**UDC633.71** **ISSN 0495-3244**

# ТУТУН

# TOBACCO


| Vol.67 | Nº 7-12 |
|---|---|


## BULLETIN OF TOBACCO SCIENCE AND PROFESION


| TUTUN TOBACCO | VOL.67 | Nº **7-12** | pp. 01-81 | PRILEP R.MACEDONIA | JULY DECEMBER | 2017 |
|---|---|---|---|---|---|---|

Тутун/Tobacco Vol. 67 No 7-12, 3-10, 2017

I ISSN 0494-3244
UDC:633.71-152.61(497.7)"2008/2016"
Original scientific paper

# VARIETY STRUCTURE AS AN ESSENTIAL FACTOR FOR SUSTAINABLE DEVELOPMENT OF THE PRODUCTION OF ORIENTAL TOBACCO IN REPUBLIC OF MACEDONIA AND MARKETING OF TOBACCO PRODUCTION COMPETITIVE IN FOREGIN MARKETS

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## ABSTRACT

In the last 10 years in Macedonia are produced only oriental types of tobacco which achieved an average annual production of unprocessed raw tobacco amounting to around 25 000 tonnes, with the exception of two years (2008 and 2015). This production is competitive and compatible with the real needs of the market. Accordingly, in this ten year period total production by year shows that the Republic of Macedonia has continued sustainable development of tobacco production and the variety structure is extremely heterogeneous, especially in type prilep and there are no direct payments (subsidies).

The analysis of the parameters it was found that the introduction of subsidies by the government 1€ per kg of tobacco and improving the variety structure as a major factor in the introduction of the variety Prilep 66-9/7 in the primary tobacco production in this period is decisive for the increase in production and achieving sustainable development in the last 10 years. The variety Prilep 66-9 / 7 in the last two years has represented an average of 88% in total production and over 97% in the production of the type prilep. This variety is not only achieve sustainable development of tobacco production, significantly increased the competitiveness of tobacco production in the market and realize a much higher export price for 1kg processed tobacco.

**Key words :** tobacco, varieties, prilep, yaka,basma, production

## СОРТНАТА СТРУКТУРА КАКО ВАЖЕН ФАКТОР ЗА ОДРЖЛИВ РАЗВОЈ НА ПРОИЗВОДСТВОТО НА ОРИЕНТАЛНИОТ ТУТУН ВО РЕПУБЛИКА МАКЕДОНИЈА И МАРКЕТИНГ НА ПРОИЗВОДСТВОТО НА ТУТУН КОНКУРЕНТЕН НА НАДВОРЕШНИТЕ ПАЗАРИ

Во последните десет години во Република Македонија се произведуваат само ориенталски типови на тутун и е остварено годишно просечно производство од 25. 000 тони со исклучок на две години ( 2008 и 2015 год. ) . Ова производство е е реално спрема потребите на странскиот пазар. Имено, десетгодишното производството на тутун во Република Македонија покажува дека производството на тутун е стабилно , структурата на сортите е хетерогена , особено кај типот прилеп но нема исплата на субвенции во овој период. Анализираќи ги податоците , констатирано е дека воведувањето на субвенциите од страна на владата , едно евро за килограм откупен тутун, подобрувањето на сортната структура како основен фактор , и воведување на сортата Прилеп 66-9/7 во примарното производство , претставуваат главен фактор за зголемување на вкупното производство на тутун во последните десет години . Сортата Прилеп 66-9/7 во последните две години е застапена со 88/% во вкупното производство и преку 97% во производството на тутун од типот прилеп. Оваа сорта значајно ја подобри конкурентноста на тутунот за извоз при што се достигна повисока просечна цена по килограм обработен тутун.

**Клучни зборови:** тутун, сорти,прилеп, јака, басма, производство

## INTRODUCTION

Tobacco has conquered the world not only for the pleasure it gives during smoking but also for the high revenues it brings to the countries in which it is grown. Tobacco was introduced in Macedonia from Turkey in 1638 and ever since it has been highly valued for its economic and social importance. According to Turkish statistical data, total tobacco production in R. Macedonia before the Balkan wars was 4-5 million kg. The production of this crop start increasing after the Second World War, to reach 36.221 tonnes in 1982.

Tobacco production takes a very important place in the economy of Macedonia due both for economic and social reasons. Tobacco cultivars grown in R. Macedonia are of oriental origin, mainly of the types Prilep, Jaka and Basma. With a share of 3%, R. Macedonia is positioned among the eight major tobacco producing countries in the world. Of the total arable land in the country tobacco occupies 3,4% in the area under energy crops around 81.1 % of the total area. The average tobacco planted area in Macedonia in the period 2008-2016 was 16025 ha. With an average yield of 1.437 kg/ha, the total production for the same period was 23 068 tons of oriental tobacco, but it is presumed that it can achieve up to 35.000 tonnes. Tobacco share in the total exports of R. Macedonia was 3.7% (Statistics Annual of RM for 2011). At first sight that seems to be an insignificant segment in the foreign trade relations of the country, but its real value can be figured out when we take into consideration that the total exports in 2015 was 24 893 tons of oriental tobacco. The importance of tobacco for Macedonian economy is even more obvious when compared to the total exports of agricultural products in 2015 , in which it participated with over 22% . All activities and processes related to production of this crop are regulated by law (The Tobacco and Tobacco Products Act). According to the provisions of this Act, tobacco producers are allowed to use only certified seed material and the only authorized institution for production of such material is Scientific Tobacco Institute , Prilep.

The aim of our investigation is to study importance of the tobacco production in Macedonia for economic and social reasons.

Presently, tobacco production in the country is managed by nine purchasing companies with foreign capital (Socotab, Alliance One, Strumica Tabak, etc.) and one domestic company - Tututnski Kombinat - Prilep. Oriental tobacco is grown in several areas: Pelagonia (which covers 46.8% of the area under tobacco in R. Macedonia), Strumica, Radovis and some minor regions. 7-10 years ago, a conglomerate of varieties could be observed in these regions, but now, due to the selection work of the experts from Scientific Tobacco Institute – Prilep, the varietal structure is far more homogenous and a share of almost 80% of the total oriental tobacco production in the country accounts for the variety Prilep P 66-9/7.

Pelagonia is the largest tobacco growing region, especially the Prilep area which accounts for 25% of the oriental tobacco production. The most dominant variety in this region is of the type Prilep, while in the other regions Yaka and Basma tobaccos can also be observed.

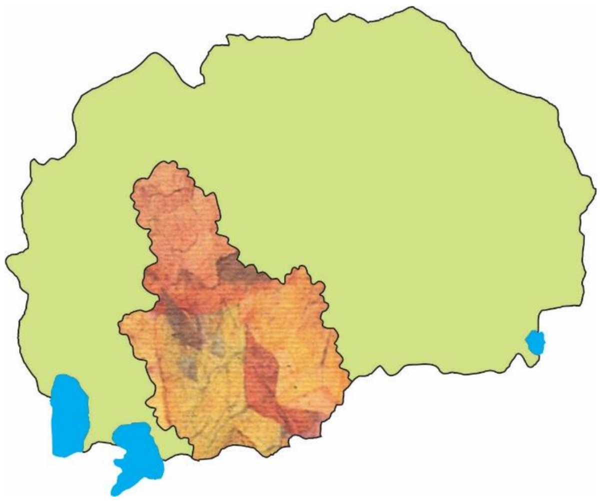

**Foto. 1 Region of Pelagonia**

This is a newly created variety, recognized in 2004 and added to the list of registered varieties of domestic agricultural plants (Official Gazette of the Republic of Macedonia No.70/2004. Authors of this variety is Prof.dr. Dimitrieski M. and prof. dr.Miceska G.).

Plants are ellipsoid-conic in shape, with height average of 65 - 75 cm, depending on conditions of growing and applied cultural practices. Average leaf number of leaves is 52 and they are uniformly distributed on the stem. Average size is in the limits of 18 - 22 cm for the largest leaf, 16-18 cm for the middle and 8 - 10 for the top leaves. Inflorescence is relatively small, moderately to tightly condensed, semi-oval in shape. (Dimitrieski M.,Miceska G., 2011).

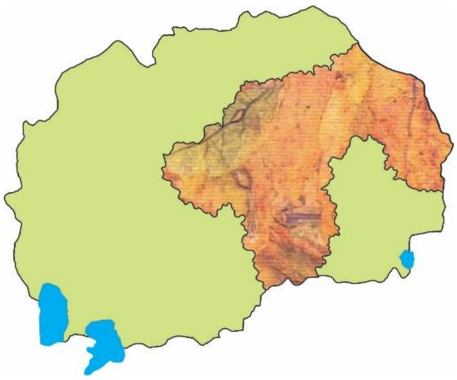

**Foto 2. Eastern region**

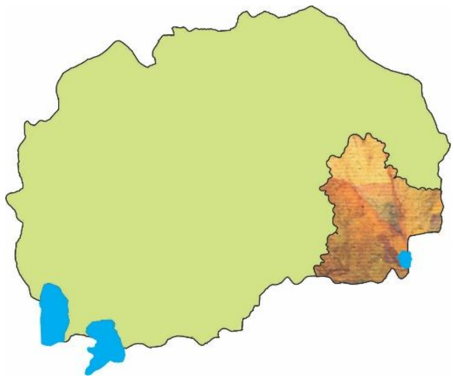

**Foto 3. Strumica – Radovis region**

Prilep 66-9/7 (Miceska G., at all. 2014) is suitable for growing at loose, light and drained soils, with poor supply of nutrient elements, showing especially good results under irrigation conditions. The variety also achieves good yields and quality in soils with medium supply of nutrients, where no possibilities for irrigation exist, yielding small-leaf aromatic and substantial tobacco typical for the type Prilep. Length of the growing season from planting to the beginning of flowering is 70 - 75 days. The variety is resistant to blue mold and bassara disease, and tolerant to viruses. The Dry tobacco yield averages **2000 - 3600 kg/ha,** depending on conditions of growing and applied cultural practices.

Prilep 66-9/7 belongs to the group of small-leaf aromatic tobaccos with uniform raw typical for the type Prilep. It has fine, soft and substantial dry leaf tissue, with yellow-orange color of the middle leaves and orange to light red of the upper. It is distinguished by high percentage of high-graded tobacco. ( Dimitieski M.), at all. 2014.

Chemical composition of this tobacco is variable and depends greatly on conditions of growing and applied agrotechnical measures. The average values of major chemical compounds range within the following limits: nicotine **1.0 % (irrigated) - 2.30 % (non-irrigated), proteins 5 % - 8%, soluble sugars 18.50% - 29.00% and Shmuk's quality index 2.5 - 5.0.**

During smoking, this tobacco is medium in strength, with full and sweetish taste and strongly expressed, intensive aroma

.


**Foto 4. Prilep 66-9/7**


**Foto 5. Seed plot of Prilep 66-9/7**

The average area under tobacco in R. Macedonia for the period 2008 - 2016 was 16 052 ha. According the oriental tobacco production in the country has increased from 16 280 tonnes purchased tobacco in 2008 to 30 997 tonnes in 2013 (Table 1).

Of course, this is primarily due to the interest shown by the foreign companies for Macedonian oriental tobacco, characterized by good quality and yield, which is included in the American blend cigarettes by 7-10% and it is known that these cigarettes account for almost 70% of the world cigarette manufacture.

The number of contracts, the areas planted with tobacco in hectares and total subsides are also presented in Table 1.

**Table 1: Production of tobacco in R. Maedonia in the period 2008-2016**

| Year | Number of contracts | Areas under tobacco/ ha | Purchased tobacco/tonnes | Total subsidies in € |
|---|---|---|---|---|
| 2008 | 30 519 | 17 185 | 16 280 | 11 799 512,20 |
| 2009 | 38 710 | 16 212 | 23 196 | 22 655 291,29 |
| 2010 | 40.743 | 18 846 | 26 393 | 25 749 268,00 |
| 2011 | 33.234 | 15.677 | 21 024 | 20 511 219,00 |
| 2012 | 29.090 | 14.609 | 27.993 | 27 310 244,00 |
| 2013 | 42.367 | 19.806 | 30.997 | 30 240 976,00 |
| 2014 | 34 445 | 14 030 | 24 857 | 23 623 171,00 |
| 2015 | 28454 | 14 127 | 18 910 | 35 035 691,00 |
| 2016 | 27 380 | 13 978 | 25 167 | 24 554 097,00 |
| 2017 | 29 132 | 14 342 | 23559,122 | |

**Source: Ministry of agriculture, forestry and water economy of R. Macedonia**

The tobacco purchase in the last year 2016 of production recorded upward movement, with noticeable decrease of the average purchase price Table 2. In 2017 the purchase amount was 85,58% higher compared to 2014, while the average price was 1,90€/kg.

The increase of production appeared as a result of the stimulative measures undertaken by the government of the Republic of Macedonia and the subsidies for tobacco of 1€ per kg (Table 1).

**Table 2. Quantity of purchased tobacco**

| Year | 2009 | 2010 | 2011 | 2012 | 2013 | 2014 | 2015 | 2016 | 2017 |
|---|---|---|---|---|---|---|---|---|---|
| Quantity (tons) | 23 196 | 26 158 | 21 024 | 27 993 | 30 997 | 24 857 | 18 910 | 25 167 | 23557 |
| Average price €/kg | 3,12 | 2,22 | 2,68 | 2,93 | 2,48 | 1,90 | 3,00 | 3,20 | 3,53 |

**Source: Ministry of agriculture, forestry and water economy of R. Macedonia**

If we analyze the prevalence of the purchased tobacco by types of tobacco (Table 3), it can be concluded that the type Prilep participates with an average of 78.56% (2010-2016 in the total purchase of tobacco.) Its participation is above all the level of uniformity and quality of tobacco raw material, as well as the varietal structure of the type Prilep represented in the tobacco production in Republic of Macedonia, which completely coincides with the results given in Table 4, for the representation of the high classes of purchased tobacco in accordance with the Rulebook on Tobacco purchased in the Republic of Macedonia.

**Table 3 . Purchase of leaf tobacco by types**

| Types/ in tonnes | Year | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 2010/2011 | % | 2011/2012 | % | 2012/2013 | % | 2014/2015 | % |
| Prilep, | 18 364 | 70,00 | 14 838 | 73,00 | 20 320 | 72,77 | 16 684 | 88,22 |
| Yaka | 6 704 | 25,56 | 5 327 | 26,41 | 6 913 | 24,75 | 2 150 | 11,36 |
| Basmak | 1161 | 4,43 | 0.730 | 0,04 | 690 | 2,47 | 77 | 0,41 |
| Total | 26 229 | 100,00 | 20 165 | 100,00 | 27 923 | 100,00 | 18 911 | 100,00 |

**Source: Ministry of agriculture, forestry and water economy of R. Macedonia**

**Table 4. Purchased amount of row tobacco in tonnes and share of Purchased classes in %**

| Class | 2014 | | 2015 | | 2016 | |
|---|---|---|---|---|---|---|
| | Tobacco tonnes | Tobacco % | Tobacco tonnes | Tobacco % | Tobacco tonnes | Tobacco % |
| I | 245 | 1,06 | 2 264 | 11,98 | 4 020 | 16,00 |
| II | 3987 | 17,39 | 11 711 | 61,98 | 17 366 | 69,00 |
| III | 12 102 | 52,80 | 4 579 | 24,23 | 3 610 | 14,30 |
| IV | 6584 | 28,73 | 337 | 1,78 | 162 | 0,6 |
| V | | | | | 618 | 0,002 |
| Total | 22 918 | 100,00 | 18 892 | 100,00 | 25 160 | 100,00 |

**Source: Ministry of agriculture, forestry and water economy of R. Macedonia**

According to the State Statistical Office, tobacco exports from the Republic of Macedonia in 2010 was 16 546 tons, which is over 69% of the total tobacco production. The share of tobacco in the total exports from the country was 3.7% (Statistical Yearbook of the Republic of Macedonia, 2011). At first sight, it might seem that tobacco takes insignificant part in international economic relations of the Republic of Macedonia. In terms of agri-exports, however, exported tobacco in 2013 amounted to 25 444 tons, which is a share of over 22% from the total exports from R. Macedonia Table 5.

**Table 5. Amount of exportet and imported of fermented tobacco in the period 2010- 2015 ( tonnes)**

| Year | fermented tobacco | Tobacco export | Tobacco import |
|---|---|---|---|
| 2010 | 26 229 | 16 546 | 2 911 |
| 2011 | 20 165 | 21 495 | 3 625 |
| 2012 | 27 923 | 22 462 | 4 986 |
| 2013 | 30 997 | 25 440 | 3 197 |
| 2014 | 24 857 | 22 588 | 3 463 |
| 2015 | 18 910 | 24 893 | 3 616 |

**Source: Ministry of agriculture, forestry and water economy of R. Macedonia**

## CONCLUSIONS

In general, the presented data for the investigation period (2006-2013) reveal an upward trend in production of oriental tobacco in R. Macedonia. Tobacco is strategic crop and I believe that all questions and disputes regarding the ban on its future production in these areas have no basis, taking into account the Global strategy on tobacco as well as the assumptions of FAO experts on the increase of its production by almost 25%. The real question for us is how to find a place in that framework and to make the best use of the comparative advantages of this area for stable production of oriental tobacco. If we want to resolve that problem, several important factors should be taken into account:

- specification of tobacco growing areas, to stop the production of varieties in regions where the inappropriate climate, pedological and other factors significantly affect the quality of tobacco.

-taking proper measures to stabilize the varietal purity by: consistent upholding of the Law on Tobacco;

- use only of certified seed, supplied from authorized breeder;

- constant work on creation of new varieties which will retain typical traits of the type;

- use of varieties with low environmental variability, i.e. higher biological plasticity;

- timely contract-signing and information on purchase prices of leaf tobacco;

- financial support for scientific-rersearch activities;

- Further subsidization of purchased tobacco per kg and for modernization of agricultural mechanization;

- investments in micro-accumulations;

- consolidation of agricultural properties;

- introducing modern technologies in the process of production.

Only with a well-coordinated cooperation among the Ministry of Agriculture, Forestry and Water Economy, Tobacco Institute, buyers, tobacco associations, manufacturers and farmers there is a real opportunity to resolve the problems.

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ISSN 0494-3244
Тутун/Tobacco Vol. 67 No 7-12, 11-17, 2017 UDC:633.71-152.61:631.574 (497.775)”2012/2013”
Original scientific paper

# MORPHOLOGICAL TRAITISTICS OF LEAVES IN BURLEY TOBACCO GENOTYPES

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## ABSTRACT

The raw materials of Burley and Virginia tobacco constitute about 80% of the composition of blend cigarettes. The early 70ties of the last century were marked by an intensive search for the best variety, primarily in terms of yield and quality. During that period of modest production of Burley tobacco, the Scientific Tobacco Institute in Prilep, R. Macedonia, created new male-sterile varieties of this type: B-96/85 CMS $F_1$, Burley CMS F1, B-2/93 CMS $F_1$ and Pelagonec CMS $F_1$, each of them carrying specific traitistics. These varieties successfully replaced the variety Chulinec, and some of them were exported to other countries. But the fact that there is no ideal variety created once and for all motivates the breeders of Tobacco Institute - Prilep to create new varieties (genotypes) with improved morpho-biological and productional traits. The aim of this paper is to present morphological traits (leaf length and width) of 3 foreign varieties (Kentucky 14 (Ø), Stella, B-1000) and three Macedonian male-sterile hybrid lines: B-198/11 CMS $F_1$, B- 204/10 CMS $F_1$ and B-205/10 CMS $F_1$. The two-year investigation was set up in randomised block design with 4 replications and morphological measurements were statistically processed. In the process of creating new lines, parental pairs which contain dominant morphological traits in their genetic constitution were used. Only intervarietal hybridization was applied to obtain male-sterile hybrid varieties. The best of them were studied for several years and they gave satisfactory results, which are presented in this paper.

**Keywords**: tobacco, Burley, type, variety, hybrid

## МОРФОЛОШКИ ОСОБИНИ НА ЛИСТОВИТЕ КАЈ ГЕНОТИПОВИ ТУТУН ОД ТИПОТ БЕРЛЕЈ

Суровината од типот берлеј заедно со вирџиниската, во составот на блендцигарите учествуваат со околу 80%. На почетокот на седумдесетите години од минатиот век, интензивно се трага по сорта која во повеќе својства ( пред сé принос и квалитет ) ќе се покаже како најдобра. Во овој период на скромно производство на тутун од овој тип, во Р. Македонија односно во Научниот институт за тутун – Прилеп се создадени и признати берлејските машкостерилни сорти: Б-96/85 ЦМС $F_1$, Берлеј ЦМС $F_1$, Б-2/93 ЦМС $F_1$ и Пелагонец ЦМС $F_1$, сите различни и со свои специфичности. Овие сорти доста успешно ја заменија сортата Чулинец, а некои од нив беа застапени во производство и надвор од Македонија. Фактот дека нема идеална сорта еднаш засекогаш создадена, ги мотивира селекционерите во НИТ - Прилеп да создаваат нови сорти. Тука пред сé се мисли на сорти (генотипови) кои ќе бидат доминантни во морфолошко-биолошките и квалитетно - производните својства. Целта на овој труд е да ги претставиме морфолошките својства ( должина и широчина на листовите ) на 3 странски сорти: Kentucky 14 (Ø), Stella, Б-1000 и три македонски машкостерилни хибридни линии: Б-198/11 ЦМС $F_1$, Б-204/10 ЦМС $F_1$ и Б-205/10 ЦМС $F_1$. Опитот беше двегодишен, поставен по мeродот Randum block system во 4 повторувања, а морфолошките мерења беа варијационо статистички обработени. Во процесот на создавање на новите линии беа користени родителски парови кои во својата генетска конституција ги содржат доминантните морфолошки својства. Хибридизацијата исклучиво е меѓусортова насочена во правец на добивање на машкостерилни хибридни сорти. Најдобрите од нив повеќе години ги испитувавме по што добивме резултати кои беа задоволителни и тие ги прикажуваме во трудот.

