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T H E S I S

A Study of the Chemistry of the 1928
Pennsylvania Tobacco Crop.

Submitted to the Committee on Graduate
Study and Advanced Degrees at The
Pennsylvania State College

By

Joseph B. Longenecker

In partial fulfillment of the require-
ments for the Degree of Master of Science.

June 1930

Approved:

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INTRODUCTION.

The quality of cigar tobacco depends upon the chemical composition of the leaf at the time it is harvested, and upon the kind of treatment it receives during the process of curing and fermentation. More is known about the effects of environments on the changes which occur in the leaf during the process of curing and fermentation than is known about the effects of the composition of the leaf at the time it is harvested, therefore, it is of especial importance to learn to what extent the leaf constituents govern chemical changes which occur during this important stage.

The inorganic composition of a tobacco leaf is determined, to a great extent, by the type of soil on which it is grown. Similar kinds of tobacco grown in different sections of the country differ greatly in composition, size, and other characteristics; but notwithstanding the fact that the nature of the soil and climate exert a dominant influence on composition and quality, fundamental changes in the ratios of important inorganic constituents have been obtained by fertilizer treatments.

The object of this experiment is to examine the effects of certain inorganic constituents on the fermentation changes, and to study the effects of these changes on

the quality of the leaf.

Review of the Literature.

The importance of fertilizer treatments is shown by Moss in his experiments with flue cured tobacco in North Carolina. This particular tobacco is used for cigarettes. The cured product should yield a thin yellow leaf, a product that cannot be procured unless the feeding conditions of the plant in the field are carefully controlled.

Moss⁽¹⁾ observed that a soil rich in nitrogen produced a leaf too dark in color, and, therefore, a soil that becomes depleted in this element during the period of maturation is desirable. In the process of curing yellow tobacco, the temperature and humidity are regulated carefully, yet, despite this, the process cannot be carried out successfully unless the tobacco is carefully grown for that purpose.

Killebrew⁽²⁾ in his studies on the curing of tobacco, discusses the probable importance of the basic elements in the leaf. He believes the final outcome of a tobacco crop may depend on the conversion and translocation of certain substances during the ripening and early part of the curing stage.

1. Fertilizer Tests With Flue Cured Tobacco, Tech. Bul. No. 12, 1927.
2. Tobacco Leaf, Book.

It is thought that the rate at which these processes take place depends upon the presence of excess basic elements, a considerable amount of basic substances in excess of those tied up with inorganic acid radicles. It is generally recognized that a shifting of the PH toward the alkaline side speeds up the processes involved in curing and fermentation.

The quantity and quality of the ash exert a great influence on the quality of tobacco, especially in regard to smoking tobacco. The variations in the ash constituents, due to climate, soil, variety and fertilizer used, run more or less parallel with variations in burn, aroma and taste. But Killebrew does not believe that one can speak definitely of the effect of inorganic constituents on the curing processes.

Nessler ⁽¹⁾ and Schloesing ⁽²⁾ believe the quality of the cured product, especially with regard to burn, is determined by the basic salts in the tissue. These salts are thought to function in two ways: they stimulate the processes which remove injurious substances during ripening and curing. In these reactions the ratio of cellulose to the other organic substances is changed to the proper proportionality for good burn. And second, the presence

1. Döngunguersuche zuTobak, Landw. ver. Sta. 29, 309, 1883.
2. Über die verhennlichkeit des Tabok, Landw. Ver. Sta. 3, 98, 1860.

of the salts in the seasoned tobacco catalize burn.

Haley⁽¹⁾ conducted a series of experiments on the catalytic effect of certain salts on combustion and reports results agree very well with the conclusions of earlier investigators.

During the curing of tobacco, the leaf is gradually drying out. In the meantime a series of chemical or physiological reactions are going on. The quality of the tobacco will be the resultant of the series of reactions. The changes that occur during the first stage are designated as the results of life processes, and the reactions which follow the living processes are thought to be oxidations. Loew⁽²⁾ believes the living and post mortem changes are the results of enzyme activity, and the degree to which these enzymes will function (assuming humidity and temperature are properly regulated) will depend upon the conditions in which the tobacco was grown. The leaves of cured tobacco grown in dry weather will be small, thick and inelastic; they will burn poorly and the smoke will be strong and pungent. According to Loew, the production of enzymes in dry weather is not sufficient, or, in other words, the physico-chemical make up of the protoplasmic material is not right for the proper enzymatic reactions to take place. Tobacco grown in a very wet season will

1. On the Relation of Potassium to Iron in the Combustion of Cigar-Leaf-Tobacco, Science, V70, P17, 1929
2. Curing and Fermentation of the Cigar Leaf, U.S. Dept. of Agri. Report No. 59, 1899.

not cure properly. In this case, the fault is attributed to the high acidity of the leaf, a condition which prevents the accumulation of enzymes. In moderately dry seasons, the protoplasmic structure has the proper physico-chemical make up to insure the carrying out of the proper reactions. A too great acidity of the juices in the leaf prevents the proper changes from taking place. Under good curing conditions, the acidity of the tissues decreases because some of the acids are oxidized, ammonia, liberated during deaminization reactions neutralizes others, and, thus, the free basic substances gradually increase.

