CENTRAL AND PERIPHERAL NERVOUS SYSTEMS

**Anticonvulsants.** **Bromide.** A loading dose of 450 mg/kg should yield concentrations of 1 mg/ml (the minimum end of the therapeutic range); for each 0.5 mg/ml increase in blood concentrations desired (maximum end of the range is 3.5 mg/ml), an additional 225-250 mg/kg loading dose should be given. If this loading dose is split over 5 days, the maintenance dose should also be given (30 mg/kg/day for 1 mg/ml; 15 mg/kg/day for each 0.25 to 0.5 mg/ml increase above that). Patients should be monitored 1 to 3 days after the loading dose and again at one month; if the two samples do not match, the maintenance dose should be changed accordingly. Our lab will increase bromide concentrations well above the recommended range if necessary to control seizures as long as the animal is not groggy or otherwise is intolerant to the drug. If groggy, our choice is to decrease phenobarbital concentrations first. Phenobarbital can be completely eradicated in some animals; in contrast, some animals will be controlled only at concentrations of both bromide and phenobarbital at the maximum end of the therapeutic range. Bromide can be made by dividing a 1kg bottle of the salt into 4 equal 250 gm parts (store in zip lock back, protect from humidity). One package can be added to a 1 liter bottle of commercial spring water: draw a line at the 1 liter mark, remove about 0.5 liter, add the bromide, and enough water to make the bromide dissolve, and fill the remaining volume to the line with either water or corn syrup for flavoring. The final solution is 250 mg/ml. Because bromide is very safe, we will increase bromide concentrations above the therapeutic range as necessary to sufficiently control seizures until the animal becomes unacceptably groggy. Potassium bromide can be loaded following rectal administration over a 24 hour period (divide the loading dose into 4 administrations). IV administration is not recommended because of the risk of potassium overload. An ongoing study comparing phenobarbital and bromide in dogs as a first choice anti-epileptic thus far suggests no difference in efficacy, although bromide is harder to begin because of the risk of gastrointestinal upset. Bromide has been studied in cats (as the potassium salt) when used at the canine maintenance dose. Although concentrations are similar to those achieved in dogs, in a retrospective study of 17 cats, 38% of seizuring cats developed signs consistent with feline bronchial asthma. The time to onset varied from 3 to 24 months and did not seem to be related to dose. Treatment with glucocorticoids may be helpful. Combination anticonvulsant therapy is a powerful tool for control of refractory seizures (defined as unacceptable seizure activity despite anticonvulsant concentrations at the maximum end of the therapeutic range). Several options exist. **Gabapentin** (Neurontin®) is a renally eliminated human anticonvulsant whose actions appear to involve gamma amino butyric acid receptors. The drug may be useful, primarily as an add-on anticonvulsant in dogs refractory to Phenobarbital. It has a short elimination half-life and thus may requiring 8 hour dosing intervals. Efficacy as sole anticonvulsant is questionable even at high doses. Recommended doses range from 10 to 30 (or up to 60) mg/kg every 8 hours po. The drug has been used effectively in both the dog and cat. Like felbamate, the drug is expensive. **Zonisamide** (Zonegran®; 8 to 12 mg/kg divided twice to three times daily, po) is a sulfonamide anticonvulsant approved for use to treat epilepsy in humans. Zonisamide appears to inhibit neuronal voltage-dependent sodium and T-type calcium channels. Additionally, ZNS modulates the dopaminergic system and
accelerates the release of γ-amino butyric acid (GABA) from the hippocampus. Like phenytoin, ZNS is less likely to affect normal neuronal activity. A potential advantage of ZNS is free radical scavenging which protects against the destructive nature of radicals, especially in neuronal membranes. As with most sulfonamides, elimination includes both a hepatic (induced by Phenobarbital) and renal elimination. Its elimination half-life in humans is 10 to 40 mcg/ml. It has been used safely in combination with Phenobarbital for control of refractory seizures in dogs. As with any sulfonamide, at high concentrations (including those necessary to control seizures in some dogs), thyroid hormone synthesis will be impaired within several weeks of start of therapy. Function will return to normal but only when therapy is d/c. The protocol for thyroid hormone replacement, which is probably indicated once hypothyroidism has been documented, has not been well established.

Levetiracetam (Keppra®) is an anticonvulsant approved for use in humans. Its mechanism of action is not known. It is renally excreted (60%) and to a lesser degree, metabolized by the liver. However, it has a short half-life that is supportive of 8 hour dosing. It is being studied at NCSU as an anticonvulsant for use in dogs. However, it has been safely used in cats as well as an add-on anticonvulsant at 20 mg/kg every 8 hrs po. A test is available to detect the MDR-1 genetic deletion that leads to P-glycoprotein deficiencies in collies and related breeds. This deficiency has been linked to CNS toxicities caused by ivermectin and loperamide. Owners of potentially affected dogs should be tested not only to be aware of possible CNS drug toxicities but also to try to minimize breeding (http://www.vetmed.wsu.edu/announcements/ivermectin/ownerinfo.asp). The sample is a buccal mucosal swab and the cost is approximately $60.

Centrally-acting analgesics. Buprenorphine (0.005-0.03 mg/kg IV,IM,SQ, epidural [D]) is to be rescheduled to Class III because of changes in availability from international sources. As a thebaine derivative, its analgesic effects are at least 25 more times potent than morphine. As a partial mu agonist / antagonist, it appears to be a more effective analgesic compared to butorphanol which is a mu antagonist. Although its onset of action takes longer than morphine, its effects may last longer. Like morphine, buprenorphine induces dose-dependent respiratory depression; however, like butorphanol, a ceiling effect is reached. Although respiratory depression has not been a problem in human patients receiving the drug, it is noteworthy that these effects are not fully reversible with antagonists such as naloxone. A potential advantage of buprenorphine (like butorphanol) is its ability to reverse opioid-induced sedation while maintaining analgesia. It has been recommended in humans as the reversal agent of choice (ie, in lieu of naloxone) in patients receiving neuroleptanalgesics. It can be used in conjunction with a fentanyl patch, but is less preferred than a pure mu opioid agonist because it will block mu receptor actions of fentanyl. Tramadol (Ultram®) is a non-opioid non-NSAID analgesic drug with opioid type properties but without respiratory depression. An added advantage of the drug is impaired reuptake of serotonin and norepinephrine and thus an added analgesic effect. Both parent compounds and metabolites are active. Clinical studies have not been performed in veterinary medicine, but anecdotally, the drug appears to be safe in dogs. It has been used on an outpatient basis for control of pain (1-2 mg/kg q 8-12 hrs orally). The role of ketamine (1-2 mg/kg [IV for burns], 0.5-1.0 mg/kg IM ) for control of pain should be considered controversial. Ketamine increasingly is being cited for its analgesic effects, but it is not particularly effective as a sole agent. It’s efficacy appears to prevent activation of NMDA (n-methyl-D-aspartate) receptors. These receptors, located in the dorsal horn of the spinal cord, interact with opioid receptors and increasingly are recognized for their role in modulating
response to pain. For example, activation of these receptors has been cited as the cause for the “winding-up” or hyperalgesia associated with chronic (and particularly neuralgic?) pain. NMDA receptors also appear to be responsible for the development of tolerance to opioids. In humans, some analgesia is provided when used in combination with other analgesics, particularly perioperatively. However, side effects (CNS) limit its use as an analgesic. Ketamine is reasonably used in combination with other analgesics for control of pain. For example, in cats, it has been combined with domitor (0.025 to 0.6 mg/kg; IM in lumbar musculature), ketamine (5 mg/kg or 10 to 20 ml/hr of a CRI prepared with 60 mg ketamine/liter fluid) and butorphanol (0.2 mg/kg) as a preanesthetic followed by buprenorphine (0.03 mg/kg or 30 µg/kg) or oxymorphone (0.05 mg/kg IM) immediately post-operatively. Another example is control of cancer pain in dogs: ketamine has been used preoperatively (0.5 mg/kg IV), intraoperatively (10 µg/kg/min) and postoperatively (2 µg/kg/min) in conjunction with fentanyl. Some oral alternatives to NMDA antagonism may include dextromethorphan (use an antitussive dose?) and amantadine.

**Amantadine** (3 mg/kg sid; 100 mg capsule or 10 mg/ml solution), a drug used to treat Parkinson’s disease (apparently it increases dopamine by a largely unknown mechanism) is an orally bioavailable drug which also appears to inhibit NMDA receptor activation. This drug also is being used as part of a combination approach for controlling pain in animals, although citations are largely anecdotal.

**Disease Modifying Agents.** Normal cartilage is avascular and tightly adhered to cortical bone. A load-bearing and gliding surface of the joint is formed such that a frictionless surface occurs throughout the range of motion of the joint. The fibrous capsule of the joint contains a layer of synovial cells which are very vascular and serve as a selective membrane, precluding passage of molecules greater than 12,000 MW. Synovial fluid produced by the cells lubricates and nourishes cartilage. Hyaline cartilage contains a small number of chondrocytes which synthesize the matrix in which they are embedded. The matrix is comprised of collagen fibers interspersed in a well structure manner with proteoglycan (PGN) aggregates of varying molecular weights (MW). Proteoglycans are comprised of glycosaminoglycans encircling a core protein. The PGN complex in turn is bound (by a link protein) to hyaluronic acid. Chondroitin sulfate is the principle PGN of mature cartilage with other sulfates (keratin, dermaten, etc) making the remainder. Chondroitin sulfates are glycosaminoglycans of varying MW composed of alternating sulfated residues of a glucuronic acid and a galactosamine. Sources of chondroitin sulfate for commercial purposes include bovine trachea (mammalian = chondroitin-4-), nasal septum and shark cartilage (chondroitin-6-). PGNs are large polar (sulfur, amino and hydro) molecules which attract water, thus maintaining the gel-like consistency of cartilage such that it acts as an elastic shock absorber. Chondrocytes are very metabolically active, constantly breaking down and resynthesizing PGN and collagen. The initial lesion in osteoarthritis occurs in cartilage. Chondromalacia (softening of the cartilage) occurs early in the course of disease. Collagen turnover markedly increases by the chondrocytes; reparation may not yield the appropriate (Type II) collagen. Ultimately, collagen loss may predominate. Species differences in the repair of collagen are likely to exist. PGNs also are lost as DJD progresses. Initially, PGN synthesis is markedly increased, but the normal ratios of high MW vs low MW PGNs may not be maintained. Eventually, PGN synthesis markedly decreases. Hyaluronic acid concentrations also decrease. The loss of cartilage matrix is mediated, in part, by proteolytic enzymes such as metalloproteases, including collagenases, stromelysin, and aggrecanase, and lysosomal enzymes released (stimulated by interleukin-I or TNF) by synovial cells or chondrocytes. Interleukins a and 6, tumor necrosis factor and nitric oxide also act as cellular or molecular mediators. Mediators (eicosanoids, interleukin 1 and tumor necrosis factor) act to up-regulate catabolic enzymes of destruction while downregulating mediators which inhibit catabolic actions.
The catabolic process of cartilage degradation worsens as these enzymes are released. The damaged cartilage attempts to repair the damage as it occurs by synthesizing new but malformed proteoglycan and collagen.

**Oral Disease Modifying Agents. Pentosan Polysulfate.** This product (PPS) is isolated from beechwood hemicellulose and synthetically modified by adding sulfates to its repeating units of xylanpyranoses. Thus, unlike previously described disease modifying agents, it is not derived from animal sources. Currently, it is available in the US approved for use to treat interstitial cystitis. In Europe, it is used to treat thrombosis and hyperlipidemia, with application for treatment of osteoarthritis being only recent. It may improve subchondral and synovial membrane blood flow. In addition, it modulates cytokine actions, stimulates hyaluronic acid synthesis and maintains PSGAG content in joints. Its oral bioavailability is not as good as Cosequin, but it can be given orally. When administered IM (2 mg/kg once weekly; oral dose should be 5 times the IM dose) in a model of osteoarthritis in dogs, cartilage damage was significantly decreased by 3 weeks. The drug appears to be safe, but like other PSGAG-like compounds, it appears to prolong clotting times, and may cause thrombocytopenia.

**Nutraceuticals.** These products represent the newest therapy for treatment of DJD in dogs and cats. Most compounds (those which are likely to modify disease) contain glucosamine and chondroitin sulfates (extracted or synthesized) in various complex forms. These products undergo no approval process. A large number of oral disease modifying agents are currently available for treatment of osteoarthritis. Note that no mechanism exist to assure quality control of the manufacture of these products. While products that contain glucosamine are relatively accurately labeled, those containing chondroitin sulfate are much more likely to be mislabeled regarding contents (84% mislabeled in one study), particularly the cheaper products. These products currently are “regulated” by state feed officials. The American Association of State Feed Officials provides guidelines for individual states regarding products not approved by the FDA, but given orally. Most states follow their guidelines. If a product has not been “approved” by AAFCO, their recommendation is that the product not be sold. None of the disease modifying agents are approved by AAFCO. AAFCO recently announced that they will be taking (in the Spring) unapproved products off the shelf and have chosen to focus their initial efforts on glucosamine.

Several products have been used by many veterinarians or pet owners with some evidence (subjective) of benefit. Examples include the mussel extract that may contain mucopolysaccharides/mixed glycosaminoglycans/chelating metals/etc found in Glyco-flex (components less certain) or synthesized glucosamine, chondroitin sulfates and manganese ascorbate) found in Cosequin (Nutramax Laboratories, Inc). Those products which contain various forms of glycosaminoglycans (aggregates form proteoglycans, the major constituent of cartilage matrix) such as glucosamines or chondroitin sulfates appear most promising based on studies that have supported their efficacy. Much of this work has been reported in horses although a large amount of literature supports the use of selected ingredients (especially glucosamine) in the treatment of OA in humans and animals. Presumably, as precursor nutrients, chondroitin sulfates and glucosamines will administered orally are extracted from the serum by chondrocytes and be used to synthesize proteoglycans. Ascorbic acid is a reducing agent for enzymes which form residues important for fibril formation and cross-linkage of collagen fibers for the articular cartilage, joint capsule, tendons, ligaments and bone. During periods in which cartilage degradation exceeds cartilage formation, the need for precursor molecules may exceed availability and the repair process is inhibited. The availability of orally administered compounds not only increases the efficiency of the ability of the chondrocytes to repair damaged cartilage as is evidenced by increased synthesis, but also leaves less
opportunity for formation of inappropriate molecules. Orally supplemented precursors also may inhibit cartilage degradation. Glucosamines appear to modulate the inflammatory process, perhaps by scavenging oxygen radicals. Cartilage degradative enzymes are also inhibited. These products appear to be safe in dogs and cats; an LD50 can not be established. The compounds may actually be gastoprotective. Platelet function may change, but the clinical relevance of this finding is no obvious. Lethal doses can not be established in some animals (indicating the drugs are safe). Oral absorption (up to 70%) of chondroitin sulfate has been reported in a number of species administered radioactive materials. Bioavailability of glucosamine salts will vary, with the HCL salt being move bioavailable (87%) compared to the sulfate salts (47%); thus doses of sulfate salts should be higher. Chondroitins are also bioavailable (70%) as long as the MW is < 17,000 (note that very few products are of this MW and labels will not state MW). Both compounds appear to be taken up into the joint. Studies which have documented the ability of these compounds to modify disease are increasing. A number of studies have used rabbit models of surgically – induced damaged. Controlled studies show clear histological and gross improvement in joints treated with a combination glucosamine-chondroitin product compared to placebo. Some evidence supports that the two components act synergistically. Note that these products will never work as well as NSAID, and NSAID will need to be used as “rescue” drugs. Time to effect is 4 to 6 weeks; a 3 month holdover should be anticipated when changing drugs. A close to 30% placebo drug will also complicate unbiased assessment. S-adenosyl methionine (20 to 30 mg/kg bid to tid) also appears to be useful for treatment of osteoarthritis. MSM (Flexagen ®), superoxide dismutase and DMSO act as oxygen radical scavengers and analgesics and as such should be used in combination with disease modifying agents. DMA should be used prophylactically (prior to elective procedures, in animals predisposed to hip dysplasia), and in any damaged joint situation.

Other potential indications for oral disease modifying agents in animals includes interstitial cystitis in cats, chronic urinary tract infections associated with deep-seated bladder infections, degenerative disc disease (discs are comprised of the component parts of PGAGs) and (early) collapsing trachea.

