Mechanism of Palmityl Coenzyme A Inhibition of Liver Glycogen Synthase*

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Palmityl-CoA inhibits free liver glycogen synthase; the concentration required for half-maximum inhibition is 3 to 4 μM. Almost complete inhibition was observed at 50 μM. Palmityl-CoA inhibition is associated with dissociation of the tetrameric enzyme into monomers, and binding of palmityl-CoA to the monomers. Glycogen-bound enzyme is also inhibited by palmityl-CoA, resulting in dissociation of the enzyme into monomers and concomitant release of the enzyme from the primer glycogen.

Palmityl-CoA inhibition of the enzyme is partially reversed by the glycogen synthase activator, glucose-6-P, whereas sodium lauryl sulfate-inhibited enzyme is not reactivated by glucose-6-P. Sodium lauryl sulfate inhibition results in the dissociation of the tetramer into the monomers.

Bovine serum albumin and cyclodextrin can prevent palmityl-CoA inhibition only when they are added prior to palmityl-CoA addition.

The possible physiological role of palmityl-CoA in glucose homeostasis is discussed.

Liver concentrations of long chain fatty acyl-CoAs fluctuate considerably under different physiological conditions. Two- to three-fold increases in the concentration of palmityl-CoA were observed in fasted and in diabetic animals (1, 2). A similar degree of decrease was observed when starved animals were refed (1). Decreased lipogenic activity in the fasted or in the diabetic conditions together with the observation that palmityl-CoA inhibits various lipogenic enzymes led to the theory that palmityl-CoA and other long chain fatty acyl-CoAs serve as negative effectors for lipogenesis (2, 3). However, such a physiological role of long chain acyl-CoAs in lipogenesis has been frequently questioned on the basis of difficulties associated with palmityl-CoA being a natural detergent; also palmityl-CoA inhibits some enzymes which are seemingly not directly related to lipogenesis, notably, glutamic dehydrogenase and malic dehydrogenase (4-6). Although more recent work strengthens the case for a physiological role of palmityl-CoA as a negative feedback inhibitor of lipogenesis (7, 8), the meaning of inhibition of other enzymes such as glutamate dehydrogenase has not been satisfactorily explained.

In the course of studies on the relationship between lipogenesis and glycogenesis, we have observed a potent inhibitory effect of palmityl-CoA on liver glycogen synthase. This observation, together with the existing literature on the pattern of palmityl-CoA action on various enzymes, suggests that palmityl-CoA inhibits most if not all of the key enzymes in decreasing blood glucose level.

In an attempt to assess the significance of the palmityl-CoA inhibition of glycogen synthase for metabolic regulation, we have examined the mechanism by which palmityl-CoA inhibits glycogen synthase. Our results show that palmityl-CoA causes the dissociation of active tetrameric synthase into monomers which are unable to bind to the primer glycogen.

MATERIALS AND METHODS

Chemicals — UDP-glucose, glucose-6-P, shellfish glycogen, bovine serum albumin, β-cyclodextrin, and glucose-1-P were obtained from Sigma Chemical Co. UDP-[1-14C]glucose (specific activity 220 mCi/mmol) was from New England Nuclear. Palmityl-CoA, decanoyl-CoA, valeryl-CoA, and acetyl-CoA were purchased from P-L Biochemicals. Sodium palmitate, coenzyme A, and Sepharose 4B were from Sigma Chemical Co.

Animals — Experimental animals were 250- to 300-g male Wistar rats from the Biochemistry Department rat colony at Purdue University. They were maintained on a balanced diet and allowed free access to food and water.

Preparation of Glycogen Synthase D — The livers of decapitated animals were homogenized at low speed in a Polytron homogenizer in 2 volumes of 0.25 M sucrose, 5 mM EDTA, pH 7.4. This and all other steps were performed at 0-4°. The crude homogenate was centrifuged at 12,100 × g for 10 min. The supernatant was then centrifuged at 78,000 × g for 60 min. The red microsomal fraction which sedimented on top of the glycogen pellet was carefully removed by washing with the sucrose medium. The glycogen pellet was resuspended by homogenization in 5 volumes of 0.25 M sucrose, 5 mM EDTA, pH 7.4, containing 30% (v/v) glycerol and stored at -4°. This procedure consistently yielded a very stable preparation of glycogen synthase bound to glycogen with greater than 95% of the enzyme activity in the D form.

