LECTURE 26

THE PATHOLOGICAL BASIS of METABOLIC DISEASE

MAMMALIAN PATHOBIOLOGY
PATB 4130 / 5130

Metabolism is the sum of all physical and chemical processes by which living, organized substance is produced and maintained – Dorland’s Medical Dictionary. The word metabolism is derived from metaballein (Greek) which means to turn about, change, or alter. Metabolism is required to maintain homeostasis – stability in the normal body states of an organism. Simple unicellular organisms such as bacteria and protozoa contain all of the metabolic machinery necessary to maintain homeostasis. Like the ability to regenerate and adapt to injury (see lecture 8), cells of complex organisms such as mammals rely on the cells, tissues, and organs of the body to work together to maintain homeostasis and a state of health.

The purpose of this lecture is to introduce you to the diverse causes of abnormal metabolism using some specific examples of metabolic disturbances that can affect the body as a whole.

The Causes of Metabolic Disease (slides 6 – 13)

The various causes of abnormal metabolism are as diverse as the underlying diseases themselves. The various causes can disrupt specific metabolic pathways or can cause damage to specific organs or tissues leading to secondary metabolic disease. One example already covered is the cytochromic C oxidase disruption in normal energy metabolism caused by copper deficiency (see lecture 25). Others will be covered in this lecture including disturbances of acid-base balance, hepatic encephalopathy (see also lecture 27 for a toxin that damages the liver), and diabetes mellitus.

- Genetic defects – hereditary defects probably constitute the largest and most diverse category. Many of these relate to faulty synthesis or deficiency of enzymes involved in metabolic pathways. An example would be pyruvate kinase, a terminal enzyme in the glycolytic pathway; deficiency in animals is inherited as a simple autosomal recessive trait and deprives erythrocytes of ATP. The end result is shortened erythrocyte lifespan. Another example would be phenylketonuria due to a deficiency of phenylalanine hydroxylase that leads to mental retardation in humans.
- Faulty nutrition can lead to abnormal metabolism by depriving the body of important nutrients. Examples include vitamins and minerals as co-factors for many enzymes and glucose for energy metabolism.
- Hormonal deficiencies and imbalances participate in many metabolic processes. Examples include deficiencies of insulin leading to diabetes mellitus,
hyperparathyroidism as a cause of metabolic bone disease and metastatic soft tissue mineralization, and the deficiency of aldosterone in Addison’s disease.

- **Drugs / pharmaceuticals** can inhibit enzymes or otherwise block important biochemical pathways. Obviously, these effects can be the intended mode of action of the drug in the treatment for disease but drugs may also have undesirable secondary effects. Cortisone is used to block unwanted inflammation by inhibiting the enzyme phospholipase A₂ in the synthetic pathway for prostaglandins and leukotrienes. Too much cortisone or related compounds can cause Cushing’s disease. The diuretic Lasix blocks absorption of sodium and water from the renal tubules and can lead to hyponatremia and dehydration.

- **Toxins / poisons** often disrupt metabolic pathways or damage tissues leading to secondary metabolic disease. The organophosphate compounds inhibit cholinesterase, the enzyme necessary to metabolize the neurotransmitter acetylcholine leading to persistent neural stimulation. A fungal toxin (mycotoxin) called aflatoxin B₁ can damage the liver leading to hepatic encephalopathy (covered later in this lecture). Aflatoxin B₁ can also disrupt transcriptional processes (see lecture27).

- **Microorganisms** can lead to metabolic disease in diverse ways. Parvovirus infection damages the intestine in dogs and cats causing vomiting and diarrhea that lead to dehydration, ionic imbalances, and acid-base disturbances. Other viruses usurp transcriptional pathways (see hepatitis B virus infection in lecture 27). Certain toxins produced by strains of the bacterium Escherichia coli cause little to no overt morphological damage to the intestine but stimulate active secretion of electrolytes and water from the intestinal epithelium leading to a secretory diarrhea, dehydration, electrolyte depletion, and acid-base disturbances.

From this, you should be able to appreciate that metabolic abnormalities can be associated with just about all diseases, no matter what the underlying cause. For the remainder of this lecture, a few specific metabolic disorders will be covered for illustrative purposes.

