TABLE 1. Influence of 30 hr of intermittent hypoxia on the phosphorus fractions of the bone marrow and spleen.

Group	No. of animals	Phosphorus content mg/g tissue		Specific activity*	
		ASP†	DNAP	ASP	DNAP
		Bone marrow			
Control	15	0.966	1.320	0.481	0.199
σ‡		± 0.088	± 0.295	± 0.064	± 0.034
30-hr hypoxia	6	1.059	1.458	0.608	0.383
σ		± 0.070	± 0.116	± 0.078	± 0.086
P§		< 0.98	< 0.80	> 0.99	> 0.99
		Spleen			
Control	15	0.969	1.294	0.468	0.034
σ		± 0.107	± 0.244	± 0.011	± 0.002
30-hr hypoxia	6	0.949	1.454	0.676	0.287
σ		± 0.020	± 0.112	± 0.080	± 0.036
P		< 0.50	< 0.90	> 0.99	> 0.99

* Percentage of dose of P^{32} injected/mg. P in fraction.
† Acid-soluble phosphorus.
‡ Standard deviation of the mean.
§ Probability.

Animals were subjected to hypoxia in a low-pressure chamber at an oxygen concentration of 7.75 percent. The methods of obtaining marrow and spleen samples, the tissue fractionation procedures, and the counting techniques have been described (*3, 4*).

The data on 15 control animals and 6 animals subjected to intermittent hypoxia for 10 hr a day for 3 consecutive days are presented in Table 1. The experimental animals were injected with radiophosphorus 15 hr after return to ground level. There was no weight loss during the experimental period.

Striking changes were found in the specific activity of the DNAP in the bone marrow and spleen. Hypoxia produced a stimulation of mitosis in both organs. The specific activity in the spleen reached 8 times that of the control, while there was a doubling of the control value in the bone marrow.

Although there was not a marked change in the DNAP concentration or cellularity in either organ, the increase in total erythropoietic mass in the spleen was notable. The spleen weights of animals exposed to 30 hr of intermittent hypoxia increased 175 percent. Taken together with the 500 percent increase in the cells of the erythropoietic series observed in the differential counts from the spleen imprints, an almost tenfold increase in the total erythropoietic tissue resulted from the increased mitotic rate following the hypoxic stimulus.

Unlike the spleen, which may increase in size to accommodate a larger number of cells, the bone marrow is restricted to the marrow cavities, which in the rat are almost entirely filled with red marrow. This would be expected to prevent large increases in the total number of cells. The small increase found was due almost entirely to hyperplasia of the red cell series. The importance of the spleen in the erythropoietic response to hypoxia in the rat is clearly shown by these experiments.

References

1. J. N. Davidson. *The Biochemistry of the Nucleic Acids.* New York: John Wiley, 1951.
2. G. Hevesy. *Radioactive Indicators.* New York: Interscience, 1948.
3. W. A. Rambach, D. R. Moomaw, H. L. Alt, and J. A. D. Cooper. *Proc. Soc. Exptl. Biol. Med.* **79**, 59 (1952).
4. W. A. Rambach, H. L. Alt, and J. A. D. Cooper. USAF School of Aviation Medicine, Project No. 21-3501-0001, Report No. 2 (1953).

Received November 20, 1953.

Detection of Microbially Produced Gaseous Hydrocarbons Other than Methane

John B. Davis and Rodney M. Squires[1]

Magnolia Petroleum Company, Field Research Laboratories, Dallas, Texas

Attempts to detect microbially produced gaseous hydrocarbons other than methane have usually failed. Notable exceptions are reports of ethylene being formed by fungi (*1, 2*). Where other gaseous hydrocarbons such as ethane have been reported there has usually been a question of the actual source, due to the use of municipal sewage in experiments, for example.

Buswell (*3*) states that efforts to detect gaseous hydrocarbons other than methane in microbial fermentations in his laboratory have failed. Furthermore, in data furnished to him by A. V. Grosse (*3*) and reported at the 113th Meeting of the American Chemical Society but not included in the abstract (*4*), a mass spectrometer analysis of purified sewage gas indicating 99.2 percent methane failed to reveal ethane or propylene. The limit of sensitivity in the measurement was 20 ppm for those gases.

By increasing sensitivity still further to the order of 0.05 ppm, we have detected numerous gaseous hydrocarbons other than methane produced in microbial fermentations, and also by the fungus, *Penicillium digitatum* A.T.C.C. No. 10030, growing upon potato-glucose agar. Details of the modification of a Westinghouse Type LV mass spectrometer to obtain high sensitivity will be reported elsewhere.

For hydrocarbon measurements in the parts per million range, the C_2 and heavier hydrocarbons were condensed in a trap at liquid nitrogen temperature while the major portions of noncondensable atmospheric gases and methane were pumped off and discarded. Carbon dioxide was removed by KOH absorption. The trapped C_2 and C_3 hydrocarbons were admitted to the mass spectrometer through a trap at $-155°$ C, which retains most of the C_4 and all the heavier hydrocarbons. The use of 500-ml samples of microbially produced gas permitted measurements of individual C_2 and C_3 hydrocarbons to within ± 0.05 ppm of total sample.

Cow dung was selected as an inoculum free from

[1] Acknowledgment is due J. P. Stanley for his assistance in the performance of experiments.

TABLE 1. Analyses of gas collected over the microbial fermentation of paper.

