

## Breath hydrogen excretion in infants with colic

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**SUMMARY** Breath hydrogen excretion as an index of incomplete lactose absorption was measured in 118 healthy infants who were either breast fed or given a formula feed containing lactose, some of whom had colic. Infants with colic (n=65) were selected on the basis of the mother's report of a history of inconsolable crying lasting several hours each day. Infants in the control group (n=53) were not reported to cry excessively by their mothers. Breath samples were collected using a face mask sampling device preprandially, and 90 and 150 minutes after the start of a feed. Normalised breath hydrogen concentrations were higher in the group with colic than in the control group at each time point. The median maximum breath hydrogen concentration in the colic group was 29 ppm, and in the control group 11 ppm. The percentage of infants with incomplete lactose absorption (breath hydrogen concentration more than 20 ppm) in the colic group was 62% compared with 32% in the control group. The clinical importance of the observed association between increased breath hydrogen excretion and infantile colic remains to be determined. Increased breath hydrogen excretion indicative of incomplete lactose absorption may be either a cause or an effect of colic in infants.

Infantile colic is a common but poorly defined condition of unknown cause that occurs in otherwise healthy infants during the first few months of life.<sup>1</sup> It is characterised by paroxysms of excessive and inconsolable crying.

Colic is important as a cause of distress to the infant and parents, it may result in premature weaning or change of formula feed, it may necessitate treatment of the infant, and be a possible stimulus for child abuse or domestic violence.<sup>1-3</sup> Children who have had colic seem to be predisposed to disturbances of sleep and have a 'difficult temperament'.<sup>2,4</sup> Despite these concerns there is no effective behavioural, nutritional, or pharmacological treatment for colic.

Mothers' reports of 'rumbling tummies', excessive flatus, and frothy stools in colicky infants suggest that a possible cause may be carbohydrate malabsorption. Furthermore, the concentration of hydrogen in the breath of healthy infants increases from birth to peak in the second month of life, declining to low concentrations by 3 to 4 months of age.<sup>5</sup> This pattern of breath hydrogen excretion parallels the pattern of crying in infants with colic. Raised concentrations are more frequently detected in the late afternoon or early evening, a time when colic is

commonly reported to occur.<sup>5</sup> Hydrogen in the breath of infants is derived exclusively from bacterial fermentation of unabsorbed carbohydrate in the intestine. Its measurement provides a sensitive, reliable, and semiquantitative index of incomplete carbohydrate absorption.<sup>6</sup> Despite the similarities between carbohydrate malabsorption and colic there has been no published study to our knowledge that has compared breath hydrogen excretion in infants with colic and a control group.

We report here a study of breath hydrogen excretion in infants, with or without colic, who were either breast fed or given a formula feed containing lactose. The hypothesis was that infants with colic absorbed lactose less efficiently than control infants.

### Subjects and methods

Healthy, full term infants between 3 and 20 weeks of age, whose growth and development had been normal since birth, and who were breast fed or given a formula feed containing lactose, were eligible for the study. Infants with colic were recruited from a Tresillian Family Care Centre (where infants and their mothers were admitted for mothercraft help),

or from baby health centres. These infants were selected if the mother reported a history of inconsolable crying lasting several hours each day for which no clinical cause could be found. Infants in the control (without colic) group were recruited from baby health centres and nursing mothers groups, and were not reported to cry excessively by their mothers. Infants were recruited to the study from July 1985 to June 1987. The following criteria would have excluded an infant from the study: treatment in the past two weeks with an oral antibiotic ( $n=0$ ), a recent attack of gastroenteritis ( $n=1$ ), use of an antacid or feed thickener ( $n=2$ ), weaning in the two weeks before the study ( $n=2$ ), negative in vitro faecal hydrogen test ( $n=4$ ), onset of colicky like behaviour after 6 weeks of age ( $n=5$ ), or incomplete collection of the series of breath samples because of inconvenience or distress caused by the procedure ( $n=7$ ).

