Veterinary Hospital Management

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What is hospital/health care administration?

- Health administration or healthcare administration is the field relating to leadership, management, and administration of public health systems, health care systems, hospitals, and hospital networks.
- Health care administrators are considered health care professionals.
- Definite role of administration other than clinical services
- From reception to the discharge of a case at each step administrative persons required
- Management of a single institution (e.g. a hospital/clinic) is also referred to as "medical and health services management", "healthcare management", or "health administration".

Why is it required?

Health systems management ensures that

- Specific outcomes are attained
- Departments within a health facility are running smoothly
- The right people are in the right jobs
- People know what is expected of them
- Resources are used efficiently and
- All departments are working towards a common goal

Historic Background

- Early hospital administrators were called patient directors or superintendents.
- At the time, many were nurses who had taken on administrative responsibilities.
- The first degree granting program in the United States was established at Marquette University in Milwaukee, Wisconsin.
- The original idea is credited to Father Moulinier, associated with the Catholic Hospital Association.
- The first modern health systems management program was established in 1934 at the University of Chicago.
- At the time, programs were completed in two years – one year of formal graduate study and one year of practicing internship.
- In 1958, the Sloan program at Cornell University began offering a special program requiring two years of formal study, which remains the dominant structure today
- Health systems management has been described as a "hidden" health profession because of the relatively low-profile role managers take in health systems, in comparison to direct-care professions such as nursing and medicine.
- However the visibility of the management profession within healthcare has been rising in recent years, due largely to the widespread problems developed countries are having in balancing cost, access, and quality in their hospitals and health systems.

Healthcare administration as a career

Health care management is usually studied through healthcare administration or healthcare management programs in a business school or, in some institutions, in a school of public health.

Although many colleges and universities are offering a bachelor's degree in healthcare administration or human resources, a master's degree is considered the "standard credential" for most health administrators.

Research and academic-based doctorate level degrees, such as the Doctor of Philosophy (PhD) in Health Administration and the Doctor of Health Administration (DHA) degree, prepare health care professionals to turn their clinical or administrative experiences into opportunities to develop new knowledge and practice, teach, shape public policy and/or lead complex organizations.There are multiple recognized degree types that are considered equivalent from the perspective of professional preparation.

The Commission on the Accreditation of Healthcare Management Education (CAHME) is the accrediting body overseeing master's-level programs

It accredits several degree program types, including

Master of Hospital Administration (MHA)
Master of Health Services Administration (MHSA)
Master of Business Administration in Hospital Management (MBA-HM)
Master of Health Administration (MHA)
Master of Public Health (MPH, MSPH, MSHPM)
Master of Science (MS-HSM, MS-HA)
Master of Public Administration (MPA)

Economics of health care administration

It was reported in September 2014, that the United States spends roughly $218 billion per year on hospital's administration costs, which is equivalent to 1.43 % of the total U.S. economy. Hospital administration has grown as a percent of the U.S. economy from .9 % in 2000 to 1.43 % in 2012, according to Health Affairs. In 11 different countries, hospitals allocate approximately 12 % of their budget toward administrative costs. In the United States, hospitals spend 25 % on administrative costs.

Veterinary hospital/clinic administration

It is extremely difficult to separate each specific level of management from the others because each level builds upon the knowledge and skills of others. Therefore, we will look at three differing levels simultaneously:

1) Veterinary Hospital Office Manager
2) Veterinary Practice Manager
3) Veterinary Hospital Administrator

A typical Veterinary Hospital Office Manager may be responsible for the following:

• The supervision of receptionists and/or front office support staff.

• The office manager may be responsible for the initial reviewing of receptionists' applications, receptionists' interviews, their training, and may perform their employment reviews.

Veterinary Practice Manager

- A veterinary practice manager is charged with the responsibilities of managing the business activities of a veterinary hospital.

- A practice manager may have extremely different responsibilities and authority depending upon the size of the hospital and the ownership or administrator's delegation.

- Practice managers will have knowledge of all the responsibilities of a veterinary office manager and have the ability to further the management of a veterinary hospital by having

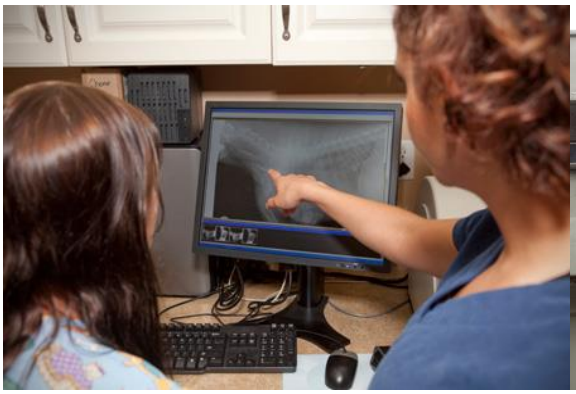
direct authority and decision-making responsibilities over all business aspects of the veterinary practice.

A typical veterinary practice manager may have the following responsibilities:

• Plans for optimal staffing to assure maximum productivity and service.

• Directly recruits, interviews, and hires support staff personnel.

• Supervises support staff personnel and acts as the direct authority regarding disciplinary procedures, discharging, and all the legal responsibilities of employment.



The veterinary hospital manager is responsible for seeing that success is achieved with each client interaction with the veterinary hospital staff.

•The hospital manager will use the tools of education, motivation, structuring, scheduling, coordinating, evaluation, and analysis to achieve optimal client satisfaction from the veterinary hospital services and staff.

•Developing and accomplishing a hospital marketing program.

• Oversees the building and equipment maintenance and housekeeping standards.

Either directly prepares or supervises preparation of all business accounting reports and transactions. Audit both the preparer and hospital personnel performance to assure that proper methods and techniques are being used.

• Review and/or prepare all accounts payable and receivables to confirm that each is handled correctly and timely.

• Periodically reviews fee schedule for services, products, and increases or changes as necessary.

• Establishes hospital budgets and projections for growth.

• Reviews and/or purchases supplies and equipment assuring that a periodic review is made to assure that optimal prices are obtained.

Veterinary Hospital Administrator

- The function of a veterinary hospital administrator is unique from all other positions in a veterinary hospital because the administrator has complete authority over the operation of the business and practice in concert with the practice owner(s) or board of directors.
- The administrator will be the coordinator and final authority of all business functions and the supervising agent of all hospital services and personnel.

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A veterinary hospital administrator will be responsible for all of the functions described for office and hospital managers with the additions of being responsible for professional staffing and supervision.
- While the administrator may not have the knowledge of a veterinarian regarding medicine, the administrator should have a general knowledge of quality assurance and performance in veterinary medicine and may act in an advisory role in helping establish and supervise medical protocols of the practice.
- A typical veterinary hospital administrator's responsibilities will include all of those listed for the office manager and practice manager with the addition of a fourth area of responsibility:
  - Either directly or in conjunction with the business owner(s) or board of directors: recruits, interviews, and hires professional staff.
  - Mediates professional staff personnel problems, maintains their employment policies and contracts, and may act to help maintain and supervise a medical protocol.
- The purpose of a hospital administrator is to serve the owner(s) or board of directors of the practice in establishing and reaching the goals and policies they desire.
- The administrator combines the elements of business and veterinary medicine to succeed in maintaining excellence and quality of care to clients and their pets.
- There are obviously degrees of veterinary practice management requiring increasingly higher levels of knowledge, skill, and expertise.
- Most veterinary practices employing a full-time person appointed to manage the business affairs of the practice can expect that administrator to have general knowledge and skills in four very general areas.
- Each practice will customize the hospital administrator's role to meet their individual needs and requirements.
- The veterinary hospital administrator uses their skills and authority to accomplish optimal staffing to assure maximum productivity and service.
- The veterinary hospital administrator uses the tools of education, motivation, coordination, evaluation,
- and analysis to achieve optimal client satisfaction from the veterinary practice services and staff.
- The veterinary hospital administrator directly or through supervision performs financial functions of the business ranging from fee structuring and application to tax preparation and debt/asset management. The common goal of all veterinary hospital administrators is their desire to achieve the best financial success possible for their businesses.

• The veterinary hospital administrator may be charged with a range of administrative esponsibilities from daily directing the business affairs of the practice to establishing the short and long term direction, goals, budgets, and protocols of the business and practice.

Schedules personnel and is responsible for support staff personnel employment benefits.

• Assures that personnel are properly trained for their position.

• Maintains a thorough set of employment policies and employee manual.

• Mediates all personnel problems, maintains employee motivation, and structures continuing education for support staff personnel.

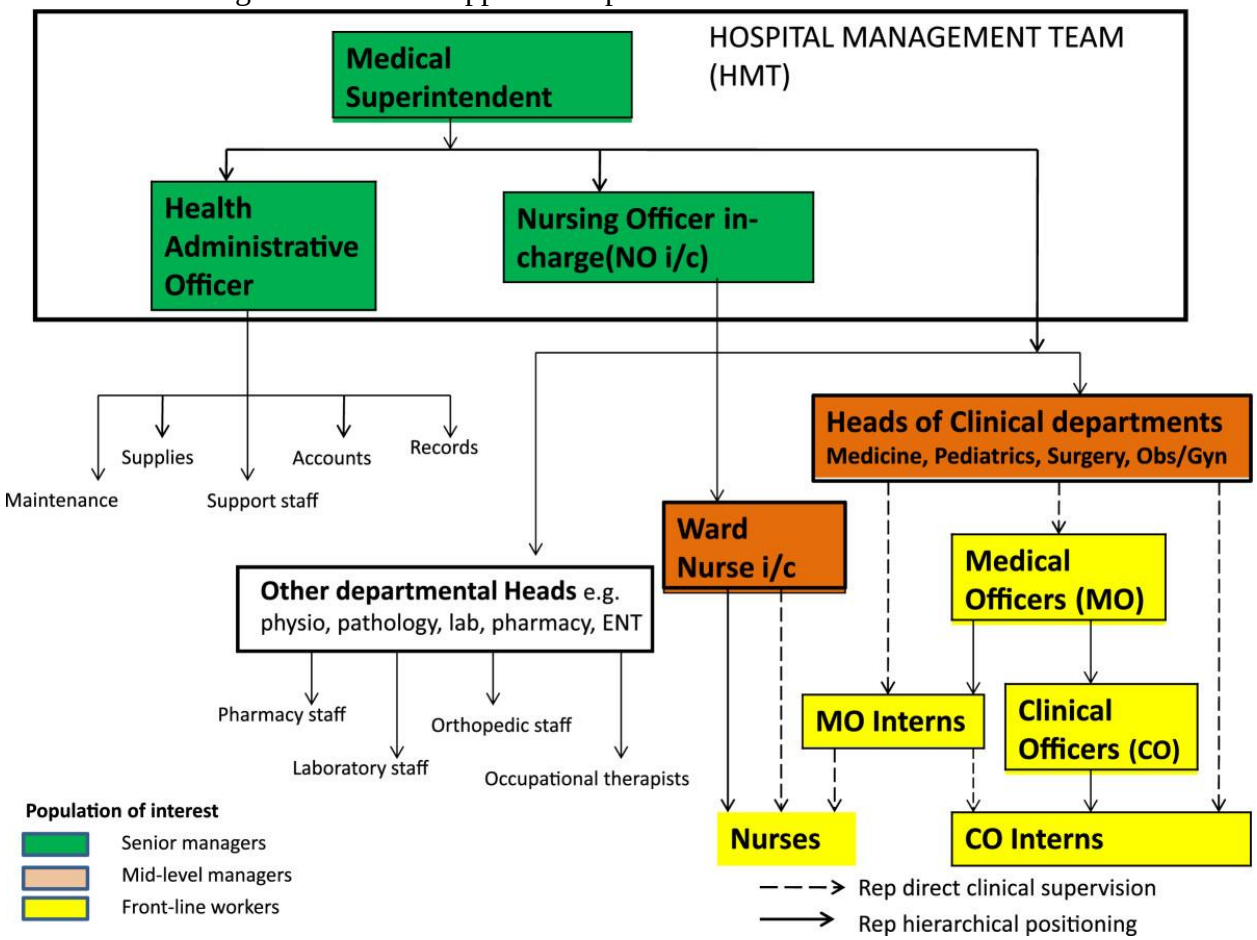
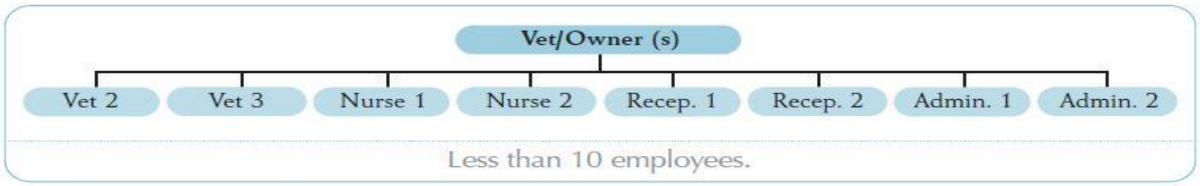
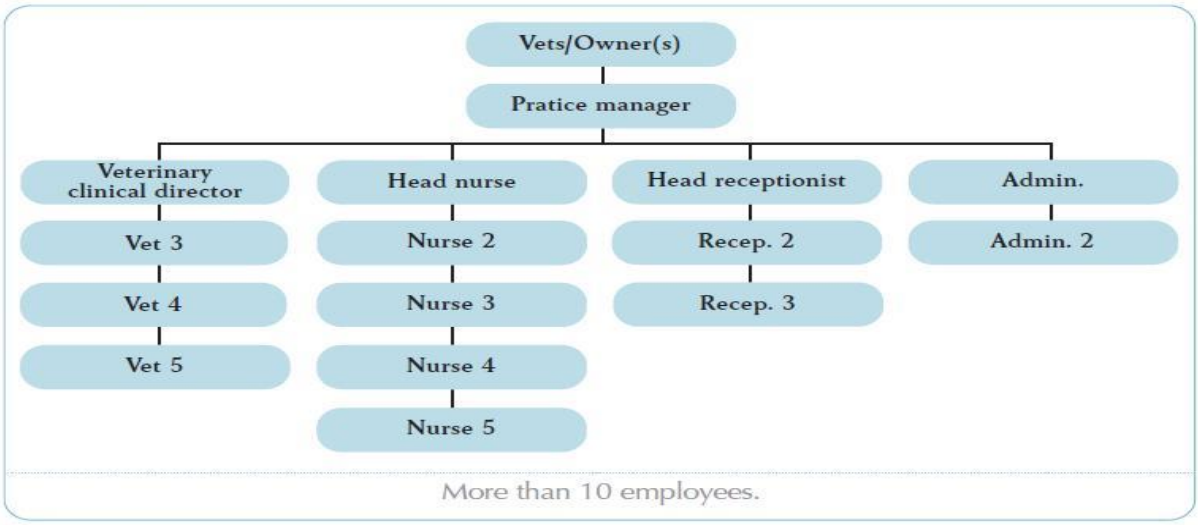


Figure 1. Evolution of a veterinary clinic's organisational chart.



NB: Job titles may vary depending on country.



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CLINICAL EXAMINATION OF PET ANIMALS

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Diagnosis of disease in an animal that cannot describe its complaints is an interesting job comparable to solving a puzzle and will be enjoyed by those who are sincere, interested in the profession and pledged for the service to the dumb. Number of animals required to be dealt with, limitations in diagnostic facilities under filed conditions, cost of treatment, public health aspect and such other considerations render the job of a veterinary physician extremely difficult. Quickness and accuracy in diagnosis is most important while dealing with an outbreak so as to take proper preventive measures and save a large number of animals at risk. Responsibility becomes still more while dealing with zoonoticinfections.

It is with these practical difficulties that a veterinary physician has to perform his duty and only a sound knowledge of physiology, pathology, microbiology, nutrition, pharmacology, medicine and its related fields together with skill of observation and interpretation that a veterinary physician can attain proficiency and reputation. Accurate diagnosis and understanding of the disease is essential for adopting proper line of treatment. One must observe the normal behaviour and habits of animals in health and develop an ability to identify the sick animal. Veterinary clinician has to depend on the objective symptoms observed, information about the history of a case and finally on the various methods of examination.

General signs of Illness

A sick animal looks dull in appearance and expression, either takes only a little feed or may be completely off-feed. If the animal has fever, in addition to the above signs, body surface warm and rough due to erect hair, breath is hot, dung usually hard, urine scanty and deep in colour, respiration accelerated and there may be redness of eyes and lacrymation.General inspection of the patient is the first step in the diagnostic approach to form an overall impression of the animal initially by observing the animal from a distance and this can be done while the patient is entering the hospital or you are viewing the patient while visiting for it's treatment.

Clinical Examination

(A) History of a Case *(Anamnesis)*

A tactful enquiry of history by taking the owner or the animal attendant into confidence and by judicious and discrete questioning is the first step and a key to accurate diagnosis. Previous History: Information about previous illness if any and it's relation with the present condition.

For example

- Chorea as an after effect of Distemper in dogs.
- Past history of dog bite to support diagnosis of rabies.

History Pertaining to Present Illness:

(a) Duration of illness	To *know the nature of the disease i.e., whether per acute, acute or chronic, Categorization of disease is based on the duration of illness and not on the severity of illness. Peracntc -few hours to 2 days, Acute-3 to*

	*7days Snbacute —*2 *to* 4 *weeks Chronic— more thnn 4 weeks,*
(b) How are the normal functions of body and behaviour.	*(defecation, urination*) History of recent change in behaviour is a very important.
(c) First symptoms noticed	*To know what system is primarilyinvolved.*
(d) Appetite and water intake	*Whether feed is changed*
(e) Whether any other animals in the herd or vicinity are affected	*Possibility of common cause of infection s disease.*
(f) Wether any treatment given earlier	*Drenching pneumonia due la faulty drenching, drug resistance due lu incomplete treatment, possibility of reaction to other drugs.*

General Inspection: Is the first step wherein you should observe general health, skin, bodily condition, behaviour, expression, respiration, posture etc.

General health, bodily condition, appearance of skin and coat	Stunted growth, malnutrition, oedema, rickets, chronic diseases, skin diseases, deficiency diseases can be diagnosed
Behaviour, expression, posture and gait (Habitus)	Diseases of C.N.S. paralysis, specific disease like rabies can be diagnosed
Respiration	Observe change in rate, depth, character of respiration, adventitious sounds and type of nasal discharge. These abnormalities are seen in primary respiratory disease or secondarily due to cardiac involvement, allergic conditions
Abdomen	Distention of abdomen may occur in impaction, ascties, intestinal obstruction, tumors and advanced pregnancy

Inspection of the patient and history taking should not be undertaken simultaneously, because it is likely that you may miss some important lesion.

General Appearance and Behavior

Attitude	Possible Diagnosis
Frenzied restlessness, butting, weakness of hindquarters.	Rabies
Moving to and fro, lifting the feet, Kicking at the belly & anxious look	Colic

General Examination

This needs approaching and handling the animal. By the time general observation and history is completed you get the idea about the general behaviour and temperament of the animal

Restraint

The animals have to be handled routinely for the purpose of clinical examination and administration of medicines. Animals vary in their behaviour and temperament from highly docile to extremely ferocious. Each animal's behavioural pattern has to be studied while approaching and handling.

One must protect himself from accidental injuries i.e. bites in case of dogs and cats. The owner or animal attendant usually knows about the habits of animal and one must explore the temperature of animal by asking appropriate questions. Sometimes animals behave in unpredictable manner due to change of place and in presence of unfamiliar persons. Proper restraint by tape muzzle in dogs is always necessary. While handling the mouth parts and examination of buccal cavity one must be more particular and the remotest, possibility of rabies should be ruled out. Never examine the buccal cavity without wearing hand gloves in case of any slightest injuries on your fingers.

It may be necessary to tranquilize the animal, if it is a very aggressive and non co-operating. Cruel methods of restraint should be avoided as they may cause accidents like fractures and sometimes strangulation and asphyxia. Always handle animals in presence of their owner.

General Clinical Examination

Recording Temperature: Record temperature while animal is at rest. Eliminate effect of high atmospheric temp., in summer. For this, dip the mercury bulb of thermometer in cold water *immediately after* recording and before reading the theromometer. Let the pet animal become quiet on the examination table. High fever is a first sign in several acute infectious diseases. Record temperature twice a day in sick animal to know the type of fever *(continuous, recurrent, intermittent etc.)* which is a very important criteria for disease diagnosis. Range of fever is also an important deciding factor to suspect a peracute condition such as acute infectious disease. Fever of moderate range or intermittent nature gives some breathing time to arrive at proper diagnosis and treatment. *(In fact this is very good guideline for the owner to decide as to how urgently the veterinary aid should be sought for only if he can record the temperature. This is the modern trend winch a owner must adopt for himself to save his precious pet.).* Fever is only the symptom and other associated symptoms need however to be considered by the efficient Veterinary clinician.

Pulse :

Site-Dog : Femoral artery on the inside of thigh.

Examine pulse at rest because excitement and exercise will affect its rate and character. One can know about the condition of heart, peripheral, circulation and certain diseases like chronic interstitial nephritis in old dogs and endocarditis by examining pulse, irregularity and loss of rhythm is common in dogs of nervous or excitable temperament..

Type of pulse	**Possible disease condition**
Weak pulse:	Myocardial asthenia, or Mitral incompetence
Strong pulse:	Hypertension
Wiry pulse:	Acute pleurisy, Pericarditis and Peritonitis.
Thready pulse:	Fatal condition, Large bounding and water hammer
Pulse:	Anaemia, Insufficiency of aortic semilunar valve or patent ductous arteriosus.

Respiration:

Count respirations while animal is at rest. Note the type *(costal, abdominal, intercostal, jerky etc.)* Resp. rate increases in fevers, in summer, after exercise, and in resp. distress due to disease. It also increases due to nervous excitement and in acute painful condition. There is a

ratio 1:3 between resp. and pulse in healthy animal. This ratio is altered to 1:1 in respiratory diseases. Note the type of dyspnoea.

Examining the Visible Mucous Membranes

Visible m.m. can be examined at conjunctiva, buccal or nasal mucosa and inside of vulva in females. Examine in bright natural day light. In normal healthy animal the mucous membrane is moist and rosy coloured. Examine at both so as to rule out local conditions.

Look for

Congestion:	Sign in fever and inflammation
Paleness:	Sing of anaemia, internal haemorrage and shock
Yellow Colouration:	Sign of jaundice and liver disease
Petecheal Haemorrhages:	Seen in septicemias and surra
Blue Colouration:	Sign of cyanosis observed in hypoxia, dyspnoea and congestive cardiac failure

Examination of Eyes

Sunken Appearance:	Sign, of Chronic. Wasting Disease and dehydration.
Pupilar Response:	Loss of pupilar response to light seen in toxaemias, shock, some poisoning and C.N.S. diseases.
Corned Opacity, Ulcers:	Usually due to injuries but also seen in diseases like Distemper,

External Body Surface and Skin

One can get idea about body temperature, provided external atmospheric influences are ruled out. Look for general lustre, pliability of skin, oedema, skin disease, loss of hair, loss of pigmentation. Degree of dehydration can be assessed by pulling a fold of skin at parts where it is loose.

Methods of Physical Examination

A systematic approach must be developed and used on every physical examination. The initial and often the most important step in diagnostic approach to the sick animal is the physical examination. A complete examination should always be performed, even though the complaint with which the animal appears is one easily recognizable.Each one of the following methods has its own importance when judiciously employed. An experienced clinician can note several important points simply by careful observation of the patient from distance and this has already been discussed under *"General Inspection".*

(1) Palpation:-

By palpation or laying hand with gentle pressure on a part of a clinician can feel the consistency of organ or tissues *(fibrous, induration)* whether the part is hot and or painful (abscess) or cold and painless *(tumour, cyst)* whether swollen and distended part of abdomen contains gases or dry impacted feed *(Impaction)* or fluid *(ascites, oedema),* By deep palpation in small animals a clinician can feel the abdominal tumors, intestinal obstruction, gravid uterus, cystic calculus etc..

(2) Percussion:

A useful diagnostic aid and can be recognized by striking on a part of body with finger tips *(immediate percussion)* or by use of plexor *(a rubber hammer)* and pleximeter a circular disc *(mediate percussion)* the organ underneath is set into vibration and from the sounds produced, a clinician can get an idea about the contents and consistency

(3) Auscultation:-

Listening of sounds produced in course of normal physiological functioning of certain vital organs by use of stethoscope is very valuable method of clinical examination and needs considerable practice and patience to attain proficiency for correct interpretations. Useful for hearing peristaltic sounds during ruminal as well as intestinal contraction, knowing the functional state of respiratory tract and lungs : *(prominent vesicular murmur in congestion, moist rales when exudate is present)* cardiac sounds *(friction sounds in early pericarditis, cardiac murmrs in valvular diseases, splashing sounds in hydropericardiurn etc.).* It is possible to diagnose respiratory and cardio vascular diseases by auscultation by an expert physician.

It is necessary to eliminate unrelated sounds from surroundings, sounds due to movements of skin, friction sounds produced due to rubbing of hair. The animals should be properly restrained, quiet and in case of long haired animals the coat should be moistened so as to eliminate due to hair friction.Sound knowledge of topographical anatomy is an essential prerequisite for percussion and auscultaion.

(4) Other Diagnostic Aids

In addition to the above methods various other specialized methods are now available in veterinary practice as are routinely practiced.Branch specialisation such as Gastroenerology, Nephrology, Cardiology, Neurology Dermatology, Surgery, Gynaecology, Opthalmology and so on are gradually developing in animal pactice in India. For developing such specialized consultation and sophisticated therapy there must be enough number of references made by the general practioners spread over in moffusil areas. These specialities have already developed in western countries like America where specific facilities are available. General practitioners refer specific cases to these speciality hospitals where cases are examined by subject matter specialist and the required investigations performed in certain cases for confirmation of the tentative diagnosis. X-ray examination, exploratory puncture, exam of urine, bacteriological exam. Serological tests, allergic tests, biopsy, biochemical tests and several other types of diagnostic tests, including E.C.G., which has been found very useful in small animal practice, Laproscopic exam, is also possible to visualise the visceral organs. Recent developments in diagnostics such as Endoscopy, Magnetic Resonance Imaging (MRI). Computerised Tomography Scanning (CT scan) and ultra sonography are some of the recent diagnostic techniques which could be used in small animal practice. These diagnostic facilities need costly equipments and trained technical staff to operate and interpret the findings. Presently these facilities are available in Veterinary college hospitals and some district level Veterinary polyclinics.

Indiscriminate use of antibiotics, incomplete and delayed treatment in cases infections has created problems of resistant bacterial strains. In such situations cultural examination and antibiotic sensitivity tests have become valuable diagnostic aids in the successful management of infections.

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CLIENT MANAGEMENT IN VETERINARY CLINICAL PRACTICE

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The success of a veterinary practice depends not only on the clinical skills of the professional veterinary but also on non-clinical skills related to organizing and managing the practice.In many veterinary practices, the non-clinical skills associated with human resource management, marketing, financial management, and service delivery are the responsibility of the practice.Management of pet owners may not be familiar with the job requirements of a practice and may need guidance in hiring qualified veterinarians. Practicing Veterinary doctor should have ability to express one's thoughts verbally in a clear and understandable manner, and the ability to actively listen and attend to what others are saying. Good group presentation skills are also important.Integrity-honesty, trustworthiness, and adherence to high standards of ethical conduct. Leadership - a willingness to lead and take charge, and the ability to motivate others and mobilize group effort toward common goals. Adaptability- being open to change, flexible work methods, and the ability to adapt behaviour to changing conditions or new information. Compliance- being reliable, thorough, and conscientious in carrying out work assignments, as well as an appreciation for the importance of organizational rules and policies. Planning and Prioritizing- the ability to effectively manage time and work load to meet deadlines; the ability to organize work, set priorities, and establish plans for achieving goals. Decision Making- the ability to make good decisions, solve problems, and decide on important matters; the ability to gather and analyze relevant data and choose decisively between alternatives. Relationship Building-the ability to develop constructive and cooperative working relationships with others and maintain them over time; must also be able to settle disputes, resolve grievances and conflicts, and negotiate with others.

In Veterinary Clinical practice client/pet owner plays very important role so veterinarians should establishes protocols for client communications and monitors client services to facilitate client retention and satisfaction. Client Services requires monitoring client retention, develop and manage new client programs, handle client complaints, obtain/report client feedback on service, respond to client questions, develop and manage client reminder system, Client Education for first aid and seriousness of heath related issues of pets and management of client education system. Remember the clients' perceptions and expectations of services from veterinary establishments are important concerns that need appropriate attention. Veterinarians in private practice have a duty to their clients and the public, as well as themselves, to inform their clients and the public, generally and up front, about their facility and its policies. This information should include choices concerning hospitalization, various methods and personnel for patient monitoring, after hours and emergency services, and the use of emergency clinics where available. This information, of course, is best conveyed via a practice brochure or the use of in-hospital closed circuit TV.Build Lasting Relationships with your clients so they can bring their pet continuously in your hospital or clinic.

Now a day's veterinary clinical practice is developed like anything we can compare our practice to the human practice. Veterinary practice is now becomes corporate means our client base is especially in small animals is from very rich peoples to common everyone keeps dog, cat

etc. Recent trends of veterinary client or pet owner wants quick response and treatment as early as possible because their pets are their family members. Pet owners are ready to pay whatever you charge for treatment of their pets but clients need punctuality, availability of veterinarians on your clinics at least which you mentioned your time of clinic unless of an emergency condition.

Client management in clinical practice is very difficult task because there are different types of mentality of pet owners came to your hospitals. Number of owners are very short tempered they doesn't have patience to wait for treatment, they required very qualified or senior vet to check their pets. For such clients you have to maintain separately by giving your comfortable free time, avoid such client in heavy rush time of your OPD time. Some owners misguide you to treat by providing wrong history that you have to collect a proper data by asking very close questions match to physical symptoms. We have to keep proper data of client based, Breed wise, disease wise, type of cases either surgery ,medicine or gynaecological case so we can manage our inventory record which helps us if there is issue of legal case. Pet owners are very forward they knows symptoms of their patient and they are updates their knowledge by using international knowledge providing search engine like www.google.com etc. so for such clients which is highly educated we must have to updates our knowledge so we can handle this type of owners in our routine veterinary practice.

Daily routine practice we must learn how to tackle client of different variety. We must listen silently their problems; symptoms collect as much as history which is very important for diagnosing the case and provide them facility as possible as you can with comfortable manner. You must check their pets with love affection handle them by touching, check properly, thoroughly with your own as a veterinary don't depend on your nursing staff it gives good feedback from client mind set. Avoid as possible as by gossips and other our colleagues veterinary doctors if clients previously to any other vet. Love our profession we serve the livestock's and proud to be a veterinary practitioner.

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Estrous cycle and its endocrinology of the Pet animals

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Sound knowledge of the bitch reproductive cycle is essential. Individual bitches may vary from normal, be presented at variable times during their estrous cycles for evaluation, and sometimes exhibit pathologic variations in cycles. Each of these scenarios requires veterinary interpretation. The normal canine reproductive cycle can be divided into 4 phases, each having characteristic behavioral, physical, and endocrinologic patterns, although considerable variation exists. Bitches with normal estrous cycles but unexpected patterns must be differentiated from those with true abnormalities. Detection of individual variation within the normal range of events in a fertile bitch can be crucial to breeding management. Evaluation of the estrous cycle for true abnormalities is an important.

PHYSIOLOGY OF THE ESTROUS CYCLE

Estrous Cycle

The onset of the first estrous cycle (puberty) of an individual bitch is expected between 6 and 10 month of age but may not begin until she has reached 2 years of age. The interestrous interval is normally 4-13 month, with 7 month the average.

Anestrus

The anestrus phase of the extrous cycle normally lasts 16 month. It is marked by ovarian inactivity, uterine involution, and endometrial repair. An anestrous bitch is not attractive or receptive to male dogs. No overt vulvar discharge is present, and the vulva is small. Vaginal cytology is predominated by small parabasal cells, with occasional neutrophils and small numbers of mixed bacteria.

The endoscopic appearance of vaginal mucosal folds is flat, thin, and red. The physiologic controls terminating anestrus are not well understood, but the deterioration of luteal function and the decline of prolactin secretion seem to be prerequisites. The termination of anestrus is marked by an increase in the pulsatile secretion of pituitarygonadotropins, follicle stimulating hormone (FSH), and luteinizing hormone (LH), induced by gonadotropin-releasing hormone (GnRH). Hypothalamic GnRH secretion is itself pulsatile, its intermittent secretion is a physiologic requirement of gonadotropin release. Mean levels of FSH are moderately elevated, and those of LH slightly elevated, during anestrus. At late anestrus, the pulsatile release of LH increases, causing the proestrous folliculogenesis. Estrogen levels are basal (2-10 pg/mL) and progesterone levels at nadir (<1 ng/mL) at late anestrus.

Table - Some aspects of the estrous cycle in the bitch				
	Duration	*Progesterone*	*Estrogen (E_2) levels*	*Notes*
Anestrus	1-6 month	Basal (<1 ng/mL)	Basal (2-10 pg/mL)	
Proestrus	days-3 weeks (9 days average)	Initial basal (<1 ng/mL); At LH surge: 2-3 (0,8-3) ng/mL At day of ovulation: 4-10 ng/ml (ovulation: range 1-8 ng/mL)	Rising to peak levels (50-100 pg/mL)	
Estrus	days-3 weeks (9 days average)		Abrubt decrease at the day of LH peak to 10-20 pg/mL Basal (2-10 pg/mL) during the following few days	Primary oocytes ovulate 2 days after LH peak; Oocyte maturation finished 2-3 days later; lifespan of secondary oocytes: 2-3 days
Diestrus	2-3 month (in the absence of pregnancy)	Peak at 15-80 ng/mL	Basal (2-10 ng/mL)	

Proestrus

During proestrus, the bitch becomes attractive to male dogs but is still not receptive to breeding, although she may become more playful. A serosanguineous to hemorrhagic vulvar discharge of uterine origin is present, and the vulva is mildly enlarged. Vaginal cytology shows a progressive shift from small parabasal cells to small and large intermediate cells, superficial-intermediate cells, and finally superficial (cornified) epithelial cells, reflecting the degree of estrogen influence. RBC are usually, but not invariably, present. The vaginal mucosal folds appear edematous, pink, and round. FSH and LH levels are low during most of proestrus, rising during the preovulatory surge. Estrogen rises from basal anestrous levels (2-10 pg/mL) to peak levels (50-100 pg/mL) at late proestrus, while progesterone remains at basal levels (<1 ng/mL)

until rising at the LH surge (2-3 ng/mL). Proestrus lasts from 3 days to 3 weeks, with 9 days average. The follicular phase of the ovarian cycle coincides with proestrus and very early estrus.

Behavior correlates with decreasing estrogen levels and increasing progesterone levels. Serosanguineous to hemorrhagic vulvar discharge may diminish to variable degrees. Vulvar edema tends to be maximal. Vaginal cytology remains predominated by superficial cells; RBC tend to decrease but may persist throughout. Vaginal mucosal folds become progressively wrinkled (crenulated) in conjunction with ovulation and oocyte maturation. Estrogen levels decrease markedly after the LH peak to variable levels, while progesterone levels steadily increase (usually 4-10 ng/mL at ovulation), marking the luteal phase of the ovarian cycle.

Estrus

Estrus lasts 3 days to 3 wk, with an average of 9 days. Estrous behavior may precede or follow the LH peak—its duration is variable and may not coincide precisely with the fertile period. Primary oocytes ovulate 2 days after the LH peak, and oocyte maturation is seen 2-3 days later; the lifespan of secondary oocytes is 2-3 days.

Diestrus

During diestrus, the normal bitch becomes refractory to breeding, with diminishing attraction of male dogs. Vulvar discharge diminishes and edema slowly resolves. Vaginal cytology is abruptly altered by the reappearance of parabasal epithelial cells and frequently neutrophils. The appearance of vaginal mucosal folds becomes flattened and flaccid.

Estrogen levels are variably low, and progesterone levels steadily rise to a peak of 15-80 ng/mL before progressively declining in late diestrus. Progesterone secretion depends on both pituitary LH and prolactin secretion. Proliferation of the endometrium and quiescence of the myometrium develop under the influence of elevated progesterone levels. Diestrus usually lasts 2-3 mo in the absence of pregnancy. Parturition terminates pregnancy 64-66 days after the LH peak. Prolactin levels increase in a reciprocal fashion to falling progesterone levels at the termination of diestrus or gestation, reaching much higher levels in the pregnant state. Mammary ductal and glandular tissues increase in response to prolactin levels.

Estrogens

Increased estrogen causes an increased turnover rate of vaginal epithelial cells, resulting in the progressive cornification seen on vaginal cytology. Progressive edema of the vaginal mucosa also develops and can be visualized with endoscopic examination. Estrogen assays are performed by many commercial laboratories; however, the information is of little value for ovulation timing because peak estrogen levels vary from bitch to bitch, and even relative changes do not correlate to ovulation or the fertile period.

Estrogen is best assessed by serial vaginal cytologies and vaginoscopy. Estrogen levels do not indicate the fertile period because ovulation is triggered by the LH surge, not an estrogen peak. Examination of the cells on the surface of the vaginal epithelium can provide information about the stage of the estrous cycle. Proper technique is important so that the cells obtained are representative of the hormonal changes occurring. The sample should be collected from the cranial vagina; cells from the clitoral fossa, vestibule, or caudal vagina are not as indicative of the stage of the cycle. Under the influence of rising estrogen levels, the number of layers composing the vaginal epithelium increases dramatically, presumably to provide protection to the mucosa during copulation. As estrogen rises during proestrus, the maturation rate of the epithelial cells increases, as does the number of keratinized, cornified epithelial cells seen on a

vaginal smear. Full cornification continues throughout estrus until the "diestral shift" occurs 7-10 days after the LH surge, signifying the first day of diestrus.

The vaginal smear then changes abruptly, with appearance of neutrophils and epithelial cells changing from full cornification to 40-60% immature (parabasal and intermediate) cells over the next 24-36 hr. If vaginal cytology is performed until the diestral shift is observed, the LH surge, ovulation, and the fertile period can be analyzed retrospectively.

Luteinizing Hormone

At the end of the follicular phase of the estrous cycle, a marked increase in LH over usual baseline values devlops over 24-48 hr, followed by a return to baseline values. This surge is thought to occur in response to the decline in estrogen levels and increase in progesterone levels. The LH surge triggers ovulation, making it the central endocrinologic event in the reproductive cycle of the bitch. Daily serial measurement of LH to identify the exact date of the LH surge is an accurate diagnostic tool for timing breedings. Affordable semiquantitative in-house kits are available for measuring serum LH levels in the dog and for identifying the preovulatory LH surge and thus the time of ovulation and the true fertile period. Blood samples must be drawn daily (at about the same time) for LH testing, as the LH surge may last only 24 hr in many bitches. The kits can be subject to variable interpretation, so the same person should run the tests if possible.

Progesterone

Progesterone levels begin to rise at approximately the time of the LH surge (prior to ovulation). Rising progesterone acts synergistically with declining estrogen to reduce edema of the vulva and vagina, which can be seen on vaginoscopic exam. Other observable clinical signs are minimal. Serial blood samples performed every 2 days may identify the initial rise in progesterone (usually >2 ng/mL), which indicates that the LH surge has occurred. Progesterone can be assayed by radioimmunoassay at most veterinary commercial laboratories. Several in-house semiquantitative kits are also available. No single absolute value of progesterone correlates to any particular stage of the cycle. Progesterone varies from 0.8-3.0 ng/mL at the point of the LH surge, from 1.0-8.0 ng/mL at ovulation, and from 4.0-20.0 ng/mL during the fertile period. However, if accurate serial quantitative progesterone assays are obtained, the LH surge may be estimated as the day a distinct increase in progesterone level is seen. While this is not as accurate as actual identification of the LH surge by assay, estimation by progesterone levels is still very useful and is often more widely available and convenient.

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Methods of oestrous detection and prediction of ovulation in canine

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The methods used to determine the optimum breeding time in bitches are as follows:

- Sexual behaviour
- Vaginal exfoliative cytology
- Vaginoscopy
- Hormone assays
- Ultrasonography

Vaginal exfoliative cytology:

A series of vaginal smear examination establishes the stage of estrous cycle with greater accuracy and help to know optimal timing of mating or artificial insemination in small animals (bitch, queen) but not in mare, ewes, cows and sows etc.

For sample collection, restrain the animal. Elevate the tail and clean the valva. Introduce sterile swab well in to vagina using lubricated speculum. Roll the swab on a pair of clean microscope slides. Air dry and stain with Wright's or Giemsa stain with same techniques used for blood. Interpret the cytological findings keeping in mind the history and clinical signs.

The canine estrous cycle consists of 4 phases: proestrus, estrus, diestrus and anestrus. Proestrus and estrus are commonly called "heat" or "season". During proestrus, the start of the estrous cycle, the bitch attracts male dogs, but is still not receptive to breeding. She may become more playful and passive as proestrus continues. A blood tinged vaginal discharge (of uterine origin) is present and the vulva is moderately enlarged and turgid. The cells from vaginal cytology smears change over a period of 4 to 7 days from non-cornified (small "parabasal" cells, small and large "intermediate" cells) to cornified cells ("superficial-intermediate" cells, and "anuclear" cells). These changes in the vaginal cytology reflect the increasing estrogen from the ovarian follicles. Red blood cells are usually, but not invariably present. Proestrus can last from 3 days to 3 weeks, with 9 days the average. Proestrus progresses to estrus.

