Disorders of Calcium: Hypercalcemia and Hypocalcemia

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CHAPTER • 6

DISORDERS OF CALCIUM: HYPERCALCEMIA AND HYPOCALCEMIA

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Calcium is required in the body for many vital intracellular and extracellular functions, as well as for skeletal support. Ionized calcium (iCa or Ca^{2+}) is required for enzymatic reactions, membrane transport and stability, blood coagulation, nerve conduction, neuromuscular transmission, muscle contraction, vascular smooth muscle tone, hormone secretion, bone formation and resorption, control of hepatic glycogen metabolism, and cell growth and division.[448] Intracellular calcium ion is one of the primary regulators of the cellular response to many agonists and serves as "an almost universal ionic messenger, conveying signals received at the cell surface to the inside of the cell."[421] In addition to serving as an intracellular messenger, the iCa concentration in the extracellular fluid (ECF) regulates cell function in many organs, including the parathyroid gland, kidney, and thyroid C cells by binding to a newly identified cell membrane–bound calcium-sensing receptor.[75] Normal homeostatic control mechanisms usually maintain the serum calcium concentration within a narrow range and guarantee an adequate supply of calcium for intracellular function. These mechanisms must be disrupted for hypercalcemia or hypocalcemia to develop. Abnormal serum calcium concentrations are of diagnostic value and contribute to the development of lesions and clinical signs. Technologic advances in the measurement of serum iCa concentration, parathyroid hormone (PTH), parathyroid hormone–related protein (PTHrP), and vitamin D metabolites have provided tools that allow greater diagnostic accuracy in the investigation of calcium disorders.

Veterinarians must frequently interpret abnormal serum calcium concentrations. Large deviations of serum calcium concentration from normal occur infrequently, but small deviations may be equally important because they also provide diagnostic clues to an underlying disease. The magnitude of altered serum calcium concentration often does not suggest a specific diagnosis or the extent of disease. Furthermore, a normal serum calcium concentration does not eliminate a disorder of calcium homeostasis.

NORMAL PHYSIOLOGY

OVERVIEW OF CALCIUM HOMEOSTASIS

Regulation of serum calcium concentration is complex and requires the integrated actions of PTH, vitamin D metabolites, and calcitonin (Fig. 6-1). PTH and calcitriol (1,25-dihydroxyvitamin D_3) are the main regulators of calcium homeostasis and have major regulatory effects on each other.[435] PTH is largely responsible for the minute-to-minute control of serum iCa concentration, whereas calcitriol maintains day-to-day control. In the fetus, the parathyroid glands and placenta produce PTHrP, which binds to PTH receptors and regulates calcium balance.[530] After birth, the parathyroid glands modify their pattern of hormone secretion and produce predominantly PTH. Other hormones, including adrenal corticosteroids, estrogens, thyroxine, growth hormone, glucagon, and prolactin, have less influence on calcium homeostasis but may play a role during growth, lactation, or certain disease states.

The intestine, kidney, and bone are the major target organs affected by calcium regulatory hormones. These interactions allow conservation of calcium in the ECF by renal tubular reabsorption, increased intestinal transport of calcium from the diet, and internal redistribution of calcium from bone (Fig. 6-2). The intestine and kidneys are the major regulators of calcium balance in health.[167] Normally, dietary calcium intake equals the amount of calcium lost in urine and feces. The enteric absorption of calcium depends on the physiologic status of the intestines (e.g., acidity, presence of other dietary components, integrity of the villi or presence of small intestinal disease, and degree of enterocyte stimulation by calcitriol). Non–protein-bound calcium is filtered by the glomerulus and undergoes extensive renal reabsorption. This process results in reclamation of more than 98% of the filtered calcium in health.[137,439]

The skeleton provides a major supply of calcium and phosphorus when intestinal absorption and renal

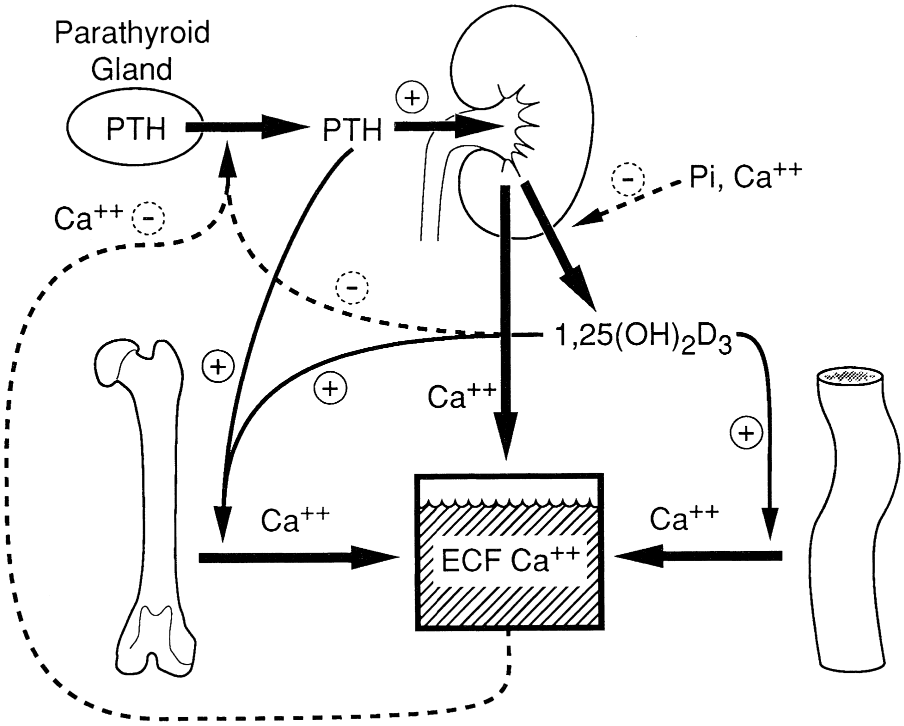

Fig. 6-1 Regulation of extracellular fluid (ECF) calcium concentration by the effects of parathyroid hormone (PTH) and calcitriol (1,25-dihydroxyvitamin D_3) on gut, kidney, bone, and parathyroid gland. The principal effect of PTH is to increase the ECF calcium concentration by mobilizing calcium from bone, increasing tubular calcium reabsorption, and, indirectly on the gut, by increasing calcitriol synthesis. The principal effect of calcitriol is to increase intestinal absorption of calcium, but it also exerts negative regulatory control of PTH synthesis and further calcitriol synthesis. (Modified from Habner JF, Rosenblatt M, Pott JT: Parathyroid hormone: biochemical aspects of biosynthesis, secretion, action, and metabolism, *Physiol Rev* 64:1000, 1984.)

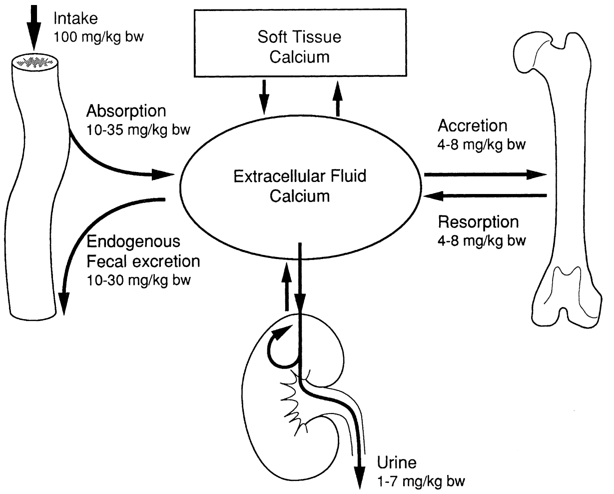

Fig. 6-2 Normal calcium balance showing the major organs that supply or remove calcium from extracellular fluid: bone, gut, and kidney. Total calcium input into extracellular fluid equals total calcium leaving the extracellular space. (Modified from Hazewinkel HAW: Dietary influences on calcium homeostasis and the skeleton. In *Purina international nutrition symposium,* Orlando, FL, Ralston Purina Company, Marriott World Center, January 15, 1991, p. 52.)

reabsorption inadequately maintain normal serum calcium concentrations. Bone calcium mobilization is important in the acute regulation of blood calcium.[395] Calcium and phosphorus can be mobilized from calcium phosphate in the bone ECF compartment, but these stores are rapidly depleted. The osteoblast is critical in limiting the distribution of calcium and phosphate between bone and ECF, and exchangeable bone water is separated from ECF water by the combined membranes of osteoblasts lining bone surfaces. For greater or prolonged release of calcium from bone, osteoclastic bone resorption must be activated. Osteoclasts secrete acid and proteases that result in dissolution of the mineralized matrix of bone and mobilize calcium and phosphorus.

Extracellular iCa concentration is the actively regulated fraction of total calcium (tCa).[76,111] When blood iCa concentration decreases, PTH secretion is stimulated. PTH exerts direct effects on bone and kidney and indirect effects on the intestine through calcitriol. PTH increases synthesis of calcitriol by activating renal mitochondrial 1α-hydroxylation of 25-hydroxycholecalciferol. Calcitriol increases calcium absorption from the intestine and acts with PTH to stimulate osteoclastic bone resorption.[99]

Calcitriol is necessary for differentiation of osteoclasts from precursor mononuclear cells. PTH increases osteoclast number and stimulates osteoclast function to increase bone resorption and the release of calcium from bone to blood. Calcitriol also induces renal transport mechanisms activated by PTH that increase tubular reabsorption of calcium from the glomerular filtrate, thus preventing calcium loss in urine.[366]

CALCIUM DISTRIBUTION WITHIN THE BODY

Approximately 99% of body calcium resides in the skeleton and is stored as hydroxyapatite, $Ca_{10}(PO_4)_6(OH)_2$. Most skeletal calcium is poorly exchangeable, and less than 1% is considered readily available. The small amount of rapidly exchangeable bone calcium arises from the ECF in bone that is present between osteoblasts and osteocytes and the bone matrix. Almost all of the nonskeletal calcium resides in the extracellular space, although small and biologically important quantities are found intracellularly.[448]

Extracellular Calcium

Calcium in plasma or serum exists in three fractions: ionized (iCa), complexed (bound to phosphate, bicarbonate, sulfate, citrate, and lactate), and protein bound (Fig. 6-3).

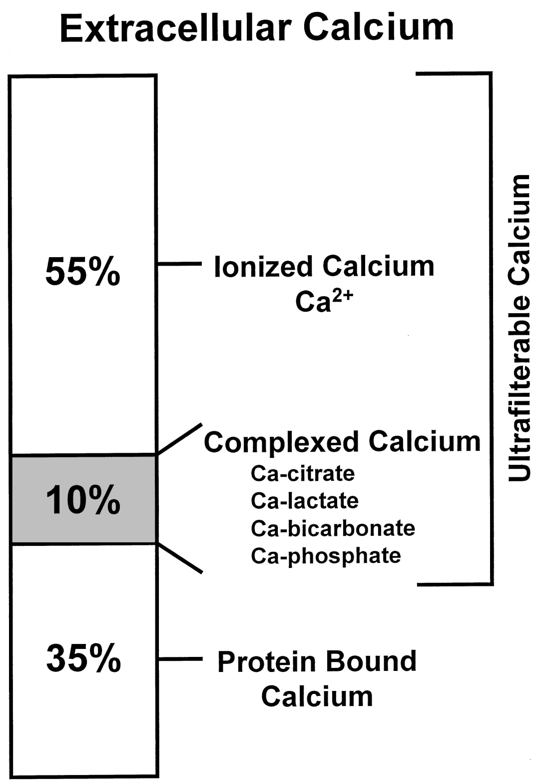

Fig. 6-3 Serum total calcium concentration consists of ionized (free), complexed, and protein-bound fractions.

In clinically normal dogs, protein-bound, complexed, and iCa account for approximately 34%, 10%, and 56% of serum tCa concentration, respectively.[472] Ionized calcium is the most important biologically active fraction in serum, although an active biologic role for complexed calcium has been suggested.[520] No biologic role for protein-bound calcium has been identified other than as a storage pool or buffering system for iCa.

Intracellular Calcium

Intracellular iCa is an important secondary messenger in the response to biochemical signals (such as hormones) transduced through the cell membrane.[420,448] Therefore intracellular iCa concentrations are maintained at a very low level (approximately 100 nM), 10,000-fold less than the serum concentration. This permits rapid diffusion into the cytoplasm from the ECF or endoplasmic reticulum. Intracellular calcium is rapidly buffered by cytosolic proteins and is transported into organelles or to the outside of the cell after an increase in intracellular iCa. If intracellular iCa is not maintained at a low concentration, it leads to toxicity and eventual cell death.

Most intracellular calcium is sequestered in organelles or bound to cellular membranes or proteins.[256] Sequestration of iCa in mitochondria blunts an increase in cytosolic iCa, whereas endoplasmic reticulum serves as a reservoir to increase cytosolic iCa when necessary. Binding of calcium to specific cytosolic or membrane proteins is an efficient method for regulation of intracellular iCa concentration. Protein binding provides intracellular iCa buffering and also may act as a messenger system when protein configuration and activity are altered. Calbindin, calmodulin, and troponin C are important intracellular calcium-binding proteins.[52]

Cell Membrane Calcium Ion Sensing Receptor. In 1993, a novel iCa-sensing receptor was cloned and sequenced.[73] The iCa receptor plays an integral role in iCa balance by regulating parathyroid chief cells, C cells, and renal epithelial cells.[72,235] In parathyroid chief cells and C cells, the iCa receptor directly regulates intracellular iCa concentration, which controls PTH and calcitonin secretion. Ionized magnesium (iMg) is also an agonist of the iCa receptor. Stimulation of the iCa receptor caused by increased extracellular iCa concentration in the kidney decreases NaCl, iCa, and iMg reabsorption in the proximal convoluted tubule and decreases water reabsorption in collecting ducts. This results in greater excretion of iCa and iMg in a more dilute urine.

Genetic diseases have been described related to both inactivating and activating mutations of the calcium receptor gene.[20] Inactivating mutations lead to severe neonatal hypercalcemia when homozygous and to familial hypocalciuric hypercalcemia when heterozygous.[512] Activating mutations of the calcium receptor produce hypoparathyroidism and hypocalcemia.[513] Autoantibodies produced against the calcium receptor may either disable it, producing hyperparathyroidism with hypercalcemia,[389,429] or activate it, producing hypoparathyroidism.[206,270] Drugs that bind the Ca^{2+}-sensing receptor may be useful to treat disorders of the parathyroid gland.

PARATHYROID HORMONE

STRUCTURE

PTH is an 84-amino acid single-chain polypeptide that is synthesized and secreted by chief cells of the parathyroid glands.[435] The amino acid sequence of PTH is known for the dog, cow, pig, rat, chicken, and human,[289,445] and most mammals appear to have very similar amino-terminal portions of the molecule.[366] Whereas the conserved amino end of PTH is vital for binding to cell membrane receptors, the role of the carboxyl terminus is to serve as a guide for PTH through the cellular secretory pathway.[303]

SYNTHESIS AND SECRETION

Synthesis, secretion, and degradation of PTH by chief cells are closely related. Little PTH is stored within the parathyroid glands,[216] and synthesis of new specific messenger RNA (mRNA) and translation to PTH are required to maintain secretion.[489] After secretion, PTH has a short half-life (3 to 5 minutes) in serum; thus a steady rate of secretion is necessary to maintain serum PTH concentrations. Circulating PTH has many forms, not all of which have bioactivity,[66,375] leading to potential confusion in assay interpretations.[464,510,569]

The amount of PTH available for secretion is a function of the balance of synthesis and degradation within chief cells (Fig. 6-4). Calcitriol, via the vitamin D receptor (VDR), and extracellular iCa concentration, via effects on the plasmalemmal calcium receptor,[103,104,427] control these parathyroid cell processes. Because calcitriol regulates expression of the calcium receptor gene,[94] calcitriol can be considered to exert overall control over PTH synthesis and secretion by the parathyroid cells. In general, the parathyroid gland has evolved most of its regulatory strategies to protect against hypocalcemia, with sensitive control of PTH synthesis and secretion being the dominant sites for regulation.[77,490] However, high serum iCa concentrations increase the rate of degradation of PTH within the gland to protect against hypercalcemia.[289]

Except for minor diurnal variation, PTH secretion is relatively constant but may have a mild pulsatile pattern in response to minor fluctuations in the concentration of serum iCa.[76] A relatively low rate of PTH secretion is needed normally to maintain serum iCa concentration. The basal secretory rate of PTH is approximately 25% of the maximal rate, and PTH is constantly secreted during normocalcemia. Complete inhibition of PTH secretion is not achieved even in the presence of severe hypercalcemia.[289]

Hypocalcemia is the principal stimulus for PTH secretion, but epinephrine, isoproterenol, dopamine, secretin, prostaglandin E_2, and stimulation of nerve endings within the parathyroid gland may have minor effects.[216] High concentrations of serum and intracellular iCa inhibit PTH secretion via increased arachidonic acid[57,96] and possibly subsequent eicosanoid production.[96] The control at PTH mRNA synthesis is also critically important.[489]

Calcitriol also plays an important role in the regulation of PTH synthesis and secretion.[492] Calcitriol inhibits PTH mRNA synthesis[491] and stimulates synthesis of the calcium receptor.[94] These relationships explain the requirement for adequate blood concentrations of calcitriol to maintain the ability of the parathyroid gland to respond to changes in extracellular calcium concentrations.[323,367] Increased intracellular iCa may also cooperate with calcitriol to reduce PTH synthesis in chief cells by inhibiting the expression of calreticulin (a blocker of VDR action).[481,548] Animals with uremia and reduced serum calcitriol concentrations have poorly regulated chief cell function that results in renal secondary hyperparathyroidism,[204,363] but a significant part of the hyperparathyroid response in uremic patients is the result of a glandular hyperplasia caused by the changes of both calcitriol and serum phosphorus.[8] Serum phosphorus concentrations are generally considered to regulate PTH secretion principally by indirect means. Renal calcitriol synthesis is reduced early in uremia by modest hyperphosphatemia, and the plasma iCa concentration may decrease because of reduced effects of calcitriol on the intestine, bone, and kidney. Markedly increased serum phosphorus concentrations (as seen in advanced renal failure) can lower the serum iCa concentration (mass law effect), resulting in an increase in PTH secretion because of the lowered calcium, but these effects do not occur early in renal failure when serum phosphorus is only moderately increased.[363]

Serum magnesium concentration has little role in the control of PTH secretion under normal conditions, but PTH secretion can be inhibited by very high concentrations of serum iMg.[435] Paradoxically, hypomagnesemia or magnesium depletion also results in inability to secrete PTH, but the cellular mechanism of this effect is unclear. This effect may be partially caused by reduced sensitivity of cell membrane receptors to iCa in the presence of low serum iMg concentrations.[216,347]

Set-point for PTH Secretion

The **set-point** for PTH secretion is defined as the ECF iCa concentration that occurs at the serum PTH concentration that is midway between maximal and minimal values of PTH obtained experimentally.[76] Normal serum iCa concentration is maintained slightly higher than the set-point; thus PTH release normally is less than half-maximal (Fig. 6-5).

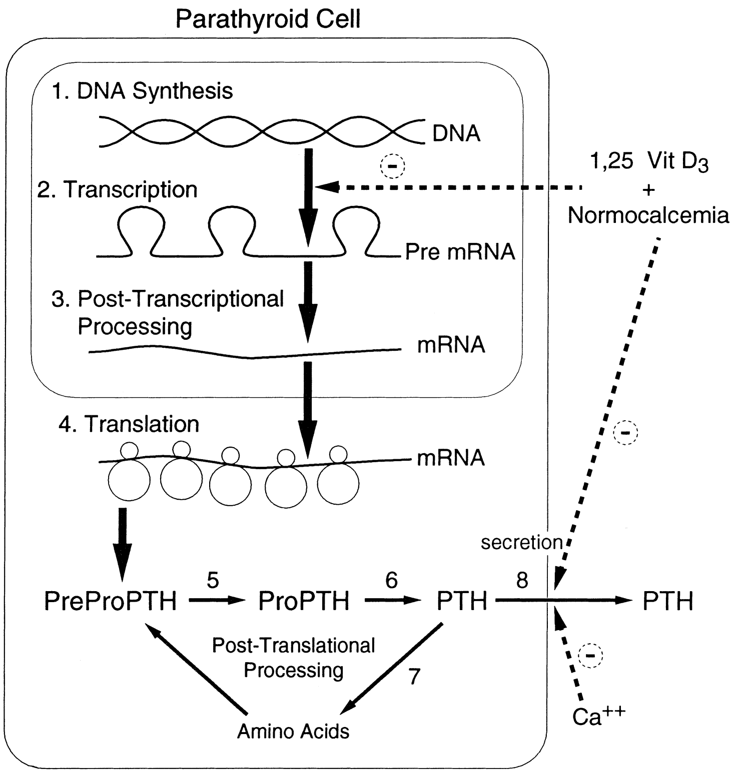

Fig. 6-4 Synthesis and secretion of parathyroid hormone. Note sites of regulation of PTH biosynthesis by extracellular ionized calcium or calcitriol (1,25-$[OH]_2$-vitamin D_3) interaction. (Modified from Habner JF, Rosenblatt M, Potts JT: Parathyroid hormone: biochemical aspects of biosynthesis, secretion, action, and metabolism, *Physiol Rev* 64:1004, 1984.)

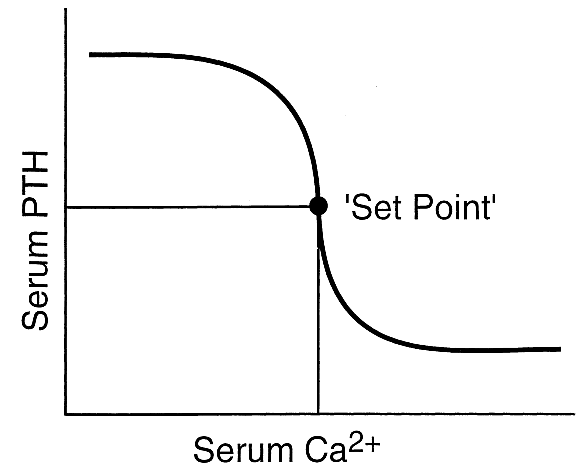

Fig. 6-5 Relationship between secretion rate of parathyroid hormone and plasma calcium concentration. Small changes in plasma calcium concentration cause large changes in parathyroid hormone secretion, but secretion is not completely suppressed by high plasma calcium concentrations.

The rate of PTH secretion is inversely proportional to the concentration of extracellular calcium, but this proportional secretion of PTH occurs only over a narrow range corresponding to a serum tCa concentration of 7.5 to 11.0 mg/dL.[216] An inverse sigmoidal curve with a steep slope results when the relationship between serum iCa concentration and PTH secretion is plotted over a larger range of calcium concentrations (see Fig. 6-5).[76] This ensures large changes in PTH secretion for relatively small changes in iCa concentration in the physiologic range and precise control of serum iCa concentration. An approximately 10% decrease in serum iCa concentration elicits a nearly maximal PTH secretory response. The rate of decrease of serum iCa concentration is also important, and rapid decreases in serum iCa result in larger increases in PTH secretion. A 2% to 3% decrease in iCa concentration, if rapid in onset, may result in a 400% increase in PTH secretion.[76]

The cell membrane calcium receptor is responsible for establishing the relationship of the set-point for PTH secretion and extracellular iCa concentration.[546]

The calcium receptor regulates PTH secretion indirectly by controlling the intracellular iCa concentration by means of (1) release of iCa from intracellular stores, and (2) cell membrane calcium channels. Calcium channels span the parathyroid chief cell membrane and are important in allowing extracellular iCa access to the interior of the cell.[176] The calcium channels are controlled by intracellular iCa concentration[77] and membrane regulatory G proteins, which interact with the cell membrane calcium receptor.[21]

Calcitriol plays an important role in controlling the parathyroid gland set-point by regulating (1) synthesis of the cell membrane calcium receptor,[71,94] (2) synthesis of cell membrane G proteins, and (3) function of cell membrane calcium channels.[366] Therefore adequate calcitriol is necessary to maintain the set-point for PTH secretion. The regulation of calcium receptor expression by calcitriol explains the observed "calcium set point" aberrations in control of PTH secretion in those with uremia.[329] These patients have deficits in calcitriol production,[112,563] as well as resistance in uremic parathyroids to calcitriol[151,396]; thus they are less able to induce synthesis of adequate numbers of calcium receptors.

Although regulations at each parathyroid cell may fail, thus producing abnormally increased PTH,[201,419] changes may also be seen in the maximal secretory capacity dependent mostly on parathyroid cell numbers.[462] It is likely that increased PTH secretion in patients with renal secondary hyperparathyroidism is primarily caused by parathyroid gland hyperplasia.[148] One important role of calcitriol therapy in these patients is to prevent or reverse the parathyroid cellular hyperplasia.[95,147,365]

Inhibition of PTH Synthesis and Secretion

This topic has become important with understanding of the toxicity of PTH in animals and humans with chronic renal failure (CRF) and accompanying secondary hyperparathyroidism.[10,330,363,398] Recently, increased awareness of PTH toxicity stems from established relations to cardiovascular disease[128] and mortality.[499] PTH secretion is inhibited by increased serum iCa concentration,[489,491] and the initial effect to decrease PTH secretion is rapid (occurring within 2 to 3 minutes), mediated by the calcium receptor with a cascade of resulting intracellular events[62,129,235] and involving mediation by arachidonate.[7] Slower effects are caused by inhibition of synthesis of PTH mRNA and its translation to hormone (Fig. 6-6).[489]

Calcitriol is an important inhibitor of PTH synthesis, and it completes a negative feedback loop from the kidney because PTH stimulates renal calcitriol synthesis. Short and long negative feedback loops complement each other to control normal secretion of PTH.[289] The long negative feedback loop is completed when an increased serum iCa concentration results from PTH stimulation of renal calcitriol production and subsequent enhanced gastrointestinal absorption of calcium. This effect takes hours to develop because calcium-binding proteins associated with calcium absorption must be induced in enterocytes.[67,549] The short negative feedback loop is mediated by the binding of calcitriol to VDRs in parathyroid cells, with inhibition of transcription of the PTH gene.[489] The calcitriol receptor (VDR) is expressed in parathyroid chief cells at concentrations equal to those in intestinal epithelial cells that regulate calcium absorption in the gastrointestinal tract. The VDR was found to be depleted in the parathyroid glands of dogs and humans with uremia because of lack of renal production of calcitriol.[70] After the VDR binds calcitriol, the VDR-calcitriol complex acts in the nucleus of the parathyroid chief cells by binding to specific regions of the PTH gene called vitamin D response elements (VDREs) and inhibiting transcription of the PTH gene (see Fig. 6-6).[289,363] For calcitriol to suppress synthesis of PTH, a normal concentration of iCa must be present because it would be inappropriate to suppress PTH synthesis in a hypocalcemic patient.

CLEARANCE AND METABOLISM OF PARATHYROID HORMONE

The intact PTH molecule (84 amino acids) circulates in the bloodstream with a half-life of 3 to 5 minutes and is removed by fixed macrophages.[289,435] A significant amount of cleavage is close to the amino terminus of the PTH molecule. Regardless of where the endopeptidase cleavage occurs, the amino-terminal portion of PTH is completely degraded within the phagocytes. Kidney and bone also participate in destruction of intact PTH.

Fragments of PTH are filtered by the glomeruli. This mechanism of excretion is most important for the excretion of the carboxyl-terminal PTH fragments because carboxyl-terminal PTH (released from either the parathyroid gland or Kupffer cells) is cleared only by glomerular filtration (Fig. 6-7). The carboxyl-terminal fragments of PTH are not important for calcium metabolism. The circulating half-life of carboxyl-terminal PTH is much longer than that of intact PTH, and serum concentrations of carboxyl-terminal PTH can be very high during primary or secondary hyperparathyroidism and can be nonspecifically increased during renal failure.

ACTIONS OF PARATHYROID HORMONE

PTH is the principal hormone involved in the minute-to-minute fine regulation of blood calcium concentration. It exerts its biologic actions directly by influencing the function of target cells primarily in bone and kidney and indirectly in the intestine to maintain plasma calcium at a concentration sufficient to ensure the optimal functioning of a wide variety of body cells.

In general, the most important biologic effects of PTH on calcium are to (1) increase the blood calcium

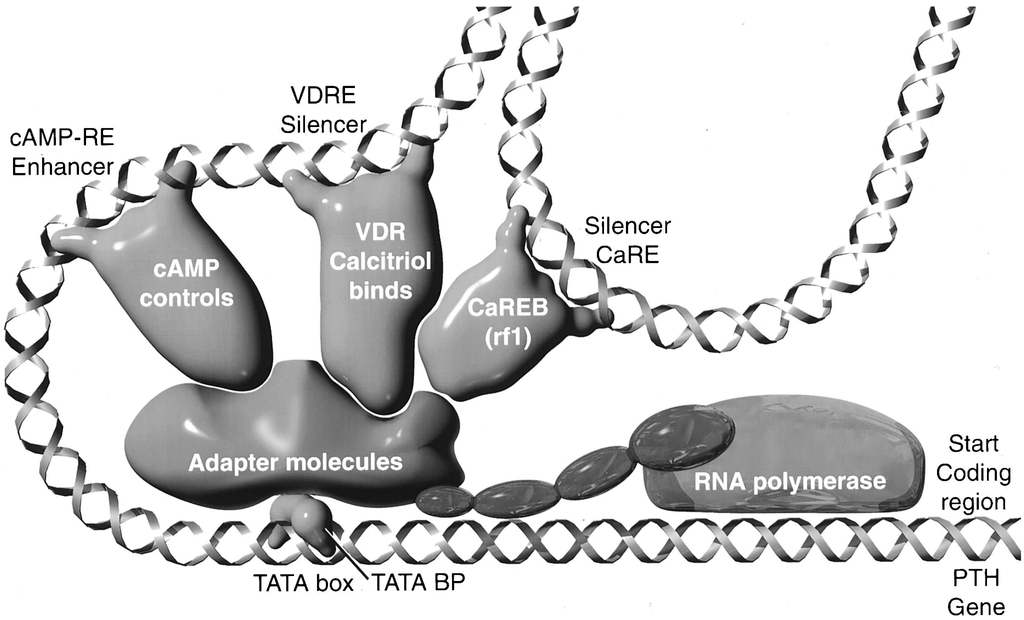

Fig. 6-6 Simplified depiction of events regulating transcription of the parathyroid hormone (PTH) gene by RNA polymerase. Only the three transcription factors best understood to interact in this regulation are shown. Cyclic AMP (cAMP) stimulates phosphorylation of a transcription factor that binds to a cAMP response element (cAMP-RE) on the gene and enhances transcription. In contrast, the vitamin D receptor (VDR)–calcitriol complex and calcium response element–binding protein (CaREB, rf1) bind to their respective vitamin D (VDRE) and calcium (CaRE) response elements of the PTH gene, which function as "silencers" or negative regulators of gene transcription. Note that for calcium to exert its negative effect by means of the CaREB transcription factor, calcitriol and the vitamin D receptor must also be present. The adapter molecules (shown as a single structure) diagrammatically represent about 30 proteins termed accessory transcription factors. The TATA box is part of the gene promoter to which the TATA box binding proteins (BPs) bind. (From Nagode LA, Chew DJ, Podell M: Benefits of calcitriol therapy and serum phosphorus control in dogs and cats with chronic renal failure, *Vet Clin North Am Small Anim Pract* 26:1293-1330, 1996.)

concentration; (2) increase tubular reabsorption of calcium, resulting in decreased calcium loss in the urine; (3) increase bone resorption and the numbers of osteoclasts on bone surfaces; and (4) accelerate the formation of the principal active vitamin D metabolite (1,25-dihydroxyvitamin D, or calcitriol) by the kidney through a trophic effect to both induce synthesis of and activate the 1α-hydroxylase in mitochondria of renal epithelial cells in the proximal convoluted tubules.

An important action of PTH on bone is to mobilize calcium from skeletal reserves into ECF.[97] The increase in blood calcium concentration results from an interaction of PTH with receptors on osteoblasts that stimulate increased calcium release from bone and direct an increase in osteoclastic bone resorption.[356]

The response of bone to PTH is biphasic. The immediate effects are the result of increasing the activity of existing bone cells. This rapid effect of PTH depends on the continuous presence of hormone and results in an increased flow of calcium from deep in bone to bone surfaces through the action of an osteocyte-osteoblast "pump" to make fine adjustments in the blood calcium concentration.[395] The later effects of PTH on bone are potentially of greater magnitude and are not dependent on the continuous presence of hormone. Osteoclasts are primarily responsible for the long-term action of PTH on increasing bone resorption and overall bone remodeling.[97,356]

PTH also has the potential to serve as an anabolic agent in bone and stimulate osteoblastic bone formation.[190,505] Intermittent administration of exogenous 1-34 PTH has been reported to increase bone mass in humans and animals.[507]

The ability of PTH to enhance the renal reabsorption of calcium is of considerable importance. This effect of PTH on tubular reabsorption of calcium is caused by, in part, a direct action on the distal convoluted tubule.[573] PTH may also increase calcium reabsorption in the ascending thick limb of Henle's loop indirectly by increasing the net positive charge in the nephron

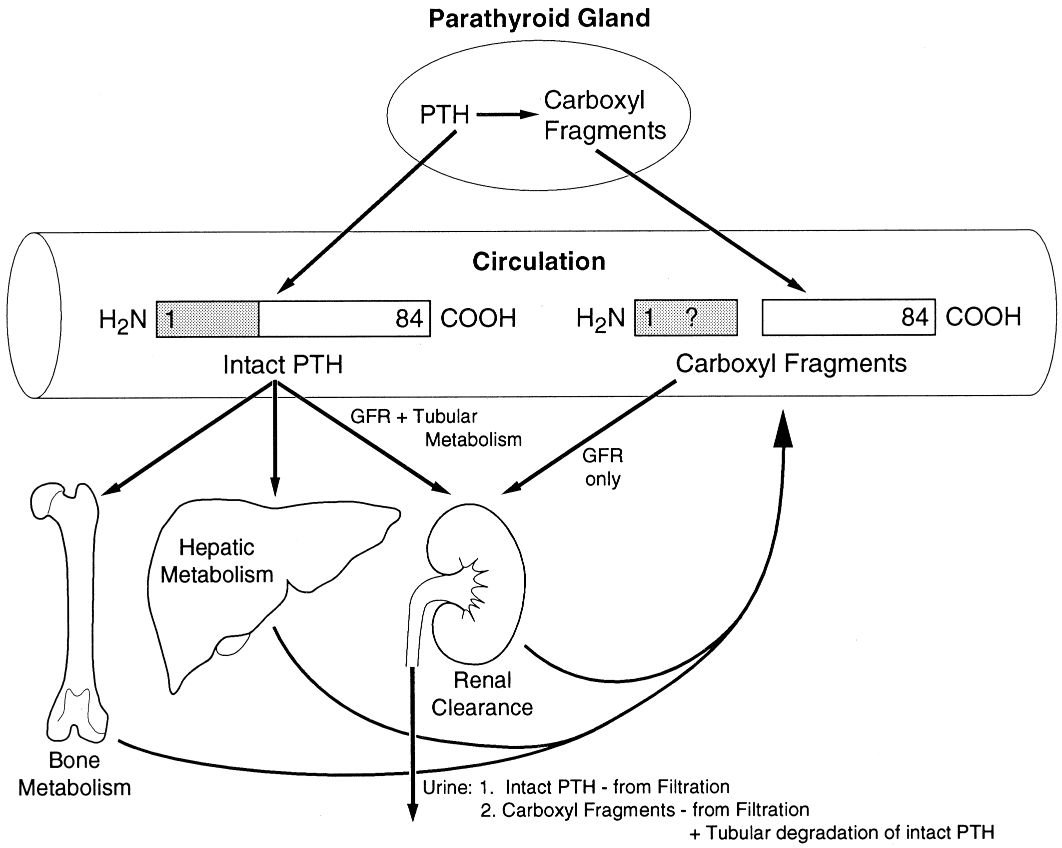

Fig. 6-7 Degradation and clearance of parathyroid hormone (PTH). PTH (1-84) is secreted intact from the parathyroid gland into the circulation. Biologically inactive carboxy-terminal (COOH) fragments of PTH are also secreted by the parathyroid gland, but amino-terminal PTH is not secreted and does not circulate in biologically relevant concentrations. Peripheral metabolism of intact PTH to carboxy-terminal PTH fragments occurs mostly in the liver but may also occur in the kidney and bone. Both intact PTH and carboxy-terminal PTH are cleared by glomerular filtration, but only intact PTH is metabolized in the liver, kidney, and bone. The half-life of intact PTH in vivo is short compared with that of the carboxy-terminal fragments of PTH. (Modified from Endres DB, Villaneuva R, Sharp CF, et al: Measurement of parathyroid hormone, *Endocrinol Metab Clin North Am* 18:614, 1989.)

lumen and creating a stimulus for diffusion out of the lumen. PTH also regulates the conversion of 25-hydroxycholecalciferol to calcitriol and other metabolites of vitamin D.

Parathyroid Hormone C-Terminal 7-84 as PTH Antagonist

It was originally thought that PTH 35-84 and other fragments cleaved between residues 24 and 43 dominated the carboxyl-terminal fragments of PTH secreted by chief cells. The C-terminal fragments can be measured using C-terminal-specific immunoassays. The function of PTH 35-84 and its receptor is unknown, but it may regulate bone cell function. The larger C-terminal fragment, PTH 7-84,[259] may be significantly increased in renal secondary hyperparathyroidism[351] and can antagonize the effects of PTH 1-84 in vivo.[297] The antagonistic action of PTH 7-84 is likely attributable to binding to an alternate PTH receptor and not to the PTH1 receptor that is used by PTH 1-34 and PTH 1-84.[139,376]

Parathyroid Hormone Receptor

The receptor for N-terminal PTH (amino acids 1 to 34), the region important in calcium regulation, has been cloned and sequenced in humans, dogs, and other species.[1,378,496] It is a seven-transmembrane domain receptor that is expressed in renal epithelial cells, osteoblasts, and some other cells. The N-terminal regions of PTH and PTHrP bind this receptor with equal affinity. The PTH receptor is also located on many cell types, such as dermal fibroblasts, that are not associated with the action of PTH. It is assumed that the receptor functions as the binding protein for PTHrP in these tissues. The currently used terminology for this receptor is the PTH1 receptor, but it is often described as the PTH/PTHrP receptor. The PTH2 receptor is present in the brain and binds to both PTH and tuberoinfundibular peptide but not to PTHrP.[233]

PARATHYROID HORMONE–RELATED PROTEIN: A POLYHORMONE

PTHrP is not strictly a calcium-regulating hormone, but it was identified in 1982 as an important PTH-like factor that plays a central role in the pathogenesis of humoral hypercalcemia of malignancy (HHM).[437] PTHrP is produced widely in the body and has numerous actions in the developing fetus and adult animal independent of its role in cancer-associated hypercalcemia.[411] This is in contrast to PTH, which is produced by the parathyroid glands and functions principally in regulation of calcium balance. PTHrP has multiple actions that are specific to the N-terminal, midregion, and C-terminal regions of the protein, making PTHrP a true polyhormone.

Some of the actions of PTHrP involve normal regulation of calcium metabolism.[448] For example, PTHrP functions as a calcium-regulating hormone in the fetus and is produced by the fetal placenta.[317] In the adult, PTHrP circulates in the blood in low concentrations (<1 pM) but is produced by many different tissues and functions principally as an autocrine, paracrine, or intracrine cellular regulator. PTHrP is produced by the lactating mammary gland and is secreted into milk. Mammary gland production of PTHrP likely facilitates mobilization of calcium from maternal bones and may play a role in the transport of calcium into milk during lactation.[571,572] PTHrP acts as an abnormal systemic calcium-regulating hormone and mimics the actions of PTH in patients with HHM. PTHrP not only plays a major role in most forms of HHM but also has been demonstrated in many normal tissues, including epithelial cells of the skin and other organs; endocrine glands; smooth, skeletal, and cardiac muscle; lactating mammary gland; placenta; fetal parathyroid glands; bone; brain; and lymphocytes.[411,435] Therefore PTHrP functions as (1) a hormone in an endocrine manner in the fetus and lactating dams, (2) a paracrine factor in many fetal and adult tissues, and (3) an abnormal hormone in an endocrine manner in adults with HHM (Fig. 6-8). PTHrP is necessary for normal endochondral bone formation in the fetus and neonate. Knockout of the PTHrP gene results in short-limb dwarfism and death at birth as a result of a failure of cartilage proliferation at the growth plates and premature ossification.[265]

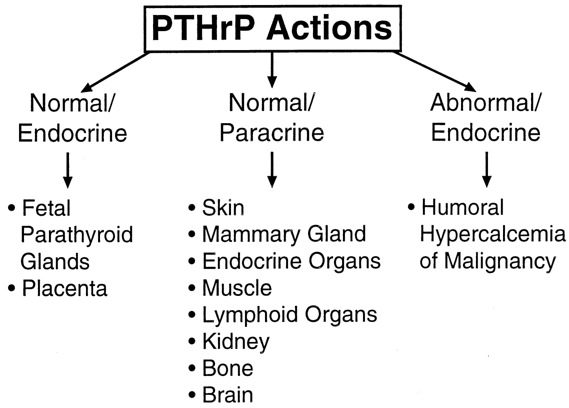

Fig. 6-8 Actions of parathyroid hormone–related protein (PTHrP).

PTHrP is a 139- to 173-amino acid peptide originally isolated from human and animal tumors associated with HHM.[437] PTHrP shares 70% sequence homology with PTH in its first 13 amino acids. The N-terminal region of PTHrP (amino acids 1 to 34) binds and stimulates PTH receptors in bone and kidney cells with affinity equal to that of PTH, so that PTHrP functions similarly to PTH in patients with HHM.[119,385] The midregion of PTHrP is responsible for stimulating iCa uptake by the fetal placenta,[317]and the C-terminal region can inhibit osteoclastic bone resorption.[171]

The complementary DNA (cDNA) for canine and feline PTHrP has been cloned and sequenced.[449,508] The sequence of canine PTHrP cDNA and gene indicated that the dog PTHrP gene is more closely related to the human PTHrP gene than are the PTHrP genes in rats, mice, and chickens.[212] The deduced amino acid sequence of the N-terminal region (amino acids 1 to 36) is identical in five mammalian species (dog, cat, human, rat, and mouse), and there is a high degree of homology of the midregion of PTHrP in these species.[324,449,504,508,575] The high degree of interspecies homology indicates the importance of the N terminus and midregion in the function of PTHrP.

There is less homology of the C-terminal region of canine PTHrP with that from other species. The function of the C-terminal region is unknown. PTHrP (107 to 111) and PTHrP (107 to 139) may inhibit osteoclastic bone resorption.[172,501] Increased urine concentrations of C-terminal PTHrP have been demonstrated in humans and mice with cancer-associated hypercalcemia[255,266] and in patients with renal failure.[84] Increased C-terminal PTH is also seen in the serum of patients with renal failure and indicates that the kidney is an important site of excretion of C-terminal PTHrP.

C-terminal PTHrP may have a longer serum half-life than N-terminal or midregion PTHrP.

PARATHYROID HORMONE–RELATED PROTEIN IN THE FETUS

Fetuses maintain higher concentrations of serum iCa than their dams. Fetal parathyroid glands produce low levels of PTH,[100] and PTHrP functions to maintain iCa balance in the fetus.[316,317] PTHrP is secreted by fetal parathyroid chief cells, and PTHrP is produced by the placenta, which is necessary for iCa uptake by the fetus.[571] The midregion of PTHrP is the most active portion that stimulates iCa and iMg transport by the placenta. The placenta expresses the iCa-sensing receptor, which may contribute to the regulation of placental calcium transport.[285] PTHrP is also produced by the uterus, where it is important in permitting relaxation of the smooth muscle of the muscularis as the fetuses grow.[514]

VITAMIN D

Vitamin D (calciferol) is classified as a secosteroid hormone.[243] In tetrapods, the role of vitamin D via the calcitriol-activated VDR has evolved into one dominated by calcium regulatory mechanisms, but the roles in primitive species, including regulation of detoxification enzymes, have commonly been retained in more evolved life forms.[544,559] These pleiotropic actions of vitamin D[304] include, among others, important roles as antiproliferative and prodifferentiative mediators[22] working in part via control of DNA replication[155] and roles as immunomodulators,[222] including effects on glomerulonephritis[393] and encephalitis.[193] A role of calcitriol to regulate expression of the insulin receptor has been described,[319] as has a role in muscle.[132] Of particular interest in uremic patients is the calcitriol increase of erythroid proliferation via burst-forming units.[18] These pleiotropic effects of calcitriol can be related to important clinical applications in patients with renal or other metabolic disease.[236] They may explain the clinical improvements noticed in dog and cat uremic patients treated preventatively with low doses of calcitriol[363] that were accomplished when calcitriol was used before any PTH elevation had occurred.

VITAMIN D METABOLISM

The cholecalciferol (parent vitamin D_3 of animal origin) metabolites 25-hydroxyvitamin D_3 (calcidiol), 1,25-dihydroxyvitamin D_3 (calcitriol), and 24,25-dihydroxyvitamin D_3 are the most important of at least 30 metabolites. In domestic mammals, the same three metabolites derived from vitamin D_2 (ergocalciferol of plant origin) are equally bioactive; thus generic use of the terms 1,25-dihydroxyvitamin D and calcitriol is assumed to include metabolites of vitamin D_3 or D_2 derived from animal or plant origin, respectively. The 25-hydroxyvitamin D that is produced in liver is the major circulating form of vitamin D[197] and serves as a pool for further activation by 1α-hydroxylation or catabolism by 24-hydroxylation.[227,383] Only 25-hydroxylation and 1α-hydroxylation are important in the function of vitamin D.[131]

Synthesis

In humans, the requirement for vitamin D can be met by consumption of vitamin D_2 or D_3 or by synthesis of vitamin D_3 (cholecalciferol) in the skin. Cholecalciferol is synthesized in the skin from 7-dehydrocholesterol after exposure to ultraviolet light. 7-Dehydrocholesterol forms previtamin D_3 in the presence of ultraviolet B light at 288 nm, followed by further thermal conversion from previtamin D_3 to vitamin D_3.[237] Dogs and cats inefficiently photosynthesize vitamin D in their skin and consequently are dependent on vitamin D in their diet.[245] Vitamin D ingested in the diet is absorbed intact from the intestine.

Vitamin D–binding protein transports vitamin D to the liver and other target sites (Fig. 6-9).[124] Hydroxylation of vitamin D occurs in the liver to produce 25-hydroxyvitamin D (calcidiol). The 25-hydroxylase activity is not influenced by calcium or phosphorus.[197] Calcidiol does not have any known action in normal animals,[131] but during vitamin D intoxication, high levels of calcidiol are produced by the liver and can induce hypercalcemia.

