BIOCHEMICAL CHANGES IN INTESTINAL MUCOSA AFTER EXPERIMENTAL SMALL BOWEL BY-PASS IN THE RAT

M. H. GLEESON, R. H. DOWLING AND T. J. PETERS

Department of Medicine and Medical Research Council Intestinal Malabsorption Group, Royal Postgraduate Medical School, Hammersmith Hospital, London

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SUMMARY

1. After experimental small bowel by-pass the excluded segment of intestine, whether jejunum or ileum, shows a decrease (expressed in units/cm of intestine) in mucosal weight, protein, RNA and activity of all eight enzymes studied (leucyl β-naphthylamidase, alkaline phosphatase, β-galactosidase, α-glucosidase, aryl sulphatase, β-glucuronidase, cytochrome oxidase and catalase). There is a fall in DNA content and there are no consistent changes in enzyme activity when expressed as units/mg of DNA.

2. The intestine remaining in continuity after small bowel by-pass shows an increase (expressed in units/cm of intestine) in mucosal weight, protein, RNA and enzyme activity. There is an accompanying rise in DNA content in the intestine in continuity.

3. The increased values of all variables measured in the intestine in continuity are much more marked in ileum than in jejunum.

4. The adaptive changes seen in the by-passed intestine and the intestine in continuity are a result of 'hypoplasia' and 'hyperplasia' respectively.

Key words: intestinal by-pass, mucosal enzyme activities, mucosal DNA and RNA content, rat.

After small bowel resection in the rat the remaining small intestine shows mucosal ‘hypertrophy’ (Loran & Althausen, 1958; Booth, Evans, Menzies & Street, 1959) and enhanced absorption (Dowling & Booth, 1967; Nygaard, 1967). The term ‘hypertrophy’ has been used to describe the macroscopic appearance of the intestine; as this paper shows, at the cellular level, ‘hyperplasia’ is the more appropriate term. These changes are much greater in the ileum after a proximal (jejunal) small bowel resection, than in the jejunum after distal (ileal) resection, although after removal of the ileum the jejunum does show significant hypertrophy and an increased absorptive capacity (Dowling & Booth, 1967). The mechanism by which

Correspondence: Dr R. Hermon Dowling, Department of Medicine, Royal Postgraduate Medical School, Ducane Road, London, W.12.
these hypertrophic changes occur is uncertain, but Dowling (1967) has suggested that the nutrient content within the intestinal lumen may be an important factor.

It seems likely that similar structural and functional changes occur in the residual intestine that remains in continuity following by-pass, as seen when segments of intestine are resected (Flint, 1912; Dowling, 1968).

The exclusion of segments of proximal and distal small bowel from normal continuity by surgical by-pass provides the opportunity not only to study the effects of deprivation of luminal nutrition from the by-passed intestine, but also to examine the changes in the intestine that remains in continuity. We have previously shown that in by-passed rat intestine, although villous architecture is preserved, with time there is a progressive deterioration in absorptive function (Gleeson, Cullen & Dowling, 1972).

The aim of the present study was to investigate whether these changes in absorptive function were accompanied by quantitative changes in mucosal protein, DNA, RNA and enzyme content, both in by-passed intestine and in small bowel that remained in continuity. To study possible enzyme changes in the different sub-cellular organelles, brush-border, mitochondrial, lysosomal and peroxisomal enzymes were studied. The changes in both jejunal and ileal segments were compared with similar measurements in normal rat small intestine. A preliminary account has been presented (Gleeson, Peters, White & Dowling, 1970).

MATERIALS AND METHODS

Operative procedures

Female Wistar rats weighing 160–200 g at the time of the original surgical procedure were used. Full details of the operative techniques are described by Gleeson et al. (1972).

Experimental groups

There were six animals with proximal and six with distal small bowel by-pass.

Throughout, the results for excluded jejunum and for ileum in continuity were obtained from the proximal by-pass group while distal by-pass provided the results for excluded ileum and for jejunum in continuity.

There were six control rats which underwent a simple transection and resuture at the mid-point of the small bowel.