During estrus, the normal bitch displays receptive or passive behavior, enabling breeding. Vaginal discharge normally diminishes. Vulvar edema tends to be maximal and the vulva is flaccid. Vaginal cytology during estrus consists of 80 to 100 percent cornified ("superficial" and "anuclear") cells. Red blood cells tend to diminish, but sometimes persist throughout estrus. Estrus can last 3 days to 3 weeks, with 9 days being the average. Receptive behavior begins when estrogen concentrations decline and progesterone concentrations increase. The duration of receptivity to male dogs is variable, and may not coincide precisely with the fertile period, which occurs during estrus. Ovulation is triggered by a surge in luteinizing hormone (LH) from the pituitary gland in the brain. Ovulation of immature, infertile primary oocytes (eggs) begins approximately 2 days after the LH surge and oocyte maturation occurs over the following 1 to 3 days. The life span of the secondary (fertile) oocytes is 2 to 3 days. Thus, the bitch's actual fertile period extends from 3 through 6 to 7 days after the LH surge. The LH surge occurs at the same time as an initial increase in progesterone concentration, enabling ovulation timing by measurement of either hormone.

Ovulation timing should be performed using a combination of serial vaginal cytologic exams and ideally, serum progesterone concentrations. Testing for LH can be used for some

cases (infertility, frozen breedings). Vaginal cytology exams are started during the first few days of proestrus and performed every 2 to 3 days. When >70% of the epithelial sells are cornified ("superficial" cells), serum progesterone concentrations are obtained every 48 hours to detect the day of the initial progesterone rise (usually between 2-3 ng/ml), which correlates with the LH surge triggering ovulation. That day is called "day zero". The bitch is most fertile, and can be bred with good conception rates, between 2 and 7 days after "day zero". The number of breedings and specific day(s) of breedings depends on the type of semen (fresh, chilled/extended, or frozen).

If LH testing is used to provide the most precise ovulation timing, daily serum samples must be acquired for testing once the vaginal cytology contains >70% "superficial" cells. The initial rise in progesterone or the occurrence of the LH surge is confirmed 48 hours later by running an additional progesterone test. To economize ovulation timing, daily serum samples can be saved (refrigerated or frozen) and selected for later LH testing based on the estimated initial rise in progesterone.

Following estrus and breeding the bitch enters diestrus. During diestrus, the normal bitch becomes refractory to breeding and less attractive to male dogs. Vaginal discharge becomes mucoid and diminishes and vulvar edema slowly resolves.

Vaginal cytology is abruptly altered by the reappearance of noncornified ("parabasal") epithelial cells and, frequently, white blood cells. Diestrus usually lasts 2 to 3 months in the absence of actual pregnancy. Bitches normally experience false pregnancy if not actually pregnant. Whelping occurs 64 to 66 days after the LH surge, or 56 to 58 days after the onset of diestrus as determined by vaginal cytology.

Following diestrus the bitch enters true anestrus. The interestrus interval (period between outwardly apparent heat cycles) consists of diestrus and anestus and normally varies from 4 1/2 to 10 months in duration, with 7 the average. The anestrus phase is characterized physically by apparent reproductive inactivity, although hypothalamic, pituitary and ovarian hormonal fluctuations are occurring. During anestrus, the uterus is undergoing recovery and repair following a false or true pregnancy. The normal bitch is neither attractive nor receptive to male dogs. Little vaginal discharge is present, and the vulva is relatively small. Vaginal cytology taken during anestrus finds small "parabasal" cells, with occasional white blood cells and small numbers of mixed bacteria representing normal flora. Anestrus normally lasts from 1 to 6 months before the bitch enters proestrus again.

Classification of Cells

- The different cell types represent stages of cell death. As healthy round vaginal cells die, they then become larger and more irregular in shape.
- The nucleus becomes progressively smaller, then pyknotic, before disintegrating, leaving a nuclear "ghost" and then anuclear cell.

Cytologic Staging of the Cycle

Stage	Red Blood Cells	Neutrophils	Bacteria	Epithelial Cells
Early Proestrus	Usually	Often	Many	Parabasal, intermediate, superficial intermediate, few superficial cells
Late Proestrus	Usually	Few or none	Many	Superficial Intermediate and Superficial
Estrus	Present, may be decreased in number or may be absent	None	Many	> 80-90% superficial or cornified cells

Diestrus	Some to none	Many	May be clearing	Parabasal, Intermediate, decreased superficial
Anoestrus	None	Few or none	None	Parabasal and Intermediate cells

Vaginal exfoliative cell types seen during different stages of the canine estrous cycle

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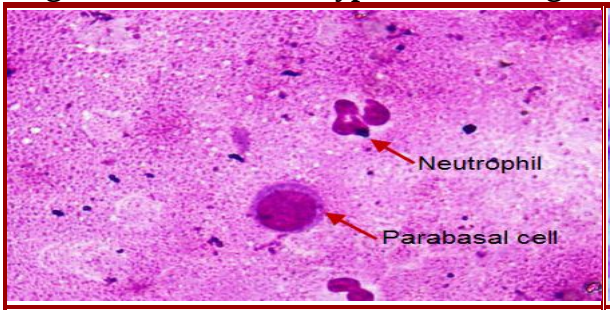
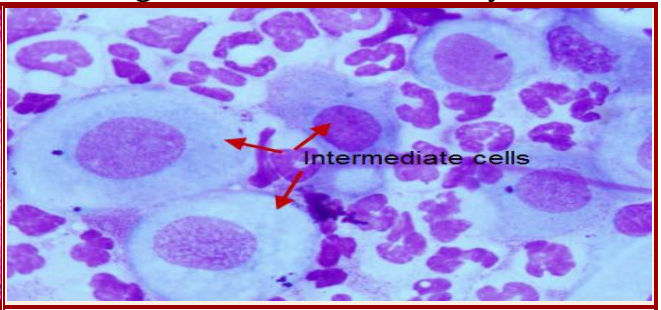
<tr><td> 
</td><td> 
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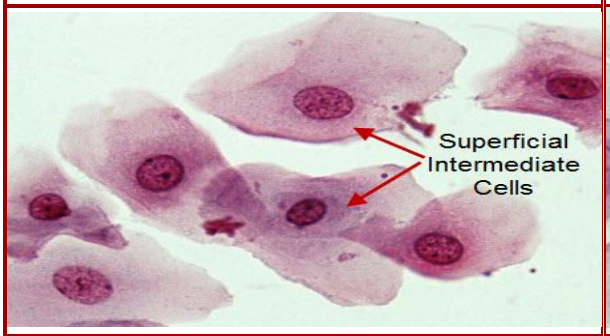
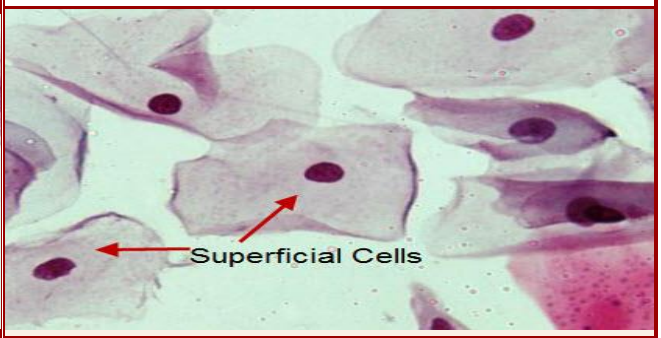
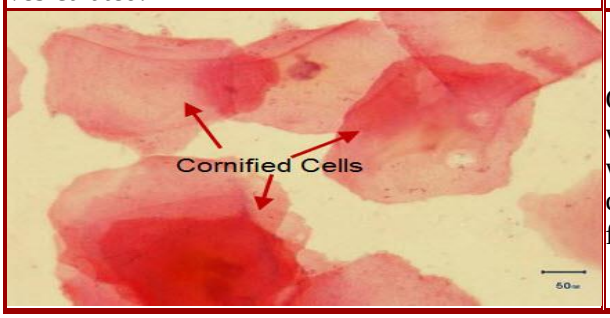
<tr><td>PARABASAL CELLS
Healthiest and smallest of vaginal cells. Round or slightly oval with large vesiculated nucleus and Relatively small amount of cytoplasm</td><td>INTERMEDIATE CELLS
Vary in size from slightly larger than parabasal cell to twice its size. Cells have oval to rounded irregular borders and vesiculated nucleus which is smaller than those found in parabasal cells. Change in cell morphology reflects the first stage in cell death.</td></tr>
<tr><td> 
</td><td> 
</td></tr>
<tr><td>SUPERFICIAL INTERMEDIATE CELLS
Cells have flat, angular cytoplasmic borders typical of superficial cells but nucleus is still vesiculated.</td><td>SUPERFICIAL CELLS
Largest cells identified in vaginal cytology. Cells have sharp, flat angular borders and small, pyknotic, fading or ghost nucleii.</td></tr>
<tr><td> 
</td><td>CORNIFIED CELLS

Cornified cells are large, dead irregular vaginal cells with no nucleus. These cells end the process that began with the round parabasal cells. These cells are also called as anuclear superficial cells, fully keratinized or fully cornified cells.</td></tr>
</table>

Vaginoscopy:

Vaginoscopy can be used to aid in timing of natural breeding. Vaginal mucosa in proestrus appears rounded and odematous. Wrinkling of mucosa is associated with LH surge. This is the time to begin breeding. Breeding should be continued throughout the phase of maximum mucosal wrinkling, seen as angulated folds of vaginal mucosa with sharp profiles.

Breeding should be discontinued when the vaginal mucosa becomes flaccid and smooth, with patchy red and white surface which is observed 6 to 10 days following LH surge.

Flexible or rigid fiber optic endoscope or pediatric proctoscopes less than 12mm in diameter can be used for Vaginoscopy. Very small bitches and queen may be examined with an otoscope. Before endoscopic examination, case history has been obtained and a general physical examination has been completed. The colour and character of any vulvar discharge are noted.

Hormone Assays:

The most accurate way to predicting ovulation in a bitch. Progesterone levels begin to rise after proestrus and increase throughout heat. They reach specific levels through ovulation, which mean that this is a quantitive way of predicting ovulation, therefore a more accurate way. Serum should be assessed beginning 3 or 4 days after onset of proestrus and should be continued every other day. Ovulation takes place when values of progesterone are between 4-10ng/ml, mating must be done at this time and second mating should be done 48 hours later to achieve maximum litter size.

Ultrasonography:

Ultrasound is another recent method of detecting ovulation. There are number of follicles are visible through ultrasound but we can't correlate the number of oocytes that actually ovulate so we do not use to monitor bitches for breeding but it is useful in diagnosis of cystic ovarian disease, pregnancy or pyometra.

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Advances in pregnancy diagnosis in Pet animals

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Bitch owners frequently wish to know whether their bitches are pregnant following a planned mating, principally out of curiosity but also so that adequate plans can be made in advance of the anticipated whelping date. For example owners can plan in advance, so that they can be present at the whelping or make suitable alternative arrangements; Prepare the whelping area at leisure; Better manage the bitch in respect of exercise, feeding and medication or, avoidance of medication

Complications:

Pregnancy diagnosis in bitches is not easy because of two complications. First, the hormone levels are very similar in pregnant and non-pregnant bitches, certainly in mid metoestrus, 4-5 wk after mating when diagnosis is most commonly required. Secondly, canine spermatozoa can remain viable in the female genital tract for up to 7 d. Thus a bitch presented for diagnosis 3 wk after mating may two wk pregnant. It is for this reason that the traditionally quoted duration of pregnancy 63 d from the date of first mating is not regularly reliable. A range of potential whelping dates 58-68 d from the date of first mating is more likely to be correct in fact only

Pregnancy diagnosis in Bitch:

Several methods are used for pregnancy diagnosis in the bitch. Some methods, such as evaluation of the teats, weight gain, and abdominal enlargement are not reliable methods of pregnancy detection. More reliable methods include abdominal palpation (at 30 days gestation), measuring the level of relaxin in the blood (after 26 days gestation), ultrasound of the abdomen (starting at 21-30 days gestation), and radiographs (after 45 days gestation). These methods vary in their ability to positively identify the number of fetuses and fetal viability.

Palpation

Small ovoid swellings can be palpated along the uterine horns 21-30 days post breeding. These swellings double in size every week until days 35 to 38; they then become confluent and difficult to differentiate. Palpation is not as reliable as other methods for evaluating pregnancy. It is difficult without experience to determine the exact number of fetuses and fetal viability cannot be verified with palpation.

Ultrasound

Ultrasound is the best method used to visualize and evaluate the pups in the womb. Ultrasound is best done 21-30 days after breeding. Ultrasound done prior to 21 days can give false negatives. Ultrasound is a good method of distinguishing between pregnancy and pyometra. Fetal viability can be determined by ultrasound through visualization of the fetal heart. The number of fetuses can be estimated, though it can be less accurate in bitches carrying larger litters, or when done later in gestation.

Radiographs

Twenty-eight days into pregnancy, the bones of the fetus begin to become calcified. At 42-45 days of pregnancy, radiographs can be taken to visualize the pup's in-utero. However, for best results, radiographs should be taken more than 47 days post breeding, when fetal skeletons are more readily visible. Radiographs can help determine the number of fetuses, though it is less

accurate in bitches carrying large litters. Fetal viability cannot be determined by radiograph unless advanced lesions exist in the fetus, such as skeletal collapse or gas within the fetus.

Relaxin

Relaxin is a hormone that is produced by the placenta and found in the bloodstream of the bitch when the fertilized egg implants in the uterine wall, about 21 days post breeding. This hormone continues to circulate through the blood throughout pregnancy, peaking around day 40 to 50. ReproCHEK is the current test on the market that measures relaxin. This test can be used to determine pregnancy and can be used continuously throughout the pregnancy to verify if a total abortion has occurred. Pseudopregnant bitches do not test positive for relaxin. This test cannot determine the number of fetuses present in the bitch. A false negative is possible if the litter size is very small.

THE DEVELOPMENT OF A NEW BIOLOGICAL TEST

As noted earlier, it is not possible to detect pregnancy in the bitch by the measurement of oestrogen, progesterone levels in the blood or urine. However, it was discovered that a major change in the levels of acute phase proteins in the blood occurs in pregnant animals but not in healthy non-pregnant bitches. Recently a pregnancy diagnosis test based on the measurement of these proteins in the blood has been developed by Cambridge.

In the early stages of an inflammatory response the concentration of a number of plasma proteins is significantly increased. These acute phase proteins (APPs) act as mediators and inhibitors of inflammation, as scavengers of cell-derived products from damaged tissues and they may also influence the immune response which accompanies inflammation. This acute phase response may be triggered by bacterial and viral infections, shock or trauma. The rise that occurs during pregnancy in bitches may be triggered by bacterial and viral infections, shock or trauma. It is postulated, in response to the blastocyst hatching which does not occur until almost 3 wk after ovulation. The reaction of the uterus to this foreign antigen appears to produce an antigenic reaction which results in uterine swellings around the blastocyst and eventually there is an elevation of acute phase protein production from the Veterinary Sciences.

The production of APPs is not pregnancy specific and it will be apparent that bitches suffering from some infection, eg metritis, could be falsely diagnosed as pregnant by this method as acute phase proteins will be raised in such circumstances. Trials have shown, however, that provided animals presented for pregnancy diagnosis are checked for obvious signs of illness and only healthy bitches are sampled, then problems do not generally arise, possibly because usually only healthy young bitches are used for breeding. Studies have shown that pyometra cases will show positive in the test but usually that condition occurs rather later post oestrus when testin is advocated.

Miscellaneous:

Abdominal girth measurement:

It is inexpensive and non-invasive method of pregnancy diagnosis in bitch by measuring the abdominal girth from same area, just posterior to last rib. Abdominal girth can be start to measure prior to breeding weekly starting at 3-4 weeks up to the week of whelping. The increase in abdominal girth at five weeks after successful mating 13 % > the girth prior to breeding then bitch can be declared as pregnant.

Infertility problems and possible interventions in Pet animals

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In comparison to the last 10 or 20 years, veterinarians are now frequently requested to solve fertility problems in the dog, mainly due to the increased popularity of purebred dogs as well as for sentimental or financial reasons. In fact, breeders may be concerned about different kinds of problems which includes their bitches produce no pups after mating or Artificial Insemination, anoestrus or a low prolificity rate.

Many different problems can lead to infertility including hormonal problems, infectious diseases, congenital or acquired defects of the genital tract. We recommend that veterinarians follow a very strict progression, commencing with the most frequent cause to the most uncommon cause. In the bitch, things become even more difficult when we realize that apparent infertility can be due to very different situations which prove difficult to distinguish, such as lack of fertilization (no union between eggs and sperm) and early embryonic death.

1. Mistimed Breeding

Mistimed breeding represents by far the most common cause of infertility in the bitch. The incidence may vary between 40% to 80% of infertile bitches. Although the following belief has been clearly proved erroneous in approximately 30% of bitches, many breeders are still convinced that a bitch will conceive when mated around the 12th day of the heat period. Actually, it has been clearly demonstrated that a bitch may ovulate as early as the 3rd or 4th day after the onset of pro-oestrus and as late as the 30th day of the heat period. Clinical factors such as the increased swelling of the vulva and the decreasing bleeding of the genital tract at the time of the ovulation period may help veterinarians determine if bitches are obviously mated at a wrong time. But, these criteria are in no way precise enough. Nowadays, numerous available techniques may be utilized to investigate the "optimal time for breeding": vaginal cytology, endoscopic appearance of vaginal folds and progesterone assays are the most commonly used methods. Ovarian ultrasonography is especially indicated in infertile bitches, as it represents the most accurate way to determine the precise date of ovulation. This method also helps quantify the number of growing follicules as well as follicules undergoing ovulation and as such, it helps to evaluate the fertility potential of the bitch.

2. Male infertility

After mistimed breeding, male infertility is the most common cause of conception failure in bitches presented with infertility.

3. Other causes of Infertility

As it is often ascertained in large animals, we suggest that veterinarians should first consider if infertile bitches show regular inter-oestrus intervals or irregular ovarian cycles.

3.1. Infertility with prolonged interestous intervals

The apparent prolongation of interestrous intervals occurs in dogs with a silent heat, defined as ovarian activity in the absence of overt physical and behavioral changes characteristic of canine estrus. Among the hormonal causes of anoestrus, hypothyroidism, hyperadrenocorticism, hyperprolactinemia, or bitches treated with hormonal compounds such as progestagens, androgens (racing dogs) or anabolic steroid compounds. Ovarian cysts that secrete progesterone may cause prolonged interestrous intervals. The surgical removal of the cyst is

often the best solution. Hormonal attempts using prostaglandins are not well documented in bitches. Bitches housed in very bad environmental conditions (including a high concentration of animals, low luminosity, low quality food) may undergo prolonged interestrous intervals.

3.2 Infertility with shortened interestrous intervals:

As it is often ascertained in large animals, we suggest that veterinarians should first consider if infertile bitches show regular inter-oestrus intervals or irregular ovarian cycles.

3.2.1. Ovarian cysts or tumors

It is important to diagnose and remove these hormonally active cysts or tumors as quickly as possible for at least two reasons. First, it is necessary to cure them and try to restore fertility. Second, the secretion of high quantity of estrogens may act on the uterus as a potentialtowards the cystic endometrial hyperplasia-pyometra complex, but also on the bone marrow in creating progressive non regenerative anemia.

Follicular cysts may be single or multiple; if multiple cysts are present in one ovary, the cysts do not communicate. The ovarian cysts in the bitch may be present in only one or both ovaries. Estradiol assays during the pro-estrus period may be useful for practitioners who do not perform ultrasonography. The patterns of estrogen secretion are often modified. Still, when possible, it is much more valuable to perform ovarian ultrasonography. Follicular cysts appear as focal hypoechoic to anechoic structures, greater than one cm are supposed to be cystic structures. The treatment of choice of ovarian follicular cysts is ovariectomy or ovario-hysterectomy when the uterus is damaged. It is recommend induction of luteinization of the cystic follicles, using GnRH or HCG. But such protocols, as they increase the progesterone plasma level after a prolonged period of estrogen secretion and this treatment very often leads to the occurrence of pyometra in the following weeks. Surgical removal of a cyst, or aspiration of a cyst with a fine sterile needle under laparotomy or ultrasonography may be new alternatives in therapy.Granulosa cells produce estrogens; and therefore, it is not surprising that **GCTumours** often lead to infertility with prolonged heat periods.

(**Note:** Exogenous estrogens administered to elderly ladies receiving treatment after the end of genital activity with estrogens containing gels can penetrate through the skin of miniature breeds when they are frequently handled on the forearm and cause prolonged estrus signs).

Premature decline in progesterone

Split heats are defined as successive short proestrus signs, at intervals of 2 weeks to 2 months, associated with short interestrus intervals. This pattern is more often observed in young bitches and leads to no real infertility in the rest of the genital life of the bitch. Anovulatory cycles are not frequent in bitches. In such anovulatory cycles, serum progesterone level never increases above 3.5 to 6 ng/ml. That's why the following heat period will often occur earlier than usual.

Bitches may also suffer from hypoluteoidism, which is the lack of progesterone secretion during pregnancy which makes the pregnancy impossible to maintain. Some breeds are well known to express **hypoluteoidism,** like Rottweilers and German Shepherds. Progesterone supply can be given parentally (progesterone in oil: 2 mg/kg every 3 days; allytrenbolone). In France, veterinarians often use oral micronised progesterone which is currently given to women.

A "**short anoestrus syndrom**" has also been described in Rottweilers and German Shepherds.Early embryonic or fetal death remains most of the time impossible to detect, as no vulvar swelling occurs in general. Possible causes include endometritis, cystic endometrial hyperplasia, embryonic defects and possibly inbreeding.

3.3 Infertility with normal interestrous intervals

3.3.1 Hormonal Problems

Hormonal defects may be suspected also in bitches with regular interestrous intervals, and veterinarians should control the hormonal status of the bitch during the heat period and also during pregnancy.

3.3.2 Infectious diseases

Many infectious agents have been suspected to induce infertility in bitches. Several **viruses** have been shown to play a potential role in canine infertility. **Canine Herpes Virus (CHV)** is well known to have a pathogenic action on neonate pups. Several elements suggest however that CHV may well act on infertility in the bitch.

Tranplacental infection by **Canine Distemper Virus** has been shown in experimental conditions. Recently, potential incidence of a parvovirus **Minute Virus of Canines (CPV1)** on resorption during the first half of pregnancy.

The incidence of b**acterial infections** on canine infertility is better documented. **Canine Brucellosis,** which is well known as an abortive agent during late pregnancy, could also generate early embryonic or fetal death through endometritis.

Other **specific bacterial diseases** have been suspected to act on canine infertility. However, **usual genital bacteria** may play a real role on infertility. **Canine Mycoplasms and Ureaplasms** are commonly isolated in the genital tract of fertile and infertile bitches. But it has been shown that there is a higher incidence of these agents in the vagina of infertile bitches. Many bacteria are commonly isolated from the uterus and the vagina of normal fertile bitches. It has been shown that in case of vaginitis, there are significant qualitative and quantitative variations. Strong evidence exists that bacteria causing vaginitis may lead to infertility. It may well have been underestimated due to lack of specific clinical signs and due to the difficulty of the clinical examination of the vagina of the bitch.

The role of **parasitic infections** on infertility is better documented. Recent experimental data suggest that Neospora caninum could cause early fetal death in the bitch.

3.3.3 Drugs induced infertility

In practice, many breeding bitches may be treated with drugs that may contribute to the decline of fertility. Steroid hormones and anti-fungal compounds may create hormonal defects in prepuberal or adult bitches. Abortive drugs such as prostaglandins, antiprogestins and antiprolactinic substances have to be avoided during pregnancy.

3.3.4. Anatomical abnormalities of the vulva, vestibule or vagina

Some bitches do not manage to mate because of **congenital abnormalities** of the posterior genital tract (vulva, vestibule or vagina). **Acquired diseases** or abnormalities of the posterior genital tract (scars after a bad parturition, episiotomy, violent mating) may also lead to the lack of copulation.

3.3.5 Uterine Pathology

Endometritis is a common cause of infertility in mares. In bitches, however, it is hard to diagnose. Endometrial smears, eventually performed after endoscopic canulation of the cervix, may be valuable. Bitches with **cystic endometrial hyperplasia** (CEH) are often infertile due to implantation failure after conception. Somehow, ultrasonography usually permits the visualization of the glandular endometrium. One succesful therapy has been described with mibolerone oral administration, 30 microgrammes per 25 lb body weight daily during 6 months. CEH often leads to pyometra, which may be treated in many cases by a mixed treatment using prostaglandins and antiprogestins (aglepristone). A healing of the endometrium seems to occur, as many bitches may have successful pregnancies at their next heat period.

3.3.6 Abnormal sexual behaviour

Many psychological factors may influence sexual receptivity in bitches. Psychology may influence factors like ovulation or early embryonic death in the bitch.

3.3.7 Miscellaneous causes

Bitches with systemic diseases like diabetes mellitus, hyperadrenocorticism or renal insufficiency may likely be infertile. Finally, breeders stress nutrition when their breeding kennel suffers from decreased reproductive results. Little is known in this regard.

Infertility in the queen

The main causes of diagnostic procedure in the queen resembles to what is done for the bitch. However, the main cause of infertility in practice is probably the mating or the ovulation failure. Infectious diseases are probably an important factor, too, although they remain often under-diagnosed. Ovarian and uterine pathology, uterine pathology, chromosomal defects and nutritional factors are other common causes of infertility or sub-fertility in this species.

Clinical approaches of infertility in the bitch

Veterinarians should consider that a high proportion of bitches with a presenting complaint of infertility probably ovulate before or after day 12. Also, onset of male receptivity may be delayed for several days after ovulation, which may complicate interpretation of clinical data.

The following data base should be recorded for all previous seasons for which information is available:

1. Date of onset of proestrual bleeding
2. Date of onset of first receptivity
3. Breeding(s): dates, out/inside tie, AI, fresh vs frozen semen
4. Date of first refusal of mating
5. Male fertility, age, semen colture
6. Brucella canis antibody status of the bitch and dog
7. Pregnancy status at 28 days
8. Previous normal whelping(s)/litter(s)
9. Previous signs of false pregnancy
10. Previous reproductive disease
11. Previous hormonal therapy

Importance of progesterone assay:

Serum progesterone assay is important to follow the canine estrus cycle as the bitch progresses towards ovulation, to confirm that she ovulated and maintained a 60-day luteal phase, to gain more information on fetal survival in case of problems during pregnancy, as well as to decide whether or not to use prostaglandin F2a in pyometra cases.

Breeding Management:

Timing ovulation is of utmost importance in achieving good conception rate as well as in solving cases of canine infertility. Canine proestrus and estrus last on average 9 days each with ovulation taking place 3 days after onset of estrus (or day 12 after onset of proestrus). However ovulation can occur as early as 5 days or as late as 27 after onset of proestrus. Therefore, it is very important to check the female's behavior, perform vaginal smears every 2-3 days starting on thefirst day of proestrus in order to catch early ovulators, and draw blood samples to measureprogesterone once behavior and/or vaginal smear indicate estrus. Estrus is indicated by acceptance of the male or by a degree of vaginal cornification of >70. Ovulation occurs 3 days after onset of estrus. Serum progesterone has a concentration of (values are approximate) 2.0-3.0

ng/ml on the day of the peak of luteinizing ormone (LH), 4.0-8.0 ng/ml on the day of ovulation, 10-25 ng/ml during the 2 days following ovulation, which is when oocytes are reaching maturity in the ampulae of the oviducts and fertilizations are taking place. Ovarian structures can be visualized with ultrasound using 5.0 to 7.5 sectorial MHz probes; follicular growth can be followed and ovulation can be estimated based on disappearance of the hypoechogenic areas representing follicles (which become luteinized) and on appereance of a hypoechogenic area at the periphery of the ovary representing follicular fluid accumulation within the ovarian bursa

Pyometra:

Apart from mismating, reproductive diseases play a big role in canine infertility. Pyometra may be treated by following drugs:

Prostaglandin F2α	Daily dosage in the bitch/queen	N° treatments/day
Natural PGF2α or Dinoprost	Bitch - 100 μg/kg (0.1 mg/kg) Queen - 500 μg/kg (0.5 mg/kg)	2 2
Cloprostenol	Bitch - 1-5 μg/kg (0.001-0.005 mg/kg) Queen - 5 μg/kg (0.005 mg/kg)	1 1
Alfaprostol	Bitch - 20 μg/kg (0.02 mg/kg)	2

Antiprolactinic drugs can also induce luteolysis if administered during the second half of diestrus.

Antiprolactinic	Daily dosage in the bitch/queen	N° treatments/day
Cabergoline	5 μg/kg	1
Bromocriptine	10-30 μg/kg	(*) 2
Metergoline	500 μg/kg	(*) 2

There is no scientific information available for the queen.Progesterone antagonist's act by blocking progesterone receptors, causing opening of the cervix and in most cases a resumption of miometrial contractility. They are reported to be very efficacious for closed-cervix pyometra. Reported dosages are 6mg/kg twice daily on the first day followed by the same dose once daily on days 2, 3 and 4. Some authors prefer to use a larger dose (10 mg/kg) once daily to be repeated after 48 hours.

KEY FACTS:

1. Canine ovulation may occur as early as day 5 days or as late as day 23 after proestrus onset.
2. Onset of receptivity to mating in normal bitches may occur during proestrus (up to 11 daysbefore ovulation) or at mid-estrus (up to 6-7 days after ovulation)
3. Ovulation should be confirmed assaying serum P4 and using vaginal smear every 2-3 days until a high P4 value and the first day of cytological diestrus are observed
4. Pyometra is characterized by cystic endometrial hyperplasia and presence of high serum P4 concentrations.
5. Treatment includes specific antibiotic and - if the cervix is open - PGF2a to be administered BID in diestrus (to cause luteolysis) or once daily in anestrus.

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Artificial collection of semen and insemination in canine

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Artificial insemination (AI) in the dog is commonly practiced when the female does not accept breeding by a specific male, when a male cannot mount due to physical problems (vertebral column disease, hind leg disease, excessive panting in brachicephalic breeds etc.), or when male and female live far apart and cannot travel. Semen collection in the dog is a simple technique which requires very little training and which can provide veterinarians with extremely important clinical information concerning the present and future fertility of their canine patients.

In Artificial Insemination (AI) the semen is collected manually from a stud male andthereafter deposited (inseminated) in the female so that fertilization can occur in the absence of natural mating. Artificial Insemination, one of the earliest techniques for assisted reproduction in animals and humans, took longer to be implemented in dogs due to species specific particularities.In past decades, progresses in the knowledge of canine physiologyand new advances in canine semen technology allowed these services to become available worldwide. Hence, subsequent to the increase in the artificial insemination demand among dog breeders and owners and the broaden of the AI to preserved semen as a management tool in canine breeding, as through international exchange of frozen semen, inbreeding within breeds can be reduced.

Therefore, with spread of canine AI dog, breeders now may select stud dogs from all over the world to improve their kennel´ genetics, without transport-associated stress to the animals. Also, it is possible to save semen from valuable dogs into sperm bank to be used in next generations, after their death or the peak of reproductive age. In addition, breeders also are aware of the sanitary benefits associated with AI. Avoiding direct contact between the male and female, AI also prevents the spread of sexually transmitted diseases, as those originated by *Brucella canis* or *Herpes virus*.

Indications for artificial insemination:

Main indications for AI in dogs include both medical and breeding-management reasons. As major potential advantage, AI may allow to reduce physical distances, the use of genetically valuable stud dog semen all over the word, fighting the stress of transportation of animals and inbreeding. It is also an important technique whenever physical and behavioral abnormalities in the male or female preventing natural mating.

Semen collection

Equipment:

latex cone (not indispensable); 10-15 cc sterile tubes; bitch in heat (not indispensable); latex gloves; microscope with a 100x ocular; miscroscope slides; Pasteur pipette; micropipette or insuline syringe; formaline; haemocitometer; stain (eosine-nigrosine).

Collection technique:

Semen collection in the dog is a relatively easy procedure, although requiring some trainingfor optimization of the technique. Semen collection and evaluation is necessary to obtain good results in canine AI. Although practitioners are often asked to collect semen and perform AI without detailed semen analysis, every sample of semen collected should be evaluated (at least progressive forward motility, total sperm count and morphology) before it is used for artificial insemination or cryopreservation. Semen evaluation prior to insemination warrants the

male potential fertility and consequently may predict the fertility potential for the AI. In addition, when preparing semen preservation, fertility certificate may be needed. In such cases andrological evaluation of the stud dog (breeding soundness evaluation or BSE) has to be performed. Semen collection should be performed before the physical exam or any stressful procedures on the stud, or can be booked to another day

Semen can be collected from most dogs in the absence of a teaser, in a quiet and isolated room, where interruptions should be prevented, although the presence of a bitch would allow better ejaculates. In reluctant males, stimulating estrus scent can be provided by the presence of a female in estrus or by using frozen-thawed swabs or gauze sponges taken from vaginal secretions of estrus bitches. Although possible, not everyone achieves the use of a chemical pheromone (methyl phydroxybenzoate, Aldrich Chemical, Milwaukee) swabbed on the perineal area and tail of an anestrus teaser.

Collection of semen should be prepared in advance, and interval between collections or between the natural mating and collection, should be registered, if the male is regularly used. Ideal intervals between collections are 2 to 5 days, whilst intervals longer than 10 days may result in an increased number of morphological abnormalities and decreased motility. In longer periods, it is advisable to perform one previous collection, if semen is to be chilled or frozen for shipment. If semen preservation is planned, semen extender should be prepared before the arrival of the animal.

The most common method for semen collection in the dog is by digital manipulation, in the presence of a female. However, bitch presence, although desirable as it facilitates procedures. It should be noticed that when the collection is achieved in the presence of the bitch ejaculates present higher concentration.

The use of manual massage is the most commonly used technique, although in the past semen was collected from dogs using an artificial vagina. The process is started with a massage of the dog prepuce at the level of the *bulbus glandis* until developing partial erection, followed by the quick retraction of the prepuce and penile expose. If the collector is right-handed, semen must be collected from the dog's left side, with the operator holding the dog's penis with the right hand and the collection container in the left hand. During pelvic thrusting, rigid vials should be kept at a distance from the penis, to avoid trauma. When pelvic movements are finished and the dog lifts its rear leg, a 180º backward rotation of the penis should beobtained and the erectile penis should then be directed into the collection cone or the funnel.Some pressure may be applied with the thumb on the apex of the *glans penis*, at the level ofthe urethral process, to stimulate ejaculation. When a crystal clear fluid (prostatic fluid) begins to flow into the collection tube, you can gently slide the collection cone off the penis. Watch for semen to flow in the collection tube. Canine ejaculate consists of 3 fractions, with the first and third fraction consisting of prostatic fluid and the second being rich in spermatozoa **(Table 1)**. The first fraction, the presperm portion, is emitted in 0.5 to 1 minute and is colourless, with a volume range of 1-5 ml. It is expelled during first stage of erection, at the moment of the presence of evident copulatory movement of male. The second fraction, the sperm-rich portion, is also rapidly completed (1-2 minutes), and is grayish-white in colour, with a volume of 1-3 ml. It is expelled when thrusting movement of the male ceases and full erection is observed. The third fraction comes from the prostate and may be up to 30-40 ml; it may take up from 5 to 30 minutes to be completed.

Semen Evaluation

Macroscopic evaluation

Volume: The volume of the ejaculate may be assessed in the calibrated tubes used for semen collection. It mainly dependends on the size of the dog, the size of the prostate gland, the animal age, the frequency of semen collection, the level of erotisation, and the volume of 3rdfraction collected. A decrease of semen volume is observed in cases of benign prostatichyperplasia, prostatic cysts, inflammatory lesions of prostate and testicles, inflammation of epididymis, vas deferens or urethra and at weak libido.

Colour: The colour of whole ejaculate depends on the volume of third fraction of ejaculate collected, on the concentration of spermatozoa per mL and the potential presence of nongerm cells in the ejaculate. When analysing the colour, one should be aware of the method of collection, as colour varies with the fraction to be analysed and the fact that analysis may been performed on the whole semen or on fractioned semen. The normal colour of whole ejaculate is greyish-white. Pathological colours include: green-greyish typical for the presence of the pus in semen; red or pink-specific for erythrocytes contamination (haemorrhages from urethra or corpora cavernosa, prostatitis); yellow specific for urine contamination; and brown, if in the presence of blood. Any kind of semen contamination, such as hair or mud, exclude the specimen from further procedures including artificial insemination or semen preservation. It is therefore important to check the region of praeputial opening before semen collection and to clean it. The presence of sediment consisting of sperm cells at the bottom of the tube is a normal feature if the semen is left for several minutes.

Microscopic evaluation

Motility: One of the most important step of conventional semen assessment is the subjective evaluation of progressively motile spermatozoa under microscope. The optimal temperature for assessment of dog sperm cell motility is 39ºC. A small drop of about 20 µl of semen is placed on in a pre-warmed slide and cover by the coverslip. The evaluation is performed under the objective of x20 to x40. The assessment is based on the evaluation of the average percentage of progressively motile spermatozoa ina few different fields of the specimen. The normal dog semen contains at least 70% of progressively motile spermatozoa. A decrease in the percentage of motile spermatozoa may results from temperature shock, contamination with water, urine, blood or lubricants but also from long sexual abstinenceand systemic or infectious diseases, such as brucellosis. Sperm agglutination is always pathological and is frequently found in cases of infectious diseases

Concentration and total sperm count: The sperm concentration in whole canine normal ejaculate usually exceeds $80x10^6$ Spz/mL. If the second fraction of ejaculate is collected separately, the sperm cells concentration in sperm-rich fraction varies usually between 200-600 x 10^6 Spz/mL. It is generally assumed that the number of motile spermatozoa necessary for successful AI should be >150 $x10^6$. Therefore, under normal conditions, the dog´s ejaculate contains far more sperm cells than those needed for a seminal dose, although sometimes, especially in miniature or toy breeds, ejaculate volume and the total number of sperm cells are relatively low (<100 $x10^6$Spz/ml). The concentration of spermatozoa in semen volume is usually assessed by cytometric method on the haemocytometer, such as the Neubauer chambers, with semen pre-diluted at 1:200. In order to find the sperm count *per* ml, the number of spermatozoa in the one or four large squares (depending of the chamber) is multiplied by 500 000. For the assessment of sperm concentration more sophisticated equipment could also be used, such as the spectrophotometer, flow cytometer or computer assisted semen analyser. The number of

spermatozoa *per* ejaculate alsovaries according to age, testicular weight, sexual activity and the size of the dog. The total number of spermatozoa in the ejaculate may be decreased in young and older dogs and in inbred males. Apprehension, absence of the teaser bitch, painful prostate and spine rear limbs may also negatively influence the number of spermatozoa ejaculated.

Sperm morphology: The percentage of morphologically normal spermatozoa in caninesemen should be greater than 70%. The morphology may be assessed under contrast-phase microscope, but usually the evaluation is performed under light microscope on stained slides. Smears of undiluted or diluted ejaculate are examined microscopically for the presence of structural abnormalities of spermatozoa. The stains used with a nigrosin-eosin stain. The semen is smeared on aglass slide in a similar manner to that of blood, air dried and stained. A drop of this stain is gently mixed with a drop of semen on a pre-warmed slide before being smeared, and allowed to air dry. Evaluation of sperm morphology should be completed microscopically using oil immersion, using an objective of x100 or x 125. A minimum number of 200 spermatozoa should be counted and evaluated for the presence of abnormalities. The percentage of cells with particular morphological defects and of normal cells are calculated. Traditionally sperm cells abnormalities are divided into primary defects-originating from abnormalities of spermatogenesis and secondary defects.

Live-dead' spermatozoa: The assessment of the percentage of live and dead spermatozoa isbased on the assumption that dead spermatozoa possess disintegrated plasma membrane allowing eosin penetration. The percentage of eosin positive cells stained with nigrosine eosin stain is considered as percentage of dead cells. The normal dog semen consists of maximal percentage of 30% of dead sperm cells. The evaluation of the percentage of live and dead spermatozoa and the percentage of morphological defects may be performed on the same nigrosin-eosin stained slides.

Table: 1 Main characteristics of the different fractions of the dog ejaculate.

Characteristics	**1st fraction**	**2nd Fraction**	**3rd Fraction**
Volume	0.1-2 mL (average 0.33 mL)	0.1-3 mL (average 1.17 ml) Sometimes larger volume	1-2 to >20 ml Quite variable depending on the animal.
Colour	clear or opaque	greyish-white white, milky-white	clear, transparent
Consistency,	watery	watery-milky	milky watery
Character	Prostate secretion with admixture ofepithelial cells, urine, bacteria and sperm cells	sperm cells suspended inseminal plasma	prostate gland secretion
pH (average)	6.37	6.10	7.20
Duration	5-90 sec. (average 13.5 sec)	5-300 sec.(average 52.4 sec.)	60 sec-20 min. (average 6 min. 55 sec.)

