INTEGRATION OF METABOLISM

DR. A. TARAB
DEPT. OF BIOCHEMISTRY
HKMU
To appreciate fully the significance of individual metabolic pathways and their regulation, we must view these pathways in the context of the whole organism.

An essential characteristic of multicellular organisms is cell differentiation and division of labour.

In addition to the central pathways of energy-yielding metabolism that occur in all cells, the tissues and organs of complex organisms have specialized functions and thus characteristic fuel requirements and patterns of metabolism.
Tissue-Specific Metabolism: The Division of Labour

- Each tissue and organ of the human body has a specialized function that is reflected in its anatomy and its metabolic activity.
- Skeletal muscles, for example, uses metabolic energy to produce motion; adipose tissue stores and releases fats, which serve as fuel throughout the body; the brain pumps ions to produce electrical signals.
• The liver plays a central processing and distributing role in metabolism and furnishes all other organs and tissues with a proper mix of nutrients via the blood stream

• The functional centrality of the liver is indicated by the common reference to all other tissues and organs as “extrahepatic” or “peripheral”
The Liver Processes and Distributes Nutrients

- During digestion in the GIT of mammals, the three major classes of nutrients undergo enzymatic hydrolysis into their monomeric subunits.
- This breakdown is necessary because the epithelial cells lining the intestinal lumen are able to absorb only relatively small molecules.
• After being absorbed, most of the sugars and amino acids and some TAG pass to the blood and are taken up by hepatocytes in the liver; the remaining TAG take a different path via the lymphatic system and enter adipose tissue.

• Hepatocytes transform the nutrients obtained from the diet into the fuels and precursors required by each of these tissues, and export them in the blood.
• The demand of extrahepatic tissues for fuels and precursors varies among organs and with the activity of the organism
• To meet these changing circumstances, the liver has remarkable metabolic flexibility
• For example, when the diet is rich in protein, hepatocytes contain high levels of enzymes for amino acid catabolism and gluconeogenesis
• Within hours after a shift to a high-carbohydrate diet, the levels of these enzymes drop and the synthesis of enzymes essential to carbohydrate metabolism begins.

• Other tissues also adjust their metabolism to the prevailing conditions, but none is as adaptable as the liver, and none is so central to the organism’s overall metabolic activities.
• **Sucars**: Glucose entering the liver is phosphorylated by glucokinase to yield G-6-P. Fructose, galactose and mannose absorbed from the small intestine, are also converted into G-6-P by enzymatic pathways

• G-6-P is at the crossroads of carbohydrate metabolism in the liver

• It may take any of five major metabolic routes, depending on the current metabolic needs of the organism
• 1) G-6-P is dephosphorylated by glucose-6-phosphatase to yield free glucose, which is exported to replenish blood glucose
• Export is the pathway of choice when the amount of G-6-P is limited, because the blood glucose concentration must be kept sufficiently high to provide adequate energy for the brain and other tissues
• 2) G-6-P not immediately needed to form blood glucose is converted into liver glycogen
• 3) G-6-P may be oxidized for energy production via glycolysis, and the citric acid cycle
• The ensuing electron transfer and oxidative phosphorylation yield ATP
4) Excess G-6-P not used to make blood glucose or liver glycogen is degraded via glycolysis and the PDH reaction into acetyl-CoA, which serves as the precursor for the synthesis of lipids: fatty acids, which are incorporated into TAG, phospholipids and cholesterol.

