*	11.55+/-2.15*
	<i>H</i> <sub>2</sub>	-3.21+/-6.58 n.s.	0.13+/-4.12*	3.90+/-0.21 n.s.	8.31+/-1.99*
	<i>F</i>	-2.40+/-6.30 n.s.	3.72+/-3.94 n.s.	9.81+/-2.10 n.s.	6.95+/-1.90*
	<i>E</i>	2.42+/-1.09*	5.63+/-6.87*	9.86+/-3.65*	1.17+/-0.33*
	<i>H</i> <sub>2</sub> / <i>4H</i> <sub>1</sub>	0.22	0.30	0.19	0.18
	$\sqrt{H_1/D}$	-	0.67	1.29	1.48
	<i>Kd/Kr</i>	-	1.27	1.27	2.59

tance for the success of the breeding procedure. Extensive application of dialleles in various forms provides a valuable information for evaluation of heritability and the manner of inheritance, participation and action of genes, additive and nonadditive components, as well as distribution of dominant and recessive alleles in parents. One of the most suitable models is that after Hayman (1954), Jinks (1954) and Mather, Jinks (1971), which was also applied in this study.

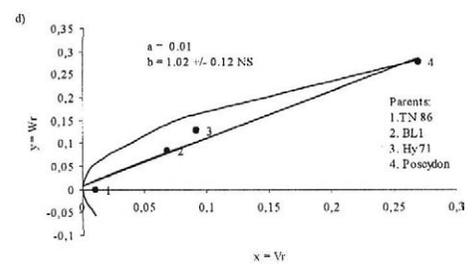
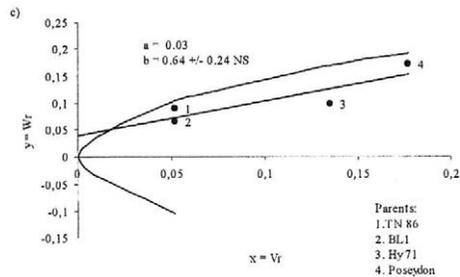
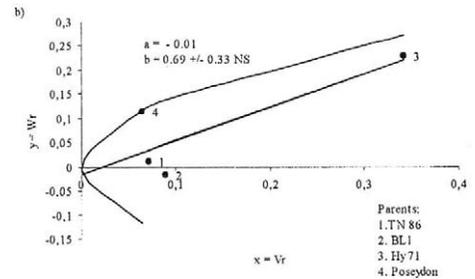
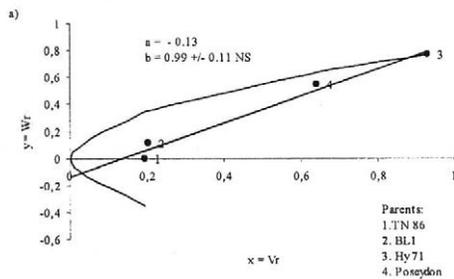
Starting from nicotine, a chemical compound from the group of alkaloids, which characterizes tobacco as a plant, according to the so far studies of the components of genetic variance significant additive and nonadditive variances (Povilaitis, 1966) were estimated, or only significant additive variance (Matzinger, Wersman, 1971; Chiang, Chen, 1985). In my own in-

vestigations, according to the components of genetic variance, nicotine is also inherited additively. The graphic method indicates, in dependence on the year, overdominant inheritance of nicotine, but also partial dominance. Pandeya et al. (1984) also estimated, using the graphic method, partial dominance in inheritance of nicotine. No interalle interaction is present, which is in agreement with my own results, however genetic divergence of parent genotypes is noticeable.

One of the main reasons for including proteins into these investigations was that the manner of their inheritance has been insufficiently studied. According to the scarce investigations of the manner of inheritance of proteins to date, only significant dominant variance has been estimated (Beljo, 1980). According to my own research into the components of genetic variance, pro-



