# The Dog as an Experimental Animal

Mary Bate Animal Welfare Officer University of Newcastle NSW, Australia 2308

# ANZCCART Facts Sheet

ANZCCART PO Box 19 Glen Osmond SA 5064 Australia Tel: 08–8303 7393 Fax: 08–8303 7113 E-mail: anzccart@waite.adelaide.edu.au

## Introduction

The domestic dog (*Canis familiaris*) is a species with a large number of breeds rather than genetically defined strains. There is a marked difference between breeds in their size, appearance, and life expectancy (e.g. the Chihuahua at one kg, St. Bernard at 90 kg). The dog has been used as a model for many human conditions in areas such as cardiovascular research, diabetes mellitus, ulcerative colitis, open heart surgery, organ transplantation, pharmacology and toxicology. This paper is intended as a guide for animal technicians and researchers involved in the care and use of the dog as an experimental animal.

## Sources

Dogs used for research or teaching may be purpose-bred (e.g., beagle). A second source may be city pounds which allow the release of stray, unidentified animals to research and teaching institutions. Regardless of the source, researchers should ensure careful selection of an animal for suitability to the experimental situation. This selection should consider the dog's temperament, health status, and physical characteristics. For example, highly excitable breeds (e.g. red setters, dalmations and German short-haired pointers), may be unsuitable for long-term experimental conditions. Likewise, long-haired breeds (e.g. afghans, saluki) may be unsuitable for cardiovascular research.

When using animals obtained from city pounds, researchers must be fully aware of the emotive issues associated with this source of dogs, and of local legislative requirements regarding the procurement, holding periods, identification and records related to these animals. All animals should be quarantined on arrival at the animal unit unless they are bought from a source of known reliability, and are of known health status.

# Housing

## Caging

Dogs are gregarious, social animals and live well in a kennel environment with a defined social order. Because of their social needs with respect to other members of their species, housing should provide an opportunity for each animal to at least see and smell other dogs, especially if no contact is possible. Dogs may be housed in indoor pens, with or without outdoor attached runs. The use of cages should be limited to short periods only. Dogs are best housed in pairs or in groups. Single housing may be necessary if the dogs are incompatible or because of the demands of an experiment (e.g., animals recovering from surgery, or with exteriorized cannulae). Where not used for breeding, females in oestrus should be housed away from males. There is considerable variation in the recommended space allowances for housing dogs in pens and runs(Table I). The *Australian Code of Practice* (NHMRC, 1990) states that pens and cages should be designed to ensure the comfort and well-being of the animals. The size and suitability of allotted housing is determined by the size and activity of the individual dog and whether an exercise run is available.

Materials used in dog facilities should be smooth and resistant to corrosion, denting, cracking or chipping. Indoor caging should provide a comfortable place for the animal to lie down and enough room for the animal to defecate away from the sleeping area. Bedding should be warm and dry, preferably raised from the floor, and composed of material which is resistant to chewing. Trampoline beds are recommended, with the size dependent upon the size of the dog. The use of cages with wire floors results in the animal's weight being borne on a relatively small area of the footpad and is therefore not recommended. Outside housing should provide a dry sleeping area, some shade and shelter from wind and rain, and should be well drained.

## Environmental enrichment

Dogs housed in restricted environments are likely to develop abnormal stereotypic behaviour (e.g. repetitive aimless movements). Because of the dog's naturally sociable nature, contact with other dogs and with people is important for their general well-being. Frequent handling, especially during the critical period between six and eight weeks of age, ensures that puppies become accustomed to human company as they mature. Likewise, purchased animals should be given similar attention during their period of quarantine and conditioning. All animals should receive regular contact, at least daily, with the animal attendant and/or researchers to ensure that they become adapted to the experimental conditions and are amenable to handling. Attempts to vary their environment may include the provision of outside runs (so that they can "watch the world go by"), provision of objects for chewing (large uncooked bones, toys which can be safely chewed or which contain food treats) and socialisation with other dogs and with Where an outside run is not available, an people. opportunity should be provided daily for dogs to leave their normal cage. The NHMRC (1996) recommends periods of at least 30 minutes each day, even in bad weather, when they are taken outside to run freely or taken for a walk on a leash.

It is important to remember that there are wide withinbreed and within-sex differences in behavioural needs. Hence, it is essential that animal attendants and researchers know the individual animal so that its needs can be met.

## Environmental conditions

While dogs are extremely adaptable to external temperature, it is important that housing be designed to avoid extreme

fluctuations in environmental conditions. In general, for inside housing, the conditions recommended for a range of laboratory animals are also suitable for dogs.

Temperature:	18 -21° C
Air Changes:	8 - 12 per hour
Lighting:	12 hour light/dark cycle.
Humidity:	35 - 70%

High humidity may be a problem in indoor housing when floors are cleaned by hosing down with water. There is no evidence that either light intensity or duration affects the breeding cycle of the bitch (MacArthur, 1987). Attention must be given to reducing noise that may be within the dog's auditory range.

## Cleaning

The frequency of cleaning cages will largely depend on their design. All faeces should be removed from both indoor and outdoor housing at least once daily. Since eating stimulates defaecation, it may be helpful to provide meals prior to cleaning. For puppies and younger dogs, cages may need to be cleaned twice or three times daily.

#### Identification

An excellent method of identification for dogs is a Microchip implant (e.g. Trovan®), inserted beneath the skin between the shoulder blades. Other methods include the use of collars, identification of characteristic markings of individual animals and the application of an ear tattoo (which must be performed using anaesthesia or heavy sedation). Clipping of hair or using colour marking with non-toxic solutions may be used as temporary identification.

