The 15 g D-xylose absorption test: its application to the study of coeliac disease

FIONA M. STEVENS, D. W. WATT, MARY A. BOURKE, B. McNICHOLL, P. F. FOTTRELL, AND C. F. MCCARTHY

From the Department of Gastroenterology, Regional Hospital, Galway and the Departments of Medicine, Paediatrics, and Biochemistry, University College, Galway

SUMMARY The absorption of xylose following an oral load of 15 g D-xylose has been studied by serial blood levels in 17 untreated adult coeliac patients, 21 treated coeliac patients, and 30 non-coeliac patients. A statistically significant difference in xylose blood levels was found between untreated coeliac and non-coeliac patients at all the times studied, but a complete separation between these two groups occurred only at the 75 minute stage. The reproducibility of absorption was assessed by repeating the test in 16 subjects. The 95% confidence limits of the standard error of estimate are narrowest at 75 and 90 minutes.

Dicke's original demonstration of the harmful effects of wheat was based on increased faecal fat excretion in coeliac patients fed this cereal (Dicke *et al.*, 1953). This method of investigation lacks sensitivity as steatorrhoea is not present in all untreated coeliac patients.

Deterioration of small intestinal morphology and absorption in treated coeliac patients following challenge with fractions of gliadin have been the methods of determining if a given fraction is toxic to the coeliac patient.

The demonstration of histological damage to the small intestinal mucosa of coeliac patients following ingestion of gliadin fractions requires that a jejunal biopsy be carried out, and patients are not always willing to have this done. Therefore, an alternative test is necessary for screening large numbers of gliadin fractions.

Decreased urinary excretion of D-xylose following an oral load has been used as an index of the toxicity of various gliadin fractions (Shmerling and Shiner, 1970; Bayless *et al.*, 1970; Kendall *et al.*, 1972a and b; Hamilton and McNeill, 1972; Rolles and Kendall 1973; Townley *et al.*, 1973; Hekkens *et al.*, 1974; Schneider *et al.*, 1974; Cornell, 1974; Cornell and Townley, 1974). Rolles *et al.* (1975) have used a one-hour blood xylose test to confirm persisting gluten intolerance in children.

In this study, D-xylose absorption has been reinvestigated to determine the reproducibility of

absorption in the individual and its potential as a screening method for occult adult coeliac disease in the population.

Material and methods

A standard dose of 15 g D-xylose (BDH 30590) in 250 ml tap water (resultant osmolality approx. 440 mOsm/kg) was given orally after a fast of at least 8 hours. No food or fluid was given until the test was completed. Serial venous blood samples were taken into fluoride oxalate tubes at 0, 30, 60, 75, 90, and 120 minutes (samples were not available from all subjects at 30, 75, and 120 minutes). Samples were stored at 4°C until analysed. Xylose levels in deproteinized blood were determined within 48 hours by the method of Roe and Rice (1948), optical density readings being made at 520 nm using a Pye Unicam SP 600 spectrophotometer. All determinations were carried out in duplicate and the mean result was recorded.

The following subjects were studied: 30 patients undergoing investigation for anaemia, diarrhoea or short stature but subsequently shown to have morphologically normal small intestinal mucosal biopsies; 17 untreated adult coeliac patients; and 21 coeliac patients who had been treated with a gluten-free diet for at least three months.

Repeat absorption studies were carried out in 6 subjects—four untreated adult coeliacs, three treated adult coeliacs, and nine volunteer controls (departmental personnel). The second test was per-

formed after an interval of at least five days and not longer than six weeks.

No patient with renal disease (serum creatinine > 160 μ mol/l), diabetes, Crohn's disease, tropical sprue or ascites was accepted into the study. No patient was receiving any drugs at the time of investigation known to reduce xylose absorption. However, some patients were taking iron, folate, B12, calcium, and vitamin supplements before the study.

Fasting xylose levels (expressed as mmol/l) were taken as a baseline for each subject and subtracted from the other results obtained.

All clinical aspects of the tests were carried out by FMS, and the blood xylose assays by DWW and MAB in the Department of Gastroenterology.

Results

Figure 1 shows the standard error of estimate (SEE) of the differences between the blood xylose levels in repeated tests in the same individual at various times. The 95% confidence limits of the SEE are least at 75 and 90 minutes.