Homogeneous glycogen-free enzyme was prepared by a modified method based on previously reported procedures (9, 10). The glycogen pellet enzyme preparation described above containing from 2 to 3 units of enzyme activity/ml was made 20 mM in glucose-6-P and adjusted to pH 8.5 with 2 M Tris. p-Hydroxymercuribenzoate was added to a final concentration of 25 μM and then the mixture was stirred for 10 min at room temperature. During this time, maltose hydrate was added (0.1 g/ml); the preparation was then centrifuged...
at 1/8,000 x g for 90 min. Glycogen synthase D in the supernatant was reactivated by incubation at 37° for 20 min in the presence of 20 mM mercaptoethanol. The reactivated enzyme solution was then re-adjusted to pH 6.9 with 0.1 M phosphoric acid. Calcium phosphate was employed in the final step in the purification, which was carried out at 0-4° in an ice bath. The gel was washed with a solution of 0.25 M sucrose, 30 mM NaF, 30 mM 2-mercaptoethanol, and 50 mM potassium phosphate, pH 6.9, then suspended at a concentration of 50 mg/ml in the same buffer. Calcium phosphate gel was added to the enzyme solution at a ratio of 1 mg/mg of protein and stirred for 10 min. The gel with adsorbed protein was collected by centrifugation at low speed. Essentially all of the glycogen synthase was bound to the gel as determined by the absence of glycogen synthase activity in the supernatant after separation from the gel. The gel was washed sequentially with 50 mM potassium phosphate, pH 6.9, containing 5% ammonium sulfate, 0.1 M potassium phosphate, pH 6.9, and finally with 0.14 M potassium phosphate, pH 6.9. All of these solutions also contained 0.25 M sucrose, 50 mM NaF, and 30 mM 2-mercaptoethanol. For each washing, the gel was suspended and stirred for 10 min. After the third washing the glycogen synthase was eluted from the gel by stirring with a solution of 0.3 M potassium phosphate, pH 8.5, 0.25 M sucrose, 30 mM 2-mercaptoethanol, and 10 mM glucose-6-P for 15 min. Usually three elutions were required. Approximately 70% of the total enzyme activity was recovered in the first elution. The enzyme solution was then concentrated to a small volume by ultrafiltration through a Diaflo membrane. On polyacrylamide gel (6%) electrophoresis, a single protein band was detected which corresponds to the glycogen synthase activity. One single protein band was also detected in the presence of sodium lauryl sulfate.

Assay of Glycogen Synthase D–The enzyme activity was assayed as described previously by determining the incorporation of radioactive glucose into glycogen from UDP-[14C]glucose (9). The standard assay mixture contained 0.67 µmol of UDP-[14C]glucose (8,000 to 9,000 cpm), 1.2 mg of shellfish glycogen, 50 µmol of glycylglycine (pH 7.4), and 10 µmol of glucose-6-P in a final volume of 0.5 ml. Reconstituted Glycogen-bound Glycogen Synthase D–High molecular weight protein-free glycogen was prepared from rat liver by a modification of the method of Bueding and Orrell (11). Glycogen synthase D was incubated with the glycogen in 50 mM glycylglycine buffer, pH 7.4, in the presence of 20 mM glucose-6-P at 37° for 10 min. The glycogen-bound enzyme was collected by centrifugation at 78,000 x g for 60 min. The pellet was resuspended in glycylglycine buffer.

Homogeneous glycogen synthase D was used in all of the experiments reported, unless otherwise indicated.

Assay of Glycogen Phosphorylase–Glycogen phosphorylase activity was assayed by determining the incorporation of radioactive glucose into glycogen from UDP-[14C]glucose (9). The standard assay system contained 2 mg of glycogen, 2 µmol of [14C]glucose-1-P (8,500 cpm/µmol), 1 µmol of EDTA, and 50 µmol of β-glycerophosphate, pH 6.8, in a final volume of 0.5 ml. The reaction was run at 30° for 5 min after the addition of enzyme preparation. Labeled glycogen was assayed as described.