• Much of the lipid synthesized in the liver is exported to other tissues, carried there by blood lipoproteins.
5) Finally, G-6-P is the substrate for the PPPPathway, yielding both reducing power (NADPH), needed for the biosynthesis of FA and cholesterol, and ribose-5-P, a precursor in nucleotide biosynthesis.
Metabolic pathways for glucose 6-phosphate in the liver
• **Amino acids**: amino acids that enter the liver have several important metabolic routes
  
  1) They act as precursors for protein synthesis in hepatocytes
  
  The liver constantly renews its own proteins, which have a very high turnover rate, with an average half-life of only a few days
• The liver is also the site of biosynthesis of most of the plasma proteins of the blood

• 2) Alternatively, amino acids may pass from the liver into the blood and thus to other organs, to be used as precursors in the synthesis of tissue proteins

• 3) Certain amino acids are precursors in the biosynthesis of nucleotides, hormones and other nitrogenous compounds in the liver and other tissues
4) Amino acids not needed for biosynthesis of proteins and other molecules in the liver or elsewhere are deaminated and degraded to yield acetyl-CoA and citric acid cycle intermediates.

Citric acid cycle intermediates so formed may be converted into glucose and glycogen via the gluconeogenic pathway.
• Acetyl-CoA may be oxidized via the citric acid cycle for ATP energy (4b), or it may be converted into lipids for storage (4c)
• The ammonia released on degradation of amino acids is converted by hepatocytes into the excretory product, urea (4d)
• Finally, the liver participates in the metabolism of amino acids arriving intermittently from the peripheral tissues
• The blood is adequately supplied with glucose just after the digestion and absorption of dietary carbohydrate or, between meals, by the conversion of some of the liver glycogen into blood glucose.

• But in the periods between meals, especially if prolonged, there is some degradation of muscle protein to amino acids (5).
Metabolism of amino acids in the liver
• These amino acids donate their amino groups (by transamination) to pyruvate, the product of glycolysis, to yield alanine, which is transported to the liver and deaminated

• The resulting pyruvate is converted by hepatocytes into blood glucose (via gluconeogenesis), and the NH$_3$ is converted into urea for excretion
• The glucose returns to the skeletal muscles to replenish muscle glycogen stores
• One benefit of this cycle process, the glucose – alanine cycle, is the smoothing out of fluctuations in blood glucose in the periods between meals
• The amino acid deficit incurred in the muscles is made up after the next meal from incoming dietary amino acids
• **Lipids**: The fatty acid components of the lipids entering hepatocytes also have several different pathways

• 1) Fatty acids are converted into liver lipids

• 2) Under most circumstances, fatty acids are the major oxidative fuel in the liver

• Free fatty acids may be activated and oxidized to yield acetyl-CoA and NADH
The acetyl-CoA is further oxidized via the citric acid cycle to yield ATP by oxidative phosphorylation.

3) Excess acetyl-CoA released on oxidation of fatty acids and not required by the liver is converted into the ketone bodies, acetoacetate and β-hydroxybutyrate, which are circulated in the blood to peripheral tissues, to be used as fuel for the citric acid cycle.
• The ketone bodies may be regarded as a transport form of acetyl groups
• They can supply a significant fraction of the energy in some peripheral tissues, up to one-third in the heart, and 60 to 70% in the brain during prolonged fasting
• 4) Some of the acetyl-CoA derived from fatty acids (and from glucose) is used for the biosynthesis of cholesterol, which is required for membrane biosynthesis
• Cholesterol is also the precursor of all steroid hormones and of the bile salts, which are essential for the digestion and absorption of lipids

• The final two metabolic fates of lipids involve specialized mechanisms for the transport of insoluble lipids in the blood
5) Fatty acids are converted to the phospholipids and TAG of the plasma lipoproteins, which carry lipids to adipose tissue for storage as TAG

- Cholesterol and cholesteryl esters are also transported as lipoproteins

6) Some free fatty acids become bound to serum albumin and are carried in the blood to the heart and skeletal muscles, which absorb and oxidize free fatty acids as a major fuel
Metabolism of fatty acids in the liver
Liver lipids → Fatty acids

1. Liver lipids → Fatty acids
2. β-oxidation
3. Acetyl-CoA
4. Citric acid cycle
5. Ketone bodies in blood
6. Cholesterol
7. Plasma lipoproteins
8. Free fatty acids in blood

