Chapter 19 Lipid Metabolism

As we saw in Chapter 9, cells contain a diverse array of lipids. The major functions of lipids include forming a barrier to the extracellular environment, maintaining membrane fluidity, and serving as an important energy store, principally in the form of triacylglycerols. This chapter focuses on the diverse metabolic pathways of cellular lipids. First, the means by which ingested dietary lipids are degraded and assimilated is considered. The chapter then presents the oxidative pathways by which long acyl chains are converted to successive two-carbon acetyl-CoA fragments, which are either further oxidized via the citric acid cycle and the electron-transport system, converted to ketone bodies, or used in biosynthesis. Next, mechanisms of fatty acid biosynthesis by successive condensations of two-carbon fragments are presented. The steps leading to the biosynthesis of membrane phospholipids and glycolipids are outlined, followed by the pathway of cholesterol biosynthesis and its regulation. The reader should contemplate the varied metabolic reactions that result in the formation or degradation of lipids in the context of the nutritive, structural, and regulatory roles that lipids fulfill in the cell.

Essential Concepts

Lipid Digestion, Absorption, and Transport

1. The enzymatic digestion of triacylglycerols occurs at the lipid–water interface and is aided by the presence of bile acids, which help solubilize the lipids. Bile acids are cholesterol derivatives that are synthesized in the liver as taurine or glycine derivatives, stored in the gall bladder, and released into the small intestine.

2. Pancreatic lipase hydrolyzes triacylglycerols first to diacylglycerols and then to 2-monoacylglycerols plus free fatty acids. Binding of the enzyme to triacylglycerol at the lipid–water interface requires colipase. The interaction of this protein with lipase produces a hydrophobic surface which promotes binding of the protein complex to the lipid. Phospholipase A_2 also degrades phospholipids to lysophospholipids and fatty acids at a lipid–water interface.

3. Micelles containing bile acids promote the absorption of triacylglycerol and phospholipid hydrolysis products by the intestinal mucosa. Inside intestinal cells, fatty acids are complexed to a fatty acid–binding protein that, in effect, increases the solubility of these hydrophobic compounds.

4. Triacylglycerols are resynthesized in intestinal cells and incorporated into chylomicrons. These lipoproteins enter the bloodstream via the lymphatic system and eventually provide triacylglycerols to peripheral tissues, chiefly skeletal muscle and adipose tissue. The delivery process converts chylomicrons to much smaller chylomicron remnants, which are taken up by the liver.

5. The liver synthesizes other lipoproteins, including very low density lipoproteins (VLDL), which transport triacylglycerols and cholesterol from the liver to skeletal muscle and adipose tissue. In the capillaries, triacylglycerols are degraded by lipoprotein lipase to yield fatty acids (which the cells can either oxidize or reincorporate into triacylglycerols) and glycerol (which can be transformed to the glycolytic intermediate dihydroxyacetone phosphate). As they lose their triacylglycerol component, VLDL are converted first to intermediate density lipoproteins (IDL) and then to low density lipoproteins (LDL).

6. Cholesterol is removed from cell surface membranes and carried as cholesteryl esters through the bloodstream to the liver by high density lipoproteins (HDL). There, the cholesteryl esters are transferred to VLDL.

Fatty Acid Oxidation

7. When energy needs dictate, triacylglycerols stored in adipose tissue are broken down (mobilized) by hormone-sensitive lipase. Released free fatty acids are transported in complex with serum albumin to the liver and other tissues.

8. Before being degraded by oxidation, fatty acids are first activated by the formation of an acyl-CoA in an ATP-dependent reaction catalyzed by thiokinase.

9. Since β oxidation takes place in the mitochondrial matrix, the acyl groups must cross the inner mitochondrial membrane, which is impermeable to fatty acyl-CoA derivatives. Therefore, the acyl group is transferred to carnitine by carnitine palmitoyl transferase I. The resulting acyl-carnitine readily crosses the membrane via a carrier protein. Once in the mitochondrial matrix, the acyl group is transferred back to a CoA molecule by carnitine palmitoyl transferase II, and the liberated carnitine crosses the membrane back to the cytosol.

10. Fatty acyl groups are degraded by a process called β oxidation, in which successive two-carbon fragments are removed as acetyl-CoA units.
    (a) Acyl-CoA dehydrogenase catalyzes formation of a *trans*-2,3 (α,β) double bond. The enzyme's bound FAD is thereby reduced to $FADH_2$.
    (b) Enoyl-CoA hydratase catalyzes the hydration of the double bond to produce a 3-L-hydroxyacyl-CoA.
    (c) 3- L-Hydroxyacyl-CoA dehydrogenase catalyzes the formation of a β-ketoacyl-CoA with the reduction of NAD^+ to NADH.
    (d) Thiolase catalyzes the thiolysis of the C2—C3 bond, releasing acetyl-CoA and forming a new acyl-CoA which is two carbons shorter than the starting substrate.
    This sequence of reactions is repeated until the acyl-CoA has been converted entirely to acetyl-CoA. For oxidation of palmitoyl-CoA, the sequence occurs 7 times to yield 8 acetyl-CoA.

11. The oxidation of fatty acids is highly exergonic. For example, palmitate's 8 acetyl-CoA can enter the citric acid cycle, and the $FADH_2$ and NADH generated by β oxidation and the citric acid cycle can be reoxidized by the electron-transport chain, which yields a total of 129 ATP.

12. The oxidation of unsaturated fatty acids requires additional enzymatic reactions to accommodate the double bond at C9 in monounsaturated fatty acids (e.g., oleic acid) and at three-carbon intervals in polyunsaturated fatty acids (e.g., linoleic acid). When a *cis*-3,4 (β,γ) double bond is encountered after several rounds of β oxidation, enoyl-CoA isomerase converts it to a *trans*-2,3 double bond. Oxidation of a polyunsaturated fatty acid also requires a reaction catalyzed by 2,4-dienoyl-CoA reductase, which removes a double bond at the expense of NADPH.

13. The final round of β oxidation of odd-chain fatty acids yields propionyl-CoA. This three-carbon compound is converted to succinyl-CoA, a citric acid cycle intermediate, by three reactions:
    (a) Propionyl-CoA carboxylase catalyzes an ATP-dependent carboxylation reaction that requires the coenzyme biotin and produces (*S*)-methylmalonyl-CoA.
    (b) Methylmalonyl-CoA racemase converts (*S*)-methylmalonyl-CoA to (*R*)-methylmalonyl-CoA.
    (c) Methylmalonyl-CoA mutase transforms (*R*)-methylmalonyl-CoA into succinyl-CoA in a reaction that requires the coenzyme 5′-deoxyadenosylcobalamin, which is derived from cobalamin (vitamin B_{12}).

