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PLANTS SPACING AND NUMBER OF LEAVES IN THE SHADE GROWN DARK TOBACCO. I. EFFECT IN GROWTH AND DEVELOPMENT

Espaciado entre plantas y número de hojas en el tabaco negro tapado. I. Efecto en el crecimiento y desarrollo

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ABSTRACT. The study was carried out in the Tobacco Experimental Station "San Juan y Martínez", in Pinar del Río province, to determine the influence of different spacing between plants and number of leaves in growth and development of dark tobacco variety "Corojo-2006" cultivated under shade. A randomized block design with four replications and sixteen treatments by the combination of four spacing among plants (0,30; 0,35; 0,40 and 0,45 m) and four number of leaves per plant (18, 20, 22 and 24) was used. In each treatment height and diameter of stem, longitude and width of central leaf, dry mass for organ and total of plant were measured. The combination of spacing of 0,30 and 0,35 m with 24 leaves per plant facilitated a bigger growth in height of stem; while the biggest magnitudes for the diameter of stem, longitude and width of the central leaf and dry mass of root corresponded to spacing of 0,45 m with the smallest number of leaves. Biomass production in leaves, stems and total was significantly higher when combined the biggest separation among plants (0,45 m) with the biggest number of leaves (24 leaves).

Key words: tobacco, distance among plants, number of leaves, growth, biomass

RESUMEN. El estudio se realizó en la Estación Experimental del Tabaco "San Juan y Martínez" provincia Pinar del Río, con el objetivo de determinar la influencia de diferentes espaciados entre plantas y el número de hojas en el crecimiento y desarrollo del tabaco negro variedad "Corojo-2006" bajo tela. Se utilizó un diseño experimental de bloques al azar con cuatro réplicas y dieciséis tratamientos producto de la combinación de cuatro espaciados entre plantas (0,30; 0,35; 0,40 y 0,45 m) y cuatro números de hojas por planta (18, 20, 22 y 24). En cada tratamiento se evaluó la altura y diámetro del tallo, longitud y anchura de la hoja central, masa seca por órgano y total de la planta. La combinación de 0,30 y 0,35 m de espaciado con 24 hojas por planta, posibilitó un mayor crecimiento en altura del tallo; mientras que las mayores magnitudes para el diámetro del tallo, longitud y anchura de la hoja central y masa seca de la raíz correspondió al tratamiento donde se empleó 0,45 m de distancia entre planta con el menor número de hojas. La producción de biomasa en hojas, tallos y total fue significativamente superior cuando se combinó la mayor separación entre plantas (0,45 m) con el mayor número de hojas (24 hojas).

Palabras clave: tabaco, distancia entre plantas, número de hojas, crecimiento, biomasa

INTRODUCTION

Crop technology of tobacco (*Nicotiana tabacum* L.) should considerably change depending on the growing variety, the environment under which it develops and the production objectives pursued (1).

In this context, some restrictions limiting the shaded growth of a new black tobacco variety "Corojo-2006" are those related to planting density, useful leaf number per plant, harvesting time, mineral fertilization and irrigation, among others, which have been managed for the commercial varieties "Criollo-98" and "Corojo-99", regardless their own features, the ability to form more useful and larger leaves, yield potential and higher agricultural cycle than commercial varieties (2).

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The study of plant growth is particularly important in any crop, since the production reached depends on it (3). In the case of tobacco, its yield is largely determined by leaf surface development. This in turn is not only affected by the behavior of different varieties and other abiotic factors, but also by the soil area that each plant may occupy, and the leaf number allowed to be fed (4, 5); they are agrotechnical practices strongly influencing the shaded black tobacco crop, which can lead to quantitative changes in some aspects of growth, development and production levels.

Based on the above mentioned, the objective of this study was to evaluate the influence of different plant spacings and leaf number on crop growth and development of shaded black tobacco, variety "Corojo-2006".

