

TENTATIVE METHOD FOR DETERMINATION OF CARBONYL  
COMPOUNDS IN CIGARETTE SMOKE

Summary of the method

The smoke from five cigarettes is led into a trapping system where the carbonyl compounds are converted into their 2:4 dinitrophenyl hydrazone derivatives. After refluxing to complete the conversion, the excess 2:4 dinitrophenyl hydrazine is removed. The solution containing the derivatives is streaked onto a silica thin layer and the streak eluted to separate the derivatives from any tars, etc., which may be present. Finally, the bands which contain carbonyl derivatives are removed from the plate, re-dissolved and the optical density compared with a standardized graph. The results are expressed as acetaldehyde.

Notes on the various stages comprising the method.

1. Plain cigarettes, weighing 1115 - 1155 mgm. were used in the development of the method. Five cigarettes were smoked in each run. A two second 35 ml. puff at 1 minute intervals was used.

2. After several experiments an efficient trapping system was devised. This consisted of:-

(a) A. T.R.C. pattern cold trap containing 50 ml. methanol at  $-65^{\circ}\text{C}$  followed by

(b) a 500 ml. round bottomed flask containing 100 ml. of a 1% methanolic solution of 2:4 dinitrophenyl hydrazine, to which 1 ml. of concentrated HCl had been added. This flask was continuously agitated mechanically during each run.

Thus the trapping system consisted of:

Cigarette	-	Methanol	-	"Shaking"	-	Smoking
		at $-65^{\circ}\text{C}$		solution		machine
				2:4 dnph		
				in methanol		

The effectiveness of this trapping system was checked in two ways.

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(i) A known weight of acetone was placed in the empty T.D.C. trap at  $-65^{\circ}\text{C}$ . Fifty puffs of air were drawn through this trap, the vapour being led into the "shaking" trap. On working up, the predicted optical density was found.

(ii) Known small volumes of acetone were injected into cigarettes below the butt-line during the smoking process. The butts were coated with a layer of silicone grease to prevent vapour losses through the paper. Providing that too rapid or too large injections were not given, the predicted optical densities were obtained after working up.

3. The contents of the two traps were combined and transferred to a flask and refluxed for two hours, to complete the condensation reaction. (Shorter periods than two hours gave rise to low recoveries).

4. After refluxing, the alcoholic solution was diluted with about 300 mls. of water, and extracted with 4 x 50 ml. of benzene. Most of the excess 2:4 dnph, and tars etc., precipitated at this stage and were removed by filtration.

5. 2:4 dinitrophenyl hydrazine present in the benzene layer was removed by 4 x 50 ml. extractions with 2N HCl. The partition coefficient of 2:4 dinitrophenyl hydrazine between benzene and 2N HCl was found gravimetrically and by optical density to be 1:9.

6. The final benzene solution was filtered, dried by washing with brine, and then diluted to 250 mls. and mixed thoroughly.

7. A 40 micro litre streak of the benzene solution was placed onto a layer of silica 250 microns thick on glass, and the streak was eluted with 10% cyclohexane in benzene. After about 30 - 40 minutes this gave a chromatogram consisting of three main yellowish-orange bands and a number of minor orange and brown bands.

Examination of the  $R_f$  values of the 2:4 dnph derivatives of 14 carbonyl compounds, most of which are known to be present in tobacco smoke<sup>2</sup>, showed that 13 of these derivatives had  $R_f$  values in benzene/cyclo-

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hexane which corresponded with one or other of the three main bands. It was therefore thought that removal and measurement of these three bands would give a fair indication of the total carbonyl content of the smoke.

The silica holding the three bands was removed from the plates and the colour removed from the silica by extraction with benzene. After centrifuging, the benzene was made up to 10 als.

Experimentally, the recovery of acetone 2:4 dnph from the silica was found to be 94%. However, the ability to detect 100% of added acetone (Note 2) suggests that recovery from the silica is 100%.

8. The optical density of this solution was measured at 355 m $\mu$  on a Unicam SP. 500. This is the  $\lambda_{\text{max}}$  for this solution and corresponds with the  $\lambda_{\text{max}}$  values for the 2:4 dnph's of acetaldehyde, propionaldehyde and butyraldehyde. Also acetone, methyl ethyl ketone, diethyl ketone and formaldehyde have  $\lambda_{\text{max}}$  in the region 350 - 365 m $\mu$ .

The molar extinction coefficients at 355 m $\mu$  for the 2:4 dnph's of  $\text{CH}_3\text{CHO}$ ,  $\text{CH}_3\text{COCH}_3$  and  $\text{CH}_3\text{COC}_2\text{H}_5$  (which comprise 70% of the carbonyls in smoke found by Hobbs<sup>1</sup>) were all found to be the same. Assuming this is true for all the carbonyls present, then a single measurement enables a total carbonyl figure to be obtained.

Comparison of the optical density obtained, with a calibration graph prepared from acetaldehyde 2:4 dnph gave an "acetaldehyde equivalent".

9. Using the method described reproducible "acetaldehyde equivalents" were found for smokes of 5 cigarettes.

10. On heavier loadings i.e. 10 cigarettes, discrepancies have been found despite the scaling up of the quantity of reagent used. Possible causes of these discrepancies are now being investigated in order to put the method on a sounder footing.

#### References

1. HOBBS, M.R. et al. Anal. Chem., (1956), 28, 910.
2. JOHNSTONE, R.A.W. and PLIMMER, J.R., Chem. Reviews, 1959, 59, 895.

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