INFLUENCE OF METHOD AND DEGREE OF SUCKER CONTROL ON THE CONCENTRATION OF DUVATRIENEDIOLS, YIELD, AND QUALITY OF FLUE-CURED TOBACCO

TOBACCO SCIENCE

Gregory D. Goins, David A. Danehower, and Albert R. Butler[1]

Certain cultural practices affect the concentration of duvatrienediols (α and β-4,8,13-duvatriene-1,3-diols) in flue-cured tobacco (*Nicotiana tabacum* L.) leaves. Suckers (axillary buds) are removed manually or chemically to enhance overall tobacco yield and quality. Relative to manually suckered tobacco, MH-treated tobacco exhibits altered leaf chemistry and decreased overall leaf quality. The purpose of this study was to examine the influence of the degree and method of sucker control on the concentration of duvatrienediols in green tobacco leaves, and their relationship to yield and quality of the cured product.

During harvest of ripe leaves from lower, middle, and upper stalk positions in 1989 and 1990, leaf disks were taken from flue-cured tobacco plants that had not been suckered, that were manually suckered, or that were chemically suckered. In both years, leaves at lower and middle stalk positions of not-suckered tobacco were significantly lower in concentrations of duvatrienediols, yields, and grade indexes relative to tobacco that had received close sucker control. At upper stalk positions, not-suckered tobacco had the highest concentrations of duvatrienediols and grade indexes, but yields remained consistently lower than in tobacco with close sucker control. In 1990 at lower and middle stalk positions, leaves from MH-treated plants were significantly lower in duvatrienediol concentrations than leaves from plants manually suckered to a high degree. Leaf yields increased at all stalk positions with the degree of sucker control, irrespective of the method of control employed. Overall, results suggested that a high degree of sucker control, whether manual or chemical, is important to consistently harvest tobacco that has high levels of duvatrienediols, yields, and quality.

Additional key words: *Nicotiana tabacum* L., sucker control, duvane diterpenes, maleic hydrazide, MH, fatty alcohol, quality.

INTRODUCTION

Removal of inflorescences (topping) and removal of axillary buds (suckering) are cultural practices that enhance the yield and quality of tobacco (*Nicotiana tabacum* L.) leaves. Generally, topping and suckering positively correlate with a desirable balance of tobacco flavor and aroma components (26). Poor sucker control has been associated with the production of neutral filler-type tobacco (20). Inflorescences and suckers behave as net metabolic sinks (26) relative to more mature leaves. Thus, early topping and good sucker control throughout the growing season allow greater accumulation of certain organic compounds in the leaves that remain on the plant, resulting in increased yield and higher quality. Such observations suggest the degree and method of sucker control may influence concentrations of leaf surface compounds, which contribute to the flavor and aroma quality of flue-cured tobacco.

Chemical compounds found on the leaf surface or associated with the trichome hairs of green flue-cured tobacco may include hydrocarbons, wax esters, fatty alcohols, duvane diterpenes, labdane diterpenes, and sucrose esters (23). Classes of leaf surface compounds produced by a specific cultivar are determined by its particular genotype (18). In commercial cultivars, the predominant component of leaf surface trichome exudates are macrocyclic diterpenes, α- and β-4,8,13-duvatriene-1,3-diols (23). Duvatrienediols are synthesized in the glandular head portion of trichomes (14), and they have been reported to influence the interactions of certain pests (13) and diseases (7,15) with the actively growing plant. They have also been shown to contribute significantly to the final flavor and aroma of the cured leaves (11).