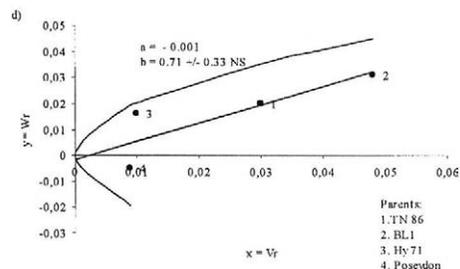
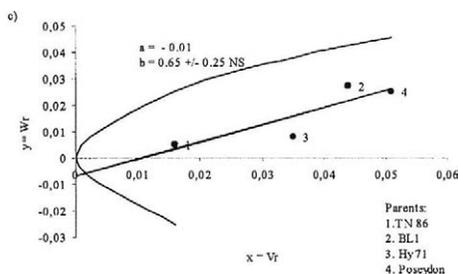
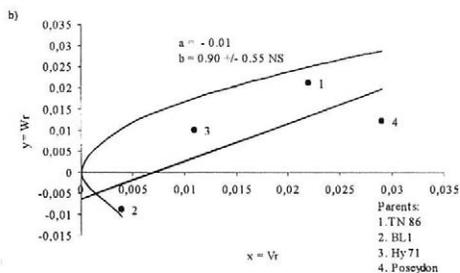
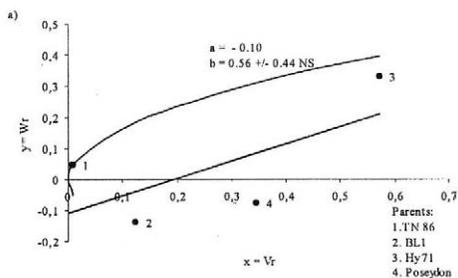
1.  $W_r V_r$  plots for nicotine content (%) in 1992 (a), 1993 (b), 1994 (c) and 1995 (d)



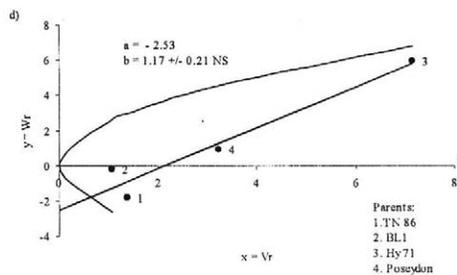
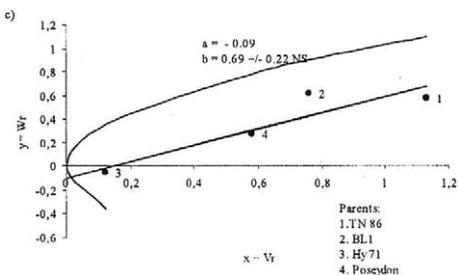
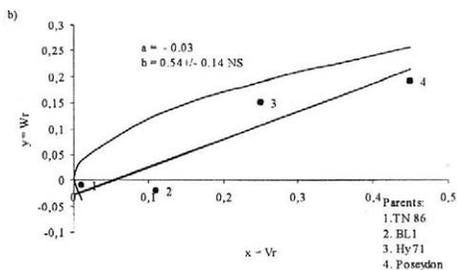
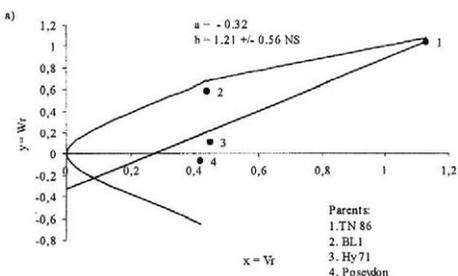
2.  $W_r V_r$  plots for protein content (%) in 1992 (a), 1993 (b), 1994 (c) and 1995 (d)

teins were inherited additively in two years and additively and nonadditively also in two years. This was confirmed by the graphic method in two years of four years. It follows that protein inheritance is more influenced by additive variance, however the role of nonadditive variance cannot be fully excluded either.

Pandeya et al. (1984) studied the inheritance of total nitrogen using the graphic method. According to their results, this chemical compound is inherited partly dominantly and there is no interallelic interaction. In my own investigations, however, overdominance was estimated by the same method in all four investigation



3.  $W_r V_r$  plots for total nitrogen content (%) in 1992 (a), 1993 (b), 1994 (c) and 1995 (d)



4.  $W_r V_r$  plots for ash content (%) in 1992 (a), 1993 (b), 1994 (c) and 1995 (d)

years. Preponderance of nonadditive variance over additive variance was confirmed also by studies of the components of genetic variance, but the role of additive variance is not negligible either.

According to my results into the components of genetic variance, ash inheritance is more influenced by additive variance, which was fully confirmed by the results of regression analysis by Pandeya et al. (1984).

In contrast to their studies, my results estimated overdominance in ash inheritance by regression analysis.

To conclude, research done by other authors as well as my own investigations into the manner of inheritance of selected chemical traits indicate that the additive/nonadditive variance ratio may vary in dependence on the genetic material, size of the trial plot, investigation conditions, year, location and the method applied. When the results achieved in my own research are summed up, they allow the conclusion that a higher value of additive variance was estimated according to the components of genetic variance. But, this is not confirmed by the results obtained by the graphic method.

