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CHAPTER

3

Clinical Biochemistry and Hematology

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OUTLINE

The Laboratory Rabbit, Guinea Pig, Hamster, and Other Rodents
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INTRODUCTION

This chapter reviews information on the clinical biochemistry and hematology of the rabbit (*Oryctolagus cuniculus*), guinea pig (*Cavia porcellus*), hamster (*Mesocricetus auratus*) and other rodents, including the gerbil (*Meriones unguiculatus*), chinchilla (*Chinchilla laniger*), degu (*Octodon degus*), deer mouse (*Peromyscus maniculatus*), dormouse (Gliridae family), kangaroo rat (*Dipodomys* spp.), cotton rat (*Sigmodon hispidus*), and sand rat (*Psammomys obesus*).

The chapter begins with a review of sample collection and preparation, and a description of commonly measured parameters and analytical techniques. The reference values, sources of variation, and unique characteristics are then presented for each species, as available. We have attempted to include the most current literature on a backdrop of historical information for each of these topics, along with supporting references.

It is important to recognize that many variables affect the parameters of clinical biochemistry and hematology that are presented here, including methods of sample collection and preparation, equipment, reagents, and methods of analysis, as well as the age, sex, breed, and environment of the animals being sampled. Reference ranges are presented according to the most recent and reliable literature available. Some clinical biochemistry and hematology parameters for the less common species covered in this volume are not yet available in the literature.

CLINICAL BIOCHEMISTRY

Introduction

Abnormal clinical conditions are usually associated with specific biochemical changes in the blood and urine, and these changes are detected using specific analytical techniques. Values obtained from a clinical case are usually compared with reference values that are either produced in the same laboratory or in a similar group of animals and cited in the literature.

A joint committee formed by several scientific societies, the International Committee for Harmonization of Clinical Pathology Testing (IHCPT), formulated a recommended core clinical chemistry panel for safety and toxicity studies in laboratory animals (Weingand et al., 1996). This core panel includes glucose, blood urea nitrogen, creatinine, total protein, albumin, globulin, calcium, sodium, potassium, chloride, and total cholesterol. Additionally recommended are at least two hepatocellular function tests, including alanine aminotransferase, aspartate aminotransferase, sorbitol dehydrogenase, glutamate dehydrogenase, or total bile acids, and two hepatobiliary function tests, including alkaline phosphatase, gamma glutamyltransferase, 5′-nucleotidase, total bilirubin, or bile acids. The U.S. EPA (2008) recommended the additional inclusion of assays for triglycerides, magnesium, protein electrophoresis, inorganic phosphate/cholinesterase, phosphorus, ornithine carbamoyl transferase, lactate dehydrogenase, and creatine (Evans, 2009).

Urinalysis tests recommended by the IHCPT and U.S. EPA include appearance (color and turbidity), volume, specific gravity, pH, microscopic sediment, protein, glucose, ketones, bilirubin, blood, nitrate/nitrite, and urobilinogen (Evans, 2009; U.S. EPA, 2008; Weingand et al., 1996).

Sample Collection

Restraint

Most small laboratory animals are prey species in the wild, and they are easily stressed by the handling and restraint needed to obtain a sample of blood or urine. Pre-conditioning has been shown to reduce plasma corticosterone levels in rats (Fluttert et al., 2000), but this practice is often impossible or impractical in many laboratory rodents, especially for the diagnosis of spontaneous disease. A restraint tube can be used to hold small rodents for obtaining samples if the tube is the appropriate size and is disinfected between animals.

Anesthesia may be needed to obtain samples from laboratory rabbits or rodents in certain situations, especially when larger volumes of blood, or sterile urine samples, are needed. However, anesthesia itself has been shown to produce changes in clinical biochemistry or hematology parameters, including decreased hematocrit, hemoglobin level, and red blood cell count

(Marini et al., 1994; Pilny, 2008). It is important to know what method of restraint was used to obtain samples when interpreting reference clinical biochemistry and hematology values.

Blood Collection

Optimal sites for blood collection vary between laboratory animals and are described in this chapter for each species for which information is available. Small animals typically lack superficial blood vessels, and deeper vessels are often obscured by fat. A variety of peripheral veins, such as the lateral saphenous vein and the cephalic vein, may be used for small samples of blood, as described for individual species. The jugular vein may be used in most species for larger blood samples, although appropriate positioning and restraint can be stressful and may require sedation or anesthesia (Thrall et al., 2004).

Blood collection from the smaller laboratory animals is challenging due to the limited blood volume available and because the restraint or sedation necessary to obtain a sample may alter the results of some assays (Pilny, 2008). A general guideline for the maximum volume of blood that can be safely withdrawn in a single sample is approximately 1% of the animal's body weight, or 10% of the circulating blood volume, in order to avoid hypovolemic shock and anemia. This volume will be replaced within 24 hours in most healthy animals, although a return to normal levels of all blood constituents may take up to 2 weeks (Thrall et al., 2004). An alternate standard states that the blood volume withdrawn should not exceed 7.5% of the total blood volume per week or 10% of the total blood volume every 2 weeks (McGuill and Rowan, 1989).

Clinical chemistry assays have become more sensitive with advances in technology, and successful analysis can now be achieved with smaller volumes of blood. Most clinical chemistry assays require a few milliliters of blood, and some assays now require only a few microliters of blood.

Serum is the preferable sample for most clinical chemistry assays to avoid interference by the actions of an anticoagulant. Some of the anticoagulants, such as ethylenediamine-tetraacetic acid (EDTA), and citrate, prevent clotting by chelating calcium ions, which are often cofactors in enzyme assays. Heparin is usually the preferred anticoagulant because it has minimal or no interference with the majority of clinical chemistry assays (Buetow et al., 1999). Heparin may, however, produce artificially elevated inorganic phosphorus values (Benjamin and McKelvie, 1978).

Serum is produced by collecting blood into tubes with no anticoagulant and allowing it to clot for 30–45 minutes at room temperature. The sample should not stand for more than 45 minutes without refrigeration because many constituents may change concentration at room temperature, including glucose, phosphates, lipids, and chloride (Mitruka and Rawnsley, 1977). The tube is then centrifuged at approximately 1000 *g* for 15 minutes to yield serum that can be analyzed for clinical chemistry parameters (Buetow et al., 1999).

Plasma is suitable for many clinical chemistry assays and is a practical necessity in many cases because of the small volume of blood available from laboratory rabbits and rodents. Blood samples may be treated with heparin and used for both hematology and clinical chemistry assays. Blood may be collected in a syringe containing 50 U of heparin per ml, or placed in a heparinized tube, and centrifuged for 2 minutes at high speed, such as 12 000 *g*, to yield platelet-free plasma that can be used for clinical chemistry assays (Lindena and Trautschold, 1986). Alternatively, miniature blood collection tubes, such as those used in human pediatric medicine, can be used for small-volume blood samples. The advantage of using these tubes is that the anticoagulant is lyophilized and does not dilute the sample (Dyer and Cervasio, 2008).

Both serum and plasma samples may be used immediately, stored at 4°C for use in 24–48 hours, or stored frozen at −20°C for up to 90 days or at −70°C for 360 days or longer (Buetow et al., 1999; Cray et al., 2009). However, some enzymes, such as lactate dehydrogenase, are not stable if the sample is frozen (Mitruka and Rawnsley, 1981).

Certain factors, such as lipemia and hemolysis, may interfere with the results of clinical biochemistry assays that are performed by spectrophotometer. The increased turbidity associated with lipemia can produce artificially high values for certain enzymes. Lipemia also interferes with the assays for glucose, bilirubin, total protein, phosphorus, and urates (Saibaba et al., 1998). Hemolysis also produces errors in clinical biochemical values because most serum enzymes are present at high concentrations in red blood cells. Hemolysis causes the release of these enzymes into the bloodstream to yield artificially high serum values. Hemolysis may produce an artificial elevation of serum potassium and inorganic phosphorus levels (Owens et al., 2005; Stankovic and Smith, 2004), as well as bilirubin, transaminases, and lactate dehydrogenase (Gad, 2007).

The method and site of sample collection may also affect resulting values. For example, enzymes found in cardiac muscle, such as creatine kinase, aspartate aminotransferase, lactate dehydrogenase, and alanine aminotransferase, may be artificially elevated in the serum of some species when blood samples are obtained by cardiac puncture (Maxwell et al., 1985).

Urine Collection

Urine samples of satisfactory volume and quality are difficult to obtain from many small laboratory animals. Urine may be collected from most animals following spontaneous urination, manual bladder expression, cystocentesis, or catheterization. Cystocentesis should be used to obtain a sterile sample when a urine culture is needed, as for a bacterial culture and antibiotic sensitivity. A metabolism cage is useful when sequential urine samples are required or when a larger volume of urine needs to accumulate over a specified period of time. The volume of urine that is excreted per day and available for sampling depends on many factors, including food and water consumption, activity level, disease, and environmental temperature.

The simplest method of urine analysis is performed with a commercial dry reagent test strip, or dipstick, which requires only a few microliters of urine per test reagent square. This assay provides semiquantified measures of urine components, such as blood, glucose, ketones, and pH.

The animal's ability to concentrate urine is evaluated with a small drop of urine placed on a refractometer. Normal specific gravity falls in a range of approximately 1.003–1.050 and varies by species and environment. Increased specific gravity may indicate dehydration, heart failure, or shock, and decreased specific gravity may indicate renal failure, diabetes insipidus, or other conditions.

A small volume of 1–2 ml of urine can be centrifuged at approximately 10 000 *g* for 10 minutes. The sediment is then analyzed under the microscope for cells, bacteria, and crystals.

Reference values for urine characteristics of the laboratory animals covered in this volume are provided in Table 3.1.

Carbohydrate Metabolism

Glucose

Glucose is the parameter most commonly evaluated as a measure of carbohydrate metabolism. Glucose concentration is affected by many factors, including nutrition, hormones, hibernation, restraint, fasting, and anesthesia. Handling of the blood sample may also affect glucose values because glucose decreases in serum when erythrocytes are present. A decrease in glucose concentration by approximately 7% is reported in blood that sits at room temperature for 1 hour (Tietz, 1976). Therefore, serum should be separated and tested for glucose as soon as possible after a blood sample is taken. Alternatively, a preservative such as thymol or fluoride should be added to inhibit glycolysis, as described for the rat (Ringler and Dabich, 1979). Glucose is usually measured using glucose oxidase/peroxidase-linked chromogenic methods or by measuring hexokinase activity (Evans, 2009).

Reference ranges for serum glucose concentration in the laboratory animals covered in this volume are provided in Table 3.2.

Lipid Metabolism

The primary lipids contained in serum are cholesterol, triglycerides, fatty acids, and phospholipids. Of these, cholesterol and triglycerides are the most commonly measured in laboratory animals. There are significant interspecies differences in the normal serum concentrations of these lipid components. Reference ranges for cholesterol, triglycerides, and total lipids for the laboratory animals covered in this volume are provided in Table 3.2.

High levels of lipids cause lipemia, which can interfere with clinical biochemistry assays performed by photometric analysis. Solubilizers may be used on

TABLE 3.1 Urine Characteristics[1]

	Volume (ml/kg/d)	pH	Specific Gravity	Normal Color	Turbidity	Protein (mg/wk)	Cells	Crystals	References
Rabbit	20–75	7.6–8.8	1.003–1.036	Yellow/straw	Cloudy	Trace	Occas. RBC, WBC	TP, $CaCO_3$	Kozma et al., 1974 Mitruka and Rawnsley, 1977 Mader, 2004
Guinea pig	N/A	8–9	N/A	Yellow/amber	Opaque	N/A	N/A	Occas. $CaCO_3$	Riggs, 2009
Hamster	7 ml/d	5.1–8.4	N/A	N/A	Cloudy	10	N/A	TP, $CaCO_3$	Fiszer et al., 1979 Heatley and Harris, 2009
Chinchilla	N/A	8–9	N/A	Yellow/amber	N/A	N/A	N/A	N/A	Riggs and Mitchell, 2009

N/A = values not available; RBC = red blood cell; WBC = white blood cell; TP = triple phosphate
[1]*Species for which no values are available are not listed.*

TABLE 3.2 Carbohydrate and Lipid Metabolism[1]

	Glucose (mg/dl)	Total Cholesterol (mg/dl)	Triglycerides (mg/dl)	Total Lipids (mg/dl)	References
Rabbit	75–155	10–80	15–160	150–400	Suckow and Douglas, 1996 Evans, 2009 Vennen and Mitchell, 2009
Guinea pig	60–130	20–80	10–70	N/A	Hrapkiewicz and Medina, 2007 Evans, 2009
Hamster	37–198	112–210	72–350	224–466	Cox and Gökcen, 1974 Heatley and Harris, 2009 Evans, 2009
Gerbil	50–135	90–150	N/A	N/A	Hrapkiewicz and Medina, 2007
Chinchilla	60–125	40–300	149–198	N/A	Hrapkiewicz and Medina, 2007 de Oliveira Silva et al., 2005
Degu	74–82	N/A	N/A	N/A	Opazo et al., 2004
Cotton rat	115–216	134–165	N/A	N/A	Dolyak and Leone, 1953
Sand rat	68–71	N/A	N/A	N/A	Frenkel and Kraicer, 1972

N/A = values not available.
[1]*Species for which no values are available are not listed.*

lipemic samples to decrease this interference (Evans, 2009). Polyethylene glycol 6000 is a precipitating agent that is often used to remove lipoproteins from a serum sample (Thomson and Kunze, 1984).

Cholesterol

Cholesterol is an essential component of cell membranes and an important precursor for the synthesis of bile acids, steroid hormones, and some fat-soluble vitamins. Cholesterol is produced in the liver or obtained in the diet, and it circulates in free and esterified forms. Cholesterol is recycled through metabolism in the liver and excretion in bile into the digestive tract. Cholesterol levels typically peak after a meal, so fasting is required for an accurate assay.

Cholesterol has been the subject of many studies in laboratory animals because high levels of circulating cholesterol are associated with the development of atherosclerosis. There are several valuable laboratory animal models of cholesterolemia and atherosclerosis.

Cholesterol is usually measured using a cholesterol esterase followed by a cholesterol oxidase linked to a peroxidase-chromogen detection system (Richmond, 1992).

Lipoproteins are macromolecules that transport lipids and cholesterol within the bloodstream and can be classified as alpha or beta, or by a density value that varies inversely with size.

- *Low-density lipoproteins (LDL)* are also known as beta lipoproteins and have a diameter of 18–28 nm, density of 1.019–1.063 grams per ml, and carry cholesterol from the liver throughout the body.
- *High-density lipoproteins (HDL)* are also known as alpha lipoproteins and have a diameter of 5–15 nm, density of less than 1.063 grams per ml, and carry cholesterol away from the tissues of the body and back to the liver.

Different lipoprotein fractions can be separated and quantified using microaffinity chromatography columns (Tschantz and Sunahara, 1993), electrophoresis or other methods, including fast protein liquid chromatography, and high-performance liquid chromatography (Evans, 2009).

Triglycerides

Triglycerides are composed of a glycerol molecule bound to three fatty acids and are digested by pancreatic lipase. Triglycerides are a major component of very-low-density lipoprotein (VLDL) and serve as a source of energy. They are broken down in the intestine, absorbed by intestinal cells, and combined with cholesterol and proteins to form chylomicrons that are transported in lymph to the bloodstream.

Triglycerides are measured using enzymatic reagents, including lipase, glycerol kinase, and glycerol-3-phosphate oxidase linked to a peroxidase-chromogen detection system (Evans, 2009).

Enzymes

Enzymes are normally contained within the cells that synthesize them, and increased enzyme levels in serum or plasma are indicative of induction or cell damage. Several

factors affect the activity of enzymes in plasma or serum, including the concentration of enzyme in cells, the extent and duration of cell damage, and the accessibility and persistence of the enzyme in the bloodstream. Diet and nutritional deficiencies can also impact the levels of enzymes measured. The laboratory methods used to measure serum enzyme activity can affect the values obtained.

Enzyme activity is usually represented in International Units (IU), where one unit is the amount of enzyme that reacts with one picomole of substrate per minute. In theory, a standardized method of representing enzyme activities should allow better comparisons between laboratories. However, the choice of enzyme substrates and the experimental conditions used have important effects on the results of enzyme assays (Evans, 2009).

Reference values for several commonly assayed enzymes for laboratory animals covered in this volume are provided in Table 3.3.

Alkaline Phosphatase (ALP)

Alkaline phosphatase (ALP) represents a group of enzymes that remove phosphate groups from molecules such as nucleotides and proteins, and they work most effectively in an alkaline environment of pH 9–10 (Evans, 2009). The highest concentrations of ALP are found in liver and bone, and lower concentrations of ALP are present in kidney tubules, intestinal epithelium, lung, and placenta. Bone growth in young animals produces elevated ALP levels. Concentration of ALP varies according to species but in general is increased by digestion, cholestasis, or injury to intestinal or biliary epithelium. Levels of ALP may be decreased by fasting, hypothyroidism, or pernicious anemia (Fernandez and Kidney, 2007).

Alkaline phosphatase is typically measured using 4-nitrophenol phosphate as a substrate and 2-amino-2-methyl-1-propanol (AMP) or diethanolamine (DEA) as a buffer, although DEA yields higher ALP values. Isoenzymes of ALP can be distinguished using electrophoresis, HPLC, and other methods (Evans, 2009).

Alanine Aminotransferase (ALT)

Alanine aminotransferase (ALT) is a transaminase enzyme that was formerly known as serum glutamate pyruvate transaminase (SGPT). Alanine aminotransferase catalyzes the transfer of an amino group from alanine to alpha-ketoglutarate in the alanine cycle to form pyruvate and glutamate. The ALT enzyme is found in serum and organ tissues, especially liver, although significant concentrations are also found in kidney, skeletal muscle, and myocardium. Lower levels of ALT are present in pancreas, spleen, and lung. Alanine aminotransferase is elevated in serum under conditions of significant cellular necrosis and is used as a measure of liver function. Levels of ALT may be elevated in cases of hepatitis, congestive heart failure, liver or biliary duct damage, or myopathy. Diet, restraint, and drug administration may also affect plasma ALT in rodents (Evans, 2009).

Concentration of ALT is quantified using the NADH oxidation method.

Aspartate Aminotransferase (AST)

Aspartate aminotransferase (AST) is a transaminase enzyme that catalyzes the conversion of aspartate and alpha-ketoglutarate to oxaloacetate and glutamate. The AST enzyme was formerly known as serum glutamate oxalate transaminase (SGOT) and is present in all tissues except bone, with highest levels in liver and skeletal muscle. Concentration of AST is elevated after bruising, trauma, necrosis, infection, or neoplasia of liver or muscle (Eugster et al., 1966). The AST enzyme is found in cerebrospinal fluid, exudates, and transudates in proportion to the amount of cellular damage. Low concentrations of AST are also found in urine but are not useful for diagnosis of renal damage (Evans, 2009).

AST is measured using the NADH oxidation method.

Lactate Dehydrogenase (LDH)

Lactate dehydrogenase (LDH) is an enzyme that catalyzes the conversion of pyruvate to lactate, along with NADH to NAD+, during glycolysis in conditions of hypoxia. The LDH enzyme is present in all cells, but it is concentrated in muscle, liver, and kidney. LDH exists as five isozymes, LDH-1 through LDH-5, each composed of four subunits. Differential LDH isozyme levels are used diagnostically (Markert et al., 1975). Isozyme LDH-1 (4H) has four heart subunits and is the major isozyme in heart. Isozyme LDH-2 (3H1M) has three heart and one muscle subunit and is the major isozyme in the macrophage-monocyte system and serum. The LDH-3 (2H2M) isozyme has two heart and two muscle subunits and is the major isozyme in lungs. Isozyme LDH-4 (1H3M) has one heart and three muscle subunits and is the primary isozyme in kidneys. The LDH-5 (4M) isozyme has four muscle subunits and is the major isozyme in liver and skeletal muscle. The concentration of LDH is elevated in serum as a result of organ infarction and significant cell death that results in loss of cytoplasm. For example, elevations of LDH result from conditions such as hepatitis, shock, hypoxia, extreme hypothermia, and meningitis, among others. The LDH enzyme has often been used in laboratory animals, along with troponin levels, to detect myocardial damage (Evans, 2009; Herman et al., 2006). However, LDH stability is highly susceptible to freezing, and values are affected by storage conditions (Mitruka and Rawnsley, 1981; Tietz, 1976).

TABLE 3.3 Enzymes[1]

	ALP (IU/l)	ALT (IU/l)	AST (IU/l)	LDH (IU/l)	CK (IU/l)	SDH (IU/l)	Amylase (IU/l)	GDH (IU/l)	References
Rabbit	10–140	14–80	14–113	30–140	150–1000	170–177	200–500	6.39	Suckow and Douglas, 1997 Evans, 2009 Vennen and Mitchell, 2009
Guinea pig	80–350	10–90	10–90	20–120	50–150	N/A	995–1239	N/A	Waner et al., 1996
Hamster	50–186	20–128	20–150	100–300	23	N/A	154–196	N/A	Mitruka and Rawnsley, 1977 Evans, 2009 Heatley and Harris, 2009
Gerbil	12–37	N/A	N/A	N/A	N/A	N/A	N/A	N/A	Hrapkiewicz and Medina, 2007
Chinchilla	10–70	10–35	15–100	N/A	0–300	N/A	N/A	N/A	Hrapkiewicz and Medina, 2007 Mitchell and Tully, 2009
Deer mouse	6–7	N/A	N/A	N/A	N/A	N/A	N/A	N/A	Meagher, 1998
Cotton rat	10–14	N/A	N/A	N/A	N/A	N/A	N/A	N/A	Dolyak and Leone, 1953

N/A = values not available.
[1]*Species for which no values are available are not listed.*

Creatine Kinase (CK)

Creatine kinase (CK) is also known as creatine phosphokinase (CPK) and is an enzyme that catalyzes the phosphorylation of creatine. Creatine kinase is a dimer that exists as isoenzymes with greatest activity in muscle (CK-MM), heart (CK-MB), and brain (CK-BB) (Lang, 1981). An additional form of the CK enzyme exists in mitochondria (CK-Mt). The distribution of CK isoenzymes varies between tissues and species. Creatine kinase is found in serum, plasma, and urine and is elevated by muscle necrosis or disease.

Sorbitol Dehydrogenase (SDH)

Sorbitol dehydrogenase (SDH) is an enzyme that converts the alcohol form of glucose, sorbitol, into fructose, in the presence of NAD. Sorbitol dehydrogenase activity has been detected in liver, kidney, and seminal vesicles of many animals. Increased levels of SDH usually indicate hepatocellular damage (Dooley, 1984), and may be a more sensitive indicator of hepatic damage than ALT or AST, but changes in SDH levels may also be minimal (Evans, 2009).

Amylase

Amylase is a small enzyme that breaks down complex carbohydrates to ultimately form monosaccharides such as glucose. The pancreas produces large amounts of alpha-amylase, which requires calcium and acts as a major digestive enzyme at neutral pH. Amylase is also present in liver and salivary glands. Thus, increased plasma amylase is indicative of tissue damage in pancreas, liver, or salivary glands (Evans, 2009).

Chromogenic and immunologic methods are typically used to measure amylase for the diagnosis of pancreatic disease (Evans, 2009).

Glutamate Dehydrogenase (GDH)

Glutamate dehydrogenase (GDH) is a mitochondrial enzyme that is involved in the metabolism of glutamate to 2-oxoglutarate. The GDH enzyme is found primarily in liver, kidney, and cardiac muscle, with lower levels in brain, skeletal muscle, and leukocytes. The majority of GDH in the serum originates from hepatocytes in healthy as well as diseased animals. The distribution of GDH in the liver is mainly centrilobular, whereas ALT is periportal or centrilobular in rats, and may indicate the site of hepatocyte damage (Carakostas et al., 1986; Merrick et al., 2006). Levels of GDH are normally low in most species, and increased serum GDH is indicative of damaged or necrotic hepatocytes due to liver disease (Evans, 2009).

Hormones

There are many hormones that can be measured in the serum of laboratory animals, but a complete listing is beyond the scope of this chapter. This section will focus on some of the more relevant hormones in laboratory animals, including adrenal, thyroid, and reproductive hormones. Reference values for hormones in laboratory animals covered in this volume are provided in Table 3.4.

Adrenal Hormones

Cortisol is a glucocorticoid produced by the zona fasciculata of the adrenal cortex and is involved in the response to stress. Cortisol acts to increase blood pressure and blood glucose and has an immunosuppressive action. Levels of cortisol are known to vary according to diurnal rhythm and other environmental influences.

Corticosterone is also produced by the adrenal cortex and is a significant glucocorticoid in some species, including rodents. Corticosterone has functions that are similar to those of cortisol.

Thyroid Hormones

Thyroxine (T4) is secreted by the follicle cells of the thyroid gland and transported in the bloodstream to control metabolic processes in the body. Thyroxine is a prohormone and is converted to the more potent form, *triiodothyronine (T3)* in the tissues of the body. Thyroid hormones are involved in growth and metabolism and act synergistically with other hormones, such as catecholamines and glucocorticoids. Circulating levels of thyroid hormones are affected by many internal and external factors to which an animal is exposed.

Reproductive Hormones

Luteinizing hormone (LH) is a glycoprotein hormone produced by the anterior pituitary gland. In the female, its sudden increase at the end of the follicular phase triggers ovulation and corpus luteum development. In the male, it is also called interstitial cell stimulating hormone, and it triggers testosterone production by the Leydig cells of the testis.

Follicle-stimulating hormone (FSH) is a glycoprotein hormone produced by the anterior pituitary gland. In both males and females, FSH stimulates germ cell maturation and helps regulate growth and sexual maturation.

Estradiol is a steroid hormone that is the predominant female sex hormone and the primary estrogenic hormone. In females, the majority of estradiol is produced by the ovaries, and smaller amounts are produced by the adrenal cortex. In males, small amounts of estradiol are produced by the testes and adrenal cortex.

Progesterone is a steroid hormone that is produced by the corpora lutea of the ovaries, the adrenal glands, and the placenta during pregnancy. Progesterone increases at the time of ovulation and helps support the maintenance of pregnancy.

Prolactin is a peptide hormone produced by the anterior pituitary gland and other organs and is primarily involved in lactation.

TABLE 3.4 Hormones[1]

	Cortisol (µg/dl)	C'sterone (µg/dl)	T3 (ng/dl)	T4 (ug/dl)	LH (ng/ml)	FSH (ng/ml)	Estradiol (pg/ml)	Progest (ng/ml)	Prolactin (ng/ml)	Oxytocin (ng/m)l	Testost (pg/ml)	References
Rabbit	2.6–3.8	1.54	130–143	1.7–2.4	10–100	2–4.5	3	<1	10	10–20	3–5	DePaolo and Masoro, 1989 Suckow and Douglas, 1997
Guinea pig	5–30	N/A	22–56	2.3–3.5	5–55	100–200	30	1	N/A	<5	N/A	DePaolo and Masoro, 1989
Hamster	2.3–3.2	5.5–9.3	30–80	3–7	20–40	100–300	5–10	1	5–10	N/A	1.5–2	Cox and Gokcen, 1974 Neve et al., 1981 DePaolo and Masoro, 1989
Chinchilla	N/A	N/A	N/A	3.4–6.4	N/A	N/A	N/A	N/A	N/A	N/A	N/A	Martin et al., 2005
Degu	15	1.5	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	Gruss et al., 2006
Kangaroo rat	N/A	N/A	N/A	0.48–1.92	N/A	N/A	N/A	N/A	N/A	N/A	N/A	Banta and Holcombe, 2002

N/A = values not available.
[1]*Species for which no values are available are not listed.*

TABLE 3.5 Liver Function[1]

	Total Bilirubin (mg/dl)	Bile Acids (mMol/l)	Total Protein (g/dl)	Albumin (g/dl)	Globulins (g/dl)	References
Rabbit	0–0.8	0–40	5.4–8.3	2.4–4.6	1.5–2.8	Hrapkiewicz and Medina, 2007 Evans, 2009 Vennen and Mitchell, 2009
Guinea pig	0–1	N/A	4–7	2–5	2–4	Waner et al., 1996 Evans, 2009 Riggs, 2009
Hamster	0.1–0.9	N/A	5.2–7.0	3.5–4.9	2.7–4.2	Heatley and Harris, 2009
Gerbil	0.2–0.6	N/A	4.3–12.5	1.8–5.5	1.2–6.0	Heatley and Harris, 2009
Chinchilla	0.6–1.3	N/A	3.8–8.0	2.3–4.1	0.9–4.2	Hrapkiewicz and Medina, 2007 Riggs and Mitchell, 2009
Deer mouse	N/A	N/A	6.3–6.9	4.1–4.7	N/A	Meagher, 1998
Cotton rat	N/A	N/A	6.1–6.9	4.2–5.2	1.4–2.2	Dolyak and Leone, 1953

N/A = values not available.
[1]*Species for which no values are available are not listed.*

Oxytocin is a peptide hormone produced in the hypothalamus and released from the posterior pituitary. Stimulation of the cervix during delivery causes release of oxytocin, which is involved in lactation and uterine contractions.

Testosterone is a steroid hormone that is the primary sex hormone in males and has anabolic effects. Testosterone is secreted primarily by the testes in males and the ovaries in females. Small amounts of testosterone are also secreted by the adrenal glands.

Most reproductive hormones are evaluated using commercial radioimmunoassay kits.

Liver Function

Enzymes

The aminotransferases, AST and ALT, are normally contained within liver cells and are released into the blood when liver cells are damaged. Increased levels of AST or ALT are therefore usually indicative of liver damage, as discussed above. Reference ranges for these enzymes in the laboratory animals covered in this volume are provided in Table 3.3.

Reference values for other indicators of liver function, including total bilirubin, bile acids, and serum proteins, in the laboratory animals covered in this volume are provided in Table 3.5.

Total Bilirubin

Bilirubin is produced in the macrophage-monocyte system by the breakdown of hemoglobin to biliverdin and then bilirubin. Bilirubin is carried in the plasma to the liver, where it is conjugated to form bilirubin diglucuronide and excreted in the bile. Bilirubin level thus serves as a measure of liver and bile tract function. Total bilirubin includes both the conjugated and unconjugated (free) forms and, if elevated, is usually indicative of liver damage or hemolysis.

Bilirubin oxidizes slowly when exposed to light, so specimens obtained for bilirubin levels should be protected from light exposure. Freezing also artificially alters bilirubin levels in plasma samples (Ringler and Dabich, 1979).

Elevated total bilirubin may be recognized by a visible yellow coloration of the plasma. A standard colorimetric assay for total bilirubin involves a diazotization reaction (Evans, 2009).

Bile Acids

Bile acids are synthesized from cholesterol and conjugated to amino acids in the liver, then stored in the gallbladder. Bile acids are released from the gallbladder during digestion to form micelles that assist in the processing of dietary fats. Bile acid levels are affected in most species by diet, gastric and intestinal transit time, enteritis, and gallbladder contraction (Center et al., 1995). Bile acid concentration can be determined by gas or liquid chromatography-mass spectrometry (Perwaiz et al., 2001).

Serum Proteins

The liver is the source of many proteins, and liver function is often evaluated by measuring total serum protein, albumin, and globulin levels (Evans, 2009). Serum proteins include albumin and globulins, and total serum proteins represent the sum of these constituents. Protein electrophoresis is used to separate serum proteins, based on their mobility, into fractions of albumin, alpha-, beta-, and gamma-globulins and determine their concentrations (Thrall et al., 2004).

ALBUMIN

Albumin is the first fraction to appear by electrophoresis and is the predominant serum protein. Albumin is produced in the liver and helps maintain osmotic pressure within the intravascular compartment.

GLOBULINS

Alpha-1 and -2 globulins and beta globulins include a group of binding and carrier proteins in the blood. Alpha globulins are increased during acute infections and demonstrate the most rapid electrophoretic mobility of the globulin fractions. Beta globulins are binding proteins in plasma and thus should be measured in plasma rather than serum. Gamma globulins are primarily antibodies and are increased in response to infection.

TOTAL PROTEIN

Total protein levels can be affected by many factors including age, diet, pregnancy, disease, and dehydration. Technical issues such as assay method and sample handling can also impact the results of this assay.

Low total protein, or hypoproteinemia, is often due to chronic malnutrition, malabsorption, or protein loss due to kidney or liver disease. A specific decrease in the albumin fraction is suggestive of liver disease, although chronic malnutrition from poor diet or dental disease may also cause hypoalbuminemia. Reduced cecotrophy in some laboratory animals may also cause a decrease in total protein, especially albumin.

Increased total protein, or hyperproteinemia, is commonly caused by dehydration, but it may also indicate chronic infection, shock, or a metabolic disturbance.