MATERIALS AND METHODS

The study was performed under covered conditions at the experimental area No. 1 from "San Juan y Martínez" Tobacco Experimental Station in Pinar del Río province, during 2008-2009 and 2009-2010 tobacco-growing campaigns, on a low-clay-activity Alitic soil (6). The black tobacco variety "Corojo-2006" was used as planting material, since it is resistant to blue mold (*Peronospora hyoscyami* de Bary f. Sp. Tabacina), black shank (*Phytophthora nicotianae* Breda de Haan), mosaic virus (TMV) and to environmental necrosis, with high productivity and good climatic adaptation. Also, considering the organoleptic quality that characterizes Cuban black tobacco (7).

To carry out the experiment, a randomized block design with factorial arrangement was employed; the causes of variation were: four plant spacings (0.30, 0.35, 0.40 and 0.45 m) and four leaf numbers per plant (18, 20, 22 and 24). Each treatment was replicated four times (Table I).

For growth characterization, five tobacco plants were randomly selected and identified per plot 30 days after plantation was established for a total of 20 plants per treatment, depending on the characteristics of the design used. The following variables were evaluated:

- Stem diameter (cm) within an inch of its basis by a ± 0.01 mm precision.
- Plant height (cm) with graduated rule of ± 0.1 mm precision from stem basis to the corresponding disbudding height.
- Central leaf length and width according to the leaf number studied (cm) with graduated rule of ± 0.1 mm precision.
- Total leaf dry mass, according to harvest time, placed in an oven at 80 oC to reach constant weight (g) by the gravimetric method, in a ± 0.1 mg precision analytical balance.
- Stem and root dry mass recorded after the last harvest and placed in an oven at 80 °C to reach constant weight (g) by the gravimetric method in a ± 0.1 mg precision analytical balance.
- Total biomass was performed by adding dry mass of each plant organ (g).

Table I. Description of treatments

Treatments	Description (plant leaves)
1	0,30 m x 18
2	0,30 m x 20
3	0,30 m x 22
4	0,30 m x 24
5*	0,35 m x 18*
6	0,35 m x 20
7	0,35 m x 22
8	0,35 m x 24
9	0,40 m x 18
10	0,40 m x 20
11	0,40 m x 22
12	0,40 m x 24
13	0,45 m x 18
14	0,45 m x 20
15	0,45 m x 22
16	0,45 m x 24

* Check production

A row spacing of 0.90 m was used whereas the other cultural practices were performed according to what was established by Crop Technical Instructions (8).

Data were statistically processed through a double classification variance analysis whereas a Duncan Multiple Range Comparison Test was applied if there were significant differences, with p≤0.05 % using the SPSS statistical program version 11.5 for Windows.

RESULTS AND DISCUSSION

When evaluating plant growth (Table II), it was evident that the largest stem growth was reached by combining 0.30 and 0.35 m with 24 leaves per plant, without significant differences between them, which was mainly due to self-shading, on account of a higher planting density, as a result of a narrower plant spacing along with a greater leaf number.

Concerning stem height difference, it was mainly determined by leaf number per plant, indicating a larger organ as well as a recognized relationship existing between a greater plant size and an increasing plant number per unit area; also, this plant response must have been associated to plant competition (poor growth) for the weakened radiation resulted from covering, which can provoke an increased concentration of auxin, due to light reduction on these tissues, causing a higher cell elongation than the other variants studied (9).