Figure 2 (a-e) shows the distribution of the blood xylose levels in untreated and treated coeliac patients and controls at the times indicated. The statistical significance of these results is given under Figure 2. There is a significant difference between untreated coeliac patients and both other groups at all times. There is no statistically significant difference between the treated coeliacs and controls at 30 minutes or 120 minutes.

Figure 3 (a-c) shows the mean ± 1 standard deviation of the mean of the 15 g D-xylose absorption curves in (a) non-coeliac patients, (b) untreated coeliac patients, and (c) treated coeliac patients. The peak blood level in controls is before 90 minutes, whereas in both treated and untreated coeliacs the peak is at or after 90 minutes.

Fig. 1 Standard error of estimate (SEE) of differences between the blood xylose levels in repeated tests in the same individual at the times indicated. *n* indicates the number of paired tests at each time. NC = non-coeliac patients; UC = untreated coeliac patients; TC = treated coeliac patients.

Fig. 2 (a-e) Rises in blood xylose levels above fasting sample at times indicated; NC = non-coeliac patients; UC = untreated coeliac patients; TC = treated coeliac patients. Student's *t* test was used for statistical analysis of the results. A *P* value of < 0.05 was considered to be significant.

Fig. 3 Mean ± 1 standard deviation of mean of 15 g D-xylose absorption tests in (a) non-coeliac patients, (b) untreated coeliac patients, and (c) treated coeliac patients.

Discussion

The 95% confidence limits of the standard error of estimate of the 15 g D-xylose absorption test in this study were smallest at 75 and 90 minutes. It appears that the 30 and 60 minute levels are dependent upon the rate of gastric emptying. Although in this study the volume and osmolality have been kept constant, other factors such as stress and mood affect the rate of gastric emptying. The two-hour value may vary as the rate of absorption is no longer linear (McCance and Madders, 1930), the xylose having passed beyond the region of maximal absorption. Nevertheless the reproducibility of the test in the same patient is such that it is reasonable to use it as a screening test when assessing the toxicity of gliadin fractions. In further studies, the changes in xylose absorption have been found to parallel intestinal biopsy morphologic changes when toxic or non-toxic gliadin fractions are fed (in preparation).

Few studies of reproducibility of xylose absorption have been reported. Rolles *et al.* (1973) found a maximal difference of 0.28 mmol/l when repeating tests in controls and coeliac children. Lamabadusuriya *et al.* (1975) found a coefficient of variation of 7% in five tests on the same individual.

In this study untreated coeliac and non-coeliac patients could be differentiated from one another on the basis of their blood D-xylose levels at 75 minutes post xylose, but the sample number is limited. Five of 31 controls fell within the limits of the untreated coeliac group at the 90 minute stage. Four of these five non-coeliac patients were post maximum values, whereas the coeliac patients were at or about the peak. Xylose estimation on a single sample of blood taken after an oral load of D-xylose is of less value than estimations on samples taken at intervals after the oral dose, as the time of the peak blood level varies from individual to individual.

Xylose blood levels in coeliac patients, treated with a gluten free diet for at least three months, are significantly higher than those found in untreated coeliac patients but remain below the levels found in non-coeliac patients. This indicates continued malabsorption in these treated coeliac patients due either to occasional dietary lapses or to the inability of the intestinal mucosa of these adult coeliac patients to respond completely to gluten withdrawal. A marked improvement in the one-hour blood xylose level has been noted within one week in coeliac children treated with a gluten free diet (Rolles *et al.*, 1973).

Hawkins (1970) found that both the one and two hour blood xylose concentration following a dose of xylose of 0.5 g/kg body weight distinguished between coeliac and non-coeliac children. Lamabadusuriya *et al.* (1975), using 0.4 g/kg body weight of D-xylose given at a constant osmolality, were unable to demonstrate such a distinction one hour post xylose. Rolles *et al.* (1973), giving a constant dose of 5 g of D-xylose to children under 30 kg body weight, did report almost complete differentiation at the one hour interval. Wolfish *et al.* (1955), giving a large dose of 1.1 g/kg body weight to infants, reported differentiation in tolerance tests.