The relative size of the albumin and globulin fractions may help determine the cause of an abnormal total protein value. The albumin to globulin ratio is often calculated as a measure of this relationship and is usually slightly greater than one. A low ratio could be the result of decreased albumin or increased globulins due to myeloma or autoimmune disease. A high ratio

TABLE 3.6 Kidney Function[1]

	BUN (mg/dl)	Creatinine (mg/dl)	References
Rabbit	10–30	0.5–2.5	Evans, 2009 Vennen and Mitchell, 2009
Guinea pig	9–32	0.6–2.2	Hrapkiewicz and Medina, 2007 Riggs, 2009
Hamster	12–26	0.4–1.0	Heatley and Harris, 2009
Gerbil	17–27	0.6–1.4	Heatley and Harris, 2009 Hrapkiewicz and Medina, 2007
Chinchilla	10–40	0.4–2.3	Hrapkiewicz and Medina, 2007 Vennen and Mitchell, 2009
Kangaroo rat	30	N/A	Intress and Best, 1990
Cotton rat	13–14	0.9–1.5	Dolyak and Leone, 1953

N/A = values not available.
[1]*Species for which no values are available are not listed.*

may be caused by low immunoglobulin levels as occurs in some leukemias.

There are several methods used to measure serum protein levels, and the specific method chosen has an effect on the values obtained. Total proteins can be measured with the biuret method, in which a cupric ion–peptide complex forms in an alkaline solution. The bromcresol green dye method produces higher total protein values than the method of cellulose acetate electrophoresis (Hall, 1992).

Kidney Function

Reference values for blood urea nitrogen and creatinine, which are typically used as indicators of kidney function, are provided in Table 3.6 for the laboratory animals covered in this volume.

Blood Urea Nitrogen (BUN)

Urea is produced by the breakdown of proteins and is excreted in the urine. Blood urea nitrogen (BUN) is often used to evaluate renal function in animals, including laboratory animals. Blood urea nitrogen level is affected by circadian rhythm, diet, liver function, hydration, and intestinal absorption (Melillo, 2007). The concentration of BUN is artificially increased by a high-protein diet or vigorous exercise, whereas BUN is decreased by low protein intake, liver failure, or treatment with anabolic steroids (McLaughlin and Fish, 1994).

Urea is measured using the urease enzyme followed by colorimetric detection methods (Evans, 2009).

Creatinine

Creatinine is a non-protein nitrogenous compound that is produced by the breakdown of creatine in muscle. Creatinine is found in serum, plasma, and urine and is excreted by glomerular filtration at a constant rate and in the same concentration as in plasma. Creatinine is a more reliable indicator of renal function than BUN because it is less influenced by other factors such as diet and hydration. Serum creatinine is increased by deficient glomerular filtration, although both BUN and creatinine remain in the reference range in most animals until 50–75% of renal function is lost (Finco, 1989). Creatinine is also increased by necrosis or atrophy of skeletal muscle, hyperthyroidism, infections, burns, or fractures.

When a renal disorder is suspected, the ratio of BUN to serum creatinine may be useful in diagnosing the location of the problem. A normal ratio is 10–20 mg/dl BUN to 1 mg/dl creatinine. A high (more than 20:1) BUN to creatinine ratio suggests pre-renal disease, whereas a low (less than 10:1) ratio is indicative of renal disease.

Creatinine is measured using an alkaline picrate colorimetric (Jaffé) method, by other enzymatic methods, or by HPLC (Evans, 2009). Endogenous chromogens such as bilirubin and ketones can artificially elevate creatinine values (Palm and Lundblad, 2005).

Electrolytes

Reference values for electrolytes in the laboratory animals covered in this volume are provided in Table 3.7.

Bicarbonate (HCO_3^-)

The bicarbonate anion (HCO_3^-) affects the acid–base balance in the body and is necessary for its buffering capacity in blood. The main influences on bicarbonate levels in the blood are respiratory production of carbon dioxide and renal excretion of bicarbonate after its conversion from carbonic acid (Evans, 2009). Increased levels of bicarbonate are indicative of respiratory acidosis and metabolic alkalosis.

Calcium (Ca)

Calcium is the most prevalent mineral in the body and is involved in bone metabolism and conduction of electrical impulses. Total calcium is the sum of bound and ionized (Ca^{2+}) forms. Approximately half of total calcium is bound to plasma proteins and the other half exists in the free cation form in serum. Ionized calcium is difficult to test but represents the more physiologically important value (Evans, 2009).

Total calcium is affected by diet, serum proteins, metabolism, and disease. Calcium is increased in animals with kidney or bone neoplasia. Calcium is decreased in animals with hypoparathyroidism, chronic renal failure, or magnesium deficiency.

TABLE 3.7 Electrolytes[1]

	HCO_3^- (mEq/l)	Ca (mg/dl)	Cl^- (mEq/l)	Mg^{2+} (mg/dl)	P (mg/dl)	K^+ (mEq/l)	Na^+ (mEq/l)	References
Rabbit	16.2–38.0	5.6–17	92–120	2.0–5.4	2.3–6.7	3.5–6.9	131–155	Melillo, 2007 Evans, 2009 Vennen and Mitchell, 2009
Guinea pig	N/A	5.3–12.0	90–115	3.5–4.1	3.0–12.0	4.0–8.0	120–150	Waner et al., 1996 Hrapkiewicz and Medina, 2007 Riggs, 2009
Hamster	N/A	5.3–12.0	93–110	N/A	3.0–9.9	3.9–6.0	128–150	Evans, 2009 Heatley and Harris, 2009
Gerbil	N/A	3.7–6.2	93–118	N/A	3.7–7.0	3.8–6.3	141–172	Hrapkiewicz and Medina, 2007 Heatley and Harris, 2009
Chinchilla	N/A	5.6–15.0	105–130	3.6–4.0	4–8	3–7	130–170	Hrapkiewicz and Medina, 2007 de Oliveira Silva et al., 2005 Riggs and Mitchell, 2009
Cotton rat	12.6–15.9	N/A	N/A	N/A	6.0–7.9	N/A	N/A	Dolyak and Leone, 1953

N/A = values not available.
[1]*Species for which no values are available are not listed.*

Total calcium is typically measured using a colorimetric assay. Measurement of ionized calcium is not as common in animals because it requires the use of a selective ion electrode.

Chloride (Cl^-)

Chloride (Cl^-) is a major extracellular anion and, along with sodium, is important for the maintenance of osmolality and acid–base balance. Chloride ions are found in serum, plasma, urine, and cerebrospinal fluid and are reabsorbed in the kidney. Elevated chloride occurs in metabolic acidosis or respiratory alkalosis. Chloride is decreased in metabolic alkalosis and conditions of high fluid loss, such as diarrhea and dehydration (Evans, 2009).

Magnesium (Mg^{2+})

The magnesium (Mg^{2+}) cation is involved in enzymatic reactions, conduction of electrical stimuli, and bone formation. Approximately half of total magnesium is ionized and the other half is bound to albumin or otherwise complexed (Evans, 2009).

Elevated magnesium may occur with dehydration, hyperthyroidism, tissue trauma, or adrenocortical insufficiency and can depress myocardial contractions. Low magnesium levels may be associated with myopathies.

Magnesium is typically measured using a colorimetric assay (Evans, 2009).

Phosphorus (P)

Phosphorus (P) is present within cells and is required for normal Krebs cycle function and formation of bone and teeth. Serum phosphorus exists mainly as the phosphate anion (PO_4^-), which is the form normally assayed. Phosphorus is increased in metabolic bone disease, healing fractures, and during anesthetic recovery. Phosphorus is decreased in animals with renal acidosis, hypopituarism with growth hormone deficiency, and Gram-negative septicemia.

The kidney is the main organ involved in phosphorus balance by regulating glomerular filtration and tubular reabsorption, so phosphate levels are indicative of renal function. Hyperphosphatemia usually indicates prerenal, renal, or post-renal effects or soft tissue trauma.

Hypophosphatemia occurs but its clinical significance is not known.

Phosphorus is typically measured using a colorimetric assay (Evans, 2009).

Potassium (K^+)

Potassium (K^+) is primarily an intracellular cation and is important for maintenance of membrane potential. Intracellular and extracellular potassium levels are regulated by a complex ion exchange mechanism and affected by several compounds, including aldosterone, insulin, and catecholamines (Evans, 2009). Elevated plasma potassium levels increase aldosterone secretion

from the adrenal glands and may also cause arrhythmias and cardiac arrest. Hypokalemia results in decreased aldosterone secretion.

Potassium is usually measured with an ion-selective electrode (Evans, 2009).

Sodium (Na^+)

Sodium (Na^+) is a cation present in all tissue spaces, and its concentration is associated with osmotic homeostasis. Plasma sodium is normally in balance with renal excretion of sodium, with most reabsorption of sodium occurring in the distal tubules and collecting ducts of the kidney. Sodium is increased when there is high fluid loss that spares electrolytes, or with inadequate water intake. Sodium is decreased in conditions of fluid loss when water replacement includes no electrolytes.

Sodium, as with potassium, is typically measured using an ion-selective electrode (Evans, 2009).

RABBIT

Sample Collection

Restraint

Rabbits are relatively docile with repeated handling, but they are easily frightened by unusual noises and movements. If rabbits are not held correctly, the combination of strong hind legs and a fragile skeleton can result in a broken back. A rabbit should be picked up by the scruff of the neck with one hand and supported under the hind end with the other hand to prevent the rabbit from kicking (Brewer, 2006).

Rabbits may be placed in a restraint device, a cat bag, or wrapped in a towel for blood collection without anesthesia.

Blood

The best site for blood collection in a rabbit depends on the volume and number of samples needed and whether or not the rabbit will be anesthetized for blood collection.

The marginal ear vein is the most common site for collection of small blood samples in the unanesthetized rabbit and can be used for single or repeat samples of 1–5 ml. The rabbit should be restrained as described above and the ear warmed by gentle stroking or using a lamp. Acepromazine maleate (1–5 mg/kg IM) may be given to calm the rabbit and aid in vasodilation. Topical oil of wintergreen, xylene, and local anesthetics have all been used to aid in vasodilation (Hrapkiewicz and Medina, 2007). Xylene especially may be irritating if not thoroughly rinsed off the ear after blood withdrawal (Suckow and Douglas, 1997). The collection site should be shaved and then wiped with alcohol or an antiseptic solution. Blood is first collected from the vein near the base of the ear, with successive samples taken from the alternate ear or by moving distally toward the tip of the ear. A 25G or 26G needle or butterfly catheter may be inserted into the vein while pressure is applied to the vein at the base of the ear. It is difficult to obtain sufficient blood volume using this method in dwarf breed rabbits due to the small size of their ears (Melillo, 2007), and catheterization of this vein in dwarf breeds may lead to vascular necrosis, thrombosis, or sloughing of the ear (Vennen and Mitchell, 2009).

Other peripheral veins that may be used in a rabbit for small blood samples without anesthesia include the lateral saphenous vein and the cephalic vein, both of which can be accessed using a 25G needle, with or without a 1 ml syringe. The lateral saphenous vein runs superficially and is easily visualized on an extended leg when held off at the stifle. Visualization of this vein is assisted by applying alcohol or clipping the overlying fur. Pressure should be placed on the vein after blood withdrawal to prevent hematoma formation (Mader, 2004). The cephalic vein may be used to collect blood samples or for intravenous catheter placement (Vennen and Mitchell, 2009).

The central ear artery can be used for collection of larger volumes of up to 30 ml of arterial blood in the unanesthetized rabbit. The rabbit should be restrained as described above and a 21G or 22G needle or butterfly catheter inserted into the artery near the distal tip of the ear. Blood may be obtained faster using a vacutainer tube or a vacuum ear bleeder (Fox and Laird, 1999). Subsequent samples can be taken from the alternate ear or by moving proximally toward the base of the ear. Several minutes of digital pressure should be applied to ensure hemostasis after blood withdrawal. Potential side effects of blood collection from the ear vessels of rabbits include thrombosis, hematoma, and vascular necrosis of the pinna.

The jugular vein is the preferred sampling site for venous blood volumes greater than 0.5 ml (Vennen and Mitchell, 2009). An unanesthetized rabbit can be positioned with its neck gently extended upward and its front feet held straight down over the edge of a table. Anesthesia is recommended for jugular venipuncture to avoid the stress of restraint. The anesthetized rabbit is placed in dorsal recumbency and the jugular vein occluded near the apex of the sternum. The vein is most easily found by clipping the overlying hair. The dewlap may partially obstruct access to the jugular vein in a large female rabbit (Mader, 2004).

Cardiocentesis (cardiac puncture) may be used as a terminal procedure to collect a large volume of blood from a rabbit under anesthesia. A large-bore (18G–21G) 1.5-inch needle can be used to enter the left ventricle through the chest wall, through the diaphragm,

or from the cranial or caudal end of the sternum, and blood should be withdrawn slowly. A blood volume of 60–200 ml may be obtained using this method, depending on the size of the rabbit.

Urine

The method used for urine collection in a rabbit depends on the need for an uncontaminated sample and whether or not the animal is anesthetized. Normal rabbit urine is dense and rich in minerals and should be centrifuged and filtered before it is analyzed. Urine samples should be assayed immediately or stored refrigerated until used.

If sample contamination is not a concern, urine may be collected from a pan placed beneath the wire grid floor of the cage in which the rabbit is housed. Alternatively, manual expression of the bladder may be performed on an awake or anesthetized rabbit and urine collected directly from the urethra or a collection pan. Manual expression must be performed carefully due to the thin wall of the rabbit bladder, which may rupture if the rabbit struggles or if there is a urethral obstruction.

Repeated urine samples may be collected over a period of time using a metabolism cage, which separates urine output from feces.

Relatively uncontaminated urine samples may be obtained directly from the urinary bladder of a rabbit by catheterization or cystocentesis under sedation or light anesthesia. A sterile lubricated 9 French flexible catheter may be inserted into the external urethral opening and directed into the urinary bladder. Urine is withdrawn by gentle aspiration with an attached syringe, or it flows out due to gentle manual pressure applied to the caudal abdomen. Cystocentesis may be performed on a rabbit placed in dorsal recumbency after aseptic preparation of the caudal ventral abdomen. The bladder is located by palpation cranial to the pelvis, and a 20G–22G needle is attached to a syringe and inserted through the skin and bladder wall at the ventral midline. Urine is withdrawn under gentle aspiration pressure (Vennen and Mitchell, 2009).

Reference Values and Sources of Variation

Blood

Total blood volume of the rabbit is discussed in the Hematology section of this chapter.

Rabbit blood clots fairly rapidly and may be separated by centrifugation at 1100 rpm after it stands for 30–45 minutes at room temperature. If inadequate time is allowed for clotting to occur, fibrin clots may form in the cell-free fraction (Fox and Laird, 1999).

Rabbit blood may require more anticoagulant than other species due to its higher calcium content (Bjoraker and Ketcham, 1981).

Urine

Characteristics and reference ranges of rabbit urine are presented in Table 3.1.

Rabbits have a wide reference range for urine volume, 20–75 ml per kg body weight, collected over 24 hours. Normal rabbit urine has an alkaline pH of 7.6–8.8 and is fairly dilute, with a specific gravity that ranges from 1.003–1.036. Rabbit urine is normally cloudy due to a high concentration of triple phosphate (ammonium magnesium phosphate), calcium carbonate monohydrate, and anhydrous calcium carbonate crystals. Crystals are identified by examining the urine sediment after centrifugation. Trace protein and a few red or white blood cells are commonly found in rabbit urine.

The color of normal rabbit urine can vary from light yellow or straw color to a reddish-brown. Reddish coloration may be due to dietary porphyrins but must be differentiated from the presence of blood. A dipstick may be used to detect hematuria, and a woods lamp will detect porphyrins due to their natural fluorescence. Clear urine suggests low excretion of calcium, which can be due to renal failure, or may be normal in a young rabbit or lactating doe. Excess dietary calcium may cause rabbit urine to be thick and white with a reddish-orange pigment (Harkness and Wagner, 1989).

Abnormalities in rabbit urine are usually identified by simple diagnostic tests using a few drops of urine. Rabbit urine with an acidic pH indicates acidosis due to anorexia, fever, pregnancy toxemia, or hepatic lipidosis.

Hematuria may be caused by several conditions in the rabbit, including uterine adenocarcinoma, uterine polyps, endometrial hyperplasia, abortion, urolithiasis, cystitis, renal infarcts, cystitis, and urinary tract infection.

Glucosuria may be due to stress hyperglycemia. True glucosuria is indicative of hepatic lipidosis or diabetes mellitus. Ketonuria is abnormal in rabbits and is indicative of anorexia, hepatic lipidosis, pregnancy toxemia, or diabetes.

Proteinuria with low specific gravity typically indicates renal disease. Ratio of urine protein to urine creatinine is normal if it is less than 0.6.

Spores of *E. cuniculi* are visualized using Gram or trichrome stain on urinary sediment obtained by centrifugation.

Carbohydrate Metabolism

GLUCOSE

The reference range for blood glucose in rabbits is 75–155 mg/dl (Table 3.2). Blood glucose remains fairly consistent due to the fact that rabbits eat throughout the day and use volatile fatty acids as their primary source of energy (Melillo, 2007). Normal glucose levels are maintained, even after a prolonged fast of up to 96 hours, probably due to coprophagy (Hunt

and Harrington, 1974; Kozma et al., 1974). However, some reports suggest that glucose levels in rabbits are affected by diurnal variation (Fox and Laird, 1970). There is some strain variation in glucose concentration, with higher levels in New Zealand White rabbits than in Dutch Belted or Polish rabbits (Kozma et al., 1974).

Glucose levels in neonatal rabbits increase to a peak level 9 hours after suckling, then decrease at 24 hours. Glycogen stores are depleted by 6 hours in newborns that do not nurse. Glycogen stores typically increase in neonatal rabbits after 9 days old (Mottaz et al., 1989). At 15 days old, rabbits that are fasted for 48 hours have no change in blood glucose, but a decline in concentration of ketone bodies.

Elevated blood glucose in rabbits is usually due to disease or environment. Hyperglycemia may occur in rabbits anesthetized with urethane, halothane, or isoflurane (Bito and Eakins, 1969; Gil et al., 2007). Rabbits with diabetes mellitus may have blood glucose concentrations higher than 500 mg/dl, accompanied by the common diabetic symptoms of polyuria, polydipsia, and polyphagia. Hyperglycemia of approximately 350 mg/dl may also be caused by acute intestinal blockage or mucoid enteropathy. Other causes of hyperglycemia in rabbits include traumatic or hypovolemic shock, cold stress, and hyperthermia (de la Fuente et al., 2007; Melillo, 2007).

Low blood glucose in rabbits may be the result of prolonged anorexia and metabolism of adipose tissue, which may lead to hepatic lipidosis. Hypoglycemia may also result from acute sepsis, or terminal mucoid enteropathy, liver failure, or other chronic disease (Melillo, 2007). Rabbits housed with unfamiliar rabbits were found to have decreased glucose concentrations (de la Fuente et al., 2007).

Low glucose values may also result from technical handling, which includes allowing blood samples to stand at room temperature for prolonged periods. Glycolytic enzymes are released from platelets and increase in serum with prolonged clotting time, which lowers glucose in the sample (Lindena and Trautschold, 1986).

Lipid Metabolism

The rabbit is recognized as a valuable model for human disturbances in lipid metabolism, such as the metabolic syndrome and hypercholesterolemia leading to atherosclerosis. The lipoprotein profile of rabbits is similar to that of humans, and rabbits have the same cholesteryl ester transfer protein as in humans, which is not present in rodents (Brousseau and Hoeg, 1999). However, the measurement of cholesterol and triglycerides in rabbits may be confounded by cecotrophy in this species.

CHOLESTEROL

The reference range for cholesterol levels in the rabbit is 10–80 mg/dl (Table 3.2), with normal variation in cholesterol levels in rabbits depending on age, sex, and time of day. Cholesterol levels in neonatal rabbits are similar to adult values and then increase to a peak at 25 days old and decline to adult values at 60–80 days old (Laird and Fox, 1970). Serum cholesterol is higher in female than in male New Zealand White rabbits, and cholesterol levels typically increase at the end of the day (Melillo, 2007).

High-cholesterol diets are particularly effective in the rabbit for producing hypercholesterolemia, and the rabbit is often used as a model to study cholesterol metabolism and atherogenesis (Fekete, 1993). Cholesterol levels are particularly elevated in certain strains of rabbit. For example, the Watanabe Heritable Hyperlipidemic (WHHL) rabbit has a single gene defect that produces 8–14 times the reference level of cholesterol in homozygotes and a slight elevation of cholesterol in heterozygotes. The WHHL strain serves as a model for human familial hypercholesterolemia (Kita et al., 1981).

A high-fat diet, obesity, or liver disease may also cause high cholesterol and triglycerides in rabbits. Hypercholesterolemia during anorexia suggests end-stage hepatic lipidosis and has a poor prognosis. High-density lipoproteins have been found to be reduced, and low-density lipoproteins increased, in pregnant rabbits and their cesarean-derived neonates (Bortolotti et al., 1989). Hypercholesterolemia may be seen in rabbits with disease conditions such as pancreatitis, diabetes mellitus, nephrotic syndrome, or chronic renal failure (Saunders and Davies, 2005). Decreased serum cholesterol may be found in rabbits with liver failure, malnutrition, or pregnancy (Matsuoka et al., 2009).

TRIGLYCERIDES

The reference range for triglyceride levels in the rabbit is 15–160 mg/dl (Table 3.2). Recent models of hypertriglyceridemia have been produced by mating WHHL strain rabbits with normal Japanese White rabbits. One resulting strain, TGH, has naturally high cholesterol levels of 1000–1500 mg/dl and triglycerides greater than 500 mg/dl, with widespread atherosclerotic plaques and xanthomas. The other resulting strain, PHT, has postprandial cholesterol of 100–150 mg/dl and triglycerides of 500–3000 mg/dl, with no atherosclerosis (Mitsuguchi et al., 2008).

Decreased triglycerides have recently been reported in late-gestational pregnant rabbits on a restricted feeding regimen (Matsuoka et al., 2009).

TOTAL LIPIDS

The reference range for total lipid levels in the rabbit is 150–400 mg/dl (Table 3.2).

Enzymes

ALKALINE PHOSPHATASE (ALP)

Reference levels of alkaline phosphatase (ALP) in the rabbit are 10–140 IU/l (Table 3.3). Levels of ALP have

been found to vary by breed and strain of rabbit (Fox and Laird, 1970; Kozma et al., 1974) but not by sex or diurnal variation (Fox and Laird, 1970; Kozima et al., 1974). Levels of ALP are 2–4 times higher in young rabbits compared to adults (Kramer, 1989).

Rabbits produce three isoenzymes of alkaline phosphatase, including one intestinal form and two different isoenzymes in the liver and kidney. This is in contrast to most other animals, which produce only two ALP isoenzymes.

Elevated ALP in the rabbit is caused by conditions associated with bile stasis in the liver, including lipidosis, coccidiosis, liver lobe torsion, abscesses, and neoplasia (Melillo, 2007). Bone lesions are also associated with elevated ALP in the rabbit.

Alkaline phosphatase is decreased in pregnant rabbits (Viard-Drouet et al., 1984), in contrast to an increase in ALP during late gestation in many other species.

ALANINE AMINOTRANSFERASE (ALT)

Reference levels of alanine aminotransferase (ALT) in the rabbit are 14–80 IU/l (Table 3.3). The ALT enzyme is not specific to the liver in herbivores, including the rabbit. The ALT enzyme has a short half-life of approximately 5 hours in the rabbit and is therefore not a very useful indicator of liver damage in this species (Melillo, 2007). However, ALT is not increased by restraint in the rabbit, so an elevated ALT value is suggestive of tissue damage in this species (Melillo, 2007). The ALT enzyme is found at high concentrations in cardiac muscle in rabbits (Hoffmann et al., 1982).

The ALT enzyme can be mildly elevated in healthy rabbits after exposure to low levels of lead or mycotoxins (Harcourt-Brown, 2002) or halothane anesthesia (Gil et al. 2007). This enzyme may also be elevated in rabbits with advanced hepatic neoplasia (Reznik and Padberg, 1991).

Liver enzymes in rabbits are not liver-specific, since ALT is found in liver and cardiac muscle, and AST is found in liver, as well as muscle, kidney, and pancreas (Vennen and Mitchell, 2009).

High levels of liver enzymes have been described in cases of viral hemorrhagic disease in rabbits (Melillo, 2007).

ASPARTATE AMINOTRANSFERASE (AST)

Reference levels of aspartate aminotransferase (AST) in the rabbit are 14–113 IU/l (Table 3.3). The AST enzyme is found in many tissues in rabbits, including the heart and liver, and it has a relatively short half-life in the rabbit of approximately 5 hours (Melillo, 2007).

Levels of AST may be elevated due to disease, environment, or technical reasons. Increased AST is found in rabbits with liver damage and in rabbits anesthetized with halothane or isoflurane (Gil et al., 2007). Experimental infection with *Eimeria stiedae* has also been shown to produce elevated AST levels in rabbits (Gomez-Bautista et al., 1987). Experimental heat stress induced by hyperthermia in rabbits causes a three-fold increase in AST (Marder et al., 1990). Decapitation, heart puncture, or aortic incision produces an artificial elevation of AST due to muscle cell damage (Lindena and Trautschold, 1986). The AST value may also be elevated by physical restraint of a rabbit or when the blood sample becomes hemolyzed (Melillo, 2007).

Liver enzymes in rabbits are not liver-specific, since ALT is found in liver and cardiac muscle, and AST is found in liver, as well as muscle, kidney, and pancreas (Vennen and Mitchell, 2009).

High levels of liver enzymes have been described in cases of viral hemorrhagic disease in rabbits (Melillo, 2007).

LACTATE DEHYDROGENASE (LDH)

Reference levels of lactate dehydrogenase (LDH) in the rabbit are 30–140 IU/l (Table 3.3). The LDH enzyme is not usually helpful in disease diagnosis because it is produced by both muscle and liver cells.

The LDH value may be increased by physical restraint of a rabbit or when the blood sample is hemolyzed because of its high concentration in erythrocytes (Melillo, 2007). Rabbits exposed to noise stress have elevated LDH levels (de la Fuente et al., 2007).

CREATINE KINASE (CK)

Reference levels of CK in the rabbit are 150–1000 IU/l (Table 3.3). Dutch rabbits are reported to have elevated levels of the MM isoform of CK compared to other rabbit breeds and other animals (Yi et al., 1991).

Creatine kinase is commonly increased in the serum by physical restraint or the process of blood collection (Melillo, 2007). The different CK isoforms may be elevated depending on the method of blood collection. For example, the CK-MM and CK-BB isoforms are predominant after blood collection from peripheral veins, and the CK-MB isoform is predominant after blood collection by cardiac puncture (Lindena and Trautschold, 1986). Rabbits exposed to stressors, such as cold, noise, and housing with unfamiliar rabbits, have elevated levels of CK (de la Fuente et al., 2007).

SORBITOL DEHYDROGENASE (SDH)

Reference levels of sorbitol dehydrogenase (SDH) in the rabbit are 170–177 IU/l (Table 3.3).

AMYLASE

Reference levels of amylase in the rabbit are 200–500 U/l (Table 3.3).

In the rabbit, amylase is produced almost exclusively in the pancreas, with little contribution from the salivary glands, intestines, or liver. Amylase levels are

lower in the serum of rabbits than in most other animals (Lindena and Trautschold, 1986).

Elevated amylase levels in rabbit serum may be caused by pancreatitis, pancreatic duct obstruction, peritonitis, or renal failure, or treatment with corticosteroids.

Amylase values may be lower in hemolyzed blood samples.

GLUTAMATE DEHYDROGENASE (GDH)

The reference level of glutamate dehydrogenase (GDH) in the rabbit is 6.39 IU/l (Table 3.3). There is little information on the diagnostic value of this enzyme in rabbits. Its concentration in the centrilobular area of the liver could potentially identify the primary site of hepatocyte damage in liver disease in this species, as in others. Rabbits, like guinea pigs, are different from most animals in that GGT is released into the serum as clotting occurs (Clifford and White, 1999).

OTHERS

Atropine esterase is present in approximately one-third of domestic rabbits. This enzyme hydrolyzes atropine and thus interferes with the pharmacologic action of atropine in many rabbits (Hrapkiewicz et al., 1998).

Hormones

ADRENAL HORMONES

Reference levels of adrenal hormones in the rabbit include 2.6–3.8 µg/dl cortisol and 1.54 µg/dl corticosterone (Table 3.4). Cortisol is the predominant steroid hormone found in rabbits (Fecteau et al., 2007).

The stress of handling and restraint in rabbits causes the release of steroids in an age- and strain-related manner (Redgate et al., 1981). Steroids can affect some biochemical assays (Lindena and Krautschold, 1986). Therefore, it is important to standardize handling methods, blood draw technique, and length of anesthesia in rabbits to reduce the impact of stress on assay results.

A study on the effect of inhalant anesthetics in New Zealand White rabbits found that halothane increases serum corticosterone levels, whereas isoflurane increases corticosterone and serotonin levels in this species (Gil et al., 2007). A recent study in New Zealand White rabbits showed that group housing produced lower white blood cell counts and higher plasma cortisol levels than in individually housed rabbits (Fuentes and Newgren, 2008). Heat stress has also been shown to produce increased cortisol levels in rabbits (de la Fuente et al., 2007). Changes in leukocytes correlated with changes in cortisol levels (Toth and Krueger, 1989).

Stress causes an increase in blood pressure due to increased release of adrenocorticotropic hormone (ACTH) and adrenaline. This results in a decreased marginal pool of neutrophils as they move to the central pool and are found at higher levels in the venous blood. Halothane anesthesia, which is now rarely used, has also been shown to increase ACTH levels in the rabbit (Gil et al., 2007).

THYROID HORMONES

Reference levels of thyroid hormones in the rabbit include 130–143 ng/dl triiodothyronine (T3), and 1.7–2.4 µg/dl thyroxine (T4) (Table 3.4).

REPRODUCTIVE HORMONES

Reference levels of reproductive hormones in the rabbit include 10–100 ng/ml luteinizing hormone (LH), 2–4.5 ng/ml follicle stimulating hormone (FSH), 3 pg/ml estradiol, <1 ng/ml progesterone, 10 ng/ml prolactin, 10–20 ng/ml oxytocin, and 3–5 pg/ml testosterone (Table 3.4). There was no difference found between adult male and female neutered rabbits in the serum concentrations of several reproductive hormones, including progesterone, 17-hydroxy-progesterone, androstenedione, and testosterone. Elevated prolactin levels and enlarged mammary glands have been described in a 4-year-old female New Zealand White rabbit with a prolactin-secreting pituitary adenoma (Sikoski et al., 2008).

Liver Function

TOTAL BILIRUBIN

Reference levels of total bilirubin in the rabbit are 0–0.8 mg/dl (Table 3.5). The rabbit produces a large amount of bile per body weight when compared with other animals. Biliverdin is the primary component of rabbit bile, comprising approximately 70% of excreted bile. There is no assay available for biliverdin and it is therefore not measured. Approximately one-third of biliverdin is converted to bilirubin, which is measured in the blood.

In the rabbit, bilirubin is secreted by the liver in the monoconjugated form, in contrast to the diconjugated form secreted in most species (McLaughlin and Fish, 1994). Bilirubin comprises approximately 30% of the excreted bile.

Elevated bilirubin concentration is typically a marker of liver dysfunction or hemolytic disease. In rabbits, hyperbilirubinemia is caused primarily by bile flow obstruction. This obstruction can be the result of hepatic coccidiosis in young rabbits or biliary neoplasia, hepatic lipidosis, or liver lobe torsion in older rabbits. Hepatic fibrosis and hyperbilirubinemia may also be a result of aflatoxicosis from ingesting moldy food (Melillo, 2007).

BILE ACIDS

Reference levels of bile acids in the rabbit are 0–40 mMol/l (Table 3.5), although this is not a standard

biochemical assay in this species. Serum bile acids are used as a measure of liver function in other animals by comparing pre- and postprandial levels, but cecotrophy makes it difficult to fast a rabbit for the pre-prandial sample.

Persistently elevated bile acid concentrations have been reported in rabbits with liver disease (Harcourt-Brown, 2002).

SERUM PROTEINS

The reference range for total serum protein levels in the rabbit is 5.4–8.3 g/dl (Table 3.5), which is higher than those in most other animals (Vennen and Mitchell, 2009). Total protein levels vary by rabbit strain, with descending reference values in Polish rabbits, Dutch Belted rabbits, and New Zealand White rabbits, respectively (Kozma et al., 1974). There is also an effect of age on total proteins in the rabbit, as these values are lowest at birth and increase with age to reach adult values by 12 weeks old (Jain, 1986).