The most important step in bioactivation of vitamin D occurs as 25-hydroxyvitamin D is further hydroxylated to calcitriol in the proximal tubule of the kidney.[227] This reaction is tightly regulated by ionic and hormonal control mechanisms that modulate the activity of the hydroxylase enzyme systems (Fig. 6-10). The two principal enzyme systems involved are 25-hydroxyvitamin D-1α-hydroxylase (resulting in active calcitriol formation) and 25-hydroxyvitamin D-24*R*-hydroxylase (the first step of catabolism to inactive vitamin D metabolites). The activities of these enzymes are reciprocally regulated.[383]

The 1α-hydroxylase enzyme activity is localized within mitochondria of the convoluted tubules and portions of the straight proximal tubule of the kidney. Little extrarenal 1α-hydroxylation of 25-hydroxyvitamin D occurs in other tissues except in human and rat placenta and skin and in some lymphoproliferative disorders.[4,150] The 24-hydroxylation can also metabolize calcitriol, generating 1,24,25-trihydroxyvitamin D as the first step in the major catabolic pathway of calcitriol to biologically inactive calcitroic acid.[243] Inactive vitamin D catabolites are excreted through the bile into feces, which is the only important excretory route; less than 4% is excreted into urine.[131]

Stimulation of Calcitriol Synthesis. Serum PTH, calcitriol, phosphorus, and calcium concentrations are

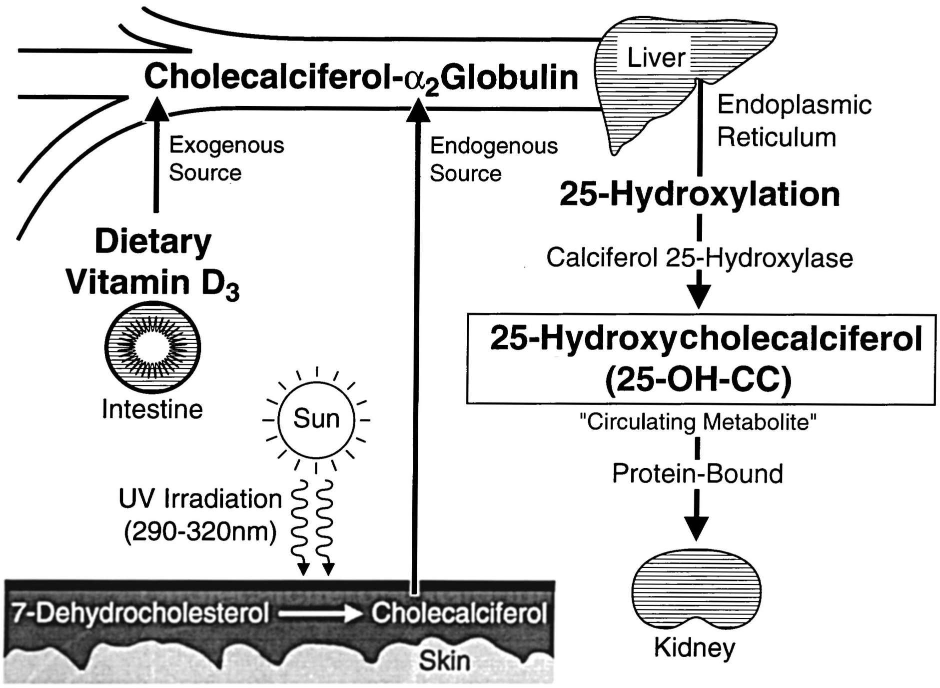

Fig. 6-9 Metabolism of vitamin D. The initial step of metabolic activation of vitamin D_3 from endogenous (photoactivation) and dietary sources is in the liver to form 25-hydroxycholecalciferol (25-hydroxyvitamin D_3). Photoactivation is poor in dogs and cats; consequently, they depend on dietary sources of vitamin D_3.

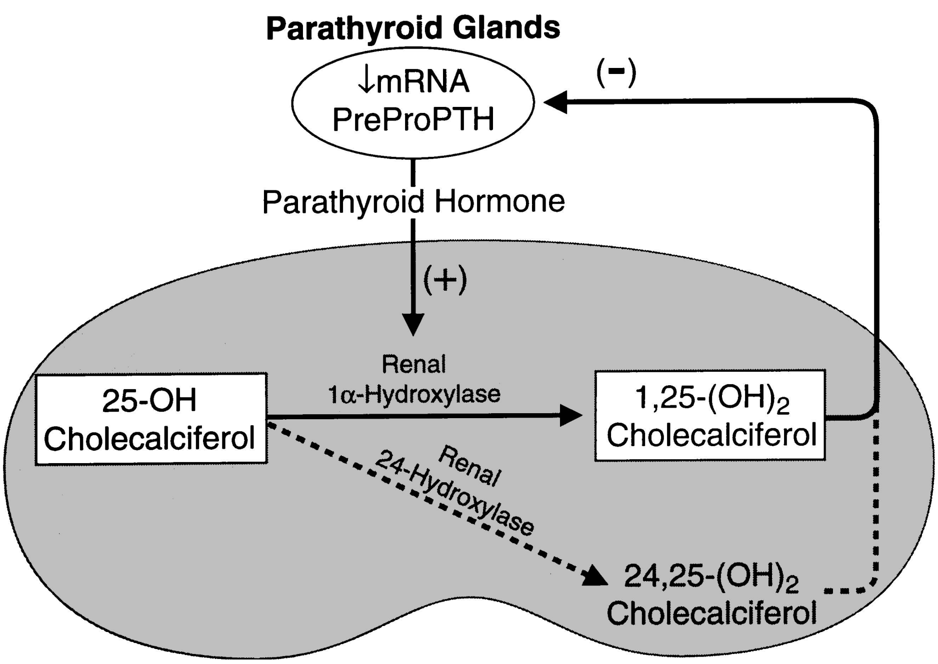

Fig. 6-10 Parathyroid hormone increases renal synthesis of 1,25-dihydroxycholecalciferol (calcitriol) by stimulating the 1α-hydroxylase activity in renal epithelial cells that converts 25-hydroxycholecalciferol to 1,25-dihydroxycholecalciferol. Negative feedback is exerted by 1,25-dihydroxycholecalciferol (calcitriol) on parathyroid chief cells to decrease the rate of PTH synthesis and secretion, which in turn decreases the rate of formation of 1,25-dihydroxycholecalciferol. Calcitriol also directly suppresses synthesis of the renal 1α-hydroxylase enzyme.

the principal regulators for renal calcitriol synthesis.[227] Chronic changes in serum calcium concentration regulate the synthesis of calcitriol, and these calcium changes can override signals from serum phosphorus and PTH concentrations.[248] Deficiencies of phosphorus, calcium, and calcitriol lead to increased calcitriol formation.[364] Low calcium or calcitriol concentrations lead to increased serum PTH concentrations. In the kidney, PTH mediates dephosphorylation of renal ferredoxin (renoredoxin) and results in increased synthesis of calcitriol.[199,487] Renoredoxin is the regulatory constituent of the 1α-hydroxylase enzyme system and is inhibited by phosphorylation in the presence of high concentrations of phosphorus or calcium in the renal tubule.[227] PTH not only activates the renal 1α-hydroxylase but also induces synthesis of the enzyme from the renal gene encoding it.[149,150]

Several drugs and hormones have effects on vitamin D metabolism, some of which are stimulatory.[60] Hypocalcemia and calcitonin directly stimulate 1α-hydroxylation independent of PTH.[65] Estrogens increase calcitriol synthesis after up-regulation of PTH receptors in the kidney,[65] and testosterone may also increase calcitriol synthesis.[582] Reduced dietary calcium intake can lead to stimulation of renal 1α-hydroxylase in the absence of detectable hypocalcemia.[582]

Inhibition of Calcitriol Synthesis. Calcitriol synthesis is inhibited by calcitriol, hypercalcemia, and phosphate loading.[65,227] Calcium directly and indirectly inhibits calcitriol synthesis.[166] The indirect action is caused by inhibition of PTH synthesis and secretion, thus removing the stimulus provided by PTH. The inhibitory effects of chronic hypercalcemia can override the stimulatory effects of increased PTH concentrations in calcitriol production, as may occur in primary hyperparathyroidism.[248] The inhibitory effects of high concentrations of phosphorus on calcitriol synthesis are important and affect the activity of existing enzyme molecules.[363,364]

Actions of Calcitriol

Calcitriol is the only natural form of vitamin D with significant biologic activity.[131,424] It is approximately 1000 times as effective as parent vitamin D and 500 times as effective as its precursor calcidiol (25-hydroxyvitamin D) in binding to the natural calcitriol receptor (VDR) in target cells.[366] Calcitriol increases serum calcium and phosphorus concentrations, and its major target organ for these effects is the intestine.[67] However, there is also an important contribution from bone,[502] and calcitriol stimulates the kidney to reabsorb both calcium and phosphorus from the glomerular filtrate. Calcitriol has multiple indirect effects on calcium balance, including up-regulation of calcitriol receptors in patients with uremia, regulation of PTH synthesis and secretion by the parathyroid gland,[577] and prevention or reversal of parathyroid gland hyperplasia in the uremic patient.[191,363]

THE CALCITRIOL RECEPTOR

The VDR for calcitriol is present in many tissues in addition to bone, kidney, intestine, and parathyroid gland.[221] The importance of calcitriol in a tissue is proportional to the abundance of the VDR in the cells, and this is highly regulated.[287] Intestinal epithelial cells and parathyroid gland chief cells have equal and high concentrations of VDR. VDR genetic polymorphisms are thought to generate variation of efficiency of the VDR.[79,105] Calcitriol initially dissociates from its serum binding protein, diffuses across the cell membrane, and binds with its receptor.

Effects of Calcitriol on the Intestine

Calcitriol enhances the transport of calcium and phosphate from the intestinal lumen to plasma across the enterocyte.[68,549] Energy in the form of adenosine triphosphate (ATP) is required to transport calcium from the enterocytes into the blood and to absorb phosphate from the intestinal lumen. Calcitriol induces synthesis of the plasma membrane calcium pump (ATPase) that removes calcium from the enterocytes[394] and the Na^+-phosphate cotransport protein that transports phosphorus into the enterocyte. In addition, calcitriol increases the brush border permeability to calcium and induces the synthesis of calbindin-D 9k.[120,516] Calbindins serve as buffers to protect enterocytes from toxic concentrations of calcium ion while ferrying calcium across the cell.[549] Calcitriol also directly stimulates rapid calcium transport (transcaltachia) across the enterocyte.[380] Normal dogs have a progressive decrease in the number of calcitriol receptors and calbindin concentrations that regulate the efficiency of calcium absorption in enterocytes from the duodenum to the ileum.[283] Longer transit times in certain portions of the intestinal tract (e.g., ileum) can still lead to significant calcium absorption despite low transport efficiency.[549]

Effects of Calcitriol on Bone

Calcitriol is necessary for bone formation and mineralization because it ensures an adequate source of calcium and phosphorus from the intestinal tract. Deficiencies in vitamin D lead to impaired bone growth, such as rickets in growing animals and osteomalacia in adults.[435] Calcitriol is necessary for normal bone development and growth because it regulates the production of multiple bone proteins produced by osteoblasts, including alkaline phosphatase (ALP), collagen type I, osteocalcin, and osteopontin.[17,497] Calcitriol is also necessary for normal bone resorption because it promotes differentiation of monocytic hematopoietic precursors in the bone marrow into osteoclasts.[502] This relationship between calcitriol and osteoclasts explains the dependence of PTH on calcitriol for optimal bone resorption.[365]

Effects of Calcitriol on the Kidney

An important effect of calcitriol in the kidney is direct inhibition of 25-hydroxyvitamin D–1α-hydroxylase in the renal tubule, preventing overproduction of calcitriol.[424] In addition, calcitriol facilitates calcium and phosphorus reabsorption from the glomerular filtrate.[294] Calcitriol is necessary to work with PTH to reabsorb urinary calcium into blood. Glomerular podocytes contain the VDR for calcitriol and respond to low doses of calcitriol with decreased injury and loss of podocytes.[292] In glomerulonephritis, low doses of calcitriol decreased mesangial proliferative nephritis, which involved calcitriol abrogation of inflammatory mediators interleukin (IL)-1α, tumor necrosis factor-α (TNF-α), and IL-6 in the mesangium.[392] Although calcitriol has generally been thought to protect the kidneys during CRF by preventing the damage from excess PTH,[365,577] it is becoming clear that calcitriol has direct beneficial effects on the diseased kidney as well.

Effects of Calcitriol on the Parathyroid Gland

Calcitriol inhibits the production of PTH in the parathyroid gland by direct and indirect means.[486,491] Binding of calcitriol to its receptor in parathyroid chief cells directly inhibits PTH synthesis. Second, calcitriol stimulates intestinal calcium absorption, which indirectly reduces PTH secretion by increasing serum iCa concentration. Calcitriol suppression of PTH synthesis is dose dependent and occurs before serum iCa concentration is increased by the delayed effects of calcitriol on intestinal calcium transport.[494] Calcitriol may be considered the primary controlling factor for transcription of the PTH gene and subsequent synthesis of PTH because suppression of PTH synthesis cannot occur in the absence of calcitriol even in the presence of hypercalcemia (see Fig. 6-6).[364,491] PTH secretion decreases 12 to 24 hours after exposure to calcitriol. Whereas PTH stimulates renal calcitriol synthesis, calcitriol is a negative regulator of PTH. Long-standing calcitriol deficiency results in chief cell hypertrophy and hyperplasia, demonstrating that calcitriol is important in limiting cellular proliferation in the parathyroid gland.[491] Calcitriol treatment of uremia in dogs and humans has resulted in regression of parathyroid gland hyperplasia.[191,366] Calcitriol can be used in a preventative manner to avoid development of hyperparathyroidism in dogs and cats with early stages of CRF.[363] This has proven to be highly successful and is consistent with developing thinking in the human medical profession.[583]

Calcitriol in Therapy of Cancer

Many studies focus on the benefits of calcitriol therapy in cancer.[208,238] Part of the great interest stems from the antiproliferative role of calcitriol,[22] with specific effects on DNA replication genes[155] and with a potentially important effect on proliferation of blood vessel endothelial cells.[39] Studies are focused on human prostate cancer[288] and also on breast and colon cancers.[238] Although a discussion is beyond the scope of this chapter, its dynamic character indicates it will be important for many years to come.

CALCITONIN

Calcitonin is a 32-amino acid polypeptide hormone that is synthesized by C cells in the thyroid gland.[352,435] An important role of calcitonin is to limit the degree of postprandial hypercalcemia. This effect, in concert with PTH, acts to maintain serum iCa concentration within a narrow range. Calcitonin is secreted in response to hypercalcemia and also to a calcium-rich meal. Calcitonin secretion increases during hypercalcemia, but the effects of calcitonin on normal calcium homeostasis are considered to be minor. The major target site for calcitonin is bone, where it inhibits osteoclastic bone resorption. The effects of calcitonin in bone are transitory, which has limited the usefulness of calcitonin as a treatment for hypercalcemia. At high doses, calcitonin may promote urinary calcium excretion.[76]

NORMAL HOMEOSTATIC RESPONSE TO HYPOCALCEMIA

Hypocalcemia elicits corrective responses that are mediated by PTH and calcitriol.[435] Acute effects occur in seconds to minutes; subacute effects occur over several hours; and chronic effects occur over days to weeks. A marked increase in PTH secretion occurs in response to mild hypocalcemia, and this response occurs in seconds. Acute secretion of preformed PTH can maintain PTH concentrations for 1 to 1.5 hours during hypocalcemia. Hypocalcemia decreases the proportion of PTH that is degraded in the parathyroid chief cells, making more PTH available for secretion. This effect is relatively rapid (approximately 40 minutes). During increased PTH secretion, renal calcium reabsorption and phosphorus excretion are increased within minutes, whereas bone mobilization of calcium and phosphate occurs within 1 to 2 hours.

After several hours of hypocalcemia, increased PTH secretion stimulates the synthesis and secretion of calcitriol. Increased intestinal transport of calcium and phosphorus into blood follows, providing an external source of calcium in addition to the internal mobilization from bone. Hypocalcemia increases transcription of the PTH gene and synthesis of PTH mRNA, enhancing the ability of the chief cells to produce PTH. This effect also occurs within hours of hypocalcemia. Over days or weeks of hypocalcemia, further increases in PTH secretion are achieved largely by hypertrophy and hyperplasia of chief cells in the parathyroid gland.[454] In addition, the proportion of chief cells actively synthesizing PTH is increased.

NORMAL HOMEOSTATIC RESPONSE TO HYPERCALCEMIA

Most of the effects that occur during hypercalcemia are the opposite of those described earlier for hypocalcemia.[435] Hypercalcemia results in decreased PTH secretion, increased intracellular degradation of PTH in chief cells, and decreased PTH synthesis. Increased calcitonin secretion is stimulated in an attempt to minimize the magnitude of hypercalcemia. In addition, hyperplasia of C cells in the thyroid gland results if the hypercalcemic stimulus is sustained, but this mechanism is ineffective for controlling hypercalcemia because of the transitory effect of calcitonin on osteoclastic bone resorption.[382,441] Calcitriol synthesis is decreased both through direct inhibition by iCa and as a result of decreased stimulation because of decreased PTH concentration.

DIAGNOSTICS

Table 6-1 lists normal values for serum tCa,[111] iCa,[110] PTH,[366,526] PTHrP,[446] and vitamin D metabolites that are useful in the diagnostic workup of patients with calcium disorders.[435]

TABLE 6-1 Normal Serum Concentrations

	Dog	Cat
Total Calcium		
mg/dL	9.0-11.5	8.0-10.5
mmol/L	2.2-3.8	2.0-2.6
Ionized Calcium		
mg/dL	5.0-6.0	4.5-5.5
mmol/L	1.2-1.5	1.1-1.4
Parathyroid Hormone (PTH)		
Intact (pmol/L)	2-13*	0-4*
N-terminal (pg/mL)	15-55	8-28
Parathyroid Hormone Related protein (PTHrP) (pmol/L) (intact or N-terminal)	<1.0*	<1.0*
25-Hydroxyvitamin D (calcidiol) (nmol/L)	60-215*	65-170*
1,25-Dihyroxyvitamin D (calcitriol) (pg/mL)		
Adults	20-50	20-40
10-12-week-old	60-120	20-80

**Data from Endocrine Diagnostic Section, Diagnostic Center for Population and Animal Health, Lansing, MI.*

TOTAL CALCIUM

Despite the fact that only the iCa fraction is physiologically active, the calcium status of animals is usually initially based on evaluation of the serum tCa concentration. Measurement of tCa concentration is more readily available than iCa measurement, but it does not always accurately reflect the iCa concentration of the patient. The serum tCa concentration has been assumed to be directly proportional to iCa, but in many clinical conditions, this may lead to erroneous interpretation of laboratory data. In humans with disorders of calcium balance, measurement of serum tCa concentrations failed to predict serum iCa concentrations in 31% of all patients[515] and in 26% of patients with renal disease.[83] In 1633 canine samples, diagnostic disagreement between serum iCa and tCa was 27%, and in dogs with CRF, this disagreement was 36%.[475] In cats, serum iCa concentrations were only moderately correlated with serum tCa concentrations,[134] and a 40% diagnostic disagreement between serum iCa and tCa measurement was noted in 434 cats.[474] In dogs, tCa measurement overestimated normocalcemia and underestimated hypocalcemia,[475] and in cats, hypercalcemia and normocalcemia were underestimated, and hypocalcemia was overestimated when using serum tCa concentration to predict iCa status.[474]

Analytical Methods

Fasting serum or heparinized plasma samples should be submitted for analysis. Oxalate, citrate, and ethylenediaminetetraacetic acid (EDTA) anticoagulants should not be used because calcium is bound to these chemicals and becomes unavailable for analysis.[567]

Serum tCa concentrations vary with the method used. Isotope dilution with subsequent mass spectrometry constitutes the definitive method for calcium measurement but is not readily available.[189] For clinical determination of serum tCa concentration, simple colorimetric reactions and spectrophotometry are usually employed using automated or manual methods. *Ortho*-cresophthalein complexone is a metal dye that is commonly used to form a color complex with calcium. This method is accurate and reproducible.[189] Hemolysis can result in formation of an interfering hemoglobin-chromogen complex that falsely increases measured calcium concentration. High concentrations of bilirubin falsely decrease, and acetaminophen and hydralazine falsely increase serum tCa concentration. Lipemia can result in spuriously high calcium concentrations,[345] with values exceeding 20 mg/dL in some instances of severe lipemia.

Caution should be exercised in the interpretation of tCa measurements performed on small serum or plasma volumes. When submitted volume is inadequate, dilution with water or saline is often performed. In an in-house commercial laboratory study, when samples were diluted 1:3, serum tCa concentrations were nearly 3 mg/dL lower than when analyzed in undiluted samples (Antech newsletter 05-1999).

Normal Values

The range for serum tCa concentration in normal dogs and cats is wide and varies among laboratories (see Table 6-1). Each laboratory should establish normal values. Variability may result from differences in age, diet, duration of fasting before sampling, and time of sampling, in addition to differences in analytical method.

Normal serum tCa concentrations in mature dogs and cats are approximately 10.0 and 9.0 mg/dL, respectively. No difference in serum tCa concentration has been ascribed to breed or sex in normal dogs and cats, but an effect of aging has been observed in the dog.[111,224] Dogs younger than 3 months of age have slightly higher mean serum calcium concentrations (approximately 11.0 mg/dL) than those for dogs older than 1 year (approximately 10.0 mg/dL), probably because of normal bone growth. In a small percentage of normal young dogs, serum tCa concentrations may be greater than 12.0 mg/dL and as high as 15.0 mg/dL.[379] Dietary calcium, phosphorus, and vitamin D supplementation should be evaluated in dogs with serum tCa concentrations greater than 12.0 mg/dL.

Adjusted Total Calcium

It has been reported that serum tCa concentrations should be "corrected" or "adjusted" relative to the total serum protein or albumin concentration to improve diagnostic interpretation.[174,341] Such correction seemed logical because binding of serum calcium to protein is substantial, and 80% to 90% of the calcium bound to proteins is bound to albumin. The correlation between serum tCa and serum albumin or total protein concentrations was moderate, and adjustment formulas were developed for use in dogs older than 1 year. These adjustment formulas were not recommended for use in cats because there was no linear relationship between serum tCa and serum albumin and total protein concentrations in this species.[179]

It has been assumed that serum tCa concentrations that correct into the normal range are associated with normal serum iCa concentration. Likewise, samples with values that fail to correct into the normal range are presumed to have abnormal serum iCa concentrations. However, these formulas were developed without verification by serum iCa measurements. Correction of serum tCa concentration for albumin did not improve the correlation between serum tCa and iCa concentrations.[350] In 1633 canine serum samples, the use of an adjustment formula to predict iCa status showed a higher diagnostic disagreement than did serum tCa measurement alone.[475] Diagnostic disagreement between tCa adjusted to total protein and iCa measurement was 37% and was 38% between tCa adjusted to albumin and iCa measurement. In 490 dogs with CRF, diagnostic disagreement between adjusted tCa and iCa measurement increased to 53%, indicating the poor performance of the adjustment formulas in the prediction of iCa status. In all dogs, hypercalcemia and normocalcemia were overestimated, and hypocalcemia was underestimated when either adjustment formula was used. In dogs with CRF, however, hypercalcemia was overestimated, and normocalcemia and hypocalcemia were underestimated. Because of the high degree of diagnostic disagreement between adjusted tCa and iCa measurement, the use of adjustment formulas to predict iCa status cannot be recommended.

IONIZED CALCIUM

Ionized calcium is the biologically active form of calcium, and its homeostasis is important for many physiologic functions.[435] Calcium ion regulates its own homeostasis directly by binding to cell membrane receptors specific for iCa.[74] The cell membrane calcium receptors are present in parathyroid chief cells and C cells of the thyroid gland, in which iCa regulates PTH and calcitonin secretion, respectively. Calcium receptors are also present on renal tubular cells, and iCa directly regulates its own tubular reabsorption rate. Therefore serum iCa concentration is controlled by interacting feedback loops that involve iCa, phosphate, PTH, calcitriol, and calcitonin. These mechanisms help maintain serum iCa concentration in a narrow range.

For accurate assessment of calcium status, iCa must be measured directly. Ionized calcium measurement has been shown to be superior to serum tCa measurements in many conditions, especially in hyperparathyroidism, renal disease, hypoproteinemia and hyperproteinemia, acid-base disturbances, and critical illnesses.[205,475,580] Changes in the magnitude of serum protein concentration, individual protein binding capacity and affinity, serum pH, and complexed calcium all interact to determine the iCa concentration, independent of the tCa concentration. Fasting serum samples collected at the same time in the morning are advised.

Analytical Methods

Use of automated equipment with a calcium ion-selective electrode allows easy and accurate measurement of iCa in blood, plasma, or serum.[59] Newly developed electrodes minimize interference by other ions (e.g., magnesium, lithium, and potassium), protein, or hemolysis.[207] Nevertheless, differences among analyzers exist, and it is recommended that reference ranges be established for each analyzer.[246]

Recently, portable clinical analyzers have been developed for cage-side analysis of iCa concentration. These analyzers use a disposable cartridge containing an impregnated biosensor for iCa and other analytes. Heparinized whole blood is used for analysis, but caution should be exercised when interpreting these results. Ionized calcium concentrations in dogs are typically 0.05 to 0.26 mmol/L lower, and 0.05 to 0.14 mmol/L lower in cats, when heparinized whole blood is compared with serum iCa measurement.[213] The greatest underestimation of iCa concentration occurred when serum iCa concentrations were greater than 1.3 mmol/L. When iCa concentration in heparinized whole blood was measured using both ion-selective electrode methodology and portable clinical analyzer methods, correlation (*r*) was only 0.71.[361] The portable clinical analyzer method resulted in an iCa concentration that was approximately 2.6% lower than that measured with an ion-selective electrode.[308] However, in a study of dogs and horses, there were no differences in iCa concentrations using heparinized whole blood measured with an ion-selective electrode and portable clinical analyzer.[311] Because the quantity and type of heparin used and volume of blood collected also have an effect on iCa measurement, it is best to establish a rigid protocol for blood collection when using a portable clinical analyzer. Reference ranges should also be established for the analyzer using this standard protocol.

Sample Handling Techniques

Concentration of iCa can be determined in samples handled under both anaerobic and aerobic conditions. The most precise determination of iCa concentration and physiologic pH requires that samples be collected and processed anaerobically to ensure that no increase in pH occurs because of loss of CO_2. The pH of blood or serum has a significant effect on serum iCa concentration. Acidic pH favors dissociation of calcium from protein and increases the amount of iCa in the sample. Alkaline pH occurs with loss of CO_2 and favors calcium binding to protein, thus decreasing the amount of iCa. Mixing serum with air results in increased pH and decreased measured iCa concentration because of loss of CO_2 from the sample.[471] Exposure to air in partially filled serum tubes also can affect iCa concentration; tubes that were only 25% or 50% filled had 0.07 or 0.04 mmol/L lower concentrations of iCa when compared with measurement from tubes that were 100% filled.[535]

Ionized calcium can be measured in whole blood or heparinized plasma, but measurement is problematic. Heparinized canine blood provided stable iCa measurements when stored up to 9 hours at 4° C, but pH was significantly increased after 3 hours.[506] In practice, it may be impossible to analyze the sample within this period. The amount and type of heparin used for whole blood or plasma samples also affect the measurement of iCa. When zinc heparin is used as an anticoagulant, iCa concentration is overestimated most likely because of a decrease in pH, which displaces calcium from proteins.[312,314] Lithium heparin causes an underestimation in iCa concentration,[312] and an electrolyte-balanced heparin may underestimate or overestimate iCa concentration depending on whether hypocalcemia, normocalcemia, or hypercalcemia is present. The amount of heparin used is critical in the measurement of iCa in blood. Using syringes containing a premeasured quantity of lithium heparin or electrolyte-balanced heparin, iCa measurement was underestimated when a less than recommended quantity of blood was collected for analysis.[312,313] When using heparinized whole blood for measurement of iCa concentration, it is imperative to collect the same volume of blood for each sample to avoid the dilutional effects of heparin. Syringes containing a premeasured amount of dry heparin are preferable to coating a syringe manually with an unknown and variable quantity of liquid heparin.

Ionized calcium and pH are more stable in serum than in whole or heparinized blood. The analysis of serum eliminates the potential interference of heparin and allows a longer storage period before analysis. Silicone separator tubes should not be used; the iCa concentration was increased in serum separated by use of silicone separator tubes because of release of calcium from the silicone gel.[298] Measured iCa in canine and equine serum was stable after storage for 72 hours at 23° C or 4° C and for 7 days at 4° C.[470,471] Use of serum collected anaerobically and stored at 4° C allows sufficient time for shipment to a reference laboratory for anaerobic measurement of iCa and pH.

Ionized calcium may also be accurately measured in samples handled aerobically. Mathematical formulas have been developed to correct the iCa concentration in samples exposed to air (with increased pH) to the actual pH of the patient or to a pH of 7.4.[305,362] In a study of serum samples from 61 dogs and 21 cats, there was good correlation between iCa measured anaerobically and again aerobically after shipment to a diagnostic laboratory (Schenck and Chew, unpublished observations). These pH correction formulas are species specific, and formulas developed in humans should not be used. A mathematical correction formula should be derived for each species in each laboratory setting. Although not as precise as anaerobic measurement, aerobic measurement under proper laboratory conditions offers a diagnostically accurate methodology for iCa determination with simplified shipping and handling requirements.

Some iCa analyzers will automatically mathematically manipulate the iCa concentration and actual pH value of the sample and yield an adjusted value for iCa concentration that theoretically would occur at a pH of 7.4. These correction formulas were developed for use in humans and should not be used in animals. When using anaerobically collected samples, corrected iCa concentrations have not been advocated for use in humans because insight into the pathophysiology of the patient is gained by evaluation of the in vivo iCa concentration and pH.[188] This may be especially true for patients with renal disease.[455] If anaerobic sampling is possible (typically in an in-house setting), there is no necessity or benefit in correcting the iCa concentration to a pH of 7.4. Only when samples are handled aerobically is there a need for correction to a standard pH.

Normal Values

The range for serum iCa concentration in normal dogs and cats varies among laboratories but is approximately 5.0 to 5.8 mg/dL (1.25 to 1.45 mmol/L) in adult dogs[472] and 4.6 to 5.4 mg/dL (1.15 to 1.35 mmol/L) in adult cats.[134] An effect of aging has been observed in both the dog and cat. Young dogs and cats (up to 2 years of age) have serum iCa concentrations that are 0.1 to 0.4 mg/dL higher than those reported in older animals.[134,350] Normal values should be established for each laboratory based on age of animal, type of sample, and analyzer used.

Fractionation of Serum Calcium

In addition to measuring the ionized concentration in serum, the protein-bound and complexed fractions of calcium can be quantified using fractionation techniques. Ionized calcium and complexed calcium are diffusable, and together are referred to as ultrafilterable calcium. To separate protein-bound from ultrafilterable serum calcium, a micropartition system based on the filtration method has been used.[164,472] The micropartition system contains a filter through which ultrafilterable calcium (complexed and ionized) passes. It is important that serum be collected anaerobically before ultrafiltration to allow accurate measurement of the calcium fractions and to prevent changes in serum pH.

Protein-bound, ionized, and complexed calcium fractions in serum were 34%, 56%, and 10% in normal dogs[472] and 40%, 52%, and 8% in normal cats, respectively (Schenck, unpublished observations). Ultrafilterable calcium (ionized and complexed fractions) in dogs,[472] horses,[239] and cats (Schenck, unpublished observations) accounted for 66%, 63%, and 60% of serum tCa, respectively. The iCa fraction has the smallest variation, with larger variations occurring in the protein-bound and complexed fractions. This observation supports the concept that the iCa fraction is tightly regulated and represents the biologically active fraction of serum calcium.

Complexed and protein-bound calcium fractions have not been assessed in metabolic disorders associated with abnormal calcium concentrations. Measurement of the protein-bound and complexed calcium fractions in addition to the iCa fraction may facilitate detection of disease processes that affect calcium metabolism. In dogs with CRF, two subgroups have been identified based on calcium fractionation. Dogs with normal to elevated serum tCa concentrations had a significantly higher concentration of circulating complexed calcium as compared with those dogs with low concentrations of tCa, even though there was no difference in iCa or protein-bound calcium between groups.[473] Further studies are needed to determine whether prognosis or effectiveness of therapy differs between these groups.

Parathyroid Hormone

PTH circulates predominantly as intact PTH (1-84) and carboxyl-terminal fragments. Only intact PTH is biologically active, and it is best to measure this form in serum or plasma. Samples should be stored and shipped frozen to prevent degradation of intact PTH. Stability is best in plasma collected with EDTA, but serum is adequate if stored frozen after separation from blood. Because of sequence homology of human and animal PTH, commercial assays developed for humans have been used successfully for some veterinary species.[113] An amino-terminal–specific radioimmunoassay (RIA) was used for more than 50 mammalian species but is no longer commercially available.[364] A two-site immunoradiometric assay (IRMA) for intact human PTH has been validated in the dog and cat.[23,526] Normal values for serum PTH concentration are 2 to 13, 0 to 4, and 0 to 2 pmol/L in the dog, cat, and horse, respectively (Endocrine Diagnostic Section, Diagnostic Center for Population and Animal Health, Lansing, MI). The two-site assays have not proved useful for measurement of PTH in reptiles. Expected response of PTH in various conditions will be discussed later (see Hypercalcemia and Hypocalcemia).

The current two-site IRMA measures both the intact PTH-(1-84) and the PTH-(7-84) fragment because the amino-terminal antibodies react near the tenth amino acid.[69,126,375] A new third generation IRMA "whole" PTH assay has been developed for use in humans that measures only PTH-(1-84).[192] This new assay could offer a better measure of whole PTH especially in patients with secondary hyperparathyroidism because the PTH-(7-84) fragment is increased in these patients.[351] High concentrations of carboxyl-terminal PTH fragments, which occur in cats with CRF, may interfere with intact PTH immunoassays.[27] Using ratios of "whole" PTH versus "intact" PTH to clarify low bone turnover renal osteodystrophy[368] or dynamics of PTH secretion[463] have been attempted.[203,281] The "whole" PTH assay may also be of better diagnostic value in dogs than the "intact" PTH assay because PTH-(7-84) fragments may be increased in dogs as compared with humans.[159] Whole PTH (1-84) and intact PTH (1-84 and 7-84) have been measured in dogs, and it was observed that the whole PTH/intact PTH ratio in dogs (about 36%) was less than in humans, and the ratio did not change during acute hypocalcemia.[159] In preliminary studies in cats, a third generation PTH-(1-84) assay resulted in higher PTH values than a second generation assay that also measures the PTH-(7-84) fragment.[127] Although this is opposite of what is found in humans, it is not unexpected because cat and other mammalian PTH is more similar to human PTH in the first few amino acids than in the region of the tenth amino acid.

PARATHYROID HORMONE–RELATED PROTEIN

Two-site IRMA and N-terminal RIA are available for the measurement of human PTHrP.[44,286] These assays are useful for measuring biologically active PTHrP in the dog (see Cancer-Associated Hypercalcemia)[113,446] because of the high degree of sequence homology of PTHrP between species, especially in the N-terminal 111 amino acids.[86] An N-terminal RIA for human PTHrP did not prove useful for measuring circulating PTHrP in a small number of horses.[447] PTHrP is susceptible to degradation by serum proteases, and PTHrP concentrations must be measured in fresh or frozen plasma using EDTA as an anticoagulant. EDTA complexes with plasma calcium, which is required for function of many proteases. The addition of protease inhibitors such as aprotinin and leupeptin may provide further inhibition of proteolysis in plasma.[391]

The circulating forms of PTHrP are not completely understood because PTHrP rapidly undergoes proteolysis intracellularly and extracellularly after secretion into blood.[391] The forms of PTHrP that are present in vivo include intact PTHrP, an N-terminal peptide, a combined N-terminal and midregion peptide, a midregion peptide, and a C-terminal peptide.[85,574] Fragments that have PTH-like biologic activity in vivo include N-terminal PTHrP (1-36), PTHrP (1-86), and intact PTHrP (1-141). The two-site immunologic assays measure intact PTHrP (1-141) and PTHrP (1-86) because antibodies bind to the N terminus and midregion. The N-terminal RIAs measure intact PTHrP (1-141), PTHrP (1-86), and N-terminal PTHrP (1-36). The C-terminal PTHrP accumulates in the serum of human patients with renal failure, which suggests that C-terminal PTHrP peptides are excreted by the kidney, as occurs with PTH.[84]

VITAMIN D METABOLITES

Measurement of vitamin D metabolites is occasionally helpful in diagnosing disorders of calcium homeostasis (see Table 6-1). 25-Hydroxyvitamin D (calcidiol) and calcitriol are the metabolites of clinical interest for detection of hypovitaminosis D, hypervitaminosis D, and abnormalities of the renal hydroxylase system (e.g., renal failure). The metabolites are stable during refrigeration and freezing, but samples should not be exposed to light for long periods.

The metabolites of vitamin D are chemically identical in all species, thus receptor-binding assays or RIAs developed for use in humans are satisfactory for the measurement of the same metabolites in animals.[240,242] Young growing dogs have higher calcitriol concentrations than mature dogs, and most mammals appear to share this attribute during rapid growth.[339]

Concentrations of 25-hydroxyvitamin D are a good indicator of vitamin D ingestion or production in vivo and can be used to diagnose hypovitaminosis D or hypervitaminosis D.[102] Calcitriol assays can be used to detect genetic errors of vitamin D metabolism, low concentrations of calcitriol in patients with renal failure, or high concentrations of calcitriol in some patients with cancer-associated hypercalcemia.[435]

BONE BIOPSY AND BONE MARROW ASPIRATION

Bone marrow aspiration or core biopsy is frequently part of the diagnostic evaluation of animals without an obvious cause of hypercalcemia. Its greatest utility is in the discovery of lymphoma, myeloproliferative disease, or multiple myeloma. Biopsy of the iliac crest is recommended for standardization, particularly when histomorphometric analysis is available for the quantitative evaluation of bone formation and bone resorption. A procedure for iliac crest bone biopsy has been described.[117,443] Direct biopsy of focal bone lesions may be diagnostic, particularly when such lesions are caused by lymphoma, multiple myeloma, or a metastatic bone tumor.

HYPERCALCEMIA

Hypercalcemia is an uncommon but important electrolyte disturbance of dogs and cats. The frequency of finding hypercalcemia based on evaluation of serum tCa in more than 10,000 canine serum samples analyzed during a 6-month period at one private veterinary diagnostic laboratory was 1.5%.[89] Of these, 28% were found to be from young growing dogs, 62% were found to be transient, and 18% were persistent and associated with pathology.

Hypercalcemia can serve as a marker of disease or can create disease. Increases in serum iCa concentration above normal often have adverse pathophysiologic consequences. Hypercalcemia represents a clinically relevant increase above an individual animal's own normal serum calcium concentration, usually defined as a fasting serum tCa concentration greater than 12.0 mg/dL in dogs or greater than 11.0 mg/dL in cats. Ionized calcium measurements can provide greater sensitivity and specificity for the diagnosis of some hypercalcemic disorders. A serum iCa concentration greater than 6.0 mg/dL (1.5 mmol/L) in dogs and greater than 5.7 mg/dL (1.4 mmol/L) in cats constitutes ionized hypercalcemia.

TOXICITY OF HYPERCALCEMIA AND CLINICAL SIGNS

Excessive calcium ions are toxic to cells,[420] and increased serum iCa concentration decreases cellular function by causing alterations in cell membrane permeability and cell membrane calcium pump activity. Increased intracellular iCa content can ultimately result in cell death caused by deranged cellular function and reduced energy production. Although all tissues may be subject to the dangerous effects of hypercalcemia, effects on the central nervous system, gastrointestinal tract, heart, and kidneys are of most importance clinically.

Polydipsia, polyuria, anorexia, lethargy, and weakness are the most common clinical signs in dogs with hypercalcemia,[109,168] but individual animals often display remarkable differences in clinical signs despite similar magnitudes of hypercalcemia. The severity of clinical signs and development of lesions of hypercalcemia depend not only on the magnitude of hypercalcemia but also on its rate of development and duration. Simultaneous disturbances in other electrolyte concentrations and in acid-base balance, as well as organ dysfunction secondary to hypercalcemia, all contribute to clinical signs, laboratory abnormalities, and lesions. Box 6-1 lists the signs and conditions associated with hypercalcemia.

Clinical signs are most severe when hypercalcemia develops rapidly, as can occur with vitamin D intoxication or during rapid infusion of calcium-containing fluids. Dogs with similar magnitudes of hypercalcemia may display minimal clinical signs when hypercalcemia has developed gradually. Regardless of the rate of increase in serum calcium concentration, clinical signs become more severe as the magnitude of hypercalcemia increases. Serum tCa concentrations of 12.0 to 14.0 mg/dL may not be associated with severe clinical signs, but most animals with concentrations greater than 15.0 mg/dL show systemic signs. Dogs with serum calcium concentrations greater than 18 mg/dL are often severely ill, and concentrations greater than 20 mg/dL may constitute a life-threatening crisis. Exceptions do occur, however, and some dogs are severely affected by mild hypercalcemia, whereas others are relatively unaffected by severe hypercalcemia. Clinical signs and histopathologic changes are more likely to develop the longer hypercalcemia has been present, regardless of its magnitude. Progressive hypercalcemia may also contribute to the severity of clinical signs, as occurs in animals with malignant neoplasia or hypervitaminosis D related to rat bait ingestion.

Box 6-1 Clinical Signs and Conditions Associated with Hypercalcemia

Common	Uncommon
Polydipsia and polyuria	Constipation
Anorexia	Cardiac arrhythmia
Dehydration	Seizures or twitching
Lethargy	Death
Weakness	Acute intrinsic renal failure
Vomiting	Calcium urolithiasis
Prerenal azotemia	
Chronic renal failure	

Changes in serum sodium and potassium concentrations can magnify the clinical signs of hypercalcemia by their effects on cell membrane excitability, particularly in nerve and muscle (see Chapter 5). Acidosis increases the proportion of serum calcium that is ionized, worsening clinical signs, whereas alkalosis lessens toxicity and clinical signs by decreasing the proportion of calcium that is ionized.

Mineralization of soft tissues (especially the heart and kidneys) is an important complication of hypercalcemia. The serum phosphorus concentration at the time hypercalcemia develops is important in determining the extent of soft tissue mineralization. Soft tissue mineralization is most severe when the calcium (mg/dL) times phosphorus (mg/dL) product is greater than 60.[111] Soft tissue mineralization occurs regardless of the serum phosphorus concentration in severe hypercalcemia.

Renal Effects of Hypercalcemia

Abnormal renal function frequently accompanies hypercalcemia, and rapid deterioration in renal function occasionally occurs. The functional effects of hypercalcemia on the kidneys are readily reversible, but structural changes may not be reversible if renal lesions are advanced. Azotemia occurred commonly in 34 dogs with hypercalcemia related to malignancy, hypoadrenocorticism, CRF, and hypervitaminosis D.[290] The frequency of azotemia was higher in dogs with malignancy (71%) than in those with hypercalcemia related to primary hyperparathyroidism (11%). Azotemia caused by hypercalcemia can result from any combination of the following mechanisms: prerenal reduction in ECF volume (anorexia, hypodipsia, vomiting, and polyuria); renal vasoconstriction from ionized hypercalcemia; decreased permeability coefficient of the glomerulus (K_f); acute tubular necrosis from the ischemic and toxic effects of hypercalcemia; and CRF caused by nephron loss, nephrocalcinosis, tubulointerstitial inflammation, and interstitial fibrosis.

Decreased urinary concentrating ability and polyuria are early functional effects of hypercalcemia in dogs. The concentrating defect is often out of proportion to the observed reduction in glomerular filtration rate (GFR) and increase in serum creatinine or blood urea nitrogen (BUN) concentration. Urine specific gravity is consistently less than 1.030 in dogs and was less than 1.020 in more than 90% of hypercalcemic dogs in one study.[290] Urinary concentration may be well preserved in some cats with hypercalcemia that do not have CRF. Defective urinary concentrating ability results from a combination of reduced tubular reabsorption of sodium and impaired action of antidiuretic hormone on tubular cells of the collecting duct. This results in a form of nephrogenic diabetes insipidus characterized by hyposthenuria if the diluting segment of the nephron (medullary thick ascending limb of Henle's loop) is unaffected. These effects are caused by intrinsic responses of the kidney to hypercalcemia. Some of these effects are mediated by calcium-sensing receptors on the renal epithelial cells,[74] whereas others may be related to effects of hypercalcemia on aquaporin expression, cell trafficking, and delivery to apical membranes of the collecting tubules.[154,418,545] Additional direct effects of hypercalcemia on the kidney include reduced tubular calcium reabsorption and antagonism of the actions of PTH. These responses by the kidney facilitate calcium excretion and help to ameliorate the clinical effects of hypercalcemia. Renal medullary blood flow is increased in dogs with experimental hypercalcemia[81] and can result in medullary washout as another mechanism contributing to hyposthenuria. Isosthenuria develops if the diluting segments have been structurally altered by long-standing hypercalcemia. Polydipsia occurs as compensation for obligatory polyuria, but there is evidence that polydipsia can be caused by direct stimulation of the thirst center by hypercalcemia.[111] Mineralization of renal tubules, basement membranes, or the interstitium; tubular degeneration; and interstitial fibrosis are structural changes that may occur in the kidney secondary to hypercalcemia and can contribute to impaired urinary concentrating ability.

Dehydration is common owing to increased fluid losses from vomiting and polyuria. Substantial contraction of the ECF volume results in reduced GFR severe enough to increase BUN and serum creatinine concentrations and cause prerenal azotemia. The clinical axiom that dilute urine in association with azotemia is caused by intrinsic renal lesions may not be true in animals with hypercalcemia because the urinary concentrating defect can occur without structural renal lesions. This condition is commonly misdiagnosed as primary renal failure when it is actually prerenal failure caused by dehydration and a renal concentrating defect early in the course of hypercalcemia.

Intrarenal causes of azotemia during hypercalcemia can be functional or structural. Hypercalcemia can induce renal vasoconstriction, resulting in decreased renal blood flow (RBF) and GFR.[107] In an acute model of hypercalcemia, reduced RBF and GFR were observed consistently in conscious dogs when serum tCa concentration exceeded 20 mg/dL, but only one half of the dogs had significant reductions in GFR and RBF when serum calcium concentration was 15 to 20 mg/dL. Little effect on RBF and GFR was observed when serum calcium concentration was less than 15 mg/dL. These findings are in contrast to those in studies of anesthetized dogs, which demonstrated much more severe functional changes during hypercalcemia.[309] Impaired renal autoregulation related to the effects of hypercalcemia may result in azotemia at early stages of dehydration because GFR would otherwise be maintained by afferent arteriolar vasodilatation.

Acute intrinsic renal failure (AIRF) occasionally develops as a consequence of hypercalcemia, but chronic intrinsic renal failure is more common. Sustained renal vasoconstriction related to hypercalcemia may result in ischemic tubular injury, promoting development of both AIRF and chronic intrinsic renal failure and potentiating the direct toxic effects of calcium on tubular cells. The toxic effects of ionized hypercalcemia are enhanced by high concentrations of PTH in animals with CRF because excess PTH increases calcium entry into cells.[365] The ascending limb of Henle's loop and distal convoluted tubule show the earliest structural lesions, but lesions in the collecting system are ultimately the most pronounced. Thickening and mineralization of tubular basement membranes are most apparent in the proximal tubule. Tubular atrophy, mononuclear cell infiltration, and interstitial fibrosis occur in the chronic stages. Degenerative and necrotic tubules also are observed. Granular and tubular cell casts contribute to intrarenal obstruction and azotemia.[107,290]

Calcium-oxalate urolithiasis occasionally occurs in animals with long-standing hypercalcemia and has been described in dogs and cats with primary hyperparathyroidism. Nephrocalcinosis and linear mineralization along the renal diverticula are nonspecific findings discovered by radiography or ultrasonography in some dogs with long-standing hypercalcemia. Increased renal echogenicity and the medullary rim sign have been described during renal ultrasonography in dogs with hypercalcemia.[25,48] These changes can occur in other normocalcemic conditions and in forms of dystrophic mineralization.

Effects of Hypercalcemia on Other Organs

Anorexia, vomiting, and constipation can result from hypercalcemia by reduction of the excitability of gastrointestinal smooth muscle and from direct effects on the central nervous system. Gastric hyperacidity and subsequent gastric ulceration caused by increased secretion of gastrin and direct stimulation of hydrogen ion secretion from parietal cells by hypercalcemia may account for some of the vomiting. Gastrin concentration was increased in four of six dogs with hypercalcemia in one preliminary report.[61] Increased gastrin concentration occurs secondary to reduced renal clearance as a consequence of the hypercalcemia. Decreased excitability of skeletal muscle contributes to generalized weakness. Lethargy is commonly observed in severe hypercalcemia because of direct effects on the central nervous system and rarely can progress to stupor and coma. Seizures and muscle twitching are unusual neuromuscular manifestations of hypercalcemia.[251]

Clinically important cardiac effects of hypercalcemia are not commonly detected in dogs and cats, but PR interval prolongation and QT interval shortening can be observed on the electrocardiogram. Serious arrhythmias (including ventricular fibrillation) can be caused by the direct effects of severe hypercalcemia or may be a consequence of mineralization of cardiac tissue. Hypertension has been demonstrated in humans and rats during both acute and chronic hypercalcemia. The increase in blood pressure is proportional to the increase in serum calcium concentration in acute studies.[92] In a study of acute hypercalcemia, hypertension was attributed to a direct effect of calcium on vascular smooth muscle and to an indirect effect of calcium to increase secretion of catecholamine with activation of adrenergic receptors.[156] Whether hypertension is a clinically relevant complication in dogs and cats with hypercalcemia is unknown.