14. The methylmalonyl-CoA mutase reaction rearranges the substrate's carbon skeleton. The reaction mechanism features homolytic cleavage of the C—Co bond in the coenzyme so that the C and Co atoms each retain one electron. Such homolytic cleavage is rare in biological systems; in biochemical reactions, bonds are usually broken by heterolytic cleavage in which one of the atoms acquires both electrons. The Co ion therefore functions as a generator of free radicals, which are essential for the reaction.

15. The peroxisome also carries out β oxidation. In animals, peroxisomes oxidize very long acyl chains (>22 carbons). These are shortened in the peroxisome, and β oxidation is then completed in the mitochondrion. Peroxisomal oxidation differs from oxidation in mitochondria in two ways:
    (a) No carnitine is required.
    (b) The first step of acyl-CoA oxidation by acyl-CoA oxidase involves transfer of electrons to O_2 with formation of hydrogen peroxide (H_2O_2) and a *trans*-2,3-enoyl-CoA. Thus, peroxisomal oxidation of fatty acids yields less energy than mitochondrial oxidation.

Ketone Bodies

16. Acetyl-CoA, in addition to undergoing oxidation via the citric acid cycle, can also undergo ketogenesis to form acetoacetate, D-β-hydroxybutyrate, and acetone. These water-soluble compounds are collectively called ketone bodies. Acetoacetate and D-β-hydroxybutyrate are important sources of metabolic energy under certain circumstances.

17. Ketogenesis, the formation of ketone bodies from acetyl CoA, occurs as follows:
    (a) Two acetyl-CoA units condense to form acetoacetyl-CoA in a reversal of the thiolase reaction.
    (b) Hydroxymethylglutaryl-CoA synthase catalyzes the condensation of acetoacetyl-CoA with a third acetyl-CoA unit to form β-hydroxy-β-methylglutaryl-CoA (HMG-CoA).
    (c) HMG-CoA lyase cleaves HMG-CoA to form acetyl-CoA and acetoacetate.
    (d) β-Hydroxybutyrate dehydrogenase can reduce acetoacetate to β-hydroxybutyrate in an NADH-dependent reaction.
    (e) Acetoacetate also undergoes spontaneous decarboxylation to form acetone.

18. Acetoacetate and β-hydroxybutyrate formed in the liver are transported in the bloodstream to peripheral tissues and there are converted into two acetyl-CoA units. Succinyl-CoA supplies the CoA for this process and is therefore not utilized in the citric acid cycle to form GTP and succinate.

Fatty Acid Biosynthesis

19. Although fatty acid biosynthesis involves the condensation of successive acetyl-CoA units, this metabolic pathway is distinct from β oxidation in several respects:
    (a) It is a reductive process.
    (b) It takes place in the cytosol.
    (c) It utilizes NADPH as hydrogen donor.
    (d) It uses the C_3 dicarboxylic acid derivative, malonyl-CoA, as its C_2 donor.
    (e) The growing acyl chain is attached to acyl-carrier protein (ACP) rather than to CoA.
    (f) It employs entirely different enzymes and is independently regulated.

20. In order for fatty acid biosynthesis to proceed, sufficient amounts of both acetyl-CoA and NADPH must be available in the cytosol. Acetyl-CoA, which is generated by pyruvate dehydrogenase in the mitochondrion, cannot cross the inner mitochondrial membrane to reach the cytosol. Instead, transport occurs by means of the tricarboxylate transport system in which acetyl-CoA reacts with oxaloacetate to form citrate, which readily crosses the membrane via a transporter. Once in the cytosol, citrate is converted to pyruvate in a series of reactions that liberates acetyl-CoA and also generates NADPH in a 1:1 ratio. Pyruvate then enters the mitochondrion and is converted to oxaloacetate.

21. The first committed step in fatty acid biosynthesis is catalyzed by acetyl-CoA carboxylase, a biotin-dependent enzyme, which converts acetyl-CoA to malonyl-CoA. In the first reaction

step, bicarbonate becomes covalently attached to the biotin prosthetic group. This "activated" CO_2 is then transferred from biotin to acetyl-CoA, forming malonyl-CoA.

22. In eukaryotic cells, acetyl-CoA carboxylase is regulated both allosterically and by covalent modification. Citrate allosterically increase the V_{max} of the enzyme, whereas long-chain acyl-CoAs inhibit the reaction. AMP-dependent kinase phosphorylates Ser 79, thereby inactivating the enzyme. Glucagon acts through a cAMP-dependent pathway to inhibit the enzyme, possibly by preventing its dephosphorylation. In contrast, insulin enhances enzyme activity by promoting its dephosphorylation.

23. Fatty acid synthesis from acetyl-CoA and malonyl-CoA takes place by successive cycles of six enzymatic reactions. In *E. coli,* these reactions are catalyzed by separate enzymes. In eukaryotic cells, the process occurs within fatty acid synthase, a protein whose sequence contains all the required enzyme activities and an ACP domain. Fatty acid synthase catalyzes the following reactions:
    (a) In the first two reactions, the synthase is primed by transferring an acetyl group from acetyl-CoA to a Cys SH group and by transferring a malonyl group from malonyl-CoA to the terminal SH of the phosphopantetheine prosthetic group of ACP.
    (b) In the third reaction, malonyl-ACP is decarboxylated and condenses with the acetyl group to form a β-ketoacyl-ACP.
    (c) In reactions 4 to 6, the β-keto group is reduced by NADPH to form a hydroxyl group, the hydroxyl group is eliminated in a dehydration reaction to form a double bond, and a second reduction by NADPH reduces the double bond to produce an alkyl group.
    (d) The four-carbon acyl chain is then transferred from the ACP to the enzyme Cys SH. The cycle starts anew by the transfer of another malonyl group to the now vacant ACP site in the synthase.

    Seven cycles are required to synthesize a C_{16} acyl chain (palmitate), a process that consumes 8 acetyl-CoA and 14 NADPH (which are provided by glucose oxidation via the pentose phosphate pathway). The completed saturated acyl chain is released by thioester cleavage catalyzed by the seventh enzyme activity associated with the synthase.

24. Mammalian fatty acid synthase consists of two identical monomers in a head-to-tail association such that the Cys-linked acyl group in one polypeptide is located close to the phosphopantetheine moiety in the other. This allows the synthase dimer to simultaneously synthesize two acyl chains.

25. Palmitate may be lengthened and desaturated by elongases and desaturases, respectively. However, because animals, unlike plants, cannot introduce a double bond beyond C9 in an acyl chain, linoleic acid (9,12-*cis*-octadecadienoic acid) must be obtained from the diet. It is therefore called an essential fatty acid.

26. The synthesis of triacylglycerols begins with successive acylations of glycerol-3-phosphate to yield lysophosphatidic acid and then phosphatidic acid. Dephosphorylation produces 1,2-diacylglycerol, which then accepts another fatty acyl group. An alternative pathway involves

acylation of dihydroxyacetone phosphate, followed by an NADPH-dependent reduction to produce lysophosphatidic acid.