All animals were maintained under identical conditions and received the same diet (Diet 41B, Crane Mead Mills, Ware, Herts.). The quantitative mucosal studies in both experimental groups and in their matched controls were carried out between 12 and 25 weeks after operation. [We had established that the maximal reduction in absorptive capacity for glucose was apparent 10 weeks after by-pass (Gleeson et al., 1972) and that the maximal increase in glucose absorption for intestine in continuity after resection was fully developed by 1 month (Dowling & Booth, 1967).]

Preparation of mucosal homogenates

The animals were starved overnight, killed by a blow on the head and the abdomen rapidly opened. In the experimental groups both the excluded segment of intestine and the portion remaining in continuity were removed and washed through with 50 ml of ice-cold 0·15 M-NaCl. The length of intestine to be homogenized was measured against a vertical scale using a
Mucosal enzymes after small bowel by-pass

standard 5 g stretch. The pieces of intestine were everted by using a metal rod and washed in four changes of ice-cold saline. After gentle blotting with tissue paper to remove excess of saline, the mucosa was scraped from the serosa and muscle coats by using two glass slides. The mucosa was then weighed rapidly and homogenized in ice-cold water (10 vol./g wet wt.) for 30 s by using a Waring Blendor. Samples of the homogenates were stored at -25°C until assay. Before all measurements the samples were sonicated for 2-4 s in a 100 Watt M.S.E. ultrasonic disintegrator.

In addition to measuring mucosal mass/unit length of intestine by simple weighing, the mucosal protein, RNA and DNA contents were measured. The mucosal enzymes which were measured may be sub-divided into brush border enzymes (leucyl β-naphthylamidase, alkaline phosphatase, α-glucosidase (maltase) and β-galactosidase (lactase), lysosomal enzymes (aryl sulphatase and β-glucuronidase), mitochondrial (cytochrome oxidase) and peroxisomal enzymes (catalase). Results were obtained for four to six animals in each group with the exception of lactase, where for technical reasons there was insufficient homogenate to complete the assay in all groups (Table 3).

Analytical techniques

All assays were done in duplicate.

RNA and DNA. These were measured by the method of Hatcher & Goldstein (1969) using yeast RNA (Koch-Light Laboratories, Colnbrook, Bucks.) and calf thymus (Koch-Light) as standards. Duplicate assays agreed within ±5%. Corrections were made for the presence of DNA in the orcinol assay for RNA.

Protein. This was estimated by the method of Lowry, Rosebrough, Farr & Randall (1951) with bovine serum albumin (Armour Pharmaceutical Co. Ltd, Hampden Park, Eastbourne, Sussex) as standard.

Leucyl β-naphthylamidase. This enzyme (aryl amidase) (EC 3.4.1.1) was assayed by the method of Goldberg & Rutenberg (1958).

Alkaline phosphatase. This enzyme (EC 3.1.3.1) was assayed by the method of Besey, Lowry & Brock (1946) with p-nitrophenol phosphate as substrate but with the addition of the following metal ion activators: 5 mM-Co²⁺, 2 mM-Zn²⁺ and 0.2 mM-Mg²⁺ (Eichholz, 1967).

β-Galactosidase (lactase) (EC 3.2.1.23) and α-glucosidase (maltase) (EC 3.2.1.20). These enzymes were assayed by the method of Dahlqvist (1964).

Aryl sulphatase. This enzyme (EC 3.1.6.1) was assayed with p-nitrophenyl sulphate as substrate by the method of Roy (1952). Triton X-100 (0-1%) was present in the incubation medium.

β-Glucuronidase. This enzyme (EC 3.2.1.31) was measured, with phenolphthalein glucuronide as substrate, by the method of Fishman (1967). Triton X-100 (0-1%) was present in the incubation medium. Colour development was by the technique of Hübscher, West & Brindley (1965).

Cytochrome oxidase. This enzyme (EC 1.9.3.1) was assayed as described by Cooperstein & Lazarow (1951).

Catalase. This enzyme (EC 1.11.1.6) was measured by the technique of Lück (1965).