Mechanisms and Differential Diagnosis of Hypercalcemia

Increased entry of calcium into ECF, decreased egress of calcium from ECF, reduced plasma volume, or a combination of these factors must occur for hypercalcemia to develop (Fig. 6-11). Increased calcium input can arise from increased intestinal absorption, increased bone resorption, or increased renal tubular reabsorption of calcium. Decreased glomerular filtration and decreased bone accretion result in decreased egress of calcium from ECF. Volume contraction is common in the presence of hypercalcemia because of the effects of anorexia, vomiting, and obligatory polyuria. The mechanisms of hypercalcemia vary with the specific causes, but much attention has been focused on the importance of increased bone resorption.

Box 6-2 provides a list of possibilities in the differential diagnosis for hypercalcemia. Characterization of the hypercalcemia as transient or persistent, pathologic or nonpathologic, mild or severe, progressive or static, and acute or chronic is helpful in determining its cause. Persistent, pathologic hypercalcemia occurs most often in association with malignancy. Most studies in dogs attribute hypercalcemia to malignancy in more than 50% of the cases,[41,157,534] although in one series malignancy accounted for only one third of the cases.[290] Hypoadrenocorticism, renal failure, primary hyperparathyroidism, hypervitaminosis D, and inflammatory disorders sporadically account for hypercalcemia in dogs. It is often difficult to determine the cause of hypercalcemia in animals with mild or transient hypercalcemia. No definitive diagnosis could be made for 2% to 9% of hypercalcemic dogs in two reports.[157,534] No definitive diagnosis was reported in 13% of cats with hypercalcemia in one report, but the actual percentage is much higher based on sample submissions to veterinary endocrinology laboratories.[467]

In serum samples from 332 hypercalcemic cats, 80% had parathyroid-independent hypercalcemia, 10% had parathyroid-dependent hypercalcemia, and 10% were equivocal.[56] Approximately 10% of these hypercalcemic cats had PTHrP levels above the reference range, suggesting malignancy as the cause. Hypercalcemic cats have parathyroid-independent hypercalcemia more commonly than do dogs. Samples from 5722 hypercalcemic dogs from the same laboratory categorized the hypercalcemia as parathyroid dependent in about 40%, parathyroid independent in 50%, and equivocal in 10%.[423]

General Approach to Diagnostic Workup of Patients with Hypercalcemia

It is important to ensure that the hypercalcemia initially detected is repeatable, especially if the magnitude of hypercalcemia is modest. The likely cause of the hypercalcemia will be obvious in some patients from findings in the history (hypervitaminosis D) or from physical examination (masses and effusions). When the cause is not immediately apparent, body cavity imaging with chest radiographs, abdominal radiographs, and abdominal ultrasound is recommended to determine whether organomegaly or infiltrative processes are present that could account for the hypercalcemia. Fine needle aspiration, needle biopsy, or wedge biopsy of abnormal tissues will often yield the cause of the hypercalcemia. Patients with cytopenias (neutropenia, anemia, and thrombocytopenia) should undergo bone marrow evaluation if the diagnosis has not already been established by other means. Bone marrow evaluation in the absence of cytopenias does not often result in a diagnosis. Radiographs of painful bones may reveal lesions associated with hypercalcemia. Aspiration of focal bone lesions may reveal the cause of the hypercalcemia. Bone survey of all bones is sometimes useful in finding lesions even in those without demonstrable bone pain (multiple myeloma). Bone scintigraphy may be considered in those in which a diagnosis is lacking despite exhaustive diagnostics.

High frequency ultrasonography of the cervical region can be performed to help determine whether the hypercalcemia is parathyroid dependent (large parathyroid glands) or parathyroid independent. In parathyroid-independent hypercalcemia, parathyroid glands are not enlarged or may not be identified; some may be atrophic if ionized hypercalcemia of malignancy or hypervitaminosis D has been long standing.

If the increase in serum tCa is minimal, measurement of serum iCa is important to determine whether the increase is clinically significant. Measurement of iCa in patients with renal failure is essential because renal failure can be associated with nonionized or ionized hypercalcemia. Serum iCa should be measured in association with PTH determination to assess the appropriateness of PTH response to serum iCa concentration.

If the cause of hypercalcemia is not apparent following history, physical examination, hematology, routine serum biochemistry, and body cavity imaging, then measurement of calcium-regulating hormones is needed to establish or suggest a definitive cause. The first step is to determine whether the hypercalcemia is parathyroid dependent (disease of the parathyroid glands is causing the hypercalcemia) or parathyroid independent (normal parathyroid glands suppress PTH secretion in response

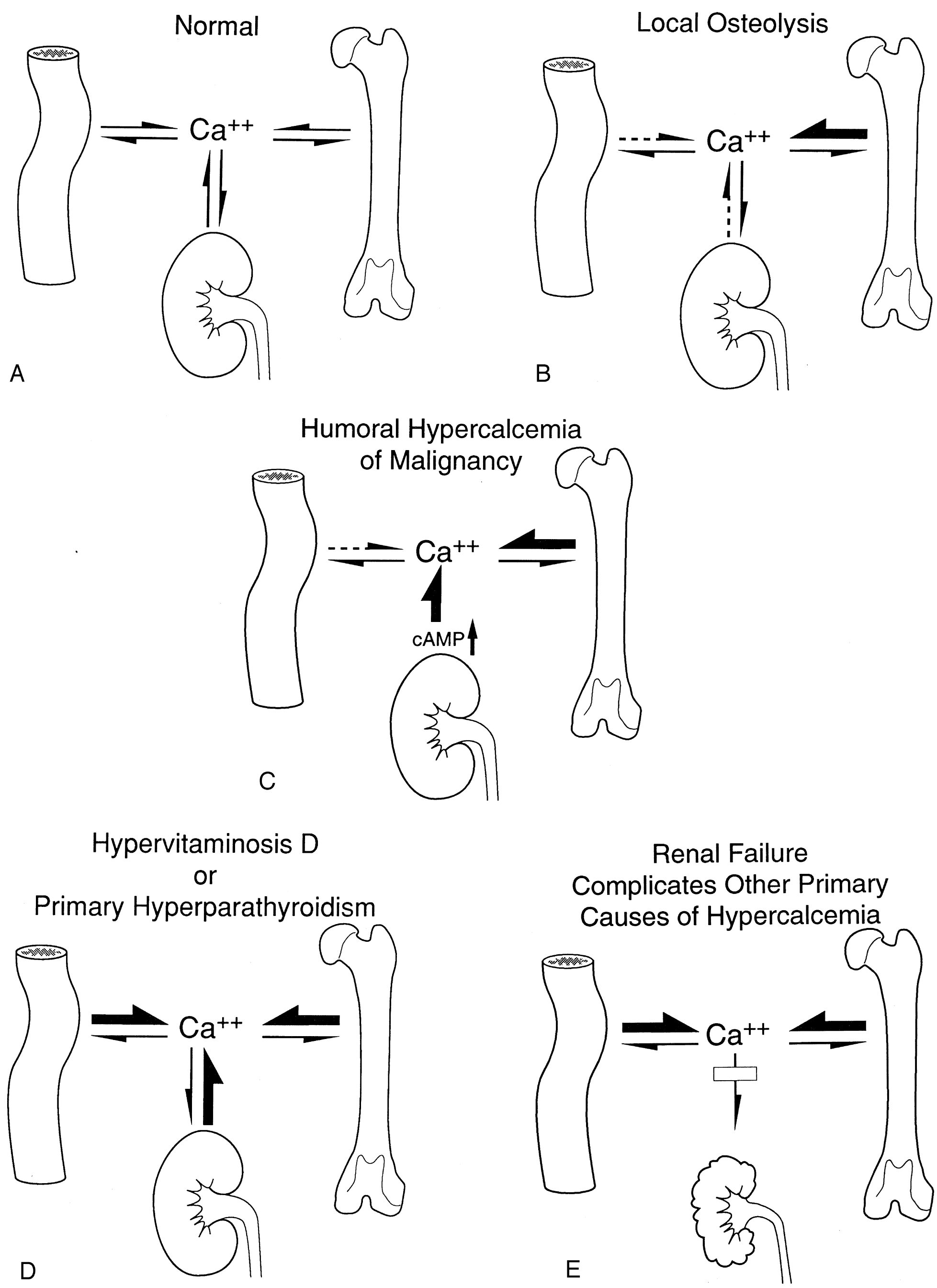

Fig. 6-11 Patterns of calcium transport between extracellular fluid and gut, kidney, and bone in various states of hypercalcemia. **A,** Normal. **B,** Osteolysis. **C,** Humoral hypercalcemia of malignancy. **D,** Hypervitaminosis D or primary hyperparathyroidism. **E,** Hypercalcemia complicated by renal failure. Size of arrows is proportional to the degree of calcium influx or efflux. Dashed arrows indicate possible response of decreased PTH secretion to hypercalcemia of nonparathyroid origin. (Modified from Mundy GR: Malignancy and hypercalcemia—humoral hypercalcemia of malignancy, hypercalcemia associated with osteolytic metastases. In Mundy GR, editor: *Calcium homeostasis: hypercalcemia and hypocalcemia,* London, 1989, Martin Dunitz, p. 65.)

Box 6-2 Conditions Associated with Hypercalcemia

Nonpathologic
- Nonfasting (minimal increase)
- Physiologic growth of young
- Laboratory error
- Spurious
  - Lipemia
  - Detergent contamination of sample or tube

Transient or Inconsequential
- Hemoconcentration
- Hyperproteinemia
- Hypoadrenocorticism
- Severe environmental hypothermia (very rare)

Pathologic or Consequential—Persistent
- Parathyroid dependent
  - Primary hyperparathyroidism
    - Adenoma (common)
    - Adenocarcinoma (rare)
    - Hyperplasia (uncommon)
- Parathyroid independent
  - Malignancy-associated (most common cause in dogs)
    - Humoral hypercalcemia of malignancy
      - Lymphoma (common)
      - Anal sac apocrine gland adenocarcinoma (common)
      - Carcinoma (sporadic): lung, pancreas, skin, nasal cavity, thyroid, mammary gland, adrenal medulla
      - Thymoma (rare)
    - Hematologic malignancies (bone marrow osteolysis, local osteolytic hypercalcemia)
      - Lymphoma
      - Multiple myeloma
      - Myeloproliferative disease (rare)
      - Leukemia (rare)
    - Metastatic or primary bone neoplasia (very uncommon)
  - Idiopathic hypercalcemia (most common association in cats)
  - Chronic renal failure (with and without ionized hypercalcemia)
  - Hypervitaminosis D
    - Iatrogenic
    - Plants (calcitriol glycosides)
    - Rodenticide (cholecalciferol)
    - Antipsoriasis creams (calcipotriol or calcipotriene)
  - Granulomatous disease
    - Blastomycosis
    - Dermatitis
    - Panniculitis
    - Injection reaction
  - Acute renal failure (diuretic phase)
  - Skeletal lesions (nonmalignant) (uncommon)
    - Osteomyelitis (bacterial or mycotic)
    - Hypertrophic osteodystrophy
    - Disuse osteoporosis (immobilization)
  - Excessive calcium-containing intestinal phosphate binders
  - Excessive calcium supplementation (calcium carbonate)
  - Hypervitaminosis A
  - Raisin/grape toxicity
  - Hypercalcemic conditions in human medicine
    - Milk-alkali syndrome (rare in dogs)
    - Thiazide diuretics
    - Acromegaly
    - Thyrotoxicosis (rare in cats)
    - Postrenal transplantation
    - Aluminum exposure (intestinal phosphate binders in dogs and cats?)

to hypercalcemia). Measurement of PTHrP is helpful if malignancy is suspected, but PTHrP concentrations are not always increased in malignancy. If extensive imaging methodologies are not available, measurement of serum iCa, PTH, and PTHrP may be performed before extensive body cavity imaging or bone marrow evaluation. Measurement of 25-hydroxyvitamin D is useful in cases of potential cholecalciferol or ergocalciferol ingestion. Measurement of 1,25-dihydroxyvitamin D (calcitriol) is occasionally useful if excess calcitriol is the cause of hypercalcemia. The anticipated changes in calcium hormones and serum biochemistry in disorders causing hypercalcemia are noted in Table 6-2.

NONPATHOLOGIC HYPERCALCEMIA

Serum calcium concentrations in animals may be mildly increased after feeding; consequently, a 12-hour fast is recommended before blood sampling. Laboratory error or detergent contamination of the serum or sample tube may result in artifactual hypercalcemia.[345] Lipemia frequently causes erroneously high serum tCa concentrations because of colorimetric interference. Normal young growing dogs may have mildly higher serum calcium concentrations than older dogs.[350]

Transient or Inconsequential Hypercalcemia

Inconsequential hypercalcemia does not cause injury, resolves rapidly, or is only mild. Dehydration can result in mild hypercalcemia attributed to hemoconcentration. Furthermore, dehydration and volume contraction stimulate increased sodium and calcium reabsorption in the kidney. An increased serum concentration of protein, especially albumin, can result in an increased serum tCa concentration as more calcium binds to protein. Dehydration in dogs is occasionally associated with serum tCa concentrations of 12.0 to 13.5 mg/dL that rapidly return to normal after dehydration is corrected. Increased serum tCa and decreased iCa concentrations can occur transiently after plasma transfusion because of excess citrate–calcium ion complexes.[350]

TABLE 6-2 Anticipated Changes in Calcemic Hormones and Serum Biochemistry Associated with Disorders of Hypercalcemia

	tCa	iCa	alb	Corr tCa	Pi	PTH	PTHrP	25(OH)-D	1,25 $(OH)_2$-D	PTG ULS, Surgery
Primary hyperparathyroidism	↑	↑	N	N	↓N	↑N	N	N	N ↑	Single ↑
Nutritional secondary hyperparathyroidism	N↓	N↓	N	N↓	N↑	↑	N	↓N	N ↓	Multiple ↑
Renal secondary hyperparathyroidism	N ↓ ↑	N↓	N	N	↑ N	↑	N	N ↓	N ↓	Multiple ↑
Tertiary hyperparathyroidism	↑	↑	N	↑	↑	↑	N	N ↓	↓ N	Multiple ↑
Malignancy Associated										
Humoral hypercalcemia	↑	↑	N ↓	↑ N	↓ N	↓ N	↑ N	N	↓N ↑	↓
Local osteolytic	↑	↑	N ↓	↑ N	N ↑	↓ N	N ↑	N	N	↓
Hypervitaminosis D										
Cholecalciferol	↑	↑	N	↑	↑ N	↓	N	↑	N ↑	N ↓
Calcitriol	↑	↑	N	↑	N ↑	↓	N	N	↑	↓ N
Calcipotriene	↑	↑	N	↑	↑ N	↓	N	N	↓ N	↓ N
Hypoadrenocorticism	↑	↑	N ↓	↑	↑ N	↓ N	N	N	↓ N	N
Hypervitaminosis A	↑	↑	N	↑	N	↓	N	N	N ↓	↓ N
Idiopathic (cat)	↑	↑	N	↑	N ↑	↓ N	N	N	N ↓ ↑	↓ N
Dehydration	↑	N ↑	↑ N	↑ N	N ↑	N ↓	N	N	N	N
Aluminum exposure (renal failure)	↑	↑	N	↑	↑ N	↓ N	N	N	N ↓	N ↑ ↓
Hyperthyroidism (cat)	↑	↑	N	↑	N ↑	N ↑ ↓	N	N	N ↓	N ↑
Raisin/grape toxicity (dog)	↑	—	N	↑	N ↑	—	—	—	—	—

↓, *Decreased concentration;* ↑, *increased concentration;* N, *normal;* tCa, *serum total calcium;* iCa, *serum ionized calcium;* alb, *albumin;* Corr tCa, *corrected total calcium;* Pi, *inorganic phosphorus;* PTH, *parathyroid hormone;* PTHrP, *parathyroid hormone–related protein;* 25(OH)-D, *25-hydroxyvitamin D;* 1,25(OH)2-D, *1,25-dihydroxyvitamin D;* PTG, *parathyroid gland;* ULS, *ultrasound.*

Hypoadrenocorticism

Hypoadrenocorticism is the second most common cause of hypercalcemia in dogs (after malignancy), accounting for 11% to 45% of cases in five studies,[111,157,290,534,560] but no cases were reported in one study.[41] Hypercalcemia was reported in 28% to 31% of dogs with glucocorticoid- and mineralocorticoid-deficient hypoadrenocorticism,[402,405] in some dogs with glucocorticoid-deficient hypoadrenocorticism,[302] and in 1 of 10 cats.[403] Hypoadrenocorticism is rarely recognized in cats, and hypercalcemia is present in only 8% of cases.[32] Hypercalcemia was present in one cat with iatrogenic secondary hypoadrenocorticism and diabetes mellitus.[495] Magnitude of hypercalcemia was greatest in the most severely affected dogs, but the mechanism is unknown. A correlation between the degree of hyperkalemia and hypercalcemia was detected when the serum potassium concentration was greater than 6.0 to 6.5 mEq/L, and serum tCa concentration was often 11.4 to 13.5 mg/dL.[168] Increases in serum iCa may or may not develop in hypoadrenocorticism.[543] Serum tCa concentration rapidly returns to normal after 1 to 2 days of corticosteroid replacement therapy in dogs,[402] and IV volume expansion can return serum calcium concentration to normal within a few hours. Hypoadrenocorticism should always be included in the differential diagnosis of hypercalcemia because clinical signs of hypoadrenocorticism and hypercalcemia are similar.

Chronic Renal Failure

The finding of hypercalcemia and primary renal azotemia poses a special diagnostic problem because hypercalcemia can cause renal failure or develop as a consequence of CRF. Serum PTH concentration is often increased in patients with hypercalcemia related to renal failure, and these animals must be differentiated from those with primary hyperparathyroidism. Serum iCa concentration is increased in primary hyperparathyroidism but is usually normal or low in patients with CRF.[114,290]

Deleterious effects of hypercalcemia occur in patients with renal failure only if it is associated with increases in serum iCa concentration. Consequently, clinical signs of hypercalcemia are uncommon in CRF patients, and measurement of serum iCa concentration to assess calcium status in CRF patients is critical. In CRF patients, the serum tCa measurement incorrectly assessed iCa status in 36% of dogs and 32% of cats.[474,475] The use of the “adjusted tCa” value incorrectly assessed iCa status in approximately 53% of dogs with CRF. In dogs, serum tCa measurement or adjusted tCa measurement overestimated hypercalcemia and underestimated hypocalcemia. In cats with CRF, serum tCa measurement overestimated normocalcemia and underestimated hypercalcemia. Thus to accurately assess calcium status in patients with CRF, iCa concentration must be directly measured.

Fewer than 10% of all dogs with CRF have increased serum iCa concentrations. In one study, approximately 6% exhibited ionized hypercalcemia.[114] In a recent study of 490 dogs with CRF, 9% exhibited hypercalcemia, 55% were normocalcemic, and 36% were hypocalcemic based on serum iCa concentrations.[475] Cats with CRF appear to have a higher incidence of ionized hypercalcemia as compared with dogs. In 102 cats with CRF, 29% were hypercalcemic, 61% were normocalcemic, and 10% were hypocalcemic based on iCa concentration.[474]

Many dogs and cats with CRF have normal serum tCa concentrations.[138,173,345] Hypercalcemia based on measurement of serum tCa concentration occurs sporadically in dogs and cats with CRF and is usually listed as second or third in frequency of causes of hypercalcemia in dogs. Elevated tCa occurs in up to 14% of dogs with CRF, with a range of 12.1 to 15.2 mg/dL.[114,173,290,367] In 71 hypercalcemic cats, CRF was noted in 38%.[467] In cats with CRF, the reported incidence of serum total hypercalcemia ranged from 11.5%[138] to 58%.[24]

The incidence of elevated tCa increases with severity of azotemia. In 73 cats with CRF, serum tCa was increased in 8%, 18%, and 32% of those with mild, moderate, or severe azotemia, respectively.[24] However, increases in serum iCa do not show a strong association with the degree of azotemia.[127] In 47 of the previous 73 cats with CRF, iCa was increased in 0%, 9%, and 6% of those with mild, moderate, or severe azotemia, respectively.[24] Hypercalcemia was also not correlated with serum phosphorus concentration in dogs with experimental renal failure.[381,531]

The parathyroid glands must be present for hypercalcemia to develop,[531] and partial parathyroidectomy ameliorates hypercalcemia in some dogs with CRF.[173] Treatment of dogs with CRF and hypercalcemia with low-dose calcitriol to reduce PTH synthesis and secretion can result in decreased iCa concentration. Low-dose calcitriol therapy does not appreciably increase intestinal calcium absorption.[363,364] In patients with CRF, increased serum PTH concentration (renal secondary hyperparathyroidism) contributes to the progression of renal disease.[364] Oral administration of low doses of calcitriol reduces toxic concentrations of PTH, improves quality of life, reduces progression of renal disease, and leads to prolongation of life.[365,479]

Some cases of ionized hypercalcemia and CRF may be associated with the use of calcium carbonate intestinal phosphate binders. In these cases, serum iCa concentration rapidly returns to normal after discontinuation of treatment. In humans with CRF, therapeutic use of calcitriol is limited by development of hypercalcemia in patients also being treated with calcium-based dietary phosphorus binders.[121,365] In veterinary medicine, use of aluminum-based phosphorus binders or sevelamer (Renagel, Genzyme Corporation, Cambridge, MA) largely precludes this problem.[9] "Noncalcemic analogues" of calcitriol have been developed for use in humans,[493] such as paricalcitol (Zemplar, Abbott Laboratories, Abbott Park, IL), 22-oxa-calcitriol (OCT), and doxercalciferol (Hectorol, Bone Care International, Middleton, WI).[146] These analogues have a very short half-life (several minutes), and this short half-life is responsible for their weak stimulation of intestinal calcium absorption. Doses of noncalcemic analogues needed to suppress PTH synthesis are approximately eightfold higher than that of calcitriol[493] and are up to 12 times the cost. If hypercalcemia develops with calcitriol therapy, a twice-weekly dosing strategy of calcitriol is used. This dosing regimen will suppress PTH but be much less effective at stimulating intestinal calcium absorption. Noncalcemic analogues are not needed and are financially impractical in veterinary medicine.

Ionized hypercalcemia occurs in patients with CRF who receive excessive doses of calcitriol. Hypercalcemia is very uncommon in animals treated with the lower dosages of calcitriol (2.5 to 4.0 ng/kg daily). If hypercalcemia is caused by excessive calcitriol, the serum tCa concentration decreases during the week after its discontinuation. Most CRF patients who develop an elevated tCa during low-dose calcitriol treatment have normal or low serum iCa concentrations. Serum tCa concentration may not decrease when calcitriol is discontinued if the increased serum tCa concentration is caused by increased complexed calcium.

The mechanisms of increased serum tCa concentration in CRF have not been well characterized.[173,290,435,531] In dogs with CRF, serum total hypercalcemia, and normal iCa concentrations, the increase in serum tCa is caused by an increase in the complexed calcium fraction.[473] In CRF, organic anions such as citrates, phosphates, lactates, bicarbonates, and oxalates are capable of complexing with calcium. Complexed calcium accounted for 24% of serum tCa in those dogs with CRF and elevated serum tCa as compared with 11% in those dogs with CRF and low serum tCa. Increased PTH-mediated bone resorption as a consequence of CRF could increase serum tCa concentration. If elevated iCa is also present, then reduced GFR caused by loss of renal mass could cause increased iCa concentration as the filtered load of calcium declines. Hyperplasia of parathyroid gland chief cells could account for increased PTH secretion and serum calcium concentration because chief cells secrete small amounts of PTH that are nonsuppressible regardless of serum iCa concentration.[201]

Tertiary hyperparathyroidism refers to the condition of a subset of patients with CRF who develop ionized hypercalcemia and excessive PTH secretion that is not inhibited by high serum iCa concentration. It is likely that such patients had high PTH concentrations in association with normal or low serum iCa concentration (renal secondary hyperparathyroidism) earlier in the clinical course of CRF. Autonomous secretion of PTH from the parathyroid gland is unlikely, but the set-point for PTH secretion may be altered in CRF such that higher concentrations of iCa are necessary to inhibit PTH secretion.[202] Decreased serum calcitriol concentrations, decreased numbers of calcitriol receptors in the parathyroid gland, and decreased calcitriol–VDR interactions with chief cell DNA caused by uremic toxins may contribute to this increase in set-point,[70,247,396] as may decreased levels of the calcium receptor, which both establishes the set-point and depends on calcitriol functionality for synthesis of its mRNA from the parathyroid cells' DNA.[94] Ten dogs with CRF and increased serum tCa concentration were compared with those with normal serum tCa concentration (Fig. 6-12). Serum amino-terminal PTH concentration was markedly increased in both groups of uremic dogs, but those with increased tCa had higher PTH concentrations. Calcitriol concentration was decreased to a similar extent in both groups. It was proposed that the hypercalcemic and more markedly hyperparathyroid uremic dogs might have had greater calcitriol receptor (VDR) deficits in their parathyroid cells, which would lead to poorly controlled PTH synthesis and chief cell hyperplasia.[367] Deficient calcitriol functionality caused by VDR deficits would also lead to calcium receptor deficits and the "set-point" elevations involved in the observed hypercalcemia.[94]

Aluminum accumulation in the development of hypercalcemia in dogs or cats with renal disease being treated with aluminum-containing intestinal phosphate binders has not been investigated despite the fact that such treatment is common. Experimental dogs exposed to aluminum developed mild hypercalcemia within minutes of a single intravenous injection. During chronic daily exposure to aluminum during a period of weeks, serum calcium concentration progressively increased, and azotemia developed.[226]

Two of 15 cats with CRF developed hypercalcemia while eating a phosphate-restricted veterinary diet designed for treatment of renal failure. Hypercalcemia in these cats was associated with a decrease in serum phosphorus and low or undetectable PTH concentrations. Serum calcium returned to normal, and PTH and phosphorus increased with the feeding of a maintenance diet.[26]

PATHOLOGIC OR CONSEQUENTIAL HYPERCALCEMIA

Cancer-Associated Hypercalcemia

The most common cause of hypercalcemia in dogs is cancer-associated hypercalcemia. Cancer is third in frequency of association with hypercalcemia in cats. There are three mechanisms (Fig. 6-13) of increased serum calcium concentration induced by neoplasms: (1) HHM, (2) hypercalcemia induced by metastases of solid tumors

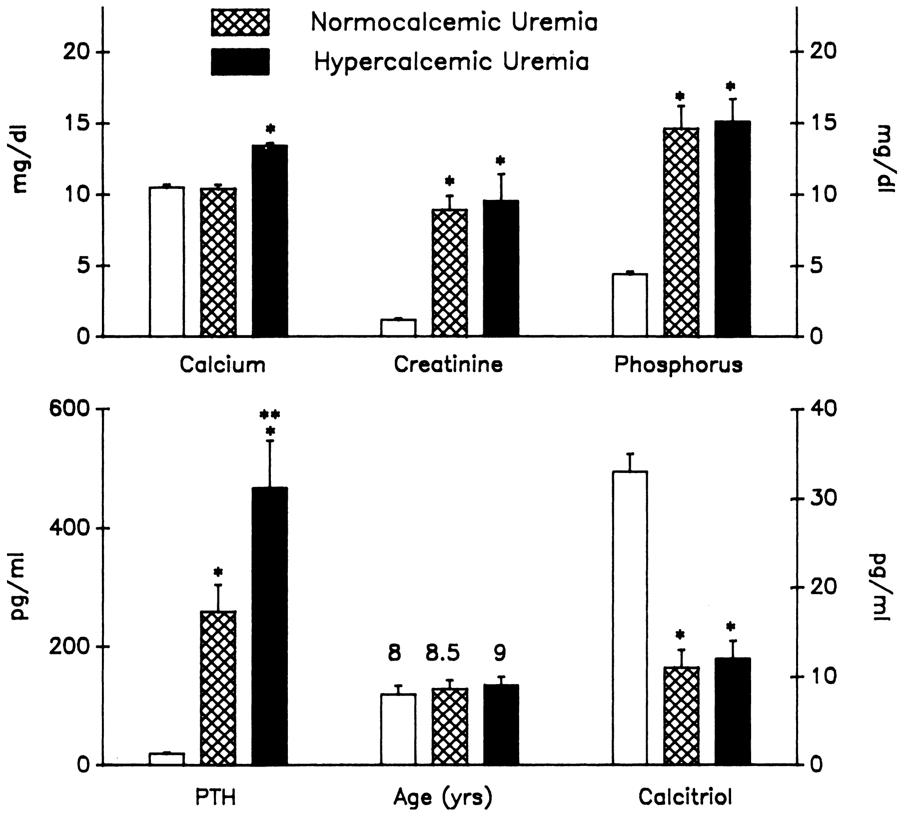

Fig. 6-12 Comparison of biochemical data for dogs with renal failure and hypercalcemia or normocalcemia. Dogs with renal failure were normalized for age and had similar concentrations of serum creatinine, phosphorus, and calcitriol. Serum concentrations of PTH were greater in the hypercalcemic dogs than in the normocalcemic dogs. Data are mean ± SEM. For normal and hypercalcemic uremic dogs, n = 10; for normocalcemic uremic dogs, n = 20. Significant differences were *P < 0.0001 (from normal) and **P < 0.02 (from normocalcemic uremia PTH) by Student's t test. (From Nagode LA, Steinmeyer CL, Chew DJ, et al.: Hyper- and normo-calcemic dogs with chronic renal failure: relations of serum PTH and calcitriol to parathyroid gland Ca^{++} set-point. In Norman AW, Schaefer K, Grigoleit HG, et al, editors: *Vitamin D 1988. Chemical, biochemical and clinical endocrinology,* Berlin, 1988, Walter de Gruyter & Co., pp. 799-800.)

to bone (local osteolytic hypercalcemia [LOH]), and (3) hematologic malignancies growing in the bone marrow (LOH).[436,437]

Humoral Hypercalcemia of Malignancy. HHM is a syndrome associated with many tumors in people and animals.[437] Characteristic clinical findings in patients with HHM include hypercalcemia, hypophosphatemia, hypercalciuria (often with decreased fractional calcium excretion), increased fractional excretion of phosphorus, increased nephrogenous cyclic adenosine monophosphate (cAMP), and increased osteoclastic bone resorption. Hypercalcemia is induced by humoral effects on bone, kidney, and possibly the intestine (Fig. 6-14).[438] Increased osteoclastic bone resorption is a consistent finding in HHM and increases calcium release from bone. The kidney plays a critical role in the pathogenesis of hypercalcemia because PTHrP stimulates calcium reabsorption, which binds and activates renal PTH-PTHrP receptors. The level of renal function in the patient may also contribute to the development of hypercalcemia. Animals with dehydration or impaired renal function are more susceptible to developing hypercalcemia or may have more severe hypercalcemia because of decreased renal excretion of calcium. In some forms of HHM, increased serum 1,25-dihydroxyvitamin D concentrations may increase calcium absorption from the intestine.[446]

Malignancies that are commonly associated with HHM in dogs include T-cell lymphoma and adenocarcinomas derived from the apocrine glands of the anal sac.[33,436,553,561] Dogs with cancer and HHM are expected to have shorter survival. In addition, sporadic cases of HHM occur in dogs with thymoma, myeloma,

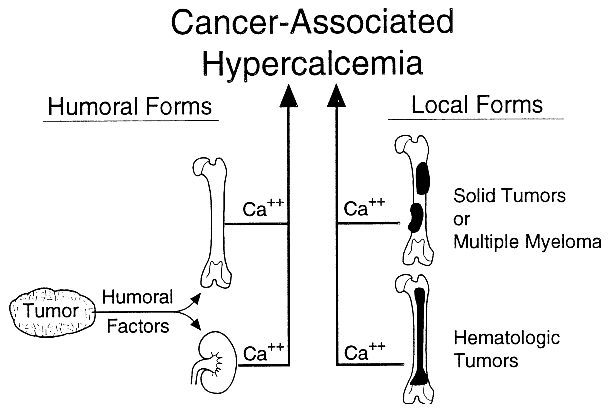

Fig. 6-13 Pathogenesis of cancer-associated hypercalcemia. Humoral and local forms of cancer-associated hypercalcemia increase circulating concentrations of calcium by stimulation of osteoclastic bone resorption and increased renal tubular reabsorption of calcium.

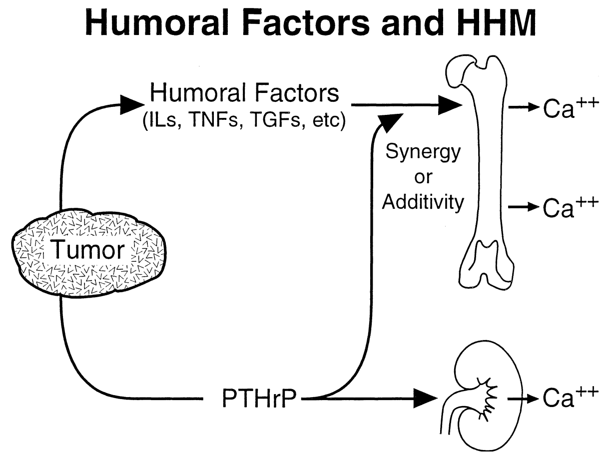

Fig. 6-14 Humoral factors such as parathyroid hormone–related protein (PTHrP), interleukin-1 (IL-1), tumor necrosis factors (TNFs), or transforming growth factors (TGFs) produced by tumors induce humoral hypercalcemia of malignancy (HHM) by acting as systemic hormones and stimulating osteoclastic bone resorption or increasing tubular reabsorption of calcium.

melanoma, or carcinomas originating in the lungs, pancreas, thyroid gland, skin, mammary gland, nasal cavity, and adrenal medulla.[56,417,436-438] Tumors associated with hypercalcemia in cats include lymphosarcoma, multiple myeloma, squamous cell carcinoma, bronchogenic carcinoma/adenocarcinoma, osteosarcoma, fibrosarcoma, undifferentiated sarcoma, undifferentiated renal carcinoma, anaplastic carcinoma of the lung and diaphragm, and thyroid carcinoma.* Lymphosarcoma and squamous cell carcinoma are the two most common causes of hypercalcemia in cats.[467] Of 11 hypercalcemic cats with lymphosarcoma, two each had renal, generalized, gastrointestinal, or mediastinal involvement, and one each had laryngeal, nasal, or cutaneous disease.[56,115,152,158,467] Squamous cell carcinoma has been found in mandibular, maxillary, pulmonary, and ear canal locations.[56,250,276,467]

Excessive secretion of biologically active PTHrP plays a central role in the pathogenesis of hypercalcemia in most forms of HHM, but cytokines such as IL-1, TNF-α, and transforming growth factor (TGF)-α and -β or calcitriol may have synergistic or cooperative actions with PTHrP (see Fig. 6-14). Before PTHrP was identified, it was recognized that tumors associated with HHM induced a syndrome that mimicked primary hyperparathyroidism with secretion of a PTH-like factor that was antigenically unrelated to PTH.[359,552]

PTHrP binds to the N-terminal PTH-PTHrP receptor in bone and kidney but does not cross-react immunologically with native PTH (Fig. 6-15). PTHrP stimulates adenylyl cyclase and increases intracellular calcium in bone and kidney cells by binding to and activating the cell membrane PTH-PTHrP receptors. This binding results in stimulation of osteoclastic bone resorption, increased renal tubular calcium reabsorption, and decreased renal tubular phosphate reabsorption. IL-1 stimulates bone resorption in vivo and in vitro and is synergistic with PTHrP.[335,437] TGF-α and -β can stimulate bone resorption in vitro and have been identified in tumors associated with HHM, including adenocarcinomas derived from apocrine glands of the anal sac in dogs.[340]

Lymphoma. Hypercalcemia is found in 20% to 40% of dogs with lymphoma (Fig. 6-16).[297,316] Most dogs with lymphoma and hypercalcemia have HHM because increased osteoclastic resorption is present in bones without evidence of tumor metastasis. Lymphoma is an uncommon cause of mild HHM in ferrets.[268] Lymphomas associated with HHM are usually of the T-cell type.[553] T-cell lymphoma occurred in 22% of dogs with lymphoma, and hypercalcemia only occurred in dogs with CD4$^+$ lymphoma in one study.[459] The pathogenesis of hypercalcemia in dogs with lymphoma and HHM resembles that occurring in humans with lymphoma or leukemia induced by human T-cell lymphotropic virus type I (HTLV-I). Neoplastic cells from humans with HTLV-I–induced lymphoma have increased PTHrP production.[428]

Most dogs with lymphoma and hypercalcemia have T-cell lymphoma.[511,553] Dogs with T-cell lymphoma were significantly more likely to have early relapse and death compared with those with B-cell lymphoma. Shorter remissions and survival times have been noted by others for T-cell lymphoma compared with B-cell lymphoma in dogs.[211] In another study, 46 (32.8%) of 140

**References 11,42,56,115,152,158,229,250,276,467,484.*

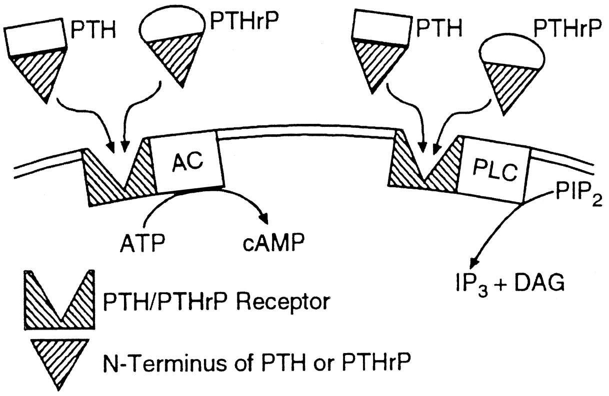

Fig. 6-15 Parathyroid hormone–related protein (PTHrP) induces many of the effects of parathyroid hormone (PTH) by interacting with the PTH receptor in bone and kidney and activating adenylyl cyclase (AC) to form cyclic AMP (cAMP) and phospholipase C (PLC) to form inositol triphosphate (IP_3) and diacylglycerol (DAG) from phosphatidylinositol (PIP_2). Stimulation of the PTH receptor results in increased osteoclastic bone resorption and renal tubular reabsorption of calcium, inhibition of renal tubular reabsorption of phosphorus, and stimulation of renal production of 1,25-dihydroxyvitamin D_3 (calcitriol).

lymphomas were classified as T cell in origin, and 16 of these dogs (35%) were hypercalcemic.[185] In 37 dogs with lymphoma and hypercalcemia, calcium concentration was not related to prognosis; mean remission was 10.4 months, and median remission was 6 months.[434] The presence of a mediastinal mass had an adverse effect on remission in these hypercalcemic dogs. Serum tCa concentration may return to normal despite minimal reduction in tumor mass following chemotherapy, as happened in 5 of 12 dogs with lymphoma and initial hypercalcemia.[556] The finding of hypercalcemia in dogs with lymphoma was not prognostic for survival or time to remission, but T-cell origin lymphoma did adversely affect prognosis.[275,511,538]

Most dogs with lymphoma and HHM have increased circulating PTHrP concentrations, but concentrations are lower than in dogs with carcinomas and HHM, and PTHrP concentrations are not correlated with serum calcium concentration (Fig. 6-17).[446] These findings indicate that PTHrP is an important marker of HHM in dogs with lymphoma but is not the sole humoral factor responsible for stimulation of osteoclasts and development of hypercalcemia. It is likely that cytokines such as IL-1 or TNF function synergistically with PTHrP to induce HHM in dogs with lymphoma (see Fig. 6-14).[436,437]

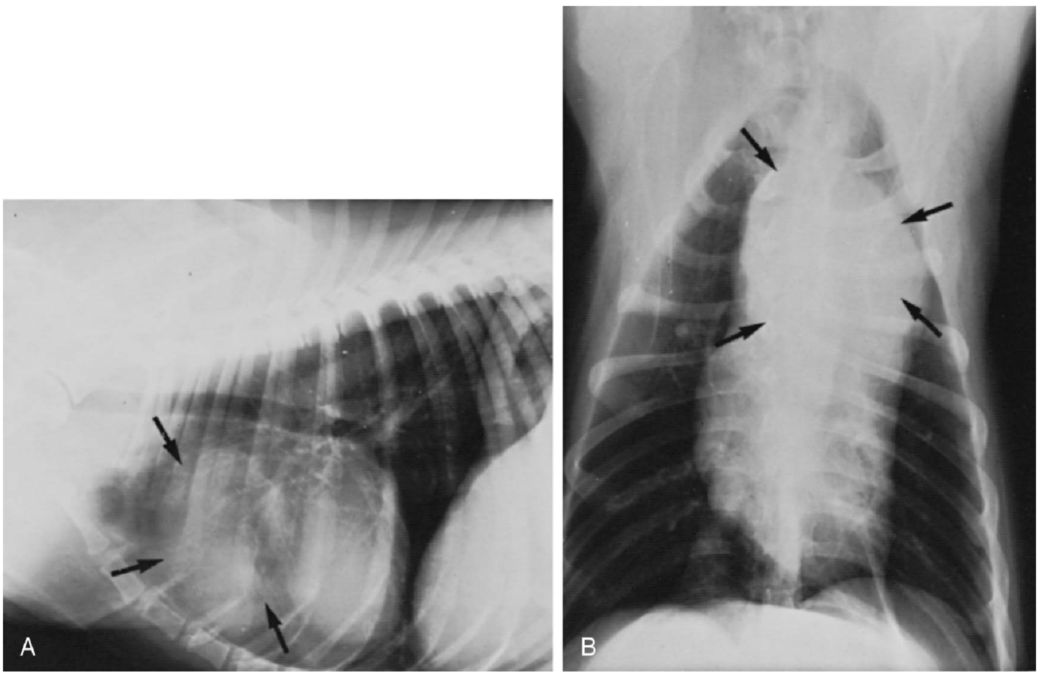

Fig. 6-16 Lateral **(A)** and ventrodorsal **(B)** thoracic radiographs of a 5-year-old boxer dog with hypercalcemia of malignancy caused by mediastinal lymphoma (arrows). Severe hypercalcemia (serum total calcium concentration, 20.6 mg/dL) was detected on initial presentation. (From Chew DJ, Carothers M: Hypercalcemia, *Vet Clin North Am* 19:272, 1989.)

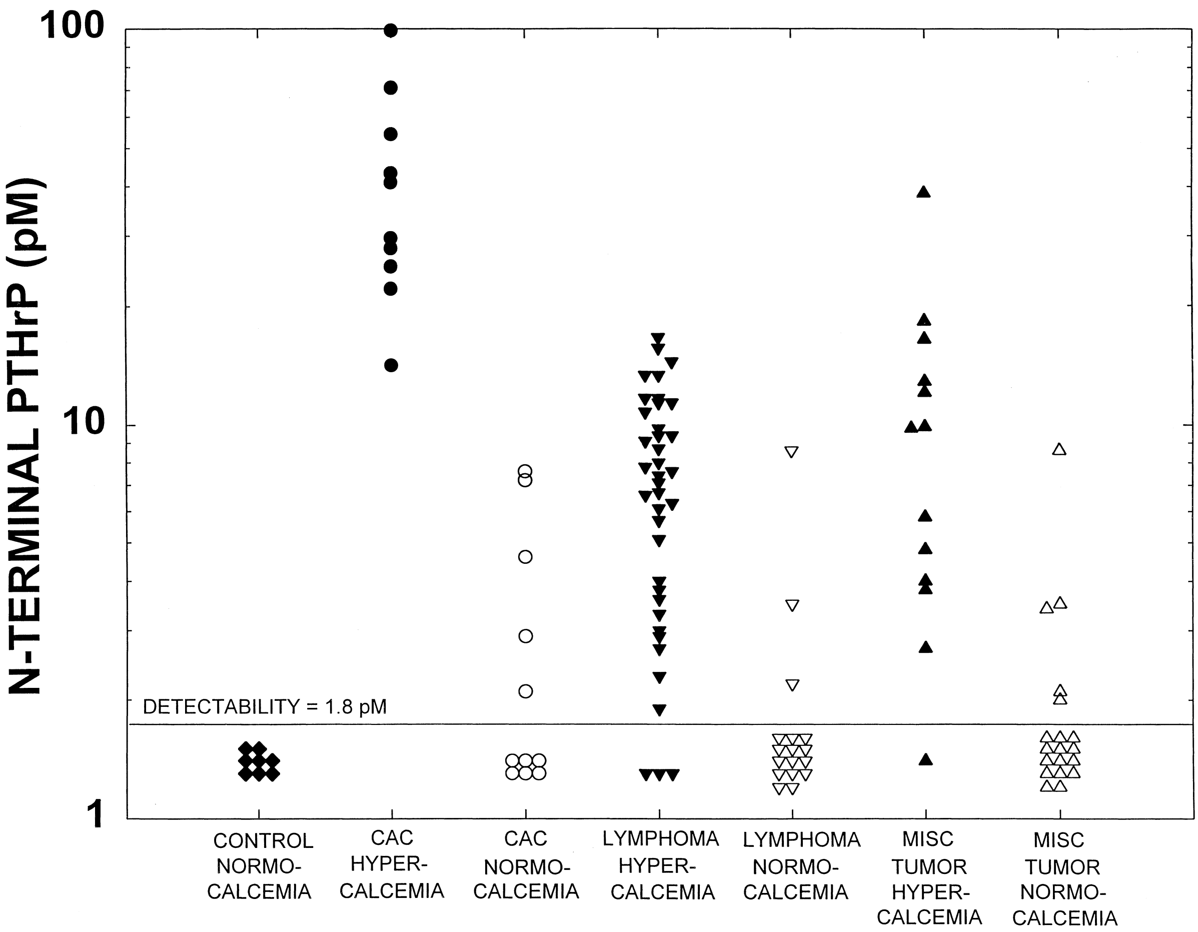

Fig. 6-17 Circulating N-terminal parathyroid hormone–related protein (PTHrP) concentrations in normal dogs (CONTROL); dogs with hypercalcemia (>12 mg/dL) and anal sac adenocarcinomas (CAC), lymphoma, or miscellaneous tumors (MISC TUMOR); and dogs with normocalcemia (<12 mg/dL) and anal sac adenocarcinomas, lymphoma, or miscellaneous tumors. (From Rosol TJ, Nagode LA, Couto CG, et al: Parathyroid hormone–related protein, parathyroid hormone, and 1,25-dihydroxyvitamin D in dogs with cancer-associated hypercalcemia, *Endocrinology* 131:1157, 1992. ©The Endocrine Society.)

Some dogs and human patients with lymphoma and hypercalcemia have increased serum calcitriol concentrations, which may contribute to the induction of hypercalcemia.[446,482] Some lymphocytes contain the 1α-hydroxylase (similar to that found in renal tubules) that converts 25-hydroxyvitamin D to the active metabolite 1,25-dihydroxyvitamin D (calcitriol). Therefore lymphomas that retain this capability may synthesize excessive calcitriol, which could increase calcium absorption from the intestinal tract and facilitate development of hypercalcemia.