Table: 2 Variation on the volume of the ejaculate with the size of the dog

Size of the dog	**Volume of the ejaculate**
< 20 kg	1-22.5 mL (average 5.38 ml)
> 20 kg	2-45 mL (average 12.75 ml)

Timing of ovulation in the bitch:

Canine proestrus and estrus last on average 9 days each with ovulation taking place 3 days after onset of estrus (or day 12 after onset of proestrus). However ovulation can occur as early as 5 days or as late as 27 after onset of proestrus. Therefore, it is very important to check the female's behavior, perform vaginal smears every 2-3 days starting on thefirst day of proestrus in order to catch early ovulators, and draw blood samples to measureprogesterone once behavior and/or vaginal smear indicate estrus. Estrus is indicated by acceptance of the male or by a degree of vaginal cornification of >70. Ovulation occurs 3 days after onset of estrus. Serum progesterone has a concentration of (values are approximate) 2.0-3.0 ng/ml on the day of the peak of luteinizing ormone (LH), 4.0-8.0 ng/ml on the day of ovulation, 10-25 ng/ml during the 2 days following ovulation, which is when oocytes are reaching maturity in the ampulae of the oviducts and fertilizations are taking place. Ovarian structures can be visualized with ultrasound using 5.0 to 7.5 sectorial MHz probes; follicular growth can be followed and ovulation can be estimated based on disappearance of the hypoechogenic areas representing follicles (which become luteinized) and on appereance of a hypoechogenic area at the periphery of the ovary representing follicular fluid accumulation within the ovarian bursa.

Fresh and Refrigerated Semen:

In most countries of the world canine AI is performed using fresh semen. When properly performed, the success of AI with fresh semen is equal to the success of natural breeding, i.e. >80%. Although shipment of fresh undiluted semen can be done provided that travel time does not exceed few hours (and provided also that prostatic fluid is normal), it is always better to dilute semen as spermatozoa lose very rapidly their fertilizing ability when maintained in seminal plasma. Semen extenders protect the sperm membrane from temperature variations as well as from mechanical trauma during transport, providing also stable pH and temperature conditions. Antibiotics such as streptomicin and penicilline should be use especially when storage is prolonged for more than a few hours especially when using egg yolk-based extenders where bacterial growth is enhanced. Canine semen extenders, one rather simple, milk-based extender good for a practice situation which maintains normal semen motility for 24-36 hours and a more complex, Tris-fructose-egg yolk based extender which requires more time to be prepared but allows a semen survival of up to 4-5 days. Properly extended and refrigerated semen placed in a plastic vial can be shipped across countries in a thermos.

Canine semen should be diluted 1:3 or 1:5 depending on its concentration. If the semen sample is too diluted it can be centrifuged at 500 g for 5 minutes to remove the excess prostatic fluid prior to adding the extender. Refrigeration can be performed in a normal refrigerator for 30-60 minutes, after which the semen sample can be shipped in a thermos. Keep in mind that a motile semen sample may already have lost its fertilizing capacity: it is generally believed that motility lasts for about twice as long as fertilizing ability. Prior to insemination let the semen sample reach slowly room temperature.

Insemination Technique:

Fresh semen can be deposited in the cranial portion of the vagina through a plastic cathether. Rigid catheters used for large animal uterine flushing work well although they need to be shortened for the use in bitches. Intravaginal insemination is easy and widely practiced and conception rates following use of fresh or refrigerated semen are good. Ideally the bitch should have an empty stomach and be contained in a standing position. The catheter is inserted from the dorsal vulvar commissure (just like the cotton swab for vaginal smear) and its positioning at the end of the vagina is verified through abdominal palpation: the cervix (easily palpable during

estrus) is identified and the tip of the catheter must be palpated just caudally. Once all the semen has been flushed from the catheter the hind legs of the bitch are elevated and kept in this position for 5-10 minutes (a procedure which is widely believed to help spermatozoa cross the cervix, although no scientific data have ever been produced).

Frozen semen must be inseminated into the uterus, as thawed spermatozoa are short-lived and cannot move vigorously enough to cross the cervix in numbers adequate to achieve a good conception rate. A Norwegian catheter (made by a steel 2.0 mm catheter with a Teflon sheath) purposedly designed for AI in foxes works well in bitches and has been used at the canine frozen semen bank of the One hand identifies and holds the cervix while the other one pushes the Teflon sheath until it reaches the par cervix, after which the steel tip of the catheter is carefully worked through the cervix into the uterus. Disadvantages of this catheter are a difficult learning process and the fact that it cannot be used in large size bitches (because the cervix cannot be palpated with one hand).

The cervix can be passed also with a rigid endoscope. A human cystoscope or cystoureteroscope is best used. A complete set of endoscopy (CO2 insufflator, light source etc.) is necessary, which makes the technique expensive. Alternatively, intrauterine insemination can be performed surgically or laparoscopically. The surgical approach is faster, both are without complications and conception rate is in the 60%-70% range.

Table: 3 Artificial insemination schedules for dogs, according to the type of semen used.

Semen	**Doses**	**Expected spz Semen survival**	**Insemination schedule**	**Expected fertility**
Fresh	150-200x106 spz/ml (extended)	4-6 days	-Every other day, when P4 rise above 4ng/mL, up to 3 times. -Day 1 to 4 post-ovulation -P4 levels between 8 & 15ng/mL	-80-90% (either with transcervical or vaginal deposition)
Chilled	150 - 200x106 spz/ml (extended)	24-72hrs	-Breeding once or twice 2-4 days post ovulation (P4 = 4 -10ng/mL). -Day 2 to 4 post-ovulation -P4 levels between 8 &15ng/mL	- 80-90% (either with Transcervical or vaginal deposition
Frozen	50-300x106 spz/ml (extended)	12-24 hrs.	-Twice, at P4 levels above 8 ng/mL and estrus vaginal cytology -Day 5 to 7 post-ovulation -P4 levels between 18& 28 ng/mL	-45% if vaginal deposition -67-84% if transcervical or intrauterine

Success rates for artificial insemination:

The key-issues to obtain good results by using canine artificial insemination are:

- Proper timing of the insemination
- The use of adequate number of viable sperm cells *per* dose
- Good semen preparation and handling
- Adequate deposition of semen in the female reproductive tract

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Understanding of basics of reproductive ultrasonography and its application in Pet animals

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Ultrasound techniques are becoming increasingly important in Pet animals, offering both a mean of diagnosis and a useful therapeutic tool and thus help to maximize the reproductive efficiency. For this reasons, understanding the use of ultrasound technology is critical in contemporary animal sciences, since ultrasound examinations are now a routine component of diagnostic workups in Pet animals. Ultrasonography has several advantages over other imaging modalities. It is non-invasive, free from radiation hazards, provides instant diagnosis and determines shape, size, location and internal consistency of structures. Further repetitive examinations can be done and it is well tolerated by the animals. Practical applications of reproductive ultrasound include follicular dynamics and ovulatory status for timed AI, treatment efficacy in problematic animals, early assessment of pregnancy, and identification of animals carrying twins, detection of ovarian and uterine pathology and determination of fetal age, viability and sex. Each of these applications presents opportunities for improving reproductive efficiency of an animal.

Principles of Ultrasonography

Ultrasound is defined as any sound frequency above the normal hearing range of the human ear; i.e. greater than 20,000 Hz. Briefly, ultrasonography utilizes high frequency sound waves to produce cross sectional images of the tissues and internal organs. The sound waves are produced by vibrations of specialized crystals (piezo-electrical crystals) housed in the ultrasound transducer. Vibrations of the crystals are produced by pulses of electric current. A proportion of sound waves reflected back to the transducer is converted to electric current and displayed as an echo on the ultrasound viewing screen. The transducer, therefore, acts as both the sender and receiver of echoes. The echoes are evident on the viewing screen as varying shades of gray (black to white). The absolute value of the acoustic impedance of any tissue is relatively unimportant, because it is the magnitude of the difference in acoustic impedance at tissue interface that determine the amount of reflection of the beam. Most ultrasound scanners used in bovine reproduction currently are B-mode (brightness modality) real-time scanners. In B-mode ultrasonography, the image is a two-dimensional display of dots (pixels). The brightness of the dots is proportional to the amplitude of the reflected echoes returning to the transducer. Real-time refers to the ability to image movements (e.g. fetal heart beat or motion) as it occurs. Dynamics of some reproductive structures or events (i.e. ovulation) could be studied by video tape recordings of real-time ultrasound examinations. Ultrasound scanners are equipped with transducers of varying frequencies. The most commonly used frequencies in bovine reproduction are 3.5, 5.0 and 7.5 MHz. The higher the frequency of the transmitted sound waves, the better the image resolution, but the shallower the depth of penetration.

There are two types of scanners; linear array and sector. Linear array transducers have piezo-electric crystals arranged in rows and as such the image produced by linear array transducer appears rectangular. Sector transducers, on the other hand, have only a few such crystals and the image produced is pie-shaped corresponding to the field of scan. Mechanical sector scanners offer multi frequency capability in a variety of scan head design with 3.0, 5.0 and

7.5 MHz crystals in a single scan head. Because of the versatility of these machines, they are also more expensive than linear- array systems. Recent ultrasound scanners have a wider frequency range and accept a full line of single and dual frequency probes. Digital image port for computer image storage is also available.Battery-powered portable ultrasound scanners are also currently available. Doppler ultrasonography which detects turbulence within blood vessels and direction of flow is also a useful diagnostic tool in bovine reproduction. The Doppler phenomenon is the change in sound frequency of a moving object as perceived by a stationary observer. Doppler ultrasound machines detect frequency change and, therefore, movement which is converted to an audible signal. The major considerations in selecting an ultrasound scanner are price, resolution quality, portability, serviceability and technical support. Other factors include memory capabilities, remote control, transducer and cable design (I or T), single or dual frequency probe and above all, the use for which the machine would be put. For routine bovine reproductive ultrasonography (early pregnancy diagnosis, pathology of the ovaries and uterus, fetal sexing etc) a 5 MHz linear rectal transducer seem to be the most versatile and effective. However, a 7.5 MHz linear transducer is recommended for follicular dynamics studies. For transvaginal oocyte recoveries, a convex-linear transducer gives better results.

Techniques

Probe selection: 5-10MHz should be adequate to scan most abdomens in small animal practice. Most common is a 5-7.5MHz curvilinear probe. Deeper dogs need lower frequency for more penetration and smaller dogs and cats can be scanned at 8-10MHz for more detail.

Patient Preparation:

Fasting: To limit the amount of food material and gas in the GIT.

Bladder: Try to scan with a full bladder if looking for bladder wall lesions. However if too distended can displace abdominal organs cranially.

Clipping: Necessary for optimal image quality. Clip close to the skin. Caudal costal arch to pubis. Use alcohol before coupling gel but care to protect probe.

Sedation: Most abdominal ultrasounds possible without sedation but animals should be kept as stress free as possible. Use quiet clippers and reassure patient. Sedation is sometimes necessary; it reduces panting and allows ultrasound of tense abdomens. Beware of vasodilation, ACP and barbiturates can enlarge the spleen.For transabdominal ultrasound scanning in Pet animals. Adequate restraint is however required.Ultrasonography was performed from the left andright shaved flank using a phase array 8.0-MHz linear scanner with a commercial ultrasound gel. The dog was placed in either left or right lateral recumbency, or during some examinations she was positioned in dorsal recumbency.

Basal requirements

To make an accurate diagnosis via an ultrasonographic examination, ambient lighting is imperative. A darkroom is ideal for viewing the monitor and helps the human eye recognize as many shades of gray as possible. When examinations are carried out in lighted conditions, some type of hood must be draped over the monitor to facilitate effective gray-shade delineation. Interposition of any contaminating stool will prevent ultrasound transmission and produce poor imaging and artefactual interference. The ultrasound screen and the human eye should be at similar level for accurate interpretation of ultrasound images.

Interpretation of ultrasound images

Interpretation of ultrasonography of the reproductive tract requires a thorough understanding of the composition of the images and an awareness of the possible artefacts which can occur and lead to a misdiagnosis. As sound waves pass through the tissues and surrounding

areas they may be modified in a number of ways. Sound waves passing through body structures will encounter tissue interfaces and the returning echoes will be of varying strengths and so produce a variety of images. The ultrasonic characteristic of a tissue depends on its ability to reflect sound waves. Liquids do not reflect sound waves (i.e. are non-echogenic or anechoic) and are represented on the viewing screen as black. The ultrasonic images of liquid containing portions of structures such as ovarian follicles, embryonic vesicles appear black. A dense tissue (e.g. bone) reflects a large proportion of the transmitted sound waves (i.e. echogenic) and is represented on the viewing screen as light gray or white. Various tissues and contents of the reproductive tract appear on the screen in varying shades of gray depending upon their echogenicity. Some common artifacts include; distant enhancement (occurs when the incident sound beam strikes the far wall of a fluid-filled structure; e.g. follicle, embryonic vesicle), refraction artifacts, specular reflection, reverberation artifacts, acoustic shadowing.

Applications of Ultrasonography in Pet animals:

The normal ovaries and uterus

The ovaries are either directly caudal, or caudal and medial, lateral or ventral to each respective kidney. The normal ovary is small (canine: 1.5 x 0.7 x 0.5 cm) and may be hypoechoic, or have anechoic follicles (normal size is up to 1 cm) within it. After ovulation, the follicles do not collapse, but develop an initial mixed echogenicity, progressing to a hyperechoic then hypoechoic corpus luteum, which produces progesterone. Ovarian neoplasia can occur and is suspected when a mass is seen just caudal to one of the kidneys. The normal uterus is difficult to visualize in the dog (normal size 0.5 – 1 cm), and usually not visualized in the cat. A full urinary bladder provides a good acoustic window. The body of the uterus lies dorsal to the bladder and ventral to the colon, and is either close to the midline, or just to one side of it. The uterine horns are difficult to visualize unless they are enlarged. The normal uterine lumen is usually not distinguishable.

Pyometra in the dog and cat

Cystic endometrial hyperplasia can develop in both dogs and cats, though it is seen more often in dogs, and this condition can contribute to infertility. Ultrasonographically, a pyometra will appear as a fluid-filled uterus. The fluid may be anechoic, but often will have echogenic particles within it. The uterine wall may initially be edematous and thickened, but with increasing uterine distension, it can become very thin.

Pregnancy diagnosis

Ultrasound is a very sensitive and specific imaging method to diagnose pregnancy if it isperformed at least 30 days after breeding in the dog and at least 16 days after breeding in the cat. Pregnancy has been diagnosed as early as 10 days after breeding in the dog and 11 days after breeding in the cat. Visualization of a gestational sac is considered to confirm pregnancy. Uterine enlargement will precede the formation of the gestational sac, but that is not a specific finding. Cardiac activity and fetal movement are predictable signs of fetal viability. Visualization of the embryo and cardiac activity usually occur on the same day. Initially, cardiac activity appears as a flutter within the embryo tissue. The fetal heart rate is usually twice the maternal heart rate, and usually is between 200 and 220 beats per minute (bpm). Fetal activity can include swallowing, hiccoughs, and body and limb movements.

Liver

Ultrasonography of the liver should be performed if liver disease is suspected even if there are no radiological abnormalities. The hepatic parenchyma, gall bladder, large hepatic and portal veins and caudal vena cava are all visible. The patient should be fasted but given free

access to water. Place the transducer on the ventral abdomen at the level of the xiphisternum and angle it craniodorsally to image the liver. Fan the beam from left to right to span the entire liver. If the liver is small, image it through an intercostal approach.

Ultrasonographic appearance of the normal liver moderately echoic with a granular appearance Lobes smooth in outline and sharply pointed Gall bladder is rounded or pear shaped. It can be dilated after prolonged periods of anorexia

Stomach

Food should be withheld for 12 hours for full assessment of the stomach. Offer water just before the scan as visualisation of the stomach walls are best seen in a moderately fluid distended stomach. Probe placed caudal to rib cage in a sagittal plane. Sweep from right to left in a cranio-dorsal direction. Deep chested dogs may need an intercostal approach. The gastric fundus is seen in left cranio-dorsal abdomen and is recognised by prominent rugal folds when empty. Follow the greater curvature ventrally and to the right and examine the body and the antrum. The pylorus and cranial duodenal flexure are caudal to the liver hilus and ventral to the portal vein. Excessive gas in the stomach makes imaging of deeper stomach structures difficult, so use positional ultrasonography or offer water.

Spleen

The head of the spleen is located in the left cranio-dorsal abdomen and is often located within the rib cage. After this has been imaged place the probe in transverse and follow the spleen down to the hilus and then the caudal extremity. Often difficult to image the entire spleen so adopt a meandering probe technique, constantly moving the probe to visualise the entire spleen. The splenic parenchyma is homogenous and of a fine echotexture. It is covered by a thin, very hyperechoic capsule. It is hyperechoic to liver and renal cortices. Branches of the splenic vein are seen as tubular anechoic structures within the parenchyma and exit the spleen at the hilus. Splenic arteries are not usually seen.

Kidney

Left Kidney Best seen with patient in right lateral recumbency and from a left lateral approach. Also possible to scan from a ventro-lateral approach. Kidneys should be scanned from cranial to caudal and lateral to medial, in several transverse and longitudinal planes. Kidneys are typically bean shaped in dogs and oval in cats. Kidney length variable in dogs and in cats typically measure between 3.0cm and 4.3cm. Renal medulla is hypoechoic in comparison to renal cortex, which is hypoechoic to liver and spleen. A hyperechoic renal cortex can also be seen in dogs with normal renal function. Medulla separated into almost anechoic segments by interlobar vessels and diverticuli. Walls of arcuate arteries are seen as paired, short, hyperechoic lines at the corticomedullary junction, can generate an acoustic shadow and don't mistake them for mineralisation. The cortex should be about twice the depth of the medulla and they should be easily distinguished (good corticomedullary definition). The renal pelvis is surrounded by the sinus which is hyperechoic (contains fat). The normal renal pelvis should be no greater than 2mm in height. Sometimes it is seen in normal animals if diuresed or on intravenous fluids.

The right kidney is more difficult to visualise than the left as it sits partially under the rib cage and may be obscured by gas. A right lateral intercostal approach is sometimes necessary. The right adrenal gland is more cranially positioned than the left. It lies cranial to the right renal artery, medial to the cranial pole of the right kidney and in close apposition to the dorsal surface of the caudal vena cava.

Bladder and Urethra

Clip the hair to the level of the pubic bone. Use 7.5-10 Mhz probe depending on size of patient. Can scan the bladder in dorsal, left or right lateral recumbency. A standing position can be useful in confirming the presence of calculi as they will fall towards the gravity-dependent wall. Scan in transverse and longitudinal planes Bladder should be moderately distended as bladder wall thickness increases if bladder is empty. Normal urine is anechoic. Echogenic urine can also be within normal limits, however urinalysis should be used to determine significance. Normal bladder wall is smooth apart from uretral papilla which can be seen extending from bladder wall and should not be confused as focal thickening. Can use colour flow doppler to check for uretral jet. Side-lobe artefacts can be confused as sludge. Only some of urethra can be visualised with ultrasound, use retrograde positive contrast ultrasound for visualising pelvic urethra.

Prostate in male dogs

The prostate surrounds the proximal urethra. More detail if use higher frequency probe. Prostate is of medium echogenicity and homogenous with medium echotexture and smooth margins. In neutered dogs ultrasound of the prostate can be challenging. It is small, inconspicuous, hypoechoic and homogenous. Age related changes in intact dogs include an increase in size and echogenicity.

Small Intestines

Withhold food for 12 hours prior to ultrasound exam. Allow free access to water. Shadowing and gas can be a problem so use positional ultrasonography. Use high frequency probes for detail of layers. Normal anatomy: Layers ¬ Luminal interface - Hyperechoic ¬ Mucosa (widest layer) – Hypoechoic ¬ Submucosa- hyperechoic ¬ Muscularis – hypoechoic ¬ Serosa – hyperechoic Assess for overall layering, thickness, echogenicity of each layer and individual width of each layer. Note if changes are focal or generalised. Assess for motility (regional and general). Assess if luminal diameter is increased generally or proximal to a lesion.

Colon

The descending colon is found easily on the left side of the doro-caudal abdomen. Follow it cranially through the transverse and ascending colon to the ilio-colic junction. Appearance varies depending on content. Gas and faeces are hyperechoic typically with distal acoustic shadowing and/or reverberation artefacts. When distended the wall is very thin (1-2mm thick), when empty the wall collapses and can appear thick and irregular with a multi-layered crumpled appearance. Ilio-colic junction is difficult to find in dogs because of gas in the caecum. It is easier to identify in cats and should be examined as it is a site for intestinal lymphoma. It has a "wagon-wheel" appearance.

Pancreas

A boomerang-shaped organ with a body, left limb and right limb. In dogs the right limb is larger than the left. In cats it is vice versa. The pancreatic parenchyma is difficult to identify. Use a high frequency probe. Normally iso-hypoechoic to surrounding mesenteric fat. Normal pancreas has a homogenous, fine, granular echotexture with smooth borders. Position the animal in right lateral recumbency. Keeping probe parallel to the table and underneath the patient, apply gentle pressure, find right kidney, pull back slightly, see duodenum. Right limb of the pancreas sits between right kidney and duodenum. Left limb of the pancreas is caudal to stomach, cranial to the colon and medial to the spleen. The body of the pancreas is ventral to the portal vein and caudal to pyloric canal. Pancreatico-duodenal vein is a useful landmark, it is embedded in the parenchyma of the right limb.

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ADVANCES IN DIAGNOSIS AND THERAPEUTICS OF CANINE DISEASES

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Dogs are affected with many diseases. It can be infectious or noninfectious in origin. This article will give a brief outline of canine disorders, its diagnosis and treatment

VIRAL DISEASES

Rabies

Changes in behavior may be one of the first clinical signs and may consist of episodes of mild or dramatic abnormalities.Rabies is neurotropic and may result in irritation or paresthesia at the site of initial exposure even though the inflicted wound may have healed. A combination of increased saliva production and a decreased ability to swallow may present as profound contamination of the mouth, chin, and forelegs with potentially infectious saliva. Cranial nerve involvement may be focal and unilateral, presenting as unequal pupil size with dysfunction, facial or tongue paresis, and changes in phonation. As the clinical period progresses, unpredictable episodes of attempts to bite may be invoked by auditory, visual, or tactile stimuli with aggression to the point of self-mutilation. In the end stage, most animals become profoundly moribund. Post bite management consists of prompt wound cleansing, infiltration of rabies immune globulin of human (or equine) origin, and five doses of vaccine administered intramuscularly on days 0, 3, 7, 14, and 28.

Canine Parvovirus

CPV-2 is the most common cause of viral enteritis in dogs. Dogs of all ages are susceptible to infection if they have no or partial immunity. Dogs 6 weeks to 6 months of age are most susceptible to infection and clinical disease due to maternal antibody interference with active immune responses to CPV-2 vaccines.Clinical signs often start with anorexia, lethargy, and fever. The condition progresses within 1 to 2 days to include vomiting and diarrhea, which may be yellow, mucoid, or hemorrhagic. More severe GI signs can be expected in puppies with intestinal parasites or other concurrent intestinal disease. Uncommonly, clinical signs of myocarditis may be observed in puppies, typically those younger than 8 weeks of age with failure of passive transfer of maternal immunity.

No specific antiviral drug, therapy of parvovirus focuses on supporting effective circulating volume, controlling secondary bacterial infections, and resting the GI tract. Administration of crystalloid fluids such as lactated Ringer's or 0.9% saline at volumes sufficient to restore and maintain hydration despite ongoing fluid losses is a key element of therapy. Supplementation of fluids with potassium and dextrose may be necessary to maintain normal serum potassium and glucose concentrations.

Colloid administration (hetastarch or dextran 70) may be indicated for hypoproteinemia, especially if peripheral edema and/or effusion develop in third spaces. Plasma transfusion has been considered as an adjunctive treatment for hypoproteinemia but may not be as effective as nonprotein colloids because a large volume is required to achieve a small increase in plasma protein at the risk of fluid overload. Dogs with vomiting and diarrhea are typically maintained NPO (nothing by mouth) to rest the GI tract; partial parenteral nutrition may be considered for nutritional

support of these animals, especially young puppies.Parenteral administration of broad-spectrum antimicrobials with good gram-negative spectra is considered a key aspect of treatment due to risk for sepsis. Good combinations include an aminoglycoside or fluoroquinolone with a beta-lactam penicillin (amoxicillin, ampicillin) or first-generation cephalosporin. Aminoglycosides should only be used in well-hydrated dogs. Once vomiting has stopped, patients may be offered water. If no more vomiting occurs, food may then be offered. Diets in the initial feeding period should be easily digestible and low in fat because villus structure and function may require a number of days to return to normal.

Canine Distemper Virus

Canine distemper is caused by an enveloped single-stranded RNA virus in the Paramyxoviridae family and is closely related to viruses that cause measles, rinderpest, and distemper in other animals.Viremia occurs 4 to 6 days later, with spread of virus to the stomach, small intestine, spleen and hepatic macrophages, bone marrow, and other lymphoid sites. The widespread increase in virus production is associated with fever and lymphopenia. A second viremia follows within a few days and is responsible for infection of epithelial cells in multiple organs, including the eyes, skin, and central nervous system (CNS).Puppies are more likely to suffer more severe and protracted illness and have the highest mortality rate.Affected dogs may be lethargic, anorexic, dehydrated, and febrile, and they frequently have respiratory signs initially. These include serous or mucopurulent oculonasal discharge and cough that progressively worsens if an inadequate immune response exists. Viral infection of the lower respiratory tract results in pneumonia that may or may not be clinically evident but can be documented via radiographs. Viral pneumonia complicated by secondary bacterial infections can be life threatening in puppies. Depending on viral strain, affected dogs may also have vomiting and mucoid or hemorrhagic diarrhea from viral replication in the GI epithelium. Virus infection of ocular tract epithelium can cause photophobia, anterior uveitis, and chorioretinitis. Recovered animals may have hyper-reflective retinal lesions that develop from retinal atrophy and scarring, as well as keratitis sicca from scarring of the lacrimal glands. Viral infection of epidermis can result in hyperkeratosis and hardening of the nasal planum and foot pads. Neurological signs starts 1 to 3 weeks after recovery from clinical signs. Treatment is supportive including fluid therapy, anticonvulsant drugs and broad spectrum antibiotics.

Adenovirus Infection

CAV-1 causes systemic infection with tropism for endothelial cells, epithelial cells, and hepatocytes. Viremia follows, leading to infection of other tissues. Direct cytopathic effects of the virus in the liver, eyes, and kidney. Clinical signs of anterior uveitis (“blue eye”) initially develop as a consequence of the inflammation following infection of corneal endothelial cells and the deposition of immune complexes as antibody responses to the virus increase.Initial clinical signs include fever, depression, and lethargy. Later, development of abdominal discomfort, mucous membrane pallor, and inflammation of the tonsils and pharynx with tonsillar and cervical lymph node enlargement occur. Abdominal fluid and hepatomegaly will be detected in some dogs. Laryngitis, tracheitis, pneumonia, coughing, vomiting, and diarrhea occur in some. In severe cases, petechial and ecchymotic hemorrhages and epistaxis may develop from coagulation abnormalities secondary to hepatic dysfunction and DIC. Icterus is uncommon. Neurologic signs may be seen as a consequence of hepatic encephalopathy or CNS infection. There may be leukopenia or leukocytosis depending on whether the patient is seen early or later in the course of disease. Thrombocytopenia is possible and could contribute to coagulopathies in the setting of DIC or abnormal platelet function. Increases in alanine aminotransferase (ALT) and alkaline phosphate (ALP) activity are expected. Therapy is directed at provision of

supportive care and managing clinical signs and complications. Intravenous fluid therapy to replace losses from vomiting or diarrhea is important, as is administration of blood products to manage the complications of hemorrhage and DIC. In patients with neurologic signs from hepatic encephalopathy, administration of lactulose via enema (or orally if the patient is not vomiting) can help reduce circulating concentrations of encephalotoxins.

CAV-2 and CPiV are part of a complex of pathogens causing canine infectious respiratory disease (CIRD) or "kennel cough."A hacking cough with a terminal retch and serous or mucoid nasal discharge are the typical clinical signs. Coughing bouts are often easily elicited with tracheal palpation and can be paroxysmal in nature. Coughing may produce foamy white phlegm.PCR assays can also be performed on swabs to rule out the presence of coinfecting viruses and *Bordetella bronchiseptica* bacteria.Treatment is largely supportive. Antibiotics are necessary for treating secondary bacterial infections evidenced by fever, purulent nasal discharge, productive cough, or pneumonia. Judicious short-term use of antiinflammatory doses of glucocorticoids or administration of cough suppressants, such as hydrocodone or butorphanol tartrate, may help ameliorate cough and improve patient comfort but are unlikely to shorten the clinical course.

Enteric Coronavirus Infection

Diarrhea (occasionally with blood) is the principle sign in clinically affected dogs. Vomiting may be seen before or after diarrhea in some animals. Anorexia and lethargy are common features, and if vomiting and diarrhea are severe, dehydration may ensue;more severe in neonatal animals, with diminishing severity of clinical disease in older animals. Clinical signs abate after 7 to 10 days in most dogs. Use of antibiotics in dogs with diarrhea, though not universally accepted, has been supported because bacteria may be primary or secondary pathogens.

BACTERIAL DISEASES

Leptospirosis

*L. interrogans*serovarsicterohaemorrhagiae and canicola were believed to be responsible for most cases of canine leptospirosis. The bacteria are maintained in the renal tubules of the reservoir host and excreted in the urine. Hosts are not typically ill and may be able to shed bacteria for their entire life. Leptospires can be transmitted directly between hosts in close contact through urine, venereal routes, placental transfer, bites, or ingestion of infected tissues as the organism penetrates mucosa or broken skin. Shedding by infected animals occurs, usually via urine. Diagnosis by serological tests. Lethargy, vomiting, anorexia, polydipsia, Icterus and fever. Azotemia, increased serum liver enzyme activity, electrolyte disturbances, and mild increases in serum bilirubin concentrations are common. Microscopic agglutination test (MAT) is the most common diagnostic method currently used for the diagnosis of canine leptospirosis.Visualization of organisms in fresh urine can be accomplished using dark-field microscopy. Organisms may be detected via culture or by detection of deoxyribonucleic acid (DNA) by polymerase chain reaction (PCR). Treatment depends on the severity of the clinical signs and whether renal or hepatic dysfunction is present. The goal of the first stage is to immediately inhibit multiplication of the organism and rapidly reduce fatal complications of infection, such as hepatic and renal failure. Doxycycline (5 mg/kg given every 12 hours) or penicillin and its derivatives are the antibiotics of choice for terminating leptospiremia. Ampicillin (22 mg/kg given IV every 8 hours) or amoxicillin, if available for IV use (22 mg/kg given every 12 hours) are preferred for vomiting, uremic, or hepatically compromised animals.

Tetanus

Tetanus is caused by a potent neurotoxin produced by the bacterium *Clostridium tetani*.All toxigenic strains produce two toxins, a tetanus neurotoxin (tetanospasmin) and a hemolysin (tetanolysin. Clinical signs of tetanus commonly occur between 3 and 18 days (up to 3 weeks) after an injury. Younger dogs are more susceptible.Clinical signs of tetanus commonly occur between 3 and 18 days (up to 3 weeks) after an injury. Younger dogs are more susceptible.Animals with localized tetanus frequently have stiffness in one limb or muscle group close to the injury site.Dogs and cats affected with generalized tetanus have generalized muscle stiffness of variable intensity, hypersensitivity to touch, lights, and sounds, but a normal mental state. Most dogs will show characteristic facial muscle spasms: wrinkling of the forehead, erect ears that are drawn together, lips that are drawn back (risussardonicus), and protrusion of the third eyelid from enophthalmus due to continuous or episodic contraction of extraocular muscles. For treatment, Tetanus antitoxin (100-300 U/Kg) should be administered to neutralize any toxin. Antimicrobial therapy should be administered in an attempt to kill any existing vegetative *C. tetani* organisms. Metronidazole and penicillin G (40,000 U/kg q4-6h IV) are the drugs of choice for treatment of tetanus.Supportive treatment includes Sedative and muscle-relaxing drugs such as Phenothiazine tranquilizers (chlorpromazine, acepromazine) in combination with phenobarbital, Benzodiazepines (diazepam, midazolam), Magnesium sulphate (subcutaneously), fluid therapy, urinary catheterization, enteric feeding and positive pressure ventillation.

The enteropathogenic bacteria most commonly incriminated in canine and feline diarrhea include *Clostridium perfringens*, *Clostridium difficile*, *Campylobacter* spp., *Salmonella* spp., and *Escherichia coli*.Antibiotics that have been recommended for the treatment of canine *Clostridium perfringens*–associated diarrhea include ampicillin (22 mg/kg q8h for 5 days), metronidazole (10 mg/kg q12h for 5 days), and tylosin (15 mg/kg q12h for 5 days). The drugs of choice for *Campylobacter*are the macrolides (erythromycin at 10 to 20 mg/kg q8h) or fluoroquinolones (enrofloxacin at 5 mg/kg q12h). Antibiotics reported to be effective against *Salmonella* include fluoroquinolones, chloramphenicol, trimethoprim-sulfonamide, and amoxicillin.Clinically stable animals can be treated with amoxicillin-clavulanate and first- or second-generation cephalosporins, until antibiotic susceptibility results are known. Dogs with life-threatening bacteremia should be treated with aminoglycosides, a third-generation cephalosporin, or enrofloxacin.

Ehrlichia, Anaplasmosis, Rocky Mountain Spotted Fever, and Neorickettsial Infection

Canine ehrlichiosis is caused by the intracellular, gram-negative, bacteria *Ehrlichiacanis*, *Ehrlichiaewingii*, and *Ehrlichiachaffeensis*.

Anaplasmataceae	*Ehrlichia*	1. *E. canis* 2. *E. chaffeensis* 3. *E. ewingii*
	Anaplasma	1. *A. phagocytophilum* 2. *A. platys*
	Neorickettsia	1. *N. helminthoeca* 2. *N. risticii*
Rickettsiaceae	*Rickettsia*	*R. rickettsia* (Rocky Mountain spotted fever)

These organisms are transmitted to dogs and cats by arthropod or trematode vectors. They also have the potential to be transmitted by blood transfusion.

Ehrlichiacanis is the cause of canine monocyticehrlichiosis. The organism is a pleomorphic bacteria that infects and forms morulae -a cluster of bacteria, within circulating monocytes.There are acute and chronic phases of diseases. Nonspecific signs are common and include depression, inappetence, fever, and weight loss. Replication of the organism in reticuloendothelial tissues is associated with generalized lymphadenopathy and splenomegaly. Ocular and nasal discharges, peripheral edema, and less commonly, petechial and ecchymotic hemorrhages may also occur. Neurologic signs, including twitching, ataxia, seizures, vestibular signs, hyperesthesia, and cranial nerve defects, may occur as a result of meningeal inflammation or hemorrhage.Thrombocytopenia and sometimes mild leukopenia and anemia occur 1 to 4 weeks after infection.Dogs may recover from the acute phase without treatment.Chronic ehrlichiosis ranges in severity from mild to life-threatening, with signs including lethargy, inappetence, bleeding tendencies, pallor, fever, weight loss, lymphadenopathy, splenomegaly, dyspnea, anterior uveitis, retinal hemorrhage and detachment, polyuria/polydipsia, and edema.Bleeding tendencies result from thrombocytopenia and platelet dysfunction. Cutaneous and mucosal petechial or ecchymotic hemorrhages, epistaxis, melena, hematochezia, hematuria, and prolonged bleeding from venipuncture sites have been reported.The treatment of choice is doxycycline (10 mg/kg PO q24h for 14-28 days.Chronic ehrlichiosis need more duration of treatment. If thrombocytopenia fails to respond to doxycycline administration, a short course (up to a week) of therapy with immunosuppressive doses of glucocorticoids may be beneficial in addition to ongoing therapy with doxycycline.

PROTOZOAL INFECTIONS

Protozoans generally cause either gastrointestinal (GI) tract disease (enteric protozoans) or polysystemic disease.

Hepatozoon canis and *H. americanum* both infect dogs. Fever, weight loss, and severe hyperesthesia over the paraspinal regions are common findings. Anorexia, pale mucous membranes from anemia, depression, oculonasal discharge, and bloody diarrhea occur in some dogs. Neutrophilic leukocytosis (20,000 to 200,000 cells/µL) with a left shift and normocytic, normochromic nonregenerative anemia are the most common hematologic findings.Imidocarbdipropionateadministered (5 to 6 mg/kg, IM or SC) once or twice 14 days apart is the drug of choice for treatment of *H. canis* and may also be effective for *H. americanum.* Administration of nonsteroidal antiinflammatory agents may lessen discomfort for some dogs.For treatment of *H. americanum,* the combination of trimethoprim-sulfadiazine (15 mg/kg PO q12h), pyrimethamine (0.25 mg/kg PO q24h), and clindamycin (10 mg/kg PO q8h) for 14 days is very successful in the acute stage.Use of decoquinate (10 to 20 mg/kg q12h) with food lessens the likelihood of recurrence of clinical disease and prolongs survival time.

Neospora caninum is a coccidian previously confused with *T. gondii* due to similar morphology.Although organism replication occurs in many tissues, clinical illness primarily reflects neuromuscular infection in dogs.Congenitally infected puppies develop ascending paralysis with hyperextension of the hindlimbs; muscle atrophy occurs in many cases. In some dogs, myocarditis, dysphagia, ulcerative dermatitis, pneumonia, and hepatitis occur. If not treated, most affected dogs die.Some have survived after treatment with trimethoprim-sulfadiazine combined with pyrimethamine, sequential treatment with clindamycin hydrochloride, trimethoprim-sulfadiazine, and pyrimethamine, or clindamycin alone.

Babesiosis

B. canis and *B. gibsoni* are most common etiological agents. In some infected dogs, the intracellular replication in red blood cells (RBCs) results intravascular hemolytic anemia. Immune-mediated reactions against the parasite or altered self-antigens worsen the hemolytic anemia. Clinical manifestations are those of acute anemia and include fever, pale mucous membranes, tachycardia, tachypnea, depression, anorexia, and weakness. Icterus, petechiation, azotemia, and hepatosplenomegaly are present in some dogs depending on the stage of infection and the presence of disseminated intravascular coagulation (DIC).Definitive diagnosis is based on organism demonstration in RBCs

using Wright's or Giemsa stains on thin blood smears. *B. canis* is typically found as paired, piriform bodies. *B. gibsoni* is typically found as single, annular bodies. Diaminazeneaceturate can be used for treatment for *Babesia canis* infection. dogs with suspected *B. canis* associated clinical illness often respond to imidocarbdiproprionate administered at 5 to 6.6 mg/kg SC or IM twice, 14 days apart or 7.5 mg/kg, SC or IM. Dogs with suspected *B. gibsoni* associated clinical illness often respond to azithromycin (10 mg/kg, PO, q24hr for a minimum of 10 days) combined with atovaquone (13.3 mg/kg, PO, q8hr for at least 10 days).If these drugs are not available, clindamycin can be administered at 12.5 mg/kg, PO, q12h for at least 10 days for controlling clinical signs.

MISCELLANEOUS CLINICAL DISORDERS

Sl. No	Disease	Symptoms	Diagnosis	Treatment
1	**Atopic dermatitis** Inherited predisposition to develop allergic symptoms.	Atopic animals usually rub, lick, chew, bite, or scratch at their feet, muzzle, ears, armpits, or groin, causing hair loss and reddening and thickening of the skin.	Noting seasonality to the skin problem.	Avoidance of the allergic substance. Antihistamines and fatty acids are two therapies. Steroids can also be used to alleviate the itch if other therapy is ineffective to control a severe itch
2	**Cushing's syndrome** By administration of corticosteroids (Prednisolone, Methylprednisolone, Triamcinolone and Dexamethasone)	Polydypsia, polyuria, Alopecia, loss of regrowth of hair commonly on back, tail, and the back of the rear legs, Muscle weakness, pot-belly, excessive panting and food intake	Radiography of abdomen to rule out tumours of adrenal gland Estimation of cortisol to creatinine ratio (normal-<10)	Surgery or oral drugs mitotane is 30 to 50 mg/kg/day, administered for 10 days Concurrent glucocorticoid supplementation with prednisone or prednisolone (0.15 to 0.25 mg/kg/day to avoid adverse affects by fall in cortisol level
3	**Hypothyroidism** Thyroid hormone deficiency	Weight gain, Cold intolerance, Lethargy, Skin problems, Abnormalities of the reproductive and ervous systems	By measurement of thyroid hormone concentrations (T3, T4, and free T4) and TSH.	Treatment involves daily or twice-daily oral medication with synthetic thyroxine (l-thyroxine 20 µg/kg)
4	**Colitis** Colitis refers to the inflammation of the colon	Straining to defecate, Bright red blood on the stool, Fecal mucus and Increased frequency of defecation	Parasitological and cultural examination Colonoscopy Radiography	An antibiotic based on fecal culture Antiparasitic agents Fiber supplementation

5	**Diabetes mellitus** An absolute or relative insulin deficiency	Polydipsia, polyuria, polyphagia and weight loss	Finding sugar in the urine and an abnormally increased blood sugar concentration.	Recombinant human NPH or pork lente insulin at an approximate dosage of 0.25 U/kg twice a day. Oral sulfonylurea drugs (e.g., glipizide, glyburide) Acarbose for treating poorly controlled diabetic dogs Diets containing increased fiber content
6	**Acute Renal Failure**	Oliguria, Vomiting, History of using any nephrotoxic drugs etc.	Hematology, Biochemistry, Ultrasonography and Radiography	Management of fluid balance Antibiotic if infectious in origin Diuretics Supportive therapy
7	**Eclampsia** Hypocalcemia in bitches	History of late pregnancy or early lactation Temperature-103-108^{0}F. Animal is frantically excited. Bitch becomes staggery, and suffers from tonoclonic convulsions	Serum calcium concentration less than 7 mg/dl QT interval of ECG prolonged Ventricular premature contractions	Separate puppies from bitch 10% calcium gluconate @0.5-1.5 ml/kg/hr IV Cortcosteroids if hypoglycemia also present. If too much excitement, sodium phenobarbitone should be administered IV

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INTRODUCTION TO FELINE DISEASES

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Cats are truly wild animal and a common pet also. Since the number of feline cases is rising, it is essential for a veterinarian to have a better knowledge on cats. Cats are adapted for utilization of protein and fat. They have only a fewer amounts of digestive enzymes of carbohydrate also. Essential aminoacids for cats include taurine, arginine, methionine and cysteine. Tyrosine is conditionally essential. Methionine and cysteine are gluconeogenic aminoacids. Essential vitamins needed for cats include Vitamin A, B (B1, B5, B6 and B12) and D. Cats can't convert β-carotene to Vitamin A and is directly got from animal tissue. They have no dermal photosynthesis and Vitamin D is derived from animal liver and fat. These all factors should be kept in mind while handling with feline diseases.