RESULTS

Properties of Inhibition–The effect of different concentrations of palmitoyl-CoA on glycogen synthase D activity is shown in Fig. 1. It requires 4 and 50 µM palmitoyl-CoA to inhibit by 50 and 100%, respectively. This is a relatively high concentration compared to the critical micellar concentration of palmitoyl-CoA (4 µM). High concentrations of palmitoyl-CoA also bind to glycogen (Fig. 2). The effect of glycogen on the elution of [1-14C]palmitoyl-CoA from a Sephadex G-50 column is shown in Fig. 2. When mixed with glycogen some [1-14C]palmitoyl-CoA is eluted in the void volume. Binding of palmitoyl-CoA to various cyclodextrins has been attributed to the formation of inclusion compounds: palmitoyl-CoA apparently tets into the interior void space of the cyclodextrins (12). Inhibition of glycogen synthase D by palmitoyl-CoA is partially reversed by the glycogen synthase activator glucose-6-P. Both glycogen and glucose-6-P are reaction components in the synthase assay. Because of the multiple roles of glycogen and glucose-6-P, it is difficult to determine the exact concentration

![Fig. 1. The specificity of inhibition by palmitoyl-CoA on rat liver glycogen synthase and phosphorylase activity. Glycogen synthase and phosphorylase from the same preparation were used to test the effect of palmitoyl-CoA. Rat livers were homogenized in 4 volumes of buffered medium containing 0.25 M sucrose, 5 mM EDTA, and 50 mM glycylglycine, pH 7.4, and centrifuged at 12,000 x g for 10 min. Supernatant was concentrated and 0.1 ml was used for the assay of glycogen synthase or phosphorylase in the presence of indicated concentrations of palmitoyl-CoA. Glycogen synthase activity was assayed in the presence of 10 mM glucose-6-P as described under "Materials and Methods." Phosphorylase activity was assayed by determining the incorporation of radioactive glucose into glycogen from [1-14C]glucose-1-P.](http://www.jbc.org/)

![Fig. 2. Gel filtration chromatography of [1-14C]palmitoyl-CoA preincubated with glycogen. The ability of glycogen to bind [1-14C]palmitoyl-CoA was examined using gel filtration elution profile. [1-14C]Palmitoyl-CoA (2.5 µM) was incubated with glycogen (24 mg) in 50 mM glycylglycine buffer, pH 7.4, at 37° for 10 min. The sample (0.5 ml) was then applied to a Sephadex G-50 column (1 x 27 cm) and fractions were collected (10 drops/fraction). The distribution of eluted [1-14C]palmitoyl-CoA was expressed as radioactivity in each fraction. Elution of glycogen was monitored by reading absorbance at 480 nm.](http://www.jbc.org/) of palmitoyl-CoA required for the inactivation of the synthase. Even in an enzymatic system where other reaction components do not interfere, it has been difficult to obtain exact stoichiometric results which relate the amount of palmitoyl-CoA binding to the functional status of protein (8). Under the
same conditions, palmitoyl-CoA shows no inhibitory effect on glycogen phosphorylase, as shown in Fig. 1.

Neither coenzyme A nor sodium palmitate showed any inhibitory effect at concentrations as high as 0.1 mM, suggesting that inhibition is specifically due to palmitoyl-CoA. Free fatty acids have been reported to be physiological regulators of various enzymes in the glycolytic pathway (13).

Studies with a limited number of fatty acyl-CoAs of different chain length indicate that long chain fatty acyl-CoAs are more effective than the shorter acyl-CoAs (Table I). At 10 μM, palmitoyl-CoA inhibits glycogen synthesis to 70%, whereas C₁₀, C₁₄, and C₁₈ acyl-CoAs showed no inhibitory effect. Decanoyl-CoA inhibits 15% and 25% at 26 and 50 μM, respectively. At these concentrations, palmitoyl-CoA inhibits glycogen synthase almost completely. For a comparison, the effect of sodium dodecyl sulfate was also tested. As shown in Table I, 50% inhibition requires approximately 20 μM.