## Handling and physical restraint

A minimum of physical restraint should be necessary in laboratory dogs if they have been selected for temperament and acclimatised to the experimental situation. Every effort should be made to accustom the dogs to their handlers and surroundings before experiments commence. For most common procedures, it is sufficient for the handler to hold the animal close to their body. Ensuring that the back of the dog is close to a wall or a corner may prevent the animal from "backing away" from the operator during the procedure. When the safety of the handler is in question, a soft bandage muzzle should be applied around the nose of the dog and tied behind its head . Signs which may indicate the necessity for a muzzle include tenseness, growling, or a "wide-eyed" or anxious facial expression.

## **Common procedures**

## Recording body temperature

The body temperature of the dog is usually recorded using a rectal thermometer (mercury or digital). In the anaesthetised animal, core temperature is measured using a rectal or oesophageal thermistor. The normal range is 39.0  $\pm$  0.5<sup>o</sup> C and may be influenced by disease, ambient temperature, exercise, and individual variation.

## Oral dosing

For the administration of tablets, (for a right-handed person) the head is elevated by placing the left hand over the muzzle such that the tips of the thumb and second finger rest in contact with the lips immediately behind the canine teeth.

With the tablet held between the thumb and second finger of the right hand, the mouth is then opened with the index finger of that hand placed against the lower incisors. Place the tablet at the back of the tongue, close the mouth and hold the dog's head tilted upwards for several seconds or until you can see swallowing movements. If medication is to be administered with food, it is best to mix it with only a small amount of the meal. Ensure that the dog eats all of this food before giving the rest of the meal.

Fluid can be given by syringe directly into the mouth with the dog's head tilted upward. Unpalatable fluids or large volumes of material may have to be administered by a stomach tube. This procedure is usually accepted by the conscious dog if it is done properly. Open the mouth as described previously. To ensure that the dog does not bite the tube, a gag may be placed into the mouth. A suitable gag may be purpose-made for the procedure. Alternatively, a partially-used roll of elastoplast may be adequate. The operator can then introduce a lubricated rubber or soft polythene tube through the gag and over the tongue into the oesophagus sufficiently slowly for the dog to swallow as the tube passes the tracheal opening.

#### Sub-cutaneous (s.c.) injection

The skin of dogs is so loose over the neck, back and flanks that injections can be made at virtually any of these sites. The skin is held up with the fingers of one hand to make an inverted "V" shaped tent. A 21-22 G needle is inserted through the skin using the other hand, directed slightly downwards. Care should be taken to ensure that the needle does not penetrate through both sides of the "tent" in the skin. Large volumes of fluid can be injected if given at multiple sites (10 - 12 ml/kg per injection site). A lesser volume per site (15-30 ml) is preferable.

## Intradermal (i.d.) injection

This is simple in dogs as the skin is relatively thick. The skin must be carefully shaved and cleaned before injection. A 26-30 G needle is inserted almost parallel to the skin. If the injection is successful, a "blister" will appear in the skin.

#### Intramuscular (i.m.) injection

The anterior thigh muscle (quadriceps) provides the safest site for injection. The belly of the muscle should be gripped between fingers and thumb of one hand. Alternatively, the triceps or paralumbar muscles can be used. If injecting into the posterior thigh muscles, care must be taken to avoid the nerves and large vessels located in that region. Intramuscular injection at any site is painful.

## Intravenous (i.v.) injection

The cephalic vein on the anterior aspect of the forelimb is the most convenient vessel for injection. The vessel is easily seen if the area is carefully clipped and swabbed with alcohol. Dilation and stabilisation of the vessel is achieved by the assistant pushing the dog's forelimb into extension using the palm of his/her hand behind the dog's elbow. The assistant's thumb is then placed over the anterior aspect of the dog's upper forelimb, dragged slightly laterally and then held firmly. The operator penetrates the vein from the antero-medial aspect. The vein may be stabilised using the thumb of the opposite hand. A 21-22 G needle is suitable for most i.v. injections. Alternatively, butterfly needles (25 mm, 21 G) or a 21-23 G Medicut® indwelling catheter can be taped in place and connected directly to a syringe or to a three-way tap for sequential injections or for withdrawal of blood. The recurrent tarsal vein on the lateral aspect of the

hind limb is less easily stabilised for venepuncture but is a useful alternative to the cephalic vein. Immediately following venepuncture, firm pressure is applied to the injection site until any bleeding has stopped.

#### Specimen collection

#### Blood collection

Up to 20 ml of blood (not exceeding approximately 1 per cent of body weight) may be collected from the cephalic vein using a syringe and needle. Volumes of 1-50 ml can be collected from this vessel with a butterfly infusion set (21G) or Medicut catheter. Pressure should be maintained above the elbow by an assistant. Alternately squeezing and flexing the foot will increase the rate of venous return and flow.

Larger volumes of blood (100 ml or even more) can readily be collected from the jugular vein percutaneously and without the need to sedate the animal unless it is particularly fractious. The site over the jugular vein should be clipped closely and swabbed with alcohol. The vein is distended by pressing the thumb into the jugular groove at the point of entry into the thorax. The needle (21 G) is introduced first into the skin then advanced almost parallel to the surface to puncture the vein. Haemostasis is achieved by firm digital pressure over the puncture site.