Steroid hormones → Bile salts → Cholesterol → Acetyl-CoA → Citric acid cycle → ATP

ADP + P_i → ATP

O_2 → H_2O → e^-

CO_2
• Serum albumin is the most abundant plasma protein; one molecule of serum albumin can carry up to 10 molecules of free fatty acids, releasing them at the consuming tissue where they are taken up by passive diffusion.
Thus, the liver serves as the body’s distribution centre: exporting nutrients in the correct proportions to the other organs, smoothing out fluctuations in metabolism caused by the intermittent nature of food intake, and processing excess amino groups into urea and other products to be disposed of by the kidneys.
In addition to the processing and distribution of carbohydrates, fats and amino acids, the liver is also active in the enzymatic detoxification of foreign organic compounds, such as drugs, food additives, preservatives and other possibly harmful agents with no food value.
Adipose Tissue Stores and Supplies Fatty Acids

- Adipose tissue, which consists of adipocytes is amorphous and widely distributed in the body; under the skin, around the deep blood vessels and in the abdominal cavity.

- It typically makes up about 15% of the mass of a young adult human, with approximately 65% of this mass being in the form of TAG.
Scanning electron micrograph of human adipocytes
• Adipocytes are metabolically very active, responding quickly to hormonal stimuli in a metabolic interplay with the liver, skeletal muscles and the heart.

• Like other cell types in the body, adipocytes have an active glycolytic metabolism, use the citric acid cycle to oxidize pyruvate and fatty acids and carry out mitochondrial oxidative phosphorylation.
During periods of high carbohydrate intake, adipose tissue can convert glucose via pyruvate and acetyl-CoA into fatty acids, from which TAG are made and stored as large fat globules.

In humans, however, most fatty acids synthesis occurs in hepatocytes, not in adipocytes.

Adipocytes store TAG arriving from the liver (carried in the blood as VLDLs) and from the intestinal tract, particularly after meals rich in fat.
When fuel is needed, TAG stored in adipose tissue are hydrolyzed by lipases within the adipocytes to release free fatty acids, which may then be delivered via the blood stream to skeletal muscles and the heart.

The release of fatty acids from adipocytes is greatly accelerated by the hormone epinephrine, which stimulates the conversion of the inactive form of TAG lipase into its active form.
• Insulin counterbalances this effect of epinephrine, decreasing the activity of TAG lipase

• Humans and many other animals, particularly those that hibernate, have adipose tissue called brown fat, which is specialized to generate heat rather than ATP during the oxidation of fatty acids
Muscle Uses ATP for Mechanical Work

- Skeletal muscles account for over 50% of the total O$_2$ consumption in a resting human being and up to 90% during very active muscular work.
- Metabolism in skeletal muscle is primarily specialized to generate ATP as the immediate source of energy.
- Moreover, skeletal muscle is adapted to do its mechanical work in an intermittent fashion, on demand.
Energy sources for muscle contraction

- **Bursts of heavy activity**
  - Muscle glycogen
  - Phosphocreatine
  - Creatine

- **Light activity or rest**
  - Fatty acids, ketone bodies, blood glucose
  - CO₂

- **ADP + Pₐ**
  - ATP
  - Muscle contraction

- Lactate
Sometimes skeletal muscles must deliver much work in a short time, as in a 100 m sprint; at other times more extended work is required, as in running a marathon or giving birth.
• Skeletal muscle can use free fatty acids, ketone bodies or glucose as fuel, depending on the degree of muscular activity.

• In resting muscle the primary fuels are free fatty acids from adipose tissue and ketone bodies from the liver.

• These are oxidized and degraded to yield acetyl-CoA, which enters the citric acid cycle for oxidation to CO$_2$. 
• The ensuing transfer of electrons to O$_2$ provides the energy for ATP synthesis by oxidative phosphorylation

• Moderately active muscles use blood glucose in addition to fatty acids and ketone bodies

• The glucose is phosphorylated, then degraded by glycolysis to pyruvate, which is converted to acetyl-CoA and oxidized via the citric acid cycle
• However, in maximally active muscles, the demand for ATP is so great that the blood flow cannot provide $O_2$ and fuels fast enough to produce the necessary ATP by aerobic respiration alone.