ENDOCRINE

Hyperglycemia. Acarbose (Precose ®) is a complex oligosaccharide that is bacterial in origin. The enzyme inhibits the degradation of alpha- amylase, and glucosidase of pancreatic and intestinal origin. As a result, enzymatic degradation of complex, oligo, di, trisaccharides is inhibited and glucose absorption is decreased. Post-prandial hyperglycemia subsequently is minimized. However, hypoglycemia is not a sequelae. Indications in humans are non-insulin dependent diabetes mellitus. Because carbohydrates that otherwise would be absorbed are metabolized in small intestine, a number of gastrointestinal side effects occur including pain, diarrhea, and flatulence (77% in humans). These clinical signs tend to resolve once accommodation of flora) has occurred. This product has been studied in diabetic dogs using 25 to 50 mg/kg depending on size. Mean serum glucose pre and 8 hr post prandial, and glycosylated hemoglobin were lowered in dogs receiving both insulin and acarbose compare to insulin and a placebo. Side effects of weight loss and watery stool were evident in normal dogs receiving 200 mg in another study. Vetsulin™ is a sterile aqueous zinc suspension of purified porcine (homologous to canine) insulin approved for use in dogs. PZI insulin is available as a compassionate use product through Blue Ridge Pharmacy of Raleigh, North Carolina; beef-
pork (919)-781-7986 or 800-374-8006 @ $65 per vial plus shipping, a subsidiary of IDEXX. Other insulin preparations to be tried include Ultralente or Lente humulin products. PZI insulin is an example of a product for which accuracy in compounding is paramount. **Glargine insulin (Lantus®)** is one example of several injectable human recombinant long acting insulin analogue that differ from human insulin by the replacement (for glargin, asparagine is substituted with two glycines) and/or addition (for glargine, two asparagines) of amino acids. The resulting insulins are an attempt to prolong action and resolve peaks. Glargine is described as “peakless” in humans, thus mimicking more closely endogenous glucose/insulinrelationships. The potency in humans is similar to insulin. Concentrations of insulin remain relatively constant for a 24 hour period following subcutaneous administration. In healthy nondiabetic cats, glargine was similar to PZI and porcine lente insulin in time to onset and nadir glucose concentration, but time to reach nadir was longer for glargine. Time to return to baseline glucose was similar for glargine and PZI which was longer than lente. Concentrations were not peakless. However, a subsequent study in diabetic cats suggested that glucose concentrations might indeed flatten. The starting dose of 0.5 U/kg ideal body weight should begin at a BID dosing interval for cats with glucose ≥ 360 mg/dl, and at 0.25 U/kg if less than 360 mg/dl. Animals should be hospitalized for 3 days and a 12 hr glucose curve is recommended with samples at 4 hr intervals. Doses should not be increased the first week. The insulin should not be mixed with anything (insulin release is pH dependent) and the product should be kept refrigerated (where it can be kept for up to 6 months). Urine glucose should remain below 2+ (assuming overdosing and hyperglycemic overswing is not occurring).

**Trilostane** is a synthetic competitive inhibitor of 3-β-hydroxysteroid dehydrogenase, which prevents formation of progesterone and subsequent formation of steroidal hormones, including endogenous corticosteroids. It has been used successfully to treat human hyperadrenocorticism of various causes. The drug has been studied in the United Kingdom in dogs with naturally occurring PDH. Doses varied with weight: 30 mg if less than 5 kg 60 mg for 5 to 19 kg and 120 mg if > 60 kg once daily. Medication was well tolerated; mean dose necessary to control PDH was approximately 20 mg/kg with larger dogs requiring more on a mg/kg basis. Response was based on resolution of clinical signs, baseline and post – stimulation cortisol and urinary cortisol creatinine ratio (although the latter did not change statistically across time). Percent of animals controlled was 40% at 10 days, 57% at 90 days, and 80% at 180 days. Time to response was initially 89 days, but as the investigators became more adept, the duration was reduced to 40 days. A starting dose of 10 mg/kg once daily was suggested. Trilostane also has been successfully used in at least one cat to control clinical signs when treated with 30 mg/kg once daily orally and a dog with an adrenal tumor.

**Urinary.** Since diethylstilbesterol (DES) has been removed from the market, Premarin®, a conjugated estrogen, has been recommended as its replacement (0.625 mg for a 60 lb animal). However, while DES is nonsteroidal, this and other estrogen products should be used cautiously. Several veterinary pharmacies can compound DES which is still available as a powder. Phenylpropanolamine or the tricyclic antidepressants should be alternatives to estrogen-based therapy. **Phenopropanolamine** has not been removed from the market and is available from some compounding pharmacies. Psuedoephedrine has been used in Australia as an alternative (15, 30, 45, 60 and 75 mg total dose for dogs ranging in size from < 10 kg to > 60 kg).Vasopressin® has been available only as a nasal spray (give subconjunctively), but now is available as a 0.1 mg and 0.2 mg tablet. Human bioavailability of the tablet is only 5 to 15% of the intranasal drug. Since each nasal drop contains about 0.1 mg (and assuming a same level of bioavailability), the oral dose in the dog would be about 1 to 2 tablets a day. Currently, the nasal drops are probably still less expensive and the oral tablets.
**Renal Disease:** Control of phosphorus is critical for slowing the development of complications associated with secondary hyperparathyroidism. Generally this is accomplished with aluminum containing phosphate binders. Either the hydroxide or carbonate anion can be used. Of the two, aluminum carbonate is available as a capsule which can be pulled apart and added to food (divide a 125 or 250 mg capsule up twice to three times daily). Constipation associated with their use can be treated with either a bulk laxative or lactulose (see gastrointestinal). **Calcitriol** (1,25-dihydroxycholecalciferol; 1,25-dihydroxy Vitamin D). Vitamin D requires two activation steps before it is capable of controlling calcium balance. The final step of activation is the formation of the 1,25 dihydroxy vitamin takes place in the kidneys. The enzyme necessary for inactivation becomes deficient with renal disease resulting in a deficiency. Vitamin D facilitates calcium absorption from the gastrointestinal tract and its absence can (2.5 to 3.5 ng/kg po/day) contribute to the calcium/phosphorous imbalances leading to secondary hyperparathyroidism of renal disease. Calcitriol therapy, if begun sufficiently early in the course of renal disease, decreases the risk of nephrocalcinosis, the magnitude of hyperparathyroidism and can slow the progression of glomerularsclerosis. Although it should be implemented as early as possible, calcitriol is not indicated in all patients. It should not be used if the product of serum calcium and phosphorus exceeds 60; phosphorus should be less than 6 mg/dl and ideally less than 4 mg/dl. Use in the presence of high serum phosphorus will increase the risk of nephrocalcinosis. Serum calcium must be measured as long as the patient is receiving the drug. Monitoring should occur weekly for 4 weeks, then bi weekly for a month. The interval can then be increased to monthly for several months followed by 3 to 6 month intervals as necessary.

Twenty-five percent of cats with renal disease also may be suffering from an iron deficiency, perhaps due to chronic blood loss associated with gastritis. Iron (ferrous sulfate) can be supplemented prophylactically (1 mg/cat/day po) or as a treatment in mildly anemic cats, prior to treatment with erythropoietin at 15 mg/cat/day. Severe iron deficiency anemia may require parenteral therapy (iron dextran 50 mg/cat/day, IM). EPO should be administered when the PCV is between 12 and 19%, as is indicated by clinical signs, and after alternate causes of anemia (eg, iron deficiency, gastrointestinal bleeding, etc) have been ruled out. This should take approximately 2 months; if more rapid, the dose s should be decreased in order to avoid polycythemia. Hypertension may occur if the PCV is too elevated and is more likely in patients already hypertensive. Treatment begins at 50 to 150 Units/kg/cat 3 times weekly until a PCV of 30 to 35% is reached. Daily therapy can be initiated with severe anemia, but not in hypertensive patients. A maintenance dose of 50 U/kg/cat 2 to 3 times per week begins once the targeted PCV is achieved. Up to 25 to 40% of cats receiving human recombinant EPO will develop antibodies as indicated by a drop in PCV several weeks to several months after therapy has increased the PCV. Anabolic steroids might be implemented in those patients that can not tolerate EPO.

**IMMUNOLOGIC**
**“Soft” Glucocorticoids.** Among the mechanisms whereby undesirable side effects of glucocorticoids can be minimized is topical administration of drugs which are potent for the glucocorticoid receptor but also rapidly metabolized should the drug be absorbed into systemic circulation. Examples include beclamethasone, budesonide and fluticasone propionate, steroids designed specifically for use in inhalant metered doses. Their potency when inhaled varies in clinical trials, with fluticasone propionate being most potent and budesonide and beclomethasone dipropionate approximately equipotent. Time of onset in humans to budesonide is approximately 10hrs based on evidence of clinical improvement at that time. Improvement can be expected over the next 1-2 days, with maximum effects potentially not being evident until 2 weeks after therapy has begun.

Budesonide is also available as an oral preparation. It is rapidly metabolized in the liver by CYP3A4. Inhalant drug is generally considered to be absorbed (as much as 25%). The side effects of glucocorticoids might be reduced with “soft” preparations, but probably will not be avoided by topical therapy. Drugs which impaire CYP3A4 (eg, CsA) may increase the plasma drug concentration of budesonide over 7 fold. Even by itself, budesonide appears to suppress the hypothalamic-adrenal-pituitary axis in dogs based on a clinical trial in dogs (n=6) with IBD. Budesonide has been used to treat CIBD in dogs. Budesonide has been recommended in dogs (1- 3 mg daily depending on dog size) although no scientific or anecdotal information has been published regarding its efficacy in animals. Means to reduce these side effects include combination drug therapies.

**Cyclosporine** is a T-cell specific immuomodulator approved for use in humans for immune suppression of graft vs host transplant rejection, autoimmune disorders, keratitis sicca and red cell aplasias. The standard preparation is very lipophilic and as such must be prepared in oil and oral absorption is bile acid dependent; oral bioavailability ranges from 20 to 50%. The newer microemulsion preparations (Atopica® or Neoral®) is not as dependent on bile acids for oral absorption and is more bioavailable. Note that the immunomodulatory effects generally will necessitate twice daily dosing (5 mg/kg twice daily) because of the short half-life of the drug in dogs and cats (3 to 9 hrs). With the exception of atopy (as per approval data), once daily dosing of cyclosporine is not likely to be preferred. In humans, peak concentrations are more predictive of area under the curve; a target of 4500-5000 ng/ml is recommended. However, trough concentrations of 600-800 ng/ml also have been recommended for transplantation patients. Similar studies have not been performed in animals. Note that often a more cost effective approach for reaching target concentrations is decreasing the interval rather than increasing the dose. Doubling concentrations will require doubling the dose, but tripling concentrations will require a 4 fold dose increase, and quadrupling the drug concentrations will require an 8 fold dose increase. Although ketaconazole has been used to increase concentrations, this approach is recommended only if both a peak and trough sample can be collected so the half-life can be monitored. We have measured half-lives of > 150 hrs; for such patients, drug concentrations do not reach steady-state for close to 4 weeks. Finally, note that CsA is involved in many drug interactions because of both its impact on drug metabolism and p-glycoprotein.

A placebo controlled clinical trial provides strong evidence that the drug is a nonsurgical alternative to treatment of perianal fistulas in dogs. When given at a dose (as little as 3 to mg/kg but up to 5 to 15 mg/kg; orally daily) which maintains trough concentrations at 400 to 600 ng/ml, improvement occurred in all treated animals within 4 weeks, with resolution in 85% of the dogs occurring by the 16th week of treatment. Trough concentrations as low as 100 to 300 ng/ml may be effective. Recrudescence of the syndrome appears to occur in a small
number of animals, with subsequent retreatment apparently effective in resolving the lesions. Ketaconazole, an inhibitor of hepatic drug metabolizing enzymes, can be used (2.2-8.5 mg/kg/day; divide and administer bid) to decrease the dose of cyclosporine (to 1.0 to 3.5 mg/kg/day; divide and administer bid) necessary to maintain effective concentrations. This has been documented in only a small number of dogs; hepatic enzymes might be monitored bi-weekly during combined therapy to assure no combined hepatotoxic effects. Vitamine E may also be used to prolong blood concentrations of cyclosporine. As an apparent inhibitor of p-glycoprotein, an efflux carrier of intracellular drugs, Vitamin E appears to increase oral absorption of cyclosporine. This may be particularly useful (10 IU/kg with each oral cyclosporine administration) for Sandimmune®, which is less bioavailable than the newer, more bioavailable microemulsion preparation, Neoral®. Therapeutic drug monitoring is strongly recommended, particularly when using adjuvant drugs to decrease the cost of cyclosporine. Tacrolimus is similar to cyclosporine in its actions, toxicities and indications. It is available as an ointment and was associated with cure of perianal fistulas in 50% of dogs treated for up to 16 weeks; 90% of animals responded. Tacrolimus is similar to cyclosporine in action, but is available (0.1%) as a topical ointment (cost about $110/60 gms). A 50% cure rate was realized (90% improvement rate) when used twice daily for 16 to 20 weeks for topical treatment of perianal fistulas. The ointment also has been used with close to 90% response rate in patients with localized disocoid lupus and pemphigus erythmatosus lesions.

Mycophenolate mofetil is a lymphocyte specific immunomodulating drug that was developed in humans for treating graft versus host rejection in lieu of cyclosporine which is too toxic. The drug impairs lymphocyte synthesis of a essential metabolite. It is a pro-drug that must be activated by the liver. The drug is very safe in humans, with side effects limited to gastrointestinal signs. The drug has been used successfully in dogs (20 mg/kg bid orally) to treat myasthenia gravis and, more recently, other immune mediated disorders that no longer respond to glucocorticoids, or if cyclophosphamide can not be tolerated. However, toxicity in 1 out of 6 experimental animals indicates that prudence is warranted with its use in clinical cases, and reservation of its use to non-responders.

The use of leukotriene receptor antagonists (see respiratory) should be considered in any inflammatory disease associated with eosinophilic (and to a lesser degree, lymphocytic) influx. In humans, therapy targets the bone marrow; leukotrienes appear to have a role in stimulating stem cells to differentiate and mature into eosinophilic precursors. They also appear to stimulate the release into systemic circulation.

**DERMATOLOGIC**

The treatment of atopy, chronic inflammatory bowel disease and respiratory inflammatory disease (ie, asthma) might be approached similarly by focusing on the commonality of the diseases (antigen exposure; initial tissue response; generation of local mediators that become systemic; impact of mediators at the level of the bone marrow; bone marrow response through cytokines such as interleukin – 5 [chemokines including eotaxin; leukotrienes may act as signal between interleukins and cells], release of inflammatory cells, movement of inflammatory cells to the cite of tissue response. Tissue responses involving eosinophils and lymphocytes are particularly conducive to this approach. Treatment might target both the tissue associated with clinical signs and response at the level of the bone marrow. Treatment also should include adjuvant therapy specific for the control of clinical signs in each of the target tissues.

**Pruritis. Misoprostol**, a PGE analogue, is one of the most potent inhibitors of mast cell degranulation known (Silverstein 1998) and recently has been studied for treatment of
chronic pruritis. When administered at 6 ug/kg PO tid, 60% of animals improved by ≥ 50% within 3 weeks. It may act synergistically when combined with the antihistamines hydroxyzine or , clemastine. The effects of misoprostol on mast cells should warrant is consideration as part of the armamentarium used for palliative therapy in dogs or cats with mast cell disease. **Pentoxyfylline** is a methylxanthine derivative (similar to theophylline) with minimal respiratory or cardiac effects. Its ability to change rheological properties (perhaps by changing red blood cell deformability, or blood viscosity) (Amrus 1990) and potential anti-inflammatory effects (including potential antiplatelet aggregation) has led to its use in dermatomyositis, atopy, and pruritis associated with atopy (Asanuma 1997). It also has been used for treatment of immune-mediated diseases of the skin. Metabolites of the compound contribute a large component of pharmacologic activity; it currently is being studied in dogs (Rees 1999) (10-15 mg/kg orally twice daily, for 2 to 6 weeks). **Tapoxalin (Zubrin®)** might also be considered.