Units of enzyme activity

One unit of activity corresponds to the hydrolysis of 1 μmol of substrate/h at 37°C for all enzymes except the disaccharidases, cytochrome oxidase and catalase. One unit of disacchari-
dase activity corresponds to the hydrolysis of 1 μmol of substrate/min. One unit of catalase activity is the amount of enzyme that liberates half the peroxide oxygen from the solution of H₂O₂ in 100 s (Lück, 1965). One unit of cytochrome oxidase is the unit change in log₁₀ E₅₅₀/min at 23°C (Cooperstein & Lazarow, 1951).

RESULTS

Mucosal weight and protein (Table 1 and Fig. 1)

The wet weight and protein content of the normal ileum was less than that of normal jejunum, but these differences were not statistically significant (paired t-test).

In excluded intestine after both proximal and distal by-pass, there were slight but statistically insignificant falls in mucosal wet weight compared with similar segments of control intestine. However, there was a pronounced and significant decrease in protein content of both excluded jejunum and ileum. By contrast, in intestine remaining in continuity (included intestine) highly significant increases in mucosal weight and protein content occurred and these changes were more marked in ileum than in jejunum.
Table 1. Mucosal wet weight, mucosal DNA and RNA content of jejunum and ileum from controls and from animals with experimental bypass. All data are expressed as mean ± SE of observations in five or six animals. P values in parentheses refer to the significance of differences between the control group and the two experimental groups. P values in square brackets refer to the significance of differences between control jejunum and ileum in the same animal, using the paired t-test.

<table>
<thead>
<tr>
<th></th>
<th>Jejunum</th>
<th>Ileum</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Excluded (Proximal by-pass)</td>
<td>Control (Distal by-pass)</td>
</tr>
<tr>
<td>Mucosal wet weight</td>
<td>40.8 ± 4.9 (P = 0.224)</td>
<td>47.2 ± 1.9</td>
</tr>
<tr>
<td>(mg/cm of intestine)</td>
<td></td>
<td>(P = 0.009)</td>
</tr>
<tr>
<td>Mucosal DNA</td>
<td>0.39 ± 0.01 (P = 0.062)</td>
<td>0.52 ± 0.06</td>
</tr>
<tr>
<td>(mg/cm of intestine)</td>
<td></td>
<td>(P = 0.108)</td>
</tr>
<tr>
<td>Mucosal RNA</td>
<td>0.18 ± 0.04 (P = 0.001)</td>
<td>0.41 ± 0.03</td>
</tr>
<tr>
<td>(mg/cm of intestine)</td>
<td></td>
<td>(P = 0.32)</td>
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</tbody>
</table>
TABLE 2. Mucosal leucyl \( \beta \)-naphthylamidase and alkaline phosphatase of jejunum and ileum from controls and from animals with experimental by-pass. All data expressed as the mean ± SE of observations in four to six animals. \( P \) values in parentheses refer to the significance of differences between the control group and the two experimental groups. \( P \) values in square brackets refer to the significance of differences between control jejunum and ileum in the same animal using the paired \( t \)-test.