Regulation of Fatty Acid Metabolism

27. Triacylglycerol metabolism, like that of glycogen, is important for the well-being of the whole organism and is regulated by hormones. Fatty acid oxidation is controlled by the rate of triacylglycerol hydrolysis in adipose tissue by hormone-sensitive triacylglycerol lipase. The lipase is stimulated by glucagon through cAMP-dependent phosphorylation, which also inhibits acetyl-CoA carboxylase, so fatty acid oxidation is enhanced and fatty acid synthesis is inhibited.

28. Insulin opposes the effects of glucagon by reducing cAMP levels, thereby inactivating triacylglycerol lipase and stimulating fatty acid synthesis. The ratio of glucagon to insulin therefore controls the status of fatty acid metabolism.

29. These mechanisms of short-term regulation are complemented by long-term hormonal regulation of fatty acid metabolism, which alters the levels of key enzymes such as acetyl-CoA carboxylase and triacylglycerol lipase.

Membrane Lipid Synthesis

30. The formation of choline- and ethanolamine-containing glycerophospholipids involves three enzymatic steps:
    (a) The phosphorylation of the nitrogen-containing base.
    (b) Activation of the phosphocholine or phosphoethanolamine by CTP to form CDP–choline or CDP–ethanolamine.
    (c) Transfer of the activated base to 1,2-diacylglycerol to form the phospholipid.

31. The synthesis of phosphatidylglycerol and phosphatidylinositol involves activation of phosphatidic acid by reaction with CTP to form CDP–diacylglycerol. The activated lipid is then transferred to glycerol-3-phosphate or inositol. Cardiolipin is synthesized from two phosphatidylglycerol.

32. Enzymes that acylate glycerol-3-phosphate show a preference for introducing a saturated fatty acid in position 1 and an unsaturated fatty acid in position 2. However, there must be additional reactions, catalyzed by phospholipases and acyltransferases, that result in the exchange of acyl groups, to account for the fatty acid compositions of all membrane phospholipids.

33. In sphingolipid synthesis, ceramide (*N*-acylsphingosine) is formed in four reactions from palmitoyl-CoA and serine. Phosphatidylcholine donates its phosphocholine group to ceramide to produce sphingomyelin. Ceramide can also be glycosylated, with UDP–glucose or UDP–galactose serving as the sugar donor, to form cerebrosides. Additional glycosylation of cerebrosides generates more complex sphingoglycolipids such as globosides and gangliosides.

Cholesterol Metabolism

34. All 27 carbon atoms of cholesterol are derived from acetate. The major stages in cholesterol formation are:
    (a) Acetate is converted to hydroxymethylglutaryl-CoA (HMG-CoA) and then via mevalonate to an isoprene unit, isopentenyl pyrophosphate.
    (b) Condensation of six isoprene units forms squalene, a linear 30-carbon compound.
    (c) Squalene is oxidized and cyclized to form lanosterol.
    (d) Further modification and removal of three carbons yields cholesterol.

35. After its synthesis in the liver, cholesterol may be transformed into bile acids or converted to cholesteryl esters. Both endogenously synthesized and dietary cholesterol are esterified, packaged in VLDL, and transported through the bloodstream. Peripheral tissues take up LDL (which are derived from VLDL) by receptor-mediated endocytosis, after which the acyl groups of cholesteryl esters are removed by hydrolysis, yielding free cholesterol. The cholesterol may become a cell membrane constituent, it may be re-esterified for intracellular storage, or it may be transported to the liver by HDL.

36. Cholesterol is essential for cell membrane integrity, yet excess cholesterol may be harmful to the organism. Thus, its biosynthesis, utilization, and cellular distribution are carefully controlled. Cholesterol metabolism is regulated in two ways:
    (a) Cholesterol biosynthesis is controlled by HMG-CoA reductase, which catalyzes the rate-limiting conversion of HMG-CoA to mevalonate. In the short term, this enzyme is inactivated by phosphorylation (catalyzed by AMP-dependent kinase, the same enzyme that inactivates acetyl-CoA carboxylase) and activated by dephosphorylation. Long-term regulation involves changes in the level of enzymes in inverse proportion to the concentrations of mevalonate and cholesterol-containing LDL.
    (b) Cholesterol transport and removal from blood is governed largely by the activity of LDL receptors on the liver cell surface, which in turn depends on the number of LDL receptors and hence on the rate of receptor synthesis.

37. High blood cholesterol results from the genetic disease familial hypercholesterolemia (which is characterized by an absence of LDL receptors) or by high dietary cholesterol intake (which tends to repress LDL receptor synthesis).

Guide to Study Exercises (text p. 609)

1. Bile acids (also called bile salts), which are amphipathic molecules, help solubilize dietary lipids so that they become accessible to intestinal lipases. Micelles containing bile acids also take up lipid-soluble vitamins and the products of lipid digestion so that these hydrophobic molecules can pass through the aqueous solution to reach the intestinal cell surface, where they are absorbed. (Section 19-1A)

2. Lipoproteins package hydrophobic and amphipathic lipids into water-soluble particles for transport in the bloodstream. Chylomicrons transport dietary lipids from the intestine through the lymphatic system to the bloodstream. They deliver triacylglycerols to skeletal muscle and adipose tissue, and cholesterol to the liver. The liver synthesizes VLDL particles, which also contain triacylglycerols and cholesterol. The triacylglycerols are degraded by lipoprotein lipase in the capillaries so that free fatty acids can be absorbed by cells. As it gives up its triacylglycerols and becomes smaller and denser, a VLDL becomes an IDL and then an LDL before being taken up by the liver. HDL particles transport cholesterol from the tissues to the liver. They are assembled in the plasma and contain mostly cholesteryl esters, which they transfer to VLDL. (Section 19-1B)

3. Fatty acids are first activated by linking them to CoA via a "high-energy" thioester bond, whose formation consumes the free energy of one phosphoanhydride bond of ATP. After transport into the mitochondrion, the acyl-CoA is degraded, two carbons at a time, in a process called β oxidation. An α,β double bond forms by dehydrogenation, and the electrons are transferred to the mitochondrial electron-transport chain. Next, water is added across the double bond to form a 3-hydroxyacyl-CoA. NAD^+-dependent dehydrogenation then yields a β-ketoacyl-CoA and NADH. Finally, the C_α—C_β bond is cleaved by attack of a second CoA, eliminating acetyl-CoA and producing an acyl-CoA two carbons shorter than the original substrate. These last four reactions are repeated until the entire acyl chain has been degraded to acetyl units. (Section 19-2)

4. Unsaturated fatty acids are degraded by the β oxidation enzymes until their double bonds prevent their binding to these enzymes. The acyl groups must then be enzymatically transformed to substrates of the β oxidation enzymes. When a β,γ double bond is encountered, it is isomerized to an α,β double bond, which is a substrate for enoyl-CoA hydratase. When a 2,4-dienoyl-CoA (with two consecutive double bonds) is encountered, the γ,δ double bond is eliminated by NADPH-dependent reduction.

