Studies in the Growing of Flue-cured Tobacco

II. Some Preliminary Observations on the Influence of Temperature and Light on the Growth and Quality of Tobacco

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Introduction

Investigations designed to throw light on the effect of climatic factors on the growth of tobacco have been performed in growth rooms under controlled environmental conditions by Camus and Went (1952), Coolhaas (1955), Okuma and Suyama (1955) and Pearse (1955). However, with the exception of the latter these workers were concerned with the influence of temperature, relative humidity and light duration on the actual growth of the plants and not on the quality of the cured leaf. Camus and Went for instance were able to show that night temperature has a marked effect on developmental processes such as growth rate, leaf characterisation, flowering and final fresh weight and their investigations also revealed that certain varietal differences exist, e. g. Cuba White was found to be more sensitive to thermal treatment than either Turkish Samsun or Trujillo.

Certain of these results were confirmed for variety Delcrest by Coolhaas who found that at 64° F flower initiation occurred earlier than at 91° F. He also showed that at high day temperature (91° F) the number of leaves produced was greater than at lower temperatures (70° and 75° F) and that optimum dry weight production took place at 86° F. This investigator also found that flower bud initiation was retarded when the plants were grown at a constant relative humidity of 80 per cent as compared with one of 50 per cent. Light duration was also found to have an effect on plant development, the number of leaves formed and dry weight production being greater with a photoperiod of 16 hours as compared with one of 12 hours.

Okuma and Suyama found with variety Bright Yellow that optimum growth was obtained over the range 81° to 99° F and that the dry weight of the plants rose as the illumination was increased from approximately 100 to 1000 ft. c.

Pearse working with a mosaic resistant variety which was a cross between Vamoor and Yellow Mammoth found that at low light intensities (200 ft. c.) there was a considerable reduction in growth and leaf development and that the cured leaf was of low quality having a low sugar and a high nitrogen content, when compared with leaves from plants grown at 600 ft. c. When the plants were grown at night temperature of 77° and 59° F a reduction in growth and size of leaves occurred at the higher temperature, and also the sugar content of the leaves was reduced while the nitrogen content was considerably higher. Pearse concluded that at low light intensities photosynthesis was limited and that the bulk of the carbohydrates synthesised were being utilised by respiratory processes. Similarly at high night temperatures the products of photosynthesis were rapidly used during the dark period by respiration.

It has already been reported from this laboratory that flue-cured tobacco of reasonable quality can be obtained from plants grown in a greenhouse [Ferguson and Weaving (1960)] and in order to carry this investigation a stage further a growth room providing control of environment has now been used.

The room which is 15 ft long by 9½ ft wide and 6½ ft high can be operated over the temperature range 50° to 100° F, with humidity control in the range 50 to 95 per cent R. H. Change over from day to night conditions is controlled by time switches. Air circulation within the room is 925 c. ft/min and fresh air is introduced at a rate which can be controlled up to six changes per hour.

Temperature is controlled by a steam heater and refrigeration

(WD-200H water-cooled "Frigidarie" *condensing unit) and humidification* is provided by steam circulated through magnetic steam valves which operate in conjunction with a humidistat. The room is controlled by a Honeywell-Brown electronic unit.

In the first two experiments now reported lighting was by sixty 125 watt 8 foot internal reflector "Mazda" flourescent warm white lamps giving an intensity of about 2000 ft. c. at plant level. In the third experiment six of these lamps were replaced by "Mazda" de luxe warm white models in an attempt to increase the amount of red radiation to the plants. All lamps are mounted above the roof of the room which consists of six double glazed units and heat from the lamps is extracted by two fans.