Bovine serum albumin and various dextrans are known to interact with palmitoyl-CoA, and these compounds have been used to ascertain the reversibility of palmitoyl-CoA inhibition of various enzymes (8). As shown in Fig. 3, the presence of β-dextrin or bovine serum albumin in the inactivation mixture effectively prevents the inhibitory effect of palmitoyl-CoA. This effect of β-dextrin and bovine serum albumin, however, was observed only when these compounds were added prior to palmitoyl-CoA.

These experiments suggest not only that palmitoyl-CoA binds to glycogen synthase but also that cycloexetrin at 2.5 mg/ml and bovine serum albumin at 5 mg/ml cannot dissociate bound palmitoyl-CoA from the enzyme. Dithiothreitol has no effect on palmitoyl-CoA inhibition.

**Mechanism of Inhibition**—To further understand the nature of palmitoyl-CoA inhibition, the effect of palmitoyl-CoA on the affinities of the activator glucose-6-P and the substrate UDP-glucose for the enzyme have been examined. The effects of different concentrations of palmitoyl-CoA on enzyme activity in the presence of various concentrations of glucose-6-P (Fig. 4) and UDP-glucose (Fig. 5) indicate that palmitoyl-CoA is a noncompetitive inhibitor with respect to both glucose-6-P and UDP-glucose. These kinetic studies suggest that the palmitoyl-CoA binding site on glycogen synthase is different from those of glucose-6-P and UDP-glucose. In addition, the noncompetitive nature of palmitoyl-CoA inhibition suggests the possibility that glycogen synthase undergoes a conformational change in the presence of palmitoyl-CoA.

Although palmitoyl-CoA is a noncompetitive inhibitor toward glucose-6-P, interestingly enough the palmitoyl-CoA-inhibited enzyme can be partially reactivated by glucose-6-P as shown in Fig. 6. In one experiment, the synthase was almost completely inactivated by using 0.1 mM palmitoyl-CoA, and in the other the enzyme was inhibited by 70% by 12.5 μM palmitoyl-CoA. In both cases, the maximum reactivation observed was approximately 40%. Thus, 30 mM glucose-6-P reactivated from almost 0 to 40% activity in one case, from 30% to almost 70% in the other.

The sodium dodecyl sulfate-inhibited enzyme cannot be reactivated at all by glucose-6-P, although both sodium dodecyl sulfate and palmitoyl-CoA dissociate the enzyme into mon-

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**Table I**

Effect of acyl-CoAs on glycogen synthase

| Inhibitor concentration (μM) | % inhibition |
|-----------------------------|-------------|
|                             | SDS | Acetyl-CoA | Valeryl-CoA | Decanoyl-CoA | Palmitoyl-CoA |
| 5                           | 20  | 0           | 0           | 0            | 60           |
| 10                          | 25  | 0           | 0           | 0            | 70           |
| 20                          | 45  | 0           | 0           | 18           | 90           |
| 50                          | 90  | 0           | 0           | 25           | 98           |

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**Fig. 3.** Effect of palmitoyl-CoA on glycogen synthase activity in the presence of bovine serum albumin, β-cyclodextrin, and dithiothreitol. In each separate assay, bovine serum albumin (BSA) (5 mg/ml), β-cyclodextrin (2.5 mg/ml), and dithiothreitol (4 mM) were added simultaneously with the indicated concentration of palmitoyl-CoA at the start of the reaction. Assay mixture contained 0.67 μM of UDP-[14C]glucose (8000 cpm), 1.2 mg of shellfish glycogen, 50 μmol of glycylglycine, pH 7.4, 10 μmol of glucose-6-P, and 3 μg of the enzyme in a final volume of 0.5 ml. The activity was expressed as percentage of the activity assayed in the absence of palmitoyl-CoA.