An early report indicated that a mediastinal mass was detected in most dogs with lymphoma and hypercalcemia.[343] However, a recent report indicates that the presence of a cranial mediastinal mass was not required for development of hypercalcemia in dogs, and mediastinal masses were not disproportionately more common in those dogs with hypercalcemia.[49]

Canine Adenocarcinoma Derived from Apocrine Glands of the Anal Sac. The adenocarcinoma derived from apocrine glands of the anal sac of dogs consistently fulfills the criteria for HHM.[342,344,430] This tumor appears primarily in middle-aged (mean, 10 years) dogs and rarely metastasizes to bone. Clinical signs are referable to hypercalcemia (polyuria, polydipsia, anorexia, and weakness), a mass in the perineum (tenesmus, ribbonlike stools, increased odor, and protruding mass), a mass in the sublumbar region, or more distant metastases. Apocrine adenocarcinomas often require rectal and anal sac palpation to confirm their presence because their size ranges from 7 mm to 6 × 8 cm (Fig. 6-18). Dogs

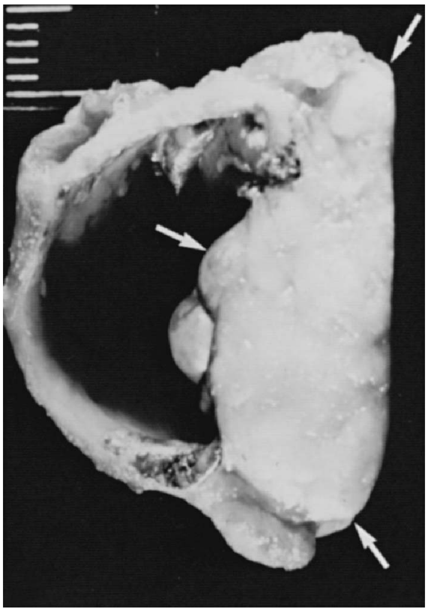

Fig. 6-18 Hypercalcemia of malignancy associated with apocrine gland adenocarcinoma of the anal sac in an elderly female dog. Transverse section of the anal sac and associated malignancy (arrows). (From Chew DJ, Meuten DJ: Disorders of calcium and phosphorus metabolism, *Vet Clin North Am* 12:417, 1982.)

with this tumor and HHM have hypercalcemia (tCa, 12 to 24 mg/dL); hypophosphatemia; decreased immunoreactive PTH concentration; increased urinary excretion of calcium, phosphorus, and cAMP; and increased osteoclastic bone resorption.[33,344,561] This tumor should not be confused with the common perianal adenomas or the uncommon perianal adenocarcinomas that arise from the circumanal glands and have entirely different biologic behavior. Perianal adenomas and adenocarcinomas affect primarily male dogs and are not associated with hypercalcemia.[539]

Hypercalcemia was present at the time of diagnosis in 80% to 100% of affected dogs in early studies.[330,331] Recent reports in dogs with earlier detection note the incidence of hypercalcemia to be lower, at 33%,[453] 27%,[561] and 53% of cases.[33] Early reports also noted a strong bias toward the occurrence of this tumor in female dogs, but equal sex distribution has been more recently noted.[561] In some instances, the finding of hypercalcemia during routine serum biochemistry testing prompts rectal palpation and subsequent discovery of an apocrine gland adenocarcinoma. Surgical removal or radiation therapy of the adenocarcinoma results in rapid return to normal of serum calcium and phosphorus concentrations, increased serum PTH concentration, and decreased calcitriol concentration.[446] Postsurgical survival of dogs with apocrine gland adenocarcinoma and hypercalcemia ranged from 2 to 21 months, with a mean of 8.8 months. Sublumbar metastases occur in a high percentage (72%) of affected dogs and are associated with recrudescence of the biochemical alterations in serum and urine.[33] In one study, dogs with hypercalcemia and anal sac adenocarcinoma had shorter survival times compared with normocalcemic dogs with this tumor (356 versus 584 days)[561]; in another study, survival was not influenced by the presence of hypercalcemia.[33]

Most dogs with HHM have increased concentrations of circulating PTHrP (see Fig. 6-17). Plasma concentrations of PTHrP are highest (10 to 100 pmol/L) in dogs with apocrine adenocarcinomas of the anal sac and sporadic carcinomas associated with HHM.[446] Serum calcium concentrations in affected dogs correlate well with circulating PTHrP concentrations, which is consistent with the concept that PTHrP plays a primary role in the pathogenesis of HHM in these dogs. Dogs with apocrine adenocarcinomas and normocalcemia may have increased plasma PTHrP concentrations (2 to 15 pmol/L), but the concentrations are lower than in dogs with hypercalcemia.

Some dogs with apocrine adenocarcinomas have inappropriate concentrations (normal or increased) of calcitriol for the degree of hypercalcemia.[446] This finding suggests that the humoral factors produced by the neoplastic cells are capable of stimulating renal 1α-hydroxylase and increasing the formation of calcitriol even in the presence of increased serum calcium concentration. PTH concentrations were not increased in hypercalcemic dogs and were significantly lower than those observed in dogs with primary hyperparathyroidism. Parathyroid glands from dogs with apocrine adenocarcinoma were atrophic or inactive, and there was nodular hyperplasia of C cells in the thyroid glands because of prolonged hypercalcemia.[344]

Hematologic Malignancies. Some types of hematologic malignancies present in the bone marrow produce hypercalcemia by inducing bone resorption locally.[436,437] This effect occurs most commonly in multiple myeloma and lymphoma. Hypercalcemia has been reported in 17% of dogs with multiple myeloma.[331] A number of paracrine factors or cytokines may be responsible for the stimulation of bone resorption in this setting. The cytokines most often implicated in the pathogenesis of local bone resorption are IL-1, TNF-α, and TNF-β (lymphotoxin).[328,360] Other cytokines or factors that may play a role include IL-6, TGF-α and -β, and PTHrP.[48] Production of small amounts of PTHrP by a tumor in bone may stimulate local bone resorption without inducing a systemic response. Prostaglandins (especially prostaglandin E_2) may also be responsible for local stimulation of bone resorption.

Some dogs with lymphoma and hypercalcemia have localized bone resorption associated with metastases to medullary cavities without evidence of increased bone resorption at sites distant from the tumor metastases.[343] Hypercalcemic dogs with lymphoma and bone metastases had decreased PTH and calcitriol concentrations, increased excretion of hydroxyproline, calcium, phosphorus, and increased concentrations of the prostaglandin E_2 metabolite 13,14-dihydro-15-ketoprostaglandin E_2. Prostaglandin E_2 may be an important local mediator of bone resorption in these dogs. Other potential mediators include IL-1 and TNFs.

Tumors Metastatic to Bone. Solid tumors that metastasize widely to bone can produce hypercalcemia by the induction of local bone resorption associated with tumor growth. This is not common in animals but is an important cause of cancer-associated hypercalcemia in humans.[436,450,451] Tumors that often metastasize to bone and induce hypercalcemia in human patients include breast and lung carcinomas. Carcinomas of the mammary gland, prostate, liver, and lung were most frequently reported to metastasize to bone in dogs, and the humerus, femur, and vertebrae were the most common sites of metastasis.[345,452] Primary bone tumors are not often associated with hypercalcemia in dogs or cats.

The pathogenesis of enhanced bone resorption is not well understood, but two primary mechanisms are secretion of cytokines or factors that stimulate local bone resorption and indirect stimulation of bone resorption by tumor-induced cytokine secretion from local immune or bone cells.[195] Cytokines or factors that may be secreted by tumor cells and stimulate local bone resorption include PTHrP,[416] TGF-α and -β, and prostaglandins (especially prostaglandin E_2). In some cases, bone-resorbing activity can be inhibited by indomethacin, which suggests that prostaglandins are either directly or indirectly associated with stimulation of bone resorption. The cytokines most often implicated in indirect stimulation of bone resorption by local immune cells include IL-1 and TNFs.

Malignant neoplasms with osseous metastases may cause moderate to severe hypercalcemia and hypercalciuria, but serum ALP activity and phosphorus concentrations are usually normal or only moderately increased. It is believed these changes are caused by release of calcium and phosphorus into the blood from areas of bone destruction at rates greater than can be cleared by the kidney and intestine. Bone involvement can be multifocal but is usually sharply demarcated and localized to the area of metastasis.

Primary Hyperparathyroidism

Primary hyperparathyroidism is an uncommon cause of hypercalcemia in dogs[37,82] and is even less common in cats.[133,263] In hypercalcemic cats, primary hyperparathyroidism was found in 4 of 71 cases.[467] Excessive and inappropriate secretion of PTH by the parathyroid glands relative to the serum iCa concentration characterizes this condition. Primary hyperparathyroidism was caused by a solitary parathyroid gland adenoma in approximately 90% of dogs, whereas parathyroid gland carcinoma and parathyroid gland hyperplasia each accounted for 5% of cases in one large series.[168] Adenomas occurred with nearly equal frequency in the external and internal parathyroid glands in one study,[37] but external gland adenomas predominated in another report in dogs.[565] Idiopathic parathyroid gland hyperplasia may affect one or more glands and has been reported in six older dogs.[135] Although remnant parathyroid tissue may be found in the cranial mediastinum near the base of the heart, neoplastic transformation has not been reported at this site in dogs or cats. An ectopic parathyroid gland adenoma cranial to the thoracic inlet has been described in one dog.[564] In cats, the underlying lesion is typically benign, owing to an adenoma, bilateral cystadenomas, or hyperplasia,[133,163,467,503] but unilateral or bilateral carcinomas have also been diagnosed.[168,263,326,412]

Primary parathyroid gland hyperplasia has been reported in two German shepherd dog puppies.[517] Diffuse hyperplasia was present in all four parathyroid glands. In retrospect, this family of German shepherd dogs probably had an inactivating mutation in the gene for the calcium-sensing receptor. Mutations in one or both of the calcium-sensing receptor genes in humans result in familial hypocalciuric hypercalcemia or neonatal severe hypercalcemia, respectively, because of inadequate ability to sense extracellular calcium concentration and coordinate the appropriate cellular response.[414] The affected puppies had a disease syndrome that mimicked neonatal severe hypercalcemia in humans. Neonatal severe hypercalcemia is lethal unless total parathyroidectomy is performed early in life to markedly reduce increased PTH concentrations.

Dogs with primary hyperparathyroidism are older, with a mean age of 10.5 years (range, 5 to 15 years).[168] The mean age in affected cats was 12.9 years (range, 8 to 15 years).[263] No sex predisposition has been noted, but keeshonds constituted 36% of affected dogs, and five of eight cats were Siamese.[345] Parathyroid gland masses usually cannot be palpated in dogs, but 50% of cats with primary hyperparathyroidism had a palpable cervical mass.[133,263] Clinical signs related to hypercalcemia are either mild (e.g., lethargy, polydipsia, polyuria, and weakness) or absent in many affected dogs.[37,168] In one study, most owners of affected dogs were not convinced that their dogs had a serious illness,[37] but some owners retrospectively recognized subtle signs after hypercalcemia resolved.[168] More prominent clinical signs and serious consequences can occur when hyperparathyroidism and severe hypercalcemia are long standing and associated with renal failure.[111,112] Clinical signs referable to the lower urinary tract have been reported to occur in

27% of dogs as a result of urolithiasis or bacterial urinary tract infection.[168] Calcium-containing uroliths (calcium phosphate, calcium oxalate, or mixtures) occurred in approximately 30% of dogs and in a cat with primary hyperparathyroidism.[168,277,326] Urolithiasis is attributed to hypercalcemia and subsequent hypercalciuria. Interestingly, hypercalcemia arising from other causes has not been associated with urolithiasis except in cats with idiopathic hypercalcemia (IHC).[476]

The diagnostic workup to confirm primary hyperparathyroidism often begins with the fortuitous finding of increased serum calcium concentration on routine clinical chemistry testing.[168] The diagnosis of primary hyperparathyroidism is easy in dogs and cats that have increased serum tCa concentration, normal renal function, and increased concentration of immunoreactive PTH. The appropriateness of the PTH concentration must be interpreted in relation to the serum iCa concentration. Additional support for the diagnosis of primary hyperparathyroidism is provided by the finding of increased serum iCa concentration, increased serum ALP, low serum phosphorus concentration, increased or normal calcitriol concentration, undetectable PTHrP, and calcium-containing uroliths. The most consistent laboratory abnormality in dogs with primary hyperparathyroidism is increased serum calcium concentration.[168]

Hypercalcemia results from a combination of effects following PTH binding to receptors in kidney and bone. PTH also acts indirectly to increase serum iCa concentration by enhancing renal conversion of 25-hydroxyvitamin D to calcitriol. Hypophosphatemia secondary to PTH-enhanced urinary excretion of phosphorus was observed in 5 of 21 dogs.[37] Serum phosphorus concentration is typically low,[168] and calcitriol concentrations were mildly increased or in the high-normal range in three of four dogs with primary hyperparathyroidism.[446]

The diagnosis of primary hyperparathyroidism is more challenging when PTH is within the reference range. A PTH concentration in the upper part of the reference range in association with hypercalcemia is inappropriate. Confirmed primary hyperparathyroidism has been noted in dogs and cats with hypercalcemia and reference range PTH concentrations.[168,263] In a cat with persistent hypercalcemia related to primary hyperparathyroidism, PTH concentration was increased on two occasions but within the reference range on five other occasions.[133] PTH concentrations measured in blood collected from either the left or right jugular vein did not differ, and sampling from a specific side was not valuable for localizing the site of an enlarged parathyroid gland.[170] Circulating PTHrP concentrations were undetectable in six dogs with primary hyperparathyroidism.[446]

Ultrasonography of the neck is helpful in the diagnosis of primary hyperparathyroidism in dogs and cats, but it requires an ultrasound unit with a high-frequency (7.5- to 10-MHz) transducer to achieve the necessary level of resolution rather than the widely available 5- or 7.5-MHz units used for abdominal studies.[168,564] With a 10-MHz linear transducer, the parathyroid glands of normal dogs can routinely be identified especially in larger dogs.[426] Parathyroid gland masses greater than 5 mm can usually be identified, and some masses as small as 2 mm may be detected. Enlarged parathyroid glands are expected to be hypoechoic or anechoic, well marginated, and easily contrasted with thyroid tissue. False-positive results are rare, but false-negative findings may occur. Ultrasonography correctly identified the presence and location of a solitary parathyroid gland mass in 10 of 11 dogs in a prospective study in which the mass was confirmed at surgery.[170] Sonography identifies the location of the parathyroid gland tumor and allows presurgical planning.

Double-phase scintigraphy of the parathyroid glands using ^{99m}Tc sestamibi was useful in the diagnosis of parathyroid gland adenoma in initial reports from two dogs.[333,570] In a study of 15 dogs with hypercalcemia, scintigraphy correctly identified 3 of 3 dogs with hypercalcemia of malignancy as negative for hyperfunctioning parathyroid glands.[332] Scintigraphy identified only one of six dogs with parathyroid gland adenoma and only one of six dogs with parathyroid hyperplasia. Based on these results, parathyroid gland scintigraphy is not recommended to identify abnormal parathyroid glands because of very poor sensitivity and specificity.

Surgical exploration of the cervical region in patients with parathyroid gland adenoma or carcinoma usually reveals enlargement of one parathyroid gland, and the remaining three are small or impossible to identify because hypercalcemia results in atrophy of normal parathyroid tissue. Primary parathyroid gland hyperplasia may affect more than one gland, and clinical signs can recur if only the largest gland is removed surgically. Parathyroid gland tumors may be difficult to identify if the tumor is embedded in fat or if it arises from the internal parathyroid gland. Failure to visualize a parathyroid gland tumor is rarely attributed to the occurrence of a tumor in ectopic parathyroid tissue. Methylene blue infusion to enhance visualization of parathyroid glands should be reserved for patients in whom a tumor is strongly suspected but not readily identified during surgery because clinically relevant side effects of methylene blue administration include hemolytic anemia and acute renal failure.[175]

Ultrasound-guided chemical ablation was used safely and effectively as an alternative treatment to surgery in eight dogs with a solitary parathyroid gland mass and hypercalcemia.[310] Serum tCa and iCa concentrations were within reference ranges 24 hours after treatment in seven dogs and within 5 days in one dog. Transient hypocalcemia developed in four dogs during the first 5 days after treatment; one dog required treatment for hypocalcemic tetany. Dysphonia was noted in two of eight dogs in this study, but Horner's syndrome, laryngeal paralysis, and death were not encountered as has been described with ethanol injection of thyroid glands of hyperthyroid cats.[200,540,558] It is likely that the low volume of ethanol injected into a single parathyroid mass provides less potential for leakage beyond the parathyroid mass.

Ultrasonographically guided radiofrequency heat ablation of parathyroid masses in dogs has become the preferred treatment at some referral hospitals. In one study, 11 dogs with either one or two masses on ultrasonography were treated by radiofrequency heat following anesthesia and insertion of a 20-gauge over-the-needle catheter into the mass.[415] Hypocalcemia developed in five of the eight successfully treated dogs, all of which required treatment. The only other adverse effect was a transient voice change in one dog.

Hypervitaminosis D

Hypervitaminosis D refers to toxicity resulting from excess cholecalciferol (vitamin D_3) or ergocalciferol (vitamin D_2). Metabolites of vitamin D can also exert toxicity, and the term hypervitaminosis D has been extended clinically to include toxicity from 25-hydroxyvitamin D, dihydrotachysterol, and 1,25-dihydroxyvitamin D (calcitriol), as well as newer analogues of calcitriol. Vitamin D toxicity is better referred to as 25-hydroxyvitamin D toxicity because vitamin D is rapidly transformed into this metabolite in vivo.[188] Vitamin D and its immediate metabolite, 25-hydroxyvitamin D, have little biologic activity at physiologic concentrations because they have low binding affinity for the VDR. Pharmacologic concentrations of 25-hydroxyvitamin D that occur during hypervitaminosis D exert hypercalcemic effects because 25-hydroxyvitamin D competes with calcitriol for binding to the VDR in target tissues.[153,366] Hypercalcemia results from increased intestinal absorption of calcium, but increased osteoclastic bone resorption and calcium reabsorption from renal distal tubules may also contribute.

Vitamin D intoxication and hypercalcemia may result from excessive dietary supplementation or may be caused iatrogenically during the treatment of hypoparathyroidism. Accurate dosing with cholecalciferol and ergocalciferol is difficult because they have a slow onset and prolonged duration of action.[37,485] Hypercalcemia developed in 7 of 16 hypoparathyroid dogs during treatment with vitamin D and calcium salt supplementation.[37] Ingestion of toxic plants that contain glycosides of calcitriol (e.g., *Cestrum diurnum, Solanum malacoxylon,* and *Trisetum flavescens*) is a potential cause of hypercalcemia in small animals.[390] Vitamin D toxicity associated with ingestion of *C. diurnum* has been reported in a cat.[142] *C. diurnum,* day-blooming jessamine, has achieved increasing popularity as a house plant and should not be confused with jasmine, which is an indoor climbing plant without active vitamin D metabolites.[80]

A diagnosis of hypervitaminosis D in dogs and cats increased with the introduction of cholecalciferol-containing rodenticides in 1985, but this source of intoxication is less common today. Cholecalciferol bait is delivered as pellets that are palatable to some animals and are very toxic when ingested. One manufacturer claimed a low hazard to dogs (oral median lethal dose, 88 mg/kg), but toxicity at a lower dosage (10 mg/kg) was demonstrated.[153,214] High-risk groups include dogs weighing 12 kg or less and those younger than 9 months. Recovery from previous cholecalciferol toxicity can be a risk factor for subsequent occurrence because removal of the source from the premises may not be possible.[108] Toxicity in four cats has also been reported.[353,401] One reason for the few reports of vitamin D toxicity in cats is that they appear to be resistant to cholecalciferol toxicity when the diet is otherwise complete and balanced.[488]

Clinical signs are usually vague and include anorexia, lethargy, vomiting, tremors, constipation, and polyuria. These signs are usually attributed to the effects of hypercalcemia. Hypercalcemia is reversible with early and aggressive therapy by providing enough time for 25-hydroxyvitamin D to be eliminated from the body.[102,140,153] Death occurred in approximately 45% of dogs after developing hypercalcemia from hypervitaminosis D in early reports,[153,214,318,457] but the survival rate was higher in dogs of a later series.[102]

Hypercalcemia usually develops within 24 hours after ingestion,[214] and hypercalcemia is often severe unless serum samples were obtained within 24 hours of ingestion. Mild hyperphosphatemia is often noted. Azotemia is initially absent but can develop subsequently. Serum creatinine concentration usually is less than 3 mg/dL unless treatment has been delayed, in which case azotemia may be marked. It may take as long as 72 hours for azotemia to develop as a result of renal lesions caused by hypercalcemia. Measurement of serum 25-hydroxyvitamin D concentration can provide conclusive evidence for hypervitaminosis D after exposure to cholecalciferol or ergocalciferol. Serum concentrations of 25-hydroxyvitamin D were increased to at least twice the upper limit of normal, with a mean concentration approximately 10 times normal in dogs with hypervitaminosis D,[102] and were increased for weeks to months in some instances.[140] In 10 episodes of cholecalciferol intoxication, concentrations of cholecalciferol were increased above the normal range for 10 to 61 days.[108] The half-life for cholecalciferol was 29 days in experimental dogs.[457] Serum calcitriol concentrations were also increased early in the syndrome,[102] but suppression of calcitriol synthesis occurs later. Hypervitaminosis D with hypercalcemia, azotemia, high concentrations of 25-hydroxyvitamin D, and/or renal calcification has been described in cats from Japan fed fish-based commercial cat food.[220,354,465] Cholecalciferol content of these diets exceeded the dietary requirements of vitamin D by more than 100 times.

Renal disease and failure occurred within 4 to 14 months in a large number of cats fed a commercial cat food containing 30 times the vitamin D requirement.[355] All commercial cat foods provide vitamin D in excess of the minimal requirements, and there is no regulated upper limit on the quantity of vitamin D that can be included. Other factors may modulate the toxicity of hypervitaminosis D, such as increased dietary calcium and phosphorus or dietary reduction in magnesium.[488]

Hypercalcemia attributed to the effects of increased calcitriol occasionally occurs during calcitriol treatment in animals with hypoparathyroidism and rarely during treatment of renal secondary hyperparathyroidism. When hypercalcemia is observed, it is usually in patients given doses more than 3.5 ng/kg daily. Discontinuation of calcitriol should result in normocalcemia within 1 week. Dosing with calcitriol at twice the daily dosage every other day up-regulates fewer intestinal epithelial cells for calcium absorption and decreases the chance for further development of hypercalcemia. Formulation errors have also been encountered in which the concentration of calcitriol in a compounded product was too high. There are no veterinary preparations of calcitriol; thus the available preparations of calcitriol must be diluted in pharmaceutical oils for appropriate dosing. Hypercalcemia has also been encountered when dosing errors have been made (mg/kg amounts given as opposed to ng/kg amounts). High serum concentrations of calcitriol have been observed in some dogs with lymphoma and hypercalcemia,[446] but it is not clear whether the excess calcitriol was synthesized by the tumor or by the kidneys under stimulation of PTHrP.

Topical ointments containing potent vitamin D analogues (calcipotriene) for treatment of human psoriasis can result in hypercalcemia when toxic quantities are ingested by dogs.* Minimal toxic dose is 10 μg/kg; minimal lethal dose is 65 μg/kg; and the oral LD50 is between 100 and 150 μg/kg in dogs.[218] In 25 dogs with calcipotriene ingestion, 28% died, and 50% experienced AIRF. Phosphorus, tCa, and iCa are elevated with calcipotriene toxicity.[215,218] The affinity of calcipotriene for vitamin D–binding protein is much lower than that of calcitriol; thus free calcipotriene is readily available for binding to VDRs. The rapid binding to VDRs accounts for the rapid onset of hypercalcemia and hyperphosphatemia and also for the rapid catabolism of calcipotriene. Hypercalcemia decreases after several days rather than being prolonged for weeks to months as seen in cholecalciferol toxicity. Exposure to calcipotriene has not yet been reported in cats, although there are two anecdotal reports (one in Ireland and one in Australia; Boyd Jones, personal communication) of cats that developed hypercalcemia after licking calcipotriene from their owner's skin. Telephone calls to animal poison control centers indicate that exposure to this ointment has been increasing in dogs.[327] Whether calcipotriene cross-reacts with calcitriol in the measurement of vitamin D metabolites has not yet been determined, but it is not detected by methods to measure 25-hydroxyvitamin D.

**References 91,163,218,231,252-254,525.*

Granulomatous Disease

Hypercalcemia can result from calcitriol synthesis by activated macrophages during granulomatous inflammation. Normal macrophages express 1α-hydroxylase activity (which converts 25-hydroxyvitamin D to calcitriol) when stimulated by interferon or lipopolysaccharide. Macrophages in granulomatous inflammation express such activity without stimulation.[150] Blastomycosis is a granulomatous disease in dogs that is occasionally (6% to 14% of cases) associated with hypercalcemia. Hypercalcemia is usually mild but can be severe.[13,141] Reports of granulomatous diseases associated with hypercalcemia include two cats with disseminated histoplasmosis[234] and dogs with coccidioidomycosis or schistosomiasis.[168,529] In one dog with schistosomiasis, PTHrP levels were undetectable,[433] but in two other dogs with schistosomiasis, PTHrP levels were increased with no malignancy found at necropsy.[186] In cats, elevated calcitriol concentrations were documented in cases of *Nocardia* and atypical mycobacteria infection.[338] Cats with blastomycosis, cryptococcosis, actinomyces, and injection site granulomas (Chew and Peterson, unpublished observations on injection site granuloma)[338,467,500] have been noted with hypercalcemia possibly because of enhanced synthesis of calcitriol.[141] Severe hypercalcemia was observed in association with noninfectious granulomatous dermatitis in two dogs in which excess synthesis of calcitriol was suspected (Kwochka and Chew, unpublished observations). PTH, PTHrP, and 25-hydroxyvitamin D concentrations were not increased. Hypercalcemia resolved as the inflammation subsided. Nodular panniculitis with hypercalcemia has been reported in dogs, and calcitriol concentrations were two to three times normal in one instance.[157,408]

Idiopathic Hypercalcemia of Cats

Hypercalcemia may be less common in cats than in dogs, although the incidence of hypercalcemia from primary care practices is not reported. Within the past 10 years, IHC has been recognized in cats[336,346] and is now the most common cause of ionized hypercalcemia in cats in the United States. Even though some suggest that IHC is a local geographic phenomenon,[168] it is widespread across the United States and is being recognized in other parts of the world.

In IHC, serum calcium concentration may be increased for months to more than 1 year. In 427 cases of feline IHC, 46% had no clinical signs, 18% had mild weight loss with no other clinical signs, 6% had inflammatory bowel disease, 5% had chronic constipation, 4% were vomiting, and 1% were anorectic.[476] Uroliths or renoliths were observed in 15%, and calcium oxalate stones were noted in 10% of cases. Cats ranged in age from 0.5 to 20 years, and longhaired cats accounted for 27% of the cases (compared with an overall submission rate of 14% from longhaired cats). There was no sex predilection. Serum iCa concentration was increased; PTH concentration was in the lower half of the reference range; and PTHrP was negative in all samples. Concentration of iMg was normal, and mean concentration of 25-hydroxyvitamin D was within the reference range. Calcitriol was measured in a small number of these cats and was suppressed. In another study, 1 of 7 cats exhibited an increased concentration of calcitriol, and 2 of 11 cats had increased PTHrP in the absence of underlying neoplasia following extensive diagnostic evaluation, survival for many months, and necropsy.[346] It appears that excessive PTH, 25-hydroxyvitamin D, or calcitriol concentration is not the cause of IHC in most cats. However, normal concentrations of calcitriol could result in hypercalcemia if there are mutations of the VDR or an increase in number of calcitriol receptors. Normal concentrations of iMg indicate that PTH secretion is not inhibited by decreased or excess iMg.[476] Renal function, based on BUN and serum creatinine concentration, is usually normal initially, but some cats develop CRF secondary to long-standing IHC.[346] Results of serology testing for feline leukemia virus and feline immunodeficiency virus have been negative, and serum thyroxine concentrations have been normal. Chronic acidosis could explain chronic elevations of iCa,[116] but venous blood gas analysis has not revealed significant acid-base disturbances. Exploration of the cervical region has not identified primary hyperparathyroidism, and subtotal parathyroidectomy has not resolved hypercalcemia in cats in which this procedure was performed.[346]

As many as 35% of cats with calcium oxalate urinary stones have hypercalcemia. Even though the specifics of the underlying diagnoses were not detailed,[386] it is likely that most had IHC. The occurrence of ureterolithiasis in cats was very uncommon before 1993. Eleven cases of calcium oxalate ureterolithiasis were recently described in cats, and four had mild to moderate hypercalcemia.[295] It appears that the frequency of hypercalcemia in calcium oxalate stone-forming cats has decreased substantially (Lulich, personal communications, 2003).

Specific treatment for IHC is impossible because the pathogenesis remains unknown. Increased bone resorption, increased intestinal absorption, or decreased renal excretion of calcium or combinations of these mechanisms could be responsible for hypercalcemia. The feeding of increased dietary fiber decreased serum calcium in some cats[336] but not in others.[346] The beneficial effect of a higher fiber diet may be because of decreased intestinal absorption of dietary calcium. The effects of fiber on intestinal absorption are complex and depend on the types and amounts of fiber in the diet and other nutrients present.

The feeding of veterinary renal diets may result in normocalcemia in some cats with IHC. These diets are generally low in calcium and phosphorus and are considered alkalinizing or at least less acidifying than maintenance diets. Some cats that show an initial decrease in serum calcium concentration following any type of dietary change will have a return to hypercalcemia over time.

In those cats that do not respond to a change in diet, prednisone therapy may result in a long-term decrease in iCa. The effects of glucocorticosteroid treatment may last for months to years in some cats with doses of 5 to 20 mg prednisone/cat/day. There is an escape from the effects of glucocorticosteroid treatment in some cats and a return to hypercalcemia despite maximal doses of prednisone. When dietary modification and treatment with prednisolone have been unsuccessful in resolving IHC, intravenous pamidronate treatment can be considered.

Beneficial effects from the chronic administration of subcutaneous fluids or oral furosemide to cats with IHC have not been evaluated. Treatment with calcimimetics could be of benefit. Calcimimetics interact with the calcium receptor and are effective in decreasing calcium, phosphorus, and PTH in human patients.[50]

Uncommon Causes of Hypercalcemia

AIRF in dogs is occasionally associated with mild hypercalcemia. Hypercalcemia may occur more commonly after conversion of oliguria to polyuria, possibly as calcium salts that were deposited during oliguria are mobilized from soft tissues. Sudden improvement in renal function also may result in rapid decrease of serum phosphorus concentration, changing mass law interactions between phosphorus and calcium and resulting in transient hypercalcemia. Mild hypercalcemia (11.5 to 12.5 mg/dL) is observed uncommonly in some dogs with severe oliguria and decreased GFR during intrinsic renal failure. Animals with severe hyperphosphatemia during AIRF usually have normal or low serum calcium concentrations.

Nonmalignant skeletal lesions are occasionally associated with hypercalcemia in dogs. Bacterial and fungal osteomyelitis can potentially result in hypercalcemia if the rate of osteolysis is sufficient.[106] Neonatal septicemia has been associated with hypercalcemia on rare occasion in puppies after septic embolization of bone and subsequent osteolysis.[106] Mild hypercalcemia occurs in some dogs with hypertrophic osteodystrophy, and the hypercalcemia may be aggravated by ascorbic acid supplementation.[509] Hypothermia has caused hypercalcemia in one cat.[391] One cat with pancreatitis and hypercalcemia has been described, even though hypocalcemia is more common in cases of pancreatitis.[231] In one report, a dog receiving intermittent calcium therapy for hypocalcemia developed hypercalcemia and acute pancreatic hemorrhage that may have been related to excessive calcium therapy.[371] Dehydration may cause mild and reversible hypercalcemia, especially with normal kidney function. Disuse osteoporosis after prolonged immobilization can rarely contribute to the development of mild hypercalcemia because weight bearing is necessary to maintain the balance between new bone formation and resorption of old bone. Serum total hypercalcemia has been noted in a small percentage of hyperthyroid cats,[25,467] but iCa concentration is normal. In cats with untreated hyperthyroidism, mild ionized hypercalcemia that resolved following conversion to euthyroidism with treatment has been uncommonly noted (Chew, unpublished observations). Overuse of calcium-containing intestinal phosphate binders can occasionally cause hypercalcemia.[106] An unusual case of hypercalcemia was attributed to the chronic ingestion of calcium carbonate in the form of limestone rocks.[273] Malignant histiocytosis in dogs was reported in association with hypercalcemia in one dog.[534]

The ingestion of large amounts of grapes or raisins may result in hypercalcemia. Seven of 10 dogs with renal failure associated with grape or raisin ingestion had increased serum tCa concentrations (12.3 to 26 mg/dL) and increased serum phosphorus (6.4 to 22 mg/dL) 24 hours to several days following ingestion.[215] In four dogs, ingestion was estimated to be from 0.41 to 1.1 ounces of grapes or raisins per kilogram of body weight. Oliguria or anuria was noted in 5 of 10 dogs, and 5 of 10 dogs survived. These cases were clustered from 1999 to 2001, and raisin/grape toxicity has not been previously reported.

Vomiting following ingestion of what appears to be a trivial quantity of raisins or grapes in some dogs leads to the development of AIRF usually within 48 hours. Not all dogs that consume grapes or raisins develop clinical signs or acute renal failure. Of 132 dogs reported with raisin or grape ingestion, 33 developed no clinical signs or azotemia, and 14 of 133 dogs developed clinical signs but no azotemia.[160] Of 132 cases, 43 dogs developed clinical signs and AIRF. The pathogenesis of nephrotoxicity associated with raisins and grapes remains unknown, but it is speculated that ochratoxin may be a toxic component.[409] Tubular degeneration and necrosis of varying severity are consistently described and most pronounced in proximal tubules.[160,357]

In some cases of grape/raisin ingestion with AIRF, mild to severe hypercalcemia develops, and in some dogs, serum tCa concentration can change dramatically from day to day during various treatments.[334] With acute renal failure following ingestion of raisins or grapes, hypercalcemia was detected in 93% of affected dogs, and tCa ranged from 8 to 26 mg/dL.[160,215] Of 40 dogs, 23 (57.5%) survived, and 17 (42.5%) failed to survive; 15 of 23 underwent complete resolution of azotemia. Initial and peak serum tCa concentration and initial and peak calcium x phosphorus product were significantly higher in those that did not survive as compared with those that did survive. Hypercalcemia was documented in 1 of 3 dogs evaluated within 24 hours of ingestion, in 2 of 8 dogs within 24 to 48 hours, and in 12 of 13 dogs evaluated for the first time 48 to 72 hours after ingestion. Total calcium concentration returned to the normal range in a median of 11 days (range, 2 to 51 days). Unfortunately, iCa measurements have yet to be reported for any dogs with raisin toxicity, AIRF, and hypercalcemia based on serum tCa. Because many dogs with severe AIRF have hyperphosphatemia, some of the increased serum tCa may be because of complex formation with phosphate. The observation that serum tCa concentration can dramatically increase or decrease daily during treatment suggests that its origin is related to extracellular or intravascular fluid volume dynamics.

A favorable outcome is possible in about 50% of cases, but several weeks of hospitalization with intensive fluid treatment is often needed in those with AIRF, especially if oliguric. About 50% of affected dogs can be expected to develop oliguria or anuria.[160,215,334] A case of AIRF with a fatal outcome occurred after ingestion of 450 g of raisins in a vizsla dog despite intensive treatment including peritoneal dialysis.[397] Aggressive treatment has been recommended for any dogs suspected of having ingested large, or even small, quantities of grapes or raisins, including induction of emesis, gastric lavage, and administration of activated charcoal, followed by intravenous fluid therapy for a minimum of 48 hours.[215] However, some dogs may consume relatively large quantities of grapes or raisins without development of ill effects.

Hypercalcemia was reported in a dog with a retained fetus and endometritis.[232] Serum PTH was suppressed, and 25-hydroxyvitamin D concentration was within the normal range. Biopsy of the removed uterus documented neutrophilic inflammation but no granulomatous inflammation as a possible cause of the hypercalcemia. Serum iCa was normal 4 days after surgical removal of the uterus, and serum tCa was normal 6 weeks later.

Humoral hypercalcemia of benignancy is a phrase used to describe the association of humoral factors such as PTHrP and hypercalcemia in the absence of malignancy.[186,279] One dog with massive mammary gland hyperplasia, severe ionized hypercalcemia, and increased PTHrP in the absence of malignancy at necropsy has been observed (Chew, unpublished observations). This phenomenon has rarely been described in humans.[258,269]

Treatment of Hypercalcemia

Philosophy of Treatment

There is no absolute serum calcium concentration that can be used as a guideline for the decision to treat hypercalcemia aggressively.[109,174] The magnitude of hypercalcemia, its rate of development, whether the serum calcium concentration is stable or progressively increasing, and the modifying effects of other electrolyte and acid-base disturbances must all be considered when deciding on a treatment plan. The clinical condition of the animal ultimately dictates how aggressive treatment should be, but a serum calcium concentration of 16 mg/dL or greater has been recommended as a basis for aggressive therapy.[174] Animals with serum calcium concentrations approaching 20 mg/dL should be considered candidates for crisis management. Animals with serum calcium concentrations less than 16 mg/dL may also require aggressive treatment, depending on the degree of neurologic, cardiac, and renal dysfunction induced by the hypercalcemia and concurrent deleterious factors. Acidosis can magnify the effects of hypercalcemia at all serum calcium concentrations by shifting more calcium to the ionized fraction. The serum phosphorus concentration at the time of hypercalcemia is also an important modulating factor in clinical decision making because soft tissue mineralization is potentiated by hyperphosphatemia. Animals with rapid and progressive development of hypercalcemia usually display serious clinical signs that require aggressive therapy.

Definitive Therapy

Removal of the underlying cause is the definitive treatment for hypercalcemia. Most animals with pathologic hypercalcemia have an associated malignancy that is quickly diagnosed but often not readily treated. Complete excision of isolated neoplasms (e.g., apocrine gland adenocarcinoma of the anal sac and parathyroid gland adenoma) abolishes hypercalcemia. In animals with disseminated metastases, multicentric neoplasia, or nonresectable primary malignancy, the tumor burden and hypercalcemia may be decreased by appropriate chemotherapy, radiation therapy, and immunotherapy. Chemotherapy may disrupt neoplastic cellular metabolism to such an extent that the tumor may no longer be able to synthesize enough humoral factors to sustain hypercalcemia. Decreased serum calcium concentrations can occur despite lack of obvious reduction in tumor size in these instances.

Antifungal treatment with amphotericin B, ketoconazole, or itraconazole effectively lowers increased serum calcium concentrations in dogs with systemic mycoses as the infectious agent is eradicated and inflammation resolves. For animals with hypercalcemia associated with hypoadrenocorticism, replacement therapy with mineralocorticoids and glucocorticoids after fluid volume replacement definitively manages the condition. Discontinuing all vitamin D supplementation in animals with hypervitaminosis D and hypercalcemia removes the external cause of intoxication, but excessive body stores of vitamin D may continue to contribute to hypercalcemia for several weeks.

Supportive Therapy

Supportive therapy is often necessary to decrease serum calcium concentration to a less toxic level while waiting for a definitive diagnosis to be established, for definitive treatment to reduce serum calcium concentration permanently, or for chronic management of hypercalcemia when the underlying cause cannot be removed. Box 6-3 and Table 6-3 list general and specific treatments for the management of hypercalcemia. Unfortunately, no single treatment protocol is consistently effective for all causes of hypercalcemia. Consequently, regimens must be tailored for the individual patient. Supportive treatments reduce the magnitude of hypercalcemia by increasing renal calcium excretion, inhibiting bone resorption, promoting soft tissue deposition of calcium, causing a shift of intravascular calcium to other body compartments, promoting extrarenal calcium loss, reducing calcium transport across the gut, or some combination of these effects.[109,291,327]

Initial Considerations for Treatment

Parenteral fluids, furosemide, sodium bicarbonate, glucocorticoids, or combinations of these treatments effectively reduce serum calcium concentrations in most animals. Repeatable serum hypercalcemia should be confirmed before prescribing aggressive treatments. It is not necessary to reduce serum calcium concentration to within normal limits, but substantial resolution of serious clinical signs may occur when serum tCa concentration decreases by as little as 1 to 3 mg/dL.

Box 6-3 General Treatment of Hypercalcemia

Definitive
- Remove underlying cause

Supportive
- Initial considerations
  - Fluids (0.9% sodium chloride)
  - Furosemide
  - Calcitonin
- Secondary considerations
  - Glucorticosteroids
  - Bisphosphonates
- Tertiary considerations
  - Sodium bicarbonate
  - Mithramycin (severe toxicity)
  - Ethylenediamine tetraacetic acid (EDTA) (severe toxicity)
  - Dialysis
- Future considerations
  - Calcium channel blockers
  - Somatostatin congeners
  - Calcium receptor agonists
  - Nonhypercalcemic calcitriol analogues

TABLE 6-3 Specific Treatment of Hypercalcemia

Treatment	Dose	Indications	Comments
Volume Expansion			
Subcutaneous saline (0.9%)*	75-100 mL/kg/day	Mild hypercalcemia	Contraindicated if peripheral edema is present.
Intravenous saline (0.9%)*	100-125 mL/kg/day	Moderate to severe hypercalcemia	Contraindicated in congestive heart failure and hypertension. Minimal decreases of calcium as single therapy when cause is severe pathologic hypercalcemia.
Diuretics			
Furosemide	2-4 mg/kg BID to TID IV, SQ, PO	Moderate to severe hypercalcemia	Volume expansion is necessary before use of this drug. Rapid onset of action.
Alkalinizing Agent			
Sodium bicarbonate	1 mEq/kg IV slow bolus; may give up to 4 mEq/kg total dose	Severe hypercalcemia	Requires close monitoring. Rapid onset of action.
Glucocorticoids			
Prednisone	1-2.2 mg/kg BID PO, SQ, IV	Moderate to severe hypercalcemia	Use of these drugs before identification of etiology may make definitive diagnosis difficult or impossible.
Dexamethasone	0.1-0.22 mg/kg BID IV, SQ		
Bone Resorption Inhibitors			
Calcitonin	4-6 IU/kg SQ BID to TID	Hypervitaminosis D	Response may be short-lived. Vomiting may occur. Rapid onset of action.
Bisphosphonates			
EHDP–didronel	15 mg/kg SID to BID	Moderate to severe hypercalcemia	Delayed onset of action.
Clodronate	20-25 mg/kg in a 4-hr IV infusion		Clodronate is approved for use in humans in Europe; availability in U.S. may be limited.
Pamidronate	1.3 mg/kg in 150 mL 0.9% saline a 2-hr IV infusion; can repeat in 1 week		Very expensive
Mithramycin	25 µg/kg IV in 5% dextrose over 2 to 4 hr every 2 to 4 weeks	Severe hypercalcemia, refractory HHM	Limited use in dogs and cats. Nephrotoxicity, hepatoxicity, thrombocytopenia.
Miscellaneous			
Sodium EDTA	25-75 mg/kg/hr	Severe hypercalcemia	Nephrotoxicity
Peritoneal dialysis	Low calcium dialysate	Severe hypercalcemia	Short duration of response. Use in hypercalcemia not reported.

**Potassium supplementation is necessary. Add 5 to 40 mEq KCl/L depending on serum potassium concentration.*

BID, *Twice daily;* TID, *thrice daily;* PO, *oral;* IV, *intravenous;* SQ, *subcutanous;* SID, *once daily;* HHM, *humoral hypercalcemia of malignancy.*

Fluid Therapy. Parenteral fluid therapy is an important first treatment for all animals with hypercalcemia. The first goal of fluid therapy is to correct dehydration because hemoconcentration contributes to increased serum calcium concentration. In addition, the kidney responds during ECF volume contraction with more avid reabsorption of sodium and calcium from the glomerular ultrafiltrate. Correction of dehydration abrogates this effect and allows calciuresis and natriuresis to occur.

Dehydration should be corrected with intravenous fluids within 4 to 6 hours of presentation in animals with severe clinical signs attributable to hypercalcemia. Additional expansion of ECF volume with parenteral fluids is then indicated, but sufficient fluid for rehydration and volume expansion is often provided simultaneously. Fluid therapy alone may be sufficient in some animals to reduce the magnitude of hypercalcemia adequately when the initial serum calcium concentration is less than 14 mg/dL, but often other treatments must be added. Normocalcemia may be restored by fluid therapy alone if hypercalcemia was initially mild (12 to 13 mg/dL).

Physiologic saline (0.9% NaCl) is the solution of choice for correction of the intravascular volume deficit and for further slight volume expansion. Slight volume expansion with 0.9% NaCl promotes calcium loss in urine secondary to increased GFR and increased filtered load of calcium, and competition from the additional sodium ions results in reduced renal tubular calcium reabsorption and enhanced calciuresis.

ECF volume expansion with lactated Ringer's solution (6 mg/dL calcium) in dogs results in decreased total protein, tCa, and iCa concentrations. Decreases in tCa concentration were greater (12.4%) than those observed for iCa concentration (3.5%).[425] Thus volume expansion with solutions that contain some calcium can be beneficial because the dilutional effect supersedes the effect of the additional calcium that is administered. However, physiologic saline (0.9% NaCl) is preferred because it is devoid of additional calcium and contains more sodium than that in lactated Ringer's solution (154 versus 130 mEq/L). Consequently, 0.9% NaCl results in a more rapid reduction in serum calcium concentration. An initial fluid volume of two to three times maintenance needs (120 to 180 mL/kg/day) usually corrects dehydration, provides maintenance needs, and results in mild volume expansion. The use of sodium phosphate is not recommended because of the potential detrimental effects of soft tissue mineralization.[174]

Diuretics (Calciuretics). Administration of furosemide follows rehydration and fluid volume expansion as second in importance for treatment of persistent hypercalcemia. Furosemide promotes enhanced urinary calcium loss, but calciuresis does not follow the use of all diuretics. In particular, thiazides should not be used because they may result in hypocalciuria and potentially may aggravate hypercalcemia. Furosemide (5 mg/kg intravenously, followed by 5 mg/kg/hr as an infusion) acutely decreases serum tCa by a maximum of approximately 3 mg/dL.[384] It is important to match the increased volume of urine lost with an increased volume of parenteral fluids to prevent dehydration and to gain maximal calciuresis. Less aggressive regimens of furosemide administration may be effective in combination with other treatments or for chronic management of hypercalcemia. Adequate hydration before and during furosemide administration is essential; otherwise, diuresis may increase serum calcium concentration through hemoconcentration. Diuresis, natriuresis, and calciuresis were greater in greyhounds given a continuous rate infusion of furosemide (0.66 mg/kg bolus, followed by 0.66 mg/kg/hr for 8 hours) compared with intermittent furosemide (3 mg/kg at 0 and 4 hours).[5]

Sodium Bicarbonate. Infusion of sodium bicarbonate has been advocated for acute or crisis management of hypercalcemia, but most often it is mentioned for use in the presence of metabolic acidosis.[5,109,290] Serum iCa concentration is reduced as acidosis is corrected or mild alkalosis is created because more calcium becomes bound to serum proteins, and there is increased binding of calcium to bicarbonate.[425] Decreases in ionized and tCa concentrations after bicarbonate infusions have been observed in dogs[345] and cats.[110] A dosage of 1 to 4 mEq/kg sodium bicarbonate has been recommended to obtain the desired reduction in calcium concentration,[110,290] but it may not be necessary to provide continuous bicarbonate infusion because the effect can last for as long as 3 hours after a single dose of bicarbonate in normal cats.[110] Reduction in serum calcium concentration is slight after administration of sodium bicarbonate alone, but the effect increases with larger doses. Sodium bicarbonate infusion is most likely to be helpful in combination with other treatments.

Steroids. Glucocorticosteroids can contribute to a significant reduction in serum iCa concentration in hypercalcemic animals with lymphoma, apocrine gland adenocarcinoma of the anal sac, multiple myeloma, thymoma, hypoadrenocorticism, hypervitaminosis D, hypervitaminosis A, or granulomatous disease, but they have little effect on serum iCa concentration in animals with other causes of hypercalcemia (Box 6-4). Some cats with IHC also have a substantial decrease in serum iCa concentration after glucocorticoid treatment. Steroids exert their effect mainly by reducing bone resorption, decreasing intestinal calcium absorption, and increasing renal calcium excretion.[107,302]

Cytotoxicity against neoplastic lymphocytes after glucocorticoids can result in a dramatic and rapid reduction in serum calcium concentration in dogs with lymphoma. Whenever possible, however, glucocorticoids should be

Box 6-4 Steroid-Sensitive Causes of Hypercalcemia

Lymphoma or leukemia
Multiple myeloma
Thymoma
Vitamin D toxicity
Vitamin A toxicity
Granulomatous disease
Hypoadrenocorticism
Idiopathic hypercalcemia in cats

withheld from animals for which a diagnosis has not yet been established because lymphocytolysis can make a definitive histopathologic diagnosis of lymphoma much more difficult or impossible. A challenge test for the diagnosis of occult lymphoma has been proposed using L-asparaginase at 20,000 IU/m^2 intravenously in an effort to disturb tumor cell metabolism but not cause cytolysis. Calcium concentrations are measured at baseline and then every 12 to 24 hours for 72 hours. A complete return of serum calcium concentration to normal suggests occult lymphoma.[168] Once a diagnosis of lymphoma has been made, prednisone is usually administered at 1 to 2 mg/kg twice daily concomitant with chemotherapy.