Hydroxyzine hydrochloride has recently become very expensive. However, a ** benzoate salt is available that is less expensive and appears to be equally bioavailable in humans (not studied in animals). The drug should be dosed as with the hydrochloride sale. In contrast to older drugs, newer antihistaminergic drugs prevent mast cell degranulation as well as block the effects of histamine at the receptor. Thus, these drugs should be more effective compared to traditional antihistamines. Examples include cetirizine (Zytrec® 2.5 to 10 mg once daily, orally), Loratidine (Claritine, 0.25 to 0.5 mg/kg sid orally) and fenoxfenadine (Allegra®, 30 mg/kg orally twice daily).

Lufeneron (65 [dog] or 75 [cat] mg/kg once; possibly repeat in 3 weeks; and terbinafine (10 mg/kg once daily orally) have been used to treat dermatophytosis. Terbinafine also is effective against sporotrichosis and aspergillosis; both drugs should be considered for combination therapy against these organisms.

**GASTROINTESTINAL**

Cisapride has been withdrawn from the human market because of its use is associated with Torsdade’s syndrome, a cardiac disrhythmia. There are no current plans to market it for animals or humans. The FDA suggests that veterinarians that use the drug import a small amount from a foreign country, which is allowed as long as the drug is not shipped commercially (that is, the veterinarian cannot sell the drug to others), the drug is for personal use (that is, the veterinarian prescribes the drug), the company selling the drug does not encourage or advertise the extra-label use and a valid client veterinary patient relationship exists. Bethanechol, a cholinergic agent used to treat hypotonic urinary bladder (5 mg for small dogs, 10 mg for larger dogs) has not been that useful in our experience. Other alternatives include the H-2 receptor antagonists nizatidine and ranitidine, which are also acetylcholinesterase inhibitors. Nazitidine (5 mg/kg once daily orally) significantly increases gastric emptying and may be the preferred drug. Erythromycin has motilin-like effects at low doses (0.5 to 1 mg/kg orally) and will accelerate gastric emptying in the dog (hence its gastrointestinal side effects when used to treat infections). However, food that is emptied may be unprocessed and thus nondigestible. Erythromycin also appears to have some colonic activity in the dog, but not cat. Misoprostol appears to stimulate colonic propulsive activity and may be useful for treatment of refractory constipation in dogs and cats (see below). Finally, two new serotonin (5-HT-4) receptor agonist products are being studied for approval in the US: tegaserod (Novartis) which has effects in the canine colon, (0.03 to 0.3 mg/kg IV) and prucalopride (Janssen Pharmaceutical) which has colonic activity in the dog and cat and increases gastric emptying in the dog (0.02 to 1.25 mg/kg).

Although **Misoprostol (2-5 µg/ kg po bid)** might be indicated for the treatment of
ulceration regardless of the lesion, specific indications would include prophylaxis or treatment of ulcerations induced by nonsteroidal anti-inflammatories; mucosal erosion accompanying liver disease may also respond to prostaglandin E$_2$. Side effects include gastrointestinal cramping and diarrhea. Although histamine receptor antagonists will contribute to healing, they control only hydrochloric acid secretion and do not contribute to other aspects of ulcer or erosion healing contributed by the prostaglandins. Thus, their use is encouraged in patients with NSAID-induced gastrointestinal. An additional reason for using misoprostol prophylactically is that it appears to facilitate the anti-inflammatory effects of NSAID drugs in the treatment of osteoarthritis. Misoprostol also is a potent inhibitor of mast cell degranulation and this effect might be useful in CIBD.

*Helicobacter felis*, like *Helicobacter pyloris* in humans, may play a role in gastrointestinal ulceration. Treatment is oriented toward increasing gastric pH with antisecretory drugs (ie, omeprazole or ranitidine); and antibiotics effective against the organism (metronidazole; amoxicillin; tetracyclines). Bismuth compounds act to disrupt the integrity of bacterial cell walls and can be an effective part of combined therapy (used as bismuth subsalicylate). Use of antisecretory drugs (most often omeprazole) is well established in veterinary medicine. Rebound hypersecretion has been reported in humans when drugs which decrease gastric acid secretion (eg, ranitidine, omeprazole) are discontinued abruptly. The rebound reflects response to high plasma gastrin concentrations stimulated by high gastric pH. Hypersecretion can be minimized by tapering the dose gradually as the drug is discontinued.

Treatment of chronic inflammatory bowel disease includes asulfasalazine, a combination of 5-amino-salicylic acid (5-ASA) with sulfapyridine. The sulfonamide component might be considered undesirable, and products have been developed in human medicine which contain 5-ASA but not a sulfonamide. These include mesalamine (10-20 mg/kg [D] PO every 6-8 hrs) and osalazine sodium (10-20 mg/kg PO every 12-24 hrs). The former is also available in an enema preparation for treatment of disease limited to the lower bowel. The aspirin in these products - like that in asulfasalazine - can be absorbed (including that in enema form) and caution is recommended to avoid aspirin overdose. The product is available in a slow release preparation that releases drug only in the colon and ileum and thus limit effects to the lower bowel. Oral absorption also may be reduced. The drug delivery system may pass with the feces apparently intact; however, drug movement out of the capsules occurs in the gastrointestinal tract. Pancreatitis is an apparently recently but rarely recognized potential side effect in humans receiving these drugs. The use of leukotriene receptor antagonists (see Respiratory) also should be considered, particularly in animals that do not respond to or can not take (eg, renal failure) glucocorticoids. Animals that continue to be unresponsive to medical management of IBD may respond to chlorambucil (if lymphocytic; 15 mg/m$^2$ days 1,2,3 and 4 of every 21 days; or 2 mg PO q4-5 days). Cyclosporine (see immunomodulators) also should be considered. Azathioprine (0.3 mg/kg every other day, orally) is more toxic in cats compared to dogs and should be used cautiously. Response may take up to 5 weeks. Adjuvant therapy for CIBD should include antisecretory drugs (eg, ranitidine, famotidine, omeprazole) as indicated to facilitate healing and prevent worsening of damages in gastroduodenal or small intestinal disease. Sucralfate should be used to facilitate healing of damaged mucosa. Folate and cobalamin also might be supplemented because they become depleted with chronic inflammatory disease of the small bowel. Treatment for *Helicobacter* spp might also be considered in animals whose condition does not respond to therapy. Treatment of this organism may become a more important target for IBD therapy as we learn more about its role.

*S-Adenosyl-L-Methionine (SAMe)* is a naturally-occurring compound distributed throughout the body, including the liver. As a major methyl donor, it activates enzymes
responsible for the synthesis and metabolism of hormones and neurotransmitters, and cellular constituents such as nucleic acids, phospholipids and proteins. Its synthesis is markedly decreased in patients with chronic liver disease. SAMe appears to be responsible for sulfation of hepatotoxic endogenous bile acids and thus promotes bile acid secretion. Its relative absence in liver disease may contribute to intrahepatic cholestasis and the progression of liver disease. In addition, SAMe may also directly protect the liver from continued damage by methylating hepatic cell membrane phospholipids, thus allowing enhanced membrane fluidity. Studies in cell cultures, experimental animals, and human patients afflicted with liver disease have documented the efficacy of SAMe when administered orally for control of liver disease associated with cholestasis. As with UCDA, scientific studies establishing the safety and efficacy of SAMe in diseased animals are indicated.

**RESPIRATORY SYSTEM**

Feline Asthma: Recent advances in the treatment of human asthma increasingly are leading to its treatment as a systemic rather than simply local allergic disease. In particular, the bone marrow response to allergens, and subsequent release of eosinophils, is recognized to be an important systemic process in allergic inflammation. Several cytokines, and in particular, the eotaxin IL-5, contribute to eosinophil movement from the bone marrow to local tissues. In humans, the number of eosinophil/basophil progenitors in the bone marrow declines with the peak allergy season, suggesting that the mature cells are circulating systemically. A central role of IL-5 has been supported through a number of studies documenting increased concentrations in asthmatics. Eosinophilia, lung damage and airway hyperresponsiveness are blunted in IL-5 deficient antigen-challenged mice. Sources of IL-5 include T helper 2 (TH2) lymphocytes, mast cells, eosinophils and bone marrow stroma. Among its actions are promotion of differentiation and maturation of progenitor cells, release of mature eosinophils, promotion of survival and inhibition of apoptosis. T-cell recruitment to the lungs may play a key role in inflammation. The role of LTs is not clear, although cysteinyl LTs are expressed on a number of bone marrow progenitor cells and appear to be involved (based on effects of antagonists) in eosinophil/basophil progenitor differentiation. Leukotrienes are very potent causes of inflammation in the lungs, causing marked edema, inflammation, and bronchoconstriction. The recent approval of drugs that specifically inhibit the formation of LTs or their actions have offered a new avenue of control of respiratory inflammatory disease (e.g., asthma) in human medicine.

Zafirlukast (Accolate) is a leukotriene receptor antagonist (LRA), whereas zileuton (Zyflo) is a lipooxygenase inhibitor. Comparative studies in animals and humans suggest that, of the two, zafirlukast (0.15 to 0.2mg/kg orally once daily) or montelukast (0.5-1.0 mg/kg po sid) is more effective and less likely to increase hepatic enzymes (and thus is safer) among different species. and can be administered less frequently (every 8 to 12 hours compared with every 6 to 8 hours for zileuton). Zafirlukast is associated with very few side effects in human patients. However, a recent report delineates severe acute hepatopathy in three human patients receiving zafirlukast for at least 5 months. In human clinical trials, LRA inhibit early and late phase bronchoconstriction and increased bronchial hyperresponsiveness in response to allergens, accumulation of inflammatory cells and mediators in bronchial lavage fluid, and acute bronchospasms stimulated by exercise, cold air and aspirin. Their use has been associated with improvement of asthma either as sole therapy (in lieu of low dose inhaled glucocorticoids) in mild to moderate asthma or as add-on therapy regardless of diseases severity in
Glucocorticoids non-responders. The role of LAR in the treatment of feline asthma has had some, albeit limited attention. Because receptors for leukotrienes have not been found in the smooth muscle of airways in cats, the use of antagonists have been questioned. However, with the recent approach to asthma as a systemic response mediated at the level of the bone marrow, the use of LRA warrants further and strong consideration.

**Glucocorticoids:** In 1997, the National Heart Lung Blood Institute Expert Panel Guidelines recommended control of mild persistent asthma with a single, long-term control medication with anti-inflammatory properties. Glucocorticoid efficacy for treatment of respiratory inflammatory disease is dependent on achieving therapeutic concentrations in and below the epithelium of all diseased airways. However, whereas systemic therapy might provide the most consistent exposure to diseased airways, it also provides the greatest exposure to tissues other than the lungs, leading to adverse effects. Administration of glucocorticoids by aerosol has decrease many of the side effects associated with systemic glucocorticoids use in humans. Indeed, the preferred treatment for humans with mild disease is low-dose inhaled glucocorticoids. However, short courses (5 to 7 days) of high doses of oral glucocorticoids are used to treat acute exacerbations of asthma.

**Inhalant Devices: Glucocorticoids:** The addition of inhaled glucocorticoids has increased asthmatic control in humans. Beclomethasone was among the first aerosol glucocorticoids developed for inhalant therapy. Systemic side effects associated with deposition of glucocorticoids on the pharynx and central airways and local side effects in the upper airway (e.g., dysphonia in up to 50% of the patients) led to the inclusion of “spacers” that removed larger particles before they penetrated the pharynx. Additionally, administration of glucocorticoids removed by first pass metabolism (e.g., budesonide or fluticasone [preferred]) decreased the risk of systemic exposure to swallowed drug. However, poor compliance of inhaled glucocorticoids in human patients led to the development of combinations of steroids with long-acting β2 agonists (e.g., salmeterol/fluticasone or formoterol/budesonide). Although compliance has improved, concern has arisen that the β2 agonists will mask clinical signs that might otherwise indicate worsening of the disease. Indeed, recent studies of the efficacy of inhaled beclomethasone dipropionate found improvements to be short-lived, probably because the inhaled drug does not control inflammation well. Reduced airway caliber will further decrease efficacy by reducing drug delivery to the peripheral airways. In human asthmatics, deposition studies reveal that the majority of drug is deposited on central airways. Yet, anti-inflammatory therapy should target both large and small airways if inflammation is to be suppressed. Thus, systemic therapy should be considered (either as sole therapy or in addition to systemic therapy) in animals with moderate to severe disease. Note that the peak effect of inhaled glucocorticoids may not occur for 1 to 2 weeks after therapy has begun.

**Beta-adrenergics:** With the advent of metered doses inhalers in the 1960s, beta-adrenergics became a common therapy for treatment of human asthma. Short-acting β2 agonists administered by aerosol include albuterol (preferred), pirbuterol, bitolterol and terbutaline whereas longer-acting (in humans) products include salmeterol (preferred) and formoterol. The drugs differ in their effect and use. Short acting products are associated with rapid symptomatic relief in human asthmatics when used at appropriate doses. However, use at high doses has been associated with an increase in mortality in humans, leading to their recommended use on an “only as needed” basis. On the other hand, improvement in pulmonary function in humans was sustained with prolonged used of long-acting beta-adrenergics and thus such use was not associated with a decrease in symptomatic relief afforded by short-acting drugs. Note that time to onset of long acting drugs may be up to one hour. Although minimally
effective by themselves for control of inflammation, long-acting beta-adrenergics appear to enhance responsiveness to glucocorticoids. Rebound hyperresponsiveness does not appear to occur with rapid discontinuation of the long-acting drugs. Tolerance may occur to the effects of beta-adrenergics, probably due to down regulation of receptors, particularly in the presence of triggering events (such as exercise in humans). Because beta-adrenergics do not provide as much anti-inflammatory control, their efficacy may be reduced in the presence of inflammation and combination therapy with an anti-inflammatory drug (eg, inhaled or systemic glucocorticoids), or use of theophylline may be indicated.

Theophylline is available in slow release preparations; however, the kinetics of release of these products are designed for humans and vary among animals. Selected products allow twice daily dosing in dogs and once daily dosing in cats (Theodur®: 25 mg/kg sid in pm po [C]; 20 mg/kg bid po [D]; Slo-bid®: 25 mg/kg bid to sid [C] 20 mg/kg sid). Because the bioavailability of these products markedly varies, therapeutic drug monitoring should be used to confirm therapeutic concentrations (10 to 20 g/ml) particularly if the patient does not appear to be responding. Theo-Dur® is among the most bioavailable in dogs. Terbutaline (Brethine®: 0.2 mg/kg po every 8-12 hrs [D]; 1.25 to 2.5 mg/cat po every 12 to 24 hrs) is a specific beta_2 agonist that can be used for bronchodilation while avoiding cardiac stimulation. It can be used in patients that have not responded to theophylline; the two can be combined for potential additive effects.

N-acetylcysteine serves as a precursor to glutathione, a major scavenger of free oxygen radicals associated with inflammation. The drug also appears to induce respiratory tract secretions, probably via a gastro-pulmonary reflex. The drug can be used orally or IV, with indications in people including toxic inhalants (including tobacco smoke), bronchitis, chronic obstructive pulmonary disease, cystic fibrosis, asthma, tuberculosis, pneumonia and emphysema and the adult respiratory distress syndrome. Installation of a 10-20% solution has also been used to clean and treat chronic sinusitis and the drug is of benefit in treatment of chronic obstructive pulmonary disease. It also appears to facilitate treatment of bacterial infections of the respiratory tract. Acetylcysteine therapy is associated with few adverse affects. In a small number of human patients, IV administration has been associated with an anaphylactoid reaction which can be minimized by administration of histamine receptor (subtype 1) antagonists. In humans, doses as high as 500 mg/kg are well tolerated, although vomition and anorexia can occur. Doses of 250 to 500 mg (total) twice daily are reported to be effective. N-acetylcysteine appears to have benefits in other inflammatory conditions, including septic shock. Because it is metabolized to sulfur containing products, it should be used cautiously in animals suffering from liver disease characterized by hepatic encephalopathy, but it can be used to help protect the liver following any drug-induced hepatotoxicity.

Rutin (50 mg/ kg po bid) is a non-anticouagulant coumarin used to treat selected causes of peripheral limb edema in human patients. The edema should be associated with protein exudation (ie, edema due to permeability changes or lymphatic blockage). The drug, available in health food stores, acts to stimulate macrophage removal of proteins and thus removes the oncotic flux of fluid into tissues. Indications include appropriate causes of peripheral limb edema and pleural effusions associated with proteinatious secretions (ie, chylothorax). Response may take several weeks to months. There appear to be no toxicities associated with the drug.