Graham and Carr's ⁽¹⁾ studies of the important changes in the ash composition following the use of certain fertilizers show similar results to those obtained by Ames and Boltz ⁽²⁾. The amount of chlorine in the leaf is not necessarily a determining factor for good burn. According to the men quoted above, it depends upon what source the chlorine is derived from. The plants receiving a heavy application of stable manure, as a supplement for commercial fertilizers, contained a large amount of chlorine, and the cured leaves burned well. It is apparent that the relative amounts of the different inorganic substances in the leaf are of far more importance, with respect to quality, than are the absolute amounts. It has been suggested by a

1. Chemical Factors Determining the Quality of Tobacco, J.A.C.A. V.46, 1924.
2. Influence of Fertilizers on Composition and Quality, Ohio Expt. Sta.No. 285, 1915.

number of investigators that the ill effect of chlorine or burn is caused by a fusion of chloride salts, but according to Ames & Boltz, this idea is not supported by experimental evidence. It is possible that the chemical combination of the chlorides in the leaf, which were absorbed from the stable manure decomposition products, may be different from those resulting from the absorption of potassium chloride. This suggests that the injury resulting from chlorine may be due to its effects on the metabolism, including curing and fermentation, and not necessarily to the presence of chlorine salts in the burning leaf. The tables containing the inorganic composition of the tobacco grown under different fertilizer treatments show that as long as there is a sufficient excess of soluble basic substances, the quantity of the chlorine in the leaf is not a determining factor for burn. Further evidence, that the inclusion of carbonaceous substances in melted masses of chloride salts is not the cause of poor burn, is available. Nessler ⁽¹⁾, Haley ⁽²⁾, and other investigators have improved the burn by adding dilute solutions of basic substances to the fermented leaves. This indicates that poor burn is not caused by the chlorine salts in the leaf, but that it is due to the absence of basic substances. It is not probable that the catalytic effect

1. Dungungversuche zu Tobaks, Landw, Ver.Station 29, 1883.
2. On the Relation of Potassium to Iron in the Combustion of Cigar-Leaf-Tobacco, Science, V 70, Pl7, 1929

of a potassium base will exert such a tremendous influence on combustion that it will completely cover up a retarding effect of the chlorine salts.

Steece ⁽¹⁾ made a survey of the work done at tobacco experiment stations and reports that differences in the ~~ether, soluble~~ substances, nitrogen free extracts and fibre are not traceable to fertilizer treatments. He attributes variations in these fractions to climate, type of soil and variety of tobacco. But he does believe that the catalase activity is associated with the mineral content. A parallelism is shown to exist between the humidity of the air in the curing sheds, and the catalase content of the Leaf ⁽²⁾. Steece says that it is really a response that is governed largely by the fertilizer treatments. Oxidative processes bring about fundamental changes during curing and fermentation. The oxidative activity is said to ascend to its highest level in the period following the yellowing stage. During this time the tobacco will gain in softness and elasticity. Behrens ⁽³⁾ believes that the softness and elasticity is dependent upon the production of such hygroscopic compounds as malates. These compounds

1. Investigations on Tobacco, with Special Reference to Quality U.S. Report on Ag. Exp. Stations 1925.
2. Penn Station Bul. #196, Report 1925.
3. Weitere Beiträge zur Kenntnis des Tobak pflanze Landw Ver. Sta. 46, 1896.

are oxidation products and this may be one reason why it is important to produce a leaf with a high catalase activity. But the production of a leaf with softness and elasticity is not a guarantee of good desirable tobacco. Furthermore, tobacco, may possess, when cured, most of the desired physical properties and still be worthless as a marketable product.

Garner⁽¹⁾ believes the ether soluble fraction of tobacco tissues is traceable to fertilizer treatments. He also believes the substances which are responsible for the agreeable taste and pleasant odor are derived from nitrogenous compounds. There is a lot of evidence to be seen that lends support to this conception. Practically all tobaccos used for cigar filler are grown on soils which produce a rank growth. The leaves are deeper green and heavier than those of the tobaccos used for cigar binders and wrappers and cigarettes. In the case of the lighter tobaccos, the nitrogen content of the soil should be sufficient to support growth; while with the heavier tobaccos, it should be sufficient to accelerate vegetative growth, and thus stimulate the production of fleshy tissues. Because of the large amount of nitrogenous substances in the heavy tobacco, it is difficult to procure a fermented product that is free from substances which tend to make the smoke pungent. During the

1. Tobacco Curing, Farmers Bul. No. 523.

curing processes, undesirable compounds are removed, and the more there are in the leaf at the time it is harvested, the more difficult the task will be. Garner suggests that the curing of cigar filler tobacco can be facilitated by harvesting it at the time when the ripening processes have partially diminished the dense masses of chlorophyll matter. Successful production of tobacco depends upon the skill with which the natural physiological processes can be substituted with pathological processes. The changes which can take place during curing are of considerable magnitude, but a failure to carry the ripening processes to the proper limit cannot be corrected by careful manipulation of the agencies involved in the curing and fermentation processes, at least our information thus far is too limited to accomplish such corrections.