In this study, the influence of sucker control practices on the accumulation of duvatrienediols was examined. Flue-cured tobacco growers rely primarily on chemical suckering agents because manual sucker removal is too labor intensive. Maleic hydrazide (MH) (1,2-dihydro-3,6-pyridazinedione) is usually applied after sprays of fatty alcohols (FA). FAs exert their activity upon contact with the developing suckers by interrupting cell membranes and desiccating axillary bud tissues (27). MH is absorbed by the plant, translocates systemically, and retards sucker growth principally by inhibition of cell division (1,19). MH-treated tobaccos have shown yield and quality differences relative to hand-suckered tobaccos (5,16,22). Such findings may be due to the physiological effects of MH per se, or they may result from the increase in available photosynthate as a consequence of the control of sucker growth (21). Differences in duvatrienediol levels that result from various sucker control practices could influence final flavor and aroma components of the cured leaves. This study was undertaken to examine the influence of the method and degree of sucker control on the concentration of duvatrienediols in green flue-cured tobacco at harvest. In addition, data were collected on the effects of these treatments on the yield and quality of the cured leaves.

MATERIALS AND METHODS

Cultural Practices and Treatments

Plot rows of 20 bordered tobacco plants, cv. McNair 373, were grown in a randomized complete block experimental design with four replications on the Border Belt Tobacco Research Station near Whiteville, North Carolina, during 1989 and 1990. General cultural procedures were in accordance with accepted flue-cured tobacco production practices. In treatments requiring topping (**Table 1**), all plants were topped at 20 leaves when two-thirds of the plots had reached the full-flower stage of development. Immediately following topping, all visible suckers were manually removed on all plants. Then sucker growth was monitored, and suckers were manually removed every 3-4 days on plants in the manually suckered plots. For manually suckered treatments, sucker length was defined as the distance from the base (point of attachment to main stem) to the tip of the longest extended leaf of the sucker. For two of the manually suckered treatments, suckers were counted and removed from the plants when they reached designated lengths of 15 cm and 30 cm (S15 and S30 treatments, respectively). The total weight of suckers from each treatment plot was determined immediately after the suckers were removed. Suckers in the "rubbed out" (RO) treatment were removed by hand using the sharpened end of a garden stake.

Immediately following topping and initial hand removal of suckers, plants in chemically suckered treatment plots received 35 mL of a 4% solution of contact-type sucker-controlling agent (Off-Shoot-T, a mixture of FAs: C_6-0.5%, C_8-

[1] Graduate Research Assistant, Associate Professor, and Research Technician respectively, Department of Crop Science, Box 7620, North Carolina State University, Raleigh, NC 27695-7620.

Contribution received February 19, 1993. Tob. Sci. 37:78-83, 1993.

42%, C_{10}-56%, C_{12}-1.5%), using a backpack sprayer equipped with boom apparatus and a single-cone nozzle calibrated at a low pressure (34.5 kPa). FAs were applied by positioning the nozzle over the apex of plants while spraying downward to allow the flow of emulsion to reach all leaf axials. This method of application did not expose leaf surfaces to appreciable amounts of contact sucker control agent. Approximately one week later, 30 mL of spray mixture at treatment rates of 42.5, 85, 170, and 340 mg MH per plant were applied using a high-clearance sprayer (calibrated for 483 kPa) with three solid-cone nozzles (Spraying Systems, TG3). This process wetted the top one-third of the leaves on each plant. All chemical sucker control agents were applied between 10:00 and 11:30 a.m. Following initial application of all chemical agents, visible suckers that had escaped initial chemical control received a direct manual application of 4% FA solution from a narrow-mouth polyethylene wash bottle. Immediately following the last harvest, suckers from the not-suckered treatments were removed, counted, and weighed. Sucker weights from topped but not-suckered (TNS treatment) treatments were used as the basis for calculating percent sucker control (**Table 1**). Plants from the TNS treatment were assigned a percent sucker control value of 0. A final treatment included in the experiment consisted of plants that were neither topped nor suckered (NTNS).

Table 1. Designations, control methods, and percent control under various sucker control treatments during the 1989 and 1990 growing seasons.

