Stimulation by 2-deoxy-\textit{D}-glucose tetraacetates of hormonal secretion from the perfused rat pancreas

VIVIANE LECLERCQ-MEYER, MARCEL M. KADIATA, AND WILLY J. MALAISSE
Laboratory of Experimental Medicine, Brussels Free University, B-1070 Brussels, Belgium

Leclercq-Meyer, Viviane, Marcel M. Kadiata, and Willy J. Malaisse. Stimulation by 2-deoxy-\textit{D}-glucose tetraacetates of hormonal secretion from the perfused rat pancreas. Am. J. Physiol. 276 (Endocrinol. Metab. 39): E689–E696, 1999.—The effects of \( \alpha \)- and \( \beta \)-2-deoxy-\textit{D}-glucose tetraacetate (1.7 and 8.5 mM) on insulin, somatostatin, and glucagon secretion from isolated rat pancreases perfused in the presence of 8.3 mM \textit{D}-glucose were compared with those of unesterified 2-deoxy-\textit{D}-glucose tested at the same two concentrations. The unesterified glucose analog caused, in a concentration-related manner, inhibition of glucose-induced insulin and somatostatin release and augmentation of glucagon secretion. The two anomers of 2-deoxy-\textit{D}-glucose tetraacetate, however, increased the secretion rate of all three hormones; this effect was also related to the concentration of the esters. No obvious anomic specificity of the secretory response to 2-deoxy-\textit{D}-glucose tetraacetate was observed. These findings indicate that the insulinotropic action of hexose esters cannot be accounted for solely by the metabolic effect of their glucidic moieties. They suggest that the A, B, and D cells of the endocrine pancreas are each equipped with a receptor system responsible for the direct recognition of monosaccharide esters as secretagogues. They further support the view that a paracrine effect of insulin on glucagon-producing cells does not represent a major component in the regulation of their secretory activity.

insulin secretion; glucagon secretion; somatostatin secretion; rat pancreas perfusion

THE RECENT INTRODUCTION of selected esters of monosaccharides in biomedical research has allowed an increase in the nutritional value or metabolic efficiency of several hexoses and their antimetabolic analogs (10). For instance, \( \alpha \)-\textit{D}-glucose pentaacetate augments glycyllic flux and insulin release in rat pancreatic islets to a larger extent than unesterified \textit{D}-glucose tested at the same concentration as its ester (21, 27). Likewise, \textit{D}-mannohexulose hexaacetate inhibits \textit{D}-glucose phosphorylation in different cell types otherwise resistant to the unesterified heptose (4, 17). Last, 2-deoxy-\textit{D}-glucose tetraacetate is more efficient than the unesterified 2-deoxy-\textit{D}-glucose tested at the same two concentrations. The unesterified glucose analog caused, in a concentration-related manner, inhibition of glucose-induced insulin and somatostatin release and augmentation of glucagon secretion. The two anomers of 2-deoxy-\textit{D}-glucose tetraacetate, however, increased the secretion rate of all three hormones; this effect was also related to the concentration of the esters. No obvious anomic specificity of the secretory response to 2-deoxy-\textit{D}-glucose tetraacetate was observed. These findings indicate that the insulinotropic action of hexose esters cannot be accounted for solely by the metabolic effect of their glucidic moieties. They suggest that the A, B, and D cells of the endocrine pancreas are each equipped with a receptor system responsible for the direct recognition of monosaccharide esters as secretagogues. They further support the view that a paracrine effect of insulin on glucagon-producing cells does not represent a major component in the regulation of their secretory activity.

Materials and Methods

The \( \alpha \)- and \( \beta \)-tetraacetate esters of 2-deoxy-\textit{D}-glucose were synthesized by a method adapted from that described elsewhere (30). 2-Deoxy-\textit{D}-glucose was purchased from Sigma (St. Louis, MO).