There is a parallelism between the nitrogenous content of a leaf and aroma, taste and nicotine content. This parallelism is the basis for Garner's belief that the compounds which are responsible for the aroma and taste are primary derivatives of protein constituents in the leaf.

Bailey and Anderson⁽¹⁾ attempt to postulate certain qualities in terms of chemical composition. They were unable to find a simple relationship between the physical and chemical standards for grading tobacco. There are a number of reasons why the physical characteristics of tobacco do

1. Chemical Analyses of Tobacco from the Nitrogen Plots
Report of Conn. Tobacco Station Bul. 10, 1928.

not check up with the chemical composition. There is an unequal distribution of substances between the midrib and web or leaf proper, and, therefore, an analysis of the whole leaf cannot be considered as a representation of the chemical influence that is exerted on the leaf proper.

it is true that an analysis of both midrib and web will tell more about the distribution of the various chemical influences, but unless the rate of movement, and the conditions regulating the movement are known, it will be impossible to state definitely the chemical influences that are exerted on a series of physiological reactions, and unless this can be done, it will be a hazardous venture to explain quality in terms of composition.

Killebrew ⁽¹⁾ and Frear ⁽²⁾ both report that they have observed a movement of inorganic substances during the ripening and curing processes. Analysis of midrib and web at the time of harvesting and again when the tobacco was cured showed a one-third increase of potash in the web and a considerable decrease in calcium. Other investigators also report a movement of salts from midrib to web and the reverse order.

It is not probable that the chemist will ever be able to get perfectly concordant results in an attempt to explain physiological processes on the basis of chemical analysis,

1. Tobacco Leaf, Book

2. ✓

for the presence of a definite quantity of a substance, in a reacting system, in one state or form may have a totally different influence than if the same quantity were present in a different state or form.

PURPOSE AND PLAN OF THE EXPERIMENT

The object of this investigation was to study the effects of fertilizer treatments on the potash content of a given strain of tobacco*, and to see if there is a correlation between the absorbtion of potash and calcium. It was also anticipated that the potash carrier, a sulphate, would have some effect on the sulphur content of the leaf. If it is possible to change the amounts of the principal minerals absorbed by the plant, then it should be possible to determine the effects of a new distribution of inorganic substances on the cured and ferments[?] leaves. Analyses of some of the inorganic and organic constituents of the cured and ferments[?] leaves were made with the idea that certain qualities of a cigar leaf can be attributed to the composition of the tissue.

The aroma and taste of a cigar leaf arises from certain organic compounds that are volatilized during combustion of the rolled leaves in the cigar. Most of the tobacco produced in Lancaster County, Pennsylvania, gives a smoke possessing pleasant aromatic^{ap} substances, but oftentimes the smoke also contains pungent substances which interfere with the enjoyment of the cigar. This pungency is due to incomplete destruction of ill smelling substances during the curing and fermentation changes, probably caused by

* Hibshman Strain, Pennsylvania Broadleaf.

improper handling during the curing season, but it is more likely caused by the conditions under which the tobacco grew and matured, conditions as effected by rainfall, temperature and fertilizer treatments. The growing conditions in the summer of 1928, in so far as weather is concerned, were favorable, therefore, the effects of the fertilizer treatments should be of some importance.

Materials Used.

Eighteen plants carefully selected from each of ten plots, one thirtieth of an acre in area, located at Ephrata, Lancaster County, Pennsylvania, were harvested at the end of the growing season in 1928 and removed to the curing sheds, where they were air cured. When the tobacco was cured, it was taken from the shed and dried at room temperature. When the leaves were thoroughly dried, they were separated from the midrib, finely ground and put into air tight containers and kept for analysis.

The chemical treatment of the plots from which the samples were taken is given in Table I.

Methods of Analysis.

The total carbon content of the leaves of the cured and fermented tobacco was determined by digesting 0.15 grams of the dried material with conc. sulphuric and Chromic acid. The chlorides and sulphuric fumes given off by the digestion mixture were trapped in a silver sulphate solution and constant boiling sulphuric acid.

Table I. Fertilizer Treatments of the Experimental Plots.

Plot No.	Manure	Cotton-seed meal	Nitrate of soda	Prec. Bone	Carbon-ate of Potash	Sulphate of Potash	Urea
	tons/a	lbs./a	lbs./a	lbs./a	lbs./a	lbs./a	lbs./a
A-1	10	400	125	130		200	
A-2	10	400	125	130		400	
A-3	10	400	125	130		600	
A-4	10	400	125	130	310		
A-5	10						
A-6		600	187	195		300	
A-7		600	187	195		600	
A-8		600	187	195		900	
A-9		600	187	195	465		
A-10		375		460	465		57

The carbon dioxide was collected in a soda lime tube.

The organic nitrogen was determined by the Kjehdal[?] method.