Because of the risk of haemorrhage into the pericardial sac, cardiac puncture should be performed only in the anaesthetised animal and as a terminal procedure. A 21-23 G needle 4-5 cm long is introduced into the left side of the chest between the fourth or fifth intercostal space over the maximum heart beat. For exsanguination, larger bore needles (9-16 G) can be used.

The femoral artery is commonly used for collection of blood for transfusion purposes. This is performed in the anaesthetised animal as a terminal procedure. The skin of the upper medial thigh is surgically prepared. Using aseptic techniques, the skin is incised over the area where the pulse is palpable. The femoral artery is surgically exposed. Blood is collected using a wide bore needle.

## Urine collection

For urinanalysis not involving microbiological studies, a "mid-stream" urine sample may be adequate, i.e. a sample collected in a clean container in the middle of the dog's normal act of urination.

Urethral catheterisation can usually be carried out in unanaesthetised dogs but aggressive or frightened dogs should be sedated or even lightly anaesthetised. To avoid introduction of micro-organisms into the bladder, sterility must be maintained when performing this procedure. Males are laid on one side, the penis is extruded from the prepuce and the tip is gently cleaned. A lubricated (K-Y or xylocaine gel) human urethral catheter (44-10 FG) is then introduced into the urethra and can usually be advanced without difficulty into the bladder. Slight resistance may be felt when the catheter passes either the caudal end of the os penis or around the pelvic flexure. This resistance may be overcome by moving the penis and prepuce towards the operator. Catheterisation of females is comparatively more difficult and is best performed with the animal standing up or in sternal recumbency. The urethral opening in the vagina is located by direct visualisation via a lubricated (K-Y Jelly) speculum. It is located in the centre of a slightly raised papilla on the ventral surface of the vagina. A lubricated catheter is inserted into the urethra and can usually be advanced into the bladder without resistance.

For cystocentesis, it is essential to be able to palpate the bladder for accurate insertion of the sampling needle. With the dog lying on its back or on its side, the skin overlying the bladder is clipped and cleaned with alcohol. If the urine sample is being collected for microbiological studies, the skin must be surgically prepared. The bladder must be palpated in the posterior abdomen and grasped by one hand through the body wall. A 21 G needle, 4-6 cm long, can then be introduced through the body wall in the midline and directed caudally at an angle of about  $45^{\circ}$  so that it punctures the posterior bladder and remains within the lumen whilst urine is aspirated into a syringe.

Table 1. Space requirements for each dog						
Source	Weight (kg)	Floor area (m <sup>2</sup> )	Minimal height (m)	Group or		
				loose housing (m <sup>2</sup> )		
CCAC (1984)	12	0.75	0.8	1.5		
	15	1.20	0.9	2.0		
ILAR Guide (1996)	15	0.72	-	-		
	Up to 30	1.08	-	-		
	30	2.16	-	-		
MacArthur (1987)	15 -20	4.2 (may be used for two animals)	-	-		

#### Table 2. Reproductive data

Age at puberty - male; - female	7-8 months, 8-14 months
Cycle length	21 - 28 days
Breeding season (average)	bi-annual
Average oestrus interval	7 - 8 months
Gestation length	63 (53-71) days
Litter size - small breed,	
large breed	2 -3; 4 - 12
Average reproductive span	
- male; female	5 - 7 years; 4 - 5 years

#### Faeces

Fresh samples can be collected immediately following defaecation, or directly from the rectum using a lubricated gloved finger, a swab or small speculum.

#### Vaginal fluid, vaginal cells

Vaginal smears are a common requirement in laboratory dogs. A sterile swab is introduced into the vagina, turned, withdrawn and smeared onto a glass slide. Alternatively, a small volume of isotonic saline is instilled into the vagina via a blunt-ended glass pipette, aspirated and dropped onto a glass slide. During oestrus, over 1 ml of fluid can be aspirated directly from the vagina using a blunt-ended glass pipette. Vaginal smear cytology at different stages of the oestrus cycle is well described (Nett and Olsen, 1983).

## **Additional procedures**

References for some additional procedures are as follows: collection of cerebrospinal fluid (CSF tap; cistern and lumbar puncture) - Green and Oliver (1983); collection of semen - Olson (1992); chronic vascular access - Lumley *et al.*, (1990); anatomy - Evans and Christensen (1993); surgical techniques - Bojrab (1990), Slatter (1993), Piermattei (1993); electrocardiogram - Tilley (1992), Edwards (1987).

# Breeding

Reproductive data for the dog are shown in Table 2. The canine ovarian cycle can best be understood in terms of the follicular and luteal phases. Bitches ovulate spontaneously following a follicular phase during which the elevated oestrogen levels cause increasingly evident external signs of pro-oestrus. This phase often lasts 5 to 10 days (range: 2 to 22 days). The first evidence of pro-oestrus may be vulval enlargement followed 1 to 4 days later by a bloody vaginal discharge. Some bitches may be so fastidious in licking their genitalia that the bloody discharge is not observed on the vulva although it is present intravaginally. During this period the bitch is attractive to, but will not accept, the male. Oestrus usually corresponds with the peak of luteinising hormone and commences when the female accepts the male with the characteristic lordosis and sidewards deviation of the tail. The behavioural change from pro-oestrus to oestrus may occur rapidly (over an 8 to 12 hour period) or slowly (over 1 to 3 days). With the behavioural change, the bloody discharge begins to subside. The transition from late pro-oestrus to oestrus can be determined by testing the bitch's response daily with the male. When mating occurs, the bitch and the dog remain "tied" for twenty to thirty minutes. Oestrus lasts from 6 to 12 days during which time the female may continue to permit mating. Ovulation, however, occurs within 1 to 2 days after first acceptance of the male and is spontaneous. Onset of metoestrus is defined behaviourally as the first day after a period of oestrus that the bitch refuses the male. This period is characterised by a gradual decrease in the size and turgidity of the vulva and usually lasts 2 to 3 months in the absence of pregnancy. Following a reproductive cycle, the bitch enters a period of ovarian quiescence called anoestrus. This period can last from two to ten months. Concannon and Lein (1989) provide extensive information on the correlation between hormonal and behavioural changes in the bitch's reproductive cycle.