**Specific Metabolic Disorders**

**ACID-BASE (pH) DISTURBANCES** (slides 15 – 22)

Disturbances in the pH of body fluids and within cells can contribute significantly to the morbidity and mortality associated with a variety of diseases; pH being a reflection of the H⁺ concentration. Carbon dioxide (CO₂) is generated by many oxidative biochemical reactions in the body. CO₂ combines with water to form carbonic acid. Carbonic acid can then dissociate to form H⁺ and bicarbonate (HCO₃⁻) (see slide 18). Other acids in the body, either absorbed from the diet or that are products of metabolism include S0₄²⁻, PO₄³⁻, HCl, lactate, and β-hydroxybutyrate (one of the ketoacids). Maintenance of pH within a relatively narrow range is critical for normal enzyme-catalyzed biochemical reactions. Normal physiological pH in dogs and most other mammals is maintained in the range of 7.4 by buffer systems; pH’s of 6.9 and 7.8 represent the life-threatening extremes. Do you understand the concept of buffers? Buffers are able to accept or donate H⁺, maintaining pH within a narrow range despite addition of acids or alkaline substances (see slide 17). The most powerful buffers are proteins throughout the body (hemoglobin in red blood cells and the protein matrix of bone are powerful buffers), and phosphate compounds. These buffers act passively to changes in pH. The third buffer system, the
bicarbonate system, is relatively weak. The utility of the bicarbonate system is in its ability to regulate the components of the equation given in slide 18 and to adapt to changes in body pH. The lungs regulate CO₂ and the kidneys H⁺ and HCO₃⁻. The respiratory rate influences the amount of CO₂ in the body fluids. The kidneys can remove H⁺, a complex process involving bicarbonate and ammonia. Ultimately, H⁺ is excreted in the urine as NH₄ and HCO₃⁻ is conserved. The response of the lung to changes in pH occurs fairly rapidly while the response in the kidney may require 2-3 days.

Obviously, in disease these buffer systems can fail for a variety of reasons resulting in changes in body pH, conditions called acidosis and alkalosis.

**Respiratory acidosis** (slide 19). As mentioned in other lectures, the major function of the lung is to exchange oxygen for other gases, especially CO₂, at the level of the alveoli. If the respiratory rate decreases (hypoventilation) or if disease of the lung prevents gas exchange, CO₂ is not eliminated resulting in acidosis (lowering of pH). As a result, the kidneys compensate by excreting more H⁺. A variety of conditions can cause respiratory acidosis. These can include:

- injury to nerves or the spinal cord that control respiratory rate or drugs that affect the functions of these nervous system tissues
- brain injury and drugs such as anesthetics and opiates
- Weakness in the muscles of respiration or abnormalities in the chest wall
- Lung diseases can include any of a variety of diverse conditions that can interfere with respiration or gaseous exchange: pneumonia, obstructive airway disease, toxins (see lecture for a toxin that can damage the lungs), etc.

**Respiratory alkalosis** (slide 20). This is the opposite that occurs with respiratory acidosis; too much CO₂ is exhaled due to increased respiratory rate (hyperventilation). As a result, pH increases (alkalosis). The kidneys compensate by resorption of H⁺. Causes of hyperventilation include anxiety, fever, pain, high ambient temperature, and disease of the nervous system, and others.

**Metabolic acidosis** (slide 21). Metabolic acidosis occurs in conditions where there is increased production of acids or by a loss of HCO₃⁻. Lactic acidosis can be associated with hypoxia and global fall in tissue perfusion. Ketoacidosis results from the use of fatty acids for glucose production (gluconeogenesis) during periods of negative energy balance such as starvation and ketoacidosis is a common complication of diabetes mellitus. Diseases of the gastrointestinal tract with diarrhea or kidney disease can result in substantial loss of or failure to conserve bicarbonate.

**Metabolic alkalosis** (slide 22). Metabolic alkalosis most commonly occurs when there is loss of H⁺ as hydrochloric acid due to severe vomiting. Other causes include diuretics. With both, there is fluid loss and dehydration that are also complicating factors.

**HEPATIC ENCEPHALOPATHY** (slides 23 – 29)

It is well recognized that liver failure can adversely affect normal brain function resulting in a condition called hepatic encephalopathy. The liver has diverse functions (see slide 24).
Failure to filter blood and to metabolize and eliminate potentially harmful substances (absorbed from the gut as well as bi-products of normal metabolism) is the function most often associated with encephalopathy. Encephalopathy can result from various types of liver failure.

- **Kernicterus** (meaning yellow nuclei) is a special form of hepatic encephalopathy. The disorder was first discovered when yellow nuclei (distinct areas of the brain containing cell bodies of neurons – see slide 25) were observed in the brains of infants suffering from neonatal jaundice. The enzymes in the liver that conjugate bilirubin for elimination through the intestinal tract are immature. Any condition in the neonate that increases the level of bilirubin in the blood such as increased destruction of erythrocytes, diseases of the biliary system, and others can result in jaundice (icterus), abnormally high levels of unconjugated bilirubin, and kernicterus. It has been demonstrated that unconjugated bilirubin is lipid soluble and passes through the blood-brain barrier. Once in the brain, the mechanisms by which unconjugated bilirubin damage the brain and why the damage is targeted to specific nuclei are poorly understood.