The total ash was determined by separating the soluble part from the insoluble and igniting and weighing the fractions separately. The soluble portion was extracted from the ash of the cautiously ignited sample with 150 cc's of hot water, evaporated to dryness and ignited to a constant weight, at a temperature not exceeding a low red heat.

The insoluble fraction was ignited in a platinum crucible at a temperature sufficiently high to drive off the combined carbon dioxide and weighed. The sum of the two portions was calculated as the total ash.

The potash was determined by the method outlined by Ames and Boltz⁽¹⁾.

The calcium and magnesium were determined by the standard methods.

The sulphur⁽²⁾ was determined by the Bomb calorimeter method.

The alkalinity of the ash was determined as outlined by Haley, Nasset and Olsen.⁽³⁾ The alkalinity is expressed as the number of ccs. of normal sulphuric acid needed to neutralize the ash of a 100 gram sample.

The moisture was determined by extraction of the water with an evacuated sulphuric dessicator, (evacuated with water suction).

Experiment I.

The web portions of the cured samples were analyzed for the total ash, the soluble and insoluble fractions, and the more important ash constituents. The organic nitrogen and the total carbon were determined; also the alkalinity of the soluble and insoluble parts

1. Ohio Agriculture Experiment Station Bul.#285.
2. Work done by W. I. Zimmerman.
3. A Study of certain constituents of leaf, and their relation to the burning quality of tobacco, Plant Physiology V 3, Pl89, 1928.

of the ash.

Experiment II.

The web portions of the fermented samples were analyzed for the total ash and the soluble and insoluble fractions of the total ash. The ash of two of the samples were analyzed for the soluble and insoluble alkalinity.

The nitrogen and carbon were determined as in the previous experiment. Table II shows the carbondioxide free ash in the cured and fermented samples; including the soluble and insoluble fractions.

Table II.

A study of the ash of the cured and fermented Pennsylvania cigar filler tobacco as modified by fertilizer treatments.

Plot No.	Soluble Ash %		Insoluble Ash %		Total Ash %	
	Cured	Fermented	Cured	Fermented	Cured	Fermented
A-1	3.99	5.49	13.47	10.45	17.47	15.94
A-2	4.64	6.01	11.95	10.63	16.57	16.65
A-3	4.76	6.38	12.02	10.10	16.82	16.49
A-4	4.51	5.70	11.89	10.61	16.40	16.31
A-5	2.79	3.77	12.85	10.28	15.63	14.55
A-6	3.57	5.40	12.33	10.67	16.41	16.07
A-7	5.00	4.90	11.48	9.94	16.47	15.39
A-8*	5.13		12.74		17.88	
A-9	4.23	4.97	12.71	10.53	16.94	15.50
A-10	3.44	5.02	12.74	10.57	16.18	15.58

* Lost the fermented sample.

Table III shows the water imbibing power of the cured and fermented tobacco. These samples were both dried at room temperature, ground and put into air tight containers. But the samples were not prepared at the same time, and it is possible that differences in the humidity at the time of preparation of the samples may have something to do with the moisture content. It, however, is of some interest to know that when the dessicated samples of the cured and fermented tobacco were exposed to room conditions, they both regained their original weight, and prolonged exposure would not induce greater absorbtion of moisture from the air of either of the samples. It is evident, as seen by the Table, that there is a slight decrease in the water embibing power brought about by fermentation.

Table III

A study of the moisture content of cured and fermented Pennsylvania cigar filler of the 1928 crop after preparation for analysis.

Plot Number	Cured %	Fermented %
A-1	5.69	4.07
A-2	5.76	3.92
A-3	5.62	4.03
A-4	5.81	4.20
A-5	4.92	4.30
A-6	5.70	4.04
A-7	5.06	3.75
A-8	5.00	
A-9	4.20	3.94
A-10	5.51	4.02

The distribution of the more important mineral constituents in the cured leaves as effected by fertilizer treatments is shown in Table IV. The alkalinity of the ash is listed in the same table in order to show how it is effected by the distribution of the ash constituents.

Table IV.

A study of the partial inorganic composition of cured cigar filler tobacco of the 1928 crop, and alkalinity of the ash, as effected by fertilizer treatments.

Plot No.	CaO %	MgO%	So ₃ %	K ₂ O%	Sol. Alk. ccs.*	Insol. Alk. ccs.*	Total Alk. *
A-1	7.95	0.84	1.14	2.57	46.7	309.6	356.3
A-2	7.55	0.75	1.16	3.16	45.7	286.5	332.5
A-3	7.45	0.80	1.32	3.35	36.6	292.5	339.1
A-4	7.76	0.81	1.16	3.21	39.5	287.0	325.5
A-5	7.65	0.96	1.18	1.74	28.1	287.0	315.1
A-6	8.09	0.92	1.40	2.81	25.1	332.4	357.5
A-7	6.97	0.82	1.47	3.26	37.3	296.0	333.3
A-8	7.01	0.85	1.68	3.27	30.1	300.0	330.6
A-9	7.23	0.81	1.05	2.84	35.4	309.1	344.5
A-10	7.67	0.85	1.49	2.23	31.3	332.3	363.6

* ccs. of normal sulphuric acid required to neutralize the alkalinity of the ash of 100 grams of material.

