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GWA/CAL/61

THE EFFECT OF AMMONIA ON THE DETERMINATION OF SUGARS IN TOBACCO

Various experiments are being carried out at R. & D.E. to test the effect of additives to tobacco on the composition of smoke. Ammonia is one of the additives being investigated and analytical results obtained so far indicate a considerable loss of sugar as a result of this treatment.

Normally sugars in tobacco are determined using Somogyi's reagent which contains copper sulphate. As ammonia also reacts with copper sulphate, tests have been carried out to check whether ammonia present in tobacco after treatment would interfere with the subsequent sugar assay or whether the actual sugar content of the tobacco had changed during ammonia treatment.

The following experiments were carried out.

1. A solution of pure glucose, with and without added ammonia was analysed.

Conc. of ammonia	Wt. glucose taken (mg)	Wt. glucose by analysis (mg)
nil	2.00	2.00
N/100	2.00	1.95

These results show that ammonia does not have an appreciable effect on the Somogyi method for the determination of reducing sugars.

2. A bulk aqueous extract of a flue-cured tobacco (CN 102) was prepared and part of the extract was treated with ammonia to give a final ammonia concentration of N/100. On addition of ammonia, the tobacco extract turned a very dark almost black colour. Both fractions of the extract were clarified

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using lead acetate and potassium oxalate. There was no difference in colour between the ammonia treated and untreated samples after clarification. The sugar content of each fraction was determined in the usual way.

Untreated sample - 17.1% reducing sugars
Ammonia Treated sample - 15.8% " "

The results show that ammonia treatment reduces the amount of reducing sugar determined by the standard method.

3. Samples of tobacco (CN 102) were extracted with a) water and b) N/100 ammonium hydroxide. The extracts were clarified and the reducing sugars determined in the usual way.

Aqueous extraction - 17.0% reducing sugars
Ammonium hydroxide extraction - 16.0% " "

These results are very similar to those obtained above (2) showing that ammonia is not affecting the actual extraction of sugars from the tobacco.

4. Another bulk aqueous extract was prepared and clarified with carbon. Part of the clarified extract was treated with ammonia to give a final ammonia concentration of N/100 and both fractions were analysed for reducing sugar content.

Untreated sample - 16.6% reducing sugars
Ammonia treated sample - 16.4% " "

The treated and untreated samples give very similar results, as was found when pure glucose was analysed (1), and indicate that ammonia does not react with sugars themselves, but forms a complex when polyphenols are also present.

5. The effect of varying the ammonia concentration of the tobacco extract before clarification was next investigated.

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Ammonia Concentration	% reducing sugars
nil	17.1
N/160	16.8
N/100	16.1
N/80	16.0
N/40	15.5

As expected, increasing the ammonia concentration of the tobacco extract, decreases the amount of reducing sugars found on analysis.

This means that samples of ammonia treated tobacco submitted for sugar analysis will have to be carefully selected otherwise the residual ammonia in the tobacco will, on extraction, react with sugars and polyphenols in the tobacco and so cause a low result to be obtained for the reducing sugar assay.

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