- **Altered blood supply** to the liver is another cause of hepatic encephalopathy (slide 26). In lecture 8, one cause of atrophy was insufficient blood supply. Atrophy of the liver occurs when shunts form between the portal vein and caudal vena cava, depriving the liver of one of its major blood supplies. The significance with regard to hepatic encephalopathy is that blood escapes around the liver and is not filtered.

- **Acute liver injury (necrosis)** sufficient to affect a large portion of the liver also causes encephalopathy (slide 27). This is because the damaged or necrotic hepatocytes cannot metabolize and eliminate potentially harmful substances allowing them to accumulate in blood.

- **Chronic liver injury** (slide 28) can also be associated with encephalopathy. This occurs when there is significant loss of functional hepatic tissue over time.

- **Hereditary enzyme deficiencies** in the urea cycle lead to a failure to metabolize ammonia to urea. The enzyme ornithine transcarbamylase is most commonly deficient but at least four other enzymes in the urea cycle can also lead to hyperammonemia (see slide 29).

Despite a pretty good in depth knowledge of the various abnormalities that occur in hepatic encephalopathy, no single universal cause for the neurological disturbances has been confirmed (with the exception of unconjugated bilirubin in kernicterus). Accumulation of ammonia (hyperammonemia) due to failure of the liver to remove NH3 from the blood or to metabolize this compound to urea is observed in most, but not all cases of hepatic encephalopathy. What then are possible mechanisms of ammonia-induced encephalopathy? The enzyme responsible for metabolizing and detoxifying ammonia in the brain is glutamine synthetase (slide 29) localized almost exclusively to astrocytes. Excessive glutamine synthesis from glutamate has been proposed to alter levels of the excitatory neurotransmitter glutamate. Additionally, the increased levels of glutamine in the brain results in an osmotic burden (organic osmolyte – see brain response to dehydration in lecture 11) leading to fluid redistribution into the brain and brain swelling. Other hypotheses have been proposed over the years to explain the encephalopathy but to date, the pathogenesis is still unclear.
DIABETES MELLITUS (slides 30 – 39)

Diabetes mellitus is the prototypical metabolic disease; the abnormalities being systemic with impacts on a variety of organs and tissues. Diabetes mellitus is one of the most common endocrine related diseases in dogs and cats but can occur in horses, cattle, sheep, pigs and laboratory animals. The incidence in dogs has been estimated at approximately 1:66 and in cats 1:800. In humans, diabetes mellitus and related co-morbidities are consistently in the top ten leading causes of death. The disorder is due to deficient production of insulin by β-cells of the pancreatic islets and/or resistance of the peripheral tissues to the actions of insulin. The coverage of diabetes mellitus in this class will necessarily be brief and superficial but this is not to understate the importance of this disease. Insulin has important functions in the body, chiefly to insure that cells have sufficient energy:

- **Insulin stimulates the uptake of glucose** into most cells. Striated muscle and fat comprise the bulk of the body’s tissues and consume considerable glucose. Some tissues and organs do not require insulin for glucose uptake such as the brain, renal tubular epithelial cells, lens of the eyes, and erythrocytes. When insulin is deficient or when the target tissues become resistant to the effects of insulin, glucose is not taken up into the tissues and, instead, increases in the blood (*hyperglycemia*). This persistent hyperglycemia can then have adverse effects on tissues that do not require insulin. Furthermore, when glucose levels in the blood increase to a certain limit, it spills over into the urine creating an osmotic *diuresis* and dehydration leading to increased thirst. Individuals with uncontrolled diabetes mellitus commonly have polyuria-polydipsia.

- **Insulin enhances glucose metabolism** for the production of energy by the cells from glucose.

- **Insulin stimulates glycogenesis**, the intracellular storage form of glucose mainly in liver and skeletal muscle.

- **Insulin stimulates amino acid uptake and protein synthesis** in insulin-dependent cells.

- **Insulin stimulates lipogenesis**, formation of fat or adipose tissue when dietary energy exceeds demands.

- **Insulin decreases fatty acid utilization (lipolysis) and gluconeogenesis** (the formation of glucose from non-carbohydrate sources) when there is a plentiful supply of carbohydrates in the diet.