Feline Viral Respiratory Disease

The two main causes of viral respiratory disease in cats are feline herpesvirus (FHV-1) (feline rhinotracheitis virus) and feline calicivirus (FCV). Bacterial pathogens such as *Bordetella bronchiseptica* and *Chlamydophilafelis* may also be involved in infectious respiratory disease in cats. FHV-1 causes upper respiratory tract (URT) disease, with oculonasal discharges, conjunctivitis, sneezing, and sometimes hypersalivation and coughing. Occasionally more severe signs including pneumonia and generalized disease may be seen, particularly in young or debilitated animals. In FCV infection, the most characteristic sign is oral ulceration, typically on the tongue, but lesions may sometimes occur elsewhere in the mouth or on the skin along with ocular and nasal discharges and conjunctivitis. Treatment involves both broad-spectrum antibacterial cover, and supportive therapy. Tetracyclines are indicated if *Bordetella bronchiseptica* or *Chlamydophilafelis* are involved.Prevention and control may be achieved through a combination of vaccination and management.

Feline Panleukopenia/Feline Parvovirus (FPV) Infection

Feline panleukopenia is caused by a parvovirus, a small, nonenveloped DNA virus similar to the canine parvovirus type 2 (CPV-2). The disease is characterized by enteritis and a panleukopenia and has a high mortality. Profuse watery diarrhoea or dysentery then develops and cats may become severely dehydrated. Vaccination is generally highly effective in controlling the disease.Treatment is largely supportive and is aimed at restoring the fluid and electrolyte imbalance and covering against secondary bacterial infection.

Feline Infectious Peritonitis and Feline Coronavirus Infection

FIP is an immune-mediated disease, triggered by infection with a feline coronavirus (FCoV) that belongs to the family Coronaviridae, enveloped, positive-stranded RNA viruses.Affected cats develop signs caused by granulomatous lesions in the target organs (central nervous system, eyes, and parenchymatous organs) and vasculitis leading to fluid redistribution into third spaces with fluid accumulation in body cavities, including pericardium. Intestinal and dermatological disorders are commonly noticed in this disease. Rivalta's test is a test used to diagnose FIP. Every cat with confirmed FIP dies from the disease.

Other Viruses causing Feline diseases: Feline Immunodeficiency Virus, Feline Leukemia Virus, Bornavirus Disease (Feline "Staggering Disease") etc.

Feline *Bartonella*

Symptoms include mild neurologic signs (nystagmus, whole body tremors, focal motor seizures, either decreased or exaggerated responses to external stimuli, behavior changes), and epaxial muscle pain in a few experimentally inoculated cats. Some cats were lethargic and anorexic during febrile periods. Enrofloxacin (5.4 to 7.6 mg/kg, given PO every 12 hours) for 14 or 28 days clears*B. henselae* or *B. clarridgeae* infection. Erythromycin, amoxicillin, amoxicillin-clavulanate, and tetracycline, rapidly decreases the level of bacteremia in infected cats. Prevention of *Bartonella* infections in cats is best accomplished by avoiding exposure to infected animals and fleas.

Feline infectious anaemia

Feline infectious anaemia is caused by *Haemobartonella felis*, belonging to the family *Anaplasmataceae*. Recovered animals may remain asymptomatic carriers. Peracute form of disease shows anemia, immune suppression with high parasitaemia; while acute form may show signs of fever, anemia, depression, weakness and occasionally, jaundice. Chronic form leads to symptoms such as anaemia, lethargy and marked weight loss. The parasite may be demonstrated on the surface of erythrocytes in Giemsa stained blood smear. Also animal may have reduced PCV. Doxycycline hydrochloride @ 5 mg/kg is the treatment of choice.

Feline Ehrlichioses

Cats are rarely found to be infected with ehrlichiae. Clinical signs including fever, lethargy, anorexia, pallor, and splenomegaly were reported in these cats, although some had concurrent infections with hemotropic mycoplasmas or feline retroviruses. Cats infected with monocyticehrlichiae should be treated with doxycycline at 10 mg/kg PO q24h for a minimum of 28 days.

Toxoplasma Gondii

Toxoplasma gondii is one of the most prevalent parasites infecting warm-blooded vertebrates. Only cats complete the coccidian life cycle and pass environmentally resistant oocysts in feces. Cats may show clinical signs such as diarrhea.Most clinical signs are systemic and can include death in dogs and cats that develop due to intracellular replication of tachyzoites after primary infection; hepatic, pulmonary, CNS, and pancreatic tissues are commonly affected by tachyzoites.Common clinical findings in cats with disseminated toxoplasmosis include

depression, anorexia, fever followed by hypothermia, peritoneal effusion, icterus, and dyspnea.Clindamycin hydrochloride, trimethoprim-sulfonamide combination, and azithromycin can be used successfully for the treatment of clinical toxoplasmosis.

Endoparasites

Endoparasites are common in cats, especially in kittens. They includes Toxocara cati, T. leonina, T. canis, Ancylostoma brazilienses and Uncineria stenocephala. Lack of growth and loss of condition of cats can occur. Diarrhea with mucus may be evident. Diagnosis is performed by microscopic examination of feces for eggs. Treatment can be based on type of worm causes the disease. Coccidiosis can be caused by *Isospora bigemia*, *I. Felis* and *I. revolta*. Treatment is same as that of dog only.

Vaccination Schedule

CORE VACCINES	PRIMARY KITTEN SERIES (≤16 WEEKS)	PRIMARY ADULT SERIES (>16 WEEKS)	BOOSTER INTERVAL
Parvovirus (panleukopenia)	Administer one dose as early as 6 weeks of age, then every 3-4 weeks until 16 weeks of age	Administer two doses, 3-4 weeks apart	Administer one dose 1 year following completion of the initial series, then every 3 years thereafter Note: Annual booster of cats against FHV-1 and FCV may be recommended in cats housed in high-risk environments
Herpesvirus-1 and Calicivirus Modified-live (nonadjuvanted), or Killed (adjuvanted) (SQ or intranasal administration)	Same as above	Same as above	Same as above
Rabies Recombinant (nonadjuvanted) (SQ injection)	Administer one dose at 12-16 weeks of age	Administer one dose	Annually
Rabies Killed—1 Year Killed—3 Year (adjuvanted) (SQ injection)	Administer one dose at 12-16 weeks of age	Administer one dose	Administer one dose 1 year following administration of the first dose, then every 3 years thereafter

Drugs Contraindicated in Cats

Acetylsalicylic acid (aspirin) and salicylates, Acetaminophen (Paracetamol), Propofol, Carprofen (long-term use) and Piroxicam are contraindicated in cats since they cause toxicity. Although Bismuth sub salicyclate (Pepto-bismol), Buterphenol, Chlorpromazine HCL, Codeine + Acetaminophen, Chlormphenicol (Kittens), Dopamine HCl, Enrofloxacin, Gentamicin, Pentobarbitol, Phenobarbitol, Tetracycline and Tiletaminealso have to be administered cautiously.

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APPLICATIONS OF ECG IN CANINE PRACTICE

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Electocardiography in clinical practice is the recording at the body surface of electrical fields generated by the heart. It is a basic and valuable diagnostic technique in veterinary medicine and is relatively easy to acquire.

PRINCIPLE

Electrical impulses in the heart are recorded and amplified on a moving strip of paper which allows to analyze pace maker of the heart (SA node). Small metal contacts called electrodes are placed on the skin of the patient to measure the flow and direction of the electrical currents in the heart during each heart beat. Each of the electrodes are connected by a wire to a machine that will produce tracing for each electrode. This tracing represents lead of the heart's electrical patterns. Surface electrodes are placed in a designated fashion to obtain standard electocardographic leads. ECG signals are typically in the range of ±2 mV and require a recording bandwidth of 0.05 to 150 Hz.

Electro cardiograph is the representation of vector principle. Galvanometer writes on paper. Paper moves from left to right. It is based on 2 essential and fundamental principle: 1. When an electromagnetic force flows, or is directed, towards the positive electrode of a lead, the electrocardiograph will record an upward or positive deflexion. 2. When an electromagnetic force flows, or is directed, away, from the positive electrode of a lead and thus towards the negative electrode of the lead, the electrocardiograph will record a downward or negative deflexion.

USES

1. Evaluation of cardiac diseases
Arrhythmia, Hypertrophy, Inadequate blood supply to the heart, inadequate oxygen supply to the heart and Pericardial effusions
2. Differentiation of nonspecific diseases that cause fatigue, lethargy, weakness, fever, collapse or seizures.
3. Using this technique, origin of abnormal rhythms in heart can be determined.
4. It detects not only heart diseases but also metabolic problems.

WAVE FORM OF ECG

Einthoven chose the letters of the alphabet from P to U to label the waves and to avoid conflict with other physiologic waves being studied at the turn of the century. As the heart beat begins with an impulse from the sinoatrial node, impulse will first activate the upper chambers of the heart or atria and produce the P wave. Then the electrical current will flow down to the lower chambers of the heart or ventricles producing the Q, R and S waves. As the electrical current spreads back over the ventricles in the opposite direction it will produce the T waves.

Feature	Description
RR interval	The interval between an R wave and the next R wave. It is the inverse of the heart rate

P wave	During normal atrial depolarization, the main electrical vector is directed from the SA node towards the AV node and spreads from the right atrium to the left atrium.
PR interval	The PR interval is measured from the beginning of the P wave to the beginning of the QRS complex. The PR interval reflects the time the electrical impulse takes to travel from the sinus node through the AV node and entering the ventricles. The PR interval is therefore a good estimate of AV node function.
PR segment	The PR segment connects the P wave and the QRS complex. This coincides with the electrical conduction from the AV node to the bundle of His to the bundle branches and then to the Purkinje Fibers.
QRS complex	The QRS complex reflects the rapid depolarization of the right and left ventricles. They have a large muscle mass compared to the atria and so the QRS complex usually has a much larger amplitude than the P-wave.
QT interval	The QT interval is measured from the beginning of the QRS complex to the end of the T wave.
T wave	The T wave represents the repolarization of the ventricles. The interval from the beginning of the QRS complex to the apex of the T wave is referred to as the *absolute refractory period*. The last half of the T wave is referred to as the *relative refractory period* (or vulnerable period). The wave is normally rounded and positive.
ST interval	The ST interval is measured from the J point to the end of the T wave. Iso electric segment. Represents the period of ventricular muscle contraction before repolarization.

P WAVE

Atrial contraction begins at about the middle of the P wave and continues during the PR segment. During normal atrial depolarization, the main electrical vector is directed from the SA node towards the AV node, and spreads from the right atrium to the left atrium. It is usually smooth and positive. A negative P-wave can indicate depolarization arising from the AV node. **Wide and tall P wave is seen in Biatrial enlargement.**

PR SEGMENT

PR segment corresponds to the time between the ends of atrial depolarization to the onset of ventricular depolarization. PR segment is an isoelectric segment, that is, no wave or deflection is recorded. During the PR segment, the impulse travels from the AV node through the conducting tissue (bundle branches, and Purkinje fibers) towards the ventricles. Most of the delay in the PR segment occurs in the AV node. Although the PR segment is isoelectric, the atrial are actually contracting, filling the ventricles before ventricular systole.

QRS COMPLEX

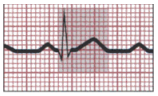
QRS complex represents the time it takes for depolarization of the ventricles. Activation of the anterioseptal region of the ventricular myocardium corresponds to the negative Q wave. Activation of the rest of the ventricular muscle from the endocardial surface corresponds to the rest of the QRS wave. Q wave is not always present. R wave is the point when half of the ventricular myocardium has been depolarized. Activation of the posteriobasal portion of the ventricles give the RS line. Normal ventricular depolarization requires normal function of the right and left bundle branches. A block in either the right or left bundle branch delays

depolarization of the ventricles, resulting in a prolonged QRS duration.**Wider or taller QRS complex are the peculiarity of Left ventricular hypertrophy.**

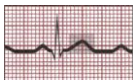
QT INTERVAL

QT interval begins at the onset of the QRS complex and to the end of the T wave. It represents the time between the start of ventricular depolarization and the end of ventricular repolarization. It is useful as a measure of the duration of repolarization. QT interval will vary depends on the heart rate, age and gender. It increases with bradycardia and decreases with tachycardia. Male has shorter QT intervals than female. This interval is influenced by electrolyte balance, drugs, and ischemia. **Prolonged QT interval is noted in Hypokalemia and hypocalcemia.**

T WAVE

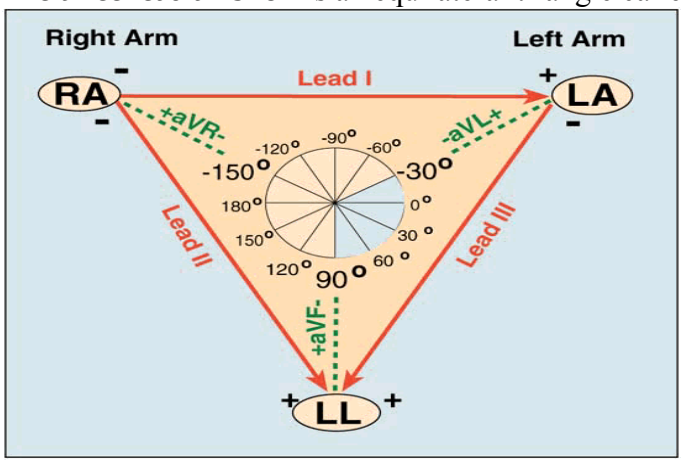
T wave corresponds to the ra  repolarization. The wave is normally rounded and positive. It can become inverte lattened due to electrolyte imbalance, hyperventilation, CNS disease, ischemia or myocardial infarction. **Tall and peaked T wave is seen in Hyperkalemia. Small and biphasic T wave occur in hypokalemia.**

STANDARD LEADS

Einthoven deliberately placed the electrodes of the  d limbs as far as away from the heart as possible i.e. on the right arm, left arm and l e three electrodes thus electrically equidistant from the heart. Standard bip ord electrical potential difference between 2 electrodes on body surface. The lead axis is oriented between these 2 points. The leads derived from three electrodes are:

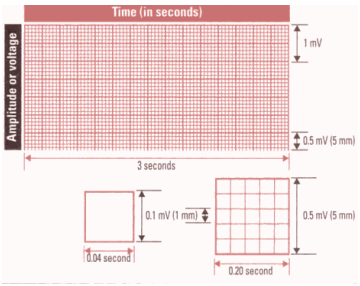
1. Standard lead I: negative electrode on the right arm and positive electrode on left arm
2. Standard lead II: negative electrode on the right arm and the positive electrode on the left foot
3. Standard lead III: negative electrode on the left arm and the positive electrode on the left foot.

The three lead axis forms an equilateral triangle called as the Einthoven's triangle.



ELECTROCARDIOGRAM

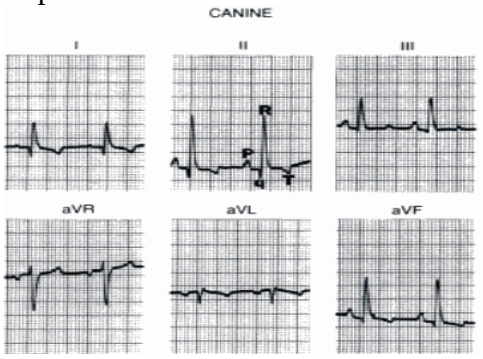
The electrocardiogram is recorded on to standard paper travelling at a rate of 25 mm/s or 50mm/s. The paper is divided into large squares. Each large square is five small squares in width. At a speed of 25 mm/s, width of small square: 1 mm equivalent to 0.04 s.At speed of 50 mm/s width of small square is equal to 0.02 s. Height of one small box is o.1 mV

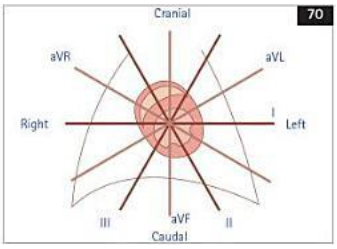


MEAN ELECTRICAL AXIS

The mean electrical axis is the principal vector of ventricular depolarization. It describes average direction of ventricular depolarization process in the frontal plane. It represents the summation of various instantaneous vectors that occur from the beginning until the end of ventricular muscle activation. It is the sum of all the waves of depolarization that are occurring simultaneously. The mean electrical axis normally points toward the left ventricle, as this is larger of the two ventricles. Deviations of the mean electrical axis result from either right ventricular hypertrophy or a block in the intraventricular conduction system.The normal range of the mean electrical axis in dogs is +40 to +100 degrees. In dogs, a right axis shift is defined as a mean electrical axis between +100 and -90 degrees. A left axis shift occurs in dogs when the mean electrical axis is between +40 and -90 degrees and in cats when the mean electrical axis is between 0 and -90 degrees.

To determine the mean electrical axis, we must obtain a six-lead ECG from a patient positioned in right lateral recumbency with the limbs perpendicular to the long axis of the animal's body. For systemic analysis of direction and magnitude of electrical axis of heart, 6 basic limb leads are arranged in circular fashion according to direction of each lead and location of positive electrodes.





BASICS OF INTERPRETATION

Amplitude of R wave is proportional to how parallel the wave of depolarization is to that lead. When a wave of depolarization is moving toward the positive pole, the more directly parallel the wave front is to the lead, the taller the R wave will be. The more perpendicular the wave of depolarization is, the smaller the resulting R wave will be. A wave of depolarization that is travelling 90 degrees perpendicular to a lead will result in an isoelectric QRS complex (Isoelectric means that the positive deflection of the R wave and the negative deflection of the Q and S waves are equivalent). Positive and negative deflections of the QRS complex sum to zero.

The electrocardiogram should be interpreted in conjunction with a complete data base. For evaluation of cardiovascular system, this database consists of history, physical examinations, radiographic studies and laboratory profile. History should include age, sex, breed, weight, medication (especially digitalis) and associated disease. Laboratory profile includes

hematology, urinalysis, and evaluation of extra vascular fluids. An echocardiography study also may be necessary. Atleast 4 features of electrocardiogram should be examined systemically: heart beat, heart rhythm, PQRST complexes and intervals, and mean electrical axis.

STEPS IN ANALYSIS OF ELECTROCARDIOGRAM

1. Calculate heart rate
2. Evaluate heart rhythm
3. Measure the complexes and interval
   a. P Wave
   b. PR Interval
   c. T wave
   d. QT Interval
   e. QRS Complex
   f. ST segment
   g. Basic limb leads (I,II, III, aVR, aVL & aVF)
4. Determine the mean electrical axis

Rate 70-160bpm for adult dogs
60-140 bpm for giant breeds
Up to 180 bpm for toy breeds
Up to 220bpm for puppies

Rhythm
Normal sinus rhythm
Sinus arrhythmia
Wandering SA pacemaker

Measurements (Lead II (50mm/sec, 1 cm=1mV))

P Wave

Width Maximum- 0.04 sec (2 box wide)
Maximum - 0.05 sec (2 ½ box wide) in giant breeds

Height Maximum - 0.4 Mv (4 boxes tall)

PR interval

Width - 0.06- 0.13 sec (3-6 1/2 boxes)

QRS Complex

Width Maximum - 0.05 sec (21/2 boxes) in small breeds
Maximum - 0.06 sec (3 boxes in large breeds)

Height of R wave Maximum - 3 Mv (30 boxes in large breeds)
Maximum - 2.5 Mv (25 boxes) in small breeds

ST Segment

No depression. Not more than 0.2 Mv (2 boxes)
No elevation. Not more than 0.15 Mv (1 ½ boxes)

T wave

Can be positive, negative or diphasic
Not greater than ¼ amplitude of R wave
Amplitude range - 0.05 to 1 Mv (1/2 to 10 boxes in any lead)

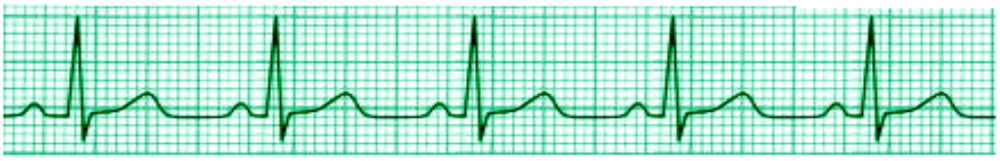
QT Interval

Width - 0.15-0.25 sec (7 ½ to 12 ½ boxes at normal heart ratevaries with heart rate) varies with heart rate (faster rates have shorter QT interval and Viz)

Electrical axis frontal plane - +40 - +100
Values of special importance (Precordial chest leads)
CV5RL (rv2): T wave positive. R wave not greater than 3 mV (30 boxes)
CV6LL (V2): Not greater than 0.8 mV (8 boxes). R wave not greater than 3 mV (30 boxes) (not valid for thin deep chested dogs under 2 years of age)
CV6LU (V4): s wave not greater than 0.7 mV (7 boxes) R wave not greater than 3 mV (30 boxes)
V10: Negative QRS Complex (T wave negative except in chihuahua)

DETERMINING HEART RATE AND RHYTHM

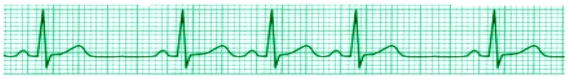
Method I



Examine the distance between QRS complexes and determine if the peaks (RR intervals) are regularly spaced. If the RR distances are regular, count the number of "small boxes" from the beginning of one QRS complex to the beginning of the next QRS complex. Then divide 1500 by the number of "small boxes" to obtain the heart rate in beats per minute.

$$\text{Heart Rate} = \frac{1500}{\text{No. small boxes}}$$

If the distances are irregular, count the number of QRS complexes within 30 large boxes (each represent 0.2 seconds) and multiply this number by 10 to obtain the heart rate in beats/minute.

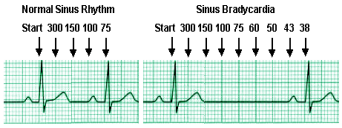


Method II

If the peaks are regular, the heart rate can be estimated using the ECG grid. To do this locate a QRS complex on a bold line. If the next QRS complex is separated by:
i. One large box, the heart rate is 300 BPM (300/1)
ii. Two large boxes, the heart rate is 150 BPM (300/2)
iii. Three large boxes, the heart rate is 100 BPM (300/3)
iv. Four large boxes, the heart rate is 75 BPM (300/4)

...and so on.

The ECG grid is used to estimate the heart rate rapidly



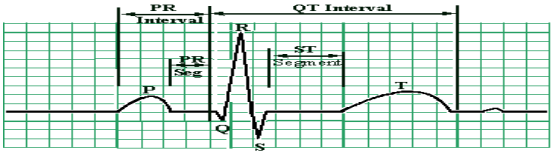
THE DIFFERENCE BETWEEN ATRIAL AND VENTRICULAR RATES
The heart rate calculated using the RR intervals is the ventricular rate. In sinus rhythm,
The ventricular rate corresponds to the atrial rate. The atrial rate can be determined from the PP interval using either of the two methods.

CARDIAC ARRHYTHMIAS

Arrhythmia is a generalized term used to denote disturbances in the heart rhythm.

Normal sinus rhythm is characterized by a regular rhythm and normal PR interval duration. The following 5 steps can be used for identifying arrhythmias.

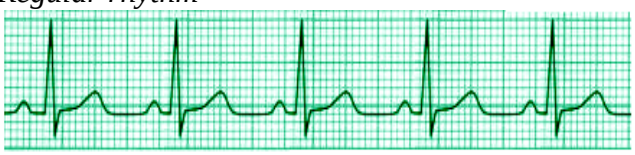
1. After finding out the patient's medical history, begin by labeling the P wave, PR interval, QRS complex, QT interval and T wave.



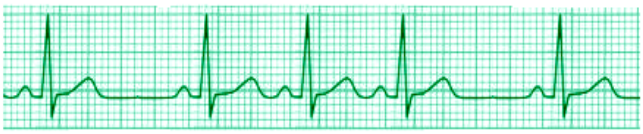
2. Calculate the atrial and ventricular heart rates. A rate of less than 60 beats per minute is slow (*Brady*), 60-160 beats per minute is normal and greater than 160 beats per minute is fast (*tachy*).

3. Determine if the rhythm is regular or irregular. This is done by accessing whether the RR intervals and PP intervals are regularly spaced.

Regular rhythm



Irregular rhythm



a. If the rhythm is irregular, determine if:

i. It is occasionally irregular
ii. Regularly irregular (there is a pattern to the irregularity)
iii. Irregularly irregular (there is no pattern to the irregularity)

4. Evaluate the waveform of the ECG in detail for additional clues:

a. Determine the shape of the P-wave.

Normally, P-waves are upright and each P wave is related to a QRS complex. If inverted, the impulse is spreading from the ventricles to the atria in a retrograde manner.

b. Determine if the PR interval is of normal length
c. Examine the QRS complexes and determine if the QRS complex is wide or narrow.

Narrow QRS complexes indicate that the rhythm is supraventricular (originating from above the ventricles). Wide QRS complexes indicate that the rhythm is originating in the ventricles or that there is an intraventicular block.

d. Determine if the ST segment is displaced from the mid-line.

5. Review the patient's medical history, assess the patient to ensure that the assessment and rhythm agree. The following ECG clues can be used to recognize cardiac arrhythmias in non sinus rhythm ECGs.

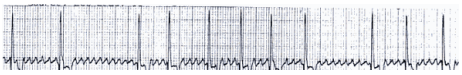
A. If the rhythm is regular but too fast or slow, it could be an indication of either:

a. Sinus bradycardia: Rhythm is regular and looks normal but is slower than 60 beats per minute.The RR interval is longer, often more than one second. P waves are present and regular and each P-wave is followed by a QRS complex in a ratio of 1:1.

b. Sinus tachycardia: Rhythm is regular and looks normal but at a rate greater than 160 beats per minute. R-R interval is shorter. P waves are present and regular and each P-wave is followed by a QRS complex in a ratio of 1:1.

B. If rhythm is irregular

a. Atrial flutter: Atrial flutter waves (F-waves) with a characteristic saw-tooth form will also be observed at a rate of 200-350 BPM.

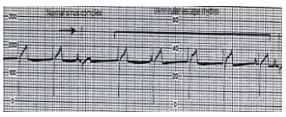
b. Atrial fibrillation: No P-waves will observab  l.

C. If there are no P-waves, it could be an indica

a. Atrial fibrillation: No P-waves will observable. Rather, a wavy base-line is recorded.

b. Sinus arrest: with junctional or ventricular escape.

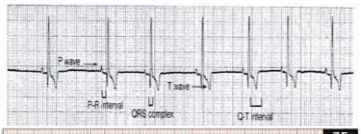
Ventricular escape rhythm

D. If P-waves are not associated with QRS complexes, it c 

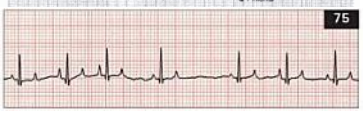
1. Ventricular tachycardia

2. Third degree AV block: Dissociation between the atriur :o complete block.

NORMAL AND ABNORMAL ELECTROCARODIOGRAMS

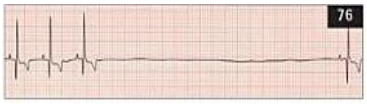


Normal canine ECG



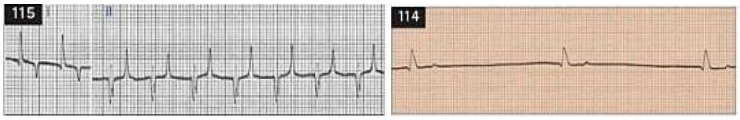
Wandering pacemaker

Cyclic changes in P wave configuration, related to shift in pacemaker location. P wave becomes spiked in inspiration and flatter in expiration. Sinus arrhythmia occur commonly in dogs especially brachycephalic dogs.



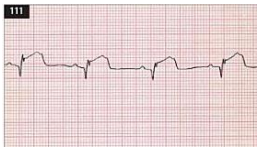
Sinus arrest

QRS-QRS interval will be twice the original.



Marked hyperkalemia

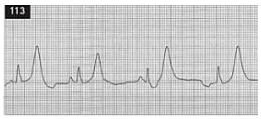
Marked hyper kalemia- peaked T Waves, loss of P waves, widened QRS complexes



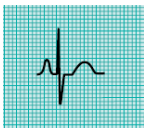
Electrical alternans

Every other beat alteration in QRS complex size, associated with large volume pericardial effusions.

Stenosis of pulmonary valve

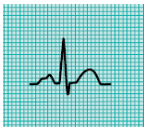


ST segment elevation



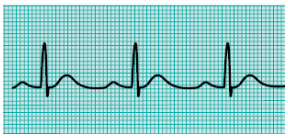
Right atrial hypertrophy

Peaked P waves (P pulmonale)



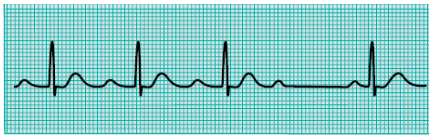
Left atrial hyper trophy

Bifid P wave



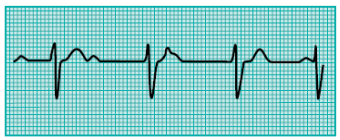
First degree AV block

A long PR interval



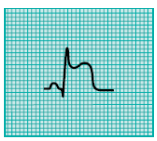
Second degree AV block

Occasional non conducted P wave



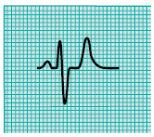
Third degree AV block

P wave fail to conduct



Myocardial infarction

ST elevation



Hyperkalemia

T wave elevation

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COMMON PARASITES FOUND IN PET ANIMALS

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Dogs and cats are not just pets. They are treated like members of the family. And like any member of your family, it's important to keep your companion animal healthy and free of parasites. It is fairly common for a dog or cat to become infected with an internal or external parasite at some point in its lifetime. Parasites can affect your pet in a variety of ways, ranging from simple irritation to causing life-threatening conditions if left untreated. Some parasites can even infect and transmit diseases to you and your family. Your veterinarian can help prevent, accurately diagnose and safely treat parasites and other health problems that not only affect your dog or cat, but also the safety of you and your family.

Parasites that may affect your pet

- Coccidia
- Giardia
- Mange Mites
- Ticks
- Ear Mites
- Heartworms
- Roundworms
- Toxoplasmosis
- Fleas
- Hookworms
- Tapeworms
- Whipworms

Common External Parasites parasites of Dogs & Cats
Biting insects (hematophagous), i.e. they suck blood

- **Fleas.** Probably the most frequent and universal parasite of dogs worldwide, and potentially very harmful. Not dangerous for humans, but extremely annoying. Widespread **resistance** to several insecticides.
- **Stable flies.**Usually a minor problem for dogs, worldwide but mainly in rural regions. May **transmit various diseases**.
- **Mosquitoes.** Usually not a serious threat by themselves, but are **vectors of several diseases**, notable heartworms (*Dirofilaria* spp).
- **Bed bugs.** An increasing problem, but less for the dogs than for their owners. Worldwide.
- **Horseflies.**Usually a minor problem for dogs, worldwide but mainly in rural regions. Their bites can be quite painful and are also **vectors of various diseases**.

Non-biting insects, do not suck blood

- **Houseflies.** Usually a minor problem for dogs, worldwide but mainly in rural regions. Widespread **resistance** to several insecticides. May **transmit some dog diseases**.
- **Filth & nuisance flies.**Usually a minor problem for dogs, worldwide but mainly in rural regions. Locally, **resistance** to several insecticides.
- **Lice.** Usually not a serious threat, neither for pets, nor for humans. Some species do suck blood.
- **Human bot flies**, *Dermatobia*. A problem in many regions of Central and South America.
- Screwworm flies. Usually not a very serious problem for dogs, unless in endemic regions in tropical and subtropical countries.

Ticks & mites

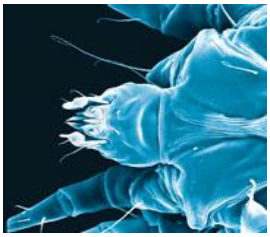
- Ticks. A considerable threat worldwide, especially in rural regions of tropical and subtropical countries. Ticks **transmit many dog diseases**.
- Mites. Occur worldwide but usually not the worse problem in dogs

Ticks

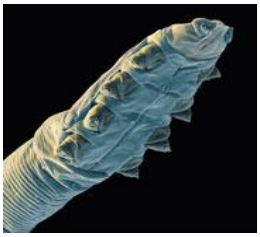
Usually grey and round, ticks have eight legs and two body parts. Ixodes ricinus is the most common tick in the UK. Ticks lie in wait in vegetation & long grass and attach to passing dogs and cats (and people). As well as producing uncomfortable bites, ticks can (rarely) transmit nasty diseases (e.g. Lyme disease). Heavy infestation in younger animals can cause anaemia.

Ticks can be prevented by regular treatment: ask us for more advice.

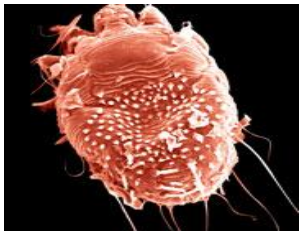
a. Tick



b. Ear mite



c. Demodex mite



d. Sarcoptic mite

Ear mites

Cats and dogs can both be affected by these 8 legged parasites. They live in the outer ear and can be a source of significant irritation. Cats and dogs can infect each other by direct contact: ear mites are very contagious. Humans are rarely affected.

Demodex

Demodecosis is mainly a disease of dogs: young and debilitated dogs are most at risk. Mites are 'cigar' shaped. Puppies are infected from their mothers by direct contact: otherwise the mites are not contagious. Low levels of demodex mites are present in a lot of dogs and cause no clinical signs. Occasionally, mite numbers spiral out of control leading to significant skin disease. The mite does not affect people.

Sarcoptic mange mite

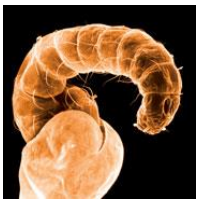
Sarcoptes is extremely contagious and is passed to dogs (and people) from other infected dogs and foxes. Sarcoptic mange is very itchy and often affects the ear tips, abdomen, elbows and hocks.

Lice

Lice are very small and are often confused with 'dander'. They are wingless insects with 6 legs and 3 body parts. They are host specific: dog lice can not affect cats (or people) and vice versa. Lice spread from dog to dog and cat to cat via direct contact. Mild louse infections often cause few clinical signs. However, heavy infections can cause skin irritations, skin damage and bacterial skin infections. Very young and debilitated patients are most at risk.

e. Louse

f. Flea



g. Flea larva

h. Lungworm

Fleas

Fleas are small wingless insects that are just visible with the naked eye. They are a few millimetres long and live on dogs and cats. They feed on blood and lay eggs that fall into the environment. Eggs develop into larvae and then pupae which, under the right conditions (e.g. heat and moisture), hatch into adult fleas and jump onto a passing dog or cat. Rabbits and people can also be affected. Uncommonly, fleas can jump from one dog or cat to another. Fleas are sometimes well tolerated by cats and dogs without causing significant skin disease. However, other pets with flea allergic dermatitis succumb to very itchy skin disease. Fleas can be prevented by regularly cleaning pets bedding. Regular vacuuming will reduce the egg burden in the environment. Treating the carpets and pets bedding with a veterinary recommended spray will also help.

Internal parasites (endoparasites, worms, helminths)
Predilection sites are indicated in braquets.

Gastrointestinal roundworms *(nematodes)*

- *Ancylostoma* spp**. Hookworms.** (Small intestine and *larva migrans*). A serious threat for dogs. Worldwide, with different regional incidence for the various species.
- *Baylisascaris procyonis.* **The raccoon roundworm.** (Small intestine and *larva migrans*). Not a major threat. Only in endemic regions with abundant raccoons.
- *Capillaria* hepatica. **Hairworms.** (Liver). Not a major problem. Worldwide, but with different regional incidence.
- *Gnathostoma* spinigerum. (Stomach and *larva migrans*). Usually a secondary problem in some endemic hot and humid regions.
- *Gongylonema* spp**.** (Esophagus and stomach). Not a major threat for dogs. Worldwide, but only regionally relevant.
- *Physaloptera* spp**.** (Stomach and small intestine). Usually a minor problem in endemic regions.

- *Spirocerca lupi***.** (Esophagus). Can be a serious threat for dogs in endemic zones, mainly in tropical and subtropical regions.
- *Strongyloides* spp**. Threadworms, pinworms.** (Small intestine and *larva migrans*). A serious threat for dogs, worldwide but especially in warm and humid rergions.
- *Toxascaris leonina.* (Small intestine). Usually a minor problem, worldwide.
- *Toxocara canis.* **The dog roundworm**. (Small intestine and *larva migrans*). One of the **most serious threats for dogs**, especially for puppies. Worldwide and very frequent everywhere.
- *Trichuris* spp**. Whipworms.** (Large intestine and *larva migrans*). Not the major problem worldwide, but occasionally serious.
- *Uncinaria stenocephala.* **The fox hookworm.** (Small intestine and *larva migrans*). A significant threat worldwide, but usually less serious than other hookworms (e.g. *Ancylostoma* spp).

Round worms (nematodes) in the eyes, skin, heart and other organs

- Angiostrongylus vasorum. ***The French heartworm.*** *(Lung arteries, occasionally heart). A serious threat for dogs, mainly in endemic zones in Europe.*
- *Dioctophyma renale.*. ***The giant kidney worm.*** *(Kidneys). Can be a significant problem in endemic zones.*
- Dirofilaria *spp.* ***Dog Heartworms****. (Lung arteries, occasionally heart). A* ***very serious threat for dogs****. Worldwide, but especially in tropical and subtropical regions with abundant mosquitoes.*
- Onchocerca lupi. *(Eyes). Occasional problem in endemic regions.*
- Pearsonema *spp (=* **Capillaria** *spp). **H-F**. **Hairworms, bladder worms.** (Bladder). usually a minor problem. Worldwide, with varying regional incidence.*
- *Thelazia* spp.***Eyeworms*** *(Eyes). Occasional problem in dogs. Worldwide but with endemic distribution.*
- *Trichinella spp****.*** *(Muscle, small intestine). A minor health problem for dogs. Worldwide, but only in endemic zones, mainly in rural regions.*

Flukes (trematodes, flatworms)

- *Alaria* spp***.*** *(Small intestine). Usually a minor issued for dogs. Worldwide, but only in endemic regions.*
- *Dicrocoelium* spp***.Lancet flukes****. (Bile ducts and gall bladder). An occasional problem for dogs. Worldwide.*
- *Fasciola hepatica*.***The common liver fluke.*** *(Biliary ducts and gallbladder). Mostly an uncommon and not very threatening problem for dogs. Worldwide, but mainly in rural regions.*
- *Heterobilharzia Americana*.***The dog Schistosome****. (Mesenteric veins). An occasional problem in North America.*
- *Opisthorchis felineus****. The cat liver fluke****. (Hepatic and biliary ducts). Very occasionally found in dogs. Worldwide but only in endemic regions.*

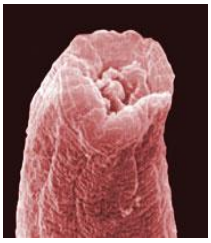
Tapeworms (cestodes)

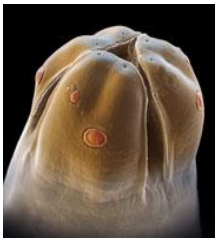
- *Dipylidium caninum*.***The flea tapeworm***. *(Small intestine). Usually not seriously harmful for dogs. But very frequent worldwide.*
- Echinococcus granulosus. ***The hydatid worm.*** *(In dogs, small intestine). Rather benign for dogs, but a serious threat for livestock and humans. Worldwide but mainly in rural zones of less developed regions.*
- *Echinococcus multilocularis*.***The small fox tapeworm.*** *(In dogs, small intestine). Rather benign for dogs, but a serious threat for livestock and humans. Worldwide but not very frequent.*
- *Mesocestoides* spp***.*** *(Small intestine). Usually not a serious threat for dogs. Worldwide, but not very frequent.*
- *Taenia* spp***.*** *(In dogs, small intestine). Usually not seriously harmful for dogs, but a significant problem for livestock (cysticercosis). Quite frequent worldwide*.

Lungworm

Please refer to our separate web pages on lungworm.

Intestinal Worms



i. Tapeworm **j. Whipworm** **k. Hookworm** **l. Roundworm**

Tapeworms

Adult tapeworms look like ribbons (divided into segments) and live in the small intestine. Adult worms release segments that are passed into the environment. The latter can sometimes be seen as small, white crawling 'rice grains' around your pet's tail base.Dogs and cats become infected by ingesting an intermediate host e.g. mice, fleas (usually accidental ingestion via grooming) and, less commonly, infected sheep carcases. Most tapeworms do not produce symptoms in your pet. Occasionally, they can cause irritation around your pet's bottom leading to 'scooting' across the ground. Rarely, tapeworms can be passed to people causing problems (zoonosis).