**Fig. 4.** Inhibition of glycogen synthase by palmitoyl-CoA as a function of glucose-6-P concentration. Enzyme activity was assayed at three concentrations of palmitoyl-CoA (2, 10, and 25 μM) in the presence of indicated concentrations of glucose-6-P. Assay mixture contained 0.67 μM of UDP-[14C]glucose, 1.2 mg of glycogen, 50 μmol of glycylglycine, pH 7.4, 3 μg of enzyme, and the indicated concentrations of glucose-6-P and palmitoyl-CoA in a total volume of 0.5 ml. The velocity was expressed as nanomoles of glucose incorporated into glycogen per 10 min.
Regulation of Liver Glycogen Synthase

[Graph: Regulation of Liver Glycogen Synthase]

Fig. 5. Inhibition of glycogen synthase by palmityl-CoA as a function of UDP-glucose concentration. Glycogen synthase activity was assayed at three different concentrations of palmityl-CoA (5, 10, and 25 μM) in the presence of indicated concentrations of the substrate UDP-glucose. Assay mixture contained 1.2 mg of shellfish glycogen, 50 μmol of glycylglycine, pH 7.4, 10 μmol of glucose-6-P, 3 μg of enzyme, and the indicated concentrations of UDP-glucose and palmityl-CoA in a total volume of 0.5 ml. Velocity of the reaction was expressed as nanomoles of glucose incorporated into glycogen per 10 min.

 unequal distribution of the radioactivity associated with the enzyme and the radioactive glucose-6-P in the void volume, indicating that the enzyme is dissociated from the glucose-6-P. When purified, glycogen-free synthase is subjected to Sepharose 4B column chromatography, the peak of enzyme activity is eluted in Fraction 40 as shown in Fig. 7B. The molecular weight of the enzyme is 330,000 as determined by sucrose density gradient centrifugation. When purified enzyme treated with 0.1 mM [1-14C]palmityl-CoA is chromatographed under the same conditions, the peak is eluted in Fraction 60. The molecular weight of this species was determined separately by sucrose density gradient centrifugation and found to be 74,000. This experiment shows that in the presence of palmityl-CoA the enzyme dissociates into monomers and that palmityl-CoA binds to the monomers.

Fig. 7A presents the results of a similar experiment carried out with purified glycogen synthase-glycogen complex. The glycogen synthase-glycogen complex was prepared by mixing the purified enzyme with high molecular weight glycogen and reisolating the glycogen pellet by centrifugation at 40,000 rpm for 60 min. In the absence of palmityl-CoA, the complex was eluted in the void volume. Treatment of the complex with 0.2 mM [1-14C]palmityl-CoA led to dissociation of the monomers. When purified, glycogen-free synthase is subjected to Sepharose 4B column chromatography, the peak of enzyme activity is eluted in Fraction 40 as shown in Fig. 7B. The molecular weight of the enzyme is 330,000 as determined by sucrose density gradient centrifugation (14). When purified enzyme treated with 0.1 mM [1-14C]palmityl-CoA is chromatographed under the same conditions, the peak is eluted in Fraction 60. The molecular weight of this species was determined separately by sucrose density gradient centrifugation and found to be 74,000. This experiment shows that in the presence of palmityl-CoA the enzyme dissociates into monomers and that palmityl-CoA binds to the monomers.

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enzyme from glycogen. The dissociated enzyme with bound \(1^{-14}C\)palmityl-CoA eluted in the fraction where the pure monomeric enzyme would be eluted. These experiments clearly establish not only that palmityl-CoA dissociates the enzyme from glycogen by converting it into the monomer but also that the monomeric form of the enzyme cannot attach itself to the primer glycogen. Small amounts of radioactive palmityl-CoA in the void volume could be due to palmityl-CoA binding to glycogen itself or to tetrameric enzyme still attached to glycogen. The small amount of \([1^{-14}C]\)palmityl-CoA found with the tetrameric form of glycogen-free enzyme (Fig. 7B) indicates that the tetramer binds palmityl-CoA without being dissociated into the monomer. Probably the number of acyl-CoA’s bound per molecule of the active enzyme is critical for dissociation into the monomer.