   Fatty acids that contain an odd number of carbon atoms yield the C_3 derivative propionyl-CoA in the final round of β oxidation. The propionyl group is carboxylated to a C_4 methylmalonyl group in an ATP-dependent reaction. Next, the configuration of the methylmalonyl group is inverted by a racemase, and then the carbon skeleton is rearranged to a succinyl group by methylmalonyl-CoA mutase in a reaction that requires coenzyme B_{12}. (Sections 19-2D and E)

5. Ketone bodies are synthesized from acetyl-CoA in liver mitochondria. First, 2 acetyl-CoA are condensed, in a reaction that is the reversal of the last step of β oxidation, to acetoacetyl-CoA. A third acetyl-CoA is added, generating the C_6 group-containing β-hydroxy-β-methylglutaryl-CoA (HMG-CoA). This compound is degraded by HMG-CoA lyase to produce the ketone body acetoacetate and acetyl-CoA. Acetoacetate can be reduced by NADH to produce the ketone body β-hydroxybutyrate, or it may be nonenzymatically decarboxylated to acetone + CO_2.

   Acetoacetate and β-hydroxybutyrate travel from the liver to tissues to be used as alternative fuels to glucose. β-Hydroxybutyrate is oxidized by NAD^+ to produce

Cholesterol Metabolism

34. All 27 carbon atoms of cholesterol are derived from acetate. The major stages in cholesterol formation are:
    (a) Acetate is converted to hydroxymethylglutaryl-CoA (HMG-CoA) and then via mevalonate to an isoprene unit, isopentenyl pyrophosphate.
    (b) Condensation of six isoprene units forms squalene, a linear 30-carbon compound.
    (c) Squalene is oxidized and cyclized to form lanosterol.
    (d) Further modification and removal of three carbons yields cholesterol.

35. After its synthesis in the liver, cholesterol may be transformed into bile acids or converted to cholesteryl esters. Both endogenously synthesized and dietary cholesterol are esterified, packaged in VLDL, and transported through the bloodstream. Peripheral tissues take up LDL (which are derived from VLDL) by receptor-mediated endocytosis, after which the acyl groups of cholesteryl esters are removed by hydrolysis, yielding free cholesterol. The cholesterol may become a cell membrane constituent, it may be re-esterified for intracellular storage, or it may be transported to the liver by HDL.

36. Cholesterol is essential for cell membrane integrity, yet excess cholesterol may be harmful to the organism. Thus, its biosynthesis, utilization, and cellular distribution are carefully controlled. Cholesterol metabolism is regulated in two ways:
    (a) Cholesterol biosynthesis is controlled by HMG-CoA reductase, which catalyzes the rate-limiting conversion of HMG-CoA to mevalonate. In the short term, this enzyme is inactivated by phosphorylation (catalyzed by AMP-dependent kinase, the same enzyme that inactivates acetyl-CoA carboxylase) and activated by dephosphorylation. Long-term regulation involves changes in the level of enzymes in inverse proportion to the concentrations of mevalonate and cholesterol-containing LDL.
    (b) Cholesterol transport and removal from blood is governed largely by the activity of LDL receptors on the liver cell surface, which in turn depends on the number of LDL receptors and hence on the rate of receptor synthesis.

37. High blood cholesterol results from the genetic disease familial hypercholesterolemia (which is characterized by an absence of LDL receptors) or by high dietary cholesterol intake (which tends to repress LDL receptor synthesis).

Guide to Study Exercises (text p. 609)

1. Bile acids (also called bile salts), which are amphipathic molecules, help solubilize dietary lipids so that they become accessible to intestinal lipases. Micelles containing bile acids also take up lipid-soluble vitamins and the products of lipid digestion so that these hydrophobic molecules can pass through the aqueous solution to reach the intestinal cell surface, where they are absorbed. (Section 19-1A)

2. Lipoproteins package hydrophobic and amphipathic lipids into water-soluble particles for transport in the bloodstream. Chylomicrons transport dietary lipids from the intestine through the lymphatic system to the bloodstream. They deliver triacylglycerols to skeletal muscle and adipose tissue, and cholesterol to the liver. The liver synthesizes VLDL particles, which also contain triacylglycerols and cholesterol. The triacylglycerols are degraded by lipoprotein lipase in the capillaries so that free fatty acids can be absorbed by cells. As it gives up its triacylglycerols and becomes smaller and denser, a VLDL becomes an IDL and then an LDL before being taken up by the liver. HDL particles transport cholesterol from the tissues to the liver. They are assembled in the plasma and contain mostly cholesteryl esters, which they transfer to VLDL. (Section 19-1B)

3. Fatty acids are first activated by linking them to CoA via a "high-energy" thioester bond, whose formation consumes the free energy of one phosphoanhydride bond of ATP. After transport into the mitochondrion, the acyl-CoA is degraded, two carbons at a time, in a process called β oxidation. An α,β double bond forms by dehydrogenation, and the electrons are transferred to the mitochondrial electron-transport chain. Next, water is added across the double bond to form a 3-hydroxyacyl-CoA. NAD^+-dependent dehydrogenation then yields a β-ketoacyl-CoA and NADH. Finally, the C_α—C_β bond is cleaved by attack of a second CoA, eliminating acetyl-CoA and producing an acyl-CoA two carbons shorter than the original substrate. These last four reactions are repeated until the entire acyl chain has been degraded to acetyl units. (Section 19-2)

4. Unsaturated fatty acids are degraded by the β oxidation enzymes until their double bonds prevent their binding to these enzymes. The acyl groups must then be enzymatically transformed to substrates of the β oxidation enzymes. When a β,γ double bond is encountered, it is isomerized to an α,β double bond, which is a substrate for enoyl-CoA hydratase. When a 2,4-dienoyl-CoA (with two consecutive double bonds) is encountered, the γ,δ double bond is eliminated by NADPH-dependent reduction.

   Fatty acids that contain an odd number of carbon atoms yield the C_3 derivative propionyl-CoA in the final round of β oxidation. The propionyl group is carboxylated to a C_4 methylmalonyl group in an ATP-dependent reaction. Next, the configuration of the methylmalonyl group is inverted by a racemase, and then the carbon skeleton is rearranged to a succinyl group by methylmalonyl-CoA mutase in a reaction that requires coenzyme B_{12}. (Sections 19-2D and E)

5. Ketone bodies are synthesized from acetyl-CoA in liver mitochondria. First, 2 acetyl-CoA are condensed, in a reaction that is the reversal of the last step of β oxidation, to acetoacetyl-CoA. A third acetyl-CoA is added, generating the C_6 group-containing β-hydroxy-β-methylglutaryl-CoA (HMG-CoA). This compound is degraded by HMG-CoA lyase to produce the ketone body acetoacetate and acetyl-CoA. Acetoacetate can be reduced by NADH to produce the ketone body β-hydroxybutyrate, or it may be nonenzymatically decarboxylated to acetone + CO_2.