The reference range for albumin levels in the rabbit is 2.4–4.6 g/dl, and the reference range for globulin levels is 1.5–2.8 g/dl (Table 3.5). The albumin fraction of the total protein value is higher in female New Zealand White rabbits than in males (Kozma et al., 1974), but lower in pregnant females than in other adults. Caesarian-derived neonates have low total protein levels, with low albumin and high alpha-globulin levels (Bortolotti et al., 1989).

Feed restriction has been shown to result in lower total protein levels, albumin, and triglycerides in late-gestation pregnant rabbits and to produce a high percentage of near-term abortions (Matsuoka et al., 2009).

Kidney and liver disease are usually associated with decreased albumin and total protein, whereas hyperthermia, dehydration, or shock cause elevated total proteins (Marder et al., 1990). Coronavirus infections have been shown to produce increased globulins in rabbits.

Hemolyzed serum samples can produce articially increased total protein values (Melillo, 2007).

Cold stress causes an increase in total plasma proteins and beta globulins, and a decrease in serum albumin in the rabbit.

Kidney Function

BLOOD UREA NITROGEN (BUN)

The reference range for blood urea nitrogen (BUN) concentratiom in the rabbit is 10–30 mg/dl (Table 3.6), and these levels vary according to several factors. Rabbit urea levels increase in late afternoon and early evening and are higher in female than in male New Zealand White rabbits (Melillo, 2007). Polish rabbits are reported to have higher BUN levels than New Zealand White or Dutch Belted strains (Kozma et al., 1974).

Some causes of elevated BUN include urolithiasis and obstruction of urine flow, or kidney tumors or ureteral ligation (Adjarov et al., 1976). Low BUN may be due to hepatic insufficiency or loss of muscle mass due to dental disease (Melillo, 2007).

CREATININE

The reference range for creatinine levels in the rabbit is 0.5–2.5 mg/dl (Table 3.6) and can be increased by renal insufficiency or muscle injury. Creatinine, and often BUN, is elevated when there is reduced blood flow to the kidneys associated with 50–70% loss in renal function (Vennen and Mitchell, 2009). Dehydration can cause pre-renal azotemia in rabbits due to the limited ability of this species to concentrate urine. Blood urea nitrogen and creatinine levels are also increased after stroke, cardiac disease, or bleeding within the gastrointestinal tract (Melillo, 2007).

A study on the effect of inhalant anesthetics in New Zealand White rabbits found that halothane and isoflurane both cause increased levels of BUN and creatinine (Gil et al., 2007).

Renal failure in rabbits can be a result of renal disease due to neoplasia, glomerular nephritis, or pyelonephritis. In addition, renal failure can result from pre-renal disease, such as dehydration, or post-renal disease, such as urethral calculi (Vennen and Mitchell, 2009). The parasite *E. cuniculi* is the most common cause of renal failure in rabbits due to granulomas and fibrosis in the renal parenchyma (Melillo, 2007).

Electrolytes

BICARBONATE (HCO_3^-)

The reference range for the bicarbonate anion in the rabbit is 16.2–38.0 mEq/l (Table 3.7). Rabbit kidneys have minimal ability to correct acid–base imbalances, making this species fairly susceptible to electrolyte alterations. Anorexia in rabbits may also result in metabolic acidosis (Melillo, 2007).

CALCIUM (Ca)

Total serum calcium (Ca) concentration is higher in normal rabbits than in other mammals and has a wide range of reference values in this species, from 5.6–17 mg/dl (Table 3.7). Calcitrol is a metabolite of vitamin D and has a major effect on serum calcium levels in most animals by regulating calcium absorption from the gut. In contrast, gut absorption in the rabbit is independent of calcitrol, and serum calcium concentration depends directly on calcium levels in the diet (Brewer, 2006). Total serum calcium concentrations up to 15 mg/dl and higher have been reported in the rabbit (Hrapkiewicz and Medina, 2007; Melillo, 2007). Young rabbits and pregnant does have a higher calcium requirement than normal adult rabbits,

resulting in lower blood calcium levels in these conditions (Melillo, 2007). Ionized calcium (Ca^{2+}) concentration in rabbit serum is similar to that of other animals.

Excess calcium is excreted in the urine of rabbits, compared to bile excretion in other animals (Hrapkiewicz and Medina, 2007), and the Ca level in urine varies with serum calcium concentration. Urinary excretion rate of calcium is 45–60% in rabbits, compared to 2% in most other mammals (Melillo, 2007). This situation predisposes the rabbit to the formation of Ca-rich uroliths. Hypercalcemia can result when rabbits are unable to eliminate excess calcium and may be a sign of renal disease (Melillo, 2007).

Hypocalcemia is rare in rabbits but may be indicative of poor nutrition, hypoalbuminemia, diarrhea, chronic renal failure, or hyperparathyroidism (Vennen and Mitchell, 2009). Parathyroid hormone levels affect blood calcium levels, and calcium is readily moved from the blood to bones in rabbits (Melillo, 2007). Seizures resulting from hypocalcemia have been reported in late-pregnant and lactating does (Kerr, 1989).

CHLORIDE (Cl^-)

Reference levels of chloride (Cl^-) levels in rabbits are 92–120 mEq/l (Table 3.7) and remain fairly constant across different ages and sexes (Fox and Laird, 1999). Differences in chloride levels have been reported when compared among different strains of rabbits (Fox et al., 1970). Chloride levels are increased in rabbits with dehydration or increased dietary salt intake, and decreased in rabbits with diarrhea or low dietary salts (Vennen and Mitchell, 2009).

MAGNESIUM (Mg^{2+})

Magnesium (Mg^{2+}) levels in normal rabbits are 2.0–5.4 mg/dl (Table 3.7) and are not significantly affected by age, sex, or strain. Increased magnesium levels are seen in rabbits with dehydration, hyperthyroidism, tissue trauma, or adrenocortical insufficiency (Fox and Laird, 1999).

In the rabbit, Mg^{2+} ions are excreted mainly in the urine instead of in the bile.

PHOSPHORUS (P)

Phosphorus (P) levels in normal rabbits are 2.3–6.7 mg/dl (Table 3.7) and show significant differences between strains of rabbits (Fox and Laird, 1970).

Increased phosphorus concentration is indicative of renal disease in rabbits, whereas decreased phosphorus occurs with urinary Ca^{2+} excretion (Vennen and Mitchell, 2009).

Hemolysis of the blood sample may cause an artificially elevated phosphorus value.

POTASSIUM (K^+)

The reference range for potassium (K^+) in normal rabbits is 3.5–6.9 mEq/l (Table 3.7) and may fluctuate physiologically in rabbits, as in other herbivores (Melillo, 2007).

Hyperkalemia in the rabbit is usually due to acute renal failure, obstructed urine flow, or metabolic acidosis. Hyperkalemia in this species can also be due to severe tissue damage and loss of potassium into the extracellular space, or by excess administration of potassium-rich fluids (Melillo, 2007). Artificially elevated potassium values may be obtained when a blood sample is hemolyzed for technical reasons.

Low potassium levels are seen in cases of acute renal failure, starvation, low-potassium diet, or after administration of excess potassium-poor fluids (Vennen and Mitchell, 2009). Hypokalemia can also result from a stress-induced increase in catecholamines. A condition referred to as "floppy rabbit syndrome" has been associated with low potassium levels in the blood (Harcourt-Brown, 2002). Potassium values may also be decreased for technical reasons, including the use of plastic tubes for blood collection (Caisey and Kling, 1980) and hemolysis of serum samples (Melillo, 2007).

SODIUM (Na^+)

The reference range for sodium (Na^+) in rabbits is 131–155 mEq/l (Table 3.7), and levels vary considerably among different strains of rabbits (Fox and Laird, 1970). Sodium level in rabbits is not very useful as a diagnostic indicator. The serum sodium value may be artificially low in rabbits with lipemia or hyperproteinemia. True hyponatremia in the rabbit may be associated with acute or chronic renal failure and polyuria (Melillo, 2007). Sodium depletion may also be the result of polydipsia in fasted rabbits that drink excessively (Cizek, 1961). Sodium is elevated in rabbits with dehydration or fluid loss due to diarrhea, peritonitis, burns, or myiasis (Melillo, 2007).

GUINEA PIG

Sample Collection

Restraint

Guinea pigs are generally docile, especially with frequent and gentle handling. However, guinea pigs are difficult to restrain for blood and urine samples because they resist being held securely, and their short legs and neck make it difficult to access peripheral vessels without anesthesia. Guinea pigs should be held under the trunk with one hand and supported under the hind end with the other hand, being careful not to constrict the thorax or abdomen (Hrapkiewicz and Medina, 2007).

Blood

Venipuncture in guinea pigs is difficult because the peripheral veins in this species are small and often covered by layers of fat. It is often necessary to shave

the skin and apply alcohol to help visualize peripheral veins in this species (Dyer and Cervasio, 2008).

Small volumes of blood can be obtained in unanesthetized guinea pigs from a variety of peripheral veins using a sterile lancet or a 23G–27G needle. A marginal ear vein can be nicked with a 25G needle and blood drops collected directly into a microhematocrit tube. The lateral saphenous vein runs across the tarsal joint and can be sampled with a 1-ml syringe on a 23G–27G needle after applying digital pressure on the thigh to hold off the vein. The cephalic vein and tarsal vein are also accessible on the guinea pig, although only small volumes can be obtained from these sites, and bruising and hemorrhage often result. Alternatively, a toenail can be cut short using a nail clipper to yield small volumes of blood (Clifford and White, 1999).

Larger volumes of blood can be obtained from anesthetized guinea pigs using large central veins, including the jugular vein, cranial vena cava, and femoral vein. The short neck of the guinea pig makes the jugular vein and cranial vena cava difficult to access, but shaving and proper positioning allow optimal exposure (Pilny, 2008). In the guinea pig, the right jugular vein is larger than the left and will yield up to 2.5 ml of blood. A 24G needle on a 1-ml syringe can be directed caudally to enter the jugular vein just cranial to the clavicle (Shomer et al., 1999). The cranial vena cava can be accessed with a 22G or 23G needle on a 3- or 6-ml syringe that is directed caudally into the sternal notch under the first rib. A ketamine–xylazine mixture has been described as the preferable method of anesthesia for this route of blood sampling (Dang et al., 2008). However, the cranial position of the heart makes cardiac hemorrhage a significant risk when blood is sampled from the cranial vena cava (Joslin, 2009). The femoral vein can be used to collect up to 3 ml of blood with the guinea pig under anesthesia, in dorsal recumbency, and with the rear leg abducted. A 23G needle on a 1–3-ml syringe can be directed at a 45–90° angle to the skin and inserted just ventral to the pulsing femoral artery within the femoral triangle (Joslin, 2009).

Alternative sites for blood collection in a guinea pig under anesthesia include an interdigital vein and the retro-orbital sinus. The interdigital vein technique in the guinea pig is adapted from that described in the rat (Snitily et al., 1991). The foot is disinfected with alcohol and stimulated by rubbing with a methyl salicylate soaked gauze. Approximately 0.4 ml of blood is collected into a capillary tube after puncturing the interdigital vein with a 20G needle. The sample is then transferred into a sodium citrate solution, and pressure is applied to the sampling site for hemostasis (Keino et al., 2002). The retro-orbital sinus will also yield small volumes of blood in an anesthetized guinea pig (Joslin, 2009).

Cardiocentesis can be performed as a terminal procedure in an anesthetized guinea pig. The animal is placed in dorsal recumbency, and a 20G–25G needle is advanced toward the heart under the xiphoid cartilage to enter the left ventricle. Alternatively, up to 15 ml of blood can be obtained from the descending aorta or caudal vena cava following a laparotomy as a terminal procedure in an anesthetized guinea pig (Joslin, 2009). Decapitation, performed by a trained individual, may yield 10–15 ml of trunk blood (Clifford and White, 1999).

Urine

Urine may be readily collected from the guinea pig using a metabolic cage, free catch after manual pressure on the bladder, or placement on a cold surface (Hrapkiewicz and Medina, 2007). If a urine culture is needed, a cystocentesis should be performed using sedation and aseptic preparation, as described for the rabbit and other animals (Riggs, 2009).

Reference Values and Sources of Variation

Blood

Total blood volume and plasma volume of the guinea pig are discussed in the Hematology section of this chapter.

Clotting of guinea pig blood occurs more slowly when collected into plastic tubes (Clifford and White, 1999).

Urine

Characteristics and reference values of normal guinea pig urine are presented in Table 3.1.

Guinea pigs are herbivores and produce alkaline urine, with a pH of 8–9 (Riggs, 2009).

Normal guinea pig urine is yellow to amber in color, and is fairly opaque. Dietary factors may cause the urine to be darker and more orange in color. It is important to distinguish dietary pigmentation from hematuria using a dipstick to detect blood or Wood's lamp to detect porphyrins. Crystalluria is not uncommon in the guinea pig and should prompt a thorough examination for urinary calculi (Riggs, 2009). A study of urinary calculi in guinea pigs found that the vast majority are composed of 100% calcium carbonate (Hawkins et al., 2009).

Urinary tract infections may occur in older female guinea pigs, and uroliths are an occasional finding in this species (Hrapkiewitz and Medina, 2007).

Carbohydrate Metabolism

GLUCOSE

Reference range for blood glucose level in guinea pigs is 60–130 mg/dl (Table 3.2).

Glucose is elevated in guinea pigs with diabetes mellitus, as in other animals. Spontaneous diabetes mellitus has been reported in guinea pigs at 3–6 months old and is associated with hyperglycemia, glucosuria, and often ketonuria (Hrapkiewicz and Medina, 2007).

Differences in serum glucose have been reported in guinea pigs treated with different anesthetics. For example, serum glucose was higher in guinea pigs anesthetized with 30 mg/kg ketamine and 2.5 mg/kg xylazine IP, compared to 37 mg/kg pentobarbital IP prior to blood draw from the cranial vena cava. This difference was thought to be due to xylazine's inhibition of insulin secretion from pancreatic beta cells in the guinea pig, as in other animals (Dang et al., 2008).

Pregnancy toxemia may occur in the guinea pig in late gestation and takes one of two forms, a fasting or metabolic form and a circulatory or toxic form. The fasting or metabolic form is referred to as ketosis and occurs especially in obese guinea pigs in the first or second pregnancy. This condition may be triggered by stress factors, such as a change in environment or feeding routine, and often results in death. Controlled food intake to prevent obesity can help prevent this condition. In the circulatory or toxic form of pregnancy toxemia, the gravid uterus compresses the descending aorta and causes ischemia of the uterus and placenta (Hrapkiewicz and Medina, 2007).

Obese old male guinea pigs may also become ketotic as a result of reduced carbohydrate diet, resulting in mobilization of fat for energy (Hrapkiewicz and Medina, 2007).

Lipid Metabolism

CHOLESTEROL

The reference range for cholesterol levels in guinea pigs is 20–80 mg/dl (Table 3.2). These values are reported to increase gradually with age in male guinea pigs of the inbred Weiser-Maples strain (Kitagaki et al., 2005).

The lipoprotein fraction in guinea pigs contains primarily LDL, with little HDL present unless cholesterol is present in the diet. Changes in dietary fiber primarily affect LDL levels (Fernandez et al., 1997).

The intestine is the primary site of cholesterol synthesis in the guinea pig, as it is in humans, whereas the liver serves this function in most species (Clifford and White, 1999). The guinea pig is very sensitive to cholesterol in the diet, with biochemical changes and lesions produced by dietary cholesterol as low as 0.1%. High dietary cholesterol in the guinea pig causes hemolytic anemia, enlarged spleen, and hyperplastic bone marrow, and may result in death prior to the production of atherogenetic plaques (Yamanaka et al., 1967). Hypercholesterolemia in the guinea pig results in fatty infiltration of the liver and other tissues. The guinea pig serves as a good model for hypercholesterolemia in humans (Sullivan et al., 1993).

TRIGLYCERIDES

The reference range for triglyceride levels in the guinea pig is 10–70 mg/dl (Table 3.2). Triglyceride levels have been found to increase gradually with age in male and female guinea pigs of the inbred Weiser-Maples strain (Kitagaki et al., 2005).

Enzymes

ALKALINE PHOSPHATASE (ALP)

The reference range for alkaline phosphatase (ALP) levels in guinea pigs is 80–350 IU/l (Table 3.3). The ALP enzyme was found to decrease with body weight, as in other species (Clifford and White, 1999). Levels of ALP are reported to decrease sharply after birth and then level off in male and female guinea pigs of the inbred Weiser-Maples strain (Kitagaki et al., 2005).

A zinc-deficient diet causes an increase in ALP activity (Clifford and White, 1999), whereas fasting and ascorbic acid deficiency cause decreased ALP levels in guinea pigs (Mahmoodian et al., 1996; Tsuchiya and Bates 1994). Hairless Dunkin-Hartley guinea pigs are reported to have lower ALP levels compared to normal Dunkin-Hartley guinea pigs, although the significance of this difference is not known (Waner et al., 1996).

ALANINE AMINOTRANSFERASE (ALT)

The reference range for alanine aminotransferase (ALT) levels in guinea pigs is 10–90 IU/l (Table 3.3), and these values are generally lower in this species than in other animals (Clampitt and Hart, 1978).

Elevated ALT is not a specific or sensitive marker of hepatocellular injury in the guinea pig compared to other animals (Clifford and White, 1999).

Hairless Dunkin-Hartley guinea pigs are reported to have higher ALT concentration compared to normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

ASPARTATE AMINOTRANSFERASE (AST)

The reference range for aspartate aminotransferase (AST) levels in guinea pigs is 10–90 IU/l (Table 3.3).

As with ALT and ALP, AST levels are reported to be lower in hairless Dunkin-Hartley guinea pigs than in normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

LACTATE DEHYDROGENASE (LDH)

The reference range for lactate dehydrogenase (LDH) levels in guinea pigs is 20–120 IU/l (Table 3.3).

CREATINE KINASE (CK)

The reference range for creatine kinase (CK) levels in guinea pigs is 50–150 IU/l (Table 3.3).

Hairless Dunkin-Hartley guinea pigs are reported to have higher CK levels than normal Dunkin-Hartley guinea pigs (Waner et al., 1996). The method of anesthesia has been shown to affect CK levels in guinea pigs. For example, guinea pigs anesthetized with 37 mg/kg pentobarbital given intraperitoneally have higher CK levels than

those anesthetized with 30 mg/kg ketamine and 2.5 mg/kg xylazine given intramuscularly (Dang et al., 2008).

Elevated CK may be seen in guinea pigs with myopathy associated with a deficiency of vitamin E and selenium (Hrapkiewicz and Medina, 2007).

AMYLASE

The reference range for amylase levels in guinea pigs is 995–1239 IU/l (Table 3.3).

Hairless Dunkin-Hartley guinea pigs are reported to have higher amylase levels than normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

Hormones

ADRENAL HORMONES

The reference range for cortisol levels in guinea pigs is 5–30 µg/dl (Table 3.4). Cortisol is the primary glucocorticoid produced by the guinea pig, instead of corticosterone, which predominates in the mouse and rat (Clifford and White, 1999). Cortisol and corticosterone are readily interconverted in the guinea pig (Manin et al., 1982). Plasma cortisol levels vary by age and sex in the guinea pig, probably due to differences in its binding to transcortin (El Hani et al., 1980). Transcortin in guinea pigs has a higher binding affinity for cortisol than for progesterone, whereas transcortin binds both hormones with similar affinity in humans (Clifford and White, 1999). Plasma cortisol levels in males are higher than in females at ages greater than 30 days, similar in males and females at 90–120 days, and higher in adult male guinea pigs (Clifford and White, 1999).

THYROID HORMONES

Reference ranges for thyroid hormone levels in guinea pigs are 22–56 ng/dl for triiodothyronine (T3) and 2.3–3.5 µg/dl for thyroxine (T4) (Table 3.4).

REPRODUCTIVE HORMONES

Reference levels of reproductive hormones in guinea pigs include 5–55 ng/ml luteinizing hormone (LH), 100–200 ng/ml follicle stimulating hormone (FSH), 30 pg/ml estradiol, 1 ng/ml progesterone, and <5 ng/ml oxytocin (Table 3.4).

Guinea pigs, like other hystricomorphs, have a progesterone-binding globulin that is present during pregnancy and binds progesterone but not cortisol (Wriston, 1984).

Liver Function

TOTAL BILIRUBIN

The reference range for total bilirubin levels in guinea pigs is 0–1 mg/dl (Table 3.5).

Blood samples exposed to sunlight demonstrate bilirubin decreases up to 50% in 1 hour (Benjamin and McKelvie, 1978). Freezing of serum samples may also cause an artificial decrease in bilirubin levels (Bayard, 1974).

SERUM PROTEINS

The reference range for total protein levels in guinea pigs is 4–7 g/dl (Table 3.5). Total protein is reported to increase sharply and level off at 150–200 days old in male and female guinea pigs of the inbred Weiser-Maples strain (Kitagaki et al., 2005).

The reference range for albumin levels in guinea pigs is 2–5 g/dl (Table 3.5). Hairless Dunkin-Hartley guinea pigs are reported to have higher total protein and albumin levels compared to normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

The reference range for globulin levels in guinea pigs is 2–4 g/dl (Table 3.5).

Kidney Function

BLOOD UREA NITROGEN (BUN)

The reference range for blood urea nitrogen (BUN) levels in guinea pigs is 9–32 mg/dl (Table 3.6). Levels of BUN were found to increase gradually with age in male and female guinea pigs of the inbred Weiser-Maples strain (Kitagaki et al., 2005).

The concentration of BUN was found to be higher in guinea pigs anesthetized with 37 mg/kg pentobarbital given intraperitoneally compared to 30 mg/kg ketamine and 2.5 mg/kg xylazine given intramuscularly. This difference was thought to be due to pentobarbital's depression of cardiac output, renal blood flow, and glomerular filtration rate (Dang et al., 2008).

Renal disease is a common chronic condition in older guinea pigs, whereas acute renal disease may occur in younger animals. For example, acute renal disease, with elevated BUN and creatinine, has been described in a 1-year-old guinea pig after ingestion of oxalate-containing plants (Holowaychuk, 2006).

CREATININE

The reference range for creatinine levels in guinea pigs is 0.6–2.2 mg/dl (Table 3.6). Creatinine levels were found to increase gradually with age in male and female guinea pigs of the inbred Weiser-Maples strain (Kitagaki et al., 2005).

Hairless Dunkin-Hartley guinea pigs are reported to have higher creatinine levels compared to normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

Electrolytes

CALCIUM (Ca)

The reference range for calcium levels in guinea pigs is 5.3–12.0 mg/dl (Table 3.7).

Hypercalcemia is rare in guinea pigs and is usually associated with poor nutrition and low phosphorus levels (O'Rourke, 2004).

Hairless Dunkin-Hartley guinea pigs are reported to have higher levels of most electrolytes measured compared to normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

CHLORIDE (Cl^-)

The reference range for chloride levels in guinea pigs is 90–115 mEq/l (Table 3.7). Chloride levels are reported to decrease gradually with age in male guinea pigs of the Weiser-Maples strain (Kitagaki et al., 2005).

MAGNESIUM (Mg^{2+})

The reference range for magnesium levels in guinea pigs is 3.5–4.1 (Table 3.7).

A dietary imbalance of minerals, such as low magnesium and high phosphorus, can cause soft tissue calcification in guinea pigs older than 1 year. Mineral deposits may be found in the elbows and ribs or more widespread, in lung, trachea, heart, aorta, liver, and kidney (Hrapkiewitz and Medina, 2007).

PHOSPHORUS (P)

The reference range for phosphorus levels in guinea pigs is 3.0–12.0 mg/dl (Table 3.7).

Phosphorus was found to be higher in guinea pigs anesthetized with 37 mg/kg pentobarbital given intraperitoneally compared to 30 mg/kg ketamine and 2.5 mg/kg xylazine given subcutaneously (Dang et al., 2008).

POTASSIUM (K^+)

The reference range for potassium levels in guinea pigs is 4.0–8.0 mEq/l (Table 3.7).

Serum potassium values may be lower when blood is allowed to clot in plastic rather than glass tubes, because of slower clotting and decreased release of potassium from platelets (Caisey and Kling, 1980).

SODIUM (Na^+)

The reference range for sodium levels in guinea pigs is 120–150 mEq/l (Table 3.7). Sodium levels are reported to increase gradually with age in female guinea pigs of the Weiser-Maples strain (Kitagaki et al., 2005).

In contrast to most electrolytes, sodium levels are reported to be lower in hairless Dunkin-Hartley guinea pigs compared to normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

HAMSTER

Sample Collection

Restraint

Hamsters are fairly docile when handled frequently but are typically more aggressive than mice. Hamsters can be restrained manually by gathering the loose scruff skin in one hand and supporting the lower body against the palm of the same hand. A hamster may also be placed in a restraint tube for blood collection without anesthesia. Such a tube should have airholes at the nose end and be cleaned frequently to reduce the risk of cross-infection or stress from pheromones (Hem et al., 1998).

Blood

Small blood samples can be obtained in an unanesthetized hamster from the lateral saphenous vein, which runs dorsally and laterally across the tarsal joint. The hamster may be restrained manually or in a restraint tube, as described above, and the vein is held off with gentle pressure above the knee joint. A 20G–25G needle can be inserted into the vein, and blood is collected from the needle hub (Hem et al., 1998). Blood collection occasionally results in bruising, hemorrhage, or temporary favoring of the sampled limb (Joslin, 2009). The lateral vein of the tarsus may also be used to collect small volumes of blood without anesthesia. A clipped toenail, or a small cut in the ear, tail, or footpad, have also been described for obtaining a small blood sample in the hamster (Buetow et al., 1999; Hrapkiewicz and Medina, 2007).

Anesthesia is needed for blood collection from the cephalic vein in a hamster. A tourniquet is placed around the proximal leg, a 25G needle is inserted in the vein, and blood is collected from the needle hub (Joslin, 2009). Other peripheral veins that can be sampled under anesthesia include the jugular vein, femoral vein, sublingual vein, and ear vein (Joslin, 2009).

The retro-orbital venous plexus is commonly used for blood sampling in the hamster and can be accessed by penetrating the conjunctiva midway along the superior border of the eye (Dyer and Cervasio, 2008). A lateral or medial approach to the retro-orbital venous plexus is also described (Buetow et al., 1999). This method may be used by experienced personnel in unanesthetized hamsters, but anesthesia is recommended to avoid stress and damage to the eye. Up to 3 ml of blood can be withdrawn from this site in an adult Syrian hamster (Buetow et al., 1999), but there is risk of ocular damage using this method (Joslin, 2009).

Hamsters can be anesthetized to obtain a 1-ml sample of blood from the cranial vena cava using a 23G–25G needle inserted lateral to the manubrium, cranial to the first rib, and directed caudally. Cardiac puncture has been used for obtaining 1–2 ml of blood in the anesthetized hamster with a 3/8-inch 25G needle (Mitruka and Rawnsley, 1981). However, this technique requires practice, and repeated sampling is associated with a higher risk of mortality (Buetow et al., 1999).

Terminal blood collection of up to 5 ml in a Syrian hamster can be performed under anesthesia by cardiocentesis, laparotomy, and venipuncture of the caudal vena cava, or by translumbar vena cava collection (Joslin, 2009).

Urine

Hamsters often urinate when they are picked up, so small urine samples can be obtained by this method. A metabolic cage may be used to collect larger samples of urine from a hamster. This method is advantageous for separating urine from fecal pellets, which are typically mixed on the floor of a conventional hamster cage.

Reference Values and Sources of Variation

Blood

Total blood volume of the hamster is discussed in the Hematology section of this chapter.

Urine

Characteristics and reference values of hamster urine are presented in Table 3.1.

Hamsters produce up to 7 ml of urine per day. Chinese hamsters with diabetes mellitus may have significantly increased urine excretion of up to 75 ml per day (Fiszer et al., 1979). The pH of hamster urine is normally alkaline and varies from 5.1–8.4.

Hamster urine is cloudy due to the normal presence of crystals and protein. Protein excretion rate in hamster urine is almost 10 mg per week, which is approximately ten times higher than that in humans (Tietz, 1976). Cholesterol is the main lipid present in hamster urine, with an excretion rate of 11.7 mg per week (Cox and Gökcen, 1973). Hamster urine has variable levels of sodium and potassium, averaging 70 mM sodium and 120 mM potassium (Fiszer et al., 1979). The primary crystals present in hamster urine include calcium carbonate and triple phosphate (Schuchman, 1980).

Carbohydrate Metabolism

GLUCOSE

The reference range for blood glucose concentration in the hamster is 37–198 mg/dl (Table 3.2). Glucose level is affected by many factors, including diet, restraint, anesthesia, hibernation, and diurnal variation, as well as certain disease states (Tomson and Wardrop, 1987). These conditions must be taken into account when evaluating blood glucose concentration.

Blood glucose levels in the hamster are affected by dietary factors, including feeding schedule, duration of fast, and the food content of the stomach (Feldman et al., 1982).

The stress of restraint is likely an important factor producing elevated blood glucose in hamsters. This is demonstrated by higher glucose concentrations in blood samples repeated after 15 minutes. In addition, hamsters sedated for blood sampling yield lower glucose values than those that are unsedated (Tomson and Wardrop, 1987).

Thiobarbiturate anesthesia has been found to increase blood glucose to 300 mg/dl in adult male Syrian hamsters, compared to approximately 145 mg/dl in unanesthetized controls. This hyperglycemia has been found to persist for several hours (Turner and Howards, 1977).

Elevated glucose levels are reported during hibernation in the hamster. The hamster also has a circadian rhythm of glucose concentrations, with the lowest level just before the start of the dark period and the highest level at the start of the light period (Cincotta and Meier, 1984). This effect may be mediated by melatonin secretion by the pineal gland (Ortega-Corona et al., 1991).

Insulin-induced hypoglycemia is also affected by diurnal factors, with no hypoglycemia at 4 hours after light onset and significant hypoglycemia at 8–20 hours after light onset.

The Chinese hamster is often used as a model of diabetes mellitus due to the high incidence of the disease in this strain. Glucose levels of up to 500 mg/dl may be seen in diabetic Chinese hamsters, and this level can be decreased with lithium supplementation in the drinking water (Hu et al., 1997).

Lipid Metabolism

CHOLESTEROL

The reference range for cholesterol concentration in the hamster is 112–210 mg/dl (Table 3.2), which represents the highest reference range of normal rodents and rabbits (Cox and Gökcen, 1973). Cholesterol levels in the hamster are affected by a number of factors, including diet, strain, environment, diurnal variation, and temperature, as well as disease.

Cholesterol levels in the hamster are significantly affected by diet. A high-cholesterol, high-triglyceride diet is associated with elevated plasma cholesterol levels in the hamster. These abnormal levels can usually be reduced by fasting (Sullivan et al., 1993). A high-fructose diet produces higher VLDL triglycerides, free cholesterol, and phospholipid levels in the hamster, but no change in apolipoprotein B levels. In addition, a high-fructose diet increases LDL triglycerides, decreases LDL cholesteryl ester, and increases HDL fractions (Wang et al., 2008).

Group housing causes increased cholesterol levels in hamsters fed an atherogenic diet when compared to singly housed controls (Yoganathan et al., 1998).

A decrease in photoperiod from 14 to 10 hours of light reduces cholesterol levels in hamsters.

Photoperiods of 10 hours or less cause a decrease in cholesterol with no effect on other plasma lipids (Gad, 2007). Low environmental temperature causes a decrease in plasma cholesterol but no change in plasma triglycerides (Vaughan et al., 1984), whereas hibernation causes an increase in serum lipids (Gad, 2007).

Disease conditions have an effect on cholesterol levels in the hamster. Chronic hepatitis and biliary disease cause hypercholesterolemia in the hamster, probably due to cholestasis (Brunnert and Altman, 1991). Diabetes in Chinese hamsters, alloxan treatment in Syrian hamsters, pertussis toxin, and amyloidosis are all associated with hypercholesterolemia (Garcia-Sainz et al., 1987; Gerritsen, 1982; Murphy et al., 1984).

TRIGLYCERIDES

The reference range for triglyceride concentration in the hamster is 72–350 mg/dl (Table 3.2). A high-carbohydrate diet increases plasma triglycerides in the hamster. Low environmental temperature is reported to produce no change in plasma triglycerides in the hamster (Vaughan et al., 1984).

TOTAL LIPIDS

The reference range for total circulating lipids in hamsters is 224–466 mg/dl (Table 3.2), which is higher than in other rodents (Cox and Gökcen, 1974; Mitruka and Rawnsley, 1977).