**Клучни зборови:** тутун, берлеј, тип, сорта, хибрид.

## INTRODUCTION

Until 2002, Burley tobacco production in R. Macedonia was located mostly in the southwest regions. Meanwhile, a number of new Burley tobacco varieties have been created in the Scientific Tobacco Institute-Prilep, which yield and quality guarantee a profitable production, with continuous work on creation of new genotypes.

Creation of new genotypes is a long-term process that is based on phenotypic and genotypic investigations of the lines obtained. Due to the effect of various environmental factors, it takes several years to complete the investigations. The presentation of our findings on morphological traits of Burley tobacco is aimed to give useful knowledge for further selection of this type. The aim of this paper is to present morphological traits (leaf length and width) of 3 foreign varieties (Kentucky 14 (Ø), Stella, B-1000) and three Macedonian male-sterile hybrid lines: B-198/11 CMS $F_1$, B- 204/10 CMS $F_1$ and B-205/10 CMS $F_1$. Kocoska, 2016, reported that the average length of the 10th leaf in Burley tobacco variety Pelagonec reached 66.60 cm. Risteski, 2007, reported that the average length of the $10^{th}$ leaf in Burley tobacco variety Pelagonec reached 54.5 cm. Results obtained by morphological traits dominance of new genotypes B-198/11 CMS $F_1$, B- 204/10 CMS $F_1$ and B-205/10 CMS $F_1$.

## MATERIAL AND METHODS

The two-year investigation (2012 - 2013) included 6 tobacco varieties (lines): three of domestic origin (B-198/11 CMS $F_1$, B-204/10 CMS $F_1$ and B-205/10 CMS $F_1$- new hybrid lines created in Tobacco Institute-Prilep) and three introduced fertile varieties (Kentucky 14 (standard), Stella and B-1000). The trial was set up on colluvial soil in the field of Tobacco Institute. The first plowing was done in the autumn at 40 cm depth. In spring, the plots were fertilized with 300 kg/ha N: P: K 8:22:20, and then ploughed two more times. Before transplanting, the plots were treated with selective herbicide and seedlings were planted in randomized block design with 4 replications at 90 × 50 cm planting density. Due to the poor supply of nitrogen, 5 g/26 % KAN was applied prior to second hoeing. 3-5 irrigations with 30-40 l/$m^2$ water were carried out, depending on the climate conditions. Morphological measurements were made on the lower and middle belt leaves ($5^{th}$, $10^{th}$, and $15^{th}$ leaf) of 5 stalks - typical representatives of the varieties, and their average value was calculated. The obtained data were processed by variational-statistical analysis and LSD test was used by the methods cited by (Najchevska, 2002).

## RESULTS AND DISCUSSION

### Length and width of the 5th leaf

This leaf is positioned in the lower belt and, due to its size, it can significantly affect the tobacco yield. Data presented in Table 1 show that maximum length was measured in the variety B-198/11 CMS $F_1$ (48.4 ± 0.98 cm in 2012 and 49.6 ± 0.54 in 2013), compared to the standard variety Kentucky 14 (37.8 ± 1.25 cm in 2012 and 37.2 ± 1.23 in 2013). In 2012 the lowest standard deviation of 0.74 and coefficient of variation of 1.65% cm was calculated in the newly created line B-205/10 CMS $F_1$, while in 2013 the lowest standard deviation of 0.33 cm and coefficient of variation of 0.66% was calculated in the new line B-198/11 CMS $F_1$.

Table 1. Length of the 5th leaf

| Genotypes | 2012 | | | 2013 | | |
|---|---|---|---|---|---|---|
| | $\bar{x} \pm s\bar{x}$ | σ | cV % | $\bar{x} \pm s\bar{x}$ | σ | cV % |
| Kentucky 14 Ø | 37.8±1.25 | 1.33 | 3.52 | 37.2±1.23 | 1.25 | 3.37 |
| Stella | 44.8**±0.26 | 0.93 | 2.09 | 45.0**±1.01 | 1.01 | 2.24 |
| B-1000 | 31.2±1.51 | 1.58 | 5.07 | 30.6±0.90 | 0.56 | 1.82 |
| B-198/11 CMS $F_1$ | 48.4**±0.98 | 1.04 | 2.15 | 49.6**±0.54 | 0.33 | 0.66 |
| B-204/10 CMS $F_1$ | 45.4**±1.22 | 1.51 | 3.35 | 44.6**±1.04 | 1.08 | 2.42 |
| B-205/10 CMS $F_1$ | 44.8**±0.86 | 0.74 | 1.65 | 44.06**±0.77 | 0.59 | 1.33 |
| LSD 5 % * | 4.20 cm | | | 1.97 cm | | |
| 1% ** | 5.81 cm | | | 2.73 cm | | |

Table 2. Width of the 5th leaf

| Genotypes | 2012 | | | 2013 | | |
|---|---|---|---|---|---|---|
| | $\bar{x} \pm s\bar{x}$ | σ | cV% | $\bar{x} \pm s\bar{x}$ | σ | cV% |
| Kentucky 14 Ø | 19.8±1.87 | 1.54 | 7.80 | 19.4±0.87 | 0.33 | 1.68 |
| Stella | 25.8**±1.21 | 0.84 | 3.27 | 28.0**±0.85 | 1.45 | 1.61 |
| B-1000 | 18.8±1.71 | 1.26 | 6.75 | 18.8±1.10 | 0.51 | 2.72 |
| B-198/11 CMS $F_1$ | 29.4**±1.08 | 0.77 | 2.63 | 29.4**±0.88 | 0.52 | 1.76 |
| B-204/10 CMS $F_1$ | 28.8**±0.98 | 0.62 | 2.15 | 27.6**±1.03 | 0.65 | 2.38 |
| B-205/10 CMS $F_1$ | 30.6**±0.80 | 0.42 | 1.41 | 31.2**±0.86 | 0.51 | 1.64 |
| LSD 5 % * | 1.88 cm | | | 2.12 cm | | |
| 1% ** | 2.61 cm | | | 2.94 cm | | |

Table 2 shows that the greatest width of the 5th leaf was measured in the line B-205/10 CMS $F_1$ (30.6 ± 0.80 cm in 2012 and 31.2 ± 0.86 in 2013). It is distinguished by lower values for standard deviation (0.42 in 2012 and 0.51 cm in 2013), while the coefficient of variation ranged from 1.41% in 2012 to 1.64% in 2013. Statistically significant difference for the trait leaf width by 1% compared to the standard variety was estimated in the variety Stella and the newly created hybrid lines B-198/11 CMS $F_1$, B-204/10 CMS $F_1$, B-205/10 CMS $F_1$ in both years of investigation.

**Length and width of the 10th leaf**

Middle belt leaves have the highest share in large-leaf tobaccos both in terms of quality and weight. To obtain the maximum of some variety, besides the genetic potential, proper cultural practices and favorable soil - climate conditions are very important. In our investigations, high genetic potential was recorded in B-198/11 CMS $F_1$ and B-204/10 CMS $F_1$. According to Table 3, the maximum length of the 10th leaf (69.4 ± 1.05 cm) in 2012 was measured in line B-198/11 CMS $F_1$ and in B-204/10 CMS $F_1$ (68.6 ± 1.12cm) in 2013. Kocoska, 2016, reported that the average length of the 10th leaf in Burley tobacco variety Pelagonec reached 66.60 cm. The lowest standard deviation (0.25 in 2012) and coefficient of variation (0.48% in 2012) were calculated in the variety B-1000. In 2013, the lowest standard deviation (0.69 cm) and coefficient of variation (1.37%) were calculated in line B-204/10 CMS $F_1$. The low values of these two parameters confirm the stability of the line. 1% statistical significance for the trait Length of the 10th leaf was calculated in the variety Stella and hybrid lines B-198/11 CMS $F_1$, B-204/10 CMS $F_1$, B-205/10 CMS $F_1$ both in 2012 and 2013. The investigated lines and their morphological traits show that they can also reach higher average yield per stalk and hectare. (Uzunoski, 1987, and Karayankov et al, 2007), on the basis of leaf size, divide all tobacco types and varieties into three basic groups: small-leaf tobaccos (up to 18 cm), semi-oriental tobaccos (19-30 cm) and large-leaf tobaccos (over 30 cm). According to literature data, the investigated varieties and lines belong to the group of large-leaf tobaccos.

Table 3. Length of the 10th leaf

| Genotypes | 2012 | | | 2013 | | |
|---|---|---|---|---|---|---|
| | $\bar{x} \pm s\bar{x}$ | σ | cV % | $\bar{x} \pm s\bar{x}$ | σ | cV% |
| Kentucky 14 Ø | 53.2±1.58 | 2.00 | 5.64 | 55.4±1.23 | 1.88 | 3.39 |
| Stella | 60.2**±0.65 | 0.59 | 0.97 | 61.4**±0.85 | 1.00 | 1.63 |
| B-1000 | 49.8± 0.46 | 0.24 | 0.48 | 51.0±0.78 | 0.69 | 1.37 |
| B-198/11 CMS $F_1$ | 69.4**±1.05 | 1.72 | 2.48 | 67.0**±0.85 | 1.09 | 1.62 |
| B-204/10 CMS $F_1$ | 68.2**±1.15 | 2.01 | 2.95 | 68.6**±1.12 | 1.69 | 2.47 |
| B-205/10 CMS $F_1$ | 63.4**±0.88 | 1.10 | 1.73 | 62.6**±0.65 | 0.59 | 0.95 |
| LSD 5 % * | 3.54 cm | | | 3.51 cm | | |
| 1% ** | 4.91 cm | | | 4.88 cm | | |

Table 4. Width of the 10th leaf

| Genotypes | 2012 | | | 2013 | | |
|---|---|---|---|---|---|---|
| | $\bar{x} \pm s\bar{x}$ | σ | cV % | $\bar{x} \pm s\bar{x}$ | σ | cV % |
| Kentucky 14 Ø | 30.0±1.48 | 1.47 | 4.90 | 30.6±0.80 | 0.43 | 1.42 |
| Stella | $38.0^{**}$±0.46 | 0.52 | 1.50 | $37.2^{**}$±0.61 | 0.98 | 2.63 |
| B-1000 | 30.2±0.68 | 0.31 | 1.04 | 30.0±0.89 | 0.53 | 1.78 |
| B-198/11 CMS $F_1$ | $44.0^{**}$±0.76 | 0.57 | 1.30 | $45.2^{**}$±0.80 | 0.65 | 1.44 |
| B-204/10 CMS $F_1$ | $43.6^{**}$±0.92 | 0.82 | 1.89 | $42.8^{**}$±1.01 | 0.98 | 2.28 |
| B-205/10 CMS $F_1$ | $40.8^{**}$±0.51 | 0.24 | 0.59 | $40.2^{**}$±0.64 | 0.37 | 0.93 |
| LSD 5 % * | 3.18 cm | | | 2.83 cm | | |
| 1% ** | 4.40 cm | | | 3.93 cm | | |

As presented in Table 4, the maximum width of the 10th leaf was measured in the line B-198/11 CMS $F_1$ (44.0 ± 0.76 cm in 2012 and 45.2 ± 0.80 cm in 2013), while the smallest width was recorded in Kentucky 14 (30.0 ± 1.48 cm in 2012 and 52.2 ± 1.01 cm in 2013). The results obtained show significant differences among the varieties and lines, which is confirmed by the LSD test.

## Length and width of the 15th leaf

According to the data presented in Table 5, the largest length of the 15th leaf was measured in the line B-198/11 CMS $F_1$ (67.0 ± 1.05 cm in 2012 and 64.2 ± 0.83 cm in 2013), while the lowest length was recorded in variety B-1000 (50.2 ± 0.61 cm in 2012 and 50.0 ± 0.64 cm in 2013). The lowest values for standard deviation (0.43 and 0.45 cm) and coefficient of variation (0.84% and 0.90%) were calculated in the variety B-1000, which is an indicator of uniformity of plants with respect to this trait. Compared to the standard variety, the hybrid lines had lower values for these two traits and according to the LSD test, the variety Stella and lines B-198/11 CMS $F_1$, B-204/10 CMS $F_1$ and B-205/10 CMS $F_1$ showed highly significant differences.

Table 5. length of the 15th leaf

| Genotypes | 2012 | | | 2013 | | |
|---|---|---|---|---|---|---|
| | $\bar{x} \pm s\bar{x}$ | σ | cV % | $\bar{x} \pm s\bar{x}$ | σ | cV % |
| Kentucky 14 Ø | 54.0±1.19 | 1.71 | 3.17 | 52.2±1.01 | 1.18 | 2.27 |
| Stella | $57.4^{**}$±0.83 | 0.87 | 1.52 | $56.3^{**}$±1.22 | 1.88 | 3.34 |
| B-1000 | 50.2± 0.61 | 0.43 | 0.84 | 50.0±0.64 | 0.45 | 0.90 |
| B-198/11 CMS $F_1$ | $67.0^{**}$±1.05 | 1.16 | 1.73 | $64.2^{**}$±0.83 | 0.98 | 1.52 |
| B-204/10 CMS $F_1$ | $66.8^{**}$±1.10 | 1.80 | 2.69 | $63.6^{**}$±1.00 | 1.44 | 2.25 |
| B-205/10 CMS $F_1$ | $62.0^{**}$±0.75 | 0.73 | 1.17 | $61.0^{**}$±0.72 | 0.70 | 1.15 |
| LSD 5 % * | 2.53cm | | | 2.71 cm | | |
| 1% ** | 3.51 cm | | | 3.75 cm | | |

The results of the investigations show that the newly obtained lines have significantly greater length and width of the 15th leaf.

According to Table 6, the greatest width in 2012 was measured in the leaves of line B-204/10 CMS $F_1$ (36.0 ± 1.10 cm) and line B-198/11 CMS $F_1$ in 2013 (34.0 ± 0.98 cm). The smallest width of the 15th leaf was measured in variety B-1000 (28.2 ± 0.77 in 2012, and 28.1 ± 1.31 in 2013). The standard deviation for this trait was lower in all varieties and lines in 2012, while in 2013 the estimated values were somewhat higher compared to the standard, which is also the case with the coefficient of variation.

Table 6. Width of the 15th leaf

| Genotypes | 2012 | | | 2013 | | |
|---|---|---|---|---|---|---|
| | $\bar{x} \pm s\bar{x}$ | σ | cV % | $\bar{x} \pm s\bar{x}$ | σ | cV % |
| Kentucky 14 Ø | 29.6±1.32 | 1.15 | 3.90 | 29.0±0.83 | 0.45 | 1.56 |
| Stella | 29.8±0.80 | 0.43 | 1.43 | 29.4±0.89 | 0.52 | 1.77 |
| B-1000 | 28.2±0.77 | 0.37 | 1.32 | 28.1±1.31 | 1.08 | 3.83 |
| B-198/11 CMS $F_1$ | $35.4^{**}$±0.64 | 0.33 | 0.90 | $34.0^{*}$±0.98 | 0.73 | 2.14 |
| B-204/10 CMS $F_1$ | $36.0^{**}$±1.10 | 0.97 | 2.69 | $32.0^{**}$±1.01 | 0.75 | 2.30 |
| B-205/10 CMS $F_1$ | $33.4^{**}$±0.92 | 0.63 | 1.87 | $31.8^{*}$±0.88 | 0.55 | 1.73 |
| LSD 5 % $^{*}$ | 2.65 cm | | | 2.58 cm | | |
| 1% $^{**}$ | 3.67 cm | | | 3.58 cm | | |

## CONCLUSIONS

The two-year trials on the morphological properties of tobacco (leaf length and width) led to the following conclusions:

- The greatest length of the 5th leaf (48.4 ± 0.98 cm in 2012 and 49.6 ± 0.54 in 2013) was measured in the variety B-198/11 CMS $F_1$, compared to the standard variety Kentucky 14 (37.8 ± 1.25 cm in 2012 and 37.2 ± 1.23 in 2013).
- The lowest standard deviation of the 5th leaf (0.74 cm) in 2012 and coefficient of variation (1.65%) were measured in the newly created line B-205/10 CMS $F_1$. In 2013, the lowest standard deviation (0.33 cm) and coefficient of variation (0.66%) were assessed in the newly created line B-198/11 CMS $F_1$, which is an indicator of the stability of these lines related to this trait.
- The width of the 5th leaf was the largest in line B - 205/10 CMS $F_1$ (30.6 ± 0.80 cm in 2012 and 31.2 ± 0.86 in 2013). This variety has lower values for standard deviation and coefficient of variation for this trait.
- The greatest length of the 10th leaf in 2012 had the line B-198/11 CMS $F_1$(69.4 ± 1.05 cm), and in 2013 the line B-204/10 CMS $F_1$ (68.6 ± 1.12cm). In 2013, B-205/10 CMS $F_1$ had the lowest values for standard deviation (0.59 cm) and coefficient of variation (0.95%). The low values for these two parameters indicate the stability of the line.

- The width of the 10$^{th}$ leaf was the largest in line B-198/11 CMS $F_1$ (44.0 ± 0.76 cm in 2012 and 45.2 ± 0.80 cm in 2013, and the smallest width was measured in the standard variety Kentucky 14 (30.0 ± 1.48 cm in 2012 and 52.2 ± 1.01 cm in 2013).

- The largest length of the 15$^{th}$ leaf was measured in B-198/11 CMS $F_1$ (67.0 ± 1.05 cm in 2012 and 64.2 ± 0.83 cm in 2013), and the smallest length in variety B-1000 (50.2 ± 0.61 cm in 2012 and 50.0 ± 0.64 cm in 2013.) In 2013, line B-204/10 CMS $F_1$ has the lowest values of the standard deviation (0.69 cm) and coefficient of variation (1.37%), which also indicates stability of the line for this trait.

- The maximum width of the 15$^{th}$ leaf in 2012 was measured in the line B-204/10 CMS $F_1$ (36.0 ± 1.10 cm), and in the line B-198/11 CMS $F_1$ in 2013 (34.0 ± 0.98) and the smallest width was found in variety B-1000 (28.2 ± 0.77 in 2012 and 28.1 ± 1.31 cm in 2013).

It can be concluded that the newly created lines and varieties have greater leaf size compared to the standard and are genetically stable, while according to the LSD test they show highly significant differences.

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Тутун/Tobacco Vol. 67 No 7-12,18-25, 2017

ISSN 0494-3244
UDC: 633.71-152.61(497.2)”2013/2015”
Original scientific paper

# INHERITANCE OF BASIC QUANTATIVE TRAITS OF ORIENTAL TOBACCO ECOTYPE DUPNITSA

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## ABSTRACT

Investigations were carried out with four oriental tobacco varieties - three of which are Bulgarian – Dupnitsa 126, Dupnitsa 160 and Sandanski 321, and Prilep 7 from the Republic of Macedonia. The trial was set up in the Tobacco and Tobacco Products Institute - Markovo, Experimental Tobacco Station - Rila in 2013-2015 in randomized block design with four replications, using traditional agricultural practices. The aim of the investigation is to study the inheritance of basic quantitative traits (plant height, leaf number, length and width of the 14-th leaf). It was found that plant height is inherited by partial dominance.

The mode of inheritance of leaf number was complete dominance, incomplete dominance and overdominance towards parent with fewer leaves.

The 14-th leaf length and width are inherited dominantly, incompletely and overdominantly towards parent with longer and wider leaves.

## НАСЛЕДУВАЊЕ НА ОСНОВНИТЕ КВАНТИТАТИВНИ СВОЈСТВА КАЈ ОРИЕНТАЛСКИОТ ТУТУН ЕКОТИП ДУПНИЦА

Истражувања беа спроведени со четири ориенталски видови тутун, од кои три бугарски - Дупница 126, Дупница 160 и Сандански 321 и Прилеп 7 од Република Македонија. Опитот е поставен во Институтот за тутун и тутунски производи - Марково, експериментална станица - Рила во 2013-2015 година, во случаен блок систем во четири повторувања, користејќи традиционални агротехнички мерки. Целта на истражувањето е да се проучи наследувањето на основните квантитативни својства (висина на растенијата, број на листови, должина и ширина на 14-тиот лист). Утврдено е дека висината на растенијата се наследува со парцијална доминантност.

Начинот на наследување на бројот на листови беше целосна доминантност, нецелосна доминантност и супердоминантност кон родителот со помалку листови.

Должината и ширината на 14-тиот лист се наследуваат со доминантност, нецелосна доминантност и супердоминантност кон родителот со подолги и пошироки листови.

## INTRODUCTION

Tobacco production has a long tradition in Bulgaria. The climatic conditions are favorable for growing high-quality oriental tobaccos, rich in aromatic oils and with well-balanced content of reducing sugars and nicotine. The variability of quantitative traits of oriental tobacco is due to the large number of genes which determine them, relatively low gene effects in the inheritance of certain signs and different environmental conditionc. The individual characteristics of the genotype need a specific study of the structure of gene effects and their influence in the inheritance of the traits (Lee and Chang,1984; Dimitrova and Vasilev, 2004).

Major signs тхат determine the productivity of Oriental tobacco are leaf number, plant height, size and absolute dry weight of the leaves, length of the flowering period etc. Unlike other crops, tobacco varieties are exclusively related to the growing conditions in which a large part of chemical compounds and specific consumer qualities of the raw material is formed (Kasheva et al.,2013; Korubin-Aleksoska, 2001).

Several authors ( (Dulgerski, 2014, Šmalcelj 1983, Shoai Daylami and Honarneja 1996, Butorac et al., 1999) reported that nonadditive variance plays somewhat greater role in the inheritance of plant height. The number of leaves is mostly inherited dominantly and partially dominant against parent with a higher leaf number (Caneva, 1980; Vassilev 2010) and overdominantly or additively towards parent with higher or lower values (Dulgerski 2014). Sastry and Rrasada Rao (1980) reported that the most important in inheritance of leaf number in Virginia tobacco are dominant and additive gene effects.