Two female (B & K Universal, Hull, UK) and 10 male (Iffa Credo, L'Arsbrele, France) fed Wistar rats were used in the present study (Table 1). The animals were anesthetized with an intraperitoneal injection of pentobarbital sodium (46 mg/kg), and the pancreas was perfused without recirculation through both the celiac and superior mesenteric arteries, as described elsewhere (18). A slight modification of this procedure was introduced. The duodenum was not excluded during the surgical procedure, and the secretions from the intestine were collected in tubes placed into a cold bath. The perfusate was discarded at the end of the experiment. The perfusate was collected at the end of the experiment and the date to indicate this fact.

Leclercq-Meyer, Viviane, Marcel M. Kadiata, and Willy J. Malaisse. Stimulation by 2-deoxy-\textit{D}-glucose tetraacetates of hormonal secretion from the perfused rat pancreas. Am. J. Physiol. 276 (Endocrinol. Metab. 39): E689–E696, 1999.—The effects of \( \alpha \)- and \( \beta \)-2-deoxy-\textit{D}-glucose tetraacetate (1.7 and 8.5 mM) on insulin, somatostatin, and glucagon secretion from isolated rat pancreases perfused in the presence of 8.3 mM \textit{D}-glucose were compared with those of unesterified 2-deoxy-\textit{D}-glucose tested at the same two concentrations. The unesterified glucose analog caused, in a concentration-related manner, inhibition of glucose-induced insulin and somatostatin release and augmentation of glucagon secretion. The two anomers of 2-deoxy-\textit{D}-glucose tetraacetate, however, increased the secretion rate of all three hormones; this effect was also related to the concentration of the esters. No obvious anomic specificity of the secretory response to 2-deoxy-\textit{D}-glucose tetraacetate was observed. These findings indicate that the insulinotropic action of hexose esters cannot be accounted for solely by the metabolic effect of their glucidic moieties. They suggest that the A, B, and D cells of the endocrine pancreas are each equipped with a receptor system responsible for the direct recognition of monosaccharide esters as secretagogues. They further support the view that a paracrine effect of insulin on glucagon-producing cells does not represent a major component in the regulation of their secretory activity.

Materials and Methods

The \( \alpha \)- and \( \beta \)-tetraacetate esters of 2-deoxy-\textit{D}-glucose were synthesized by a method adapted from that described elsewhere (30). 2-Deoxy-\textit{D}-glucose was purchased from Sigma (St. Louis, MO).

Two female (B & K Universal, Hull, UK) and 10 male (Iffa Credo, L’Arbrésle, France) fed Wistar rats were used in the present study (Table 1). The animals were anesthetized with an intraperitoneal injection of pentobarbital sodium (46 mg/kg), and the pancreas was perfused without recirculation through both the celiac and superior mesenteric arteries, as described elsewhere (18). A slight modification of this procedure was introduced. The duodenum was not excluded during the surgical procedure, and the secretions from the intestine were collected in tubes placed into a cold bath. The perfusate was discarded at the end of the experiment. The perfusate was collected at the end of the experiment.
and exocrine pancreas were diverted through a plastic tubing that was secured in the upper part of the duodenum. The basal salt-balanced solution (18) contained d-glucose (8.3 mM), dextran (clinical grade; 40 g/l; Sigma), and bovine serum albumin (RIA grade; 5 g/l; Sigma). It was supplemented, as required, with 2-deoxy-D-glucose or its tetraacetate esters (1.7 and 8.5 mM), with separate reservoirs. All solutions were continuously gassed (95% O2-5% CO2), with an extended scale (see Fig. 4). The results were not statistically different from one another (ANOVA, P = 0.896) and, when pooled, averaged 8 ± 1 ng/min (n = 12).

At similar times, the output of somatostatin amounted to 20 ± 6, 14 ± 6, and 16 ± 7 pg/min, respectively (Fig. 2; see Fig. 4, middle; Table 1). Again, these secretory rates were comparable (ANOVA, P = 0.798) and, when pooled, averaged 16 ± 3 pg/min (n = 12).

Concomitantly, the output of glucagon amounted to 95 ± 6, 80 ± 5, and 75 ± 6 pg/min, respectively (Fig. 3; Fig. 4, bottom; Table 1). These secretory rates were not statistically different (ANOVA, P = 0.105) and, when pooled, averaged 86 ± 4 pg/min (n = 12).

