Increased pentane and carbon disulfide in the breath of patients with schizophrenia

M Phillips, M Sabas, J Greenberg

Abstract

***Aims*—To determine the concentrations of pentane (a marker of lipid peroxidation) and other volatile organic compounds in the breath of patients with schizophrenia.**

***Methods*—Volatile organic compounds were assayed by gas chromatography/mass spectroscopy (GC/MS) in 88 subjects—25 with acute schizophrenic psychosis, 26 with psychiatric disorders other than schizophrenia, and 37 normal volunteers.**

***Results*—The mean alveolar gradients of pentane and carbon disulfide (CS_2) were significantly higher in the patients with schizophrenia than in the control groups.**

***Conclusions*—Schizophrenia may be accompanied by accelerated lipid peroxidation in cell membranes, as well as increased manufacture of CS_2, a known neurotoxin.**

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Department of Medicine, St Vincent's Medical Center of Richmond, Staten Island, New York 10310-1699 USA
M Phillips
J Greenberg

Department of Medicine, New York Medical College, New York
M Phillips

Department of Psychiatry, St Vincent's Medical Center of Richmond, Staten Island
M Sabas

Correspondence to:
Dr Michael Phillips, Department of Medicine, St Vincent's Medical Center of Richmond, 355 Bard Avenue, Staten Island, NY 10310-1699, USA.

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Schizophrenia is a common and devastating psychotic illness: it affects nearly 1% of the population of all cultures, and often culminates in severe disability and premature death.[1 2] The aetiology of schizophrenia is still unknown, and researchers have used many different investigative techniques to determine a biochemical basis for the disordered cognition and behaviour that characterise the disease. Kovaleva *et al* assayed the breath of acutely psychotic schizophrenic patients and found raised concentrations of pentane which varied with the clinical severity of the condition, then rapidly returned to normal during a course of treatment.[3 4]

Alkanes in the breath (principally ethane and pentane) result from cellular injuries which cause an intracellular accumulation of oxygen free radicals and accelerated peroxidation of polyunsaturated fatty acids.[5 6] The peroxidation of lipids may result in membrane injury, with dysfunction and death of the affected cells. Raised alkane concentrations in the breath have been reported in patients with a number of conditions, including acute myocardial infarction,[7] rheumatoid arthritis,[8] and nutritional deficiencies of vitamin E.[9] The toxic effects of oxygen free radicals may represent the final common pathway of several different pathogenic agents, including chemical toxicity, inflammation, and ischaemia.

Methods

The method for collection of volatile organic compounds has been described before.[10] A mobile apparatus was used which sampled 10 litres of alveolar breath over 5 minutes while the donor was breathing in chemically purified air. The breath was drawn through a stainless steel trap in which the volatile organic compounds were captured by adsorption to activated carbon and molecular sieve.

The volatile organic compounds captured from alveolar breath were thermally eluted from the trap in a microprocessor controlled automatic desorber, concentrated by two stage cryofocusing at −150°C, then assayed by gas chromatography and mass spectroscopy using an ion-trap detector. Each volatile organic compound was identified by its mass spectrum, quantified by area under curve, and its alveolar gradient was determined (concentration in alveolar air minus concentration in inspired air). Standard curves for volatile organic compounds were obtained using adsorptive traps loaded with vapour standards prepared by the method of Morris *et al*.[11]

Table 1 Diagnostic categories of non-schizophrenic psychiatric disorders

DSM-III-R Category	*Diagnosis*	*No of subjects*
296·3	Major depression, recurrent unspecified	5
296·33	Major depression, recurrent, severe, without psychotic features	4
296·44	Bipolar disorder, manic, with psychotic features	3
296·53	Bipolar disorder, depressed, severe, without psychotic features	1
296·54	Bipolar disorder, depressed, with psychotic features	1
296·62	Bipolar disorder, mixed, moderate	1
300·40	Dysthymic	1
305·00	Alcohol abuse	3
309·00	Adjustment disorder with depressed mood	4
311·00	Depressive disorder not elsewhere classified	3
	Total	26

Table 2 Characteristics of subjects

	Normal controls	*Psychiatric controls*	*Patients with schizophrenia*
Number	37	26	25
Mean age (SD)	37·4 (9·0)	41·0 (10·0)	35·8 (12·3)
Sex (% males)	50	53·1	48
Race:			
White	17	17	18
Black	2	4	3
Hispanic	11	5	3
Asian	7	0	1

No significant differences among the three groups were observed.