## REFERENCES

- Beljo J. (1980): Prilog proučavanju nasljeđivanja kemijskih svojstava duhana. *Tutun/Tobacco*, 11-12: 5-14.
- Beljo J. (1984): Proučavanje heterozisa i nasljednosti važnih komponenata prinosa i kvaliteta duhana (*Nicotiana tabacum*) u F<sub>1</sub> i F<sub>2</sub> generaciji. [Dissertation.]
- Beljo J. (1992): Oplemenjivanje duhana-dostignuća i izazovi. *Tutun/Tobacco*, 7-12: 179-190.
- Chang E. Y., Shyu C. C. (1980): The separation and relative importance of gene effects for seven agronomic characters and three chemical constituents in tobacco. I. Vamfen-Hicks x Burley 21. *Bull. Taiwan Tob. Res. Inst.*, 13: 19-31.
- Chiang T. H., Chen S. Y. (1985): Genetic study and development of low alkaloid tobacco varieties. *Tob. Ab.*: 235-236.
- Devčić K., Bolsunov I. (1975): Čulinec, nova sorta burleya stvorena u Duhanskom institutu Zagreb. 9. Simp. duhanskih stručnjaka Jugoslavije: 10-12.
- Devčić K., Triplat J., Benković F. (1984): Sorte burleya selekcionirane u Duhanskom institutu Zagreb. *Tutun/Tobacco*, 1-2: 1-10.
- Hayman B. I. (1954): The theory and analysis of diallel crosses. *Genetics*, 39: 789-809.
- Jinks J. L. (1954): The analysis of continuous variation in a diallel cross of *Nicotiana rustica* varieties. *Genetics*, 39: 767-788.
- Kozumplik V., Devčić K., Vuletić N. (1991): Suvremeni ciljevi i metode oplemenjivanja duhana tipa virginia i burley. *Poljopr. Znan. Smotr.*, 56: 169-180.
- Legg P. D., Collins G. B. (1971): Inheritance of percent total alkaloids in *Nicotiana tabacum* L. II. Genetic effects of two loci in Burley 21 x LA Burley 21 populations. *Can. J. Genet. Cytol.*, 13: 287-291.
- Legg P. D., Chaplin J. F., Collins G. B. (1969): Inheritance of percent total alkaloids in *Nicotiana tabacum* L. *J. Hered.*, 60: 213-217.
- Mather K., Jinks J. L. (eds) (1971): *Biometrical genetics*. London, Chapman and Hall.
- Matzinger D. F., Wersman E. A. (1971): Inheritance and relationships among plant characters and smoke constituents in flue-cured tobacco. *Int. Tob. Sci. Congr.*: 68-76.
- Miller R. D. (1987): Registration of TN 86 burley tobacco. *Crop. Sci.*, 27: 365-366.
- Pandeya R. S., Dirks V. A., Poushinsky G., Zilkey B. F. (1984): Quantitative genetic studies in flue-cured tobacco (*Nicotiana tabacum* L.). II. Certain physical and chemical characters. *Can. J. Genet. Cytol.*, 27: 92-100.
- Povilaitis B. (1966): Diallel cross analysis of quantitative characters in tobacco. *Can. J. Genet. Cytol.*, 8: 336-346.
- Vasilj Đ. (1973): Komparativna istraživanja metoda procjene heritabiliteta nekih svojstava klipa *Zea mays* L. *Arh. Poljopr. Nauke*, 95: 68-96.
- AOAC (1984): *Official methods of analysis*. Arlington, Va.
- Coresta (1969): Standard method No. 20. Determination of alkaloids in manufactured tobacco.

Received on March 17, 1998

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## BIOORGANICKÁ CHEMIE

K. Waiser

*Praha, Karlova univerzita 1995. 263 s.*

Náš knižní trh v oblasti organické chemie a biochemie po delší přestávce obohacuje knižní titul *Bioorganická chemie*. Publikace se instruktivně zabývá studiem organických látek tvořených v živých organismech, jejich strukturou, zejména topografickou architekturou biomakromolekul, syntézou biologicky účinných látek, enzymovou katalýzou *in vitro*, modelováním chemických procesů probíhajících v přírodě a uměle vytvářenými chemickými systémy, které tyto procesy simulují.

V současné době je někdy obtížné nalézt hranici mezi bioorganickou chemií a biochemií, to však nemění nic na skutečnosti, že bioorganická chemie se postupně vykrystalizovala v samostatnou vědní disciplínu, které se věnuje stále větší počet specialistů ve světě i u nás. Toto relativně mladé odvětví chemie má již své vlastní zázemí v podobě několika specializovaných časopisů, ale i v řadě vědeckých setkání, která netrpí nedostatkem odborných příspěvků. Jako obor je tato disciplína

rozvíjena na řadě pracovišť a je jistě potěšitelné, že i u nás má už svou tradici (prof. Vodrážka, doc. Krechl, prof. Večeřa). Dosud zde však zcela chyběla učebnice bioorganické chemie, která by mohla uspokojit všechny zájemce o tento obor.