Decreased bone resorption after administration of glucocorticoids may be the result of impaired osteoclast maturation and decreased numbers of calcitriol receptors in bone.[498] Cortisol antagonizes the effects of vitamin D on the intestine in rats.[219] In dogs, chronic oral administration of prednisone (1.2 to 1.5 mg/kg/day) resulted in decreased serum calcitriol concentrations but caused no change in the number of calcitriol receptors or calcium-binding proteins in enterocytes.[283] Granulomatous diseases associated with increased calcitriol synthesis and hypercalcemia are often sensitive to the effects of glucocorticoids in reducing the serum calcium concentration.[424,483] However, caution is advised because the underlying disease (e.g., systemic mycosis) may be worsened. Hypercalcemia associated with hypervitaminosis A can also be steroid responsive.[38]

Calcitonin. Calcitonin treatment may be useful in animals with severe hypercalcemia. Calcitonin should be considered instead of prednisone for treatment of animals without a definitive diagnosis. Calcitonin rapidly decreases the magnitude of hypercalcemia primarily by reducing the activity and formation of osteoclasts. A maximal decrement in serum tCa concentration of approximately 3 mg/dL can be expected.[111] The only known adverse effects of calcitonin are anorexia and vomiting, but relatively few treated dogs and cats have been evaluated. Calcitonin treatment is expensive; the magnitude of its effect is unpredictable; its effects may be short-lived (hours); and resistance often develops in a few days. Receptor down-regulation is thought to be responsible for development of resistance, a phenomenon that may be delayed by concurrent glucocorticoid treatment. The effectiveness of calcitonin may be restored after discontinuing treatment for 24 to 48 hours.[327] Despite these limitations, calcitonin in combination with pamidronate is considered the best therapy for severe malignancy-associated hypercalcemia in humans.[118,480]

The dosage of calcitonin in animals has been extrapolated from that used in humans (4 IU/kg intravenously, followed by 4 to 8 IU/kg subcutaneously once or twice daily).[291] Calcitonin is listed as an antidote on packages of cholecalciferol-containing (vitamin D) rat poison, and treatment with calcitonin has been reported in dogs with hypercalcemia resulting from cholecalciferol toxicity. The dosage of calcitonin used in these dogs was 8 IU/kg subcutaneously every 24 hours,[183] 5 IU/kg subcutaneously every 6 hours,[194] and 4 to 7 IU/kg subcutaneously every 6 to 8 hours.[140] Short-term calcitonin treatment (6 U/kg subcutaneously every 8 hours for 2 days) was not effective in controlling hypercalcemia in dogs when measured 4 days after ingestion of cholecalciferol.[457] Vomiting was common within 2 hours of calcitonin administration. Calcitonin (4 U/kg every 4 hours for the first day and then 8 mg/kg twice daily for the next 3 days) decreased serum tCa from nearly 18 mg/dL to 13 to 15 mg/dL, but the effect only lasted 4 to 8 hours.[232] Calcitonin has also been used as part of combination therapy for treatment of hypercalcemia in a cat with granulomatous disease.[338]

Bisphosphonates. Bisphosphonates (formerly misnamed diphosphonates) are drugs (pyrophosphate analogues) that have been developed to inhibit bone resorption.[53,431] The hypocalcemic effects of bisphosphonates during malignancy are bone related because there is no effect on tumor mass. Bisphosphonates decrease osteoclast activity and function, despite increased numbers of osteoclasts present as a result of local or humoral mechanisms of osteolysis. Inhibition of resorption requires 1 to 2 days. Long-term bisphosphonate administration can lead to decreased osteoclast numbers through lethal injury of osteoclasts and decreased recruitment of new osteoclasts. Etidronate was the first bisphosphonate to be used clinically, and the activity of newer bisphosphonates is often compared with that of etidronate. Etidronate and clodronate are non-amino bisphosphonates. The addition of an amine group to one of the side chains increases the antiresorptive action in bone (alendronate, residronate, ibandronate, and zoledronate). The greatest potency to date has been obtained in those compounds containing a tertiary amine (zoledronate).[349] Clodronate, pamidronate, alendronate, and residronate have potencies 10, 100, 1000, and 5000 times as great as that of etidronate, respectively.[181] Ibandronate is approximately 5000 times and zoledronate is more than 10,000 times the potency of etidronate.[372,374] Zoledronate is 100 to 850 times more active than pamidronate.[54]

Inhibition of bone resorption by pamidronate occurs earlier and is maintained longer than that induced by etidronate. Intravenous infusion of pamidronate has been the treatment of choice for severe hypercalcemia associated with malignancy in humans,[55,118,125] controlling cancer-induced hypercalcemia in more than 70%[466] to 90% of human patients.[55] The use of intravenous zoledronate is the new treatment of choice because of its increased potency over pamidronate, as well as the more convenient infusion protocol of only 15 minutes.* Serum tCa decreases more rapidly, and maintenance of normocalcemia is nearly twice as long when treated with zoledronate compared with pamidronate.[557]

Bisphosphonate treatment occasionally has been associated with the development of renal impairment in humans and AIRF in experimental animals.[315] This effect was seen after multiple doses and in some with preexisting renal disease.[22,315,325,399] Renal toxicity in dogs may be more likely when doses of 10 mg/kg or more of pamidronate are given.[456] The rate of infusion and the particular bisphosphonate chosen influence the possibilities for nephrotoxicity.[3,349] Dehydration should be corrected before bisphosphonates are administered to lessen chances of renal injury. Depending on the bisphosphonate used, several hours of 0.9% saline infusion may be required to attenuate potential adverse effects. Pamidronate infusion in humans with hypercalcemia and underlying renal failure was shown to be safe in some studies.[36,315,533]

In a model of cholecalciferol-induced hypercalcemia, dogs treated with pamidronate (1.3 mg/kg in 150 mL saline administered intravenously over 2 hours) starting 1 day following ingestion lost less weight and had significantly lower serum concentrations of phosphorus, tCa, and iCa than those treated with saline or calcitonin. Mean serum tCa decreased to within the reference range, and values for iCa decreased but not to the same degree as that for tCa following pamidronate treatment.[457] In a subsequent study, three different doses of pamidronate were given to dogs after a single dose of cholecalciferol.[456] Clinical signs were fewest in dogs given the two higher doses of pamidronate. All dogs given any dosage of pamidronate were alert and lost less weight compared with saline treatment. The decreases in serum tCa were dose dependent. Pamidronate lessened the reduction in GFR in a dose-dependent manner, but GFR was still reduced by 20% to 25% on day 14 (end of study). Minimal histopathologic lesions were seen in dogs treated with the low and intermediate doses of pamidronate; no lesions were detected in dogs treated with the high dose of pamidronate. It appears that doses of pamidronate at 2.0 mg/kg are most effective in dogs with cholecalciferol toxicity.

Clodronate was used clinically to treat hypercalcemia of malignancy in one dog and hypervitaminosis D in another dog.[408] Serum iCa and tCa concentrations were normal at 36 and 48 hours after a 4-hour infusion of clodronate (20 to 25 mg/kg), but long-term results were not reported. In a dog with severe hypercalcemia associated with adenocarcinoma of the anal sac, a single 2-hour infusion of pamidronate rapidly reduced serum tCa and iCa that had not previously responded to intravenous fluids, calcitonin, and furosemide.[262] In seven dogs with clinical calcipotriene toxicity, pamidronate (1.3 to 2.0 mg/kg intravenously) resulted in a decrease in tCa, phosphorus, and creatinine.[215] In another clinical report, seven dogs and two cats were given pamidronate (1.05 to 2.0 mg/kg intravenously) for a variety of disease processes, and treatment rapidly decreased serum calcium without evidence of toxicosis.[244] In dogs with bone tumors, intravenous pamidronate (1 mg/kg given over 2 hours as a constant rate infusion) was administered every 28 days depending on progression of the bone tumor.[162] One hundred thirty-three doses of intravenous pamidronate were given to this group of 33 dogs. Only one dog developed renal toxicity 16 days following the second pamidronate treatment; this dog also had paraneoplastic hypercalcemia. Based on these findings, pamidronate at multiple doses may safely and effectively lower both serum total and iCa concentrations in patients with hypercalcemia resulting from various disease processes.

Oral bisphosphonate therapy is generally designed for maintenance treatment after a course of intravenous bisphosphonates has been effective in the control of hypercalcemia. Less than 5% of orally administered bisphosphonate is absorbed from the gastrointestinal tract,[182] which limits the usefulness of oral forms of etidronate, clodronate, and alendronate.[210] Food in the stomach markedly reduces the oral absorption of some bisphosphonates.[198] Increasing the dose can slightly increase the oral absorption of bisphosphonates.[349] Etidronate is generally administered orally to dogs at 10 to 40 mg/kg/day in divided doses, and it has had some effectiveness in reduction of hypercalcemia associated with lymphoma, myeloma, primary hyperparathyroidism, and hypervitaminosis D in dogs (Chew and Couto, unpublished observations). A puppy with hypercalcemia and primary hyperparathyroidism was also successfully treated using etidronate.[517] There is concern about the oral administration of some bisphosphonates to humans because nausea, vomiting, abdominal pain, dyspepsia, esophagitis, and esophageal reflux can be adverse effects.[47] Both clodronate and alendronate have been used orally in humans,[34,249] but

References 35,78,321,322,373,399,557.

there are no clinical studies in dogs or cats using these drugs orally.

Both clodronate and pamidronate have safely and effectively been given subcutaneously for the control of hypercalcemia in people.[149,432] Use of subcutaneous clodronate was better tolerated than subcutaneous pamidronate.[541,542] The subcutaneous route has not yet been investigated for use in dogs or cats with hypercalcemia.

Other Miscellaneous Treatments. Mithramycin is a potent inhibitor of osteoclastic bone resorption.[440,443] Significant toxicity, including thrombocytopenia, hepatic necrosis, renal necrosis, and hypocalcemia, unfortunately has been reported with the use of this drug.[111,174,290] Mithramycin was safe when two doses of 0.1 mg/kg were administered intravenously 1 week apart to eight normal beagle dogs. Mithramycin decreased serum iCa concentration in these normal dogs without adverse side effects such as hepatotoxicity, nephrotoxicity, or bone marrow hypoplasia, but some shivering occurred during the infusion. Osteoclastic bone resorption was significantly reduced.[443] Mithramycin was used to treat cancer-associated hypercalcemia in client-owned dogs.[444] A single infusion of 0.1 mg/kg to two dogs resulted in normal serum tCa concentration within 24 hours, but severe hepatocellular necrosis associated with marked vomiting, diarrhea, and fever resulted in death shortly thereafter. To decrease additional episodes of toxicity, the dosage of mithramycin was decreased to 25 μg/kg for the remaining dogs in this study. Serum calcium concentration returned to the normal range in six of nine dogs within 24 to 48 hours of treatment. Toxicity at this dosage was minimal, but the calcium-lowering effect lasted only 24 to 72 hours in three dogs. PTHrP concentrations and tumor size remained unchanged after treatment, and the lowering of serum calcium concentration was attributed to decreased osteoclastic bone resorption. Mithramycin is seldom prescribed because of its toxicity in hypercalcemic dogs at higher dosages and the short-lived effect at lower dosages.

During a hypercalcemic crisis, EDTA can be infused at a dosage of 25 to 75 mg/kg/hr. Administered EDTA combines with circulating calcium to form a soluble complex that then is excreted by the kidneys.[111] This treatment is considered a rescue method designed to allow other modalities time to take effect. Use of EDTA should be reserved for crisis situations because EDTA is nephrotoxic at higher dosages. A 2-hour infusion of EDTA in normal dogs at 25 mg/kg/hr did not have detrimental effects on the kidneys.[527]

Hemodialysis or peritoneal dialysis with calcium-free dialysate may be used to lower serum calcium concentration when other methods fail.[93,282] Dialysis may be particularly helpful in animals with severe intrinsic renal failure caused by hypercalcemia. Clinical experience with this method of treatment in animals is limited.

Future Considerations

Calcimimetics are a new class of compounds that are able to activate the calcium receptor, stopping PTH secretion.[161,536] Cinacalcet (Sensipar, Amgen Inc., Thousand Oaks, CA) has been marketed for use in human renal secondary hyperparathyroidism.[50,187,369] This drug is expensive, and it is available only as a solid tablet, making its use in small animals problematic because creating smaller doses is very difficult. Despite their action on calcium receptors throughout the body rather than exclusively on the calcium receptors of the parathyroid glands, calcimimetics may have promise in treating hypercalcemias of any type, including idiopathic hypercalcemia of cats. In the future, calcimimetics for veterinary use may be developed.

The calcium channel blocker diltiazem reduces the magnitude of hypercalcemia and soft tissue mineralization in vitamin D toxicosis in chicks[153] and may be effective in hypercalcemia of other causes. The toxic effects of hypercalcemia on the cardiovascular system of dogs can be blunted by verapamil,[30,579,581] and this drug may prove useful for stabilizing dogs and cats with severe hypercalcemia until other measures to decrease serum calcium concentration become effective.

Most treatments for HHM have focused on counteracting the effects of excess PTHrP rather than inhibiting PTHrP secretion. Somatostatin congeners inhibit secretion of certain hormones, and one congener, lanreotide, successfully reduced serum calcium and PTHrP concentrations in a human patient with HHM.[12] Similar results were observed in other tumors in humans treated with octreotide.[348,358,410,532]

Nonhypercalcemic analogues of calcitriol have been reported to inhibit cell proliferation and PTHrP production by neoplastic tissue in vitro.[337,576] These new modalities for treating hypercalcemia in conditions associated with increased PTHrP appear to be safe, are easy to use, and are effective.[290]

Gallium nitrate is an antineoplastic, radioprotectant drug that has hypocalcemic properties related to its ability to reduce the solubility of hydroxyapatite in bone and inhibit osteoclast function. Gallium nitrate has been considered for treatment of refractory hypercalcemia, but it requires constant infusion.[43,291,382,547] Gallium nitrate was more effective in control of hypercalcemia for longer periods than etidronate or pamidronate in a recent study of humans. Treatment with gallium nitrate may be more effective than bisphosphonates in cancer-related hypercalcemia in those with the highest concentrations of PTHrP.[301] The cytoprotectant amifostine (investigational drug WR-2721) inhibits PTH secretion and may have effectiveness in animals with hyperparathyroidism.[551] Use of amifostine has been limited to humans, and its adverse effects include nausea, vomiting, somnolence, and hypotension.[43]

Additional Specific Treatments for Hypervitaminosis D

In hypervitaminosis D associated with cholecalciferol intoxication, treatment may be necessary for several weeks because of the long half-lives of cholecalciferol and vitamin D metabolites. Consequently, aggressive fluid therapy for 1 week or more may be required to correct the severe hypercalcemia that is often encountered. Prednisone and furosemide therapy should be continued as maintenance therapy for 1 month. In addition, a low-calcium diet is important to reduce intestinal absorption of calcium. The diet provided can be a commercially available veterinary food or a homemade diet consisting mostly of macaroni and lean ground beef. Dairy products should be strictly avoided. Non–calcium-containing intestinal phosphorus binders may also be beneficial to counteract the effects of hyperphosphatemia. This treatment may be particularly important because the magnitude of soft tissue mineralization is most severe in animals with hypercalcemia induced by vitamin D toxicosis. Aluminum hydroxide at 30 to 90 mg/kg/day in divided doses is recommended during the first 2 weeks, with dosage and duration of treatment adjusted based on serial measurements of serum phosphorus concentration. Other unproven methods for treatment include anticonvulsants to increase hepatic metabolism of cholecalciferol, intestinal calcium binders to reduce intestinal calcium absorption, and calcium channel blockers to decrease the toxic intracellular effects of persistent hypercalcemia.[153]

When hypervitaminosis D is caused by excess calcitriol in patients with granulomatous disease, chloroquine, hydroxychloroquine, and ketoconazole may be used as supplemental therapeutic agents or as substitutes for glucocorticoids because they impair conversion of 25-hydroxyvitamin D to 1,25-dihydroxyvitamin D by macrophages.[155,435]

HYPOCALCEMIA

INTRODUCTION

Hypocalcemia based on serum tCa is a relatively common laboratory abnormality and was observed in 13.5% of serum biochemical profiles of dogs in one clinical study.[111] Based on serum iCa measurement in 1633 sick dogs, the prevalence of hypocalcemia was 31%,[475] and in 434 sick cats, the prevalence was 27%.[192] On the basis of serum tCa concentration, hypocalcemia is usually defined as a concentration less than 8.0 mg/dL in dogs and less than 7.0 mg/dL in cats. When serum iCa concentration is used, hypocalcemia is generally defined as a concentration less than 5.0 mg/dL (1.25 mmol/L) in dogs and less than 4.5 mg/dL (1.1 mmol/L) in cats. The most likely reason for submission of samples to measure calcium regulatory hormones in animals with hypocalcemia is for those with persistent hypocalcemia that is moderate to severe in magnitude and for which a known cause cannot be identified; most will be submitted with suspicion for a diagnosis of primary hypoparathyroidism.

In human patients, large and unexplained differences between ionized and tCa concentrations have been found in hypocalcemic conditions.[296] This discordance is also seen in dogs and cats and is not predictable. Based on serum tCa measurement in 1633 sick dogs, 27% were classified hypocalcemic, but when iCa was measured, 31% were hypocalcemic.[475] Using serum tCa measurement in 434 sick cats, 49% were classified hypocalcemic, but when iCa was measured, only 27% were actually hypocalcemic. Thus in dogs, tCa measurement underestimated ionized hypocalcemia, and in cats, hypocalcemia was overestimated when using serum tCa concentration to predict iCa status.

CONSEQUENCES OF HYPOCALCEMIA AND CLINICAL SIGNS

Clinical signs related to hypocalcemia are identical regardless of the underlying cause (Box 6-5). Low serum iCa increases excitability of neuromuscular tissue, which accounts for many of the clinical signs of hypocalcemia.

Box 6-5 Clinical Signs Associated with Hypocalcemia

Common
- None
- Muscle tremors or fasciculations
- Facial rubbing (paresthesia?)
- Muscle cramping
- Stiff gait
- Behavioral change
  - Restlessness or excitation
  - Aggression
  - Hypersensitivity to stimuli
  - Disorientation

Occasional
- Panting
- Pyrexia
- Lethargy
- Anorexia
- Prolapse of third eyelid (cats)
- Posterior lenticular cataracts
- Tachycardia or electrocardiographic alterations (prolonged QT interval)

Uncommon
- Polyuria or polydipsia
- Hypotension
- Respiratory arrest or death

Animals with mild decreases in iCa concentration may display no obvious clinical signs. The duration and magnitude of ionized hypocalcemia and the rate of decline in iCa concentration interact to determine the severity of clinical signs. Clinical signs in dogs often are not obvious until serum tCa concentration is less than 6.5 mg/dL, and some dogs show surprisingly few signs despite severe hypocalcemia (serum tCa concentration, <5.0 mg/dL), especially if the underlying disease has been chronic and there has been sufficient time for physiologic adaptation. Acute development of hypocalcemia is usually associated with severe clinical signs. In its most severe forms, hypocalcemia can cause death as a result of circulatory effects (e.g., hypotension and decreased myocardial contractility) and respiratory arrest from paralysis of respiratory muscles. Serum tCa concentration less than 4.0 mg/dL can cause left-sided myocardial failure[145] and death,[169] especially if the decline in serum calcium concentration was rapid.

Other electrolyte and acid-base abnormalities can either magnify or diminish the signs of hypocalcemia. Correction of hypokalemia in cats with concurrent hypocalcemia may precipitate the onset of clinical signs of hypocalcemia.[136,370] Patients with chronic hypocalcemia often display intermittent clinical signs despite seemingly stable serum tCa concentrations. Although unpredictable, clinical signs often follow periods of exercise or excitement that may be associated with respiratory alkalosis and subsequent decreases in iCa concentration. Rapid infusion of alkali to correct metabolic acidosis can cause seizures in animals with marginal or previously compensated hypocalcemia through further reduction in iCa concentration.

Clinical signs in dogs with chronic hypocalcemia (primary hypoparathyroidism) include seizures, muscle tremors or fasciculations, muscle cramping, stiff gait, and behavioral changes (e.g., restlessness, excitation, aggression, hypersensitivity to stimuli, and disorientation).[82,111,136,485] Seizures often begin as focal muscle tremors that become more widespread. Most dogs in one series had a seizure during the initial 24 to 48 hours of hospitalization, a much higher frequency than encountered with idiopathic epilepsy.[169] Seizure activity associated with hypocalcemia may not be similar to that in idiopathic epilepsy because affected dogs may remain partially conscious and retain urinary continence during the seizure.[169,406] Seizures are often preceded by apprehension or nervousness. The seizures may be as short as 60 seconds or as long as 30 minutes in some dogs. Most seizures resolve without treatment but often recur despite treatment with anticonvulsants. Growling attributable to pain or behavior change occurred in approximately 40% of dogs, and intense rubbing of the face with the paws or on the ground was observed in more than 50% of dogs. These signs were attributed to either paresthesias or pain from facial muscle spasms.[82,169]

Pyrexia may be caused by increased muscular activity with or without seizures. Lethargy and weakness are seen in approximately 33%, and polyuria and polydipsia occur in about 25% of cases, as a result of psychogenic mechanisms or renal injury (nephrocalcinosis) from hypercalciuria associated with PTH deficiency in animals with hypoparathyroidism.[460,485]Anterior and posterior lenticular cataracts occurred in more than 33% of affected dogs[82,284] and also in cats.[169,404] Tachycardia and electrocardiographic abnormalities (increased QT interval) may also be encountered. Both hypertension and hypotension have been reported during hypocalcemia in humans.[92,145]

Neuromuscular signs in cats with chronic hypocalcemia associated with primary hypoparathyroidism are similar to those in dogs (e.g., muscle tremors, weakness, and generalized seizures).[404] Anorexia and lethargy appear to be more common in cats than in dogs with primary hypoparathyroidism, but seizures have not been reported to be induced by excitement, as occurs in dogs. Prolapse of the third eyelid is occasionally observed in cats with acute hypocalcemia but is not a prominent finding during chronic hypocalcemia.

Clinical signs associated with acute postoperative hypocalcemia are similar in dogs and cats and are related to neuromuscular excitability. Focal twitching of facial muscles and vibrissae may be noticed before more generalized muscle tremors or seizures develop. Tetany or facial twitching has not been observed in cats after thyroidectomy until serum tCa concentration is less than 6.9 mg/dL.[169,404,406] Severe hypocalcemia (<6.5 mg/dL) is often associated with muscular twitching, tetany, or seizures. Anorexia and lethargy are not often considered primary signs of hypocalcemia, but both signs decrease in cats during calcium infusion after thyroidectomy, suggesting a relationship between hypocalcemia and these signs.

Approach to Hypocalcemia

Hypocalcemia develops when bone mobilization of calcium is reduced, skeletal calcium accretion is enhanced, urinary losses of calcium are increased, gastrointestinal absorption of calcium is reduced, calcium is translocated intracellularly, or as a result of a combination of these mechanisms. Much like the initial approach to hypercalcemia, it is helpful to make the initial distinction as to whether hypocalcemia is parathyroid dependent or parathyroid independent. Ionized calcium concentration must be evaluated in conjunction with PTH concentration to determine whether PTH production is appropriate. Patients with low iCa and low PTH concentrations have absolute hypoparathyroidism (parathyroid dependent). A normal reference range PTH when iCa is low is inappropriate because normal parathyroid glands should respond with increased PTH. Hypocalcemic patients with increased PTH are classified as having parathyroid-independent hypocalcemia. Normograms to determine the adequacy of the increased response of PTH to low iCa have not been established for dogs or cats. In cases of parathyroid-independent hypocalcemia, hypocalcemia exists from redistribution of calcium into other body spaces, excess phosphorus effects, or from deficiencies of vitamin D or dietary calcium. Patients with persistent moderate to severe hypocalcemia based on serum tCa should be evaluated for iCa and PTH concentrations; measurement of 25-hydroxyvitamin D and serum phosphorus is also helpful, and in rare circumstances, measurement of calcitriol may help provide a definitive diagnosis. The conditions associated with hypocalcemia in dogs and cats are listed in Box 6-6 according to their relative frequency regardless of clinical signs or severity of decreased serum calcium concentration. The anticipated changes in calcium hormones and serum biochemistry in disorders causing hypocalcemia are noted in Table 6-4.

Box 6-6 Conditions Associated with Hypocalcemia

Common
- Hypoalbuminemia
- Chronic renal failure
- Puerperal tetany (eclampsia)
- Acute renal failure
- Acute pancreatitis
- Undefined cause (mild hypocalcemia)

Occasional
- Soft tissue trauma or rhabdomyolysis
- Hypoparathyroidism
  - Primary
    - Idiopathic or spontaneous
    - Postoperative bilateral thyroidectomy
  - After sudden reversal of chronic hypercalcemia
  - Secondary to magnesium depletion or excess
- Ethylene glycol intoxication
- Phosphate enema
- After $NaHCO_3$ administration

Uncommon
- Laboratory error
- Improper sample anticoagulant (EDTA)
- Infarction of parathyroid gland adenoma
- Rapid intravenous infusion of phosphates
- Acute calcium-free intravenous infusion (dilutional)
- Intestinal malabsorption or severe starvation
- Hypovitaminosis D
- Blood transfusion (citrated anticoagulant)
- Hypomagnesemia
- Nutritional secondary hyperparathyroidism
- Tumor lysis syndrome

Human
- Pseudohypoparathyroidism
- Drug-induced
- Hypercalcitonism
- Osteoblastic bone neoplasia (prostate cancer)

DIFFERENTIAL DIAGNOSIS AND MECHANISMS OF HYPOCALCEMIA

Hypoalbuminemia

Hypoalbuminemia is the most common associated condition but perhaps the least important for clinical consequences, and it occurs in nearly one half of the dogs with hypocalcemia.[111] Hypocalcemia associated with hypoalbuminemia is usually mild (serum tCa concentration, 7.5 to 9.0 mg/dL in dogs), and no signs referable to the functional effects of low serum calcium concentration are observed. Application of calcium correction formulas to serum tCa concentrations in dogs or cats with hypoproteinemia or hypoalbuminemia has been advocated in the past. However, these correction formulas do not improve the prediction of actual iCa concentration and in many cases increase the level of diagnostic discordance.[475] Use of correction formulas to adjust serum tCa concentration to serum total protein or albumin concentration is not recommended.

Renal Failure

Renal failure is the second most common disorder associated with hypocalcemia in dogs.[111] Decreased calcitriol synthesis by diseased kidneys and, to a lesser extent, mass law interactions of calcium with markedly increased serum phosphorus concentration are probable causes of the hypocalcemia observed in dogs and cats with CRF. To decrease iCa concentration by 0.1 mg/dL, serum phosphorus concentration must increase by 3.7 mg/dL.[6] Calcitriol deficits are more important because hypocalcemia results from reduced intestinal calcium absorption and increased skeletal resistance to PTH.[366] Animals with CRF and decreased serum tCa concentration are most often asymptomatic, possibly because of an increase in iCa concentration that accompanies metabolic acidosis.

Serum tCa concentration was 8.0 mg/dL or less in 10% of 268 dogs with clinical CRF, whereas low serum iCa concentrations were detected in 40% of affected dogs.[114] In 23 dogs with CRF, iCa represented 40% of tCa as compared with 51% of tCa in normal dogs.[280] Serum iCa was low in 56%, normal in 26%, and high in 17% of the dogs with CRF. Thus iCa concentration was low in the majority of dogs despite the presence of metabolic acidosis in 83% of dogs, which would be expected to increase iCa.[280] Hypocalcemia was diagnosed more frequently in a study of 489 dogs with CRF when determined by iCa measurement. Based on serum tCa measurement, hypocalcemia was noted in only 19% of dogs with CRF; when iCa concentration was measured, hypocalcemia was observed in 29% of dogs with CRF.[475]

TABLE 6-4 Anticipated Changes in Calcemic Hormones and Serum Biochemistry Associated with Disorders of Hypocalcemia

	tCa	iCa	alb	Corr tCa	Pi	PTH	PTHrP	25(OH)-D	1,25 $(OH)_2$-D	PTG ULS, Surgery
Primary hypoparathyroidism	↓	↓	N	↓	↑ N	↓ N	N	N	N ↓	Multiple ↓
Pseudohypoparathyroidism	↓	N ↓	N	↓	↑ N	↑	N	N	N ↑	N ↑
Sepsis/critical care	↓ N	↓	N	↓ N	N ↑	↑ N	N	N	N	N
Ethylene glycol toxicity	↓	↓	N	↓	↑ N	↑	N	N	↓ N	N
Paraneoplastic	↓	↓	N	↓	↓	↑ N	N	N	N	N ↑
Phosphate enema	↓	↓	N	↓	↑	↑	N	N	N ↓ ↑	N
Eclampsia	↓	↓	N	↓	↓	Mild ↑, N	N	N	N ↓	N
Hypoalbuminemia	↓	↓ N	↓	N	N	N ↑	N	N	N ↑	N ↑

↓, *Decreased concentration;* ↑, *increased concentration;* N, *normal;* tCa, *serum total calcium;* iCa, *serum ionized calcium;* alb, *albumin;* Corr tCa, *corrected total calcium;* Pi, *inorganic phosphorus;* PTH, *parathyroid hormone;* PTHrP, *parathyroid hormone related protein;* 25(OH)-D, *25-hydroxyvitamin D;* 1,25(OH)2-D, *1,25-dihydroxyvitamin D;* PTG, *parathyroid gland;* ULS, *ultrasound.*

In 74 cats with clinical CRF, 15% were hypocalcemic based on serum tCa.[138] In cats with CRF, hypocalcemia was found more commonly with higher magnitudes of azotemia.[24] In 73 cats with CRF, none had hypocalcemia based on tCa, but 3% of cats with moderate CRF and 23% of cats with advanced CRF did have hypocalcemia. In 47 cats with CRF, 14% with moderate CRF and 56% with advanced CRF had ionized hypocalcemia. Mean iCa for cats with advanced CRF was significantly lower than values from normal cats or cats with mild and moderate CRF. Hypocalcemia was underappreciated when based on results of tCa measurement, especially with advancing azotemia.

AIRF and postrenal failure can result in hypocalcemia that is more likely to be symptomatic because the degree of hyperphosphatemia is often greater than that observed in CRF. Dogs with AIRF had a mean serum tCa concentration of 9.8 ± 1.7 mg/dL, but iCa was not reported.[537]

Emergency and Critical Care

Ionized hypocalcemia is common in critically ill humans in the intensive care setting and is more common in septic patients.[101,307,584] The magnitude of hypocalcemia is correlated to severity of illness. Hypocalcemia with critical illness probably also occurs in veterinary patients.[136] The causes of hypocalcemia in critical illness appear to be multifactorial because sepsis, systemic inflammatory response syndrome, hypomagnesemia, blood transfusions, and AIRF have been associated with hypocalcemia.[136,306,578,584] In humans, hypocalcemia associated with critical illness involves decreased PTH secretion, hypercalcitonism, and altered calcium binding to proteins.[422] The cause of the hypocalcemia is not related to enhanced urinary calcium excretion, decreased bone mobilization, or blunted secretion of PTH in septic patients.[307] The presence of proinflammatory cytokines during sepsis is related to the development of hypocalcemia in septic patients.[307] PTH is commonly elevated in this population even when normocalcemia exists.[101,307]

Up to 88% of hospitalized human patients had decreased iCa that correlated to severity of illness but not any specific diagnosis.[584] The impact of hypocalcemia on patient survival has not yet been determined, although in one study, hypocalcemia and higher levels of PTH were more frequently associated with fatality.[101]

Ionized calcium concentration decreased in experimental dogs with hemorrhage-caused hypotension and continued to decline during replacement of blood volume with citrated whole blood.[51] Hemorrhage also decreases iCa concentration in clinical dogs. Massive transfusions in 10 dogs resulted in significant ionized hypocalcemia.[261]

Cardiopulmonary resuscitation (CPR) may result in hypocalcemia. Dogs developed ionized hypocalcemia within 5 minutes of starting CPR in dogs with prolonged cardiac arrest and continued to decrease after 20 minutes.[88,377] Serum tCa was not concordant with changes in iCa because mean serum tCa did not change, and iCa concentrations were negatively associated with lactate concentrations. Decreased iCa was most likely related to formation of complexes with lactate.

In horses with enterocolitis, decreased iCa was identified in nearly 80% of patients.[524] Ionized hypocalcemia was associated with decreased iMg, increased serum phosphorus, decreased fractional urinary excretion of calcium, and increased PTH in 71% of cases. Hypocalcemia in 29% of these horses was a result of inadequate secretion of PTH, although impaired mobilization of calcium from bone and loss or sequestration of calcium within the gastrointestinal tract could not be excluded.

Acute pancreatitis may be associated with hypocalcemia. In 46 cats with acute pancreatitis, iCa concentration was low in 61% of cats.[272] Suggested mechanisms to account for low iCa in acute pancreatitis include sequestration of calcium into peripancreatic fat (saponification), increased free fatty acids, increased calcitonin secondary to increased glucagonemia, and PTH resistance or deficit resulting from the effects of hypomagnesemia.[40,136,257,461]

In dogs with diabetes mellitus, 47% had ionized hypocalcemia.[228] Normal iCa concentrations were noted in 49.4% of dogs from this study, and 3.5% had ionized hypercalcemia. Acute pancreatitis was diagnosed in 13% of these dogs, which could be the mechanism in some but not all of those with hypocalcemia.

Puerperal tetany (eclampsia) typically occurs between 1 and 3 weeks postpartum in females of small dog breeds and is attributed to loss of calcium into milk during lactation, although parathyroid gland dysfunction has not been conclusively excluded.[19,169] Proposed mechanisms for hypocalcemia include a poor dietary source of calcium, major loss of calcium during lactation, fetal skeletal ossification, and abnormal parathyroid gland function, including parathyroid gland atrophy. Hypophosphatemia may accompany the hypocalcemia, and clinical signs rarely occur before whelping.[98] In 31 dogs with periparturient hypocalcemia, iCa was less than the reference range, and small breed dogs with large litters were typical.[143] Median time from whelping to detection of clinical signs was 14 days, but variation was wide. Clinical signs most often included seizures, trembling, twitching, shaking, and stiffness. Nontypical signs included panting, behavioral changes, collapse, and whining; vomiting, diarrhea, and choking were rare. Rectal temperature was elevated, attributable to increased muscle activity. After treatment with intravenous calcium gluconate (mean dose, 115 mg/kg), iCa concentration normalized within 25 minutes in 90% of dogs. Most dogs received more than one injection of calcium gluconate, but the total calcium dose given did not correlate to initial iCa concentration. In one lactating bitch, severe hypocalcemia and hypomagnesemia occurred in association with acute onset of gastric and bladder atony, congestive heart failure, weakness, and paresis without muscle fasciculation or seizures.[14]

Puerperal tetany is rare in cats.[550] Eclampsia was described in four cats in which hypocalcemia developed 3 to 17 days before parturition.[165] Signs of depression, weakness, tachypnea, and mild muscle tremors were most common; vomiting and anorexia were less common, and prolapse of the third eyelid occurred in some cats. Hypothermia, instead of hyperthermia as seen in dogs, was observed. All cats responded to parenteral calcium gluconate initially and to oral calcium supplementation throughout gestation and lactation.

Ionized hypocalcemia is common in cats with urethral obstruction and is likely to develop in cats that also have hyperkalemia and metabolic acidosis. Cats with severe ionized hypocalcemia can exhibit compromised vital functions, although most survive with relief of urethral obstruction. In 199 cats with urethral obstruction, iCa was below the reference range in 34%, normal in 47%, and above the reference range in 19%.[299] Of those with low iCa, 14% had moderate and 6% had severe hypocalcemia. In an earlier study, 75% of cats with urethral obstruction exhibited low iCa.[144] Most of these cats had elevated tMg probably from reduced renal function at the time of obstruction. Hypomagnesemia is not likely to account for the development of hypocalcemia in these cats, but iMg was not measured. Calcium regulatory hormones were not evaluated in either of these studies. Alkalinizing infusions designed to correct metabolic acidosis or for translocation of potassium into cells are often considered for treatment of cats with urethral obstruction, but these can decrease tCa and iCa concentrations.[110]

Rhabdomyolysis is sometimes associated with hypocalcemia, but clinical signs of hypocalcemia are rare. Mild hypocalcemia in dogs and cats with severe vehicular muscle trauma is occasionally observed (Chew, personal observations). Hypocalcemia likely occurs as a consequence of translocation of calcium into the damaged muscles. Symptomatic hypocalcemia resulting in death of three dystrophin-deficient cats occurred following anesthesia or mild exertion during restraint and subsequent acute rhabdomyolysis.[196] Hypocalcemia was documented along with hyperphosphatemia, increased liver transaminases, and massive increases in creatine kinase. Hypocalcemia has been described in some dogs with fatal *Vipera xanthina palestinae* envenomation.[15] The origin of the hypocalcemia may be multifactorial, including muscle necrosis. Renal transplantation in 14 cats resulted in decreased iCa in the 5-day postoperative period.[568] All cats also had decreased serum iMg but normal tMg.

Small Intestinal Diseases

Hypocalcemia may occur in association with gastrointestinal disease. Ionized calcium concentration was below the reference range (mean, 0.99 ± 0.19 mmol/L; reference range, 1.13 to 1.33 mmol/L) in 12 dogs with intestinal lymphangiectasia.[293] Ten of 13 dogs had hypoalbuminemia with a mean of 2.12 ± 0.70 g/dL, and "corrected" serum tCa was discordant with iCa measurement. Mechanisms for hypocalcemia could include calcium/fatty acid complexes in the intestinal lumen that could decrease intestinal calcium absorption. Hypovitaminosis D from malabsorption or hypomagnesemia may have contributed to hypocalcemia but was not evaluated. No dogs had clinical signs associated with hypocalcemia.

In five Yorkshire terriers and a shih tzu with protein-losing enteropathy, iCa and tMg concentrations were moderately to severely low.[87,271] Concentration of PTH was increased (secondary hyperparathyroidism), and 25-hydroxyvitamin D concentration was below the reference range. It is not clear whether the apparent elevation in PTH was increased to an appropriate level in the face of low iCa, or whether maximum production was suppressed because of the effects of hypomagnesemia. Intravenous supplementation with fluids containing magnesium salts resulted in increases in PTH and iCa; 25-hydroxyvitamin D remained below the reference range.[87] Following 8 weeks of treatment for inflammatory bowel disease, calcium homeostasis was normal based on normal iCa, PTH, tMg, and 25-hydroxyvitamin D concentrations. Magnesium repletion apparently resulted in resolution of hypocalcemia largely because of increased PTH secretion, whereas 25-hydroxyvitamin D concentration was still low. Resolution of weakness may have been the result of correction of hypocalcemia, hypomagnesemia, or both.

Alkali Administration

The administration of alkaline agents may result in the development of hypocalcemia. Symptomatic hypocalcemia was documented in a cat treated for salicylate intoxication with sodium bicarbonate.[2] Muscle fasciculation increased during treatment with sodium bicarbonate, and serum tCa was low. A single dose of intravenous sodium bicarbonate at 4 mEq/L to cats resulted in a maximal decrease of iCa 10 minutes following infusion; iCa remained below baseline for 3 hours.[110] Part of the decrease in iCa was attributed to dilution and part to increased pH of serum, but most of the decrease was the result of unidentified factors. Similar findings were noted in dogs receiving sodium bicarbonate infusion.[345] Twitching has been observed on rare occasion during or shortly after infusion of sodium bicarbonate solutions to cats with urethral obstruction and to dogs or cats with renal failure (Chew, personal observations) presumably because of decreases in serum iCa.

Acute Reversal of Chronic Hypercalcemia

The sudden correction of chronic hypercalcemia can result in hypocalcemia as a result of parathyroid gland atrophy and inadequate ability to synthesize and secrete PTH. This happens frequently in dogs with primary hyperparathyroidism caused by parathyroid gland adenoma following surgical excision of the affected parathyroid gland(s). The degree of parathyroid gland atrophy depends on the magnitude of hypercalcemia and its duration before correction. Two dogs with spontaneous infarction of a parathyroid gland adenoma have been reported with the development of hypocalcemia and clinical signs.[442] Rapid correction of hypercalcemia following chemotherapy for lymphosarcoma or surgical excision of anal sac adenocarcinoma often results in mild hypocalcemia that is usually not associated with clinical signs, but clinical signs of hypocalcemia may occur.[241] Persistent hypocalcemia has been observed in dogs following parathyroidectomy in association with hypomagnesemia. In three dogs, hypocalcemia resolved following supplementation with magnesium salts, but calcium regulatory hormones were not measured (Chew, unpublished observations).

Tumor Lysis Syndrome

Tumor lysis syndrome occurs when there is rapid destruction of sensitive tumor cells (usually lymphoid or bone marrow tumors) following chemotherapy.[400] Release of intracellular products can result in hyperkalemia, hyperphosphatemia, and hyperuricemia. Hypocalcemia can develop as calcium-phosphate salts are deposited into soft tissues by mass-law effects from markedly increased serum phosphorus[90,388,413] and may be associated with the development of AIRF. Tumor lysis syndrome is a rarely reported cause of symptomatic hypocalcemia in dogs,[388,413] although it may be more common than previously reported because The Ohio State University oncology service has documented seven cases (Couto, personal communication, 2004).

Nutritional Secondary Hyperparathyroidism

Vitamin D deficiency and nutritional secondary hyperparathyroidism associated with low calcium and/or high phosphorus concentrations in the diet results in low serum iCa and phosphorus concentrations, with an increase in PTH secretion. Nutritional secondary hyperparathyroidism may also occur when severe gastrointestinal disease is present, limiting the absorption of calcium and vitamin D. Increased PTH secretion tends to return serum iCa concentration to normal, but decreases serum phosphorus concentration.[567] The occurrence of nutritional secondary hyperparathyroidism has decreased dramatically since the advent of feeding commercially available, nutritionally complete and balanced pet food.[264] Nutritional secondary hyperparathyroidism was induced in adult beagles by feeding a diet high in phosphorus and low in calcium, with a calcium to phosphorus ratio of 1:10.[123] A significant increase in PTH production was seen at 10 weeks of feeding, and cancellous bone volume was reduced by 20% to 30%. Under experimental conditions, puppies fed a low-calcium, normal phosphorus content diet exhibited increased concentrations of PTH and calcitriol, with a decrease in 24,25-dihydroxyvitamin D concentration.[223] In five German shepherd dog puppies fed a diet consisting of 80% steamed rice and 20% raw meat, nutritional secondary hyperparathyroidism was observed.[267] This diet apparently had an adequate calcium concentration but contained an excess of phosphorus. All puppies showed moderate to marked fibrous osteodystrophy.

Serum iCa and phosphorus concentrations were below the reference range in six young cats with nutritional secondary hyperparathyroidism.[522] Clinical signs referable to hypocalcemia (excitation, muscle twitching, seizures) and spontaneous fractures of bones were present in most cats. Renal secondary hyperparathyroidism preferentially affects the bones of the face (fibrous osteodystrophy), whereas nutritional secondary hyperparathyroidism tends to cause osteopenia of the long bones and vertebrae. Calcitriol concentration was mildly increased in three of four cats in which it was measured, whereas 25-hydroxyvitamin D was mildly decreased in three of three cats. PTH concentrations were increased in all cats and ranged from a minimal increase in one cat to a marked increase of 4 to 9.7 times the upper range in the remaining five cats. Cats had been fed meat only (three cats), meat combined with vegetables (two cats), or vegetables only (one cat). Dietary calcium intake was less than one tenth of the minimal nutritional requirement; dietary intake of phosphorus was mildly below the minimal requirements in five of six cats. An unfavorable calcium to phosphorus ratio existed for all diets. A case of type 2 vitamin D–dependent rickets was described in a 4-month-old cat examined because of vomiting, diarrhea, muscle tremors, and mydriasis of acute onset.[478] Serum tCa and tMg concentrations were decreased, and serum phosphorus, calcitriol, and PTH concentrations were increased, excluding hypoparathyroidism as the cause of hypocalcemia. Calcitriol and calcium salt supplementation resulted in the return to normocalcemia.

Exotic animals may be at increased risk for the development of nutritional secondary hyperparathyroidism because nutritional requirements are not always known. Nutritional secondary hyperparathyroidism was documented in a 3-month-old tiger cub that was fed only beef with no calcium or vitamin supplementation.[566] This tiger cub was reluctant to walk, exhibited osteodystrophy of the lumbosacral vertebrae, and had an elevated serum PTH concentration. Clinical signs improved after administration of vitamin D and calcium.

With the feeding of BARF (biologically appropriate raw food, or bones and raw food) and other homemade diets, the occurrence of nutritional secondary hyperparathyroidism is more likely. In a recent report, 6-week-old, large-breed puppies from two litters were fed a BARF diet on weaning.[130] Puppies were weak, exhibited pain, and had abnormal-appearing joints, and some were unable to stand. In puppies that were radiographed, osteopenia was noted, with pathologic fractures apparent in multiple long bones. In euthanized puppies, the long bones were pliable, and cortices were thin. Parathyroid glands were prominent, and histologically, fibrous osteodystrophy was present in bones. Nutritional secondary hyperparathyroidism was attributable to a diet low in calcium and an inappropriate calcium to phosphorus ratio.

Effects of Drugs

Drug administration may cause a decrease in iCa. A significant decrease in iCa was observed in dogs administered enrofloxacin at 5 mg/kg intramuscularly once daily for 14 days.[130] Mean iCa decreased to its nadir on day 3, remained below normal at day 10, and normalized by day 14 despite continued administration of enrofloxacin.

The administration of mithramycin or bisphosphonates can cause mild hypocalcemia as a side effect in humans, but symptomatic hypocalcemia is rare.[125,519] The potential for development of hypocalcemia exists in dogs following mithramycin administration because normal dogs and those with malignancy-associated hypercalcemia undergo significant decreases in serum iCa and tCa.[443,444] Use of mithramycin is reserved for emergency management of hypercalcemia refractory to other treatments because of severe toxicity in some dogs.

Phosphate enema administration can result in hypocalcemia after rapid absorption of phosphate, hyperphosphatemia, and subsequent mass law interaction with serum calcium. This is particularly a problem in cats and small dogs in which death can occur.[16,260,468,523] Serum tCa decreased within 45 minutes of administration of a hypertonic phosphate enema to cats and persisted for 4 hours.[16] Mean serum phosphorus was more than 14 mg/dL within 15 minutes, and increases persisted for 4 hours. Serum tCa concentrations were negatively correlated to serum phosphorus. Mild hypernatremia, severe hyperphosphatemia (mean, 37.6 mg/dL), and hypocalcemia were noted in five cats. Phosphate enemas should not be used in small dogs, cats, or in debilitated patients of any size.

Hypoparathyroidism

Hypoparathyroidism is an absolute or relative deficiency of PTH secretion that can be permanent or transient. Hypocalcemia and clinical signs referable to low iCa concentration are the hallmarks of advanced hypoparathyroidism. Hypoparathyroidism in dogs is most commonly idiopathic, whereas surgical removal of or injury to the parathyroid gland during thyroidectomy to correct hyperthyroidism is the most common cause in cats.