• Under these conditions, the stored muscle glycogen is broken down to lactate, with a yield of two molecules of ATP per glucose unit degraded.
• Lactic acid fermentation thus provides extra ATP energy quickly, supplementing the basal ATP production resulting from the aerobic oxidation of other fuels via the citric acid cycle

• The use of blood glucose and muscle glycogen as emergency fuels for muscular activity is greatly enhanced by the secretion of epinephrine, which stimulates the formation of blood glucose from glycogen in the liver and the breakdown of glycogen in muscle tissue
• Skeletal muscle does not contain glucose-6-phosphatase and cannot convert glucose-6-P to free glucose for export to other tissues.

• Consequently, muscle glycogen is completely dedicated to providing energy in the muscle, via glycolytic breakdown.

• After a period of intense muscular activity, heavy breathing continues for sometime.

• Much of the O$_2$ thus obtained is used for the production of ATP by oxidative phosphorylation in the liver.
Metabolic cooperation between skeletal muscle and the liver
Muscle: ATP produced by glycolysis for rapid contraction.

Liver: ATP used in synthesis of glucose (gluconeogenesis) during recovery.
• This ATP is used for gluconeogenesis from lactate, carried in the blood from the muscles to the liver
• The glucose thus formed returns to the muscles to replenish their glycogen, completing the Cori cycle
• Skeletal muscles contain considerable amounts of phosphocreatine, which can rapidly regenerate ATP from ADP by the creatine kinase reaction
Phosphocreatinine $\xrightleftharpoons{\text{during activity}}{\text{during recovery}}$ ATP + Creatine
• **Heart muscle** differs from skeletal muscle in that it is continuously active in a regular rhythm of contraction and relaxation

• Mitochondria are much more abundant in heart muscle than in skeletal muscle; they make up almost half the volume of the cells

• The heart uses as fuel a mixture of glucose, free fatty acids and ketone bodies arriving from the blood
• These fuels are oxidized via the citric acid cycle to deliver the energy required to generate ATP by oxidative phosphorylation
Electron micrograph of heart muscle
The Brain

• The metabolism of the brain is remarkable in several aspects
• First, the brain of adult mammals normally uses only glucose as fuel
• Second, the brain has a very active respiratory metabolism; it uses almost 20% of the total $O_2$ consumed by a resting human adult
• The use of $O_2$ by the brain is fairly constant in rate and does not change significantly during active thought or sleep
Because the brain contains very little glycogen, it is continuously dependent on incoming glucose from the blood.

If the blood glucose should fall significantly below a certain critical level for even a short period of time, severe and sometimes irreversible changes in brain function may occur.
Although the brain cannot directly use free fatty acids or lipids from the blood as fuels, it can, when necessary, use β-hydroxybutyrate (a ketone body) formed from fatty acids in hepatocytes. The capacity of the brain to oxidize β-hydroxybutyrate via acetyl-CoA becomes important during prolonged fasting or starvation, after essentially all the liver glycogen has been depleted, because it allows the brain to use body fat as a source of energy.
• The use of β-hydroxybutyrate by the brain during severe starvation also spares muscle proteins, which become the ultimate source of glucose for the brain during severe starvation
Glucose metabolism in the brain

• The technique of positron emission tomography (PET) scanning shows metabolic activity in specific regions of the brain

• PET scans allow visualization of isotopically labeled glucose in precisely localized regions of the brain of a living person, in real time
• The scans compare glucose metabolism (in mg/100 g/min) when the experimental subject (a) is rested and (b) has been deprived of sleep for 48 hours.
Hormonal Regulation of Fuel Metabolism

• The adjustments that keep the blood glucose level near 4.5 mM involve the combined actions of insulin, glucagon and epinephrine on metabolic processes in many body tissues, but especially in liver, muscle and adipose tissue.