Materials and Methods

Seed was sterilized and seedlings raised by methods previously reported [Ferguson and Weaving (1960)]. Two nutrient solutions were used, compounded from $Ca(NO_3)_2$, KH_2PO_4. KNO_3. KC1. $MgSO_4 \cdot 7H_2O$ and NaC1. and their chemical compositions expressed in parts per million were as follows:—

	Ca	Mg	Na	K	N	P	S	Cl
Experiments 1 & 2	240	68	39	195	168	155	90	61
Experiment 3	200	68	—	285	150	155	90	47

Trace elements [Hoagland's A-Z Supplement—Hoagland and Snyder (1933)] and iron citrate (0.0245 g/litre) were included in the solutions which were supplied by the drip-feed method. The ten inch pots in which the plants were grown were mounted on stands whereby the level could be adjusted so that the stem apex received a constant amount of light. In all the experiments the strength of the nutrient solution was reduced after topping [Ferguson and Weaving (1960)], while fresh and cured weights and leaf areas were recorded at harvest.

The following environmental conditions were used:—

Experiment	Photoperiod	Temperature Day	Temperature Night	Relative Humidity Day	Relative Humidity Night
1	16 hours	70°-82°F	60°-72°F	50%	90%
2	"	74°-82°F	54°-62°F	50%	90%
3	"	75°-82°F	54°-62°F	60%	90%

In each experiment both the day and night temperatures were raised from the minimum employed (e.g. 70°F) by 2° per week until the maximum was reached.

Table 1—Yield Data (per plant) from the Three Experiments.

Experiment	Variety	Total Fresh Wt. (G)	Total Cured Wt. (G)	Total Leaf Area (Sq. M.)
1	Hicks	737.4	52.8*	1.51
2	Hicks	931.2	117.8	1.80
3	D.B.244	728.2	107.2	1.27

* *Leaves 11 to 20.*

Results

The yield data for the three experiments are summarized in **Table 1.** In the first experiment healthy plants with short internodes developed. Flower buds appeared 45 *days after transplanting and topping* to 20 leaves was carried out four days later. Although the leaves were harvested as they reached maturity, it was found to be impossible to carry out the yellowing stage of the cure for the first ten leaves from each plant. Instead the leaves turned brown and developed into trashy tobacco. Microscopic examination revealed that these leaves had a very low starch content. On the other hand leaves 11 to 20 contained numerous starch grains and cured normally although the final colour was somewhat darker than desired. The fact that the lower leaves could not be cured could have been due to (a) a reduction in photosynthetic activity due to the shading effect of the upper leaves or (b) to the fact that the difference between day and night temperature was too small and respiratory activity was proceeding at a rapid rate at night.

The results of chemical analysis on the curable leaf from this experiment are given in **Table 2.** The percentage ash, CaO, chloride and protein N contents were high, total sugars were low. Petrol ether extract (P.E.E.) and equilibrium moisture contents (E.M.C.) were in the range expected for U.S. flue-cured leaf.

In Experiment 2 an attempt was made to improve the amount of light reaching the lower leaves of the plants by reducing the number of plants in the room from three rows of six to two rows of six. At the same time the difference between day and night temperatures was increased from 10°F to 20°F in order to slow down respiration during the night period. Two weeks after the supply of nutrients had been stopped the uppermost leaves exhibited symptoms of potassium deficiency. Downward and inward curling of the leaf margin took place until it was at *right angles to the plane of the leaf.* Evidently the curtailment in the supply of potassium after topping had been too drastic and reserves in the plant were inadequate to support normal growth for the remainder of the experiment.

The plant yields as measured by leaf weights and areas were higher than in Experiment 1 **(see Table 1)** but once again plants with short internodes, especially at the base of the stem, developed. The color of the cured leaf was darker than desired but the leaves could in fact be cured.

The results of the chemical analysis on the lamina from the various harvests are given in **Table 3.** The percentage ash, CaO, chloride and protein nitrogen contents were again high but there was an improvement in the total sugar content in harvests 3 to 7 inclusive.

At this stage it was realized that the quantity of light reaching the lower leaves was probably too low and it was felt that this could be improved by having the walls of the room coated with a reflective material. The walls were, therefore, covered with "Melinex", a metalized Terylene foil which has high reflective properties. At the same time the floor and plant stands were painted with white gloss paint.

As has already been stated the plants which had been grown in the first two experiments had very short internodes especially at the base of the plant. One possible explanation of this was a deficiency of red light from the warm white fluorescent tubes. Six de luxe warm white lamps

Table 2—Mean Chemical Analyses on Lamina from Experiment 1.