Changes in exfoliated vaginal epithelial cells occur as a result of changing secretory patterns of ovarian hormones. The daily or alternate daily examination of vaginal smears can be helpful in analysing phases of the oestrus cycle and in determining optimum breeding times. The cell types found in vaginal smears taken during the oestrus cycle have been described by a number of authors (Nett and Olsen, 1983, MacArthur 1987, Concannon, 1989).

## Detection for pregnancy

Pregnancy can be detected by palpating the abdomen of the bitch (unreliable in mid-pregnancy), by ultrasound (from about 17 to 21 days after ovulation) or by X-ray (in the last 3 weeks of pregnancy).

Some bitches show signs of pseudo-pregnancy (or false pregnancy) beginning 6 to 8 weeks after the oestrus period. These animals may exhibit one or all of the following signs: distended abdomen, engorged uterus, development of mammary glands, lactation, and various behaviours such as nervousness, nesting, guarding enclosed areas, and mothering objects.

## Parturition

The most consistent physical sign of impending parturition is palpable relaxation of pelvic and abdominal musculature accompanied by increased friendliness and loss of appetite. The first stage of parturition may last several hours during which the bitch may appear agitated. At the end of this stage, the bitch may begin to exert voluntary pressure. The allantochorion or "water bag" of the first foetus appears at the vulva and is often ruptured by the bitch. Shortly after, the first foetus is presented in the cervix and strong abdominal contractions will occur. The expulsion of the first foetus may take from only a few minutes to a couple of hours. A longer delay indicates that assistance is required. The bitch will chew the umbilical cord and eat the placenta. Her continual licking of the puppy stimulates respiration during the first minutes of independent life. Further puppies will follow at intervals of up to one hour.

## Breeding systems

A colony ratio of 1 male to 10 females is acceptable for most breeding systems. Dogs may be bred in packs of 1 male with up to 12 bitches. Once a bitch is obviously pregnant, she is removed to separate accommodation for whelping. This system has the advantage that oestrus detection by the animal technician is unnecessary. However, conception rates may be reduced if several bitches are in oestrus simultaneously. A second system involves bringing the bitch to the location of the male when the bitch is fertile. This practice is appropriately protective of the male's libido and territoriality but requires daily examination of each bitch for signs of pro-oestrus. A bitch is put with the male approximately 10 days following prooestrus detection and remains with the male for about 5 days, or the dog and bitch may be put together daily over the same 5 days, for at least 1 hour each day.

# Hormonal manipulation of reproduction

Naturally produced and synthetically prepared reproductive hormones are frequently used in canine reproduction to improve reproductive efficiency or to treat specific disorders (e.g. oestrus induction, induction of ovulation, oestrus suppression and pregnancy prevention, reduction of signs of pseudo-pregnancy, and termination of unwanted pregnancy). The potential side-effects of these drugs in intact bitches include endometritis and pyometron (Braakman *et al.*, 1993; Cain, 1995).

# Development of the newborn

Newborn puppies commence sucking immediately after they have been licked clean by the bitch. It is important that puppies suck adequately during the first 24 hours of life to ensure absorption of maternal antibodies from the colostrum. Puppies are born with their eyes and ears closed, with eyes fully open and ears becoming patent by day 10-14. Puppies are usually walking steadily by day 21 and achieve voluntary control of micturition and defecation between days 16 to 21. By about 3 weeks of age, puppies will begin eating solid food. Separation from the bitch may occur as early as 6 weeks of age. However, some studies have shown that disease susceptibility and mortality is higher in puppies with a short maternal contact period and have recommended that maternal contact be maintained until 12 weeks (Slabbert and Rasa, 1993). Hand rearing of puppies is best achieved by fostering on to a lactating female. Alternatively, puppies can be successfully hand reared using a commercial milk substitute (for example — Divetalac ®, Animalac ®). For further reading on orphan feeding and care, see Lewis *et al.*, (1987).

#### Feeding

Dogs vary enormously in both adult size and growth rate. The nutritional requirements of dogs are provided in detail in Lewis et al., (1984) and IVS Annual (1994). The requirements of the average adult non-breeding dog can be met by feeding a good quality canned, semi-moist or dry commercial dog food. Home-made diets may require supplementation with essential vitamins, minerals or fatty acids. Some canned diets contain added cereals so that they constitute a complete diet, whilst others with a higher protein content are intended to be fed together with a cereal biscuit. The semi-moist diets usually constitute a complete diet and should be fed with an ample supply of drinking water. Dry diets should also be fed with ample drinking water. Dry diets can be fed from a food hopper where the food can remain fresh for several days. This ad lib availability enables the dog to develop a nibbling pattern of feeding. In some animals this may lead to obesity, so that restricted meals offered twice daily become necessary. The temperature at which food is fed may affect its acceptability. The sudden intake of very cold foods or sudden changes in diet may be poorly tolerated and result in diarrhoea. Intermittent large meals should be avoided, as acute gastric dilatation may result.