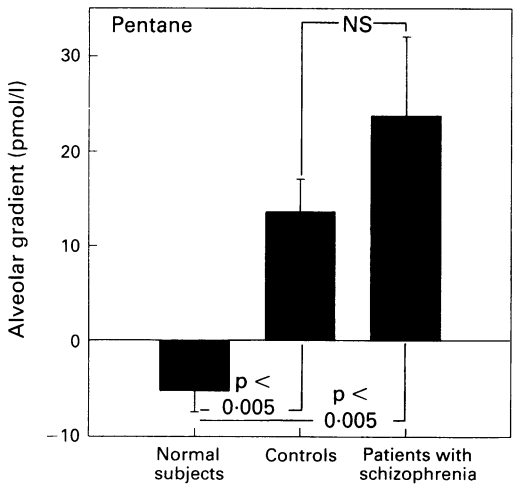

Figure 1 Alveolar gradients of pentane. Solid bar = group mean, error bar = standard error of mean.

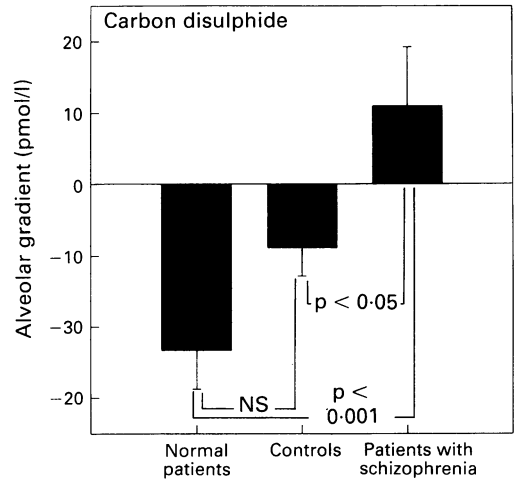

Figure 2 Alveolar gradients of carbon disulfide. Solid bar = group mean, error bar = standard error of mean.

Eighty eight volunteers were studied at St Vincent's Medical Center of Richmond and Bayley Seton Hospital in Staten Island, New York. Breath collections were performed between 07 00 h–1100 h. Criteria for acceptance to the study included: (1) willingness to cooperate with the breath collecting procedure; (2) age between 18 and 70 years; and (3) no known acute or chronic medical illness. All subjects gave their signed informed consent to participate in this research, which was approved by the institutional review boards of both institutions.

A group of 37 normal volunteers was drawn from the medical and nursing staff of both institutions. None gave any history of psychiatric illness.

Twenty six subjects were recruited from the psychiatric inpatient units of both hospitals. All had been diagnosed as having a psychiatric illness other than schizophrenia by DSM-III-R criteria (table 1).[12]

Breath samples were collected from 25 subjects who had fulfilled DSM-III-R criteria for the diagnosis of schizophrenia during previous hospital admissions. All had experienced a recent acute psychotic exacerbation that was sufficiently severe to require admission to a psychiatric unit, where they shared the same environment and diet as the psychiatric controls. All had been treated with neuroleptic drugs for at least 24 hours before the breath collection.

Using multiple regression, the alveolar gradient of each volatile organic compound was treated as a dependent variable, and the diagnostic group (schizophrenic, normal, or psychiatric controls) as the independent variable. Where significant differences between groups were observed, the effects of cigarette smoking, race, neuroleptic drugs, sex and age were evaluated as potential contributory factors using multiple correlations. A subgroup of treated patients with schizophrenia (n = 8) was studied a second time before discharge; the two assays were compared using Student's paired *t* test.