Harvest	Ash %	CaO %	K_2O %	SO_3 %	Chloride %	Nicotine %	Protein N. %	P.E.E. %	Total Sugar %	E.M.C. %	Sugar/ Nicotine
4	18.6	4.45	3.18	2.52	0.65	1.98	1.13	6.70	15.0	12.7	7.6
5	13.4	3.27	2.34	1.88	0.70	2.56	1.03	6.86	19.2	13.1	7.5
6	12.9	3.03	2.51	1.50	0.86	3.53	1.03	8.25	16.0	13.3	4.5
7	12.0	2.99	2.35	1.64	1.22	4.23	1.14	8.27	16.1	13.6	3.8

Table 3—Mean Chemical Analyses on Lamina from Experiment 2.

Harvest	Ash %	CaO %	K_2O %	SO_3 %	Chloride %	Nicotine %	Protein N. %	P.E.E. %	Total Sugar %	E.M.C. %	Sugar/ Nicotine
1	31.4	6.61	7.56	2.48	1.74	1.47	1.17	4.81	11.6	11.1	7.9
2	23.1	4.99	4.24	2.35	1.72	1.58	1.29	6.26	2.1	12.6	1.3
3	15.2	3.37	3.28	1.61	1.22	1.83	0.89	5.77	25.6	14.4	14.0
4	13.6	2.68	3.17	1.39	1.36	1.55	0.74	6.24	25.4	14.4	16.4
5	12.5	2.81	2.60	1.48	1.11	2.78	0.79	7.92	29.7	14.2	10.7
6	10.9	2.14	2.90	1.30	1.16	3.98	0.82	7.48	24.7	13.5	6.2
7	11.2	2.48	2.39	1.49	1.06	2.11	0.91	9.44	17.5	12.0	8.3

Table 4—Mean Chemical Analyses on Lamina from Experiment 3.

Harvest	Ash %	CaO %	K_2O %	SO_3 %	Chloride %	Nicotine %	Protein N. %	P.E.E. %	Total Sugar %	E.M.C. %	Sugar/ Nicotine
1	27.9	6.64	5.89	1.96	1.40	1.32	0.76	5.07	9.4	14.7	7.1
2	19.7	4.50	4.97	1.63	1.33	1.57	0.87	5.52	15.2	15.2	9.7
3	12.3	2.76	3.52	1.14	0.82	1.45	0.75	6.11	22.5	15.7	15.5
4	9.8	2.21	2.82	0.85	0.69	1.55	0.65	7.18	21.0	14.8	13.5
5	8.1	1.98	2.44	0.72	0.69	1.98	0.65	7.44	21.2	13.9	10.7
6	7.1	1.83	2.04	0.69	0.72	2.11	0.65	8.44	19.3	13.5	9.1
7	6.8	1.75	1.87	0.68	0.77	2.01	0.64	9.20	22.0	14.4	10.9
8	7.1	1.60	1.88	0.66	1.00	2.84	0.77	10.29	20.0	14.7	7.1

which have a higher red light output were, therefore, incorporated into the lighting system. The chemical composition of the nutrient solution was also altered for Experiment 3. The amount of calcium, nitrogen and chloride in the solution was reduced and the potassium content increased. Through these alterations it was hoped to reduce the ash, calcium, chloride and protein nitrogen contents, increase the total sugars and perhaps improve the color of the cured leaves.

Variety D.B.244 was grown in Experiment 3 and although the yields were somewhat reduced **(see Table 1)** the chemical composition of the lamina was more in line with commercial tobacco **(see Table 4).** The total sugar content of the lamina from the lower reapings was greatly improved and the color of the cured leaf was lemon or orange like commercial grades. The introduction of the de luxe fluorescent lamps did not, however, improve the overall development of the plants and the basal internodes were still very short.

