Rat Liver Peroxisomes Catalyze the Initial Step in Cholesterol Synthesis

THE CONDENSATION OF ACETYL-CoA UNITS INTO ACETOACETYL-CoA*

(Received for publication, November 21, 1988)

Sara L. Thompson and Skaidrite K. Krisans†
From the Department of Biology and Molecular Biology Institute, San Diego State University, San Diego, California 92182

In the last few years, it has been demonstrated by this group and others that rat liver peroxisomes participate in cholesterol synthesis. It has been shown that the key regulatory enzyme of isoprenoid biosynthesis, 3-hydroxy-3-methylglutaryl coenzyme A reductase, is present in liver cells not only in the endoplasmic reticulum but also within peroxisomes. It has been also demonstrated that rat liver peroxisomes in the presence of cytosolic proteins in vitro are able to convert [14C]mevalonic acid to [14C]cholesterol. In addition, a recent study demonstrated that the largest cellular concentration of sterol carrier protein-2 is inside peroxisomes.

It is of interest, therefore, to inquire whether other proteins known to be involved in cholesterol biosynthesis are also present in peroxisomes. In this study we investigated the first step in cholesterol synthesis, the condensation of two acetyl-CoA units to acetoacetyl-CoA. It was demonstrated that peroxisomal thiolase, purified by DEAE-phosphocellulose chromatography from gemfibrozil-treated rats, is active not only toward acetoacetyl-CoA and 3-ketoacyl-CoA, consistent with literature reports, but is also capable of converting acetyl-CoA units to acetoacetyl-CoA. This is the first demonstration of condensation activity in rat liver peroxisomes.

In the last few years, it has been demonstrated by this group and others that rat liver peroxisomes participate in cholesterol synthesis. What is currently known about peroxisomal cholesterol synthesis began with the discovery that peroxisomes can convert 3-hydroxy-3-methylglutaryl (HMG) coenzyme A to mevalonic acid due to the presence of a peroxisomal HMG-CoA reductase, the rate-limiting enzyme in cholesterol synthesis (1, 2). Following those studies, it was demonstrated that peroxisomes can convert mevalonic acid to cholesterol in the presence of cytosolic proteins (3). And most recently, it was reported that hepatic peroxisomes contain the thiolase activity necessary for the initial step in cholesterol synthesis, the condensation of acetyl-CoA units into acetoacetyl-CoA.

This paper presents the first evidence that rat liver peroxisomes catalyze the initial step in cholesterol synthesis. The condensation of two molecules of acetyl-CoA into one molecule of acetoacetyl-CoA is strongly favored in the cleavage direction.

Peroxisomes contain a thiolase that catalyzes reactions with long chain 3-ketoacyl-CoA substrates. The primary function of this enzyme appears to be the cleavage of the two-carbon acetyl-CoA unit from medium and long chain 3-ketoacyl-CoA substrates (11–15). It is structurally and immunologically different than the mitochondrial 3-ketoacyl-CoA thiolase (EC 2.3.1.16). Acetoacetyl-CoA can be used as a substrate for both the peroxisomal and mitochondrial enzymes, though with much less specificity (12). The purified mitochondrial 3-ketoacyl-CoA thiolase is not able to convert acetyl-CoA to acetoacetyl-CoA (16).

This paper presents the evidence that rat liver peroxisomes catalyze the initial step in cholesterol synthesis, the condensation of acetyl-CoA units into acetoacetyl-CoA. 

EXPERIMENTAL PROCEDURES

Animals

Male Sprague-Dawley rats (180–250 g) were kept on a 12-h light-dark cycle from birth. Standard lab chow and water were given ad libitum. A standard laboratory diet was also supplemented with 0.2% gemfibrozil (Lopid, 5-(2,5-dimethylphenoxy)-2,2-dimethylpentanoic acid; Warner-Lambert Company) and fed ad libitum 1–4 weeks prior to killing. All rats were fasted overnight and killed by decapitation 2 h into their light cycle.

Preparation of Rat Liver Peroxisomes, Cytosol, and Mitochondria

The buffer for homogenization and differential centrifugation contained the following: 0.25 M sucrose, 5 mM MOPS, 1 mM EDTA, and 0.1% ethanol at pH 7.5. The buffer for all gradient media was 3 mM imidazole, pH 7.5.