Whipworms

Whipworms have a thick head and whip like tail. They live in the large intestine of dogs & foxes only. Rarely, they can be passed to people. Dogs become infected by eating eggs in the

environment that have been passed in the faeces of other, infected dogs. Infections in adult dogs can cause diarrhoea. Heavy worm burdens in puppies can cause anaemia and poor growth.

Hookworms

About 10-15mm long, they live in the small intestine of dogs and foxes: infections in cats are very rare. They have cutting plates & teeth within their mouthparts that are used to feed on blood from the intestinal wall. Again, dogs become infected by eating eggs in the environment that have been passed in the faeces of other, infected dogs and foxes. Diarrhoea, lethargy and anorexia can result. Dogs can also become infected by ingestion of intermediate hosts (e.g. mice). Occasionally, hookworms can penetrate the skin around dog's feet causing infection.

Roundworms

Roundworms are long, cylindrical, white worms that live in the small intestines of dogs and cats. They feed on intestinal contents and pass eggs into the environment. Cats and dogs become infected either by ingestion of paratenic hosts (e.g. mice & birds) or through accidental ingestion via contaminated soil. Puppies and kittens can be infected from their mother's milk. Finally, puppies can be infected from their mother whilst developing in the womb. Infections in adult dogs and cats often cause no (or only minor) clinical signs (e.g. diarrhoea). Heavy burdens in puppies and kittens can cause anaemia, a 'pot belly' and poor growth.Humans are sometimes affected and infection can be serious.

Coccidia

Coccidia are single celled organisms that infect the intestines of dogs and cats. They can
only be detected by microscopic exam of your pet's feces. Your pet becomes infected by ingesting contaminated dirt/feces or intermediate hosts such as mice. This parasite is especially common in young and stressed animals. It causes bloody, watery diarrhea and sometimes death, especially in young animals. There are no products to kill the organism. Treatment is aimed at stopping reproduction of the organism so it is easier for the patient's immune system to take over and fight off the infection. Treatment usually lasts severalweeks.

Giardia

Giardia is a protozoal (one-celled) organism that parasitizes the small intestine of dogs and cats. It can also infect humans. Pets/people are infected by ingesting the cyst form of this organism from contaminated food and water. Diagnosis is made by microscopic exam of feces. It can be very difficult to detect. Symptoms of disease are chronic soft stools to profuse watery diarrhea. There is often mucous mixed in. Weight loss, lethargy, dehydration and anorexia may also occur. Treatment can be difficult as there is a high degree of recontamination of the patient and resistance to several drugs. Bathing the animal during treatment is recommended to help prevent reinfection by your pet licking cysts off of its fur while grooming.

Physaloptera (stomachworms)

Stomach worms are parasites of both cats and dogs. As the name implies, they live in the stomach. They are often treated for by your veterinarian if your pet has chronic vomiting. Eggs are not readily detected in stool samples. Your pet may occasionally vomit up a worm. Pets are infected by eating an intermediate host such as a cricket, cockroach or another animal which has ingested one of these insects. These parasites are also sometimes diagnosed by endoscopy. These parasites are not infectious to humans.

How to Prevent Parasites in Dogs and Cats

If your pet frequently goes outside and enjoys the freedom to roam, make sure you are doing everything you can to prevent intestinal parasites, or "worms" in your pet. Common parasites found in cats and dogs include:

- Roundworms
- Hookworms
- Whipworms
- Tapeworm**How to Diagnose Intestinal Worms in your Pet**

Your veterinarian can usually provide you with a deworming schedule that will prevent worms in a cat or dog before they become a problem. However, if your pet has any of the following symptoms, it could mean that your pet has become infected with worms once again, which is not unusual.Cats and dogs like to roam around in the woods, in creeks or streams, and where other animals have left droppings or carcasses. Here are common symptoms of intestinal worms in dogs or cats:

- Diarrhea
- Vomiting
- Bloody stools
- Distended stomach, or a "pot-belly" appearance

How to Protect your Pet from Intestinal Worms

Healthy pets may not show outward signs of a worm infection. However, if you notice a change in your pet’s ppetite or coat, diarrhea, or excessive coughing, see your veterinarian. In most cases, a simple fecal test can detect the presence of worm eggs or adults and, if present, your veterinarian will recommend a deworming program.Dewormers containing piperazine or pyrantel are effective at killing hookworms and roundworms. Visit your local Tractor Supply Co. store to ask an expert team member about deworming your dog or cat.To treat whipworm or tapeworm in your dog or cat, you will need to get a dewormer that contains fenbendazole and praziquantel from your veterinarian.Treat intestinal worms in your pet quickly to reduce

discomfort in your dog or cat and to reduce the risk of your pet infecting another animal or a human.Most worm infections can be prevented by regular treatment: ask us for more advice. In addition, try not to let your dog eat other dog's poo. Sniffing around other dog's bottoms should also be prevented. Keeping your pets bedding clean and regularly cleaning litter trays will also help. Keep pets well groomed and free of fleas. Try to stop your pet eating wildlife (e.g. a cat safe collar with a bell on it to hinder hunting behaviours).

Nursing female dogs and cats and their litters are also major sources for the spread of infective eggs and larvae. If you have a new puppy or kitten, or a pregnant pet, consult with your veterinarian about a deworming program that will reduce your family's risk of infection. Worm infections in humans can be easily prevented by practicing good hygiene and sanitation. Children should be discouraged from eating dirt and should not be allowed to play in areas that are soiled with pet feces. Sandboxes should be covered when not in use. Adults and children should always ash their hands after handling soil and after contact with pets. Shoes should be worn when outside to protect feet from larvae present in the environment, and raw vegetables should be thoroughly washed because they may contain parasites from infected soil. Dog droppings should be immediately picked up from public areas and from your yard to reduce the chances of contaminating the soil. Keeping cats indoors is an effective way to limit their risk of exposure to roundworms.

Treatment

Over the years, the number and types of parasite treatments for small animals have grown remendously. There are two main approaches to parasite treatment--preventative treatments and curative treatments. Preventative treatments are given on a regular interval in an effort to keep animals from getting a large burden of parasites. Curative treatments are given only in response to the positive identification of parasites. As with most things, prevention is easier and better than treating a problem once it is present. Many of the newer anti-parasite medications aim at preventing a wide variety of parasites while many of the curative treatments only kill one or two types of parasites. A complete discussion of all of the available parasite medications is beyond the limitations of this article, thus only the newest, preventative, prescriptionproducts will be discussed. These include Advantage, Frontline, Revolution, Heartgard, Interceptor, Program, Sentinel, and Proheart.

Advantage and Frontline are monthly topicals that kill ectoparasites. Advantage will kill adult fleas and flea larva and Frontline will kill adult fleas and ticks. A new version of Frontline, called Frontline Plus, ill soon be available. This product will kill adult fleas, larva, and eggs.

Revolution is a monthly topical that will kill adult fleas, eggs, ear mites, roundworms and hookworms in cats and adult fleas, eggs, ticks, ear mites, and sarcoptic mange in dogs.

Program is an oral monthly that will kill flea eggs only. Program is also available in a long lasting injectable form.

Heartgard, Interceptor, and Sentinel are oral monthly tablets that will kill a variety of parasites. Heartgard will kill immature heartworms, roundworms, and hookworms. Interceptor will kill immature heartworms, roundworms, hookworms, and whipworms. Sentinel is the

combination of Interceptor and Program and thus will kill immature heartworms, roundworms, hookworms, whipworms, and flea eggs.

Proheart is the newest heartworm medication availalble for use in dogs. It is an injection that is given twice a year. Proheart will kill immature heartworms and hookworms.

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EXAMINATION OF THE COMMON PARASITES OF PET ANIMALS FROM STOOL SAMPLE

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Intestinal parasites are one of the most common problems veterinarians see in dogs. Although pets of any age can carry them, they are a health a problem primarily in young dogs, dogs whose life style increases their risk of exposure, dogs living in sub-standard conditions and dogs with other health issues. Dogs and their parasites have had millions of years to evolve together. During that time, they have, begrudgingly, learned to tolerate each other. There are a large number of these worms and assorted freeloaders. Parasites are often "silent" and you will not know you pet has them. The more common intestinal parasites have adapted so well to their hosts that they are living in balance and cause no observable health issues. But that can always change. It is when the parasites become too numerous for one reason or another that pet's health is affected. Because of their silent stealthy nature, our best approach is to try to keep your pets completely free of them before the balance becomes disturbed.

The most common early signs of intestinal parasites in puppies are poor growth (dull hair coat, scrawniness, lack of playful energy and diarrhea. Many of these puppies have bony bodies but potbellied, big tummies. Many are anemic. Puppies with parasite over-burden invariably grew up in poor sanitary conditions.The most common signs in older pets are lack luster, brittle hair coat, boniness, listlessness and diarrhea. Some of these older pets become picky eaters, in some, the opposite occurs - they are ravenous and some show no changes in appetite at all. In adult dogs with parasite problems, multiple health issues are common. This is because the poor sanitation that leads to parasite overload also increases their risk of exposure to other diseases and because their parasite burden lessens their resistance to other diseases.

Faeces intended for parasitological examination should be collected from the rectum unless the animal is observed in the act of defecation when the sample may be collected from the ground. In dogs and cat can usually be induced to defecate by inserting a moistened finger into the rectum and gently massaging with a rotary motion until the external sphincter relaxes. For dogs, moist cotton swabs, introduced into the rectum and rotated well to collect the maximum possible faecal matter and dipped in saline in a test tube, are useful for examination of eggs. Suitable containers for the dispatch of samples to the laboratory are: 30 ml wide mouthed screw capped bottles of glass or preferably of plastic, which should be filled to the top if possible so as to exclude air as much as possible and diminish the rate of development and hatching of the eggs. Faecal samples collected from the ground in a field in which the animals have been running, are less useful for diagnostic purposes, but where rectal samples cannot be obtained, these should be examined. Such samples should be selected from the top of the most recently dropped faeces, and several samples should be collected.

Method which are used for diagnosis of faecal sample are belowed

Direct smear method

It is possible to demonstrate the presence of eggs or larvae of helminths, by the examination of a thin smear of emulsified faeces. A small quantity of faeces is placed on a slide, mixed with some droplets of water and a cover slip is placed on the fluid. The slide is

investigated using a magnification of 10x and 40x. In good smear a person should be able to read small print through it (but not too clearly). If it is too thick or too thin, observation of the elements in the mount may be difficult.

Parasites stages found

- Coccidia and helminth eggs (Only when high numbers are present).
- Cestode and trematode eggs.

Concentration by sedimentation technique

Procedure:

Place a lump of faeces (5-10g or 5-10 faecal pellets of sheep or goats) in a cup or glass container.Add approximately 50-100 ml of tap water and mix thoroughly with a spatula/ glass rod until all the faecal material is broken down.The mixture is poured through a wire mesh sieve with an aperture of 500-800 μm to remove coarse large lumps. The strained fluid is collected in a bowl. The sieve is rinsed with water and the debris left on the sieve is discarded.Transfer the suspension to centrifuge tubes and centrifuge at 2000 rpm for 2 min.Discard the supernatant.Mix the sediment well and take a small quantity of it and mix it with a drop of water on a clean slide.Apply a cover slip and examine under low power objective of the microscope.Thickness of the smear should be such that if the slide is placed on a newspaper, you should be able to read the fine print through the smear.

Parasite stages found: Eggs of trematodes. Larvae of lung worms

PARASITESEGGS FOUND IN FAECES OF DOG AND CAT

Cestodes

Dipylidium caninum, Echinococcus spp. Taenia spp.

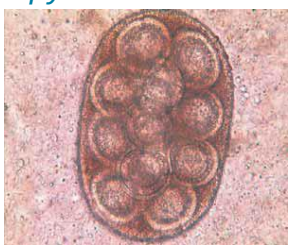
Nematodes

Ancylostoma caninum, Ancylostoma tubaeforme, Uncinaria stenocephala, Physaloptera spp., Toxascaris leonine, Toxocara canis, Toxocara cati, Baylisascaris procyonis,Trichuris vulpis

Protozoa

Giardia spp., Isospora (Cystoisospora) spp.,Toxoplasma gondii,Cryptosporidium spp.

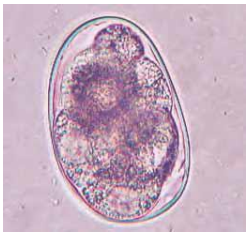
Dipylidium caninum



Echinococcus spp.

Taenia spp

Ancylostoma caninum



Toxascaris leonina

Toxocara canis

Toxocara cati

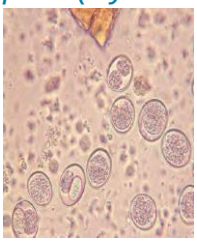
Capillaria

Paragonimus kellicotti

Trichuris vulpis

Giardia spp.

Isospora (Cystoisospora) spp.



Toxoplasma gondii

Cryptosporidium spp.

Prevention and control of Pet diseases

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Prevention and control of disease consist of measures taken for disease prevention, as opposed to disease treatment. Just as health encompasses a variety of physical and mental states, so do disease and disability, which are affected by environmental factors, genetic predisposition, disease agents, and lifestyle choices. Health, disease, and disability are dynamic processes which begin before pets realize they are affected. Disease prevention relies on anticipatory actions that can be categorized as primary, secondary, and tertiary prevention.There are many methods for prevention of disease. It is recommended that pets aim to visit their veterinarian for regular check-ups, even if they feel healthy, to perform disease screening, identify risk factors for disease, discuss tips for a healthy and balanced nutrition, stay up to date with immunizations and boosters, and deworming.

Level	Definition
Primary prevention	Methods to avoid occurrence of disease either through eliminating disease agents or increasing resistance to disease. Examples include immunization against disease, maintaining a healthy diet and exercise regimen.
Secondary prevention	Methods to detect and address an existing disease prior to the appearance of symptoms. Examples include cancer screenings.
Tertiary prevention	Methods to reduce negative impact of symptomatic disease, such as disability or death, through rehabilitation and treatment. Examples include surgical procedures that halt the spread or progression of disease.
Quaternary prevention	Methods to mitigate or avoid results of unnecessary or excessive interventions in the health system.

Vaccination scheduled:

Cat Vaccination				
Disease	**Vaccine**	**Age**	**Dose**	**Immunity**
Rabies	Antirabies Vaccine	12-14 weeks of age	1 ml I/M	1 Year
Feline Panleucopenia		9-10 weeks of age. Booster - 12-15 weeks	1 ml S/C	

Canine Vaccination				
Disease	**Vaccine**	**Age**	**Dose**	**Immunity**
Rabies	Antirabies Vaccine	12- 14 weeks of age as preventive **Post Bite:**	1 ml I/M 1 ml I/M on 0,3,7,14,28,90 days	1 year
Leptospirosis	Canine Leptospirosis	7-9 week & all ages	1 ml S/C, I/M Booster:12-14 wk	1 year
Corona Virus	Canine Corona Vaccine	6-8 weeks	1 ml S/C, I/M Booster:12-14 wk	1 year
Canine Distemper	Canine Distemper Vaccine	6-8 weeks	1 ml S/C, I/M Booster:12-14 wk	1 year
Infectious Canine Hepatitis	Infectious Canine Hepatitis Vaccine	6-8 weeks	1 ml S/C, I/M Booster:12-14 wk	1 year
Parvo Virus Infection	Anti Parvo Virus Vaccine	6-8 weeks & all ages	1 ml S/C Booster: After 4 Wk	1 year

Deworming Schedule for dog and cat:

	Deworming schedule	**Commonly Used Drugs**
Pup	Pup initiate treatment 6-8 weeks of age and then monthly until 6 months of age.	Albendazole: 25-50 mg/kg for 3-5 days (Oral). Levamisole: 5-8 mg/kg (Oral). Morantel: 7.5 mg/kg (Oral). Piperazine: 200-300 mg/kg (Oral).
Kitten	Kitten initiate treatment at 3 weeks; Repeat at 5, 7 & 9 weeks of age, and then monthly until 6 months of age	
Dam	Treat at the same time as Kittens/Puppies	
Adult	Have a fecal test performed 2-4 times per year & treat appropriately.	
Newly Acquired Animals	Deworm immediately, after 2 weeks and then follow above deworming schedule.	

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REPRODUCTIVE SURGERY IN PET ANIMALS

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SURGICAL AFFECTION OF FEMALEREPRODUCTIVE SYSTEM

1) Uterus - Ovariohysterectomy (OHE), Pyometra, Caesarean section, metritis, uterine torsion, uterineprolapse, uterine rupture, uterine neoplasia.
2) Vagina - Vaginal hyperplasia, vaginal prolapse, vaginal trauma.
3) Vulva - Vulvar hypertrophy, vulvar trauma, episiotomy.

Preoperative concerns

1) Perform abdominal palpation to detect uterine enlargement, a mass or pain.
2) Evaluate abnormal vulvar skin folds, conformation, discharge or enlargement.
3) Use an otoscope/vaginal speculum to view vestibule and caudal vagina and endoscope for cranial vagina and cervix.
4) Radiography and ultrasonography.

Anaesthetic considerations

1) Premedicate with atropine / glycopyrolate to help prevent bradycardia induced by visceral manipulation.
2) Use opiods for preoperative and postoperative analgesia.
3) Use fluid administration to compensate for evaporation during abdominal surgery.
4) Maintain body temperature.

OVARIOHYSTERECTOMY (OHE)

- Removal of ovaries and uterus.

Indication

Elective sterilization. Usual treatment for many of uterine diseases like pyometra, vaginal prolapse/hyperplasia, uterine prolapse and uterine rupture, vaginal yperplasia. Animals with diabetes or epilepsy may be sprayed to prevent hormonal changes that can interfere with medication. Ovariohysterectomy before the first estrous cycle decreases the incidence of mammary gland tumour to less than 0.5% compared with intact bitches.

Procedure

A midline abdominal incision is made extending from umbilicus approximately 5 cm caudally in dog. In cat, the incision is started approximately 1 cm caudal to umbilicus and extended caudally for 3 cm. The tissue just underneath the skin is called the subcutaneous layer. It consists mostly of fat and small blood vessels, and is the next layer we cut into after the skin. If these small blood vessels don't clot within a few minutes they are clamped with instruments. The uterine horn is traced into the body cavity until the ovary is found. It has to be gently teased from its location near the kidneys in order to be able to pull it out through the abdominal incision. In older dogs this part of the procedure is much more difficult. The ovary (arrow) is usually covered with fat. The blood supply to the ovary is extensive so a special technique is utilized to prevent hemorrhage. This technique involves the use of 3 clamps. The tissue is cut with a scissors between the second and third clamp. Two sutures are securely placed under the first two clamps. This whole process is repeated for the other ovary that female dogs have in their abdomen. Both ovaries with their attached clamps have been removed from the abdominal cavity. Two clamps are placed at the cervix and the

remaining body of the uterus with its two attached ovaries is cut away. The uterus is sutured in the same manner at the ovaries, with two secure stitches placed under the clamps the linea alba, subcutaneous tissue and skin are sutured separately either by simple interupted suture pattern or simple continuous suture pattern

Complications of ovariohysterectomy

Haemorrhage, Recurrent estrus – due to residual ovarian tissue. Uterine stump pyometra can develop if uterine horn or body present. Fistulous tracts can develop from tissue reaction to ligature material. Hydronephrosis due to accidental ligation of ureter. Urinary incontinence following ovarioh-ysterectomy can be caused by adhesions or granulomas of uterine stump that interfere with urinary bladder spincture function.

PYOMETRA

Literally means pus in the uterus. Describes the clinical condition of a pus filled uterus, ovarian changes and extra genital lesions occurring secondary to the uterine changes. It occurs in the mature bitch or queen and unrelated to pregnancy. Pyometra is a disease of diestral phase of the estrous cycle, when corpus leuteum is actively secreting progesterone.

Types/ Classification

Pathological changes during pyometra are collectively called cystic hyperplasia pyometra complex. It can be divided into 4 types.

Type I - Cystic endometrial hyperplasia in middle aged dogs and not related to estrous cycle. Thickened endometrium lined by numerous thin walled transluscent cysts.

Type II - Diffuse plasma cell infiltration of endometrium in addition to cystic endometrial hyperplasia. Occurs only in diestrus.

Type III - Cystic endometrial hyperplasia is accompanied by acute inflammatory reaction of endometrium.

Type IV - Chronic endometritis with cervix open or closed. Myometrial hypertrophy and fibrosis.Endometrium atrophied and infiltrated with lymphocytes and plasma cells.

Etiology

Infection: E. coli is the most common organism identified in canine and feline pyometra. In progesterone primed uterus, an inhibition of the leukocyte response to infection may predispose the uterus to infection. If cervix is open, vaginal discharge occurs. If cervix is closed, discharge is prevented and more serious disease results. Septicemia and endotoxemia can develop if pyometra is untreated.

Diagnosis:

A) **Clinical presentation** –

1) Older (7 to 8 years) intact bitches and queens are typically affected.
2) Younger animals given exogenous estrogen or progestins may be affected.
3) The animal may present because of a purulent, sometimes bloody, vaginal discharge. Others have obvious abdominal distension, fever, partial to complete anorexia, lethargy, polyuria, polydipsia, vomiting, and diarrhoea and weight loss.

B) Physical Examination –

Purulent blood tinged vaginal discharge if cervix is open. Dehydration is frequently present. Fever is in-frequently present.

C) Radiography / Ultrasonography

Uterus as a fluid dense tubular structure in the caudoventral abdomen. Rule out pregnancy – Radiographically foetal calcification after 45 days. Ultrasonographically foetal structures after 21 days

D) Laboratory findings

Neutrophilia with a left shift, monocytosis, WBC toxicity, clothing abnormalities, hyperproteinemia, hyperglobulinemia and azotemia vaginal cytology to confirm a septic exudate with open pyometra

Treatment

Most common treatment is ovariohysterectomy which is already discussed earlier in this chapter.

Post Operative Care

Provide analgesics, fluid therapy and antibiotics. Diuretics given if urine production is reduced.

Complications:

Septecemia, endotoxemia, peritonitis and cervical or stump pyometra may occur.

CAESAREAN SECTION (HYSTEROTOMY)

Major operation chiefly performed when the pelvic passage is for some reason unable to accommodate and discharge the foetus.

Indications:

- For prolonged gestation (**not likely to occur after 70 days gestation)**
- For incomplete primary uterine inertia.(**dachshunds and scotties)**
- For secondary uterine inertia when several foetus remain.(**St.Bernard**)
- Abnormalities of maternal pelvis or soft tissue.
- For relative and absolute fetal **oversize(Brachycephalic foetus tend to have large head or shoulders)**
- For foetal monstrosities.
- For foetal malpositioning.
- For foetal death and putrefaction.(**Grey hounds**)

Surgical Procedure

Two approaches – Ventral midline abdominal and flank approach.

Ventral abdominal → provide better exposure of uterus, require less hemostasis, result in lessscarring

Flank approach → doesn't interfere lactation, reduce chance of wound dehiscence.

Ventral abdominal

Animal placed in dorsal recumbancy. Surgical site prepared and ventral midline abdominal incision is made. Uterus extracted and moistened. A longitudinal incision is made on dorsal midline of uterine body gently squeezing of uterine horn to extract foetus. Ensure all foetuses are removed before closing uterine incision. Uterine incision closed with inverted continuous suture like Lembert or Cushing pattern. Uterus surface cleaned with warm saline. Laparotomy incision routinely closed with simple interrupted sutures followed by subcutaneous and skin sutures. In case of dystocia where uterus is devitalised, hysterectomy may be necessary. In that case, uterine body incision closed prior to removal of uterus to avoid spillage of uterine fluid

Complications of caesarean

Peritoneal infection, Excessive haemorrhage, Wound dehiscence, uterine scar which prevent future placentation or cause abnormal foetal development.

METRITIS

Inflammation of uterus. Occurs most commonly in the immediate postpartum period and is usually associated with dystocia, obstetrical manipulations or retained placenta. Clinical signs include mucopurulent vaginal discharge, fever, aneurexia and vomitting. Enlarged uterus is palpable. **Ovariohysterectomy** is recommended if the owner do not want to breed the animal again or if the animal has severe systemic sign.

UTERINE TORSION

Uncommon in dogs and cats. One or both uterine horns can twist along long axis or around the opposite horn or entire uterine body can rotate. Torsion of gravid and non-gravid uteri has been reported.

Causes: Jumping or running late in pregnancy. Active foetal movement. Premature uterine contraction. Partial abortion. Variation in length and mobility of proper ovarian ligament. Abnormalities of uterus.

Signs: Abdominal pain and distension, radiographic examination discloses a large air or fluid filled tubular structure.

Treatment: Ovariohysterectomy. Caesarean performed first if there are viable foetuses.

UTERINE PROLAPSE

Is an eversion and protrusion of a portion of the uterus through the cervix into the vagina during or near parturition.

Causes: Over relaxation and stretching of pelvic musculature. Uterine atony due to metritis. Incomplete separation of placental membranes. Severe tenesmus.

Diagnosis:

One or two tubular masses protruding from the vulva. Everted tissue is discoloured from venous congestion, trauma and debris. Typical history of excessive straining during parturition.

Treatment:

Manual reduction attempted if animal is in good physical condition and uterus is healthy. Extensive devitalisation of the uterus necessitates ovariohysterectomy after reduction of the prolapse. If reduction is impossible, uterus amputated and stump is reduced. Ovaries removed through abdominal incision.

UTERINE RUPTURE

Grand uterus may rupture spontaneously during parturition or as a result of severe trauma. Foetuses expelled into the peritoneal cavity may die immediately and may resorb or cause peritonitis. Sometimes, foetus may live to term and extrauterine location not recognized until whelping when foetus is not delivered. Acute rupture is usually treated by ovariohysterectomy. A unilateral ovariohysterectomy may preserve breeding ability but repaired area may be more likely to rupture during subsequent pregnancies.

UTERINE NEOPLASIA

- Tumours that may occur in uterus are leiomyoma, leiomyosarcoma, adenocarcinoma, lipoma, fibroma, adenoma and fibrosarcoma.
- Leiomyomas are most common uterine tumours which are benign and protrude into uterine lumen and cause wall to bulge externally.
- German shepherd dogs have a syndrome characterized by multiple uterine leiomyomas.
- Leiomyosarcomas are most common malignant tumors or bitches.
- Adenocarcinomas are most common malignant tumors of queens.

VAGINA

VAGINAL HYPERPLASIA

Normal estrogenic stimulation causes vaginal mucosa to become edematous. The edematous mucosa may prolapse with hyperestrogenism or weakness of vaginal connective tissue during pro-estrus and estrus. Do not breed the animal because vaginal prolapse appears to be familial.

Diagnosis: Young large breed dogs are commonly affected. Most commonly, a mass will be seen protruding between the vulvar lips or the perineum will bulge. Acute prolapse characterized by glistening, edematous, pale pink surface and chronic appears leathery, corrugated and sometimes ulcerated.

Treatment:

Treat Transmissible Venereal Tumours with Vincristine (0.025 mg/kg upto 1 mg. or 0.5 mg/m^2 weekly for 3-6 weeks). In sometimes TVT regresses spontaneously.

Surgical treatment - Perform ovariohysterectomy to prevent recurrence and injury to the everted mucosa. Perform OHE and biopsy the mass to rule out neoplasia. Perform an

episiotomy to allow biopsy. Replace protruding mass into the vagina or vestibule. Lavage, lubricate and reduce the prolapsed tissue by digital manipulation. Maintain reduction by placing two or three stay sutures.

VAGINAL PROLAPSE

Doughnut shaped eversion of the complete vaginal circumference protrudes through the labia. Brachycephalic breeds such as boxers and Boston terriers appear predisposed to vaginal prolapse.

Causes: Constipation, forced separation during coitus and size discrepancy between breeding partners may play a role in vaginal prolapse. Also when estrogen production predominates (estrus) and with pathological hyperestrogenism (eg: - cystic ovaries)

Surgical Treatment: Under general anaesthesia. Everted tissue cleaned with saline or diluted antibiotic. A lubricated plastic syringe case can be used to push the everted tissue back into place. After reposition recurrent prolapse can be minimized in some cases by suturing uterine body on broad ligament to the abdominal wall. In long standing vaginal prolapse surgical resection of the devitalised tissue is necessary to prevent further sepsis and self mutilation and to restore vaginal lumen.

EPISIOPLASTY

Is defined as plastic repair of the vulva. Used to treat a recessed or juvenile vulva in bitches. Deep perivulvar tissue folds may cause the perineal skin to overlap the vulva, obscuring it from view.Retension of fluid within the vulva and perivulvar folds combined with frictional irritation predispose the animal to bacterial growth, infection and ulceration.

SURGICAL AFFECTIONS OF MALE GENITAL SYSTEM

DISORDERS/AFFECTIONS OF PENIS

HYPOSPADIAS: Developemental anomaly. External urethral orifice occur anywhere on the ventral aspect of penis from the normal opening to the perineal region; glandular, penile, scrotal region. Most frequently in Boston terriers

PENILE WOUNDS: It may occur during mating, dogfights, and fence jumping or from automobile accident or gunshot. Severity may involve urethra and sometime fracture of penis may occur. Wounds are treated with topial antimicrobial agent and allow it to heal by second intention. In severe cases haemorrage is controlled, debridement and suturing is carried out for rapid healing.

FRACTURE OF OS PENIS: Clinical signs depends on the degree of soft tissue damage and fracture displacement. Crepitus may be present, dysuria and haematuria may be present. Usually conservative treatment is carried out.

BALANOPOSTHITIS: Infection of penis and prepuce both. Preputial discharge is copius yellow and blood tinged. Affected dog frequently licks at the penis. Enlarged lymphoid nodules near the fornix. Adhesions May follow penile injury, accompany phimosis, preputial foreign bodies, or neoplasia Examination should be carried out under general anaesthesia Treatment is by flushing with dilute povidone iodine or chlorhexidine solution followed by antimicrobial ointment.

STRANGULATION OF PENIS

It may occur by malicious application of rubber band or by constriction of a ring of preputial hairs. Dog exhibit pain and frequently lick the prepuce, dysuria with penile mucosa swollen with necrotic circle. Treatment in minor cases is topical application of antimicrobial agent. Partial amputation is indicated when the distal portion is gangrenous or when the urethra is severely damaged.

PENILE TUMORS

In male cats it is rare. In dogs include mainly TVT, papilloma, and squamous cell carcinoma. Clinical signs include licking and serosanguineous preputial discharge. It responds to radiation therapy, chemotherapy and surgery. Tumors involving distal part of

penis may require partial penile amputation; more extensive and proximal tumors may require ablation of the external genitalia.

PERSISTENT PENILE FRENULUM

In this the epithelial surface of the penis and prepuce are fused together. It occurs in cocker spaniels, miniature poodles, Peckingese, and mixed breeds. Pain during sexual excitement and ventral deviation of penis may be evident. Surgical severing of minimally vascular connective tissue is carried out with good prognosis

PARAPHIMOSIS

In this the penis protrudes from the preputial sheath but cannot replaced to its normal position. It has both congenital and acquired causes. Congenital are a narrowed preputial orifice and abnormally shortened prepuce. Acquired are trauma, infection and priapism. In treatment lubricants, hyperosmolar solutions and local heat or cold can be used to reduce the size of penis and to premitt replacement. Long standing cases with necrosis can be undergone partial penile amputation.

PRIAPISM

Persistent erection not associated with sexual excitement. It is usually secondary to spinal cord injury accompany with constipation and genitourinary infection.

URETHRAL PROLAPSE

Protrusion of the urethral mucosa from the tip of the penis. Most often seen in young male brachiocephalic dogs and Yorkshire terrier. It is due to the sexual excitement or urethral infection or both for correction, prolapsed urethral mucosa is incised at the point of its reflection from the tip of penis and is continued through 180^0C of circumference of the urethra.

DISORDERS/ AFFECTIONS OF PREPUCE

PHIMOSIS

It is the inability to protrude penis beyond the preputial orifice. In congenital phimosis there is a distended prepuce and inability to urinate normally. Acquired phimosis occur due to preputial trauma and neoplasia. In this the surgical correction is made under general anaesthesia by taking triangular incision over the preputial orifice and the skin, s/c tissue and preputial mucosa are excised over the dorsal surface.

PREPUTIAL FOREIGN BODIES

Foreign bodies such as grass awns, plant seeds, pieces of straw, and urinary caliculi can lodge in preputial cavity. Blood tinged preputial discharge is seen and animal frequently licks the prepuce. The animal is usually painfull, listless and mildly pyretic and walks stiffly. Surgically foreign bodies are removed and preputial cavity is irrigated with antiseptic solution and systemic antimicrobials may be indicated.

DISORDERS/ AFFECTION OF SCROTUM

SCROTAL INJURY

Infection of the Scrotal Skin: Scrotal dermatitis caused by *brucella canis* is evident. Treatment is to avoid self mutilation and local application of emolients and antimicrobials.

Varicosity of Scrotal Blood Vessels: Treatment involves either stimulation of thrombosis of the varicose vessels with styptics or surgical removal of the involved scrotal skin and blood vessels.

Chronic Hyperplasia of the Srotum: It occurs due to chronic irritation. Scrotum is thickened, wrinkled and heavily pigmented on ventral side. Simple cases are not treated but extensive hyperplasia with infection may require orchiectomy and scrotal ablation.

Scrotal neoplasm: Mastocyomas are mainly reported.

DISORDERS/AFFECTION OF TESTES

Anorchism and Monorchism: Congenital absence of both the testis is anarchism. In monorchism, left testis is usually absent.

Testicular hypoplascia: It may be unilateral or bilateral. It is usually incapable of spermatogenesis but leydig cell function may be present. Treatment is orchiectomy.

CRYPTORCHIDISM

It is the failure of one or both the testis to descend into the scrotum at the usual time. Normally it occur at the time of birth or at any time up to six month of age. It can be unilateral, bilateral and the position of the ectopic testis may be prescrotal, inguinal, or abdominal

Treatment

The surgical technique for removal of cryptorchid testes varies with the location. In extra-abdominal ectopic testes removal, skin incision is directly made over the testes.

ORCHITIS

Infection of testes in dog which is frequently accompanied by epididymitis. In acute orchitis clinical signs include testicular pain, tenseness, and scrotal edema. Acute orchitis is usually suppurative, with the formation of one or more abscess in the testes and epididymis with systemic signs are present.

Treatment

Severely traumatised or abscesssed testes are surgically removed. Chronically affected testis are removed to prevent continuing episodes of acute inflammation.

TESTICULAR TUMORS

Commonly in older dogs. Clinical sign are increased firmness and nodular enlargement of the scrotal or ectopic testis. The three common neoplasms seen are seminoma, interstitial cell tumor and sertoli cell tumor.

ORCHIECTOMY

CANINE ORCHIECTOMY

It is carried out by two method, close and open. Both the methods midline prescrotal incision is given, and the testes is pushed cranially to the skin incision, and the s/c tissue and spermatic fascia are incised over the testis to expose the parietal vaginal tunic. The testis is exteriorized and freed from its scrotal attachment by incising the spermatic fascia and scrotal ligament close to the testis. In the **open method**, an incision is made through the parietal vaginal tunic covering the spermatic cord where it is doubly ligated by transfixation. The parietal vaginal tunic and cremaster muscle are ligated in an encircling or transfixation ligature distal to the spermatic cord ligation. Alternatively, the parietal vaginal tunic is incised before exteriorization of the testis. The tunics are seperated from the remainder of the spermatic cord and these two structure are doubly ligated seperately. In **closed method,** the intact spermatic cord and vaginal tunics are doubly ligated by a transfixation ligation. The spermatic cord is transected distal to the ligature and return to the inguinal region. If necessary than the s/c tissues are closed with 3-0 or 4-0 absorbable suture material. The skin is closed by the intradermal pattern of 4-0 absorbable suture pattern.

ORCHIECTOMY IN THE CAT.

It is performed by making a separate longitudinal scrotal incision over each testis. The testis is pushed caudally to the skin incision, and the s/c tissue and parietal vaginal tunic is grasped with haemostats, seperated from testis and excised. The components of the spermatic cord are occluded by ligation, overhand or figure of eight knot in the spermatic cord, or square knot of the ductus deferens to the testicular vessels. The spermatic is transected distal to these and the scrotal is left unsutured.

SCROTAL ABLATION.

It is recommended when trauma to the scrotum is severe and suggests ischemia. It also preferred in old dogs with pendulous scrotum for avoidance of postoperative problems such as scrotal irritation and edema. Surgically, curvilinear incision is made on both the sides of the scrotum at its base and the incision are curved toward the scrotum to provide adequate

skin for closure. Trauma to the urethra or penis is avoided during dissection. After transection of the scrotal septum, the skin is closed in usual manner.

DISORDERS/AFFECTIONS OF EPYDIDYMIS

EPIDIDYMITIS

It results from ascending infection of the genital tract, canine distemper virus infection, or haematogenous infection. Diagnosis of the causative organism often requires microbiological testing of fluid within the vaginal tunics, semen, urine and possibly blood. Treatment of acute cases is orchiectomy. Antibacterial drugs are used to control infection.

EPIDIDYMAL TUMORS

They are seen rare in small animals. Fibromas have been observed. Treatment is orchiectomy.

DISORDERS/AFFECTION OF PROSTATE

PROSTATECTOMY

It is indicated in benign prostatic hyperplasia, suppurative prostatitis, prostatic cysts, prostatic neoplasia and prostatic trauma. There are two types of prostectomy. Excisional prostectomy removes the entire prostate. Partial or subtotal can eliminate major portion of diseased tissue.

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Judicious application of antimicrobial agents in Veterinary Practices

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Introduction

In the past years, antibiotics have been critical in the fight against many diseases and infections. Their discovery was one of the leading causes for rise of average life expectancy in the current century and their significance to health would be incredible to exaggerate. Upon the introduction of penicillin into general clinical practice in 1944, formerly deadly illnesses in animals and human became instantly curable. A return to the "pre-antibiotic era" would render many routine infections untreatable and would seriously affect current practice in veterinary and human medicine through major increases in morbidity and mortality. Judicious use of antimicrobial drugs is an essential to prevent the bacterial resistance during the therapeutic approach in uterine infection. Resistance development has resulted in perpetual research and development in the search of new antibiotics in order to maintain a pool of effective drugs at all times. While the development of resistant strains is unavoidable. Resistance to antimicrobials is a natural biological phenomenon. The introduction of every antimicrobial agent into clinical practice has been followed by the detection in the laboratory of strains of microorganisms that are resistant. Such resistance may either be a characteristic associated with the entire species or emerge in strains of a normally susceptible species through mutation or gene transfer.

Mechanism of resistance

1. *Reduced drug accumulation:* by decreasing drug permeability and/or increasing active efflux (pumping out) of the drugs across the cell surface
2. *Drug inactivation or modification:* e.g., enzymatic deactivation of *Penicillin* in some penicillin-resistant bacteria through the production of β-lactamases.
3. *Alteration of target site:* e.g., alteration of PBP — the binding target site of penicillins — in MRSA and other penicillin-resistant bacteria.
4. *Alteration of metabolic pathway:* e.g., some sulfonamide-resistant bacteria do not require para-aminobenzoic acid (PABA), an important precursor for the synthesis of folic acid and nucleic acids in bacteria inhibited by sulfonamides. Instead, like mammalian cells, they turn to utilizing preformed folic acid.

Types of resistance

1. **Natural Resistance:** Inherently or genetically resistant due to lake of penetration of drug into bacterial cell, absence of metabolic pathway or target site or rapid inactivation of drug in bacterial cell.
2. **Acquired resistance:** Resistance against drug to which bacteria was previously sensitive. It is due to inappropriate use of antimicrobials. It is done by mutation or gene transfer.

Table 1: Reports of resistance developed in bacteria from India

Bacteria	Resistance to	Reference
Streptococcus pneumoniae	Penicillin, cotrimoxazole, tetracycline, erythromycin, ciprofloxacin	Goyal *et al.*, 2007 Chawala *et al.*, 2010

S. pyogenes	Penicillin, erythromycin, trimethoprim	Capoor *et al.,* 2009 Bergmann *et al.,* 2012
Staphylococcus aureus	Clindamycin, Vancomycin	Gupta *et al.,* 2009 Thati *et al.,* 2011
E. Coli	Ampicillin, tetracycline, co-trimazole, trimethoprime, carbenicillin	Sukumaran *et al.,* 2012
Salmonella spp.	Nalidixic acid, ciprofloxacin, ampicillin, chloramphenicol, ampicillin and trimethoprim	Rowe *et al.,* 1997 Nagshetty *et al.,* 2010
K. pneumoniae	Ceftizoxime , cefotaxime, carbenicillin	Sikarwar & Batra, 2011 Nagaraj *et al.,* 2012
Shigella spp.	Newer gen. fluoroquinolones, 3rd gen. Cephalosporins	Bhattacharya *et al.,* 2012
Pseudomonas spp.	Ciprofloxacin, ceftazidime, cefepime, gentamicin, amikacin	Chaudhary *et al.,* 2013

Consequences of resistance

The emergence of antimicrobial resistance has an impact on the cost of animal and human health care worldwide. *Resistant strains of bacteria are found around the world. Organisms that are resistant to one drug are more likely to become resistant to others.* Resistant pathogens are expensive to control and extremely difficult to eradicate. Ineffective therapy can seriously affect the progress and outcome of disease Resistant animal pathogens in food products may cause infections in humans that are difficult to treat.