The aim of the investigation was to determine the mode of inheritance of basic traits of $F_1$ tobacco hybrids of Dupnitsa ecotype.

## MATERIALS AND METHODS

The experiment was conducted in the period 2013-2015 in the field of the Tobacco Experimental Station–Rila. The trial was set up according to the randomised complete block design at four replications. Standard agrotechnical practices for this tobacco type were applied during tobacco growing. Four tobacco genotypes were included in investigations - Sandanski 321, Dupnitsa 126, Dupnitsa 160 and Prilep 7.

The following variants were included in the study: $P_1$, $P_2$, $F_1$.

The following traits were subject of investigation:

- Plant height - measurements were made from the base of the stem to the first branch of the color wrist (cm).
- Number of leaves - the total number of leaves was counted.
- Size of the leaves - the length and width of the 14th leaf (cm) were measured.

Data were taken on a sample of 80 plants for each genotype and for each year. The acquired data for all studied traits and for each year were statistically processed by the analysis of variance and LSD test was performed. Inheritance of quantitative signs in $F_1$was applied to the data for each year according to the methods of Genchev (1979) and Mather and Jinks (1971).

The soils in the valley are alluvial and alluvial-meadow, with a significant amount of sand and gravel. They are light and skeletal.

The highest average monthly temperatures during the vegetation of tobacco were recorded in 2013 (total - 106,2 °C at a norm of 99,2 °C); the year with the largest amount of rainfall was 2014 (total 341 mm rainfall, at a norm of 243 mm) and the year with the highest relative humidity was 2014 (334%, at 312 % humidity).

## RESULTS AND DISCUSSION

Quantitative traits are characterized by continuous distribution and are strongly affected not only by effects of genes but also by environmental effects and the effects resulting from the genotype × environment interaction.

Since most tobacco traits important for successful breeding are of quantitative nature, the estimation of the mode of inheritance contributes to their better understanding, thereby enabling rational and targeted combining of desirable genes into future cultivars (Aleksoski J. and A. Korubin-Aleksoska, 2011).

## Inheritance of plant height

Data of the analysis (Table 1) show that in hybrid combination (Prilep 7 x Sandanski 321), the inheritance of plant height is partially dominant. In 2013 and 2015, the degree of dominance (h/d) was (- 0,54) and (- 0,57), the direction of the gene effects is towards the parent with lower height and in 2014 to the parent with a higher stem.

Table 1. Inheritance of plant height

| Hybrid combinations | $P_1$ $\bar{x}\pm S\bar{x}$ | $P_2$ $\bar{x}\pm S\bar{x}$ | $F_1$ $\bar{x}\pm S\bar{x}$ | MP | h | d | h/d |
|---|---|---|---|---|---|---|---|
| **2013** | | | | | | | |
| (Pr.7 x Sand. 321) | 75,00 ±1,53 | 132,7 ± 0,9 | 88,2 ±2,05 | 103,85 | -15,65 | 28,85 | -0,54 |
| (Pr.7 x Dupn. 160) | 75,00 ±1,53 | 130,6 ±1,86 | 105,5 ±1,23 | 102,80 | 2,72 | 27,78 | 0,10 |
| (Pr. 7 xDupn.126) | 75,00 ±1,53 | 161,3 ±1,66 | 108,3 ±2,34 | 118,15 | -9,85 | 43,15 | -0,23 |
| **2014** | | | | | | | |
| (Pr.7 x Sand. 321) | 60,98 ±0,85 | 112 ±1,14 | 89,1 ±1,57 | 86,50 | 2,61 | 25,51 | 0,10 |
| (Pr.7 x Dupn. 160) | 60,98 ±0,85 | 129,5 ±0,98 | 84,8 ±1,31 | 95,20 | -10,44 | 34,26 | -0,31 |
| (Pr. 7 xDupn.126) | 60,98 ±0,85 | 132,8 ±2,15 | 98,5 ±1,75 | 96,90 | 1,61 | 35,91 | 0,50 |
| **2015** | | | | | | | |
| (Pr.7 x Sand. 321) | 53,4 ± 1,11 | 119,5 ±1,40 | 67,5 ± 1,36 | 86,45 | -19,00 | 33,05 | -0,57 |
| (Pr.7 x Dupn. 160) | 53,4 ± 1,11 | 114,2 ±1,82 | 65,1 ± 1,96 | 83,80 | -18,70 | 30,40 | -0,62 |
| (Pr. 7 xDupn.126) | 53,4 ± 1,11 | 160,6 ±2,87 | 94,2 ± 2,69 | 107,00 | -12,80 | 53,60 | -0,24 |

In hybrid combinations Prilep 7 x Dupnitsa 160 and Prilep 7 x Dupnitsa 126, the same mode of inheritance was observed - incomplete dominance. In 2015, the direction was towards the variety Prilep 7 - parental form with lower height. According to most studies (Yancheva 1987, Stoilova 1988, Dulgerski 2014, Masheva, 2016) the inheritance of plant height is incompletely dominant.

## Inheritance of the number of leaves

The number of leaves in tobacco is a major morphological indicator that affects both the yield of tobacco varieties and the homogeneity of tobacco products. Leaf number is one of the traits strongly influenced by genetics. The results are presented in Table 2. For hybrid combinations with parental varieties Prilep 7 x Sandanski 321, inheritance was incompletely dominant in two years of the study and the direction of gene effects was on the parent with a smaller or greater number of leaves. These results are in agreement with those reported by Tomov (1975) and Mincheva (1980) for oriental tobacco and Dyulgerski et al. (2012,2014) and Chen (1976) for Burley tobacco.

In 2015 the inheritance was overdominant towards parent with smaller number of leaves. These results are in agreement with those of Noveva et al. (1990), Dimitrova, Vasilev (2004), who reported that inheritance of this trait was overdominant and leaf number was controlled by dominant gene effects.

Table 2. Inheritance of number of leaves

| Hybrid combinations | $P_1$ $\bar{x}\pm S\bar{x}$ | $P_2$ $\bar{x}\pm S\bar{x}$ | $F_1$ $\bar{x}\pm S\bar{x}$ | MP | h | d | h/d |
|---|---|---|---|---|---|---|---|
| **2013 г.** | | | | | | | |
| (Pr.7 x Sand. 321) | 39,20 ±0,26 | 31,9±0,46 | 32,4±0,48 | 35,60 | -3,20 | 3,65 | -0,88 |
| (Pr.7 x Dupn. 160) | 39,20 ±0,26 | 32,6±0,45 | 33,5±0,36 | 35,90 | -2,40 | 3,30 | -0,73 |
| (Pr. 7 xDupn.126) | 39,20 ±0,26 | 36,3±0,35 | 33,8±0,46 | 37,80 | -4,00 | 1,45 | -2,76 |
| **2014 г.** | | | | | | | |
| (Pr.7 x Sand. 321) | 36,40 0,36 | 28,2 ±0,6 | 33,0±0,43 | 32,3 | 0,71 | 4,1 | 0,17 |
| (Pr.7 x Dupn. 160) | 36,40 ±0,36 | 31,6±0,29 | 33,8±0,36 | 33,96 | 0,16 | 2,41 | 0,07 |
| (Pr. 7 xDupn.126) | 36,40 ±0,36 | 33,3±0,32 | 35,3 ±0,9 | 34,8 | 1,49 | 1,56 | 1,0 |
| **2015 г.** | | | | | | | |
| (Pr.7 x Sand. 321) | 37,00 ±0,52 | 32,4±0,82 | 30,0±0,62 | 34,7 | -4,6 | 2,3 | -2,20 |
| (Pr.7 x Dupn. 160) | 37,00 ±0,52 | 33,9±0,77 | 41,9±1,29 | 35,45 | 6,4 | 1,55 | 4,13 |
| (Pr. 7 xDupn.126) | 37,00 ±0,52 | 39,9 ±0,5 | 37,6±0,96 | 38,5 | -0,9 | 1,45 | -0,62 |

In the hybrid combination Prilep 7 x Dupnitsa 160, the inheritance varied within the range of additive, incompletely dominant and overdominant. Such mode of inheritance was established by Stoilova (1988) for hybrids of the Ustina ecotype and Dyulgerski et al. (2016) for Burley tobaccos.

By comparing the average values of the sign in hybrid combination Prilep 7 x Dupnitsa 126, dominant inheritance prevailed over the three years. In 2013 h/d = -2,86 > -1, indicating that the gene action is overdominant and the direction of the effect is towards the parent with lower number of leaves. In 2014, h/d = 1, indicating total dominance of $P_1$ - Prilep 7. In the third year, the trait is inherited with incomplete dominance, towards the parent with lower number of leaves - Prilep 7.

## Inheritance of the 14-th leaf length

The data on the inheritance of the 14-th leaf length are presented in Table 3. The table shows that the trait is inherited incompletely and overdominantly.

In 2013, incompletely dominant mode of inheritance was present in the hybrid combination Prilep 7 x Sandanski 321, in the direction of the parent with longer leaves. Inheritance of this type and direction was found by Dyulgerski, Radukova, (2015), in hybrid combinations of Virginia and Burley tobaccos, and Stankev (1988) in the Oriental tobacco ecotypes Dupnitsa and Nevrokop. In 2014 and 2015, the dominant gene effects were of higher importance and gene action was overdominant.

In the hybrid combination Prilep 7 x Dupnitsa 160, in the first year of investigation the inheritance was overdominant in direction of the parent with higher 14-th leaf length. In 2014, the action of the genes was incompletely dominant and the sign was inherited in direction of variety Dupnitsa 160, h/d = 0,50. In 2015 complete dominance was present, with a value of $F_1$ = (15,6 ± 0,33) = $P_1$ (15,7 ± 0,31) cm and h/d = 1.

Table 3. Inheritance of the 14-th leaf length

| Hybrid combinations | $P_1$ $\overline{x}\pm S\overline{x}$ | $P_2$ $\overline{x}\pm S\overline{x}$ | $F_1$ $\overline{x}\pm S\overline{x}$ | MP | h | d | h/d |
|---|---|---|---|---|---|---|---|
| **2013 г.** | | | | | | | |
| (Pr.7 x Sand. 321) | 19,8 ±0,43 | 22,5±0,47 | 21,4±0,52 | 21,15 | 0,25 | 1,35 | 0,19 |
| (Pr.7 x Dupn. 160) | 19,8 ±0,43 | 24,2±0,56 | 25,7±0,49 | 22,03 | 3,67 | 2,20 | 1,67 |
| (Pr. 7 xDupn.126) | 19,8 ±0,43 | 28,8±0,48 | 26,0±0,41 | 24,34 | 1,66 | 4,49 | 0,37 |
| **2014 г.** | | | | | | | |
| (Pr.7 x Sand. 321) | 15,0 ± 0,21 | 18,8±0,39 | 21,2±0,46 | 16,98 | 4,21 | 1,99 | 2,12 |
| (Pr.7 x Dupn. 160) | 15,0 ± 0,21 | 21,3±0,33 | 19,7±0,42 | 18,12 | 1,57 | 3,13 | 0,50 |
| (Pr. 7 xDupn.126) | 15,0 ± 0,21 | 22,2±0,43 | 22,5±0,54 | 18,6 | 3,93 | 3,59 | 1,09 |
| **2015 г.** | | | | | | | |
| (Pr.7 x Sand. 321) | 15,6±0,33 | 15,6±0,31 | 18,4±0,49 | 15,62 | 2,78 | 0,01 | 2,78 |
| (Pr.7 x Dupn. 160) | 15,6±0,33 | 20,2±0,36 | 15,7±0,31 | 17,9 | -2,2 | 2,3 | 1 |
| (Pr. 7 xDupn.126) | 15,6±0,33 | 22,2±0,31 | 21,7±0,32 | 18,9 | 2,8 | 3,3 | 0,85 |

In the hybrid combination Prilep 7 x Dupnitsa 126 in the first and third year of investigation, the inheritance was incompletely dominant towards the parent with a longer leaf length - variety Dupnitsa 126. In 2014 the degree of inheritance of the trait was complete dominance ($F_1$ = $P_2$, 22,5 ± 0,54 = 22,2 ± 0,43 cm).

Dominant, incomplete dominant and over-dominant inheritance of leaf length in oriental tobacco was reported by Kaneva (1980), Mincheva (1980), Yancheva (1987), Stankev 1988), Butorac et al. (2004) and Dyulgerski (2011).

## Inheritance of the 14-th leaf width

Data on the inheritance of the 14-th leaf width are presented in Table 4. In the hybrid combinations used for the Dupnitsa ecotype, the sign is inherited in completely dominant, incompletely dominant and overdominant mode.

In the hybrid combination Prilep 7 x Sandanski 321, dominant genetic effects were leading during the three-year study. The inheritance was overdominant in direction of the parent with a lower leaf width in 2013 and in direction of the parent with a longer width in the other two years. Similar results were obtained in the studies of Gelemerov (1988) and Stankev (1988), in experiments with Nevrocope and Dupnitsa tobacco varieties.

In the second cross (Prilep 7 x Dupnitsa 160) inheritance was determined by dominant gene effects. The degree of inheritance was incompletely dominant, with direction of the effect on the parent with smaller width in 2015 (h/d = -0.90) and on the parent with wider leaves in variety Dupnitsa 160 (2013 and 2014).

In the hybrid combination Prilep 7 x Dupnitsa 126, the inheritance was completely dominant in 2014 and incompletely dominant in direction of the the parent with wider leaves in 2015 and to the parent with narrower leaves (Prilep 7) in 2013. In the studies of Radukova et al. (2015), such type of inheritance was observed in hybrid combinations of Virginia tobacco.

Table 4. Inheritance of the trait 14th leaf width

| Hybrid combinations | $P_1$ $\bar{x}\pm S\bar{x}$ | $P_2$ $\bar{x}\pm S\bar{x}$ | $F_1$ $\bar{x}\pm S\bar{x}$ | MP | h | d | h/d |
|---|---|---|---|---|---|---|---|
| **2013 г.** | | | | | | | |
| (Pr.7 x Sand. 321) | 11,2±0,35 | 12,7 ±0,42 | 10,99±0,41 | 11,90 | -0,93 | 0,75 | -1,24 |
| (Pr.7 x Dupn. 160) | 11,2±0,35 | 14,2 ±0,2 | 14,02±0,17 | 12,70 | 1,32 | 1,5 | 0,88 |
| (Pr. 7 xDupn.126) | 11,2±0,35 | 16,94±0,47 | 13,3 ±0,17 | 14,07 | -0,77 | 2,87 | -0,27 |
| **2014 г.** | | | | | | | |
| (Pr.7 x Sand. 321) | 8,1± 0,14 | 10,7 ± 0,28 | 12,3 ± 0,48 | 9,38 | 2,91 | 1,27 | 2,29 |
| (Pr.7 x Dupn. 160) | 8,1± 0,14 | 14,2 ± 0,26 | 12,6 ± 0,27 | 11,15 | 1,41 | 3,03 | 0,47 |
| (Pr. 7 xDupn.126) | 8,1± 0,14 | 13,7 ± 0,35 | 13,7 ± 0,28 | 10,90 | 2,80 | 2,80 | 1,00 |
| **2015 г.** | | | | | | | |
| (Pr.7 x Sand. 321) | 8,6± 0,19 | 9,36 ± 0,26 | 10,5 ± 0,33 | 8,98 | 1,52 | 0,38 | 4,00 |
| (Pr.7 x Dupn. 160) | 8,6± 0,19 | 12,4 ± 0,25 | 8,8 ± 0,21 | 10,50 | -1,70 | 1,90 | -0,90 |
| (Pr. 7 xDupn.126) | 8,6± 0,19 | 12,8 ± 0,21 | 11,8 ± 0,21 | 10,70 | 1,10 | 2,0 | 0,52 |

## CONCLUSIONS

The inheritance of plant height in three hybrid crosses was incompletly dominant and direction of the gene effects was prevalling towards the parent with a lower height - Prilep 7.

The mode of inheritance in the number of leaves was completely dominant, incompletely dominant and overdominant towards the parent with lower leaf number. The modes of inheritance of the 14-th leaf length are dominance, incomplete dominance and overdominance, towards the parent with longer and wider leaves.

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ISSN 0494-3244
Тутун/Tobacco Vol. 67 No 7-12, 26-34, 2017 UDC:633.71-152.75(497.2)"2014/2016"
Original scientific paper

# MANIFESTATION OF HETEROSIS AND INFLUENCE OF THE DIRECTION OF CROSSING UPON SOME BIOMETRICAL CHARACTERS IN HYBRID COMBINATIONS OF BURLEY TOBACCO

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## ABSTRACT

Manifestations of true and hypothetical heterosis were studied in terms of plant height and leaf number, length and width in twenty hybrid combinations of Burley tobacco. Each of them is represented by its straight and back crossing. Heterosis for plant height was poorly developed and has no significant importance for this character. Higher heterosis was observed for leaf number and it has practical significance in the selection of Burley tobacco. It becomes even more pronounced when parental components of domestic line and introduced variety are used in the crosses. Heterotic effect was insignificant in relation to leaf length. Heterosis for leaf width and length was higher than for leaf length, but lower compared to leaf number. If heterosis occurs for leaf number, it cannot be observed for leaf size, and vice versa. The results show that heterosis appears to be promising for increasing the width and especially the number of leaves in Burley tobacco. There was no significant effect of the direction of crossing upon the manifestations of heterosis for the studied characters and some low effect was observed only for leaf width.

**Keywords:** Burley tobacco, heterosis, plant height, leaf number, leaf length and width, direction of crossing

## ВЛИЈАНИЕ НА ХЕТЕРОЗИСОТ И НА НАСОКАТА НА ВКРСТУВАЊЕ ВРЗ НЕКОИ БИОМЕТРИСКИ ПОКАЗАТЕЛИ КАЈ ХИБРИДНИТЕ КОМБИНАЦИИ НА БЕРЛЕЈСКИ ТУТУН

Испитувани се ефектите на вистинскиот и хипотетичкиот хетерозис за својствата висина на растенијата, број на листови и должина и ширина на листот на дваесет хибридни комбинации. Секоја хибридна комбинација е претставена и со нејзината реципрочна комбинација. Слабо изразен хетерозис се јавува во однос на висината на растенијата и тој нема големо значење за овој показател. Хетерозисот за бројот на листови е посилно изразен и има посилно влијание и поголемо практично значење во селекцијата на типот берлеј. Тој е уште поизразен кога за родители во крстоските се користат локални и интродуцирани сорти. Во поглед на должината на листот, хетерозисот е многу слабо изразен и нема никакво значење. Многу поизразено хетеротично влијание има за ширината/должината на листот отколку за должината, но тоа е сепак помало во споредба со бројот на листови. Таму каде што се појавува хетерозис во однос на својството број на листови, не може да се забележи за својството димензии на листот, и обратно. Резултатите покажуваат дека хетерозисот е перспективен за зголемување на ширината, а најмногу за бројот на листови кај берлејските тутуни. Насоката на вкрстување нема значајно влијание врз појавата на хетерозис во однос на проучуваните својства, а послабо влијание е забележано само за ширината на листот.

**Клучни зборови**: берлејски тутун, хетерозис, висина на растението, број на листови, должина и ширина на листот

## INTRODUCTION

Dynamics of the foreign markets and the increasing demands for tobacco varieties constantly put new tasks before the tobacco selectionists (Masheva, 2007; Risteski et al., 2012). Heterosis provides very good and effective opportunities in creation of new varieties (Pophristev, 1977).

Heterosis refers to the biological phenomenon that hybrids of $F_1$ progeny exhibit greater output, viability and productivity than their initial parental forms (Yankulov, 1996). Beside general increase, it is associated with accelerated growth and development, increased yields, change of chemical composition, etc. (Delchev et al., 1996; Docheva, 2016; Aycock and McKee, 1985).

Heterosis has been determined in all types of tobacco throughout the world (Gelemerov, 1990). Manifestation of heterosis in tobacco is a subject to a number of studies (Ramanarao et al., 1993, Kara and Esendae, 1995; Prasannasimha Rao, 1995; Zi Cheng Xu and Jun Zhu, 1999; Gixhari and Canllari, 2004).

Most of them reported that heterosis mainly increases the vegetative mass and the growth rate (Lee and Chang, 1984). For large-leaf varieties of tobacco it is very important to have rapid initial growth and development, in view of the shortened stage of rosette, when tobacco is most vulnerable (Pophristev, 1977; Shabanov and Tomov, 1989).

There is little information on the influence of the direction of crossing upon the occurrence of heterosis in tobacco and even those that are available are very devergent and contradictory (Butorac, 2000; Vassilev and Dimitrova 2010; Stankev , 1988; Dimitrova and Vasilev, 2010).

In large-leaf tobacco, heterosis contributes to the intensification of selection process and therefore it deserves further investigation (Manolov, 2000; Shabanov and Tomov, 1989; Prasannasimha Rao et al., 1990).

The aim of this research is to study heterosis and to assess the influence of the direction of crossing upon the most important biometric characters in some hybrid combinations of Burley tobacco.

## MATERIAL AND METHODS

The experimental work was carried out in the period 2014 – 2016 at the trial field of TTPI – Markovo, with 20 hybrid combinations in F1 created in the Institute: Hybrid 1571 (L 1410 x B 1344) and Hybrid 1571 A (B 1344 x L1410); Hybrid 1572 (L 1410 x Ky 17) and Hybrid 1572 (Ky 17 x L 1410); Hybrid 1573 (L 1349 x B 1344) and Hybrid 1573 (B 1344 x L 1349); Hybrid 1574 (L 1349 x Tn 90) and Hybrid 1574 (Tn 90 x L 1349); Hybrid 1575 (L 1349 x Tn 86) and Hybrid 1575 (Tn 86 x L 1349); Hybrid 1576 (L 1399 x Ky 907) and hybrid 1576 (Ky 907 x L 1399); Hybrid 1577 (B 1344 x Ky 17) and Hybrid 1577 (Ky 17 x B 1344); Hybrid 1578 (L 1409 x Ky 17) and Hybrid 1578 (Ky 17 x L 1409); Hybrid 1579 (Tn 90 x B 1344) and Hybrid 1579 (B 1344 x Tn 90); Hybrid 1580 (L 1322 x Tn 90) and Hybrid 1580 (Tn 90 x L 1322).