Liver homogenates were first fractionated by differential centrifugation to obtain a peroxisome-enriched fraction (similar to deDuve’s light mitochondrial fraction) and a cytosolic fraction (2). Peroxisomes and mitochondria were then further purified by equilibrium density
Peroxisomes Catalyze the Initial Step in Cholesterol Synthesis

centrifugation of the peroxisome-enriched fraction in a TV-850 vertical rotor (40,000 rpm, 50 min) on a linear sucrose gradient. A total of 20–25 fractions were collected from the bottom of the tube with a two-way needle.

Assay of Marker Enzymes

All fractions were analyzed for marker enzyme activity and protein concentration. The activity of catalase was measured according to Leighton et al. (17). The activity of glutamate dehydrogenase was measured according to Schmidt (18). Phosphoglucone isomerase was used as a marker for the cytosolic fraction and measured according to the method of Nothmann (19). Esterase was used to measure microsomal activity according to the method of Beaufay et al. (20). Acid phosphatase, a lysosomal marker, was measured according to Bergmeyer et al. (21). Protein was determined by the biinchoninic acid method (Pierce Chemical Co.) using bovine serum albumin as a standard.

In all experiments, the isolated peroxisomes were approximately 94% pure. They contained less than 1% mitochondria as estimated by glutamate dehydrogenase activity. There was no measurable cytosolic or lysosomal contamination as measured by phosphoglucone isomerase and acid phosphatase, respectively. Microsomal protein was essentially the only contaminant at about 5–6%.

Mitochondrial fractions were approximately 85% pure. The major contaminant was microsomal, with some contamination by peroxisomes. Cytosolic fractions were approximately 90% pure.

Thiolase Assays

Clavage—Acetoacetyl-CoA thiolase activity in the cleavage direction was determined by following the decrease in absorbance at 303 nm at 30 °C, based on the protocol of Staak et al. (22). The reaction mixture containing the following was then added for a final volume of 1 ml: 100 mM Tris-HCl, pH 8.3, 25 mM MgCl₂, 100 μM EDTA, and 52 μM acetoacetyl-CoA. Endogenous activity was allowed to run out for exactly run at 30 °C which took approximately 2–3 min. The reaction was then started with the addition of CoA (250 mM, final concentration) and followed for 1 min. With these conditions, the change in the slope following the addition of CoA was greater than 0.1 OD/min. The molar coefficient of 21,400 M⁻¹ for acetoacetyl-CoA as reported by Staak et al. (22) was used for the calculations. The units of activity are defined as micromoles/min/ml. This assay was accurate with initial subcellular fractions as well as partially purified proteins and activity was not affected by freezing (−70 °C, 20% glycerol).

3-Oxocotanoyl-CoA activity was determined according to Seubert et al. (23) with two modifications. 3-Oxocotanoyl-CoA was synthesized by the addition of octanoyl-CoA (35 μM, final concentration) and acetyl-CoA oxidase (0.02 unit) to the 1 ml of reaction mixture. Also, the Mg²⁺ concentration was increased to 25 mM. The molar coefficient for the thiolase reaction was 14,400 M⁻¹ as determined by Staak et al. (22).

Condensation—Condensation activity was determined by measuring the increase in absorbance at 303 nm at 30 °C according to Clinkenbeard et al. (6). The reaction mixture contained the following: 100 mM Tris-glycine, pH 8.8, 50 mM MgCl₂, and sample protein (1–10 μg). Endogenous cleavage activity was allowed to run out at 30 °C which took approximately 1–2 min with a change in absorbance of less than 0.1 OD/min. The reaction was started with the addition of 1 mM acetyl-CoA and was followed for 1 min. Under these conditions, the change in the slope following the addition of acetyl-CoA was greater than 0.1 OD/min. This reaction could only be measured in partially purified protein samples and was not affected by freezing (−70 °C, 20% glycerol). The molar extinction coefficient for the condensation reaction is 20,700 M⁻¹ cm⁻¹ (6).