Enzymes

ALKALINE PHOSPHATASE (ALP)

The reference range for alkaline phosphatase (ALP) concentration in the hamster is 50–186 IU/l (Table 3.3). Levels of ALP are affected by the age, sex, and strain of hamster (Maxwell et al., 1985). Immature hamsters have higher ALP levels than adults because of its release from growing bone, as in other animals (Dent, 1977).

Hamster ALP includes isoenzymes from bone, liver, and intestine (Cox and Gökcen, 1973), and placenta (Manning et al., 1970). Bone provides the main isoenzyme in hamster, and intestinal ALP provides 10–15% of the total ALP activity in this species. The liver ALP isoenzyme is sensitive to heat and declines from weaning to undetectable levels in older hamsters (Tomson and Wardrop, 1987).

Increased ALP in the hamster is a sensitive indicator of liver damage resulting from viral-induced hepatitis, as well as chronic hepatitis with biliary disease due to intrahepatic cholestasis and increased ALP synthesis (Brunnert and Altman, 1991). Increased ALP in hamsters has been reported in association with leukemias, prostate gland tumors, and bone and liver cancers (Eugster et al., 1966).

ALANINE AMINOTRANSFERASE (ALT)

The reference range for alanine aminotransferase (ALT) levels in the hamster is 20–128 IU/l (Table 3.3) and may vary due to a variety of factors, including disease or technical issues.

The ALT enzyme is elevated in hamsters with hepatic necrosis caused by virus or acetaminophen toxicity (El-Hage et al., 1983; Rollinson and White, 1983). Elevated ALT is seen in Syrian hamsters with chronic hepatitis and is correlated with the number of affected hepatocytes. Chronic hepatitis can also result in decreased ALT due to decreased hepatocyte numbers (Duncan and Prasse, 1986). Hamsters experimentally infected with pirital virus (Arenaviridae) have increased ALT and AST 5–8 days post-infection (Sbrana et al., 2006).

The cardiac puncture method may result in higher serum levels of several enzymes, including ALT, AST, LDH, and CK, compared to blood sampled from other sites (Tomson and Wardrop, 1987).

ASPARTATE AMINOTRANSFERASE (AST)

The reference range for aspartate aminotransferase (AST) levels in the hamster is 20–150 IU/l (Table 3.3).

The AST enzyme is increased in hamsters with liver neoplasia and metabolic alterations (Eugster et al., 1966).

LACTATE DEHYDROGENASE (LDH)

The reference range for lactate dehydrogenase (LDH) levels in the hamster is 100–300 IU/l (Table 3.3). Levels of LDH are increased in hamsters with hereditary myopathy (Homburger et al., 1966) and after inoculation with some viruses (Eugster et al., 1966).

Freezing of hamster serum samples has been shown to decrease LDH activity (Tietz, 1976).

CREATINE KINASE (CK)

The reference value for creatine kinase (CK) level in the hamster is 23 IU/l (Table 3.3).

Creatine kinase has been found in hamster striated muscle and nervous tissue, and its level is used to diagnose myopathies, muscular dystrophies, and cardiac necrosis in this species. The Syrian hamster has a hereditary myopathy characterized by increased CK levels up to 730 IU/l compared to 23.2 IU/l in controls (Bajusz and Homburger, 1966).

AMYLASE

The reference range for amylase levels in the hamster is 154–196 IU/l (Table 3.3).

Amylase is present in the hamster pancreas, intestine, and liver, as in other animals (Takahashi et al., 1981).

Hormones

ADRENAL HORMONES

The reference range for cortisol levels in the hamster is 2.3–3.2 µg/dl (Table 3.4), which is lower than in many other rodent species. Male hamsters generally have larger adrenal glands and higher cortisol levels than female hamsters.

Pregnancy is associated with increased cortisol levels in hamsters, with elevations from 0.3–30 µg/dl as parturition approaches (Brinck-Johnsen et al., 1981).

Cortisol fecal metabolites have been extracted and measured by radioimmunoassay in hamsters undergoing surgery. Cortisol metabolites were found to increase immediately following surgery and decrease thereafter (Chelini et al., 2006). Plasma cortisol levels were also elevated after chronic stress in the hamster (Ottenweller et al., 1985).

The reference range for corticosterone levels in the hamster is 5.5–9.3 µg/dl (Table 3.4).

The hamster adrenal cortex contains distinct glomerulosa, fasciculata, and reticularis layers, as in other species, and ACTH administration increases adrenal weight (Alpert, 1950).

Adrenal gland activity is sensitive to diurnal variation, with maximal secretion approximately 3 hours before the onset of the dark period (Frenkel et al., 1965).

Cold temperatures do not stimulate ACTH release in the hamster (Frenkel et al., 1965).

THYROID HORMONES

The reference ranges for thyroid hormone levels in the hamster are 30–80 ng/dl for triiodothyronine (T3) and 3–7 µg/dl for thyroxine (T4) (Table 3.4). Several factors affect T3 and T4 levels in the hamster, including hibernation, age, strain, and temperature. Chronic exposure of hamsters to shortened photoperiod and lower temperature, as occurs during periods of hibernation, results in a decrease in levels of TSH, T3, and T4 (Hoffman et al., 1982; Vaughan et al., 1982). Increasing age in the hamster, from 3 to 20 months, is accompanied by deceasing levels of T3 and T4 (Neve et al., 1981). There is also a smaller response to TSH in older hamsters.

Hamsters have decreased protein-bound iodine during pregnancy, which may alter thyroid hormone metabolism during this condition (Galton and Galton, 1966).

REPRODUCTIVE HORMONES

The adult female hamster has a 4-day estrous cycle associated with predictable variations in the reproductive hormones, including follicle stimulating hormone (FSH), luteinizing hormone (LH), and progesterone (PG). The luteal phase includes day 1 (ovulation) and day 2, while the follicular phase, or proestrus, occurs on day 4 of the cycle. Hormonal fluctuations occur daily in the immature female hamster and shift to the mature 4-day cycle in the adult female (Donham and Stetson, 1991). Reproductive hormone levels have been well characterized in the hamster and are described below.

Reproductive hormone levels for both male and female hamsters are sensitive to photoperiod, with short periods of light producing decreased testosterone levels in males (Furuta et al., 1994), and decreased FSH, LH, and prolactin levels in females (Chandrashekar and Bartke, 1989).

LUTEINIZING HORMONE (LH) The reference range for LH levels in the hamster is 20–40 ng/ml (Table 3.4).

Normal adult female hamsters have one LH peak on the afternoon of proestrus (day 4), approximately 10 hours before ovulation. LH levels are fairly stable during days 1–3 of the estrous cycle in the Syrian hamster (Buetow et al., 1999). A similar pattern for LH has been reported in the Djungarian hamster (Erb and Wynne-Edwards, 1994).

FOLLICLE-STIMULATING HORMONE (FSH) The reference range for FSH levels in the hamster is 100–300 ng/ml (Table 3.4).

In the adult female hamster, FSH normally peaks at the time of the LH peak on the afternoon of proestrus (day 4) and then again on the morning of estrus (day 1), stimulating follicular growth for the next cycle. Levels of FSH are fairly stable during days 2 and 3 of the estrous cycle in the Syrian hamster (Buetow et al., 1999). A similar pattern for FSH has been reported in the Djungarian hamster (Erb and Wynne-Edwards, 1994).

Female Turkish hamsters demonstrate daily FSH variations when in anestrus and at immature stages (Ogilvie et al., 1992).

ESTRADIOL The reference range for estradiol levels in the hamster is 5–10 pg/ml (Table 3.4).

Estradiol levels are low during the luteal phase of the reproductive cycle of the hamster, showing an inverse relationship to levels of progesterone.

PROGESTERONE The reference value for progesterone level in the hamster is 1 ng/ml (Table 3.4).

Progesterone is the predominant reproductive hormone during the luteal phase, or day 1 and 2 of the hamster reproductive cycle. Progesterone then decreases and increases again on day 4, or proestrus (Buetow et al., 1999).

PROLACTIN The reference range for prolactin levels in the hamster is 5–10 ng/ml (Table 3.4).

Prolactin levels in the female hamster typically correspond with the persistence of corpora lutea during pregnancy and the onset of maternal behaviors

at parturition. During the estrous cycle, prolactin peaks each afternoon, with highest concentrations in proestrus. This pattern continues into gestation, but peak concentrations decrease as pregnancy advances (Buetow et al., 1999). In Djungarian hamsters, prolactin levels decrease in midgestation (Edwards et al., 1994).

Prolactin levels in most hamsters respond to photoperiod, although this response is less apparent in the Djungarian hamster (Ebling, 1994).

Prolactin increases in the female hamster in response to ether anesthesia and to the stress of handling (Matt et al., 1983), although similar stresses decrease prolactin and testosterone levels in the male hamster (Huhman et al., 1995).

TESTOSTERONE The reference range for testosterone levels in the hamster is 1.5–2.0 pg/ml (Table 3.4).

Liver Function

TOTAL BILIRUBIN

The reference range for bilirubin levels in the hamster is 0.1–0.9 mg/dl (Table 3.5), which are lower values than bilirubin in humans (Mitruka and Rawnsley, 1981).

Hamsters with hepatic dysfunction do not usually have abnormal serum bilirubin levels (Wardrop and Van Hoosier, 1989).

SERUM PROTEINS

The reference range for total protein concentration in the hamster is 5.2–7.0 g/dl (Table 3.5) and is affected by age, disease, and other factors. There are significant levels of fibrinogen in hamster plasma, making it necessary to accurately measure liver proteins in serum samples. Fibrinogen is linked to alpha-globulin rather than beta-globulin in the hamster.

The reference ranges for albumin and globulin concentrations in the hamster are 3.5–4.9 g/dl and 2.7–4.2 g/dl, respectively (Table 3.5). Albumin concentration decreases during the first year of life, whereas alpha-2-globulin increases by 6 months old, beta-globulin decreases at 8 weeks old, and levels of gamma-globulin and fibrinogen are variable between 8 weeks and 1 year of age (House et al., 1961). The albumin-to-globulin ratio decreases with age in the hamster.

Pregnancy in hamsters alters serum proteins, with decreased albumin and increased alpha-globulins in near-term hamsters. Hibernating hamsters have increased beta-globulin, albumin, and total protein, and decreased levels of gamma-globulin (South and Jeffay, 1958).

Disease states can also produce changes in protein concentrations. Experimentally induced inflammation produces an elevation of gamma-globulins (Betts et al., 1964). Hamsters experimentally infected with pirital virus (Arenaviridae) have decreased albumin 5–8 days post-infection (Sbrana et al., 2006).

Diabetes mellitus is associated with increased alpha-2 globulin levels in Chinese hamsters (Green and Yerganian, 1963). Older hamsters commonly have amyloidosis, which produces decreased levels of albumin and increased levels of gamma-globulin (Gleiser et al., 1971; Murphy et al., 1984).

Kidney Function

BLOOD UREA NITROGEN (BUN)

The reference range for blood urea nitrogen (BUN) concentration in the hamster is 12–26 mg/dl (Table 3.6) and is affected by diet, sex, and disease. Blood urea nitrogen is affected by the quality and quantity of the diet, as well as duration of fasting in the hamster. A diet containing 18–24% protein has been shown to produce a higher BUN level in the hamster than one with 12% protein (Feldman et al., 1982).

Some studies have demonstrated higher BUN values in the female hamster (Feldman et al., 1982; Maxwell et al., 1985), although findings have been variable.

Renal disease is common in older hamsters and is associated with increased BUN (Feldman et al., 1982). *Leptospira pomona* infection in the hamster is associated with elevated BUN, as high as 424 mg/dl, along with renal lesions and renal failure (Abdu and Sleight, 1965). Hamsters experimentally infected with pirital virus (Arenaviridae) were found to have increased BUN, and other abnormalities, 5–8 days post-infection (Sbrana et al., 2006).

CREATININE

The reference range for creatinine concentration in the hamster is 0.4–1.0 mg/dl (Table 3.6) and is affected by sex and age (Murphy et al., 1984) as well as disease states.

Elevated serum creatinine has been found in hamsters with renal amyloidosis and the nephrotic syndrome (Murphy et al., 1984). However, hamsters with chronic hepatitis were found to have no change in serum creatinine (Brunnert and Altman, 1991). Similarly, there is no difference in creatinine levels in inbred hypertensive (CHF 148) hamster strains compared to controls (Thomas et al., 1997).

Electrolytes

In general, electrolyte values in hamsters are similar to those of other rodents (see Table 3.7).

CALCIUM (Ca)

The reference range for calcium concentration in the hamster is 5.3–12.0 mg/dl (Table 3.7).

CHLORIDE (Cl^-)

The reference range for chloride concentration in the hamster is 93–110 mEq/l (Table 3.7).

PHOSPHORUS (P)

The reference range for phosphorus concentration in the hamster is 3.0–9.9 mg/dl (Table 3.7). These values are higher in younger hamsters than older hamsters (Dent, 1977). Phosphorus levels in blood can be affected by the age, sex, and strain of the hamster (Maxwell et al., 1985).

POTASSIUM (K)

The reference range for potassium concentration in the hamster is 3.9–6.0 mEq/l (Table 3.7).

Hemolysis of blood samples can result in artifactually elevated serum potassium levels (Bannon and Friedell, 1966).

SODIUM (Na^+)

The reference range for sodium concentration in the hamster is 128–150 mEq/l (Table 3.7).

OTHER RODENTS

Gerbil

Sample Collection

RESTRAINT

The gerbil may be restrained by grasping the base of the tail with one hand and the scruff of the neck with the other hand. Alternatively, the gerbil may be enclosed within one hand and held firmly in an upright position. Gerbils should not be picked up by the tip of tail due to the risk of degloving. Gerbils are not aggressive but will readily jump in an attempt to escape (Hrapkiewicz and Medina, 2007).

BLOOD

Blood samples can be collected more frequently in the gerbil than in other laboratory animals because of their rapid rate of erythrocyte turnover. The lateral tail vein can be used for blood collection in an unanesthetized gerbil, although caution should be taken because of the increased risk of tail degloving injury in this species (Joslin, 2009). Anesthesia should be used in the gerbil for blood collection from sites other than the tail vein. The saphenous vein or metatarsal vein can be distended using a tourniquet proximal to the stifle and a small blood sample is collected into the hub of a 22G needle. A 0.1–0.3-ml blood sample can be collected from the retro-orbital venous sinus of a gerbil by advancing a microhematocrit tube through the lateral canthus and holding off the external jugular vein behind the mandible (Joslin, 2009). The medial canthus is also a viable approach to the retro-orbital sinus in the gerbil (Dyer and Cervasio, 2008). Alternatively, small volumes of blood may be obtained from the lateral metatarsal vein, or from a clipped toenail. Large volumes of blood can be obtained by cardiac puncture under anesthesia as a terminal procedure in the gerbil, as in other species (Hrapkiewicz and Medina, 2007).

URINE

The gerbil is a desert species and produces small quantities of concentrated urine. However, a sample can usually be obtained when a gerbil is picked up (Hrapkiewicz et al., 1998). Other methods of urine collection, such as urinary catheterization or cystocentesis, require sedation or anesthesia.

Reference Values and Sources of Variation

BLOOD

Total blood volume of the gerbil is discussed in the Hematology section of this chapter.

CARBOHYDRATE METABOLISM

GLUCOSE The reference range for glucose concentration in the gerbil is 50–135 mg/dl (Table 3.2).

Gerbils are susceptible to obesity and may develop reduced glucose tolerance and diabetes mellitus.

LIPID METABOLISM

CHOLESTEROL The reference range for cholesterol concentration in the gerbil is 90–150 mg/dl (Table 3.2).

Cholesterol metabolism of the gerbil is similar to that of humans, so this species has been used to study the effects of dietary cholesterol and fats (Gordon and Cekleniak, 1961). Gerbils fed a diet with no cholesterol have higher alpha lipoprotein fractions than pre-beta or beta lipoprotein fractions (Forsythe, 1986). This finding has been confirmed with micro-affinity chromatography (Tschantz and Sunahara, 1993). Gerbils fed high-cholesterol diets often develop gallstones.

Gerbil serum is often lipemic, especially in older gerbils of either gender, although this condition is more common in males. Sunflower seeds in the diet may contribute to lipemic serum in gerbils (Heatley and Harris, 2009). Repeatedly bred male and female gerbils may develop hyperadrenocorticism with associated elevation of serum triglycerides and development of fatty livers (Percy and Barthold, 2007).

ENZYMES

ALKALINE PHOSPHATASE (ALP) The reference range for ALP concentration in the gerbil is 12–37 IU/l (Table 3.3).

HORMONES

ADRENAL HORMONES The gerbil adrenal gland is very large and secretes primarily cortisol.

Repeatedly bred male and female gerbils may develop hyperadrenocorticism with associated mineralization in the aorta and peripheral arteries, as well as

pathology of the pancreas, thymus, and adrenal glands. This condition is also associated with obesity and diabetes (Percy and Barthold, 2007).

LIVER FUNCTION

TOTAL BILIRUBIN The reference range for bilirubin concentration in the gerbil is 0.2–0.6 mg/dl (Table 3.5).

SERUM PROTEINS The reference range for total protein concentration in the gerbil is 4.3–12.5 g/dl (Table 3.5).

The reference ranges for albumin and globulin concentrations in the gerbil are 1.8–5.5 and 1.2–6.0 g/dl, respectively (Table 3.5).

KIDNEY FUNCTION

BLOOD UREA NITROGEN (BUN) The reference range for BUN concentration in the gerbil is 17–27 mg/dl (Table 3.6).

CREATININE The reference range for creatinine concentration in the gerbil is 0.6–1.4 mg/dl (Table 3.6).

ELECTROLYTES

CALCIUM (Ca) The reference range for calcium concentration in the gerbil is 3.7–6.2 mg/dl (Table 3.7).

CHLORIDE (Cl^-) The reference range for chloride concentration in the gerbil is 93–118 mEq/l (Table 3.7).

PHOSPHORUS (P) The reference range for phosphorus concentration in the gerbil is 3.8–7.0 mg/dl (Table 3.7).

POTASSIUM (K^+) The reference range for potassium concentration in the gerbil is 3.8–6.3 mEq/l (Table 3.7).

SODIUM (Na^+) The reference range for sodium concentration in the gerbil is 141–172 mEq/l (Table 3.7).

Chinchilla

Sample Collection

RESTRAINT

Chinchillas are relatively easy to handle and restrain for collection of blood and urine samples, and they rarely bite unless agitated. Chinchillas should be restrained by grasping the base of the tail with one hand and placing the other hand over the shoulders and thorax. Fur slip, or loss of a patch of fur, can occur if a chinchilla is handled roughly (Hrapkiewicz and Medina, 2007).

BLOOD

Blood collection in the chinchilla is best performed using anesthesia to reduce stress, unless the animal is very accustomed to being handled. Peripheral blood vessels are very small in the chinchilla and yield volumes of blood that are sufficient for assays such as blood glucose and packed cell volumes (Riggs and Mitchell, 2009). Small blood samples can be obtained from several peripheral veins, including the lateral saphenous vein, cephalic vein, lateral abdominal vein, and ventral tail vein (Joslin, 2009). A 23G–27G needle on a 1-ml syringe can be used at these sites. Best success is achieved using a tourniquet or holding off the vein proximally, as in other animals. A toenail clip or needle stick between the toes may also be used for a small blood sample in the chinchilla (Hrapkiewicz and Medina, 2007).

The retro-orbital sinus may be used for blood collection in the anesthetized chinchilla, as in other rodents. Alternatively, the transverse sinus in the chinchilla runs superficially around the auditory bulla and can also serve as a site for blood collection. A 25G butterfly catheter attached to a 1-ml syringe can be used to penetrate this sinus from the dorsal surface of the head near the ear (McClure, 1999).

Larger blood samples, sufficient for a complete blood count or clinical biochemistry panel, may be obtained from the femoral vein, jugular vein, or cranial vena cava in the anesthetized chinchilla, using methods described for other animals (Joslin, 2009). A 25G needle on a 1-ml or 3-ml syringe can be used to collect a blood sample from these sites (Riggs and Mitchell, 2009). The jugular veins are very superficial in the chinchilla, making them visible and particularly accessible for blood sampling in this species (Pilny, 2008).

Cardiocentesis may be used as a terminal procedure under anesthesia for collection of a large volume of blood in the chinchilla, as in other animals.

URINE

Urine for routine urinalysis can be collected from a chinchilla by clean-catch, from the cage floor, using a metabolic cage, or by manual pressure on the bladder. Urine to be cultured should be collected by cystocentesis, with or without anesthesia. For cystocentesis, the animal is placed in dorsal recumbency, the caudoventral abdomen disinfected, and urine collected percutaneously with a 25G needle attached to a 3-ml syringe (Riggs and Mitchell, 2009).

Reference Values and Sources of Variation

BLOOD

Total blood volume of the chinchilla is discussed in the Hematology section of this chapter.

URINE

Reference values and characteristics of chinchilla urine are listed in Table 3.1.

Normal chinchilla urine is yellow to amber, although dietary porphyrins can produce dark orange urine

that must be distinguished from hematuria. Chinchilla urine is normally alkaline, with a pH of 8–9 (Riggs and Mitchell, 2009).

An overweight female chinchilla with diabetes mellitus has been described with hyperglycemia and severe glucosuria and ketonuria (Marlow, 1995).

CARBOHYDRATE METABOLISM

GLUCOSE The reference range for glucose concentration in the chinchilla is 60–125 mg/dl (Table 3.2).

Serum glucose is lower in blood obtained from chinchillas anesthetized with ketamine and xylazine than in blood obtained by cardiac puncture post-mortem (de Oliveira Silva et al., 2005).

Diabetes mellitus has been described in an overweight female chinchilla that was accompanied by hyperglycemia of greater than 400 mg/dl (Marlow, 1995).

LIPID METABOLISM

CHOLESTEROL The reference range for cholesterol concentration in the chinchilla is 40–300 mg/dl (Table 3.2).

TRIGLYCERIDES The reference range for triglyceride concentration in the chinchilla is 149–198 mg/dl (Table 3.2).

Serum triglycerides are lower in blood obtained from chinchillas anesthetized with ketamine and xylazine than in blood obtained by cardiac puncture post-mortem (de Oliveira Silva et al., 2005).

ENZYMES

ALKALINE PHOSPHATASE (ALP) The reference range for ALP concentration in the chinchilla is 10–70 IU/l (Table 3.3).

ALANINE AMINOTRANSFERASE (ALT) The reference range for ALT concentration in the chinchilla is 10–35 IU/l (Table 3.3).

ASPARTATE AMINOTRANSFERASE (AST) The reference range for AST concentration in the chinchilla is 15–100 IU/l (Table 3.3).

CREATINE KINASE (CK) The reference range for CK concentration in the chinchilla is 0–300 IU/l (Table 3.3).

HORMONES

THYROID HORMONES The reference range for T4 in the chinchilla is 3.4–6.4 µg/dl.

Thyroparathyroidectomy performed on chinchillas to create an animal model of hypothyroidism resulted in 9–89% reduction in thyroxine (T4) levels in 80% of animals at 6–14 days after surgery. This was followed by rapid regrowth of the thyroid gland and a return of T4 to baseline levels in most animals studied (Martin et al., 2005). The average baseline T4 level was 5.25 µg/dl, with no difference between males and females. Sham-operated controls had a significant increase in T4 levels, suggesting a non-specific response to surgery (Martin et al., 2005).

LIVER FUNCTION

TOTAL BILIRUBIN The reference range for bilirubin concentration in the chinchilla is 0.6–1.3 mg/dl (Table 3.5).

SERUM PROTEINS The reference range for total protein concentration in the chinchilla is 3.8–8.0 g/dl (Table 3.5).

The reference ranges for albumin and globulin concentrations in the chinchilla are 2.3–4.1 g/dl and 0.9–4.2 g/dl, respectively (Table 3.5).

KIDNEY FUNCTION

BLOOD UREA NITROGEN (BUN) The reference range for BUN concentration in the chinchilla is 10–40 mg/dl (Table 3.6).

Serum urea is lower in blood obtained from chinchillas anesthetized with ketamine and xylazine than in blood obtained by cardiac puncture post-mortem (de Oliveira Silva et al., 2005).

CREATININE The reference range for creatinine concentration in the chinchilla is 0.4–2.3 mg/dl (Table 3.6).

ELECTROLYTES

CALCIUM (Ca) The reference range for calcium concentration in the chinchilla is 5.6–15 mg/dl (Table 3.7).

CHLORIDE (Cl^-) The reference range for chloride concentration in the chinchilla is 105–130 mEq/l (Table 3.7).

MAGNESIUM (Mg^{2+}) The reference range for magnesium concentration in the chinchilla is 3.6–4.0 mg/dl (Table 3.7).

PHOSPHORUS (P) The reference range for phosphorus concentration in the chinchilla is 4–8 mg/dl (Table 3.7).

POTASSIUM (K^+) The reference range for potassium concentration in the chinchilla is 3–7 mEq/l (Table 3.7).

SODIUM (Na^+) The reference range for sodium concentration in the chinchilla is 130–170 mEq/l (Table 3.7).

Degu

Sample Collection

There is no published information available on methods of restraint or sample collection in the degu. In addition, blood volume and urine characteristics have not been reported in the literature.

Reference Values and Sources of Variation

CARBOHYDRATE METABOLISM

GLUCOSE The reference range for glucose concentration in the degu is 74–82 mg/dl (Table 3.2).

The degu is very intolerant of sugar in the diet and is very susceptible to developing diabetes mellitus when exposed to increased dietary glucose (Opazo et al., 2004). Cases of spontaneous diabetes mellitus in the degu may also be exacerbated by a carbohydrate-rich diet (Najecki and Tate, 1999; Nishi and Steiner, 1990). The protein structure of degu insulin and glucagon is similar to that of the guinea pig but varies from that of most other laboratory rodents, suggesting a difference in carbohydrate metabolism in histricomorph rodents (Nishi and Steiner, 1990).

A case of transitional cell carcinoma in a degu was found to be associated with hyperglycemia, increased ALT, and decreased total protein and albumin levels (Lester et al., 2005).

HORMONES

ADRENAL HORMONES The reference value for cortisol concentration in the degu is 15 µg/dl and the reference value for corticosterone in the degu is 1.5 µg/dl (Table 3.4).

The degu has been referred to as semi-precocial and has a more advanced hypothalamic–pituitary–adrenal axis at birth compared to that of more altricial rodents such as rats and mice (Gruss et al., 2006). The adrenal glands of the degu secrete both cortisol and corticosterone, with cortisol showing the greatest increase in response to stress (Kenagy et al., 1999). Plasma corticosterone levels in the degu are approximately 10–20% of cortisol levels (Gruss et al., 2006). Both male and female degus have elevated cortisol and corticosterone levels at birth, low levels at days 3–21, and higher, adult-like levels at days 45 and later (Gruss et al., 2006). There are no gender-dependent differences in these levels at any age.

The stress of parental separation causes elevated cortisol and corticosterone in male and female degus between 3 and 21 days of age. The absence of measurable ACTH activity is thought to be due to lack of degu-specific antibodies (Gruss et al., 2006).

Diurnal cycle affects cortisol levels in male degus, with a peak at the beginning of the light cycle and a trough near the end of a 12-hour light cycle (Mohawk et al., 2005).

KIDNEY FUNCTION

A case of transitional cell carcinoma in a degu was found to be associated with increased BUN, creatinine, and phosphorus levels (Lester et al., 2005).

Deer Mouse

Sample Collection

RESTRAINT

Peromyscus species readily adapt to the laboratory environment and may be handled in a similar manner to *Mus musculus.*

BLOOD

Blood can be collected from tail veins of the deer mouse under light anesthesia (Sealander, 1961). Larger blood samples may be collected directly from the heart in animals under deep terminal anesthesia (Gough and Kilgore, 1964).

There is very little information available in the literature regarding clinical biochemistry parameters in the deer mouse.

Reference Values and Sources of Variation

ENZYMES

ALKALINE PHOSPHATASE (ALP) The reference range for ALP concentration in the deer mouse is 6–7 IU/l (Table 3.3).

Deer mice infected with *Capillaria hepatica* were found to have increased concentrations of alkaline phosphatase compared with uninfected controls (Meagher, 1998).

LIVER FUNCTION

SERUM PROTEINS Deer mice infected with *Capillaria hepatica* were found to have increased total proteins and decreased albumin levels compared with uninfected controls (Meagher, 1998).

Dormouse

Sample Collection

BLOOD

Little information is available on obtaining blood samples from the dormouse. Blood samples of 1 ml were obtained by cardiac puncture in the anesthetized dormouse (Jallageas and Assenmacher, 1986).

Very little information is available in the literature regarding clinical biochemical parameters in the dormouse.

Reference Values and Sources of Variation

CARBOHYDRATE METABOLISM

GLUCOSE Glucose-induced insulin release was studied in the perfused pancreas of the edible dormouse (*Glis glis*) and demonstrated seasonal variation in this species. Exposure of the dormouse to 302 mg/dl glucose caused highest insulin release in the fall and winter and decreased glucose sensitivity in the spring and summer (Castex and Sutter, 1981).

HORMONES

THYROID HORMONES Artificially controlled photoperiod pattern and constant warm temperatures were found to interfere with the normal annual cycle of thyroxine levels in the dormouse (Jallageas and Assenmacher, 1986).

REPRODUCTIVE HORMONES The annual cycle of plasma testosterone levels in the dormouse is parallel and correlated to thyroxine concentrations (Jallageas and Assenmacher, 1983). Testosterone levels are decreased compared to reference levels in the dormouse when the thyroid gland is removed in June, at the beginning of the breeding season (Jallageas et al., 1992). Constant pattern of photoperiod and warm temperatures were found to interfere with the normal annual cycle of testosterone levels in this species (Jallageas and Assenmacher, 1986).

Kangaroo Rat

Sample Collection

RESTRAINT

The kangaroo rat has a fragile tail that should not be used for restraint. The recommended method of restraining a kangaroo rat is by grasping the loose skin at the nape of the neck (Fine et al., 1986).

BLOOD

Blood withdrawal from the kangaroo rat is best accomplished using sedation or light anesthesia. Blood sampling from the infraorbital sinus has been described in this species under light anesthesia (Banta and Holcombe, 2002). Blood samples can be obtained by tail clipping in the unanesthetized kangaroo rat (Scelza and Knoll, 1982).

URINE

Kangaroo rats rarely drink water in the wild and use internal metabolically derived water and concentration of urine to survive in an arid or semiarid environment. Kangaroo rats will drink water in captivity, but it is difficult to obtain a sufficient urine sample from this species to run a urinalysis (Donnelly and Quimby, 2002). A metabolism cage has been used successfully to evaluate urine volume and concentration in the kangaroo rat in one study (Banta and Holcombe, 2002).

Reference Values and Sources of Variation

URINE

Urine produced by kangaroo rats is very concentrated, which represents an adaptation to their desert environment in nature (Tracy and Walsberg, 2002). One species (*D. panamintinus*) reportedly excretes urine that is concentrated to 3.28M (Intress and Best, 1990).

HORMONES

THYROID HORMONES The reference range for T4 in the kangaroo rat is 0.48–1.92 µg/dl (Table 3.4). Levels of circulating T4 are lower in the kangaroo rat than predicted based on its size and may explain the low metabolic rate in this species (Hulbert et al., 1985; Yousef and Johnson, 1975).

Kangaroo rats implanted subcutaneously with slow-release thyroxine pellets producing 0 (placebo) to 111.1 µg per day showed a dose-dependent increase in plasma thyroxine and basal metabolic rate, with no increase in urinary water loss (Banta and Holcombe, 2002).

KIDNEY FUNCTION

BLOOD UREA NITROGEN (BUN) The reference value for BUN in the kangaroo rat is 30 mg/dl (Table 3.6).

The kangaroo rat produces uric acid as the final product of purine metabolism, and it is excreted in the feces. This process is metabolically complex but advantageous for a desert-dwelling rodent because it conserves water.

It is possible to produce a condition similar to diabetes insipidus in the kangaroo rat if it is allowed to drink water ad libitum (Eisenberg and Isaac, 1963).

Cotton Rat

Sample Collection

RESTRAINT

Cotton rats are challenging to restrain for sample collection because they move quickly, jump vertically, and may attempt to bite when picked up. The tail skin is susceptible to degloving, so the tail should not be used to pick up a cotton rat (Donnelly and Quimby, 2002). Restraint with a leather glove followed by anesthesia is recommended to attain sufficient control for sample collection (Prince, 1994).