Each hybrid combination is represented by its straight and backcross variant, so that the mother component in the straight cross is a father component in the backcross. The crosses were composed of parental components both from approved varieties and from our perspective lines. Assessment was made of all variants for plant height, leaf number, leaf length and width.

The obtained data were analysed using the analysis of variance (ANOVA). Arithmetic mean (x) and arithmetic error (Sx%) were also calculated and hypothetical and true heterosis were determined by Omarov (1975).

## RESULTS AND DISCUSSION

According to the results on heterotic effect on plant height, the values for true heterosis in all hybrid combinations are lower than those for hypothetical heterosis (Table №1). Differences in values between the two indicators, however, were insignificant in all crosses. True heterosis is more accurate model of selection practice, so its occurrence is more important. High values for true heterosis were obtained in only 5 of the 20 crosses tested and it was the most significant in Hybrid 1579 (B 1344 x T H 90). In 1574 Hybrid (T H 90 x L 1349) and Hybrid 1577 (B 1344 x Ky 17) heterosis is on the borderline of significance. In several variants, the values of hybrids in $F_1$ generation are almost equal to those of the parents. Heterosis for the character plant height is poorly developed and has no significant importance.

The influence of the direction of crossing upon plant height was almost negligible.

**Table 1. Manifestations of heterosis in relation to plant height**

| **Parents / Crosses** | **$P_1$** x±Sx% | **$P_2$** x±Sx% | **$F_1$** x±Sx% | **HP** hypothetical % | **HP** real % |
|---|---|---|---|---|---|
| Hybrid 1571 (L 1410 x B 1344) | 163,7±0,59 | 166,3±0,60 | 168,1±0,61 | 1,9 | 1,1 |
| Hybrid 1571 A (B 1344 x L1410) | 166,3±0,60 | 163,7±0,59 | 166,8±0,60 | 1,1 | 0,3 |
| Hybrid 1572 (L1410 x Ky 17) | 163,7±0,59 | 164,6±0,59 | 169,6±0,61 | 3,3 | 3 |
| Hybrid 1572A (Ky 17 x L 1410 ) | 164,6±0,59 | 163,7±0,59 | 171,3±0,61 | 4,4 | 4,1 |
| Hybrid 1573 (L 1349 x B1344) | 168,3±0,61 | 166,3±0,60 | 168,4±0,61 | 0,7 | 0,1 |
| Hybrid 1573A (B 1344 x L 1349) | 166,3±0,60 | 168,3±0,61 | 169,2±0,61 | 1,1 | 0,5 |
| Hybrid 1574 (L 1349 x Tn 90) | 168,3±0,61 | 164,1±0,59 | 174,6±0,63 | 5,1 | 3,7 |
| Hybrid 1574A (Tn 90 x L 1349) | 164,1±0,59 | 168,3±0,61 | 176,8±0,64 | 6,4 | 5,1 |
| Hybrid 1575 (L1349 x Tn 86) | 168,3±0,61 | 161,8±0,58 | 170,7±0,61 | 3,4 | 1,4 |
| Hybrid 1575A (Tn 86 x L 1349) | 161,8±0,58 | 168,3±0,61 | 173,8±0,63 | 5,3 | 3,3 |
| Hybrid 1576 (L1399 x Ky 907) | 160,7±0,58 | 169,7±0,61 | 172,2±0,18 | 4,2 | 1,5 |
| Hybrid 1576A | 169,7±0,61 | 160,7±0,58 | 174,2±0,63 | 5,4 | 2,7 |

| | | | | | |
|---|---|---|---|---|---|
| (Ky 907 x L 1399) | | | | | |
| Hybrid 1577 (Б 1344 x Ky 17) | 166,3±0,60 | 164,6±0,59 | 174,6±0,63 | 5,5 | 5 |
| Hybrid 1577A (Ky 17 x B 1344) | 164,6±0,59 | 166,3±0,60 | 171,8±0,62 | 3,9 | 3,3 |
| Hybrid 1578 (L 1409 x Ky 17) | 162,5±0,59 | 164,6±0,59 | 174,3±0,63 | 6,6 | 5,9 |
| Hybrid 1578A (Ky 17 x L 1409) | 164,6±0,59 | 162,5±0,59 | 176,1±0,63 | 7,7 | 7 |
| Hybrid 1579 (Tn 90 x B 1344) | 164,1±0,59 | 166,3±0,60 | 177,3 ±0,64 | 7,3 | 6,6 |
| Hybrid 1579A (B 1344 x Tn 90) | 166,3±0,60 | 164,1±0,59 | 178,7±0,64 | 8,2 | 7,5 |
| Hybrid 1580 (L 1322 x Tn 90) | 167,7±0,60 | 164,1±0,59 | 176,0±0,63 | 6,1 | 4,9 |
| Hybrid 1580A (Tn 90 x L 1322) | 164,1±0,59 | 167,7±0,60 | 175,6±0,63 | 5,8 | 4,7 |

With regard to leaf number, the values for true heterosis in all hybrid combinations were lower than those for hypothetical heterosis (Table 2). In this case, however, the differences between the two indicators were more pronounced, especially in Hybrid 1571 (L 1410 x B 1344) and Hybrid 1571 A (B 1344 x L1410). True heterosis with significant values was obtained in 9 of 20 tested crosses but the most pronounced it was in Hybrid 1578 (Ky x 17 L 1409). Beside this, heterosis of over 10% was estimated in two other crosses. Manifestation of heterosis for the number of leaves corresponds to certain degree to that for plant height, which is logical given the strong positive correlation between these two characters. With respect to this character, heterotic effect is more pronounced and has a practical importance in the selection of Burley tobacco.

Heterosis is more pronounced when both domestic line and introduced variety are used as parental components in the cross.

The influence of the direction of crossing upon the number of leaves is very low. The results show that there is no significant influence of the direction of crossing upon the manifestation of characters plant height and leaf number in Burley tobacco crosses.

**Table 2. Manifestations of heterosis in relation to leaf number**

| **Parents / Crosses** | **P₁** x±Sx% | **P₂** x±Sx% | **F₁** x±Sx% | **HP** hypothetical % | **HP** real % |
|---|---|---|---|---|---|
| Hybrid 1571 (L 1410 x B 1344) | 23,8±0,11 | 29,6±0,14 | 29,9±0,14 | 12 | 1 |
| Hybrid 1571 A (B 1344 x L1410) | 29,6±0,14 | 23,8±0,11 | 29,7±0,14 | 11,2 | 0,3 |
| Hybrid 1572 (L1410 x Ky 17) | 23,8±0,11 | 25,5±0,12 | 27,2±0,13 | 10,3 | 6,6 |
| Hybrid 1572A (Ky 17 x L 1410 ) | 25,5±0,12 | 23,8±0,11 | 27,8±0,13 | 12,8 | 9 |

| | | | | | |
|---|---|---|---|---|---|
| Hybrid 1573 (L 1349 x B1344) | 27,7±0,13 | 29,6±0,14 | 29,8±0,14 | 4 | 0,7 |
| Hybrid 1573A (B 1344 x L 1349) | 29,6±0,14 | 27,7±0,13 | 30,2±0,14 | 5,4 | 2 |
| Hybrid 1574 (L 1349 x Tn 90) | 27,7±0,13 | 27,4±0,13 | 30,4±0,14 | 10,3 | 9,7 |
| Hybrid 1574A (Tn 90 x L 1349) | 27,4±0,13 | 27,7±0,13 | 30,6±0,14 | 11,1 | 11 |
| Hybrid 1575 (L1349 x Tn 86) | 27,7±0,13 | 26,2±0,12 | 28,0±0,13 | 3.9 | 1,1 |
| Hybrid 1575A (Tn 86 x L 1349) | 26,2±0,12 | 27,7±0,13 | 28,4±0,13 | 5,4 | 2,5 |
| Hybrid 1576 (L1399 x Ky 907) | 25,4±0,12 | 28,0±0,13 | 28,6 ±0,13 | 7,1 | 2,2 |
| Hybrid 1576A (Ky 907 x L 1399) | 28,0±0,13 | 25,4±0,12 | 29,1±0,14 | 9 | 3,9 |
| Hybrid 1577 (Б 1344 x Ky 17) | 29,6±0,14 | 25,5±0,12 | 30,7±0,14 | 11,4 | 3,7 |
| Hybrid 1577A (Ky 17 x B 1344) | 25,5±0,12 | 29,6±0,14 | 30,2±0,14 | 9,6 | 2 |
| Hybrid 1578 (L 1409 x Ky 17) | 26,3±0,12 | 25,5±0,12 | 29,1±0,14 | 12,8 | 10,6 |
| Hybrid 1578A (Ky 17 x L 1409) | 25,5±0,12 | 26,3±0,12 | 29,5±0,14 | 14,3 | 12,2 |
| Hybrid 1579 (Tn 90 x B 1344) | 27,4±0,13 | 29,6±0,14 | 31,3±0,15 | 9,8 | 5,7 |
| Hybrid 1579A (B 1344 x Tn 90) | 29,6±0,14 | 27,4±0,13 | 31,5±0,15 | 10,5 | 6,4 |
| Hybrid 1580 (L 1322 x Tn 90) | 27,8±0,13 | 27,4±0,13 | 29,4±0,14 | 6,5 | 5,8 |
| Hybrid 1580A (Tn 90 x L 1322) | 27,4±0,13 | 27,8±0,13 | 29,1±0,14 | 5,4 | 4,7 |

The results on the influence of heterosis upon the leaf length show that the values for true heterosis in all hybrid combinations are lower than those for hypothetical heterosis (Table 3). Differences, however, between the two indicators in all tested crosses are insignificant. True heterosis with significant values was obtained only in 1 of the 20 crosses tested - Hybrid 1577 (B 1344 x Ky 17), where it is on the borderline of significance. Generally, the heterotic effect for leaf length was very poor and insignificant.

The effect of straight cross and backcross was much poorly manifested with regard to leaf length.

**Table 3. Manifestations of heterosis in relation to leaf length**

| Parents / Crosses | $P_1$ x±Sx% | $P_2$ x±Sx% | $F_1$ x±Sx% | HP hypothetical % | HP real % |
|---|---|---|---|---|---|
| Hybrid 1571 (L 1410 x B 1344) | 60,8±0,19 | 62,1±0,20 | 63,9±0,20 | 4 | 2,8 |
| Hybrid 1571 A (B 1344 x L1410) | 62,1±0,20 | 60,8±0,19 | 64,6±0,21 | 5,1 | 4 |
| Hybrid 1572 (L1410 x Ky 17) | 60,8±0,19 | 60,3±0,19 | 62,3±0,20 | 2,9 | 2,5 |
| Hybrid 1572A (Ky 17 x L 1410 ) | 60,3±0,19 | 60,8±0,19 | 61,7±0,20 | 1,9 | 1,5 |
| Hybrid 1573 (L 1349 x B1344) | 61,8±0,19 | 62,1±0,20 | 64,2±0,21 | 3,6 | 3,4 |
| Hybrid 1573A (B 1344 x L 1349) | 62,1±0,20 | 61,8±0,20 | 65,1±0,21 | 5,1 | 4,8 |
| Hybrid 1574 (L 1349 x Tn 90) | 61,8±0,20 | 61,6±0,20 | 62,4±0,20 | 1,1 | 1 |
| Hybrid 1574A (Tn 90 x L 1349) | 61,6±0,20 | 61,8±0,20 | 62,2±0,20 | 0,8 | 0,6 |
| Hybrid 1575 (L1349 x Tn 86) | 61,8±0,20 | 60,9±0,19 | 63,3±0,20 | 3,2 | 2,4 |
| Hybrid 1575A (Tn 86 x L 1349) | 60,9±0,19 | 61,8±0,19 | 63,0±0,20 | 2,7 | 1,9 |
| Hybrid 1576 (L1399 x Ky 907) | 61,1±0,19 | 61,7±0,20 | 64,3 ±0,21 | 4,7 | 4,2 |
| Hybrid 1576A (Ky 907 x L 1399) | 61,7±0,20 | 61,1±0,19 | 63,7±0,20 | 3,7 | 3,2 |
| Hybrid 1577 (Б 1344 x Ky 17) | 62,1±0,20 | 60,3±0,19 | 65,3±0,21 | 6,7 | 5,2 |
| Hybrid 1577A (Ky 17 x B 1344) | 60,3±0,19 | 62,1±0,20 | 64,5±0,21 | 5,4 | 3,9 |
| Hybrid 1578 (L 1409 x Ky 17) | 61,5±0,20 | 60,3±0,19 | 61,7±0,20 | 1,3 | 0,3 |
| Hybrid 1578A (Ky 17 x L 1409) | 60,3±0,19 | 61,5±0,20 | 62,1±0,20 | 2 | 1 |
| Hybrid 1579 (Tn 90 x B 1344) | 61,6±0,20 | 62,1±0,20 | 64,2±0,21 | 3,8 | 3,4 |
| Hybrid 1579A (B 1344 x Tn 90) | 62,1±0,20 | 61,6±0,20 | 64,8±0,21 | 4,8 | 4,3 |
| Hybrid 1580 (L 1322 x Tn 90) | 62,6±0,20 | 61,6±0,19 | 63,1±0,20 | 1,6 | 0,8 |
| Hybrid 1580A (Tn 90 x L 1322) | 61,6±0,20 | 62,6±0,20 | 62,8±,0, 20 | 1,1 | 0,3 |

With regard to leaf width, the true heterosis in all tested variants was lower than the hypothetical heterosis (Table 4), in which case the differences in the values of the above indicators are insignificant.

Heterotic effects for leaf width are more pronounced than for leaf length. Significant positive heterosis was observed in 7 out of 20 tested combinations in $F_1$, indicating that heterosis is a promising mode for increasing the leaf width in Burley tobacco. The most pronounced heterosis was observed in Hybrid 1577 (B 1344 x Ky 17). In general, however, the heterosis for the character leaf width is less developed than for the number of leaves. If heterosis occurs for the leaf number, it does not occur for leaf size, and vice versa.

In cases where Burley1344 variety was used as a parent component, the values of heterosis were higher.

The influence of the true heterosis and backcross variant in this case is slightly more pronounced. Significant heterotic effect was observed only in the first cross, compared to the reciprocal cross in two of the investigated hybrid pairs, although the difference in values is not strongly expressed.

In all crosses with Burley 1344 variety, heterotic effect is more pronounced when it occurs as parental component - pollinator.

**Table 4. Manifestations of heterosis in relation to leaf width**

| **Parents / Crosses** | **$P_1$** x±Sx% | **$P_2$** x±Sx% | **$F_1$** x±Sx% | **HP** hypothetical % | **HP** real % |
|---|---|---|---|---|---|
| Hybrid 1571 (L 1410 x B 1344) | 31,2±0,14 | 33,1±0,15 | 34,2±0,15 | 6,4 | 3,3 |
| Hybrid 1571 A (B 1344 x L1410) | 33,1±0,15 | 31,2±0,14 | 34,6±0,15 | 7,6 | 4,5 |
| Hybrid 1572 (L1410 x Ky 17) | 31,2±0,14 | 30,8±0,14 | 33,2±0,15 | 7,1 | 6,4 |
| Hybrid 1572A (Ky 17 x L 1410 ) | 30,8±0,14 | 31,2±0,14 | 32,9±0,14 | 6,1 | 5,4 |
| Hybrid 1573 (L 1349 x B1344) | 32,5±0,14 | 33,1±0,15 | 34,4±0,15 | 4,9 | 3,9 |
| Hybrid 1573A (B 1344 x L 1349) | 33,1±0,15 | 32,5±0,14 | 35,0±0,15 | 6,7 | 5,7 |
| Hybrid 1574 (L 1349 x Tn 90) | 32,5±0,14 | 31,9±0,14 | 32,7±0,14 | 1,6 | 0,6 |
| Hybrid 1574A (Tn 90 x L 1349) | 31,9±0,14 | 32,5±0,14 | 32,8±0,14 | 1,9 | 0,9 |
| Hybrid 1575 (L1349 x Tn 86) | 32,5±0,14 | 33,4±0,15 | 35,2±0,15 | 6,8 | 5,4 |
| Hybrid 1575A (Tn 86 x L 1349) | 33,4±0,15 | 32,5±0,14 | 35,5±0,16 | 7,7 | 6,3 |
| Hybrid 1576 (L1399 x Ky 907) | 31,7±0,14 | 33,2±0,15 | 33,6±0,15 | 3,5 | 1,2 |
| Hybrid 1576A (Ky 907 x L 1399) | 32,7±0,14 | 31,7±0,14 | 33,9±0,15 | 4,5 | 2,1 |

| | | | | | |
|---|---|---|---|---|---|
| Hybrid 1577 (Б 1344 x Ky 17) | 33,1±0,15 | 32,7±0,14 | 35,1±0,15 | 6,7 | 6 |
| Hybrid 1577A (Ky 17 x B 1344) | 30,8±0,14 | 33,1±0,15 | 34,6±0,15 | 5,2 | 4,5 |
| Hybrid 1578 (L 1409 x Ky 17) | 32,3±0,14 | 30,8±0,14 | 33,5 ±0,15 | 6,2 | 3,7 |
| Hybrid 1578A (Ky 17 x L 1409) | 30,8±0,14 | 32,3±0,14 | 33,3±0,15 | 5,5 | 3,1 |
| Hybrid 1579 (Tn 90 x B 1344) | 31,9±0,14 | 33,1±0,15 | 34,6±0,15 | 6,5 | 4,5 |
| Hybrid 1579A (B 1344 x Tn 90) | 33,1±0,15 | 31,9±0,14 | 35,6±0,16 | 9,5 | 7,6 |
| Hybrid 1580 (L 1322 x Tn 90) | 32,2±0,14 | 31,9±0,14 | 32,8±0,14 | 2,3 | 1,9 |
| Hybrid 1580A (Tn 90 x L 1322) | 31,9±0,14 | 32,2±0,14 | 32,4 ±0,14 | 1,1 | 0,6 |

The obtained results on leaf size indicate that the direction of crossing has a poor effect upon manifestation of heterosis on the investigateded characters in hybrid combinations of Burley tobacco and has no practical importance.

## CONCLUSION

Poorly expressed heterosis was observed for the character plant height and it is not of significant importance.

Significantly more pronounced heterosis was observed for leaf number and it has practical importance in the selection of Burley tobacco.

In hybrid combinations of Burley tobacco, the heterotic effect for leaf width is of economic importance and is much higher than for leaf length.

The use of heterosis is a promising method for increasing the leaf size, but only in respect of their width.

In crosses where heterotic effect was observed for the character leaf number, no such effect was observed on leaf size, and vice versa.

The direction of crossing has almost no influence on the manifestation of heterosis in relation to the investigated characters and it shows some significance only for leaf width.

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ISSN 0494-3244
Тутун/Tobacco Vol. 67 No 7-12, 35-45, 2017 UDC:633.71-153.037:632.48]:632.937.1(497.775)
Original scientific paper

# TOBACCO SEEDLINGS PROTECTION FROM DAMPING OFF DISEASE WITH *TRICHODERMA HARZIANUM* T22 (BIOPRODUCT TRIANUM P)

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## ABSTRACT

Modern tobacco protection follows the principles of sustainable agricultural production and supports the use of environment-friendly methods.

*Trichoderma harzianum* T22 is the most powerful biocontrol agent. The aim of our research was to study its ability to control the causing agents of damping off disease in tobacco seedlings.

Trianum P is biological fungicide which contains spores of *T. harzianum* T22. It was applied in a rate of 15 g/10m$^2$ through three modes of application and with two additional treatments by the time of planting. Investigations were made in biolaboratory and in seedbeds, with natural infestation. The biofungicide was applied by storing the seed in suspension 48h before sowing, fungicide application over the soil and first application 15 days after sowing. All variants in seedbeds were set up with and without herbicide. In the biolab tests, treatment of infested soil was included.

Generally, the best results in the control of damping off were achieved in variants where the seed was kept in the product (with and without herbicide-in seedbeds). The poorest results were obtained with the fungicide first application 15 days after sowing. It can be concluded that *T. harzianum* T22 is biocontrol agent which can be used in protection of tobacco seedlings from the damping off disease. Biological product Trianum P has a good perspective in sustainable tobacco production.

**Keywords:** *Trichoderma harzianum* T22, Trianum P, damping off disease, intensity of attack

## ЗАШТИТА НА ТУТУНСКИОТ РАСАД ОД БОЛЕСТА СЕЧЕЊЕ СО *TRICHODERMA HARZIANUM* T22 (БИОПРЕПАРАТ TRIANUM P)

Современото производство на тутун ги следи принципите на одрживото земјоделско производство и ја поддрржува примената на еколошко-прифатливи методи.

*Trichoderma harzianum* T22 е најмоќниот биоконтролен агенс. Нашата цел беше да се испита неговата способност во сузбивањето на предизвикувачите на болеста сечење на тутунскиот расад.

Trianum P е биопрепарат кој содржи живи спори на *T. harzianum* T22. Тој беше аплициран во доза од 15 g/m$^2$ на три начини и две дополнителни третирања до времето на расадување. Испитувањата беа извршени во биолошка лабораторија и тутунски леи –природна инфекција. Биофунгицидот беше аплициран преку: семе чувано во препаратот 48 часа пред сеидба, апликација на препаратот врз почвата и прва апликација по 15 дена по сеидба. Сите варијанти во леите беа поставени со и без хербицид. Во биолошка лабораторија беше вклучена и варијанта со инфицирана почва.

Најдобри резултати во заштитата од болеста сечење беа постигнати кај варијантите каде семето беше чувано во препаратот (во леи - со и без хербицид), а најслаби при неговата прва апликација 15 дена по сеидбата.