Idiopathic chronic inflammation of parathyroid tissue occurs sporadically in both dogs and cats but more commonly in dogs. It is presumed that parathyroiditis has an immune-mediated mechanism. Histopathologic study of affected parathyroid glands reveals inflammatory cell infiltration (lymphocytes, plasma cells, and neutrophils), fibrosis, and loss of secretory cells.[82,169,404,406,485] Clinical signs occurred 1 to 26 weeks (mean, 7 weeks) before diagnosis of primary hypoparathyroidism in cats[404] and 1 day to 25 weeks (mean, 3 weeks) before diagnosis in dogs.[82] Primary hypoparathyroidism and parathyroiditis occur in dogs and cats of any age but more frequently in female dogs and male cats. In 735 dogs with primary hypoparathyroidism, 62% were female and 38% were male.[423] Mean age was 7.0 ± 3.9 years, with 71% of diagnoses occurring in purebred dogs. The highest odds ratios for hypoparathyroidism correcting for breed popularity occurred in the standard schnauzer, Scottish terrier, miniature schnauzer, West Highland white terrier, and dachshund. Reduced risk was identified for the German shepherd dog, shih tzu, and Labrador retriever. In another study, 357 dogs were diagnosed with primary hypoparathyroidism over a 2-year period.[477] Mixed-breed dogs accounted for 25% of the cases, with 13% schnauzers, 7% Labrador retrievers, 5% dachshunds, 4% Yorkshire terriers, 4% poodles, 3% golden retrievers, and 3% Scottish terriers without correction for breed popularity. There were 59 other dog breeds represented with an incidence of less than 3% each.

Serum tCa concentration is usually less than 6.5 mg/dL (often 4.0 to 4.9 mg/dL) in dogs with primary hypoparathyroidism. Dogs that have episodes of tetany or seizures often have serum tCa concentration less than 6.0 mg/dL. Serum phosphorus concentration is greater than serum calcium concentration in nearly all affected dogs and cats, and most have hyperphosphatemia. Parathyroid gland biopsy may confirm the diagnosis of lymphocytic parathyroiditis as the cause of primary hypoparathyroidism, but the parathyroid glands can be difficult or impossible to locate during surgical exploration because of atrophy and fibrosis. Parathyroid gland biopsy is not recommended to confirm hypoparathyroidism since the advent of validated PTH assays for use in the dog and cat.

Diagnosis of Hypoparathyroidism. Inappropriately low concentrations of PTH result in hypocalcemia, hyperphosphatemia, and decreased concentrations of 1,25-dihydroxyvitamin D (calcitriol). Hypocalcemia results from increased urinary loss of calcium (hypercalciuria), reduced bone resorption, and decreased intestinal absorption of calcium. Hyperphosphatemia results from decreased urinary loss of phosphorus (hypophosphaturia) that overrides the effects of decreased bone resorption and decreased intestinal absorption of phosphorus (secondary to calcitriol deficit) on serum phosphorus concentration. PTH is a potent stimulator and phosphorus is a potent inhibitor of the 25-hydroxyvitamin D–1α-hydroxylase enzyme system in renal tubules. Consequently, the absence of PTH and the presence of hyperphosphatemia act together to decrease renal synthesis of calcitriol. Decreased concentrations of calcitriol contribute to hypocalcemia via decreased intestinal calcium absorption. Hypocalcemia unrelated to low PTH concentrations may arise from increased uptake of calcium by bone after rapid correction of long-standing hyperparathyroidism or hyperthyroidism, both of which are associated with loss of bone calcium before treatment ("hungry bone" syndrome).[501,516,563]

Definitive diagnosis of primary hypoparathyroidism is based on the combination of clinical signs (see Box 6-5), low iCa concentration, and PTH concentration inappropriately low to the magnitude of ionized hypocalcemia. Hypoparathyroidism is the only possible diagnosis when low serum calcium concentration, high serum phosphorus concentration, normal renal function, and low PTH concentration are present in combination. Low serum calcium and high serum phosphorus concentrations can be encountered during nutritional and renal secondary hyperparathyroidism, after phosphate-containing enemas, and during tumor lysis syndrome, but PTH is increased in all of these conditions.

PTH should be measured in patients with chronic hypocalcemia of undetermined etiology. Primary hypoparathyroidism requires lifelong treatment, and confirmation of the diagnosis with PTH measurement is recommended. It is not necessary to measure PTH routinely in patients with postsurgical hypocalcemia because this effect is usually transient and the cause obvious. PTH concentrations should be determined for patients in which hypocalcemia does not resolve. Absolute hypoparathyroidism is present if a PTH concentration below the reference range is detected simultaneously with hypocalcemia. Relative hypoparathyroidism is present if PTH concentration is inappropriately low but remains within the normal reference range. Increased serum phosphorus and decreased calcitriol concentrations provide further support for a diagnosis of hypoparathyroidism.[217]

Causes of Hypoparathyroidism. The causes of hypoparathyroidism can be divided into three categories: (1) suppressed secretion of PTH without parathyroid gland destruction,[111,136] (2) sudden correction of chronic hypercalcemia, and (3) absence or destruction of the parathyroid glands. The most common category of hypoparathyroidism in dogs and cats is absence or destruction of the parathyroid glands.

Postoperative hypocalcemia develops 1 to 3 days after thyroidectomy in approximately 20% to 30% of cats.[46,177,180,209,555] Some cats developed hypocalcemia as late as 1 to 2 weeks after surgery. The surgical technique used for thyroidectomy influences the chances that hypocalcemia will develop, and hypocalcemia occurred in more than 80% of cats when original extracapsular technique was used.[177] Bilateral thyroidectomy results in loss of the two internal parathyroid glands, and hypoparathyroidism is permanent in patients in which the external parathyroid glands are completely removed during bilateral thyroidectomy. Hypocalcemia and hypoparathyroidism do not develop if the two external parathyroid glands are not excised or damaged during thyroidectomy. Normocalcemia can be maintained with one completely functional parathyroid gland.

Hypoparathyroidism is usually transient when the external parathyroid glands are retained but have their blood supply disrupted (parathyroid gland ischemia after physical trauma, vessel stretching, suture, cautery, or transection) during surgery. Permanent hypoparathyroidism is rare, but it may take as long as 3 months to be certain whether remaining parathyroid tissue can recover by hyperplasia.[46,406,460] Similar injury to parathyroid glands can occur during any extensive surgery of the neck in dogs[225,278] or cats or after exploration of the neck for unilateral parathyroid gland removal. Restored vascular supply to damaged parathyroid tissue seems unlikely as the mechanism for recovery from hypocalcemia. It is more likely that hyperplasia and hypertrophy of parathyroid gland remnants left behind during surgery or ectopic parathyroid tissue achieve sufficient mass to synthesize adequate amounts of PTH. Experimental cats subjected to parathyroidectomy predictably developed hypocalcemia and low serum PTH concentration, but, interestingly, the hypocalcemia resolved, although the PTH concentrations remained low.[178] Autotransplantation of parathyroid tissue after bilateral thyroparathyroidectomy was associated with reduced morbidity and rapid return of serum calcium concentrations to normal in experimental cats.[387]

Long-standing ionized hypercalcemia causes normal parathyroid tissue to atrophy. If hypercalcemia is nonparathyroid in origin, PTH concentrations will already be low. Rapid correction of hypercalcemia results in hypocalcemia because the atrophic parathyroid glands cannot respond immediately to the need for increased PTH secretion. Surgical removal of a single parathyroid gland tumor (usually an adenoma) commonly causes postoperative hypocalcemia in this manner. Hypocalcemia severe enough to require treatment is likely to develop within 24 to 48 hours. Nearly 50% of dogs with primary hyperparathyroidism can be expected to develop clinical signs of hypocalcemia 3 to 6 days after surgical removal of a parathyroid gland tumor. Hypocalcemia is more likely to develop in dogs with higher presurgical iCa concentrations. More than one half of hyperparathyroid dogs exhibit a rapid decrease in serum iCa concentration that normalizes within 24 hours of surgery. Serum iCa concentrations in the remaining dogs usually normalize by

2 or 3 days after surgery, but some require as long as 5 days. Hypoparathyroidism resolves for most affected dogs in 8 to 12 weeks. Cats develop hypocalcemia less frequently than dogs after surgical correction of primary hyperparathyroidism.[133,263]

Hypoparathyroidism following spontaneous infarction of a parathyroid gland tumor previously causing hypercalcemia is a rare condition that can result in acute hypocalcemia in dogs.[442] The rapid correction of cancer-associated hypercalcemia (e.g., with tumor excision and chemotherapy) can be associated with hypocalcemia and low PTH concentration, but hypocalcemia is usually minor and transient.

Both acute hypermagnesemia and severe magnesium depletion may suppress PTH secretion.[58,422,521] As with hypocalcemia, mild acute hypomagnesemia stimulates PTH secretion, but severe magnesium depletion decreases PTH secretion, increases end-organ resistance to PTH, and may impair calcitriol synthesis. The end-organ resistance to PTH that develops during magnesium depletion may persist for days after magnesium repletion and resumption of normal PTH concentrations in humans. Until recently, hypomagnesemia has been reported rarely in dogs and cats with hypoparathyroidism. Normal serum tMg does not guarantee a normal iMg concentration because there is substantial discordance between these two measurements.

Magnesium depletion can cause functional hypoparathyroidism, and measurement of serum iMg concentration is recommended to exclude or identify this form of hypoparathyroidism. Serum tMg concentrations in dogs and cats with primary hypoparathyroidism usually have been normal when measured.[82,169] In 357 dogs with primary hypoparathyroidism, mean iCa and mean PTH concentrations were below the reference range.[477] The iMg concentration was below the reference range in 39%, within the reference range in 55%, and above the reference range in 6% of dogs with hypoparathyroidism. Of the 55% of dogs with iMg within the reference range, 69% had an iMg concentration within the lower half of the reference range, and only 31% had an iMg concentration within the upper half of the reference range.

Despite the relative paucity of published reports from cats, hypoparathyroidism was diagnosed in 27 cats during a 2-year period.[477] Of cats with hypoparathyroidism, 59% were domestic shorthairs, 22% were an unspecified breed, and 15% were Siamese. Mean serum iCa concentration was below the reference range, and mean PTH concentration was in the lower half of the reference range. The iMg concentration was below the reference range in 37%, within the reference range in 59%, and above the reference range in 4%. Of the 59% of cats with iMg within the reference range, 88% had an iMg concentration within the lower half of the reference range, and only 12% had an iMg concentration within the upper half of the reference range. These results suggest that a large number of dogs and cats with hypoparathyroidism also exhibit subnormal or marginal iMg concentrations. The impact of magnesium supplementation in the treatment of hypoparathyroidism should be investigated. Although primary hypoparathyroidism is usually diagnosed in older cats, it has been reported in a 6-month-old kitten initially evaluated for lethargy, inappetence, muscle tremors, and seizures.[31]

Most causes of primary hypoparathyroidism have been attributed to immune destruction of parathyroid tissue. Early reports of hypoparathyroidism in dogs and cats did not consistently evaluate magnesium status and used tMg when it was reported. Based on discordance of magnesium status using iMg versus tMg, hypomagnesemia based on tMg assessments may have underestimated a role for hypomagnesemia in the genesis of hypoparathyroidism in animals. Hypomagnesemia may decrease cell membrane receptor sensitivity to iCa and PTH, as well as decrease PTH synthesis.[300] Serum iMg concentration should be measured when iCa and PTH concentrations are determined.

The potential role of magnesium depletion in the development of postthyroidectomy hypocalcemia in cats has not been explored. Magnesium depletion could play a role in the development of postoperative hypocalcemia in cats with hyperthyroidism because hyperthyroidism can be associated with magnesium depletion.[169]

Canine distemper virus (CDV)-induced parathyroid hypofunction may contribute to development of hypocalcemia. Dogs infected with CDV had reduced serum tCa concentrations associated with ultrastructural evidence of parathyroid gland inactivity, degeneration, and viral inclusions.[554]

Miscellaneous Causes of Hypocalcemia

Metabolites of ethylene glycol can chelate calcium and become deposited in soft tissues, resulting in hypocalcemia. Both dogs and cats exhibit hypocalcemia after ethylene glycol ingestion.[518] Seizures have been observed in dogs within hours of ingestion; renal function was normal at this time (Chew, personal observations). Hypocalcemia often develops later when renal function is severely reduced from acute renal failure and when hyperphosphatemia is severe.

Acute decreases in iCa concentrations are most commonly caused by acute respiratory alkalosis in humans.[422] It is likely that this phenomenon also occurs in dogs and cats subjected to the stresses of hypocalcemia and a visit to a veterinary clinic. This could explain the phenomenon of mild stress-induced seizures or tetany in dogs that have hypocalcemia, as the alkalosis shifts some calcium to the protein-bound state, causing more severe ionized hypocalcemia.

TREATMENT OF HYPOCALCEMIA

Puerperal tetany is the condition most likely to require correction of hypocalcemia acutely, but chronic treatment is not needed. Hypoparathyroidism is the only condition requiring acute and chronic treatment to alleviate clinical signs of hypocalcemia. Other conditions associated with hypocalcemia are transient or result in minimal decreases in serum calcium concentration, do not cause obvious clinical signs, and only occasionally necessitate calcium replacement therapy. No treatment is indicated for hypocalcemia attributable entirely to hypoalbuminemia or hypoproteinemia, assuming that the iCa fraction is normal.

Treatment is individualized based on severity of clinical signs, magnitude of hypocalcemia, rapidity of decline in serum calcium concentration, and trend of serial serum calcium measurements (i.e., further decrease or stability). Aggressive treatment is prescribed for patients with severe clinical signs of hypocalcemia, patients with severe ionized hypocalcemia with or without signs, and patients in which serum calcium concentration is steadily or rapidly declining. Acute, subacute, and chronic rescue treatment regimens are available using supplementation with calcium salts and vitamin D metabolites. The goal of therapy is to increase serum calcium concentration to a level that alleviates the signs of hypocalcemia, minimizes the likelihood of the development of hypercalcemia, and reduces the magnitude of hypercalciuria (especially in patients with hypoparathyroidism). It is usually not necessary or desirable to return serum calcium concentration completely to normal because many clinical signs improve dramatically with slight increases in serum calcium concentration, and the consequences of overcorrection can be serious. For suspected temporary postsurgical hypoparathyroidism, it is desirable to keep the serum calcium concentration relatively low to maximize compensatory hypertrophy of remaining parathyroid glands.

In patients with hypoparathyroidism, no treatment regimen completely compensates for the full range of physiologic actions of the absent PTH. Vitamin D metabolite treatment corrects the low intestinal absorption of calcium but does not completely protect the kidneys from hypercalciuria as would occur in the presence of PTH. Similarly, vitamin D metabolites do not exert as powerful an effect on bone in the absence of PTH. Replacement therapy with once-daily subcutaneous injections of human PTH (1-34) in human subjects was highly effective in providing good 24-hour control of serum calcium concentration.[562] Use of synthetic human amino-terminal PTH for treatment of veterinary patients is possible because the amino-terminal portions of PTH are highly conserved, function *in vivo* in animals, and would be unlikely to elicit an immune response.

Hypocalcemia severe enough to cause clinical signs should be anticipated in dogs undergoing parathyroidectomy as treatment for hypercalcemia related to a parathyroid gland adenoma. Animals with very high concentrations of serum calcium, PTH, and serum ALP may be at greater risk of developing postoperative hypocalcemia. Postoperative hypocalcemia in this instance is the consequence of acute hypoparathyroidism resulting from chronic suppression of remaining parathyroid glands and calcium uptake into "hungry" bones. Hypocalcemia should be anticipated in cats that undergo bilateral thyroidectomy because up to 30% of cats can be expected to have transiently lowered serum calcium concentrations.

Therapy should be instituted before the development of tetany. Preemptive therapy to increase serum calcium concentration may be a good choice for animals with marked hypocalcemia with no apparent clinical signs or for those in which serum calcium concentration is steadily or rapidly declining. Prophylactic therapy to prevent hypocalcemia in dogs undergoing surgery for hyperparathyroidism should be considered, especially in dogs with severe hypercalcemia. Active vitamin D metabolites should be started before surgery in these instances because there is a lag time until maximal effect is achieved. Vitamin D metabolites given at the time of surgery or just after surgery fail to prevent development of hypocalcemia.

Autotransplantation of normal parathyroid glands is a treatment option to minimize postoperative hypocalcemia when it is obvious that damage has been done to the parathyroid glands during surgery (bilateral extracapsular thyroidectomy). Autotransplantation of normal parathyroid glands was studied in experimental cats following bilateral extracapsular thyroparathyroidectomy.[387] External parathyroid glands were harvested and dissected from thyroid tissue, and small pieces of parathyroid tissue were embedded into the sternohyoideus muscle. Cats showed an average decrease of 44% in serum tCa with the nadir occurring 1.9 days following surgery. Hypocalcemia was present a median of 14 days in cats having parathyroidectomy and autotransplantation in this study compared with a median of 71 days in cats of a previous report involving parathyroidectomy without autotransplantation.[178] Seven of eight cats with autotransplantation of parathyroid glands regained normocalcemia within 20 days without oral calcium salt supplementation.[387]

Acute Management of Hypocalcemia Causing Tetany or Seizures

Tetany or seizures caused by hypocalcemia require treatment with intravenously administered calcium salts. Calcium is administered to effect, at a dosage of 5 to 15 mg/kg of elemental calcium (0.5 to 1.5 mL/kg of 10% calcium gluconate) over a 10- to 20-minute period.[111,169,406,407] The calcium content of different calcium salts varies considerably (Table 6-5). There is no difference in effectiveness of calcium salts administered intravenously to correct hypocalcemia when the dose is based on elemental calcium content. Calcium gluconate is often the calcium salt of choice because it is nonirritating if the solution is inadvertently injected perivascularly. In contrast, calcium chloride is extremely irritating to tissues but provides more elemental calcium in each milliliter of solution (see Table 6-5).

The heart rate and electrocardiogram should be monitored during acute infusions of calcium salts. Bradycardia may signal the onset of cardiotoxicity arising from excessively rapid infusion of calcium. Sudden elevation of the ST segment or shortening of the QT interval also may indicate cardiotoxicity resulting from the calcium infusion. Not all clinical signs abate immediately after acute correction of hypocalcemia; some may persist for 30 to 60 minutes. Nervousness, panting, and behavioral changes may persist despite return of normocalcemia during this period, perhaps reflecting a lag in equilibration between cerebrospinal fluid and ECF calcium concentrations.[169,274,460] Hyperthermia that resulted from increased muscle activity or seizures may also take time to dissipate.

Subacute Management of Hypocalcemia

The initial bolus injection of elemental calcium can be expected to decrease signs of hypocalcemia for as little as 1 hour to as long as 12 hours if the underlying cause of hypocalcemia has not been corrected. Vitamin D metabolites should be administered as soon as possible because some of them require a few days before intestinal calcium transport is maximized. Calcitriol exerts initial effects on the intestine within 3 to 4 hours.[549] Additional parenteral calcium salt administration is necessary until therapy with vitamin D metabolites is effective at maintaining serum calcium concentration at an acceptable level.

Multiple intermittent intravenous injections of calcium salts can be administered to control clinical signs, but this method is not recommended because wide fluctuations in serum calcium concentration are observed. Instead, continuous intravenous infusion of calcium is recommended at 60 to 90 mg/kg/day elemental calcium (2.5 to 3.75 mg/kg/hr) until oral medications provide control of serum calcium concentration.[82,169,406,407] Initial doses in the higher range are administered to patients with more severe hypocalcemia, and the dose decreased according to the serum calcium concentration achieved. The intravenous dose of calcium is further reduced as oral calcium salts and vitamin D metabolites become more effective.

Ten milliliters of 10% calcium gluconate provides 93 mg of elemental calcium. A convenient method for infusing calcium is available when intravenous fluids are given at a maintenance volume of 60 mL/kg/day (2.5 mL/kg/hr). Approximately 1, 2, or 3 mg/kg/hr elemental calcium is provided by adding 10, 20, or 30 mL of 10% calcium gluconate, respectively, to each 250-mL bag of fluids. Calcium salts should not be added to fluids that contain lactate, acetate, bicarbonate, or phosphates because calcium salt precipitates can occur. Alkalinizing fluids that contain or generate bicarbonate should be avoided because they can decrease iCa and may unmask clinical signs of hypocalcemia in animals with borderline hypocalcemia.

Subcutaneous administration of calcium gluconate has been regarded as safe for use in dogs with hypocalcemia when diluted to at least 1:1 by volume. The use of calcium chloride is too caustic to ever be given subcutaneously. However, a recent report raises concerns about the safety of calcium gluconate administration subcutaneously. A 6-month-old border collie with hypoparathyroidism was initially treated with intravenous calcium gluconate, followed by oral calcitriol and calcium carbonate.[469] This dog then received subcutaneous calcium gluconate three times daily for 2 days, and calcium gluconate was diluted as previously recommended. Fever and pain, swelling, and erythema of the ventral abdomen were obvious after 2 days of subcutaneous calcium gluconate treatments. Initial skin biopsy revealed calcinosis cutis with pyogranulomatous dermatitis and dermoepidermal separation. The dog's condition worsened; ulceration involving about 80% of the skin developed over the trunk; and the dog was euthanized. A second skin biopsy revealed severe pyogranulomatous panniculitis with mineralization of adipocytes.

Reports of this reaction to the subcutaneous administration of calcium gluconate had not previously been reported in dogs despite its extensive use by some institutions (Feldman, personal communication, 2005). Unfortunately, we are aware of at least three other dogs with similar severe reactions to the subcutaneous administration of properly diluted calcium gluconate as treatment for primary hypoparathyroidism, resulting in euthanasia for most (Chew, personal communications, 2003, 2004). Differences in an individual animal's susceptibility to the effects of calcium salts on subcutaneous tissues could account for severe reactions in some dogs. All dogs with this severe tissue reaction were also receiving calcitriol, which may potentiate more local dramatic effects in the subcutaneous tissues as compared with less active vitamin D metabolites (cholecalciferol, ergocalciferol, and dihydrotachysterol) commonly used in the past.

There are only two reports of cats with primary hypoparathyroidism that were treated with subcutaneous administration of calcium gluconate. No adverse effects were noted in one report.[184] Iatrogenic calcinosis cutis, skin necrosis, and scarring occurred at sites of diluted calcium gluconate injection and sites where injected

TABLE 6-5 Treatment of Hypocalcemia

Drug	Preparation	Calcium Content	Dose	Comment
Parenteral Calcium*				
Calcium gluconate	10% solution	9.3 mg of Ca/mL	a. Slow IV to effect (0.5-1.5 mL/kg IV)	Stop if bradycardia or shortened QT interval occurs
			b. 5-15 mg/kg/hr IV	Infusion to maintain normal Ca
			c. SQ diluted calcium salts	SQ calcium salts can cause severe skin necrosis/mineralization; no longer recommended as safe
Calcium chloride	10% solution	27.2 mg of Ca/mL	5-15 mg/kg/hr IV	Only given IV because extremely caustic perivascularly
Oral Calcium†				
Calcium carbonate	Many sizes	40% tablet	25-50 mg/kg/day	Most common calcium supplement
Calcium lactate	325- and 650-mg tablets	13% tablet	25-50 mg/kg/day	
Calcium chloride	Powder	27.2%	25-50 mg/kg/day	May cause gastric irritation
Calcium gluconate	Many sizes	10%	25-50 mg/kg/day	

Vitamin D	Dose	**Time for maximal effect to occur:**	**Time for toxicity effect to resolve:**
Vitamin D_2 (ergocalciferol)	Initial: 4000-6000 U/kg/day; Maintenance: 1000-2000 U/kg once daily to once weekly	5-21 days	1-18 weeks
Dihydrotachysterol	Initial: 0.02-0.03 mg/kg/day Maintenance: 0.01-0.02 mg/kg every 24-48 hours	1-7 days	1-3 weeks
1,25-$(OH)_2D_3$ (calcitriol)	Initial: 20-30 ng/kg/day for 3-4 days Maintenance: 5-15 ng/kg/day	1-4 days	2-14 days

**Do not mix calcium solutions with bicarbonate-containing fluids as precipitation may occur.*

†Calculate dose on elemental calcium content. IV, *Intravenous;* SQ, *subcutaneous.*

fluids pooled in one cat.[458] This cat survived. Because of the severity of adverse reactions that have recently been observed in dogs and a cat, the administration of subcutaneous fluids containing calcium gluconate is no longer recommended as a safe and predictable treatment.

Subacute and Chronic Maintenance

Supplemental elemental calcium is administered orally (see Table 6-5) to guarantee adequate calcium for intestinal absorption after treatment with vitamin D metabolites. Oral calcium administered by pill or slurry is most important during initial treatment, especially if the animal is not eating. Active intestinal transport of calcium is under the control of calcitriol when calcium intake is low, but vitamin D–independent (passive) intestinal absorption of calcium occurs when calcium intake is high. The passive mechanisms for intestinal calcium transport can be used therapeutically before the actions of vitamin D take effect in the intestine. In most patients, normal dietary intake of calcium is sufficient to maintain adequate serum calcium concentrations in the presence of vitamin D metabolite treatment. Consequently, oral calcium salt supplementation can be tapered and discontinued in many instances as vitamin D compounds reach maximal effect.

Calcium carbonate is the most widely used oral preparation of the calcium salts because it contains the greatest percentage of elemental calcium. This approach allows fewer pills to be administered. The degree of calcium ionization from its salt and its bioavailability for absorption vary for each calcium salt and with conditions in the intestine. Consequently, it is not a simple matter to determine the bioavailable elemental calcium content of a specific oral calcium salt. Oral calcium is usually administered at 25 to 50 mg/kg/day elemental calcium in divided doses. Oral calcium carbonate serves as an intestinal phosphate binder in addition to providing calcium for intestinal absorption. It is advisable to continue oral calcium carbonate therapy for its intestinal phosphate-binding effects if serum phosphorus concentration remains increased. Lower serum phosphorus concentrations may allow increased endogenous synthesis of calcitriol because phosphate inhibits renal synthesis of calcitriol.

Vitamin D preparations (see Table 6-5) include ergocalciferol, cholecalciferol, dihydrotachysterol (DHT), 25-hydroxycholecalciferol (calcidiol), 1α-hydroxycholecalciferol, and calcitriol. Ergocalciferol, DHT, and calcitriol are the preparations most commonly used in veterinary medicine. Lifelong treatment with some form of vitamin D metabolite is necessary for patients with primary hypoparathyroidism or postoperative hypocalcemia that fails to resolve spontaneously.

Ergocalciferol is favored by some because of its low cost,[422] but it has several features that make it the least attractive agent for treatment of hypocalcemia. Ergocalciferol and its immediate metabolite, 25-hydroxyergocalciferol, have low VDR avidity; thus high doses are necessary. Ergocalciferol is highly lipid soluble, and several weeks are required to saturate body stores and achieve a maximal effect. It also has a long half-life. Consequently, prolonged periods of hypercalcemia occur after overdose with ergocalciferol. In addition, there is extreme individual variation in the dose of ergocalciferol required to achieve a target serum calcium concentration. Use of loading doses reduces the time required to achieve a maximal effect (see Table 6-5).

DHT is a synthetic vitamin D analogue with onset of maximal effect and biologic half-life between those of ergocalciferol and calcitriol. The polarity and lower dose requirements of DHT limit its storage in fat compared with ergocalciferol. Toxicity resulting from hypercalcemia still can be prolonged (up to 30 days), and there is wide variation in the dose required to achieve a target serum calcium concentration. Use of loading doses reduces the time to maximal effect.

Calcitriol is the vitamin D metabolite of choice to provide calcemic actions because it has the most rapid onset of maximal action and the shortest biologic half-life. Calcitriol is approximately 1000 times as effective as parent vitamin D and 500 times as effective as its precursor, calcidiol (25-hydroxyvitamin D), in binding to the VDR. The dose of calcitriol can be adjusted frequently because of its short half-life and rapid effects on serum calcium concentration. If hypercalcemia occurs, it abates quickly after dose reduction. The half-life of calcitriol in blood is 4 to 6 hours, whereas its biologic half-life is 2 to 4 days. Loading protocols for use of calcitriol in animals have not been reported, but it is logical to use a loading protocol when more rapid correction of serum calcium concentration is desirable. A calcitriol dosage of 30 to 60 ng/kg/day has been recommended.[82,169] This dosage may be satisfactory as a loading dose, but in our experience it is too high for chronic maintenance therapy. Calcitriol dosages for chronic maintenance therapy in humans range from 10 to 40 ng/kg/day, and doses are divided and given twice daily.[217,422,562] We have used loading dosages of 20 to 30 ng/kg/day for 3 to 4 days and maintenance dosages of 10 to 20 ng/kg/day in most patients. The dose of calcitriol is divided and given twice daily to ensure sustained priming effects on intestinal epithelium for calcium transport. Calcitriol is commercially available in 0.25- and 0.50-μg capsules (250 and 500 ng per capsule, respectively; Rocaltrol, Hoffman-LaRoche, Basel, Switzerland). It is likely that reformulation of calcitriol in doses suitable for a variety of animal sizes will be necessary. It may be useful to prescribe calcitriol in liquid formulation so that small adjustments in dosage can be made accurately. A number of specialty pharmacies reformulate human drugs for veterinary use and can create any calcitriol dose needed.

CLINICAL FOLLOW-UP AND POTENTIAL COMPLICATIONS

Periods of hypocalcemia and hypercalcemia occur sporadically in patients during initial efforts to manage serum calcium concentration. Daily measurement of serum tCa concentration during stabilization is necessary. Weekly serum calcium measurements should suffice during maintenance therapy until target serum calcium concentration has been achieved and maintained. Measurement of serum tCa concentration is recommended every 3 months thereafter in animals with permanent hypoparathyroidism. Serum calcium concentration should be adjusted to just below the reference range. This not only lessens the likelihood that hypercalcemia will develop but also reduces the magnitude of hypercalciuria that occurs in patients with PTH deficiency. Maintaining a mildly decreased serum calcium concentration also ensures a continued stimulus for hypertrophy of the remaining parathyroid tissue in patients with postoperative hypoparathyroidism.

A change in dosage of vitamin D metabolites should only occur after maximal effect has occurred and should be altered gradually. The time lag for maximal effect varies with the different vitamin D metabolites (see Table 6-5). Dosage increases of 10% to 25% are recommended when serum calcium concentration is still below the target level.[406,407] Vitamin D metabolite and calcium salt supplementation should be discontinued temporarily in patients that develop hypercalcemia.

Hypercalcemia is a serious adverse effect of treatment that can result in death or renal damage causing acute or CRF.[106,111,290] Early signs of hypercalcemia should be explained to owners, who should be instructed to seek veterinary attention immediately if clinical signs suggest hypercalcemia. Clinical signs of hypercalcemia that clients are likely to recognize include polydipsia, polyuria, anorexia, vomiting, and lethargy. Animals with severe hypercalcemia require hospitalization. Fluids, furosemide, corticosteroids, bisphosphonates, calcitonin, or some combination may be required. All patients with symptomatic, vitamin D metabolite-induced hypercalcemia should be given a calcium-restricted diet because increased intestinal absorption of calcium contributes substantially to the development of hypercalcemia in hypervitaminosis D.

Patients that maintain serum iCa concentrations in the target zone are often managed successfully for years. Twenty-four of 25 dogs with primary hypoparathyroidism were managed successfully for more than 5 years,[169] and long-term management was successful in a small number of cats.[404] Patients that develop episodic or prolonged hypercalcemia during treatment have a poor prognosis. Management with calcitriol is easier and more successful in inducing and maintaining serum iCa concentrations in the target zone than are older therapeutic approaches.

Hypercalciuria, nephrocalcinosis, urolithiasis, and reduced renal function have occurred in humans treated for chronic hypoparathyroidism.[217,521,562] As many as 80% of human patients treated for 2 years or longer have decreased creatinine clearance.[562] These abnormalities can be attributed to episodes of hypercalcemia and hyperphosphatemia and to hypercalciuria that occurs in the absence of the actions of PTH on the renal tubules. In the absence of PTH, hypercalciuria occurs more readily at all serum iCa concentrations and is especially severe as iCa concentrations approach the normal range, which increases the filtered load of calcium. Nephrocalcinosis, reduced renal function, and CRF have also been suspected in veterinary patients receiving long-term treatment for hypoparathyroidism, but the risk for these disorders has not been critically evaluated.[406]

Vitamin D metabolite treatment is gradually tapered and then discontinued in patients with postsurgical hypoparathyroidism because hypocalcemia is usually transient. Most cats are able to maintain normal serum iCa concentrations 2 weeks after thyroidectomy, although some may take as long as 3 months. Dogs with hypocalcemia usually require 6 to 12 weeks of treatment after removal of a parathyroid gland adenoma. A reduction in dose of vitamin D metabolites is usually begun 1 month after initiation of therapy. If serum iCa concentration declines substantially, the previous dose is resumed, and reduction is attempted again 1 or 2 months later. Permanent hypoparathyroidism is likely if failure to maintain acceptable serum iCa concentration occurs after reduction of the vitamin D metabolite dose at 3 months.

REFERENCES

1. Abou-Samra AB, Juppner H, Force T, et al: Expression cloning of a common receptor for parathyroid hormone and parathyroid hormone-related peptide from rat osteoblast-like cells: a single receptor stimulates intracellular accumulation of both cAMP and inositol trisphosphates and increases intracellular free calcium, *Proc Natl Acad Sci U S A* 89:2732-2736, 1992.
2. Abrams KL: Hypocalcemia associated with administration of sodium bicarbonate for salicylate intoxication in a cat, *J Am Vet Med Assoc* 191:235-236, 1987.
3. Adami S, Zamberlan N: Adverse effects of bisphosphonates. A comparative review, *Drug Saf* 14:158-170, 1996.
4. Adams JS, Sharma OP, Diz MM, et al: Ketoconazole decreases the serum 1,25-dihydroxyvitamin D and calcium concentration in sarcoidosis-associated hypercalcemia, *J Clin Endocrinol Metab* 70:1090-1095, 1990.
5. Adin DB, Taylor AW, Hill RC, et al: Intermittent bolus injection versus continuous infusion of furosemide in normal adult greyhound dogs, *J Vet Intern Med* 17:632-636, 2003.
6. Adler AJ, Ferran N, Berlyne GM: Effect of inorganic phosphate on serum ionized calcium concentration in vitro: a reassessment of the "trade-off hypothesis," *Kidney Int* 28:932-935, 1985.
7. Almaden Y, Canalejo A, Ballesteros E, et al: Regulation of arachidonic acid production by intracellular calcium in parathyroid cells: effect of extracellular phosphate, *J Am Soc Nephrol* 13:693-698, 2002.
8. Almaden Y, Felsenfeld AJ, Rodriguez M, et al: Proliferation in hyperplastic human and normal rat parathyroid glands: role of phosphate, calcitriol, and gender, *Kidney Int* 64:2311-2317, 2003.
9. Almirall J, Lopez T, Vallve M, et al: Safety and efficacy of sevelamer in the treatment of uncontrolled hyperphosphataemia of haemodialysis patients, *Nephron Clin Pract* 97:c17-22, 2004.

10. Amin M, Fawzy A, Hamid MA, et al: Pulmonary hypertension in patients with chronic renal failure: role of parathyroid hormone and pulmonary artery calcifications, *Chest* 124:2093-2097, 2003.
11. Anderson TE, Legendre AM, McEntee MM: Probable hypercalcemia of malignancy in a cat with bronchogenic adenocarcinoma, *J Am Anim Hosp Assoc* 36:52-55, 2000.
12. Anthony LB, May ME, Oates JA: Case report: lanreotide in the management of hypercalcemia of malignancy. *Am J Med Sci* 309:312-314, 1995.
13. Arceneaux KA, Taboada J, Hosgood G: Blastomycosis in dogs: 115 cases (1980-1995), *J Am Vet Med Assoc* 213:658-664, 1998.
14. Aroch I, Ohad DG, Baneth G: Paresis and unusual electrocardiographic signs in a severely hypomagnesaemic, hypocalcaemic lactating bitch, *J Small Anim Pract* 39:299-302, 1998.
15. Aroch I, Segev G, Klement E, et al: Fatal Vipera xanthina palestinae envenomation in 16 dogs, *Vet Hum Toxicol* 46:268-272, 2004.
16. Atkins CE, Tyler R, Greenlee P: Clinical, biochemical, acid-base, and electrolyte abnormalities in cats after hypertonic sodium phosphate enema administration, *Am J Vet Res* 46:980-988, 1985.
17. Aubin JE, Heersche JN: Vitamin D and osteoblasts. In Feldman D, editor: *Vitamin D*, New York, 1997, Academic Press, pp. 313-328.
18. Aucella F, Scalzulli RP, Gatta G, et al: Calcitriol increases burst-forming unit-erythroid proliferation in chronic renal failure. A synergistic effect with r-HuEpo, *Nephron Clin Pract* 95:c121-127, 2003.
19. Austad R, Bjerkas E: Eclampsia in the bitch, *J Small Anim Pract* 17:793-798, 1976.
20. Bai M, Quinn S, Trivedi S, et al: Expression and characterization of inactivating and activating mutations in the human Ca2+o-sensing receptor, *J Biol Chem* 271:19537-19545, 1996.
21. Bai M: Structure-function relationship of the extracellular calcium-sensing receptor, *Cell Calcium* 35:197-207, 2004.
22. Banerjee D, Asif A, Striker L, et al: Short-term, high-dose pamidronate-induced acute tubular necrosis: the postulated mechanisms of bisphosphonate nephrotoxicity, *Am J Kidney Dis* 41:E18, 2003.
23. Barber PJ, Elliott J, Torrance AG: Measurement of feline intact parathyroid hormone: assay validation and sample handling studies, *J Small Anim Pract* 34:614-620, 1993.
24. Barber PJ, Elliott J: Feline chronic renal failure: calcium homeostasis in 80 cases diagnosed between 1992 and 1995, *J Small Anim Pract* 39:108-116, 1998.
25. Barber PJ, Elliott J: Study of calcium homeostasis in feline hyperthyroidism, *J Small Anim Pract* 37:575-582, 1996.
26. Barber PJ, Rawlings JM, Markwell PJ, et al: Effect of dietary phosphate restriction on renal secondary hyperparathyroidism in the cat, *J Small Anim Pract* 40:62-70, 1999.
27. Barber PJ, Torrance AG, Elliott J: Carboxyl fragment interference in assay of feline parathyroid hormone, *J Vet Intern Med* 8:168, 1994.
28. Barr FJ, Patterson MW, Lucke VM, et al: Hypercalcemic nephropathy in three dogs: sonographic appearance, *Vet Radiol* 30:169-173, 1989.
29. Barrett S, Sheafor SE, Hillier A, et al: Challenging cases in internal medicine "What's your diagnosis?" *Vet Med* 93:35-44, 1998.
30. Basoglu A, Sevinc M, Sen I, et al: The blocking effect of verapamil in hypercalcemic dogs, *Turkish J Vet Anim Sci* 21:331-333, 1997.
31. Bassett JR: Hypocalcemia and hyperphosphatemia due to primary hypoparathyroidism in a six-month-old kitten, *J Am Anim Hosp Assoc* 34:503-507, 1998.
32. Behrend EN, Kemppainen R: Adrenocortical disease. In August J, editor: *Consultations in feline internal medicine 4,* Philadelphia, 1980, WB Saunders, pp. 159-168.
33. Bennett PF, DeNicola DB, Bonney P, et al: Canine anal sac adenocarcinomas: clinical presentation and response to therapy, *J Vet Intern Med* 16:100-104, 2002.
34. Bereket A, Erdogan T: Oral bisphosphonate therapy for vitamin D intoxication of the infant, *Pediatrics* 111:899-901, 2003.
35. Berenson J, Hirschberg R: Safety and convenience of a 15-minute infusion of zoledronic acid, *Oncologist* 9:319-329, 2004.
36. Berenson JR, Rosen L, Vescio R, et al: Pharmacokinetics of pamidronate disodium in patients with cancer with normal or impaired renal function, *J Clin Pharmacol* 37:285-290, 1997.
37. Berger B, Feldman EC: Primary hyperparathyroidism in dogs: 21 cases (1976-1986), *J Am Vet Med Assoc* 191:350-356, 1987.
38. Bergman SM, O'Mailia J, Krane NK, et al: Vitamin-A-induced hypercalcemia: response to corticosteroids. *Nephron* 50:362-364, 1988.
39. Bernardi RJ, Johnson CS, Modzelewski RA, et al: Antiproliferative effects of 1alpha,25-dihydroxyvitamin D(3) and vitamin D analogs on tumor-derived endothelial cells, *Endocrinology* 143:2508-2514, 2002.
40. Bhattacharya SK, Luther RW, Pate JW, et al: Soft tissue calcium and magnesium content in acute pancreatitis in the dog: calcium accumulation, a mechanism for hypocalcemia in acute pancreatitis, *J Lab Clin Med* 105:422-427, 1985.
41. Bienzle D, Jacobs RM, Lumsden JH: Relationship of serum total calcium to serum albumin in dogs, cats, horses and cattle, *Can Vet J* 34:360-364, 1993.
42. Bienzle D, Silverstein DC, Chaffin K: Multiple myeloma in cats: variable presentation with different immunoglobulin isotypes in two cats, *Vet Pathol* 37:364-369, 2000.
43. Bilezikian JP, Singer FR: Acute management of hypercalcemia due to parathyroid hormone and parathyroid hormone-related protein. In Bilezikian JP, Levine MA, Marcus R, editors: *The parathyroids,* New York, 1994, Raven Press, pp. 359-372.
44. Bilezikian JP: Clinical utility of assays for parathyroid hormone-related protein, *Clin Chem* 38:179-181, 1992.
45. Biller DS, Bradley GA, Partington BP: Renal medullary rim sign: ultrasonographic evidence of renal disease, *Vet Radiol Ultrasound* 33:286-290, 1992.
46. Birchard SJ, Peterson ME, Jacobson A: Surgical treatment of feline hyperthyroidism: results of 85 cases, *J Am Anim Hosp Assoc* 20:705-709, 1984.
47. Biswas PN, Wilton LV, Shakir SA: Pharmacovigilance study of alendronate in England, *Osteoporos Int* 14:507-514, 2003.
48. Black KS, Mundy GR: Other causes of hypercalcemia: local and ectopic secretion syndromes. In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* New York, 1994, Raven Press, pp. 341-358.
49. Blackwood L, Sullivan M, Lawson H: Radiographic abnormalities in canine multicentric lymphoma: a review of 84 cases, *J Small Anim Pract* 38:62-69, 1997.
50. Block GA, Martin KJ, de Francisco AL, et al: Cinacalcet for secondary hyperparathyroidism in patients receiving hemodialysis, *N Engl J Med* 350:1516-1525, 2004.

51. Blumenthal SR, Williams TC, Barbee RW, et al: Effects of citrated whole blood transfusion in response to hemorrhage, *Lab Anim Sci* 49:411-417, 1999.
52. Boden SD, Kaplan FS: Calcium homeostasis, *Orthop Clin North Am* 21:31-42, 1990.
53. Body JJ, Coleman RE, Piccart M: Use of bisphosphonates in cancer patients, *Cancer Treat Rev* 22:265-287, 1996.
54. Body JJ: Clinical research update: zoledronate, *Cancer* 80(suppl):1699-1701, 1997.
55. Body JJ: Hypercalcemia of malignancy, *Semin Nephrol* 24:48-54, 2004.
56. Bolliger AP, Graham PA, Richard V, et al: Detection of parathyroid hormone-related protein in cats with humoral hypercalcemia of malignancy, *Vet Clin Pathol* 31:3-8, 2002.
57. Bourdeau A, Souberbielle JC, Bonnet P, et al: Phospholipase-A2 action and arachidonic acid metabolism in calcium-mediated parathyroid hormone secretion, *Endocrinology* 130:1339-1344, 1992.
58. Bourke E, Delaney V: Assessment of hypocalcemia and hypercalcemia, *Clin Lab Med* 13:157-181, 1993.
59. Bowers GN Jr, Brassard C, Sena SF: Measurement of ionized calcium in serum with ion-selective electrodes: a mature technology that can meet the daily service needs, *Clin Chem* 32:1437-1447, 1986.
60. Bowman AR, Epstein S: Drug and hormone effects on vitamin D metabolism. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 797-829.
61. Bregazzi VS, Fettman MJ, Twedt DC: Hypergastrinemia associated with hypercalcemia in the dog, *J Vet Intern Med* 14:389, 2000.
62. Breitwieser GE, Miedlich SU, Zhang M: Calcium sensing receptors as integrators of multiple metabolic signals, *Cell Calcium* 35:209-216, 2004.
63. Brenza HL, DeLuca HF: Regulation of 25-hydroxyvitamin D3 1alpha-hydroxylase gene expression by parathyroid hormone and 1,25-dihydroxyvitamin D3, *Arch Biochem Biophys* 381:143-152, 2000.
64. Brenza HL, Kimmel-Jehan C, Jehan F, et al: Parathyroid hormone activation of the 25-hydroxyvitamin D3-1alpha-hydroxylase gene promoter, *Proc Natl Acad Sci U S A* 95:1387-1391, 1998.
65. Breslau NA: Normal and abnormal regulation of 1,25-(OH)2D synthesis, *Am J Med Sci* 296:417-425, 1988.
66. Bringhurst FR: Circulating forms of parathyroid hormone: peeling back the onion, *Clin Chem* 49:1973-1975, 2003.
67. Bronner F: Mechanisms and functional aspects of intestinal calcium absorption, *J Exp Zoolog A Comp Exp Biol* 300:47-52, 2003.
68. Bronner F: Mechanisms of intestinal calcium absorption, *J Cell Biochem* 88:387-393, 2003.
69. Brossard JH, Cloutier M, Roy L, et al: Accumulation of a non-(1-84) molecular form of parathyroid hormone (PTH) detected by intact PTH assay in renal failure: importance in the interpretation of PTH values, *J Clin Endocrinol Metab* 81:3923-3929, 1996.
70. Brown AJ, Dusso A, Lopez-Hilker S, et al: 1,25-(OH)2D receptors are decreased in parathyroid glands from chronically uremic dogs, *Kidney Int* 35:19-23, 1989.
71. Brown AJ, Zhong M, Finch J, et al: Rat calcium-sensing receptor is regulated by vitamin D but not by calcium, *Am J Physiol* 270:F454-460, 1996.
72. Brown EM, Conigrave A, Chattopadhyay N: Receptors and signaling for calcium ions, In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* San Diego, 2001, Academic Press, pp. 127-142.
73. Brown EM, Gamba G, Riccardi D, et al: Cloning and characterization of an extracellular Ca(2+)-sensing receptor from bovine parathyroid, *Nature* 366:575-580, 1993.
74. Brown EM, Hebert SC: Calcium-receptor-regulated parathyroid and renal function, *Bone* 20:303-309, 1997.
75. Brown EM, Pollak M, Seidman CE, et al: Calcium-ion-sensing cell-surface receptors, *N Engl J Med* 333:234-240, 1995.
76. Brown EM: Extracellular Ca2+ sensing, regulation of parathyroid cell function, and role of Ca2+ and other ions as extracellular (first) messengers, *Physiol Rev* 71:371-411, 1991.
77. Brown EM: Homeostatic mechanisms regulating extracellular and intracellular calcium metabolism. In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* New York, 1994, Academic Press, pp. 15-54.
78. Brown JE, Neville-Webbe H, Coleman RE: The role of bisphosphonates in breast and prostate cancers, *Endocr Relat Cancer* 11:207-224, 2004.
79. Brown MA, Haughton MA, Grant SF, et al: Genetic control of bone density and turnover: role of the collagen 1alpha1, estrogen receptor, and vitamin D receptor genes, *J Bone Miner Res* 16:758-764, 2001.
80. Brownie CF: Confusion over jasmine and jessamine, *J Am Vet Med Assoc* 191:613-614, 1987.
81. Brunette MG, Vary J, Carriere S: Hyposthenuria in hypercalcemia. A possible role of intrarenal blood-flow (IRBF) redistribution, *Pflugers Arch* 350:9-23, 1974.
82. Bruyette DS, Feldman EC: Primary hypoparathyroidism in the dog. Report of 15 cases and review of 13 previously reported cases, *J Vet Intern Med* 2:7-14, 1988.
83. Burritt MF, Pierides AM, Offord KP: Comparative studies of total and ionized serum calcium values in normal subjects and patients with renal disorders, *Mayo Clin Proc* 55:606-613, 1980.
84. Burtis WJ, Brady TG, Orloff JJ, et al: Immunochemical characterization of circulating parathyroid hormone-related protein in patients with humoral hypercalcemia of cancer, *N Engl J Med* 322:1106-1112, 1990.
85. Burtis WJ, Dann P, Gaich GA, et al: A high abundance midregion species of parathyroid hormone-related protein: immunological and chromatographic characterization in plasma, *J Clin Endocrinol Metab* 78:317-322, 1994.
86. Burtis WJ: Parathyroid hormone-related protein: structure, function, and measurement, *Clin Chem* 38:2171-2183, 1992.
87. Bush WW, Kimmel SE, Wosar MA, et al: Secondary hypoparathyroidism attributed to hypomagnesemia in a dog with protein-losing enteropathy, *J Am Vet Med Assoc* 219:1732-1734, 1708, 2001.
88. Cairns CB, Niemann JT, Pelikan PC, et al: Ionized hypocalcemia during prolonged cardiac arrest and closed-chest CPR in a canine model, *Ann Emerg Med* 20:1178-1182, 1991.
89. Caldin M, Tommaso F, Lubas G, et al: Incidence of persistent hypercalcemia in dogs and its diagnostic approach. In *European Society of Veterinary Internal Medicine Congress,* Dublin, Ireland, 2001.
90. Calia CM, Hohenhaus AE, Fox PR, et al: Acute tumor lysis syndrome in a cat with lymphoma, *J Vet Intern Med* 10:409-411, 1996.
91. Campbell A: Calcipotriol poisoning in dogs, *Vet Rec* 141:27-28, 1997.
92. Campese VM: Calcium, parathyroid hormone, and blood pressure, *Am J Hypertens* 2:34S-44S, 1989.