Attention to nutritional requirements is especially important in the following situations:

- the pregnant bitch, especially during the last three to four weeks of gestation and throughout lactation (e.g. prevention of lactation tetany);
- puppies before and after weaning;
- during the growth period (e.g. prevention of secondary nutritional hyperparathyroidism); and
- during periods of stress (e.g. following surgery).

The reader is referred to Lewis *et al.*, (1987) and Donoghue (1992) for further information.

## Health and disease

## Signs of health

The normal healthy dog is active, alert and responsive to human presence. Appetite and body condition should be good and the animal should not be obese. Eyes and ears should be clear of any discharge. Teeth should be clean. Gums are normally smooth and moist, and pink in colour. The coat should be clean and shiny and no lesions should be visible on the skin. Urine and faeces should be passed without difficulty. The urine of a dog is normally clear or straw coloured. While the faeces are normally dark brown, the colour and consistency varies with the diet. Pale, bulky faeces are abnormal. Body weight should be monitored regularly (e.g. weekly). Dogs should be clinically examined by a veterinarian on a regular basis, especially if the animal is part of an experimental protocol. Because of enormous variations in behaviour, appearance and clinical signs of individual animals, it is important to record baseline data for each animal so that abnormal signs may be detected more easily. Any dog exhibiting abnormal clinical signs should be referred to a veterinarian for examination and diagnosis.

#### **Disease prevention**

#### Vaccination

Regular vaccination protocols should be followed in breeding colonies, and dogs obtained from local pounds should be vaccinated prior to release to an experimental protocol. Vaccinations are recommended against distemper virus, hepatitis virus, parvovirus, and two organisms implicated in infectious tracheobronchitis (parainfluenza virus and *Bordatella bronchiseptica*). To ensure high levels of maternal antibodies in the colostrum, breeding bitches should be vaccinated annually. Veterinary advice should be obtained on the vaccination protocol required.

#### Intestinal parasites

There are a large number of drugs available for the treatment of internal parasites (see IVS Annual, 1996). The most common intestinal parasites in dogs are roundworm, hookworm, whipworm and tapeworm. It is common practice to treat dogs either with a broad-spectrum anthelmintic active against the most common parasites (e.g. Drontal ®, Popantel ®, Canex ®), an anthelmintic active against the common parasite for the age of the dog (e.g. Canex Puppy Suspension ® for roundworms and hookworms), or a specific anthelmintic based upon the results of a faecal analysis. Infestation with intestinal parasites in breeding colonies is prevented by adequate cage/run cleaning protocols, regular faecal floats of a sample population, and an appropriate de-worming schedule of all animals. Roundworm is the most common problem in puppies and breeding bitches, while tapeworm infection is usually concomitant with flea infestation. Recommended de-worming schedules for puppies normally concentrate on possible roundworm and hookworm infestation (pyrantel embonate every 2 weeks until 12 weeks of age; every month between 3-6 months of age, then a 3-6 monthly intervals). Regular broad-spectrum treatment of adult dogs is recommended at 3-6 monthly intervals (e.g. Drontal®). In adult dogs of unknown history, initial treatment should consist of two doses given 3 weeks apart. Breeding bitches should be treated in the last trimester of pregnancy so that transplacental and transmammary transfer of roundworms is prevented.

#### Heartworm prevention

Dogs are infected with heartworm when the immature stages of the parasite are injected into the blood stream via the bite of the mosquito. The presence of the adult worms in the right ventricle and pulmonary artery of the heart can result in a long period of sub-clinical disease, followed by clinical disease and death. All dogs with an unknown history should be tested for heartworm disease prior to release to an experimental protocol. Detection of heartworm infestation is best done using a highly specific and sensitive antigen test (SNAP ®, Vetred ®) which detects antigen specific to mature or immature worms. Due to a lower sensitivity when compared with antigen tests, high false-negative rates are possible when testing only for the presence of microfilaria (larvae) in the peripheral blood. Dogs which test positive for the presence of heartworm should not be used as experimental animals. In dogs which are heartworm negative, the disease is easily prevented by the administration of appropriate compounds which kill the

immature tissue stages of the parasite. These drugs can be administered either daily (diethylcarbamazine) or monthly (ivermectin, milbemycin oxime, moxidectin). The newer forms of the monthly preparation also act against intestinal parasites (milbemycin oxime - Endovet ®), or include compounds which act against selected intestinal parasites (Ivermectin and pyrantel - Heartguard Plus ®).

## External parasites

Without doubt, the most common external parasite of the dog is the flea. Regular treatment with insecticidal washes (e.g. organophosphate preparations at 7-14 day intervals), sprays (Frontline® at 8-12 weekly intervals), oral organophosphate dosing (Proban ® every 2-3 days) or spot on preparations (Advantage ® once monthly), is recommended if flea infestation is observed. However, good hygiene within the kennel environment and defleaing of any new dog entering the colony will largely prevent this problem. Another common and often endemic infestation in colony dogs is ear mites (Otodectes cynotis). Scratching of the ear or a dark brown crusting discharge from the ear canal is highly suggestive. Treatment with Ivermectin® (200-400 µg/kg s/c at two weekly intervals for 2-3 treatments) is very effective. Other, less common external parasites include sarcoptes, demodex, and chyletellia mites, ticks and lice. In such cases, clinical signs such as scratching, hair loss or visible presence of the parasite should warrant examination and diagnosis by a veterinary surgeon.

