Breath methane and large bowel cancer risk in contrasting African populations

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SUMMARY Breath methane has been measured in 1016 people from four populations resident in Southern Africa which experience widely different risks of bowel cancer and other colonic diseases. Highly significant differences in the proportion of subjects with detectable methane in breath were found; % producers – rural black 84, urban black 72, white 52, Indian 41 (χ^2 121 p<0.001 3 df). There was a slight preponderance of female producers over male (female producers 63%, males 57%) and an age trend with fewer producers in the older age groups in the urban blacks and Indians, these comparisons being significant when tested by stepwise logistic regression analysis. Bowel cancer risk, determined from a variety of sources, was lowest in rural blacks, greatest in whites, with intermediate rates for urban blacks and Indians. Methane production in the human colon shows significant interethnic differences but which bear no relation to bowel cancer risk in these populations.

Although it has been known for more than 150 years that methane is produced in the human large intestine, nevertheless the physiology and importance of methanogenesis in man remains something of an enigma. Methane is one of the principal end products of fermentation in anaerobic systems such as the rumen,¹ whilst in man active fermentation occurs in the large intestine² and methanogenic bacteria such as *Methanobrevibacter smithii* have been found in faeces.^{3–7} Methane instilled into the human colon rapidly appears in breath⁸ as do other fermentation gases such as hydrogen.⁹ In most published reports of population studies, however, only 35–61% of healthy adult subjects have detectable methane in their breath^{8 10–17} although two reports give higher values.^{18–19} In detailed studies by Levitt and Ingelfinger of individuals, using intestinal perfusion with air or nitrogen, about half of the subjects tested showed no sign whatsoever of methane evolution in the colon.⁹

A group of people with unexpectedly high breath methane levels are those with large bowel cancer. In two separate studies 80 and 91% of these patients had methane detectable in their breath and the proportion of methane producers was also increased in patients with premalignant conditions of the bowel such as polyps.^{13 14} Physiologically methane production is associated with slow gut transit time, small faecal weight, and high faecal pH²⁰ although these differences in faecal weight have not been found consistently.^{12 15}

Many clues to the aetiology of bowel disease have come from population studies, especially those in Africa where major differences in the risks of bowel cancer, appendicitis, and diverticular disease exist^{21–24} often in people living in closely related geographical areas. We have therefore measured breath methane in four population groups in Southern Africa who show marked differences in bowel disease risk in the belief that the group with the lowest risk of bowel disease and fastest gut transit time would have least methane in their breath and *vice versa*. The results show that there are major differences in methane producer status amongst the populations, but these do not relate to bowel cancer risk.

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Methods

SUBJECTS

One thousand and sixteen outwardly healthy subjects from four population groups were studied (Table 1). One hundred rural black adults were volunteers from the villages of Hekpoort and Pankop situated in western and northern Transvaal 80 and 120 km respectively from Johannesburg. One hundred pupils attending the village schools also participated. One hundred and forty eight urban blacks were recruited from the townships of Alexandra and Soweto and 120 pupils from schools in these towns; 134 whites were staff and students from Baragwanath Hospital and the South African Institute of Medical Research in Johannesburg, and 86 pupils were recruited from local Catholic Convent schools; the Indians, 128 adults and 200 schoolchildren were resident in Johannesburg and Lenasia. All the subjects were fit and none had been taking antibiotics or laxatives for three months before the study. Smoking was not allowed for one hour before breath samples were taken.

METHANE

Duplicate end expiratory breath samples were collected *via* a Wiggins end expiratory breath sampler²⁵ into 20 ml plastic syringes. Methane concentration was determined by gas chromatography using a Pye Unicam PU-4500 equipped with a hydrogen ionisation detector. The carrier gas was nitrogen. A sample of room air was taken at the time of each breath sample and the value (usually in the range 1.7–2.2 parts per million (ppm)) was subtracted from the average level of the duplicate breath samples. Methane producers were defined as those producing more than 1 ppm of methane above the level in the ambient air.¹⁰

This study was approved by the Ethical Committee of the Medical School the University of the Witwatersrand, Johannesburg.