CARDIOVASCULAR

ACE Inhibitors and Renal Disease A study in dogs with CHF found life expectancy to
increase from 300 days to 800 days when treated with benazepril. However, the efficacy of ACE inhibitors in treatment of canine CHF remains controversial. In patients in which cardiac reserve threatens renal perfusion, renal disease may be more likely. Renal blood flow in such patients is dependent upon the ability of the kidney to auto-regulate: constriction of the efferent arteriole forces blood flow from the renal artery into the glomerulus in the presence of reduced renal blood flow, thus maintaining the glomerular filtration rate (GFR). Treatment with an ACEI precludes autoregulation. However, if cardiac output increases in response to decreased afterload sufficiently to compensate for the loss of efferent arteriolar constriction, GFR is minimally impacted. Sodium wasting (including excessive use of diuretics), and drugs which modify (decrease) renal prostaglandins (NSAIDs, aminoglycosides) may predispose the patient to renal disease once ACE inhibitors are begun. Tests indicative of renal function should be monitored the first several weeks of therapy and the ACEI dose or interval should be adjusted to the minimum effective dose.

**Benazepril**, like enalapril, is a prodrug, and must be converted to its active metabolite, benazeprilate. Approved for use in dogs in Canada, it is administered once daily (0.25 to 0.5 mg/kg orally). In contrast to enalapril, altered renal function does not appear to alter the kinetics of benazepril in dogs. **Lisinopril** is a renally-eliminated ACE inhibitor approved for human use. In contrast to enalapril, it requires no pro-drug activation by the liver. Its elimination is impacted only with marked decrease in GFR. Indications in animals would include liver disease. Another advantage is that it can be given once daily (0.5 mg/kg orally). A recently recognized indication of ACEI is proteinuria. The mechanism by which ACE inhibitors decrease proteinuria is not known, but may include changes in hydrostatic pressure or direct action on the glomerular membrane. Decreasing protein exposure to renal tubular cells appears to decrease the progression of glomerular disease to glomerulonephritis and prolong or reduce the development of glomerulosclerosis. A renal protective effect has also been described for generalized renal disease. However, benazepril is approved for use in cats in Australia for treatment of chronic renal insufficiency. A study of 201 cats in Sweden found the drug to prolong life expectancy to 402 days compared to 126 days in cats that did not receive the drug (2.5 to 5 mg a day).

**Carvedilol**. Is a “third” generation beta blocker that also is characterized by alpha adrenergic blockade. Blockade is relatively selective for beta-1 receptors. Its combined effects result in decreased total peripheral resistance and preload without compromise of cardiac output or reflex tachycardia. Decreased heart rate may not be as dramatic as with other beta-blockers. In human patients with left ventricular failure, it has reduced mortality. A marked advantage of this drug is potential anti-oxidant antiproliferative effects in the heart as well as decreased myocardial cell fibrosis, apoptosis and remodeling (as occurs with other beta-blockers, such as **metaprolol**; 0.2-0.4 mg/kg bid po). As such, the drug may decrease progression of myocardial failure; indeed, it is approved for such use in humans. The dose has not been well established, but dogs are started out at ½ of a 3.125 mg/tablet once to twice daily. If the dose is well tolerated (blood pressure does not decrease), then the dose is doubled in two weeks. Among the diuretics, **spironolactone** also has been associated with a beneficial effect on myocardial remodeling in patients with dilated cardiomyopathy and its use should be considered early in the pathophysiology, including as part of combination therapy. **Pimobendan** is a positive inotropic drug that targets phosphodiesterase (III). As such it not only increased contractility (more effectively than currently available cardiac glycosides) but also helps ameliorate neurohumoral responses by causing peripheral vasodilation and thus reduced preload & afterload. The increase in renal perfusion helps ameliorate some activation of the rennin-angiotensin-aldosterone system. Marketed by Boeringer INgelheim as Vetmiden® in other countries, the drug is not available in the US. It thus far has proven equal to or superior to ACE inhibitors in dogs with congestive heart failure.
Antimicrobials

Antibiotics

In human medicine, few antimicrobials are under consideration for approval. However, the few that are in line tend to focus on microbes that are particularly problematic in humans, such as methicillin-resistant *Staph aureus* (MRSA) and vancomycin-resistant *Enterococcus* (VRE). The spectrum of such drugs can be expected to be very narrow; further, the drugs are likely to be more toxic. In human medicine, experts in the field are reconsidering the need of approving new antibiotics versus more judicious use of those currently available. MRSA, which has been a common hospital acquired nosocomial pathogen, has now become a common community acquired organism. Further, and disconcertingly, it has acquired virulence factors that have led to increased morbidity and mortality. *Clostridium difficile* is rapidly joining MRSA as an epidemiologic concern. The role of antimicrobial use in the advent of resistance is well recognized. Among the different methods which might control the advent of resistance is de-escalation. De-escalation –differ from “restrictive” use that precludes the use of certain antimicrobials (eg, aminoglycosides) hospital environents. The latter is probably an inappropriate approach if it precludes the use of the most appropriate antimicrobial for the patient. The former focuses on 1. avoiding indiscriminant use in the face of limited evidence of infection; 2. narrowing the spectrum of the drug such that microbes are not broadly exposed to drugs; 3. limiting the duration (but not dose) of the drug such that exposure time is less. The variable bioavailability of ciprofloxacin (mean of 40% in dogs; range not known) suggests that the dose of the drug be double or tripled compared to a comparable dose of enrofloxacin. Oral bioavailability is worse in cats (0 to 20%). All currently veterinary approved FQs are associated with retinal degeneration in cats. The mechanism is not clear. Bayer Animal Health has demonstrated safety of enrofloxacin when administered at 5 mg/kg and has demonstrated changes in the retina (based on ERG) in 1/6 cats receiving 20 mg/kg and all cats receiving 50 mg/kg. At labeled doses, neither orbifloxacin nor marbofloxacin appear to be associated with retinal lesions. However, as with enrofloxacin, as doses are increased, the risk of retinal damage also occurs in cats with these FQs. Some early evidence supports (but does not confirm) that changes may be associated with the effects of UV radiation on the retina in cats treated with FQs. Until the mechanism of damage is identified, dosing cats at night, or keeping animals out of sunlight when receiving fluorinated quinolones might be considered. Metronidazole hydrochloride often requires recompounding; however, its bitter taste is hard to mask. Metronidazole benzoate is an alternative medication that is not nearly as bitter. Caution is indicated when using products containing benzoate or benzoic acid in cats because of their susceptibility to methemoglobinemia and other side effects caused by metabolism of benzoate to toxic metabolites. However, the amount of benzoate is far less than the dose that will cause toxicity in cats. When using the benzoate salt of metronidazole, the dose should be about 60% higher (divide the dose by 0.6) than the metronidazole hydrochloride salt if dosing is based on the total ingredient (ie, 10 mg metronidazole hydrochloride = 16.7 mg metronidazole benzoate). Cefpodoxime is an orally bioavailable 3rd generation cephalosporin that is a pro-drug. Its spectrum includes gram negative (most Enterobacteriaceae but not *Pseudomonas*), and Gram positive but not anaerobic organisms. As with many cephalosporins, it is relatively resistant to beta-lactamases destruction. However, it is susceptible to extended-spectrum beta-lactamases which may not be evident with culture data (if present, these paraticular beta-lactamases do not target Clavamox®). Most of the newer drugs are characterized by a narrow spectrum and specifically address organisms for which resistance has developed (eg, vancomycin-resistant...
Staphylococcus or Enterococcus). Included are members of the streptogranins (eg quinupristin; virginiamycin has been used as a growth promotent in food animals), the oxazolidinones (eg, linezolid), a novel ribosomal inhibitor, telithromycin (a synthetic marolide) and newer fluorinated quinolones. Because of the narrow spectrum of the drugs, inconvenient route of administration, and for selected drugs, safety concerns, the use of these drugs has not been addressed yet in veterinary medicine.

**Voriconazole** a synthetic derivative of fluconazole, is the first of the “second generation” triazole compounds to be approved by the FDA. Compared to fluconazole, voriconazole contains a fluorine molecule and a methyl group which greatly enhances its spectrum compared to fluconazol. As with other azoles, its spectrum includes a variety of infecting fungal organisms. Voriconazole also is effective against *C. neoformans, Trichosporon beigelii* and *Saccharomyces cerevisiae*. Killing studies with *Aspergillus* demonstrate that, in general, amphotericin B is less efficacious than voriconazole. Pharmacokinetics of voriconazole apparently have not been performed in either the dog in the cat, but in humans, are complicated by a marked variable half-life and drug interactions typical of ketoconazole (ie, inhibition of drug metabolizing enzymes). Because this drug is associated with both adverse reactions (including visual defects, hepatotoxicity), care must be taken with extrapolating drug metabolizing enzymes. The long halflife (6 to 24 hr depending on the dose in humans) will require 5-6 days of dosing; a loading dose consisting of a double daily dose is recommended for the first day of therapy (the drug is dosed at 3 to 6 mg/kg twice daily to yeiled 3 to 6 µg/ml in the plasma). Extrapolations to animals must be made cautiously because the drug undergoes non-linear pharmacokinetics in humans, probably due to saturation of drug metabolizing enzymes. The drug is cleared primarily by hepatic metabolism to inactive metabolites by CYP 2C19 (the primary isoenzyme), 2C9, and 3A4 being involved. Selected humans are considered “poor metabolizers” of the drug because of variation in CYP 2C19. The drug has been used apparently successfully by the author in combination therapy with terbinafine to treat a retrobulbar opportunistic fungal disorder in a dog. **Terbinafine** (20-40 mg/kg once daily) is an allylamine antifungal that inhibits squalene epoxidase and ultimately formation of ergosterol. It is particularly efficacious against dermatophytes, but is also efficacious against *Sporothrix schenckii* and *Apergillus*. Because of its different mechanism of action, it might be used in combination with other antifungals effective against these two organisms. Recent evidence suggests that it also may be efficacious against histoplasmosis; combination with traditional antifungals should be considered for such therapies, including treatment of coccidiodomycosis. The role of **lufeneronm**, a benzoylphenylurea that inhibits synthesis and deposition of chitin (a flea preventative) for treatment of fungal disorders is not clear. Recently, its use as an immunomodulator (5 mg/kg [D] and 15 mg/kg [cat] once daily) has been suggested.

**Interpreting MIC: From Benchtop to Bedside**

Dawn Merton Boothe, DACVIM, DACVCP,Auburn University,
Auburn, Alabama, USA

**Introduction**

As in human medicine, the advent of resistance has and is increasingly impacting successful therapy in veterinary patients. In human medicine, antibiotic stewardship has become the focus for reducing resistance. Prudent veterinarian and veterinary practices will implement decision making processes (antimicrobial use paradigms) that minimize the temptation to use
antimicrobials as alternative therapies. The age of designing a dosing regimen based on cost and convenience rather than pharmacodynamics and pharmacokinetics must end. Antimicrobial stewardship begins by recognizing the problems and issues leading to antimicrobial resistance, and successfully implementing procedures that reasonably minimize the impact of antimicrobial use in the patient while not forfeiting the likelihood of therapeutic success.

**Advent Antimicrobial Resistance**

Development of antimicrobial resistance is facilitated by several factors; among the most important is exposure to antibiotics; indeed, the most important risk factor for fluoroquinolone resistance is probably previous fluoroquinolone use. Environmental microbes maintain an ecological niche through suppression of the competition by section of antibiotics. As such, commensal organisms are constantly being exposed to antibiotics. However, the microbe producing the antibiotic, as well as surrounding normal flora, are resistant to the antibiotic. Thus, genes for resistance develop along with genes directing antibiotic production and microbes among the normal flora are “primed” to develop resistance. Alteration of the environment – including the use of antimicrobials that alter the anaerobic population – disrupts this balance, increasing the risk of infection. Rapid microbial turnover in the gastrointestinal tract supports the development of resistance by assuring active DNA and thus mutation potential. Microflora of the GI tract can serve as reservoir of resistance genes; a single drug, via integrons, plasmids and transposons facilitate the rapid transfer of multiple drug resistance among organisms. Transposons move resistance genes back and forth between chromosomes to plasmids. As such, bacterial resistance is extremely mobile, and can spread rapidly. Narrowing the spectrum of the chosen antibiotic will help limit, but is not likely to prevent, the development of resistance. The use of synthetic antimicrobials is less likely to induce resistance, perhaps because there is less risk of previous exposure of the flora to these drugs.

**The Avoidance of Antimicrobial Resistance**

Avoidance of antimicrobial resistance is an overwhelming consideration and will require a different paradigm than has driven antimicrobial use in the past. Probably the single most important first step in judicious antimicrobial use is confirming the need. This is no small task, being fraught with the lack of effective diagnostic aids. Probably the most common–and least correct mindset is that our directive of “above all else do no harm” is being followed with the use of antimicrobials in the absence of infection. It is paramount that actions be taken that stay the hand. Once the decision to use the antimicrobial is made, efforts should focus on assuring that concentrations adequate to kill the infecting microbe are achieved at the site of infection. Thus, the second step in antimicrobial is identifying the susceptible target and the third, the site of infection, that is, the site to which the drug must be distributed. Historically, empirical therapy has been based on the most likely organisms to infect the site of infection, and the subsequent (presumed) response of the microorganisms to first choice antimicrobials. However, the historical data upon which these choices are base, quite frankly, is lacking in scientific scrutiny. More to the point, as has been addressed, microbes will not sit idly by when faced with exposure to antimicrobials and will develop mechanisms of resistance.

Antimicrobial therapy is most likely to be successful when the target (and thus spectrum of antimicrobial activity) is known, thus allowing a marriage of the pharmacodynamics of the organisms – susceptibility data – to the pharmacokinetics of the drug in the (healthy or otherwise) patient’s. Among the guides for antimicrobial therapy is Culture and Susceptibility testing. Despite its pitfalls (to be discussed), C&S is an important - albeit one of several- guides to antimicrobial selection. Cultures always should be collected for serious (life or organ threatening) infections. For serious /life threatening infections, cultures should be collected prior to initiation of antimicrobial therapy. Other indications for culture include but are not limited to chronic infections, recurrent
infections, a history of previous antimicrobial therapy and non-responsive infections. The C&S data is most useful if the clinician compares what is needed to be effective against the microbe (pharmacodynamics) with what is achieved in the patient when using the chosen drug and dosing regimen. C&S not only identifies a potential pathogen, but also narrows the selection of drugs, offering support for selection of a less toxic or (with care) less expensive drug. Additionally, C&S can identify resistant organisms, and if tube dilution data is available, organisms that have undergone first step resistance although still "susceptible". Finally, C&S can be used to design a dosing regimen specific to the individual patients.

Pharmocodynamic / Susceptibility Data: What you need.

Simplistically, susceptibility data represents "what is needed" in the patient to facilitate antimicrobial efficacy. Care must be taken with this simplistic approach: susceptibility data is generated from in vitro methodologies, yet it is applied to in vivo (and abnormal) conditions. As long as this caveat is foremost in the clinicians mind, the data can be useful to antimicrobial selection. Package inserts for newer antimicrobials include susceptibility data and as such, offer a means of understanding the role of susceptibility data in antimicrobial selection. Microbiological data for labels generally includes data generated both from agar gel disc diffusion (eg, Kirby Bauer: zone diameters) as well as tube dilution (MIC) methods of susceptibility testing. However, the presentation of the "numbers" differ because the methodology differs.

The disk diffusion method (e.g., Kirby Bauer) uses disks impregnated with a known amount of drug that diffuses into the agar at a known rate. The agar is inoculated with a standardized number of organisms. The concentration of the drug decreases with the distance (zone) diameter from the disk. Microbial growth that is inhibited by the drug will result in a zone of no growth (zone of inhibition, diameter, in millimeters) that surrounds the disk. Antimicrobial growth that is inhibited at a large diameter indicates a susceptible organism because it is likely that the drug concentration in the agar that inhibited growth is low enough that it can be achieved safely in the patient. Growth inhibited only in close proximity to the disk indicates the need for a higher drug concentration in the patient. If it is unlikely that this concentration can be reached in the patient at the recommended dose, an "R" is designated for the drug. The zone diameters at which an organism is considered resistant is established by the National Committee for Clinical Laboratory Standards (NCCLS: recently renamed to Committee on Laboratory Standards International: CLASI) and generally can be correlated with an MIC of the drug (see tube dilution method). The disk diffusion method thus might be considered a qualitative method in that drug concentrations achieved in the agar surrounding the disk can be correlated roughly with concentrations achieved in the patient serum, but only "SIR" indicators are provided.. Disc diffusion data on package inserts will include zone diameters indicative of organism susceptibility (S), intermediate (I), or resistance (R) (SIR). For example, current zone diameters for enrofloxacin at a dose of 2.5 mg/kg are ≥ 20 mm = S (susceptible), ≤ 16 = R (resistant), and between 17-19 mm = I (intermediate).