Practices responsible for failure of antimicrobials in disease state:

- Improper diagnosis (Viral not Bacterial infection)
- Improper selection of drug (Causative organisms are not sensitive to drug).
- The microorganisms have developed resistance to drug.
- Mixed infection & narrow spectrum drug
- Penetration of drug into site infection is not proper due to pus, debris, exudates etc.
- The host defense mechanism is impaired
- Improper route of administration with inadequate duration of treatment
- Interaction of drug with other administered drugs.
- Late administration of antimicrobial drug
- Use of expired drug
- The owner or attendant of animal does not comply with therapeutic regimen
- Improper nursing and feeding

How can we select antibiotics?

- Requires clinical judgment and detailed knowledge of pharmacological and microbiological factors.
- Antibacterials: empirical therapy, definitive therapy, and prophylactic therapy.
- Empirical therapy: infecting organism has not been identified - Combination therapy/broad-spectrum agent
- Infecting microorganism is identified : Narrow-spectrum AB
- Select an antibacterial based on indication
- The diagnosis may be masked if therapy is started before cultures are obtained.
- Antibacterials may be used immediately if disease is severe

- Initiation of optimal empiric antibacterial therapy: knowledge of most likely infecting organisms and their antibacterial susceptibilities.
- Simple and rapid laboratory tests may permit more rational selection of initial antibacterial therapy.
- Blood should be taken prior to the institution of drug therapy.
- *For definitive therapy, Use* specific & narrow-spectrum antibacterial once an organism has been identified & its susceptibility is known.

Successful Antimicrobial Therapy depends on:

- For definitive therapy, recommend a narrow-spectrum drug
- Keep the broad spectrum drug reserve for life threatening infection
- Prefer bactericidal over bacteriostatic drug with less toxicity
- Prefer drug requires administration at long interval
- For less severe infections prefer an oral administration in small animals
- For severe infection→ parenteral administration
- Always use antimicrobial agent in proper dose
- Proper duration of time
- Do not combine antimicrobials without valid cause
- Do not use antimicrobial indiscriminately
- Avoid overuse of newer agent if older is effective
- Use drug manufactured by reliable pharmaceutical firm.
- Do not use antimicrobials to treat slight, self-limiting or unbeatable infections.

How can we fight back?

- Maintain good hygiene and infection control measures – particularly hand washing.
- Strict infection control measures should be monitor in hospitals
- Don't use antibacterial in minor or self limiting viral infections
- Farmers should not use antibacterials of previous prescription
- Educate farmers: help them to understand about cost of unnecessary use of antibacterials
- Communicate with farmers about progression of disease after initiation of therapy
- Use laboratory tests to support your diagnosis & select the right antibacterial.
- Record of vaccinations must be generated.
- Develop and implement guidelines, protocols and drug utilization reviews to ensure that use of antibacterial drug is optimized
- Ensure surveillance for changes in the occurrence and pattern of antimicrobial resistance in different bacteria.
- Emphasise good animal husbandry practices (adequate and clean quarters)
- Work with governments to move away from using antibacterials as growth promoters.
- Collaborate in monitoring of antibacterial use and resistant pattern with institutes
- Educate the public and health professionals about the antibacterial resistance
- Coordinate the development and implementation of regional programs to optimize antibacterial use and to prevent the spread of resistant organisms.
- Develop the rapid affordable systems for diagnosis and susceptibility testing.
- Ensure that antibacterials remain available through prescription only, rather than as over-the-counter medications.

Table 1: Type, mode and spectrum of activity of different antimicrobial agents

Group of antimicrobials	Type of action	Mode of action	Spectrum of activity
Penicillins	Bactericidal	Inhibition of cell wall synthesis	Narrow

Cephalosporins	Bactericidal	Inhibition of cell wall synthesis	Broad
Carbapenems	Bactericidal	Inhibition of cell wall synthesis	Broad
Polypeptide Antibacterials	Bactericidal	Inhibition of cell wall synthesis	Narrow
Quinolones	Bactericidal	Inhibits DNA synthesis	Broad
Metronidazole	Bactericidal	Inhibits DNA synthesis	Narrow
Rifamycins	Bactericidal	Inhibitions of RNA transcription	Narrow
Aminoglycosides	Bacteriostatic/ Bactericidal	Inhibition of protein synthesis	Narrow
Lincosamides	Bacteriostatic	Inhibition of protein synthesis	Narrow
Macrolides	Bacteriostatic	Inhibition of protein synthesis	Narrow
Tetracyclines	Bacteriostatic	Inhibition of protein synthesis	Broad
Chloramphenicol	Bacteriostatic	Inhibition of protein synthesis	Broad
Sulfonamides	Bacteriostatic	Competitive inhibition	Broad

Table 2: Spectrum of activity of commonly used antimicrobials

G+ve activity	G-ve activity	Anaerobic activity
Penicillins	Fluoroquinolones	Penicillins
Macrolides	Aminoglycosides	Metronidazole
Cephalosporins	Cephalosporins	Cephalosporins
Sulpha + TMP	Sulpha + TMP	Sulpha + TMP
OTC	OTC	OTC
Imipenem	Imipenem	Imipenem
Chloramphenicol	Chloramphenicol	Chloramphenicol
Rifampin		Clindamycin

Table 2: Last-resort antibacterials

Drug	Why last resort?
1) Meropenem and other carbapenem	Potency & lack of resistance
2) Vancomycin	Anti-MRSA
3) Co-trimoxazole	Powerfull
4) Piperacillin/Tazobactam	Broad coverage
5) Levofloxacin	Broad spectrum& PO
6) Linezolid	Anti-MRSA
7) Cefepime	Broad spectrum
8) Polymyxin B (Colistin)	Potent
9) Tigecycline	Anti-MRSA
10) Aztreonam	Anti-pseudomonal

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Importance of nutrition in Pet and laboratory animals

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A laboratory animal's nutritional status influences its ability to reach its genetic potential for growth, reproduction, and longevity and to respond to pathogens and other environmental stresses. A nutritionally balanced diet is important both for the welfare of laboratory animals and to ensure that experimental results are not biased by unintended nutritional factors. Laboratory animals require about 50 nutrients in appropriate dietary concentrations.

Factors Affecting Nutrient Requirements

1. Genetics

Genetic differences among species, breeds, strains, stocks, sexes, and individuals may affect nutrient requirements. For example, the lack of L-gulonolactone oxidase (a key enzyme required for the synthesis of ascorbic acid) in some species is apparently the consequence of genetic mutation (Chatterjee, 1978). L-gulonolactone oxidase activity differs among rodent species, among rat strains, and between sexes within rat strains (Jenness et al., 1980).

2. Stage of Life

Nutrient requirements change during stages of the life cycle, especially in response to growth, pregnancy, or lactation. Synthesis of tissues or products requires amino acids, fatty acids, minerals, glucose, or other substrates as well as increased amounts of vitamins and associated cofactors.

3. Environmental impacts

Nutrient requirements are usually studied under controlled conditions with minimal diurnal or seasonal variation in temperature, light cycle, or other environmental conditions. Marked modification in these conditions may alter nutrient requirements. For example, exposure to temperatures below the lower threshold of the thermoneutral zone increases energy requirements as animals are obliged to expend energy to maintain a constant body temperature

4. Microbiological Status

Under normal rearing conditions, laboratory animals harbor populations of microorganisms in the digestive tract. These microorganisms generate various organic constituents as products or by-products of metabolism, including various water-soluble vitamins and amino acids. The extent to which these nutrients contribute to the nutrition of the host may be substantial but varies according to species, diet composition, and rearing conditions. In the rat and mouse, most of the microbial activity is in the colon, and many of the microbially produced nutrients are not available to the host unless feces are consumed, as is common for rats and other rodents (Stevens, 1988). Prevention of coprophagy may require an increase in the nutrient concentrations that must be supplied by the diet. The loss of some or all microbial symbionts in animals free of specific pathogens and germ-free animals, respectively, may also alter microbial nutrient synthesis and, thereby, influence dietary requirements.

5. Research Conditions:

Experimental procedures may produce stress or otherwise alter food intake. For example, surgical procedures or test substances in diets may lead to anorexia, necessitating the provision of more palatable diets or diets with elevated nutrient concentrations.

6. Nutrient Interactions:

Alterations in dietary energy density usually cause a change in feed intake. If high-energy diets are used, it may be necessary to increase nutrient concentrations in the diet to compensate for decreased food consumption. Other interactions occur between nutrients, such as competition for absorption sites among certain minerals that share common active transport systems.

7. Formulation of Diet Types

Diet formulation is the process of selecting the kinds and amounts of ingredients (including vitamin and mineral supplements) to be used in the production of a diet containing planned concentrations of nutrients. Choice of ingredients will be influenced by the species to be fed and the experimental or production objectives. Target nutrient concentrations must take into account estimated nutrient requirements, possible nutrient losses during manufacturing and storage (National Research Council, 1973; Harris and Karmas, 1975), bioavailability of nutrients in the ingredients, and potential nutrient interactions. Various types of diets are available for use with laboratory animals. Selection of the most appropriate type will depend on the amount of control required over nutrient composition, the need to add test substances, potential effects of feed microbes, diet acceptance by the animals, and cost.

Feeding of rabbits:

Feeding of baby rabbits starts at at the age of 15- 21 days, while coprophagy starts at 4-6 weeks

Age (Days)	Weight of food	Water consumption
0-15	nil	Twice the amount of Food DM
15-21	0-20	
21-35	10-50	
35-42	40-80	
42-49	70-110	
49-63	100-160	

The DM intake of adult rabbit is about 5.5 % of live weight. They requires total quantity of feeds as 4 weeks- 40 g, 10 weeks- 100 g and Adults- 140-160 g. For adult doe restricted feeding should be done with around 140 g feed. During lactation and pregnancy requirement increases.

Nutrient requirement of rabbits:

Nutrient	Maintenance	Gestation	Growth	Lactation
Digestible Energy (Kcal)	2100	2500	2500	2500
CP (%)	12	15	16	17

DCP (%)	9	11	12	13
TDN (%)	55	58	65	70
Crude Fibre (%)	14	10-12	10-12	10-12
Fat (%)	2	2	2	2

Caecotrophy in rabbits:

- This is the eating of faecal-like pellets produced in the caecum. These caecal pellets are sometimes called soft faeces. To do this, the rabbit sucks in the soft faeces as they emerge from the anus, then swallow without chewing.
- Consumption of the soft faeces starts when the rabbit is about 4 weeks old. Note that rabbit can survive without practicing caecotrophy for many days but death is usual if they are prevented from eating their soft faeces for several months.
- Soft faeces are higher in crude protein and lower in crude fibre than hard faeces. Their higher protein level is due to their content of bacteria.
- Caecotrophy is a very important part of the rabbit's digestive processes. It recycles some unabsorbed nutrients as well as returning protein and vitamin B rich bacteria for enzyme digestion in the small intestine.

Feeds of rabbits:

It includes hay of alfalfa, maize, clover, lucerne , berseem etc., grains of maize, wheat, barley, sorhgum and Soyabean meal, GNC, DORB.

Feeding guinea pigs:

They are strict herbivores and hind gut fermenters – cecum. Different microbes are present in cecum and out of different microbes Lactobacilli sp. dominates the most. Primary fatty acid produced in cecum is propionic acid. Stomach lined entierly with glandular epithelium and they have Semicircular cecum with lateral pouches. Energy requirement is- 136 Kcal/kg Metabolic BW and require vitamin C @200mg per day.They have higher folic acid requirement and sensitive to excess Ca, Vit. A, Vit. D which may leads to metastatic calcification and mineralization of soft tissues

Feed Consumption in guinea pigs-

- Growing guinea pig- 20-30 g/ day
- Adult guinea pig- 30-50g/ day
- Pregnant and lacating- 40-60 g/ day

Nutrient requirements of guinea pigs:

Sr. No.	Proximate principles (% by weight)	Requirement

1.	Moisture	Max 10
2.	CP	Min 22
3.	EE	Min 4
4..	CF	9-14
5.	Total Ash	9
6.	AIA	1.0
7.	Ca	1.2
8.	P	0.6
9.	Vitamin C	200 MG

Feeding of Rat and mice:

Feed requirement of growing Rats is 15 g/ day, while it is 15-20 g/day for adult and pregnant rats and 30- 40 g/ day for lactating rats. Maintenance Energy Requiement of rat and mice is 114 Kcal/ kg metabolic body weight. While, requirement of CP- 24 %, EE- 5 %, CF- 6 %, Total Ash- 9 %, Ca- 0.6 % and P- 0.3 % of diet

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ADVANCES IN THERAPEUTIC MANAGEMENT OF DIABETES MELLITUS IN DOGS

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INTRODUCTION

The endocrine pancreas is composed of the islets of Langerhans, which are dispersed as "small islands" ina "sea" of exocrine-secreting acinar cells. Four distinct cell types have been identified within these islets onthe basis of staining properties and morphology: *alpha cells*, which secrete glucagon; *beta cells*, whichsecrete insulin; *delta cells*, which secrete somatostatin; and *F cells*, which secrete pancreatic polypeptide.Dysfunction involving any of these cell lines ultimately results in either an excess or a deficiency of thatrespectivehormone. In the dog, the most common disorder of the endocrine pancreas is diabetes mellitus, which results from an insulin deficiency caused by destruction of beta cells.

Diabetes mellitus comes from the Greek word "diabainein" meaning "to pass through," and the Latin word "mellitus" meaning "sweetened with honey." Put the two words together and you have "to pass through sweetened with honey."

Diabetes mellitus (DM) is disorder of the endocrine pancreas resulting in metabolic disorders of carbohydrate, fat and protein metabolism characterized by an absolute or relative deficiency of insulin resulting in hyperglycemia.

CLASSIFICATION OF DIABETES MELLITUS:

1) Insulin dependent diabetes (IDD)/ Type I diabetes:

Primary IDD is characterized by a progressive loss of pancreatic beta cells. The etiology of beta cell deficiency/destruction in diabetic dogs is currently unknown but a number of diseases processes are thought to be involved:

- Congenital beta cell hypoplasia/abiotrophy
- Beta cell loss associated with exocrine pancreatic diseases
- Immune-mediated beta cell destruction
- **Idiopathic:** In Type I DM, endogenous insulin is low or absent. So, Diabetic dog should have to dependent on exogenous insulin for life. This most common form of diabetes strikes 1 in 500dogs.

2) Insulin resistance diabetes (IRD)/type II diabetes:

IRD usually results from antagonism of insulin function by other hormones.

- Dioestrous/gestational diabetes
- Secondary to other endocrine disorders
- Hyperadrenocorticism
- Acromegaly
- Iatrogenic
- Synthetic glucocorticoids
- Synthetic progestagens

INCIDENCE:

Diabetes mellitus is one of the most frequent endocrine disease affecting middle-aged and older dogs and the prevalence is increasing. Diabetes mellitus most commonly occurs in middle age to older dogs and cats, but occasionally occurs in young animals. In addition, it occurs more commonly in female dogs. Juvenile onset diabetes is uncommon in dogs.

CLINICAL SIGNS:

Generally there is a gradual onset of the disease over a few weeks, and it may escape unnoticed for a while. The main symptoms are:

- Excessive water consumption–Polydipsia
- Frequent and/or excessive urination–Polyuria–possible house "accidents"
- Greater than average appetite–Polyphagia–with either weight loss or maintenance of current weight
- Cloudy eyes–Cataracts

DIAGNOSIS

- History and clinical signs
- Clinically, DM is diagnosed on the basis of persistent glycosuria and persistent hyperglycemia.
- Testing the fasting blood glucose concentration (BGC) by glucometer.
- Urine examination for ketone bodies and glucose.

THERAPEUTIC MANAGEMENT

As per the FDA, U 40 pork lente (porcine zinc insulin suspension) has been the first choice recommendation for dogs. Since dogs are insulin-dependent, oral drugs are not effective for them.

Types of Insulins:

1. **Long-acting Insulins**
   Ultralente insulins • 100 percent crystalline insulin.
   PZI insulins• Contain protamine and zinc.
2. **Intermediate-acting Insulins**
   Lente insulins • Mixtures of 30 percent amorphous and 70 percent crystalline insulin in aqueous suspension. e. g. Vetsulin
   NPH insulins• Contain protamine. e.g. Wosulin
3. **Short-acting Insulins**
   • Soluble insulin and semilente insulin
   • Intravenous administration possible. e.g. Wosulin-S

When porcine zinc insulin is not available, U-100 human recombinant Neutral Protamine Hagedorn (NPH) insulin is a good initial alternative, although its duration of action is often <12 hours in many dogs. At third option, human PZI is likely to be a better choice for dogs due to its slow release from subcutaneous tissues.

Determining Insulin Dose and Frequency of Administration

In dogs with uncomplicated diabetes it is recommended to initiate treatment with an intermediate duration of action insulin product such as recombinant human NPH or porcine lente (Vetsulin) at a dose of **0.25 U/kg** every 12 hours subcutaneously starting at time in the morning that will fit the client's schedule. Blood glucose is measured two or three times at 3-hour intervals during the first day to ensure that the dose is not so high as to create hypoglycemia. If the blood glucose drops below 100 mg/dl at any time during the first day, the dose should be reduced by 25%. Because it can take several days for the dog to adjust to any change of insulin dose or product type, it is sent home to receive insulin injections for 5 to 7 days before the first recheck to monitor the response to the insulin. In an attempt to prevent tissue irritation from injecting insulin at the same site while maintaining consistent absorption of insulin from each injection, it is recommended to alternate injections at approximately 1 to 2 inches on either side of the midline. The suggested level is over the thorax from just caudal to the scapulae to the last rib, alternating from.

Oral medications:

Oral hypoglycemic drugs are primarily used for the treatment of Type 2 DM, a form of diabetes that is extremely rare in dogs. Oral hypoglycemic drugs work by stimulating pancreatic insulin secretion, enhancing tissue sensitivity to insulin, or slowing postprandial intestinal glucose absorption. Oral sulfonylurea drugs (e.g. Glipizide, glyburide) directly stimulate insulin secretion by beta cells and are the most commonly used oral hypoglycemic drugs for the treatment of diabetes mellitus in humans and cats. Oral sulfonylurea drugs are ineffective in diabetic dogs, presumably because dogs lack functional pancreatic beta cells at the time diabetes is diagnosed. Acarbose is a complex oligosaccharide of microbial origin that competitively inhibits pancreatic α-amylase and α-glucosidases (glucoamylase, sucrase, maltase, and isomaltase) in the brush border of the small intestinal mucosa, which delays digestion of complex carbohydrates, delays absorption of glucose from the intestinal tract, and decreases postprandial blood glucose concentrations. Although acarbose is beneficial in improving control of glycemia in some dogs with IDDM, the high prevalence of adverse effects (diarrhea and weight loss) resulting from carbohydrate malassimilation and the expense of the drug limit its usefulness.Acarbose is reserved for treating poorly controlled diabetic dogs where the cause for poor control of glycemia cannot be identified and insulin treatment and dietary manipulations are ineffective in preventing clinical signs of diabetes. The initial dose should be kept low (i.e., 12.5 to 25 mg/dog at each meal) and always administered at the time of feeding. A stepwise increase to 50 mg/dog and, in large dogs (> 25 kg), a further increase to 100 mg/dog can be considered in dogs that fail to show improvement in control of glycemia after 2 weeks at lower doses.

Advances:

In surgical therapies, the first successful islet transplant was credited to Rundles and Swan. Use of encapsulate islet in diabetes management, as well as successful intravascular xenotransplantation of macro-encapsulated pancreatic islet cells in Type 1 DM reported in human patients. Micro-capsules using alginate or similar hydrogels encapsulate individual islets is also useful. Viral vector based gene therapy using adeno-associated virus encoding for glucokinase and insulin in canine diabetes had desirable effects consistently for consecutive 4 years of study. The same study has also been successfully undertaken in mouse's model.

DIET, FEEDING SCHEDULE, AND EXCERCISE

Diets that are high in fiber, low in simple sugars, and moderately restricted in fat and protein are recommended for diabetic dogs. However, the most important aspect of diet for diabetic dogs is that it be a balanced diet that the dog will eat consistently. If dogs receive insulin twice daily, it is recommended to feed two equal meals at the time of insulin administration. Some clinicians recommend that the dog be fed before injecting insulin and withholding the insulin injection anytime the dog does not eat its entire meal. Although this may work well for a dog that is a gluttonous eater, it is less effective for finicky eaters or those dogs that eat small amounts throughout the day. Even when insulin is given just once daily, it is still recommended that the dog be fed twice daily. The second meal should coincide with the glucose nadir (depending on when the owner can be home to feed) or alternatively should be presented 12 hours after the insulin injection. Feeding a larger portion of the daily diet around the time of the glucose nadir may lead to a less fluctuating glucose curve. However, if lente insulin given once daily provides adequate control, the glucose response can be improved in some cases by feeding the larger meal at the time of the insulin injection. This is because lente insulin contains 30% short acting-insulin (Semilente) and 70% long-acting insulin (Ultralente) and is absorbed in many dogs in a manner such that there are two peaks of insulin activity, with the earlier peak leading to the higher insulin concentration. Exercise affects both the absorption of insulin and the metabolic use of

glucose. Consequently, exercise levels should be kept constant with activity provided at the same times every day.

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Collection, preservation and dispatch of samples for laboratory investigations of various diseases of Pet animals

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Introduction

Each veterinary diagnostic laboratory offers a unique set of diagnostic tests that is subject to frequent changes as better tests become available. The increasing availability of tests based on newer molecular biology techniques is an excellent example of this trend. The protocols for sample collection and submission are therefore also subject to change. The practitioner and diagnostic laboratory staff must maintain good communication to complete their diagnostic efforts efficiently and provide optimal service to the client. Practitioners must be specific and clear in their test requests. The laboratory staff can provide guidance when there are questions regarding sample collection and handling, as well as offering assistance in interpretation of test results. Most diagnostic laboratories publish user guidelines with preferred protocols for sample collection and submission, but the following broad recommendations are fairly standard.

Regardless of the type of submission, a detailed case history should be included with the samples to assist laboratory personnel in determining a diagnosis. The information should include owner, species, breed, sex, age, animal identification, clinical signs, gross appearance (including size and location) of the lesion(s), previous treatment (if any), time of recurrence from any previous treatment, and morbidity/mortality in the group. If a zoonotic disease is suspected, this should also be clearly indicated on the submission sheet to alert laboratory personnel. The submission form should be placed in a waterproof bag to protect it from any fluids that might be present in the packaged materials. Waterproof markers should be used when labeling specimen bags and containers. When packaging samples, the use of breakable containers should be avoided, but properly padded glass tubes are commonly shipped. The basic principles of good shipping practices include the use of sturdy containers, clearly labeled to contain biologic diagnostic specimens; ideally, this should be a Styrofoam container within a cardboard box. Any coolant packs should be sealed within plastic bags and accompanied by adsorbent material within the container. If dry ice is used, this should be noted on the cardboard box label, and the stryofoam lid should not be sealed with tape. A "triple barrier" approach can be applied to most diagnostic specimens. One barrier is the outer cardboard box; another is the inner Styrofoam container or perhaps sealable plastic bags (with adsorbent material). Note that if air shipment of samples is anticipated, then International Air Transport Association (IATA) requirements include specialized bags capable of withstanding 95 kilopascals of pressure. The third barrier is around the sample itself. Liquid samples should not be shipped in plastic bags; a sealable tube or jar should be used. The top of these fluid-filled containers should be sealed with tape. The tube/jar is then placed in a sealable plastic bag with some adsorbent material in case of leakage. Fresh tissue samples are similarly placed within a second bag (third barrier) containing some adsorbent material. Use of appropriate padding material within the box will protect sample integrity while preventing coolant packs from crushing samples. Regulations regarding shipping of biologic samples vary according to country but in the USA are mainly in the purview of the Department of Transportation, Hazardous Materials Regulations. Also, IATA restricts the volume of formalin that can be shipped in any single shipping container; <1 L in total and<30 mL per jar.

For Microbiological investigation

Any specific agents that are of interest in the diagnostic investigation should be mentioned on the submission form; some agents have requirements (eg, anaerobic culture, special media) that would not be used in most laboratories unless the pathogen was cited as a differential diagnosis. Laboratory techniques and capabilities for microbiologic examination vary; available tests include bacteriologic culture, fungal culture, virus isolation, in-situ hybridization, a variety of PCR methods, fluorescent antibody tests, latex agglutination tests, Western blotting, ELISA, and many others. Most tests, including the newer molecular biology techniques, rely on either the growth/visualization of intact viable organisms or the detection of the nucleic acids and proteins of these pathogens. Therefore, unfixed specimens (tissue, fluid, etc) should be collected aseptically and shipped promptly to avoid degradation. If PCR testing is to be performed, it is particularly important to avoid cross-contamination between multiple animals in a submission; this applies to tissues, fluids, and even dissection instruments. Furthermore, swabs destined for PCR analysis should not be placed in agar or charcoal-based transport media. Calcium alginate swabs should be avoided. Instead, cotton or Dacron swabs should be shipped in a tube with a few drops of sterile saline or viral transport media.

Some test protocols may permit pooling of organ specimens from an individual, but for the vast majority, it is preferable that each tissue be collected into separate sterile, clearly labeled bags or tubes for shipping. Gut samples must never be pooled in a container with other tissue samples. Tissues and fluids for most microbiologic assays may be frozen before shipment, but generally freezing is undesirable if samples can be chilled and delivered directly to the laboratory within 24 hr. Exceptions to this rule include analysis for certain toxins, such as those of Clostridium perfringens and C botulinum, in which degradation of the toxin must be prevented by prompt freezing after collection. Adequate refrigerant should be provided so that samples remain chilled (or frozen) until they reach the laboratory.

For Histo-pathological investigation

Microscopic examination tissues collected either via biopsy or during necropsy can be critical to obtaining a diagnosis. Use of this relatively rapid and inexpensive diagnostic technique can often result in substantial savings in time, money, and animal life. The increasing number of immunohistochemical (IHC) tests that can be applied to formalin-fixed tissue has further reinforced the utility of this diagnostic technique.

Autolyzed tissues are generally useless for histopathologic examination; prompt necropsy examination and organ sampling are critical. Tissue should not be frozen before fixation. Other than CNS tissues (see below), samples collected for histology should never be >1 cm thick (preferably 5–7 mm) and must be placed immediately into ≥10 times their volume of phosphate-buffered 10% formalin to ensure adequate fixation. Tissues collected for histologic examination should be representative of any lesions present and, in the case of cutaneous punch biopsies and biopsies obtained via endoscopic collection, should be centered directly on the grossly visible lesions. Wedge biopsies or tissue samples collected at necropsy should include some of the apparently normal surrounding tissue; the interface between normal and abnormal may provide key information. Excisional biopsies of small tumors (<1.5 cm) may be cut in half. Larger tumors may be sliced like bread so that formalin can penetrate to the face of each slice. Alternatively, several representative samples (7 mm wide, including the interface of normal and abnormal) may be collected. The tissues should remain in fixative for ≥24 hr; after this initial fixation, the samples may be placed in a smaller volume of fresh formalin for shipment if necessary. Prolonged fixation can adversely affect IHC testing, so samples should be shipped promptly if IHC tests are anticipated. Histologic samples should be shipped in unbreakable containers and packed in a manner that prevents spillage during shipment. Fixed tissues should be protected from freezing.

Specific tissues collected at necropsy require additional attention. Because the GI mucosa decomposes rapidly, short sections of gut collected at necropsy must be opened lengthwise to allow adequate fixation. If spinal cord is to be submitted, the dura mater should be carefully incised lengthwise to permit more rapid penetration to the cord. Fixing the brain poses a special dilemma, especially if a neuroanatomic location of the lesion(s) within the organ could not be determined antemortem. Ideally, a whole, intact fixed brain is required for complete histopathologic analysis. Immersion of the brain for many days in a very large volume of formalin is required to adequately fix such a specimen, so brains are commonly transported in an only partially fixed state. If the specimen can be shipped by overnight delivery, it may be acceptable to send a chilled, carefully packaged, unfixed brain, which can then be processed at the diagnostic laboratory. Often, the brain is halved longitudinally and one-half sent unfixed (fresh), properly refrigerated, for microbiologic tests, while the other half partially fixes in transit. This method can prove unsatisfactory if a solitary unilateral lesion is involved. Slicing the brain into widths suitable for rapid fixation introduces considerable fixation artifact and should be avoided if possible; fixing the intact/halved brain in a large volume of formalin for >24 hr is preferred.

For toxicological investigation

If a known toxin is suspected, a specific analysis should always be requested—laboratories cannot just "check for poisoning." A complete description of clinical and epidemiologic findings may help differentiate poisoning from infectious diseases that can simulate poisoning. Tissues or fluids for chemical analysis should be as fresh as possible and kept refrigerated. For some analyses, freezing is critical to prevent degradation of volatile chemicals, and in rare instances a chemical preservative is required. If legal action is a possibility, all containers for shipment should be either sealed so that tampering can be detected or hand-carried to the laboratory and a receipt obtained. The chain of custody must be accurately documented. If feed or water is suspected as the source of poisoning, samples of these and any descriptive feed tag should accompany the tissue samples. If at all possible, a representative composite sample of the feed should be submitted from the suspect lot or shipment. In some instances, if an adequate amount of involved feed is available, some of it may be fed to experimental animals in an effort to reproduce the signs and lesions seen in the field cases.

For Hematology investigations

Routine studies require anticoagulated whole blood and several blood smears. Blood smears should be prepared immediately after the sample has been collected to minimize cell deterioration. Anticoagulated blood should be kept refrigerated; blood smears should not. EDTA is the anticoagulant of choice for a CBC because it best preserves the cellular components of the blood and prevents platelet aggregation. Blood for coagulation testing should be collected into a blue top tube, which contains sodium citrate. After mixing, the sample should be centrifuged for 5 min, and then plasma should be removed and transferred to a clean tube without anticoagulant. The plasma should be kept frozen until the time of analysis. Whole blood should not be frozen because this causes cell lysis and gross hemolysis, which interfere with testing.

For Clinical Chemistry investigations

Most clinical chemistry tests require serum, but an occasional test may require plasma. Anticoagulants present in plasma may interfere with tests; therefore, serum should always be submitted unless plasma is specifically requested. Because lipemia can interfere with a number of chemistry tests, dogs and cats should be fasted for 12 hr before samples are collected.

For serum samples, the blood should be drawn into a red top tube or a separator tube. The sample should be held at room temperature for 20–30 min to allow complete clot

formation and retraction. Incomplete clot formation may cause the serum to gel due to latent fibrin formation. The clot should be separated from the glass by gently running an applicator stick around the tube walls ("rimming"). The sample should then be centrifuged at high speed (~1,000 g; 2,200 rpm) for 10 min. Rough handling of the sample or incomplete separation of erythrocytes from serum may promote hemolysis, which can interfere with certain tests.

If the sample has been collected into a serum separator tube, centrifugation will cause a layer of silicon gel to lodge between the packed cells and the serum. The gel layer should be inspected to ensure the integrity of the barrier, and re-centrifugation is recommended if there is a visible crack in this layer. If a red top tube has been used, the serum should be removed and transferred to a clean tube. Serum should be refrigerated or frozen until analyzed. Many commercial laboratories provide sample containers and mailers.

For Cytology

Air-dried smears are usually acceptable. Rapid air drying of smears minimizes cell distortion, thereby enhancing diagnostic quality. However, depending on the method of staining used, some laboratories prefer alcohol-fixed smears. Samples can be obtained by fine-needle aspiration or by scraping. Imprints (touch preparations) of external lesions can also be used, although these tend to have a greater degree of contamination. Aspirated material should always be smeared before air drying. Smears of fluid can be prepared using a traditional blood smearing technique. Highly cellular fluids may be smeared directly; fluids of low cellularity should be centrifuged to concentrate the cells. Thick material or viscous fluid is more readily smeared using a squash technique in which a second glass slide is placed over the aspirated material and then slid rapidly and smoothly down the length of the lower slide. Blood or cytologic smears should never be mailed to the laboratory in the same package with formalin-fixed tissues because formalin vapors will produce artifacts in the specimen. Many laboratories now offer immunocytochemical testing, and proper handling of cytologic submissions is required for reliable results. Usually air-dried, unfixed smears will suffice, but in some instances shipping of samples in tubes containing a transport media is recommended.

For Fluid Analysis

Analysis of various effusions and fluids usually includes determination of protein content, total cell count, and cytologic examination. Other tests may be performed depending on the source or appearance (eg, chylous fluid) of the effusion. A sample of effusion/fluid should be collected into an EDTA (purple top) tube for routine analysis. A second sample should be collected into a serum (red top) tube if any biochemical analyses (eg, triglyceride, cholesterol, lipase for chylous effusions) are to be performed or if a bacterial culture is desired (eg, joint fluid). These samples should be shipped chilled but not frozen. Smears for cytologic examination (see above) should be prepared from a drop of the fluid immediately after the sample has been collected to minimize cell deterioration and other in vitro artifacts. Samples of CSF should be collected into small EDTA tubes and shipped immediately with high priority; the cytologic value of CSF samples degrades rapidly and the low cellularity makes examination of direct smears unrewarding. If sufficient CSF is available, then a red top tube sample may be useful for serology or culture attempts.

For Genetic Analysis

Tests based on the detection of specific genetic features range from karyotype analysis to the identification of specific genes. The laboratory offering the test should be contacted to determine the specifics of sample collection and handling; required samples range from hair to skin or blood. Many blood-based analyses require collection into yellow-topped acid-citrate-dextrose tubes and overnight shipment of the chilled tubes to the laboratory. Tissue samples for genetic analysis should be unfixed and shipped immediately

after collection. As with most molecular techniques, aseptic collection and the prevention of cross-contamination between samples is critical for reliable test results.

Apart from these general guidelines the specific clinical materials which are to be collected in some of the important infectious diseases of pet animals are given in the following tables. The ideal material for skin infections (fungal or parasitic) are optimum amount of skin scrappings along with hairs covering the both healthy and affected areas.

Table 1: Sample collection in important viral diseases of pets

Rabies	Half of the brain in 50% glycerine saline on ice packed carefully sent through courier. Brain fixed in 10% formalin, smear of brain tissue fixed in methyl alcohol
Infectious canine Hepatitis	Lung, liver, kidney and urinary bladder in transport media and 10 % formalin. Paired sera samples
Canine distemper	Conjunctival swabs and scrapings from gums in Hank's medium, gum smears on slides, pieces of lung, liver, bladder, trachea, stomach in Hank's medium and in 10% formalin
Canine parvo virus	Pieces of liver, spleen, intestines, stomach, mesenteric lymph nodes in 50% glycerine saline

Table 2: Sample collection in important bacterial infections of pets

Leptospirosis	*Leptospira icterohaemorrhagiae* and *L. canicola* occur in dogs and produce jaundice. *L.pomona* has been incriminated in bovines. Horses and swine are also affected.	Serum, urine	Microscopical Dark field microscopical examination of serum and urine. Microscopical agglutination test (MAT), Slide or Plate agglutination test.
Pyaemia	*Pyaemia means the presence of pus producing (pyogenic) organisms in the blood. This results in abscess formation. Several microorganisms caused this condition.*	Pus smears, Pus swab.	Microscopical examination after staining with Grams and another with acid fast method of staining. Isolation and identification of the organisms for confirmation.
Streptococcal Infections	*Streptococcus agalactiae* *S. dysgalactiae, S. uberis. These three are called as mastitic Streptococci.* *S. zooepidemicus produces severe infection often ending in septicaemia and death.*	Swabs and smears from lesions, milk smears and milk in case of mastitis.	Isolation and identification of the organism.

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Introduction of different dog and cat breeds

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Dog belongs to the family Canidae. Dogs are believed to be direct discendent of wolf. Scientific name of dog is *Canis familiaris* whereas that of wolf is *Canis lupus.* It is estimated that there are around 500 different dog breeds in world. Domestication of dog is practiced since as long as few thousand years. Dog serve humans via protection, security, sight, hearing and companionship. In Greece dogs were used for the purpose of herding large and small livestock, guarding humans and their properties and assistance in hunting. In Egypt, dogs are also reported to be used in wars.

Dogs are bred based on climate, environment and master's preference viz. herding, guarding or hunting. This has stimulated various dog industries, accessory manufacturers, veterinarians, pharmaceutical industry, breeders, racers, trainers and herders.

Average life expectancy of dogs is 9 to 15 years, however some live upto the age of 20 also. Small dogs live longer than larger dogs. They shed hair once in a year. Other common features includes non-retractable claws, 42 adult teeth, pointed canine teeth, panting and sweat glands on nose and feet. They can hear twice better than human beings. Sound of higher frequencies are also audible to them. Their ability of smelling is popularly used in tracking and hunting.

Classification of dogs are based on breed (morphology, form and structure), variety (hair length and color, body size and type, instinct traits) and utility. Following table shows some of the popular dog breeds of world.

Group	Examples of Breeds	Uses
Terrier	Airedale Terrier, Bedlington Terrier, Bull Terrier, Calrn Terrier, Irish Terrier, Fox Terrier, Jack Russell Terrier	Originally used in catching prey such as foxes, badgers, and rabbits
Working	Collie, Boxer, Alaskan Malamute, German Shepherd, and St. Bernard	Guards, guides, and herders such as muscular, even-tempered, and obedient
Sporting	Pointers, Retrievers, Setters, and Spaniels	Hunt by air scent
Hound	Beagles, Foxhounds, and Bloodhounds	Track their prey by ground scent
	Greyhound type such as Whippets, Borzois, and Salukis	Hunt mainly by sight
Herding	Collie, Australian Shepherd, Corgi	Drive livestock and keep farm animals from straying
Toy	Pekinese, Pomeranian, and Pug	Pets and companions
Non-sporting	Boston Terrier, Bulldog, Chow, Dalmatian, and Poodle	Bred principally as pets and companions

Some important dog breeds are:

Pointers

These are bred several hundred years to "point" birds and small game such as rabbits. It is a versatile field dog and excellent family dog. It excels in field, dog shows and obedience. Males are 25 to 28 inches at the shoulder and weight 25 to 35 kg.

Retrievers

One of the most popular dog breeds. The retrievers are bred to be both friendly companion and useful working dog. These work as retriever for hunters, assistance to handicapped, show competitor, search and rescue dog among other canine jobs.

Spaniels

Spaniel breeds are bred to be all round hunting dog. Specialized in water fowl, these are skilled swimmers and has a water resistant double coat. The dog breed has high energy to chase and retrieve game, but can also make a great family companion. This breed has an average height of 1 foot 3 inch to 1 foot 6 inch tall at shoulder and weight of 11 to 20 kg. Average lifespan is 12 to 15 years.

Fox terrier

Fox terriers were originally bred to flush foxes out of their hiding places during fox hunts. Today they are family companion and show dogs. They are upto 1 foot 3 inch height at shoulder and weight 7 to 9 kg. They live around 12 to 15 years.

Boxer

Boxers were originally medium sized guard dogs. Nowadays they are part of home as loving family companion. Height of this breed is 1 foot nine inch to 2 feet one inch and weight is 27 to 31 kg. These are large, muscular, square headed dog. With minimum grooming needs, patience and gentleness with children, boxers are great family companion.

German shepherd

This is one of the most popular breed. This breed is intelligent and capable working dog, excellent at anything he has trained to perform viz. guide and assistant work for handicapped, police and military service, herding, search and rescue, drug detection, competitive obedience and faithful companionship. Height 1 foot 10 inch to 2 feet 2 inch and body weight 34 to 43 kg. Average lifespan is 10 to 14 years.

Saint Bernard

Originally the breed was used to guard the grounds of Switzerland's hospice Saint Bernard as well as to help find and save lost and injured travelers. Today the St. Bernard enjoys the family life in many homes across the world. This breed is versatile and excels in the show ring and obedience trials, drafting and weight pulling competitions. Height: 2 feet 2 inch to 2 feet 6 inch. Body weight: 54 to 81 kg. Average lifespan is 8 to 10 years.

Bloodhounds

The bloodhound's ancestors were developed in France to trail deer and boar. This breed is highly active and intelligent whose keen sense of smell has found him a special place in law enforcement and search and rescue operations. Height: 1 foot 11 inch to 2 feet 3 inch. Weight: 36 to 50 kg. Average lifespan is 11 to 15 years.

Pomeranian

The foxy headed little dog is compact and active, capable of competing in obedience and agility. They can also be a simple family friend. Pomeranian are loyal to their family. They are curious and alert about the world around them. Height: 7 inch to 1 foot tall. Weight: 1 to 4 kg. Average lifespan is 12 to 16 years.

Pug

Pugs are square and thickest, usually weight no more than 10 kg. Their heads are large and round, with large round eyes. They have deep and distinct wrinkles on their faces.

These are sturdy compact dogs and are known as clowns of canine world. Originally bred to be a lap dog, the pug thrives on human companionship.

Poodle

The dog was originally a water retriever, a job that require jumping into the water to fetch waterfowl for the hunters. These are people friendly dog who want to stay close to their families. Height: 1 foot 10 inch at the shoulder. Weight: 20 to 32 kg. Average lifespan: 12 to 15 years.

Dogs found in India

Indian Pariah

Being Indian native breed, these dogs are widely found all across India. Double coated medium sized dog with about 20-25 inches height. Common colours are spotted brown or dark and reddish brown. It is also found in black colours.