BOWEL DISEASE STATISTICS

Because of the difficulties in obtaining morbidity data for some bowel diseases, the analysis has been limited to large bowel cancer. Data have been compiled from Cancer Incidence in Five Continents, Vol II and III^{26, 27} and various published and unpublished sources.^{21, 22, 28–36}

STATISTICAL ANALYSIS

Statistical analysis was carried out using SPP (a statistical package for personal computers – by P Royston, Supersoft Ltd, Harrow, UK). Because of the distribution of age and breath methane data all values were transformed to $\log_e$. Intergroup differ-

ences were assessed by analysis of variance and frequencies of methane producer *versus* non-producer status by χ^2 . The relation of age, location and sex to methane producer status was tested by stepwise logistic regression analysis using GENSTAT. Results are given as median and range unless otherwise stated.

Results

The total population of 1016 subjects was roughly equally divided between men (511) and women (505) and between schoolchildren (499) and adults (517) (Table 1). Median age of adults was 30 (range 14–80) years and adolescents 16 (14–21). There was a greater proportion of the population over the age of 45 amongst the whites but the oldest subjects (70+) were in fact all rural blacks. Analysis of variance for age showed a significant difference amongst the adult population ($F\ 6.48$, $p\ 4.9 \times 10^{-4}$) mainly because the white males who were slightly older (median 32 years). Modifying this group by excluding some of the older subjects made no difference to the conclusions, however, and interpretation of any of the data so they have been left in.

For the whole population 60% had detectable methane in breath (greater than 1 ppm above background), but as Table 2 shows there were highly significant differences amongst the four ethnic groups. Seventy two to 84% of black populations were producers, while only 52% of the whites and 41% of the Indians were.

A more detailed examination (using stepwise logistic regression) of methane producer status in relation to age, location, and sex (Fig. 1) showed that for all subjects the relative risk of being a producer was in the order rural black, urban black, white, Indian and was greater in the younger age groups, this age trend being significant in the urban blacks and Indians. More women than men were producers; 57% men and 63% women ($\chi^2\ 3.8$, $p=0.04\ 1\ df$) but breath methane concentrations were similar; 11.1 (1.1–55.1) ppm men, 11.95 (1.1–56.1) ppm women.

Table 1 Age, sex, and number of subjects

	Rural black	Urban black	White	Indian
<i>n</i>	Total (M/F)	Total (M/F)	Total (M/F)	Total (M/F)
School pupils	100 (53/47)	120 (58/62)	86 (44/42)	200 (95/105)
Adults	100 (50/50)	148 (75/73)	134 (67/67)	128 (65/59)
Age*				
School pupils	16 (14–21)	17 (14–21)	16 (14–18)	16 (14–18)
Adults	28 (16–80)	28 (18–57)	31.5 (16–68)	31 (14–58)

*Median (range).

Table 2 *Breath methane: all subjects*

Population	% Producers (n)	% Non-producers (n)	Breath methane (Median+range)
Rural Black	84 (168)	16 (32)	15.5 (1.1–56.1)
Urban black	72 (193)	28 (75)	10.6 (1.1–48.2)
White	52 (114)	48 (106)	10.0 (1.1–44.4)
Indian	41 (133)	59 (195)	8.5 (1.1–52.3)
All cases	60 (608)	40 (408)	11.4 (1.1–56.1)

χ^2 121.8 $p < 0.001$ 3 df; ANOVA F 12.2 $p < 0.001$.

The concentration of methane in breath followed the same order as methane producer status amongst the various populations (Table 2). Median breath methane for all producer subjects was 11.4 (1.1–56.1) ppm. There was no general relationship between age and breath methane concentration (r 0.06, p 0.17, $n=608$) despite the significant association with methane producer status and age (Fig. 1). Breath methane levels were highest in rural black school children; 17.7 (1.2–51.0) ppm, 95% of whom were producers, and lowest in Indian adults; 5.8 (1.4–52.3) ppm, of whom 21% were producers. Figure 2 shows the distribution of breath methane levels in

Fig. 1 Relative risk of being a methane producer in relation to age and location of all subjects. Slope of line (% per year): rural black -0.5 ; urban black -6.4 ; white $+0.1$; Indian -5.8 .

each population group. Distributions were all skewed but similar in each ethnic group.