   Acetoacetate and β-hydroxybutyrate travel from the liver to tissues to be used as alternative fuels to glucose. β-Hydroxybutyrate is oxidized by NAD^+ to produce

acetoacetate. The acetoacetate is then linked to CoA donated by succinyl-CoA. A free CoA group then attacks the acetoacetyl-CoA to produce 2 acetyl-CoA. (Section 19-3)

6. A fatty acid, in the form of acyl-CoA, is transported out of the mitochondrion by a shuttle system. The acyl portion of acyl-CoA is transferred to carnitine, and the acyl-carnitine is transported across the inner mitochondrial membrane by a carrier protein. In the matrix, the acyl group is transferred back to CoA from the mitochondrial pool of CoA to produce the original acyl-CoA, and carnitine returns to the cytosol.

   Acetyl-CoA produced in the matrix is shuttled back to the cytosol, where it can be used for fatty acid synthesis. First, acetyl-CoA reacts with mitochondrial oxaloacetate to produce citrate (the first step of the citric acid cycle). Citrate is transported across the inner mitochondrial membrane by a transport protein. In the cytosol, ATP-citrate lyase catalyzes the conversion of citrate back to oxaloacetate and acetyl-CoA, in a reaction that consumes ATP. The shuttle system is regenerated when cytosolic oxaloacetate is reduced to malate and then oxidatively decarboxylated to pyruvate (this reaction sequence has the net effect of converting a reducing equivalent from NADH to NADPH, the form required for fatty acid synthesis). After transport back into the matrix, the pyruvate is carboxylated to regenerate oxaloacetate. (Sections 19-2B and 4A)

7. Fatty acid oxidation and synthesis are both four-step cyclic pathways that proceed in increments of C_2 units, and many of their intermediates are chemically similar. The differences are: (a) Oxidation occurs in the mitochondrion and synthesis in the cytosol; (b) the acyl group is linked to CoA for oxidation and to ACP for synthesis; (c) FAD and NAD^+ are electron acceptors in oxidation, whereas NADPH is the electron donor in synthesis; and (d) synthesis but not oxidation involves a C_3 unit, malonyl-CoA (this is critical for the regulation of fatty acid synthesis). (Section 19-4 and Figure 19-19)

8. Fatty acid metabolism is regulated by controlling the availability of fatty acids and by controlling the activity of acetyl-CoA carboxylase. The concentration of fatty acids in the blood depends on the rate of triacylglycerol hydrolysis by hormone-sensitive lipase. Glucagon, epinephrine, and norepinephrine (which signal low glucose levels) lead to activation of the lipase and thereby increase the supply of fatty acids for energy production by β oxidation. At the same time, these hormones, via the cAPK pathway, inactivate acetyl-CoA carboxylase, which prevents fatty acid synthesis from acetyl-CoA. Insulin, which reflects high glucose levels, reverses the effects of the cAPK pathway. This results in inhibition of hormone-sensitive lipase (which decrease the mobilization of fatty acids) and activates acetyl-CoA carboxylase (which promotes fatty acid synthesis).

   Long-term regulation of fatty acid metabolism is accomplished through changes in enzyme concentrations. Insulin stimulates the synthesis of acetyl-CoA carboxylase and fatty acid synthase, whereas starvation inhibits the synthesis of these enzymes. (Section 19-5)

9. Triacylglycerols can be synthesized from acyl-CoA and glycerol-3-phosphate to produce first a lysophosphatidic acid. Triacylglycerol synthesis that begins with dihydroxyacetone phosphate rather than glycerol-3-phosphate requires a reduction step. Addition of a second

acyl group to lysophosphatidic acid and removal of the phosphate group yields diacylglycerol. Addition of a third acyl group yields a triacylglycerol.

Diacylglycerols can be converted to glycerophospholipids in two ways. Phosphatidylethanolamine and phosphatidylcholine are synthesized by adding the respective head group, in the form of a CDP adduct, to 1,2-diacylglycerol. Phosphatidylserine is produced by exchanging the ethanolamine group of phosphatidylethanolamine for serine. In phosphatidylinositol and phosphatidylglycerol synthesis, the diacylglycerol group is activated by linkage to CDP before being linked to inositol or glycerol-3-phosphate.

Sphingolipid synthesis begins with the condensation of palmitoyl-CoA with serine to yield 3-ketosphinganine. Reduction and transfer of an acyl group from acyl-CoA produce dihydroceramide, which is then oxidized to produce the double bond in ceramide. A head group (phosphocholine in sphingomyelin or a glycosyl unit in cerebrosides) is then added. Additional glycosylation yields the elaborate head groups of gangliosides and globosides. (Sections 19-4E and 19-6)

10. Cholesterol is synthesized entirely from acetyl-CoA. HMG-CoA, formed from acetyl-CoA by cytosolic counterparts of the mitochondrial enzymes that synthesize ketone bodies, is converted to the isoprenoid derivative isopentenyl pyrophosphate in a series of four reactions that consume 2 NADPH and 3 ATP and include a decarboxylation. Two C_5 isopentenyl pyrophosphate (one isomerized to dimethylallyl pyrophosphate) condense to form the C_{10} compound geranyl phosphate. Addition of another isopentenyl group yields the C_{15} compound farnesyl pyrophosphate. Two of these molecules condense to form the linear C_{30} hydrocarbon squalene. Squalene epoxidase oxidizes squalene to produce an epoxide, then squalene oxidocyclase catalyzes a series of cyclizations that convert the epoxide to the four-ring lanosterol. Further oxidation and the elimination of three carbons, in 19 steps, yields cholesterol. (Section 19-7A)

11. Serum cholesterol occurs in the form of lipoproteins. These are removed from the circulation via receptor-mediated endocytosis when a lipoprotein binds to the LDL receptor in liver cells. Consequently, the number of LDL receptors and therefore the rate of uptake of lipoproteins regulates the level of circulating cholesterol. (Section 19-7C)

Questions

1. From a chemical perspective, why is the energy content of fats so much greater than that of carbohydrates or proteins?

Lipid Digestion, Absorption, and Transport

2. Why do individuals who have their gall bladders removed sometimes encounter difficulties in digesting fats?

3. How do pancreatic lipase and phospholipase A_2 differ in the mechanism by which they promote hydrolysis of glycerolipids at interfaces?

4. Match each term on the left with its description on the right.

____ Bile acid	A. Helps bind lipase to the lipid–water interface
____ Intestinal fatty acid–binding protein	B. Hydrolyzes phospholipids to yield lysophospholipids and free fatty acids
____ Albumin	C. Forms micelles that take up nonpolar lipid degradation products and transports them through the intestinal wall
____ Phospholipase A_2	D. Transports lipid digestion products through the lymphatic system and then the bloodstream to the tissues
____ Colipase	E. Transports through the bloodstream free fatty acids released from adipose tissue stores
____ Chylomicrons	F. Forms complexes with free fatty acids to shield intestinal cells from their detergent-like effects

5. Determine the order of the following events involving lipoprotein-mediated transport of dietary triacylglycerols and cholesterol.