Table 3 Effects of smoking and neuroleptic drugs on alveolar gradients of pentane and CS_2

	Normal controls	*Psychiatric controls*	*Acutely psychotic schizophrenic patients*
Smokers:			
n =	11	17	12
pentane	−10·90 (7·16)	11·55 (4·09)	37·27 (18·45)
CS_2	−22·61 (9·49)	−9·04 (3·90)	11·74 (7·17)
Non-smokers:			
n =	26	9	13
pentane	−2·86 (3·58)	17·48 (10·63)	11·33 (3·86)
CS_2	−23·57 (6·46)	−8·61 (8·18)	10·34 (14·12)
Neuroleptic drugs			
n =	0	12	25
pentane		18·80 (7·37)	23·78 (8·31)
CS_2		−12·44 (5·99)	11·01 (8·10)
No neuroleptic drugs			
n =	37	14	0
pentane	− 5·25 (2·19)	9·15 (5·40)	
CS_2	−23·28 (5·34)	−5·85 (4·72)	

The mean concentration of pentane and CS_2 in each subgroup is shown, expressed in pmol/l, with the standard error of the mean in parentheses. There were no significant differences among the three groups between the smokers and the non-smokers, or between the psychiatric controls treated or not treated with neuroleptic drugs.

Results

Breath samples were obtained from all subjects without any adverse effects. Characteristics of the three groups are shown in table 2; no significant differences were observed in the distribution of age or sex.

The alveolar gradients of five volatile organic compounds in the breath were significantly higher in the patients with schizophrenia than in the controls: pentane ($p < 0·005$) (fig 1); carbon disulfide (CS_2) ($p < 0·001$) (fig 2); benzene, 2-methylbutane, and tetrachloroethene ($p < 0·05$). Only CS_2 was significantly increased when the patients with schizophrenia were compared with the psychiatric controls ($p < 0·05$). No significant difference in the alveolar gradient of either pentane or CS_2 was associated with tobacco smoking, treatment with neuroleptic drugs (table 3), or racial group. No significant changes in any volatile organic compounds were observed in the subjects with schizophrenia who were studied a second time.

Discussion

Microanalysis of the breath opens a non-invasive window on to the chemical composition of the blood. Volatile organic compounds diffuse passively across the pulmonary

alveolar membrane; like water running down a hill, they flow rapidly from the compartment with the higher vapour pressure to the compartment where it is lower.[13] A positive alveolar gradient is evidence that the vapour pressure of a volatile organic compound is higher in the venous blood than in the inspired air, while a negative alveolar gradient shows the opposite.

The mean alveolar gradient of pentane was highest in the acutely psychotic schizophrenic patients, a finding consistent with the study by Kovaleva *et al.*[3 4] The lack of a significant difference between the schizophrenic patients and the psychiatric controls may have been a consequence of experimental design. We studied schizophrenic patients who had been treated with neuroleptic drugs for 24 hours or longer, while Kovaleva *et al* studied untreated patients who may have been more acutely psychotic with higher concentrations of pentane in their breath.

In addition, the mean alveolar gradient of CS_2 was positive in the patients with schizophrenia and negative in the two control groups. These observations indicate that the vapour pressure of CS_2 in venous blood was significantly increased in the schizophrenic patients. This was an unexpected finding which, to our knowledge, has not been reported before. It is unlikely that the differences in breath CS_2 could have been due to environment or diet, because the patients with schizophrenia and the psychiatric controls all resided in the same ward areas and ate similar meals. Tobacco smoke and some neuroleptic drugs (thioridazine and mesoridazine) contain sulfur, but neither significantly affected the alveolar gradient of CS_2. Nor was breath CS_2 significantly affected by age, sex, or racial group.

It has been known for more than 100 years that industrial workers exposed to high levels of CS_2 can develop acute toxicity with psychiatric manifestations, including acute psychotic episodes with mania, rapid mood changes, extreme irritability, uncontrollable anger, and suicidal tendencies.[14–16] Magos has suggested that these toxic effects may be due to the reaction of CS_2 with amines or thiols[14]; the resulting chelation of metals (mainly zinc and copper) may inhibit the activity of dopamine β hydroxylase and cause disturbances of cathecholamine metabolism.

The negative alveolar gradient of CS_2 observed in the two control groups was consistent with normal catabolism of CS_2, which can occur via several different pathways, including conjugation with glutathione, formation of dithiocarbamates, and by monooxygenase catalysed generation of reactive sulfur.[16] Conversely, the positive alveolar gradient observed in the patients with schizophrenia indicated that more CS_2 had been expired than inspired through the lungs. In the absence of any apparent exposure to CS_2 from other sources, this finding was consistent with endogenous manufacture of CS_2.