BLOOD

Cotton rats have tail veins that are not accessible for blood sampling, although blood samples can be obtained from the cut tip of the tail without anesthesia (Dolyak and Leone, 1953). A blood sample of up to 500 µl (per 100-g animal) may be collected from the retro-orbital venous sinus of an anesthetized cotton rat (Prince, 1994; Webb et al., 2003). Blood flow may be increased by applying pressure to the ipsilateral jugular vein (Prince, 1994). Blood sampling directly from the

heart of anesthetized cotton rats has also been reported (Dolyak and Leone, 1953).

Reference Values and Sources of Variation

CARBOHYDRATE METABOLISM

CHOLESTEROL The reference range for glucose level in the non-fasted cotton rat is 115–216 mg/dl (Table 3.2), or 78–124 mg/dl after a 24-hour period of fasting (Dolyak and Leone, 1953).

LIPID METABOLISM

GLUCOSE The reference range for cholesterol in the cotton rat is 134–165 mg/dl (Table 3.2).

ENZYMES

ALKALINE PHOSPHATASE (ALP) The reference range for ALP in the cotton rat is 10–14 IU/l (Table 3.3).

LIVER FUNCTION

SERUM PROTEINS The reference range for total protein in the cotton rat is 6.1–6.9 g/dl (Table 3.5).

The reference ranges for albumin and globulins in the cotton rat are 4.2–5.2 g/dl and 1.4–2.2 g/dl, respectively (Table 3.5).

KIDNEY FUNCTION

BLOOD UREA NITROGEN (BUN) The reference range for BUN in the cotton rat is 13–14 mg/dl (Table 3.6).

CREATININE The reference range for creatinine in the cotton rat is 0.9–1.5 mg/dl (Table 3.6).

ELECTROLYTES

BICARBONATE (HCO_3^-) The reference range for bicarbonate anion concentration in the cotton rat is 12.6–15.9 mEq/l (Table 3.7).

PHOSPHORUS (P) The reference range for phosphorus concentration in the cotton rat is 6.0–7.9 mg/dl (Table 3.7).

Sand Rat

Sample Collection

BLOOD

Blood samples of 100 µl or more may be collected from the sand rat by cutting the tip of the tail (Frenkel and Kraicer, 1972).

URINE

Urine samples from sand rats may be collected using metabolic cages (Frenkel and Kraicer, 1972).

Very little information is available on the clinical biochemistry parameters of the sand rat.

Reference Values and Sources of Variation

CARBOHYDRATE METABOLISM

GLUCOSE The reference range for glucose concentration in the sand rat is 68–71 mg/dl (Table 3.2). This value does not vary significantly during a 60-hour period of fasting (Frenkel and Kraicer, 1972).

HEMATOLOGY

Introduction

Hematology is the study of blood and blood-forming organs, including the diagnosis, treatment, and prevention of diseases of the blood, bone marrow, and immunologic, hemostatic, and vascular systems. Hematologic analysis is often used for the diagnosis and treatment of animal diseases. As analytical methods have become more sensitive, requiring smaller sample volumes, hematology has become more important for disease recognition and treatment in the smaller laboratory animals.

Several physiologic and physical factors can affect the results of hematologic assays and make it difficult to establish reference values. Physiologic factors include age, breed and strain, gender, season and time of day, nutrition, disease, stress, and trauma. Important physical factors include site of blood withdrawal, presence and type of anticoagulant, and sample handling and preparation (Dyer and Cervasio, 2008). Interpretation of results must take these factors into consideration.

A joint committee formed by several scientific societies formulated a recommended core panel of hematology tests for safety and toxicity studies in laboratory animals (Weingand et al., 1996). This core panel includes total leukocyte count, differential leukocyte count, erythrocyte count, erythrocyte morphology, platelet count, hemoglobin concentration, hematocrit or packed cell volume, mean corpuscular volume, mean corpuscular hemoglobin, and mean corpuscular hemoglobin concentration. Blood smears can be used for reticulocyte counts, and bone marrow cytology should be collected and evaluated if indicated. Tests of hemostasis recommended by the committee include prothrombin time, activated partial thromboplastin time, and platelet count (Weingand et al., 1996).

Blood Collection and Sample Preparation

Blood Collection

RESTRAINT

General approach and methods of restraint for blood collection in laboratory animals are discussed above in the Clinical Chemistry section of this chapter.

SAMPLE VOLUME

The size of the animal determines the total volume of blood that can be safely collected in a single sample. The standard safe volume of a single sample is 1% of the animal's body weight every 14 days, although a smaller volume is recommended in older animals or those with suspected anemia or hypoproteinemia. Blood volume is usually restored within 24 hours, but the return of some blood components to normal levels can take up to 2 weeks after blood collection (Joslin, 2009).

COLLECTION SITES

Many of the same peripheral vessels used for blood collection in larger animals can be used in laboratory animals, although the unique anatomy of certain species determines the success of one site over another. For example, the tail vein is an appropriate site for blood sampling in the mouse and gerbil but is not useful in the hamster. Blood collection sites for the respective species are discussed in the Clinical Biochemistry section of this chapter.

Regardless of the site used for blood collection, the overlying skin should be clipped or shaved and cleaned with an aseptic solution such as chlorhexidine gluconate or povidone iodine. A topical anesthetic, such as lidocaine and prilocaine, may also be applied to the skin prior to blood collection (Joslin, 2009).

Sample Preparation

Hematology parameters are best evaluated on a blood sample that is freshly collected and not hemolyzed or clotted. The blood sample may be collected into a tube with or without an anticoagulant.

Routine hematology assays are usually performed using an anticoagulant such as ethylenediaminetetraacetic acid (EDTA) or lithium heparin. EDTA is preferable to heparin as an anticoagulant because of its minimal alteration of cell morphology and staining characteristics. Coagulation studies are typically performed using sodium citrate.

If no anticoagulant is used, the blood should be well mixed by manually inverting or electronically rotating the tube, to ensure thorough mixing of blood components before removing an aliquot for evaluation. Blood clotting occurs more slowly in plastic tubes than in glass tubes.

Erythrocyte and leukocyte morphology and staining characteristics may be altered by anticoagulants. For this reason, blood smears should be made prior to mixing blood samples with anticoagulant (Dyer and Cervasio, 2008). A small volume of blood can be collected in the hub of a needle and used for blood smear preparation without exposure to anticoagulants.

Blood Volume

Total Blood Volume

Published values for total blood volume in a species can vary up to 10%, depending on the method used for calculation. Blood volume is also affected by physiologic factors such as age and lean body mass. In general, as an animal increases in size, the percentage of blood volume decreases (Joslin, 2009).

Blood volume is measured by the T-1824 or Evans blue dye method due to the high affinity of this azo compound dye to serum albumin.

Plasma Volume

In most animals, plasma volume is approximately 50–60% of the total blood volume. The plasma volume can be estimated from the size of the clear layer above the packed red cells and buffy coat in a microhematocrit tube after centrifugation.

Plasma volume can be calculated using radiolabeled albumin (McGuill and Rowan, 1989).

Reference ranges for blood and plasma volumes in the laboratory animals covered in this volume are provided in Table 3.8.

Peripheral Blood Cells

Erythrocytes

Red blood cells are anucleate and have a biconcave disc shape, which minimizes the distance that hemoglobin must travel to the surface of the red blood cell for gas exchange. The biconcave disc shape also increases the flexibility of the cell for movement through vessels for oxygen delivery.

The number of erythrocytes may be represented by red blood cell count, red blood cell concentration, hematocrit, or packed cell volume.

RED BLOOD CELL COUNT

An automated hematology instrument typically provides the red blood cell count in an isotonic dilution of the blood sample and calculates the red blood cell concentration from this value and the sample volume. Hematocrit and packed cell volume values both indicate the percentage of red blood cells in whole blood and are essentially equivalent but are derived by different methods.

Hematocrit (Hct) is usually calculated in an automated hematology instrument from the red blood cell concentration and mean cell volume (MCV) of red blood cells. Hematocrit is calculated using the formula Hct = (MCV × red blood cell concentration)/10 and is represented as percent.

Packed cell volume (PCV) is usually determined using the microhematocrit method (Goldenfarb et al.,

TABLE 3.8 Blood Values[1]

	Total Blood Volume		Single Sample Vol (ml/2 wks)	Exsanguination Vol (ml)	Plasma Volume (ml/kg BW)	Bone Marrow (M:E ratio)	References
	(ml)	(ml/kg BW)					
Rabbit	160–480	44–70	20–40	60–160	28–50	1.1:1	Suckow and Douglas, 1997 Hrapkiewicz and Medina, 2007
Guinea pig	40–80	67–92	4–12	15–30	39	1.5–1.9:1	Dineen and Adams, 1970 Harkness and Wagner, 1995 Longley et al., 2008
Hamster	6.8–12	65–80	0.5–1.3	3–5	N/A	0.9:1	Gad, 2007 Hrapkiewicz and Medina, 2007 Brown and Donnelly, 2004
Gerbil	4.4–8	67	0.4–1.3	2–4	N/A	0.75–2.35:1	Dineen and Adams, 1970 Brown and Donnelly, 2004 Pilney, 2008
Chinchilla	20–32	N/A	2–7	8–12	N/A	N/A	Hrapkiewicz and Medina, 2007 Riggs and Mitchell, 2009

N/A = values not available.
[1]*Species for which no values are available are not listed.*

1971). A microhematocrit tube is filled from the hub of the sampling needle or from the tube of anticoagulant-treated blood. The microhematocrit tube is filled at one end with clay sealant and centrifuged at high speed for 2–3 minutes in a microhematocrit centrifuge. The top clear layer is plasma, the middle white band is the buffy coat representing predominantly leukocytes and platelets, and the bottom red layer is packed erythrocytes. Normal plasma is clear, whereas icteric plasma is yellow, lipemic plasma is white, and hemolytic plasma is red. The PCV value is measured on a microhematocrit card reader by placing the tube on the card so that the sealant boundary with erythrocytes is on the zero line and the top of the plasma column is on the 100 line. The top of the red cell column then represents the percentage of the sample that is composed of red blood cells.

Errors of up to 8% can occur due to improper force or duration of centrifugation (Mitruka and Rawnsley, 1977).

PCV is fairly constant among mammals, whereas total red blood cell count and mean cell size vary among species (Campbell and Ellis, 2007).

HEMOGLOBIN (Hgb)

Hemoglobin is the protein that transports oxygen and carbon dioxide in the blood stream. There are several methods used to determine hemoglobin levels (Mitruka and Rawnsley, 1977).

Hemoglobin level is fairly constant among mammals and is usually approximately one-third of the hematocrit value, or one-third of the packed cell volume (Campbell and Ellis, 2007).

The cyanmethemoglobin method is usually performed by an automated hematology instrument, which uses a lytic dilution of the blood sample to release hemoglobin. Potassium cyanide converts hemaglobin to the stable pigment cyanmethemoglobin. Light absorbed at a specific wavelength is measured by spectrophotometry in a hemoglobinometer. Light is absorbed in proportion to the hemoglobin concentration, using a known concentration as a reference. Hemoglobin concentration is expressed as g/dl.

ERYTHROCYTE LIFESPAN

The lifespan of circulating red blood cells varies somewhat between species but typically ranges from 50–70 days. A variety of methods have been used to estimate this duration, including radiolabeled compounds and alveolar carbon monoxide.

ERYTHROCYTE SIZE AND STAINING

Mean cell volume (MCV) is the average size of red blood cells counted electronically. It is expressed in femtoliters or μ^3.

Mean cell hemoglobin (MCH) is calculated using the hemoglobin concentration and red blood cell

concentration and therefore provides no additional clinical information. MCH is expressed in pg per cell.

Mean cell hemoglobin concentration (MCHC) is calculated from the hemoglobin concentration and hematocrit and provides the index of Hgb volume in grams relative to PCV in g per dl or percent.

MCHC (g/dl) = [Hgb (g/dl)/PCV (%)] × 100. Since Hgb is typically one-third the hematocrit value, the MCHC value is usually approximately 30 g/dl.

In an automated hematology instrument, hemoglobin and packed cell volume are determined using different aliquots of a blood sample that are diluted separately.

A hemolyzed sample causes a falsely increased MCHC value.

Regenerative anemia causes decreased MCHC if the reticulocyte fraction is >20%. Severe iron-deficiency anemia or microcytic anemia also decreases MCHC due to decreased Hgb content.

MCHC <28 g/dl is usually due to instrument error.

RETICULOCYTES

Reticulocytes provide an assessment of the bone marrow's response to anemia. The rate of reticulocyte release from the bone marrow indicates the erythroid component of the bone marrow. A reticulocyte stain measures aggregates of residual ribosomes and mitochondria that form clumped granular material called reticulum.

Reticulocytes appear as polychromatophilic cells seen on a Wright- or Wright-Giemsa-stained blood film.

New methylene blue or brilliant cresyl blue are mixed with several drops of blood and incubated for 10 minutes in a tube before making a blood film. Reticulocytes are counted out of 1000 RBCs in a blood film and expressed as percentage of the RBCs. In cases of anemia, the number will be altered, so reticulocyte number per µl is calculated as RBCs/µl × % reticulocytes.

Reticulocytes are increased as the bone marrow responds to anemia. A reticulocyte number of less than 10 000 per µl is considered to represent no or minimal regenerative response; 10 000–60 000 per µl is a poor regenerative response; 60 000–200 000 per µl is considered a moderate response; and 200 000–500 000 reticulocytes per µl is a maximal regenerative response.

Reference ranges for erythrocyte parameters in the laboratory animals covered in this volume are provided in Table 3.9.

Leukocytes

TOTAL WHITE BLOOD CELL (WBC) COUNT

Total leukocyte concentration is measured using the Unopette dilution method, automated nucleated particle counter, or by expanded buffy coat analysis. The Unopette system is a commercial kit that dilutes a blood sample 1:100 in acetic acid, resulting in membrane lysis and removal of erythrocytes and platelets. The remaining nucleated particles are leukocytes and these are quantified within a grid under a microscope. Electronic cell counters use a similar approach, except that red blood cells are lysed with a detergent solution before nucleated particles are quantified. A quantitative buffy coat analysis system (QBC, Becton Dickinson) expands the buffy coat layer in a specialized microhematocrit tube and quantifies the leukocytes by optical scanning. An automated hematology instrument uses lytic dilution of the blood sample to provide white blood cell numbers.

BLOOD SMEAR

The optimal blood smear is a single layer of dispersed cells that accurately represents the distribution of cells in whole blood. The most common method of blood smear preparation is the wedge technique, in which a drop of blood is placed near the end of one glass slide. A second slide is held at a 30–45° angle, backed into the drop, and quickly pushed forward to the end of the first slide. The best area for cell counting is the monolayer of cells (counting area) between the thick region and the feathered edge (Thrall et al., 2004).

BLOOD STAINING

Blood staining is needed to perform a differential leukocyte count and to identify morphologic abnormalities. Romanowsky stains, such as Wright's or Wright-Giemsa stains, are employed in diagnostic laboratories to stain blood smears. Rapid hematology staining kits that mimic the Wright stain are commonly used in clinical settings.

DIFFERENTIAL LEUKOCYTE COUNT

The concentration of individual white blood cell types is the most important indicator of disease, and this is determined in a well-prepared and stained blood smear. The stained counting area is examined under high power to perform a differential count of leukocytes, evaluate blood cell morphology, and determine platelet number. The differential count of leukocytes is performed by classifying 100 consecutively encountered white blood cells as neutrophil, band neutrophil, lymphocyte, monocyte, eosinophil, and basophil. Morphology of white and red blood cells should be noted, and number of platelets per high-powered field recorded.

Flow cytometry can be used to count cell populations based on phenotypic markers and measuring cell functions (Kania, 2008). Antibodies are used that bind specifically to cell surface markers called cluster of differentiation (CD) antigens. The markers may correlate with specific cell functions.

Cell counting is performed with instruments that use light-scattering technology. The degree and angle

TABLE 3.9 Erythrocyte Values[1]

	RBC Count ($\times 10^6$/µl)	Hct/PCV (%)	Hgb conc (g/dl)	RBC Lifespan (days)	RBC diam (microns)	MCV (μ^3)	MCH (pg/cell)	MCHC (g/dl)	Reticulocytes (%)	References
Rabbit	4.9–7.8	31–50	10–17.4	57	5.0–7.8	57.5–75	17.1–23.9	28.2–37	1.7–6.3	Thrall et al., 2004 Brewer, 2006 Melillo, 2007 Vennen and Mitchell, 2009
Guinea pig	4–11	30–50	11–17	N/A	6.6–7.9	70–95	23–27	25–40	0–6.1	Thrall et al., 1974 Riggs, 2009
Hamster	2.7–12.3	30–59	10–19.2	50–78	5–7	64–78	20–26	28–37	N/A	Thrall et al., 2004 Heatley and Harris, 2009
Gerbil	7–10	35–52	10–17.9	10	N/A	46.6–60	16.1–19.4	30.6–33.3	N/A	Donnelly and Quimby, 2002 Heatley and Harris, 2009
Chinchilla	5–10	30–55	9–15	N/A	N/A	32.1–69.2	10.4–19.8	20–38.5	N/A	Donnelly and Quimby, 2002 Riggs and Mitchell, 2009
Deer mouse	10–12	38–52	12–16	N/A	N/A	36–46	11–15	30–35	N/A	Sealander, 1960, 1961 Gough and Kilgore, 1964 Meagher, 1998
Kangaroo rat	7–9	47–53	15–17	N/A	N/A	57.3–66.0	18.6–25	30–35	N/A	Scelza and Knoll, 1982 Intress and Best, 1990
Cotton rat	N/A	N/A	13–14	N/A	N/A	N/A	N/A	N/A	N/A	Dolyak and Leone, 1953

N/A = values not available.
[1]*Species for which no values are available are not listed.*

of light scatter is used to differentiate different types of cells. Automated differential WBC counting is fairly accurate for normal blood but can be inaccurate if the blood sample is abnormal and can be verified with a manual differential count.

Coulter technology is more commonly used for cell counting. This method is used to measure the size of RBCs, distinguish RBCs from platelets, and differentiate types of WBCs. Suspended cells impede the medium's ability to conduct current. Current deflection is measured as cells and current pass through an opening, and the amount of the deflection is proportional to the size of the cell.

LYMPHOCYTES Lymphocytes are found in blood, bone marrow, lymph nodes, gut-associated lymphoid tissue (GALT), and are associated with immunologic response.

NEUTROPHILS Mammalian neutrophils are phagocytic and function in the destruction of microorganisms. Inflammation, especially that caused by microorganisms, results in an increase in circulating neutrophils.

EOSINOPHILS Eosinophils are involved in the inactivation of histamine in response to allergens and some chronic diseases of the skin, lungs, gastrointestinal tract, and uterus, organs with high concentrations of mast cells.

Eosinophilic granules contain alkaline proteins that stain intensely pink with eosin dye. The granules have a crystalline shape that varies between species.

Eosinophils have phagocytic activity, but this function is much less effective than neutrophils.

Eosinophilia is typically associated with infections with helminth larvae, allergic inflammation, and antigen–antibody complexes (Thrall et al., 2004). Eosinophils primarily act to destroy parasitic microorganisms with the alkaline proteins contained in their granules.

BASOPHILS Basophils are typically the least common of the granulocytes and contain large cytoplasmic granules that stain with basophilic dyes. Basophils function in inflammatory reactions associated with allergies. Basophils contain heparin and histamine and are often found at sites of ectoparasite infection.

MONOCYTES Monocytes have a large nucleus with two lobes. Monocytes function in the immune system to restore the macrophages and dendritic cells in the tissues and respond to inflammatory mediators by differentiating into macrophages and denritic cells as part of an immune response.

Platelets (Thrombocytes)

Platelets are cytoplasmic fragments from megakaryocytes in the bone marrow and contain cytoplasmic organelles. Platelets form the initial hemostatic plug during clot formation and prevent hemorrhage after damage to small blood vessels. Platelets often appear as clumps in blood smears. Large platelets represent those released early from the bone marrow. Platelets are much smaller than erythrocytes but are typically counted simultaneously by hematology analyzers, according to particle size. An experienced technician can count platelets using a hemocytometer and Unopette kit.

Reference ranges for leukocyte and platelet numbers in the laboratory animals covered in this volume are provided in Table 3.10.

Coagulation

Coagulation of blood is a defense mechanism to prevent excessive blood loss after vascular injury. It involves the formation of a clot at the site of damaged endothelium in response to thrombin, which converts fibrinogen into the fibrin matrix of the clot. After the endothelium has been repaired, the clot dissolves by fibrinolysis.

Prothrombin is the circulating precursor of thrombin, and there are several pro-coagulant proteins that interact to enhance thrombin formation. Endothelial damage initiates a cascade of events. A variety of tests are used to determine the functioning of the coagulation system.

Bleeding Time (BT)

Bleeding time is the time it takes, in minutes, for bleeding to stop after a standardized cut is made in a hairless area of skin. Bleeding stops when a platelet plug forms and BT thus assesses platelet function.

Prothrombin Time (PT)

Prothrombin time is a measure of the function of the extrinsic coagulation pathway. Blood is collected in a tube containing the anticoagulant sodium citrate and centrifuged to yield plasma. Tissue factor (III) is added and the time to clot formation is measured in seconds.

Activated Partial Thromboplastin Time (APTT)

Activated partial thromboplastin time is a measure of the function of the intrinsic and common coagulation pathways. A blood sample is collected into a tube containing citrate, which stops coagulation by binding calcium, and then centrifuged. A phospholipid, activator, and calcium are added to the plasma sample and time to clot formation is measured in seconds.

TABLE 3.10 Leukocytes[1]

	Total WBC ($\times 10^3$ cells/μl)	Lymphocytes (%)	Neutrophils (%)	Eosinophils (%)	Basophils (%)	Monocytes (%)	Platelets ($\times 10^6$/μl)	References
Rabbit	5.2–12.5	30–85	20–75	0–5	0–10	0–10	200–1000	Hrapkiewicz and Medina, 2007 Vennen and Mitchell, 2009
Guinea pig	6–17	30–80	20–60	0–7	0–3	1–12	250–850	Hrapkiewicz and Medina, 2007 Riggs, 2009
Hamster	3–15	50–96	17–35	0–5	0–5	0–5	200–590	Heatley and Harris, 2009
Gerbil	4.3–21.6	32–97	2–41	0–4	0–2	0–9	400–830	Hrapkiewicz and Medina, 2007 Heatley and Harris, 2009
Chinchilla	4–25	19–98	9–78	0–9	0–11	0–6	300–600	Donnelly and Quimby, 2002 Riggs and Mitchell, 2009
Deer mouse	1–4	N/A	N/A	N/A	N/A	N/A	N/A	Gough and Kilgore, 1964
Kangaroo rat	3–9	N/A	N/A	N/A	N/A	N/A	N/A	Scelza and Knoll, 1982 Intress and Best, 1990
Cotton rat	11–13	50–54	36–40	5–6	0–0.2	4–5	N/A	Webb et al., 2003

N/A = values not available.
[1]*Species for which no values are available are not listed.*

TABLE 3.11 Coagulation Tests[1]

	Bleeding Time (min)	Prothrombin Time (s)	APTT (s)	Thrombin Time (s)	Fibrinogen (g/dl)	References
Rabbit	1.1–2.7	7.2–7.8	35	7.8–12.0	0.17–0.31	Lee and Clement, 1990 Lewis, 1996
Hamster	1.5–2.4	10.3–10.7	N/A	N/A	188–316	Gad, 2007 Dodds et al., 1977 Tomson and Wardrop, 1987

N/A = values not available.
[1]*Species for which no values are available are not listed.*

Thrombin Time (TT)

Thrombin time is also known as thrombin clotting time (TCT) and is a test that measures the time for a clot to form in plasma in a blood sample placed in anticoagulant to which an excess of thrombin is added. This test evaluates the conversion of soluble fibrinogen to insoluble fibrin. Failure to form a clot within 10–15 seconds is indicative of a possible fibrinogen deficiency.

Fibrinogen Concentration

Fibrinogen, or Factor I, originates in the liver and is converted to fibrin by the enzyme thrombin during coagulation. The fibrinogen test evaluates fibrinogen levels when there are suspected abnormalities in this protein that contribute to clotting disorders.

Reference ranges for coagulation tests in the laboratory animals covered in this volume are provided in Table 3.11.

Bone Marrow

Pluripotent stem cells in the bone marrow produce progenitor cells that differentiate into the erythrocyte, granulocyte, megakaryocyte, and monocyte/lymphocyte cell lines. These cells mature and are released into the circulation (Marshall, 2008).

Evaluation of the bone marrow is indicated for hematologic conditions that cannot be completely described by examination of peripheral blood elements. Conditions that may require bone marrow evaluation include non-regenerative anemias, thrombocytopenia, gammopathy, leukemias, and staging of lymphomas and mast cell tumors (Pilny, 2008).

General anesthesia is required for bone marrow aspiration in laboratory animals, and topical anesthetic should also be infiltrated at the sampling site. The proximal femur is the most common sampling site, but the proximal tibia, proximal humerus, and the ilium can also be used in larger animals (Pilny, 2008). Surgical exposure and resection of a bone to recover bone marrow may also be used, but this procedure is more invasive and has a greater risk of complications.

For bone marrow aspiration, the animal is placed in lateral recumbency, and the site to be sampled is shaved and aseptically prepared. A small incision is made in the overlying skin, and then a spinal needle is advanced into the medullary cavity and the stylet removed. A syringe is attached to the needle, and bone marrow is withdrawn. A smear can be prepared from a drop of the sample, and the remainder is usually stored in EDTA to prevent clotting (Pilny, 2008).

A bone marrow smear is prepared by placing a drop of marrow sample on one slide and covering it briefly with a second slide to spread the drop. The second slide is removed and the smear is air dried and stained with Wright-Giemsa or a rapid hematology stain. The smear is examined for normal cell morphology and myeloid vs. erythroid cell types. The myeloid to erythroid (M:E) ratio is calculated as an indication of bone marrow activity, and this ratio varies between 0.5:1 and 3:1 depending on the species. The maturation index, or ratio of proliferating to non-proliferating cells, is also calculated as an assessment of hematopoiesis (Pilny, 2008).

Bone marrow hypoplasia can result from infectious disease, estrogen or chemical toxicity, myelofibrosis, or immunosuppressive therapy or disease. Bone marrow aplasia indicates lack of cells from all lines (Pilny, 2008).

Bone marrow hyperplasia may represent a regenerative response to loss of myeloid or erythroid cells in the peripheral circulation, or it may be due to a lymphoproliferative or myeloproliferative neoplastic disease (Pilny, 2008).

Reference ranges for the myeloid to erythroid (M:E) ratio in the laboratory animals covered in this volume are provided in Table 3.8.

RABBIT

Sample Collection

Blood

Methods of restraint and acceptable sites and volumes of blood samples in the rabbit are described in the Clinical Biochemistry section of this chapter.

Bone Marrow

Bone marrow aspirates should be performed if there is a suspicion of anemia or other abnormalities of blood cell production. Collection of bone marrow should be performed with the rabbit under anesthesia and the sampling site prepared aseptically. Percutaneous aspiration may be used to sample bone marrow from the humerus or the femur with a Rosenthal pediatric needle (Horan et al., 1980) or from the proximal end of the tibia with a 15G needle (Sundberg and Hodgson, 1949). The proximal femur is generally the preferred site in most species, including the rabbit. A spinal needle with stylet is typically used and inserted in the trochanteric ridge of the femur (Vennen and Mitchell, 2009). The femur may also be surgically resected for direct access to the bone marrow (Powsner and Fly, 1962), but this approach is not recommended due to the importance of the rear limb for weight-bearing in the rabbit (Suckow and Douglas, 1997).

Reference Values and Unique Characteristics

Several factors can affect the results of hematology assays in the rabbit, including physiologic factors, such as age, breed or strain, sex, or pregnancy (Murray, 2000), as well as physical factors, such as sample handling and analytical techniques.

Blood Volume

Reference ranges for blood and plasma volumes in the rabbit are provided in Table 3.8.

TOTAL BLOOD VOLUME

The total blood volume of a normal adult New Zealand White rabbit is approximately 160–480 ml, or 44–70 ml per kg body weight. Rabbits have a higher blood volume during pregnancy (Prince, 1982). Blood volume per body weight in the young rabbit decreases significantly between birth and 4 months old.

Using the general guidelines for blood withdrawal based on body weight, approximately 20–40 ml may be safely removed every 2 weeks.

The use of radio-iodinated albumin is preferable for estimating blood volume in a rabbit, instead of the dye T-1824 method often used in dogs (Brewer, 2006).

PLASMA VOLUME

Plasma volume in the rabbit is approximately 28–50 ml per kg body weight and decreases significantly between birth and 4 months old.

Peripheral Blood Cells

Peripheral blood cells and elements of hematopoiesis in the rabbit are nicely illustrated in color photomicrographs in a recent publication (Marshall, 2008).

ERYTHROCYTES

There are no known blood groups in rabbits, and blood transfusions are well tolerated in this species (Marshall, 2008).

Reference values for erythrocyte parameters in the rabbit are provided in Table 3.9.

RED BLOOD CELL COUNT The reference range for red blood cell count in rabbits is 4.9–7.8 $\times$ 10^6 cells per µl (Table 3.9) and may be affected by several factors. For example, lower erythrocyte counts are reported in young rabbits of less than 12 weeks old (Melillo, 2007). Interestingly, the red blood cell count in a 28-day fetus is similar to that of the doe (Kriesten et al., 1987). The sex of the rabbit has been reported to have little or no effect on hematologic values (Jain, 1986).

An increase in red blood cell number has been reported in response to cold stress (de la Fuente et al., 2007), whereas hemolysis of the blood sample can result in a decreased red blood cell count (Melillo, 2007).

HEMATOCRIT (Hct) OR PACKED CELL VOLUME (PCV) The reference range for hematocrit in rabbits is 31–50% (Table 3.9) and may vary by age and strain. Hematocrit is reported to be high at birth and then falls by 20 days old, followed by a gradual increase to adult levels by 90 days old (Abelson and Simon, 1991). However, other studies report a PCV in neonatal rabbits that is similar to adult rabbits (Bortolotti et al., 1989). Differences exist in hematocrit values between different breeds and strains of rabbits but probably have little clinical significance (Kozma et al., 1974). There also appears to be little or no effect of sex on hematocrit values in the rabbit (Jain, 1986).

A PCV of less than 30% is indicative of anemia. A non-regenerative anemia may suggest the presence of a chronic disease. A regenerative anemia is associated with reticulocyte production and may occur in cases of blood loss due to external or internal hemorrhage, intravascular hemolysis, autoimmune hemolytic anemia, or lead toxicity (Melillo, 2007).

Hematocrit values of 13–31% were described in New Zealand White rabbits with hemolytic anemia resulting from systemic infection with *Achromobacter xylosoxidans* in contaminated intravenous fluids (Allison et al., 2007).

A PCV of more than 45% may indicate dehydration. Elevated PCV has also been reported as a consequence of cold stress and heat stress (de la Fuente et al., 2007).

HEMOGLOBIN (Hgb) The reference range for hemoglobin levels in rabbits is 10–17.4 g/dl (Table 3.9). Similar to hematocrit levels, hemoglobin concentrations are high at birth and then fall by 20 days old, followed by a gradual increase to adult levels by 90 days old (Abelson, 1991). As with hematocrit,

hemoglobin differences between breeds and strains of rabbits exist but probably have little clinical significance (Kozma et al., 1974). Gender appears to have little or no effect on hemoglobin values in the rabbit (Jain, 1986).

Hemoglobin levels demonstrate diurnal variation in the rabbit, with hemoglobin levels highest in the morning and lowest in the late afternoon and early evening (Fox and Laird, 1970).

ERYTHROCYTE LIFESPAN Rabbit red blood cells have a short lifespan of 57 days (Table 3.9) and a high turnover rate. This results in polychromasia, which is not clinically significant in the rabbit. A few nucleated red blood cells (1–2%) and occasional Howell-Jolly bodies, which are basophilic nuclear remnants, are normal in the rabbit due to the release of young red blood cells into the circulation (Melillo, 2007).

ERYTHROCYTE SIZE AND STAINING Rabbit erythrocytes have an average diameter of 6.8 microns, although there is significant anisocytosis, with a diameter range of 5.0–7.8 microns (Table 3.9) (Melillo, 2007). Erythrocyte diameter decreases with age from a diameter of approximately 9 microns at birth to adult size by 20–120 days old (Kozma et al., 1974).

It is reported that red blood cells are not as deformable in the rabbit as in humans (Amin and Sirs, 1985). This is thought to be due to higher concentrations of oxygen-free radicals and plasma fibrinogen in the rabbit (Hirayama et al., 1986).

Mean Cell Volume (MCV) The reference range for mean cell volume (MCV) in rabbits is 57.5–75 μ^3 (Table 3.9).

This value is reported to be higher in pregnant rabbits (Prince, 1982).

