ACCLIMATIZATION OF BURLEY TOBACCO GERMPLASM UNDER AGRO-ECOLOGICAL CONDITIONS OF SWAT VALLEY

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ABSTRACT

This study was performed to identify desirable Brazilian Burley tobacco (Nicotiana tabacum) genotypes for commercial cultivation under agro-ecological conditions of Swat Valley of North West Frontier Province (NWFP), Pakistan. Four Brazilian Burley tobacco genotypes; Bag-16101, Bag-161128, CSC-206, and CSC-227 along with adapted Burley-64 as check cultivar were planted in randomized block design at the Pakistan Tobacco Company Research Station, Khawza Khela, Swat during 2003-04 growing season. Data were recorded on yield and some other important plant parameters. Highly significant differences ($p<0.01$) among the tobacco genotypes were observed for plant height, cured leaf yield, grade index and reducing sugar levels whereas differences among the genotypes were only of significant nature ($p<0.05$) for leaf area, number of leaves plant^{-1} and nicotine content. The genotype, Bag-16101, yielded the maximum number of leaves plant^{-1} and produced the maximum leaf yield as well. It also manifested resistance against the blank shank, brown spot diseases and viruses. Among all the introductions, CSC-206 and CSC-227 produced the best quality tobacco with grade indices of 70.8 and 68.5, respectively. Burley-64, the commercial cultivar used in the present study, however, produced the shortest plants with the least number of leaves plant^{-1}. This genotype displayed minimum nicotine content, the lowest yield and grade index and was the most susceptible genotype against blank shank disease and viruses. Bag-16101, CSC-206 and CSC-227, thus, displayed immense potential to replace Burley-64 in Swat valley of North West Frontier Province (NWFP), Pakistan. Moreover, the genetic potential of these genotypes can be exploited in future tobacco breeding programmes.

Key Words: Acclimatization, Burley tobacco, Disease/Pest resistance, Yield and quality traits

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INTRODUCTION

Burley tobacco, *Nicotiana tabacum* L., belongs to the family *Solanaceae* and genus *Nicotiana*. Only two species of the genus *Nicotiana* viz. *N. tabacum* L. and *N. rustica* L. are grown widely all over the world. The former is processed for manufacturing of cigarettes, cigars and bidis,whereas the later is utilized for snuff, local hukka and chewing purposes. Some other species of this genus are utilized for ornamental purposes (Taj, 1994).

Tobacco industry in Pakistan makes a significant contribution in different sectors of the economy; from farming through manufacturing and to retailing the end product. The industry is also a major purchaser of supplies from other industries. In Pakistan it is cultivated on an area of 56.4 thousand hectares with production of 112.6 thousand tones. Tobacco is an important cash crop of North West Frontier Province (NWFP) where the agro climatic conditions are highly suitable for its cultivation. In NWFP it is planted on area of 36.5 thousand hectares with production of 87.9 thousand hectares (MINFAL, 2006). Next to sugarcane and sugar beet, tobacco is the major source of income for farmers in Peshawar valley. Peshawar, Mardan, Hazara and some parts of Malakand Agency are among the largest tobacco producing areas.

Burley is an air-cured tobacco and is produced in Swat Valley of North West Frontier Province of Pakistan. It requires 80 % humidity and a temperature of less than 40 ^{0}C for good quality leaf. When compared with flue-cured Virginia (FCV), it is less costly and more bulky and is used as cheap filler. Burley is semi-flavored tobacco and is considered as building block of future blends for consumer's (PTC, 2003).

Continuous increase in cost of production of tobacco crop demands the development of high yielding tobacco genotypes with desirable morphological and chemical attributes. To improve the yield of tobacco, it is vital to know the important plant traits such as plant height, total number of leaves plant^{-1}, cured leaf yield and total yield of the different genotypes to be used in a particular study. Further, tobacco quality plays an important role in marketing of the tobacco as tobacco leaf is marketed by the physical characteristics like body, color, texture and aroma (Woras, 1996). Various biochemical traits like nicotine content and reducing sugars affect the quality of tobacco. Genetic variability among burley tobacco genotypes for yield and quality traits has previously been reported many researchers (Burton *et al.*, 1994; Pamukov and Mutafchiefa, 1998; Lukipudis *et al.*, 1998; Carotenuto *et al.*, 1999; Butorac, 2000). The present study was, therefore, conducted to identify desirable burley genotypes for yield and quality traits for commercial cultivation.

