A Titrimetric Method for the Determination of Nicotine in Tobacco Tissues

RAY F. DAWSON, DePauw University

A great many accurate and valuable methods have been developed for the quantitative estimation of nicotine in samples of tobacco tissue. The procedure most generally employed is that recommended by the Association of Official Agricultural Chemists in which nicotine is precipitated as the silicotungstate from a steam distillate of the tobacco tissues. This method is time-consuming, however, and requires careful temperature control during the ashing process. A new method has been devised which permits the rapid separation of nicotine from the remainder of the volatile bases which pass over with nicotine during the steam distillation. In its principal features, this method combines the steam distillation portion of the official method[1] with the ammonia distillation procedure described by Vickery and his associates.[2]

At ordinary pressures nicotine passes quantitatively into the vapor phase when steam is passed through its aqueous solution. In this state it is generally associated with ammonia and with small amounts of unknown bases. The vapor phase, when passed into standard acid, loses its basic constituents which, consequently, may be determined by direct titration with alkali. Since nicotine is nearly as strong a base as sodium hydroxide, it titrates quite readily. Under reduced pressure and at the boiling point of water, ammonia and the remainder of the lower boiling bases pass from the aqueous phase into the vapor phase; nicotine remains behind. The former may be trapped in standard acid and again titrated. The nicotine equivalent of the difference between the two titrations may then be calculated. If a positive determination is desired, the nicotine may be redistilled from the residues and titrated directly.

The advantages of the method are the rapidity with which individual analyses may be completed and its ease of application to semi-micro procedures. If suitable apparatus is available, duplicate determinations may be completed parallel with one another in a period of one hour or less. Furthermore, samples of tissue weighing 0.20 gm. may be employed, provided the volume of the distillation system is sufficiently small. Duplicate determinations usually check within 3% or less of one another. Very frequently the agreement is exact. Repeated tests have shown that, under the conditions prescribed for the conduction of the vacuum distillation, nicotine is not volatilized in sufficient amounts to be detected by qualitative reagents such as silicotungstic and picric acids even when the distillate is concentrated to a small volume. When compared with the usual silicotungstic acid quantitative method, the

[1]Official and tentative methods of analysis of the association of official agricultural chemists, 1935. Fourth ed. P. 60.

[2]Pucher, G. W., H. B. Vickery, and C. S. Leavenworth, 1935. *Ind. Eng. Chem., Anal. Ed.* 7:152.

new procedure gives results which are in reasonable agreement. In Table I are listed the results of a few repetitive and comparative analyses in which the new method has been employed.

TABLE I.—Percentage of Nicotine in Dry Tobacco Samples by Titrimetric Method and by Gravimetric Method

Sample	Titrimetric Method	Gravimetric Method
	Percentage	Percentage
1	9.57	
	9.49	
2	3.96	
	3.94	
3	0.90	
	0.90	
4	0.24	
	0.25	
5	9.50	9.72
6	5.85	5.98
7	0.63	0.65
8	0.42	0.40

Since the new method represents merely the combination of two procedures which have been described very thoroughly elsewhere in the literature[1,2], a brief outline of its adaptation to semi-micro analysis will be included here.

The Titrimetric Method

The sample of dry, finely powdered tissue is well mixed, and 0.20 gm. of the powder is transferred from a weighing bottle to a 125 cc. Erlenmeyer flask and suspended in 20 cc. of water. If an aqueous extract of tobacco tissues is employed, the aliquot to be used for analysis should contain not less than 0.5 mgm. of nicotine. Three drops of paraffin oil and 0.2 cc. of a concentrated nitrogen free solution of sodium hydroxide are added, and the flask is attached to a steam distillation apparatus. The receiver should be charged with an appropriate volume of 0.0200 *N* HCl delivered from a micro burette which is calibrated in units of 0.02 cc. Distillation is continued with bottom heat until about 50-75 cc. of distillate are collected, depending upon the nicotine content of the tissues. Titration to the end-point of a methyl red-methylene blue indicator yields the total volatile base content of the tissues.

The distillate is then quantitatively transferred to a 200 cc. round bottom flask, and a few drops of a sodium hydroxide-sodium borate solution (5 gm. of borax dissolved in 100 cc. of 0.5 *N* nitrogen free sodium hydroxide)[2] are added. Standard acid is placed in the suction flask-

receiver, and the vacuum pump is turned on while the nicotine solution is still at room temperature. When the solution begins to discharge its dissolved gases vigorously, a water bath previously warmed to 42°C. is raised around the flask and distillation continued for 20 minutes. At the end of this time, the water bath is lowered, the vacuum released very slowly, the suction flask removed from the apparatus, and the distillate titrated as before. The difference between the two titrations corrected for the apparatus blank (usually between 0.02 and 0.04 cc. of 0.0200 *N* acid) and multiplied by the nicotine equivalent of the standard acid yields the quantity of nicotine contained in the sample of tissue.

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