Effects of α-2-deoxy-D-glucose tetraacetate, β-2-deoxy-D-glucose tetraacetate, and 2-deoxy-D-glucose on insulin release. Both the α- and β-2-deoxy-D-glucose tetraacetate esters stimulated, in a concentration-dependent manner, the release of insulin (Fig. 1, top and middle; Table 1).

In response to 1.7 mM α-2-deoxy-D-glucose tetraacetate (Fig. 1, top), the output of insulin increased to a mean value of 28 ± 3 ng/min (minutes 28-43), which represented 310 ± 13% of that recorded during the 5-min prestimulatory period (minutes 23-28, 9 ± 1 ng/min). The pattern of release disclosed regular

### Table 1. Hormonal release from perfused rat pancreas at selected times of the experiments

<table>
<thead>
<tr>
<th></th>
<th>α-DGTA</th>
<th>β-DGTA</th>
<th>DOG</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Insulin release</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Basal period (min 20-28)</td>
<td>9 ± 1</td>
<td>8 ± 2</td>
<td>8 ± 2</td>
</tr>
<tr>
<td>1.7 mM stimulation period (min 28-43)</td>
<td>28 ± 3</td>
<td>21 ± 3</td>
<td>5 ± 2</td>
</tr>
<tr>
<td>8.5 mM stimulation period (min 68-83)</td>
<td>89 ± 6</td>
<td>109 ± 20</td>
<td>2 ± 1</td>
</tr>
<tr>
<td>End of experiments (min 100-105)</td>
<td>17 ± 2</td>
<td>39 ± 12</td>
<td>8 ± 2</td>
</tr>
<tr>
<td><strong>Glucagon release</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Basal period (min 20-28)</td>
<td>20 ± 6</td>
<td>14 ± 6</td>
<td>16 ± 7</td>
</tr>
<tr>
<td>1.7 mM stimulation period (min 28-43)</td>
<td>44 ± 10</td>
<td>41 ± 9</td>
<td>10 ± 4</td>
</tr>
<tr>
<td>8.5 mM stimulation period (min 68-83)</td>
<td>87 ± 6</td>
<td>85 ± 5</td>
<td>4 ± 1</td>
</tr>
<tr>
<td>End of experiments (min 100-105)</td>
<td>13 ± 5</td>
<td>76 ± 25</td>
<td>11 ± 5</td>
</tr>
<tr>
<td><strong>Somatostatin release</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Basal period (min 20-28)</td>
<td>89 ± 6</td>
<td>95 ± 6</td>
<td>75 ± 6</td>
</tr>
<tr>
<td>1.7 mM stimulation period (min 28-43)</td>
<td>107 ± 12</td>
<td>90 ± 7</td>
<td>72 ± 8</td>
</tr>
<tr>
<td>8.5 mM stimulation period (min 68-83)</td>
<td>365 ± 46</td>
<td>405 ± 82</td>
<td>69 ± 7</td>
</tr>
<tr>
<td>End of experiments (min 100-105)</td>
<td>59 ± 3</td>
<td>64 ± 3</td>
<td>53 ± 8</td>
</tr>
</tbody>
</table>

Values are means ± SE for each series of experiments; n = 4 rats. α-DGTA, α-2-deoxy-D-glucose tetraacetate; β-DGTA, β-2-deoxy-D-glucose tetraacetate; DOG, 2-deoxy-D-glucose.
oscillations. Thus, after a first peak at minutes 33-34, two further secretory cycles of 4.9 ± 0.4 min each were identified. During the late part of the stimulatory period (minutes 35-43), the output of insulin averaged 32 ± 3 ng/min. The later administration of 8.5 mM α-2-deoxy-α-glucose tetraacetate induced a rapid and prominent stimulation of insulin release, the secretory rates reaching a value of 89 ± 6 ng/min (minutes 68-83), which represented 686 ± 82% of that recorded during the immediate 5-min prestimulatory period (minutes 63-68, 13 ± 1 ng/min). As was the case with the 1.7 mM concentration, the pattern of release ob-