MATERIALS AND METHODS

This research was conducted at the Pakistan Tobacco Company Research Station, Khawazakhela, Swat during 2003-2004 tobacco crop growing season. Germplasm of four different Burley Tobacco genotypes (Bag-16101, CSC-206, CSC-227 and Bag-16128) of Brazil origin along with one adapted check (Burley-64) were used. First, nursery was raised and seedbeds of 12 m each were made in an East West direction for better exposure of the crop to sunlight. Seedbeds were top dressed with 3.5 cm of well-decomposed and rotten farm yard manure. Germplasm were sown with a seed rate of 1.3g/bed. The beds were covered with plastic sheets till germination. After emergence, plastic sheets were removed in sunny days and replaced at night. Healthy seedlings were transplanted during the 4th week of March, 2004. The experiment was laid out in randomized complete block design with four replications. Each genotype was planted in a 14 row plot with plant to plant and row to row distance of 60 and 90 cm, respectively. Basal dose of 50 N: 60 P_2O_5: 80 K_2O kg ha^{-1} was applied in the form of compound fertilizer i.e. NPK (12:15:20). Standard cultural practices and insect control measures were followed from transplantation till picking and curing of leaves.

Plant height was measured from ground level to the tip of upper most leaf in each treatment at the time of physiological maturity. Number of leaves plant^{-1} was recorded by counting the number of leaves from bottom to the top of ma in stalk of each plant. For leaf area, length and breadth of 5th, 10th and 15th leaf in each treatment was measured and leaf size was calculated by using the following formula (Idrees and Khan, 2001):

Leaf Area (cm^2) = Average leaf length (cm) × Average leaf breadth (cm) × 0.634

Where 0.634 is Correction Factor

Total weight of cured leaves in each treatment was determined after each picking and summed after four pickings. Yield ha^{-1} for each treatment was obtained as under (Idrees and Khan, 2001):

$$\text{Cured leaf yield (kg ha}^{-1}\text{)} = \frac{\text{Total cured weight (kg)}}{\text{Net area harvested}} \times 10000\,\text{m}^2$$

Grade index is based on the percentage of Mature and Ripe grades. Colors of cured leaves of each treatment in all pickings were carefully observed and graded as Mature and Ripe grades. Grade index (%) was calculated by the following formula (Idrees and Khan, 2001):

$$\text{Grade index (\%)} = \frac{\text{Weight (kg) of upper grade cured leaves in a treatment}}{\text{Total weight (kg) of cured leaves in a treatment}} \times 100$$

Samples were analyzed for nicotine percentage in the Chemistry Section of Pakistan Tobacco Company, Akora Khattak Factory (AKF). Nicotine content was calculated by the following formula as used by Idrees and Khan (2001):

$$\text{Nicotine content (\%)} = \frac{V_1 \times N \times 32.45}{\text{Weight of the sample}}$$

V_1 = Volume of titrant for non-alcholic aliquot
N = Normality of perchloric acid

Reducing sugars were analyzed using the procedure suggested by Lane and Eynon (1986) with the help of following formula:

$$\text{Reducing sugars (\%)} = \frac{25 \times 100 \times 0.05}{\text{Amount of titrate} \times \text{Weight of the Sample}}$$

All the genotypes used in the study were visually observed for the presence or absence of symptoms of blank shank disease, viruses, nematodes and brown spot disease. The presence or absence of diseases was also confirmed at Crop Diseases Research Program Laboratories of National Agricultural Research Centre, Islamabad. Disease/pest percentage was determined using the following formula:

$$\text{Disease/Pest Percentage} = \frac{\text{Number of affected plants in a treatment}}{\text{Number of plants in a treatment}} \times 100$$

The data after compiling was statistically analyzed using MSTATC package version 1.2 (Freed, 1990) and least significant difference (LSD) test was applied to test the significance of genotypic mean differences.

RESULTS AND DISCUSSION

Plant Height, Leaf Area and Number of Leaves Plant $^{-1}$

Plant height is directly related to the number of leaves that are borne on a tobacco plant; hence this character may be used as an indicator for the potential number of leaves (Ali *et al.*, 1984). Analysis of variance revealed highly significant differences ($p<0.01$) among the genotypes for plant height (Table I). Plant height among the genotypes ranged between 101 and 121 cm. Bag-16101 displayed maximum plant height (121 cm) followed by CSC-206 (114 cm), Bag-16128 (113 cm) and CSC-227 (111 cm). Check cultivar Burley-64, however, attained minimum plant height of 101 cm (Table II). Bag-16101 thus displayed its superiority for this plant trait. These results are compatible with the findings of Hashmi *et al.* (1985) and Butorac *et al.* (1999). They also observed significant mean differences among tobacco genotypes for plant height.