Treatment Designation	Method of Control	1989 Mean ± SD	1990 Mean ± SD
		---------% sucker control[a]---------	
Not-Suckered			
NTNS	Not-topped and not-suckered	0.0	0.0
TNS	Topped but not-suckered	0.0	0.0
Manually Suckered			
S 30	Topped and 30-cm suckers removed	29.8 ± 10.3	42.4 ± 9.9
S 15	Topped and 15-cm suckers removed	60.2 ± 4.6	59.5 ± 4.1
R O	Topped and suckers rubbed out	99.3 ± 0.2	99.4 ± 0.2
Chemically Suckered			
FA	Topped and fatty alcohols only	97.4 ± 2.2	98.5 ± 0.1
MH 42.5 mg/plt	Topped and MH - 42.5 mg/plt	-----	98.8 ± 0.5
MH 85 mg/plt	Topped and MH - 85 mg/plt	97.9 ± 0.9	99.1 ± 0.2
MH 170 mg/plt	Topped and MH - 170 mg/plt	98.6 ± 1.7	99.5 ± 0.1
MH 340 mg/plt	Topped and MH - 340 mg/plt	99.2 ± 0.9	99.4 ± 0.1

[a] Sucker weights from TNS treatment used as basis for calculating percent sucker control. Each value is an average of four replications ± standard deviation.

[b] All chemically suckered plants received FA at time of topping. MH rates expressed as active ingredients; MH = potassium salt of maleic hydrazide.

Sampling and Analyses

Ripe leaves were harvested from the lower (4th, 5th, 6th leaves), middle (10th, 11th, 12th leaves), and upper (16th, 17th, 18th leaves) stalk positions. Using a stainless steel leaf punch (1.6-cm diam), leaf disks were obtained from each plot. A complete sample from each stalk position consisted of 18 disks (one disk was taken from each of 18 randomly selected leaves during each harvest). Disks were taken 2-3 cm from the midrib approximately half-way between the base and the tip of the leaf. Disks were immediately placed into scintillation vials that were capped with foil-lined caps and placed on ice in a cooler for transport to the laboratory. Leaves from other stalk positions also were harvested as they ripened, but they were not sampled or included in data for this experiment. Leaves from a given stalk position were mixed with the other leaves from that plot and cured according to standard practices on the research station. Cured leaves from each plot were weighed by station personnel and graded by a USDA tobacco grader. Grade index values (2) for each plot were determined according to government grades.

Leaf surface components were extracted by vortexing the leaf disks in each scintillation vial for 30 seconds in 10 mL of HPLC-grade methylene chloride (CH_2Cl_2). The extract was then decanted into a clean scintillation vial. This procedure was repeated and the extracts were combined. Approximately 0.5 g of anhydrous sodium sulfate (Na_2SO_4) was added to dehydrate the extract. These extracts were stored at -10°C until they could be filtered through glass wool into 20-mL teflon-lined screw-cap test tubes. These samples were stored at -10°C until further preparation. After equilibration to room temperature, the volume of CH_2Cl_2 was reduced by half by means of an N_2 stream and gentle heating (40°C) on a Pierce Reactitherm heating block. At this point, 1.00 mL of an internal standard solution (2.0 mg heptadecane/mL of toluene) was added by pipette and the remainder of the solvent was removed. Samples were derivatized by adding a 100-mL portion of 1:1 N,O-bis(trimethylsilyl)trifluoroacetamide and dimethylformamide (BSTFA/DMF) with a gas-tight syringe, introducing an N_2 atmosphere, capping the test tube with a Teflon cap, and heating for 30 min at 75°C. After cooling to room temperature, a 100-mL aliquot of 1:1 mixture of N,O-bis(trimethylsilyl)acetamide and pyridine (BSA/pyridine) was added with a gas-tight syringe to prevent precipitation of hydrocarbons. The derivatized sample was transferred to a Hewlett-Packard microautosampler vial, capped, and placed into an autosampler.