![Fig. 1. Insulin responses to 1.7 and 8.5 mM α-2-deoxy-α-glucose tetraacetate (α-DOGTA, top), β-2-deoxy-α-glucose tetraacetate (β-DOGTA, middle), and 2-deoxy-α-glucose (DOG, bottom) from rat pancreas perfused in presence of 8.3 mM D-glucose. Values are means ± SE and refer to 4 individual experiments. Vertical lines show correction for dead space of perfusion device.](http://ajpendo.physiology.org/)

![Fig. 2. Somatostatin responses to 1.7 and 8.5 mM α-2-deoxy-α-glucose tetraacetate (top), β-2-deoxy-α-glucose tetraacetate (middle), and 2-deoxy-α-glucose (bottom) from rat pancreas perfused in presence of 8.3 mM D-glucose. Presentation as in Fig. 1.](http://ajpendo.physiology.org/)

served in response to 8.5 mM α-2-deoxy-α-glucose tetraacetate was oscillatory, three secretory cycles of 4.6 ± 0.2 min being identified after the initial peak at minutes 70-71. The output of insulin averaged 102 ± 6 ng/min at the end of the stimulatory period (minutes 75-83). The stimulatory effects of 1.7 and 8.5 mM α-2-deoxy-α-glucose tetraacetate were both rapidly reversible, the insulin secretory rates returning to 12 ± 3 ng/min during the interstimulatory period (minutes 50-68) and 17 ± 2 ng/min at the end of the experiments (minutes 100-105). Although higher than those seen in the early basal period, the later secretory rates were in the range of those that normally may be expected during a constant exposure to 8.3 mM D-glucose.
In response to 1.7 mM \( \beta \)-2-deoxy-D-glucose tetraacetate (Fig. 1, middle), the output of insulin increased to a value of 21 ± 3 ng/min (minutes 28-43), which represented 278 ± 42% relative to the 5-min prestimulatory period (minutes 23-28, 8 ± 2 ng/min). The secretory pattern, as in the case of 1.7 mM \( \alpha \)-2-deoxy-D-glucose tetraacetate, disclosed oscillations, two secretory cycles of 4.6 ± 0.4 min being observed after the first peak value at minutes 33-34. The release of insulin reached a mean value of 24 ± 3 ng/min during the late part of the stimulatory period. The stimulatory effect of 1.7 mM \( \beta \)-2-deoxy-D-glucose tetraacetate, as that of 1.7 mM \( \alpha \)-2-deoxy-D-glucose tetraacetate, was readily reversible, the output of insulin decreasing to 12 ± 3 ng/min during the interstimulatory period (minutes 50-68). At the higher 8.5 mM concentration, \( \beta \)-2-deoxy-D-glucose tetraacetate, similarly to 8.5 mM \( \alpha \)-2-deoxy-D-glucose tetraacetate, provoked a rapid and prominent stimulation of insulin release to a value of 109 ± 20 ng/min (minutes 68-83), which represented 925 ± 194% of the immediate 5-min prestimulatory period (minutes 63-68, 8 ± 2 ng/min). However, in contrast to the pattern seen with 8.5 mM \( \alpha \)-2-deoxy-D-glucose tetraacetate, the secretory rates progressively increased throughout the administration of 8.5 mM \( \beta \)-2-deoxy-D-glucose tetraacetate, reaching the value of 132 ± 25 ng/min at the end of the stimulatory period (minutes 75-83). An oscillatory pattern was still identified in

Fig. 3. Glucagon responses to 1.7 and 8.5 mM \( \alpha \)-2-deoxy-D-glucose tetraacetate (top), \( \beta \)-2-deoxy-D-glucose tetraacetate (middle), and 2-deoxy-D-glucose (bottom) from rat pancreas perfused in presence of 8.3 mM D-glucose. Presentation as in Fig. 1.