Leaf area and number of leaves plant^{-1} are the major yield components of a tobacco genotype (Woras *et al.*, 1989). Usually high prices are offered for long and broad leaves. Significant differences ($p<0.05$) among the genotypes were observed for leaf area. This trait ranged among the genotypes between 662 and 728 cm^2. Burley-64 displayed maximum leaf area (728 cm^2) followed by CSC-227 (711 cm^2) while Bag-16101 manifested the minimum leaf area (662 cm^2) (Table II). Significant genetic differences among the tobacco genotypes for leaf area as observed in the present study is compatible with the findings of Butorac (1998) and Butorac *et al.* (2000). Mean squares also revealed significant differences ($p<0.05$) among the genotypes for number of leaves plant^{-1} (Table I). Average number of leaves plant^{-1} among the genotypes varied from 23 to 28. Bag-16101 had the highest number of leaves (28) followed by Bag-16128 (26). Burley-64, however, produced the lowest number of leaves (23) (Table II). Bag-16101 and Bag-16128 thus excelled in performance for number of leaves plant^{-1}. These results are in agreement with the findings of Butorac *et al.* (1999) and Liu *et al.* (1999). These researchers also reported significant genetic variation among tobacco genotypes for number of leaves.

Cured Leaf Yield and Grade Index

Cured leaf yield is one of the most important plant characteristics in tobacco crop as it is directly related to farmer's profit. It is the final product of tobacco crop after passing the green leaves through different curing procedures (Woras, 1993). Highly significant differences ($p<0.01$) among the genotypes were observed for cured leaf yield hectare^{-1} (Table I). Butorac (1994), Subbian *et al.* (1994), Dimitrova (1998), Hanoomanjee *et al.* (1998) and Liu *et al.* (1999) also reported significant genetic differences among the tobacco genotypes for cured leaf yield. The value for this trait among the genotypes ranged between 2044 kg and 2836 kg ha^{-1}. Genotype Bag-16101 produced maximum yield (2836 kg ha^{-1}) while the check cultivar Burley-64 had minimum cured leaf yield (2044 kg ha^{-1}) (Table II). Tobacco genotype Bag-16101 thus exhibited superiority for this important plant characteristic.

Tobacco leaf is marketed by its physical characteristics like body, color, texture, size and aroma etc. which when grouped together, represent grade index (Woras, 1996). Statistical analysis manifested highly significant differences ($p<0.01$) among the genotypes for percent grade index (Table I). CSC-206 and CSC-206 were topping the list of tobacco genotypes used in the study with the grade indices values of 70.8 and 68.5%, respectively while Burley-64 displayed the lowest grade index value of 59.6% (Table II). These results are supported by the findings of Triplat *et al.* (1994), Rao (1998), and Spirov and Lukipudis (1999) who also reported significant mean differences among tobacco genotypes for this important commercial plant trait.

Table I. ***Mean squares for plant height (PH), number of leaves plant^{-1} (LPP), leaf area (LA), yield ha^{-1}(YLD), grade index (GI), nicotine content (NIC) and reducing sugars (RS) of Burley tobacco genotypes during 2003-04 at Swat***

SOV	df	PH	LPP	LA	YLD	GI	NIC	RS
Replications	3	36.9	0.05	421	389	17.9	0.07	0.05
Genotypes	4	197**	9.3*	2790*	332391**	99.1*	0.15*	0.35**
Error	12	20.6	2.2	718	27689	25.1	0.05	0.02

*= Significant at 5% level of probability
**= Significant at 1% level of probability

Table II. ***Mean values for plant height (PH), number of leaves plant^{-1} (LPP), leaf area (LA), yield ha^{-1} (YLD), grade index (GI), nicotine content (NIC), reducing sugars (RS) and diseases/pest percentage of Burley tobacco genotypes during 2003-04 at Swat***

Genotypes	PH (cm)	LPP	LA (cm^2)	YLD (kg)	GI (%)	NIC (%)	RS (%)	Disease/Pest (%)			
								Blank Shank	Brown Spot	Viral	Nematode
Bag-16101	121	28	662	2836	60.1	2.3	1.1	4	15	11	0
Bag-16128	113	26	702	2501	63.9	2.1	1.0	12	24	18	0
CSC-206	114	25	676	2590	70.8	2.5	1.3	18	54	21	0
CSC-227	111	25	711	2425	68.5	2.3	1.8	17	43	27	0
Burley-64	101	23	728	2044	59.6	2.0	1.2	18	31	35	0

Chemical Characteristics

Nicotine content and reducing sugars are the important chemical characteristics of a particular tobacco genotype determining its quality. Hence a thorough assessment of these traits is imperative while studying tobacco genotypes of differential genetic backgrounds.