Rajapalayam

This breed have heavier boned, large dog found in milk white, solid black and brown colors. These are found in Southern part of India.

Mudhol Hound

Found in various colours like fawn, fallow, red, cream, black, and mouse-grey. This Indian dog breed has long and narrow head broad between the ears. This breed is found in Karnataka, Maharashtra and Andhra Pradesh.

Rampur Greyhound

Found in northern India, these dogs are 24 to 27 inch in length, deep chest, sprung ribs with a long tail slightly curving upwards.

Chippiparai

This breed in indigenous to South India. This is a medium sized dog having a fawn, reddish brown, slight black tinged coat, silver-grey, with limited white markings. They have a long curved tail.

Kanni

Native breed of Tamil Nadu, these dogs are black and tan in colour but limited white on chest and feet. Looks similar to Doberman pinscher.

Gaddi Kutta

Muscular, deep chested Gaddi is athletic in nature with great speed. Thay are found usually with cropped ears. They are well suited to cold and temperate climate of Northern India.

Combai

Looks more like Rajapalayam, this dog breed is red brown in colour with a black mask. They have strong jaws, savage temper and a tendency to be much more active. This breed is found in South of India

Cats belongs to family Felidae. These were domesticated around a thousand year in Egypt and eastern world. Cats are worshiped in Egypt, whereas in eastern world certain they are consumed as human food. Cats are beneficial to human as they check the number of mouse and rat, act as companion and aid disable people. Cats are also responsible for growth and income of cat food companies, accessory manufacturers, veterinarians, pharmaceutical industry and breeders.

Average body weight of cat ranges from 2 to 10 kg. Their average lifespan is 10 to 15 years. They contain retractable claws, 30 permanent teeth and pointed canine teeth. Cats have excellent night vision and they can see upto 120 feet distance. They can hear 1.5 times better than dogs. Their smelling capacity is 14 times better than human.

Natural standard classification of cats is performed on the basis of breed (morphology, form structure), Hair length and color, body size and type and length of limbs. Some important cat breeds of world are listed below.

Short haired cats

Siamese, Burmese, Abyssinian, Color Point, Havanah Brown, Exotic Shorthair, American Shorthair, Bengal (Hybrid).

Long haired cats

Balinese, Somali, American Curl, Maine Coon, Scottish Fold, Persian/Himalayan, Birman.

Rex

Cornish Rex, Devon Rex, Sphynx (Hairless)

List of cat breeds in India

1. **Rusty spotted cat**
   This breed is found in Southern India. This world's smallest wild cat is grey in color with rusty spots over the back and thigh. They are having dark streaks on each side of head, cheeks and forehead. They are about 14 to 19 inches in height and having weight around 0.9 to 1.6 KG.
2. **Leopard cat**
   Native of eastern India, this cat is having leopard-like spots and having size of a domestic cat. Their long legs and small head is marked with two prominent dark stripes make them different from others.
3. **Himalayan Cat**
   This cat is originated from United States and United Kingdom but is very popular in India.Himalayan cats are having short legs and round body. They are having different types of coats like Blue point, Lilac point and cream point etc. There face types can be divided into two types: doll-face and the peke-faced.
4. Bombay Cat
   Originated from United States, this social breed cat is black in color with short hair. They are having yellow eyes and glossy coats.
5. **Maine Coon Cat**
   From Maine, United States, Maine Coon Cat has large paws and long haired cat. They are found in various colors like chocolate, lavender etc. Their coat is soft and silky.
6. **Siamese Cat**
   Siamese cat has muscular body and a triangular head. This large ear cat has long neck and glossy furs with a long tail. Native of this breed is Thailand.

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Surgical methods for controlling of dog population

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The high density of human population in Indian cities and accompanying dogs provide ample opportunity for zoonotic diseases to be transmitted. Indeed, dogs are associated with more than fifty zoonotic disease agents of which Rabies is the most prominent.It is thus clear that the control of street dogs is important for the control of Rabies. Further it is clear that the control of Rabies should be a multi-disciplinary activity involving veterinary departments, health departments and those agencies concerned with civic infrastructure particularly urban cleanliness, and NGOs concerned with animal welfare. No one agency will achieve the results that could be achieved by honest endeavour of all these agencies working together to ensure a rabies free environment for citizens and humane control of dogs.The abundance of dogs is dependent on the habitat, especially the availability of resources such as food, water and shelter. Access to these resources depends on settlement patterns, rubbish and waste disposal, rules for keeping animals and other cultural practices. To understand the population biology of the species, it is important to keep in mind the differences in ownership status, degrees of restriction on their movement, social interaction, reproduction and levels of dependence on human care.

Why Bitches ? Why Not Male Dogs?

One unspayed female could give birth to as many as 20 pups a year (although our figures indicate a bitch is most likely to give birth to only one litter per year and that of 6 pups only). One unneutered male could mate with several females resulting in hundreds of unwanted births. A programme concentrating on males rather than females can be rendered completely ineffective if only a few males escape sterilisation whereas the same number of missed females will have a very limited effect. Therefore a female-focussed programme is a more effective use of limited resources.

A female-focussed programme has the following additional benefits -

- If there are fewer bitches in heat there is less aggression in male dogs in dispute over females.
- Unneutered males can more effectively protect the territory of the group, reducing inward migration of dogs from outside possibly carrying rabies and other infectious diseases.
- Spayed females are more able to maintain body condition on a limited food source as they are not supporting pregnancy and lactation.

Pre-surgical Checks, Pre-operative Preparation and Fluid Therapy

Ensuring that the stray dog selected to undergo the sterilization is ‘fit for surgery’ is essential if the surgery has to be successful. Dogs suffering from a serious bacterial or viral infection can put all the animals in the shelter at risk of being infected. As a rule, if any stray dog that has been caught by the dog catcher displays visible clinical signs of illness, such as extreme emaciation, pallor, weakness or skin conditions like mange, the dog should be first treated for the condition in the veterinarian. Giving the dogs a thorough checkup prior to surgery can help filter out the dogs that are unhealthy and are not fit for surgery, thus minimizing the chances of post-surgical deaths and delayed post-surgical healing. Careful adherence to the pre-operative procedure can minimize the dose of the anesthetic needed as well as ensure that the operated dog has a safe recovery from the effects of anesthesia.

Fluid loss during surgery can cause a great deal of stress to the animal and may cause severe dehydration and shock, even leading to death if there has been severe hemorrhage

from any of the ligated blood vessels. Giving an adequate quantity of intravenous fluids (NSS-Ringer lactate) during the surgical procedure will help ensure that the dogs' tissues are adequately perfused and thus minimize the risk of surgical shock.

Prior to commencing sterilization surgery, the veterinary surgeon should check

- That the records of dogs and the dogs in the kennels are in agreement;
- The clinical condition of dogs for surgery;
- The preparedness of the operation theatre and preparation room;
- The sterility of surgical instruments and equipment;
- The availability of the required medications and;
- The physical environment in which the anaesthetized animals will recover (hypothermia is a severe problem in anaesthetized animals and it is essential that the dogs are kept warm during the immediate post-operative recovery period). If any of the above is found wanting or deficient, steps should be taken to improve the situation, or the surgeries should be postponed until the conditions are made professionally acceptable.

Pre-Surgical Checks

Every dog at the shelter that will receive anesthesia and undergo surgery should be given a thorough pre-surgical check by an experienced veterinary surgeon. It is essential to carefully examine each dog prior to surgery to ensure that the concerned animal is in a state of fitness to undergo surgery.

The key clinical parameters to be monitored are as below:

- Temperature
- Respiration
- Pulse
- Color of the mucus membranes
- Palpation of regional lymph nodes
- Auscultation of chest to rule out any infection of the lungs as well as to identify cardiac rate and rhythm abnormalities.
- Signs of external injury. e.g. fractures and wounds, skin conditions like mange etc.
- Abdominal palpation to rule out pregnancy, ascites, liver and splenic condition.
- It is only after the veterinary surgeon has confirmed that the above parameters have been checked and found to be normal, that the dog can be considered, 'ready for surgery'.
- Incurably sick and mortally wounded dogs should be considered for euthanasia.

Preparation of the Patient prior to surgery

Withholding of food

The dogs selected to undergo surgery should not be given food for 12 hours to reduce the dogs' risk of vomiting and pulmonary aspiration while undergoing general anaesthesia. A shorter fasting time for weak dogs and puppies is recommended. Water should be available to the dogs.

Pre-medication

Prior to anaesthesia, the dogs should receive pre-medication with a sedative agent. Doing so will help to reduce the total amount of anaesthetic that is required and will also help to keep the animal quiet and suitable for induction.

Analgesia

Prior to surgery, pre-emptive analgesia such as meloxicam should be administered. This is because pain relief given before a painful stimulus is experienced is more effective than pain relief given after pain has begun.

Antibiotic use

Pre-operative use of antibiotics can be considered. For sterilization surgery done under suitable conditions of asepsis, the use of antibiotics may not be necessary. In less than ideal conditions, a long acting antibiotic could be considered. The use of antibiotics has to be done judiciously and should be decided on a case by case basis by the veterinary surgeon.

General Anaesthesia

General anaesthesia should be administered and the dog must be monitored continuously, to ensure that an adequate depth of anesthesia is reached so that the surgery can be safely performed. Once anaesthetized, and throughout the anaesthesia, the patient should, if necessary, be protected against hypothermia.

Patient Preparation for Surgery

- Anaesthetic induction, shaving and prepping must be performed on a separate table other than the surgery table, to minimize contamination.
- If intravenous fluids are to be administered, the catheter site should be shaved and prepped as described for the surgical site below. The catheter is then inserted and the primed intravenous line connected.
- The bladder should be palpated and expressed if necessary and genitalia examined for presence of Transmissible Venereal Tumour (TVT).
- The surgical site should be widely and carefully shaved, avoiding trauma to the area; even small cuts can lead to wound infection.
- The site should be thoroughly cleaned with Chlorhexidine solution. Multiple pieces of cotton wool should be used in turn, commencing at the centre of the area and moving towards the periphery of the shaved area, and never back into the centre, otherwise the wound will be re-contaminated.
- Avoid wetting non-shaved areas of the patient.
- Once the shaved area appears free of gross dirt and hair, and the pieces of cotton wool used come off the skin with no staining, then the site can be considered clean, but NOT disinfected at this point.
- Disinfection of the site is achieved using three spray applications of surgical spirit - one minute between applications. A final spray of Povidone iodine solution may also be applied, but only once after the spirit has evaporated and the skin is dry. Do not touch the skin during this process, otherwise adequate disinfection will not be achieved. Once again, avoid wetting the non-clipped areas as this may lead to 'run-off' and contamination of the site.
- The patient is then transferred to the surgery table: in seeding, take care not to contaminate the prepped area with your hands or non-disinfected parts of the patient.
- The prep table should then be carefully cleaned with an appropriate disinfectant, such as Lysol solution.

Surgical Scrub:

An acceptable germicidal preparation, e.g. Chlorhexidine or Betadine, must be used and scrubbing should be carried out for a minimum of 5 minutes with Chlorhexidine, followed by scrubbing with Povidone Iodine.

Scrubbing:

- The hands and arms are washed first with the scrub mixture to remove any gross contamination.
- The nails are cleaned next, before the scrubbing procedure begins

- A sterile brush is used to scrub:

1. The fingers
2. The hands
3. Finally, the arms

In that order, scrubbing should over a period of no less than 3 minutes. Once the brush has been used on the arms, it should not return to the fingers. Each finger should receive ten strokes on each surface, making a total of forty strokes per finger. The finge mails and both surfaces of the hands should receive twenty strokes. The number of scrubbing strokes is far more important than the time spent scrubbing.

Rinsing

When scrubbing is completed, the hands, arms and the brush should be rinsed in water, allowing the water to drip from the elbows to prevent contamination of the hands with drips from upper arms.

Drying of hands

Two sterile hand towels are provided. The first towel is unfolded and used to dry thoroughly the fingers, hand and forearm (in that order) of one arm, taking care that the fingers of the hand holding the towel do not contact the skin of the other arm. The second towel is used to dry the other hand and forearm in identical fashion.

Alcohol Spray

With the hands held above the level of the elbows, surgical spirit should then be sprayed on the hands and then the forearms, and allowed to dry.

Anesthetic & Surgical Protocols

The particular combination of anesthetic and pre-medicant to be used is a choice that should be made by the Veterinary Surgeon.

Good anesthetic protocol should achieve the following:

1. Loss of consciousness that permits surgical procedures to be carried out
2. Sufficient degree of sedation, analgesia and muscle relaxation
3. Maintenance of adequate cardiac function at optimal levels
4. Adequate ventilatory and respiratory support

The cephalic vein of the forelimb or the saphenous vein of the hindlimb may be used to give intravenous anesthesia while medications to be given intramuscularly may be given in the cranial thigh muscles, so as to avoid sciatic nerve injury. Administration of Meloxicam @ 0.1 - 0.2 mg/kg bw by intravenous route 20 minutes prior to inducation of anesthesia can help to significantly reduce post-operative pain.

Anaesthetic protocols

Some recommended combinations are listed as below:

Anesthetic Protocol 1

Xylazine-Atropine-Ketamine-Diazepam

Pre-medication

- Xylazine @ 1mg / kg bw (administered intramuscularly - maximum dose 1 ml)
- Atropine @ 0.04 mg / kg bw (however, there is increasing evidence that atropine should not be given as a premedicant and should only be administered folllowing induction to maintain cardiac output)

Induction: To be given ten minutes after administration of Xylaxine and Atropine

- Ketamine @ 2.5 mg / kg bw + Diazepam @ 0.25 mg / kg bw Mix equal volumes of ketamine (50 mg/ml) and diazepam (5mg/ml) and in the same syringe

Maintenance

Increments to be given at half the induction dose

Fluid Therapy

- Ringer's Lactate should be administered by I/V route throughout the surgical procedure.

Respiration

Open mouthed with gag and spontaneous respiration / via endotracheal tube endotracheal tube inserted and cuff inflated if necessary.

Anesthetic Protocol

Triflupromazine / Atropine / Thiopentone or Xylazine / Atropine / Thiopentone

Pre-medication

Triflupromazine @ 1mg / kg BW or Xylazine @ 1 mg / kg BW

Atropine @ 0.04 mg / kg BW

Note: The combination of Xylazine-atropine-thiopentone is not considered safe for old, weak and young patients and it is recommended that Protocol 2 be used only by an experienced vet.

Induction

Thiopentone @ 25 mg / kg BW I/V

(Note: peri-venous administration of thiopentone sodium will cause severe local reaction and must be treated by local infusion of at least three times the volume of sterile saline; this risk can be reduced by the use of a 2.5% solution and by ensuring that thiopentone sodium is given by intravenous route only)

Maintenance

I/V Thiopentone at half the induction dose may be repeated as small I/V boluses but will lead to prolonged anesthesia and longer recovery time.

Fluid Therapy

Ringer's Lactate should be administered by I/V throughout the surgical procedure.

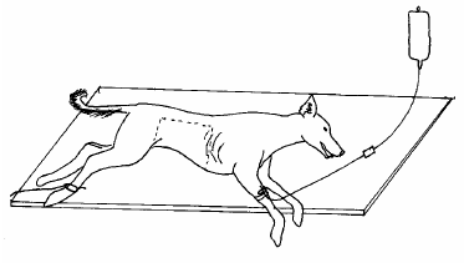
Respiration

Open mouthed with gag and spontaneous respiration

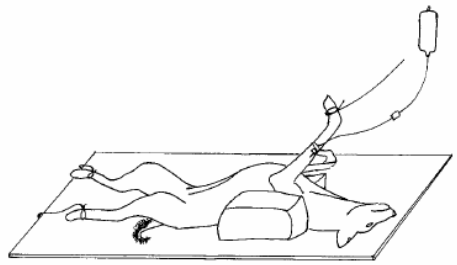
SURGICAL PROTOCOL

The dog is then carried carefully into the operations' theatre. Care must be taken when carrying anaesthetised patients that the prepared operation site is not inadvertantly contaminated. The animal is then:

- Positioned on the operation table. It is important to have patients, particularly bitches, correctly positioned for easy surgery.
- Surgical spirit is applied to operation site.
- An infusion set is attached to the i/v catheter and normal saline is slowly administered throughout the operation. In addition to improving the hydration status, this prevents blood clotting within the catheter, and allows easy administration of extra anaesthetic or other drugs as required.

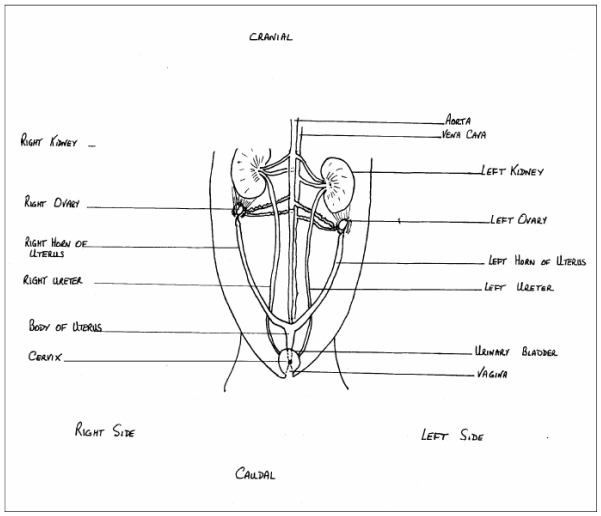


Position of bitch for Flank Spaying



Position of Bitch for Mid-Line Surgery

The surgeon and assistant scrub up using disinfectant soap and povidine-iodine solution (Betadine) and tap water, then, when the hands are dry, don a pair of sterile gloves in normal aseptic fashion without touching the outer surface of the gloves. Formerly chlorhexidine solution (Citalon) was used for scrubbing up but was found to cause skin reaction amongst some staff. It should be self evident that it is not possible to scrub up satisfactorily while wearing any kind of jewellery on the hands or arms, including string bracelets. All such should be removed beforehand. Help in Sufering allows no exceptions to this rule under any circumstances. Finger nails should be kept short and clean. Effective scrubbing up, which is different to washing hands, is dependent on the length of time that the cleansing solutions are in contact with the hands. This is particularly so with povidine iodine solutions.



FLANK APPROACH - ADVANTAGES AND DISADVANTAGES

Advantages

- Easy to check the wound and to apply any topical medicine in fractious animals.
- Wound are of three separate muscle layers each individually sutured (catgut can safely be used in this site). Wounds are not under not under the weight of abdominal contents.
- Less tension in incision area and increased vascularity can reduce healing time.
- In young lean animals the spay can easily be performed through a very small incision.
- Animals can be released earlier than with midline.

Disadvantages

- Cutting through the 3 muscle layers can cause bleeding which may be sufficient to obscure the surgical field and can lead to increased risk of post-operative infection.
- Recovery of a dropped or bleeding ovarian stump may be difficult.
- It may be difficult to expose the opposite ovary and uterine bifurcation if the original incision was incorrectly placed.
- Severe reactions to catgut can occur. Degradation sometimes produces swellings within the muscle layers. These need to be monitored as they are a favourable site for infection.
- Approach more traumatic (i.e.: through three muscle layers) than midline, and therefore increased post-operative pain is possible.

MIDLINE APPROACH - ADVANTAGES AND DISADVANTAGES

Advantages

- Incising through fascia and connective tissue causes very little bleeding.
- Less trauma to abdominal wall - decreased post-op pain.
- If any haemorrhage or dropped pedicles, can easily extend incision a little to locate and clamp bleeding vessel.
- In operations requiring a longer incision, e.g.: pregnant bitches, pyometras, it is less traumatic than a flank approach. There is decreased bleeding, and also possibly less post-operative pain.
- Minimal / no reaction to monofilament nylon suture used in the abdominal wall.

Disadvantages

- The linea alba, through which the midline incision should be made, may be difficult to identify.
- Wound is more inaccessable and thus harder to check in fearful animals.
- Risk of wound breakdown and herniation.
- Dogs must be kept longer to allow adequate healing, as the healing rate of the fibrous linea alba is slower than muscle.
- Nylon remains in the body for the life of the dog and can become a focus for infection.

CRITERIA FOR CHOOSING APPROACH

The choice of approach is influenced greatly by the surgeon's experience and preference. As the HIS ABC programme has progressed the flank approach has become the prefered surgical approach in all but the most exceptional cases.
A midline approach may be preferred by some surgeons in:

1) Heavily pregnant bitches

2) Pyometras

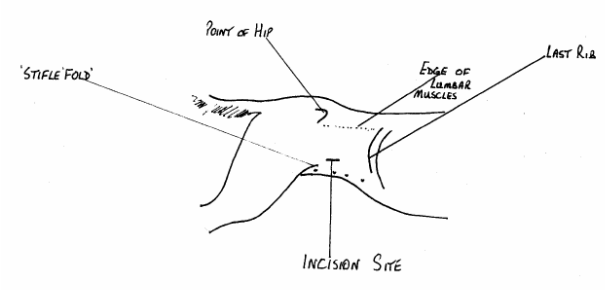
3) Fat and heavy, well-muscled bitches, such as pet dogs.

FLANK SPAY TECHNIQUE

Approach

Dog is positioned lying on its left side and abdomenal cavity is entered via the right flank with the ventral aspect of the dog towards the surgeon. The incision is made about 4 cms behind the most caudal curve of the last rib, parallel to the spine and about 9 cms ventral to the transverse processes of the lumbar vertabrae. The incision often falls at the cranial end of the stifle fold of skin. In young bitches (under 6 months) the incision is placed more caudally. Failure to do this in young dogs results in difficulties in exteriorising the cervical stump.
Note: The right ovary is more closely adhered to the right kidney and body wall than the left ovary and thus easier to exteriorise if incision is made in right flank.

The skin is cut with a scalpel. Subsequent layers are separated using scissors and blunt dissection. Incising the 3 muscle layers can cause haemorrhage. Splitting the muscles along their fibres reduces bleeding, causes less trauma and faster healing, but may results in a smaller aperture through which to work.



Location of Incision Site for Flank Spay

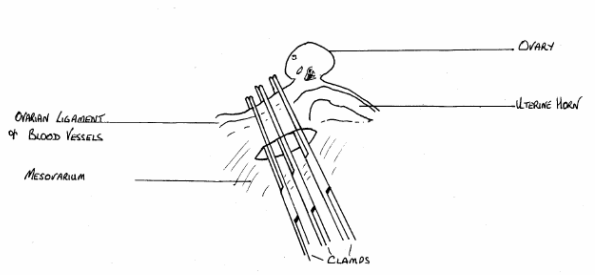
Spay

The right uterine horn is located with spay hook. The horn is elevated so that the ovary is grasped between the thumb and index finger of one hand. The suspensory ligament is stretched or broken with the second finger of that hand. When breaking the suspensory ligament direct the tension caudally to protect and avoid tearing the ovarian vascular complex and subsequent haemorrhage.Locate the ovarian vascular complex. Make a window in the mesovarium immediately adjacent to the vasculature.The ovarian vascular complex is clamped with artery forceps. The surgeon should keep hold of the ovary when applying the first clamp to ensure the clamp is placed below the ovary and thus that entire ovary is removed.

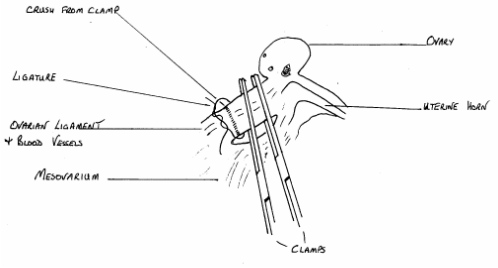
An absorbable suture, eg. Chromic catgut should be used for all ligatures. We use chromic cat gut size 5 or 6 metric (1 or 2 imperial). Choice of size of suture material depends on the size of the vascular bundle being ligated. Catgut no. 2 is used for adult bitches, but no.1 for puppies and adolescent animals.A circumferential suture is placed loosely around the pedicle at the clamp furthest from the ovary. The clamp is removed as the suture is tightened so that the suture lies in the groove of the crushed tissue created by the clamp ensuring greater ligature security. A transfixing suture (i.e. one where the suture material passes through the tissues rather than just around them) may be placed proximal to the ligature. This may be prudent in very fat bitches.

Securely Tightened Ligature in place around the Ovarian Vessels

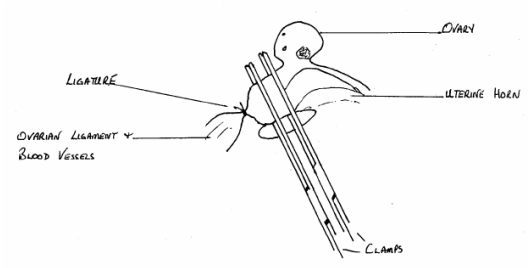
The ovarian stump is cut with scissors between the 2 clamps closest to the ovary ensuring that all ovarian tissue is excised.



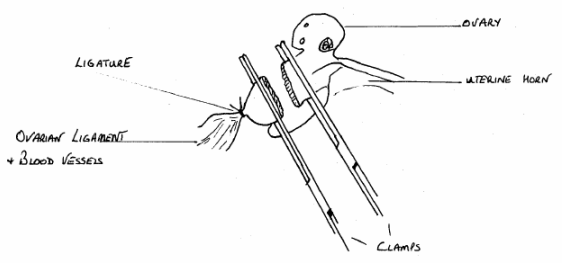
Clamping the Ovarian Blood Vessels



Placing Ligature into Crush caused by Clamp



Securely Tightened Ligature in place around the Ovarian Vessels



The Ovarian Vessels are cut from the Ovary

The Ovarian Vessels are cut from the Ovary

The stump is grasped (without grasping the ligature) with thumb (rat toothed) forceps. The clamp on the stump is released. The stump is inspected for bleeding. If none occurs the stump is replaced in the abdomen. Care must be taken to ensure that a section of body wall has not been inadvertently incorporated in the ligature.The second (left) uterine horn is located by following the right horn distally to the bifurcation. Repeat procedure as for first ovary.Both ovaries and both horns of the uterus are exteriorised, along with the attached mesovarium and associated uterine blood vessels.

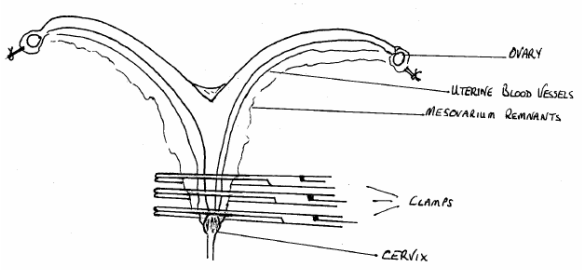
The Exteriorised Uterine Horns and Ovaries

A window is then made in the mesovarium adjacent to the uterine artery and vein, and much of the mesovarium, broad ligament and associated fat is broken from the uterus and ovary. This is best done in a controlled manner towards the now free ovary. This procedure is done with both uterine horns. Following this the uterus is seen separate from other tissues except from the vascular structures which run parallel to the uterus. The remnants of the mesovarium, broad ligament and associated fat are returned to the abdominal cavity.

The uterine body is exteriorised. The cervix is located. Various techniques may be used to ligate and remove the uterine body depending on size of uterus and the surgeon's preference. The triple clamp technique is generally used (as for ovarian attachments). Care is required, particularly with bitches in season or which have recently whelped, as the uterine tissue may be friable and the clamps may cut rather than crush the tissue.The three clamps are placed on the uterine side of the cervix. In smaller / non-pregnant dogs it is possible to mass ligate uterine vasculature with one ligature as for ovarian vascular pedicle.

Clamping of the Uterus and Blood Vessels just above the Cervix

In pregnant dogs where the uterine vessels are of greater size, the uterine arteries and veins can be individually ligated between the cervix and the closest clamp. A circumferential suture is loosely placed around this clamp, the clamp is removed, and a suture tightened in to the groove of crushed tissue. A transfixation suture can also be placed if desired. This will ensure greater security of the ligature.



Clamping of the Uterus and Blood Vessels just above the Cervix

In pregnant dogs it is sometimes easier, and may result in a smaller surgical wound than otherwise, if the uterine body is ligated and removed (as described above) before the second ovary is removed.The uterine body is severed between the remaining 2 clamps. The uterine stump is then evaluated for bleeding and returned to the abdomen. In cases where the uterine stump is very large, or if there is evidence of intra-uterine infection the stump may be oversown using catgut in a Lambert's or Cushing's suture pattern, and/or a piece of mesentry wrapped around it.

Closure

On abdominal closure each muscle layer is sutured individually i.e. 3 separate layers (the peritoneum is incorporated with the closure of the transverse abdominus muscle). In puppies the peritoneum, transverse abdominus and internal abdominal oblique are sutured with one suture and the external abdominal oblique separately with another suture.The suture material used is chromic catgut; size 5 metric (1 imperial) in all dogs. Vicryl is good for this site but expense precludes use.For longer incisions i.e. more than 2 cms in length, a continuous suture pattern can be used, such as Ford interlocking.For smaller incisions i.e. up to 2 cm in length, a horizontal mattress suture may be used. We have found the horizontal mattress suture to cause far fewer visible swellings, probably due to the reduction in the amount of catgut in the muscle layers.When suturing the abdominal muscles, it is easier to work with an assistant who gently isolates the individual muscle layers.Allis tissue forceps may be placed on the very edge of the muscle layers but it is better to use Babcock forceps or rat tooth forceps as these are less traumatic to the tissues.

The subcutaneous tissues are closed, tension and dead space eliminated with 3-0 catgut in either a horizontal mattress pattern or a continuous pattern. The skin is sutured using 3-0 vicryl with suaged-on needle, in a continuous intradermal suture. The adoption of intradermal sutures has significantly reduced the recovery times of dogs in our programme. This technique requires careful attention to aseptic techniques. The suture and knots are so arranged as to be buried. The initial suture is placed inverted so as to bury the knot. The concluding knot is a chain knot which is drawn through the incision beneath the skin.

MIDLINE SPAY TECHNIQUE

If electing to perform surgery through a mid-line approach it is important to ensure that it is the fibrous linea Alba which is incised and not the adjacent muscles.

Spay

Routine spay is performed as described above for Flank Spay Technique.

Closure

Abdominal closure is in 1 layer. A simple interrupted suture pattern is used in the linea Alba. Sterile heavy gauge monofilament nylon is used. Subcutaneous tissue and skin are closed routinely as before.

IMMEDIATE POST-OPERATIVE CARE

- After sterilisation surgery the dog or bitch is placed in a warm area to recover from anaesthesia. This is only of concern in the winter months when dogs are allowed to recover in the sunshine. For most of the year the dogs are returned directly to their kennels. If allowing anaesthetised dogs to recover in an open compound care must be taken to ensure that they do not fall prey to the attention of crows or other predators, and also that they do not over heat if in direct sunshine. It is also necessary that each animal can be easily identified so it may be returned to the correct kennel. We do this by putting a small piece of tape on the animal's head on which is written its kennel number.

Small dogs/puppies are most at risk of hypothermia, even in moderate environmental ambient temperatures. Steps must be taken to keep these animals warm by using rubber mats, 'bubble wrap' plastic sheeting, warmed intravenous fluids etc.

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Rabies in Animals and its Zoonotic Importance

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Rabies is an enzootic disease in India and has a serious public health and economic implication in our country. With the exception of Andaman and Nicobar and Lakshadweep and to some extent in Nagaland, the disease is present throughout the country. Rabies continues to be endemic in Asia for various reasons. The large population of stray dogs together with a lack of effective control strategies has been responsible for increase in number of deaths from rabies. A recent national survey by the Association of the Prevention and Control of Rabies in India (APCRI) estimated that in India a total of 20,000 human deaths occur as a result of rabies each year (Sudarshan *et al*., 2006). In India, rabies occurs mainly in the urban form, although the existence of a sylvatic cycle cannot be ruled out. In the urban form, dogs play an important role as the reservoir and transmitter of the disease to humans and domestic animals, while jackals and foxes maintain the virus in sylvatic form. Worldwide, transmission from dogs accounts for more than 90% of human cases. In developed countries, bats, foxes, coyotes, raccoons, and skunks are major reservoirs.

Etiology:

It is caused by Lyssa virus, a RNA virus of the family *Rhabdoviridae*. The virus has bullet **shaped morphology and is neurotropic in nature**. The virus is **highly fragile** and susceptible to standard disinfectants. Sunlight and moderate heat destroy the virus.

Epidemiology:

Rabies is usually maintained and transmitted by particular mammalian reservoir hosts. Two epidemiologically important infectious cycles are recognized, urban rabies in dogs and sylvatic rabies in wildlife. More than 95% of human cases are the result of bites from rabid dogs. Racoons, skunks, foxes and bats are important reservoirs of rabies virus in North America. Rabies is endemic in India and considered serious concern for veterinary and medical fraternity. In India, about 15 million people are bitten by animals, mostly dogs, and estimates put the number of deaths in India at 25,000 - 30,000 annually. In the Indian sub-continent, dogs are mainly responsible for maintaining the virus in urban areas while jackals, wolves and foxes, etc. maintain the virus in sylvatic areas. In developed countries, the control of stray dogs and vaccination programmes has reduced the prevalence of urban rabies and the focus is now on wildlife reservoirs.

Transmission:

A very high concentration of virus is released from the salivary gland secretions before the onset of clinical signs. Virus in **fresh saliva** is transmitted via bite, scratch or abrasion by an rabid animal (rabid dogs shed virus in saliva 5-7 days before showing signs and cat does so for only 3 days before signs). Contamination of skin wounds by fresh saliva from infected animals. Aerosol transmission has been documented in the laboratory and in caves where bats inhabit (requires a high concentration of suspended viral particles).

Pathogenesis:

There is long and variable incubation period in animal and human rabies, usually lasting 20 to 90 days, but sometimes it lasts longer than one year (Smith *et. al*., 1991). The infection of muscle fibers at the site of bite may be a critical pathogenetic step for the virus to

gain access to peripheral nervous system. RV binds to nicotinic acetyl choline receptors at the neuromuscular junction (NMJ) and NMJ is the major site of entry into neurons. Two other RV receptors which were found out recently are neural cell adhesion molecule (Thoulouze *et al.,* 1998) andp75 neurotropic receptor (Tuffereau *et al*., 1998). The final death of the neurons and the cells in the central nervous system are due to the process of apoptosis, which is said to be undergoing in these cells (Graffin and Hardwick, 1999).

The RV is transported to the CNS by retrograde transportation, possibly by binding to axoplasmic dynein. The phosphoprotein of rabies virus binds to dynein light chain for the transportation to the CNS (Raux *et al.,* 2000). CD56 neural cell associated molecule (NCAM) is expressed on neurons, astrocytes, myoblasts, myotubes, activated T cells and NK cells. The NCAM is expressed in three major isoforms and expression occurs in adult muscle and at the neuromuscular junction. RV propagates and spreads from sites of peripheral inoculation to CNS by fast axonal transport at the rate of 12 to 100 mm per day in retrograde direction (Kucera *et al.,* 1985). Glycoprotein is important for the trans-synaptic spread of rabies virus between neurons. Glycoprotein also exerts a very important influence on distribution of RV infection in CNS (Yan *et al.,* 2002). Despite of dramatic and severe clinical neurological signs in rabies, the neuropathological findings under natural conditions are relatively mild and degenerative neuronal changes are not prominent (Iwasaki and Tobita, 2002). Now it is said that the clinical signs in rabies are due to neuronal dysfunction and not due to neuronal death, which was the earlier hypothesis (Jackson and Rossiter 1997). This neuronal dysfunction is either due to action of acetylcholine receptors, or due to action of nitric oxide or due to apoptosis or necrosis.

Oxidative or reductive status of NO is responsible for its neurotoxic or neuroprotective actions. The oxidative state (NO^+) is proposed to mediate neuroprotective effects through S-nitrosylation of thiol groups. The reductive state, NO^-, reacts with superoxide (O_2^-) to form peroxynitrate, a highly reactive molecule that can initiate non-specific protein and lipid peroxidation. It is possible that the reduced state of NO is dominant in rabies virus infected brains (Ubol *et al.,* 2001). Both innate and acquired immunity play a vital role in rabies virus infection, but it is the innate immunity which plays major role, since there are few MHC molecules for adaptive immunity to play in the central nervous system. Innate immunity is the first line of defense, activated by interferon pathways which are released from virus infected cells in CNS. Macrophages (Microglia cells) and the NK cells are mainly responsible for the production of nitric oxide in brain. Toll like receptor (TLR) 3 present on the infected cells is activated by viral surface glycoprotein (Boehme and Compton 2004) and this TLR 3 helps stimulating the expression of interferon (IFN) α/β pathways in infected cells. TLR3 along with TLR 7 and 8 are the members of TLR 9 subfamily, which are mainly concerned with the recognition of nucleic acids and related structures.

Clearance of virus from non-neural tissues often involves cytolytic elimination of the infected cells, whereas for neural tissues, specific antibody response with non-cytolytic elimination is important (Griffin, 2003). So it has been suggested that induction of specific cell mediated immunity (CMI) is a crucial factor in the protection of the host from rabies (Kawano *et al.,* 1990).

The incubation period may last from two weeks to six months. Incubation period is variable and depends upon site of bite (distance from brain), the amount of virus introduced, age and immune status of the victim.

Clinical Symptoms:

Animal Rabies: Rabies is infectious to mammals. Three stages of rabies are recognized in dogs and other animals. The first stage is a one to three day period characterized by behavioral changes and is known as theprodromal stage. The secondstage is the excitative stage, which lasts about three to four days. This stage is often known as furious rabies due to tendency of the affected dog to be hyper reactive to external stimuli and bite at anything near, wander here and there, furiously attacking other dogs and animals. The third stage is the paralytic stage and is caused by damage to motor neurons. Incoordination is seen owing to rear limb paralysis and drooling of saliva and difficulty in swallowing is caused by paralysis of facial and throat muscles. Death is usually caused by respiratory arrest. Wild animals may be abnormally tame or appear sick- beware of approaching or picking up such an animal ("dumb rabies").

Cattle/Buffalo*:* Drooling of saliva, bellowing, swaying of hind quarters while walking, anorexia, frequent micturition, paralysis of penis and death occur usually 48 hours after recumbency or after a course of 6-7 days in dumb form. In furious rabies tense, alert appearance, hyper-sensitive, violently attack other animals or inanimate objects, loud bellowing, sexual excitement and death is often sudden.

Human Rabies

A) Furious rabies: When the virus reaches the CNS, the patient presents prodromal stage with headache, fever, irritability, restlessness and anxiety. This may progress to muscle pain, salivation and vomiting. After a few days to a week the patient may experience a stage of excitement and be wracked with painful muscle spasms, triggered sometimes by swallowing of saliva or water. Hence they drool and learn to fear water (hydrophobia). The patients are also excessively sensitive to air blown on the face. The stage of excitement lasts only a few days (2-7 days) before the patient lapses into paralysis, coma and death. Once clinical disease manifests, there is a rapid, relentless progression to invariable death, despite all treatment.

B) Dumb rabies: Starts in the same way, but instead of progressing into excitement, the subject retreats steadily and quietly downhill, with someparalysis to death. In this form, diagnosis may easily be missed.

Diagnosis:

A. By assessment of:

1. **Bite**: Geographical area, type of animal, severity and site of bite.
2. **Animal**:
   Live - observe in cage: If survives > 8 days, then probably NOT rabies.
   Dead - brain sample sent to laboratory: impression smear and histopathology
3. **Man:**
   Live - difficult diagnosis: clinical picture, skin biopsy, corneal impression.
   Dead - "Negri bodies" in cytoplasm of brain cells, immune-fluorescence, virus isolation

B. Diagnostic Techniques:

1. Identification of the agent: Clinical observation may only lead to a suspicion of rabies because signs of the disease are not characteristic and may vary greatly from one animal to another. The only way to undertake a reliable diagnosis of rabies is to identify the virus or some of its specific components using laboratory tests. As rabies virus is rapidly inactivated, refrigerated diagnostic specimens should be sent to the laboratory by the fastest means available.

Precautions should be taken when handling central nervous system tissues from suspected rabies cases. Gloves should always be worn and precautions must be taken to prevent aerosols. Cutting tools, scissors and scalpels, should be used with care to prevent injury and contamination.

a) Collection of brain samples

Usually the brain is collected following the opening of the skull in a necropsy room, and the appropriate samples collected are Ammon's horn, thalamus, cerebral cortex and medulla oblongata. Under some conditions (e.g. in the field or when sampling for large epidemiological studies, this step may be impractical. In such cases, there are two possible methods of collecting some brain samples without opening the skull.

Occipital foramen route for brain sampling: A 5 mm drinking straw or a 2 ml disposable plastic pipette is introduced into the occipital foramen in the direction of an eye. Samples can be collected from the rachidian bulb, the base of the cerebellum, hippocampus, cortex, and medulla oblongata. Brain specimens from cattle can also be sampled using the 'brain scoop or tool'.

Retro-orbital route for brain sampling: In this technique (Montano Hirose *et al.*, 1991), a trocar is used to make a hole in the posterior wall of the eye socket, and a plastic pipette or straw is then introduced through this hole. The sampled parts of the brain are the same as in the former technique, but they are taken in the opposite direction.

b) Shipment of samples: Samples in 50% glycerol/PBS mixture should be kept refrigerated. As the virus is not inactivated by glycerol/PBS, all laboratory tests can be used on these samples. When it is not possible to send refrigerated samples, other preservation techniques may be used. The choice of the preservative is dependent on the tests to be used for diagnosis.