## Physiology, biochemistry, haematology

There is a wealth of physiological data on the dog. Jacobs et al (1995) provide the most comprehensive reference values, including cerebrospinal fluid analysis, gastrointestinal and endocrine function tests and coagulation screening tests. Other references include Lumley et al (1990) and Loeb and Quimby (1989). Physiological, biochemical and haematological data in greyhounds differs significantly from other breeds of dogs. For information on this breed, see Lording (1983).

## Sedation, analgesia and anaesthesia

Common injectable agents for premedication, analgesia and analgesia are listed in table 3. For general references on anaesthesia, sedation and analgesia in the dog, see Bednarski (1992); Flecknell (1996); Lumley *et al.*, (1990); Green (1982;) Mandsager and Raffe (1989) and Quandt and Rawlings (1996).

## Sedation

Sedation is often used as a chemical restraint for many diagnostic and therapeutic procedures when animals are fractious or unco-operative, or because the procedures produce discomfort or pain or require lack of motion. Sedatives are also used as part of pre-anaesthetic medication to reduce the anxiety of the animal and assist in its restraint and to ensure a smooth induction of anaesthesia and provide a quiet and gradual recovery. Following the use of a sedative, the dose of some anaesthetic agents may need to be reduced. Sedatives may be used alone while others are more effective in combination.

# Analgesia

Analgesic agents can be used in the treatment of postoperative pain, in combination with sedatives to produce effective chemical restraint, or as part of pre-anaesthetic medication. When given pre-operatively, they act to inhibit nociceptive input to the central nervous system, thus providing a degree of prevention as opposed to treatment of pain. Single agents used for this purpose include pethidine, buprenorphine and carprofen. The last two have the advantage of long periods of effect (up to 12 hours and 24 hours respectively) when compared with pethidine (2 hours).

## Anaesthesia

Animals subjected to anaesthesia should be healthy and free from hepatic, renal, cardiac or respiratory disorders. To reduce the incidence of regurgitation during induction, food should be withheld for a period of 12 hours. Premedication is usually given 30-40 minutes prior to induction of anaesthesia, and may consist of a combination of sedative, anticholinergic and analgesic agents. Anticholinergic agents (e.g. atropine 0.05 mg/kg i.m. or s.c.) will reduce the side effects of many anaesthetic drugs, e.g. the stimulation of respiratory secretions and the parasympathetic stimulation of the cardiopulmonary system. Acclimatisation of the animal to handling will reduce the effects of stress and the possibility of injury to animal and personnel during induction of anaesthesia.

## Injectable anaesthetic agents

Anaesthesia can be achieved via i.v. administration of the drug using the cephalic vein. Short acting anaesthetic drugs (e.g. thiopentone, methohexitone and propofol) are most commonly used as induction agents for general anaesthesia. Pentobarbitone may be used for its longer acting effects (45-60 minutes of light anaesthesia following a single intravenous dose) with long periods of anaesthesia being achieved using a continuous infusion. This drug has the disadvantage of providing inadequate analgesia for major surgical procedures, unless dangerously high doses are administered. Even at lower dose rates, it produces respiratory and cardiovascular system depression. The use of barbiturates in hound breeds (e.g. greyhound, saluki) results in prolonged periods of recovery and should be avoided.

Propofol can be used both as an induction agent and to maintain anaesthesia by constant infusion (Robinson *et al.*, 1995). However, neither sedation nor anaesthesia produced by propofol are associated with complete pain relief. Lack of reflex response may only be complete at deep levels of anaesthesia. The drug must be administered slowly to avoid respiratory arrest and apnea.

Chloralose is not suitable for routine use as an anaesthetic but is of value in providing long lasting (6-10 hours), stable, light anaesthesia with minimal cardiovascular and respiratory depression. The degree of analgesia is usually inadequate to allow surgical procedures to be undertaken. For this reason, it is often combined with pentothal or urethane to produce sufficient analgesia to allow surgical procedures. Urethane is carcinogenic and its use should be discouraged.

# Inhalational agents

Long periods of stable anaesthesia can be induced using a face mask in the sedated animal, or by an i.v. short-acting anaesthetic agent. Following induction, the animal is intubated and maintained using a volatile inhalational agent (e.g. halothane 1-2%, isoflurane 2-3% or enflurane 0.8-2%). For small dogs (10 kg) an Ayres T-piece or a Bain coaxial circuit should be used. Intubation is easily performed with the animal lying on its side with its jaws held open by an assistant. The larynx can be visualised using a

