LETTER TO THE EDITOR

NOTE ON SIMPLIFIED
AUTOMATED VOLATILE FATTY ACIDS ANALYSIS

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The improved automatic method of volatile fatty acid analysis reported by us (Harwood and Huys 1968) required the separate analysis of a sample and a blank determination. This procedure was liable to inaccuracies arising from changes in sensitivity between analyzing the sample and the blank, and because different aliquots were used.

These difficulties could be partly overcome by the simultaneous analysis of each sample and blank. Originally two separate manifolds were built up, the one for the analysis and the other for the blank. This did not prove completely satisfactory due to an unstable base-line. The method could be improved by the use of a combined manifold based on splitting the sample stream for the analysis and blank determinations after esterification so ensuring that portions of the same aliquot were used for sample and blank analyses. This note describes the construction of such a manifold and the results obtained with it.

FIG. 1.
EXPERIMENTAL

Reagents
As described earlier (Harwood and Huyser 1968). The hydroxylamine should be prepared fresh.

Apparatus
Standard Technicon equipment consisting of Sampler II, Pump II, two colorimeters and a 2-pen recorder with the Range Expander were used.

Manifold
The manifold is shown in Fig. 1

Method
The sample was withdrawn and after esterification split into two streams. The one was analyzed while the other was used for the blank analysis. The colours developed were measured by the colorimeters, the output being recorded via the Range Expander. After processing all the samples, the two channels were calibrated by analyzing sodium acetate standards. For this purpose the water on the blank manifold was replaced by hydroxylamine.

RESULTS
Figure 2 is a calibration graph, with the Range Expander set on X4 and 30-1:2 sampling.

A series of 30 samples analyzed manually and with this manifold (Fig. 1), gave a correlation coefficient of \( r = 0.98 \).

DISCUSSION
Hydroxylamine appeared to be the best reagent to omit for the blank analysis, since the sample and blank would then have received the same esterification, be at the same pH values, and have equal
concentrations of ferric ion present. Any changes in optical density in the blank stream should be due
to the sample.

Samples containing carbonate have given trouble, due to the release of CO$_2$ on adding the sulphuric
acid, which caused a complete break-up of the bubble pattern. A preliminary manual acidification has
solved this problem. Tests on standards have shown that the pH of the sample over the range 2-12
did not affect the peak height.

The coefficient of variation of the peak heights was about 2.5 per cent, although this was apparently
markedly affected by the condition of the pump tubing.

Glucose a common substrate in fundamental anaerobic digestion studies, was checked to see if it
interfered in the method. A sample of 10,000 mg glucose per litre gave a response of 200 mg acetic
acid per litre. This is only 2 per cent, agreeing with the value of 2.14 per cent noted by Wagner (1967).
Glucose interference is unimportant in digestors operating satisfactorily, when glucose levels are
likely to be only a few milligrams per litre.

A batch of 60 samples were analyzed using this manifold, and gas chromatographically for the
individual acids and these acids totalled. A correlation coefficient of $r = 0.953$ was obtained between
the two sets of total volatile fatty acids.

A further improvement in this manifold would be the use of a differential colorimeter (Technicon).
This would result in an output of one peak (instead of two) for each sample, and would halve the time
spent on data processing.

By using the system described here, we have been able to double our output of total volatile fatty
acids analyses per day.

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REFERENCES

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