• **Insulin** signals these tissues that the blood glucose concentration is higher than necessary; as a result, the excess glucose is taken up from the blood into cells and converted to storage compounds, glycogen and TAG.
• **Glucagon** carries the message that blood glucose is too low, and the tissues respond by producing glucose through glycogen breakdown and gluconeogenesis and by oxidizing fats to reduce the use of glucose

• **Epinephrine** is released into the blood to prepare the muscles, lungs and heart for a burst of activity

• Insulin, glucagon and epinephrine are the primary determinants of the metabolic activities of muscle, liver and adipose tissue
The endocrine system of the pancreas
In addition to the exocrine cells which secrete digestive enzymes in the form of zymogens, the pancreas contains endocrine tissue, the islets of Langerhans. The islets contain α, β, and δ cells (also known as A, B, and D cells, respectively), each cell type secreting a specific polypeptide hormone.
Glucose regulation of insulin secretion by pancreatic β cells

- When the blood glucose level is high, active metabolism of glucose in the cell raises intracellular [ATP], which leads to closing of K channels in the plasma membrane, depolarizing the membrane.
In response to the change in membrane potential, voltage-gated Ca$_2^+$ channels in the plasma membrane open, allowing Ca$_2^+$ to flow into the cell; this raises the cytosolic [Ca$_2^+$] enough to trigger insulin release by exocytosis.
Glucose transporter GLUT2

Glucose transporter GLUT2

Pancreatic β cell

Glucose

hexokinase IV (glucokinase)

Glucose 6-phosphate
glycolysis
citric acid cycle
oxidative phosphorylation

[ATP]↑

V_m

K^+

ATP-gated K^+ channel

[Ca^{2+}]↑

Insulin granules

Insulin secretion

depolarization

Voltage-dependent Ca^{2+} channel

Ca^{2+}
Epinephrine Signals Impending Activity

• When an animal is confronted with a stressful situation that requires increased activity – fighting or fleeing, in the extreme case – neuronal signals from the brain trigger the release of epinephrine and norepinephrine from the adrenal medulla.

• Both hormones increase the rate and strength of the heart beat and raise the blood pressure, thereby increasing the flow of O$_2$ and fuels to the tissue, and dilate the respiratory passages, facilitating the uptake of O$_2$. 

• In its effects on metabolism, epinephrine acts primarily on muscle, adipose tissue and liver
• It activates glycogen phosphorylase and inactivates glycogen synthase, thus stimulating the conversion of liver glycogen into blood glucose, the fuel for anaerobic muscular work
• Epinephrine also promotes the anaerobic breakdown of the glycogen of skeletal muscles into lactate by fermentation, thus stimulating glycolytic ATP formation
The stimulation of glycolysis is accomplished by raising the concentration of fructose-2,6-bisphosphate, a potent allosteric activator of the key glycolytic enzyme phosphofructokinase-1.

Epinephrine also stimulates fat mobilization in adipose tissue, activating the TAG lipase.

Finally, epinephrine stimulates the secretion of glucagon and inhibits the secretion of insulin, reinforcing its effect of mobilizing fuels and inhibiting fuel storage.
Glucagon Signals Low Blood Glucose

• Several hours after the intake of dietary carbohydrate, blood glucose levels fall to below 4.5mM because of the continued oxidation of glucose by the brain and other tissues

• Lowered blood glucose triggers secretion of glucagon and decreases insulin release

• Glucagon causes an increase in blood glucose concentration in two ways
• Like epinephrine, glucagon stimulates the net breakdown of liver glycogen by activating glycogen phosphorylase and inactivating glycogen synthase; both effects are the result of phosphorylation of the regulated enzymes.