Component measured*	Concentration†	
	Dec. 5, 1951	Feb. 7, 1952
Nitrogen	21.7 mole %	3.48 mole %
Oxygen	2.0 " "	1.38 " "
Argon	0.13 " "	0.07 " "
Carbon dioxide	33.7 " "	46.9 " "
Methane	42.6 " "	49.0 " "
Ethane	3.2 ppm	7.0 ppm
Ethylene	3.9 "	4.7 "
Propane	0.14 "	0.06 "
Propylene	0.13 "	0.21 "

* Nitrogen gas was used to flush the system initially. Although the presence of oxygen and argon suggests atmospheric contamination of the samples in handling, air has been found generally to contain only a few parts per billion of gaseous hydrocarbons other than methane.
† Hydrogen-free basis.

petroliferous contamination. A portion of the inoculum was mixed with mineral salts medium and paper (Nu-wipe tissue) in a Waring Blendor; this mixture was then added to a 20-liter bottle filled almost to capacity with freshly boiled and cooled mineral salts medium. Tank nitrogen was passed through the liquid for ½ hr to flush oxygen from the system. Analysis of the gas formed as a result of microbial fermentation for several weeks is given in Table 1.

Aliquots from the digested sludge in the 20-l bottle were added to liter quantities of 1 percent ethanol, sodium acetate, sodium butyrate, and glucose, respectively, in mineral salts medium. Active fermentation ensued in all cases at a much greater rate than the digested sludge alone. The ethanol fermentation yielded the most methane, 90 percent. Gas issuing from each of the fermentation systems was collected and analyzed in five successive 500-ml increments.

Ethane was found in all gas samples. The largest quantities were produced by the ethanol fermentation where concentrations increased from 1.2 ppm in the first gas increment to 6.8 ppm in the fourth increment. Ethane concentrations in gas from the glucose fermentation also increased from 0.1 ppm in the first gas increment to 0.7 ppm in the fifth increment. All gas samples from the acetate and butyrate fermentation systems contained about 0.2 ppm ethane. Ethylene was found in the gas from the ethanol and glucose fermentations in concentrations ranging from 0.1 to 2.6 ppm. It was also present in early gas samples from the acetate and butyrate fermentation systems but disappeared in the gas samples collected later. Acetylene was present in all gas samples from both the butyrate and acetate fermentation systems in a concentration of approximately 0.2 ppm. It was also detected in gas from the glucose fermentation system at an intermediate stage of the fermentation, but no acetylene was detected in gas from the ethanol fermentation system.

There were no C_3 hydrocarbons found in gas from the acetate or ethanol fermentation systems. Propylene was present, 0.9 and 0.4 ppm, respectively, in the first gas increments from the butyrate and glucose fermentation systems, but its concentration decreased to 0.0 ppm in the fourth gas increments. The fifth gas increment from the glucose system contained 0.1 ppm of propane. Propane could not be positively identified in any of the other samples.

Using a different source of microbial inoculum, namely 100 ml of municipal sewage sludge, fermentation of paper and 1 percent ethanol in a mineral salts medium was carried out. The system used was a stainless steel 8-l tank fitted with a pressure gage. The tank was thoroughly evacuated after the addition of 4 l of the ethanol medium containing 5 g of homogenized Nu-wipe tissue. Pressure began to increase in the fermentation system after about 2 wk and was allowed to reach 45 psi above atmospheric pressure. The gas formed consisted of approximately 20 percent carbon dioxide and 80 percent methane. Ethane and acetylene were each found in a concentration of 0.3 ppm and ethylene was found in a concentration of 0.1 ppm. Propane and propylene were absent.

Penicillium digitatum A.T.C.C. No. 10030 was grown upon potato glucose agar in desiccators, and the atmosphere, which initially consisted of 20 percent tank oxygen and 80 percent tank nitrogen, was analyzed. The principal gas constituents were found by mass spectrometer analyses to be carbon dioxide and nitrogen; however, gaseous hydrocarbons were detected as in the bacterial fermentation systems. Thus, acetylene concentrations ranged from 0.0 to 0.6 ppm, ethylene, 0.2 to 1.4 ppm, propylene, 0.0 to 1.2 ppm, ethane, 0.1 to 0.6 ppm, and propane, 0.0 to 0.1 ppm, respectively. None of these components could be identified in a control system where *Penicillium digitatum* inoculum was omitted.

References

1. NICKERSON, W. J. *Arch. Biochem.* **17**, 225 (1948).
2. WILLIAMSON, C. E. *Phytopathology* **40**, 205 (1950).
3. BUSWELL, A. M., and MUELLER, H. R. *Ind. Eng. Chem.* **44**, 550 (1952).
4. GROSSE, A. V., and LIBBY, W. F. Abstracts, 113th Meeting Am. Chem. Soc. pp. 13R-14R (1948).

Received January 11, 1954.

The Quantification of Hostility in Dreams with Reference to Essential Hypertension

Leon Saul, Edith Sheppard, Dorothy Selby, William Lhamon, David Sachs, and Regina Master

Department of Psychiatry, School of Medicine, University of Pennsylvania

In investigating methods for the measurement of emotional forces, a preliminary hostility scale was derived from a content analysis of 500 manifest dreams obtained from 200 subjects presenting wide variation in personality structure (*1*). This scale was found to differentiate hypertensive from nonhypertensive subjects.

Although most of the studies using dream material