____ Triacylglycerols are removed from circulating VLDL by lipoprotein lipase.
____ Chylomicrons are transported through the lymphatic system and enter the bloodstream.
____ Chylomicrons are formed in the intestinal mucosa.
____ Cells take up cholesterol via receptor-mediated LDL endocytosis.
____ Chylomicrons are degraded by lipoprotein lipase to chylomicron remnants.
____ LDL components are rapidly degraded by lysosomal enzymes.
____ VLDL are synthesized in liver.

Fatty Acid Oxidation

6. What products would be isolated from urine when dogs are fed (a) phenylheptanoic acid and (b) phenyloctanoic acid? How does this experiment, originally performed by Knoop, shed light on the process of fatty acid oxidation?

7. A patient develops an enlarged fatty liver and low blood glucose. These symptoms can be partially overcome only when massive amounts of carnitine are included in the diet. What enzyme defect might be responsible for this problem?

8. Which of the following statements is(are) correct? In the β oxidation of fatty acids,
   (a) The activation of fatty acids by acyl-CoA synthetase is driven by the hydrolysis of pyrophosphate.
   (b) The reaction catalyzed by acyl-CoA dehydrogenase uses FAD as electron acceptor.
   (c) The reaction catalyzed by enoyl-CoA hydratase produces a 3-D-hydroxyacyl-CoA.

9. Calculate the yield of ATP when one mole of stearic acid is completely oxidized to CO_2 and H_2O.

10. In the β oxidation of oleic acid, the presence of the double bond at the _____ position necessitates modification of the β oxidation cycle. The modification occurs after the _____ round of β oxidation because the _____ enoyl-CoA is not a substrate for _____. The problem is overcome by the action of the enzyme _____ which converts the _____ bond to a _____ bond so that β oxidation can continue.

11. In the β oxidation of linoleic acid, a second difficulty presents itself because of the additional double bond in the _____ position. In this case, after the fifth round of oxidation, a_____ CoA is formed, which is a poor substrate for _____. In mammals, two reactions are necessary for β oxidation to resume. In the first, NADPH-dependent _____ reductase reduces one double bond to form a _____ , and then the action of a _____ isomerase yields a _____ CoA that can participate in β oxidation.

12. Describe the role of cobalamin (vitamin B_{12}) in the methylmalonyl-CoA mutase reaction.

13. Explain why succinyl-CoA arising from the oxidation of odd-chain fatty acids cannot be directly oxidized by the citric acid cycle. How can it be further degraded?

14. Why does the β oxidation of fatty acids in peroxisomes yield less ATP than the corresponding process in mitochondria?

Ketone Bodies

15. Infants have high levels of ketone bodies in their blood and abundant 3-ketoacyl-CoA transferase in their tissues (except in liver) prior to weaning. What nutritional advantage does this confer?

16. Calculate the ATP that is produced when linoleic acid (9,12-octadecadienoic acid; 18:2) is (a) oxidized to CO_2 and H_2O or (b) converted to the ketone body acetoacetate in the liver and then oxidized to CO_2 and H_2O in the peripheral tissues.

4. Match each term on the left with its description on the right.

___ Bile acid	A. Helps bind lipase to the lipid–water interface
___ Intestinal fatty acid–binding protein	B. Hydrolyzes phospholipids to yield lysophospholipids and free fatty acids
___ Albumin	C. Forms micelles that take up nonpolar lipid degradation products and transports them through the intestinal wall
___ Phospholipase A_2	D. Transports lipid digestion products through the lymphatic system and then the bloodstream to the tissues
___ Colipase	E. Transports through the bloodstream free fatty acids released from adipose tissue stores
___ Chylomicrons	F. Forms complexes with free fatty acids to shield intestinal cells from their detergent-like effects

5. Determine the order of the following events involving lipoprotein-mediated transport of dietary triacylglycerols and cholesterol.

___ Triacylglycerols are removed from circulating VLDL by lipoprotein lipase.
___ Chylomicrons are transported through the lymphatic system and enter the bloodstream.
___ Chylomicrons are formed in the intestinal mucosa.
___ Cells take up cholesterol via receptor-mediated LDL endocytosis.
___ Chylomicrons are degraded by lipoprotein lipase to chylomicron remnants.
___ LDL components are rapidly degraded by lysosomal enzymes.
___ VLDL are synthesized in liver.

Fatty Acid Oxidation

6. What products would be isolated from urine when dogs are fed (a) phenylheptanoic acid and (b) phenyloctanoic acid? How does this experiment, originally performed by Knoop, shed light on the process of fatty acid oxidation?

7. A patient develops an enlarged fatty liver and low blood glucose. These symptoms can be partially overcome only when massive amounts of carnitine are included in the diet. What enzyme defect might be responsible for this problem?

8. Which of the following statements is(are) correct? In the β oxidation of fatty acids,
   (a) The activation of fatty acids by acyl-CoA synthetase is driven by the hydrolysis of pyrophosphate.
   (b) The reaction catalyzed by acyl-CoA dehydrogenase uses FAD as electron acceptor.
   (c) The reaction catalyzed by enoyl-CoA hydratase produces a 3-D-hydroxyacyl-CoA.

9. Calculate the yield of ATP when one mole of stearic acid is completely oxidized to CO_2 and H_2O.

10. In the β oxidation of oleic acid, the presence of the double bond at the _____ position necessitates modification of the β oxidation cycle. The modification occurs after the _____ round of β oxidation because the _____ enoyl-CoA is not a substrate for _____. The problem is overcome by the action of the enzyme _____ which converts the _____ bond to a _____ bond so that β oxidation can continue.

11. In the β oxidation of linoleic acid, a second difficulty presents itself because of the additional double bond in the _____ position. In this case, after the fifth round of oxidation, a_____ CoA is formed, which is a poor substrate for _____. In mammals, two reactions are necessary for β oxidation to resume. In the first, NADPH-dependent _____ reductase reduces one double bond to form a _____ , and then the action of a _____ isomerase yields a _____ CoA that can participate in β oxidation.

12. Describe the role of cobalamin (vitamin B_{12}) in the methylmalonyl-CoA mutase reaction.

13. Explain why succinyl-CoA arising from the oxidation of odd-chain fatty acids cannot be directly oxidized by the citric acid cycle. How can it be further degraded?

14. Why does the β oxidation of fatty acids in peroxisomes yield less ATP than the corresponding process in mitochondria?

Ketone Bodies

15. Infants have high levels of ketone bodies in their blood and abundant 3-ketoacyl-CoA transferase in their tissues (except in liver) prior to weaning. What nutritional advantage does this confer?