<table>
<thead>
<tr>
<th></th>
<th>Excluded (Proximal by-pass)</th>
<th>Control</th>
<th>In continuity (Distal by-pass)</th>
<th>Excluded (Distal by-pass)</th>
<th>Control</th>
<th>In continuity (Proximal by-pass)</th>
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<tbody>
<tr>
<td>Leucyl ( \beta )-naphthyl amidase (units/cm of intestine)</td>
<td>9.6 ± 1.2 ( (P = 0.002) )</td>
<td>19.2 ± 1.7</td>
<td>35.2 ± 3.1 ( (P = 0.001) )</td>
<td>6.7 ± 0.7 ( (P &lt; 0.001) )</td>
<td>18.5 ± 1.8 ( [P = 0.54] )</td>
<td>43.3 ± 6.5 ( (P = 0.003) )</td>
</tr>
<tr>
<td>(units/mg of DNA)</td>
<td>30.0 ± 3.9 ( (P = 0.155) )</td>
<td>40.2 ± 5.3</td>
<td>51.1 ± 5.8 ( (P = 0.218) )</td>
<td>33.9 ± 4.4 ( [P = 0.166] )</td>
<td>42.9 ± 4.0 ( [P = 0.23] )</td>
<td>33.8 ± 6.1 ( (P = 0.229) )</td>
</tr>
<tr>
<td>Alkaline phosphatase (units/cm of intestine)</td>
<td>51.4 ± 16.8 ( (P = 0.004) )</td>
<td>168.9 ± 24.3</td>
<td>231.9 ± 17.2 ( (P = 0.073) )</td>
<td>11.3 ± 3.3 ( (P = 0.007) )</td>
<td>30.8 ± 4.3 ( [P = 0.003] )</td>
<td>85.2 ± 5.2 ( (P &lt; 0.001) )</td>
</tr>
<tr>
<td>(units/mg of DNA)</td>
<td>129.2 ± 38.0 ( (P = 0.041) )</td>
<td>335.6 ± 80.5</td>
<td>338.2 ± 63.8 ( (P = 0.981) )</td>
<td>62.0 ± 21.7 ( (P = 0.673) )</td>
<td>71.9 ± 10.3 ( [P = 0.046] )</td>
<td>65.9 ± 11.3 ( (P = 0.706) )</td>
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Mucosal DNA and RNA (Table 1)

As with mucosal weight and protein, the DNA and RNA content of normal ileum was less than that of normal jejunum, and again the differences were not statistically significant.

By-passed jejunum showed a decrease in DNA content but in the small number of animals studied, the difference from control jejunum did not reach conventional levels of statistical significance. In excluded ileum there was a highly significant fall in DNA content. Again, jejunum and ileum showed significant reductions in RNA content when excluded from normal continuity.

Jejunum in continuity (after distal by-pass) showed only a small increase in DNA and a lesser increase in RNA. By contrast, following proximal by-pass the ileum in continuity showed a three-fold increase in DNA and a rise of similar magnitude in RNA, when compared to controls.

Brush-border enzymes

The results for leucyl β-naphthylamidase, alkaline phosphatase, α-galactosidase and α-glucosidase are shown in Tables 2 and 3. Table 2 shows the results of measurements of leucyl β-naphthylamidase and alkaline phosphatase. The values for leucyl β-naphthylamidase were similar in normal rat jejunum and ileum, but there was a large and significant decrease in alkaline phosphatase activity, whether expressed as units/cm of intestine or as units/mg of DNA, in control ileum as compared with control jejunum.

In terms of units/cm of intestine, excluded small bowel (both jejunum and ileum) showed a similar decrease in both leucyl β-naphthylamidase and alkaline phosphatase. But expressed as units/mg of DNA, only alkaline phosphatase activity in excluded jejunum showed a statistically significant decrease when compared with control intestine.

The intestine in continuity showed marked increases in both enzyme activities (expressed as units/cm of intestine) and again the increase in included ileal segments after by-pass was greater than that in included jejunum. The changes in activity of the enzymes relative to the DNA content do not show a consistent pattern.

Table 3 shows the data for β-galactosidase and α-glucosidase. There was no significant difference between normal rat jejunum and ileum in the total activity of maltase or its activity relative to the DNA content. β-Galactosidase activity, however, was lower whether expressed as units/cm of intestine or in units/mg of DNA in normal ileum, compared with jejunum.

β-Galactosidase was the only enzyme studied which did not show a highly significant increase in activity, when expressed as units/cm of intestine, in ileum in continuity, or a significant decrease in excluded jejunum. Again, when the activity was expressed relative to DNA content there was no significant difference.

α-Glucosidase activity showed a very similar pattern of change to that seen with leucyl β-naphthylamidase except that the fall in activity (expressed in units/cm of intestine) in excluded ileum did not reach statistical significance. The activity of maltase expressed relative to DNA content was reduced in both by-passed jejunum and ileum, though it was not statistically significant. As before, there was no significant change in activity relative to DNA content in intestine in continuity.