**Effect of Glucose-6-P on Palmityl-CoA Inhibition** — The effect of glucose-6-P on reactivating palmityl-CoA-inhibited enzyme is shown in Fig. 6. Although complete reactivation was not obtained even at very high concentrations of glucose-6-P (40 mM), this effect of the specific activator of the enzyme is of interest and, therefore, was examined further. Previously, we reported that the effect of glucose-6-P on the synthase subunits was to promote aggregation to forms capable of binding to primer glycogen (15). If the inhibitory effect of palmityl-CoA is primarily due to dissociation of the tetramer into a monomeric form incapable of binding to glycogen, it is reasonable to expect that reactivation by glucose-6-P is due to aggregation of monomers into oligomers which can bind to primer glycogen.

When purified glycogen synthase is incubated with 15 \(\mu\)M \([1^{-14}C]\)palmityl-CoA and subjected to sucrose density gradient centrifugation, the labeled compound was associated with the enzyme species with a molecular weight of 74,000 (Fig. 8). However, if glucose-6-P is added to the palmityl-CoA-inhibited enzyme, the labeled palmityl-CoA sediments with the dimeric form of the enzyme which has a molecular weight of 166,000. This experiment indicates that glucose-6-P promotes aggregation of monomers into dimers. Furthermore, \([1^{-14}C]\)palmityl-CoA remained bound to the enzyme and was not released upon aggregation mediated by glucose-6-P. The top panel of Fig. 8 shows the relative sedimentation distances of purified glycogen synthase D and phosphorylase \(\alpha\) which was used as the standard.

To determine whether the \([1^{-14}C]\)palmityl-CoA-containing dimers formed in the presence of glucose-6-P can bind to primer glycogen, we have examined the binding of palmityl-CoA-inhibited enzyme to the primer in the presence and absence of glucose-6-P.

In the first column of Table II, the distribution of the purified enzyme between soluble and glycogen fractions is shown. The two fractions were separated by centrifugation at 40,000 rpm for 120 min. More than 90% of the enzyme is normally found in the glycogen fraction. However, when synthase was inhibited to 28% activity by 12 \(\mu\)M \([1^{-14}C]\)palmityl-CoA, only one-third of the residual activity was associated with glycogen, while two-thirds of the residual activity remained in the supernatant fraction. Under the

### Table II

| Addition                         | Glycogen synthase activity | Activity distribution | Activity distribution | Glycogen synthase activity | Activity distribution | Activity distribution |
|----------------------------------|----------------------------|-----------------------|-----------------------|----------------------------|-----------------------|-----------------------|
|                                  | %                          | %                     | %                     | %                          | %                     | %                     |
| Enzyme + glycogen                | 100                        | 8.8                   | 91.2                  | 28                         | 65.3                  | 34.7                  |
| Enzyme + glycogen + \([1^{-14}C]\)palmityl-CoA (12 \(\mu\)M) | 50                         | 14.5                  | 85.5                  | 16                         | 84                    |                        |
| Enzyme + glycogen + \([1^{-14}C]\)palmityl-CoA (12 \(\mu\)M) + glucose-6-P (40 mM) | 50                         | 14.5                  | 85.5                  | 16                         | 84                    |                        |

**Fig. 8.** Sucrose density gradient sedimentation of palmityl-CoA-inhibited glycogen synthase in the presence and absence of glucose-6-P. The effect of glucose-6-P on the state of aggregation of palmityl-CoA-inhibited enzyme was examined using linear sucrose density gradient of 0 to 12.5% in buffer containing 12.5% glycerol, 10 mM mercaptoethanol, 50 mM glycylglycine, pH 7.4, and 1% maltose. The enzyme (12 \(\mu\)g) was incubated with 15 \(\mu\)M \([1^{-14}C]\)palmityl-CoA (5,000 cpm/0.1 ml) at 37° for 10 min. To the incubated enzyme was then added 50 mM glucose-6-P and further incubated at 37° for another 10 min. The sample (0.4 ml) was centrifuged at 40,000 rpm for 17 h with a Beckman SW-50 rotator. The fractions were collected and determined for radioactivity associated with the enzyme forms. Rabbit muscle phosphorylase \(\alpha\), used as standard, was run in the same gradient as the control glycogen synthase and the enzyme activity distributed in each fraction was expressed as percentage of the total activity.
same conditions the distribution of [1-14C]palmityl-CoA between the supernatant and the glycogen pellet was 39 and 61%, respectively. Addition of glucose-6-P to the inhibited enzyme results in partial reactivation and the binding of enzyme activity in the glyogen pellet is raised to 85.5%. Likewise, 84% of the [1-14C]palmityl-CoA is recovered in the glycogen pellet under these conditions. These experiments indicate that glucose-6-P promotes binding of the palmityl-CoA-inhibited enzyme to primer glycogen. It is, therefore, reasonable to conclude that the partial reactivation by glucose-6-P is due to the binding of palmityl-CoA-inhibited enzyme to glycogen.