Table V shows the nitrogen and carbon content of the cured and fermented leaves. There apparently is a small change in the N/C ratio brought about by an in-

crease in the nitrogen content during fermentation.

Table V.

A study of the N/C ratio of cured and fermented Pennsylvania cigar filler tobacco of the 1928 crop as modified by fertilizer treatments .

Plot Number	Total Carbon %		Total Nitrogen %		N/C	
	Cured	Fer-mented	Cured	Fer-mented	Cured	Fer-mented
A-1	40.25	40.74	4.42	5.17	1:9.12	1:7.88
A-2	42.06	40.01	4.38	5.00	1:9.58	1:8.19
A-3	40.96	41.16	4.29	4.81	1:9.54	1:8.56
A-4	42.05	41.62	4.80	4.71	1:8.76	1:8.84
A-5	41.87	41.17	4.47	4.95	1:9.36	1:8.32
A-6	40.92	41.07	4.51	4.93	1:9.10	1:8.33
A-7	40.95	41.56	4.57	5.14	1:8.96	1:8.10
A-8*	42.11		4.44		1:9.48	
A-9	40.07	41.36	4.51	5.18	1:8.88	1:8.00
A-10	42.24	42.03	4.24	4.45	1:9.96	1:9.44

* Lost fermented sample

Experiment III.

Samples of the fermented tobacco were used for the filler of cigars rolled with a Connecticut binder and Sumatra wrapper. These cigars were scored according to their smoking qualities. The factors considered were burn, fire holding capacity, color of ash, coherence of ash, aroma and taste. The writer scored the cigars carefully, but feels that the results obtained are rather unconvincing. The

method of scoring the smoking qualities does not bring out certain factors that should be emphasized.

Table VI

The scoring of cigars prepared from Pennsylvania cigar filler tobacco as modified by fertilizer treatments and rotation of crops.

	Burn	Fire Holding Capacity	Color of Ash	Coherence of Ash	Aroma	Taste	Total
Plot Number	20 points	10 points	10 points	10 points	25 points	25 points	100 points
A-1*	18	10	10	10	14	12	74
A-2	14	10	7	6	20	18	75
A-3	14	10	6	7	18	18	73
A-4	12	10	5	8	5	10	50
A-5	17	10	8	8	12	12	67
A-6	16	10	8	5	20	17	76
A-7	14	10	6	8	15	12	65
A-8	14	10	6	8	12	12	62
A-9	16	10	8	10	10	12	66
A-10	18	10	10	10	10	10	68
G-1	16	10	8	6	14	14	68
G-2	17	10	8	8	16	15	74
G-3	14	10	8	7	14	10	63
G-4	14	10	10	6	16	15	71
G-5	14	10	8	6	15	17	70
G-6	16	10	7	8	15	16	72
G-7	16	10	6	8	17	15	72
G-8	17	10	6	7	18	17	75
G-9	16	10	8	8	20	20	82
G-10	16	10	10	8	15	12	71

* The tobaccos grown in the A series were grown on a three year rotation plan. Wheat - sod - tobacco.

4 Four year rotation. Wheat - sod - corn - tobacco.

Weather conditions and rainfall at Ephrata, Pennsylvania, for the year of 1928.

During the months of April and May alternate cool and warm weather prevailed; there was very little rainfall. The month of June was cool and wet. During the time of transplanting it was very wet. The physical condition of a wet soil is very much altered by exerting a pressure equal to the weight of a horse or tobacco planter upon it. Evidence of this has been seen in greatly reduced yields, fifty per cent reductions, when the tobacco has been transplanted in very wet soil. Transplanting by hand in wet soils will not have a bad effect on the growth of the crop. During the month of July the weather was warm and frequent thunder storms supplied sufficient moisture for good growth. From the latter part of July until the second week in September it was very warm, and, although very little rain fell during this period, occasional thunder storms modified the drought. Cooler and dry weather prevailed during the latter part of September. The month of October was also cool and dry. Table VII shows the rainfall per month during the year of 1928.

Table VII.

Rainfall during the year of 1928.

January	2.71	inches	
February	3.89	"	
March	2.74	"	
April	4.98	"	
May	1.92	"	
June	9.43	"	
July	3.65	"	
August	4.75	"	
September	3.37	"	
October	0.77	"	
November	1.59	"	
December	0.76	"	
Total	40.56	"	(Normal - 42.96)

Discussion of Results.