Classification of Diabetes in Humans

**Type 1** has also been called insulin-dependent or juvenile diabetes mellitus. The latter is somewhat of a misnomer as more and more cases of Type 2 diabetes are being found in primarily obese children. In type 1 diabetes, there is an absolute deficiency of insulin. The causes of type 1 diabetes include the combined effects of hereditary factors (genetic predisposition), environment, and autoimmune destruction of the pancreatic β-cells. There is some indication that previous viral infection may precipitate autoimmunity.

**Type 2** has also been called insulin-independent or adult onset diabetes mellitus. In type 2 diabetes, there is no absolute deficiency of insulin. Insulin levels may be low (but not to
levels that would account for the severe hyperglycemia), within the normal range, or elevated. The number of insulin-secreting \( \beta \)-cells in the pancreatic islets may be decreased but this is generally limited. Additionally, \( \beta \)-cells in these individuals do not respond normally to a glucose challenge. **Importantly, the peripheral tissues seem to be resistant to the effects of insulin.** Resistance of the tissues is seen in obesity and obesity is a major risk factor for the development of type 2 diabetes. The mechanisms of peripheral resistance to the actions of insulin have not been fully clarified.

**Diabetes Mellitus in Animals**

It has been problematic to use and apply the human classification schemes universally to diabetes in animals. Certainly in dogs and cats there is a genetic predisposition. In dogs, obesity is a major risk factor suggesting an association with insulin resistance. Cases are associated with chronic relapsing pancreatitis that destroys not only endocrine but exocrine pancreatic tissue as well. Certain viral infections in cattle including bovine virus diarrhea (BVD) and foot and mouth disease have been associated with inflammation of the pancreatic islets and diabetes. Islet amyloidosis-associated diabetes in cats has some features similar to type 2 in humans. There are also many cases of diabetes where no morphological destruction of the islets is demonstrable.

**Metabolic Dysfunction and Complications of Diabetes**

The metabolic disturbances and complications of diabetes mellitus are related to the systemic disturbances in glucose metabolism:

- **Dehydration** occurs when the glucose concentration in the blood exceeds the threshold of the renal glomeruli. When this happens, the excess glucose spills into the urine, increasing the osmolarity of urine and causing an osmotic diuresis. One of the classical symptoms of uncontrolled diabetes mellitus is **polyuria-polydipsia** (osmotic diuresis leading to dehydration, increased thirst, and drinking a lot of fluids). It is not unusual for many diabetics to suffer from some degree of dehydration, especially the elderly.

- **Ketoacidosis** is a potentially life-threatening complication of uncontrolled type 1 diabetes mellitus characterized by an absolute deficiency of insulin. Because of this deficiency, many of the tissues in the body are unable to utilize glucose for energy despite the hyperglycemia. Instead, there are lipolysis and utilization of free fatty acids to meet energy demands. If and when the rate of lipolysis and fatty acid oxidation exceeds the rate at which acetoacetic and \( \beta \)-hydroxybutyric acids (ketone bodies, ketoacids) can be utilized for energy, ketonemia and ketonuria develop.

- **Nonketotic hyperosmolar coma** is also a life-threatening complication that develops mainly in patients with uncontrolled type 2 diabetes. As previously mentioned, osmotic diuresis due to glucosuria is common in diabetics. In mainly the elderly who may not be able to drink sufficient water to counteract the dehydration that subsequently develops, a hyperosmolar state develops that potentially leads to neurological dysfunction and coma.

- **Nonenzymatic glycosylation of proteins** underlies many of the complications of diabetes that certainly contribute to morbidity and mortality. Nonenzymatic glycosylation is related to chronic or persistent hyperglycemia. Glucose attaches
nonenzymatically to proteins in the interstitium and blood vessel walls. Initially, the glycosylation is reversible but with time following a slowly progressive series of rearrangements, these glycosylation reactions become irreversible forming advanced glycation end products (AGEs) that accumulated in a variety of tissues including basement membranes and blood vessel walls leading to diabetic angiopathy, etc. Apart from merely accumulating at these sites resulting in structural and functional abnormalities, AGEs also have other effects due to receptor binding on a variety of cell types (see slide 37).

- **Alternate metabolic pathways for glucose metabolism** are involved in tissues that are not dependent on insulin for glucose uptake. In cells of these tissues, the persistent hyperglycemia leads to increased intracellular glucose. The excess is metabolized via the polyol pathways to sorbitol and fructose. One effect is intracellular elevation of sorbitol and fructose creating an osmotic imbalance and intracellular fluid accumulation. Other effects are decreased phosphoinositide metabolism, diacyl glycerol, protein kinase C, and Na-K-ATPase activity. In the lens of the eye and nerves, glucose metabolism via the polyol pathway is likely responsible for formation of cataracts and polyneuropathy that complicate the diabetic state.