In contrast to disk diffusion, tube dilution methods involve inoculation of a series of test tubes with a standard number of organisms. The test tubes contain increasing concentrations of the drug of interest in two-fold dilutions (eg, 0.125, 0.25, 0.5, 1, 2, 4, 8 µg/ml etc) (Figure 1). Generally and ideally, the concentrations to be tested include the breakpoint MIC (MIC<sub>BP</sub>; see below) and at least several fold dilutions of this MIC. Note, however, that some laboratories do not test at concentrations below the MIC<sub>BP</sub>. Following a standard time, the tubes are evaluated for detectable growth. The test tube that contains the lowest concentration of drug and no visible growth contains the minimum amount of drug necessary to inhibit (not kill) the growth of the organism cultured from the patient (the MIC). Ideally, this concentration must be achieved at the site of infection. Adaptation to computerized/automated systems allow much more accurate testing in short time
For either method of susceptibility testing, simplistically, the likelihood of a drug being effective in the patient is based on whether or not the recommended dose on the label is likely to generate plasma drug concentrations (PDC) that equal or surpass the MIC of the infecting organism.

Diagnostic laboratories indicate the likelihood of susceptibility by the “SIR” letter designation. Understanding the basis of that designation will facilitate antimicrobial selection. The SIR designation reflects whether or not the MIC of the infecting organism is less than (“S”), close or equal to (“I”) or greater than (“R”) the breakpoint MIC (MIC$_{BP}$) of the drug. Breakpoint MICs (as are interpretive zone diameters of agar gel diffusion) should be determined by the CLASI and are the basis for determining the resistance or susceptibility of an organism to a drug. The MIC$_{BP}$ is a characteristic of the drug, not the microbe, and as such, should be the same for all laboratories. The MIC$_{BP}$ is determined by at least three criteria: the pharmacokinetics of the drug, particularly peak PDC (maximum concentration; Cmax;) that will be achieved in the targeted patient population, using the recommended dose of the drug (which should surpass MIC$_{BP}$); susceptibility (MIC) of the organisms which are included in the spectrum of the drug (eg, the MIC 90, the MIC at or below which 90% of the organisms tested were susceptible; this should be below the MIC$_{BP}$); and clinical response to the drug as observed in a large number of patients. In Figure 2, the MIC$_{BP}$ for each drug is noted in parenthesis to the right of the drug. If the MIC of the organism (noted for each drug for two organisms in Figure 2) is sufficiently lower than the MIC$_{BP}$, the organism is considered susceptible (S). If the MIC of the organism equals or surpasses the MIC$_{BP}$, the organism is considered resistant (R). Some laboratories also offer an “I” or “MS” (medium susceptibility), indicating that the MIC of the organisms is approaching (generally, within 1 tube dilution) the MIC$_{BP}$. Occasionally, for some drugs, more than one MIC$_{BP}$ will be offered because selected species of organisms are more likely to be resistant. For example, it is not unusual for interpretive standards of beta lactams to be somewhat more restrictive (ie, the MIC$_{BP}$ is lower) for organisms which produce beta-lactamases. Beta lactamases destroy the drug, so the concentration at the site is actually much lower than it would be for non-beta lactamase producers. Hence, the breakpoint MIC is much lower for Staph compared to others. Examples can be seen in Table 1 for amoxicillin / clavulanic acid. *Staphylococcus* is considered resistant at a lower MIC compared to gram negative organisms, indicating it is much more likely for those organisms to receive an "R" than and "S" designation. The difference is much more dramatic for ampicillin because it is much more susceptible to beta-lactamase destruction compared to amoxicillin with clavulanic acid. Note that some literature also provides a “susceptible” and a “resistant” MIC$_{BP}$. However, the two breakpoints are the same. For example, in Table 1, for amikacin, an organism is considered susceptible if the MIC is $\leq 32$ µg/ml (the “susceptible” MIC$_{BP}$) but resistant if the MIC $\geq 64$ µg/ml (the “resistant” MIC$_{BP}$). However, there is no number between 32 and 64 µg/ml. For amoxicillin/clavulanic acid, an MIC $\leq 8$ µg/ml is considered susceptible (for *Staph*, $\leq 4$), but $\geq 32$ µg/ml (≥ 8 for *Staph*) is considered resistant. Organisms with an MIC of 16 µg/ml (the tube dilution in between 8 and 32) would be designated as an “I” or “MS” and extreme precaution should be taken when using the drug.

Clinicians should be aware of the method of reporting microbial response for C&S reports for their laboratory. Some reports of susceptibility data provide an MIC of $< X$ (where X = a drug concentration; see $< 2$ µg/ml for amikacin for *Staph intermedius* in Figure 2). For such organisms, no growth occurred in the lowest concentration tested by the laboratory, which generally (and hopefully), but not always, is several fold dilutions below the MIC$_{BP}$. An MIC of $> X$ is accompanied by an “R” because the organism was not susceptible to the highest concentration tested (which generally is the MIC$_{BP}$). Prudent clinicians should be aware of the MIC$_{BP}$ for drugs selected for treatment. To complicate interpretation further, some laboratories provide an interpretive “code” rather than MIC data that indicates a level of susceptibility of the culture organism. Provision of
data which indicates the distance of the infecting organisms MIC from $\text{MIC}_{\text{BP}}$ is particularly useful to the clinician in order to narrow the choice of “susceptible” drugs and to design the most appropriate dosing regimen for the patient.

The CLASI updates $\text{MIC}_{\text{BP}}$ particularly as new data is provided regarding organism susceptibility. Increasing resistance to organisms may lead to changes in the $\text{MIC}_{\text{BP}}$; ideally, doses of antimicrobials to which resistance is developing also should change, but unless a drug company is willing to subject the drug to approval (required for label changes) at the new dose, changes in labeled doses are not likely to occur. For older antibiotics approved decades ago, originally labeled doses may be inappropriate for all except very sensitive organisms. Enrofloxacin offers an example of how the CLASI might change an $\text{MIC}_{\text{BP}}$. Enrofloxacin was originally approved at 2.5 mg/kg. Peak drug concentrations at that dose are approximately 1.2 $\mu$g/ml. The CLASI established the $\text{MIC}_{\text{BP}}$ at 2 $\mu$g/ml. Thus, any organisms whose MIC was less than 2 $\mu$g/ml was considered susceptible; any organism with an MIC of 2 $\mu$g/ml or more was considered resistant. However, this concentration was probably too low for many organisms considered susceptible to enrofloxacin. Bayer Animal Health successfully sought a label with a higher dose that ranges from 5 to 20 mg/kg. The highest dose of 20 mg/kg yields PDC of approximately 4 $\mu$g/ml. Accordingly, Bayer sought and the CLASI established a new $\text{MIC}_{\text{BP}}$: "S" = MIC $\leq$ 1 $\mu$g/ml, "R" = $\geq$4 $\mu$g/ml, and "F" (or "I") indicates an MIC between 1 and 4 $\mu$g/ml (new zone diameters for agar gel diffusion also were established). The “F” indicates “flexibility” in the selection of a dose. For the clinicians, based on MIC data alone, an S might indicate the lower dose should be considered, and an F, the mid to higher dose, depending on the proximity of the MIC of the infecting organism to the $\text{MIC}_{\text{BP}}$. The original $\text{MIC}_{\text{BP}}$, which many laboratories still use, does not reflect the higher dose and organisms identified as "R" might actually be susceptible at the higher dose. Veterinary microbiology laboratories should be encouraged by practitioners to implement changes in interpretive susceptibility data which reflect the flexibility in dosing regimens.

Tube dilution susceptibility will be presented on package inserts as minimum inhibitory concentrations (MIC). The MIC of an organism for a drug refers to the minimum amount of drug necessary to inhibit visible growth of an organism using standardized culturing methods as guided by the CLASI which publishes guidelines for the methods of susceptibility testing as well as the interpretive criteria for each drug. The MIC data on a label may include: 1. the range of MIC for susceptible organisms; 2. the mode of MIC (the most frequently cited MIC); 3. or the $\text{MIC}_{50}$ and the $\text{MIC}_{90}$. The data are population statistics; the latter two reflect, respectively, the MIC below which 50% and 90% of the isolates (by genus and species) are inhibited (not killed). However, the $\text{MIC}_{50}$ and $\text{MIC}_{90}$ should be based on a large number of microorganisms to assure accurate sample representation of the population (ideally >300).

The Epsilon test (“E-test”) represents a mix of agar gel diffusion and tube dilution. In this system, a several cm strip containing increasing concentrations of drug is applied to a plate inoculated with the infecting organism. The zone of inhibition (tear drop shape) surrounding the strip indicates the MIC of the infecting organism. Although expensive (one drug per plate; E-strips are expensive), an advantage of this system is that the range of MIC’s is much larger than that offered by tube dilution. The author has used this system for drugs not included in automated assays and to identify the MIC of organisms noted as "R" using tube dilution when the infection is at a site where drug is or can be concentrated.

The different information provided by agar gel versus tube dilution methods may impact drug selection. For disc diffusion data, (and the E-test), the application to the clinical patient requires more hurdles than the application of MIC data in that zone diameters of disc diffusion correlate with MIC data.

**Pharmacokinetic Data: What you get**
The selection of an antimicrobial should be based on the likelihood that therapeutic (effective) concentrations will be achieved at the tissue site. What is needed for therapeutic efficacy for infections is determined largely by the susceptibility (pharmacodynamic data) of the organism. For populations of microbes, the MIC\textsubscript{90} provides an indication of what is needed; for organisms cultured from patients, the MIC as provided in C&S data is an indication of what is needed. Efficacy of an antibiotic is most likely to occur when the pharmacodynamic data is coupled with what is achieved in the patient. This in turn is best defined in pharmacokinetic data of the drug in the target species. Again, the availability on package inserts of pharmacokinetic data describing the disposition of drug in the target species can be a powerful tool for guiding design of a dosing regimen. For laboratories which provide MIC testing, the coupling of pharmacodynamic and pharmacokinetic data has already begun through provision of an S,I or R designation. However, the clinician can use the coupling of this information further to narrow the selection of appropriate antimicrobials and to design the dosing regimen.

C\text{max} or peak plasma drug concentration is generally determined following administration of a single (maximum) labeled dose using the labeled route in the target species. The value of this parameter is one of the 3 criteria for CLASI determination of the MIC\text{BP} of the drug. For the clinician seeking to improve antimicrobial efficacy, the further the MIC of the infecting organism is from the MIC\text{BP} of the drug, the more likely effective concentrations will be reached at the site of infection. If a number of drugs are designated as “S”, the selection might be narrowed by focusing on those drugs for which the MIC is furthest from the MIC\text{BP}. Some labs do not provide the MIC\text{BP} on their report. As noted before, a < X \mu g/ml designation indicates that the drug was susceptible at a concentration lower than the lowest concentration tested, indicating excellent susceptibility (/i/) the laboratory tests at concentrations several fold lower than the MIC\text{BP}. Alternatively, if the lab uses a numbering or lettering system, those drugs designated as having the lowest MIC might be the initial focus of selection. Finally, the MIC\text{BP} of many drugs might be found on package inserts (including the Physician’s Desk Reference or Veterinary Products and Biologics) or in the literature (as is provided in Table 1).

C&S data often reflects drugs not commonly used in animals. This reflects, in part, the availability of automated systems developed in human medicine because of economic considerations. Even for human drugs, cost of testing precludes generation of C&S data for all potential drugs of interest. Often, a single drug is tested as a representative of a class or subclass of drugs. Thus, ampicillin often serves as a model for amoxicillin, cephalothin for first generation cephalosporins, etc. Often, data of the representative drug fairly reflects that of similar drugs in the class. This is true for veterinary fluorinated quinolones. Enrofloxacin often serves as the model drug (as the first to be approved in the US). In general, bugs susceptible to enrofloxacin will be susceptible to other veterinary FQs and those resistant to one will be resistant to all. However, as seen in Figure 2, gentamicin could not serve as the model drug for amikacin (for both antibiotics). Third generation cephalosporins can not reflect susceptibility data for all third generation cephalosporins.

For many drugs, C&S testing simply is not available because the manufacturer has not submitted data to CLASI for determination of interpretive standards. This is particularly true for animal drugs, again, probably for economic reasons. Indeed, for drugs approved for use in both humans and animals, the interpretive standards used by a veterinary diagnostic laboratory may be those generated for human drug therapy, not animal drug therapy. Differences in C\text{max} (or in the pathology/physiology of the infecting organisms) may result in less clinical application in animals compared to humans. Clearly, susceptibility data for drugs for which no approved version exists in (non-human) animals is based on interpretive standards in humans. Finally, some laboratories provide only agar gel diffusion data. For such cases, data reported in the literature can be helpful in
selecting both a drug and a dose. Assuming the organism is known, the MIC$_{90}$ can be used as an alternative to MIC of the organism in the patient; and the reported Cmax of a pharmacokinetic study can be used in lieu of the MIC$_{BP}$. Table 1 provides Cmax or peak PDC at specific doses for selected antimicrobial drugs in dogs or cats when using the dose accompanying the Cmax. As expected, this approach has limitations. For example, using the MIC$_{90}$ will probably underestimate the efficacy of the drug because chances are the MIC of the infecting organism is below the MIC$_{90}$. The MIC$_{50}$ or, even better, MIC mode (the MIC most frequently reported) might be a more clinically relevant MIC to which the Cmax could be compared when considering in vitro susceptibility of an organism to a drug. However, using the MIC$_{90}$ will allow a conservative approach to antimicrobial selection.

**Designing a Dosing Regimen**

For selection of a drug for which multiple drugs are noted as “S”, the MIC of the infecting organism can be compared to the reported MIC$_{BP}$ to assess “relative” susceptibility. Those drugs for which the distance between the two parameters is greatest might be the initial focus of drug selection. Note that comparisons can not be made by comparing MIC of different drugs. Just as drug concentrations vary in the patient, MIC concentrations also will vary. Drug susceptibility must be compared using relative susceptibility: the distance of the MIC of the infecting organism from the MIC$_{BP}$ of the drug of interest. For example, in Figure 2, if C&S data were to be the only criteria for selection, amikacin might be the best choice to treat *Staph* since its MIC is 1/32 of the MIC$_{BP}$. Enrofloxacin might be a close second. Clearly, other considerations (especially ability to penetrate tissue) should narrow the initial list of susceptible drugs further and might, for some infections, be the initial basis for selection.

Once the drug has been selected, MIC data also can be used to design the dosing regimen, and specifically, in selecting a dose. A review of the package inserts of the veterinary FQs can exemplify this point. A review of the MIC$_{90}$ of the various organisms for these drugs reveals considerable differences in susceptibility among the organisms (although MIC for the drugs are often very similar). For example, *Pastuerella multocida* generally is characterized by very low MICs (eg, 0.06 µg/ml), whereas *Pseudomonas aeruginosa* generally is characterized by higher MICs (eg, 1 µg/ml) for the FQs. Intuitively, when treating the former organism with an FQ, a lower dose is more likely to be effective. However, when treating the latter organism, a higher dose generally should be selected. The same approach can be used when using the MIC of an infecting organism provided by a diagnostic laboratory. The closer the MIC of the infecting organism is to the MIC$_{BP}$, the more likely the maximum dose (or higher) should be used. Although less ideal, the same approach can be used when basing dose selection on agar gel diffusion data or when the organism is suspected but not known. For disc diffusion data, only "S", "I" and "R" designations are given; no evidence of relative susceptibility is provided. However, the MIC$_{90}$ data for the infecting organism on the PFL can help guide initial selection of a dosing regimen when only disc diffusion data is available. An “I” (or an MIC that approximates the MIC$_{BP}$) indicates the need for a higher dose, unless the infection is located at a site in which the drug is concentrated (urinary tract infection) or the drug can be applied topically. Indeed, for such infections, C&S may markedly underestimate efficacy because the MIC$_{BP}$ is based on drug concentrations achieved in plasma and does not reflect concentration of the drug.