16. Calculate the ATP that is produced when linoleic acid (9,12-octadecadienoic acid; 18:2) is (a) oxidized to CO_2 and H_2O or (b) converted to the ketone body acetoacetate in the liver and then oxidized to CO_2 and H_2O in the peripheral tissues.

17. Write equations for ketone body synthesis and degradation. What is the net result of combined synthesis and degradation?

Fatty Acid Biosynthesis

18. A rat liver cytosol preparation is incubated with acetate and all cofactors necessary for the biosynthesis of palmitate. Which carbon atoms of palmitate will be isotopically labeled when the preparation contains (a) $H^{14}CO_3^-$ or (b) $^{14}CH_3COO^-$?

19. What are the advantages of the multifunctional "head-to-tail" dimer structure of animal fatty acid synthase in the formation of long-chain fatty acids?

20. Which of the following statements is(are) correct? The transport of acetyl-CoA from the mitochondrial matrix to the cytosol for fatty acid biosynthesis:
(a) is necessary because the inner mitochondrial membrane is impermeable to acetyl-CoA.
(b) uses the tricarboxylate transport system, which operates in both directions.
(c) can generate equal numbers of acetyl-CoA and NADPH molecules in the cytosol.

Regulation of Fatty Acid Metabolism

21. Match each term on the left with its function in the short-term regulation of fatty acid metabolism.

____ Citrate	A. Activates acetyl-CoA carboxylase
____ cAMP-dependent phosphorylation	B. Inhibits carnitine palmitoyl transferase
____ Palmitate	C. Activates hormone-sensitive lipase
____ Malonyl-CoA	D. Inhibits acetyl-CoA carboxylase

Membrane Lipid Synthesis

22. Match each term on the left with its metabolic role on the right.

____ CDP–diacyylglycerol	A. Precursor of cardiolipin
____ Ceramide	B. Precursor of phosphatidylcholine
____ Phosphatidylglycerol	C. Contains a vinyl ether linkage
____ Plasmalogen	D. Precursor of sphingomyelin
____ CDP–Choline	E. Precursor of phosphatidylinositol

23. When tissues are incubated *in vitro* with ^{32}P-labeled P_i, the incorporation of radioactivity into phospholipids can be readily demonstrated. High concentrations of the drug propranolol dramatically alter the pattern of phospholipid labeling, such that radioactivity in phosphatidylcholine and phosphatidylethanolamine markedly declines while that in phosphatidylinositol and phosphatidylglycerol is greatly elevated. From this information and your knowledge of phospholipid biosynthetic pathways, determine which enzymatic reaction

is principally affected by propranolol and explain how the drug alters phospholipid biosynthesis.

24. A metabolic disease may result from an enzyme deficiency that prevents the synthesis of an essential metabolite or that prevents the normal breakdown of a metabolite. Which type of defect is exhibited in Tay–Sachs disease and what enzyme is affected?

Cholesterol Metabolism

25. What are the two principal ways in which the cholesterol needs of many tissues are met?

26. Place the following intermediates in cholesterol biosynthesis in the correct order: squalene, farnesyl pyrophosphate, 2,3-oxidosqualene, hydroxymethylglutaryl-CoA (HMG-CoA), lanosterol, mevalonate, geranyl pyrophosphate.

27. Explain how the regulation of HMG-CoA reductase, the principal control site for cholesterol synthesis, can conserve cellular ATP.

28. Which of the following statements is(are) correct? Inhibitors of HMG-CoA reductase are used to decrease serum cholesterol levels. Such inhibitors would also:
(a) reduce the intracellular level of mevalonate.
(b) reduce the synthesis of LDL receptors.
(c) reduce the synthesis of ubiquinone.

Answers to Questions

1. The energy content of fats is greater because the carbon atoms of fatty acids are more reduced and therefore release more free energy upon oxidation than the carbons of carbohydrates or proteins. In addition, fats can be stored in large amounts in anhydrous form. In contrast, proteins cannot be stored in large amounts, and carbohydrates require a large volume for hydration.

2. The gall bladder stores and secretes bile acids, which are essential for emulsifying triacylglycerols and thereby promoting their hydrolysis. In addition, bile acids and released fatty acids are components of mixed micelles that are absorbed across the intestinal wall.

3. Both pancreatic lipase and phospholipase A_2 hydrolyze their substrates at the lipid–water interface, where the enzymes undergo interfacial activation. Pancreatic lipase binds to the interface only when complexed with pancreatic colipase. In the presence of a lipid micelle, the lipase undergoes a structural change that exposes the active site, forms an oxyanion hole, and, in conjunction with colipase, creates a large hydrophobic surface near the active site that helps to bind the lipase–colipase complex to the lipid.

Phospholipase A_2 does not alter its conformation but instead has a hydrophobic channel that enables a micellar phospholipid to gain access to the enzyme active site without having to pass through the aqueous phase.

4. C Bile acid
F Intestinal fatty acid–binding protein
E Albumin
B Phospholipase A_2
A Colipase
D Chylomicrons

5. 5 Triacylglycerols are removed from circulating VLDL by lipoprotein lipase.
2 Chylomicrons are transported through the lymphatic system and enter the bloodstream.
1 Chylomicrons are formed in the intestinal mucosa.
6 Cells take up cholesterol via receptor-mediated LDL endocytosis.
3 Chylomicrons are degraded by lipoprotein lipase to chylomicron remnants.
7 LDL components are rapidly degraded by lysosomal enzymes.
4 VLDL are synthesized in liver.

6. (a) Hippuric acid (a benzoic acid derivative). (b) Phenylaceturic acid (a phenylacetic acid derivative). This experiment suggested that fatty acids are degraded by oxidation and removal of successive two-carbon fragments.

7. The patient probably has a carnitine palmitoyl transferase I deficiency. As a result, acyl-CoA transport across the mitochondrial membrane is inadequate. Fatty acids released from adipose tissue stores would accumulate in the liver. Glucose levels would fall because it would be the primary metabolic fuel in the absence of oxidizable fatty acids. Treatment with high doses of carnitine would elevate tissue carnitine levels and hence enable some acyl-carnitine to form and enter the mitochondrion so that the acyl group could be oxidized by β oxidation.

8. (a) and (b) are correct. (c) is incorrect because the product is 3-L-hydroxyacyl-CoA.