In adults following a 25 g D-xylose dose, Benson *et al.* (1957) found blood xylose levels distinguished between coeliac and non-coeliac patients at 30 and 60 minutes but not at 90 and 120 minutes. Roberts *et al.* (1960), using the same dose, found overlap at 30 and 120 minutes but complete distinction at 60 minutes. Sladen and Kumar (1973) found overlap at 60, 120, and 150 minutes, although the mean values of the two groups of patients were statistically significantly different at all times. Thaysen and Müllertz (1962) found complete separation between the maximal

blood levels of coeliac and non-coeliac patients after 25 g of D-xylose. Chanarin and Bennett (1962), using a weight-related dose, found an overlap in peak blood D-xylose levels after 0.3 g/kg but complete differentiation in peak blood levels after 0.4 g/kg.

Thaysen and Müllertz (1962) noted that the maximal blood xylose levels occurred later in coeliac patients than in controls. This finding was confirmed in the present study and is probably an indication of the more severe damage to the proximal duodenum and the more efficient absorption in the less affected distal mucosa. The same delayed pattern of absorption exists in treated coeliac patients, indicating continuing malfunction in the proximal duodenum.

As xylose is found in the normal diet (being one of the sugars in fruits and cereal products) (Johnstone, 1906), blood levels after D-xylose administration are given as the rise above the fasting sample. In addition, other aldopentoses present in the blood produce furfurals in the presence of acetic acid and give a positive colour reaction with the *p*-bromoaniline used in the measurement of blood xylose (Roe and Rice, 1948). The range for fasting samples in this study is 0.0-0.7326 mmol/l (mean 0.1065, SD 0.1398).

The rate of absorption of xylose is related to the rate of gastric emptying, which in turn is influenced by the osmolality of the gastric contents, mediated by osmoregulators in the duodenum (Hunt and Pathak, 1960; Hunt, 1963; Elias *et al.*, 1968; Meeroff *et al.*, 1973). Both hyperosmolar and hyposmolar solutions of monosaccharides and disaccharides delay gastric emptying (Elias *et al.*, 1968). However, osmolality is not the only factor as hyperosmolar ethanol (1300 mOsm/kg) does not delay gastric emptying (Cooke, 1970). Webster and Leeming (1975) showed no delaying of gastric emptying following a hyperosmolar solution of xylose and Micropaque (approx. 1100 mOsm/kg). The osmolality of the test solution is important for another reason. Hyperosmolar solutions (osmolality >1500 mOsm/kg) give increased rates of absorption by rendering normal mucosa more permeable, increased permeability occurring at lower osmolalities in coeliac disease (Menzies, 1972a and b; Creamer, 1974).

Urinary excretion of D-xylose following a 25 g oral dose has been used by Bayless *et al.* (1970) during the study of the effects of gluten on jejunal mucosa, and a decrease in urinary excretion was found to accompany mucosal morphologic and biochemical deterioration. A dose of 25 g of D-xylose is frequently associated with diarrhoea, nausea, and abdominal discomfort. This is due to bacterial degradation of the pentose in the colon, as more than 40% of the 25 g oral dose remains unabsorbed (Fordtran *et al.*, 1962). An oral dose of 5 g of D-xylose is insufficient to test intestinal absorptive

capacity, as even with some mucosal damage the whole of the dose may be absorbed. In addition, any bacterial overgrowth in the upper small bowel will lessen still further the dose presented to the mucosa for absorption (Goldstein *et al.*, 1970).

A dose of 15 g of D-xylose in 250 ml of water is without side effects in non-coeliacs and most coeliac patients in this study. There was some increase in diarrhoea in coeliac patients with gross steatorrhoea before the study.

Many workers have used impairment of the urinary excretion of xylose after an oral load as a basis for the detection of small intestinal mucosal absorptive defects. Urinary collections were not made in this study because during a pilot study both the 0.2 and 2.5 urinary excretions were found to vary considerably when repeated in the same individual (unpublished data).

An absorption test of 15 g D-xylose in 250 ml of water has been studied. This volume was acceptable to the patients, and the test was free from side effects except for increased diarrhoea in two coeliac patients. An essential part of the test was found to be the fasting sample, as this provided a base line for the individual patient. It is important to determine the patient's renal function status and to note any drug therapy which may impair xylose absorption before interpreting the result.