A dose can also be calculated based on the MIC of the drug, as long as the VD of the drug is known. For example, for treatment of *Pseudomonas aeruginosa* with enrofloxacin (VD = 2.7 L/kg), a dose can be calculated based on an MIC$_{90}$ of 1 µg/ml (1 mg/L): Dose = 2.7 L/kg * 1 mg/L = 2.7 mg/kg. However, this dose will simply achieve the MIC in plasma and may not be sufficient for efficacy, particularly for concentration dependent drugs.

**Conclusion:** Culture and susceptibility testing is one of several tools that can be used to
guide antimicrobial therapy in the individual patient. As microbes increasingly are becoming resistant to drugs, the importance of data will similarly increase. To be most useful, however, MIC data is paramount. Culture and susceptibility testing is particularly important in patients with a history of previous antimicrobial therapy. However, such data is predicated on the assumption that the MIC is achieved at the site of infection, but is based on plasma drug concentrations. The application of the in vitro data to the patient should be based on microbial, host and drug factors.

Table 1. Interpretive criteria for selected antimicrobial drugs

<table>
<thead>
<tr>
<th>Drug</th>
<th>S</th>
<th>R</th>
<th>icol</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>*Amikacin</td>
<td>&lt; 16</td>
<td>&gt; 64</td>
<td>Ciprofloxacin (&lt; 1</td>
<td>8)</td>
</tr>
<tr>
<td>Amoxicillin</td>
<td>&lt; 0.5</td>
<td>32</td>
<td>Clarithromycin ≤ 8</td>
<td>32</td>
</tr>
<tr>
<td>*Amoxicillin with clavulanic acid</td>
<td>&lt; 8/4</td>
<td>32/16</td>
<td>Clindamycin ≤ 4</td>
<td>&gt; 16</td>
</tr>
<tr>
<td>*Ampicillin</td>
<td>&lt; 0.5</td>
<td>5</td>
<td>Difloxacin 0.5</td>
<td>4</td>
</tr>
<tr>
<td>*Ampicillin with clavulanic acid</td>
<td>&lt; 8/4</td>
<td>32/16</td>
<td>Doxycycline &lt; 4</td>
<td>&gt; 16</td>
</tr>
<tr>
<td>Azithromycin</td>
<td>≤ 8</td>
<td>&gt; 8</td>
<td>Erythromycin &lt; 0.5</td>
<td>&gt; 4</td>
</tr>
<tr>
<td>*Cefazolin</td>
<td>≤ 8</td>
<td>32</td>
<td>Gentamicin* ≤ 4</td>
<td>&gt; 16</td>
</tr>
<tr>
<td>Cefotaxime</td>
<td>≤ 8</td>
<td>&gt; 64</td>
<td>*Imipenem/cilastin</td>
<td>≤ 4</td>
</tr>
<tr>
<td>Cefoxitin</td>
<td>≤ 8</td>
<td>≥ 32</td>
<td>Fluorfenicol</td>
<td>2</td>
</tr>
<tr>
<td>Cefpodoxime</td>
<td>≤ 2</td>
<td>&gt; 8</td>
<td>Kanamycin* ≤ 16</td>
<td>&gt; 64</td>
</tr>
<tr>
<td>Ceftazidime</td>
<td>32</td>
<td>Lincomycin</td>
<td>&lt; 1</td>
<td>&gt; 8</td>
</tr>
<tr>
<td>*Ceftiofur</td>
<td>≤ 2</td>
<td>&gt; 8</td>
<td>Marbofloxacin</td>
<td>2</td>
</tr>
<tr>
<td>Ceftizoxime</td>
<td>≥ 32</td>
<td>0.5</td>
<td>Meropenem</td>
<td>≤ 8</td>
</tr>
<tr>
<td>Ceftriaxone</td>
<td>≤ 8</td>
<td>&gt; 32</td>
<td>Metronidazole</td>
<td>≤ 8</td>
</tr>
<tr>
<td>Cefuroxime</td>
<td>≤ 8</td>
<td>&gt; 32</td>
<td>Nitrofurantoin</td>
<td>&lt; 32</td>
</tr>
<tr>
<td>*Cephalothin</td>
<td>≤ 8</td>
<td>&gt; 32</td>
<td>Orbachloxacin</td>
<td>&lt; 1</td>
</tr>
<tr>
<td>*Chloramphen</td>
<td>≤ 8</td>
<td>&gt; 32</td>
<td>Oxacillin</td>
<td>&lt; 2</td>
</tr>
</tbody>
</table>

* Interpretive criteria in animals
1 For Staphylococcus sp
2 For other organisms

Figure 1. A diagram of tube dilution methodology
Figure 2. An example of C&S data

To MIC and Beyond: Minimizing Antimicrobial Resistance
(Or “Dead Bugs Don’t Mutate)

Dawn Merton Boothe DVM, PhD, DACVIM, DACVCP
Auburn University, AL boothdm@auburn.edu
INTRODUCTION

"Even experienced practitioners may not realize that giving a patient antibiotics affects not just that patient but also the other people that come into contact with that environment". With that statement Dancer (2001) summarized antimicrobial therapy in veterinary (and human) medicine. The National Foundation for Infectious Disease estimated the cost of treating resistant bacteria to be as high as $4.5 billion annually; and to be responsible for over 19,000 (human) deaths per year. This important issue should be no surprise. In the United States alone, approximately 350 million pounds of antibiotics are used in veterinary medicine; 40% of this is "unnecessary" according to the human health sector.

In addition to the impact in human medicine, the advent of resistance has and is increasingly impacting veterinary patients. In human medicine, antibiotic stewardship has become the focus for reducing resistance. Prudent veterinarians implement decision making processes ( antimicrobial use paradigms) that minimize the temptation to use antimicrobials indiscriminately. The advent of designing a dosing regimen based on cost and convenience rather than pharmacodynamics and pharmacokinetics begins by recognizing the problems and issues leading to antimicrobial resistance, and successfully and reasonably minimize the impact of antibiotic use in the patient while not forfeiting the likelihood of therapeutic success.

The most important considerations for selection of an antimicrobial are, in order of priority: 1. confirming infection (indiscriminantly); 2. identifying the target organisms; and 3. identifying the target site. The 4th through nth step should take into account the host, and microbial factors that can negatively impact the selection of the dosing regimen. Among the more relevant considerations are identifying mechanisms of resistance, the likelihood of a site-related infection, the site, and time- versus concentration- dependency as it relates to convenience. Toxicity (because these drugs are usually given chronically) and cost is less important. Cost should be the last consideration that influences selection. Selection should strive for treatment success, just as failing to use C&S as a basis for selection (or selecting a drug characterized by "R" on the data) implies that approximately 90% of infections treated based on C&S are likely to respond if an "S" drug is selected, even if an "R" drug is selected. The most likely situations where the latter is true is if the infection is at a site in which a much higher than the minimum inhibitory concentration (MIC).

PITFALLS OF CULTURE AND SUSCEPTIBILITY TESTING: IN VITRO CONSIDERATIONS

Although culture and susceptibility data (C&S) can be a powerful tool to guide selection, it nonetheless conditions; over-reliance on the information can contribute to therapeutic failure. As early as the collection procedures beyond the scope of this manuscript to delineate proper techniques of specimen culture collection, but C&S data collection; close adherence to recommended procedures including but not limited to site selection, site preparation, and proper interpretation. Anaerobic infections are particularly problematic. Obligate anaerobes are exquisitely sensitive to oxygen. No growth may be mistakenly interpreted as lack of anaerobic infection. More anaerobes, capable of growth in anaerobic environments. Aerobic cultures may yield their growth, but the anaerobes often grow slowly limiting response to antimicrobials (particularly aminoglycosides). Organisms without cell walls (Mycoplasma, L-forms of bacteria, selected Gram positives, Nocardia, atypical Mycobacteria and others are examples)

Just as absence of growth does not indicate absence of infection, isolation of an organism is not necessarily evidence of infection. The C&S procedures themselves are fraught with potential errors. For practices that provide in-house susceptibility testing, it is important to follow guidelines established and published by (or comparable to) the Clinical and Laboratory Standards Institute.

Pitfalls of susceptibility testing also reflect the drugs selected for testing. Not all companies are interested in every drug exception is less important. Cost should be the last consideration that influences selection. Selection should strive for treatment success, just as failing to use C&S as a basis for selection (or selecting a drug characterized by "R" on the data) implies that approximately 90% of infections treated based on C&S are likely to respond if an "S" drug is selected, even if an "R" drug is selected. The most likely situations where the latter is true is if the infection is at a site in which a much higher than the minimum inhibitory concentration (MIC).
potential drugs used to treat an infection, one drug often is tested as a model for other drugs in the class. (ie, cephalosporins; enrofloxacin for the fluorinated quinolones). For some classes of drugs, cross-reactivity can be high. Animals that are resistant to one drug often are resistant to many other drugs in the same class. The organism that is resistant to one drug also is likely to be resistant to another drug in the same class. However, the organisms generally are not susceptible to these enzymes. Laboratories currently are generating methods intended to detect resistance that has developed in the infecting organism to drug to which the organism is generally susceptible. Another example of concern: they are susceptible to extended spectrum beta-lactamase (ESBL) that will be present. Despite the presence of ESBL, therapeutic failure may occur. Cefazolin is often used as a test for the presence of ESBL.

BRIDGING PHARMACOKINETICS AND PHARMACODYNAMICS

Interpreting C&S (pharmacodynamic) data is most helpful when considered in the context of the clinical situation. The MIC of the isolate for the drug of interest – to what is achieved. Even if sample C&S data is inherently deficient because in vitro methods cannot mimic in vivo conditions. For example, the in vitro concentration of drug throughout the in vitro incubation period; in vitro methods can not take into account active microorganisms. Among the most problematic concerns is interpretation of MIC. The breakpoint MIC (MICp) is equivalent to or less for many organisms. Susceptibility data also does not take into account active microorganisms. If recommended doses change, the manufacturer should provide CLASI with updated pharmacokinetic data. MICp criteria may change accordingly, and automated systems should incorporate those changes in their methods. Application of appropriate circumstances (eg, topical therapy) a drug designated as “R” might actually be effective. The more difficult to achieve the anticipated PDC at the site of infection. Most infections occur in extracellular spaces (ie, skin, brain, testicles, cartilage) will be difficult to penetrate. In humans, recommended doses of beta-lactams drugs (Vd generally ≥ 0.6 L/kg) may be more likely to distribute to target effective concentrations. Certainly tissues characterized by non-fenestrated capillaries, antimicrobials can move unimpeded from plasma into ECF. However, water soluble drugs (volume of distribution [Vd] generally ≥ 0.6 l/kg) may be more likely to distribute to target effective concentrations. Certain tissues characterized by non-fenestrated capillaries (eg, skin, brain, testicles, cartilage) will be difficult to penetrate. In humans, recommended doses of beta-lactams drugs (Vd generally ≥ 0.6 L/kg) may be more likely to distribute to target effective concentrations. Antimicrobials that describe how much tissue dilutes a drug, but provides no indication as to where the drug distributes. Inflammation, use of a drug that accumulates in phagocytes (eg FQs, macrolides, lincosamides) is likely to increase PDC.

Bactericidal versus bacteriostatic drugs: The term “bactericidal” is somewhat abused: clinicians understand it to mean that the drug to kill rather than simply inhibit an organism will increase the time to complete killing. This may be true, but only if concentrations of the drug achieved at the site of infection are sufficient to kill the microbe. The definition and based on killing rates (eg, 99.9% reduction in bacterial inoculum within a 24 hr period of exposure).
minimum bactericidal concentration (MBC) of a drug to the MIC. The MBC is determined following tube dilution where growth are inoculated on agar gel. If no organism grows on the agar, the organisms were killed in the test tube. The concentration of drug that yields no growth on the agar gel contains the MBC of drug. For drugs considered “ba
terially important that occur for several tube dilutions above the MIC, indicating that organisms were not killed. However, bacteriostatic organisms are exquisitely sensitive to the effects of selected drugs; some “static” drugs are accumulated to concentrations [eg, macrolides and lincosamides in phagocytes; urine concentration]. However, killing concentrations are generally higher for bactericidal drugs compared to a static concentration. On the other hand, a “cidal” drug may not kill if concentrations are not sufficient to preclude its actions (eg, slow growth in an anerobic environment; combination with growth inhibitors). Thus, killing concentrations (ie, MIC/MBC) are achieved at the site. The bactericidal nature of a drug often reflects its mechanism: ribosomes (eg, tetracyclines, macrolides, lincosamides, chloramphenical) often simply inhibit the growth of the organism, whereas drug concentration is necessary to kill the organism, in vitro; the MIC is distant from the MBC. Clinically, host defense and the natural history of the infection play a critical role in the use of these drugs. Dosing regimens for following treatment with these drugs unless exceptionally high concentrations (ie, the MBC) of these drugs are associated with adverse effects (eg, encephalopathy, nephrotoxicity, liver damage). In contrast, concentration dependent drugs are generally better tolerated. A number of investigators have examined several PK and pharmacodynamic (PD) indices as predictors of clinical antimicrobial efficacy. Amo
cipenem and ceftazidime for empirical therapy of severe infections (eg, meningitis, osteomyelitis; peritonitis, bacteremia/sepsis, many chronic infections).

### Relationship between MIC, Plasma and tissue Drug Concentrations

The bridging of pharmacokinetic and pharmacodynamic data can begin by comparing the MIC established by in vitro testing and the concentrations of drug at the site of infection when the drug is administered at the labeled dose. A number of investigators have examined several PK and PD indices as predictors of clinical antimicrobial efficacy. Among the most useful indices are the ratio of C\text{max} (peak PDC) to MIC, defined here as IQ; the area under the inhibitory curve (AUC) related to MIC; the percent time that PDC are above the MIC, and the relationship between MIC and the magnitude of plasma drug concentration (C\text{max} ) compared to the inhibitory quotient or IQ. For such drugs, the magnitude of the IQ generally should be 8-10 but ideally is higher than 10. For Pseudomonas aeruginosa, or infections caused by multiple organisms. The duration that PDC above the MIC are maintained, the efficacy may be enhanced by a drug-free period (i.e., a long interval between doses). For concentration dependent drugs, particularly detrimental. In a mouse model of E. coli gastrointestinal infections, the antibiotic efficacy of ciprofloxacin, but not clindamycin, is limited to infections in immune compromised animals (eg, viral infections [parvovirus, panleukopenia, FIV, FeLV], and in systems characterized by derangements in local immunity (ie, CNS infection for which an marked inflammatory response is not necessary)), or infections caused by multiple organisms. The duration that PDC above the MIC are maintained, the efficacy may be enhanced by a drug-free period (i.e., a long interval between doses). For concentration dependent drugs, particularly detrimental. In a mouse model of E. coli peritonitis, the antibacterial efficacy of ciprofloxacin, but not clindamycin, is limited to infections in immune compromised animals (eg, viral infections [parvovirus, panleukopenia, FIV, FeLV], and in systems characterized by derangements in local immunity (ie, CNS infection for which an marked inflammatory response is not necessary)).
three half-lives onto the dosing interval, the dose would have to be increased 8 fold. For example, for a Staphylococcus aureus with a peak concentration of 4 mcg/ml, assume PDC achieve the breakpoint MIC (32 mcg/ml) when a recommended dose (first dose) of 8 mcg/ml is given. After one half-life (1.5 hr), PDC have dropped to 16; in 3 hr, to 8 and in 4.5 hr, to 4 mcg/ml. Another 25% of the dose is eliminated during this time. The animal should be redosed. If the dose is doubled, this can be extended to 8 hours; the dose would have to be increased 8 fold to reach the standard 12 hrs. This is for an organism with a relatively low MIC; the problem is much more complex (compared to increasing the dose) of time dependent drugs; as such, efficacy of time dependent drugs may be enhanced for time dependent drugs which persist in selected tissues (again, assuming the MIC is surpassed). A more conservative strategy would be to accumulate in active (unbound) form in tissues (i.e., macrolides) or drugs that accumulate in phagocytes.