*T. harzianum* T22 е биоконтролен агенс кој може да се употребува во заштитата на тутунскиот расад од болеста сечење. Биопрепаратот Trianum P има добра перспектива во одрживото производство на тутун.

**Клучни зборови**: *Trichoderma harzianum* T22, Trianum P, сечење, интензитет на напад

## INTRODUCTION

Production of tobacco seedlings can be seriously affected, mostly by fungal diseases such as damping off. The causing agents of the disease are the pathogenic fungi *Rhizoctonia solani* and *Pythium debarianum*. The control is difficult because of their ability to survive in the soil, the large number of hosts etc. Therefore, biological control is important segment in protection from this disease, but also the only acceptable measure in the case of dual infection and inadequate choice of fungicides.

Biological control is an environmentally acceptable measure and a real alternative to the use of pesticides.

Biological control offers an environment-friendly approach to the management of plant disease and can be incorporated into cultural and physical measures with limited usage of chemicals for effective integrated managament (IPM) system (Monte, 2001).

Biological control is a potential alternative to the use of chemicals which are harmful to the environment, humans and other living organisms including beneficial natural enemies (Chet and Inbar, 1994). The most detailed description of the use of biological control is the supression of pathogens by a single antagonist in a single crop system. Biological control means application of biological processes to reduce the level of inoculum in order to reduce the risk of disease intensity.

There are numerous literature data and reliable, accepted solutions for biological control of diseases caused by phytopathogenic fungi by application of the fungi of the genus *Trichoderma*, which is considered a reliable alternative that can replace the chemical control (Cuervo-Parra et al., 2015).

*Trichoderma* spp. are found in soil and root system and are beneficial to plants (Goes, 2002; Contreras-Cornejo et al., 2009). The most important benefit of *Trichoderma* is that they directly attack and control the causing agents of the diseases (Chet et al., 2006).

They are opportunistic, avirulent plant symbionts as well as parasites of other fungi. Colonizing the root, *Trichoderma spp*. activates numerous mechanisms that attack pathogens and stimulate the growth of plants and roots. On the other hand, those agents have a stimulating effect on the development of root and plant. In the complex of actions, antibiosis, mycoparasitism and food competence are considered the main mechanisms for biological control (Harman, 2004, 2006; Benitez et al., 2004; Howell, 2003).

*Trichoderma* is effective in the control of numerous plant pathogens (Harman, 2004, 2006).

It acts against seed borne and soilborne fungal pathogens, including the causing agents of seed rot, damping off and root rot disease (Heydari and Pessarakli, 2010).

Nowadays there are advanced techniques for application of *Trichoderma* in agriculture. There are numerous preparations, but the best known among those that use antagonistic microorganisms are the preparations based on various strains of *Trichoderma*. *T.harzianum* T22. is widely accepted because of its characteristics: efficient control of a large number of soil diseases, improved growth and development of plant root and increased absorption of water and nutrients from the soil, rapid growth on roots of all cultivated plants, good development in various soil types, tolerance to wide temperature and pH range and compatibility with a large number of active ingredients. According to Harman (2004), it is more secure for the farmers and provides longer and cheaper protection compared to pesticides application.

Biological product Trianum P is based on spores of *Trichoderma harzianum* T22, in form of water soluble powder or granules for direct use. It is registered as a biostimulator and plant protection agent in many countries, including the Republic of Macedonia.

The appropriate time and model of application of the biocontrol agent is very important for successful biocontrol. Development of feasible and efficient delivery system for application of appropriate microorganisms in the ecosystem is an important component of biocontrol technology (Lewis et al., 1998).

Handelsman and Stabb (1996) reported that colonization is essential in biocontrol and they found a relationship between the population size of the biocontrol agent and the degree of disease suppression. According to Heydari and Pessarakli (2010), one of the criteria for successful biocontrol is to determine the factors of successful colonization and expression of biocontrol traits. Delivery system must ensure that biocontrol agent will grow well and achieve its purpose. So, delivery and application processes must be developed for each crop (Harman, 1996). Therefore, our aim was to determine how long the biocontrol agent retains in the soil, to evaluate the colonisation by various modes of application and how its quantity affects the damping off disease in seedlings.

The purpose of this study is to determine the effect of *T. harzianum* T22 on tobacco, i.e. the effect of the biofungicide Trianum P in the control of damping off disease. The research should also identify the most efficient model of application in the standard way of sowing.

The results of these investigations will contribute to commercial use of this biocontrol agent in protection of tobacco seedlings, IPM of tobacco and sustainable tobacco production.

## MATERIAL AND METHODS

Investigations of the influence of Trianum P on the intensity of damping off disease in tobacco seedlings were carried out in Scientific Tobacco Institute-Prilep, in biolaboratory and field conditions.

The intensity of the attack was monitored in conditions of natural infestation.

Assessment of disease severity was made by measuring the infested area in each replication of the variants. The intensity of disease is expressed in percentage of infested area- the mean value of the three replications.

### Biolaboratory tests

The experiment in the biological laboratory was set up in 3 replications for each variant including the check. The area of one replication was $0.3m^2$. Seed was sown in the norm of $0,6\ g/m^2$. The experiment in the biological laboratory was performed twice.

Trianum P biological fungicide was applied at a recommended rate of 15 g/10 $m^2$ (0,5 g for $0,3m^2$). According to the instructions, the preparation was previously dissolved in 1:5 ratio and added to the water by mixing. 800 ml water was used per replication.

The soil in Variant 4 was previously infested with a pure culture of the causing agents of disease - *Rhizoctonia solani and Pythium debarianum (*one Petri dish per pathogen for $0,3m^2$).

Seed for Variant 3 was prepared for sowing the previous day. 5 g of the product was dissolved in 25 ml water. The seed was stored in the suspension for 24 hours.

The occurrence of damping off was observed after the second treatment, but according to the applied method of work, the third treatment was done prior to evaluation.

**Intensity of the damping off disease** i.e. the percentage of infested area was estimated after the third treatment.

## Seedbeds tests

Investigations in seedbed conditions were performed with the following variants:

- Ø Check - standard treatment with herbicide and nutrition; no fungicide application
1. Treatment after 15 days
2. Treatment after 15 days (post-sowing - herbicide)
3. Pre-sowing
4. Pre-sowing (post-sowing - herbicide)
5. Seed treated with Trianum P
6. Seed treated with Trianum P (post-sowing - herbicide)

Seed amount of 6.75 g/10 $m^2$ i.e. 0.67 g/$m^2$ was used for planting. All variants were set up in 3 replications and each replication was 3.33 $m^2$.

Since the purpose of this research was to determine not only the effect of the preparation, but also the possible application in the standard way of sowing (with herbicide), all the variants were replicated, i.e. set up with the use of herbicide.

Herbicide Gamit (a.i. clomazone) in a rate 0.07 ml/$m^2$ was used after sowing.

**The application of Trianum P biopreparation**

Trianum P biological fungicide was applied at a recommended rate of 15 g/10 $m^2$ (1.5 g/$m^2$ ) using 2.5-5l water/$m^2$.

- Soil in variants 3 and 4 was treated prior to sowing with Trianum P at the same rate, i.e. 5 g/3.33$m^2$ using 10 l of water.

- Seed in variants 5 and 6 was prepared for sowing the previous day as well as in the biolaboratory tests.

Variants 1 and 2 were treated 15 days after sowing, at the same rate as other variants.

During the growing season, two more treatments were carried out on the seedling at 15-20 day intervals, by which all stages of seedling growth were covered. Thus, seedling of the variants 1 and 2 received 2 treatments and that of other variants 3 treatments.

Health condition of tobacco seedling was monitored throughout the growing season.

**The intensity of damping off disease**

The first assessment on the intensity of disease was made 10 days after the third treatment and the second assessment a few days before planting.

**The effectiveness of Trianum P in the control of damping off disease** was calculated by Abbott's formula (from the values for the intensity of disease attack in variants and check in the second estimation).

## RESULTS AND DISSCUSION

### Biolaboratory tests

The highest intensity of attack in the first experiment was recorded in the check. Among the variants with application of Trianum P, the lowest value was found in the variant where the seed was stored in the preparation (Table 1). The highest value was found in the variant where the first treatment was done after 15 days of sowing.

**Table 1. The influence of Trianum P on the intensity of damping off disease (Ist experiment)**

| | Variant | Percentage of infested area | | | Mean value |
|---|---|---|---|---|---|
| | | Replication | | | |
| | | I | II | III | |
| Ø | Check | 1,20 | 8,23 | 6,7 | 5,38 |
| 1 | First treatment after 15 days | 2,56 | 1,53 | 6,00 | 3,36 |
| 2 | Treatment of soil before sowing | 2,23 | 3,53 | 3,90 | 3,22 |
| 3 | Seed treated with the biopreparation | 0,3 | 1,43 | 5,4 | 2,37 |
| 4 | Treatment of infested soil | 8,06 | 0,4 | 0 | 2,82 |

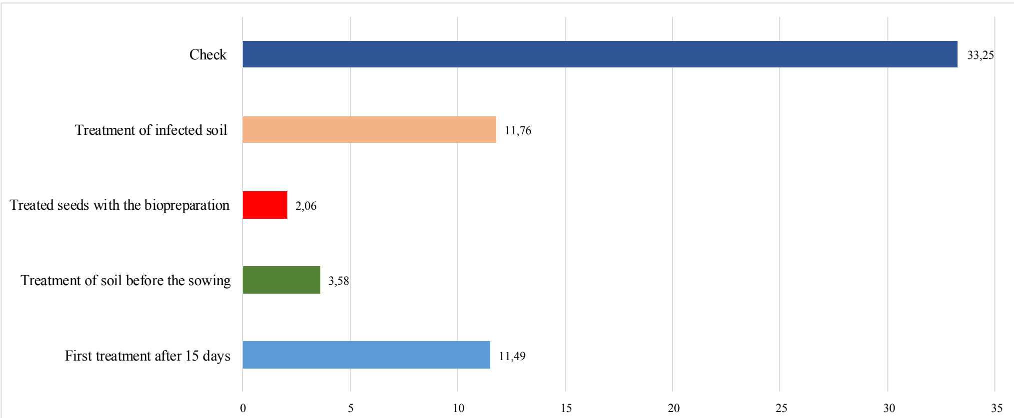
In the second experiment, the intensity of attack was much higher than in the first experiment (Table 2). However, the lowest percentage of infested area was detected in variant 3, when the seed was stored in the preparation before sowing. Also, when the soil was treated before sowing, the intensity of disease was low (3.93%). Delayed treatment of seedlings did not produce good results (20.15%). The same situation was observed in Variant 4, when the preparation was applied in infested soil. Results obtained for the variants in biolaboratory test are presented in Figures 1-4.


**Figures 1-4. Variants in biolaboratory test**

**Table 2. Influence of Trianum P on the intensity of damping off disease (IInd experiment)**

| | Variant | Percentage of infested area | | | Mean value |
|---|---|---|---|---|---|
| | | Replication | | | |
| | | I | II | III | |
| Ø | Check | 13,33 | 80,00 | 90,00 | 61,11 |
| 1 | First treatment after 15 days | 7,66 | 2,80 | 50,00 | 20,15 |
| 2 | Treatment of soil before the sowing | 7,46 | 4,33 | 0 | 3,93 |
| 3 | Treated seeds with the biopreparation | 4,80 | 0,30 | 0,13 | 1,74 |
| 4 | Treatment of infested soil | 7,66 | 2,80 | 50,00 | 20,15 |

The effectiveness of the biopreparation Trianum P applied by various application models in the biolaboratory test (mean value of the two trials) is presented in Graph 1.




**Graph. 1 The effectiveness of variants in biolaboratory test**

Treatment of infested soil didn't give satisfactory results. This is understandable bearing in mind the principle of action of this preparation. Actually, its activity is particularly pronounced in the root zone and is closely related to the development of the root system. For effective control, antagonist must be previously established in plant ecosystems and remain active in target pathogens during periods favourable for plant infection. (Heydari and Peskarli, 2010).

If species of the genus *Trichoderma* are applied directly in the soil or by seed, they grow up simultaneously by the root system of treated plants (Harman, 2000, Howell et al., 2000). There is abundant and constant quantity of root exudations from the root tips during root development but there was no increase after shoots emerged and initiated the photosynthesis .(Handelsman and Stabb, 1996). Constant maintenance of the population of the biocontrol agent is necessary for full expression of the biocontrol mechanisms and realization of biofungicidal activity. Hence, the greater quantity of the biocontrol agent *Trichoderma* has greater efficiency in reducing the damping off disease in tobacco seedlings (Gveroska, 2013 a).

## Seedbeds tests

In seedbed conditions, the disease is manifested with very low intensity in variants where the biocontrol agent, i.e. biological preparation was applied (Table 3). The disease does not occur when biocontrol agent was used in treating the soil before sowing as well as in treatment of seed (without the use of herbicide after sowing).

The disease is manifested with increased intensity in the second estimation (Table 4). However, the influence of the biocontrol agent can be observed, too. The highest intensity of attack was recorded in the check (nontreated variant). The absence of the use of fungicides, as well as nitrogen nutrition (which increases the susceptibility to the pathogen) contribute to susceptibility to pathogens and spread of infection.

Treatment with the biopreparation 15 days after sowing showed a poor effect in seedlings protection from the disease. Delayed treatment with Trianum P has negative effect on multiplication of *T. harzianum* T22 and on the expression of biocontrol mechanisms, resulting in highest percentage of infested area, which was even higher in Variant 2 than in the check. Gveroska (2013 a), comparing the *Trichoderma* models of applications prior to sowing and 15 days after sowing, determined the advantage of the first two modes over the third one. When the biocontrol agent was applied after seedlings had grown, there was a smaller amount of BCA than in application before sowing, as mentioned previously. Our results are in accordance with those of Izzati and Abdullah (2008), who reported that delayed time of application increased the index of disease severity.

**Table 3. Influence of Trianum P on the intensity of damping off disease in seedbeds (Ist estimation)**

| Mark | Variant | Percentage of infested area | | | |
|---|---|---|---|---|---|
| | | Replication | | | |
| | | I | II | III | Mean value |
| Ø | Check (herbicide,saltpeter; without fungicide) | 0,19 | 0,39 | 11,22 | 3,93 |
| 1 | Sowing without herbicide; Treatment after 15 days | 1,71 | 0 | 0 | 0,58 |
| 2 | Sowing with herbicide; Treatment after 15 days | 0,53 | 0,61 | 2,23 | 1,12 |
| 3 | Sowing without herbicide; Pre-sowing soil treatment | 0 | 0 | 0 | 0 |
| 4 | Sowing with herbicide; Pre-sowing soil treatment | 0 | 0,15 | 0 | 0,05 |
| 5 | Sowing without herbicide; Seed treated with Trianum P | 0 | 0 | 0 | 0 |
| 6 | Sowing with herbicide; Seed treated with Trianum P | 0 | 0,11 | 0 | 0,04 |

The variants where treatment began 15 days after sowing had one treatment less, which certainly had some influence on creating the conditions for growth and multiplication of the biocontrol agent population. In the other variants, the intensity of attack was lower as a result of the two additional treatments with biofungicide.

**Table 4. Influence of Trianum P on the intensity of damping off disease in the seedbeds (2nd estimation)**

| Mark | Variant | Percentage of infested area | | | |
|---|---|---|---|---|---|
| | | Replication | | | |
| | | I | II | III | Mean value |
| **Ø** | Check (herbicide,saltpeter; without fungicide) | 7,15 | 4,50 | 9,91 | 7,19 |
| **1** | Sowing without herbicide; Treatment after 15 days | 4,12 | 2,15 | 8,23 | 4,83 |
| **2** | Sowing with herbicide; Treatment after 15 days | 18,02 | 3,04 | 4,61 | 8,56 |
| **3** | Sowing without herbicide; Pre-sowing soil treatment | 6,42 | 1,46 | 0,59 | 2,82 |
| **4** | Sowing with herbicide; Pre-sowing soil treatment | 1,50 | 2,22 | 1,28 | 1,67 |
| **5** | Sowing without herbicide; Seed treated with Trianum P | 1,35 | 0,79 | 1,22 | 1,12 |
| **6** | Sowing with herbicide; Seed treated with Trianum P | 0,59 | 0,77 | 1,19 | 0,85 |

The lowest values for disease attack were found in the variants where seed was stored in suspension of the preparation and with two additional treatments. In the variant with treated seed and with herbicide use, the values were insignificant (Table 4). These results are confirmed by Gveroska (2013 a), who reported that application by seed has the best results in the control of damping off in seedlings. Majority of biocontrol products used against seedborne and soilborne fungal pathogens, including the causal agents of seed rot, damping off and root rot diseases, when it is used as seed treatment have been effective in protecting against fungal pathogens (Heydari and Pessarakli, 2010).

This situation is affected by the greater possibility of development and multiplication of *Trichoderma* population due to the possibility of seed colonization by following the growth of root system, stimulated by root exudates (Handelsman and Stabb, 1996; Harman 2000; Howell et al., 2000).


**Figures 5-7. Variants in seedbeds test**

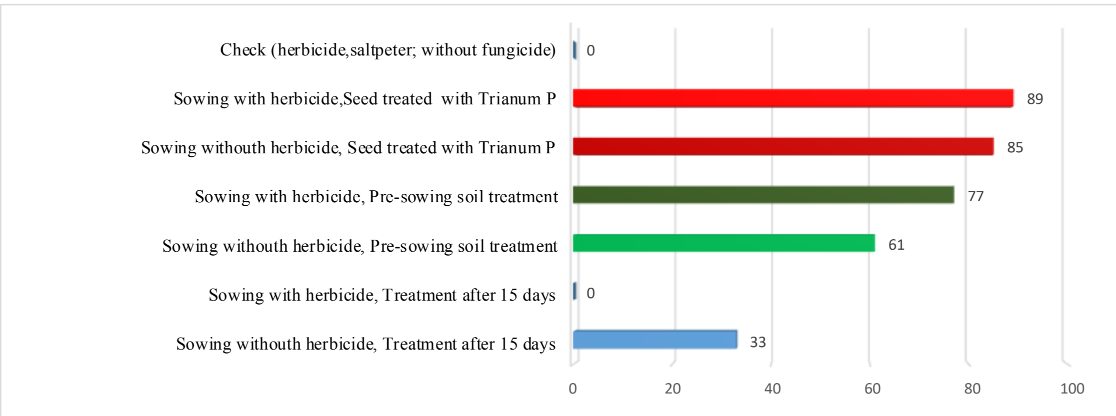
The values for disease intensity were also lower in variants where the preparation was applied in soil, compared to the check and the variants treated after 15 days (Table 4, Figures 5-7).

However, among variants 5 and 6 and 3 and 4, smaller intensity of damping off disease was observed in variants 4 and 6 that were treated with herbicide.

*T. harzianum* is multiplied in the soil as a natural environment where the possibility of herbicide influence is lower because it is bound to the organic matter from the fertilizer. Such results for herbicide effect on the population of *T. harzianum* are in agreement with previous studies (Gveroska, 2014 a), according to which the small decrease of the population caused by herbicide application is compensated or even increased during the additional treatments.

*According to investigations, T.harzianum T22* has fungicidal effect on the causing agent of damping off disease on tobacco seedlings. This is in agreement with the results of Chacon et al. (2006) and Devaki et al. (2008), who reported antagonistic action of *T. harzianum* on *Pythium* sp. It also has biocontrolling effect on *Rhizoctonia solani*, as confirmed by Monte (2001), Harman (2000, 2004, 2006), Chet et al. (2006), Wilson et al. (2008) and Gveroska (2013 b, 2014 b).

Therefore, *T. harzianum T22 (the basic component of* the biopreparation *Trianum P) is effective in reducing the damping off disease. Its* effectiveness depending on the application mode is presented in Graph 2.




**Graph. 2 Effectiveness of variants depending on the mode of application**

## CONCLUSIONS

* The application of *T. harzianum* T22 as the basic component of the biofungicide Trianum P confirmed the previous *in vitro* and *in vivo* studies on the impact of this biocontrol agent on the control of pathogenic fungi on tobacco.
* *T. harzianum* T22 is a biocontrol agent that can be used in the control of causing agents of the dampig off disease in tobacco seedling.
* In the biolaboratory tests, the best results in controlling the disease were obtained by treatment of seeds with suspension of the preparation and two additional treatments.
* Soil treatment showed good results, but still weaker compared to field conditions.
* The treatment of infested soil showed no satisfactory results in prevention of the infection, which is understandable knowing the principle of action of this preparation.
* In field investigations, the highest effectiveness in the control of the disease was recorded in variants with treated seed, with and without herbicide application.
* Pre-sowing soil treatment can be, to a certain extent, an alternative to the treated seed.
* The herbicide application did not show any adverse effects in these variants.
* Delayed treatment gives the worst results. In variants where the first treatment was made 15 days after sowing, the lowest fungicidal effect was found both in biolaboratory and in field condition
* The best model of application is to treat the seed with the preparation (storage in suspension) before sowing, with minimum two additional treatments of the seedling.
* Trianum P biofungicide confirmed its fungicidal effect. It has a good perspective in the sustainable tobacco production.

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ISSN 0494-3244
Тутун/Tobacco Vol. 67 No 7-12, 46-55, 2017 UDC: 633.71-272.7:632.951(497.775)"2016/2017"
Original scientific paper

# DOCIOSTAURUS MAROCCANUS ON TOBACCO

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## ABSTRACT

*Dociostaurus maroccanus* Thunb. is polyphagous and highly adaptable locust. In severe infestations, it may devastate tobacco fields completely.

During 2016/2017 its quantitative representation was studied. A sweep net method was used around the tobacco plots, at 10-day intervals, during June and July. The method of survey was applied on 100 randomly selected tobacco plants, at 10-day intervals, during July, August and September. Five insecticides were applied for its control, with active ingredients Deltamethrin, Esfenvalerate, Lambda-cyhalothrin, Chlorpyrifos+cypermethrin and Chlorpirifos.