93. Camus C, Charasse C, Jouannic-Montier I, et al: Calcium free hemodialysis: experience in the treatment of 33 patients with severe hypercalcemia, *Intensive Care Med* 22:116-121, 1996.
94. Canaff L, Hendy GN: Human calcium-sensing receptor gene. Vitamin D response elements in promoters P1 and P2 confer transcriptional responsiveness to 1,25-dihydroxyvitamin D, *J Biol Chem* 277:30337-30350, 2002.
95. Canalejo A, Almaden Y, Torregrosa V, et al: The in vitro effect of calcitriol on parathyroid cell proliferation and apoptosis, *J Am Soc Nephrol* 11:1865-1872, 2000.
96. Canalejo A, Canadillas S, Ballesteros E, et al: Importance of arachidonic acid as a mediator of parathyroid gland response, *Kidney Int Suppl* June:S10-13, 2003.
97. Canalis E, Hock JM, Raisz LG: Anabolic and catabolic effects of parathyroid hormone on bone and interactions with growth factors. In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* New York, 1994, Raven Press, pp. 65-82.
98. Capen CC, Martin SL: Calcium metabolism and disorders of parathyroid glands, *Vet Clin North Am* 7:513-548, 1977.
99. Capen CC, Rosol TJ: Hormonal control of mineral metabolism. In Bojrab MJ, editors: *Disease mechanisms in small animal surgery,* Philadelphia, 1993, Lea & Febiger, pp. 841-857.
100. Care AD: The placental transfer of calcium, *J Dev Physiol* 15:253-257, 1991.
101. Carlstedt F, Lind L, Rastad J, et al: Parathyroid hormone and ionized calcium levels are related to the severity of illness and survival in critically ill patients, *Eur J Clin Invest* 28:898-903, 1998.
102. Carothers M, Chew DJ, Nagode LA: 25-OH-cholecalciferol intoxication in dogs. In *Proceedings Am Coll Vet Intern Med Forum*, 1994.
103. Chang W, Shoback D: Extracellular Ca2+-sensing receptors—an overview, *Cell Calcium* 35:183-196, 2004.
104. Chen RA, Goodman WG: Role of the calcium-sensing receptor in parathyroid gland physiology, *Am J Physiol Renal Physiol* 286:F1005-1011, 2004.
105. Cheteri MB, Stanford JL, Friedrichsen DM, et al: Vitamin D receptor gene polymorphisms and prostate cancer risk, *Prostate* 59:409-418, 2004.
106. Chew DJ, Capen CC: Hypercalcemic nephropathy and associated disorders. In Kirk RW, editor: *Current veterinary therapy VII,* Philadelphia, 1980, WB Saunders, pp. 1067-1072.
107. Reference deleted in pages.
108. Chew DJ, Carothers M, Nagode LA, et al: 25-OH-cholecalciferol intoxication in 12 dogs (14 episodes). In *Proceedings of the 39th Annual Congress of World Small Animal Veterinary Association*, Berlin, Germany, 1993.
109. Chew DJ, Carothers M: Hypercalcemia, *Vet Clin North Am Small Anim Pract* 19:265-87, 1989.
110. Chew DJ, Leonard M, Muir W 3rd: Effect of sodium bicarbonate infusions on ionized calcium and total calcium concentrations in serum of clinically normal cats, *Am J Vet Res* 50:145-150, 1989.
111. Chew DJ, Meuten DJ: Disorders of calcium and phosphorus metabolism, *Vet Clin North Am Small Anim Pract* 12:411-438, 1982.
112. Chew DJ, Meuten DJ: Primary hyperparathyroidism. In Kirk RW, editor: *Current veterinary therapy VIII,* Philadelphia, 1983, WB Saunders, pp. 880-884.
113. Chew DJ, Nagode L, Rosol TJ, et al: Utility of diagnostic assays in the evaluation of hypercalcemia and hypocalcemia: parathyroid hormone, vitamin D metabolites, parathyroid hormone-related protein, and ionized calcium. In Bonagura JD, editor: *Kirk's current veterinary therapy XII: small animal practice,* Philadelphia, 1995, WB Saunders, pp. 378-383.
114. Chew DJ, Nagode LA: Renal secondary hyperparathyroidism. In *Proc Soc Comp Endocrinol*, 1990.
115. Chew DJ, Schaer M, Liu S-K, et al: Pseudohyperparathyroidism in a cat, *J Natl Cancer Inst* 11:46-52, 1975.
116. Ching SV, Fettman MJ, Hamar DW, et al: The effect of chronic dietary acidification using ammonium chloride on acid-base and mineral metabolism in the adult cat, *J Nutr* 119:902-915, 1989.
117. Ching SV, Norrdin RW: Histomorphometric comparison of measurements of trabecular bone remodeling in iliac crest biopsy sites and lumbar vertebrae in cats, *Am J Vet Res* 51:447-450, 1990.
118. Chisholm MA, Mulloy AL, Taylor AT: Acute management of cancer-related hypercalcemia, *Ann Pharmacother* 30:507-513, 1996.
119. Chorev M, Alexander JM, Rosenblatt M: Interactions of parathyroid hormone and parathyroid hormone-related protein with their receptors. In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* San Diego, 2001, Academic Press, pp. 53-92.
120. Christakos S, Beck JD, Hyliner SJ: Calbindin-D 28K. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 209-221.
121. Coburn JW, Maung HM: Use of active vitamin D sterols in patients with chronic kidney disease, stages 3 and 4, *Kidney Int Suppl* June:S49-53, 2003.
122. Coburn JW: An update on vitamin D as related to nephrology practice: 2003, *Kidney Int Suppl* November: S125-130, 2003.
123. Cook SD, Skinner HB, Haddad RJ: A quantitative histologic study of osteoporosis produced by nutritional secondary hyperparathyroidism in dogs, *Clin Orthop Relat Res* May:105-120, 1983.
124. Cooke NE, Haddad JG: Vitamin D binding protein. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 87-101.
125. Coukell AJ, Markham A: Pamidronate. A review of its use in the management of osteolytic bone metastases, tumour-induced hypercalcaemia and Paget's disease of bone, *Drugs Aging* 12:149-168, 1998.
126. D'Amour P, Brossard JH, Rousseau L, et al: Amino-terminal form of parathyroid hormone (PTH) with immunologic similarities to hPTH(1-84) is overproduced in primary and secondary hyperparathyroidism, *Clin Chem* 49:2037-2044, 2003.
127. Davainis GM, Chew DJ, Nagode LA, et al: Calcium regulation in the cat with chronic renal failure. In *European Society of Veterinary Internal Medicine/European Society of Veterinary Endocrinology Annual Meeting,* Dublin, Ireland, 2001.
128. De Boer IH, Gorodetskaya I, Young B, et al: The severity of secondary hyperparathyroidism in chronic renal insufficiency is GFR-dependent, race-dependent, and associated with cardiovascular disease, *J Am Soc Nephrol* 13:2762-2769, 2002.
129. De Luisi A, Hofer AM: Evidence that Ca(2+) cycling by the plasma membrane Ca(2+)-ATPase increases the "excitability" of the extracellular Ca(2+)-sensing receptor, *J Cell Sci* 116:1527-1538, 2003.
130. DeLay J, Laing J: Nutritional osteodystophy in puppies fed a BARF diet, *AHL Newsletter* 6:23, 2002.

131. DeLuca HF, Krisinger J, Darwish H: The vitamin D system: 1990, *Kidney Int Suppl* 29:S2-8, 1990.
132. Demay M: Muscle: a nontraditional 1,25-dihydroxyvitamin D target tissue exhibiting classic hormone-dependent vitamin D receptor actions, *Endocrinology* 144:5135-5137, 2003.
133. den Hertog E, Goossens MM, van der Linde-Sipman JS, et al: Primary hyperparathyroidism in two cats, *Vet Q* 19:81-84, 1997.
134. Deniz A, Mischke R: [Ionized calcium and total calcium in the cat], *Berl Munch Tierarztl Wochenschr* 108:105-108, 1995.
135. DeVries SE, Feldman EC, Nelson RW, et al: Primary parathyroid gland hyperplasia in dogs: six cases (1982-1991), *J Am Vet Med Assoc* 202:1132-1136, 1993.
136. Dhupa N, Proulx J: Hypocalcemia and hypomagnesemia, *Vet Clin North Am Small Anim Pract* 28:587-608, 1998.
137. DiBartola SP, Chew DJ, Jacobs G: Quantitative urinalysis including 24-hour protein excretion in the dog, *J Am Anim Hosp Assoc* 16:537-546, 1980.
138. DiBartola SP, Rutgers HC, Zack PM, et al: Clinicopathologic findings associated with chronic renal disease in cats: 74 cases (1973-1984), *J Am Vet Med Assoc* 190:1196-1202, 1987.
139. Divieti P, John MR, Juppner H, et al: Human PTH-(7-84) inhibits bone resorption in vitro via actions independent of the type 1 PTH/PTHrP receptor, *Endocrinology* 143:171-176, 2002.
140. Dougherty SA, Center SA, Dzanis DA: Salmon calcitonin as adjunct treatment for vitamin D toxicosis in a dog, *J Am Vet Med Assoc* 196:1269-1272, 1990.
141. Dow SW, Legendre AM, Stiff M, et al: Hypercalcemia associated with blastomycosis in dogs, *J Am Vet Med Assoc* 188:706-709, 1986.
142. Drazner FH: Hypercalcemia in the dog and cat, *J Am Vet Med Assoc* 178:1252-1256, 1981.
143. Drobatz KJ, Casey KK: Eclampsia in dogs: 31 cases (1995-1998), *J Am Vet Med Assoc* 217:216-219, 2000.
144. Drobatz KJ, Hughes D: Concentration of ionized calcium in plasma from cats with urethral obstruction, *J Am Vet Med Assoc* 211:1392-1395, 1997.
145. Drop LJ: Ionized calcium, the heart, and hemodynamic function, *Anesth Analg* 64:432-451, 1985.
146. Drueke TB, McCarron DA: Paricalcitol as compared with calcitriol in patients undergoing hemodialysis, *N Engl J Med* 349:496-499, 2003.
147. Drueke TB: Cell biology of parathyroid gland hyperplasia in chronic renal failure, *J Am Soc Nephrol* 11:1141-1152, 2000.
148. Drueke TB: Parathyroid gland hyperplasia in uremia, *Kidney Int* 59:1182-1183, 2001.
149. Duncan AR: The use of subcutaneous pamidronate, *J Pain Symptom Manage* 26:592-593, 2003.
150. Dusso AS, Finch J, Brown A, et al: Extrarenal production of calcitriol in normal and uremic humans, *J Clin Endocrinol Metab* 72:157-164, 1991.
151. Dusso AS: Vitamin D receptor: mechanisms for vitamin D resistance in renal failure, *Kidney Int Suppl* June:S6-9, 2003.
152. Dust A, Norris AM, Valli VEO: Cutaneous lymphosarcoma with IgG monoclonal gammopathy, serum hyperviscosity and hypercalcemia in a cat, *Can Vet J* 23:235-239, 1982.
153. Dzanis DA, Kallfelz FA: Recent knowledge of vitamin D toxicity in dogs. In *Proceedings Am Coll Vet Intern Med Forum*, vol 6, 1988.
154. Earm JH, Christensen BM, Frokiaer J, et al: Decreased aquaporin-2 expression and apical plasma membrane delivery in kidney collecting ducts of polyuric hypercalcemic rats, *J Am Soc Nephrol* 9:2181-2193, 1998.
155. Eelen G, Verlinden L, van Camp M, et al: The effects of 1alpha,25-dihydroxyvitamin D3 on the expression of DNA replication genes, *J Bone Miner Res* 19:133-146, 2004.
156. Eiam-Ong S, Punsin P, Sitprija V, et al: Acute hypercalcemia-induced hypertension: the roles of calcium channel and alpha-1 adrenergic receptor, *J Med Assoc Thai* 87:410-418, 2004.
157. Elliott J, Dobson J, Dunn J, et al: Hypercalcemia in the dog: a study of 40 cases, *J Small Anim Pract* 32:564-571, 1991.
158. Engelman RW, Tyler RD, Good RA, et al: Hypercalcemia in cats with feline-leukemia-virus-associated leukemia-lymphoma, *Cancer* 56:777-781, 1985.
159. Estepa JC, Lopez I, Felsenfeld AJ, et al: Dynamics of secretion and metabolism of PTH during hypo- and hypercalcaemia in the dog as determined by the "intact" and "whole" PTH assays, *Nephrol Dial Transplant* 18:1101-1107, 2003.
160. Eubig PA: Acute renal failure in dogs subsequent to the ingestion of grapes or raisins: a retrospective evaluation of 43 dogs (1992-2002), *J Vet Intern Med* in press, 2005.
161. Falchetti A: Calcium agonists in hyperparathyroidism, *Expert Opin Investig Drugs* 13:229-244, 2004.
162. Fan TM, de Lorimier LP, Charney SC, et al: Evaluation of intravenous pamidronate administration in 33 cancer-bearing dogs with primary or secondary bone involvement, *J Vet Intern Med* 19:74-80, 2005.
163. Fan TM, Simpson KW, Trasti S, et al: Calcipotriol toxicity in a dog, *J Small Anim Pract* 39:581-586, 1998.
164. Farese G, Mager M, Blatt WF: A membrane ultrafiltration procedure for determining diffusible calcium in serum, *Clin Chem* 16:226-228, 1970.
165. Fascetti AJ, Hickman MA: Preparturient hypocalcemia in four cats, *J Am Vet Med Assoc* 215:1127-1129, 1999.
166. Favus MJ, Langman CB: Evidence for calcium-dependent control of 1,25-dihydroxyvitamin D3 production by rat kidney proximal tubules, *J Biol Chem* 261:11224-11229, 1986.
167. Favus MJ: Intestinal absorption of calcium, magnesium, and phosphorus. In Coe FL, Favus MJ: *Disorders of bone and mineral metabolism*, New York, 1992, Raven Press, pp. 57-81.
168. Feldman EC, Nelson RW: Hypercalcemia and primary hyperparathyroidism. In Feldman EC, editor: *Canine and feline endocrinology and reproduction*, Philadelphia, 2004, WB Saunders, pp. 660-715.
169. Feldman EC, Nelson RW: Hypocalcemia and primary hypoparathyroidism. In Feldman EC, editor: *Canine and feline endocrinology and reproduction*, Philadelphia, 2004, WB Saunders.
170. Feldman EC, Wisner ER, Nelson RW, et al: Comparison of results of hormonal analysis of samples obtained from selected venous sites versus cervical ultrasonography for localizing parathyroid masses in dogs, *J Am Vet Med Assoc* 211:54-56, 1997.
171. Fenton AJ, Kemp BE, Hammonds RG Jr, et al: A potent inhibitor of osteoclastic bone resorption within a highly conserved pentapeptide region of parathyroid hormone-related protein; PTHrP[107-111], *Endocrinology* 129:3424-3426, 1991.
172. Fenton AJ, Kemp BE, Kent GN, et al: A carboxyl-terminal peptide from the parathyroid hormone-related

protein inhibits bone resorption by osteoclasts, *Endocrinology* 129:1762-1768, 1991.
173. Finco DR, Rowland GN: Hypercalcemia secondary to chronic renal failure in the dog: a report of four cases, *J Am Vet Med Assoc* 173:990-994, 1978.
174. Finco DR: Interpretations of serum calcium concentration in the dog, *Comp Contin Educ* 5:778-787, 1983.
175. Fingeroth JM, Smeak DD: Intravenous methylene blue infusion for intraoperative identification of parathyroid gland tumors in dogs. Part III: Clinical trials and results in three dogs, *J Am Anim Hosp Assoc* 24:673-678, 1988.
176. Fitzpatrick LA, Brandi ML, Aurbach GD: Control of PTH secretion is mediated through calcium channels and is blocked by pertussis toxin treatment of parathyroid cells, *Biochem Biophys Res Commun* 138:960-965, 1986.
177. Flanders JA, Harvey HJ, Erb HN: Feline thyroidectomy. A comparison of postoperative hypocalcemia associated with three different surgical techniques, *Vet Surg* 16:362-366, 1987.
178. Flanders JA, Neth S, Erb HN, et al: Functional analysis of ectopic parathyroid activity in cats, *Am J Vet Res* 52:1336-1340, 1991.
179. Flanders JA, Scarlett JM, Blue JT, et al: Adjustment of total serum calcium concentration for binding to albumin and protein in cats: 291 cases (1986-1987), *J Am Vet Med Assoc* 194:1609-1611, 1989.
180. Flanders JA: Surgical therapy of the thyroid, *Vet Clin North Am Small Anim Pract* 24:607-621, 1994.
181. Fleisch H: Bisphosphonates. Pharmacology and use in the treatment of tumour-induced hypercalcaemic and metastatic bone disease, *Drugs* 42:919-944, 1991.
182. Fleisch H: Mechanisms of action of the bisphosphonates, *Medicina (B Aires)* 57(suppl 1):65-75, 1997.
183. Fooshee SK, Forrester SD: Hypercalcemia secondary to cholecalciferol rodenticide toxicosis in two dogs, *J Am Vet Med Assoc* 196:1265-1268, 1990.
184. Forbes S, Nelson RW, Guptill L: Primary hypoparathyroidism in a cat, *J Am Vet Med Assoc* 196:1285-1287, 1990.
185. Fournel-Fleury C, Ponce F, Felman P, et al: Canine T-cell lymphomas: a morphological, immunological, and clinical study of 46 new cases, *Vet Pathol* 39:92-109, 2002.
186. Fradkin JM, Braniecki AM, Craig TM, et al: Elevated parathyroid hormone-related protein and hypercalcemia in two dogs with schistosomiasis, *J Am Anim Hosp Assoc* 37:349-355, 2001.
187. Franceschini N, Joy MS, Kshirsagar A: Cinacalcet HCl: a calcimimetic agent for the management of primary and secondary hyperparathyroidism, *Expert Opin Investig Drugs* 12:1413-1421, 2003.
188. Fraser D, Jones G, Kooh SW, et al: *Calcium and phosphate metabolism*, Philadelphia, 1986, WB Saunders, pp. 1317-1372.
189. Fraser D, Jones G, Kooh SW: Calcium and phosphate metabolism. In Tietz NW, editor: *Fundamentals of clinical chemistry,* Philadelphia, 1987, WB Saunders, pp. 705-728.
190. Frolik CA, Black EC, Cain RL, et al: Anabolic and catabolic bone effects of human parathyroid hormone (1-34) are predicted by duration of hormone exposure, *Bone* 33:372-379, 2003.
191. Fukagawa M, Kitaoka M, Kurokawa K: Renal failure and hyperparathyroidism. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 1227-1239.
192. Gao P, Scheibel S, D'Amour P, et al: Development of a novel immunoradiometric assay exclusively for biologically active whole parathyroid hormone 1-84: implications for improvement of accurate assessment of parathyroid function, *J Bone Miner Res* 16:605-614, 2001.
193. Garcion E, Sindji L, Nataf S, et al: Treatment of experimental autoimmune encephalomyelitis in rat by 1,25-dihydroxyvitamin D3 leads to early effects within the central nervous system, *Acta Neuropathol (Berl)* 105:438-448, 2003.
194. Garlock SM, Matz ME, Shell LG: Vitamin D3 rodenticide toxicity in a dog, *J Am Anim Hosp Assoc* 27:356-360, 1991.
195. Garrett IR: Bone destruction in cancer, *Semin Oncol* 20(suppl 2):4-9, 1993.
196. Gaschen F, Gaschen L, Seiler G, et al: Lethal peracute rhabdomyolysis associated with stress and general anesthesia in three dystrophin-deficient cats, *Vet Pathol* 35:117-123, 1998.
197. Gascon-Barre M: The vitamin D 25-hydroxylase. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 41-56.
198. Gertz BJ, Holland SD, Kline WF, et al: Studies of the oral bioavailability of alendronate, *Clin Pharmacol Ther* 58:288-298, 1995.
199. Ghazarian JG: The renal mitochondrial hydroxylases of the vitamin D3 endocrine complex: how are they regulated at the molecular level? *J Bone Miner Res* 5:897-903, 1990.
200. Goldstein RE, Long C, Swift NC, et al: Percutaneous ethanol injection for treatment of unilateral hyperplastic thyroid nodules in cats, *J Am Vet Med Assoc* 218:1298-1302, 2001.
201. Goodman WG, Belin T, Gales B, et al: Calcium-regulated parathyroid hormone release in patients with mild or advanced secondary hyperparathyroidism, *Kidney Int* 48:1553-1558, 1995.
202. Goodman WG, Veldhuis JD, Belin TR, et al: Calcium-sensing by parathyroid glands in secondary hyperparathyroidism, *J Clin Endocrinol Metab* 83:2765-2772, 1998.
203. Goodman WG: New assays for parathyroid hormone (PTH) and the relevance of PTH fragments in renal failure, *Kidney Int Suppl* November:S120-124, 2003.
204. Goodman WG: Recent developments in the management of secondary hyperparathyroidism, *Kidney Int* 59:1187-1201, 2001.
205. Gosling P: Analytical reviews in clinical biochemistry: calcium measurement, *Ann Clin Biochem* 23:t146-156, 1986.
206. Goswami R, Brown EM, Kochupillai N, et al: Prevalence of calcium sensing receptor autoantibodies in patients with sporadic idiopathic hypoparathyroidism, *Eur J Endocrinol* 150:9-18, 2004.
207. Gouget B, Gourmelin Y, Blanchet F, et al: Ca2+ measurement with ion selective electrodes. The French coordinated evaluation of seven analyzers, for a better clinical relevance and acceptance, *Ann Biol Clin (Paris)* 46:419-434, 1988.
208. Grant WB, Garland CF: Evidence supporting the role of vitamin D in reducing the risk of cancer, *J Intern Med* 252:178-179; author reply 179-180, 2002.
209. Graves TK: Complications of treatment and concurrent illness associated with hyperthyroidism in cats. In Bonagura JD: *Kirk's current veterinary therapy XII: small animal practice,* Philadelphia, 1995, WB Saunders, pp. 369-372.
210. Green MD: Oral bisphosphonates and malignancy, *Med J Aust* 167:211-212, 1997.
211. Greenlee PG, Filippa DA, Quimby FW, et al: Lymphomas in dogs. A morphologic, immunologic, and clinical study, *Cancer* 66:480-490, 1990.

212. Grone A, McCauley LK, Capen CC, et al: Cloning and sequencing of the 3′-region of the canine parathyroid hormone-related protein gene and analysis of alternate mRNA splicing in two canine carcinomas, *Domest Anim Endocrinol* 22:169-177, 2002.
213. Grosenbaugh DA, Gadawski JE, Muir WW: Evaluation of a portable clinical analyzer in a veterinary hospital setting, *J Am Vet Med Assoc* 213:691-694, 1998.
214. Gunther R, Felice LJ, Nelson RK, et al: Toxicity of a vitamin D3 rodenticide to dogs, *J Am Vet Med Assoc* 193:211-214, 1988.
215. Gwaltney-Brant S, Holding JK, Donaldson CW, et al: Renal failure associated with ingestion of grapes or raisins in dogs, *J Am Vet Med Assoc* 218:1555-1556, 2001.
216. Habener JF, Rosenblatt M, Potts JT Jr: Parathyroid hormone: biochemical aspects of biosynthesis, secretion, action, and metabolism, *Physiol Rev* 64:985-1053, 1984.
217. Halabe A, Arie R, Mimran D, et al: Hypoparathyroidism—a long-term follow-up experience with 1 alpha-vitamin D3 therapy, *Clin Endocrinol (Oxf)* 40:303-307, 1994.
218. Hare WR, Dobbs CE, Slayman KA, et al: Calcipotriene poisoning in dogs, *Vet Med* 95:770-778, 2000.
219. Harrison HE, Harrison HC: Transfer of Ca45 across intestinal wall in vitro in relation to action of vitamin D and cortisol, *Am J Physiol* 199:265-271, 1960.
220. Haruna A, Kawai K, Takab T, et al: Dietary calcinosis in the cat, *J Anim Clin Res Round* 1:9-16, 1992.
221. Haussler MR, Haussler CA, Jurutka PW, et al: The nuclear vitamin D receptor: from clinical radioreceptor assay of the vitamin D hormone to genomics, proteomics and a novel ligand, *J Clin Ligand Assay* 25:221-228, 2002.
222. Hayes CE, Nashold FE, Spach KM, et al: The immunological functions of the vitamin D endocrine system, *Cell Mol Biol (Noisy-le-grand)* 49:277-300, 2003.
223. Hazewinkel HA, Tryfonidou MA: Vitamin D3 metabolism in dogs, *Mol Cell Endocrinol* 197:23-33, 2002.
224. Hazewinkel HA: Dietary influences on calcium homeostasis and the skeleton. In *Proceedings of the 1st Purina Int Nutr Symp*, 1991.
225. Henderson RA, Powers RD, Perry L: Development of hypoparathyroidism after excision of laryngeal rhabdomyosarcoma in a dog, *J Am Vet Med Assoc* 198:639-643, 1991.
226. Henry DA, Goodman WG, Nudelman RK, et al: Parenteral aluminum administration in the dog: I. Plasma kinetics, tissue levels, calcium metabolism, and parathyroid hormone, *Kidney Int* 25:362-369, 1984.
227. Henry H: The 25-hydroxyvitamin D 1-alpha-hydroxylase. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 57-68.
228. Hess RS, Saunders HM, Van Winkle TJ, et al: Concurrent disorders in dogs with diabetes mellitus: 221 cases (1993-1998), *J Am Vet Med Assoc* 217:1166-1173, 2000.
229. Hickford FH, Stokol T, vanGessel YA, et al: Monoclonal immunoglobulin G cryoglobulinemia and multiple myeloma in a domestic shorthair cat, *J Am Vet Med Assoc* 217:1029-1033, 1007-1008, 2000.
230. Hilbe M, Sydler T, Fischer L, et al: Metastatic calcification in a dog attributable to ingestion of a tacalcitol ointment, *Vet Pathol* 37:490-492, 2000.
231. Hill RC, Van Winkle TJ: Acute necrotizing pancreatitis and acute suppurative pancreatitis in the cat. A retrospective study of 40 cases (1976-1989), *J Vet Intern Med* 7:25-33, 1993.
232. Hirt RA, Kneissl S, Teinfalt M: Severe hypercalcemia in a dog with a retained fetus and endometritis, *J Am Vet Med Assoc* 216:1423-1425, 1412, 2000.
233. Hoare SR, Usdin TB: Molecular mechanisms of ligand recognition by parathyroid hormone 1 (PTH1) and PTH2 receptors, *Curr Pharm Des* 7:689-713, 2001.
234. Hodges RD, Legendre AM, Adams LG, et al: Itraconazole for the treatment of histoplasmosis in cats, *J Vet Intern Med* 8:409-413, 1994.
235. Hofer AM, Brown EM: Extracellular calcium sensing and signalling, *Nat Rev Mol Cell Biol* 4:530-538, 2003.
236. Holick MF: Noncalcemic actions of 1,25-dihydroxyvitamin D3 and clinical applications, *Bone* 17(suppl):107S-111S, 1995.
237. Holick MF: Photobiology of vitamin D. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 33-40.
238. Holick MF: Vitamin D: importance in the prevention of cancers, type 1 diabetes, heart disease, and osteoporosis, *Am J Clin Nutr* 79:362-371, 2004.
239. Holley DC, Evans JW: Determination of total and ultrafilterable calcium and magnesium in normal equine serum, *Am J Vet Res* 38:259-262, 1977.
240. Hollis BW, Kamerud JQ, Kurkowski A, et al: Quantification of circulating 1,25-dihydroxyvitamin D by radioimmunoassay with 125I-labeled tracer, *Clin Chem* 42:586-592, 1996.
241. Horn B, Irwin PJ: Transient hypoparathyroidism following successful treatment of hypercalcaemia of malignancy in a dog, *Aust Vet J* 78:690-692, 2000.
242. Horst RL, Reinhardt TA, Hollis BW: Improved methodology for the analysis of plasma vitamin D metabolites, *Kidney Int Suppl* 29:S28-35, 1990.
243. Horst RL, Reinhardt TA: Vitamin D metabolism. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 13-31.
244. Hostutler RA, Chew DJ, Jaeger JQ, et al: Uses and effectiveness of pamidronate disodium for treatment of dogs and cats with hypercalcemia, *J Vet Intern Med* 19:29-33, 2005.
245. How KL, Hazewinkel HA, Mol JA: Dietary vitamin D dependence of cat and dog due to inadequate cutaneous synthesis of vitamin D, *Gen Comp Endocrinol* 96:12-18, 1994.
246. Hristova EN, Cecco S, Niemela JE, et al: Analyzer-dependent differences in results for ionized calcium, ionized magnesium, sodium, and pH, *Clin Chem* 41: 1649-1653, 1995.
247. Hsu CH, Patel SR: Altered vitamin D metabolism and receptor interaction with the target genes in renal failure: calcitriol receptor interaction with its target gene in renal failure, *Curr Opin Nephrol Hypertens* 4:302-306, 1995.
248. Hulter HN, Halloran BP, Toto RD, et al: Long-term control of plasma calcitriol concentration in dogs and humans. Dominant role of plasma calcium concentration in experimental hyperparathyroidism, *J Clin Invest* 76:695-702, 1985.
249. Hung SH, Tsai WY, Tsao PN, et al: Oral clodronate therapy for hypercalcemia related to extensive subcutaneous fat necrosis in a newborn, *J Formos Med Assoc* 102:801-804, 2003.
250. Hutson CA, Willauer CC, Walder EJ, et al: Treatment of mandibular squamous cell carcinoma in cats by use of mandibulectomy and radiotherapy: seven cases (1987-1989), *J Am Vet Med Assoc* 201:777-781, 1992.
251. Ihle SL, Nelson RW, Cook JR Jr: Seizures as a manifestation of primary hyperparathyroidism in a dog, *J Am Vet Med Assoc* 192:71-72, 1988.

252. Imaizumi T, Tsuruta M, Kitagaki T, et al: [Single dose toxicity studies of calcipotriol (MC903) in rats and dogs], *J Toxicol Sci* 21(suppl 2):277-285, 1996.
253. Imaizumi T, Tsuruta M, Koike Y, et al: [A 26-week repeated percutaneous dose toxicity study of calcipotriol (MC903) in dogs], *J Toxicol Sci* 21(suppl 2):365-387, 1996.
254. Imaizumi T, Tsuruta M, Koike Y, et al: A 4-week repeated percutaneous dose toxicity study of calcipotriol (MC903) followed by a 4-week recovery test in dogs, *J Toxicol Sci* 21(suppl 2):309-323, 1996.
255. Imamura H, Sato K, Shizume K, et al: Urinary excretion of parathyroid hormone-related protein fragments in patients with humoral hypercalcemia of malignancy and hypercalcemic tumor-bearing nude mice, *J Bone Miner Res* 6:77-84, 1991.
256. Irvine RF: Calcium transients: mobilization of intracellular Ca2+, *Br Med Bull* 42:369-374, 1986.
257. Izquierdo R, Bermes E Jr, Sandberg L, et al: Serum calcium metabolism in acute experimental pancreatitis, *Surgery* 98:1031-1037, 1985.
258. Jackson IT, Saleh J, van Heerden JA: Gigantic mammary hyperplasia in pregnancy associated with pseudohyperparathyroidism, *Plast Reconstr Surg* 84:806-810, 1989.
259. John MR, Goodman WG, Gao P, et al: A novel immunoradiometric assay detects full-length human PTH but not amino-terminally truncated fragments: implications for PTH measurements in renal failure, *J Clin Endocrinol Metab* 84:4287-4290, 1999.
260. Jorgensen LS, Center SA, Randolph JF, et al: Electrolyte abnormalities induced by hypertonic phosphate enemas in two cats, *J Am Vet Med Assoc* 187:1367-1368, 1985.
261. Jutkowitz LA, Rozanski EA, Moreau JA, et al: Massive transfusion in dogs: 15 cases (1997-2001), *J Am Vet Med Assoc* 220:1664-1669, 2002.
262. Kadar E, Rush JE, Wetmore L, et al: Electrolyte disturbances and cardiac arrhythmias in a dog following pamidronate, calcitonin, and furosemide administration for hypercalcemia of malignancy, *J Am Anim Hosp Assoc* 40:75-81, 2004.
263. Kallet AJ, Richter KP, Feldman EC, et al: Primary hyperparathyroidism in cats: seven cases (1984-1989), *J Am Vet Med Assoc* 199:1767-1771, 1991.
264. Kallfelz FA: Nutritional supplements in small animal practice: boon or bane? In *Proceedings of the 8th Am Coll Vet Intern Med Forum*, Washington, DC, 1990.
265. Karaplis AC, Luz A, Glowacki J, et al: Lethal skeletal dysplasia from targeted disruption of the parathyroid hormone-related peptide gene, *Genes Dev* 8:277-289, 1994.
266. Kasahara H, Tsuchiya M, Adachi R, et al: Development of a C-terminal-region-specific radioimmunoassay of parathyroid hormone-related protein, *Biomed Res* 13:155-161, 1992.
267. Kawaguchi K, Braga IS 3rd, Takahashi A, et al: Nutritional secondary hyperparathyroidism occurring in a strain of German shepherd puppies, *Jpn J Vet Res* 41:89-96, 1993.
268. Kawasaki T: Creatinine unreliable indicator of renal failure in ferrets, *J Small Anim Exotic Med* 1:28-29, 1991.
269. Khosla S, van Heerden JA, Gharib H, et al: Parathyroid hormone-related protein and hypercalcemia secondary to massive mammary hyperplasia, *N Engl J Med* 322:1157, 1990.
270. Kifor O, McElduff A, LeBoff MS, et al: Activating antibodies to the calcium-sensing receptor in two patients with autoimmune hypoparathyroidism, *J Clin Endocrinol Metab* 89:548-556, 2004.
271. Kimmel SE, Waddell LS, Michel KE: Hypomagnesemia and hypocalcemia associated with protein-losing enteropathy in Yorkshire terriers: five cases (1992-1998), *J Am Vet Med Assoc* 217:703-706, 2000.
272. Kimmel SE, Washabau RJ, Drobatz KJ: Incidence and prognostic value of low plasma ionized calcium concentration in cats with acute pancreatitis: 46 cases (1996-1998), *J Am Vet Med Assoc* 219:1105-1109, 2001.
273. Kirby R, Iverson W, Schaer M: Hypercalcemic nephropathy in a young dog resembling human milk alkali syndrome, *J Am Anim Hosp Assoc* 28:119-123, 1992.
274. Kirk GR, Breazile JE, Kenny AD: Pathogenesis of hypocalcemic tetany in the thyroparathyroidectomized dog, *Am J Vet Res* 35:407-408, 1974.
275. Kiupel M, Teske E, Bostock D: Prognostic factors for treated canine malignant lymphoma, *Vet Pathol* 36:292-300, 1999.
276. Klausner JS, Bell FW, Hayden DW, et al: Hypercalcemia in two cats with squamous cell carcinomas, *J Am Vet Med Assoc* 196:103-105, 1990.
277. Klausner JS, Fernandez FR, O'Leary TP, et al: Canine primary hyperparathyroidism and its association with urolithiasis, *Vet Clin North Am Small Anim Pract* 16:227-239, 1986.
278. Klein MK, Powers BE, Withrow SJ, et al: Treatment of thyroid carcinoma in dogs by surgical resection alone: 20 cases (1981-1989), *J Am Vet Med Assoc* 206:1007-1009, 1995.
279. Knecht TP, Behling CA, Burton DW, et al: The humoral hypercalcemia of benignancy. A newly appreciated syndrome, *Am J Clin Pathol* 105:487-492, 1996.
280. Kogika MM, Lustoza MD, Notomi MK, et al: Serum ionized calcium evaluation in healthy dogs and in dogs with chronic renal failure. In *Proceedings of the World Small Anim Vet Assoc*, 2002.
281. Koller H, Zitt E, Staudacher G, et al: Variable parathyroid hormone(1-84)/carboxylterminal PTH ratios detected by 4 novel parathyroid hormone assays, *Clin Nephrol* 61:337-343, 2004.
282. Koo WS, Jeon DS, Ahn SJ, et al: Calcium-free hemodialysis for the management of hypercalcemia, *Nephron* 72:424-428, 1996.
283. Korkor AB, Kuchibotla J, Arrieh M, et al: The effects of chronic prednisone administration on intestinal receptors for 1,25-dihydroxyvitamin D3 in the dog, *Endocrinology* 117:2267-2273, 1985.
284. Kornegay JN: Hypocalcemia in dogs, *Compend Contin Educ* 4:1785-1792, 1982.
285. Kovacs CS, Ho-Pao CL, Hunzelman JL, et al: Regulation of murine fetal-placental calcium metabolism by the calcium-sensing receptor, *J Clin Invest* 101:2812-2820, 1998.
286. Kremer R, Goltzman D: Assays for parathyroid hormone-related protein. In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* New York, 1994, Raven Press, pp. 321-340.
287. Krishnan AV, Feldman D: Regulation of vitamin D receptor abundance. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 179-200.
288. Krishnan AV, Shinghal R, Raghavachari N, et al: Analysis of vitamin D-regulated gene expression in LNCaP human prostate cancer cells using cDNA microarrays, *Prostate* 59:243-251, 2004.
289. Kronenberg HM, Bringhurst FR, Segre GV, et al: Parathyroid hormone biosynthesis and metabolism. In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* San Diego, 2001, Academic Press, pp. 17-30.

290. Kruger JM, Osborne CA, Nachreiner RF, et al: Hypercalcemia and renal failure. Etiology, pathophysiology, diagnosis, and treatment, *Vet Clin North Am Small Anim Pract* 26:1417-1445, 1996.
291. Kruger JM, Osborne CA, Polzin DJ: Treatment of hypercalcemia. In Kirk RW, editor: *Current veterinary therapy IX,* Philadelphia, 1986, WB Saunders, pp. 75-90.
292. Kuhlmann A, Haas CS, Gross ML, et al: 1,25-Dihydroxyvitamin D3 decreases podocyte loss and podocyte hypertrophy in the subtotally nephrectomized rat, *Am J Physiol Renal Physiol* 286:F526-533, 2004.
293. Kull PA, Hess RS, Craig LE, et al: Clinical, clinicopathologic, radiographic, and ultrasonographic characteristics of intestinal lymphangiectasia in dogs: 17 cases (1996-1998), *J Am Vet Med Assoc* 219:197-202, 2001.
294. Kumar R: Vitamin D and the kidney. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 275-292.
295. Kyles AE, Stone EA, Gookin J, et al: Diagnosis and surgical management of obstructive ureteral calculi in cats: 11 cases (1993-1996), *J Am Vet Med Assoc* 213:1150-1156, 1998.
296. Ladenson JH, Lewis JW, McDonald JM, et al: Relationship of free and total calcium in hypercalcemic conditions, *J Clin Endocrinol Metab* 48:393-397, 1979.
297. Langub MC, Monier-Faugere MC, Wang G, et al: Administration of PTH-(7-84) antagonizes the effects of PTH-(1-84) on bone in rats with moderate renal failure, *Endocrinology* 144:1135-1138, 2003.
298. Larsson L, Ohman S: Effect of silicone-separator tubes and storage time on ionized calcium in serum, *Clin Chem* 31:169-170, 1985.
299. Lee JA, Drobatz KJ: Characterization of the clinical characteristics, electrolytes, acid-base, and renal parameters in male cats with urethral obstruction, *J Vet Emerg Crit Care* 13:227-233, 2003.
300. Levi J, Massry SG, Coburn JW, et al: Hypocalcemia in magnesium-depleted dogs: evidence for reduced responsiveness to parathyroid hormone and relative failure of parathyroid gland function, *Metabolism* 23:323-335, 1974.
301. Leyland-Jones B: Treating cancer-related hypercalcemia with gallium nitrate, *J Support Oncol* 2:509-516; discussion 516-520, 2004.
302. Lifton SJ, King LG, Zerbe CA: Glucocorticoid deficient hypoadrenocorticism in dogs: 18 cases (1986-1995), *J Am Vet Med Assoc* 209:2076-2081, 1996.
303. Lim SK, Gardella TJ, Baba H, et al: The carboxy-terminus of parathyroid hormone is essential for hormone processing and secretion, *Endocrinology* 131:2325-2330, 1992.
304. Lin R, White JH: The pleiotropic actions of vitamin D, *Bioessays* 26:21-28, 2004.
305. Lincoln SD, Lane VM: Serum ionized calcium concentration in clinically normal dairy cattle, and changes associated with calcium abnormalities, *J Am Vet Med Assoc* 197:1471-1474, 1990.
306. Lind L, Bucht E, Ljunghall S: Pronounced elevation in circulating calcitonin in critical care patients is related to the severity of illness and survival, *Intensive Care Med* 21:63-66, 1995.
307. Lind L, Carlstedt F, Rastad J, et al: Hypocalcemia and parathyroid hormone secretion in critically ill patients, *Crit Care Med* 28:93-99, 2000.
308. Lindemans J, Hoefkens P, van Kessel AL, et al: Portable blood gas and electrolyte analyzer evaluated in a multiinstitutional study, *Clin Chem* 45:111-117, 1999.
309. Lins LE: Renal function in hypercalcemic dogs during hydropenia and during saline infusion, *Acta Physiol Scand* 106:177-186, 1979.
310. Long CD, Goldstein RE, Hornof WJ, et al: Percutaneous ultrasound-guided chemical parathyroid ablation for treatment of primary hyperparathyroidism in dogs, *J Am Vet Med Assoc* 215:217-221, 1999.
311. Looney AL, Ludders J, Erb HN, et al: Use of a handheld device for analysis of blood electrolyte concentrations and blood gas partial pressures in dogs and horses, *J Am Vet Med Assoc* 213:526-530, 1998.
312. Lyon ME, Bremner D, Laha T, et al: Specific heparin preparations interfere with the simultaneous measurement of ionized magnesium and ionized calcium, *Clin Biochem* 28:79-84, 1995.
313. Lyon ME, Guajardo M, Laha T, et al: Electrolyte balanced heparin may produce a bias in the measurement of ionized calcium concentration in specimens with abnormally low protein concentration, *Clin Chim Acta* 233:105-113, 1995.
314. Lyon ME, Guajardo M, Laha T, et al: Zinc heparin introduces a preanalytical error in the measurement of ionized calcium concentration, *Scand J Clin Lab Invest* 55:61-65, 1995.
315. Machado CE, Flombaum CD: Safety of pamidronate in patients with renal failure and hypercalcemia, *Clin Nephrol* 45:175-179, 1996.
316. MacIsaac RJ, Caple IW, Danks JA, et al: Ontogeny of parathyroid hormone-related protein in the ovine parathyroid gland, *Endocrinology* 129:757-764, 1991.
317. MacIsaac RJ, Heath JA, Rodda CP, et al: Role of the fetal parathyroid glands and parathyroid hormone-related protein in the regulation of placental transport of calcium, magnesium and inorganic phosphate, *Reprod Fertil Dev* 3:447-457, 1991.
318. MacKenzie CP: Poisoning in four dogs by a compound containing warfarin and calciferol, *J Small Anim Pract* 28:433-445, 1987.
319. Maestro B, Davila N, Carranza MC, et al: Identification of a vitamin D response element in the human insulin receptor gene promoter, *J Steroid Biochem Mol Biol* 84:223-230, 2003.
320. Mahgoub A, Hirsch PF, Munson PL: Calcium-lowering action of glucocorticoids in adrenalectomized-parathyroidectomized rats. Specificity and relative potency of natural and synthetic glucocorticoids, *Endocrine* 6:279-283, 1997.
321. Major P, Lortholary A, Hon J, et al: Zoledronic acid is superior to pamidronate in the treatment of hypercalcemia of malignancy: a pooled analysis of two randomized, controlled clinical trials, *J Clin Oncol* 19:558-567, 2001.
322. Major P: The use of zoledronic acid, a novel, highly potent bisphosphonate, for the treatment of hypercalcemia of malignancy, *Oncologist* 7:481-491, 2002.
323. Malberti F, Surian M, Cosci P: Improvement of secondary hyperparathyroidism and reduction of the set point of calcium after intravenous calcitriol, *Kidney Int Suppl* 41:S125-130, 1993.
324. Mangin M, Ikeda K, Broadus AE: Structure of the mouse gene encoding parathyroid hormone-related peptide, *Gene* 95:195-202, 1990.
325. Markowitz GS, Fine PL, Stack JI, et al: Toxic acute tubular necrosis following treatment with zoledronate (Zometa), *Kidney Int* 64:281-289, 2003.
326. Marquez GA, Klausner JS, Osborne CA: Calcium oxalate urolithiasis in a cat with a functional parathyroid adenocarcinoma, *J Am Vet Med Assoc* 206:817-819, 1995.