Lysosomal enzymes

The results for aryl sulphatase and β-glucuronidase are given in Figs. 2 and 3. The activity
Table 3. Mucosal $\beta$-galactosidase (lactase) and $\alpha$-glucosidase (maltase) of jejunum and ileum from controls and from animals with experimental by-pass. For details see Table 2.

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<tr>
<td></td>
<td>Excluded (Proximal by-pass)</td>
<td>Control</td>
</tr>
<tr>
<td>Lactase</td>
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<tr>
<td>(units/cm of intestine)</td>
<td>0.06 ± 0.01</td>
<td>0.05 ± 0.005</td>
</tr>
<tr>
<td>(units/mg of DNA)</td>
<td>0.14 ± 0.027</td>
<td>0.12 ± 0.014</td>
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<tr>
<td></td>
<td>$P = 0.748$</td>
<td>$P = 0.513$</td>
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<tr>
<td>Maltase</td>
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<tr>
<td>(units/cm of intestine)</td>
<td>0.58 ± 0.11</td>
<td>0.96 ± 0.11</td>
</tr>
<tr>
<td>(units/mg of DNA)</td>
<td>1.51 ± 0.28</td>
<td>2.25 ± 0.43</td>
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<td>$P = 0.042$</td>
<td>$P = 0.016$</td>
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**Mucosal enzymes after small bowel by-pass**

**β-Glucuronidase (lysosomes)**

Per unit length

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<tr>
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<tr>
<td>B-P</td>
<td>C</td>
<td>IC</td>
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Per mg of DNA

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![Graph showing β-Glucuronidase activity per unit length and per mg of DNA in jejunum and ileum from control rats and after proximal or distal Thiry-Vella by-pass.](image)

**Aryl sulphatase (lysosomes)**

Per unit length

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Per mg of DNA

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![Graph showing Aryl sulphatase activity per unit length and per mg of DNA in jejunum and ileum from control rats and after proximal or distal Thiry-Vella by-pass.](image)

**Fig. 2.** β-Glucuronidase activity per unit length of intestine (units/cm) and per unit cell (units/mg of DNA) in jejunum and ileum from control rats and after proximal or distal Thiry-Vella by-pass. (Abbreviations as in Fig. 1.)

**Fig. 3.** Aryl sulphatase activity per unit length of intestine (units/cm) and per unit cell (units/mg of DNA) in jejunum and ileum from control rats and after proximal or distal Thiry-Vella by-pass. (Abbreviations as in Fig. 1.)
### Table 4. Mucosal cytochrome oxidase and catalase of jejunum and ileum from controls and from animals with experimental by-pass.

For details see Table 2.

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<td></td>
<td>Excluded</td>
<td>In continuity</td>
</tr>
<tr>
<td></td>
<td>(Proximal by-pass)</td>
<td>Control</td>
</tr>
<tr>
<td>Cytochrome oxidase (units/cm of intestine)</td>
<td>2.2 ± 0.30</td>
<td>3.8 ± 0.32</td>
</tr>
<tr>
<td></td>
<td>(P = 0.005)</td>
<td>(P = 0.17)</td>
</tr>
<tr>
<td>Cytochrome oxidase (units/mg of DNA)</td>
<td>5.8 ± 0.78</td>
<td>6.7 ± 1.44</td>
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<td>(P = 0.015)</td>
<td>(P = 0.17)</td>
</tr>
<tr>
<td>Catalase (units/cm of intestine)</td>
<td>1.5 ± 0.18</td>
<td>2.5 ± 0.27</td>
</tr>
<tr>
<td></td>
<td>(P = 0.012)</td>
<td>(P = 0.015)</td>
</tr>
<tr>
<td>Catalase (units/mg of DNA)</td>
<td>3.9 ± 0.51</td>
<td>5.0 ± 0.72</td>
</tr>
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<td>(P = 0.033)</td>
<td>(P = 0.033)</td>
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of both lysosomal enzymes was higher in control ileum than in control jejunum, both in terms of units/cm of intestine and also in units/mg of DNA.