Analyses were carried out using a Hewlett-Packard 5890A gas chromatograph (GC) equipped with a Hewlett-Packard 7673A auto-sampler, a flame ionization detector, and a 15 m x 0.53 mm i.d. (1.5 mm film thickness) J & W Scientific Durabond-5 fused-silica bonded-phase megabore column. The GC was operated with a temperature program of 160-310°C at 10°C/min, followed by a 10 min hold at 310°C. The linear gas velocity was 27.5 cm/s helium, the injection port temperature was 240°C, and the detector temperature was 375°C. Integration and reporting of data were by an IBM Instruments System 9000 computer and associated chromatography application software package. Concentrations ($\mu g/cm^2$ of leaf surface) of α- and β-duvatrienediols were calculated by an internal standard quantitation method. Total duvatrienediols was calculated from the sum of the α- and β-duvatrienediol concentrations.

All data were evaluated by analyses of variance for randomized complete block design (25). Data for experiments conducted in different years at the same stalk position could not be combined due to significant treatment by year interactions. Comparisons among treatments that were of particular interest to the study were analyzed by orthogonal contrasts.

RESULTS AND DISCUSSION

Percent Sucker Control

The overall physiological state of a tobacco plant may be affected by the direct influence of a given chemical suckering agent as well as by alterations in source-sink relationships due

Tobacco Science, 1993, 37-18, p. 78-83, ISSN. 0082-4523.pdf

Table 2. Effect of sucker control treatments on the concentration of duvatrienediols on leaves at lower, middle, and upper stalk positions in 1989.

Treatment[a]	Lower Mean ± SD	Middle Mean ± SD	Upper Mean ± SD
	----------μg/cm²----------		
Not-Suckered			
NTNS	19.2 ± 11.2	68.1 ± 25.9	76.9 ± 10.6
TNS	22.5 ± 5.1	66.3 ± 29.9	44.3 ± 3.9
Manually Suckered			
S 30	21.2 ± 7.5	73.5 ± 11.1	31.6 ± 14.4
S 15	27.5 ± 5.1	79.9 ± 7.6	34.1 ± 11.9
R O	18.9 ± 7.1	81.4 ± 23.1	34.8 ± 8.3
Chemically Suckered			
FA	28.6 ± 7.3	78.5 ± 6.1	42.0 ± 3.1
MH 85 mg/plt	22.7 ± 11.0	76.7 ± 3.3	31.1 ± 6.6
MH 170 mg/plt	22.4 ± 8.3	87.7 ± 5.1	30.4 ± 9.8
MH 340 mg/plt	19.5 ± 6.0	71.7 ± 15.0	32.5 ± 8.6
---------Contrasts[b]---------			
Manually Suckered vs NTNS	ns	ns	***
Manually Suckered vs TNS	ns	ns	**
S30 vs S15	ns	ns	ns
S30 vs RO	ns	ns	ns
S15 vs RO	ns	ns	ns
FA vs RO	ns	ns	ns
MH-treated vs RO	ns	ns	ns
MH-treated vs FA	ns	ns	ns
MH-treated Linear	ns	ns	ns
MH-treated Quadratic	ns	ns	ns

[a] Treatment designations as outlined in Table 1.

[b] ***, **, & ns denote significance at P ≤ 0.01, P ≤ 0.05, and not significant, respectively, as determined by F values for contrasts.

Table 3. Effect of sucker control treatments on the concentration of duvatrienediols on leaves at lower, middle, and upper stalk positions in 1990.