The species is present on tobacco from July until the end of September in "solitary phase". The locusts damage tobacco leaves and make irregular holes. In 2016 and 2017, the Moroccan locust was found in increased number (805 and 724, respectively), compared to its usual occurrence.

Chemical protection must be based on treatments given in due time and on restriction of treatment to the smallest possible area. The applied insecticides provide a high level of control.

**Keywords:** *Dociostaurus maroccanus*, tobacco, damages, protection

## *DOCIOSTAURUS MAROCCANUS* НА ТУТУНОТ

*Dociostaurus maroccanus* Thunb. е полифаген и многу прилагодлив вид. При силен напад скакулците може целосно да ги уништат тутунските растенија.

Во текот на 2016/2017 година ја проучувавме неговата квантитативна застапеност. Го употребивме методот на ловење со кечер околу тутунските парцели, во интервали од 10 дена, во текот на јуни и јули. Во текот на јули, август и септември го применивме метод на преглед на 100 случајно избрани тутунски растенија, на интервали од 10 дена. За контрола на овој штетен вид применивме пет инсектициди со активна материја: Deltamethrin, Esfenvalerate, Lambda-cyhalothrin, Chlorpyrifos+cypermethrin and Chlorpirifos.

Видот е присутен на тутунот од јули до крајот на септември во "солитарна фаза". Скакулците ги оштетуваат листовите од тутунот и прават неправилни дупки. Мароканскиот скакулец беше утврден во зголемена квантитативна застапеност, од вообичаено. Во 2016 година се утврдени 805 скакулци и 724 во 2017 година.

Хемиската заштита мора да се заснова на навременост на третманите и на ограничување на третманот на најмала можна област. Применетите инсектициди обезбедуваат високо ниво на контрола.

**Клучни зборови:** *Dociostaurus maroccanus*, тутун, штети, заштита

## INTRODUCTION

Species of the order Orhopthera known to occur on tobacco in R. Macedonia are: *Dociostaurus maroccanus* Thumb. (Caelifera, Acrididae) -Moroccan locust, *Calliptamus italicus* L. (Caelifera, Acrididae) -Italian locust, *Tettigonia viridissima* L. (Ensifera, Tettigonidae) - Great green bush-cricket, *Oecanthus pellucens* Scop. (Ensifera, Gryllidae) - Italian tree cricket, and *Gryllotalpa gryllotalpa* L. (Ensifera, Gryllotalpidae) - European mole cricket ( Dimeska *et al.,* 2004).

In some period locusts form dense groups comprising huge numbers, bands of hoppers, and/or swarms of winged adults which migrate (Song, 2011).

The Moroccan locust is distributed in northern Africa, Southern and Eastern Europe and Western and Central Asia. It swarms and migrates, but not over long distances.

These insects are abundant in natural and anthropogenic habitats (rangelands, wetlands, agricultural fields, lawns, etc. (Latchininsky *et al.,* 2011).

The list of food plants used by *D. maroccanus* compiled from Russian sources alone consists of more than 150 species belonging to 33 families, including approximately 50 different crops (Latchininsky and Launois-Luong, 1992, loc. cit. Latchininsky, 1998).

In its gregarious phase the locust *Dociostaurus maroccanus* Thunberg (Orthoptera Acrididae) has periodically caused significant yield losses in many Mediterranean and Asian countries, and alarm in the general public (Baldacchino, 2012).

The Moroccan locust, *D. marrocanus* Thunb. is the only species of the genus known to express an extreme form of density-dependent phase polyphenism in color and morphology in both nymphal and adult stages (Uvarov, 1977, loc. cit. Song, 2011).

Its broad polyphagy, extreme voracity, enormous fecundity and capability to migrate in swarms made it a major enemy of agriculturists from the Canary Islands to Afghanistan (Latchininsky, 1998).

Locusts are historically known for wiping out fields of crops in a day, they are mentioned in the Bible and in poems.

In its gregarious phase, swarms of this locust have often occurred in Mariovo region in Prilep (Vasilev, 1984).

Control of the Moroccan locust during its outbreaks requires a massive campaign because vast infested territories need to be treated in a very short interval, dictated by the insect's brief developmental period (Latchininsky, 1998).

According to Cheke *et al.,* (1999), an alternative to full coverage insecticide spraying is the barrier technique in which insecticides are sprayed on to strips of vegetation, separated by wide spaces which are not sprayed. Marching hoppers encounter the sprayed barriers as they move around searching for food, and acquire a lethal dose of the insecticide.

Due to the escalating costs of locust control, the economic option of suspending locust control in the remote outbreak areas and only concentrating on direct crop protection instead is an idea that has been considered as a cost-effective option in other locust affected regions (Herok and Krall, 1995 loc cit. Cheke *et al.,* 1999).

Significant progress has been made in the search for locusticides with low environmental impact, spanning from organochlorine compounds to spinosyns, and on the use of entomopathogenic fungi such as *Metarhizium anisopliae* (Metschnikoff) (Baldacchino *et al.,* 2004, loc cit. Baldacchino *et al.,* 2012).

## MATERIAL AND METHOD

During 2016/2017 the Moroccan locust was found in enlarged numbers in the Experimental field of the Scientific Tobacco Institute - Prilep.

Standard methodology was applied for laboratory investigations on their morphological characteristics.

A sweep net is used to collect insects from grass and weeds around the tobacco plots. It is a cost-effective way to monitor for the presence of locusts.The sweep net is swept back and forth through vegetation quickly turning the opening from side to side, for 180º. A common practice is to take a sweep from right to left, walk a step, and take another sweep, left to right. The collector walks forward while sweeping, and the net is moved through plants, weeds and grasses with force. We made ten sweeps, in ten replications arround tobacco plots. The presence of larvae and adults was followed on the ten days intervals, during June and July.

After the locusts migrated on the tobacco plants, we counted its quantitative representation in tobacco plots. We applied the method of 100 randomly selected tobacco plants, at 10- day intervals, during July, August and September. In July we made investigations around tobacco plots and in tobacco plots.

Five insecticides were applied in the control of this harmful species: pyrethroids with active substance Deltamethrin, Esfenvalerate, Lambda-cyhalothrin and organophosphorus insecticides with active substance Chlorpyrifos + cypermethrin and Chlorpirifos. One decare of tobacco was divided into 5 plots where the insecticides were applied and the other decare was left as a check plot.

## RESULTS AND DISCUSSION

### Morphology and biology of the pest

*Dociostaurus maroccanus* (Thunberg, 1815) is a univoltine species, herbivore. The pest passes "incomplete metamorphosis" (*egg - nymph – adult).* Both nymph and adult often share the same food source and make damages on tobacco.

There is density-dependent phenotypic plasticity in *D. maroccanus*. The nymphs differ in color, morphometrics ratios, physiology and behavior. Group mating, group oviposition and swarm formations are present in adults (Song, 2011).

Understanding of the morphology and biology of *D. maroccanus* populations can be of great help in predicting potentially severe infestations.

*D. maroccanus* spends more than a half of its life cycle period in the embryonic stage. In autumn, the female induced by short days and fall of temperatures, lays eggs in the upper layers of the soil. The eggs are laid in oothecae. They are placed on dry and compacted surfaces near tobacco plots, on unprocessed surfaces, neglected meadows, rocky terrain, etc. If the surfaces are cultivated and irrigated, locusts do not lay eggs and they are looking for more suitable places.

This species spend the winter in egg pods in soil. In spring, April/May, under favorable conditions the nymph eclodes.

The nymphs resemble miniature adults, with wing buds. Nymphs moult five times. They differ in size, proportions and volume of the body, color and markings. After each successive instar, nymphs have larger wings and antenna. Newly hatched larva is whitish. At the end of the L1 it becomes dark brown. The wing buds are visible from third-instar. Throught development the larva becomes olive-pink with black spots on the body. The pests are fully formed at the final moult into adults.

According to Tanasijevic *et al.* (1969), the female are long 22-33 mm and male 20-28 mm.

In tobacco biocenosis the adults are 20 to 33 millimetres long, with light brown color and dark spots on the body and front wings. Most of the captured adults were 28 mm in size. The head is hypognathous and mouthparts directed downward and specialized for biting and chewing. The complex eyes are large, well developed and ocellae are present. The antennae have multiple joints but they are not longer than prothorax (Figure 1) .

Figure 1. Adult of *D. maroccanus*

Pronotum is shield like, covering much of thorax. There is a creamy coloured cross-shape (X) on the prothorax. The adults are powerful fliers. The front wings are large, elliptical and narrower than the hind wings. They are completely parchment. The hind wings are membranous, translucent, with pronounced veines.

They have a cylindrical body, with elongated hind legs and musculature adapted for jumping and single-segmented cerci. The hind legs are powerful, the femur is yellowish in color and there are three dark spots while the tibia is usually red. The ovipositor consists of two shovel shaped valves. The female uses the ovipositor to dig a deep egg chamber in the ground, to manipulate the eggs, and to cap the eggs with frothy moist protection substance.

Adults produce sound ("stridulation") by rubbing their wings with their legs, which is heard at night from the meadows around the tobacco plantations. The tympanum or ear is located on the first abdominal segment. These organisms use sound and vibrations to locate other individuals.

Locusts in “gregarious phase” differ from locusts in “solitary phase”, in many characteristics (morphological, biological et.c), esspecially in behavior. The gregaria nymphs become darker, more mobile, with increased tactile stimulation and with voracious appetite. They are attracted to each other and behave like cohesive units. The adult body colour darkens, their muscles grow stronger and they can travel great distances.

During insvestigations locusts appear on tobacco in “solitary phase” and they make irregular holes on tobacco leaves (Figure 2).

Individual locusts are brighter in color, compared with the samples of the "gregarious phase" of the locusts that are found in the Entomological collection and the images of the Institute.

Figure 2. Damaged tobacco plants

Under certain circumstances they become more abundant and change their behaviour and habits, becoming gregarious.

According Latchininsky, 1998, population increase is usually associated with hot and dry weather.

In past, in Mariovo the swarming behaviour was a response to overpopulation. The locusts are consuming most of the vegetation wherever they settle. In "gregarious phase", they destroy whole tobacco plants and whole plantations of tobacco (Vasilev, 1984).

The Moroccan locust is distributed everywhere. It is a polifagous species and it feeds with above-ground plant parts. If the surface is not cultivated, under favorable conditions they may increase their population and cause tremendous agricultural damage. In tobacco biocenosis they live separately or in small groups and eat almost all forms of vegetation.

The initial swarming of locusts may occur as a consequence of human agricultural practices that make them more successful in their new habitats: increasing locust densities are correlated with human-induced changes in vegetation structure (Benfekih *et al.*, 2002).

According Vasilev, 1984, locusts are fed with weeds (*Cynodon dactylon* L., *Poa* sp., *Medicago* sp., *Trifolium* sp.).

**Presence of locust on tobacco**

In spring *D. maroccanus* feeds in the neglected meadows, on weeds from the surrounding unprocessed land, as well as weeds around tobacco plots (Table 1).

Accroding to quantitative representation of locusts on weeds around tobacco plots, 238 locusts were recorded in 2016 and 218 in 2017.

**Table 1.** Quantitative representation of locusts on weeds around tobacco plots 2016/2017 (method: insect's sweep net)

| Replication | No. of locusts /decade- 2016 | | | | | | No. of locusts /decade- 2017 | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Junu | | | July | | | Junu | | | July | | |
| | I | II | III | I | II | III | I | II | III | I | II | III |
| I | - | 2 | 5 | 2 | 9 | 1 | 1 | - | - | 8 | 4 | 1 |
| II | 11 | - | 11 | 13 | 8 | - | 1 | - | 3 | 8 | 5 | 4 |
| III | 1 | 18 | 5 | 3 | - | 5 | 2 | 8 | 2 | - | - | 6 |
| IV | 2 | 7 | 21 | 2 | - | - | - | 5 | 7 | 5 | 11 | - |
| V | - | 6 | 6 | 7 | 5 | - | - | 7 | 6 | 4 | - | 3 |
| VI | - | 9 | - | - | 5 | 7 | - | - | 5 | - | 4 | 3 |
| VII | 8 | 2 | - | 4 | - | - | 1 | 5 | 6 | - | 8 | 2 |
| VIII | - | 3 | 7 | - | - | 6 | 4 | 3 | 9 | 12 | 9 | 1 |
| IX | 6 | 4 | 6 | 12 | - | - | - | 3 | 8 | 12 | - | 1 |
| X | - | - | - | - | 9 | - | 2 | 3 | 6 | - | 6 | 4 |
| **Total** | 28 | 51 | 61 | 43 | 36 | 19 | 11 | 34 | 52 | 49 | 47 | 25 |

After planting the tobacco, and especially after the weed vegetation had dried, the locusts moved on to tobacco plants. They are established on tobacco from July until the end of September, 2016/2017.

Larvae and adults cause damage to tobacco plants and sometimes three locusts per plant were occurred. During the fourth larval stage, they are nourished more intensively, and larvae can consume all the above-ground parts of the plant. Also, the immature sexual adults need additional nutrition. In the middle of August we noticed pairs that were copulating on tobacco plants.

The locusts were most numerous from the end of July to the end of August 2016. After the rains in early September and temperature drop at the end of September their number decreased.

In 2017, most numerous locust population was recorded in mid July and early August. The locust population stayed on tobacco to the end of September. In 2016 the number of locusts was 567 and in 2017 it was 506 (Table 2 and 3).

**Table 2.** Quantitative representation of locusts in plot margins 2016/2017 (method: survey of 100 tobacco plants)

| Decade | No. of tobacco stalks | No. of locusts in plot margins /2016 | | | No. of locusts in plot margins /2017 | | |
|---|---|---|---|---|---|---|---|
| | | July | August | September | July | August | September |
| **I** | 100 | 23 | 42 | 27 | 24 | 48 | 15 |
| **II** | 100 | 32 | 53 | 13 | 54 | 22 | 14 |
| **III** | 100 | 51 | 52 | 4 | 77 | 29 | 13 |
| **Total** | 300 | 106 | 147 | 44 | 155 | 99 | 42 |

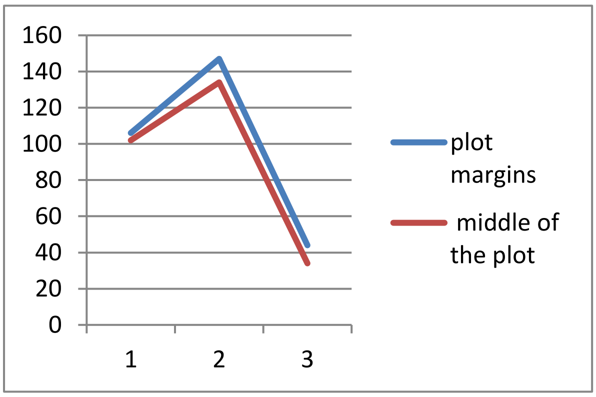
**Table 3.** Quantitative representation of locusts in the middle of the plot 2016/2017 (method: survey of 100 tobacco plants)

| Decade | No. of tobacco stalks | No. of locusts in the middle of the plot /2016 | | | No. of locusts in the middle of the plot /2017 | | |
|---|---|---|---|---|---|---|---|
| | | July | August | September | July | August | September |
| I | 100 | 19 | 41 | 15 | 18 | 43 | 10 |
| II | 100 | 36 | 51 | 16 | 35 | 19 | 5 |
| III | 100 | 47 | 42 | 3 | 59 | 15 | 6 |
| Total | 300 | 102 | 134 | 34 | 112 | 77 | 21 |

Moroccan locust is a daily and temperature-depending insect. By increasing the temperature, its activity grows. In the morning we observed small groups of locusts warming in the sun (Figure 3).

During the day, the most active period was from 8 to 11, when they were on tobacco leaves. Early in the afternoon, as a result of high temperatures, locusts activity decreased and they were hiding among tobacco plants.

Figure 3. Moroccan locus



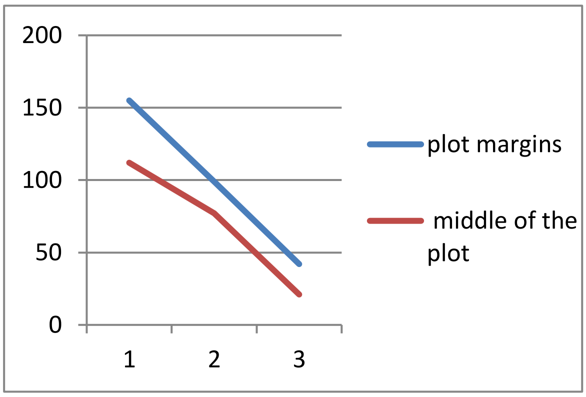




Figure 4. Number of locusts-2016

Figure 5. Number of locusts-2017

There was not much difference in locusts number along the margins of tobacco plots and in the middle of the plot (Figure 4 and 5).

The ability to feed with almost every kind of vegetation and to develop a large population and migrate as a "gregarious phase", make locusts a serious agricultural threat.

Locust control is complex. Climate factors should be also considered. In spring, during March/May the amount of precipitation is low (130 mm in 2016 and 126 mm in 2017).

According to Uvarov, 1957, loc cit. Tanasijevic, Ilic, 1969, precipitation in spring is of great importance for the developmental cycle of *D. maroccanus*. The optimum rainfall for occurrence and development of the Moroccan locust from March to May is 100 millimetres.

This factor was optimal and the number of young locusts increased on surrounding unprocessed land near the place of our investigations.

In May/June, dry weather with transitory rainfalls has favorable effect on its population. Irrigation of tobacco plots has caused green vegetation only around tobacco plots, and locusts start to appear abundantly on tobacco plants.

**Measurements for pest control**

The optimal locust control strategy is timely control. *D. marrocanus* is difficult to control *анд* there are different strategies for its management (biological methods, cultural methods, chemical methods etc). The method of control is selected depending on the concrete conditions of the terrain.

When disturbed, *D. maroccanus* is moving and jumping rapidly from one point of tobacco plant to another. It also has a pronounced mimicry, so the exact number of locusts on tobacco can not be determined.

Invasions from locusts could be avoided by taking preventive measures during autumn and early spring, such as: ploughing the soil in order to bring egg pods to the surface, weed control, rotation of tobacco with another culture, soil processing, utilization of undisturbed surfaces and neglected meadows, timely insecticidal aplication, control of locusts on strips of vegetation, control of locusts in localized area e.c.t.

It is necessary to identify the locations where locusts lay their eggs and to take soil samples. In the autumn, soil samples should be taken from the surrounding unprocessed fields in order to determine the number of egg pods per $m^2$. Also monitoring of larvae and adult movements could provide an early warning of its invasion of an area or crop.

Tanasijevic *et al.,* (1985), state that 200 eggs /$m^2$ create conditions for a serious attack of locusts and according to Camprag *et al.,* (1983), 50 larvae/ $m^2$ is a warning to apply measures of control.

For the control of this harmful species, there we applied three pyrethroid with contact and digestic action and two organophosphorus insecticides with contact, digestic and volatile action. We used the traditional means of control, based on the use of insecticides from the ground, in localized area in order to protect the environment.

The best moment for treatment with an insecticide is while the larvae are young. All applied insecticides showed high effectiveness in locust control (Figure 6). After ten days of aplication, we did not found the presence of locusts on the treated plots. In the plot that served as a check, the locusts continued to cause damage.

Figure 6. Effectiveness of insecticides in locust control

This locust has traditionally been considered a serious agricultural threat. Population outbreaks in Macedonia in recent years (2013-2014) required the application of control measures. Chemical control of locusts is more effective when insecticides are applied on young larvae. The control must be performed in due time and on localized area in order to prevent their gregarisation.

The intensity of infestation varies from year to year. It sets the goals of further monitoring of locust populations in subsequent years.

## CONCLUSSION

During the insvestigations, locusts appear on tobacco in "solitary phase" and they make irregular holes on tobacco leaves.

After planting the tobacco, and especially after the weed vegetation had dried, the locusts moved to tobacco. They stayed on tobacco plants from July until the end of September, 2016/2017.

The locusts were most numerous from the end of July to the end of August 2016 and from mid July to early August in 2017.

There was not much difference in the number of locusts along the margins of tobacco plots and in the middle of the plots.

Its large numbers, voracious appetite, ability to eat almost all forms of vegetation and ability to migrate in "gregoarious forms" make this species a feared pest.

Chemical protection must be based on the timeliness of treatments and on restriction of treatment to the smallest possible area.

All investigated insecticides: Deltamethrin, Esfenvalerate, Lambda-cyhalothrin, Chlorpyrifos+cypermethrin and Chlorpirifos had a high level of control.

For successful management greater attention should be payed on monitoring of the number of egg pods, larvae and adult movements.

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ISSN 0494-3244
Тутун/Tobacco Vol. 67 No 7-12, 56-63, 2017 UDC:633.71-153.037:632.48]:632.952(497.775)"2016"
632.952:[633.71-153.037:632.48(497.775)"2016"
Original scientific paper

# TOBACCO SEEDLINGS PROTECTION FROM DAMPING OFF DISEASE WITH STROBILURIN FUNGICIDES

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## ABSTRACT

The damping off disease is often present in tobacco seedbeds, causing severe damage to the breeding process. The causing agents of the disease are the soil phytopathogenic fungi *Pythium debaryanum* and *Rhizoctonia solani*. The aim of the investigation is to study the effects of strobilurin preparations in protection of tobacco seedlings. The following strobilurin-based products were applied in the investigation: Signum 0.1% (boscalid 267 g/kg + piraclostrobin 67 g/kg), Quadris 25SC 0.1% (azoxystrobin 250 g/l), Stroby WG 0.1% (kresoxim-methyl 500 g/kg) and the standard fungicides Proplant 0.25% (722 g/l propamocarb-hydrochlorid) and Top M 0.1% (70% thiophanate methyl). Seedlings were treated with 1 l/ $m^2$ suspension in the 4th-leaf stage and 15 days after. The intensity of the disease reached 95.00% in the check variant while with the standard fungicides and strobilurin-based fungicides the disease intensity was lower. The highest effectiveness was reached with Signum 0.1% (90.23%) and somewhat lower effectiveness with the other fungicides.