Table II. Treatment effect on plant growth and development

Treatments (leaves/plant)	Stem height (cm)	Stem diameter (mm)	Central leaf length (cm)	Central leaf width (cm)
0,30 m x 18	144,7 g	22,3 cdef	59,7 cdefg	32,3 bcdef
0,30 m x 20	163,6 f	22,0 cdef	58,7 defgh	30,7 def
0,30 m x 22	184,3 e	21,0 ef	56,3 ghi	30,0 ef
0,30 m x 24	219,3 a	20,3 f	54,0 i	29,0 f
0,35 m x 18	147,0 g	23,3 bcd	61,0 bcde	33,3 bcd
0,35 m x 20	166,7 f	22,7 cde	60,0 bcdef	31,7 bcdef
0,35 m x 22	190,0 de	22,0 cdef	57,7 efgh	31,0 def
0,35 m x 24	214,3 ab	21,0 ef	55,3 hi	29,0 f
0,40 m x 18	143,7 g	25,3 ab	63,3 ab	34,3 abc
0,40 m x 20	165,3 f	24,0 bc	61,3 bcd	33,0 bcde
0,40 m x 22	188,0 de	23,0 cde	60,0 bcdef	31,3 cdef
0,40 m x 24	210,7 bc	21,7 def	57,3 fghi	30,3 def
0,45 m x 18	144,0 g	26,7 a	65,3 a	36,3 a
0,45 m x 20	159,0 f	25,3 ab	63,0 abc	34,7 ab
0,45 m x 22	195,3 d	24,0 bc	61,0 bcde	33,0 bcde
0,45 m x 24	206,0 c	22,7 cde	59,0 defg	31,3 cdef
Es X (+/-)	1,583	0,408	0,677	0,583
CV (%)	14,187	7,882	5,205	6,829

Some studies made on tobacco crop indicated the close relationship existing between leaf number and plant spacing with stem length (1, 10, 11, 12), since plants are exposed to different conditions of water and nutrient availability as well as solar radiation capture throughout their growing season; thus, significant variations occur in their ecophysiological components due to plant competition for the weakened solar radiation (30 %) that covering involves.

Similar effects of planting density on plant growth have been identified in *Capsicum annuum*, L. (13); *Citrus paradisi* Maca (14); *Musa* spp (15) and *Glycine max*, L. (16), when reporting the response of these species under different soil and climatic conditions of the country.

When analyzing stem diameter, it was observed that a plant spacing reduction and an increased leaf number adversely influenced stem width; the largest sizes were obtained when plants were grown at 0.45 m with 18 and 20 leaves and at 0.40 m with 18 leaves per plant without statistical differences between them. These results can be attributed to the fact that plants occupy a vital space that meets their development, which permits larger stem reserves as a result from greater efficiency and supply in the use of resources, especially the soil, coupled with fewer leaves to be fed, which tends to increase the thickness of this organ compared to a higher disbudding, besides contributing to the formation of more conductive vessels, which improves nutrient translocation and accumulation in plant tissues.

Several studies demonstrate the physiological factors associated to those studied in tobacco crop for coats (4, 17), the authors suggest that tobacco plant response to changes in plant number per area and disbudding height have a significant effect on stem development. Furthermore, these values exceed those obtained when growing the varieties "Criollo-98" and "Corojo-99" in Pinar del Río province (18) and the varieties "Habana-92"; "Habana Vuelta Arriba" and “Habana-2000” under the conditions of Granma province (19).

Plant response to changes in spacing and number of useful leaves per plant on leaf growth was significant, as analyzed through leaf size, since by combining 0.45 m between plants along with the lowest disbudding height, the highest values are reached for the central leaf length and width, without significant differences with treatments (0.45 m x 20 leaves per plant and 0.40 m x 18 leaves per plant). Such results infer that this plant response includes two effects: the top result of a more efficient photosynthetic activity, guaranteeing a greater solar radiation capture by the plant due to fewer leaves and less inner shading, and an increased net nutrient availability, as a result of available vital plant space under these growing conditions.

The importance of planting density variations on leaf morphology in tobacco crop for coats (10, 20) notes that the length and width decrease when there is a vital area reduction. When leaf number per plant increases, the size tends to decrease (4, 21, 22), because this development control management has the main purpose to guide plant development towards the leaves from the quantitative point of view; it is more noticeable as fewer leaves are left in the plant. On the other hand, it has also been found that development depends on the amount of leaves left in the plant, where assimilate distribution in this organ increases as leaf number per plant decreases, promoting a lower energy expense in the physiological, biochemical and molecular processes of plant metabolism. In addition, since this development control activity occurs earlier in tobacco, it increases gibberellin levels, stimulating lateral leaf growth when removing the apical bud. Similar results were recorded in *Solanum tuberosum* L (23) and *Glycine max*, L. (24), where the presence of fewer leaves is related to greater assimilate movement, distribution and accumulation in consumer sites, in this case the organs to be harvested.