Fig. 4. Effects of 1.7 and 8.5 mM 2-deoxy-D-glucose on insulin (top), somatostatin (middle), and glucagon release (bottom) from rat pancreas perfused in presence of 8.3 mM D-glucose. Results (drawn on an enhanced scale) represent those reported in Figs. 1–3, bottom.
three out of four individual experiments, with cycles of 4.5 ± 0.3 min after the first secretory peak at minute 71.7 ± 0.7. There was a tendency for a slower reversibility in the experiments conducted with 8.5 mM β-2-deoxy-D-glucose tetraacetate, a higher rate of insulin release (39 ± 12 ng/min) being recorded at the end of the perfusions (minutes 100-105) in the former experiments. This difference, however, was not statistically significant, and the observed secretory pattern was merely related to the very high insulin output recorded in response to 8.5 mM β-2-deoxy-D-glucose tetraacetate in one of the four perfusions performed. As a whole, whether expressed in terms of absolute secretory rates or percentages, there was no statistically significant difference between the insulin stimulatory effects of α- and β-2-deoxy-D-glucose tetraacetate, whether at 1.7 or 8.5 mM. The mean 8.5 mM-to-1.7 mM ratio in insulin output (ng/min, minutes 68-83 and minutes 28-43) was somewhat higher, albeit not significantly so, in the β- (5.3 ± 0.9) compared with α-2-deoxy-D-glucose tetraacetate experiments (3.2 ± 0.2).

At variance with its esters, 2-deoxy-D-glucose dose dependently inhibited the secretion of insulin (Fig. 1, bottom; Fig. 4, top; Table 1). The B cells appeared exquisitely sensitive to 2-deoxy-D-glucose because, already at the low 1.7 mM concentration, this glucose analog decreased the 5-min prestimulatory insulin secretory rates (minutes 23-28, 8 ± 2 ng/min) to a value of 5 ± 2 ng/min (minutes 28-43, P < 0.01). At the concentration of 8.5 mM, 2-deoxy-D-glucose decreased the immediately preceding 5-min prestimulatory insulin secretory rates (minutes 63-68, 9 ± 3 ng/min) to the value of 2 ± 1 ng/min (minutes 68-83, P < 0.05). In terms of percentages, the secretion of insulin was inhibited 38 ± 15 and 79 ± 1% by 1.7 and 8.5 mM 2-deoxy-D-glucose, respectively. Those inhibitions were expressed in terms of percentages, the secretion of insulin was inhibited 38 ± 15 and 79 ± 1% by 1.7 and 8.5 mM 2-deoxy-D-glucose, respectively. Those inhibitions were expressed in terms of percentages, the secretion of insulin was inhibited 38 ± 15 and 79 ± 1% by 1.7 and 8.5 mM 2-deoxy-D-glucose, respectively.

The stimulatory effects of α- and β-2-deoxy-D-glucose tetraacetate on somatostatin release were comparable. Thus 1.7 mM α- and β-2-deoxy-D-glucose tetraacetate increased the output of somatostatin to values of 44 ± 10 and 41 ± 9 pg/min (minutes 28-43), respectively, which represented 255 ± 36 and 546 ± 243% of those recorded during the 5-min prestimulatory basal period (minutes 23-28, 19 ± 7 and 13 ± 5 pg/min, respectively). The secretory rates of somatostatin progressively increased during the stimulation, reaching values of 58 ± 11 and 55 ± 10 pg/min at the end of the stimulatory period (minutes 35-43) for the α- and β-esters, respectively. In response to the higher 8.5 mM α- and β-ester concentrations, the output of somatostatin increased to 87 ± 8 and 85 ± 9 pg/min (minutes 68-83), which represented 839 ± 263 and 1,073 ± 405% relative to that recorded during the immediate 5-min prestimulatory period (minutes 63-68, 16 ± 7 and 15 ± 7 pg/min, respectively). The secretory rates stabilized at values of 92 ± 7 and 97 ± 12 pg/min, respectively, at the end of the stimulatory period (minutes 75-83). The 8.5 mM-to-1.7 mM ratios in somatostatin output (pg/min, minutes 68-83 and minutes 28-43) were comparable in the α- and β-2-deoxy-D-glucose tetraacetate experiments, amounting to 2.2 ± 0.3 and 2.3 ± 0.4, respectively. Oscillations in somatostatin output were apparent as in the case of insulin release. Thus, in at least three of four individual experiments, a single secretory cycle of 4.3 ± 0.4 min was observed during exposure to 1.7 mM α- or β-2-deoxy-D-glucose tetraacetate after the first peak at minutes 37-38, whereas two cycles of 4.9 ± 0.2 min were identified during administration of 8.5 mM α- or β-2-deoxy-D-glucose tetraacetate after a more rapid first peak at minutes 71-72, i.e., 6 min earlier than in response to the low concentration (1.7 mM) of the same esters. However, some differences in the secretory patterns of somatostatin and insulin were noteworthy. First, the rates of somatostatin release, in contrast to those of insulin, rose steadily in the presence of the low, but not high, concentration of the esters. Second, the 8.5 mM-to-1.7 mM ratios in output were, for both the α- and β-2-deoxy-D-glucose tetraacetate esters, lower in the case of somatostatin than in that of insulin (P < 0.05). Last, a poor reversibility of the secretory responses was noted upon the arrest of the high 8.5 mM α- or β-2-deoxy-D-glucose tetraacetate infusions, an "off response" even being observed in three of the four experiments performed with the β-ester. As a result, the secretory rates at the end of the perfusions performed with β-2-deoxy-D-glucose tetraacetate remained at a value of 76 ± 25 pg/min (minutes 100-105), which was significantly higher (P < 0.05) than that seen at a comparable time in the α-2-deoxy-D-glucose tetraacetate experiments.