Drug	Tradename	Dosage	Route	Comments
Atropine	Atrosine mitis	0.05 mg/kg	S.C.	
			i.m.	-
Acepromazine	Promez ACP	up to 0.25 mg/kg	S.C.	
			i.m.	S√
Diazepam	Valium			
	Pamlin	0.2-0.6 mg/kg	i.v.	SVV
Xylazine	Rompun	2.0 mg/kg	i.m.	AC, A ✓ , S ✓ ✓ , M ✓ ✓ , R, V
Pethidine	-	1.0 mg/kg	i.m.	A ✓ ✓ , S ✓
Buprenorphine	Temgesic	0.01 mg/kg	i.m.	
			i.v.	A ✓ ✓ ✓,S ✓
Carprofen	Zenecarp	4 mg/kg (initial)g	i.v.	
		2mg/k (top-up to 24 hours	S.C.	AA 🗸 🗸 🏑, NSAID
Medetomidine	Domitor	0.01-0.08 mg/kg	i.m.	
			i.v.	A ✓ ✓, S ✓ ✓, M ✓ ✓, R
Droperidol - fentanyl	Leptan, Innovar - ve	: 1.0 ml/10 kg	i.m.	AC, A ✓ ✓ ✓, S ✓ ✓
			i.v.	R (fentanyl only)
Fentanyl- fluanisone	Fentaz, Hypnorm	0.2 ml/kg	i.m.	AC, A ✓ ✓ ✓, S ✓ ✓, R
Tiletamine / zolazepam	Zolitil, Telazol	7mg/kg	i.m.	AC, S ✓ ✓, A ✓ ✓, M ✓
				(Recovery may be prolonged or
				rough)
Thiopentone	Pentothal	10 - 20 mg/kg	i.v.	A,✓, M ✓, T✓ ✓
2.5 - 5% solution	Intraval			
Methohexitone	Brietal	4 - 8 mg/kg	i.v.	A✓,H✓,T✓✓
Pentobarbitone	Nembutal	20 - 30 mg/kg	i.v.	A poor, M ✓
6% solution				
-chloralose	-	80 - 100 mg/kg	i.v.	A poor , NR
1% solution				
- / · · · · · · · · · · · · · · · · · ·		00	·	
-chioraiose	-		I.V.	A ✔, IVI ✔, INK
- uretnane	<b>D</b>	500 mg/kg	I.V.	
Propofol	Diprivan	2 - 8 mg/kg	i.v.	A poor, H 🗸

# Table 3. Common injectable agents for premedication, analgesia and anaesthesia in the dog.

AC = prior administration of an anticholinergic drug recommended ; A = analgesia; S = sedation;

**M** = muscular relaxation; **R** = reversible or capable of reversing agonists; **V** = Vomiting common;

**NSAID** (Non-steroidal antiinflammatory drug); **H** = Useful in hound breeds and

puppies (10 weeks of age); NR = Use in non-recovery experiments only; ✓ = Degree of action

laryngoscope or a strong overhead light. Some operators prefer to hold the animal's tongue while manipulating the endotracheal (ET) tube. The size of the ET tube used varies with the size of the animal (5mm - chihuahua, 16mm - St Bernard). A cuffed ET tube prevents the aspiration of fluid/stomach contents into the lungs. At the completion of the anaesthesia, the ET tube must not be removed until the swallowing reflex has returned, and the cuff deflated.

## Local anaesthesia

To reduce the side effects of anaesthetic and analgesic agents, anaesthetic drugs may be administered locally instead of systemically. For procedures involving the caudal half of the body, an epidural anaesthetic may be considered. Analgesia following thoracic surgery can be produced using an intercostal nerve block or the interpleural administration of local anaesthetic agent (Conzemius *et al.*, 1994; Pascoe and Dyson, 1993). A good review of local anaesthetic and analgesic techniques is provided by Quandt and Rawlings (1996).

## Anaesthetic monitoring

All animals undergoing general anaesthesia must be monitored to ensure an adequate level of anaesthesia for the procedure being performed, and to minimise peri-operative mortality and morbidity. Factors which should be monitored may include depth of anaesthesia (e.g., eye reflex, pedal reflex), respiratory function, cardiovascular function, body temperature and, when using a blocking neuromuscular agent and ventilation, neuromuscular function. Nicholson (1996) provides an overview of monitoring techniques.

## Euthanasia

The most humane manner of performing euthanasia in dogs is by rapid i.v. injection of a concentrated solution (300 mg/ml) of pentobarbitone at 150 mg/kg. Dangerous or fractious animals may need prior sedation (e.g., medetomidine or xylazine i.m.). Euthanasia in the anaesthetised dog can be carried out by exsanguination, or by i.v. or i.c. injection of pentobarbitone.

#### **References and further reading**

- Bednarski, R.M. (1982). Recent advances in injectable chemical restraint. In: Kirk, R.W. and Bonagura, J.D. (eds). *Current Veterinary Therapy XI. Small Animal Practice*. W.B. Saunders, Philadelphia, pp. 27-31.
- Bojrab, M.J. (ed). (1990). *Current Techniques in Small Animal Surgery*. Lea and Febiger, Philadelphia.
- Braakman, A., van Haafaften Okkens, A.C. (1993). Medical methods to terminate pregnancy in the dog. *Comp Cont Edu Prac Vet*, 15: 1505-1512.
- Branson, K.R., Gross, M.E. (1994). Propofol in veterinary medicine. J Am Vet Med Assoc, 204: 1888-1890.
- Cain, J.L. (1995). The use and misuse of reproductive hormones on canine reproduction. In: Bonagura JD (ed). Current Veterinary Therapy XII. Small Animal Practice. WB Saunders, Philadelphia, pp. 1069-1075.
- CCAC (Canadian Council on Animal Care). (1984). Guide to the Care and Use of Experimental Animals. Ottawa.
- Clark, G.N. (1992). Epidural analgesia. In: *Current Veterinary Therapy XI*. Kirk, R.W. and Bonagura, J.D. (eds). W.B. Saunders, Philadelphia. pp. 92-98.
- Concannon, P.W. (1983). Reproductive physiology and endocrine patterns of the bitch. In: Kirk R.W. (ed). *Current Veterinary Therapy VIII. Small Animal Practice.* W.B. Saunders, Philadelphia. pp. 886-901.
- Concannon, P.W. and Lein, D.H. (1989). Hormonal and clinical correlates of ovarian clycles, ovulation, pseudo pregnancy and pregnancy in dogs. In: Kirk, R.W. and Bonagura, J.D. (eds). *Current Veterinary Therapy X. Small Animal Practice*. W.B. Saunders, Philadelphia. pp. 1269-1282.
- Conzemius, M.G., Brockman, D.J., King, L.G. and Perkowski, S.Z. (1994). Analgesia in dogs after intercostal thoracotomy: a clinical trial comparing intravenous buprenorphine and interpleural bupivacaine. *Vet Surgery*, 23: 291-298.
- Donoghue, S. (1992). Nutritional recommendations for reproductive performance. In: Kirk, R.W. and Bonagura, J.D. (eds). *Current Veterinary Therapy XI. Small Animal Practice*. WB Saunders, Philadelphia. pp. 971-980.
- Edwards, N.J. (1987). Bolton's handbook of canine and feline electrocardiography. W.B. Saunders, Philadelphia.
- Evans, H.E. and Christensen, G.C. (1993). *Miller's Anatomy of the Dog.* W.B. Saunders, Philadelphia.
- Flecknell, P.A. (1996). Laboratory animal anaesthesia. Academic Press, Orlando.
- Green, C.E. and Oliver, J.E. (1993). Neurologic examination. In: Ettinger S.J. (ed). *Textbook of Veterinary Internal Medicine*. W.B. Saunders, Philadelphia. pp. 1698-1711.
- Green, C.J. (1982). Animal anaesthesia. Laboratory Animals Ltd, London.
- Index of Veterinary Specialties (IVS) Annual. (1994). Badewitz-Dodd, L.F. (Managing Editor), Kyodo Printing Co, Singapore.
- ILAR (Institute of Laboratory Animal Resources). (1996). *Guide* for the Care and Use of Laboratory Animals. National Academy Press, Washington DC.
- Jacobs, R.M., Lumsden, J.H. and Vernau, W. (1995). Canine and feline reference values. In: Bonagura, J.D. and Kirk, R.W. (eds). *Current Veterinary Therapy XII. Small Animal Practice*. W.B. Saunders, Philadelphia. pp. 1395-1417.