Breath samples were collected in duplicate or triplicate preprandially, and 90 and 150 minutes after the start of the feed. No alteration was made to the feeding regimen. A sampling device comprising an infant face mask (Laerdal No 0, 1, or 2) and a two way valve and port system was used for the collection of breath samples.<sup>7</sup> During sampling the face mask was held firmly over the infant's nose and mouth. Small aliquots ( $<2$  ml) of each tidal volume from late in the expiratory phase were aspirated into a 20 ml syringe fitted with a stopcock until 20 ml had been collected. The breath sample was then transferred to a 20 ml evacuated blood collection tube (Venocject) and stored in a refrigerator until analysis. Breath samples can be stored in these tubes for at least seven weeks.<sup>8</sup> Tubes were marked with a code so that the analysis was done without knowing which group the baby was in. The time of sample collection was also recorded.

Breath samples were analysed for hydrogen and oxygen by gas chromatography within four weeks of collection.<sup>8</sup> The gas chromatograph (Hewlett Packard Model No 5710) was fitted with a thermal conductivity detector. The copper column (3.0 m $\times$ 6 mm) was packed with molecular sieve 5A 60/80 mesh. The operating temperature was 50°C and the carrier gas was argon (20 ml/minute). A sample of the breath gas was removed from the Venocject tube through a 20 ml gas tight syringe fitted with a locking device. After the syringe had been filled and locked off, the needle was detached and the plunger depressed to the 6 ml mark to raise the gas pressure within the syringe to above atmospheric pressure. The sample was then injected into the 5 ml sampling loop of the gas chromatograph. The instrument was calibrated using a commercial standard of hydrogen (mean (SD) 105 (2) ppm, Commonwealth Industrial

Gases) and ambient air (20.9% oxygen). The coefficients of variation for the analysis of the hydrogen standard and air were 4% and  $<1\%$ , respectively. The detection limit was 2 ppm hydrogen.

Hydrogen concentration was normalised to 18.9% oxygen (the mean in the first 50 subjects) to reduce variability related to sample collection.<sup>7</sup> Normalised hydrogen values of more than 20 ppm were taken to indicate incomplete lactose absorption on the day of sampling.<sup>9</sup> Samples with an oxygen concentration of more than 20% were discarded as technically inadequate.

Fresh stool samples were collected for testing the capacity of the faecal flora to produce hydrogen from lactose in vitro. About 200 mg of faeces were added to a 20 ml Venocject tube containing 1 ml 0.1 M phosphate buffered lactose (pH 7.2) 1.25% w/v, and 2 g sterile glass beads. The tube was closed with its stopper, shaken vigorously for 30 seconds, and incubated at 37°C for one hour. Headspace gas was collected by displacement with water into a 20 ml syringe, transferred to an evacuated Venocject tube, and analysed for hydrogen by gas chromatography. Hydrogen in excess of 100 ppm was chosen arbitrarily as an indicator of in vivo hydrogen producing capability (positive test).

Mothers were asked to complete a questionnaire regarding infant, maternal, and family history. The study protocol was approved by the Royal Society for the Welfare of Mothers and Babies (Tresillian) and the New South Wales Department of Health. Signed consent was obtained from the mother of each infant.

The breath hydrogen data were analysed as a nested factorial design by analysis of variance. Subsequent statistical analysis used the highest mean breath hydrogen value at any time point for each infant (maximum hydrogen) as there was no consistent temporal relationship between breath hydrogen excretion and the start of the feed. The maximum hydrogen values of the infants in the two groups were compared using a two sample Mann-Whitney U test. The effects of age and time on breath hydrogen excretion were examined by linear regression using the maximum hydrogen values. Other data were compared using two sample *t* tests, the equivalent two sided non-parametric test, or an appropriate contingency table.

## Results

The study comprised 118 infants, 65 with colic and 53 who acted as controls.