As was the case for insulin, 2-deoxy-D-glucose dose dependently inhibited the secretion of somatostatin (Fig. 2, bottom; Fig. 4, middle; Table 1). Thus the concentration of 1.7 mM decreased the 5-min prestimulatory somatostatin secretory rates (minutes 23-28, 16 ± 7 pg/min) to 10 ± 4 pg/min (minutes 68-83). In response to 8.5 mM 2-deoxy-D-glucose, the 5-min prestimulatory secretory rates (minutes 63-68, 15 ± 7 pg/min) were reduced to 4 ± 1 pg/min (minutes 68-83). Such inhibitions amounted to 42 ± 7 and 61 ± 11%, respectively. Regular oscillations in secretory rates
were present, which persisted in the presence of 2-deoxy-D-glucose. Thus two secretory cycles of 4.8 ± 0.6 min were observed at the low concentration of 2-deoxy-D-glucose (1.7 mM) after the first nadir recorded at minute 33.3 ± 0.9, and one to two secretory cycles of 4.0 ± 0.6 min were identified at the high concentration of 2-deoxy-D-glucose (8.5 mM) after the initial nadir at minute 74.0 ± 0.4. The secretory pattern of somatostatin differed from that of insulin, mainly in that the reversal from inhibition, upon the arrest of either the 1.7 or 8.5 mM 2-deoxy-D-glucose infusions, was not accompanied by any early phase of somatostatin secretion.

Effects of α-2-deoxy-D-glucose tetraacetate, β-2-deoxy-D-glucose tetraacetate, and 2-deoxy-D-glucose on glucagon release. Both α- and β-2-deoxy-D-glucose tetraacetate stimulated the secretion of glucagon, but such a stimulation was only prominent in the presence of the higher 8.5 mM concentration of the esters (Fig. 3, top and middle; Table 1).