Discussion

The findings in the present study show widely differing proportions of methane producers in different ethnic groups but do not support the view that methane production in the large intestine is a risk factor for large bowel cancer.

Table 3 summarises current information on the mortality and incidence of colonic and rectal cancer in the four population groups. Although there are a number of problems in estimating the reliability of these data, nevertheless some general conclusions can be drawn. Bowel cancer is uncommon in both urban and rural blacks as observed by the authors of many papers on the subject.^{21 22 28 29 31–33 35 36} As a proportion of all cancer deaths in the rural populations bowel is between one fifth and one tenth less common than in the United Kingdom. Incidence and mortality data for urban blacks show rates that are again about one tenth of those seen in England and Wales and in United States populations whilst in the white population of South Africa rates are much higher and similar to those in the west. Firm details for the Indian population are more difficult to obtain, probably because they are only a small fraction of the total population (0.9 M of a total of 26 M). Amongst Indians rates appear to be intermediate between whites and blacks. These data do not relate in any consistent way to methane status. The high proportion of methane producers found in patients with

Fig. 2 Frequency distribution of breath methane concentration (ppm) for all ages (producers only).

Table 3 Large bowel cancer (colon+rectum) incidence and mortality in South African populations

Population description	Age	Incidence (In) or mortality (Mo)	Rate/100 000	Sex	Source
Rural black					
Rural Bantu	All ages		7.5% of all cancers	M	Higginson and Oettlé 1960 ²¹
		In	0.8% of all cancers	F	
Shangaan, Swazi, and Sotho, N Transvaal	All ages	In	2.4% of all cancers	M	Sutherland 1960 ³⁶
'Rural'	All ages	In 'rare'	No recorded cases	F	Burkitt 1971 ³³
			1-2.5% of all cancers		
Urban Black Johannesburg	All ages	In	1.5	M	UICC 1960 ²⁶
			2.3	F	
Bantu-Johannesburg	All ages	Mo	2.3% of all cancers	M	Higginson and Oettlé 1960 ²¹
			3.1% of all cancers	F	
Bantu-Johannesburg	All ages standardised (African)	In	2.0	M	Higginson and Oettlé 1966 ³⁵
		In	3.5	F	
Cape Province	Age standardised (world)	In	6.9	M	UICC 1970 ²⁷
			8.8	F	
Natal	Age standardised (world)	In	3.1	M	UICC 1970 ²⁷
			6.0	F	
Soweto	45 cases over 12 year period (1954-1966) from population of 600 000				Bremner and Ackermann 1970 ³²
Johannesburg	35-64 'age standardised'	Mo	6.4	M	Burkitt 1971 ³³
Soweto	All ages	In	1.53	M	Isaacson <i>et al</i> 1978 ³¹
			1.0	F	
	60 yr +	In	7.71	M	
			7.51	F	
Soweto	All ages	Mo	0.8	M+F	Segal <i>et al</i> 1981 ²⁸
	45-64	Mo	2.2	M+F	
	65+	Mo	6.5	M+F	
Indian 'Asians'	Age standardised to US population	Mo	9.7	M	Oettlé 1964 ²²
			10.3	F	
Natal	Age standardised (world)	In	6.2	M	UICC 1970 ²⁷
			17.9	F	
'Asians' Durban	Age standardised (world)	Mo	5.0	M+F	Bradshaw and Harrington 1975 ²⁹
	6 deaths in 1983. Total population 450 000				City of Durban 1983 ³⁴
Whites National	Age standardised to US population	Mo	17.4	M	Oettlé 1964 ²²
			16.1	F	
Cape Province	Age standardised (world)	In	18.3	M	UICC 1970 ²⁷
			22.3	F	
National Johannesburg	Age standardised (world)	Mo	9-12	M+F	Bradshaw and Harrington 1975 ²⁹
	All ages	Mo	13.1	M+F	Segal <i>et al</i> 1981 ²⁸
	45-64	Mo	24.5	M+F	
	65+	Mo	55.0	M+F	