Due to their high effectiveness, strobilurin-based preparations can have wide practical application in protection of tobacco seedlings from *P. debaryanum* and *R. solani*.

**Keywords:** seedling, pathogens, disease intensity, fungicides, effectiveness

# ЗАШТИТА НА ТУТУНСКИОТ РАСАД ОД БОЛЕСТА СЕЧЕЊЕ СО ФУНГИЦИДИ ОД ГРУПАТА СТРОБИЛУРИНИ

Болеста сечење на тутунскиот расад која е редовно присутна во леите, му нанесува големи штети на расадопроизводството. Причинители на болеста се почвените фитопатогени габи *P. debaryanum* и *R. solani*. Основна цел во ова испитување е примената на препаратите од групата стробилурини за заштита на тутунскиот расад. За испитување се користени препаратите од групата стробилурини, Signum 0,1% (boscalid 267 g/kg+pyraclostrobin 67 g/kg), Quadris 25SC 0,1% (azoxystrobin 250 g/l), Stroby WG 0,1% (kresoxim-methyl 500 g/kg) и стандардните фунгициди Proplant 0,25% (722 g/l propamocarb-hydrochlorid) и Top M 0,1% (70% thiophanate methyl). Расадот е третиран со полевање, со по 1 литар раствор на $m^2$, во фаза вкрстување, и после 15 дена. Интензитетот на болеста кај контролата изнесува 95,00%, а кај стандардните фунгициди и фунгицидите од групата стробилурини има послаб интензитет. Највисока ефикасност е постигната со фунгицидот Signum 0,1%, од 90,23%, а нешто послаба е ефикасноста кај другите фунгициди.

Заради високата ефекасност што ја покажаа препаратите од групата стробилурини, истите може да најдат и поширока практична примена во заштитата на тутунскиот расад од овие почвени патогени.

**Клучни зборови:** расад, патогени, интензитет на болеста, фунгициди, ефикасност

## INTRODUCTION

Damping off is a common disease which often attacks tobacco seedlings and causes severe damage to the breeding process. The microclimate conditions in the seedbeds (high humidity and temperature) are favorable both for seedlings and pathogen development. The damping off disease in tobacco is caused by the soil phytopathogenic fungi *Pythium debaryanum* and *Rhizoctonia solani,* which often cause severe damage to the seedlings. Some years one of the pathogens occurs at one site and another pathogen at another site, but it doesn't exclude the possibility both pathogens to appear at the same site, even in the same seedbed. To prevent this, it is recommended to apply various cultural practices and chemical preparations (fungicides), as supported by a number of scientific investigations.

Due to the small number of fungicides that show some effectiveness in the control of these pathogens, research has also been made on the use of strobilurin-based preparations. Herms et al. (2002) reported that strobilurin fungicides include many synthetic plant-protective compounds with a wide range of effects. The authors confirmed that the F500 fungicide (pyraclostrobin) has not only highly fungicidal effect on the seedlings but it also improves tobacco resistance against Tobacco mosaic virus (TMV) and wildfire disease on tobacco (*Pseudomonas syringae pv. tabaci*). Plants treated with strobilurin are healthier and have a better growth and intense green color, unlike plants that are not treated with such fungicides. According to Kanungo and Joshi (2014), plants treated with strobilurin have characteristic dark green color, higher biomass and higher yield, and Venancio et al. (2003) reported 20% increase of biomass after plant treatment. Some physiological changes in strobilurin-treated tobacco and wheat have been confirmed by Grossmann et al. in 1999 (quoted by Swoboda and Pedersen, 2005).

Tashkoski (2015) and Tashkoski (2016) performed trials with Signum fungicide in protected area, containing two active ingredients (boscalid and pyraklostrobin), and reported high effectiveness in the control of damping off disease in tobacco seedlings caused by pathogenic fungi *Pythium debaryanum* and *Rhizoctonia solani*. 100% protection was reached in seedlings planted on pre-treated soil with pure pathogenic culture and then treated with Signum. High effectiveness in protection from damping off disease at extremely high intensity of infection was also recorded by Tashkoski et al. (2016), with some strobilurin fungicides applied in protected area.

The aim of this investigation was to check whether the efectiveness of strobilurin fungicides in the control of damping off disease on seedlings obtained in biological laboratory can also be confirmed in trials set up in field conditions.

## MATERIAL AND METHODS

Trials were carried out in 2016 in field conditions, as a continuation to the research conducted in Biological laboratory of the Scientific Tobacco Institute - Prilep previous years. The tested fungicides were actually intended for control of pathogens that attack fruits and leaves of plants, but in our laboratory tests they were applied on tobacco seedlings that were previously artificially inoculated with cultures of *P. debaryanum* and *R. solani*. For the sake of greater reliability and confirmation of the fungicide effectiveness in biological laboratory, investigations were also conducted in field conditions.

The trials were set up in cold frames covered with polyethylene and sown with the oriental variety P-66-9/7 on 11.4.2016 (10 g seed/10 $m^2$). After sowing, seedbeds were treated with the herbicide Gamit 1 ml/10 $m^2$. The trial was performed in three replicates and the results were presented as an average value.

The trial included standard fungicides Proplant and Top M, but particular attention was paid to the strobilurin fungicides Signum, Quadris and Stroby. Each fungicide was applied in 10 $m^2$ seedbed and Proplant and Top M were applied together in the same seedbed, because of the possible presence of both pathogens. All tested fungicides, active ingredients and concentrations are presented in Table 1.

Treatment was performed by applying 1 liter solution/$m^2$ with watering can and the check was hydrated with pure water. Usual cultural practices were applied during the seedlings growth, including regular irrigation, two treatments with nitrogen fertilizer and application of insecticides against harmful insects. The first fungicide treatment was done on 25.4.2016 in the 4th leaf stage and the second treatment was performed 15 days after.

Table 1. Fungicides testing

| Fungicide | a.i. | Concentration % |
|---|---|---|
| Proplant 722 SL | 722 g/l propamocarb hydrochlorid | 0,25 |
| Top M | 70% thiophanate methyl | 0,1 |
| Signum | 267 g/kg boscalid +<br>67 g/kg pyraclostrobin | 0,1 |
| Quadris 25SC | 250 g/l azoxystrobin | 0,1 |
| StrobyWG | 500g/kg kresoxim-methyl | 0,1 |

The health condition of tobacco seedlings was assessed according to the number of infected plants and the percentage of infected area. 10-15 days after the second treatment, two assessments of seedlings health status were made. During investigations, seedlings were checked for the presence of infection and the size of the infested area was measured. From the data obtained, the percentage of bold area was estimated and disease severity determined. According to the disease intensity measured after the second assessment, the coefficient of fungicide effectiveness was calculated by the Abbott’s formula (1925).

## RESULTS AND DISCUSSION

The results obtained in different variants revealed various levels of intensity of damping off disease in tobacco seedlings caused by *P. debaryanum* and *R. solani* (Table 2). In the check variant the disease intensity ranged from 81.66% at the first assessment to 95.00% in the second assessment, which is considerably high infestation (Figure 1). In seedlings treated with fungicides, the intensity of disease was lower. Thus, in the variant treated with combination of standard fungicides Proplant 0.25% + Top M 0.1%, the intensity of infection ranged 37.50% in the first assessment and 39.84% in the second assessment. Seedlings treated with strobilurin fungicides (Signum 0.1%, Quadris 0.1% and Stroby 0.1%) were not infested until the first assessment. In the second assessment low percentage of infection was recorded (9.28% in seedlings treated with Signum 0.1% and 15.25% with Quadris 0.1%) and the highest intensity (19.32%) was recorded in seedlings treated with Stroby 0.1%. Even this percentage, however, is low compared to the infection measured with standard fungicides and in the check variant.

Table 2. Intensity of damping off disease on seedlings growth in field conditions

| Variant | Infested area % | |
|---|---|---|
| | I assessment | II assessment |
| Check | 81,66 | 95,00 |
| Proplant 0,25% + Top M 0,1% | 37,50 | 39,84 |
| Signum 0,1% | 0,00 | 9,28 |
| Quadris 0,1% | 0,00 | 15,25 |
| Stroby 0,1% | 0,00 | 19,32 |

Fig. 1. Untreated seedlings (check)

Data on the incidence and intensity of the disease obtained in the second assessment were used to calculate the effectiveness of tested fungicides (Table 3).

Table 3. The effectiveness of fungicides in the control of damping off disease on tobacco seedlings in field conditions

| Variant | Infested area % | Effectiveness % |
|---|---|---|
| Check | 95,00 | - |
| Proplant 0,25% + Top M 0,1% | 39,84 | 58,06 |
| Signum 0,1% | 9,28 | 90,23 |
| Quadris 0,1% | 15,25 | 83,95 |
| Stroby 0,1% | 19,32 | 79,66 |

The high intensity of disease in the check variant (95.00%) and lower intensity in the treated variants denote a high effectiveness of the tested fungicides. The highest in field conditions was achieved with Signum 0.1% - 90.23% (Figure 2). Slightly lower effectiveness (83.95%) was obtained with the Quadris fungicide 0.1%, (Fig. 3) and the lowest effectiveness (79.66%) was recorded with Stroby 0.1% (Figure 4). As can be seen from the results obtained, the strobilurin fungicides showed far higher effectiveness in the control of damping off disease compared to the standard fungicide Proplant 0.25% + Top M 0.1%, which showed a very poor effectiveness (58.06%).

Fig. 2. Seedlings treated with Signum 0,1%

Fig. 3. Seedlings treated with Quadris 0,1%

Fig. 4. Seedlings treated with Stroby 0,1%

The research confirmed the high effectiveness of the strobilurin fungicides in the control of damping off disease on tobacco seedlings. The data were obtained in the trials conducted in protected area, where the seedlings are not only healthy but are in good condition and have a more intensive green color (Tashkoski et al., 2016). This is also confirmed in the studies of Herms et al. (2002) and Kanungo and Joshi (2014), who reported that plants treated with pyraclostrobin had intensive green color and greater biomass than untreated plants. Significant differences in the incidence of damping off disease caused by *P. debaryanum* were recorded in the treatment with preparations azoxystrobin and etridiazole (Gutierrez et al., 2012), where the highest percentage of healthy plants was obtained and the percentage of infection was weak. LaMondia (2012) confirmed that the

Quadris (azoxystrobin) fungicide significantly reduces the severity of the disease caused by *R. solani* in tobacco seedlings compared to the untreated check and at the same time it can have a direct impact on the reduction of fungus inoculation in the soil. Very high effectiveness in protection of tobacco seedlings from the pathogens *P. debaryanum* and *R. solani* was reported not only in this study, but also in tests conducted in previous years with Signum 0,1% fungicide (Tashkoski, 2015 and Tashkoski, 2016).

Based on the obtained results, it can be stated that the strobilurin fungicides can be widely applied in practice for control of damping off disease on tobacco seedlings due to their high effectiveness. These fungicides can be also applied in protection of many other horticultural crops with seedling production, which are attacked by the same soil pathogens.

## CONCLUSION

Field trials were carried out on tobacco seedlings with application of strobilurin fungicides, which showed high effectiveness in the control of soil pathogens *P. debaryanum* and *R. solani* – the causing agents of damping off disease. Such effectiveness was achieved at extremely high intensity of infection, which reached up to 95.00% in the check variant.

The lowest intensity of infection (9.28%) was recorded with Signum 0.1% and the highest intensity (39.84%) with the standard fungicides Proplant 0.25% + Top M 0.1% used in combination. The intensity of disease with Quadris 0.1% was 15.25% and with Stroby 0.1% it was 19.32%.

The effectiveness of 58.06% reached by combined formulation of the standard fungicides Proplant 0.25% + Top M 0.1% is not sufficient and is much lower than the one obtained by other fungicides.

The highest effectiveness was achieved by the strobilurin-based fungicides. Signum 0.1% reached the highest effectiveness (90.23%), slightly weaker effectiveness was reached by Quadris 0.1% (83.95%), while the poorest effectiveness was obtained with Stroby 0.1% (79.66%). This effectiveness, however, is still far higher than the effectiveness shown by standard preparations.

The high level of effectiveness achieved by strobilurin fungicides in the control of soil pathogenic agents of damping off disease provides the possibility for their wider practical application in seedlings protection from these pathogens.

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ISSN 0494-3244
Тутун/Tobacco Vol. 67 No 7-12, 64-70, 2017 UDC:633.71-152.61(497.7)
Original scientific paper

# INTENZIFICATION OF TOBACCO PRODUCTION IN THE REPUBLIC OF MACEDONIA

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## ABSTRACT

Intensification of agricultural production can be defined as an increase in agricultural production per unit of measurement (labor, means of production etc). Intensification at the level of a whole agricultural holding or its production unit represents a degree of change in the structure of production and the means engaged in the production.

The degree of agricultural intensification depends on environmental, technical-technological and economic conditions. The variability of these conditions requires appropriate adjustment of the degree of intensification. Intensification of an agricultural holding should be economically efficient, within the limits of the economic optimum for increase of assets and labor.

The final forms of intensification are extensive and intensive agricultural production, and between them there are different degrees of intensity. The increase in yields leads to a reduction of the fixed costs, which partially mitigates the increase of variable costs. When deciding on the economically optimal level of intensity, it is important to consider the total investments (sum of the fixed and variable costs per unit of measurement). In general, intensification can be expressed as obtaining a product at a lower cost, which is an indicator of the economic efficiency of production.

**Keywords:** intensification, tobacco production, economic efficiency, agricultural areas, tobacco producers(co-operators)

## ИНТЕНЗИФИКАЦИЈА НА ТУТУНОПРОИЗВОДСТВОТО ВО РЕПУБЛИКА МАКЕДОНИЈА

Под интензивност на земјоделското производство се подразбира одредено вложување на средства и труд на единица капацитет на одредени средства на трудот.Интензификација на ниво на одделни земјоделски производи претставува степенот на вложувања на труд и средства на единица капацитет на средствата на трудот. Интензификација на ниво на цело земјоделско стопанство или негова производна единица претставува степен на промени во структурата на производството и ангажираните средства за производство.

Степенот на интензификација на земјоделството претставува функција на природните, техничко-технолошките и економските услови. Променливиот карактер на овие услови условува соодветно прилагодување на степенот на интензивноста.Степенот на интензивност на едно земјоделско стопанство треба да биде економски ефикасен, односно да е во граници на економскиот оптимум за вложување на средства и труд.

Крајните облици на интензификација се екстензивно и интензивно земјоделско производство, а помеѓу нив се наоѓаат различни степени на интензивност. Зголемувањето на приносите доведува до намалување на фиксните трошоци на единица капацитет, со што делумно се ублажува зголемувањето на варијалбилните трошоци.При одлучувањето за економски оптималното ниво на интензивност треба да се земат во предвид вкупните вложувања (збир од фиксните и варијабилните трошоци на единица капацитет).Во основа,интензификацијата треба да се изрази како добивање на производ со пониска цена на чинење која е показател на економската ефикасност на производството.

**Клучни зборови:** интензификација, тутунопроизводство, економска ефикасност, земјоделски области, тутунопроизводители(коопeранти)

## INTRODUCTION

In the production of raw tobacco, three phases and sixteen operations have been identified so far:

- seedling production ,
- field production of raw tobacco (11 operations)
- domestic manipulation of tobacco (5 operations).

The above mentioned phases and operations were determined through special research and analysis back in 1982, when they were performed exclusively by hand, which made the production difficult and labor-intensive.(5) The research revealed labor-intensive tendencies in almost all phases and operations of tobacco production. The current intensification of the production is due to the use of improved and more available agrotechnics and cultural practices, especially in the phases and operations that can accept the changes. The constant engagement of technical and agrotechnical measures not only increases the production of tobacco per unit area and per household but simultaneously humanizes the labor. Humanization of labor is even more important when there is a continuous reduction of household members and the use of technique and agrotechnical measures can compensate for this deficiency.

## TOBACCO-PRODUCING AGRICULTURAL AREAS

Survey of tobacco-producing areas for a four decades period (1976-2015) are presented in the following table:

Table 1. Areas under tobacco in R. Macedonia (in hectares)

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 1976 | 30.000 | 1986 | 25.000 | 1996 | 30.000 | 2006 | 15.000 |
| 1977 | 28.000 | 1987 | 24.000 | 1997 | 17.000 | 2007 | 17.000 |
| 1978 | 27.000 | 1988 | 24.000 | 1998 | 22.000 | 2008 | 17.000 |
| 1979 | 28.000 | 1989 | 20.000 | 1999 | 20.000 | 2009 | 16.000 |
| 1980 | 27.000 | 1990 | 14.000 | 2000 | 25.000 | 2010 | 19.000 |
| 1981 | 25.000 | 1991 | 20.000 | 2001 | 20.000 | 2011 | 16.000 |
| 1982 | 27.000 | 1992 | 33.000 | 2002 | 21.000 | 2012 | 15.000 |
| 1983 | 27.000 | 1993 | 32.000 | 2003 | 15.000 | 2013 | 20.000 |
| 1984 | 26.000 | 1994 | 34.000 | 2004 | 15.000 | 2104 | 14.000 |
| 1985 | 26.000 | 1995 | 32.000 | 2005 | 16.000 | 2015 | 14.000 |
| Average | 27.100 | | 25.800 | | 20.100 | | 16.300 |

Source: Statistical Yearbook of SFR Yugoslavia, Association of Producers of R. Macedonia and Ministry of Agriculture, Forestry and Water Economy of R. Macedonia

Soil is a natural resource and most important factor in production of raw tobacco. It cannot be physically exhausted and has an unlimited period of use. The use of professional and scientifically based methods can improve soil quality. The soil quality, however, is not reflected in the increase of tobacco production Unlike other crops, tobacco growing areas do not increase but in the latest period they have even decreased. Presently, world's total arable land is 1,405,000,000ha and only 0.25% of that area belongs to tobacco.(8) Besides this, tobacco is grown on poorly productive soils with low land capability and on small fragmented areas which provide the producer significant economic effects compared to the production of other crops. These soils are not suitable for other crops except for tobacco. Although the size of the surfaces of this quality is not known, however, they exist and allow maximum tobacco production if its demand is increased.For illustration, the area planted with wheat is 700 times larger and the area under cotton is 10 times larger than that under tobacco, etc.(6) Despite the small coverage of the agricultural land, the value of tobacco yield is much higher and multiplied. Data on areas planted with tobacco in R. Macedonia by decades are as follows: 1976-1985 - 27.100 ha, 1986-1995 - 25.800 ha, 1996-2005 - 20.100 ha and 2006-2015 - 16.300 ha. In relation to 360.000 ha of arable land in the country, the share of agricultural land under tobacco is: 1976-1985 - 7.5%, 1986-1995 - 6.6%, 1996-25005 - 5.4%, 2006-2015 - 4.2%. The participation of areas under tobacco is constantly decreasing in relation to the arable land, but the production of raw tobacco remains relatively stable. Data on the reduction of areas planted with tobacco by decades are presented through the following indices:

Table 2 . Planted tobacco areas per decades

| Decade | Area | Index |
|---|---|---|
| 1976-1985 | 27.100 | 100 |
| 1986-1995 | 25.800 | 95 |
| 1996-2005 | 20.100 | 74 |
| 2006-2015 | 16.300 | 60 |

The above indices show the dynamics of planted areas and their decrease of almost 40% between the first and the last decade analyzed.

## TOBACCO PRODUCTION IN THE REPUBLIC OF MACEDONIA IN THE PERIOD 1976-2016

Tobacco is grown in almost the whole territory of the Republic of Macedonia, except in the western region. In other regions it is present with bigger or smaller deviations. The largest concentration of tobacco production is in the south-western region (municipalities Prilep, Bitola, Krusevo, Makedonski Brod, Demir Hisar, Dolneni, etc.) and central-eastern part (Veles, Negotino, Sveti Nikole, Vinica, Kocani etc.).(6) In other producing regions, tobacco is less represented crop, especially in the south-eastern, north-eastern and the western region. In the areas planted with tobacco (Table 2), the following yields were obtained:

Table 3. Tobacco yield by years

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 1976 | 34.000 | 1986 | 35.000 | 1996 | 15.000 | 2006 | 23.000 |
| 1977 | 32.000 | 1987 | 29.000 | 1997 | 25.000 | 2007 | 20.000 |
| 1978 | 30.000 | 1988 | 22.000 | 1998 | 20.100 | 2008 | 16.000 |
| 1979 | 29.000 | 1989 | 27.000 | 1999 | 32.000 | 2009 | 23.000 |
| 1980 | 23.000 | 1990 | 16.000 | 2000 | 28.000 | 2010 | 26.000 |
| 1981 | 31.000 | 1991 | 25.000 | 2001 | 20.000 | 2011 | 21.000 |
| 1982 | 36.000 | 1992 | 27.000 | 2002 | 23.000 | 2012 | 30.000 |
| 1983 | 22.000 | 1993 | 24.000 | 2003 | 23.000 | 2013 | 31.000 |
| 1984 | 31.000 | 1994 | 19.000 | 2004 | 20.000 | 2104 | 25.000 |
| 1985 | 30.000 | 1995 | 16.000 | 2005 | 23.000 | 2015 | 19.000 |
| Average | 29.800 | | 24.000 | | 22.910 | | 23.400 |

Source: Statistical Yearbook of SFR Yugoslavia, Association of Producers of R. Macedonia and Ministry of Agriculture, Forestry and Water Economy of R. Macedonia

The average production of tobacco ranged 29,800 t in the decade 1976-1985, 24,000 t in 1986-1995, 22,910 t in 1996-2005 and 23,400 t in 2006-2015. The growth of raw tobacco production in the above period can be represented through the following indices:

Table 4. Tobacco yield per decades

| Decade | Average tobacco yield (tons) | Index |
|---|---|---|
| 1976-1985 | 29.800 | 100 |
| 1986-1995 | 24.000 | 80 |
| 1996-2005 | 22.910 | 77 |
| 2006-2015 | 23.400 | 78 |

The above data on areas planted with tobacco and the resulting tobacco yields expressed through the indices are in correlation with the fact that tobacco production has 22% lower index of reduction compared to the planted areas. It confirms that the productional capacity of the planted areas is significantly increased. Unlike the above statement, the production of tobacco has a tendency of pronounced stability with an average yield of 25.0270 t, which is close to the production of raw tobacco in the pre-transitional period. Comparison between these two parameters confirms the existing intensification in tobacco production.