Treatment[a]	Lower Mean ± SD	Middle Mean ± SD	Upper Mean ± SD
	----------μg/cm²----------		
Not-Suckered			
NTNS	19.9 ± 7.6	33.8 ± 13.0	57.4 ± 12.5
TNS	26.1 ± 16.1	53.8 ± 9.3	72.0 ± 26.9
Manually Suckered			
S 30	31.0 ± 13.5	64.4 ± 10.6	41.7 ± 3.7
S 15	41.0 ± 11.2	108.8 ± 17.9	33.2 ± 10.7
R O	48.1 ± 11.2	110.3 ± 13.7	31.5 ± 8.0
Chemically Suckered			
FA	42.3 ± 9.8	77.8 ± 9.4	12.2 ± 5.9
MH 42.5 mg/plt	30.6 ± 4.1	59.5 ± 11.7	30.5 ± 10.6
MH 85 mg/plt	40.4 ± 17.8	70.4 ± 13.4	15.2 ± 2.3
MH 170 mg/plt	32.1 ± 17.6	74.7 ± 18.0	20.4 ± 10.7
MH 340 mg/plt	28.0 ± 10.6	72.8 ± 15.3	34.3 ± 29.7
---------Contrasts[b]---------			
Manually Suckered vs NTNS	**	***	**
Manually Suckered vs TNS	*	***	***
S30 vs S15	ns	***	ns
S30 vs RO	*	***	ns
S15 vs RO	ns	ns	ns
FA vs RO	ns	**	ns
MH-treated vs RO	**	***	ns
MH-treated vs FA	ns	ns	ns
MH-treated Linear	ns	ns	ns
MH-treated Quadratic	ns	ns	ns

[a] Treatment designations as outlined in Table 1.

[b] ***, **, *, & ns denote significance at P ≤ 0.01, P ≤ 0.05, P ≤ 0.10, and not significant, respectively, as determined by F values for contrasts.

to changes in sucker growth, which these chemicals affect (21). Both manually and chemically suckered treatments were included so that the effects of the chemical treatments per se (i.e., direct effects of MH) on diterpene production could be distinguished from the effects exerted by one physical consequence of those treatments (i.e., reduction in sucker growth). Over 99% sucker control was achieved in the RO treatments, which was much higher than the sucker control achieved in the other manually suckered treatments (**Table 1**). Very good sucker control (>97%) also was obtained with the chemical treatments. For comparisons of the effects of chemical and manual suckering, chemical treatments (FA and MH) were compared only with the RO treatment or with each other (MH vs FA). Therefore, any differences observed in chemical treatments were considered a consequence of alterations in other physiological processes of the plant due to the applied chemical rather than to the physical effect of altered sucker growth. Data from both years indicated that plants from S15 treatments received nearly twice the sucker control relative to plants from S30 treatments. Thus, treatment effects within manually suckered plots were based on incremental degrees of sucker control.

Concentration of Duvatrienediols

In 1989 (**Table 2**), there were no observed differences among treatments in concentrations of duvatrienediols at lower and middle stalk positions. The failure to detect treatment effects in this crop year was attributed to unusually high rainfall amounts following topping (data not shown). Bottom leaf-grade characteristics, which typically include lower levels of leaf surface gum (10), were observed to extend high up the stalk in 1989. Tobacco grown under excessive moisture is usually thin-bodied, less gummy, and matures faster (17). Therefore, wet growing conditions in 1989 probably had a greater influence on the physiological status of the tobacco plants than did any of our treatments.

Overall in 1990 (**Table 3**), duvatrienediols were higher than in 1989. In 1990, concentrations of duvatrienediols at lower and middle stalk positions were highest in the manually suckered treatments with the best sucker control. Plants in the RO treatments had the highest duvatrienediol levels among all treatments in 1990. Suckers may act as net metabolic sinks and assimilate photosynthate that otherwise could have been directed toward duvatrienediol biosynthesis. Interestingly, not-topped and poorly-suckered tobaccos have been noted for their neutral flavor characteristics (20,26). Such observations may partially be a result of low duvatrienediol content at harvest due to a low degree of sucker control.