Dry mass values of different plant organs and the overall biomass accumulation are shown in Table III, where significant differences were observed between the treatments studied. There was a direct relationship between the increased plant spacing and leaf number with the total biomass, leaf and stem dry matter production, where the combination of 0.45 m plant spacing with 24 leaves showed higher values than the other treatments, whereas the treatment using 0.30 m plant spacing with 18 leaves, the worst performance was observed for these variables with significant differences with the other variants studied.

Considering these results, it can be deduced that when there is a leaf number and vital space reduction per plant in the same row, dry matter accumulation is affected, since this production is a quantitative change which includes increased leaf length, dry mass and its area. This negative plant response could be due to a negative balance of substances produced by photosynthesis, as well as respiration and photorespiration expenditure (9). Also, it is due to competitive plant interactions for water, light, nutrients and physical space, which will have a more or less efficient effect on photoassimilate production and photosynthetic capacity, since biomass production is based on photosynthesis, respiration and carbohydrate conversion efficiency to dry matter (25).

Combining 0.45 m plant spacing with the lowest leaf number per plant influenced significantly root biomass accumulation, showing statistical differences with the other variants. This result highlights the close relationship existing between the sources of variation studied with root system development, mainly due to the effect of development control management (disbudding) at renewing growth and development of plant root system, since when they are exposed to larger soil area had better nutritional behavior. Similar considerations have been raised (1, 26), as a response to different types of tobacco to these growing conditions.

In general, results prove that variations in plant spacing and leaf number lead to qualitative and quantitative changes in the growth variables studied

Table III. Treatment effect on total biomass accumulation and per organ

Treatments (plant leaves)	Dry leaf mass (g)	Dry stem mass (g)	Root dry mass (g)	Total biomass (g)
0,30 m x 18	298,7 k	80,7 h	57,0 ghi	436,3 k
0,30 m x 20	315,7 j	88,0 g	53,3 ij	457,0 j
0,30 m x 22	328,3 hi	104,7 f	50,7 k	483,7 i
0,30 m x 24	346,0 fg	116,3 e	44,3 l	506,7 fg
0,35 m x 18	315,7 j	87,3 g	65,3 de	468,3 j
0,35 m x 20	334,0 hi	103,3 f	61,3 ef	498,7 gh
0,35 m x 22	352,3 ef	117,3 de	54,0 ij	523,7 e
0,35 m x 24	367,7 cd	128,7 c	50,3 k	546,7 d
0,40 m x 18	324,7 ij	91,3 g	71,3 bc	487,3 hi
0,40 m x 20	345,0 fg	105,3 f	66,3 d	516,7 ef
0,40 m x 22	363,7 d	121,7 de	60,0 fg	545,3 d
0,40 m x 24	379,3 b	135,7 b	56,7 hi	571,7 c
0,45 m x 18	337,7 gh	100,0 f	79,7 a	517,3 ef
0,45 m x 20	358,0 de	123,0 cd	73,0 b	554,0 d
0,45 m x 22	378,0 bc	139,3 b	67,0 cd	587,3 b
0,45 m x 24	395,3 a	160,0 a	61,3 ef	616,7 a
Es X (+/-)	2,060	1,127	0,862	2,605
CV (%)	7,558	18,888	15,269	9,216

for the new black tobacco variety "Corojo-2006" grown under covering. When shorter plant spacing (0.30 and 0.35 m) were combined with the greater leaf number (24), the maximum values for stem height were obtained. When increasing plant spacing to 0.45 m with 18 leaves left after disbudding, stem diameter, central leaf length and width as well as root biomass got maximum values. The best results in terms of leaf and stem dry mass, and total biomass were observed when using the greatest height of apical bud suppression and plant spacing.

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