The MCV and erythrocyte diameter both decrease between neonatal and adult ages (Kozma et al., 1974).

Mean Cell Hemoglobin (MCH) The reference range for mean cell hemoglobin (MCH) in rabbits is 17.1–23.9 pg per cell (Table 3.9), and this value is reportedly higher in neonatal rabbits than in adult rabbits (Bortolotti et al., 1989).

Mean Cell Hemoglobin Concentration (MCHC) The reference range for mean cell hemoglobin concentration (MCHC) in rabbits is 28.2–37 g/dl (Table 3.9), and this value is reportedly similar in neonatal and adult rabbits (Bortolotti et al 1989).

RETICULOCYTES The reference range for reticulocyte numbers in the rabbit is 1.7–6.3% (Table 3.9). Reticulocytes are present at 2–4% in young rabbits (Jain, 1986) and after repeated bleedings or hemolytic anemia in adult rabbits (Brunner et al., 1983).

Reticulocytes are distinguishable from mature red blood cells by their larger size and more intense basophilic staining (Marshall, 2008). Reticulocyte count is elevated in neonates and declines with age to stable adult levels at 1 year old (Jain, 1986).

Nucleated red blood cells are found during endothelial changes and regenerative responses (Vennen and Mitchell, 2009).

LEUKOCYTES

Reference ranges for leukocyte parameters in the rabbit are provided in Table 3.10.

TOTAL WHITE BLOOD CELL (WBC) COUNT The reference range for total white blood cell count in rabbits is 5.2–12.5 $\times$ 10^3 cells per µl (Table 3.10) and varies with factors such as age, diurnal rhythm, and stress. Leukocyte counts are low in rabbits less than 12 weeks old and then gradually increase to maximum values by 3–6 months old (Jain, 1986; Melillo, 2007). Interestingly, the total white blood cell count in the 28-day rabbit fetus is also much lower than that of adult rabbits (Kriesten et al., 1987). A bimodal increase in leukocytes has been reported in rabbits, with the first peak at 3 months old, representing primarily lymphocytes, and the final peak at over 1 year old, due to an increase in neutrophils (Jain, 1986).

Total leukocyte numbers demonstrate diurnal variation, with lowest counts in the late afternoon and evening (Melillo, 2007).

Stress has been found to produce variable effects on total white blood cell count in rabbits. Some reports state that stress causes leukopenia in the rabbit (Brewer, 2006; Melillo, 2007). Application of electrical current in rabbits was found to produce a 15–30% decrease in the white blood cell count (Kaplan and Timmons, 1979). A recent study found lower white blood cell counts and higher plasma cortisol levels in group-housed compared with individually housed New Zealand White rabbits used for antibody production (Fuentes and Newgren, 2008).

Most studies, however, report that stress induces leukocytosis (Melillo 2007), and changes in leukocyte numbers have been found to correlate with changes in cortisol levels (Toth and Krueger, 1989). The stress associated with routine sample collection causes a physiological leukocytosis in rabbits (Harkness, 1987). It is reported that increased leukocyte numbers following stress consist primarily of mature neutrophils and lymphocytes and decreased eosinophils (Jain, 1986).

Increased white blood cell count is not a common response to infection in the rabbit. Instead, acute infection in rabbits has been shown to result in a

leukopenia of less than 5000 cells per ml and a normal differential count, or a normal white blood cell count with increased heterophils and decreased thrombocytes (Vennen and Mitchell, 2009).

LYMPHOCYTES The reference range for lymphocyte numbers in the rabbit is 30–85% (Table 3.10) and varies with several factors, including diurnal rhythm, stress, and disease. The lymphocyte nucleus is round or oval, acentrically located, and stains deep blue to purple. The cytoplasm of the lymphocyte stains a medium to deep blue, and there is typically a perinuclear halo of nonstaining cytoplasm (Jain, 1986).

Lymphocytes are the predominant leukocyte in healthy rabbits, and both small and large lymphocytes are found in circulation. Small lymphocytes are 7–10 microns in diameter, and large lymphocytes are 10–15 microns in diameter. In addition to their presence in the bloodstream, lymphocytes are also found in the spleen, bone marrow, lymph nodes, and the lymphatic tissue of the gastrointestinal tract.

Lymphocyte numbers are typically highest in the early morning and lowest in late afternoon and evening (Fox and Laird, 1970; Melillo, 2007).

Acute stress and the associated increase in adrenaline may cause lymphocytosis, whereas prolonged stress and increased cortisol may induce lymphopenia (Melillo, 2007). However, other reports suggest that the epinephrine release associated with acute stress causes a mature heterophilia and relative lymphopenia (Marshall, 2008).

The stress induced by loud noises can cause an increase in the neutrophil to lymphocyte (NP:LC) ratio in rabbits that persists for 24–48 hours (Melillo, 2007).

Infection in the rabbit may also be accompanied by an increased NP:LC ratio, which is typically 1:1 in a normal adult rabbit (Melillo, 2007).

Increased lymphocyte numbers may indicate an immunologic response, such as occurs after a viral infection. Lymphocytosis may also be caused by lymphoma and lead toxicity (Melillo, 2007). A reversal in the NP:LC ratio due to elevated lymphocyte number was found to occur after experimental inoculation with several bacteria, as well as in spontaneously ill animals.

NEUTROPHILS (HETEROPHILS) The reference range for neutrophil numbers in the rabbit is 20–75% (Table 3.10) and is affected by several factors, including diurnal rhythm, stress, and disease.

Rabbit neutrophils have a diameter of 9–15 microns and have acidophilic cytoplasmic granules that stain with eosin and give them the appearance of eosinophils. For this reason, rabbit neutrophils are often referred to as pseudoeosinophils or, more commonly, heterophils. The neutrophil nucleus has a polymorphous shape and stains light purple to light blue. The neutrophil cytoplasm stains pink because of numerous small granules, with a reddish hue due to several larger dark-staining granules (Campbell and Ellis, 2007). By electron microscopy, these granules are identified as primary or azurophilic granules and secondary or specific granules, respectively (Jain, 1986). Primary and secondary granules are believed to arise from two different sites on the Golgi apparatus (Marshall, 2008). The small pink granules comprise 80–90% of the neutrophil granules and contain peroxidase, alkaline phosphatase, lipase, and antibacterial activity (Benson and Paul-Murphy, 1999).

Neutrophils of female rabbits and other species may contain a drumstick configuration, which is referred to as the Pelger-Huet anomaly and is likely due to abnormal maturation of these cells (Kozma et al., 1974). The homozygotic form of this disease in rabbits is usually lethal (Jain, 1993).

Neutrophil numbers in the rabbit are lowest in the early morning and increase to peak numbers in late afternoon and evening (Fox and Laird, 1970; Melillo, 2007).

Prolonged stress induces an increase in neutrophils and a concomitant decrease in lymphocytes and eosinophils (Melillo, 2007). This may result in an overall leukopenia in the rabbit (Brewer, 2006). Stress due to loud noises can also cause an increase in the NP:LC ratio in rabbits that persists for 24–48 hours (Melillo, 2007). Acute stress is associated with catecholamine release that results in a mature neutrophilia and relative lymphopenia (Marshall, 2008).

Acute bacteria-induced inflammation causes an increase in neutrophils, which migrate to the site of infection and engulf pathogens. Lysosomal granules in the neutrophil cytoplasm fuse with phagosomes to kill and degrade infectious organisms (Campbell and Ellis, 2007).

Infection in the rabbit is usually associated with an increased NP:LC ratio, although the presence of band neutrophils and a left shift is uncommon. A reversal in the NP:LC ratio has also been reported after experimental inoculation with several bacteria, as well as in spontaneously ill animals. A neutrophil count of 60–80% can occur as a result of inflammation, although there is often a concomitant decrease in lymphocytes, with little or no change in total white blood cells. Alternatively, acute infection in the rabbit has been reported to result in increased neutrophils and decreased thrombocytes (Vennen and Mitchell, 2009).

EOSINOPHILS The reference range for eosinophil numbers in rabbits is 0–5% (Table 3.10) and is affected by several factors, including diurnal rhythm, stress, and disease. A low or absent eosinophil count is common in healthy rabbits.

The rabbit eosinophil has a diameter of 12–16 microns and is larger than the neutrophil. The eosinophil contains large acidophilic granules that fill the cytoplasm and are 3–4 times the size of neutrophil granules. These granules give the cytoplasm of the eosinophil a pink-orange appearance (Benson and Paul-Murphy, 1999). The eosinophil nucleus is bi-lobed or horseshoe-shaped (Vennen and Mitchell, 2009).

Eosinophil numbers are lowest in the early morning and increase to highest numbers in the afternoon or evening (Fox and Laird, 1970; Melillo, 2007).

Stress has been reported to induce a mature eosinophilia in rabbits. However, other reports suggest that glucocorticoid release is associated with eosinopenia.

Diseases of the skin, lungs, gastrointestinal tract, or uterus can result in eosinophilia in rabbits. For example, eosinophilia may be associated with abscesses or healing wounds in the rabbit or in cases of atopy and pyoderma (Melillo, 2007). Elevated eosinophil numbers may be caused by a parasitic disease such as *Encephalitozoon cuniculi* in the rabbit, although eosinophilia is reportedly an uncommon response to parasites or hypersensitivity in this species (Melillo, 2007).

BASOPHILS The reference range for basophil numbers in normal rabbits is 0–10% (Table 3.10), but can be up to 30% of the white blood cell differential count in healthy rabbits (Benson and Paul-Murphy, 1999; Vennen and Mitchell, 2009). The rabbit has higher basophil numbers than most species and may be the only laboratory animal with a significant number of basophils in circulation (Benson and Paul-Murphy, 1999). Circulating basophils are usually inversely proportional to the number of tissue mast cells. Rabbit basophils are 8–12 µm in diameter, which is similar in size to the neutrophil. The nucleus is light purple and is less segmented than the nuclei of eosinophils or neutrophils. The rabbit basophil has many purple-black cytoplasmic granules (Campbell and Ellis, 2007).

The function of the basophil is not well understood. Basophils demonstrate diurnal variation, with lowest numbers in the early morning and highest numbers in late afternoon (Fox and Laird, 1970). Cases of atopy and pyoderma in the rabbit are associated with elevated numbers of basophils and eosinophils (Melillo, 2007).

MONOCYTES The reference range for monocyte numbers in normal rabbits is 1–10% (Table 3.10) and varies according to diurnal rhythm and disease.

The monocyte is the largest white blood cell in the rabbit peripheral circulation. The monocyte nucleus has a horseshoe or bean shape and is lightly stained due to the diffuse nuclear chromatin. The monocyte cytoplasm is blue, with few vacuoles or granules in the healthy rabbit (Jain, 1986). In conditions of non-specific toxicity, large dark-red granules may be seen in the cytoplasm of the rabbit monocyte (Benson and Paul-Murphy, 1999).

There is diurnal variation of monocytes in the rabbit, with lowest numbers in the early morning and highest numbers in late afternoon (Fox and Laird, 1970).

Elevated monocyte numbers in the rabbit may be caused by chronic inflammation from an abscess, mastitis, or tympanic bulla empyema, although monocyte counts are not always increased in these conditions (Melillo, 2007).

PLATELETS (THROMBOCYTES)

The reference range for platelet numbers in the rabbit is 200–1000 $\times$ 10^6 per µl (Table 3.10) and varies according to age, stress, and disease. Thrombocytes are present singly and in groups and appear as oblong or oval bodies measuring 1–3 microns in diameter. Thrombocytes have a pale periphery and a dark purple center when stained with Wright's stain (Kozma et al., 1974). Rabbit platelets have very high serotonin content (Fudge, 2000).

The platelet count in a 28-day rabbit fetus is significantly lower than that of the doe (Kriesten et al., 1987). Cold stress may cause an increase in platelets in rabbits (de la Fuente et al., 2007).

Decreased thrombocyte numbers may be a consequence of acute infection in rabbits (Vennen and Mitchell, 2009).

Coagulation

Rabbit blood clots rapidly at room temperature and must be treated with anticoagulants to evaluate complete blood count and plasma parameters.

Rabbits have high levels of intrinsic clotting factors, including V, VIII, IX, X, XI, and XIII, and low levels of factor I (fibrinogen), II (prothrombin), and XII, compared to other laboratory animals (Fudge, 2000).

Reference ranges for coagulation tests in the rabbit are provided in Table 3.11.

BLEEDING TIME (BT)

Bleeding time (BT) for rabbits is 1.1–2.7 minutes (Table 3.11).

PROTHROMBIN TIME (PT)

Prothrombin time (PT) varies with method but is 7.2–7.8 seconds in the rabbit (Table 3.11). Rabbits have approximately 89% prothrombin levels compared to dog (Kozma et al., 1974). Cold stress causes an increase in clotting time and a decrease in prothrombin time in rabbits (de la Fuente et al., 2007).

ACTIVATED PARTIAL THROMBOPLASTIN TIME (APTT)

Activated partial thromboplastin time (APTT) for the rabbit is approximately 35 seconds (Table 3.11).

THROMBIN TIME (TT)

Thrombin time (TT) for rabbits is 7.8–12.0 seconds (Table 3.11).

FIBRINOGEN CONCENTRATION

Fibrinogen concentration for rabbits is 0.17–0.31 g/dl (Table 3.11).

In the rabbit, fibrinogen degradation products do not stimulate the production of plasma fibrinogen, as occurs in humans (Kessler and Bell, 1980).

Bone Marrow

The maturation sequence and cell types in the rabbit bone marrow are similar to those seen in other species. Color photomicrographs of rabbit bone marrow are available (Sanderson and Phillips, 1981). The myeloid to erythroid ratio in normal adult rabbit bone marrow is approximately 1.1:1 (Sanderson and Phillips, 1981) (Table 3.8). This ratio is approximately 0.9–1 in 1-week-old rabbit kits and increases to adult levels by 4 weeks old (Sabin et al., 1936). Extramedullary hematopoiesis in the rabbit occurs in the liver and spleen.

GUINEA PIG

Sample Collection

Blood

A discussion of blood sample methods and collection sites in the guinea pig is presented above in the Clinical Biochemistry section of this chapter.

Bone Marrow

Bone marrow in guinea pigs is usually obtained from the proximal end of the femur, as in other species (Feldman et al., 1982).

Reference Values and Unique Characteristics

Blood Volume

Reference ranges for blood and plasma volumes in the guinea pig are provided in Table 3.8.

TOTAL BLOOD VOLUME

The reference range for total blood volume of the guinea pig is 40–80 ml, or 67–92 ml per kg body weight, (Table 3.8) and varies with age. The highest blood volume is reported at birth, followed by a steady decline in blood volume until the animals weigh approximately 900 grams (Sisk, 1976). The volume of a single sample of blood is 4–12 ml, and exsanguination yields 15–30 ml (Harkness and Wagner, 1995).

PLASMA VOLUME

The reference value for plasma volume of the guinea pig is 39 ml per kg body weight (Table 3.8) and varies with age (Sisk, 1976).

Peripheral Blood Cells

Color photomicrographs of representative peripheral blood cells of the guinea pig are presented in a recent publication (Thrall et al., 2004).

ERYTHROCYTES

Reference values for erythrocyte parameters in the guinea pig are provided in Table 3.9.

RED BLOOD CELL COUNT The reference range for total red blood cell count of guinea pigs is 4–11 $\times$ 10^6 per µl (Table 3.9), which is typically lower than in other rodents (Pilny, 2008). In general, most red blood cell indices are lower in the guinea pig than in other laboratory animals (Feldman et al., 1982).

Hematocrit (Hct) or Packed Cell Volume (PCV) The reference range for hematocrit of guinea pigs is 30–50% (Table 3.9).

Hairless Dunkin-Hartley guinea pigs were found to have higher PCV, hemoglobin concentration, MCV, MCH, and MCHC compared to normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

A low hematocrit, or anemia, in the guinea pig is characterized by polychromasia and macrocytosis (Pilny, 2008). Anemia may be associated with cavian leukemia (Hrapkiewitz and Medina, 2007).

HEMOGLOBIN (Hgb) The reference range for hemoglobin level of guinea pigs is 11–17 g/dl (Table 3.9).

ERYTHROCYTE SIZE AND STAINING The reference range for diameter of guinea pig erythrocytes is 6.6–7.9 microns (Table 3.9), with normal anisocytosis and occasional microcytes of 3.5 microns in diameter.

Polychromasia varies with age, with approximately 25% in neonates, 4.5% in juveniles, and 1.5% in adults.

Mean Cell Volume (MCV) The reference range for MCV value in guinea pigs is 70–95 μ^3 (Table 3.9).

Mean Cell Hemoglobin (MCH) The reference range for MCH value in guinea pigs is 23–27 pg/cell (Table 3.9).

Mean Cell Hemoglobin Concentration (MCHC) The reference range for MCHC value in guinea pigs is 25–40 g/dl (Table 3.9).

RETICULOCYTES The reference range for reticulocyte number in guinea pigs is 0–6.1% (Table 3.9).

Reticulocytes were found to decrease in young male Weiser-Maples guinea pigs, then level off between 200 and 300 days old. There was no age-related change in female reticulocytes (Kitagaki et al., 2005).

LEUKOCYTES

Reference values for guinea pig leukocyte differential counts are provided in Table 3.10.

TOTAL WHITE BLOOD CELL (WBC) COUNT The reference range for white blood cell count in guinea pigs is 6–17 $\times$ 10^3 cells per µl (Table 3.10) and varies with age, strain, stress, and disease. Several studies have reported an age-related increase in total white blood cells (Kitagaki et al., 2005). Hairless Dunkin-Hartley guinea pigs were found to have a lower total leukocyte number compared to normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

Stress can produce leukocytosis in the guinea pig. In addition, cavian leukemia is usually accompanied by a severe leukocytosis of 25 000–500 000 cells per mm^3 and is associated with high mortality and poor response to treatment (Pilny, 2008).

LYMPHOCYTES The reference range for lymphocyte number in guinea pigs is 30–80% (Table 3.10), making the lymphocyte the predominant circulating white blood cell in normal guinea pigs. There are small and large lymphocytes in the guinea pig, and the appearance of these cells is similar to lymphocytes in other species. The small well-differentiated lymphocytes predominate and are only slightly larger than red blood cells. These lymphocytes have a round dark nucleus surrounded by a narrow band of cytoplasm. The less common larger lymphocytes are approximately twice the size of the small lymphocytes, have a less densely stained more oval-shaped nucleus, and usually have small to large azurophilic granules.

Lymphocytes were found to decrease in number with increasing age in both male and female guinea pigs of the inbred Weiser-Maples strain (Kitagaki et al., 2005).

Hairless Dunkin-Hartley guinea pigs were found to have a lower number of lymphocytes, compared to normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

NEUTROPHILS (HETEROPHILS) The reference range for neutrophil number in guinea pigs is 20–60% (Table 3.10) and varies with age and disease. The guinea pig neutrophil is the predominant circulating granulocyte in this species and has the same function as the neutrophil in other animals. As in several other laboratory animals, neutrophils are also known as heterophils or pseudoeosinophils in guinea pigs because of their staining characteristics. Guinea pig neutrophils are 10–12 microns in diameter and have more visible cytoplasm than eosinophils that contains small acidophilic granules. The neutrophil nucleus contains dense chromatin, is usually purple, and can have five or more segments (Feldman et al., 1984). In female guinea pigs, some neutrophils have a drumstick-shaped sex chromatin lobe. Guinea pig neutrophils have moderate alkaline phosphatase activity (Tamburlin and Glomski, 1988).

Neutrophils lack myeloperoxidase, which is responsible for liquid purulent exudates, so abscess content in guinea pigs is caseous rather than liquid (Vennen and Mitchell, 2009).

The total neutrophil number was found to increase with increasing age in both male and female Weiser-Maples guinea pigs (Kitagaki et al., 2005).

The inflammatory response in the guinea pig is typically characterized by an increase in number of neutrophils while total leukocytes may remain constant (Riggs, 2009). Experimental infection with the skin mite, *Trixacarus caviae*, was found to cause an increase in neutrophils and other leukocytes (Rothwell et al., 1991).

EOSINOPHILS The reference range for eosinophil number in guinea pigs is 0–7% (Table 3.10). Guinea pig eosinophils are 10–15 microns in diameter, which is slightly larger than the diameter of neutrophils. The eosinophil nucleus is indented but less segmented than that of the neutrophil. The cytoplasm of eosinophils is completely full of large brightly eosinophilic granules. Cytoplasmic granules of eosinophils are larger than those of neutrophils and are round, rod-shaped, or trapezoidal and almost completely fill the cytoplasm (Jain, 1984).

Experimental infection with the skin mite, *Trixacarus caviae*, caused increased eosinophils and other white blood cells (Rothwell et al., 1991). Similarly, infestation with the tick, *Amblyomma americanum,* caused increased eosinophils in guinea pigs (Brown and Askenase, 1982). Syphilis infections induced with *Treponema pallidum* has been associated with eosinophilia in guinea pigs (Wicher et al., 1998).

BASOPHILS Basophils are rarely found in guinea pig differential counts, where the reference range for basophil number is 0–3% (Table 3.10). The basophil is approximately the same size as the neutrophil and has a purple-stained lobulated nucleus and round purple cytoplasmic granules of varying size.

Basophils were found to decrease sharply by 1 year of age in female Weiser-Maples guinea pigs using automated hematology analysis (Kitagaki et al., 2005). Other reports show no change in basophils due to age or sex using manual counts (Feldman et al., 2000).

Experimental infection with the skin mite, *Trixacarus caviae*, caused an increased number of basophils and other leukocytes (Rothwell et al., 1991). Tick infestation with either *Amblyomma americanum* or *Rhipicephalus sanguineus* produced basophilia in guinea pigs (Brown and Askenase, 1982; Szabo et al., 2003).

MONOCYTES The reference range for monocyte number in guinea pigs is 1–12% (Table 3.10). The

monocyte is the largest white blood cell in guinea pig circulation. The monocyte nucleus is oval and has less condensed chromatin and a variable shape, surrounded by blue-gray cytoplasm that is darker than the cytoplasm of lymphocytes (Jain, 1974).

Monocyte numbers were found to increase gradually with increasing age in male Weiser-Maples guinea pigs older than 300 days of age (Kitagaki et al., 2005).

Experimental infection with the skin mite, *Trixacarus caviae*, caused increased monocytes and other white blood cells (Rothwell et al., 1991).

FOA-KURLOFF CELLS Foa-Kurloff cells are a type of specialized mononuclear cell that are unique to guinea pigs and make up 3–4% of the differential leukocyte count in this species (Moore, 2000). These cells are approximately the same size as large lymphocytes and have a large granular intracytoplasmic inclusion called a Kurloff body. The Kurloff body is composed of sulfated proteoglycan that is rich in mucopolysaccharide and appears homogeneous, finely granular, or slightly vacuolated (Jain, 2004). The Kurloff body stains with toluidine blue and is PAS-positive. The nucleus of Foa-Kurloff cells is large and round and is usually eccentrically placed.

The exact origin and function of Foa-Kurloff cells are not known, but they lack lysosomes and phagocytic activity and may serve as natural killer (NK) cells in circulation (Pouliot et al., 1996). Foa-Kurloff cells are rarely seen in young animals or in males, and their presence in females is correlated with estrogen levels (Percy and Barthold, 2007). These cells are common in pregnant guinea pigs and may be induced by estrogen to shift from the lungs and red pulp of spleen to the thymus and placenta (Moore, 2000). It is thought that Foa-Kurloff cells may help form a barrier between mother and fetus during pregnancy by protecting fetal antigens in the placenta (Pilny, 2008).

PLATELETS (THROMBOCYTES)

The reference range for platelet number in guinea pigs is 250–850 $\times$ 10^6 per µl (Table 3.10), although the values are variable in the literature (Feldman et al., 1982). Guinea pig platelets have an irregular oval shape of approximately 2–3 microns in length, a pale periphery, and a darkly staining center.

Platelet numbers in guinea pigs are affected by factors such as age, strain, and disease. Platelet numbers were found to decrease between 100 and 200 days of age, followed by an increase, in male Weiser-Maples guinea pigs (Kitagaki et al., 2005). The hairless strain of Dunkin-Hartley guinea pigs were found to have higher platelet numbers compared to normal Dunkin-Hartley guinea pigs (Waner et al., 1996).

Inflammation in the guinea pig is associated with an increase in platelets, which can exceed one million per microliter without an increase in total leukocyte numbers (Riggs, 2009).

Bone Marrow

The reference range for myeloid to erythroid ratio in normal guinea pigs is 1.5:1 to 1.9:1 (Table 3.8).

HAMSTER

Sample Collection

Blood

Blood collection in the hamster is described in Clinical Biochemistry section above.

Reference Values and Unique Characteristics

Blood Volume

Reference values for blood and plasma volumes in the hamster are provided in Table 3.8.

TOTAL BLOOD VOLUME

The reference range for blood volume of the Syrian hamster is 6.8–12 ml or 65–80 ml per kg body weight (Table 3.8).

Due to the hamster's ability to conserve water, 72 hours of water deprivation in this species causes decreased whole blood volume and plasma volumes with little or no change in hematocrit (Kutscher, 1968). Hibernation in the hamster causes an increase in blood volume and other hematologic parameters (Hrapkiewicz and Medina, 2007). However, the site of blood collection reportedly has little effect on hematologic parameters in the hamster.

The volume of a single blood sample in the hamster is 0.5–1.3 ml per 2 weeks (Table 3.8).

The total exsanguination volume in the hamster is 3–5 ml (Table 3.8).

PLASMA VOLUME

Water deprivation for 72 hours was found to decrease plasma volume in the hamster (Kutscher, 1968).

Peripheral Blood Cells

ERYTHROCYTES

Reference values for erythrocyte parameters in the hamster are provided in Table 3.9.

RED BLOOD CELL COUNT The reference range for total number of red blood cells in the hamster is 2.7–12.3 $\times$ 10^6 cells per µl (Table 3.9).

Hibernation in the hamster causes an increase in red blood cell number (Heatley and Harris, 2009; Hrapkiewicz et al., 1998). Castration decreases circulating RBCs by 25–30% in male hamsters due to decreased testosterone levels (Heatley and Harris, 2009). There have been no red blood cell antigen differences identified in hamsters.

Hematocrit (Hct) or Packed Cell Volume (PCV) The reference range for hematocrit value in the hamster is 30–59% (Table 3.9) and is affected by several factors (Burns and De Lannoy, 1966). For example, hematocrit increases with age in hamsters. The neonatal hamster is relatively anemic, with a hematocrit of 34–39%. At this age red cells are characterized by hypochromia and macrocytosis, with 10–30% normoblasts (Gad, 2007). Ten to 30% of circulating red blood cells are nucleated in neonatal hamsters, compared to less than 2% in adults (Jain, 1974).

Hibernation in the hamster causes an increase in PCV and other hematologic parameters, accompanied by decreased red blood cell destruction (Hrapkiewicz and Medina, 2007).

HEMOGLOBIN (Hgb) The reference range for hemoglobin level in the hamster is 10–19.2 g/dl (Table 3.9) and is affected by factors such as age and hibernation state. Hemoglobin increases to a maximum concentration in hamsters at 8–9 weeks of age (Gad, 2007), although some studies report little change in hemoglobin levels between 1 and 27 weeks old (Rennie et al., 1981).

Hibernation causes increased Hgb concentration (Heatley and Harris, 2009), and hypothermia causes more complete oxygen saturation of hemoglobin than normothermia (Volkert and Musacchia, 1970).

ERYTHROCYTE LIFESPAN The reference range for erythrocyte lifespan in the hamser is 50–78 days (Table 3.9).

Hibernation causes increased life span of erythrocytes in the hamster (Mitchell and Tully, 2009).

ERYTHROCYTE SIZE AND STAINING The reference range for erythrocyte diameter in the hamster is 5–7 microns (Table 3.9).

Normal young hamsters (4–8 weeks old) have microcytic normochromic red blood cells with some variation in size and shape, as well as occasional spherocytes and target cells. Polychromasia and Howell-Jolly bodies are found normally in healthy hamsters (Pilny, 2008). Polychromasia is seen more often in hamsters than in humans (Gad, 2007). Hamster red blood cells are nucleated 2% of the time.

Mean Cell Volume (MCV)

The reference range for MCV in the hamster is 64–78 μ^3 (Table 3.9).

Most hamster erythrocyte values are similar to those of the rat, although MCV is slightly higher in the hamster (Dent, 1977).

Mean Cell Hemoglobin (MCH)

The reference range for MCH in the hamster is 20–26 pg per cell (Table 3.9).

Mean Cell Hemoglobin Concentration (MCHC)

The reference range for MCHC in the hamster is 28–37 g/dl (Table 3.9).

RETICULOCYTES The end of hibernation in the hamster is associated with an increase in reticulocyte numbers (Hrapkiewicz and Medina, 2007).

LEUKOCYTES

Reference values for leukocyte and platelet numbers in the hamster are provided in Table 3.10.

TOTAL WHITE BLOOD CELL (WBC) COUNT The reference range for total leukocyte count in the hamster is $3–15 \times 10^3$ cells per µl (Table 3.10) and varies with several factors including age, sex, strain, diurnal rhythm, hibernation, and disease.

Neonatal Syrian hamsters have a total leukocyte count of approximately 2000 cells per microliter. White blood cells increase with age in European hamsters, from 3500 per microliter at 13 days to 8200 per microliter at 65 days and are higher in males than females (Emminger et al., 1975).

Total white blood cell count in Chinese hamsters is higher in the middle of the light cycle than in the middle of the dark cycle (Lappenbusch, 1972).

Hibernation causes a decrease in total white blood cell count in Syrian hamsters from 5000–10 000 per microliter to 2500 per microliter (Heatley and Harris, 2009; Mitruka and Rawnsley, 1977), although the NP:LC ratio remained at 1:1 (Hrapkiewicz and Medina, 2007).

In European hamsters, hibernation is also associated with a lower total white blood cell count and platelet count, with unchanged differential distribution (Reznik et al., 1975). White cell numbers fall to a low of 100 per microliter during hibernation in this strain (Heatley and Harris, 2009). After hibernation, a leukocytosis characterized by neutrophilia occurs, with 70–90% neutrophils and a total leukocyte count of 10 000–20 000 cells per microliter (Heatley and Harris, 2009; Pilny, 2008).

Hamsters experimentally infected with piri-tal virus, a novel arenavirus isolated from the cotton rat (*Sigmodon astoni*) in Venezuela, demonstrate

leukocytosis, including neutrophilia, eosinophilia, and basophilia on days 5–6 post-infection (Sbrana et al., 2006).

LYMPHOCYTES The reference range for total lymphocyte count in the hamster is 50–96% (Table 3.10). Lymphocytes are the predominant circulating leukocyte in hamsters, comprising 60–80% of the differential leukocyte count (Heatley and Harris, 2009). Most hamster lymphocytes are small round cells with a dark-blue nucleus that occupies the majority of the cell, surrounded by a rim of lighter-blue cytoplasm. Hibernation in the hamster causes decreased WBC numbers, with a 1:1 NP:LC ratio (Hrapkiewicz and Medina, 2007).

NEUTROPHILS (HETEROPHILS) The reference range for total neutrophil count in the hamster is 17–35% (Table 3.10). As in many other rodents, hamster neutrophils are also called heterophils because they resemble eosinophils, with a lobular nucleus and dense pink cytoplasmic granules. Hamster neutrophils have a diameter of approximately 10–12 microns. Hamster neutrophils have moderate alkaline phosphatase activity (Tamburlin and Glomski, 1988) but lack lysozyme activity (Thrall et al., 2004).

Hibernation in the hamster causes decreased WBC numbers, with a 1:1 NP:LC ratio (Hrapkiewicz and Medina, 2007). At the end of hibernation, neutrophilic leukocytosis occurs, with 70–90% neutrophils and total leukocyte count of 10 000–20 000 cells per microliter (Heatley and Harris, 2009).

EOSINOPHILS The reference range for total eosinophil count in the hamster is 0–5% (Table 3.10). Hamster eosinophils are similar in appearance to those of other rodent species.

BASOPHILS The reference range for total basophil count in the hamster is 0–5% (Table 3.10). Hamster basophils are similar in appearance to those of other rodent species.

MONOCYTES The reference range for total monocyte count in the hamster is 0–5% (Table 3.10). Hamster monocytes are similar in appearance to those of other rodent species.

PLATELETS (THROMBOCYTES)

The reference range for total platelet count in the hamster is 200–590 $\times$ 10^6 per µl (Table 3.10) and varies with hibernation and disease. Hibernation causes decreased platelet counts in European hamsters (Reznik et al., 1975) and Syrian hamsters (Heatley and Harris, 2009).