The origin of the endogenous CS_2 is still unknown. One possible source is the large bowel, where volatile organic compounds containing sulfur may be produced by the metabolism of micro-organisms. The breath of patients with advanced hepatic cirrhosis and fetor hepaticus contains increased concentrations of such volatile organic compounds, principally methyl mercaptan and dimethyl sulfide.[17 18] Porto-systemic shunts in patients with cirrhosis have also been associated with increased breath concentrations of short chain fatty acids originating in the large bowel, principally acetic and propionic acids.[19] The manufacture of pentane and CS_2 in the large bowel merits further study as a possible explanation for our observations.

The air inspired from the breath collecting apparatus was partially but not completely purified. Even with a large filtration bed of activated carbon, it was not possible to extract completely all traces of airborne volatile organic compounds present in room air in picomolar concentrations. In practice, this did not present a serious problem, because the level of volatile organic compound background in the inspired air was reduced to a fairly consistent low concentration which neither exhibited any significant fluctuation during the course of the study, nor between the three groups of human subjects. The presence of these low yet consistent background concentrations of pentane and CS_2 was ultimately beneficial, because it showed the presence of negative alveolar gradients which would not have been observed had the air been completely purified.

Neuroleptic drugs introduced an extrinsic chemical source into this study which could have affected the findings. It was not ethically possible to withhold these drugs from the patients with schizophrenia, who were admitted as acute psychiatric emergencies. It probably also would not have been possible to secure their cooperation for breath testing without treatment. About half of the psychiatric control patients, however, were also treated with neuroleptic drugs, and there were no significant differences in either pentane or CS_2 concentrations between the treated and the untreated patients (table 3). Consequently, drug treatment did not seem to influence the findings.

In conclusion, patients with schizophrenia exhibited increased production of CS_2, a known neurotoxin, and pentane, a marker of lipid peroxidation. Further studies are required to determine if these observations were a consequence of the schizophrenic illness or some factor other than the disease itself.

We thank Sheldon Blackman, PhD, for advice on the statistical analysis of data.

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brief descriptions of geographical distribution, morphology, and life cycle.

Section 2 contains over 250 colour photographs covering a wide range of parasite morphology, pathology (including stained sections), and clinical pictures with captions on the facing page. Although the overall quality of the photographs is excellent, I was disappointed to see a lack of size markers on all but a handful. In the clinical laboratory size is of vital importance for identifying ova and cysts.

The third section contains black and white electron micrographs, radiographs, and other illustrations, separated from the colour section for reasons of economy. This does not detract from the atlas in any way, and indeed some of the scanning electron microscopic images are quite breathtaking. I would, however, like to have seen some indication of size on the photographs.

This atlas has a spacious and orderly feel to it, and I am impressed by the overall quality. Clinical microbiologists, particularly those in training, will find it useful.

AJ HAY

Atlas of Ovarian Tumors. L Deligdisch, A Altchek, CJ Cohen. (Pp 182; £94.) Igaku-Shoin. 1994. ISBN 0-89640-240-1.

This sumptuously produced atlas is subdivided into two main sections with three chapters devoted to "clinical aspects" and seven chapters allocated to "pathology". This strategy may be convenient in a multiauthor text, but it has resulted in a clinical section which is pathologically naive and a pathological section impoverished by the paucity of clinicopathological correlation.

The wide-ranging introductory chapter, which covers epidemiology, genetics, molecular biology, early diagnosis, and screening for ovarian cancer, provides a useful overview of the subject, although the emphasis placed on ultrasonography is excessive. The two ensuing chapters, both rather lengthy and repetitive, are devoted to management of ovarian carcinoma and nonepithelial tumours, respectively.

The pathology section comprises four chapters devoted to primary epithelial neoplasms including a whole chapter on the interesting but controversial subject of ovarian intraepithelial neoplasia. Other chapters deal with sex cord-stromal tumours, germ cell tumours, and metastatic tumours. This section is well illustrated with adequate photomicrographs and gross photographs of excellent quality. However, many entities are skimpily and uncritically described with no attempt to evaluate the taxonomic overenthusiasm displayed by recent authors in this field. On the other hand, many rare but well established entities are not included. An even more serious drawback for a book aimed at the practising histopathologist is the lack of consideration given to possible differential diagnoses.