Postantibiotic Effect: The post antibiotic effect (PAE) describes the continued inhibition of microbial growth after the drug is removed from the site of infection. The impact of the PAE on antimicrobial efficacy can be profound, particularly for concentration-dependent drugs, and would be heightened if some of these drugs to be administered at long intervals. The PAE may be absent for some organisms or some time periods (e.g., immunocompromised patients). In general, concentration-dependent drugs appear to exhibit longer PAE; further, the PAE may be enhanced for time dependent drugs which persist in selected tissues (again, assuming the MIC is surpassed). As with concentration-dependent drugs, the PAE may be enhanced for time dependent drugs which persist in selected tissues (again, assuming the MIC is surpassed). A more conservative strategy would be to accumulate in active (unbound) form in tissues (i.e., macrolides) or drugs that accumulate in phagocytes.

Mutant Prevention Concentration. Based on natural mutational frequencies, a mutation that leads to a single step increase in MIC is not likely to be detected on C&S, yet, if the inoculum is large enough, they will be in the patient. Simply achieving a concentration that inhibit the growth of all isolates except those whose MIC exceed that reported, including the first step mutants. As immunocompromised the patient, the more likely resistant mutants will survive antimicrobial therapy administered to achieve the reported MIC. Subsequent growth of these mutant organisms will yield a population still considered “S” but with MIC that approach the breakpoint MIC. Adding a 3rd or 4th dose is a much more costly option (compared to increasing the dose) of time dependent drugs; as such, efficacy of time dependent drugs may be enhanced for time dependent drugs which persist in selected tissues (again, assuming the MIC is surpassed). A more conservative strategy would be to accumulate in active (unbound) form in tissues (i.e., macrolides) or drugs that accumulate in phagocytes.

Host Factors: The impact of host response to infection can be profound. Problems contributing to therapeutic failure (design a dosing regimen that will assure bactericidal concentrations of the chosen drug reach the site of infection if one dose is otherwise appropriate clean/drain accessible infections, select a drug that distributes into tissues well and ideally increase the dose appropriately). Even tissues traditionally considered “well perfused” might be of concern. For example, in bronchial secretions well, despite the fact that the lungs are well perfused. Amoxicillin is often used to treat respiratory infections of the amoxicillin that is in plasma is distributed to bronchial secretions. Theoretically, one must dose amoxicillin appropriately to assure bactericidal concentrations of the drug reach the site of infection should be sufficiently high to inhibit the growth of all isolates except those whose MIC exceed that reported. In general, time dependent drugs appear to exhibit longer PAE; further, the PAE may be enhanced for time dependent drugs which persist in selected tissues (again, assuming the MIC is surpassed). A more conservative strategy would be to accumulate in active (unbound) form in tissues (i.e., macrolides) or drugs that accumulate in phagocytes.
achieve targeted PDC in bronchial secretions. Most water soluble drugs (beta-lactams and aminoglycosides) reach bronchial secretions whereas over 50% of lipid soluble drugs reach bronchial secretions. Dosing adjustments also are needed for intracellular or complicated by host response to infection.

**Microbial Factors:** Materials released from microbes facilitate invasion, impair cellular phagocytosis, and staphylococci associated with canine pyoderma produce "slime," a material that facilitates bacterial adhesion to tissue. Other organisms (hemolysin, epidermolytic toxin, leukocidin) may damage host tissues or alter host response. Staphylococcal "biofilm" which impairs antibody response, activates complement, and causes chemotaxis. *Nocardia* stimulates the formation of "Nocardia granules" that impair drug penetration to the organisms *Pseudomonas* and other gram-negative organisms producing extracellular proteases protect the normal flora from otherwise sterile tissues (which can be facilitated by the presence of foreign bodies) mucus associated with biofilm and accompanying inflammatory response. Persistent, chronic bacterial infections may cause persistent inflammation associated with immune complexes contributes to clinical signs. Unfortunately, bacteria growing in the normal flora killing and immune defenses of the host. In addition to debridement or other methods of cleansing, penetration; dose modification (increase) may be indicated to compensate for debris.

**Antimicrobial Resistance** Development of antimicrobial resistance is facilitated by several factors; among the antibiotics. The normal flora of the gastrointestinal tract is extremely diverse, with anaerobes predominating. A major Gram negative and *Enterococcus* the major Gram positive organisms. Environmental microbes maintain a level of the competition by section of antibiotics. As such, commensal organisms are constantly being exposed to and producing the antibiotic, as well as surrounding normal flora, are resistant to the antibiotic. Thus, genes for resistance directing antibiotic production and organisms are “primed” to develop resistance. The use of synthetic antimicrobials resistance, perhaps because there is less risk of previous exposure of the flora to these drugs. Rapid microbial transfer supports the development of resistance by assuring active DNA and thus mutation potential. Chromosomal (DNA replication, division) are DNA mistakes that have been missed by bacterial repair mechanisms. These mistakes occur spontaneously not the antibiotic is present. Every time an antibiotic is administered, the normal flora is exposed to varying concentrations that confers resistance occurs in the presence of the antibiotic, the surviving mutant can progress to a single strain (see Mutant Potential Concentration). The MIC of the organism is likely to increase. Further microbial mutations and rapid emergence of high-level resistance (eg, unlimited regulation of beta lactamase production) MIC. Stepwise mutations can lead to more specific resistance such as clinical resistance to fluorinated quinolones DNA gyrase. Information for non-specific mechanisms that is shared among organisms can result in multiple or resistance. Microflora of the GI tract can serve as reservoir of resistance genes; a single drug, via integrons, protects the rapid transfer of multiple drug resistance among organisms. Although present for eons, acquired antimicrobial resistance is problematic, although not uniformly so among organisms. The ability of organisms to develop resistance to an antibiotic strain. Many organisms remain predictably susceptible to selected drugs (eg, *Brucella, Chlamydia*); whereas others *Pasteurella multocida*. Even others have proven to be a therapeutic challenge because of resistance that rapidly develops to antimicrobials (*E coli, Klebsiella pneumoniae, Salmonella, Staphylococcus aureus, Streptococcus pneumoniae*). *Pseudomonas aeruginosa* is the prototypic example. A retrospective study of canine urinary tract infections caused by *E.coli* found 33% of to be resistant to all antimicrobials on the antibiogram. The three most commonly recommended clavulanic acid, enrofloxacin, and cephalexin, showed resistance of 47%, 44%, and 51% respectively. In addition to the extended spectrum penicillins (see abstract this proceedings).

**Enhancing Antimicrobial Efficacy.** Examining the relationship between MIC and PDC more closely en
an adequate dose. If one assumes the recommended dose is designed to achieve the MIC\(_{\text{BP}}\) of the drug (a reasonable assumption based, in part, on the Cmax), clearly this concentration will be insufficient for concentration-dependent drugs (f MIC) unless the MIC of the infecting organisms is approximately 1/10\(^{\text{th}}\) of the MIC\(_{\text{BP}}\). For example, whereas a dose of 20 mg/kg, which achieves PDC of 1 \(\mu\)g/ml may be sufficient to treat an E coli with an MIC of 0.06 \(\mu\)g/ml (0.06 \(\mu\)g/ml * 8 = 0.48 \(\mu\)g/ml), even 20 mg/kg, which achieves PDC of approximately 4 \(\mu\)g/ml may not be sufficient to treat a Pseudomonas aeruginosa strain with an MIC of 8 \(\mu\)g/ml even though a C&S report might indicate “S” (1 \(\mu\)g/ml * 8 = 8 \(\mu\)g/ml). Organisms whose MIC is approximately 1/10\(^{\text{th}}\) of the MIC\(_{\text{BP}}\) have undergone the first step mutation leading to low level resistance. Achieving an AUIC necessary to kill 125 is paramount to therapeutic efficacy, particularly in patients afflicted with infections in immunocompromised hosts. Doubling the dose twice daily may be important even for concentration-dependent drugs for treatment of first step mutants. For organisms whose MIC is close to the MIC\(_{\text{BP}}\), PDC will rapidly drop below the MIC. For example, if an organism has an MIC of 8 \(\mu\)g/ml for a beta-lactam, with an MIC\(_{\text{BP}}\) of 32 \(\mu\)g/ml, only two half-lives of the drug can elapse before the MIC (PDC = 16 \(\mu\)g/ml after the first half-life, and 8 \(\mu\)g/ml after the second). Most beta-lactams have half-lives of six hours or less between dosing intervals. Doubling the dose of the drug will add on more half-life to the dosing interval. Thus, the dose (increased) and the interval (shortened) may need to be altered for organisms whose MIC is close to the MIC\(_{\text{BP}}\). Concentrations; modifications become more important and potentially greater for infections further complicated by resistance. Design of doses increasingly will require doses that are higher than recommended. Duration of therapy also is of utmost importance. Duration of therapy is likely to support the advent of resistance; high doses for shorter periods of time (3 to 4 days) may be miminized. Because of this, antimicrobial therapy increasingly will require doses that are higher than recommended. Duration of therapy also is of utmost importance. Duration of therapy is likely to support the advent of resistance; high doses for shorter periods of time (3 to 4 days) may be miminized.

**To MIC and Beyond: Minimizing Antimicrobial Resistance**

**(Or “Dead Bugs Don’t Mutate)**

Dawn Merton Boothe DVM, PhD, DACVIM, DACVCP

Auburn University, AL boothdm@auburn.edu

**INTRODUCTION**

“Even experienced practitioners may not realize that giving a patient antibiotics affects not just that patient, but the other people that come into contact with that environment”. With that statement Dancer (2001) summarized antimicrobial therapy in veterinary (and human) medicine. The National Foundation for Infectious Disease estimates the cost of infections caused by antibiotic resistant bacteria to be as high as $4.5 billion annually; and to be responsible for over 19,000 (human) deaths per year. Thus, this important issue should be no surprise. In the United States alone, approximately 350 million pounds of antibiotics are used in medicine; 40% of this is “unnecessary” according to the human health sector.

In addition to the impact in human medicine, the advent of resistance has and is increasingly impacting veterinary patients. In human medicine, antibiotic stewardship has become the focus for reducing resistance. Prudent veterinarians implement decision making processes (antimicrobial use paradigms) that minimize the temptation to use antimicrobials; for example, the ushering in of designing a dosing regimen based on cost and convenience rather than pharmacodynamics and pharmacokinetics. In veterinary medicine, antimicrobial stewardship begins by recognizing the problems and issues leading to antimicrobial resistance, and successfully implementing strategies that reasonably minimize the impact of antimicrobial use in the patient while not forfeiting the likelihood of therapeutic success.

The most important considerations for selection of an antimicrobial are, in order of priority: 1. confirming the diagnosis (and not indiscriminantly); 2. identifying the target organisms; and 3. identifying the target site. **The 4th through n**
take into account the host, and microbial factors that can negatively impact the selection of the dosing regimen. Among the more relevant considerations are identifying mechanisms of resistance, the likelihood of antibiotic failure at the site, and time-versus-concentration dependency as it relates to convenience. Toxicity (because these drugs generally are not susceptible to these enzymes. Laboratories currently are generating methods intended to detect resistance that has developed in the infecting organism to drug to which the organism is generally susceptible. Cross-reactivity can be an important consideration. Potentially cross-reactive drugs include: ampicillin cephalosporins; enrofloxacin for the fluorinated quinolones). For some classes of drugs, cross-reactivity can be marked. Selection of potential drugs used to treat an infection, one drug often is tested as a model for other drugs in the class. (ie, not all drugs are available for testing. Because automated systems can not accommodate and laboratories must follow guidelines established and published by (or comparable to) the Clinical and Laboratory Standards Institute). Pitfalls of susceptibility testing also reflect the drugs selected for testing. Not all companies are interested in each such, not all drugs are available for testing. Because automated systems can not accommodate and laboratories must follow guidelines established and published by (or comparable to) the Clinical and Laboratory Standards Institute. Materials, including interpretive standards, should be validated by the appropriate agency. Minor changes can profoundly affect results. Personnel should be trained specifically in culture techniques and hospitals that provide C&S (pharmacodynamic) procedures (ie, culture labs) should maintain well designed and adequately collected quality control data to validate their procedures (ie, culture labs). C&S data is inherently deficient because in vitro methods can not mimic in vivo conditions. For example, the inhibitory concentration (MIC). PITFALLS OF CULTURE AND SUSCEPTIBILITY TESTING: IN VITRO CONSIDERATIONS

Although culture and susceptibility data (C&S) can be a powerful tool to guide selection, it nonetheless provides a basis for designing a dosing regimen most appropriate for the patient. Basing antimicrobial selection on the most likely situations where the latter is true is if the infection is at a site in which the organism is R to one fluorinated quinonolone (including ciprofloxacin) is likely to be R to all). However, the former is less important. Cost should be the last consideration that influences selection. Selection should strategically provide a basis for selecting a drug characterized by “R” on the data. The “rule” implies that approximately 90% of infections treated based on C&S are likely to respond if an “S” drug is selected, even if an “R” drug is selected. The most likely situations where the latter is true is if the infection is at a site in which a higher concentration of MIC.

BRIDGING PHARMACOKINETICS AND PHARMACODYNAMICS

Interpreting C&S (pharmacodynamic) data is most helpful when considered in the context of the clinical circumstances. Comparing what is needed – the MIC of the isolate for the drug of interest – to what is achieved. Even if sample would provide a dose regimen. A call to the diagnostic laboratory with suggestions as to how the patient should be dosed would be helpful. Concerns about the infecting organism can profoundly affect results. Personnel should be trained specifically in culture techniques and hospitals that provide C&S procedures (ie, culture labs) should maintain well designed and adequately collected quality control data to validate their procedures (ie, culture labs).
detract from efficacy. Among the most problematic concerns is interpretation of MIC. The breakpoint MIC (MIC	extsubscript{BP}) of a drug is determined in the species of interest, and tested against pathogens. Drugs used by veterinarians are approved for use in humans. Although interpretive MIC data has been determined for many animals, many have not and interpretation may be inappropriate. Ciprofloxacin (CIP) is an excellent example: its breakpoint MIC for Gram positives (MIC	extsubscript{BP}) is equivalent to or less for many organisms. Susceptibility data also does not take into account active metabolism of drug which is metabolized to CIP: both C\textsubscript{max} and area under the curve (AUC) of bioactivity of ENR may increase up to 40% of that in humans, and despite its increased potency compared to enrofloxacin (ENR) toward Gram negatives (MIC	extsubscript{BP}) is equivalent to or less for many organisms. MIC\textsubscript{BP} generally are based on the highest labeled dose, but higher doses might be used in veterinary practice. If recommended doses change, the manufacturer should provide CLASI with updated pharmacokinetic criteria may change accordingly, and automated systems should incorporate those changes in their methods. Against 4-quinolines approved at 2.5 mg/kg, 1 µg/ml (< 1 = S; ≥ 2 = R) was the MIC\textsubscript{BP}; the current dose is up to 20 mg/kg and new breakpoints are R.

**Penetrating the site of infection:** Interpretation of C&S is based on the assumption that the MIC shown by susceptibility testing is relevant to clinical use. Interpretation on plasma drug concentrations (PDC) might result in over or under estimation of drug efficacy. For drugs which are not absorbed systemically (or if the drug can be applied topically), and for drugs which can be concentrated by phagocytes and thus transported at higher concentrations in tissues, appropriate circumstances (eg, topical therapy) a drug designated as "R" might actually be effective. The more relevant concern is to evaluate if drug concentrations are sufficient for killing rates (eg, 99.9% reduction in bacterial inoculum within a 24 hr period of exposure) are achieved at the site. The minimum bactericidal concentration (MBC) of a drug to the MIC. The MBC is determined following tube dilution of the MIC, meaning, the organisms were not simply inhibited, but rather, were killed. For "bactericidal" drugs compared to a static concentration. On the other hand, a "cidal" drug may not kill if concentrations are below MIC and preclude its actions (eg, slow growth in an anerobic environment; combination with growth inhibitors). Thus, clinical breakpoints (ie, MIC/MBC) are achieved at the site. The bactericidal nature of a drug often reflects its mechanism (eg, tetracyclines, macrodilides, lincosamides, chloramphenical) often simply inhibit the growth of the organism, whose ribosomal inhibition is so effective that the organism dies. Drugs which target host membrane (eg, penicillins and cephalosporins; vancomycin), cell membranes (bacitracin, polymixin and colistin), and DNA (enrofloxacin).
are defined in vitro as bactericidal. Combinations of static drugs can often result in cidal actions. For example, some syntheses are static, but when used in combination with diaminopyrimidines (eg, trimethoprim), the combination of bactericidal concentrations of an antimicrobial is critical for those infections for which host killing is likely to be important. PDC is limited to infections in immune compromised animals (eg, viral infections [parvovirus, panleukopenia, FIV, FeLV], osteoarthritis, or osteomyelitis; peritonitis, bacteremia/sepsis, many chronic infections).