The pattern of change in by-passed small bowel and in remaining segments in continuity was similar to that found for the brush border enzymes (Tables 2 and 3). However, it is interesting that the fall in enzyme activity (units/cm of intestine) was not as marked in excluded jejunum as in excluded ileum. Similarly, the increase in jejunum in continuity was much less striking than that in ileum in continuity. Again, there were no consistent changes in activity of the enzymes relative to the DNA content. However, it is noteworthy, particularly in the ileum, that although enzyme activity/unit length of intestine increased from by-passed ileum to control ileum to ileum in continuity, there was actually a decrease in activity/mg of DNA.

Mitochondrial and peroxisomal enzymes

The results for cytochrome oxidase and catalase are given in Table 4. There were no significant differences in normal jejunum and ileum in the activities of these enzymes when expressed as units/cm of intestine or units/mg of DNA.

Both enzymes showed significant increases in terms of units/cm of ileum in continuity, but cytochrome oxidase showed only a small rise in activity and catalase activity did not change in jejunum in continuity. Cytochrome oxidase activity decreased significantly in by-passed intestine, but the decrease in units of catalase activity/cm of intestine was less marked. There was no consistent pattern of change in the activities of the enzymes relative to the DNA content.

Overall it is noteworthy that there was a small but consistent pattern of decrease in the activity, relative to DNA content of all the enzymes studied in ileum in continuity.

DISCUSSION

The results show that the reduced absorptive capacity seen after small bowel by-pass in the rat (Gleeson et al., 1972) is accompanied by biochemical changes in the small bowel mucosa. Further, if one assumes that the 'compensatory' hypertrophy and enhanced absorption seen in the residual small bowel after resection are similar to the changes in intestine in continuity found after by-pass, the present findings also provide an explanation for the compensatory phenomena found after small bowel resection (Booth et al., 1959; Dowling & Booth, 1967).

The enzymic activities of the small intestine are expressed as units/cm of intestine to reflect the digestive function of the small bowel in the control and experimental situations, and as units/mg of DNA to measure the amount of enzyme/cell.

The present study also provides comparative results on normal enzyme activity in proximal and distal small bowel. The comparison of normal jejunal and ileal β-galactosidase activities confirms observations of higher β-galactosidase activity in normal rat jejunum compared with ileum (Dahlqvist & Thomson, 1964). Similarly, there is higher alkaline phosphatase activity in the normal jejunum than in ileum (Triantaphyllopoulos & Tuba, 1959). Conversely, the activities of the lysosomal aryl sulphatase and β-glucuronidase were higher in normal rat ileum than jejunum, as for aryl sulphatase in the human foetal small intestine (Heringova, Koldovska, Yaffe, Jirsova & Uher, 1969).

Although not statistically significant, the higher content of cytochrome oxidase activity in jejum than ileum is consistent with the findings of Wilson & Wiseman (1954) and of
Bamford & Holmes (1971) who noted a greater degree of oxygen uptake and aerobic glycolysis in jejunum than in ileum.

In segments of excluded intestine there was a fall in total enzyme content (units/cm intestine) but there was no consistent change in activity/mg of DNA. As there was a decrease in DNA content (mg/cm of intestine) there must, therefore, have been a reduction in total number of cells (hypoplasia). Such a process would be expected to affect small bowel function, and studies by Riecken, Menge, Bloch & Schaumlöffel (1970) and Gleeson et al. (1972) confirmed that there is a decreased absorption of glucose in by-passed jejunum. Further, changes in cell turnover might also be expected in hypoplastic intestine and preliminary observations by radioautography show that cell turnover is decreased (Gleeson et al., 1972).

In segments of intestine remaining in continuity after by-pass, the DNA content/cm of intestine was increased, indicating an increased number of cells. In small bowel resection Skala & Konradova (1969) found no enlargement either in individual epithelial cells or in the microvilli of the residual small bowel mucosa. Hyperplasia, therefore, is a more accurate term than hypertrophy, which has been used to describe the macroscopic and villous changes seen in the residual intestine following resection (Loran & Althausen, 1958; Booth et al., 1959; Dowling & Booth, 1967).