Discussion

These experiments had the limited objective of discovering whether tobacco of good quality could be obtained from plants which were grown under completely artificial conditions. As far as size, color and chemical composition were concerned, leaf which compared favorably with commercial flue-cured tobacco was obtained in Experiment 3. However, the actual form of the plants did not resemble that of plants grown in the field or in the greenhouse. The internodes, particularly at the base of the plants, were very short. The trashiness of leaf obtained from the bottom half of the plant in Experiment 1 probably resulted from a general lack of starch as a storage carbohydrate. These leaves had insufficient readily available respiratory substrate at the commencement of the cure and so deteriorated to trashy material. Johanson (1951) in his investigations into the problem of trashy leaf in the Australian flue-cured tobacco industry found that this phenomenon would be caused by (a) high nitrogen fertilization, (b) relatively high night temperatures and (c) low light intensities, that is, factors which can influence the laying down of reserve carbohydrates. Each of these factors could have played a part in the production of trashy leaf in Experiment 1.

When Experiment 2 was planted out the chemical examination of the leaf from Experiment 1 had not been completed so that there was no indication whether or not the nitrogen supply had been too high. It was, therefore, decided to examine the effect of increasing the differential between day and night temperatures and of improving the quantity of light reaching the lower leaves by reducing the number of plants housed in the room. These alterations had the desired result, as the quality of the cured leaf was improved and no trashy tobacco was produced.

The increase in the difference between day and night temperature would have a twofold effect. Firstly there would be a reduction in the rate of respiration during the dark period and so carbohydrate utilization would be reduced and secondly translocation of metabolites from the lower and more mature leaves to areas of rapid development would take place more slowly. Both these factors would assist in the build up of reserve carbohydrates in the lower leaves. Microscopic examination confirmed this view. Starch grains were present in the lower leaves in Experiment 2 although they were not so numerous as in the leaves from the top of the plant.

The effect of decreasing the number of plants in the room would be to increase photosynthetic activity as the lower leaves in particular would receive more light by reflection. It has been shown by Bohning and Burnside (1956) that light saturation in tobacco leaves is reached around 2000 ft.c. and that a compensation point occurs at 100-150 ft. c. Photosynthetic activity in the lower leaves in Experiment 2 would not be at a high rate as they were receiving only approximately 400 ft.c. but this would be sufficient to permit carbohydrates synthesis at a reduced rate. The low total sugar content and high protein nitrogen (**Table 3,** Harvest 1 and 2) indicated that carbohydrate synthesis had been reduced by too high a rate of nitrogen supply.

The color of the cured leaf in Experiments 1 and 2 was unsatisfactory, being dark instead of the bright lemon and orange associated with good flue-cured tobacco. This could have been due to insufficient potassium as Lovett (1959) has shown that where there is a deficiency of this element some alterations in cell metabolism occurs and brown pigments are produced more rapidly.

The alterations to the chemical composition of the nutrient solution, the composition of the light source and the reflective properties of the walls, which have already been noted, which were made for Experiment 3 resulted in an overall improvement in the quality of the leaf. The color was comparable with commercial grades and the percentage ash, calcium, chloride and protein nitrogen contents throughout were reduced by lowering the amount of calcium, chloride and nitrogen in the nutrient solution. The total sugars, particularly in the lower leaves, were increased.

There was, however, no change in the form of the plants, the characteristic short internodes still being present at the base even although the red light output from the lamp system had been increased and variety DB244 which tends to have longer internodes than Hicks had been used. It is possible that this "rosette" form of the plants in the early stages of growth is not in fact due to a deficiency in red radiation but to a temperature effect. Camus and Went (1952) found in the early stages of their experiments that the higher the night temperature the faster was stem elongation but as the experiment progressed it was found that the optimal night temperature decreased. The objective of these preliminary experiments has, however, been reached.

It has been shown that cured tobacco of reasonable commercial quality can be obtained from plants grown under completely artificial conditions and that this method of approach in studying the influence of climatic factors on leaf quality should prove to be a useful tool.

Summary

Sand culture experiments were carried out in a growth room using two varieties of flue-cured tobacco to see whether good quality cured leaf could be obtained from plants grown under completely artificial conditions. It was shown that this was possible but that both light intensity and night temperatures have a marked effect on the quality of the tobacco obtained.

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