In 1990 at lower and middle stalk positions, MH-treated plants had significantly lower duvatrienediol levels than plants in RO treatments (**Table 3**). Because these treatments had an equivalent degree of sucker control, this suggests that MH per se diminished accumulation of duvatrienediols in 1990. MH-treated plants had duvatrienediol levels similar to manually suckered tobacco with lower degrees of sucker control. The apparent suppressive effect of MH on duvatrienediol levels suggests that, although MH affords a high degrees of sucker control, this might come at the partial expense of lowered duvatrienediol levels. Previous researchers have shown that MH-treated leaves were lower in volatile compounds (26), petroleum ether extractables (5), and flavor (16) relative to closely hand-suckered tobacco. In addition to MH affecting certain enzyme systems (1), MH alters many physiological processes of plants including photosynthesis (4), respiration (3), and vascular function (8). MH may well affect physiological and biochemical processes of tobacco plants associated with the accumulation of duvatrienediols.

In both years at upper stalk positions, tobaccos from the NTNS and TNS treatment plots had higher amounts of duvatrienediols (**Tables 2 & 3**) than manually suckered tobacco. This finding contrasts with the results from lower and middle stalk positions of both treatments in both years. At upper stalk positions, leaves were much smaller on not-suckered treatment plants relative to leaves from other treatments. Although total leaf area was not measured, smaller leaf surface area at the upper stalk position of not-suckered treatment plants may result in greater trichome density relative to fully expanded leaves from upper stalk positions of suckered plants. The duvatrienediols are synthesized in glandular trichomes (14), and a greater density of trichomes per unit area on the smaller leaves of not-suckered plants may have contributed to higher duvatrienediol content per unit of area (18).

Comparisons of data among different stalk positions gave some indication of the importance of stalk position and plant morphology to duvatrienediol levels. In both years, duvatrienediol levels were highest at middle stalk positions (except in the not-

Table 4. Effect of sucker control treatments on yield of cured leaves harvested from lower, middle, and upper stalk positions in 1989.

Treatment[a]	Lower Mean ± SD	Middle Mean ± SD	Upper Mean ± SD
	kg/ha		
Not-Suckered			
NTNS	381 ± 14	202 ± 6	206 ± 45
TNS	453 ± 25	229 ± 27	193 ± 10
Manually Suckered			
S 30	483 ± 37	299 ± 49	268 ± 83
S 15	507 ± 43	314 ± 33	334 ± 17
R O	502 ± 50	336 ± 39	505 ± 8
Chemically Suckered			
FA	530 ± 36	341 ± 39	546 ±176
MH 85 mg/plt	536 ± 28	374 ± 30	581 ± 91
MH 170 mg/plt	558 ± 90	374 ± 17	590 ±112
MH 340 mg/plt	531 ± 25	373 ± 23	657 ±187
Contrasts[b]			
Manually Suckered vs NTNS	***	***	**
Manually Suckered vs TNS	ns	***	***
S30 vs S15	ns	ns	ns
S30 vs RO	ns	ns	***
S15 vs RO	ns	ns	**
FA vs RO	ns	ns	ns
MH-treated vs RO	ns	*	ns
MH-treated vs FA	ns	ns	ns
MH-treated Linear	ns	ns	ns
MH-treated Quadratic	ns	ns	ns

[a] Treatment designations as outlined in Table 1.

[b] ***, **, *, & ns denote significance at P ≤ 0.01, P ≤ 0.05, P ≤ 0.10, and not significant, respectively, as determined by F values for contrasts.

Table 5. Effect of sucker control treatments on yield of cured leaves harvested from lower, middle, and upper stalk positions in 1990.

Treatment[a]	Lower Mean ± SD	Middle Mean ± SD	Upper Mean ± SD
	kg/ha		
Not-Suckered			
NTNS	337 ± 8	311 ± 20	209 ± 20
TNS	366 ± 31	297 ± 4	161 ± 59
Manually Suckered			
S 30	411 ± 52	409 ± 40	409 ± 48
S 15	432 ± 20	411 ± 45	460 ± 32
R O	454 ± 43	452 ± 40	557 ± 36
Chemically Suckered			
FA	457 ± 52	469 ± 33	601 ±107
MH 42.5 mg/plt	501 ± 21	464 ± 21	583 ± 37
MH 85 mg/plt	472 ± 37	481 ± 37	566 ± 82
MH 170 mg/plt	495 ± 34	500 ± 34	585 ± 44
MH 340 mg/plt	458 ± 39	443 ± 39	561 ± 16
Contrasts[b]			
Manually Suckered vs NTNS	***	***	***
Manually Suckered vs TNS	***	***	***
S30 vs S15	ns	ns	ns
S30 vs RO	ns	ns	***
S15 vs RO	ns	ns	*
FA vs RO	ns	ns	ns
MH-treated vs RO	ns	*	ns
MH-treated vs FA	ns	ns	ns
MH-treated Linear	ns	ns	ns
MH-treated Quadratic	ns	ns	ns