• But unlike epinephrine, glucagon inhibits glucose breakdown by glycolysis in the liver and stimulates glucose synthesis by gluconeogenesis.
• Both of these effects result from lowering the level of fructose-2,6-bisphosphate, an allosteric inhibitor of the gluconeogenic enzyme fructose-1,6-bisphosphatase and an activator of phosphofructokinase.

• Glucagon also inhibits the glycolytic enzyme pyruvate kinase, thus the conversion of phosphoenolpyruvate to pyruvate and preventing oxidation of pyruvate via the citric acid cycle; the resulting accumulation of phosphoenolpyruvate favours gluconeogenesis.
• Glucagon thus enables the liver to export glucose to the blood, restoring blood glucose to its normal level

• Although its primary target is the liver, glucagon (like epinephrine) also affects adipose tissue, activating TAG lipase

• This lipase liberates free fatty acids, which are exported to the liver and other tissues as fuel, thus sparing glucose for the brain
• The net effect of glucagon is therefore to stimulate glucose synthesis and release by the liver and to cause the mobilization of fatty acids from adipose tissue, to be used instead of glucose as fuel for tissues other than the brain.
During Starvation, Metabolism Shifts to Provide Fuel for the Brain

• The fuel reserves of a normal adult human are of three types: glycogen stored in the liver and in muscle in relatively small quantities; large quantities of TAGs in adipose tissue; and tissue proteins, which can be degraded when necessary to provide fuel

• After an overnight fast, almost all of the liver glycogen and most of the muscle glycogen have been depleted

• Within 24 hours, the blood glucose concentration begins to fall, insulin secretion slows and glucagon secretion is stimulated
Sources of blood glucose after ingestion of 100 g of glucose
• These hormonal signals result in the mobilization of TAG, which become the primary fuels for muscle and liver

• To provide glucose for the brain, the liver degrades certain proteins (those most expendable in an organism not ingested food)

• Their amino groups are converted into urea in the liver; the urea is exported via the bloodstream to the kidney and is excreted
• Also in the liver, the carbon skeletons of glucogenic amino acids are converted into pyruvate or intermediates of the citric acid cycle
• These intermediates, as well as the glycerol derived from TAG in adipose tissues, provide the starting materials for gluconeogenesis in the liver, yielding glucose for the brain
Eventually the use of citric acid cycle intermediates for gluconeogenesis depletes OAA, preventing the entry of acetyl-CoA into the cycle.

Acetyl-CoA produced by fatty acid oxidation accumulates, favouring the formation of acetoacetyl-CoA and ketone bodies in the liver.

After a few days of fasting, the level of ketone bodies in the blood rise as these fuels are exported from the liver to heart and skeletal muscle and the brain, which use them instead of glucose.
• Concentrations of fatty acids, glucose, and ketone bodies in the plasma during the first week of starvation:
• Despite the hormonal mechanisms for maintaining the level of glucose in the blood, it begins to diminish after two days of fasting
• The level of ketone bodies, almost immeasurable before the fast, rises dramatically after 2 to 4 days of fasting
• These water-soluble ketones, acetoacetate and β-hydroxybutyrate, supplement glucose as an energy source during a long fast
• Fatty acids cannot serve as a fuel for the brain; they do not cross the blood-brain barrier
• The TAG stored in the adipose tissue of an adult of normal weight provide enough fuel to maintain a basal rate of metabolism for about three months; a very obese adult has enough stored fuel to endure a fast of more than a year
• However, such a fast would be extremely dangerous; it would almost certainly lead to severe overproduction of ketone bodies, and perhaps to death
• When fat reserves are gone, the degradation of essential proteins begin; this leads to loss of heart and liver function, and death
Insulin Signals High Blood Glucose

• When glucose enters the blood stream from the intestine after a carbohydrate-rich meal, the resulting increase in the blood glucose causes increased secretion of insulin and decreased secretion of glucagon

• Insulin stimulates glucose uptake by muscle tissue, where the glucose is converted to G-6-P
• Insulin also activates glycogen synthase and inactivates glycogen phosphorylase, so that much of the G-6-P is channeled into glycogen