Hamsters experimentally infected with pirital virus had increased platelet numbers on days 5 and 6 post-infection (Sbrana et al., 2006).

Coagulation

Reference values for coagulation tests in the hamster are provided in Table 3.11.

BLEEDING TIME (BT)

The reference range for bleeding time in the hamster is 1.5–2.4 minutes (Table 3.11).

PROTHROMBIN TIME (PT)

The reference range for prothrombin time in the hamster is 10.3–10.7 seconds (Table 3.11).

Female hamsters have shorter prothrombin times than males (Dodds et al., 1977).

Hamsters experimentally infected with pirital virus had increased PT by 8 days post-infection (Sbrana et al., 2006).

FIBRINOGEN CONCENTRATION

The reference range for fibrinogen concentration in the hamster is 188–316 g/dl (Table 3.11). Female hamsters have higher concentrations of fibrinogen and plasminogen than males (Dodds et al., 1977).

Bone Marrow

The adult hamster has a highly cellular bone marrow, with a myeloid to erythroid ratio of approximately 0.9:1 (Table 3.8) (Gad, 2007). The largest cells in the hamster bone marrow are megakaryocytes. There are also significant numbers of eosinophils, plasma cells, histiocytes, and a few mast cells (Gad, 2007). The neonatal hamster bone marrow is primarily erythroblastic. The spleen does not play a major role in erythropoiesis in the hamster and becomes erythropoietically active only when stimulated by T3 (Boussios et al., 1982).

OTHER RODENTS

Gerbil

Sample Collection

BLOOD

Gerbil restraint techniques and blood collection are described in the Clinical Biochemistry section of this chapter.

Reference Values and Unique Characteristics

BLOOD VOLUME

Reference values for blood volume in the gerbil are provided in Table 3.8.

TOTAL BLOOD VOLUME The reference range for blood volume in the gerbil is 4.4–8 ml, or 67ml per kg body weight (Table 3.8).

A single blood sample volume in the gerbil is 0.4–1.3 ml per 2 weeks (Table 3.8).

The exsanguination volume of blood in the gerbil is 2–4 ml (Table 3.8).

PERIPHERAL BLOOD CELLS

ERYTHROCYTES Reference values for erythrocyte parameters in the gerbil are provided in Table 3.9.

Red Blood Cell (RBC) Count The reference range for red blood cell count in the gerbil is 7–10 $\times$ 10^6 cells per µl (Table 3.9).

Neonatal gerbils have erythrocyte counts that are approximately half the adult values but increase to adult values by about 8 weeks of age (Pilny, 2008).

Hematocrit (Hct) or Packed Cell Volume (PCV) The reference range for hematocrit in the gerbil is 35–52% (Table 3.9).

Male gerbils have higher hematocrit values than females (Heatley and Harris, 2009).

Hemoglobin (Hgb) The reference range for hemoglobin concentration for the gerbil is 10–17.9 g/dl (Table 3.9).

Erythrocyte Lifespan The erythrocyte lifespan in the gerbil averages 10 days (Table 3.9), which represents a much faster turnover rate than seen in other laboratory animals (Joslin, 2009).

Gerbils up to 20 weeks old have a large number of circulating reticulocytes and erythrocytes with basophilic stippling and polychromasia. These cells are also abundant in older gerbils and are probably associated with the short erythrocyte lifespan (Heatley and Harris, 2009; Pilny, 2008).

Erythrocyte Size and Staining Neonatal gerbils have macrocytic erythrocytes, but this condition resolves by approximately 8 weeks of age (Pilny, 2008).

Mean Cell Volume (MCV) The reference range for MCV in the gerbil is 46.6–60 μ^3 (Table 3.9) and is elevated in neonatal gerbils until approximately 8 weeks old (Pilny, 2008).

Male gerbils have higher MCV values than females (Heatley and Harris, 2009).

Mean Cell Hemoglobin (MCH) The reference range for MCH in the gerbil is 16.1–19.4 pg per cell (Table 3.9).

Mean Cell Hemoglobin Concentration (MCHC) The reference range for MCHC for the gerbil is 30.6–33.3 g/dl (Table 3.9).

MCHC values are higher in male gerbils than in female gerbils (Heatley and Harris, 2009).

Reticulocytes Gerbils up to 20 weeks old have a large number of circulating reticulocytes and erythrocytes with basophilic stippling and polychromasia. These cells are also abundant in older gerbils and are probably associated with the short erythrocyte lifespan (Heatley and Harris, 2009; Pilny, 2008).

LEUKOCYTES Reference values for leukocyte and platelet numbers in the gerbil are provided in Table 3.10.

Total White Blood Cell (WBC) Count The reference range for white blood cell count in the gerbil is 4.3–21.6 $\times$ 10^3 cells per µl (Table 3.10).

Neonatal gerbils have panleukocytosis that resolves by approximately 8 weeks old (Pilny, 2008). Male gerbils have higher total leukocyte counts than females (Heatley and Harris, 2009).

Lymphocytes The reference range for lymphocyte count in the gerbil is 32–97% (Table 3.10).

The predominant leukocyte in gerbils is the lymphocyte, which is much more abundant than granulocytes in this species (Dillon and Glomski, 1975).

Male gerbils have higher total leukocyte count due to a higher number of lymphocytes. The lymphocyte-to-heterophil ratio in the male is 6:1 compared to 3:1 in the female (Heatley and Harris, 2009).

Neutrophils (Heterophils) The reference range for neutrophil count in the gerbil is 2–41% (Table 3.10).

Gerbil neutrophils have moderate alkaline phosphatase activity (Tamburlin and Glomski, 1988).

Eosinophils The reference range for eosinophil count in the gerbil is 0–4% (Table 3.10).

Basophils The reference range for basophil count in the gerbil is 0–2% (Table 3.10).

Monocytes The reference range for monocyte count in the gerbil is 0–9% (Table 3.10).

PLATELETS (THROMBOCYTES) The reference range for platelet number in the gerbil is 400–830 $\times$ 10^6 per µl (Table 3.10).

BONE MARROW

The reference range for myeloid to erythroid ratio in the gerbil is 0.75:1 to 2.35:1 (Table 3.8).

Chinchilla

Sample Collection

BLOOD

Blood collection in the chinchilla is described above in the section on Clinical Biochemistry.

Reference Values and Unique Characteristics

BLOOD VOLUME

Reference values for blood volume in the chinchilla are provided in Table 3.8.

TOTAL BLOOD VOLUME The reference range for blood volume in the chinchilla is 20–32 ml (Table 3.8).

Volume of a single blood sample in the chinchilla is 2–7 ml per 2 weeks (Table 3.8).

Exsanguination volume in the chinchilla is 8–12 ml (Table 3.8).

PERIPHERAL BLOOD CELLS

ERYTHROCYTES Reference values for erythrocyte parameters in the chinchilla are provided in Table 3.9.

Red Blood Cell Count The reference range for red blood cell count in the chinchilla is 5–10 $\times$ 10^6 cells per µl (Table 3.9).

Hematocrit (Hct) or Packed Cell Volume (PCV) The reference range for hematocrit in the chinchilla is 30–55% (Table 3.9).

The packed cell volume, hemoglobin concentration, and MCHC values are higher in blood obtained from chinchillas anesthetized with ketamine and xylazine than in blood obtained by cardiac puncture post-mortem (de Oliveira Silva et al., 2005).

Hemoglobin (Hgb) The reference range for hemoglobin concentration for the chinchilla is 9–15 g/dl (Table 3.9).

Erythrocyte Size and Staining

Mean Cell Volume (MCV) The reference range for MCV in the chinchilla is 32.1–69.2 μ^3 (Table 3.9).

Mean Cell Hemoglobin (MCH) The reference range for MCH in the chinchilla is 10.4–19.8 pg/cell (Table 3.9).

Mean Cell Hemoglobin Concentration (MCHC) The reference range for MCHC in the chinchilla is 20–38.5 g/dl (Table 3.9).

LEUKOCYTES Reference values for leukocyte and platelet numbers in the chinchilla are provided in Table 3.10.

Total White Blood Cell (WBC) Count The reference range for leukocyte number in the chinchilla is 4–25 $\times$ 10^3 cells per µl (Table 3.10). Total leukocyte number is higher in blood obtained from male chinchillas anesthetized with ketamine and xylazine compared to blood obtained by cardiac puncture post-mortem (de Oliveira Silva et al., 2005).

Lymphocytes The reference range for lymphocyte number in the chinchilla is 19–98% (Table 3.10).

The lymphocyte is the most common circulating leukocyte in the chinchilla. The chinchilla responds to inflammation with an early increase in neutrophils and decrease in lymphocytes, resulting in little change in the total leukocyte count (Riggs and Mitchell, 2009).

Neutrophils The reference range for neutrophil number in the chinchilla is 9–78% (Table 3.10). The neutrophil is the second most common circulating leukocyte in the chinchilla (Riggs and Mitchell, 2009). The chinchilla neutrophil has a nucleus that may be hyposegmented, similar to the Pelger-Huet anomaly in the rabbit. Heterophils lack myeloperoxidase, the enzyme that liquefies purulent material, causing chinchilla pus to be thick and caseous (Riggs and Mitchell, 2009).

Eosinophils The reference range for eosinophil number in the chinchilla is 0–9% (Table 3.10). These cells are uncommon in the circulation of normal chinchillas (Riggs and Mitchell, 2009).

Basophils The reference range for basophil number in the chinchilla is 0–11% (Table 3.10). These cells are uncommon in the circulation of normal chinchillas (Riggs and Mitchell, 2009).

Basophil and monocyte numbers were lower in blood obtained from male chinchillas anesthetized with ketamine and xylazine compared to blood obtained by cardiac puncture post-mortem (de Oliveira Silva et al., 2005).

Monocytes The reference range for monocyte number in the chinchilla is 0–6% (Table 3.10). These cells are uncommon in the circulation of normal chinchillas (Riggs and Mitchell, 2009).

PLATELETS (THROMBOCYTES) The reference range for platelet number in the chinchilla is 300–600 $\times$ 10^6 per µl (Table 3.10). A dramatic increase in platelets, of more than 1000 per ml, may occur in response to inflammation in the chinchilla (Riggs and Mitchell, 2009).

Degu

There is little specific information available on the hematologic characteristics of the degu.

The degu is a diurnal rodent and has been the subject of many studies of the effect of circadian rhythm on physiology, behavior, and disease states.

Hematologic and serum protein values for degus of both sexes and all ages studied are reported to be

similar to those of other laboratory rodents (Murphy et al., 1978).

Deer Mouse

Sample Collection

BLOOD

Blood collection in the deer mouse is described above in the section on Clinical Biochemistry.

There is limited information available in the literature regarding hematology parameters in the deer mouse.

Reference Values and Unique Characteristics

PERIPHERAL BLOOD CELLS

ERYTHROCYTES Reference values for erythrocytes in the deer mouse are provided in Table 3.9.

Red Blood Cell Count The reference range for red blood cell count in the deer mouse is 10–12 $\times$ 10^6 cells per µl (Table 3.9).

Hematocrit (Hct) or Packed Cell Volume (PCV) The reference range for hematocrit in the deer mouse is 38–52% (Table 3.9).

Deer mice infected with *Cuterebra* sp. were found to have lower packed cell volumes than uninfected deer mice (Sealander, 1961). By contrast, deer mice infected with *Capillaria hepatica* had elevated hematocrits compared with uninfected controls (Meagher, 1998).

Hemoglobin (Hgb) The reference range for hemoglobin concentration in the deer mouse is 12–16 g/dl (Table 3.9).

Deer mice infected with *Cuterebra* sp. were found to have a lower hemoglobin concentration than uninfected deer mice trapped during the same period (Sealander, 1961).

Erythrocyte Size and Staining

Mean Cell Volume (MCV) The reference range for MCV in the deer mouse is 36–46 μ^3 (Table 3.9).

Mean Cell Hemoglobin (MCH) The reference range for MCH in the deer mouse is 11–15 pg per cell (Table 3.9).

Mean Cell Hemoglobin Concentration (MCHC) The reference range for MCHC in the deer mouse is 30–35 g/dl (Table 3.9).

LEUKOCYTES Reference values for leukocyte numbers in the deer mouse are provided in Table 3.10.

Total White Blood Cell (WBC) Count The reference range for total leukocyte number in the deer mouse is 1–4 $\times$ 10^3 cells per µl (Table 3.10).

Dormouse

There is no information available in the literature regarding hematology values in the dormouse.

Kangaroo Rat

Sample Collection

BLOOD

A discussion of blood sample methods and collection sites in the kangaroo rat is presented above in the Clinical Biochemistry section of this chapter.

Reference Values and Unique Characteristics

PERIPHERAL BLOOD CELLS

ERYTHROCYTES Reference values for erythrocyte characteristics in the kangaroo rat are provided in Table 3.9.

Red Blood Cell Count The reference range for red blood cell count in the kangaroo rat is 7–9 $\times$ 10^6 per µl (Table 3.9). Values are lowest in the winter and highest in the summer (Intress and Best, 1990).

Hematocrit (Hct) or Packed Cell Volume (PCV) The reference range for hematocrit in the kangaroo rat is 47–53% (Table 3.9). Values are lowest in the summer and highest in the winter (Scelza and Knoll, 1982).

Hemoglobin (Hgb) The reference range for hemoglobin concentration in the kangaroo rat is 15–17 g/dl (Table 3.9). Values are fairly consistent throughout the year (Scelza and Knoll, 1982).

Erythrocyte Size and Staining

Mean Cell Volume (MCV) The reference range for MCV in the kangaroo rat is 57.3–66^3 (Table 3.9).

Mean Cell Hemoglobin (MCH) The reference range for MCH in the kangaroo rat is 18.6–25 pg per cell (Table 3.9). Values are lowest in the summer and variably higher during the other seasons (Intress and Best, 1990).

Mean Cell Hemoglobin Concentration (MCHC) The reference range for MCHC in the kangaroo rat is 30–35 g/dl (Table 3.9). Values are fairly consistent throughout the year (Scelza and Knoll, 1982).

LEUKOCYTES Reference values for leukocyte numbers in the kangaroo rat are provided in Table 3.10.

Total White Blood Cell (WBC) Count The reference range for total leukocyte number in the kangaroo rat is 3–9 $\times$ 10^3 per µl (Table 3.10). Values are highest in the spring and variable during the other seasons (Intress and Best, 1990; Scelza and Knoll, 1982).

Cotton Rat

Sample Collection

BLOOD

Blood collection in the cotton rat is described in the Clinical Biochemistry section of this chapter.

Reference Values and Unique Characterstics

PERIPHERAL BLOOD CELLS

ERYTHROCYTES Reference values for erythrocyte characteristics in the cotton rat are provided in Table 3.9.

Cotton rats exposed to 1000 ppm lead in drinking water showed decreased erythrocyte indices in a dose-dependent manner (McMurry et al., 1995).

Hemoglobin (Hgb) The reference range for hemoglobin concentration in the cotton rat is 13–14 g/dl (Table 3.9).

LEUKOCYTES Reference values for leukocyte numbers in the cotton rat are provided in Table 3.10.

Total White Blood Cell (WBC) Count The reference range for total leukocyte number in the cotton rat is 11–13 $\times$ 10^3 cells per µl (Table 3.10).

Several studies have examined the effect of diet and environmental conditions on hematology and immune function of the cotton rat.

Immunocompetence has been suggested as an important influence on survival in herbivore populations (Robel et al., 1996). Supplementation of food quantity and quality (enhanced methionine) did not have a consistent effect on hematology or immune function in adult male cotton rats, although total leukocyte number was increased (Webb et al., 2003).

Cotton rats exposed to 1000 ppm lead in drinking water showed decreased total leukocytes in a dose-dependent manner (McMurry et al., 1995).

Lymphocytes The reference range for lymphocyte number in the cotton rat is 50–54% (Table 3.10).

Neutrophils (Heterophils) The reference range for neutrophil number in the cotton rat is 36–40% (Table 3.10).

Eosinophils The reference range for eosinophil number in the cotton rat is 5–6% (Table 3.10).

Basophils The reference range for basophil number in the cotton rat is 0–0.2% (Table 3.10).

Monocytes The reference range for monocyte number in the cotton rat is 4–5% (Table 3.10).

PLATELETS (THROMBOCYTES) Supplementation of food quantity and quality (enhanced methionine) increased platelet number in the cotton rat (Webb et al., 2003).

Sand Rat

Sample Collection

BLOOD

Blood collection in the sand rat is described above in the section on Clinical Biochemistry.

Reference Values and Unique Characteristics

There is very little information available on hematology parameters in the sand rat.

A study in sand rats found that glucose concentration within red blood cells is higher than in plasma of this species, as it is in other mammals. This phenomenon was explained by loose binding of glucose to macromolecules within the erythrocyte (Halperin and Adler, 1985).

References

Abdu, F., Sleight, S.D., 1965. Etiology of experimental Leptospira pomona infection in hamsters. Cornell Vet. 55, 74–86.

Abelson, J.N., Simon, M.I. (Eds.), 1991. Methods in Enzymology: Hemoglobins, Part B: Biochemical and Analytical Methods. Elsevier, London.

Adjarov, D., Popov, S., Ivanov, E., 1976. Studies on the mechanism of the changes in serum and liver gamma-glutamyl transpeptidase activity. I. Experimental extrahepatic cholestasis in rabbits. Enzyme 21, 1–7.

Allison, S.O., Artwohl, J.E., Fortman, J.D., Pogwizd, S., Jeanes, J., Koske, S., et al. 2007. Iatrogenic hemolytic anemia and endocarditis in New Zealand White rabbits secondary to Achromobacter xylosoxidans infection. J. Am. Assoc. Lab. Anim. Sci. 46, 58–62.

Alpert, M., 1950. Observations on the histophysiology of the adrenal gland of the golden hamster. Endocrinology 46, 166–176.

Amin, T.M., Sirs, J.A., 1985. The blood rheology of man and various animal species. Q. J. Exp. Physiol. (January), 70, 37–49.

Bajusz, E., Homburger, F., 1966. Myopathies. Science 152, 1112–1113.

Bannon, P.D., Friedell, G.H., 1966. Values for plasma constituents in normal and tumor bearing golden hamsters. Lab. Anim. Care 16, 417–420.

Banta, M.R., Holcombe, D.W., 2002. The effects of thyroxine on metabolism and water balance in a desert-dwelling rodent, Merriam's kangaroo rat (*Dipodomys merriami*). J. Comp. Physiol. B. 172, 17–25.

Bayard, S.P., 1974. Another look at the statistical analysis of changes during storage of serum specimens. Health Lab. Sci. 11, 45–49.

Benjamin, M.M., McKelvie, D.H., 1978. Clinical biochemistry. Benirschke, K., Garner, F.M., Jones, T.C. Pathology of Laboratory Animals, vol. 2. Springer-Verlag, New York, pp. 1749–1768.

Benson, K.G., Paul-Murphy, J., 1999. Clinical pathology of the domestic rabbit: acquisition and interpretation of samples. Vet. Clinics North Am. Exot. An. Pract. 2, 539–551.

Betts, A., Tanguay, R., Friedell, G.H., 1964. Effect of necrosis on hemoglobin, serum protein profile, and erythroagglutination reaction in golden hamsters. Proc. Soc. Exp. Biol. Med. 116, 66–69.

Bito, L.Z., Eakins, K.E., 1969. The effect of general anesthesia on the chemical composition of blood plasma of normal rabbits. J. Pharmacol. Exp. Therapeut. 169, 277–286.

Bjoraker, D.G., Ketcham, T.R., 1981. 3.8% sodium citrate (1:9) is an adequate anticoagulant for rabbit blood with high calcium. Thrombosis Res. 24, 505–508.

Bortolotti, N., Castelli, D., Bomati, M., 1989. Hematology and serum chemistry values of adult, pregnant and newborn New Zealand rabbits (*Oryctolagus cuniculus*). Lab. Anim. Sci. 39, 437–439.

Boussios, T., McIntyre, W.R., Gordon, A.S., Bertles, J.F., 1982. Receptors specific for thyroid hormones in nuclei of mammalian erythroid cells: involvement in erythroid cell proliferation. Br. J. Haematol. 51, 99–106.

Brewer, N.R., 2006. Biology of the rabbit. J. Am. Assoc. Lab. Anim. Sci. 45, 8–24.

Brinck-Johnsen, T., Brinck-Johnsen, K., Kilham, L., 1981. Gestational changes in hamster adrenocortical function. J. Steroid Biochem. 14, 835–839.

Brousseau, M.E., Hoeg, J.M., 1999. Transgenic rabbits as models for atherosclerosis research. J. Lipid Res. 40, 365–375.

Brown, S.J., Askenase, P.W., 1982. Blood eosinophil and basophil responses in guinea pigs parasitized by *Amblyomma americanum* ticks. Am. J. Trop. Med. Hyg. 31, 593–598.

Brown, C.J., Donnelly, T.M., 2004. Rodent husbandry and care. Vet. Clin. North Am. Exot. Anim. Pract. 7, 201–225.

Brunner Jr., A., Martins, I.S., Mitsutani, C.Y., Bilotta, J.A., Araújo Peres, C., 1983. A relative morphological evaluation of hemoglobin biosynthesis in peripheral blood reticulocytes of normal and anemic rabbits. Comp. Biochem. Physiol. A Comp. Physiol. 74, 755–760.

Brunnert, S.R., Altman, N.H., 1991. Laboratory assessment of chronic hepatitis in Syrian hamsters. Lab. Anim. Sci. 41, 559–562.

Buetow, B.S., Treuting, P.M., Van Hoosier, G.L., 1999. The hamster. In: Loeb, W.F., Quimby, F.W. (Eds.), The Clinical Chemistry of Laboratory Animals. Taylor & Francis, Philadelphia, pp. 49–63.

Burns, K.F., De Lannoy Jr., C.W., 1966. Compendium of normal blood values of laboratory animals with indication of variations. I. Random-sexed populations of small animals. Toxicol. Appl. Pharmacol. 8, 429–437.

Caisey, J.D., Kling, D.J., 1980. Clinical chemical values for some common laboratory animals. Clin. Chem. 26, 1877–1879.

Campbell, T.W., Ellis, C.K., 2007. Avian and Exotic Animal Hematology and Cytology, third ed.. Blackwell, Ames IA.

Carakostas, M.C., Gossett, K.A., Church, G.E., Cleghorn, B.L., 1986. Evaluating toxin-induced hepatic injury in rats by laboratory results and discriminant analysis. Vet. Pathol. 23, 264–269.

Castex, C., Sutter, B.C.J., 1981. Dynamics of insulin release by perfused edible dormouse (*Glis glis*) pancreas. Diabetologia 20, 489–494.

Center, S.A., Erb, H.N., Joseph, S.A., 1995. Measurement of serum bile acids concentrations for diagnosis of hepatobiliary disease in cats. J. Am. Vet. Med. Assoc. 207, 1048–1054.

Chandrashekar, V., Bartke, A., 1989. The influence of short photoperiod on testicular and circulating precursors in the adult golden hamster. Biol. Reprod. 40, 300–306.

Chelini, M.O., Souza, N.L., Cortopassi, S.R., Felippe, E.C., Oliveira, C.A., 2006. Assessment of the physiological stress response by quantification of fecal corticosteroids. J. Am. Assoc. Lab. Anim. Sci. 45, 8–11.

Cincotta, A.H., Meier, A.H., 1984. Circadian rhythms of lipogenic and hypoglycemic responses to insulin in the golden hamster (*Mesocricetus auratus*). J. Endocrinol. 103, 141–146.

Cizek, L.J., 1961. Relationship between food and water ingestion in the rabbit. Am. J. Physiol. 201, 557–566.

Clampitt, R.B., Hart, R.J., 1978. The tissue activities of some diagnostic enzymes in ten mammalian species. J. Comp. Path. 88, 607–621.

Clifford, C.B., White, W.J., 1999. The guinea pig. In: Loeb, W.F., Quinby, F.W. (Eds.), The Clinical Chemistry of Laboratory Animals (second ed.). Taylor and Francis, Philadelphia, pp. 65–70.

Cox, R.A., Gökcen, M., 1973. A study of golden hamster (*Cricetus cricetus*) alkaline phosphatase isoenzymes. Comp. Biochem. Physiol. 46B, 99–103.

Cox, R.A., Gökcen, M., 1974. Circulating lipids in Golden Hamster (Mesocricetus auratus). Comp. Biochem. Physiol. 49, 655–661.

Cray, C., Rodriguez, M., Zaias, J., Altman, N.H., 2009. Effects of storage temperature and time on clinical biochemical parameters from rat serum. J. Am. Assoc. Lab. Anim. Sci. 48, 202–204.

Dang, V., Bao, S., Ault, A., Murray, C., McFarlane-Mills, J., Chiedi, C., et al. 2008. Efficacy and safety of five injectable anesthetic regimens for chronic blood collection from the anterior vena cava of guinea pigs. J. Am. Assoc. Lab. Anim. Sci. 47, 56–60.

de la Fuente, J., Diaz, M.T., Ibanez, M., Gonzalez de Chavarri, E., 2007. Physiological response of rabbits to heat, cold, noise and mixing in the context of transport. Anim. Welfare 16, 41–47.

de Oliveira Silva, T., Kreutz, L.C., Barcellos, L.J.G., Borella, J., Soso, A.B., Souza, C., 2005. Reference values for chinchilla (*Chinchilla laniger*) blood cells and serum biochemical parameters. Ciencia Rural. 35, 602–606.

de Paolo, L.V., Masoro, E.J., 1989. Endocrine hormones in laboratory animals. In: Loeb, W.F., Quimby, R. (Eds.), The Clinical Chemistry of Laboratory Animals, pp. 279–308.

Dent, N.J., 1977. The use of the Syrian hamster to establish its clinical chemistry and hematology profile. Duncan, W.A., Leonard, B.J. Clinical Toxicology, vol. XVIII. Excerpta Medica, Amsterdam-Oxford, pp. 321–323.

Dillon, W.G., Glomski, C.A., 1975. The Mongolian gerbil: qualitative and quantitative aspects of the cellular blood picture. Lab. Anim. 9, 283–287.

Dineen, J.K., Adams, D.B., 1970. The effect of long term lymphatic drainage on the lympho-myeloid system in the guinea pig. Immunology 19, 11–30.

Dodds, W.H., Raymond, S.L., Moynihan, A.C., McMartin, D.N., 1977. Spontaneous atrial thrombosis in aged Syrian hamsters: II. Hemostasis. Thrombos Haemostas 38, 457–464.

Dolyak, F., Leone, C.A., 1953. Some standard values of the blood and serum of the cotton rat *Sigmodon hispidus texianus*. Trans. Kansas Acad. Sci. 56, 242–245.

Donham, R.S., Stetson, M.H., 1991. The peripubertal golden hamster and the transition between daily and estrous cycle hormone rhythms. Biol. Reprod. 44, 1108–1112.

Donnelly, T.M., Quimby, F.W., 2002. Biology and diseases of other rodents. In: Fox, J.G. et al. (Eds.), Laboratory Animal Medicine. Elsevier, San Diego, (Chapter 7).

Dooley, J., 1984. Sorbitol dehydrogenase and its use in toxicology testing in laboratory animals. Lab. Anim. 13, 20–21.

Duncan, J.R., Prasse, K.W., 1986. Veterinary Laboratory Medicine, Clinical Pathology. Iowa State University Press, Ames Iowa.

Dyer, S.M., Cervasio, E.L., 2008. An overview of restraint and blood collection techniques in exotic pet practice. Vet. Clin. North Am. Exotic Anim. Pract. 11, 423–443.

Ebling, F.J., 1994. Photoperiodic differences during the development in the dwarf hamsters *Phodopus sungorus* and *Phodopus campbelli*. Gen. Compar. Endocrinol. 95, 475–482.

Edwards, K.E., Jenkins, K.L., Mucklow, L.C., Erb, G.E., Wynne-Edwards, K.E., 1994. Endocrinology of the pregnant Djungarian hamster *Phodopus campbelli*. J. Reprod. Fertil. 101, 1–8.

Eisenberg, J.F., Isaac, D.E., 1963. The reproduction of heteromyid rodents in captivity. J. Mammol. 44, 61–67.

El-Hage, A.N., Herman, E.H., Ferrans, V.J., 1983. Examination of the protective effect of ICRF-187 and dimethyl sulfoxide against acetaminophen-induced heptatotoxicity in Syrian golden hamsters. Toxicology 28, 295–303.

El Hani, A., Dalle, M., DeLost, P., 1980. Sexual dimorphism in binding and metabolism of cortisol during puberty in guinea pigs. J. Physiol. (Paris) 76, 25–28.

Emminger, A., Reznik, G., Reznik-Sculler, H., Mohr, U., 1975. Differences in blood values depending on age in laboratory-bred European hamsters (*Cricetus cricetus* L.). Lab. Animal 9, 33–42.

Erb, G.E., Wynne-Edwards, K.E., 1994. Prolactin, follicle-stimulating hormone, and luteinizing hormone levels during preimplantation in the Djungarian hamster (*Phodopus campbelli*). Biol. Reprod. 50, 1328–1333.

Eugster, A.K., Albert, P.J., Kalter, S.S., 1966. Multiple enzyme determinations in sera and livers of tumor-bearing hamsters. Proc. Soc. Exp. Biol. Med. 123, 327–331.

Evans, G.O., 2009. Animal Clinical Chemistry. CRC Press, Boca Raton FL.

Fecteau, K.A., Deeb, B.J., Rickel, J.M., Kelch, W.J., Oliver, J.W., 2007. Diagnostic endocrinology: blood steroid concentrations in neutered male and female rabbits. J. Exot. Pet Med. 16, 256–259.

Fekete, S., 1993. Animal models in experimental atherosclerosis: a critical review. Acta Vet. Hung. 41, 3–9.

Feldman, D.B., McConnell, E.E., Knapka, J.J., 1982. Growth, kidney disease, and longevity of Syrian hamsters (*Mesocricetus auratus*) fed varying levels of protein. Lab. Anim. Sci. 32, 613–618.

Fernandez, N.J., Kidney, B., 2007. Alkaline phosphatase beyond the liver. Vet. Clin. Path. 36, 223–232.

Fernandez, M.L., Vergara-Jimenez, M., Conde, K., Behr, T., Abdel-Fattah, G., 1997. Regulation of apolipoprotein B-containing lipoproteins by dietary soluble fiber in guinea pigs. Am. J. Clin. Nutr. 65, 814–822.

Finco, D.R., 1989. Kidney function. In: Kaneko, J.J. (Ed.), Clinical Biochemistry of Domestic Animals. Academic Press, New York, pp. 496–542.

Fine, J., Quimby, F.W., Greenhouse, D.D., 1986. Annotated bibliography on uncommonly used laboratory animals: mammals. ILAR J. 24, 3–38.

Fiszer, M., Stankiewicz, D., Adamczyk, A., 1979. Normal concentration and excretion values of sodium and potassium ions in urine of Syrian hamsters. Folia Biol. (Krakow) 27, 51–57.

Fluttert, M., Dalm, S., Oitzl, M.S., 2000. A refined method for sequential blood sampling by tail incision in rats. Lab. Anim. 34, 372–378.

Forsythe III, W.A., 1986. Comparison of dietary casein or soy protein effects on plasma lipids and hormone concentrations in the gerbil (*Meriones unguiculatus*). J. Nutr. 116, 1165–1171.

Fox, R.R., Laird, C.W., 1970. Diurnal variations in rabbits: hematological parameters. Am. J. Physiol. 218, 1609–1612.

Fox, R.R., Laird, C.W., 1999. The rabbit. In: The clinical chemistry of laboratory animals. Loeb, W.F., Quimby, F.W. (eds) Taylor and Francis, Philadelphia, pp. 71–83.

Frenkel, J.K., Cook, K., Grady, H.J., Pendleton, S.K., 1965. Effects of hormones on adrenocortical secretion of golden hamsters. Lab. Invest. 14, 142–156.

Frenkel, G., Kraicer, P.F., 1972. Metabolic pattern of sand rats (Psammomys obesus) and rats during fasting. Life Sci. 11, 209–222.

Fudge, A.M., 2000. Laboratory medicine: avian and exotic pets. Saunders, Philadelphia

Fuentes, G.C., Newgren, J., 2008. Physiology and clinical pathology of laboratory New Zealand White rabbits housed individually and in groups. J. Am. Assoc. Lab. Anim. Sci. 47, 35–38.

Furuta, I., Porkka-Heiskanen, T., Scarbrough, K., Tapanainen, J., Turek, F.W., Hsueh, A.J., 1994. Photoperiod regulates testis cell apoptosis in Djungarian hamsters. Biol. Reprod. 51, 1315–1321.

Gad, S.C., 2007. Animal Models in Toxicology. Taylor & Francis, Philadelphia.