[a] Treatment designations as outlined in Table 1.

[b] ***, **, *, & ns denote significance at P ≤ 0.01, P ≤ 0.05, P ≤ 0.10, and not significant, respectively, as determined by F values for contrasts.

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Table 6. Effect of sucker control treatments on grade index of cured leaves harvested from lower, middle, and upper stalk positions in 1989.

Treatment[a]	Lower Mean ± SD	Middle Mean ± SD	Upper Mean ± SD
	----------grade index value----------		
Not-Suckered			
NTNS	60 ± 0	70 ± 5	60 ± 14
TNS	65 ± 10	70 ± 4	65 ± 0
Manually Suckered			
S 30	70 ± 12	70 ± 0	70 ± 0
S 15	65 ± 10	70 ± 0	73 ± 3
R O	75 ± 10	76 ± 5	75 ± 0
Chemically Suckered			
FA	75 ± 10	73 ± 5	75 ± 4
MH 85 mg/plt	70 ± 12	78 ± 5	76 ± 3
MH 170 mg/plt	65 ± 10	79 ± 3	83 ± 3
MH 340 mg/plt	60 ± 0	78 ± 5	80 ± 6
----------Contrasts[b]----------			
Manually Suckered vs NTNS	**	ns	***
Manually Suckered vs TNS	ns	ns	**
S30 vs S15	ns	ns	ns
S30 vs RO	ns	**	ns
S15 vs RO	*	**	ns
FA vs RO	ns	ns	ns
MH-treated vs RO	**	ns	ns
MH-treated vs FA	**	**	ns
MH-treated Linear	*	ns	ns
MH-treated Quadratic	ns	ns	ns

[a] Treatment designations as outlined in Table 1.

[b] ***, **, *, & ns denote significance at $P \leq 0.01$, $P \leq 0.05$, $P \leq 0.10$, and not significant, respectively, as determined by F values for contrasts.

Table 7. Effect of sucker control treatments on grade index of cured leaves harvested from lower, middle, and upper stalk positions in 1990.

Treatment[a]	Lower Mean ± SD	Middle Mean ± SD	Upper Mean ± SD
	----------grade index value----------		
Not-Suckered			
NTNS	45 ± 6	70 ± 0	70 ± 0
TNS	56 ± 9	68 ± 3	46 ± 12
Manually Suckered			
S 30	48 ± 6	75 ± 0	46 ± 7
S 15	55 ± 4	75 ± 0	43 ± 5
R O	53 ± 2	76 ± 3	40 ± 0
Chemically Suckered			
FA	54 ± 4	71 ± 8	44 ± 7
MH 42.5 mg/plt	53 ± 2	76 ± 3	40 ± 0
MH 85 mg/plt	51 ± 2	76 ± 3	44 ± 7
MH 170 mg/plt	53 ± 2	76 ± 3	40 ± 0
MH 340 mg/plt	51 ± 2	75 ± 0	40 ± 0
----------Contrasts[b]----------			
Manually Suckered vs NTNS	**	ns	***
Manually Suckered vs TNS	ns	*	ns
S30 vs S15	*	ns	ns
S30 vs RO	ns	ns	ns
S15 vs RO	ns	ns	ns
FA vs RO	ns	ns	ns
MH-treated vs RO	ns	ns	ns
MH-treated vs FA	ns	ns	ns
MH-treated Linear	ns	*	ns
MH-treated Quadratic	ns	ns	ns

[a] Treatment designations as outlined in Table 1.