• As a consequence of accelerated uptake of glucose from the blood, the blood glucose concentration falls to the normal level, slowing the rate of insulin release from the pancreas
• Thus there is a closely adjusted relationship between the rate of insulin secretion and the blood glucose concentration

• The effect of this regulation is to hold the blood glucose concentration nearly constant in the face of large fluctuations in the dietary intake of glucose
• Insulin also stimulates the storage of excess fuel as fat
• It activates both the oxidation of G-6-P to pyruvate via glycolysis and the oxidation of pyruvate to acetyl-CoA
• Acetyl-CoA not oxidized further for energy production is used for fatty acid synthesis in the liver, and these fatty acids are exported as the TAG of plasma lipoproteins (VLDLs) to the adipose tissue
• Insulin stimulates TAG synthesis in adipocytes, using fatty acid released from the VLDL TAG
• These fatty acids are ultimately derived from the excess glucose taken from the blood by the liver
• In summary, the effect of insulin is to favour the conversion of excess blood glucose into two storage forms; glycogen (in the liver and muscle) and TAG (in adipose tissue)
Diabetes Is a Defect in Insulin Production or Action

• **Diabetes mellitus**, caused by the deficiency in the secretion or action of insulin, is a relatively common disease

• There are two major clinical classes of the disease: insulin-dependent diabetes mellitus (IDDM) and non-insulin-dependent diabetes mellitus (NIDDM)

• In the former, the disease begins early in life and quickly becomes severe

• The latter is slow to develop, milder, and often goes unrecognized
IDDM requires insulin therapy and careful, lifelong control of the balance between glucose intake and insulin dose.

Characteristic symptoms of diabetes are excessive thirst and frequent urination (polyuria), leading to the intake of large volumes of water (polydipsia).

These changes are due to the excretion of large amounts of glucose in the urine, a condition known as glucosuria.

The term diabetes mellitus means “excessive excretion of sweet urine.”
Another characteristic metabolic change resulting from the defect in insulin action in diabetes is excessive but incomplete oxidation of fatty acids in the liver, resulting in an overproduction of the ketone bodies - acetoacetate and $\beta$-hydroxybutyrate, which cannot be used by the extra hepatic tissues as fast as they are made in the liver.
In addition to β-hydroxybutyrate and acetoacetate, the blood of diabetics also contains acetone, which results from the spontaneous decarboxylation of acetoacetate:

- Acetoacetate + H$_2$O $\rightarrow$ acetone + HCO$_3^-$

Acetone is volatile and is exhaled, giving the breath of untreated diabetic a characteristic odour sometimes mistaken for ethanol.

A diabetic experiencing mental confusion because of high blood glucose is occasionally misdiagnosed as intoxicated, an error that can be fatal.
• The overproduction of ketone bodies, called **ketosis**, results in their appearance in greatly increased concentrations in the blood (**ketonemia**) and urine (**ketonuria**)

• The oxidation of TAG to form ketone bodies produces carboxylic acids, which ionize, releasing protons

• In uncontrolled diabetes this can overwhelm the capacity of the bicarbonate buffering system of blood and produce a lowering of blood pH called **acidosis**, a potentially life-threatening condition
Biochemical measurements on the blood and urine are essential in the diagnosis and treatment of diabetes, which causes profound changes in metabolism.

A sensitive diagnostic criterion is provided by the **glucose-tolerance test**.

After a night without food, the patient drinks a test dose of 100g of glucose dissolved in a glass of water.

The blood glucose concentration is measured before the test dose and at 30 minutes intervals for several hours thereafter.
• A normal individual assimilates the glucose readily, the blood glucose rising to no more than about 9 or 10 mM; little or no glucose appears in the urine

• Diabetic individuals show a marked deficiency in assimilating the test dose of glucose

• The blood glucose level increases far above the kidney threshold, which is about 10 mM, causing glucose to appear in the urine