**Relationship between MIC, Plasma and tissue Drug Concentrations.** The bridging of pharmacokinetic data can begin by comparing the MIC established by *in vitro* testing and the concentrations of PDC at the time of infection when the drug is administered at the labeled dose. A number of investigators have examined several pharmacokinetic (PK) and pharmacodynamic (PD) indices as predictors of clinical antimicrobial efficacy. Among these, potentially useful are the ratio of $C_{\text{max}}$ (peak PDC) to MIC, defined here as IQ; the area under the inhibitory curve (AUC) to MIC; and the percent time that PDC are above the MIC (PAC). Together, these relationships, two generally categories of drugs have been described.

**Time versus Concentration Dependent Drugs.** The relationship between MIC and the magnitude and time course of drug effect is categorized as to either concentration-dependent (sometimes referred to as dose dependent) or time-dependent drugs. Time-dependent drugs are particularly detrimental. In a mouse model of *E. coli* peritonitis, the antibacterial efficacy of ciprofloxacin, but not penicillin, is dependent on the dose. As such, concentration-dependent drugs generally can be administered at longer intervals, ie, once a day. Time-dependent drugs, assuming drug release (from slow release products) is sufficiently fast to allow $C_{\text{max}}$ to surpass the MIC, can be less likely to develop for FQ characterized by longer half-lives (or for ENR, by the production of an active metabolite). For example, an FQ might be indicated for organisms already characterized by low level resistance (see MPC below); however, due to their long half-lives, it might be necessary to double the dose and redose once a day. In contrast to concentration dependent drugs, efficacy of time-dependent drugs (eg ciprofloxacin-clavulanic acid of 4 mcg/ml, assume PDC achieve the breakpoint MIC (32 mcg/ml) when a recommendation is made to double the dose immediately. With time-dependent drugs, increasing the IQ also may be beneficial, even though efficacy may not be as pronounced as with concentration-dependent drugs because increasing the dose will increase time above the MIC. Since drug concentrations decrease by 50% every one half-life (1.5 hr), PDC have dropped to 16; in 3 hr, to 8 and in 4.5 hr, to 4 mcg/ml. Another 25% of the dose is cleared and the animal should be redosed. If the dose is doubled, this can be extended to 8 hours; the dose would have to be increased 8 fold to reach the standard 12 hrs. This is for an organism with a relatively low MIC; the problem is more pronounced with organisms that are still considered “S” but with MIC that approach the breakpoint MIC. Adding a 3rd or 4th dose is a much more costly treatment (compared to increasing the dose) of time dependent drugs; as such, efficacy of time dependent drugs may be enhanced for time dependent drugs which persist in selected tissues (again, assuming the MIC is surpassed). A time dependent drug can accumulate in active (unbound) form in tissues (ie macrolides) or drugs that accumulate in phagocytes.

**Postantibiotic Effect:** The post antibiotic effect (PAE) describes the continued inhibition of microbial growth after the drug. The impact of the PAE on antimicrobial efficacy can be profound, particularly for concentration-dependent drugs and in systems characterized by derangements in local immunity (ie, CNS infection for which an anatomic inflammatory response may not be observed). In general, concentration-dependent drugs appear to exhibit longer PAE; further studies are needed.
with the magnitude of the peak PDC (ie, longer with higher PDC) and is enhanced by combination antimicrobial concentrations. For each organism.

**Mutant Prevention Concentration.** Based on natural mutational frequencies, a mutation that leads to a single step increase in MIC is usually expected in populations whose density is $\geq 10^7$ colony forming units (CFU). Because most C&S procedures are but not likely to be detected on C&S, yet, if the inoculum is large enough, they will be in the patient. Simply achieving the MIC will not inhibit the growth of all isolates except those whose MIC exceed that reported, including the first step mutants. In immunocompromised the patient, the more likely resistant mutants will survive antimicrobial therapy administered to achieve the reported MIC. Subsequent growth of these mutant organisms will yield a population still considerably smaller than the original (cultured) population. Once this new population of organisms with higher MIC achieves $\geq 10^7$ CFU, they will be characterized by a high level of resistance. The MIC of this second population of organisms will no longer be achievable with therapeutic doses. In vitro evidence is supported by in vivo experiences describing this scenario; further, clinical experiences in animals with chronic UTI, otitis externa or other infections characterized by previous antimicrobial use. A first step in the development of bacterial prostatitis in humans receiving only three days of ciprofloxacin at doses designed to achieve the MIC; prostatismological procedures was associated with the advent of resistant *E coli*. Not surprising, a novel approach intended to address high level resistance has been proposed in the design of dosing regimens in humans. The goal of therapy is to achieve the Mutant Prevention Concentration (MPC), rather than the MIC, of the drug (at the site) in the patient. Achieving concentrations in the MPC will avoid overdosage; the bottom of the window is the MIC reported by culture techniques (based on $10^5$ CFU) whereas the MPC is defined as the highest MIC identified in a population ($\geq 10^7$) of susceptible organisms, thus including the first step mutation; alternatively, it is defined as the drug concentration that would require an organism to develop the ability to grow in the presence of the drug. Whereas the MIC indicates the concentration of drug which inhibits the growth of all isolates except those whose MIC exceed that reported, the MPC indicates the concentration of drug that would minimize the advent of resistance. To prevent the advent of resistant organisms at the site of infection should be sufficiently high to inhibit the growth of the organisms that have undergone step mutations. Achieving drug concentrations below the MIC of the infecting organism is not likely to facilitate the advent of resistant isolates to be applied to the infecting organism. Unfortunately, determining the MPC of an isolate cultured from a patient will be based on $\geq 10^7$ organisms, currently cost prohibitive. Insufficient information is available to allow a reasonable prediction of the MPC and MIC of an organism, which is likely to vary with the drug and the organism. For veterinary FQs, Wetzler et al reported the MPC to MIC ratio to be approximately 6 to 10 for *E coli* (ATCC 8739) but as high as 23 to 50 for *Staphylococcus aureus*. When making dosing recommendations in humans, the MPC to MIC ratio of *E coli* (ATCC 8739) is usually considered "well perfused" might be of concern. For patients in whom bronchial secretions well, despite the fact that the lungs are well perfused. Amoxicillin is often used to treat resistant infections of the amoxicillin that is in plasma is distributed to bronchial secretions. Theoretically, one must dose amoxicillin to achieve targeted PDC in bronchial secretions. Most water soluble drugs (beta-lactams and aminoglycosides) reach bronchial secretions whereas over 50% of lipid soluble drugs reach bronchial secretions. Dosing adjustments also are necessary for intracellular or complicated by host response to infection.

**Host Factors:** The impact of host response to infection can be profound. Problems contributing to therapeutic failure are often unappreciated, and deciding upon an antimicrobial regimen that will assure bactericidal concentrations of the chosen drug reach the site of infection and will be effective against the infecting organism. With the increased use of otherwise appropriate clean/drain accessible infections, select a drug that distributes into tissues well and ideally increase the dose appropriately). Even tissues traditionally considered "well perfused" might be of concern. For example, bronchial secretions well, despite the fact that the lungs are well perfused. Amoxicillin is often used to treat resistant infections, but the MIC of the amoxicillin that is in plasma is distributed to bronchial secretions. Therapeutically, one must dose amoxicillin to achieve targeted PDC in bronchial secretions. Most water soluble drugs (beta-lactams and aminoglycosides) reach bronchial secretions whereas over 50% of lipid soluble drugs reach bronchial secretions. Dosing adjustments also are necessary for intracellular or compounded by host response to infection.

**Microbial Factors:** Materials released from microbes facilitate invasion, impair cellular phagocytosis, and protect against antimicrobial killing. Staphylococci associated with canine pyoderma produce "slime," a material that facilitates bacterial adhesion to host tissues. Certain organisms (hemolysin, epidermolytic toxin, leukocidin) may damage host tissues or alter host response. Staphyloccus aureus, which impairs antibody response, activates complement, and causes chemotaxis. *Nocardia* stimulates the formation of granules" that impair drug penetration to the organisms *Pseudomonas* and other gram-negative organisms producing a biofilm which protects the organism. Biofilms are microcolonies of pathogenic and host microbes embedded in a polysaccharide matrix that impairs drug penetration. Normal microflora of the skin or mucous membranes are characterized by a high level of resistance. The MIC of this second population of organisms will no longer be achievable with therapeutic doses. In vitro evidence is supported by in vivo experiences describing this scenario; further, clinical experiences in animals with chronic UTI, otitis externa or other infections characterized by previous antimicrobial use. A first step in the development of bacterial prostatitis in humans receiving only three days of ciprofloxacin at doses designed to achieve the MIC; prostatismological procedures was associated with the advent of resistant *E coli*. Not surprising, a novel approach intended to address high level resistance has been proposed in the design of dosing regimens in humans. The goal of therapy is to achieve the Mutant Prevention Concentration (MPC), rather than the MIC, of the drug (at the site) in the patient. Achieving concentrations in the MPC will avoid overdosage; the bottom of the window is the MIC reported by culture techniques (based on $10^5$ CFU) whereas the MPC is defined as the highest MIC identified in a population ($\geq 10^7$) of susceptible organisms, thus including the first step mutation; alternatively, it is defined as the drug concentration that would require an organism to develop the ability to grow in the presence of the drug. Whereas the MIC indicates the concentration of drug which inhibits the growth of all isolates except those whose MIC exceed that reported, the MPC indicates the concentration of drug that would minimize the advent of resistance. To prevent the advent of resistant organisms at the site of infection should be sufficiently high to inhibit the growth of the organisms that have undergone step mutations. Achieving drug concentrations below the MIC of the infecting organism is not likely to facilitate the advent of resistant isolates to be applied to the infecting organism. Unfortunately, determining the MPC of an isolate cultured from a patient will be based on $\geq 10^7$ organisms, currently cost prohibitive. Insufficient information is available to allow a reasonable prediction of the MPC and MIC of an organism, which is likely to vary with the drug and the organism. For veterinary FQs, Wetzler et al reported the MPC to MIC ratio to be approximately 6 to 10 for *E coli* (ATCC 8739) but as high as 23 to 50 for *Staphylococcus aureus*. When making dosing recommendations in humans, the MPC to MIC ratio of *E coli* (ATCC 8739) is usually considered "well perfused" might be of concern. For patients in whom bronchial secretions well, despite the fact that the lungs are well perfused. Amoxicillin is often used to treat resistant infections, but the MIC of the amoxicillin that is in plasma is distributed to bronchial secretions. Theoretically, one must dose amoxicillin to achieve targeted PDC in bronchial secretions. Most water soluble drugs (beta-lactams and aminoglycosides) reach bronchial secretions whereas over 50% of lipid soluble drugs reach bronchial secretions. Dosing adjustments also are necessary for intracellular or complicated by host response to infection.
penetration; dose modification (increase) may be indicated to compensate for debris.

Antimicrobial Resistance. Development of antimicrobial resistance is facilitated by several factors; among the major Gram negative and Enterococcus the major Gram positive organisms. Environmental microbes maintain a competition of the competition by section of antibiotics. As such, commensal organisms are constantly exposed to antibiotics producing the antibiotic, as well as surrounding normal flora, are resistant to the antibiotic. Thus, genes for resistance directing antibiotic production and organisms are “primed” to develop resistance. The use of synthetic antimicrobials resistance, perhaps because there is less risk of previous exposure of the flora to these drugs. Rapid microbial transfer supports the development of resistance by assuring active DNA and thus mutation potential. Chromosomal (DNA division) are DNA mistakes that have been missed by bacterial repair mechanisms. These mistakes occur sporadically, not the antibiotic is present. Every time an antibiotic is administered, the normal flora is exposed to varying conditions that confers resistance occurs in the presence of the antibiotic, the surviving mutant can progress to a single step resistance (see Mutant Potential Concentration). The MIC of the organism is likely to increase. Further microbial stepwise mutations and rapid emergence of high-level resistance (eg, unlimited regulation of beta lactamase production) near the MIC. Stepwise mutations can lead to more specific resistance such as clinical resistance to fluorinated quinolones. DNA gyrase. Information for non-specific mechanisms that is shared among organisms can result in multiple organism resistance. Microflora of the GI tract can serve as reservoir of resistance genes; a single drug, via integrons, provides rapid transfer of multiple drug resistance among organisms. Although present for eons, acquired antimicrobial resistance is becoming problematic, although not uniformly so among organisms. The ability of organisms to develop resistance to an antibiotic strain. Many organisms remain predictably susceptible to selected drugs (eg, Brucella, Chlamydia); whereas others (Pasteurella multocida). Even others have proven to be a therapeutic challenge because of resistance that rapidly develops multidrug resistance (MDR). Multidrug resistance is now considered the normal response to antibiotic pressure pneumococci, enterococci and staphylococci. Among these, Staphylococcus is considered most problematic: it is not to many different environmental conditions, and it tends to be associated with life threatening infections.

Our laboratory has documented what appears to be increasing rates of antimicrobial resistance among feline and canine isolates indicative of a range of activities. A retrospective study of canine urinary tract infections caused by Escherichia coli (E.coli, Klebsiella pneumoniae, Salmonella, Staphylococcus aureus, Streptococcus pneumonia). In addition, we have developed multidrug resistance (MDR). Multidrug resistance is now considered the normal response to antibiotic pressure pneumococci, enterococci and staphylococci. Among these, Staphylococcus is considered most problematic: it is not to many different environmental conditions, and it tends to be associated with life threatening infections.

Enhancing Antimicrobial Efficacy. Examining the relationship between MIC and PDC more closely enforces the need for an adequate dose. If one assumes the recommended dose is designed to achieve the MIC of the drug (a reasonable assumption, in part, on the Cmax), clearly this concentration will be insufficient for concentration-dependent drugs (eg, beta-lactams) unless the MIC of the infecting organisms is approximately 1/10th of the MIC. For example, whereas a dose of 1 µg/ml achieved PDC of 1 µg/ml (0.06 µg/ml * 8 = 0.48 µg/ml) for a beta-lactam, with an MIC of 0.06 µg/ml (0.06 µg/ml * 8 = 0.48 µg/ml) may not be sufficient to treat a Pseudomonas aeruginosa even 20 mg/kg, which achieves PDC of approximately 4 µg/ml even though a C&S report might indicate “S” (1 µg/ml * 8 = 8 µg/ml). Organisms whose MIC is approaching the MIC of the drug, for organisms whose MIC is close to the MIC, PDC will rapidly drop below the MIC. For example, if the MIC of the drug, for organisms whose MIC is less than 8 µg/ml for a beta-lactam, with an MIC of 32 µg/ml, only two half-lives of the drug can elapse before the MIC (PDC = 16 µg/ml after the first half-life, and 8 µg/ml after the second). Most beta-lactams have half-lives of 2-6 hours; therefore, dosing twice daily may be important even for concentration-dependent drugs for treatment of first step mutants. The MIC of the drug, for organisms whose MIC is close to the MIC, PDC will rapidly drop below the MIC. For example, if the MIC of the drug, for organisms whose MIC is less than 8 µg/ml for a beta-lactam, with an MIC of 32 µg/ml, only two half-lives of the drug can elapse before the MIC (PDC = 16 µg/ml after the first half-life, and 8 µg/ml after the second). Most beta-lactams have half-lives of 2-6 hours; therefore, dosing twice daily may be important even for concentration-dependent drugs for treatment of first step mutants.
concentrations; modifications become more important and potentially greater for infections further complicated by antimicrobial therapy increasingly will require doses that are higher than recommended. Duration of therapy also increases the opportunity for the development of resistance. Shorter duration of therapy is likely to support the advent of resistance; high doses for shorter periods of time (3 to 4 days) may be required to compensate for the slower growth of some strains of infecting microbes.

In conclusion, increasingly, rationale use of antimicrobial therapy should focus on short term therapy with compounds that are active against the infecting microbes, thus assuring that mutation to resistant organisms does not occur. Design of doses are most important when susceptibility data that indicates the MIC; the closer the MIC is to the breakpoint MIC, the more important increasements of the MIC to the breakpoint MIC, the more important increasements of the MIC to the breakpoint. Reduction of the interval between doses also needs to be miminized.

Combination therapy is another method that may be employed to minimize the evolution of resistance. Combination therapy is another method that may be employed to minimize the evolution of resistance.