In conclusion, this new atlas is unlikely to fulfil the need for a comprehensive, authoritative, and up to date reference text on ovarian neoplasms. It cannot be recommended as a bench book for the reporting room.

SM ISMAIL

Notices

Postgraduate course: Current concepts in surgical pathology

November 14–18 1994

Massachusetts General Hospital, Harvard Medical School

This course is designed for pathologists at resident and practitioner levels. It will provide an in-depth review of diagnostic surgical pathology with emphasis on morphological features, newly recognised entities, and new techniques, presented by the faculty of the Department of Pathology, Massachusetts General Hospital. Instruction will be primarily by lecture, but will also include discussion periods. Each participant will receive a comprehensive course syllabus.

The course has category 1 accreditation for about 35 hours CME credit by the American Medical Association. The fee for the course is $785·00 (residents and fellows $575·00).

For further information contact: Department of Continuing Education, Harvard Medical School, 25 Shattuck Street, Boston, MA 02115 USA (Tel: 0101 (617) 432 1525).

Update on Cerebrovascular Pathology

Thursday 8 December 1994 (one-day) to be held at The Royal College of Pathologists, 2 Carlton House Terrace, London SW1Y 5AF.

The meeting is open to members and non-members of the College. Further details and application forms can be obtained from the Scientific Meetings Officer, RCPath, 2 Carlton House Terrace, London SW1Y 5AF (Tel: 071 930 5862 ext: 24/26).

Cytopathology for histopathologists Northwick Park Hospital

30 January-3 February 1995

This is an intensive course in cytopathology suitable for candidates preparing for the MRCPath examination in histopathology, and for established histopathologists requiring revision. It is given by the Department of Cellular Pathology, Northwick Park Hospital (Dr Elizabeth A Hudson) and the Department of Cytopathology, St Mary's Hospital Medical School, University of London (Professor Dulcie Coleman).

The programme will consist of lectures, microscopy sessions, and discussions. Topics will include cytopathology of the cervix, urine, the respiratory tract, serous effusions and fine needle aspiration cytology of breast and other sites. The course is limited to 30 participants. The course fee is £300 excluding accommodation.

Applications and enquiries should be made to: Dr Elizabeth Hudson, Department of Cellular Pathology, Northwick Park Hospital, Harrow, Middlesex HA1 3UJ (Tel: 081–869 3312).

Corrections

J Clin Pathol 1994;47:205–8; Tillyer *et al.* The title of the correspondence should read "zinc protoporphyrin assays in patients with α and β thalassaemia trait." The title at present implies that zinc assays were performed which was not the case.

In paragraph 2, the second sentence should read ". . . not only were the drugs causing substantial interference extremely unlikely in the outpatient and general practice population we studied, . . .". Paragraph 3 second sentence should read ". . .Paul and Brumfitt's is 15 μmol/mol haem lower."

DR ML TILLYER

Estimation of haemoglobin concentrations using spectrophotometric tests. *J Clin Pathol* 1994;47:681.

The name of the author was given incorrectly as J Larner rather than AJ Larner.

ANDREW J LARNER

J Clin Pathol 1993;46:1116–9. (Darjee R, Gibb AP. Serological Investigation into the association between *Streptococcus bovis* and colonic cancer.) The methods section refers to "NCTC10449 (*Enterococcus fecalis*), but this should read ATCC19433 (*Enterococcus fecalis*). NCTC10449 is in fact the reference number of the type strain of *S mutans*.

AP GIBB

Increased pentane and carbon disulfide in the breath of patients with schizophrenia *J Clin Pathol* 1993;46:861–4. The concentrations of pentane and carbon disulfide were reported incorrectly. All values of pentane should be multiplied $\times 50$; all values of carbon disulfide $\times 0{\cdot}05$. The statistical analyses and conclusions of the paper are not affected by these corrections.

MICHAEL PHILLIPS

Chu CM, Liaw YF. Coexpression of intercellular adhesion molecules and class I major histocompatibility compression antigens on hepatocyte membrane in chronic viral hepatitis. *J Clin Pathol* 1993;46: 1004–8. The correct version of fig 2D is reproduced below.

CHIA-MING CHU