[b] ***, **, *, & ns denote significance at $P \leq 0.01$, $P \leq 0.05$, $P \leq 0.10$, and not significant, respectively, as determined by F values for contrasts.

suckered treatments for reasons previ ously described). Court (6) showec that duvatrienediols increased witl ascending stalk position except fo uppermost primings. He attributec this to harvesting upper stalk leave before they reached optimum maturi ty. Following rainy periods, a Soutl American cultivar, Galpao Comun had higher levels of z-abienol (a glan dular trichome-synthesized diter pene) at middle stalk positions than a upper stalk positions (9). Heeman e al. (12) concluded that the amounts o duvatrienediols, but not the amount of the total wax layer, are controllec by environmental factors. Darkis e al. (10) could not conclude at whicl stalk position petroleum ethe extracts were highest, due to large influences of environmental condi tions on leaf surface chemistry durin leaf development. Thus, by nature o their positioning on topped plants upper stalk leaves appeared more vul nerable to environmental exposur (especially rainfall) than leaves at th lower stalk positions.

Sucker Control and Duvatrienediol Concentration in Relation to Yield and Quality

In both years, yields consistently increased with higher levels of sucker control at each stalk position (**Tables 4 & 5**). Poor sucker control appeared to suppress yields the greatest at the upper stalk position. At each stalk position, not-suckered treatments had significantly ($P \leq 0.05$) lower yields than manually suckered tobacco, except in the TNS treatment at the lower stalk position in 1989. Yield differences among manually suckered treatments were also the most pronounced at the upper stalk position. In both years, yields were consistently higher in chemically suckered tobacco than in tobacco from RO treatments, particularly at the upper stalk position. MH-treated tobacco leaves have been shown to have greater dry weights over untreated controls, and this has been attributed primarily to MH inhibiting the translocation of photosynthate from treated leaves (3). It also has been postulated that the effects of MH after topping are maximal in actively growing areas like the upper leaves (5). However, in this experiment, yield increases observed in MH-treated tobacco relative to tobacco from the RO treatment could not be attributed totally to the effects of MH, because yields of FA-treated tobacco were not significantly different from MH-treated tobacco. This suggests that yields can be enhanced by good sucker control irrespective of the methods employed.

Except in not-suckered tobacco

reatments, the method or degree of sucker control did not)roduce consistent differences in cured leaf grade index val-)es across years or stalk positions (**Tables 6 & 7**). At lower stalk positions in both years, grade index was significantly ower ($P \leq 0.05$) in tobacco from the NTNS treatment relative o manually suckered tobacco. Conversely, grade index was significantly higher in NTNS treatments relative to manually suckered tobacco at the upper stalk position in both years. This was consistent with the observation that the highest concentration of duvatrienediols was in not-suckered tobacco reatments at the upper stalk position. In treatments involv-ng a close degree of sucker control, the concentration of luvatrienediols and grade index both were higher at the mid-lle stalk position.

In summary, it appears that the highest concentration of luvatrienediols, greatest yields, and best quality of flue-cured obacco are obtained when a high degree of sucker control is naintained, regardless of whether suckering is carried out chemically or manually. Although MH treatments may sig-nificantly lower levels of duvatrienediols compared with nanually suckered plants, labor reductions and a high degree of sucker control afforded by MH appear to offset the possible drawback of MH-related suppression of duvatrienediol accu-mulations.

ACKNOWLEDGEMENTS

This study was supported in part by funds from the N.C. Agricultural Research Service and the N.C. State University African-American Graduate Assistant Grant Program. The authors thank Thomas Bartholomew and Kimble Brock for their excellent technical assistance. Appreciation is also extended to Heinz Seltmann for assistance and helpful dis-cussions during the course of this study.

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