Partial Purification of Peroxisomal Thiolase Activity

Solubilization—Peroxisomes from gemfibrozil-treated rat livers were used for the purification. Highly purified peroxisomes were prepared as described above. The samples were diluted 4:1 with ice-cold water, mixed, allowed to sit for 10 min at 0 °C, and then centrifuged in a TI-60 Beckman rotor for 3 min at 25,000 rpm, using a Beckman ultracentrifuge (24). The supernatant was collected, measured for protein concentration and cleavage activity using acetoacetyl-CoA as substrate and then subjected to the following chromatographic procedures.

Chromatography—All chromatography was performed at 4 °C. The supernatant was applied to a DEAE-cellulose column equilibrated with 10 mM potassium phosphate buffer (1 mM EDTA, pH 7.0) with 20% glycerol. The column was washed with the same buffer. The eluant was then applied to a phosphocellulose column (equilibrated with the above buffer). The proteins were eluted with a potassium phosphate gradient (10–400 mM potassium phosphate, 1 mM EDTA, pH 7.0).

Immunoblotting Techniques

Proteins were separated on 10% polyacrylamide, 0.1% SDS slab gels (1.5 mm) according to Laemmli (25), followed by the Bio-Rad Western blot protocol. Rabbit anti-thiolase was the generous gift of Dr. Rachubinski, McMaster University, Ontario, Canada. Rabbit anti-catalase was the generous gift of Dr. Tager, Amsterdam, the Netherlands. Visualization was with affinity-purified goat anti-rabbit IgG-horseradish peroxidase conjugate.

Materials

Acetyl-CoA, acetoacetyl-CoA, and octanoyl-CoA as well as all other biochemicals were purchased from Sigma. Chromatography and electrophoresis supplies were purchased from Bio-Rad.

RESULTS AND DISCUSSION

Since the enzymes involved in cholesterol metabolism are present in the liver in trace amounts at best (both in endoplasmic reticulum and in the peroxisomes), we have invested some time in administering to rats a variety of drugs that potentially could influence the levels of these enzymes in peroxisomes. We have found that cholestyramine (a bile acid-sequestering drug) increased peroxisomal HMG-CoA reductase activity about eight fold over control levels without producing any change in peroxisome number in the liver (2). Clofibrate, a commonly used peroxisome proliferator and hypolipidemic drug (26, 27), produces a decrease in peroxisomal HMG-CoA reductase activity. Fortunately, we have identified a drug, gemfibrozil, that not only induces peroxisomal HMG-CoA reductase levels and activity, but also produces a striking proliferation in the number of liver peroxisomes.

Gemfibrozil is a hypolipoproteinemic drug and a known peroxisome proliferator (28–30). Cholesterol synthesis in the rat liver has also been reported to be increased after gemfibrozil administration (31–33). We have performed a detailed morphological analysis of the effects of this drug on the rat liver (34). This study confirmed the increase in peroxisome number and also illustrated that the size and shape of peroxisomes, the matrical inclusions, and the catalase content demonstrate a zonal variation within the gemfibrozil-treated liver. Since gemfibrozil induces cholesterol synthesis, increases peroxisomal HMG-CoA reductase levels and activity, and produces a striking proliferation in peroxisomes, we employed gemfibrozil-treated rats to study the thiolase activity in peroxisomes.

Effect of Gemfibrozil on Subcellular Distribution of Thiolase Activity—A time course study was performed to determine the number of weeks of gemfibrozil feeding required for optimal peroxisomal thiolase activity. Thiolase activity in peroxisomes, mitochondria, and the cytosolic fraction from both control rats and gemfibrozil-treated rats was assayed using acetoacetyl-CoA as substrate. Fig. 1A shows the steady increase in thiolase specific activity in peroxisomes (> 40-fold) with increasing duration of the gemfibrozil diet. By the end of 1 week of the diet, the thiolase specific activity in peroxisomes was equal to the thiolase specific activity in the mitochondria and by 3-4 weeks was roughly double that of the activity in mitochondria. Cytosolic thiolase specific activity increased approximately 10-fold, and the mitochondrial thiolase specific activity increased approximately 2-fold.