Tuto mezeru na knižním trhu vyplňuje recenzovaná kniha. Poskytuje solidní přehled o bioorganické chemii a jí používaných metodách práce a přináší takové množství přehledně uspořádaných údajů o přírodních organických látkách, že jistě nalezne uplatnění u řady chemicky a biologicky orientovaných pracovníků. Věcný rejstřík, kterým je kniha vybavena, obsahuje kolem 550 hesel a umožňuje rychlé vyhledání potřebné informace. Zájemci o hlubší studium mohou čerpat ze seznamu asi 50 knižních monografií. Vezmeme-li v úvahu, že v publikaci jsou popsány i četné mechanismy biologického účinku řady látek, je zřejmé, že jde o užitečnou příručku.

*Doc. RNDr. Ing. Josef Zahradníček, CSc.*

# CZECH HOP CHARACTERIZATION USING RAPD METHOD AND GENETIC DISTANCE ANALYSIS OF SELECTED GENOTYPES

## CHARAKTERIZACE ČESKÝCH ODRŮD CHMELE POMOCÍ RAPD METODY A GENETICKÁ ANALÝZA VYBRANÝCH GENOTYPŮ

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**ABSTRACT:** DNA isolated from 32 hop (*Humulus lupulus* L.) cultivars from a world collection in reactions with one hundred primers were analyzed. Besides RAPD primers also nine microsatellite-based primers were tested, one primer derived from a M13 phage specific sequence and two STS primers. The low level of hop DNA polymorphism between cultivars using PCR based techniques were confirmed. Only five random primers and two microsatellite-based primers producing polymorphic bands were identified. OPA-11, OPV-17, OPC-08, OPC-09 RAPD primers were used for distinction of the Czech hop cultivar Osvald's clone 72 and Premiant, Bor and Sládek hybrids from recent Czech hop breeding program. In evaluation of some methodological aspects of the RAPD technique utilization for the genetic distance analysis, the effect of magnitude of the initial data set on the results of the analysis and also differences in results owing to a software used were confirmed. The results of PCR based techniques for the complete analysis of genetic distance of hop cultivars were used. The complete genetic distance analysis is suitable to determine the relationship between hop cultivars in respect to the origin in breeding program as well as to some main chemical genotype features.

**Keywords:** hop; *Humulus lupulus* L.; varieties; DNA fingerprinting; RAPD; PCR; STS; microsatellite sequences; genetic distance analysis

**ABSTRAKT:** Analýza DNA izolované ze 32 odrůd světového sortimentu chmele (*Humulus lupulus* L.) proběhla v reakcích se sto primery. Kromě RAPD primerů bylo testováno též devět mikrosatelitně založených primerů, jeden primer odvozený od specifické sekvence fága M13 a dvě kombinace STS primerů. PCR analýzy potvrdily nízkou míru polymorfismu DNA odrůd chmele. Bylo identifikováno pouze pět RAPD primerů a dva mikrosatelitně založené primery produkující polymorfni fragmenty. RAPD primery OPA-11, OPV-17, OPC-08, OPC-09 byly použity pro odlišení české odrůdy chmele Osvaldův klon 72 a hybridů Premiant, Bor a Sládek z nového českého šlechtitelského programu. Hodnocením některých metodologických aspektů RAPD metody, užití ke genetické analýze, byl potvrzen vliv souboru vstupních dat na výsledky analýzy i rozdílnost výsledků při použití různých softwarových programů. Výsledky PCR analýz byly použity k celkové genetické analýze odrůd chmele, pomoci níž lze stanovit příbuznost mezi jednotlivými genotypy, s ohledem na původ i na některé hlavní chemické genotypové charakteristiky.

**Klíčová slova:** chmel; *Humulus lupulus* L.; odrůdy; DNA fingerprinting; RAPD; PCR; STS; mikrosatelitní sekvence; genetická analýza

### INTRODUCTION

Hop (*Humulus lupulus* L.) is a crop cultivated for its important role in the brewing industry. There are about 250 hop cultivars worldwide. The hop industry is a very important part of agriculture in the Czech Republic. Czech hop is one of the most valued hops in the world market. Hop cultivars originated during positive

selection from the ancestral population, and were distributed from the place of origin. The more recent hybrid cultivars have been bred by crossing. The hop plants are propagated vegetatively and the genetic stability of cultivars is therefore guaranteed. The distinction of hop cultivars is based on the content and composition of volatile compounds (e.g. Kralj, Zupanec, 1991; Peacock, McCarty, 1992) and phenotype evaluation.