## ENGAGEMENT OF TOBACCO GROWERS IN THE PRODUCTION OF TOBACCO

The main resource in tobacco production is the engaged labor force, which is represented by the number of engaged co-operators in the following table:

Table 5. Co-operators engaged in tobacco production of R. Macedonia by years

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 1976 | 79.400 | 1986 | 80.250 | 1996 | 27.500 | 2006 | 29.200 |
| 1977 | 74.300 | 1987 | 57.800 | 1997 | 33.000 | 2007 | 29.700 |
| 1978 | 67.536 | 1988 | 54.400 | 1998 | 54.700 | 2008 | 31.000 |
| 1979 | 59.700 | 1989 | 49.100 | 1999 | 43.600 | 2009 | 39.000 |
| 1980 | 55.300 | 1990 | 38.800 | 2000 | 34.250 | 2010 | 41.000 |
| 1981 | 50.300 | 1991 | 40.700 | 2001 | 33.900 | 2011 | 33.200 |
| 1982 | 60.300 | 1992 | 49.350 | 2002 | 27.000 | 2012 | 15.000 |
| 1983 | 58.760 | 1993 | 53.800 | 2003 | 27.300 | 2013 | 29.100 |
| 1984 | 53.670 | 1994 | 35.400 | 2004 | 38.500 | 2104 | 42.400 |
| 1985 | 71.000 | 1995 | 24.250 | 2005 | 29.200 | 2015 | 28.450 |
| Average | 63.026 | | 48385 | | 34.895 | | 31.805 |

Source: Statistical Yearbook of SFR Yugoslavia, Association of Producers of R. Macedonia and Ministry of Agriculture, Forestry and Water Economy of R. Macedonia

Data on the number of tobacco producers (co-operators) for the analyzed period (1976-2015) show a 50% reduction in the engagement of labor force in the production of raw tobacco, which also confirms the intensification of tobacco production. This phenomenon suggests that the intensification of labor is quite advanced due to the application of modern equipment and agrotechnics in tobacco production. The average labor force engagement in the observed period and the dynamics of its noticeable decrease by decades is as follows: 1976-1986 - 63.026 co-operators, 1986-1995 - 48.385 co-operators, 1996-2005- 34.895 co-operators and 2006-2015 - 31.805 co-operators. This dynamics can also be represented through the following index of the engaged co-operators:

Table 6. Co-operators engaged in tobacco production of R. Macedonia per decades

| Decade | Co-operators | Index |
|---|---|---|
| 1976-1985 | 63.026 | 100 |
| 1986-1995 | 48.385 | 77 |
| 1996-2005 | 34.895 | 55 |
| 2006-2015 | 31.805 | 50 |

The intensity of tobacco production is even more evident through the index of co-operators engaged in this field. The number of co-operators is continuously decreasing, despite certain oscillations during this long period of time, which is primarily influenced by the natural conditions in tobacco plant growth. The fact is that the trend of intensification of this resource will continue in the following period. The intensity of tobacco production per engaged co-operator is presented in Table 4:

Table7.Production of tobacco by co-operators in the period 1976-2015 (kg)

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 1976 | 428 | 1986 | 436 | 1996 | 568 | 2006 | 790 |
| 1977 | 430 | 1987 | 496 | 1997 | 743 | 2007 | 622 |
| 1978 | 444 | 1988 | 409 | 1998 | 380 | 2008 | 512 |
| 1979 | 485 | 1989 | 561 | 1999 | 743 | 2009 | 594 |
| 1980 | 415 | 1990 | 424 | 2000 | 821 | 2010 | 643 |
| 1981 | 616 | 1991 | 619 | 2001 | 592 | 2011 | 613 |
| 1982 | 597 | 1992 | 587 | 2002 | 833 | 2012 | 962 |
| 1983 | 374 | 1993 | 446 | 2003 | 842 | 2013 | 731 |
| 1984 | 577 | 1994 | 532 | 2004 | 515 | 2104 | 728 |
| 1985 | 422 | 1995 | 633 | 2005 | 794 | 2015 | 668 |
| Average | 478 | | 514 | | 683 | | 686 |

Source: Statistical Yearbook of SFR Yugoslavia, Association of Producers of R. Macedonia and Ministry of Agriculture, Forestry and Water Economy of R. Macedonia

The data show that tobacco production per co-operator was constantly increasing, so that in the last decade (2006-2015) it was almost 200 kg higher compared to the decade 1976-1985. In figures, tobacco yield per co-operator amounted from 478 kg in the decade 1976-1985, 514 kg in 1985-1995, 683 kg in 1996-2005 to 686 kg in 2006-2015. The index of the achieved production per co-operator is as follows:

Table 8. Index of achived production per co-operator

| Decade | Tobacco yield per co-operator (kg) | Index |
|---|---|---|
| 1976-1985 | 478 | 100 |
| 1986-1995 | 514 | 107 |
| 1996-2005 | 683 | 143 |
| 2006-2015 | 686 | 143 |

According to data from the Ministry of Agriculture, Forestry and Water Economy, the yield from 2016 shows a further trend of intensification of tobacco production. Namely, in 2016, 25,443 tons of tobacco from 27,380 are produced, which according to the co-operative is 929 kilograms of tobacco, which is higher compared to the decade 2006-2015 for 243 kilograms of tobacco per co-operative.This further confirms the tendency of intensification of tobacco through the segment tobacco producer-co-operator, which is a result of the increased use of technique, technology and good cultural practices.

## CONCLUSION

All observed parameters involved in tobacco production in R. Macedonia for a period of four decades show a constant intensification, both in terms of planted area, yield per unit area, yield by co-operator and number of co-operators by years. This trend in tobacco production requires special analysis of the factors that have enabled such intensification in order to see what rationalization has been made in different stages and processes of production. The data and indices presented in the tables show that the process of intensification of tobacco production continues, with a tendency to increase in future.

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Тутун/Tobacco Vol. 67 No 7-12, 71-78, 2017

ISSN 0494-3244
UDC: 331.101.26:338.312]:005.96
Original scientific paper

# THE ROLE OF HUMAN RESOURCES IN IMPROVING THE PRODUCTIVITY OF HUMAN LABOR

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## ABSTRACT

The overall development of society is directly related to the development of human capital. The quality of human potential is the main factor of competitiveness and a driving factor for the economy that is based on workers own knowledge and skill. Today, in this constantly changing business environment, the advantages of companies are temporary and variable. The driver of this constant competitiveness are the workers themselves and therefore they are the basic resource in production.

The strategic competitiveness of companies is related to the current interpretation of market success. It can be said that human capital is responsible for successful business marketing. This is a real challenge for the leaders who form the competitive strategy.

The study of human capital , the management of human resources, labor productivity and its evaluation are the main components of maintaining the business competitiveness of companies.

The aim of this paper is to emphasize the importance of labor in overall production, to link productivity to human potential and to emphasize the role of human resources in increasing productivity.

**Keywords**: Human capital, sustainable corporate competitiveness, human resource management

# УЛОГАТА НА ЧОВЕЧКИТЕ РЕСУРСИ ВО ПОДОБРУВАЊЕ НА ПРОДУКТИВНОСТА НА ТРУДОТ

## АПСТРАКТ

Целокупниот општествен развој е во директна врска со развојот на човечкиот капитал. Квалитетниот човечки потенцијал претставува основен фактор за конкурентноста и движечка сила за економијата која се темели на знаењето и вештините на човекот. Денес, во време на променливо бизнис опкружување, предностите на компаниите се привремени и варијабилни. Двигателот на оваа континуирана конкурентност се луѓето и затоа тие значат основен ресурс во производството.

Концептот на стратешка конкурентност на компаниите е поврзан со модерното толкување на успехот на пазарот. Мора да се каже дека за стекнување на успешен деловен маркетинг се’ позаслужен е човечкиот капитал. Неговиот развиток представува вистински предизвик за лидерите кои ја формираат конкурентната стратегија.

Проучувањата за човечкиот капитал, начинот на управување со човечките ресурси, продуктивноста на трудот и нејзината калкулација го чинат главниот извор на одржување на бизнис конкурентноста на компаниите.

Целта на овој труд е да се потенцира важноста на трудот во целокупното производство, да се поврзи продуктивноста со човечкиот потенцијал и да се истакне улогата на човечките ресурси во зголемувањето на продуктивноста.

***Клучни зборови*:** Човечки капитал, одржлива компаниска конкурентност, менаџмент на човечките ресурси

## INTRODUCTION

Companies and businesses operate thanks to the existence of appropriate production resources and their employees, with their capability and skill, are the most important part of those resources. Human resource management means directing staff to activities in firms. It is based on a multitude of important categories of organizational behavior, such as: motivation, relationships of individuals and groups, organizational socialization, organizational culture, etc. Treatment of people as resources is a desire to equalize this potential with the treatment of other work resources in companies, both in terms of costs and in terms of functional link with the work process.

Human resources are a treasury of knowledge and skills that are necessary for the performance of the work activities and the progress of the companies. Investing in human resources through the reform of the education system, applying the concept of lifelong education and promoting health and safety at work, by gradually reducing employment in the informal economy (Gray Economy, Informal Economy - part of the economic activity which is not registered in official statistics [8], are key mechanisms for improving quality and increasing labor productivity.

The aim of this article is to emphasize the importance of labor in overall production, to link productivity to human potential and to emphasize the role of human resources in increasing productivity through a given example of cigarette production in the Tutunski kombinat – Prilep[6].

### Definitions

- Productivity is the measure of the quantity and quality of work performed, taking into account the cost of used resources [1].
- Productivity is the relation between output - goods and services, and input - resources: labor, capital, and management [2].
- Single productivity is the relationship between one input (one resource) and output - produced goods and services offered [2].
- Multiple productivity is the relation between multiple or all input - more or all of the resources, and output - produced goods and services offered [2].
- Human resources is an expression of the overall spiritual and physical potential of employees, which can be hidden or exploited [4].
- Human Resource Management is the design and application of formal systems in an organization to ensure the effective and efficient use of human talent to meet the organizational goals [Mathis & Jackson, quoted by Griffin (4)].

## RESULTS AND DISCUSSION

## 1. PRODUCTIVITY

### Productivity and Operational Management

Productivity is the production of goods and services. Production is not a constant size, but a process that constantly requires changes in resources, directed by input and output. **Input**, from an economic point of view, means labor, capital and management integrated in the production system. **Output,** in the economy, refers to goods and services. The effectiveness of changes in resources depends the volume of productivity, as well as the value of the produced goods and the services offered. By improving productivity we also improve efficiency. Company managers are responsible for improving productivity. Their task is to increase the value that results from the **output : input** ratio. There are two ways of managing, in order to increase efficiency, as follows:

- Reduce input, and maintain constant output.
- Increase output, and maintain constant input.

Higher production means the employment of many people in companies, which does not mean high productivity. If the goal is to ensure higher production, then it is necessary to provide a way to measure, ie calculate. The measurement of production assesses the ability of a state to improve the standard of living of its people [2].

Increased productivity resulting from the good functioning of labor, capital, and management will increase employee salaries. If labor, capital and management are increased without increasing productivity, prices rise, while increasing productivity reduces prices as more is produced from the same resources.

### Measuring Productivity

Measuring productivity can be single (direct) – when only one input resource is used to measure productivity, and multiple times.

- ❖ **Single productivity** is calculated by working hours per produced units of a product (pieces, liters, kilograms, etc.), for example:

  **Productivity = Number of produced tonnes of cigarettes : used input**

**EXAMPLE:** If the number of manufactured cigarettes is 5000 tons, and the goods were produced in 4800 hours (for a period of one year, with 16 effective hours per day for a period of 300 days), then:

Productivity = 5000 : 4800 = 1,04167 tons of cigarettes / hour

or 1041,67 kg cigarettes / hour

- ❖ **Multiple productivity** is calculated by combining service units, such as:

**Productivity = output (labor + materials + capital + other)**

**EXAMPLE of single reproduction :** Ten employees work 8 hours a day and their total cost per payroll is 8000 denars / day, while the general costs are 5000 den /

day. Their productivity is 2750 kg of cigarettes / day (10 workers x 275 kg cigarettes / daily = 2750 kg cigarettes per day).

Productivity (with old technology) = 2750 kg cigarettes per day **:** 80 working hours / day (10 workers x 8 hours = 80 hours) = 34,38 kg cigarettes / hour.

A new technology has been introduced by which ten employees with unchanged working hours and the same total costs produce 8486,7 kg of cigarettes / day, and the general costs are increased to 6000 den. / day.

Productivity (with new technology) = 8486,7 kg cigarettes per day **:** 80 working hours / daily = 106,0838 kg cigarettes / hour.

**EXAMPLE for multiple reproduction:** Ten employees work 8 hours / day, and their total payroll costs are 8000 den / day, while the general costs are 6000 den / day.

Productivity (with old technology) = 2,75 tons / day **:** 13000 den / day. (total costs 8000 den / day + general costs 5000 den / day) = 0,00021 tonnes / denar or 0,21154 kg / denar = 211,5 g / denar.

Productivity (with new technology) = 8,48665 tons / day : 14000 den / (total costs 8000 den / day + general costs 6000 den / day) = 0,00060619 tonnes / denars or 0,60619 kg / den = 606,2 g / denar.

As an example, labor productivity increases from 34,38 kg of cigarettes per hour to 106,1 kg cigarettes per hour, which means that the increase in work productivity is 2,1%.

[(106,1 – 34,38) **:** 34,38 = 2,08606 ≈ 2,1%]

Multiple productivity increases from 211,5 g / day to 606,2 g / day, which means that the increase is 1,9%.

[(606,2 – 211,5) : 211,5 = 1,86562 ≈ 1,9%]

The increase in productivity (single / working by 2,1% and multiple by 1,87%) is due to the improvement of the existing technology, ie the introduction of new modern technology which is primarily a result of human resources, i.e. of their intellectual and creative work.

### Variable values of the productivity

The increase in productivity, as already mentioned, depends on three variables values: labor (work), capital and management.

- **Labor (work)**

The increase in productivity as a result of invested work is based on a healthier, more educated and better-nourished working power (often also on a shorter working week). In the developed countries, special importance is given to maintaining and increasing the working skills of the employees, which enables rapid dissemination of new technologies, knowledge and skills.

- **Capital**

Capital is an economic value that needs to be fertilized to create added value. It can be physical (production space, machines and equipment, raw materials, semi-products, final products, etc.) and financial (cash, securities, etc.). Almost every person in work uses tools provided by capital investments. Inflation and cuts increase the cost of capital. If invested capital per employee declines, productivity decline can be expected. A greater use of labor, and the smaller the capital, reduces unemployment in the short-term, but the production makes it less productive (which is also true of agriculture), and thus decreases the daily wage in the long run.

Capital investment is often necessary, but not the only weapon in the struggle to increase productivity. Efficiency in capital utilization usually means balancing between fixed assets and those in human resources.

- **Management**

Management is an economic resource and a factor of production. The manager is responsible for the effective use of work and capital, and aims to increase productivity in front of them. In that context it includes various advances that have been achieved with the knowledge and application of technologies. New technologies require additional education, trainings etc., which creates high costs. The manager has the task of bringing all operating processes to the optimum level, and to use knowledge, technology and capital more efficiently, which will increase productivity.

## Measures to increase labor productivity

Measures taken to increase labor productivity consist mainly of using the positive influence of the factors that act in the direction of its increase.This means that the factors of labor productivity are objectively given opportunities whose realization represents the activities for increasing productivity.Implementation of these activities simultaneously represents the subjective efforts of the team employed in a company to fulfill the objective opportunities contained in the plan to increase labor productivity. These activities relate to the following:

- Improving the technical equipment of labor,
- Development and improvement of the specialization of the equipment in the company,
- Improving the qualifications and work experience of the employees,
- Rational use of working hours,
- Improving the general organizational conditions of the company,
- Individual and group stimulation of all participants in the work process,
- Control of applied methods in order to increase labor productivity,
- Audit of financial and accounting operations.

The implementation and realization of these measures and the increase in labor productivity at the level of one company (micro level) leads to:

- Reduction of production costs,
- Reduction of selling prices,
- Shortening of the working time,
- Increased production,
- Greater profit,
- Real salaries of employees,
- Greater competitiveness and representation of the company on the market, etc.

In the context of the foregoing, it can be emphasized that the overall struggle for development and progress of the productive forces comes down to using less effort in achieve greater results. Given that increasing labor productivity affects the development of productive forces and the improvement of employees' living standards, it is necessary to be the primary aim in the operating and functioning of each company.

## 2. HUMAN RESOURCES

In the past there were staffing services within working organizations that were responsible for employments and salaries. Today there are HR services (human potential) whose function is much larger (evaluation of the individual's ability in the firm, employment and dismissal, organization of trainings and other employee-related activities). The introduction of the term human resources was followed by sharp criticism because it was interpreted as identifying people as objects, which was degrading to the human being. But soon this interpretation changed and the notion has gained new importance by encompassing employees and their overall power.

### Human Resources Management

The human potential of the employees is the most valuable tool of the company. The main goal of the HR service is to improve labor productivity and therefore its activities are focused on increasing and developing this potential. In order to achieve the goal it is necessary to conduct efficient organization and management of human resources.

The term **management with human resources (human resources management)** encompasses a number of interconnected activities of which the most important are:

- Planning a job offer and demand of human resources,
- Analysis of jobs,
- Recruitment of potential candidates for employment,
- Selection among the applicants for employment,
- Socialization of newly employed,
- Training and development of employees,
- Managing the activities and behavior of employees,
- Motivation,
- Compliance with legal regulations,
- Getting out of work.

The implementation of these activities can be carried out by the Human Resources Service or an external partner of the firm whose roll is human resources. The effectiveness of the implementation of the activities is related to productivity and directly affects the financial profit of the firm. The general goal of managing human resources is to achieve success through employees. In the overall production process, employees should be treated as assets in which to invest as they are a source of competitive advantages.

There are two ways of managing human resources: hard and soft management. According to Cook and Cripps [3], hard management is trying to maximize productivity and profit, to treat workers as resources, and managers as responsible persons in front of shareholders (in the case of reward and promotion, the best worker is selected); while the soft management of human resources is directed towards the development of employees, it usually does not allow giving up dismissals and does not neglect the interests of employees (in the case of reward and promotion, the best worker does not have to be selected because there is hope that the selected person will develop in the near future). Today, hard human resource management is increasingly common, especially in private companies. Soft management is inherent in public enterprises.

Without investing in human potential and skillfully managing it, there is no competition in the market. Investing in human resources is the most cost-effective investment of investing in any other resource.

### Strategic importance of human resources management

Human resource management has a strategic importance that today is reflected in the obligation of companies to the staff. The collective of companies represents human capital composed of a combination of knowledge, experience, skill and ability of the staff. Human capital expresses the investment of companies in attracting, maintaining and motivating an effective workforce [5].

By building human capital, human resources management has the task to develop strategies for selecting competent people and keeping them at work. The staff must meet the strategic goals of the parent company [7].

Behind the success of the firm's strategy for employing or managing human resources stands a capable manager who knows quickly, boldly and decisively to make crucial decisions. Any managerial decision taken, regardless of whether it concerns the management of the firm or human resources management, is of strategic importance since it aims to maximize the existing resources of the company while providing activities with the lowest level of risk and at the same time providing maximum profits for the company. Anticipating the need for human resources with specific knowledge and skills is an important component in the strategy of human resource development.

The human resource development strategy should be conceived for possible perspectives, to increase productivity and increase competitiveness. The labor market also affects this strategy. If the labor market can easily source human resources with the knowledge and skills that the company needs, it will easily find this as a result. Otherwise, it will have to organize the development of its own staff, with previous analytical choice.

Employee development is a continuous process that, in addition to education, work experience and relationships with other people, also includes the ability of staff to prepare for new things. The difference between development and training is that training enables employees to do permanent work, and through development the employees are trained for performing activities in other jobs, including in things that have not existed before that are planned to be introduce in the future. In this way, developmental activity can serve as a strategic tool for raising the motivation and dedication of employees to work.

The involvement of employees in development programs is carried out through two types of strategies:

- Development Program Strategy designed only for higher levels of management and
- Development Program Strategy involving all employees.

Experience has shown that a human resource development strategy is not just a human resource sector but should be the goal of the whole company. The human resources department should play the role of expert and coordinator [5].

### Purpose and function of strategic human resources management

According to Aaker [1], strategic human resources management has three main goals:

- To attract an effective workforce,
- To develop the overall potential of the workforce and
- To keep qualified staff for an extended period.

The same author to strategic management prescribes several functions, of which the most common are the following: planning with job analysis, recruitment of candidates and their selection, integration of new employees, professional orientation, training, motivation, team work, promotion of communication, organizational culture and business ethics, etc.

## CONCLUSION

- The importance of human resources for work and the development of production in order to improve labor productivity insures the necessary need to create a plan that will determine the position of human resource management in relation to the overall management process in enterprises and companies.
- The specificity of human resource management indicates the necessity to carry out adequate and quality management of the personnel selection policy and a permanent investment in their development, in order to ensure their qualification and training for the successful potential of all activities and future work obligations.
- The selection of personnel is based on developed appropriate methods and techniques that enable the detection and collection of professionals who are capable and competent in an adequate and defined way to respond to the requirements of modern operation and business. Only with professional staff companies will definitely achieve success and competitive advantage, which they aspire from the beginning of their existence and functioning.
- The wide range of activities that human resource management deals with is necessary for the function and implementation in all organizations and institutions that want to sustain and improve labor productivity and become competitive in the field of their business.

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