Galton, V.A., Galton, M., 1966. Thyroid hormone metabolism in the pregnant Syrian hamster (*Mesocricetus auratus*). Acta Endocrinol. 53, 130–138.

Garcia-Sainz, J.A., Juarez-Ayala, J., Valles, V.E., 1987. Pertussis toxin induces fatty liver, hyperlipemia, and ketosis in hamsters. Toxicon 25, 603–609.

Gerritsen, G.C., 1982. The Chinese hamster as a model for the study of diabetes mellitus. Diabetes 31, 14–25.

Gil, A.G., Silvan, G., Illera, J.C., 2007. Pituitary-adrenocortical axis, serum serotonin and biochemical response after halothane or isoflurane anaesthesia in rabbits. Lab. Anim. 41, 411–419.

Gleiser, C.A., Van Hoosier, G.L., Sheldon, W.G., Read, W.K., 1971. Amyloidosis and renal paramyloid in a closed hamster colony. Lab. Anim. Sci. 21, 197–202.

Gomez-Bautista, M., Rojo-Vazquez, F.A., Alunda, J.M., 1987. The effect of the host's age on the pathology of *Eimeria stiedae* infection in rabbits. Vet. Parasitol. 24, 47–57.

Gordon, S., Cekleniak, W.P., 1961. Serum lipoprotein pattern of the hypercholesterolemic gerbil. Am. J. Physiol. 201, 27–28.

Gough, B.J., Kilgore, S.S., 1964. A comparative hematological study of *Peromyscus* in Louisiana and Colorado. J. Mammol. 45, 421–428.

Green, M.N., Yerganian, G., 1963. Serum proteins in Chinese hamsters with spontaneous hereditary diabetes mellitus. Diabetes 12, 369.

Gruss, M., Westphal, S., Luley, C., Braun, K., 2006. Endocrine and behavioral plasticity in response to juvenile stress in the semi-precocial rodent *Octodon degus*. Psychoneuroendocrinol. 31, 361–372.

Hall, R.E., 1992. Clinical pathology of laboratory animals. In: Gad, S.C., Chengelis, C.P. (Eds.), Animal Models of Toxicology. Marcel Dekker Inc, New York, pp. 179–184.

Halperin, M., Adler, J.H., 1985. Studies on the "labile-bound" glucose compartment in erythrocytes: studies on *Psammomys obesus* (Sand rat) and preliminary studies on human erythrocytes. Clin. Chem. 31, 1219–1221.

Harcourt-Brown, F., 2002. Textbook of rabbit medicine. In: Butterworth-Heinemann, (Ed.), Oxford, UK.

Harkness, J.E., Wagner, J.E., 1995. The Biology and Medicine of Rabbits and Rodents, third ed.. Lea & Febiger, Philadelphia.

Hawkins, M.G., Ruby, A.L., Drazenovich, T.L., Westropp, J.L., 2009. Composition and characteristics of urinary calculi from guinea pigs. J. Am. Vet. Med. Assoc. 234, 214–220.

Heatley, J.J., Harris, M.C., 2009. Hamsters and gerbils. In: Mitchell, Tully, (Eds.), Manual of Exotic Pet Practice. Elsevier, pp. 406–432.

Hem, A., Smith, A.J., Solberg, P., 1998. Saphenous vein puncture for blood sampling of the mouse, rat, hamster, gerbil, guineapig, ferret and mink. Lab. Anim. 32, 364–368.

Herman, E., Zhang, J., Knapton, A., Lipshultz, Rifai, N., Sistare, F., 2006. Serum cardiav troponin T as a biomarker for acute myocardial injury induced by low doses of isoproterenol in rats. Cardiovasc. Toxicol. 6, 211–221.

Hirayama, T., Folmerez, P., Hanson, R., Jonsson, O., Pettersson, S., Roberts, D., Schersten, T., 1986. Effect of oxygen-free radicals on rabbits and human erythrocytes. Scand. J. Thorac. Cardiovasc. Surg. 20, 247–252.

Hoffman, R.A., Davidson, K., Steinberg, K., 1982. Influence of photoperiod and temperature on weight gain, food consumption, fat pads, and thyroxine in male golden hamsters. Growth 46, 150–162.

Homburger, F., Nixon, C.W., Eppenberger, M., Baker, J.R., 1966. Hereditary myopathy in the Syrian hamster: studies on pathogenesis. Ann. N.Y. Acad. Sci. 138, 14–27.

Holowaychuk, M.K., 2006. Renal failure in a guinea pig (*Cavia porcellus*) following ingestion of oxalate containing plants. Can. Vet. J. 47, 787–789.

Horan, P.K., Muirhead, K.A., Gorton, S., Irons, R.D., 1980. Aseptic aspiration of rabbit bone marrow and enrichment for cycling cells. Lab. Anim. Sci. 30, 76–79.

House, E.L., Pansky, B., Jacobs, M.S., 1961. Age changes in blood of the golden hamster. Am. J. Physiol. 200, 1018–1022.

Hrapkiewicz, K., Medina, L., 2007. Clinical Laboratory Animal Medicine, second ed.. Blackwell Publishing, Ames Iowa.

Hu, M., Wu, Y.-S., Wu, H.-W., 1997. Effects of lithium deficiency in some insulin-sensitive tissues of diabetic Chinese hamsters. Biol. Trace Elem. Res. 58, 91–102.

Huhman, K.L., Mougey, E.H., Moore, T.O., Meyerhoff, J.L., 1995. Stressors, including social conflict, decrease plasma prolactin in male golden hamsters. Hormones Behav. 29, 581–592.

Hulbert, A.J., Hinds, D.S., MacMillen, R.E., 1985. Minimal metabolism, summit metabolism and plasma thyroxine in rodents from different environments. Comp. Biochem. Physiol. 81A, 687–693.

Hunt, C.E., Harrington, D.D., 1974. Nutrition and nutritional diseases of the rabbit. In: Weisbroth, S.H., Flatt, R.E., Kraus, A.L. (Eds.), The Biology of the Laboratory Rabbit. Academic Press, New York, pp. 403–433.

Intress, C., Best, T.L., 1990. Dipodomys panamintinus. Mamm. Spec. 354, 1–7.

Jain, N.C., 1986. Hematologic techniques. In: Jain, N.C. (Ed.), Veterinary Hematology. Lea & Febiger, Philadelphia, pp. 20–86.

Jallageas, M., Assenmacher, I., 1983. Annual plasma testosterone and thyroxine cycles in relation to hibernation in the edible dormouse, *Glis glis*. Gen. Comp. Endocrinol. 50, 452–462.

Jallageas, M., Assenmacher, I., 1986. Suppression of annual plasma testosterone and thyroxine cycles in the edible dormouse *Glis glis* under constant photoperiod at 24°C. Cell. Molec. Life Sci. 42, 171–173.

Jallageas, M., Mas, N., Gautron, J.-P., Saboreau, M., Roussel, J.-P., 1992. Seasonal changes in thyroid-gonadal interactions in the edible dormouse, *Glis glis*. J. Comp. Physiol. B. 162, 153–158.

Joslin, J.O., 2009. Blood collection techniques in exotic small animals. J. Exotic Pet Med. 18, 117–139.

Kania, S.A., 2008. Flow cytometry applications for exotic animals. Vet. Clinics North Am. Exot. Anim. Pract. 11, 583–595.

Kaplan, H.M., Timmons, E.H., 1979. The Rabbit: a Model for the Principles of Mammalian Physiology and Surgery. Academic Press Inc., New York, p. 86.

Keino, M., Hirasaw, K., Watari, T., Kanno, M., Mikawa, M., Saito, K., et al. 2002. Minimally invasive method for collection of blood from guinea pigs. Jpn. J. Infect. Dis. 55, 27–28.

Kenagy, G.J., Place, N.J., Veloso, C., 1999. Relation of glucocorticoids and testosterone to the annual cycle of free-living degus in semi-arid central Chile. Gen. Comp. Endocrinol. 115, 236–243.

Kerr, M., 1989. Veterinary Laboratory Medicine: Clinical Biochemistry and Haematology. Blackwell Publishing, Ames Iowa.

Kessler, C.M., Bell, W.R., 1980. Stimulation of fibrinogen synthesis: a possible functional role of fibrinogen degradation products. Blood 55, 40–47.

Kita, T., Brown, M.S., Watanabe, T., Goldstein, J.L., 1981. Deficiency of low-density lipoprotein receptors in liver and adrenal gland of the WHHL rabbit, an animal model of familial hypercholesterolemia. Proc. Nat. Acad. Sci. (USA) 78, 2268–2272.

Kitagaki, M., Yamaguch, M., Nakamura, M., Sakurada, K., Suwa, T., Sasa, H., 2005. Age-related changes in haematology and serum chemistry of Weiser-Maples guineapigs (*Cavia porcellus*). Lab. Anim. 39, 321–330.

Kozma, C., Mackin, W., Cummins, L.M., Mauer, R., 1974. The anatomy, physiology and the biochemistry of the rabbit. In: Weisbroth, S.H., Flatt, R.E., Kraus, A.L. (Eds.), The Biology of the Laboratory Rabbit. Academic Press, San Diego, pp. 59–64.

Kramer, J.W., 1989. Clinical enzymology. In: Kaneko, J.J. (Ed.), Clinical Biochemistry of Domestic Animals (fourth ed.). Academic Press, San Diego, pp. 338–363.

Kriesten, K., Murawski, U., Schmidtmann, W., 1987. Haematological values during normal reproduction of the maternal and the fetal rabbit. Comp. Biochem. Physiol. 87A, 479–485.

Kutscher, C., 1968. Plasma volume change during water-deprivation in gerbils, hamsters, guinea pigs and rats. Comp. Biochem. Physiol. 25, 929–936.

Laird, C.W., Fox, R.R., 1970. The effect of age on cholesterol and PBI levels in the 3-3c hybrid rabbit. Life Sci. II 9, 1243–1253.

Lang, H., 1981. Creatine Kinase Isoenzymes. Springer-Verlag, Berlin.

Lappenbusch, W.L., 1972. Effect of circadian rhythm on the radiation response of the Chinese hamster (*Cricetulus griseus*). Radiation Res. 50, 600–610.

Lee, M.J., Clement, J.G., 1990. Effects of soman poisoning on hematology and coagulation parameters and serum biochemistry in rabbits. Mil. Med. 155, 244–249.

Lester, P.A., Rush, H.G., Sigler, R.E., 2005. Renal transitional cell carcinoma and choristoma in a degu (Octodon degus). Contemp. Topics Lab. Anim. Sci. 44, 41–44.

Lewis, J.H., 1996. Rabbits. In: Lewis, H.J. (Ed.), Comparative Hemostasis in Vertebrates. Springer, New York, pp. 182–193.

Lindena, J., Trautschold, I., 1986. Catalytic enzyme activity concentration in plasma of man, sheep, dog, cat, rabbit, guinea pig, rat and mouse. Approach to a quantitative diagnostic enzymology. J. Clin. Chem. Clin. Biochem. 24, 11–18.

Longley, L., Fiddes, M., O'Brien, M., 2008. Anaesthesia of exotic pets. Elsevier, London.

Mader, D.R., 2004. Basic approach to veterinary care. In: Queesenberry, K.E., Carpenter, J.W. (Eds.), Ferrets, Rabbits, and Rodents: Clinical Medicine and Surgery (second ed.). Saunders, St. Louis., pp. 147–154.

Mahmoodian, F., Peterkofsky, B., 1996. Vitamin C deficiency in guinea pigs ... caused by scurvy in guinea pigs. Arch. Biochem. Biophys. 325, 295–303.

Manin, M., Tournaire, C., Delost, P., 1982. Measurement of the rate of secretion, peripheral metabolism and interconversion of cortisol and cortisone in adult conscious male guinea-pigs. Steroids 39, 81–88.

Manning, J.P., Inglis, N.R., Green, S., Fishman, W.H., 1970. Characterization of placental alkaline phosphatase from the rabbit, guinea pig, mouse, and hamster. Enzymologia. 39, 307–318.

Marder, J., Eylath, U., Moskovitz, E., Sharir, R., 1990. The effect of heat exposure on blood chemistry of the hyperthermic rabbit. Comp. Biochem. Physiol. A 97, 245–247.

Marini, R.P., Jackson, L.R., Esteves, M.I., Andrutis, K.A., Goslant, C.M., Fox, J.G., 1994. Effect of isoflurane on hematological variables in ferrets. Am. J. Vet. Res. 55, 1479–1483.

Markert, C.L., Shaklee, J.B., Whitt, G.S., 1975. Evolution of a new gene. Multiple genes for LDH isoenzymes provide a model of the evolution of gene structure function and regulation. Science 189, 102–114.

Marlow, C., 1995. Diabetes in a chinchilla. Vet. Rec. 136, 595–596.

Marshall, K.L., 2008. Rabbit hematology. Vet. Clinics North Am. Exotic Anim. Pract. 11, 551–567.

Martin, L.B., Chidambaram, R.M., Schroeder, K.E., McFadden, S.L., 2005. Thyroparathyroidectomy procedures and thyroxine levels in the chinchilla. Contemp. Topics Lab. Aniom. Sci. 44, 31.

Matsuoka, T., Mizuguchi, H., Mizoguchi, Y., Endho, T., Kamata, R., Fukuda, K., et al. 2009. Effects of restricted feeding on blood parameters in pregnant rabbits. J. Toxicol. Sci. 34, 129–137.

Matt, K.S., Soares, M.J., Talamantes, F., Bartke, A., 1983. Effects of handling and ether anesthesia on serum prolactin levels in the golden hamster. Proc. Soc. Exp. Biol. Med. 173, 463–466.

Maxwell, K.O., Wish, C., Murphy, J.C., Fox, J.G., 1985. Serum chemistry reference values in two strains of Syrian hamsters. Lab. Anim. Sci. 35, 67–70.

McClure, D.E., 1999. Clinical pathology and sample collection in the laboratory rodent. Vet. Clin. North Am. Exotic Anim. Pract. 2, 565–590.

McGuill, M.W., Rowan, A.N., 1989. Biological effects of blood loss; implications for sampling volumes and techniques. ILAR J. 31, 5–20.

McLaughlin, R.M., Fish, R.E., 1994. Clinical biochemistry and hematology. In: Manning, P.J., Ringler, D.H., Newcomer, C.E. (Eds.), The Biology of the Laboratory Rabbit (second ed.). Academic Press, San Diego, pp. 111–127.

McMurry, S.T., Lochmiller, R.L., Chandra, S.A., Qualls Jr, C.W., 1995. Sensitivity of selected immunological, hematological, and reproductive parameters in the cotton rat (*Sigmodon hispidus*) to subchronic lead exposure. J. Wildlife Dis. 31, 193–204.

Meagher, S., 1998. Physiological responses of deer mice (*Peromyscus maniculatus*) to infection with *Capillaria hepatica* (*nematoda*). J. Parsitol. 84, 1112–1118.

Melillo, A., 2007. Rabbit clinical pathology. J. Exot. Pet Med. 16, 135–145.

Merrick, B.A., Bruno, M.E., Madenspacher, J.H., Wetmore, B.A., Foley, J., Pieper, R., et al. 2006. Alterations in the rat serum proteome during liver injury from acetaminophen exposure. J. Pharmcol. Exp. Therapeut. 318, 792–802.

Mitruka, B.M., Rawnsley, H.M., 1977. Clinical, Biochemical and Hematological Reference Values in Normal Experimental Animals. Masson Publishing, New York.

Mitruka, B.M., Rawnsley, H.M., 1981. Clinical, Biochemical and Hematological Reference Values in Normal Experimental Animals and Normal Humans. Masson Publishing, New York.

Mitsuguchi, Y., Ito, T., Ohwada, K., 2008. Pathologic findings in rabbit models of hereditary hypertriglyceridemia and hereditary postprandial hypertriglyceridemia. Comp. Med. 58, 465–480.

Mohawk, J.A., Cashen, K., Lee, T.M., 2005. Inhibiting cortisol response accelerates recovery from a photic phase shift. Am. J. Physiol. Regul. Integr. Comp. Physiol. 288, R221–R228.

Moore, D.M., 2000. Hematology of Rabbits and Hematology of the Guinea Pig. In: Feldman, B.F., Zinkl, J.G., Jain, N.C. (Eds.), Schlam's Veterinary Hematology, fifth ed. Lippincott Williams & Wilkins, pp. 1100–1110.

Murphy, J.C., Niemi, S.M., Hewes, K.M., Zink, M., Fox, J.G., 1978. Hematologic and serum protein reference values of the Octodon degus. Am. J. Vet. Res. 39, 713–715.

Murphy, J.C., Fox, J.G., Niemi, S.M., 1984. Nephrotic syndrome associated with renal amyloidosis in a colony of Syrian hamsters. J. Am. Vet. Med. Assoc. 185, 1359–1362.

Murray, M.J., 2000. Rabbit and ferret sampling and artifact considerations. In: Fudge, A.M. (Ed.), Laboratory Medicine: Avian and Exotic Pets. Saunders, Philadelphia, pp. 265–268.

Najecki, D., Tate, B., 1999. Husbandry and management of the degu. Lab Anim. (NY) 28, 54–62.

Neve, P., Authelet, M., Goldstein, J., 1981. Effect of aging on the morphology and function of the thyroid gland of the cream hamster. Cell Tissue Res. 220, 499–509.

Nishi, M., Steiner, D.F., 1990. Cloning of complementary DNAs encoding islet amyloid polypeptide, insulin, and glucagon precursors from a New World rodent, the degu, *Octodon degus*. Mol. Endocrinol. 4, 1192–1198.

Ogilvie, K.M., Donham, R.S., Stetson, M.H., 1992. Daily rhythms of follicle-stimulating hormone in adult anestrous and prepubertal female Turkish hamsters (*Mesocricetus brandti*). Biol. Reprod. 46, 279–283.

Opazo, J.C., Soto-Gamboa, M., Bozinovic, F., 2004. Blood glucose concentration in caviomorph rodents. Comp. Biochem. Physiol. Part A 137, 57–64.

O'Rourke, D.P., 2004. Diseases and problems of guinea pigs. In: Queesenberry, K.E., Carpenter, J.W. (Eds.), Ferrets, Rabbits, and Rodents (second ed.). Saunders, St. Louis, pp. 245–254.

Ortega-Corona, B.G., Esparza-Avalos, N., Benitez-King, G., Pratz-Guitrado, S., Anton-Tay, S., 1991. Effects of melatonin administration on plasma glucose levels. Proc. Western Pharmacol. Soc. 34, 75–77.

Ottenweller, J.E., Tapp, W.N., Burke, J.M., Natelson, B.H., 1985. Plasma cortisol and corticosterone concentrations in the golden hamster (*Mesocricetus auratus*). Life Sci. 37, 1551–1558.

Owens, H., Siparsky, G., Bajaj, L., Hampers, L., 2005. Correction of factitious hyperkalemia in hemolyzed specimens. Am. J. Emerg. Med. 23, 872–875.

Palm, M., Lundblad, A., 2005. Creatinine concentration in plasma from dog, rat and mouse: a comparison of three different methods. Vet. Clin. Path. 34, 232–236.

Percy, D.H., Barthold, S.W., 2007. Pathology of Laboratory Rabbits and Rodents, third ed.. Blackwell, Ames Iowa.

Perwaiz, S., Tuchweber, B., Mignault, D., Gilat, T., Yousef, I.M., 2001. Determination of bile acids in biological fluids by liquid chromatography-electrospray tandem mass spectrometry. J. Lipid Res. 42, 114–119.

Pilny, A.A., 2008. Clinical hematology of rodent species. Vet. Clin. North Am. Exotic. Anim. Pract. 11, 523–533.

Pouliot, N., Maghni, K., Blanchette, F., et al. 1996. Natural killer and lectin-dependent cytotoxic activities of Kurloff cells: Target cell selectivity, conjugate formation, and Ca^{++} dependency. Inflammation 20, 647–671.

Powsner, E.R., Fly, M.N., 1962. Aseptic aspiration of bone marrow from the living rabbit. J. Appl. Physiol. 17, 1021–1022.

Prince, G.A., 1994. The cotton rat in biomedical research. AWIC Newsletter 5, 1–6.

Redgate, E.S., Fox, R.R., Taylor, F.H., 1981. Strain and age effects on immobilization stress in JAX rabbits. Proc. Soc. Exp. Biol. Med. 166, 442–448.

Rennie, J.S., MacDonald, D.G., Douglas, T.A., 1981. Haemoglobin, serum iron and transferrin values of adult male Syrian hamsters (Mesocricetus auratus). Lab. Anim. 15, 35–36.

Reznik, G., Reznik-Schüller, H., Emminger, A., Mohr, U., 1975. Comparative studies of blood from hibernating and nonhibernating European hamsters (Cricetus cricetus L). Lab. Anim. Sci. 25, 210–215.

Reznik, G.K., Padberg, G., 1991. Diethylnitrosamine-induced metastasizing hepatocellular carcinomas in New Zealand white rabbits: a tumor model for clinical investigations. J. Cancer Res. Clin. Oncol. 117, 123–129.

Richmond, W., 1992. Analytical reviews in clinical biochemistry: the quantitative analysis of cholesterol. Ann. Clin. Biochem. 29, 577–597.

Riggs, S.M., 2009. In: Mitchell & Tully (Eds.), Exotic Pet Practice. W.B. Saunders, St. Louis MO.

Riggs, S.M., Mitchell, M.A., 2009. Chinchillas. In: Mitchell, M.A., Tully Jr., T.N. (Eds.), Manual of Exotic Pet Practice. W.B. Saunders, St. Louis, pp. 474–492.

Ringler, D.H., Dabich, L., 1979. Haematology and clinical biochemistryBaker, H.J.Lindsey, J.R.Weisbroth, S.H. The Laboratory Rat, vol. 1. Academic Press, San Diego, pp. 105–121.

Robel, G.L., Lochmiller, R.L., McMurry, S.T., Qualls Jr., C.W., 1996. Environmental, age, and sex effects on cotton rat (Sigmodon hispidus) hematology. J. Wildl. Dis. 32, 390–394.

Rollinson, E.A., White, G., 1983. Relative activities of acyclovir and BW 759 against Aujeszky's disease and equine rhinopneumonitis viruses. Antimicrob. Agents Chemother. 24, 221–226.

Rothwell, T.L.W., Pope, S.E., Rajczyk, Z.K., Collins, G.H., 1991. Haematological and pathological responses to experimental *Trixacarus caviae* infection in guinea pigs. J. Comp. Pathol. 104, 179–185.

Sabin, F.R., Miller, F.R., Smithburn, K.C., Thomas, R.M., Hummel, L.E., 1936. Changes in the bone marrow and blood cells of developing rabbits. J. Exp. Med. 64, 97–120.

Saibaba, K.S.S., Bhaskar, M.V., Srinivasa Rao, P.V.L.N., Ramana, G.V., Dakshinamurty, K.V., 1998. Interferences in clinical chemistry analysis. Indian J. Clin. Biochem. 13, 55–62.

Sanderson, J.H., Phillips, C.E., 1981. An atlas of laboratory animal haematology. Clarendon Press, Oxford, England.

Saunders, R.A., Davies, R.R., 2005. Notes on Rabbit Internal Medicine. Blackwell, Oxford UK.

Sbrana, E., Mateo, R.I., Xiao, S.-Y., Popov, V.L., Newman, P.C., Tesh, R.B., 2006. Clinical laboratory, virologic, and pathologic changes in hamsters experimentally infected with pirital virus (Arenaviridae): a rodent model of Lassa fever. Am. J. Trop. Med. Hyg. 74, 1096–1102.

Scelza, J., Knoll, J., 1982. Seasonal variation in various blood indices of the kangaroo rat, *Dipodomys panamintinus*. Comp. Biochem. Physiol. 71A, 237–241.

Schuchman, S.M., 1980. Individual care and treatment of rabbits, mice, rats, guinea pigs, hamsters, and gerbils. In: Kirk, R.W. (Ed.), Current Veterinary Therapy, VII, Small Animal Practice. Saunders, Philadelphia, pp. 741–767.

Sealander, J.A., 1960. Hematological changes in deer mice acclimated at different ambient temperatures. Am. J. Physiol. 198, 195–199.

Sealander, J.A., 1961. Hematological values in deer mice in relation to botfly infection. J. Mammol. 42, 57–60.

Shomer, N.H., Astrofsky, K.M., Dangler, C.A., Fox, J.G., 1999. Biomethod for obtaining gastric juice and serum from the unanesthetized guinea pig (*Cavia porcellus*). Contemp Top Lab. Anim. Sci. 38, 32–35.

Sikoski, P., Trybus, J., Cline, J.M., Muhammad, F.S., Eckhoff, A., Tan, J., et al. 2008. Cystic mammary adenocarcinoma associated with a prolactin-secreting pituitary adenoma in a New Zealand White rabbit (*Oryctolagus cuniculus*). Comp. Med. 58, 297–300.

Sisk, D.B., 1976. Physiology. In: Wagner, J.E., Manning, P.J. (Eds.), The Biology of the Guinea Pig. Academic Press, Inc., New York, pp. 63–98.

Snitily, M.U., Gentry, M.J., Mellencamp, M.A., Preheim, L.C., 1991. A simple method for collection of blood from the rat foot. Lab. Anim. Sci. 41, 285–287.

South, F.E., Jeffay, H., 1958. Alterations in serum proteins of hibernating hamsters. Proc. Soc. Exp. Biol. Med. 98, 885–887.

Stankovic, A.K., Smith, S., 2004. Elevated serum potassium values: the role of preanalytical variables. Am. J. Clin. Pathol. 121, S105–S112.

Suckow, M.A., Douglas, F.A., 1997. The Laboratory Rabbit. CRC Press, Boca Raton.

Sullivan, M.P., Cerda, J.J., Robbins, F.L., Burgin, C.W., Beatty, R.J., 1993. The gerbil, hamster, and guinea pig as rodent models for hyperlipidemia. Lab. Anim. Sci. 43, 575–578.

Sundberg, R.D., Hodgson, R.E., 1949. Aspiration of bone marrow in laboratory animals. Blood 4, 557–561.

Szabo, M.P.J., Aoki, V.L., Sanches, F.P.S., Aquino, L.P.T.C.T., Garcia, M.V., Machado, R.Z., et al. 2003. Antibody and blood leukocyte response in *Rhipicephalus sanguineus* tick-infected dogs and guinea pigs. Vet. Parasitol. 115, 49–59.

Takahashi, M, Nagase, S., Takahashi, M., Kokubo, T., Hayashi, Y., 1981. Changes of amylase during experimental pancreatic carcinogenesis in hamsters. Gann 72, 615–619.

Tamburlin, J.H., Glomski, C.A., 1988. Leucocyte alkaline phosphatase in the Mongolian gerbil and other species: a comparative view. Lab. Anim. 22, 202–205.

Thomas, C.L., Artwohl, J.E., Suzuki, H., Gao, X., White, E., Saroli, A., et al. 1997. Initial characterization of hamsters with spontaneous hypertension. Hypertension 30, 301–304.

Thomson, M.B., Kunze, D.J., 1984. Polyethylene glycol-6000 as clearing agent for lipemic serum samples from dogs and the effects on 13 serum assays. Am. J. Vet. Res. 45, 2154–2157.

Thrall, M.A., Baker, D.C., Campbell, T.W., DeNicola, D., Fettman, M.J., Lassen, E.D., et al. 2004. Veterinary Hematology and Clinical Chemistry. Lippincott Williams & Wilkins, Baltimore.

Tietz, N.W., 1976. Fundamental Clinical Chemistry. W.B. Saunders, Philadelphia.

Tomson, F.N., Wardrop, K.J., 1987. Clinical chemistry and hematology. In: VanHoosier, G.L., McPherson, C.W. (Eds.), Laboratory Hamsters. Academic Press, San Diego, pp. 43–60.

Toth, L.A., Krueger, J.M., 1989. Effects of microbial challenge on sleep in rabbits. FASEB J. 3, 2062–2066.

Tracy, R.L., Walsberg, G.E., 2002. Kangaroo rats revisited: re-evaluating a classic case of desert survival. Oecologia 133, 449–457.

Tsuchiya, H., Bates, C.J., 1994. Ascorbic acid deficiency in guinea pigs: contrasting effects of tissue ascorbic acid depletion and of associated inanition on status indices related to collagen and vitamin D. Brit. J. Nutr. 72, 745–752.

Turner, T.T., Howards, S.S., 1977. Hyperglycemia in the hamster anesthetized with inactin (5-ethyl-5(1-methyl propyl)-2-thiobarbiturate. Lab. Anim.Sci. 27, 380–382.

Tschantz, J.-C., Sunahara, G.I., 1993. Microaffinity chromatographic separation and characterization of lipoprotein fractions in rat and Mongolian gerbil serum. Clin. Chem. 39, 1861–1867.

U.S. EPA, 2008. Health effect testing guidelines subpart D. Chronic exposure. No. 40 CFR Part 798. Environmental Protection Agency, Washington, D. C.

Vaughan, M.K., Brainard, G.C., Reiter, R.J., 1984. The influence of natural short photoperiod and temperature conditions on plasma thyroid hormones and cholesterol in male Syrian hamsters. Int. J. Biometeriol. 28, 201–210.

Vaughan, M.K., Powanda, M.C., Richardson, B.A., King, T.S., Johnson, L.Y., Reiter, R.J., 1982. Chronic exposure to short photoperiod inhibits free thyroxine index and plasma levels of TSH, T4, triiodothyronine (T3), and cholesterol in female Syrian hamsters. Compar. Biochem. Physiol. 71A, 1061–1987.

Vennen, K.M., Mitchell, M.A., 2009. Rabbits. In: Mitchell, M.A., Tully Jr., T.N. (Eds.), Manual of Exotic Pet Practice. B. Saunders, St. Louis, pp. 375–405.

Viard-Drouet, F., Provot, F, Coudert, P., 1984. Changes in plasma parameters in rabbit does as a function of their physiological state and feed rationing. Ann. Rech. Vet. 15, 417–424.

Volkert, W.A., Mussachia, X.J., 1970. Blood glases in hamsters during hypothermia by exposure to He-O2 mixture and cold. Am. J. Physiol. 219, 919–922.

Waner, T., Avidar, Y., Peh, H.-C., Zass, R., Bogin, E., 1996. Hematology and clinical chemistry values of normal and euthymic hairless adult male Dunkin-Hartley guinea pigs (*Cavia porcellus*). Vet. Clin. Path. 25, 61–64.

Wang, L., Yu, J., Walzem, R.L., 2008. High-carbohydrate diets affect the size and composition of plasma lipoproteins in hamsters (*Mesocricetus auratus*). Comp. Med. 58, 151–160.

Wardrop, J.K., Van Hoosier, G., 1989. The hamster. In: Loeb, W.F., Quimby, F.W. (Eds.), The Clinical Chemistry of Laboratory Animals. Pergamon Press, New York

Webb, R.E., Leslie Jr., D.M., Lochmiller, R.L., Masters, R.E., 2003. Immune function and hematology of male cotton rats (*Sigmodon hispidus*) in response to food supplementation and methionine. Comp. Biochem. Physiol. Part A 136, 577–589.

Weingand, K., Brown, G., Hall, R., Davies, D., Gossett, K., Neptun, D., et al. 1996. Harmonization of animal clinical pathology testing in toxicity and safety studies. Fund. Appl. Toxicol. 29, 198–201.

Wicher, V., Scarozza, A.M., Ramsingh, A.I., Wicher, K., 1998. Cytokine gene expression in skin of susceptible guinea-pig infected with *Treponema pallidum*. Immunology 95, 242–247.

Wriston Jr., J.C., 1984. Comparative biochemistry of the guinea pig: a aprtial checklist. Comp. Biochem. Physiol. B77, 253–278.

Yamanaka, W., Ostwald, R., French, S., 1967. Histopathology of guinea pigs with cholesterol induced anemia. Proc. Soc. Exp. Biol. Med. 125, 303–306.

Yi, T.M., Walsh, K., Schimmel, P., 1991. Rabbit muscle creatine kinase: genomic cloning, sequencing, and analysis of upstream sequences important for expression in myocytes. Nucl. Acids Res. 19, 3027–3033.

Yoganathan, S., Wilson, T.A., Nicholosi, R.J., 1998. Housing conditions effect plasma lipid concentration and early atherogenesis independent of treatment in hamsters. Nutr. Res. 18, 83–92.

Yousef, M.K., Johnson, H.D., 1975. Thyroid activity in desert rodents: a mechanism for lowered metabolic rate. Am. J. Physiol. 229, 427–431.

Yu, L., Pragay, D.A., Chang, D., Wicher, K., 1979. Biochemical parameters of normal rabbit serum. Clin. Biochem. 12, 83–87.