Formalin inactivates the virus, thus virus isolation tests cannot be used and diagnosis depends on using a modified direct fluorescent antibody test (FAT), polymerase chain reaction (PCR), (less sensitive than these tests on fresh tissue), immunohistochemistry or histology (Warner *et al.*, 1997).

An alternative for the transport of samples for molecular techniques is the use of FTA Gene Guard system (Picard-Meyer *et al.*, 2007). The FTA paper preserves rabies virus RNA within the fiber matrix allowing the transport of samples at ambient temperature without specific biohazard precautions for further characterization of rabies strains.

c) Laboratory tests

i) Immunochemical identification of rabies virus antigen

Fluorescent antibody test (FAT): The most widely used test for rabies diagnosis is the FAT, which is recommended by both WHO and OIE. This 'gold-standard' test may be used directly on a smear, and can also be used to confirm the presence of rabies antigen in cell culture or in brain tissue of mice that have been inoculated for diagnosis. The FAT gives reliable results on fresh specimens within a few hours in more than 95-99% of cases. The FAT is sensitive, specific and cheap. Aggregates of nucleocapsid protein are identified by specific fluorescence of bound conjugate.

Immunochemical tests: Immunoperoxidase methods can be used as an alternative to FAT with the same sensitivity (Lembo *et al.,* 2006), but attention should be paid to the risk of nonspecific false-positive results. Peroxidase conjugate may also be used on fresh brain tissue or sections of formalin-fixed tissue for immunohistochemical tests.

Enzyme-linked immunosorbent assay (ELISA): An ELISA that detects rabies antigen is a variation of the immunochemical test. It is useful for large epidemiological surveys (Xu *et al.,* 2007). The specificity and sensitivity of such tests for locally predominant virus variants should be checked before use. In case of human contact, these tests should be used in combination with confirmatory tests such as FAT or virus isolation.

Rapid immunodiagnostic test (RIDT): A rapid immunodiagnostic test was developed recently (Kang *et al.,* 2007). This simple test can be used under field conditions and in developing countries with limited diagnostic resources. Generally, tests other than the gold standard FAT should only be used after validation in multiple laboratories.

ii) Detection of the replication of rabies virus after inoculation: These tests detect the infectivity of a tissue suspension in cell cultures or in laboratory animals. They should be used if the FAT gives an uncertain result or when the FAT is negative in the case of known human exposure. Wherever possible, virus isolation on cell culture should be considered in preference to the mouse inoculation test (MIT). Cell culture tests are as sensitive as MIT (Rudd & Trimarchi, 1989) but are less expensive, give more rapid results and avoid the use of animals.

iii) Molecular techniques: Various molecular diagnostic tests, e.g. detection of viral RNA by reverse transcription PCR (RT-PCR), PCR-ELISA, hybridization *in situ* and real-time PCR are used as rapid and sensitive additional techniques for rabies diagnosis (Fooks *et al.,* 2009). The principle of *Lyssa* virus-specific PCRs is a reverse transcription of the target RNA (usually parts of the N gene) into complementary DNA followed by the amplification of the cDNA by PCR. Although those molecular tests have the highest level of sensitivity, their use is currently not recommended for routine post-mortem diagnosis of rabies (WHO Expert Committee on Rabies, 2005) due to high levels of false positive or false negative results without standardization and very stringent quality control. Nevertheless, they are useful for confirmatory diagnosis, as a first step in virus typing.

iv) Histological identification of characteristic cell lesions: Negri bodies correspond to the aggregation of viral proteins, but the classical staining techniques detect only an affinity of these structures for acidophilic stains. Techniques that stain sections of paraffin embedded brain tissues (e.g. Mann's technique) are time consuming, less sensitive and more expensive than FAT.

Seller's method on unfixed tissue smears has a very low sensitivity and is only suitable for perfectly fresh specimens. These methods are no longer recommended for routine diagnosis. Immunohistochemical tests are the only histological methods specific to rabies.

d) Other identification tests: The tests above describe methods to accurately diagnose rabies and to isolate and identify the virus. Typing of the virus can provide useful epidemiological information and should be undertaken in specialized laboratories (such as OIE or WHO Reference Laboratories). These techniques would include the use of MAbs, nucleic acid probes, or the PCR, followed by DNA sequencing of genomic areas for typing the virus (Bourhy *et al.,* 1993). These characterizations enable, for instance, a distinction to be made between vaccine virus and a field strain of virus, and possibly identify the geographical origin of the latter.

2. Serological tests: The main application of serology for classical rabies is to determine responses to vaccination, either in domestic animals prior to international travel, or in wildlife populations following oral immunization. In accordance with the WHO recommendations 0.5 IU per ml of rabies antibodies is the minimum measurable antibody titer considered to

represent a level of immunity in humans that correlates with the ability to protect against rabies infection. The same measure is used in dogs and cats to confirm a satisfactory response to vaccination. As neutralizing antibodies are considered a key component of the adaptive immune response against rabies virus (Hooper *et al.*, 1998) the gold standard tests are virus neutralization (VN) tests. However, indirect ELISAs have been developed that do not require high-containment facilities and produce rapid results.

Treatment:

No specific drug is available. thorough washing ofbite wound with soap and water, local application of 1% cetrimonium bromide, carbolic acid, silver nitrate and tincture of iodine, avoid suture of wound; muscle relaxant (scopalamide hydrobriomide), high doses of vitamin C, administration of rabies immunoglobulin and antirabies vaccine are helpful in the management of rabies.

Rabies Vaccine:

Human Vaccine: A good but expensive killed virus vaccine (Human Diploid Cell Vaccine, HDCV)grown in human fibroblasts or purified chick embryo cell vaccine (PCECV) is available for safe use in man. The unusually long incubation period of the virus permits the effective use of active immunization with vaccinepost-exposure (0, 3, 7, 14, 28 and 90th day). When used, vaccine has dramatically cut the rabies death rate.

Prophylaxis: High-risk persons, eg. Veterinarians may be immunized before exposure and then merely require one or two booster doses if they may be exposed to suspected cases of rabies.

***Animal Vaccines*:** A range of live and killed virus vaccines are available for domestic animals (farm animals, cats and dogs). In dog and cat, first vaccine is given at 3 months, the booster after 3 weeks followed by annual vaccination.

For animals, live and recombinant vaccines are effective by the oral route and can be distributed in baits in order to immunize wild (or domestic) animals.

Prevention & Control

1. Immediate treatment of wound caused by a scratch or bite of a rabid animal or wildanimal.
2. Compulsory registration and licensing of all pet dogs.
3. Collection and destruction of stray or unwanted dogs as it is the main reservoir in India.
4. Destruction of unvaccinated dogs bitten by known rabid animal.
5. Keep on leash on dogs in congested areas.
6. Preventive vaccination of all dogs with Raksharab (Indian Immunological) or Rabisan (Serum Institute.)
7. Imported dogs should have vaccination certificate and in the absence of which keep the animal under quarantine for six months.
8. Free of subsidized vaccination of all dog population.
9. Oral immunization of wildlife against rabies.
10. Provide free post bite vaccination to man and animals.
11. Prophylaxis vaccination of high risk groups like kennel staff. Veterinarian, dog catcher, cave explorer, hunter, animal handler and laboratory worker.
12. Submission of intact head of suspected rabid animal under refrigeration to a laboratory for the confirmation of rabies.
13. Person with skin lesions should not attend the rabies patient.

14. Keep the animal (who has bitten a man) under observation for 7-10 days.
15. Person should wear protective clothings (rubber glove, apron, gum boots) while attending a sick animal or cleaning saliva of the patient.
16. Intensification of mass education about the mode of transmission and prevention through media of radio, television, newspaper, and poster etc.
17. Reporting of rabies cases both in man and animals.
18. Close collaboration and coordination between veterinary and medical authorities at all level i.e. local, state and central.

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Advances in Diagnosis of Haemoprotozoan Diseases in Pet Animals

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Haemoparasitic infection are parasitic infection, where the parasite itself, or in stage of its development, circulates in the blood stream. Haemoparasites include the families Haemsporidia (*Plasmodium*, *Haemoproteus*, *Leucocytozoon*), Piroplasms (*Babesia*), Haemogregarine (*Hepatozoon*, *Haemogregarina*), Rickettsias (*Aegyptianella*, *Anaplasma*, *Ehrlichia*, *Neorickettsia*, *Rickettsia*, *Theileria*), Trypanosomes (*Trypanosoma*, *Leishmania*), *Mycoplasma*, *Haemobartonella*, *Dirofilaria* and *Eperythrozoon*. Diagnosis of these diseases often poses a serious problem. Identification of the parasite in the blood and other tissues of the infected pets is the conventional method used for the diagnosis of these diseases. In these methods characteristic staining reaction with certain dyes like Romanowsky's stains are frequently used for these purpose. Stained bodies in the tissues and cells measuring between 0.5 to 20mm in size help in the detection of the organism hence the result obtained are often presumptive. The most common technique of diagnosis of haemoprotozoan infection involves observing pear or ring-shaped and amoeba-like forms of the parasite in the erythrocytes of the blood, stained according to the Giemsa or Wright's method. Sometimes, in cases of low parasitaemia, infected cells can remain undetected. Moreover, the identification of different species of Babesia and Theileria, on the basis of morphology is not only difficult but also erroneous. The inoculation of laboratory pets with blood from suspected cases of babesiosis is time consuming because the first symptoms occur after 10 days. Carrier pets, in which low numbers of erythrocytes remain infected, are important contributors to the transmission of the infection by tick bite. Hence, detection of piroplasms in carrier pets is very important to control the infection. However, detection of piroplasms by microscopy is not easy and it is generally not possible to distinguish pathogenic species from nonpathogenic species that may occur simultaneously within the same host. This is specially true in low grade infection when only a few organisms are present in the circulation. It is therefore, a number of serological tests are used as a tool for diagnosis of these diseases. Serological tests, which are commonly used for the diagnosis of haemoprotozoan diseases are Fluorescent Antibody Test (FAT), Indirect Fluorescent Antibody Test (IFAT), Complement Fixation Test (CFT) etc. Serological tests like Radioimmuno Assay (RIA), and Enzyme Linked Immunosorbent Assay (ELISA) have become more popular because of their high sensitivity. Immunological and serological methods are characterized by their high specificity and sensitivity. Although serological tests can be used to detect circulating antibodies, cross-reactivity with antibodies directed against other species of piroplasms has been reported. Moreover, antibodies tend to disappear in long-term carriers, whereas piroplasms persist. Therefore, pets with a negative serological test can infect ticks and be the source of the infection for other pets. Another drawback with the serological tests is that the antibodies can still be detected years after recovery even though the parasite is not present in the circulation. Again a false positive result may occur with the serum sample obtained from such pets. Most of serological tests employ crude parasite antigen and /or polyclonal antiserums as a test reagent. Such reagents generally produce poor specificity and lack uniformity in results. It is therefore the traditional methods have been complemented or even ousted by the molecular ones. New developments in molecular biology have generated exciting possibilities for

improved diagnosis of parasitic diseases. Molecular diagnosis of haemoprotozoan diseases involves several PCR-based diagnosis procedures, which help in the identification of the parasite up to the species or even strain level. Though parasite antigens for sero-diagnosis can be produced in vitro gene cloning and expression and peptide synthesis, the nuclear hybridization techniques offer a vastly improved approach for identification of parasites in the tissue specimens of infected hosts as a means of diagnosis. Furthermore, the advent of the polymerase chain reaction technique has made it possible to increase the sensitivity of nuclear hybridization techniques, through amplification of target DNA sequences of the parasites in test material, by in situ synthesis of these sequences prior to hybridization with the diagnostic probe. Finally, through the use of monoclonal antibody technology, it is possible to design highly specific and sensitive serological assays, as well as assays for parasite antigen detection in tissue fluids and in the excreta of infected hosts, as a means of diagnosis. However, increased sensitivity and specificity can be achieved by combining PCR with a specific hybridisation by means of reverse line blot (RLB), a macroarray that is also capable of identifying mixed infections.

Polymerase Chain Reaction:

Polymerase Chain Reaction (PCR) is a laboratory technique evolved by Dr. Kary B. Mullis, in the year 1985 for which he has been awarded Nobel Prize in the year 1993. PCR's popularity comes from its ability to specifically amplify a target DNA sequence more than a million fold within several hours. Furthermore, PCR is so sensitive that a single DNA molecule can be amplified out of complex mixtures of genomic sequences and visualized as a distinct band on an agarose gel. The PCR is an in vitro method for enzymatic synthesis of specific DNA sequences, using oligonucleotide primers that hybridize to opposite strands that flank the region of interest in the target DNA. A repetitive series of cycles involving template denaturation, primer annealing, and the extension of a annealed primers by DNA polymerase results in the exponential accumulation of specific fragments. These newly synthesized fragments can then serve as template in the next cycle, approximately doubling the number of target DNA sequences in every cycle. Theoretically after every 20 cycles of PCR about a millionfold (220) amplification occurs. In a simplified way it can be said that PCR merely involves combining a DNA sample with oligonucleotide primers, deoxynucleotide triphosphates and thermostable Taq polymerase in a suitable buffer, then repeatedly heating and cooling the mixture for several hours until a desired amount of amplification is achieved. The PCR product then can be analysed by several methods, such as Dot blots, Southern hybridization, and Gel electrophoresis.

Polymerase Chain Reaction (PCR) or PCR Restriction Fragment Length Polymorphism (PCRRFLP) is the most promising diagnostic tool and one of the most reliable tools for the detection and identification of protozoan parasites but for the efficacy of the test, selection of appropriate genetic markers of the parasite DNA is extremely important. Different molecular targets were tested for PCR and genes encoding the rRNA of the small ribosomal subunit were found to be the most useful. Within the ribosome, regions conserved in evolution can be distinguished, i.e. having the nucleotide sequences similar to the majority or all parasite species and to others closely related to them. Such an organisation of the gene enables the design of primers complementary to conserved sites for PCR and which can detect a large group of related organisms. In case of Babesia, the gene encoding the E-tubulin protein is being widely used as molecular marker for the accurate identification of the

parasite. There are two introns within this gene; the first one exhibits much variability with regard to length as well as to the nucleotide sequence. Therefore, the PCR products are of varied lengths depending on the Babesia species. However, these differences are too small for the identification of some species and so, confirmatory methods that extend the duration of the diagnosis are essential. The other genes whose sequences can be used as molecular targets for the detection and differentiation of Babesia species are genes encoding the Heat Shock Proteins, in particular, hsp70. However, the hsp70 gene is largely conserved in its nucleotide sequence even between non-related organisms. Therefore, this method, based on the amplification of the whole genome or its fragments, applies mainly to molecular phylogenetic analysis. In conclusion, the selection of a genetic marker for PCR is very important for the sensitivity of this technique.

Real – Time PCR

Real-time PCR which was developed in early 1990 is a type of quantitative PCR which measures the amount of cDNA or mRNA in a sample, either from a population of cells (tissue or cell culture), or recently even from a single cell. The main advantage of the real-time PCR over conventional PCR is that it allows high throughput analysis in a close tube format and does not require handling after the amplification. The principle of the Real-time PCR is to incorporate a specific, intercalating dye (e.g: ethidium bromide) into the PCR to measure the change in fluorescence after each cycle using a digital camera and a fluorometer attached to the reaction tube. Real-time is used commonly to determine the expression of a gene's mRNA, and its expression levels (copy number of mRNA) during certain conditions such as treating cells with a drug. Real-time PCR can be used to compare normal (control) samples to disease samples, giving an idea as to expression changes, which occur with pathogenesis. Real-time PCR due to its sensitivity is also used in the detection of parasite in the blood. The technique has been successfully used in the diagnosis of Cryptosporidium, Leishmania and Trypanosoma parasites successfully.

Microchip Electrophoresis

The application of microfabrication technology to microchip electrophoresis (ME) has been increasing in the interdisciplinary field in analytical chemistry. ME separation is significantly faster than conventional gel electrophoresis, and is usually completed in 10 to 200 seconds and consumes only a few microliters of reagents. They suggested that a combination method using whole blood PCR and ME for the diagnosis of haemoprotozoan diseases would be a very simple and ultrafast methodology for use in a clinical diagnostic laboratory. King *et.al.*, (2005) evaluated a novel strategy for fast diagnosis by microchip electrophoresis (ME), using programmed field strength gradients (PFSG) in a conventional glass double-T microfluidic chip. The ME-PFSG allows for the ultrafast separation and enhanced resolving power for target DNA fragments. These results are based on electric field strength gradients (FSG) that use an ME separation step in a sieving gel matrix (polyethylene oxide). The gradient can develop programmed shapes FSG over the time. The PFSG method could be easily used to increase separation efficiency and resolution in ME separation of specific size DNA fragments. Compared to ME that uses a conventional and constantly applied electric field (isoelectrostatic) method, the ME-PFSG achieved about 15-fold faster analysis time during the separation of 100 bp DNA ladder. The ME-PFSG was also applied to

the fast analysis of the PCR products, 591 and 1191 bp DNA fragments from the 18S rRNA of Babesia gibsoni and Babesia caballi.

Reverse Line Blot macroarray (RLB)

Sanmartin et.al (2006) for the first time reported the detection of Babesia using Reverse line blot macroarray techniques in subclinical and carrier pets. The technique allowed the simultaneous detection and identification of different Babesia species using oligonucleotide probes whose specificity has been previously determined. Moreover, the combination of a generic Babesia PCR targeting the V4 region of the 18S rRNA gene and a hybridization with specific probes provided high sensitivity. Since detection of the parasite in Giemsa-stained blood smears is the technique that has been traditionally used for diagnosis of piroplasmosis, whenever possible RLB and microscopy examination were performed in parallel. Besides, positive microscopy allowed us to identify pets with parasitaemia.

Critical care and its management in Pet animals

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CRITICAL CARE OF THE DOG

COMMON CAUSES OF CRITICAL ILLNESS IN DOGS

There are a number of different causes that may lead to critical illness in patients. However, the supportive treatment and the nutritional support may be very similar for these patients.

- Trauma
- Neoplasia
- Anemia
    - Hemolysis
    - Hemorrhage
    - Bone marrow failure
- Organ failure
    - Heart
    - Liver
    - Kidneys
    - Lungs
- Endocrinopathies
    - Diabetic ketoacidosis
    - Hypoadrenocorticism
- Sepsis/Infection
- CNS disease
- Toxins
- Immune-mediated disease

BENEFITS OF NUTRITIONAL SUPPORT

The therapeutic benefits of nutritional support in critically ill patients are well established:

- Decreased morbidity and mortality
- Improved tolerance to invasive procedures
- Shorter hospitalization periods
- Decreased incidence of infections
- Earlier ambulation
- Rapid wound healing
- Fewer complications

RESPONSE TO INJURY

Critically ill dogs, which are unable or unwilling to eat, must rely on their endogenous stores to supply energy and protein for recovery.

Glycogen reserves in the liver are mobilized but are quickly depleted within the first 12 to 24 hours. Thereafter glucose must be synthesized from lactate, glycerol, and amino acids to provide fuel for those obligate tissues, which require glucose for energy such as the brain and kidney.

Lipids are mobilized from adipose stores and metabolized to ketones, which may be used as an energy source in some tissues.

There are no storage forms of protein in the body and therefore, endogenous muscle proteins are catabolized to energy. It is this subsequent negative nitrogen balance that can lead to proteincalorie malnutrition and is most difficult for the critical patient. The demand for new proteins in healing and replacement tissues is high and a lack of amino acids results in delayed healing and decreased production of defense proteins such as immunoglobulins, clotting factors, and acute phase reactants.

Furthermore, these adaptive processes in the critically ill patient are not as efficient due to excess stress hormones, such as glucocorticoids, catecholamines, and vasoactive substances such as cytokines, and other inflammatory mediators.

Providing an animal with nutritional support may soften this hypercatabolic response to injury and preserve endogenous tissue resources. And in chronically ill patients with longstanding malnutrition, nutritional support may be vital to recovery and survival.

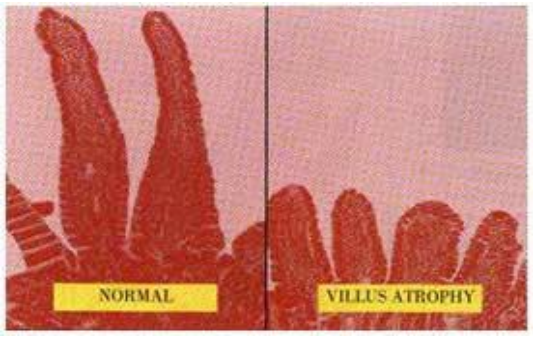
CLINICAL CONSEQUENCES OF PROTEIN-CALORIEMALNUTRITION

Prolonged protein-calorie malnutrition affects every major organ system, and this process is further accelerated in the hypermetabolic patient. Failure to provide nutritional supplementation to critically ill dogs may lead to the induction of multiple organ failure, and can be directly related to morbidity and mortality. The consequences of poor nutritional status may affect individual organ systems or conditions, including:

- • Gastrointestinal tract
- • Muscle
- • Liver
- • Kidneys
- • Cardiopulmonary tract
- • Immune system
- • General recovery

Gastrointestinal Tract

The amino acid glutamine is the primary energy source of rapidly dividing cells, including enterocytes. A deficiency of glutamine results in small intestinal villus atrophy and a compromise in the mucosal barrier, which may ultimately lead to bacterial translocation and sepsis. Brush border enzymes, which aid in digestion, down-regulate their own activity, making the digestive process much less efficient.



Muscle

Protein stores are mobilized, reducing muscle strength and mass, resulting not only in weight loss and weakness of the skeletal muscles, but also in deterioration of smooth and cardiac muscles.

Liver

Impaired function of the reticuloendothelial system allows bacteria in the portal system access to general circulation, thus increasing the risk of sepsis; this is particularly dangerous in combination with decreased antibody production due to protein malnutrition. Glycogen stores become depleted, as do the amino acid building blocks necessary to meet the protein demands of the stressed patient, which may lead to hypoproteinemia, decreased wound healing, third-spacing of fluid, and edema.

Immune System

Decreased antibody production, impaired chemotaxis, phagocytosis, and oxidative/respiratory burst of white blood cells further increase the risk of infections and sepsis. Death may result despite the use of powerful and expensive antibiotics.

Kidneys

Patients may develop a diminished capacity for renal gluconeogenesis, amino acid metabolism, and regulation of acid-base balance.

Cardiopulmonary Tract

Critically ill, malnourished dogs may show compromised gas exchange and acid-base balance, decreased metabolism of hormones and surfactant.

General Recovery

These patients may show increased susceptibility to shock, delayed wound, and fracture healing, increased incidence of dehiscence and visceral wound leakage, and diminished tolerance to hospital stress. Recovery may therefore be delayed.

PATIENT SELECTION

In human medicine, multiple parameters are used to assess the patient's need for nutritional support including anthropomorphic measurements, clinical nutrition history, and analytical laboratory measurements. In veterinary medicine, there are fewer practical and less reliable means of determining which patients require nutritional support. There is no "gold standard" test; however, a combination of the following subjective and objective means of assessment are often helpful to identify those patients in need of nutritional support.

Subjective assessment: history and physical examination

- • Previous history of illness or weight loss
- • Current poor body condition or acute loss of >5% body weight
- • Dogs that have been anorexic or inappetent for >3 days (real or anticipated)
- • Severity and nature of injuries which prevent adequate oral intake:
    - ✓ Facial injuries
    - ✓ Prolonged or unmanaged pain
    - ✓ Injuries requiring surgical correction
    - ✓ Conditions of excessive protein loss (peritoneal drainage; open, discharging skin wounds; liver or renal failure; protein-losing nephropathy or enteropathy)

Objective assessments: laboratory results

• Lymphopenia
• Low serum albumin; hypoalbuminemia
• Low creatine kinase; nonspecific increase in the absence of muscular disease or damage
• Decline in total iron binding capacity (TIBC)
• Low transferrin
• Serum insulin-like growth factor 1 (IGF-1)

NUTRITIONAL REQUIREMENTS

The nutrient requirements of critical patients have not been clearly determined, however, normal dietary constituents should be adjusted to suit the animals' disease requirements including energy, fat, carbohydrate, and protein. Additionally, there are many other essential nutrients including select amino acids, minerals, and vitamins, but it is yet not fully determined how critical illness will affect these micronutrient requirements. A number of specific nutrients have properties that may be beneficial in the nutritional management of critical care patients, including glutamine, arginine, branched chain amino acids, B vitamins, and zinc.

ENERGYEEVergyEnergyEnergye

The calculation of energy requirements of critically ill patients is difficult to establish and has therefore been subject of some controversy. As it is usually not possible to measure

the patient's energy consumption directly, equations have been established to estimate the requirement. Recommendations have been made using either resting energy requirements (RER), basal energy requirements (BER), or maintenance energy requirements (MER). Basal energy requirements describe the energy that is needed to keep the body "ticking over," i.e., the energy to meet the needs of cells and organs under certain set conditions, such as a thermoneutral environment, no stress, and 12 hour dietary rest).

The resting energy requirements (RER) involve the energy required by the animal in a resting state and account for physiologic influences and the assimilation of nutrients.

The maintenance energy requirements (MER) encompass all the energy required for maintaining normal body condition in a normal pet.

BER / RER= 70 x W 0.75 kcal/day

MER = 110 x W 0.75 kcal/day

Many authors recommend using either RER or MER and multiplying these with an illness factor to account for extra hypermetabolic requirements. It has been suggested that critically ill patients have requirements ranging from 0.5-1.5, possibly 2.0 x RER/MER.

Other authors suggest that the RER of critical patients, determined with indirect calorimetry, indicates that their energy expenditure is only slightly increased from normal.

Additionally, feeding excessive calories may have a number of negative effects, such as gastrointestinal problems, electrolyte imbalances and hepatic dysfunction and it is generally recommended to avoid overfeeding and associated complications. The practical recommendation is to ensure that all patients that require nutritional support are fed at a minimum level of their RER. Close monitoring of the patient's body weight and body condition can then be used to help to adjust the calorie intake for each individual patient, and this may mean increasing RER as discussed above.Calories are provided by a balance of fat, protein, and carbohydrates.

FAT

High fat diets have been recommended in the feeding of critical patients because triglycerides rather than glucose provides the principal fuel for increased metabolism in the catabolic patient. Fat provides more than twice the energy density per unit weight than protein or carbohydrates, and increasing the fat level will help to make a diet more concentrated. In the stressed or traumatized patient. the administration of reduced volumes of highly energy dense foods may be critical, as feeding lower volumes means less stress to the patient.High fat diets also tend to be more palatable and digestible, and patients are more likely to begin eating sooner on their own.

CARBOHYDRATESarbohydrates

There is no individual requirement for carbohydrates other than as an alternate and readily available source of energy other than fat. Supplementation with carbohydrates helps to preserve endogenous protein from breakdown and subsequent conversion to glucose via gluconeogenesis. Oversupplementation of simple carbohydrates such as glucose, however, can predispose patients to hyperglycemia. The resultant excessive release of insulin can lead to hypophosphatemia, hypokalemia, and other metabolic derangements. Complex indigestible carbohydrates such as fiber are rarely included in diets formulated for critical care, as fiber may increase the feeding volume, reduce the overall digestibility and decrease the availability of essential nutrients, such as zinc.

PROTEIN

Protein requirements can increase significantly in the critical patient. The amount of protein required reflects the number of amino acids needed for protein synthesis. This is determined by the amino acids required to replace those that have been degraded in the catabolic process, those lost due to trauma or injury, and those required for the build-up of new body tissue.To abolish negative nitrogen balance in a severely hypermetabolic and

hypercatabolic patient, it is necessary to supply protein in amounts considerably in excess of normal minimum requirements, however, because of risks associated with overfeeding hospitalized patients, a more moderate approach is recommended. For enteral feeding in dogs, protein should comprise at least 20-30% of calories (2 to 3 g/kg BW). To maintain wound healing and immune function in severe protein-losing disease conditions such as extensive burns or peritoneal drainage, as much as 25-48% of the metabolizable energy (ME) may be needed in form of protein (Wills and Simpson 1994). Dietary restriction of protein, however, may be indicated in individuals with certain concurrent conditions such as portosystemic shunts, chronic kidney or liver disease; in these critically ill patients, protein intake needs to be carefully balanced.

The dietary source of protein should be highly digestible and contain all the essential amino acids, such as those found in egg and milk proteins.

Essential Amino Acid Requirements for Maintenance

Amino Acid	*Dog*[a]	*Cat*
Arginine	68	478[b]
Histidine	71	ND
Isoleucine	155	ND
Leucine	271	ND
Lysine	161	158[c]
Methionine and cystine	97	155[c]
Phenylalanine and tyrosine	277	ND
Threonine	142	ND
Tryptophan	42	ND
Valine	193	ND

ND = No research data.
[a]Data from NRC (1985).
[b]Data from NCR (1986) for the near-adult cat.
[c]Data from Burger and Smith (1990).

BRANCHED CHAIN AMINO ACIDranc Branched Chain Amino Ached Chain A

Branched chain amino acids (BCAAs) are thought to have a beneficial effect on nitrogen balance. Studies have documented an increase in nitrogen retention and hepatic protein synthesis when BCAAs are supplemented in the traumatized or stressed patient. A decrease in the ratio of BCAAs to aromatic amino acids has been implicated in the development of encephalopathy in septic patients but not proven. A definitive need therefore for BCAA supplementation has yet to be defined in the critically ill patient.

GLUTAMINE

Glutamine is a ubiquitous amino acid, which is found in abundant quantities in blood and other tissues. This amino acid has multiple functions including:

- • Playing a role in acid-base balance
- • Being a precursor of purine and pyrimidine nucleotides
- • Playing a role in detoxification
- • As a nitrogen carrier between tissues
- • As a regulator of hepatic protein synthesis

- • As a respiratory fuel in certain tissues

The role of glutamine as an important and major substrate in rapidly dividing cells, such as those of the gastrointestinal tract and the immune system (lymphocytes, macrophages, and thymocytes), is well known. Glutamine is responsible for maintaining the IgA-secreting cells of the gut mucosa, and an adequate supply is therefore required to ensure the integrity of the intestinal mucosal barrier.

Glutamine is a nonessential amino acid in the dog; however, during stress or trauma, the synthesis of glutamine is not sufficient to match the increase in uptake and metabolism by the gastrointestinal tract. Glutamine has therefore been described as a "conditionally essential amino acid" (Lacey and Wilmore 1990, Mobrahan 1992). This increased demand and concurrent poor supply in trauma patients may result in a compromise of the gut mucosal barrier, with subsequent bacterial translocation and systemic infection (Souba et al. 1990).
Specific recommendations for levels of this amino acid in critical patients are lacking; however, the benefits of supplementation have been demonstrated (Souba et al. 1990). Milk protein is a rich source of glutamine.

ARGININE

Arginine is an essential amino acid in the dog, but not in humans. Arginine has been shown to enhance cellular immunity, wound healing, and nitrogen balance (Barbul 1986). There are no specific indications for this amino acid in the traumatized or stressed dog; however, as requirements for arginine may be increased in the critically ill patient, a level of arginine to support normal growth is recommended in diets intended for these patients.

B VITAMINS

Although specific levels have not been determined for the critically ill dog, water-soluble Bcomplex vitamins may have an increased requirement in the hypermetabolic patient due to fluid losses and increased energy expenditure. B vitamins are not stored in the body; subsequently. they may be easily diluted and depleted in the anorexic or inappetent critical patient. Fortunately, they are easily replaced in fluids or in well-balanced enteral or parenteral formulas.

Thiamin

Vitamin	*Function*	*Imbalance Causes*
Thiamin (Vitamin B1)	Involved in carbohydrate metabolism. Requirement is dependent on the carbohydrate content of the diet.	**Deficiency** - Anorexia, neurologic disorders (especially of the postural mechanisms) followed ultimately by weakness, heart failure, and death. Can occur, especially in cats, as a result of feeding large amounts of certain types of raw fish, which contain the enzyme thiaminase. In addition, the vitamin is progressively destroyed by high temperatures and under certain conditions of processing. **Toxicity** - Low toxicity.

ZINC

Zinc may be important to the hospitalized patient by virtue of its role in protein and nucleic acid metabolism and in promoting wound healing. Zinc deficiency can result in impaired protein synthesis, increased protein catabolism, depressed wound healing and depressed immune function. Although it is difficult to establish zinc status from circulatory levels, a decrease in plasma zinc level is observed following injury (McClain et al. 1986). This has been attributed to a combination of increased urinary zinc loss and tissue

redistribution of zinc. It has therefore been suggested that zinc supplementation may be beneficial in the critical canine patient.

METHODS OF SUPPLEMENTATION

Anorexia and inappetence are common in the critical patient. Normal voluntary feeding is the preferred means of supplementation, as it is the least stressful method for both patient and caregiver. Initial attempts to increase palatability of the food may include providing small, frequent meals of wet, warmed, odiferous foods. Assisted feeding may be attempted by gently syringing a liquid food into the corner of the patient's mouth. It is, however, important not to stress the patient excessively while force-feeding.

Appetite stimulants may be used initially to help "jumpstart" the appetite; however, if the dog refuses these efforts, nutritional support may be supplemented via the gastrointestinal tract(enteral feeding) or by intravenous infusion of an energy-dense solution (parenteral feeding).

Appetite Stimulants

Drug	*Dose*	*Side Effects*
Diazepam	1-2mg IV, IM or PO per cat	Sedation, idiosyncratic liver necrosis
Oxazepam	5-10 mg PO BID per cat	Same as diazepam
Cyproheptadine	8 mg/m^2 or 2-4 mg/cat PO 5-20 mg/dog PO	Excitability, aggression, vomiting
Steroids		
Nandrolone	5 mg/kg IM weekly (dogs)	Uncommon
Stanozolol	1-2 mg PO BID (cats) 1-4 mg PO BID (dogs	Uncommon
Prednisolone	0.5-1 mg/kg (cats & dogs)	Polyuria/polydipsia, decreased wound healing, may interfere with therapy for disease

Enteral Feeding

Enteral feeding is considered more physiologically sound than intravenous feeding, as it maintains the health of the gastrointestinal tract; therefore, if the gut works, use it! The intact intestinal mucosa acts as an important barrier to bacteria, and it is therefore important to maintain the health of the gastrointestinal lining by supplying nutrients enterally. When the gut is starved, bacteria can translocate from the intestine into the circulation, leading to sepsis; therefore even patients receiving parenteral nutritional support may benefit from enteral feeding.

Routes of Enteral Nutritional Support

Method	*Advantage*	*Disadvantage*
Assisted feeding	Simple, less stressful	Not effective in many patients
Chemical stimulants	Simple, "reminds" patient of the taste of foods	May induce sedation Short term (2-3 days)
Nasoesophageal tube (3.5-5 Fr, cat) (3.5-8 Fr, dog)	Easy to place, least invasive Minimal sedation required Use up to one week Low cost	Not always well tolerated Must use an Elizabethan collar
Pharyngostomy tube (12-18 Fr, dog)	No special equipment required Can be used long term	Requires general anesthesia Malpositioning may lead to aspiration

Enteral feeding is most often accomplished by use of feeding tubes, which are ideally placed as proximally as the animal's clinical state and temperament will permit. These include nasooesophageal, pharyngostomy, esophagostomy, gastrostomy and enterostomy feeding tubes. The utilization and care of these tubes is usually straightforward with few complications.

Nasoesophageal Tube Feeding

This is an excellent option for short-term feeding (<14 days) of hospitalized patients. Most patients will tolerate this kind of tube very well, provided it is protected by an Elizabethan collar. Placing a nasoesophageal feeding tube involves choosing a tube that will fit snugly into the ventral meatus and passing it to the level of the lower esophageal sphincter. Most critically ill patients will tolerate tube placement, but some individuals may require sedation.

The tube is secured in place with sutures at the nares with one or two others as needed on the face and head. Tubes passed into the stomach may allow for reflux of acid juices, causing esophagitis and contributing to vomiting and irritation; it is therefore recommended to place the tube end into the esophagus

Dog with nasoesophageal tube wearing an Elizabethan collar

Contraindications for the use of nasooesophageal tubes include patients that have had severe facial trauma involving the nares, those that are already experiencing protracted vomiting and/or regurgitation, animals that are semiconscious, or those patients that have laryngeal, pharyngeal, or esophageal physical or functional abnormalities.

Pharyngostomy Tube Feeding

Pharyngostomy tubes are placed into the pharynx under a general anesthetic and threaded down into the esophagus. These should not be used in smaller dogs (<10 kg BW) due to the limitations of space and potential interference with laryngeal function. With the advent and simplicity of gastrostomy and esophagostomy tube placement, pharyngostomy tubes are used less frequently.

Esophagostomy Tube Feeding

These are large bore tubes that can be easily placed under a light anesthetic with minimal equipment requirements in dogs of all sizes. The only major associated complication is the potential for infection at the entry site and meticulous care of the surgical wound is essential to maintain the tube.

Gastrostomy Tube Feeding

Gastrostomy tubes have become invaluable for the long-term nutritional support of critically ill patients. Gastric feeding tubes may be placed surgically, endoscopically or by a "blind" percutaneous technique. As with other ostomy tubes, they must remain in place for a minimum of 7-10 days to allow a seal to form with the abdominal wall. These tubes may be easily maintained for many weeks to months in the chronically ill or anorexic patient. Most patients tolerate the tubes quite well as long as the site of entry is adequately wrapped to prevent the wound from becoming infected, and Elizabethan collars are fitted to prevent the dog from inadvertently removing the tube prematurely.

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Enterostomy Tube Feeding

The placement of feeding tubes beyond the stomach is rarely indicated, but in cases of pancreatitis, diffuse gastric mucosal disease, protracted vomiting or delayed gastric emptying, an enterostomy tube may be life-saving. Enterostomy tubes are most commonly placed surgically, although they may be alternatively introduced via a gastric tube and then directed down through the pylorus with an endoscope.

Contraindications include ascites, peritonitis, immunosuppression and distal small bowel obstructions. Feeding through an enterostomy tube must be facilitated by a continuous infusion pump due to the narrow diameter of the tube and the volume necessary to meet energy demands.

The Care and Use of Enteral Feeding Tubes

- • Once in place, the accurate placement of feeding tube should be checked (e.g., by flushing the tube with sterile water or taking a radiograph.
- • Gastrostomy and enterostomy tubes should not be used for the first 24 hours to allow a seal to form with the body wall.
- • Liquid concentration diets may be used for all sizes of feeding tubes; with very small tubes (>8 Fr), diets may have to be blenderized further with water to allow passage.

- • One-third of the calculated energy requirements is divided into several (4-6) small feedings (5-20 ml, depending upon the size of the dog) and administered through the tube on the first day.
- • The food is warmed to body temperature and injected slowly over several minutes; if the patient begins to retch or swallow, slow down the rate of administration.
- • The tube must be flushed with 3-5 ml of water before and after each feeding to clear debris and maintain tube patency; should the tube become blocked, a cola product or enzymatic solution may be incubated in the tube overnight to help clear organic debris; if the tube remains blocked, it should be replaced.
- • The volume of feedings are increased by one-third each day until the patient is able to tolerate its full caloric intake; once the patient has been on full feedings for several days the volume of each feeding may be increased, and the frequency of meals decreased.
- • For gastrostomy tube feedings, the tube should be aspirated prior to each feeding to ensure that the stomach has emptied; if gastric emptying is delayed, and more than half of the previous meal persists, skip the next feeding.
- • Gastrostomy and enterostomy tubes must be wrapped, the wounds cleaned and bandages changed every two to three days as necessary.
- • Elizabethan collars should be used to prevent inadvertent removal of the tubes.
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Parenteral Feeding

Parenteral feeding is the administration of essential nutrients (fat, carbohydrates, and protein, vitamins and minerals, and water) by continuous intravenous infusion. This is also called total parenteral nutrition (TPN). TPN should be used only when enteral nutrition is not possible due to severe gastrointestinal dysfunction, since its application can be fraught with potentially fatal complications, and the feeding solutions can be both expensive and difficult to obtain. When solutions designed for use in humans are applied, supplementation with essential amino acids such as arginine is necessary; also, protein levels ought to be adjusted to canine requirements

Potential Complications arising from TPN

Sepsis

The nutrient-rich solutions provide an ideal growth media for contaminating bacteria. Therefore, to prevent transferring infections, these solutions must be mixed and administered under sterile conditions through a dedicated catheter, preferably in the jugular vein, which is cared for meticulously.

Additionally, there may be an increased risk of bacterial translocation and subsequent sepsis from the gastrointestinal tract. The intact intestinal mucosa acts as an important barrier to bacteria, and when the gut is starved, bacteria can translocate from the intestine into the circulation; therefore, even patients receiving parenteral nutritional support may benefit from concurrent enteral feeding (Wills and Simpson 1994).

Metabolic Derangements

The nutrients in the solution are concentrated to provide as much dextrose, lipids, and amino acids as necessary to maintain an adequate nutritional plane. However, the patient may not be able to quickly assimilate these nutrients, leading to problems such as hyperglycemia, glucosuria, hypo- or hyperkalemia, and lipemia which may necessitate adjusting the nutrient ratios, slowing the rate of infusion, or administering insulin or potassium supplements.

Monitoring

Routine physical examinations including body temperature, heart rate, respiratory rate, twice daily weight measurements, assessment of hydration status, and attitude should be performed on all critically ill patients receiving nutritional support.

Laboratory values frequently assessed include total proteins, albumin, packed cell volume, blood glucose, blood urea nitrogen, and urine specific gravity. Alterations in these parameters may indicate improved patient status due to correction and support of the underlying disease or, conversely, may signal early warnings of complications associated with nutritional supporting tubes

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