Since palmityl-CoA has some affinity toward glycogen in addition to affinity toward synthase, and since the effect of palmityl-CoA is partially reversed by glucose-6-P, it is difficult to relate the amount of fatty acyl-CoA binding to changes in enzyme activity. However, it is clear from these experiments that palmityl-CoA binding which results in the dissociation of the synthase into the monomer is the mechanism of fatty acyl-CoA inhibition.

**DISCUSSION**

The possible involvement of long chain fatty acyl-CoA as a negative feedback inhibitor of lipogenesis has been debated for a long time. Two arguments cited against such involvement of long chain acyl-CoA have been that (a) long chain fatty acyl-CoAs are natural detergents and their inhibitory effect may be attributed to this detergent property and (b) although many of the lipogenic enzymes are inhibited by long chain acyl-CoAs, there are also some enzymes whose functions are not directly related to lipogenesis, which are inhibited by acyl-CoAs, suggesting that the inhibitory effect is not specific enough to be physiologically significant.

Glycogen synthase has now been added to the list of enzymes affected by palmityl-CoA. A summary of the inhibitory pattern of the metabolic processes is shown in Fig. 9. From this figure, it can be seen that the effect of palmityl-CoA is not restricted to the lipogenic enzymes, but rather that many key enzymes which lower blood glucose levels are affected by palmityl-CoA. Indeed, in this context, palmityl-CoA inhibition of glutamate dehydrogenase or malic dehydrogenase makes sense. It appears that palmityl-CoA inhibited enzymes are generally oligomeric proteins and that palmityl-CoA dissociates them into subunit (7). However, this effect is not totally nonspecific, since glycogen phosphorylase and fructose diphosphatase are not affected by palmityl-CoA (6). These two reactions are rate-limiting enzymes which contribute to increases in the glucose level. Thus, in the diabetic animal, increases in palmityl-CoA could inhibit all of the glucose-utilizing enzymes without affecting those enzymes involved in the production of glucose. Likewise, in the fasting animal, in which gluconeogenesis activity should occur, only the glucose-draining pathways would be inhibited by palmityl-CoA without affecting gluconeogenic activity.

Although proof of the physiological significance of the present work requires further studies, it is extremely interesting to note that the inhibited enzyme can be reactivated by glucose-6-P. Previously, we reported that formation of mixed disulfides between the synthase and reduced glutathione dissociates the enzyme into the monomer and this dissociation results in the release of the enzyme from glycogen (15). Such monomers could be reaggregated into oligomers in the presence of glucose-6-P, and these oligomers could bind to glycogen. The effect of palmityl-CoA is very similar to that of mixed disulfide formation. Studies on the effect of glucose-6-P on palmityl-CoA-inhibited glycogen synthase indicated that glucose-6-P promotes oligomer formation from the subunits concurrent with increased binding to the primer glycogen.

Recently Kawaguchi and Bloch (16) compared the actions of palmityl-CoA and sodium lauryl sulfate on various enzymes. They noted that palmityl-CoA dissociates the tetrameric enzymes into dimers, while sodium lauryl sulfate effects dissociation into monomers. Furthermore, only the palmityl-CoA-dissociated enzymes could be reactivated. It appears that palmityl-CoA’s effect on glycogen synthase is very similar to those observations.

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