Breath hydrogen excretion was higher in the colic group than in the control group at each time point ( $p<0.025$ ); the maximum hydrogen values are

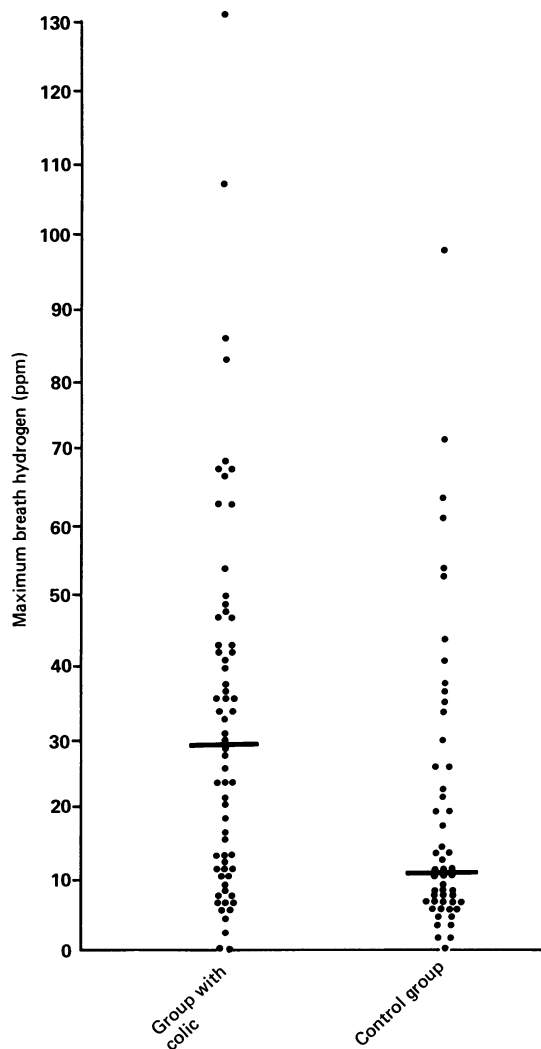


Figure Maximum breath hydrogen concentrations (ppm) in infants with and without infantile colic. Bars represents the medians (29 in the group with colic and 11 in the control group).

shown in the figure. The median maximum hydrogen value in the colic group was 29 ppm and in the control group 11 ppm ( $p < 0.001$ ). The mean (SD) oxygen concentrations in the samples from the two groups were 19.1 (0.7)% and 19.2 (0.6)%, respectively.

The percentage of infants with incomplete lactose absorption (breath hydrogen concentration of more than 20 ppm) in the colic group (62%) was significantly higher ( $\chi^2 = 10.15$ ,  $p < 0.01$ ) than in the control group (32%).

The characteristics of the infants in the study and aspects of maternal and family history are shown in tables 1 and 2. The infants with colic were younger than the control infants, but there was no correlation between breath hydrogen excretion and age in either group ( $r = -0.02$ ). Significantly fewer infants with colic were exclusively breast fed. Eight subjects in the colic group, however, had been weaned on to formula feed in an attempt to alleviate their colicky behaviour. The proportion of first born infants, and of infants whose sibling(s) or parent(s) were reported to have had colic, was significantly higher in the colic group. There were no significant differences between the two groups in family history of allergy, maternal age, obstetric history, or family income, education, or structure.

There were no significant differences between the colicky infants recruited from Tresillian Family Care ( $n = 39$ ) or baby health centres ( $n = 26$ ), or between non-colicky infants from baby health centres ( $n = 43$ ) or nursing mothers groups ( $n = 10$ ).

Preprandial samples were collected at a mean time of 1415 (range 0730–1930) in the colic group, and at 1100 (range 0830–1745) in the non-colic group ( $p < 0.001$ ). There was no correlation, however, between breath hydrogen excretion and time of sample collection ( $r = 0.06$ ).

## Discussion

In this study breath hydrogen excretion was significantly greater in infants with colic than those

Table 1 Characteristics of 65 infants with colic and 53 control infants

	Group with colic (n=65)	Control group (n=53)	p Value
No (%) of boys	36 (55)	32 (60)	NS
No (%) of girls	29 (45)	21 (40)	NS
Median (95% CI) age (weeks)	8 (6 to 9)	9 (7 to 12)	<0.025
Mean (95% CI) birth weight (g):			
Boys	3390 (3290 to 3490)	3670 (3530 to 3820)	<0.025
Girls	3300 (3170 to 3430)	3480 (3370 to 3590)	NS
No (%) breast fed	50 (77)	48 (91)	<0.05
No (%) first born	41 (63)	17 (32)	<0.001

Table 2 Maternal and family history in 65 infants with colic and 53 control infants