Total large bowel cancer incidence and mortality data for rural and urban black, white, and Indians in South Africa covering the period 1953-1971 approximately.^{21 26-36} For comparison: England and Wales (1985) colon and rectal cancer accounted for 11.2% of all cancer deaths in men, and 13.5% in women.³⁷ Average age standardised (world) incidence for six cancer registries in the UK (Birmingham, Oxford, Sheffield, South West, Liverpool, and Ayrshire) 1968-1972 is 29.8/100 000 for men, and 23.8 for women (colon+rectum).^{38 39}

bowel cancer by Haines *et al*¹³ and by Pique *et al*¹⁴ of 80% and 91% respectively must represent a change from normal due to the cancer rather than leading to its cause. This view is supported by the observations that methane producer status returns to that of the control population after surgery to remove the tumour.¹⁴

Breath methane data in the present study show marked ethnic differences and an association with age and sex. There is no consistency in the literature

at present with regard either to the effects of age or sex. In three studies^{8 10 15} no effect of sex was seen whilst in three others more women were producers than men.^{11 13 16} No study has convincingly shown men to be greater producers. With regard to age in adults again three studies^{8 10 15} show no effect whilst in the study of Haines *et al*⁴⁰ involving 1398 subjects there was an increase in the proportion of methane producers with age, the opposite of the present study. This was, however, entirely an urban population in

northwest London. Young children usually produce less methane than adults.^{10 17 19}

Few interethnic comparisons have been made. It is worth noting, however, that whilst Drasar *et al.*¹⁹ observed that 77% of 159 adult rural Nigerians were producers, Pitt *et al.*¹¹ found only 45% of black hospital staff in Toronto, Canada, were producers (total n=69) and similarly Haines *et al.*⁴⁰ observed 43% of black men and 33% of black women in a total sample of 99 persons in northwest London were producers. Pitt *et al.* also looked at other ethnic groups in Toronto and reported 24% of orientals, 48% of whites, and 32% of Indians were methane producers.

These interethnic comparisons are not as discordant as might seem at first sight. In general up to 61% (mostly 40–50%) of healthy urbanised adults of any race are methane producers whilst for rural blacks the proportion is 77–84%. Urbanised blacks in Africa are lower (72%) than rural blacks (84%) and approach the same proportion as whites (52%) in long established urban populations which have become fully acculturated – for example, Toronto and northwest London. In Soweto, however, which retains a more traditional diet and lifestyle the ethnic differences remain. Indians seem invariably to be low at 31% or less. How can these differences be explained?

Methane is produced in the gastrointestinal tract, both rumen¹ and caecum⁴¹ by oxidation of hydrogen by methanogenic bacteria. This process produces energy for the bacteria and an overall mechanism for terminal electron disposal thus permitting continued breakdown of fermentation intermediates to acetic acid. Methanogenesis is an example of interspecies hydrogen transfer which allows carbohydrate and amino acid fermenting species to oxidise more completely their substrates. The net effect of hydrogen oxidation is to increase substrate use and shift fermentation to more oxidised end products.⁴²

Despite the absence of methane in the breath of many people, methanogenic bacteria can be cultured from faeces in the majority^{3 4 6} and methane production from faecal cultures is almost universal in some studies^{15 19 43} regardless of breath concentrations, although reasonable concordance between faecal incubation and breath methane status has been observed by Levitt and Ingelfinger⁹ and Bjorneklett and colleagues.⁸ Wolin's group⁶ has suggested that methane appears in breath only when the numbers of methanogenic bacteria in the colon reach a critical level, about 10⁸/g dry weight contents, and that carriage of these bacteria is probably universal. Dividing a population into producers and non-producers may therefore be artefactual as everyone is a potential producer. What is unknown at present are

the factors controlling the number of methanogens and the amount of methane they produce.

Methanogenesis may be promoted by ensuring adequate hydrogen production from fermentation – that is, having sufficient substrate for fermentation available, by slow transit time²⁰ and mechanical factors as suggested by Weaver *et al.*⁶ Inhibition could arise through competition from sulphate reducers and the presence of sulphate⁴² or nitrate.⁴⁴ From the present study, however, slow transit would not seem to be an essential feature because whole gut transit time is known to be much faster in the populations (urban and rural blacks) which produce most methane.^{45–46} Conditions which favour methane production and their attendant health implications require further study.

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