The total ash listed in Table II, under Experiment I, shows lower values than do most tables of total ash analysis. The reason for the low ash content is due to a modification in the method of procedure for the total ash analysis. It was learned, during the attempt to determine the ash by the usual method, that it is very difficult to get a constant weight by cautiously igniting the combined water soluble and insoluble fractions. Most of the carbonaceous material can be driven off at a low temperature. When this is done, the soluble is separated from the insoluble ash and the latter fraction can then be ignited thoroughly without danger of driving off any of the volatile salts, or inclusion of carbon in the fusible ash constituents. According to the usual method of analysis, after the insoluble portion was thoroughly ignited in a platinum crucible, the soluble portion was added to the ignited residue, and the combined fractions were cautiously ignited (low red heat) and weighed as the total or crude ash. The Pennsylvania cigar filler tobacco contains much lime, and the greater part of it, when ignited, is present in the oxide form. When the soluble portion of the ash is added to the ignited residue, the oxides become hydrated, and perhaps during the evaporation of the water and low

ignition of the ash considerable carbondioxide combines with the hydrated lime. Because of the presence of soluble salts of potassium and sodium, the mixture cannot be ignited to a sufficiently high temperature to drive off the water of hydration and the combined carbondioxide. The heat applied is probably just sufficient to drive off some of the water of hydration and carbondioxide, and this is the reason why it is difficult to get a constant weight when the combined fractions are ignited. It was thought advisable to determine the two portions separately, and the total ash content obtained by this method is about four per cent lower than the ash obtained by the previous method. There can be no objections to the procedure on the grounds that some of the ash constituents were driven off by ignition at a high temperature, for the temperatures applied are the same as in previous methods.

The original purpose of the total ash determination of the cured and fermented samples was to obtain a method for measuring the loss in dry weight which occurs during fermentation. It was found that the total ash content of both samples is about the same, a condition that one would not expect to find under the circumstances. It is apparent that a loss in dry matter cannot be accounted for on the total ash basis, yet the heat generated and the strong odors

evolved during fermentation indicate that losses do occur, and the fact that there is no increase in total ash is not conclusive evidence that no losses occur; but it does suggest that the changes which accompany fermentation are more involved than were imagined. The ash content shows a slight decrease in the fermented samples; this difference may not be of any significance, but something has happened to the proportionality which exists between the soluble and insoluble ash in the cured samples during the process of fermentation. The insoluble fraction decreased while the soluble fraction increased. The simplest explanation for this behavior is that part of the insoluble ash of the cured leaf was converted to a soluble form during fermentation. This may be true. It seems more logical to account for the change on this basis than to seek an explanation on the grounds that there is a movement of ash constituents to different parts of the leaf during fermentation. But a number of investigators reported that they have observed a translocation of ash constituents, a redistribution of potash, calcium and other elements during the period from the time of harvesting to the end of the bulk sweating stage. In this period, Killebrew ⁽¹⁾ states the potash content in the leaf increased one-third and the calcium decreased

1. Tobacco Leaf, Book.

considerably. Frear⁽¹⁾ reports a movement of the same elements in the same direction corresponding to the direction observed by Killebrew. A possible explanation for the reverse movement of the salts is suggested. The web portion of the leaf consists of thin layers of tissues and the loss of moisture is considerably more rapid than from the fleshy tissues composing the midrib and the stem. During the drying processes chemical changes of a catabolic nature are taking place, and some of the products formed are acidic in nature. The calcium salts of these acids are less soluble than the salts existing in the web previous to the acid formation, and the acids are precipitated as the calcium salts. This condition continues until the web becomes dry and the rate of acid formation approaches zero. In the meantime the midrib and stem are drying out much more slowly, and their acid formation will continue long after the reactions in the leaf have been slowed by dessication. The concentration of the transportable calcium salts in the midrib and stem will fall below that of the web, and diffusion toward the midrib will occur. In the meantime, it does not seem reasonable that there will be a movement of salts in air dries tissues in a post mortem state. But the movement of salts need not extend more than one inch from the midrib in order

1. Pennsylvania Station Reports for 1916-1922.

to produce noticeable differences in the ash content of the leaf.

Some attempts have been made to study the soluble and insoluble fractions of the ash. The alkalinity studies made on the ash of cured and fermented leaves are of some interest. The alkalinity of the soluble portion of the ash in fermented leaves was one-third higher than that of the cured leaves. This agrees very well with the increase in the soluble ash of fermented tobacco. The soluble alkaline substances, when titrated with a strong acid, behave as carbonates, that is, phenol phthalein titrates one-half of the alkalinity. The alkalinity is due to a di-acid base. If the basic substance is calculated as potassium carbonate, the weight of the carbonate was equal to two-thirds of the weight of the soluble ash. One is justified in concluding, from the above results, that two-thirds of the soluble ash is alkaline in nature, while the rest must be tied up with neutral radicles. The soluble portions of the ash of fermented tobacco contains practically all the potash that is in the tissue. If the potash found in the soluble ash is calculated as a carbonate, it is equal to three-fourths of the total soluble ash. Since the part of the soluble ash that contributes toward the alkalinity (on a theoretical basis) is two-thirds of the total ash and the highest amount of potassium carbonate that can be present in the fraction is three-fourths of the

total amount, it can be definitely stated that at least twelve per cent of the potash is present in the form of a neutral salt.