Fig. 1B shows the calculated percent contribution of each

S. L. Thompson and S. K. Krisans, unpublished data.
Peroxisomes Catalyze the Initial Step in Cholesterol Synthesis

Weeks on Gemfibrozil Diet

80
60
40
20
0 1 2 3 4
Weeks on Gemfibrozil Diet

FIG. 1. Effect of gemfibrozil on the subcellular distribution of thiolase specific activity in the rat liver. Thiolase activity was measured with acetoacetyl-CoA as substrate. A, comparison of the variations in thiolase specific activity of peroxisomal (solid bars), mitochondrial (striped bars), and cytosolic (shaded bars) fractions during the time course of gemfibrozil treatment. Specific activity is defined as units/mg (micromoles/min/mg protein). B, percent contribution of each subcellular fraction to the total liver thiolase activity.

subcellular fraction to the total liver thiolase activity. These percentages were derived from the specific activity of thiolase in each fraction multiplied by the milligrams of protein/g of liver each subcellular fraction contributes to the total liver protein. The percent of total liver protein values for each subcellular fraction are well established for normal male rats (17). For the gemfibrozil-treated rats, we determined by marker enzyme analysis (35) that there was an increase in peroxisomal protein to 10% of the total liver protein (data not shown). This represents a 5-fold increase in peroxisome protein over normal levels. These data are supported by the morphological studies (34). There was no apparent change in the cytosolic percentage (30% of total) and a small increase in the mitochondrial contribution (25% of total) was observed. From Fig. 1B it can be seen that peroxisomal thiolase activity accounts for about 1–2% of the total in normal rats, whereas after four weeks of gemfibrozil treatment the peroxisomal contribution is greater than 40% of the total. The mitochondrial contribution decreases from 85% of the total in normal rats to 40% with gemfibrozil treatment. The cytosol contribution remains consistently within the range of 15–22% of the total.

However, the true contribution of the peroxisomal thiolase activity from both normal and gemfibrozil-treated animals is very likely underestimated. It has been demonstrated that peroxisomal thiolase, as measured by acetoacetyl-CoA thiolase activity, leaks out of the isolated peroxisomes more easily than any other peroxisomal protein tested (24, 36). It was shown that by simply diluting peroxisomes in water or 0.25 M sucrose buffer up to 86% of the thiolase activity was found in the supernatant. We have confirmed these results by determining the loss of thiolase and catalase from the purified peroxisomal fractions during isolation by immunoblotting. The sucrose gradient fractions were separated on SDS-polyacrylamide gel electrophoresis and the proteins transferred to nitrocellulose; the lower section of nitrocellulose was incubated with an antibody specific for peroxisomal 3-ketoacyl-CoA thiolase and the upper section of nitrocellulose was incubated with an antibody against catalase. Fig. 2 illustrates the results. Represented are the peak fractions from the light

![Fig. 2. Immunoblotting analysis of catalase and thiolase from isolated gradient fractions. Gradient fractions were separated by SDS-polyacrylamide gel electrophoresis (25 μg of protein) and transferred to nitrocellulose. The upper section of nitrocellulose was incubated with a catalase antibody and the lower section was incubated with an antibody to peroxisomal 3-ketoacyl-CoA thiolase. These antibodies are specific for peroxisomal proteins. The densities given are the densities for the peak fraction of solubilized proteins (1.08), the peak mitochondrial fraction (1.21) and the peak peroxisomal fraction (1.24). Molecular weight (M.W.) standards are shown.](http://www.jbc.org/)

![Fig. 3. Elution profile of peroxisomal thiolase activity following phosphocellulose column chromatography (as described under "Experimental Procedures"). The volume of each fraction is 2 ml. Thiolase activity was measured with acetoacetyl-CoA as substrate.](http://www.jbc.org/)
Peroxisomes Catalyze the Initial Step in Cholesterol Synthesis

TABLE I

| Sample                             | Total protein | Total activity | Specific activity | Purification |
|------------------------------------|---------------|----------------|-------------------|--------------|
| Intact peroxisomes (sucrose gradient) | 21.31         | 36.28          | 1.70              | 7            |
| Supernatant (after original dilution) | 4.92          | 17.51          | 3.56              | 2            |
| Phosphocellulose elution peak      | 0.34          | 4.03           | 11.67             | 7            |

Approx. M.W. 41.00

FIG. 4. Upper section, comparison of the phosphocellulose column elution profiles for thiolase cleavage activity using acetoacetyl-CoA (O) and 3-oxooctanoyl-CoA (+) as substrates (units/ml). Lower section, immunoblot of peroxisomal 3-ketoacyl-CoA thiolase of the peak elution fractions.