Thus the low 1.7 mM concentration of α-2-deoxy-D-glucose tetraacetate increased the output of glucagon to a value of 107 ± 12 pg/min (minutes 28-43), which only represented 123 ± 10% relative to that recorded during the 5-min prestimulatory period (minutes 23-28, 87 ± 8 pg/min). The 1.7 mM concentration of β-2-deoxy-D-glucose tetraacetate did not influence the secretion of glucagon, the secretory rates of 90 ± 7 pg/min (minutes 28-43) being comparable with those recorded during the 5-min prestimulatory period (minutes 23-28, 94 ± 6 pg/min). The glucagon responses to the 8.5 mM concentration of α- and β-2-deoxy-D-glucose tetraacetate were comparable, the secretory rates being increased to values of 365 ± 46 and 405 ± 82 pg/min (minutes 28-43), respectively, which represented 588 ± 100 and 670 ± 158% relative to those recorded during the immediate 5-min prestimulatory period (minutes 63-68, 64 ± 4 and 63 ± 4 pg/min, respectively). The secretory pattern of glucagon release was, to some extent, comparable with that of insulin. Thus the secretory rates at the end of the 8.5 mM stimulatory period stabilized at a value of 471 ± 69 pg/min in the presence of α-2-deoxy-D-glucose tetraacetate, whereas they steadily increased to a value of 575 ± 135 ng/min in the presence of β-2-deoxy-D-glucose tetraacetate (minutes 75-83). Also, the 8.5 mM-to-1.7 mM ratios in glucagon output, which amounted to 3.5 ± 0.6 and 4.6 ± 0.9 in the α- and β-2-deoxy-D-glucose tetraacetate experiments (pg/min, minutes 68-83 and minutes 28-43), respectively, were comparable with those obtained in the case of insulin. However, the glucagon release pattern differed in two respects from both the insulin and somatostatin patterns. First, the onset of the response to 8.5 mM α- and β-2-deoxy-D-glucose tetraacetate was clearly slower in the case of glucagon compared with that of insulin or somatostatin. Second, the stimulatory effects of both α- and β-2-deoxy-D-glucose tetraacetate were readily and entirely reversed upon the arrest of the 8.5 mM ester infusion.

Depending on its concentration, 2-deoxy-D-glucose either did not influence or did stimulate the secretion of glucagon (Figs. 3 and 4, bottom; Table 1). Thus, in the presence of 1.7 mM 2-deoxy-D-glucose, the output of glucagon amounted to a value of 72 ± 8 pg/min (minutes 28-43), which was comparable with that recorded during the 5-min prestimulatory period (minutes 23-28, 74 ± 6 pg/min). Upon the administration of 8.5 mM 2-deoxy-D-glucose, the secretion of glucagon was significantly increased to values of 69 ± 7 (minutes 68-83) and 75 ± 7 pg/min (minutes 75-83), which represented 114 ± 2 and 123 ± 3% relative to that recorded during the immediate 5-min prestimulatory period (minutes 63-68, 61 ± 7 pg/min, P < 0.01). The stimulation of glucagon release was rapidly and fully reversed upon the arrest of the 8.5 mM 2-deoxy-D-glucose infusion.

Effects of α-2-deoxy-D-glucose tetraacetate, β-2-deoxy-D-glucose tetraacetate, and 2-deoxy-D-glucose on perfusion pressure. α- And β-2-deoxy-D-glucose tetraacetate at 1.7 mM and 2-deoxy-D-glucose did not modify the perfusion pressure (data not shown). At the concentration of 8.5 mM, α- and β-2-deoxy-D-glucose tetraacetate induced a transient increase in perfusion pressure. The increase was comparable for the α- and β-esters and, at its peak value at minute 74, did not exceed 1.4 ± 0.3 and 1.1 ± 0.6 mmHg, respectively.

**DISCUSSION**

The present results confirm prior observations on a concentration-related inhibitory action of 2-deoxy-D-glucose on glucose-stimulated insulin secretion (20, 21). This suppressing action coincides with and is probably attributable to inhibition of D-glucose metabolism in pancreatic islet cells (27). Our results also document that unesterified 2-deoxy-D-glucose inhibits somatostatin secretion and enhances glucagon release by isolated perfused rat pancreases exposed to 8.3 mM D-glucose, and these effects are concentration related. A partial relief by 2-deoxy-D-glucose from the inhibitory action of D-glucose on glucagon release was already reported in either rat pancreatic islets exposed to 10 mM L-arginine (3) or the isolated perfused rat pancreas (31). To our knowledge, no information is available on the effect of 2-deoxy-D-glucose on somatostatin release. The present results are compatible with the view that D-glucose metabolism stimulates hormonal release from both B and D cells, while it inhibits glucagon secretion from A cells (2, 22, 23).

