Experimental Study of Dibutyryl Cyclic AMP; Its Metabolic Effects Observed in Anesthetized Human Subjects

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SUEMORI, I. Experimental Study of Dibutyryl Cyclic AMP; Its Metabolic Effects Observed in Anesthetized Human Subjects. Tohoku J. exp. Med., 1975, 117 (2), 111-118 — Metabolic effects of N°, O°-dibutyryl adenosine 3’,5’-monophosphate (DBcAMP) were studied in 10 anesthetized patients who were divided at random into two groups each consisting of 5 patients. DBcAMP dissolved in 200 ml of physiological saline was administered intravenously at a rate of 10 mg/min for 20 min in one group and 20 mg/min for 20 min in the other group. DBcAMP infusion at either rate increased levels of blood glucose, immunoreactive plasma insulin, blood pyruvate and blood redox-potential while it reduced levels of glycerol, non-esterified fatty acid and inorganic phosphate. These findings suggest that 200 and 400 mg of DBcAMP stimulates glycogenolysis and glycolysis but inhibits lipolysis in man. ——— DBcAMP; glycogenolysis; glycolysis; lipolysis

Since adenosine 3’,5’-monophosphate (cAMP) was discovered as an intracellular mediator of glycogenolysis induced by epinephrine and glucagon in the liver by Sutherland and Rall (1958), it has been generally accepted that this nucleotide was a second messenger mediating a variety of hormonal effects at each target organ. Exogenous cAMP, however, was found to be impermeable in all cell membranes except for liver (Levine et al. 1969), though there is still some controversy (Heersche et al. 1971; Fujii and Okuda 1972). Although dibutyryl cyclic AMP, one of the cAMP derivatives, can penetrate the cell membrane more readily than cAMP does (Henion et al. 1967) and has almost the same biochemical properties in animal experiments, little is known on the metabolic and pharmacological responses of man to DBcAMP. In this study, metabolic effects of DBcAMP were investigated in anesthetized human subjects.

METHODS

Ten adult subjects weighing 42–68 kg who were to undergo a variety of operative procedures were chosen. Patients with any abnormal metabolic condition in preoperative

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The following abbreviations are used: cAMP, adenosine 3’,5’-monophosphate; DBcAMP, N°, O°-dibutyryl cAMP; NEFA, nonesterified fatty acid; Pi, inorganic phosphorus.
TABLE 1. Description of subjects

<table>
<thead>
<tr>
<th>Sex</th>
<th>Age (kg)</th>
<th>Diagnosis</th>
<th>Operation</th>
</tr>
</thead>
<tbody>
<tr>
<td>DBcAMP 200 mg infusion group</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>M.Y.</td>
<td>F</td>
<td>36</td>
<td>Cervical cancer</td>
</tr>
<tr>
<td>U.T.</td>
<td>F</td>
<td>58</td>
<td>Cervical cancer</td>
</tr>
<tr>
<td>K.T.</td>
<td>M</td>
<td>25</td>
<td>Buerger’s disease</td>
</tr>
<tr>
<td>H.S.</td>
<td>M</td>
<td>17</td>
<td>Buerger’s disease</td>
</tr>
<tr>
<td>A.R.</td>
<td>F</td>
<td>52</td>
<td>Breast cancer</td>
</tr>
<tr>
<td>DBcAMP 400 mg infusion group</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>M.A.</td>
<td>F</td>
<td>44</td>
<td>Uterine myoma</td>
</tr>
<tr>
<td>T.K.</td>
<td>F</td>
<td>40</td>
<td>Uterine myoma</td>
</tr>
<tr>
<td>S.S.</td>
<td>M</td>
<td>49</td>
<td>Gastric cancer</td>
</tr>
<tr>
<td>Y.H.</td>
<td>F</td>
<td>55</td>
<td>Breast cancer</td>
</tr>
<tr>
<td>H.F.</td>
<td>F</td>
<td>43</td>
<td>Uterine myoma</td>
</tr>
</tbody>
</table>

DBcAMP infusion caused a significant elevation of blood glucose level, which reached the maximum between 20 and 40 min after termination of infusion in both groups (Fig. 1). The maximum blood glucose level was slightly higher but not significant with the higher doses of DBcAMP (197 mg/100 ml following 400 mg infusion and 189 mg/100 ml following 200 mg infusion). Then the blood glucose level started to fall, but did not reach the preinfusion level even 120 min after infusion. Plasma insulin (IRI) rose sharply by the DBcAMP infusion and reached the maximum level at the end of infusion in both groups, which declined somewhat
Fig. 1. Changes in blood glucose, plasma insulin and inorganic phosphorus following the administration of DBcAMP in man. Each value is expressed as mean±S.E. ○, blood glucose; ●, plasma insulin; ●, plasma inorganic phosphorus.