TABLE II

| Substrate                   | Specific activity |
|-----------------------------|-------------------|
| Acetyl-CoA                  | 2.09              |
| Acetoacetyl-CoA             | 11.67             |
| 3-Oxooctanoyl-CoA           | 127.89            |

density end of the gradient (soluble proteins), the middle density regions (peak mitochondrial and microsomal fractions), and the heavy density end (the peak peroxisomal fractions). Each lane contains an equal amount of protein (25 µg). The immunoblot shows that a significant amount of the peroxisomal thiolase protein has leaked out of peroxisomes and is found at the soluble end of the gradient. Based on the amount of each fraction loaded on the gel and the resulting signals after immunoblotting, it appears that between one-third and one-half of the peroxisomal thiolase activity is lost. The results with peroxisomes isolated from control animals were similar (data not shown). Our measured specific activity of catalase in purified peroxisomal fractions is consistent with literature values: for normal peroxisomes, the specific activity range was 14-16 units/mg; with gemfibrozil, the specific activity range was 6-8 units/mg. These results show that the specific activity of thiolase measured in peroxisomes (normal and gemfibrozil) could be underestimated by as much as a factor of 2. The data also suggest that the specific activity of the thiolase in the cytosolic fraction may be increased due to the leakage of peroxisomal thiolase into the cytosol during differential centrifugations. These factors would significantly change the percent of peroxisomal contribution to total liver thiolase activity.

Partial Purification of the Peroxisomal Thiolase Activity—Peroxisomal thiolase activity was partially purified as described under "Experimental Procedures." Thiolase activity was eluted in one sharp peak as seen in Fig. 3. This elution profile is very similar to other published reports (12, 37) in which the peroxisomal thiolase shows one peak and is eluted at about 15-25 mM phosphate. The profile also confirms the purity of the peroxisomal preparation as there is no measurable activity where either the cytosolic thiolase or the mitochondrial thiolases would be expected to be eluted (in the range of 200-400 mM phosphate) (7, 8, 38). The degree of purification of thiolase activity is presented in Table I. As can be seen, there was a 7-fold increase in specific activity following the separation on the phosphocellulose column.

Cleavage Substrate Specificities—The fractions eluted from the phosphocellulose column were also tested for activity using 3-oxooctanoyl-CoA as a substrate (Fig. 4). Greater specificity was obtained with 3-oxooctanoyl-CoA as compared to acetoacetyl-CoA. The ratio of specific activities was 10:1 (Table II). The greater specificity for 3-oxooctanoyl-CoA is in agreement with previous studies using thiolase isolated from di(2-ethylhexyl)phthalate-treated rats (12). In addition, an immunoblot (thiolase) of the peak fractions is shown in Fig. 4. The intensity of the signal is in excellent agreement with the activity peak. Coomassie blue staining of the peak fraction confirms a 7-10-fold purification of the sample (data not shown).

Condensation Activity—Thiolase activity was also measured in the condensation direction. Once again, there was only one peak of activity which was eluted with the peak thiolase activity measured with acetoacetyl-CoA (Fig. 5). In terms of absolute activity, the ratio of acetoacetyl-CoA cleavage to condensation is 5-6-fold (Table II). As this is the first time condensation activity has been reported in peroxisomes, this value cannot be compared to other peroxisomal studies. The acetoacetyl-CoA-specific thiolase in rat liver mitochondria (partially purified) demonstrates a similar ratio (16). Again, the mitochondrial 3-ketoacyl-CoA thiolase does not...
have condensation activity (16).