Both the α- and β-anomer of 2-deoxy-D-glucose tetraacetate, however, augmented insulin, somatostatin, and glucagon secretion from the pancreases exposed to 8.3 mM D-glucose. This positive tropic action of the tetraacetate esters failed to display any obvious anomeric specificity and was, in all cases, more pronounced at a high (8.5 mM), rather than a low (1.7 mM) concentration of the esters. Under vastly different experimental conditions, namely in isolated pancreatic islets concomitantly, but not sequentially, exposed throughout a prolonged incubation of 90 min to 8.3 mM D-glucose and 2-deoxy-D-glucose tetraacetate, the ester was found to enhance insulin release, when tested at a low concentration of 1.7 mM, and to inhibit glucose-
stimulated insulin output, when tested at a much higher concentration of 10.0 mM, both the enhancing and inhibitory action of the ester displaying α-anomeric preference (14).

The apparent disparity between the two series of experiments is probably accounted for, in part at least, by the fact that the rate of 2-deoxy-D-glucose tetraacetate hydrolysis in islet homogenates, which indeed displays preference for the α-anomer, requires prolonged exposure of intact islet cells to the ester to generate an amount of the unesterified D-glucose analog sufficient to inhibit glycolysis (24). The present results are likely, therefore, to refer mainly to the postulated direct action of the esters themselves on hormonal release, thought to be mediated by activation of a specific receptor system.

If so, the present findings suggest that the three major cell types of the endocrine pancreas, despite their different responsiveness to unesterified D-glucose, are all equipped with the previously mentioned receptors for 2-deoxy-D-glucose tetraacetate. Because the latter ester augments glucagon secretion, as well as insulin and somatostatin output, the second messenger(s) generated by such receptors should, at the first glance, belong to those few coupling factors that may exert a comparable positive secretory effect in A, B, and D cells. Alternatively, the binding of 2-deoxy-D-glucose tetraacetate to its receptor could conceivably lead to the production of distinct messengers in these three cell types.

Whatever the identity of such messengers, the present comparison between the effects of 2-deoxy-D-glucose and its ester on insulin, somatostatin, and glucagon release convincingly documents, in our opinion, that the functional response of the endocrine pancreas to monosaccharide esters cannot be fully accounted for by the metabolism of or metabolic response to their carbohydrate moiety. Moreover, the finding that 2-deoxy-D-glucose tetraacetate augmented glucagon release, despite concomitant stimulation of insulin release, provides further support to our contention that the paracrine effect of insulin plays little, if any, role in the regulation of glucagon secretion (8, 28).

The receptor system postulated to be activated by the esters of 2-deoxy-D-glucose remains to be identified. The effects of these esters on insulin, somatostatin, and glucagon release are comparable with those of β-l-glucose pentaacetate in the perfused rat pancreas (7). In the latter case, it was proposed that the stimulation of insulin release may result from the direct interaction of the ester itself with a receptor system similar to that involved in the recognition of bitter compounds by taste buds. Indeed, β-l-glucose pentaacetate displays a bitter taste (19). It causes depolarization of the plasma membrane and resulting spike activity in single isolated rat B cells, in a manner comparable with that documented in taste buds exposed to bitter compounds (11). Moreover, purified islet B cells were recently found to express the α-gustducin G protein involved in the recognition of such bitter compounds by taste buds (J. Rasschaert and W. J. Malaisse, unpublished observations). The β-anomer of L-glucose also increases cytosolic Ca2+ concentration in mouse islets (26). The sequence of cationic and secretory events evoked by β-l-glucose pentaacetate, and presumably other esters of nonmetabolized monosaccharides, in islet B cells is thus reminiscent of that operative in response to stimulation by D-glucose. This analogy could conceivably account for the present finding that the anomers of 2-deoxy-D-glucose tetraacetate did not suppress the oscillatory pattern of insulin release previously documented in rat pancreases perfused in the sole presence of 8.3 mM D-glucose (5).

We are grateful to J. Marchand for technical assistance and C. Demesmaeker for secretarial help.

This study was supported by a Concerted Research Action (94/99–183) of the French Community of Belgium.

Address for correspondence and reprint requests: W. J. Malaisse, Laboratory of Experimental Medicine, Brussels Free University, 808 Route de Lenkin, B-1070 Brussels, Belgium (E-mail: malaisse@med.uib.ac.be).

Received 12 August 1998; accepted in final form 15 December 1998.

REFERENCES