Nicotine is the principal alkaloid in tobacco defining tobacco quality (Shmuk and Nauk, 1953). Higher contents of nicotine negatively affect different physiological functions of the smoker while very low contents offer no satisfaction to the smoker (Hashmi *et al.*, 1990). The genotypes exhibited significant differences ($p<0.05$) for the nicotine content of their leaves (Table I). Nicotine content among the genotypes varied from 2 to 2.5%. CSC-206 had the leaves of maximum nicotine content (2.5%) whereas, Burley-64 leaves showed minimum nicotine (2.0%) in its leaves (Table II). These results are in agreement with the findings of Triplat *et al.* (1994), Pathak *et al.* (1996) and Liu *et al.* (1999). They also observed significant differences among tobacco genotypes for nicotine content. Nicotine content was low in all the genotypes used in the present study.

Reducing sugars exercise the most favorable influence on the tobacco taste and aroma during smoking and are one of the most important quality parameters of tobacco crop. Its higher contents impart sweetness to aroma (Hashmi *et al.*, 1990). Highly significant differences ($p<0.01$) among the genotypes were observed for reducing sugar percentage in the leaves (Table I). Reducing sugar contents in the leaves of tobacco genotypes ranged between 1 and 1.8 (Table II). Bag-16128 displayed minimum value (1) for this trait, whereas Burley-64 had maximum value (1.8) for reducing sugar content. Of all the genotypes used in the study, Burley-64 thus manifested its superiority for reducing sugars content. These results are compatible with the findings of Triplat *et al.* (1994) and Pathak *et al.* (1996). They also reported significant genetic differences among tobacco genotypes for this trait.

Disease Resistance

The genotypes showed differential response to various diseases and other pests like blank shank, brown spot, viruses and nematodes. Blank shank affected 4, 12, 18 and 17% of total plants of Bag-16101, Bag-16128, CSC-206 and CSC-227, respectively. Brown spot had impact on 15, 24, 54 and 43% of total plants of Bag-16101, Bag-16128, CSC-206 and CSC-227, respectively. Viral diseases manifested its effect on 11, 18, 21 and 27% of total plants of Bag-16101, Bag-16128, CSC-206 and CSC-227, respectively. Bag-16101 thus appeared comparatively more resistant against all the blank shank, brown spot and viruses. All the genotypes used in the study, however, displayed resistance against the nematodes. The adapted control genotype Burley-64 was observed susceptible to blank shank (18%), brown spot (31%) and viruses (35%). Genetic differences among tobacco genotypes for disease/pest resistance have also been previously reported by Burton *et al.* (1994), Triplat *et al.* (1994), Hanoomanjee *et al.* (1998). The differences in the above mentioned morphological traits, yield potential, chemical and disease/pest resistance characteristics of the tobacco genotypes used in the present study could be attributed to their genetic constitution.

CONCLUSION

In the present study, four Brazilian Burley tobacco genotypes i.e. Bag-16101, Bag-161128, CSC-206, and CSC-227 along with adapted Burley-64 as check cultivar were evaluated for yield and quality parameters of tobacco crop. Data were recorded on plant height, number of leaves plant^{-1}, leaf area, yield, grade index, nicotine content, reducing sugar and resistance to various diseases and pests. Genotypes revealed sufficient amount of genetic variation for all these studied traits. Bag-16101 produced maximum number of leaves plant^{-1} with the highest cured leaf yield. It also had desirable levels of resistance against various diseases and pests. Exotic tobacco genotypes CSC-206 and CSC-227 displayed superiority for leaf area, grade index and nicotine content. On the other hand, commercial check Burley-64 used in the present study had the lowest values for number of leaves plant^{-1}, grade index and cured leaf yield. It also exhibited susceptibility to blank shank, brown spot and viral diseases. On the basis of the observations recorded during the present study it is apparent that the genotypes Bag-16101, CSC-206 and CSC-227 proved superior for most of desired yield and quality attributes of tobacco crop. These genotypes are, therefore, recommended for commercial cultivation in Swat valley of North West Frontier Province (NWFP), Pakistan and for utilization subsequent future tobacco breeding pro

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