- Lewis, L.D., Morris, M.L. and Hand, M.S. (1987). *Small Animal Clinical Nutrition III*. Mark Morris Associates, Kansas.
- Loeb, W.F. abd Quimby, F.W. (eds). (1989). *The Clinical Chemistry* of Laboratory Animals. Pergamon Press, New York.
- Lording, P.M. (1983). Haematology and biochemistry profiles. In: Post-graduate committee in veterinary science. *Proceedings No. 64. Refresher Course on Greyhounds*. University of Sydney. pp. 491-506.
- Lumley, J.S.P., Green, C.J., Lear, P. and Angell-James, J.E. (1990). *Essentials of Experimental Surgery*. Butterworths, London.
- MacArthur, J.A. (1987). The dog. In: Poole T (ed). *The UFAW Handbook on the Care and Management of Laboratory Animals.* 6th ed. Longman Scientific and Technical, London. pp. 456-475.
- Mandsager, R.E. and Raffe, M.R. (1989). Chemical restraint techniques in dogs and cats. In: Kirk, R.W. (ed). Current Veterinary Therapy X. Small Animal Practice. W.B. Saunders, Philadelphia. pp. 63-70.
- Nett, T.M. and Olson, P.N. (1983). Reproductive physiology of dogs and cats. In: Ettinger, S.J. (ed). *Textbook of Veterinary Internal Medicine*. W.B. Saunders, Philadelphia. pp. 1698-1711.
- NHMRC. 1996. *Policy on the Care of Dogs in Medical Research Draft*. National Health and Medical Research Council, Canberra.
- NHMRC Animal Welfare Committee. (1994). *Ways of Minimising Pain and Distress in Animals in Research*. Australian Government Publishing Service, Canberra.
- NHMRC, CSIRO, AAC. (1990). Australian Code of Practice for the Care and Use of Animals for Scientific Purposes. Australian Government Publishing Service, Canberra.
- Nicholson, A. (1996). Monitoring techniques and equipment for small animal anaesthesia. *Australian Veterinary Journal*. 74: 114-123.
- Olson, P.N. Collection and evaluation of canine semen. 1992. In: Kirk, R.W. and Bonagura, J.D. (eds). *Current Veterinary Therapy XI. Small Animal Practice.* W.B. Saunders, Philadelphia. pp. 938-943.
- Pascoe, P.J. and Dyson, D.H. (1993). Analgesia after lateral thoracotomy in dogs. Epidural morphine Vs intercostal bupivocaine. *Veterinary Surgery*, 22: 141-147.
- Piermattei, D.L. (1993). An Atlas of Surgical Approaches to the Bones and Joints of the Dog and Cat. W.B. Saunders, Philadelphia.
- Quandt, J.E., Rawlings, C.R. (1996). Reducing postoperative pain for dogs: local anaesthetic and analgesic techniques. *Comp Cont Edu Prac Vet*, 18: 101-111.
- Robinson, E.P., Sanderson S.L. and Machon, R.G. (1995). Propofol: a new sedative - hypnotic anaesthetic agent. In: Bonagura, J.D. and Kirk, R.W. (eds). Kirk's Current Veterinary Therapy XII. Small Animal Practice. W.B. Saunders, Philadelphia. pp. 77-81.
- Slabbert, J.M. and Rasa, O.A.E. (1993). The effect of early separation from the mother on pups in bonding to humans and pup health. *J Sth Afr Vet Assoc*, **64**: 4.
- Slatter, D.(ed). (1993). *Textbook of Small Animal Surgery*, (2nd ed). W.B. Saunders, Philadelphia.
- Tilley, L.P. (1992). Essentials of Canine and Feline Electrocardiography. Lea and Febiger, Philadelphia.