There remain many questions to be answered about the thiolase activity in peroxisomes. First, is there more than one thiolase in peroxisomes, one for \( \beta \)-oxidation and one for formation of acetoacetyl-CoA? This is the case in rat liver mitochondria (7, 8). It is of significance to note a recent study which demonstrates the presence of a second thiolase in rat liver peroxisomes (39). Sequencing revealed that, though there is a high degree of identity, the second thiolase has a sequence distinctly different from the previously described sequence for the peroxisomal \( \beta \)-ketoady-CoA thiolase. The function of this thiolase is not known. There is also a report of two thiolases in peroxisomes from yeast (37), one specific for 3-ketoacyl CoA substrates and the other for acetoacetyl-CoA.

Second, what is the purpose of acetoacetyl-CoA thiolase activity in peroxisomes? It is well established that the mitochondrial acetoacetyl-CoA thiolase, catalyzing the condensation reaction, produces acetoacetyl-CoA for ketogenesis. It is also postulated that the mitochondrial cleavage activity of mitochondrial \( \beta \)-ketoady-CoA thiolase (16). Is that also the role for the peroxisomal acetoacetyl-CoA thiolase activity? It seems highly unlikely since peroxisomal \( \beta \)-oxidation in mammalian cells does not degrade its substrates completely, going through only 2-5 cycles (11). Another explanation is that the condensation activity is used for chain elongation of fatty acids. A recent report indicated the presence of acetyl-CoA-dependent chain elongation in intact rat liver peroxisomes from di(2-ethylhexyl)phthalate-treated rats. The chain elongation is postulated to take place essentially via a reversal of the peroxisomal \( \beta \)-oxidation system, with the final step catalyzed by enoyl-CoA reductase (40). This implies a condensation activity on the part of the \( \beta \)-oxidation thiolase.

Finally, as mentioned earlier, there is substantial evidence that peroxisomes are involved in cholesterol synthesis (1-5). In light of the present study it is very possible that peroxisomes also participate in the initial step of cholesterol synthesis by conversion of acetyl-CoA units (probably produced by peroxisomal \( \beta \)-oxidation) to acetoacetyl-CoA.