Hyperplasia was particularly evident in ileum in continuity after a proximal by-pass where there were striking increases in the amounts of DNA (mg/cm of intestine). The magnitude of the increase in DNA content was proportionately greater than the increases in activity of any of the enzymes measured. Consequently, although the enzyme activity/unit length increased, there was a slight but consistent decrease in the activity/mg of DNA of the enzymes in hyperplastic ileum. As ultrastructural studies have shown (Skala & Konradova, 1969), these functional changes were accompanied by decreased differentiation in cell morphology. However, as there is an increased epithelial cell migration rate in residual small intestine after resection (Loran & Althausen, 1960), one may postulate that this leads to a less-mature cell with an accompanying decrease in enzyme content. This concept is supported by the results of Weser & Hernandez (1971) who showed that the net transport of glucose and leucine per cell was decreased in the distal remnant after small bowel resection. In jejunum in continuity after a distal by-pass, the rises in enzyme activities were similar to the change of DNA content; thus no uniform change in activity/mg of DNA was apparent.

Dowling & Booth (1967) showed that after small intestinal resection in the rat, the jejunum and ileum differ in their capacity to develop villous 'hypertrophy' and to increase their absorptive capacity for glucose. After proximal resection the ileum showed a marked increase in villous height and glucose absorption, but after distal resection the jejunum showed less-marked structural and functional increases. The present results demonstrate similar differences in the magnitude of response between jejunum and ileum after small bowel by-pass. For all the variables studied, when expressed as units/cm of intestine (except β-galactosidase) the values were approximately twice as great in ileal segments after proximal by-pass as in the jejunum which remained in continuity after exclusion of the distal small bowel. Ileum, therefore, has a greater adaptive capacity than does jejunum.

Interestingly lactase activity did not show the same pattern of increase in ileum in continuity after proximal by-pass. This may be because lactase activity is normally much lower in ileum than in jejunum. If lactase activity may be induced in response to dietary lactose, as suggested by Fischer (1957), Rosensweig & Herman (1968) and Bolin, Pirola & Davis (1969),
then little change in ileal lactase content after proximal by-pass would be expected if the diet
did not contain significant amounts of lactose. In fact, the rat chow in these experiments
contained only 3% dried skimmed milk. However, the lactase assay may measure other
galactosidases within the cell and the observed changes may not solely reflect brush-border
enzyme activity.

The other brush-border and lysosomal enzymes also showed consistent and significant
increases in enzyme activity in both jejunal and ileal segments which remained in continuity
after by-pass, so that the digestive activity/unit length of intestine was increased.

Previous histochemical studies failed to demonstrate any change in enzyme activity in the
residual jejunum after distal resection (Dowling, 1968). At first sight, this observation appears
to conflict with the increased enzyme activity found in the present study. However, since there
was no change in the enzyme content of individual cells, the intensity of staining reactions
(at best a semi-quantitative technique) would not be expected to change since the staining
reaction is related more to activity/cell than to amount/unit length of intestine.

The association of hypoplasia in one segment of intestine by-passed from normal con-
tinuity, with simultaneous hyperplasia in another segment of small bowel in the same animal,
is interesting. It provides further evidence to support the suggestion that intraluminal factors
are important in regulating small-intestinal cell turnover. These factors may be some component
of the animal's diet, as implied in the concept of 'intraluminal nutrition of small-intestinal
epithelium' (Smyth, 1962; Dowling & Booth, 1967), or may be related to intraluminal pH
(Altmann & Leblond, 1970) or even some component of gastric biliary or pancreatic secretions.
These observations do not support the concept of a circulating 'intestinal epithelial hormone'
to explain the hyperplasia in the residual small bowel after resection or by-pass (Loran &
Althausen, 1960). If such a hormone were present, one would expect hyperplasia in the excluded
segment as well. However, it is possible that if this hormone exists, it may require an additional
intraluminal factor for its action.

Clearly further studies are necessary before assuming that the concept of 'intraluminal
nutrition' is fully validated, but the present study suggests that the intraluminal environment
must play a fundamental role in the regulation of mucosal cell turnover which, in turn, is
reflected by changes in the metabolic and digestive activities of the enterocyte.

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