9. 146 ATP. Stearic acid (an 18:0 fatty acid) is first activated by conversion to stearoyl-CoA, with the consumption of 2 equivalents of ATP. Stearoyl-CoA is then degraded by eight rounds of β oxidation to form 9 acetyl-CoA, 8 $FADH_2$, and 8 NADH. Oxidation of each acetyl-CoA by the citric acid cycle yields GTP, NADH, and $FADH_2$. Reoxidation of the $FADH_2$ and NADH, with the transfer of electrons to O_2 to form H_2O, yields ATP by oxidative phosphorylation. The stoichiometry of ATP production can be summarized as follows:

Process	*Reduced coenzymes formed*	*ATPs formed*
Fatty acid activation		–2
8 rounds of β oxidation	8 $FADH_2$	16
	8 NADH	24
9 rounds of citric acid cycle		9 (GTP)
	27 NADH	81
	9 $FADH_2$	18
Total		146

10. The missing terms are: cis Δ^9; third; cis Δ^3; enoyl-CoA hydratase; enoyl-CoA isomerase; cis Δ^3; trans Δ^2.

11. The missing terms are: cis Δ^{12}; 2,4-dienoyl-; enoyl-CoA hydratase; 2,4-dienoyl-CoA; *trans*-Δ^3-enoyl-CoA; 3,2-enoyl-CoA; *trans*-Δ^2-enoyl.

12. Cobalamin is essential for converting (*R*)-methylmalonyl-CoA to succinyl-CoA. The cobalt alternates between the Co(III) and Co(II) states and therefore functions as a free radical generator. The weak carbon—cobalt(III) bond of the coenzyme undergoes homolytic cleavage such that the carbon and cobalt each acquire one electron. This yields a deoxyadenosyl radical that can abstract a hydrogen atom from methylmalonyl-CoA, which then rearranges to form succinyl-CoA.

13. Since citric acid cycle intermediates function as catalysts and are continuously regenerated, the net oxidation of succinyl-CoA can occur only if it is converted to another compound that is oxidized by the operation of the cycle, such as pyruvate or acetyl-CoA. This is accomplished by transforming succinyl-CoA to malate and then transporting the malate to the cytosol, where it is oxidatively decarboxylated to pyruvate and CO_2 in a reaction catalyzed by malic enzyme. Pyruvate can then be transported into the mitochondrion and oxidized via pyruvate dehydrogenase and the citric acid cycle.

14. The first step in β oxidation in peroxisomes is catalyzed by acyl-CoA oxidase rather than acyl-CoA dehydrogenase as occurs in mitochondria. In the acyl-CoA oxidase reaction, electrons from acyl-CoA are transferred directly to O_2 to form H_2O_2 and therefore do not pass through the electron-transport chain with concomitant formation of ATP. For this reason, peroxisomal β oxidation yields less ATP than mitochondrial β oxidation.

15. The large intake of milk means that fat supplies a major portion of the calories consumed by infants prior to weaning. The oxidation of fatty acids produces abundant acetyl-CoA which is partly converted to ketone bodies in the liver. The ketone bodies, like glucose, are water-

soluble and easily transported. They are therefore readily available as metabolic fuels to support the rapid growth and development of the infant.

16. (a) The net production of ATP from the complete oxidation of linoleic acid can be calculated as in Problem 9. The total yield is 141 ATP. This is 5 less than for the oxidation of stearate because the first double bond of linoleate does not need an FAD to reduce it ($FADH_2$ is equivalent to 2 ATP) and the reduction of its second double bond consumes an NADPH (which is equivalent to ~3 ATP).

(b) If the 9 acetyl-CoA generated by the β oxidation of linoleic acid were instead converted to ketone bodies, 4.5 molecules of acetoacetate would be formed. The transformation of acetoacetate back into acetyl-CoA does not directly consume ATP. However, it involves the consumption of 4.5 succinyl-CoA, which would otherwise provide the energy for a substrate level phosphorylation. Therefore, the net yield of ATP for this metabolic route can be considered to be 141 – 4.5 = 136.5 ATP.

17. ***Ketogenesis:***

2 Acetyl-CoA + H_2O → acetoacetate + 2 CoASH

Ketone body breakdown:

Acetoacetate + succinyl-CoA + CoASH → 2 acetyl-CoA + succinate

Combined synthesis and breakdown:

Succinyl-CoA + H_2O → succinate + CoASH

18. (a) $H^{14}CO_3^-$ is incorporated into malonyl-CoA by the action of acetyl-CoA carboxylase, but the labeled carboxyl group is subsequently eliminated as $^{14}CO_2$ during condensation of malonyl-ACP with the growing fatty acyl chain. Therefore, the biosynthesized palmitate will be unlabeled.

(b) The radioactive methyl carbon in acetate will label each even-numbered carbon atom in the completed fatty acid.

19. The proximity of multiple enzyme activities involved in fatty acid synthesis on a single polypeptide chain may enhance the efficiency of the process. Moreover, the head-to-tail orientation of the two synthase subunits may enable two fatty acyl chains to be synthesized simultaneously: At each end of the dimer, an acyl group attached to the enzyme–Cys residue on one subunit is extended by addition of an acetyl group from malonyl-ACP on the other subunit.

20. (a) and (c) are correct. (b) is incorrect because the tricarboxylate transport system is irreversible, that is, unidirectional. This is because both the ATP-citrate lyase reaction (in the cytosol) and the pyruvate carboxylase reaction (in the matrix) involve ATP hydrolysis. Thus, the net result of one turn of the cycle is the hydrolysis of 2 ATP to 2 ADP + 2 P_i and the transport of acetyl-CoA from the matrix to the cytosol (assuming that the NADH and NADPH in the malate dehydrogenase and malic enzyme reactions are equivalent).

21. A Citrate
C cAMP-dependent phosphorylation
D Palmitate
B Malonyl-CoA

22. E CDP–diacyylglycerol
D Ceramide
A Phosphatidylglycerol
C Plasmalogen
B CDP–Choline

23. Propranolol primarily inhibits phosphatidic acid phosphatase, which converts phosphatidic acid to 1,2-diacylglycerol. Since diacylglycerol is an intermediate in the synthesis of phosphatidylcholine and phosphatidylethanolamine, propanolol decreases the production of these lipids. Inhibition of phosphatidic acid phosphatase also increases the concentration of phosphatidic acid, which is then converted in greater amounts to phosphatidylinositol and phosphatidylglycerol.

24. Tay–Sachs disease results from a defect in the enzyme hexosaminidase A, which breaks down ganglioside G_{M2} to ganglioside G_{M3}.

25. Cholesterol can be obtained either by synthesis *de novo* from acetyl-CoA or from circulating lipoproteins, primarily LDL, that enter cells by receptor-mediated endocytosis.

26. The order is: HMG-CoA, mevalonate, geranyl pyrophosphate, farnesyl pyrophosphate, squalene, 2,3-oxidosqualene, lanosterol.

27. HMG-CoA reductase catalyzes the first unique step in the synthesis of cholesterol, the conversion of HMG-CoA to mevalonate. This reaction is followed by three reactions that consume ATP. By regulating the activity of HMG-CoA reductase so that it operates only when cholesterol is needed, the cell avoids wasting metabolic energy on the production of cholesterol precursors.

28. (a) and (c) are correct. (b) is incorrect because inhibition of cholesterol biosynthesis tends to increase the synthesis of LDL receptors.