It was surprising to learn that the loss in the soluble ash during fermentation was not accompanied by a corresponding decrease in the insoluble alkalinity. This would lead one to believe that there is no movement of calcium from the web to midrib, or a conversion of insoluble alkaline substances to a soluble form during fermentation. If either one occurs, the final result of the complex changes must compensate in some way for an increase in the soluble alkalinity at the expense of the insoluble alkalinity. The difficulty of explaining what happened to part of the insoluble ash during fermentation might easily have been averted, if analysis of the midrib before and after fermentation had not been neglected. One explanation for the loss in insoluble ash which has not been considered thus far is a removal of sand and dirt from the exterior of the leaf during the handling of the tobacco in preparation for bulking. The cured leaf is coated with a gummy substance. It is natural that sand and dirt would stick to the gummy substance. During the heating of the tobacco, the gummy substances are destroyed, the surface becomes less sticky, and particles which had been adhering to it will drop off. On the other hand, there may be salts

in the gummy secretions and these would be lost to the leaf when the gum is disintegrated.

The alkalinity of the insoluble ash is due to the calcium in the tissues. If the calcium content of the leaf is known, it should be possible to predict what percentage of the total calcium is contributing toward the alkalinity by making use of the alkalinity number. The amount of calcium oxide that is equivalent to the quantity of acid used is nearly equal to the total calcium content. There is, however, a small amount of magnesium in the leaf also, and it is possible that it is responsible for a small amount of the alkalinity. But, on taking everything into consideration, it can be definitely stated that about ninety per cent in the leaf exists in such a form which, upon ignition, will be alkaline in nature. It is evident that only a small percentage of the calcium is in equilibrium with non-combustible acid residues.

Considerable work has been devoted to the study of the effects of certain salts in the nutrient solutions on the alkalinity of the ash of the plant tissue. Certain salts, especially chlorides, lower the alkalinity of the ash; whereas others, such as sulphate, do not have much effect on the alkalinity. Chlorides are absorbed in proportion to their concentration in the nutrient solution. In soils of high concentrations of chlorides the basic

radicles in the plant are tied up with chlorine, especially the soluble basic substances, and the alkalinity of the ash will be low. In the case of sulphates, the absorption of this ion is not proportional to the concentrations in the soil solution. The absorption of sulphates is apparently regulated by the synthetic processes within the leaves, for plants showing similar rates of growth absorb equal amounts of sulphur regardless of the quantity of available sulphates in the soil. This is true only insofar as availability of the sulphate depends upon the amount that is applied to the soil. Reactions in the soil may tend to keep the quantity of available sulphates more or less constant, regardless of the quantity applied.

In previous papers it is stated that the calcium content of the leaf is inversely proportional to the potash content. Nasset studied the composition of the 1925 and 1926 crops of Pennsylvania tobacco. In the dry summer of 1925 the potash content was very low and the calcium content was rather high. In the following year, with a wet summer, the potash content increased materially and the calcium content decreased considerably. If the small increase in the potash absorbed is responsible for the decrease in the calcium absorbed, then it must have a profound effect on the quantity of the calcium in the tissue. In the year of 1926, the potash content was about twenty per cent higher and the calcium content about fifteen per cent lower

than in the previous year. The same absorption behavior was not observed in 1928. A one hundred per cent increase in the amount of calcium absorbed did not lower the calcium content at all. Table IV shows that even though it is possible to increase the potash from 1.74 to 3.35 per cent, the high availability of lime in the Hagerstown limestone soil overrules what effect the potash may have on the absorption ratio.

An inspection of Tables showing the inorganic content of many kinds of tobacco grown on widely different soils shows a range in the ratio of potash to calcium from 1:4 to 1:0.71. There is some evidence that an inverse proportionality exists between potash and calcium, for in most tobaccos the sum of these two elements in the leaf is more or less constant, being about ten per cent of the total ash. The extent to which the potash in the leaf can be increased by fertilizer applications depends upon the type of soil on which it is grown. The absorption equilibrium may be a function of the concentration of the sum of the two elements in the plant sap, for there is considerable evidence available from absorption studies that the amount of substances taken up by a plant is not governed by the quantity of the substance needed for maximum growth. On the other hand there is a possibility that the quantity of potassium absorbed is governed by its availability.

An application of nine hundred pounds of potash per acre is no guarantee that there is plenty of it available throughout the growing season; it may become "fixed" in the soil. If this is true, applications of potash at definite intervals during the growing season might bring about a response similar to that obtained by applications of nitrates at different intervals during growth.

It will be noted that the nitrogen content of the tobacco leaves is very high, and this may explain why the Lancaster County cigar filler produces a pungent smoke. The nitrogen content is constant for all the samples. It would be expected to see a parallelism between yield and nitrogen content. The only constituent that varied with yield was potash. Apparently there is no noticeable accumulation of the more important constituents, such as nitrogen, calcium, magnesium and sulphur resulting from potash deficiency, for the distribution of these substances is about the same, regardless of yield.

The main purpose of this investigation was to study the effects of the inorganic composition on the changes which occur during fermentation. There are three changes in the chemical composition which accompanied fermentation.