REFERENCES

1. Keller, G. A., Barton, M. C., Shapiro, D. J., and Singer, S. J. (1986) Proc. Natl. Acad. Sci. U. S. A. 83, 770-774
2. Keller, G. A., Pazirandeh, M., and Krisans, S. (1986) J. Cell Biol. 103, 875-896
3. Thompson, S. L., Burrows, R., Laub, R. J., and Krisans, S. K. (1987) J. Biol. Chem. 262, 17420-17425
4. Tsuneoka, M., Yamamoto, A., Fujiki, Y., and Tashiro, Y. (1988). J. Biochem. (Tokyo) 104, 560-564
5. Keller, G. A., Scallen, T. J., Clarke, D., Maher, P. A., Krisans, S. K., and Singer, S. J. (1989) J. Cell Biol. 108, 1353-1361
6. Clinkenbeard, K. D., Sugiyama, T., Moss, J., Reed, W. D., and Lane, M. D. (1973) J. Biol. Chem. 248, 2275-2284
7. Middleton, B. (1973) Biochem. J. 132, 717-730
8. Middleton, B. (1973) Biochem. J. 132, 731-737
9. Chapman, M. J., Miller, L. R., and Ontko, J. A. (1973) J. Cell. Biol. 58, 284-306
10. Clinkenbeard, K. D., Reed, W. D., Mooney, R. A., and Lane, M. D. (1975) J. Biol. Chem. 250, 3108-3114
11. Lazarow, P. B. (1978) J. Biol. Chem. 253, 1522-1528
12. Miyazawa, S., Osami, T., and Hashimoto, T. (1980) Eur. J. Biochem. 103, 589-596
13. Miyazawa, S., Purata, S., Osami, T., Hashimoto, T., and Ui, N. (1981) J. Biochem. (Tokyo) 90, 511-519
14. Fujiki, Y., Kuchibunshki, K. A., Mortensen, R. M., and Lazarow, P. B. (1985) Biochem. J. 226, 697-704
15. Hinkata, M., Ishii, N., Kaamiskyama, H., Osami, T., and Hashimoto, T. (1987) J. Biol. Chem. 262, 8151-8158
16. Reed, W. D., Ozand, P. T., Tildon, J. T., and Cornblath, M. (1977) Biochem. J. 164, 27-32
17. Leighton, F., Poole, B., Beaufay, H., Bauldhuin, P., Coffey, J. W., Fowler, S., and de Duve, C. (1968) J. Cell. Biol. 37, 482-513
18. Schmidt, B. (1974) in Methods of Enzymatic Analysis (Bergmeyer, H. U., ed) 2nd English Ed. Vol. 2. pp 650-656. Verlag-Chemie, Weinheim, West Germany
19. Noltmann, E. A. (1966) Methods Enzymol B, 501-506
20. Beaufay, H., Amar-Costes, A., Feytman, E., Thines-Sempoux, D., Wibo, M., Robbi, M., and Berthet, J. (1974) J. Cell. Biol. 61, 188-200
21. Bergmeyer, H. U., Gawehn, K., and Grassl, M. (1974) in Methods of Enzymatic Analysis (Bergmeyer, H. U., ed) 2nd English Ed. Vol. 1. pp 495-496. Verlag-Chemie, Weinheim, West Germany
22. Staack, H., Binstock, J. F., and Schulz, H. (1978) J. Biol. Chem. 253, 1827-1831
23. Seubert, W., Lamberts, I., Kramer, R., and Ohly, B. (1968) Biochim. Biophys. Acta 164, 498-517
24. Alexaon, S. E. II, Fujiki, Y., Shio, H., and Lazarow, P. B. (1985) J. Cell Biol. 101, 294-304
25. Laemmli, U. K. (1970) Nature 227, 680-685
26. Reddy, J. K., and Krishnan, T. N. (1965) Biochim. Biophys. Acta 101, 498-517
27. Reddy, J. K. (1980) in Drugs Affecting Lipid Metabolism (Fumagalli, R., Kutchekovsky, D., and Paolotti, R, eds) pp 301-310. Elsevier/North Holland Biomedical Press, Amsterdam, Netherlands
28. Reddy, J. K., and Lalwani, N. D. (1983) CRC Crit. Rev. Toxicol. 12, 1-58
29. Fukuda, K., Shindo, H., Yamashina, S., and Mizuhira, V. (1978) Acta Histochem. Cytochem. 11, 432-442
30. Gray, R. H., and de la Iglesia, F. A. (1984) Hepatology 4, 520-530
31. Maxwell, R. E., Nawrocki, J. W., and Uhlendorf, P. D. (1990) Clin. Res. Forums 4, 43-53
32. Maxwell, R. E., Nawrocki, J. W., and Uhlendorf, P. D. (1983) Atherosclerosis 48, 190-203
33. Newton, R. S. (1986) Today's Ther. Sups. 3, 13-22
34. Gorgas, K., and Krisans, S. K. (1989) J. Lipid Res. 30, 1899-1875
35. Lazarow, P., and Duve, C. (1976) Proc. Natl. Acad. Sci. U. S. A. 73, 2043-2046
36. Hayashi, H., Hiro, S., and Yamasaki, F. (1981) Eur. J. Biochem. 120, 47-51
37. Kurihara, T., Ueda, M., and Tanaka, A. (1988) FEBS Lett 229, 215-218
38. Aronson, J. L., and Lowenstein, J. M. (1983) J. Biol. Chem. 258, 4735-4738
39. Bodnar, A. G., and Rachubinski, R. A. (1989) J. Cell Biol. 109, 47 (abstr.)
40. Horie, S., Suzuki, T., and Suga, T. (1989) Arch. Biochem. Biophys. 274, 64-73
Rat liver peroxisomes catalyze the initial step in cholesterol synthesis. The condensation of acetyl-CoA units into acetoacetyl-CoA.

S L Thompson and S K Krisans

*J. Biol. Chem.* 1990, 265:5731-5735.

Access the most updated version of this article at [http://www.jbc.org/content/265/10/5731](http://www.jbc.org/content/265/10/5731)

Alerts:
- When this article is cited
- When a correction for this article is posted

Click here to choose from all of JBC's e-mail alerts

This article cites 0 references, 0 of which can be accessed free at [http://www.jbc.org/content/265/10/5731.full.html#ref-list-1](http://www.jbc.org/content/265/10/5731.full.html#ref-list-1)