327. Martin LG: Hypercalcemia and hypermagnesemia, *Vet Clin North Am Small Anim Pract* 28:565-585, 1998.
328. Martin TJ, Grill V: Hypercalcemia in cancer, *J Steroid Biochem Mol Biol* 43:123-129, 1992.
329. Martin-Salvago M, Villar-Rodriguez JL, Palma-Alvarez A, et al: Decreased expression of calcium receptor in parathyroid tissue in patients with hyperparathyroidism secondary to chronic renal failure, *Endocr Pathol* 14:61-70, 2003.
330. Massry SG: Pathogenesis of uremic toxicity, Part 1. Parathryoid hormone as a uremic toxin. In Massry SG, Glassock RJ, editors: *Textbook of nephrology,* Baltimore, 1989, Williams & Wilkins, pp. 1126-1144.
331. Matus RE, Leifer CE, MacEwen EG, et al: Prognostic factors for multiple myeloma in the dog, *J Am Vet Med Assoc* 188:1288-1292, 1986.
332. Matwichuk CL, Taylor SM, Daniel GB, et al: Double-phase parathyroid scintigraphy in dogs using technetium-99M-sestamibi, *Vet Radiol Ultrasound* 41:461-469, 2000.
333. Matwichuk CL, Taylor SM, Wilkinson AA, et al: Use of technetium Tc 99m sestamibi for detection of a parathyroid adenoma in a dog with primary hyperparathyroidism, *J Am Vet Med Assoc* 209:1733-1736, 1996.
334. Mazzaferro S, Barberi S, Scarda A, et al: Ionised and total serum magnesium in renal transplant patients, *J Nephrol* 15:275-280, 2002.
335. McCauley LK, Rosol TJ, Stromberg PC, et al: Effects of interleukin-1 alpha and cyclosporin A in vivo and in vitro on bone and lymphoid tissues in mice, *Toxicol Pathol* 19:1-10, 1991.
336. McClain HM, Barsanti JA, Bartges JW: Hypercalcemia and calcium oxalate urolithiasis in cats: a report of five cases, *J Am Anim Hosp Assoc* 35:297-301, 1999.
337. McElwain MC, Modzelewski RA, Yu WD, et al: Vitamin D: an antiproliferative agent with potential for therapy of squamous cell carcinoma, *Am J Otolaryngol* 18:293-298, 1997.
338. Mealey KL, Willard MD, Nagode LA, et al: Hypercalcemia associated with granulomatous disease in a cat, *J Am Vet Med Assoc* 215:959-962, 946, 1999.
339. Meller Y, Kestenbaum RS, Yagil R, et al: The influence of age and sex on blood levels of calcium-regulating hormones in dogs, *Clin Orthop Relat Res* Jul-Aug:296-299, 1984.
340. Merryman JI, Rosol TJ, Brooks CL, et al: Separation of parathyroid hormone-like activity from transforming growth factor-alpha and -beta in the canine adenocarcinoma (CAC-8) model of humoral hypercalcemia of malignancy, *Endocrinology* 124:2456-2463, 1989.
341. Meuten DJ, Chew DJ, Capen CC, et al: Relationship of serum total calcium to albumin and total protein in dogs, *J Am Vet Med Assoc* 180:63-67, 1982.
342. Meuten DJ, Cooper BJ, Capen CC, et al: Hypercalcemia associated with an adenocarcinoma derived from the apocrine glands of the anal sac, *Vet Pathol* 18:454-471, 1981.
343. Meuten DJ, Kociba GJ, Capen CC, et al: Hypercalcemia in dogs with lymphosarcoma. Biochemical, ultrastructural, and histomorphometric investigations, *Lab Invest* 49:553-562, 1983.
344. Meuten DJ, Segre GV, Capen CC, et al: Hypercalcemia in dogs with adenocarcinoma derived from apocrine glands of the anal sac. Biochemical and histomorphometric investigations, *Lab Invest* 48:428-435, 1983.
345. Meuten DJ: Hypercalcemia, *Vet Clin North Am* 14:891-910, 1984.
346. Midkiff AM, Chew DJ, Randolph JF, et al: Idiopathic hypercalcemia in cats, *J Vet Intern Med* 14:619-626, 2000.
347. Miki H, Maercklein PB, Fitzpatrick LA: Effect of magnesium on parathyroid cells: evidence for two sensing receptors or two intracellular pathways? *Am J Physiol* 272:E1-6, 1997.
348. Miller D, Edmonds MW: Hypercalcemia due to hyperparathyroidism treated with a somatostatin analogue, *CMAJ* 145:227-228, 1991.
349. Milner RJ, Farese J, Henry CJ, et al: Bisphosphonates and cancer, *J Vet Intern Med* 18:597-604, 2004.
350. Mischke R, Hanies R, Lange K, et al: [The effect of the albumin concentration on the relation between the concentration of ionized calcium and total calcium in the blood of dogs], *Dtsch Tierarztl Wochenschr* 103:199-204, 1996.
351. Miwa N, Nitta K, Kimata N, et al: An evaluation of 1-84 PTH measurement in relation to bone alkaline phosphatase and bone Gla protein in hemodialysis patients, *Nephron Clin Pract* 94:c29-32, 2003.
352. Mol JA, Kwant MM, Arnold IC, et al: Elucidation of the sequence of canine (pro)-calcitonin. A molecular biological and protein chemical approach, *Regul Pept* 35:189-195, 1991.
353. Moore FM, Kudisch M, Richter K, et al: Hypercalcemia associated with rodenticide poisoning in three cats, *J Am Vet Med Assoc* 193:1099-1100, 1988.
354. Morita T, Awakura T, Shimada A, et al: Vitamin D toxicosis in cats: natural outbreak and experimental study, *J Vet Med Sci* 57:831-837, 1995.
355. Morris JG: Vitamin D synthesis by kittens, *Vet Clin Nutr* 3:88-92, 1996.
356. Morris SA, Bilezikian JP: Signal transduction in bone physiology: messenger systems for parathyroid hormone. In Bilezikian JP, Raisz LG, Rodan GA, editors: *Principles of bone biology,* New York, 1996, Academic Press, pp. 1203-1215.
357. Morrow CMK, Valli VE, Volmer PA, et al: Canine renal pathology associated with grape or raisin ingestion: 10 cases, *J Vet Diagn Invest* 17:223-231, 2005.
358. Mosdell KW, Visconti JA: Emerging indications for octreotide therapy, part 1, *Am J Hosp Pharm* 51:1184-1192, 1994.
359. Moseley JM, Kubota M, Diefenbach-Jagger H, et al: Parathyroid hormone-related protein purified from a human lung cancer cell line, *Proc Natl Acad Sci U S A* 84:5048-5052, 1987.
360. Mundy GR, Oyajobi B: Other local and ectopic hormone syndromes associated with hypercalcemia. In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* San Diego, 2001, Academic Press, pp. 691-706.
361. Murthy JN, Hicks JM, Soldin SJ: Evaluation of i-STAT portable clinical analyzer in a neonatal and pediatric intensive care unit, *Clin Biochem* 30:385-389, 1997.
362. Nachreiner RF, Refsal KR: The use of parathormone, ionized calcium and 25-hydroxyvitamin D assays to diagnose calcium disorders in dogs. In *Proc Am Coll Vet Intern Med Forum*, vol 8, 1990.
363. Nagode LA, Chew DJ, Podell M: Benefits of calcitriol therapy and serum phosphorus control in dogs and cats with chronic renal failure. Both are essential to prevent of suppress toxic hyperparathyroidism, *Vet Clin North Am Small Anim Pract* 26:1293-1330, 1996.
364. Nagode LA, Chew DJ, Steinmeyer CL: The use of low doses of calcitriol in the treatment of renal secondary hyperparathyroidism. In *Proc 15th Waltham Symposium (Endocrinology)*, Columbus, OH, 1992.
365. Nagode LA, Chew DJ: Nephrocalcinosis caused by hyperparathyroidism in progression of renal failure: treat-

ment with calcitriol, *Semin Vet Med Surg (Small Anim)* 7:202-220, 1992.

366. Nagode LA, Chew DJ: The use of calcitriol in treatment of renal disease of the dog and cat. In *Proc 1st Purina Int Nutr Symp*, 1991.
367. Nagode LA, Steinmeyer CL, Chew DJ, et al: Hyper- and normo-calcemic dogs with chronic renal failure: relations of serum PTH and calcitriol to PTG Ca++ set-point. In Norman AW, Schaefer K, Grigoleit HG, et al, editors: *Vitamin D. Molecular, cellular and clinical endocrinology,* Berlin, 1988, Walter de Gruyter, pp. 799-800.
368. Negri AL, Alvarez Quiroga M, Bravo M, et al: [Whole PTH and 1-84/84 PTH ratio for the non invasive determination of low bone turnover in renal osteodysthrophy], *Nefrologia* 23:327-332, 2003.
369. Nemeth EF, Heaton WH, Miller M, et al: Pharmacodynamics of the type II calcimimetic compound cinacalcet HCl, *J Pharmacol Exp Ther* 308:627-635, 2004.
370. Nemzek JA, Kruger JM, Walshaw R, et al: Acute onset of hypokalemia and muscular weakness in four hyperthyroid cats, *J Am Vet Med Assoc* 205:65-68, 1994.
371. Neuman NB: Acute pancreatic hemorrhage associated with iatrogenic hypercalcemia in a dog, *J Am Vet Med Assoc* 166:381-383, 1975.
372. Neves M, Gano L, Pereira N, et al: Synthesis, characterization and biodistribution of bisphosphonates Sm-153 complexes: correlation with molecular modeling interaction studies, *Nucl Med Biol* 29:329-338, 2002.
373. Neville-Webbe H, Coleman RE: The use of zoledronic acid in the management of metastatic bone disease and hypercalcaemia, *Palliat Med* 17:539-553, 2003.
374. Neville-Webbe HL, Holen I, Coleman RE: The antitumour activity of bisphosphonates, *Cancer Treat Rev* 28:305-319, 2002.
375. Nguyen-Yamamoto L, Rousseau L, Brossard JH, et al: Origin of parathyroid hormone (PTH) fragments detected by intact-PTH assays, *Eur J Endocrinol* 147:123-131, 2002.
376. Nguyen-Yamamoto L, Rousseau L, Brossard JH, et al: Synthetic carboxyl-terminal fragments of parathyroid hormone (PTH) decrease ionized calcium concentration in rats by acting on a receptor different from the PTH/PTH-related peptide receptor, *Endocrinology* 142:1386-1392, 2001.
377. Niemann JT, Cairns CB: Hyperkalemia and ionized hypocalcemia during cardiac arrest and resuscitation: possible culprits for postcountershock arrhythmias? *Ann Emerg Med* 34:1-7, 1999.
378. Nissenson RA: Receptors for parathyroid hormone and parathyroid hormone-related protein: signaling and regulation. In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* San Diego, 2001, Academic Press, pp. 93-104.
379. *Normal blood values for cats and normal blood values for dogs*, St. Louis, 1975, Ralston-Purina Co.
380. Norman AW: Rapid biological responses mediated by 1,25-dihydroxyvitamin D3: a case study of transcaltachia (rapid hormonal stimulation of intestinal calcium transport). In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 233-256.
381. Norrdin RW, Miller CW, LoPresti CA, et al: Observations on calcium metabolism, 47Ca absorption, and duodenal calcium-binding activity in chronic renal failure: studies in Beagles with radiation-induced nephropathy, *Am J Vet Res* 41:510-515, 1980.
382. Okada H, Merryman JI, Rosol TJ, et al: Effects of humoral hypercalcemia of malignancy and gallium nitrate on thyroid C cells in nude mice: immunohistochemical and ultrastructural investigations, *Vet Pathol* 31:349-357, 1994.
383. Omdahl JL, May B: The 25-hydroxyvitamin D 24-hydroxylase. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 69-86.
384. Ong SC, Shalhoub RJ, Gallagher P, et al: Effect of furosemide on experimental hypercalcemia in dogs, *Proc Soc Exp Biol Med* 145:227-233, 1974.
385. Orloff JJ, Reddy D, de Papp AE, et al: Parathyroid hormone-related protein as a prohormone: posttranslational processing and receptor interactions, *Endocr Rev* 15:40-60, 1994.
386. Osborne CA, Lulich JP, Thumchai R, et al: Feline urolithiasis. Etiology and pathophysiology, *Vet Clin North Am Small Anim Pract* 26:217-232, 1996.
387. Padgett SL, Tobias KM, Leathers CW, et al: Efficacy of parathyroid gland autotransplantation in maintaining serum calcium concentrations after bilateral thyroparathyroidectomy in cats, *J Am Anim Hosp Assoc* 34:219-224, 1998.
388. Page RL: Acute tumor lysis syndrome, *Semin Vet Med Surg (Small Anim)* 1:58-60, 1986.
389. Pallais JC, Kifor O, Chen YB, et al: Acquired hypocalciuric hypercalcemia due to autoantibodies against the calcium-sensing receptor, *N Engl J Med* 351:362-369, 2004.
390. Panciera DL: Diagnostic approach to disorders of calcium homeostasis. In August J, editor: *Consultations in feline internal medicine 2,* Philadelphia, 1994, WB Saunders.
391. Pandian MR, Morgan CH, Carlton E, et al: Modified immunoradiometric assay of parathyroid hormone-related protein: clinical application in the differential diagnosis of hypercalcemia, *Clin Chem* 38:282-288, 1992.
392. Panichi V, Migliori M, Taccola D, et al: Effects of 1,25(OH)2D3 in experimental mesangial proliferative nephritis in rats, *Kidney Int* 60:87-95, 2001.
393. Panichi V, Migliori M, Taccola D, et al: Effects of calcitriol on the immune system: new possibilities in the treatment of glomerulonephritis, *Clin Exp Pharmacol Physiol* 30:807-811, 2003.
394. Pannabecker TL, Chandler JS, Wasserman RH: Vitamin-D-dependent transcriptional regulation of the intestinal plasma membrane calcium pump, *Biochem Biophys Res Commun* 213:499-505, 1995.
395. Parfitt AM: Bone and plasma calcium homeostasis, *Bone* 8(suppl 1):S1-8, 1987.
396. Patel SR, Ke HQ, Vanholder R, et al: Inhibition of calcitriol receptor binding to vitamin D response elements by uremic toxins, *J Clin Invest* 96:50-59, 1995.
397. Penny D, Henderson SM, Brown PJ: Raisin poisoning in a dog, *Vet Rec* 152:308, 2003.
398. Perkovic V, Hewitson TD, Kelynack KJ, et al: Parathyroid hormone has a prosclerotic effect on vascular smooth muscle cells, *Kidney Blood Press Res* 26:27-33, 2003.
399. Perry CM, Figgitt DP: Zoledronic acid: a review of its use in patients with advanced cancer, *Drugs* 64:1197-1211, 2004.
400. Persons DA, Garst J, Vollmer R, et al: Tumor lysis syndrome and acute renal failure after treatment of non-small-cell lung carcinoma with combination irinotecan and cisplatin, *Am J Clin Oncol* 21:426-429, 1998.
401. Peterson EN, Kirby R, Sommer M, et al: Cholecalciferol rodenticide intoxication in a cat, *J Am Vet Med Assoc* 199:904-906, 1991.
402. Peterson ME, Feinman JM: Hypercalcemia associated with hypoadrenocorticism in sixteen dogs, *J Am Vet Med Assoc* 181:802-804, 1982.

403. Peterson ME, Greco DS, Orth DN: Primary hypoadrenocorticism in ten cats, *J Vet Intern Med* 3:55-58, 1989.
404. Peterson ME, James KM, Wallace M, et al: Idiopathic hypoparathyroidism in five cats, *J Vet Intern Med* 5:47-51, 1991.
405. Peterson ME, Kintzer PP, Kass PH: Pretreatment clinical and laboratory findings in dogs with hypoadrenocorticism: 225 cases (1979-1993), *J Am Vet Med Assoc* 208:85-91, 1996.
406. Peterson ME: Hypoparathyroidism. In Kirk RW, editor: *Current veterinary therapy IX: small animal practice,* Philadelphia, 1986, WB Saunders, pp. 1039-1045.
407. Peterson ME: Treatment of canine and feline hypoparathyroidism, *J Am Vet Med Assoc* 181:1434-1436, 1982.
408. Petrie G: Management of hypercalcemia using dichloromethylene bisphosphonate (clodronate). In *Proc Cong Eur Soc Vet Intern Med*, vol 6, 1996.
409. Petzinger E, Ziegler K: Ochratoxin A from a toxicological perspective, *J Vet Pharmacol Ther* 23:91-98, 2000.
410. Pezzilli R, Billi P, Barakat B, et al: Octreotide for the treatment of hypercalcemia related to B cell lymphoma, *Oncology* 54:517-518, 1997.
411. Philbrick WM, Wysolmerski JJ, Galbraith S, et al: Defining the roles of parathyroid hormone-related protein in normal physiology, *Physiol Rev* 76:127-173, 1996.
412. Phillips DE, Radlinsky MG, Fischer JR, et al: Cystic thyroid and parathyroid lesions in cats, *J Am Anim Hosp Assoc* 39:349-354, 2003.
413. Piek CJ, Teske E: [Tumor lysis syndrome in a dog], *Tijdschr Diergeneeskd* 121:64-66, 1996.
414. Pollak MR, Brown EM, Chou YH, et al: Mutations in the human Ca(2+)-sensing receptor gene cause familial hypocalciuric hypercalcemia and neonatal severe hyperparathyroidism, *Cell* 75:1297-1303, 1993.
415. Pollard RE, Long CD, Nelson RW, et al: Percutaneous ultrasonographically guided radiofrequency heat ablation for treatment of primary hyperparathyroidism in dogs, *J Am Vet Med Assoc* 218:1106-1110, 2001.
416. Powell GJ, Southby J, Danks JA, et al: Localization of parathyroid hormone-related protein in breast cancer metastases: increased incidence in bone compared with other sites, *Cancer Res* 51:3059-3061, 1991.
417. Pressler BM, Rotstein DS, Law JM, et al: Hypercalcemia and high parathyroid hormone-related protein concentration associated with malignant melanoma in a dog, *J Am Vet Med Assoc* 221:263-265, 240, 2002.
418. Procino G, Carmosino M, Tamma G, et al: Extracellular calcium antagonizes forskolin-induced aquaporin 2 trafficking in collecting duct cells, *Kidney Int* 66:2245-2255, 2004.
419. Ramirez JA, Goodman WG, Belin TR, et al: Calcitriol therapy and calcium-regulated PTH secretion in patients with secondary hyperparathyroidism, *Am J Physiol* 267:E961-967, 1994.
420. Rasmussen H, Barrett P, Smallwood J, et al: Calcium ion as intracellular messenger and cellular toxin, *Environ Health Perspect* 84:17-25, 1990.
421. Rasmussen H: The cycling of calcium as an intracellular messenger, *Sci Am* 261:66-73, 1989.
422. Reber PM, Heath H 3rd: Hypocalcemic emergencies, *Med Clin North Am* 79:93-106, 1995.
423. Refsal KR, Provencher-Bolliger AL, Graham PA, et al: Update on the diagnosis and treatment of disorders of calcium regulation, *Vet Clin North Am Small Anim Pract* 31:1043-1062, 2001.
424. Reichel H, Koeffler HP, Norman AW: The role of the vitamin D endocrine system in health and disease, *N Engl J Med* 320:980-991, 1989.
425. Renoe BW, McDonald JM, Ladenson JH: Influence of posture on free calcium and related variables, *Clin Chem* 25:1766-1769, 1979.
426. Reusch C: [Ultrasonography of the parathyroid glands in dogs—a review], *Schweiz Arch Tierheilkd* 143:55-62, 2001.
427. Riccardi D: The role of extracellular calcium in the regulation of intracellular calcium and cell function (II). Some answers and more questions, *Cell Calcium* 35:179-181, 2004.
428. Richard V, Lairmore MD, Green PL, et al: Humoral hypercalcemia of malignancy: severe combined immunodeficient/beige mouse model of adult T-cell lymphoma independent of human T-cell lymphotropic virus type-1 tax expression, *Am J Pathol* 158:2219-2228, 2001.
429. Rickels MR, Mandel SJ: Hypocalciuric hypercalcemia and autoantibodies against the calcium-sensing receptor, *N Engl J Med* 351:2237-2238; author reply 2237-2238, 2004.
430. Rijnberk A, Elsinghorst TA, Koeman JP, et al: Pseudohyperparathyroidism associated with perirectal adenocarcinomas in elderly female dogs, *Tijdschr Diergeneeskd* 103:1069-1075, 1978.
431. Rodan GA, Balena R: Bisphosphonates in the treatment of metabolic bone diseases, *Ann Med* 25:373-378, 1993.
432. Roemer-Becuwe C, Vigano A, Romano F, et al: Safety of subcutaneous clodronate and efficacy in hypercalcemia of malignancy: a novel route of administration, *J Pain Symptom Manage* 26:843-848, 2003.
433. Rohrer CR, Phillips LA, Ford SL, et al: Hypercalcemia in a dog: a challenging case, *J Am Anim Hosp Assoc* 36:20-25, 2000.
434. Rosenberg MP, Matus RE, Patnaik AK: Prognostic factors in dogs with lymphoma and associated hypercalcemia, *J Vet Intern Med* 5:268-271, 1991.
435. Rosol TJ, Capen CC: Calcium-regulating hormones and diseases of abnormal mineral (calcium, phosphorus, magnesium) metabolism. In Kaneko JJ, Harvey JW, Bruss ML, editors: *Clinical biochemistry of domestic animals,* San Diego, 1997, Academic Press, pp. 619-702.
436. Rosol TJ, Capen CC: Cancer-associated hypercalcemia. In Feldman BF, Zinkl JG, Jain NC, editors: *Schalm's veterinary hematology,* Philadelphia, 2000, Lippincott Williams & Wilkins, pp. 660-666.
437. Rosol TJ, Capen CC: Mechanisms of cancer-induced hypercalcemia, *Lab Invest* 67:680-702, 1992.
438. Rosol TJ, Capen CC: Pathogenesis of humoral hypercalcemia of malignancy, *Domest Anim Endocrinol* 5:1-21, 1988.
439. Rosol TJ, Capen CC: Pathophysiology of calcium, phosphorus, and magnesium metabolism in animals, *Vet Clin North Am Small Anim Pract* 26:1155-1184, 1996.
440. Rosol TJ, Capen CC: The effect of low calcium diet, mithramycin, and dichlorodimethylene bisphosphonate on humoral hypercalcemia of malignancy in nude mice transplanted with the canine adenocarcinoma tumor line (CAC-8), *J Bone Miner Res* 2:395-405, 1987.
441. Rosol TJ, Capen CC: Tumors of the parathyroid gland and circulating parathyroid hormone-related protein associated with persistent hypercalcemia, *Toxicol Pathol* 17:346-356, 1989.
442. Rosol TJ, Chew DJ, Capen CC, et al: Acute hypocalcemia associated with infarction of parathyroid gland

adenomas in two dogs, *J Am Vet Med Assoc* 192:212-214, 1988.
443. Rosol TJ, Chew DJ, Couto CG, et al: Effects of mithramycin on calcium metabolism and bone in dogs, *Vet Pathol* 29:223-229, 1992.
444. Rosol TJ, Chew DJ, Hammer AS, et al: Effect of mithramycin on hypercalcemia in dogs, *J Am Anim Hosp Assoc* 30:244-250, 1994.
445. Rosol TJ, McCauley LK, Steinmeyer CL, et al: Nucleotide sequence of canine preproparathyroid hormone. In Dacke C, Danks J, Caple I, et al, editors: *The comparative endocrinology of calcium regulation,* Bristol, UK, 1996, Journal of Endocrinology Ltd., pp. 201-203.
446. Rosol TJ, Nagode LA, Couto CG, et al: Parathyroid hormone (PTH)-related protein, PTH, and 1,25-dihydroxyvitamin D in dogs with cancer-associated hypercalcemia, *Endocrinology* 131:1157-1164, 1992.
447. Rosol TJ, Nagode LA, Robertson JT, et al: Humoral hypercalcemia of malignancy associated with ameloblastoma in a horse, *J Am Vet Med Assoc* 204:1930-1933, 1994.
448. Rosol TJ, Chew DJ, Nagode LA, et al: Pathophysiology of calcium metabolism, *Vet Clin Pathol* 24:49-63, 1995.
449. Rosol TJ, Steinmeyer CL, McCauley LK, et al: Sequences of the cDNAs encoding canine parathyroid hormone-related protein and parathyroid hormone, *Gene* 160:241-243, 1995.
450. Rosol TJ, Tannehill-Gregg SH, Corn S, et al: Animal models of bone metastasis, *Cancer Treat Res* 118:47-81, 2004.
451. Rosol TJ, Tannehill-Gregg SH, LeRoy BE, et al: Animal models of bone metastasis, *Cancer* 97(suppl):748-757, 2003.
452. Rosol TJ: Pathogenesis of bone metastases: role of tumor-related proteins, *J Bone Miner Res* 15:844-850, 2000.
453. Ross JT, Scavelli TD, Matthiesen DT, et al: Adenocarcinoma of the apocrine glands of the anal sac in dogs: a review of 32 cases, *J Am Anim Hosp Assoc* 27:349-355, 1991.
454. Roth SI, Capen CC: Ultrastructural and functional correlations of the parathyroid gland, *Int Rev Exp Pathol* 13:161-221, 1974.
455. Rudnicki M, Frolich A, Haaber A, et al: Actual ionized calcium (at actual pH) vs adjusted ionized calcium (at pH 7.4) in hemodialyzed patients, *Clin Chem* 38:1384, 1992.
456. Rumbeiha WK, Fitzgerald SD, Kruger JM, et al: Use of pamidronate disodium to reduce cholecalciferol-induced toxicosis in dogs, *Am J Vet Res* 61:9-13, 2000.
457. Rumbeiha WK, Kruger JM, Fitzgerald SF, et al: Use of pamidronate to reverse vitamin D3-induced toxicosis in dogs, *Am J Vet Res* 60:1092-1097, 1999.
458. Ruopp JL: Primary hypoparathyroidism in a cat complicated by suspect iatrogenic calcinosis cutis, *J Am Anim Hosp Assoc* 37:370-373, 2001.
459. Ruslander DA, Gebhard DH, Tompkins MB, et al: Immunophenotypic characterization of canine lymphoproliferative disorders. *In Vivo* 11:169-172, 1997.
460. Russo EA, Lees GE: Treatment of hypocalcemia. In Kirk RW, editor: *Current veterinary therapy IX: small animal practice,* Philadelphia, 1986, WB Saunders, pp. 91-94.
461. Ryzen E, Rude RK: Low intracellular magnesium in patients with acute pancreatitis and hypocalcemia, *West J Med* 152:145-148, 1990.
462. Salusky IB, Goodman WG: Parathyroid gland function in secondary hyperparathyroidism, *Pediatr Nephrol* 10:359-363, 1996.
463. Santamaria R, Almaden Y, Felsenfeld A, et al: Dynamics of PTH secretion in hemodialysis patients as determined by the intact and whole PTH assays, *Kidney Int* 64:1867-1873, 2003.
464. Santini SA, Carrozza C, Vulpio C, et al: Assessment of parathyroid function in clinical practice: which parathyroid hormone assay is better? *Clin Chem* 50:1247-1250, 2004.
465. Sato R, Yamagishi H, Naito Y, et al: Feline vitamin D toxicosis caused by commercially available cat food, *J Jpn Vet Med Assoc* 46:577-581, 1993.
466. Saunders Y, Ross JR, Broadley KE, et al: Systematic review of bisphosphonates for hypercalcaemia of malignancy, *Palliat Med* 18:418-431, 2004.
467. Savary KC, Price GS, Vaden SL: Hypercalcemia in cats: a retrospective study of 71 cases (1991-1997), *J Vet Intern Med* 14:184-189, 2000.
468. Schaer M, Cavanaugh P, Hause W, et al: Iatrogenic hyperphosphatemia, hypocalcemia, and hypernatremia in a cat, *J Am Anim Hosp Assoc* 13:39, 1977.
469. Schaer M, Ginn PE, Fox LE, et al: Severe calcinosis cutis associated with treatment of hypoparathyroidism in a dog, *J Am Anim Hosp Assoc* 37:364-369, 2001.
470. Schenck PA, Chew DJ, Brooks CL: Effects of storage on serum ionized calcium and pH from horses with normal and abnormal ionized calcium concentrations, *Vet Clin Pathol* 25:118-120, 1996.
471. Schenck PA, Chew DJ, Brooks CL: Effects of storage on serum ionized calcium and pH values in clinically normal dogs, *Am J Vet Res* 56:304-307, 1995.
472. Schenck PA, Chew DJ, Brooks CL: Fractionation of canine serum calcium, using a micropartition system, *Am J Vet Res* 57:268-271, 1996.
473. Schenck PA, Chew DJ: Determination of calcium fractionation in dogs with chronic renal failure, *Am J Vet Res* 64:1181-1184, 2003.
474. Schenck PA, Chew DJ: Diagnostic discordance of total calcium and adjusted total calcium in predicting ionized calcium concentration in cats with chronic renal failure and other diseases. In *Proceedings of the 10th Congress of the International Society of Animal Clinical Biochemistry*, Gainesville, FL, 2002.
475. Schenck PA, Chew DJ: Prediction of serum ionized calcium concentration by serum total calcium measurement in dogs, *Am J Vet Res* 66:1330-1336, 2005.
476. Schenck PA, Chew DJ, Refsal K, et al: Calcium metabolic hormones in feline idiopathic hypercalcemia, *J Vet Intern Med* 18:442, 2004.
477. Schenck PA: Serum ionized magnesium concentrations in dogs and cats with hypoparathyroidism. In *Proceedings of the Am Coll Vet Intern Med Meeting*, Baltimore, MD, 2005.
478. Schreiner CA, Nagode LA: Vitamin D-dependent rickets type 2 in a four-month-old cat, *J Am Vet Med Assoc* 222:337-339, 315-316, 2003.
479. Schwarz U, Amann K, Orth SR, et al: Effect of 1,25 (OH)2 vitamin D3 on glomerulosclerosis in subtotally nephrectomized rats. *Kidney Int* 53:1696-1705, 1998.
480. Sekine M, Takami H: Combination of calcitonin and pamidronate for emergency treatment of malignant hypercalcemia, *Oncol Rep* 5:197-199, 1998.
481. Sela-Brown A, Russell J, Koszewski NJ, et al: Calreticulin inhibits vitamin D's action on the PTH gene in vitro and may prevent vitamin D's effect in vivo in hypocalcemic rats, *Mol Endocrinol* 12:1193-1200, 1998.

482. Seymour JF, Gagel RF: Calcitriol: the major humoral mediator of hypercalcemia in Hodgkin's disease and non-Hodgkin's lymphomas, *Blood* 82:1383-1394, 1993.
483. Sharma OP: Vitamin D, calcium, and sarcoidosis, *Chest* 109:535-539, 1996.
484. Sheafor SE, Gamblin RM, Couto CG: Hypercalcemia in two cats with multiple myeloma, *J Am Anim Hosp Assoc* 32:503-508, 1996.
485. Sherding RG, Meuten DJ, Chew DJ, et al: Primary hypoparathyroidism in the dog, *J Am Vet Med Assoc* 176:439-444, 1980.
486. Sherwood LM, Cantley L, Russell J: Effects of calcium and 1,25-(OH)2D3 on the synthesis and secretion of parathyroid hormone. In Cohn DV, Martin TJ, Meunier PJ, editors: *Calcium regulation and bone metabolism: basic and clinical aspects,* Amsterdam, 1987, Elsevier Science Publishers, pp. 778-781.
487. Siegel N, Wongsurawat N, Armbrecht HJ: Parathyroid hormone stimulates dephosphorylation of the renoredoxin component of the 25-hydroxyvitamin D3-1 alpha-hydroxylase from rat renal cortex, *J Biol Chem* 261:16998-17003, 1986.
488. Sih TR, Morris JG, Hickman MA: Chronic ingestion of high concentrations of cholecalciferol in cats, *Am J Vet Res* 62:1500-1506, 2001.
489. Silver J, Kilav R, Naveh-Many T: Mechanisms of secondary hyperparathyroidism, *Am J Physiol Renal Physiol* 283:F367-F376, 2002.
490. Silver J, Kronenberg HM: Parathyroid hormone—molecular biology and regulation. In Bilezikian JP, Raisz LG, Rodan GA, editors: *Principles of bone biology,* San Diego, 1996, Academic Press, pp. 325-337.
491. Silver J, Naveh-Many T: Vitamin D and the parathyroid glands. In Feldman D, editor: *Vitamin D,* San Diego, 1997, Academic Press, pp. 353-367.
492. Silver J, Yalcindag C, Sela-Brown A, et al: Regulation of the parathyroid hormone gene by vitamin D, calcium and phosphate, *Kidney Int Suppl* 73:S2-7, 1999.
493. Slatopolsky E, Finch J, Brown A: New vitamin D analogs, *Kidney Int Suppl* June:S83-87, 2003.
494. Slatopolsky E, Lopez-Hilker S, Delmez J, et al: The parathyroid-calcitriol axis in health and chronic renal failure, *Kidney Int Suppl* 29:S41-47, 1990.
495. Smith SA, Freeman LC, Bagladi-Swanson M: Hypercalcemia due to iatrogenic secondary hypoadrenocorticism and diabetes mellitus in a cat, *J Am Anim Hosp Assoc* 38:41-44, 2002.
496. Smock SL, Vogt GA, Castleberry TA, et al: Molecular cloning and functional characterization of the canine parathyroid hormone/parathyroid hormone related peptide receptor (PTH1), *Mol Biol Rep* 28:235-243, 2001.
497. St. Arnaud R, Glorieux FH: Vitamin D and bone development. In Feldman D, editor: *Vitamin D,* San Diego, 1997, Academic Press, pp. 293-303.
498. Stern PH: Vitamin D and bone, *Kidney Int Suppl* 29:S17-21, 1990.
499. Stevens LA, Djurdjev O, Cardew S, et al: Calcium, phosphate, and parathyroid hormone levels in combination and as a function of dialysis duration predict mortality: evidence for the complexity of the association between mineral metabolism and outcomes, *J Am Soc Nephrol* 15:770-779, 2004.
500. Storms TN, Clyde VL, Munson L, et al: Blastomycosis in nondomestic felids, *J Zoo Wildl Med* 34:231-238, 2003.
501. Strewler GJ: Physiological actions of PTH and PTHrP: Skeletal actions. In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* San Diego, 2001, Academic Press, pp. 213-226.
502. Suda T, Takahashi N: Vitamin D and osteoclastogenesis. In Feldman D, editor: *Vitamin D,* San Diego, 1997, Academic Press, pp. 329-340.
503. Sueda MT, Stefanacci, JD: Ultrasound evaluation of the parathyroid glands in two hypercalcemic cats, *Vet Radiol Ultrasound* 41:448-451, 2000.
504. Suva LJ, Winslow GA, Wettenhall RE, et al: A parathyroid hormone-related protein implicated in malignant hypercalcemia: cloning and expression, *Science* 237:893-896, 1987.
505. Swarthout JT, D'Alonzo RC, Selvamurugan N, et al: Parathyroid hormone-dependent signaling pathways regulating genes in bone cells, *Gene* 282:1-17, 2002.
506. Szenci O, Brydl E, Bajcsy CA: Effect of storage on measurement of ionized calcium and acid-base variables in equine, bovine, ovine, and canine venous blood, *J Am Vet Med Assoc* 199:1167-1169, 1991.
507. Takahashi HE, Tanizawa T, Hori M, et al: Effect of intermittent administration of human parathyroid hormone (1-34) on experimental osteopenia of rats induced by ovariectomy, *Cell Mater* 113-117, 1991.
508. Tannehill-Gregg S, Kergosien E, Rosol TJ: Feline head and neck squamous cell carcinoma cell line: characterization, production of parathyroid hormone-related protein, and regulation by transforming growth factor-beta, *In Vitro Cell Dev Biol Anim* 37:676-683, 2001.
509. Teare JA, Krook L, Kallfelz FA, et al: Ascorbic acid deficiency and hypertrophic osteodystrophy in the dog: a rebuttal, *Cornell Vet* 69:384-401, 1979.
510. Terry AH, Orrock J, Meikle AW: Comparison of two third-generation parathyroid hormone assays, *Clin Chem* 49:336-337, 2003.
511. Teske E, van Heerde P, Rutteman GR, et al: Prognostic factors for treatment of malignant lymphoma in dogs, *J Am Vet Med Assoc* 205:1722-1728, 1994.
512. Tfelt-Hansen J, Chattopadhyay N, Yano S, et al: Calcium-sensing receptor induces proliferation through p38 mitogen-activated protein kinase and phosphatidylinositol 3-kinase but not extracellularly regulated kinase in a model of humoral hypercalcemia of malignancy, *Endocrinology* 145:1211-1217, 2004.
513. Thakker RV: Diseases associated with the extracellular calcium-sensing receptor, *Cell Calcium* 35:275-282, 2004.
514. Thiede MA, Daifotis AG, Weir EC, et al: Intrauterine occupancy controls expression of the parathyroid hormone-related peptide gene in preterm rat myometrium, *Proc Natl Acad Sci U S A* 87:6969-6973, 1990.
515. Thode J, Juul-Jorgensen B, Bhatia HM, et al: Comparison of serum total calcium, albumin-corrected total calcium, and ionized calcium in 1213 patients with suspected calcium disorders, *Scand J Clin Lab Invest* 49:217-223, 1989.
516. Thomasset M: Calbindin-D 9K. In Feldman D, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 223-232.
517. Thompson KG, Jones LP, Smylie WA, et al: Primary hyperparathyroidism in German shepherd dogs: a disorder of probable genetic origin, *Vet Pathol* 21:370-376, 1984.
518. Thrall MA, Grauer GF, Mero KN: Clinicopathologic findings in dogs and cats with ethylene glycol intoxication, *J Am Vet Med Assoc* 184:37-41, 1984.
519. Thurlimann B, Waldburger R, Senn HJ, et al: Plicamycin and pamidronate in symptomatic tumor-related hypercalcemia: a prospective randomized crossover trial, *Ann Oncol* 3:619-623, 1992.

520. Toffaletti J: Ionized calcium measurement: analytical and clinical aspects, *Lab Management* July:31-35, 1983.
521. Tohme JF, Bilezikian JP: Hypocalcemic emergencies, *Endocrinol Metab Clin North Am* 22:363-375, 1993.
522. Tomsa K, Glaus T, Hauser B, et al: Nutritional secondary hyperparathyroidism in six cats, *J Small Anim Pract* 40:533-539, 1999.
523. Tomsa K, Steffen F, Glaus T: [Life threatening metabolic disorders after application of a sodium phosphate containing enema in the dog and cat.], *Schweiz Arch Tierheilkd* 143:257-261, 2001.
524. Toribio RE, Kohn CW, Chew DJ, et al: Comparison of serum parathyroid hormone and ionized calcium and magnesium concentrations and fractional urinary clearance of calcium and phosphorus in healthy horses and horses with enterocolitis, *Am J Vet Res* 62:938-947, 2001.
525. Torley D, Drummond A, Bilsland DJ: Calcipotriol toxicity in dogs, *Br J Dermatol* 147:1270, 2002.
526. Torrance AG, Nachreiner R: Human-parathormone assay for use in dogs: validation, sample handling studies, and parathyroid function testing, *Am J Vet Res* 50:1123-1127, 1989.
527. Torrance AG, Nachreiner R: Intact parathyroid hormone assay and total calcium concentration in the diagnosis of disorders of calcium metabolism in dogs, *J Vet Intern Med* 3:86-89, 1989.
528. Tras B, Maden M, Bas AL, et al: Investigation of biochemical and haematological side-effects of enrofloxacin in dogs, *J Vet Med A Physiol Pathol Clin Med* 48:59-63, 2001.
529. Troy GC, Forrester D, Cockburn C, et al: Heterobilharzia americana infection and hypercalcemia in a dog: a case report, *J Am Anim Hosp Assoc* 23:35-40, 1987.
530. Tucci J, Hammond V, Senior PV, et al: The role of fetal parathyroid hormone-related protein in transplacental calcium transport, *J Mol Endocrinol* 17:159-164, 1996.
531. Tuma SN, Mallette LE: Hypercalcemia after nephrectomy in the dog: role of the kidneys and parathyroid glands, *J Lab Clin Med* 102:213-219, 1983.
532. Tweedy CR, Rees GM: Octreotide acetate in the treatment of hypercalcemia accompanying small cell carcinoma, *South Med J* 85:561, 1992.
533. Tyrrell CJ, Collinson M, Madsen EL, et al: Intravenous pamidronate: infusion rate and safety, *Ann Oncol* 5(suppl 7):S27-29, 1994.
534. Uehlinger P, Glaus T, Hauser B, et al: [Differential diagnosis of hypercalcemia—a retrospective study of 46 dogs], *Schweiz Arch Tierheilkd* 140:188-197, 1998.
535. Unterer S, Lutz H, Gerber B, et al: Evaluation of an electrolyte analyzer for measurement of ionized calcium and magnesium concentrations in blood, plasma, and serum of dogs, *Am J Vet Res* 65:183-187, 2004.
536. Urena P, Frazao JM: Calcimimetic agents: review and perspectives, *Kidney Int Suppl* June:S91-96, 2003.
537. Vaden SL, Levine J, Breitschwerdt EB: A retrospective case-control of acute renal failure in 99 dogs, *J Vet Intern Med* 11:58-64, 1997.
538. Vail DM, Kisseberth WC, Obradovich JE, et al: Assessment of potential doubling time (Tpot), argyrophilic nucleolar organizer regions (AgNOR), and proliferating cell nuclear antigen (PCNA) as predictors of therapy response in canine non-Hodgkin's lymphoma, *Exp Hematol* 24:807-815, 1996.
539. Vail DM, Withrow SJ, Schwarz PD, et al: Perianal adenocarcinoma in the canine male: a retrospective study of 41 cases, *J Am Anim Hosp Assoc* 26:329-334, 1990.
540. Walker MC, Schaer M: Percutaneous ethanol treatment of hyperthyroidism in a cat, *Feline Practice* 28:10-12, 1998.
541. Walker P, Watanabe S, Lawlor P, et al: Subcutaneous clodronate, *Lancet* 348:345-346, 1996.
542. Walker P, Watanabe S, Lawlor P, et al: Subcutaneous clodronate: a study evaluating efficacy in hypercalcemia of malignancy and local toxicity, *Ann Oncol* 8:915-916, 1997.
543. Walser M, Robinson BHB, Duckett JW Jr: The hypercalcemia of adrenal insufficiency, *J Clin Invest* 42:456-465, 1963.
544. Walters MR: Newly identified effects of the vitamin D endocrine system: update 1995. In Bikle DD, Negrovilar A, editors: *Hormonal regulation of bone mineral metabolism,* Bethesda, 1995, Endocrine Society, pp. 47-56.
545. Wang W, Li C, Kwon TH, et al: AQP3, p-AQP2, and AQP2 expression is reduced in polyuric rats with hypercalcemia: prevention by cAMP-PDE inhibitors, *Am J Physiol Renal Physiol* 283:F1313-1325, 2002.
546. Ward DT: Calcium receptor-mediated intracellular signalling, *Cell Calcium* 35:217-228, 2004.
547. Warrell RP Jr, Murphy WK, Schulman P, et al: A randomized double-blind study of gallium nitrate compared with etidronate for acute control of cancer-related hypercalcemia, *J Clin Oncol* 9:1467-1475, 1991.
548. Waser M, Mesaeli N, Spencer C, et al: Regulation of calreticulin gene expression by calcium, *J Cell Biol* 138:547-557, 1997.
549. Wasserman RH: Vitamin D and the intestinal absorption of calcium and phosphorus. In Feldman BF, editor: *Vitamin D,* New York, 1997, Academic Press, pp. 259-273.
550. Waters CB, Scott-Moncrieff JC: Hypocalcemia in cats, *Compend Contin Educ* 14:497-506, 1992.
551. Weaver ME, Morrissey J, McConkey C Jr, et al: WR-2721 inhibits parathyroid adenylate cyclase, *Am J Physiol* 252:E197-201, 1987.
552. Weir EC, Burtis WJ, Morris CA, et al: Isolation of 16,000-dalton parathyroid hormone-like proteins from two animal tumors causing humoral hypercalcemia of malignancy, *Endocrinology* 123:2744-2751, 1988.
553. Weir EC, Norrdin RW, Matus RE, et al: Humoral hypercalcemia of malignancy in canine lymphosarcoma, *Endocrinology* 122:602-608, 1988.
554. Weisbrode SE, Krakowka S: Canine distemper virus-associated hypocalcemia, *Am J Vet Res* 40:147-149, 1979.
555. Welches CD, Scavelli TD, Matthiesen DT, et al: Occurrence of problems after three techniques of bilateral thyroidectomy in cats, *Vet Surg* 18:392-396, 1989.
556. Weller RE, Theilen GH, Madewell BR: Chemotherapeutic responses in dogs with lymphosarcoma and hypercalcemia, *J Am Vet Med Assoc* 181:891-893, 1982.
557. Wellington K, Goa KL: Zoledronic acid: a review of its use in the management of bone metastases and hypercalcaemia of malignancy, *Drugs* 63:417-437, 2003.
558. Wells AL, Long CD, Hornof WJ, et al: Use of percutaneous ethanol injection for treatment of bilateral hyperplastic thyroid nodules in cats, *J Am Vet Med Assoc* 218:1293-1297, 2001.
559. Whitfield GK, Dang HT, Schluter SF, et al: Cloning of a functional vitamin D receptor from the lamprey (Petromyzon marinus), an ancient vertebrate lacking a calcified skeleton and teeth, *Endocrinology* 144:2704-2716, 2003.
560. Willard MD, Schall WD, McCaw DE, et al: Canine hypoadrenocorticism: report of 37 cases and review of 39 previously reported cases, *J Am Vet Med Assoc* 180:59-62, 1982.

561. Williams LE, Gliatto JM, Dodge RK, et al: Carcinoma of the apocrine glands of the anal sac in dogs: 113 cases (1985-1995), *J Am Vet Med Assoc* 223:825-831, 2003.
562. Winer KK, Yanovski JA, Cutler GB Jr: Synthetic human parathyroid hormone 1-34 vs calcitriol and calcium in the treatment of hypoparathyroidism, *JAMA* 276:631-636, 1996.
563. Winkelmayer WC, Levin R, Avorn J: The nephrologist's role in the management of calcium-phosphorus metabolism in patients with chronic kidney disease, *Kidney Int* 63:1836-1842, 2003.
564. Wisner ER, Nyland TG: Ultrasonography of the thyroid and parathyroid glands, *Vet Clin North Am Small Anim Pract* 28:973-991, 1998.
565. Wisner ER, Penninck D, Biller DS, et al: High-resolution parathyroid sonography, *Vet Radiol Ultrasound* 38:462-466, 1997.
566. Won DS, Park C, In YJ, et al: A case of nutritional secondary hyperparathyroidism in a Siberian tiger cub, *J Vet Med Sci* 66:551-553, 2004.
567. Woo J, Cannon DC: *Metabolic intermediates and inorganic ions,* ed 17, Philadelphia, 1984, WB Saunders, pp. 133-179.
568. Wooldridge JD, Gregory CR: Ionized and total serum magnesium concentrations in feline renal transplant recipients, *Vet Surg* 28:31-37, 1999.
569. Worth GK, Vasikaran SD, Retallack RW, et al: Major method-specific differences in the measurement of intact parathyroid hormone: studies in patients with and without chronic renal failure, *Ann Clin Biochem* 41:149-154, 2004.
570. Wright KN, Breitschwerdt EB, Feldman JM, et al: Diagnostic and therapeutic considerations in a hypercalcemic dog with multiple endocrine neoplasia, *J Am Anim Hosp Assoc* 31:156-162, 1995.
571. Wysolmerski JJ, Stewart AF, Martin JT: Physiological actions of PTH and PTHrP: epidermal, mammary, reproductive, and pancreatic tissues. In Bilezikian JP, Marcus R, Levine MA, editors: *The parathyroids,* San Diego, 2001, Academic Press, pp. 275-292.
572. Wysolmerski JJ: The evolutionary origins of maternal calcium and bone metabolism during lactation, *J Mammary Gland Biol Neoplasia* 7:267-276, 2002.
573. Yanagawa N, Lee DBN: Renal handling of calcium and phosphorus. In Coe FL, Favus MJ, editors: *Disorders of bone and mineral metabolism,* New York, 1992, Raven Press, pp. 3-40.
574. Yang KH, dePapp AE, Soifer NE, et al: Parathyroid hormone-related protein: evidence for isoform- and tissue-specific posttranslational processing, *Biochemistry* 33:7460-7469, 1994.
575. Yasuda T, Banville D, Rabbani SA, et al: Rat parathyroid hormone-like peptide: comparison with the human homologue and expression in malignant and normal tissue, *Mol Endocrinol* 3:518-525, 1989.
576. Yu J, Papavasiliou V, Rhim J, et al: Vitamin D analogs: new therapeutic agents for the treatment of squamous cancer and its associated hypercalcemia, *Anticancer Drugs* 6:101-108, 1995.
577. Yudd M, Llach F: Current medical management of secondary hyperparathyroidism, *Am J Med Sci* 320:100-106, 2000.
578. Zaloga GP, Chernow B: The multifactorial basis for hypocalcemia during sepsis. Studies of the parathyroid hormone-vitamin D axis, *Ann Intern Med* 107:36-41, 1987.
579. Zaloga GP, Malcolm D, Holaday J, et al: Verapamil reverses calcium cardiotoxicity, *Ann Emerg Med* 16:637-639, 1987.
580. Zaloga GP, Willey S, Tomasic P, et al: Free fatty acids alter calcium binding: a cause for misinterpretation of serum calcium values and hypocalcemia in critical illness, *J Clin Endocrinol Metab* 64:1010-1014, 1987.
581. Zawada ET Jr, Saelens DA, Lembke JM: Influence of calcium infusion on plasma atrial natriuretic peptide in conscious dogs: intervention with calcium antagonist, verapamil, *Miner Electrolyte Metab* 16:369-377, 1990.
582. Zelikovic I, Chesney RW: Vitamin D and mineral metabolism: the role of the kidney in health and disease, *World Rev Nutr Diet* 59:156-216, 1989.
583. Zitterman A: Vitamin D in preventive medicine: are we ignoring the evidence? *Br J Nutr* 89:552-572, 2003.
584. Zivin JR, Gooley T, Zager RA, et al: Hypocalcemia: a pervasive metabolic abnormality in the critically ill, *Am J Kidney Dis* 37:689-698, 2001.