1. There was an increase in the nitrogen content. The nitro-carbon ratio changed from about 1:9.5 to 1:8.5.
2. There was an increase in the soluble ash and a corresponding decrease in the insoluble ash.
3. There was an increase

in the alkalinity of the soluble ash. The magnitude of all the changes was about the same, regardless of the variation in the potash content. It was hoped that the rate and magnitude of the changes which occur could be traced to certain inorganic constituents, and the reason why there is not a noticeable difference traceable to any particular constituent may be due to the fact that the process was not continued long enough for the particular constituent to exert its full powers on the changes. It may be of interest to note that the tobacco grown on plots receiving carbonate of potash instead of sulphate of potash show a smaller increase in total nitrogen during fermentation than do the others.

The nitrogen-carbon ratio becomes more narrow during fermentation, but it cannot be explained as influenced by certain fertilizer treatments. In most cases, there is a close parallelism between increase in nitrogen and increase in soluble ash, but it is evident that the one is not dependent upon the other. For example, A-7 showed no increase in nitrogen, but an increase in soluble ash.

An increase in the percentage of nitrogen is not followed by a decrease in the percentage of carbon. If the loss in dry matter is calculated on the basis of nitrogen increase, it is equivalent to ten per cent of the original dry matter. It does not follow that a ten per cent

loss in dry matter should result in a decrease in the percentage of carbon, for all of the organic compounds contain much carbon, and no matter which compounds are oxidized to carbondioxide the original percentage of carbon would remain about the same. Smirnov⁽¹⁾ measured the oxygen consumed and the carbondioxide evolved from piles of fermenting tobacco, and reports that the evolution of carbondioxide is greater than can be accounted for by the oxygen consumed. Some of the compounds must be reduced in order to supply oxygen necessary to oxidize other compounds.

Smoking tests made on the cigars do not show a clear relationship between the composition and quality. The tobaccos used in these tests, however, were not aged; they were not put through a series of heats, but sweated once and then rolled into cigars, and, consequently, it was "green". The cigars were somewhat strong, bitter and pungent. As tobacco ages, the harshness and pungency decreases. The chemical changes which accompany a reduction of harshness are not definitely known, but it is obvious that the nitrogen changes exert an important influence. It may involve a reduction in the total nitrogen content, or merely a conversion to different forms. Despite the "greenness" of the cigars, such qualities as burn, fire-holding capacity, color of ash and coherence of the ash could be satisfactorily scored.

1. Zur Kenntnis der Tabak fermentation Beuchte des Centralin
Institute fur Tabak forschung, Krasnodar Lief 34, 1927

All the cigars burned well, and the fire-holding capacity was good. There seems to be an inverse relationship between the coherence of the ash and the aroma. The coherence of the ash must be a function of the nature of the inorganic constituents in the burning tissue, for it appears that equal amounts of the inorganic constituents will not exert the same influence on the ash formation. The difference in the coherence of the ash may indicate rates or degrees of combustion that cannot always be distinguished by the color of the ash. Aroma depends upon the rate and degree of combustion. Coherence probably has a lot to do with the degree of combustion. If conditions are right for causing considerable coalescing of the partially ashed particles, good coherence will result, combustion will be less complete, and the aromatic substances may not be liberated. The chemistry of a burning cigar must be explained in terms of the systems that are operating during combustion. The end products of the burning mass are determined by the relative amounts of the substances and the degree of the dispersion of these substances before and during combustion. Good coherence is not conducive of maximum evolution of aromatic compounds. The maximum values assigned to coherence and color of ash should be determined by the coherence and color of ash which coincide with the aromatic smoke.

The sum of the points allowed for each quality factor does not do justice to the cigar as a whole. Cigars 2, 3 and 6 of the A Series are much better than the rest, yet the sum of the total points shows very little difference. 7, 8 and 9 are pungent. The terms pungency and poor aroma are not synonymous. The smoke of a cigar may be very pungent and still possess a good aroma. Pungency is not noticeable unless a cloud of smoke is breathed into the nostrils, whereas aromatic substances can be detected throughout a whole room. The substances producing the aroma are much more volatile than those causing pungency.

Summary

If two-thirds of the total potash exists in the leaf in such a form that upon ashing will show its full basic powers, a variation of the potash content from 1.74 to 3.35 per cent will not have much effect on the burn, fire-holding capacity and color of the ash. A cigar scoring high in these three qualities is not a guarantee for good aroma.

The calcium content in the leaf is not²⁸ effected by a variation of the potash content within the limits that it was possible to vary it, by fertilizer treatments, under the growing conditions for the summer of 1928.

The fermented tobacco containing the highest percentage of soluble ash produced the most desirable smoke.

Cigars made of tobacco which increased the least in soluble ash during fermentation were extraordinarily strong.

The absolute amounts of the inorganic constituents are of less importance, with respect to burn, than are the relative amounts. But if the relative amounts of the inorganic constituents were held constant from year to year there would still be a great variation in the quality of the tobacco; therefore, it is obvious that the physico-chemical state of the protoplasm, which is largely dependent upon the environment, of the growing and curing season, has a lot to do with the quality of the final product.

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