It is doubtful whether the test would be of use in population screening of asymptomatic coeliac relatives with villous atrophy and only minimal impairment of absorption, as these patients would probably fall in the overlap zone between overt coeliacs and non-coeliacs.

The standard error of estimate of the readings of 15 g D-xylose absorption in the same individual are sufficiently small for the test to be used to demonstrate changes in intestinal absorptive function. This test may be useful in the preliminary screening of fractions before selecting specific fractions to be fed to patients willing to undergo repeat intestinal biopsy.

We wish to thank Dr Helen Grimes and her staff for serum creatinine assays; the physicians and surgeons of the Regional Hospital, Galway for sending their patients for study; Una Cradock for secretarial assistance; and the Western Health Board and the Wellcome Trust for generous financial support.

References

- Bayless, T. M., Rubin, S. E., Topping, T. M., Yardley, J. H., and Hendrix, T. R. (1970). Morphologic and functional effects of gluten feeding on jejunal mucosa in celiac disease. In *Celiac Disease*, edited by C. C. Booth and R. H. Dowling, pp. 76-89. Churchill Livingstone, Edinburgh and London.

- Benson, J. A., Jr., Culver, P. J., Ragland, S., Jones, C. M., Drumme, G. D., and Bougas, E. (1957). The D-xylose absorption test in malabsorption syndromes. *New England Journal of Medicine*, **256**, 335-339.
- Chanarin, I. and Bennett, M. C. (1962). Absorption of folic acid and D-xylose as tests of small-intestinal function. *British Medical Journal*, **1**, 985-989.
- Cooke, A. R. (1970). The simultaneous emptying and absorption of ethanol from the human stomach. *American Journal of Digestive Diseases*, **15**, 449-454.
- Cornell, H. J. (1974). Gliadin degradation and fractionation. In *Coeliac Disease*, edited by W. T. J. M. Hekken and A. S. Peña, pp. 74-75. Stenfort Kroese, Leyden.
- Cornell, H. J. and Townley, R. R. W. (1974). The toxicity of certain cereal proteins in coeliac disease. *Gut*, **15**, 862-869.
- Creamer, B. (1974). Intestinal permeability as a screening test for mucosal damage. In *Coeliac Disease*, edited by W. T. J. M. Hekken and A. S. Peña, pp. 348-349. Stenfort Kroese, Leyden.
- Dicke, W. K., Weijers, H. A., and van de Kamer, J. H. (1953). Coeliac disease II. The presence in wheat of a factor having a deleterious effect in cases of coeliac disease. *Acta Paediatrica*, **42**, 34-42.
- Elias, E., Gibson, G. J., Greenwood, L. F., Hunt, J. N., and Tripp, J. H. (1968). The slowing of gastric emptying by monosaccharides and disaccharides in test meals. *Journal of Physiology*, **194**, 317-326.
- Fordtran, J. S., Soergel, K. H., and Ingelfinger, F. J. (1962). Intestinal absorption of D-xylose in man. *New England Journal of Medicine*, **267**, 274-278.
- Goldstein, F., Karacag, S., Wirts, C. W., and Kowlessar, O. D. (1970). Intraluminal small-intestinal utilization of D-xylose by bacteria. *Gastroenterology*, **59**, 380-386.
- Hamilton, J. R. and McNeill, L. K. (1972). Childhood coeliac disease: response of treated patients to a small uniform daily dose of wheat gluten. *Journal of Pediatrics*, **81**, 885-893.
- Hawkins, K. I. (1970). Pediatric xylose absorption test: measurements in blood preferable to measurements in urine. *Clinical Chemistry*, **16**, 749-752.
- Hekken, W. T. J. M., van den Aarsen, C. J., Gilliams, J. P., Lems-Van Kan, P. H., and Bouma-Frölich, G. (1974). α Gliadin structure and degradation. In *Coeliac Disease*, edited by W. T. J. M. Hekken and A. S. Peña, pp. 39-45. Stenfort Kroese, Leyden.
- Hunt, J. N. (1963). The duodenal regulation of gastric emptying. *Gastroenterology*, **45**, 149-156.