	Group with colic (n=65)	Control group (n=53)
Mean (95% CI) maternal age (years)	30 (29 to 31)	30 (29 to 31)
Mean (95% CI) weight gain during pregnancy (kg)	12.6 (11.4 to 13.8)	12.3 (10.6 to 14.0)
Mean (95% CI) duration of labour (hours)	8 (7 to 10)	8 (6 to 11)
No (%) of vaginal deliveries	54 (83)	42 (79)
No (%) having epidural anaesthesia	28 (43)	23 (43)
No (%) having forceps deliveries	19 (29)	9 (17)
No (%) with history of colic in sibling(s) or parent(s), or both	22 (34)	7 (13)*
No (%) with family history of allergy	30 (46)	21 (40)
No (%) of families with good income \$25-35 000	19 (29)	18 (34)
No (%) of families in which either parent had tertiary education	48 (74)	35 (66)
No (%) of single parent families	1 (2)	2 (4)

\* $p < 0.001$ .

without. Incomplete lactose absorption was twice as common in the colicky infants as in the control group. In contrast, Liebman found no association between infantile colic and lactose intolerance assessed by faecal pH and reducing substances.<sup>10</sup>

Incomplete lactose absorption in infants with colic may reflect the delayed development of adequate lactase capacity or mucosal damage—for example, being secondary to milk protein allergy. Other factors affecting the delivery of lactose to the colon could also be concerned. These factors include the duration and frequency of feeding, the rate of gastric emptying, and the transit time through the small intestine. Differences in breath hydrogen excretion between colicky and control infants may also be associated with factors other than the amount or rate of delivery of lactose to the colon. The nature of the bowel flora, colonic bacterial metabolic pathways, the partial pressure of hydrogen in the colon, the buffering capacity of the colon, gut perfusion, and incomplete monosaccharide absorption may all play a part.

Colic may result directly from the hyperperistaltic stimulus of the fluid load imposed by the osmotic action of unabsorbed lactose in the small intestine. Alternatively, colonic bacterial metabolism of unabsorbed lactose forming distending gas or pharmacologically active metabolites may be responsible for the signs of colic. Absorption and metabolism of the short chain fatty acid products of bacterial fermentation of unabsorbed lactose in the colon may compensate for the apparent energy loss associated with lactose malabsorption.<sup>11</sup> Absorption of short chain fatty acids stimulates the uptake of water from the colon and is coupled with secretion of bicarbonate into the colon.<sup>12</sup> These factors may explain why an infant who absorbs the lactose in a feed incom-

pletely can continue to thrive and not show any signs of intolerance.

Colic was significantly associated with being first born and with a family history of colic. It was not associated with maternal age, a family history of allergy, or socioeconomic status, this is consistent with other reports.<sup>13</sup> Prolonged labour, forceps delivery, and epidural anaesthesia have also been linked to the subsequent development of colic.<sup>14</sup> These factors and other aspects of obstetric history were not significantly different in the two groups of this study.

There is substantial overlap of breath hydrogen excretion values between the two groups of infants. More than a third (38%) of the infants in the colic group had low values of breath hydrogen while 32% of the infants in the control group had increased breath hydrogen excretion. This may reflect the aetiological heterogeneity of infantile colic. Alternatively, assignment of infants to the colic or control group simply by the mothers' perception of the amount of crying may not clearly distinguish colicky and non-colicky infants.

The clinical relevance of the association between the increased breath hydrogen excretion and infantile colic shown in this study remains to be determined. It may be that elevated breath hydrogen excretion is secondary to colic and is not a reflection of the cause of the behaviour. Studies designed to test whether reduced lactose intake prevents the development of colic or ameliorates established colic are therefore needed.

We thank matrons Miss M Newman and Mrs P Fikar and the staff of Tresillian Willoughby Residential Unit, the nursing staff of the Sydney inner metropolitan baby health centres, Sister A Mitchell, Karitane Mothercraft Society, and members of the the Sydney group of Nursing Mother's Association of Australia for their

cooperation; Dr D Bau for allowing us to study his patients; Mr T Robb for the supply of the breath collection apparatus; and Mrs F Duncan for typing the manuscript.

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Accepted 29 November 1988