Fig. 2. Changes in I-G ratio following the administration of DBcAMP in man. Each value is expressed as mean±S.E.

but still remained at the elevated level even 120 min after infusion. The plasma IRI response curve did not correlate with that of blood glucose. Plasma Pi fell immediately reaching the minimum at about one half of its preinfusion level within 20 min after infusion and then returned to the preinfusion level. Insulin/glucose ratio (I/G ratio) rose significantly and reached a peak at the end of the infusion, then fell promptly even lower the preinfusion level, and elevated again before returning the initial level in both groups (Fig. 2). The blood lactate values remained
Fig. 3. Changes in blood pyruvate, lactate and L-P ratio following the administration of DBcAMP in man. Each value is expressed as mean±S.E. ○, blood pyruvate; ●, blood lactate; *, L-P ratio.

Fig. 4. Changes in blood redox-potential and excess lactate following the administration of DBcAMP in man. Each value is expressed as mean±S.E. ○, blood redox-potential; ●, excess lactate.

essentially unchanged with 200 mg infusion, however, those values increased significantly and remained higher during a period of observation with 400 mg infusion (Fig. 3). The blood pyruvate values increased significantly and reached a peak at 40 min after infusion in both groups. As a result, the lactate/pyruvate
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Fig. 5. Changes in plasma glycerol and nonesterified fatty acids following the administration of DBcAMP in man. Each value is expressed as mean ± S.E. o, plasma glycerol; *, plasma free fatty acid.

Fig. 6. Changes in blood pH, pCO_2 and base excess (B.E.) following the administration of DBcAMP in man. Each value is expressed as mean ± S.E. o, blood pH; *, base excess; *, arterial pCO_2.

The ratio (L/P ratio) fell promptly to a lower value and remained at the low level even 120 min after 200 mg infusion, but the decline in L/P ratio was smaller with 400 mg infusion. The pattern of change in excess lactate correlated well with the L/P ratio pattern in both groups (Fig. 4). The elevation in the blood redox potential was induced by the DBcAMP infusion in both groups. The DBcAMP infusion also induced about 50 per cent decline in NEFA and glycerol in both groups (Fig. 5). The effects of DBcAMP infusion on the acid-base parameters were
Fig. 7. Changes in serum electrolytes following the administration of DBcAMP in man. Each value is expressed as mean±s.e. ○, Na; ●, Ca; ●, K; ●, Cl.

shown in Fig. 6. Arterial P_{aCO_2} remained constant because the subject’s ventilation was controlled artificially during study. After 200 mg infusion, the arterial pH value increased progressively, and the primary cause of that was an increase in base excess. The changes in acid-base parameters following 400 mg infusion were less consistent. The levels of serum Na⁺, K⁺, and Cl⁻ remained essentially unchanged in both groups. Serum Ca^{++} levels were significantly decreased at 80 min after 200 mg infusion, but no significant changes were observed following 400 mg infusion.

DISCUSSION

Sutherland and Rall (1960) originally demonstrated the role of cAMP as an activator of phosphorylase, and Posternak et al. (1962) showed that the synthetic analogue, DBcAMP, possessed similar properties. In this study, the findings that DBcAMP induced hyperglycemia which last for more than 60 min after infusion in human subjects, suggest that exogenous DBcAMP activates a glycogenolysis in human subjects as has been reported by Hension et al. (1967) in dogs.

DBcAMP also induced striking elevations in the plasma IRI. The elevation in the plasma IRI concentrations seemed to be induced by the direct stimulating action of DBcAMP, rather than by the glucose-stimulated insulin secretion. Though the exact mechanism for the action of DBcAMP on the insulin secretion remained unclear, it can be suggested that the increased cAMP concentration in β-cells induced by exogenous DBcAMP might play an important role in the secretion of insulin, as has been previously suggested by Heersche et al. (1971).

Significant increase in the blood pyruvate value accompanying with slightly increased blood lactate level caused by the DBcAMP infusion indicates the
activation of glycolysis, probably as a result of the activation of phosphofructokinase, one of the key glycolytic enzymes, in a manner similar to that of cAMP as reported by Mansour and Mansour (1962). Since both pyruvate and lactate are known to penetrate readily cell membranes, the changes in L/P ratio calculated from the blood concentrations of these metabolites suggest that of the intracellular L/P ratios. The declines in L/P ratios shown in Fig. 3 possibly mean the shift of the redox state of the NAD/NADH system to oxidation and elevated phosphorylation state, i.e. [ATP]/[ADP][Pi], in cytoplasm (Krebs 1967). This is supported by the elevation in blood redox-potential, depression in the Pi concentration and metabolic alkalosis following DBcAMP infusion. These changes induced by the DBcAMP infusion might be ascribed to an enhanced flux of hydrogen equivalent from cytoplasm into mitochondria via the “glycerol phosphate cycle” as considered by Mullhofer et al. (1974).

The infusion of 200 mg and 400 mg of DBcAMP into human subjects produced progressive and parallel declines in NEFA and glycerol concentrations in plasma for more than 2 hr after infusion. Blether (1967) observed in isolated rat epididymal fat cells that a higher concentration of DBcAMP stimulated lipolysis while a lower concentration of DBcAMP inhibited. Infusion of DBcAMP at a rate of 10–20 mg/min for 20 min probably inhibited lipolysis.

From these results, it is concluded that DBcAMP in dose ranging from 200 to 400 mg stimulates glycogenolysis and glycolysis while lipolysis is inhibited.

Acknowledgment

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References