- Hunt, J. N. and Pathak, J. D. (1960). The osmotic effects of some simple molecules and ions on gastric emptying. *Journal of Physiology*, **154**, 254-269.
- Johnstone, R. W. (1906). Pentosuria, chronic and alimentary. *Edinburgh Medical Journal*, **20**, 138-148.
- Kendall, M. J., Cox, P. S., Schneider, R., and Hawkins, C. F. (1972a). Gluten subfractions in coeliac disease. *Lancet*, **2**, 1065-1067.
- Kendall, M. J., Nutter, S., and Hawkins, C. F. (1972b). Testing gluten sensitivity by the xylose test. *Lancet*, **1**, 667-668.
- Laabadusuriya, S. P., Packer, S., and Harries, J. T. (1972). Limitations of xylose tolerance test as a screening procedure in childhood coeliac disease. *Archives of Disease in Childhood*, **50**, 34-39.
- McCance, R. A., and Madders, K. (1930). The comparative rates of absorption of sugars from the human intestine. *Biochemical Journal*, **24**, 795-804.
- Meeroff, J. C., Go, V. L. W., and Phillips, S. F. (1972). Measurement of gastric emptying by duodenal aspiration: effects of intraduodenal osmolarity (Abstract). *Gastroenterology*, **64**, 770.
- Menzies, I. S. (1972a). Alimentary disacchariduria in adults related to the osmolality of ingested solutions. *Biochemical Journal*, **126**, 19-20.
- Menzies, I. S. (1972b). Intestinal permeability in coeliac disease (Abstract). *Gut*, **13**, 847.
- Roberts, J. G., Beck, I. T., Kallos, J., and Kahn, D. S. (1966). D(+)xylose blood-level time-curve as an index of intestinal absorption with a description of a simplified method for estimation of blood xylose levels. *Canadian Medical Association Journal*, **83**, 112-117.
- Roe, J. H. and Rice, E. W. (1948). A photometric method for the determination of free pentoses in animal tissues. *Journal of Biological Chemistry*, **173**, 507-512.
- Rolls, C. J., Anderson, C. M., and McNeish, A. S. (1972). Confirming persistence of gluten intolerance in children diagnosed as having coeliac disease in infancy. Usefulness of one-hour blood xylose test. *Archives of Disease in Childhood*, **50**, 259-263.
- Rolls, C. J., and Kendall, M. J. (1973). Gluten provocation and the xylose test in adolescents (Letter). *Lancet*, **2**, 380.
- Rolls, C. J., Kendall, M. J., Nutter, S., and Anderson, C. M. (1973). One-hour blood-xylose screening-test for coeliac disease in infants and young children. *Lancet*, **2**, 1043-1044.
- Schneider, R., Kendall, M. J., and Hawkins, C. F. (1974). Gliadin subfractionation. In *Coeliac Disease*, edited by W. T. J. M. Hekken and A. S. Peña, pp. 72-73. Stenfort Kroese, Leyden.
- Shmerling, D. H., and Shiner, M. (1970). The response of the intestinal mucosa to the intraduodenal instillation of gluten in patients with coeliac disease during remission. *Coeliac Disease*, edited by C. C. Booth and R. H. Dowling, pp. 64-74. Churchill Livingstone, Edinburgh and London.
- Sladen, G. E., and Kumar, P. J. (1973). Is the xylose test still a worth-while investigation? *British Medical Journal*, **2**, 223-226.
- Thaysen, E. H. and Müllertz, S. (1962). The D-xylose absorption tolerance test. *Acta Medica Scandinavica*, **171**, 521-529.
- Townley, R. R. W., Bhathal, P. S., Cornell, H. J., and Mitchell, J. D. (1973). Toxicity of wheat gliadin fractions in coeliac disease. *Lancet*, **1**, 1362-1364.
- Webster, S. G. P., and Leeming, J. T. (1975). Assessment of small bowel function in the elderly using a modified xylose tolerance test. *Gut*, **16**, 109-113.
- Wolfsch, M. G., Hildick-Smith, G. J., Ebbs, J. H., Conner, M. L., and Sass-Kortsak, A. (1955). The d-xylose tolerance test: a measure of intestinal absorption in normal and dystrophic infants (Abstract). *American Journal of Diseases of Children*, **90**, 609-610.