Anatomy and physiology of the injection site – implications for extended release parenteral systems

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# Introduction

There are many extended and controlled release injectable systems used to deliver drugs in human and veterinary medicine (Brannon-Peppas & Blanchette 2004; Medlicott et al 2004; Winzenburg et al 2004; Berges & Bello 2006; Kreye et al 2008). These systems are prepared from a variety of biocompatible materials and aim to release drug for an extended period following injection or implantation. Drug release is governed by the design of the dosage form, although the biological environment often influences drug release (Anderson et al 1981; Anderson et al 1993; Daugherty et al 1997; Higgins et al 2009). Understanding how the biological environment contributes to drug release following administration is increasingly becoming a focus for drug delivery research.

Extended release parenteral delivery systems range from the relatively simple aqueous suspensions that prolong drug release due to slow dissolution at the injection site to more sophisticated *in situ* gelling implants and polymeric biodegradable microparticulate systems (Medlicott & Tucker 1999; Medlicott et al 2004; Traitel et al 2008). For example, long acting intramuscular aqueous suspensions of penicillin have been available since the 1950s (Fletcher & Knappett 1953) and oily formulations of neuroleptic drugs since the 1970s (Marder et al 1989). More recently, sustained release microparticulate polymeric formulations of leuprolide have been developed for prostate cancer (Okada 1997; Perez-Marrero & Tyler 2004; Berges & Bello 2006). For all implanted extended release delivery systems, balancing the *in vivo* drug release from the delivery system with drug absorption, distribution, metabolism and elimination processes is key to achieving target drug levels in the body (**Figure 1**). Hence, careful consideration of drug pharmacokinetics and knowledge of target plasma and tissue drug concentrations can guide development of extended and controlled release parenteral drug delivery systems. One useful way to categorise parenteral delivery systems has been suggested by Washington et al. for intramuscular injections (Washington et al 2001). According to this, injectable delivery systems can be divided into those in which the pharmacokinetics is predominately controlled by the implant (or device) and those controlled by the process of absorption into blood or lymphatic capillaries at the implant site (i.e. perfusion limited pharmacokinetics), as shown in **Figure 2**. If drug release from the device is slow and drug absorption from the tissue is fast, then a situation results where the appearance of drug in the blood is closely controlled by the release characteristics of the extended release device (Type I). At the other extreme, an aqueous solution of a water-soluble drug provides its dose immediately following injection and in this case, the absorption processes (ka) may limit the appearance of drug in the blood (Type II). Many extended release parenteral delivery systems fall between these two extremes so it is important to consider the biological processes at injection sites that contribute to drug absorption. A further example of extended release systems are those designed to deliver drugs locally at the implantation site. For these local delivery systems a balance between drug release and drug absorption that maintains a constant local drug concentration may be the goal. The previous Chapter has shown that while implantable extended release systems must be biocompatible, they cannot be considered biologically inert because the patient will react to the presence of the implant.

The aims of this Chapter are to review the anatomy and physiology of injection sites (intravenous, intramuscular and subcutaneous – **Figure 3**) and to summarise the biological variables that affect drug release and absorption at these sites.

# Intravenous route

The intravenous route of administration for injectable products provides direct access into the blood stream and so therapeutic agents delivered by this route are available immediately in the systemic circulation. The plasma concentration is determined by the initial dose injected, the drug distribution and rates of metabolism and elimination according to Equations 1 and 2 for first-order elimination kinetics.

Equation 1

Equation 2

where: Co = initial plasma concentration, V = volume of distribution, Ct = plasma concentration at time t and k = first order elimination rate constant.

Controlled drug release can be achieved using the intravenous route when particulate systems such as liposomes or polymeric nanoparticles are used (Brannon-Peppas & Blanchette 2004; Cournarie et al 2004; Goel et al 2009). The prolonged effect of these dosage forms is primarily attributed to the time taken for the bioactive to be released from the circulating particles. Properties of nanoparticles, such as: method of preparation, method of drug association (e.g. encapsulated or surface-adsorbed) and the type of polymer used can be manipulated to alter the drug release profile and the *in vivo* fate of the drug. For example, nanoparticles that have the drug dispersed uniformly throughout a matrix generally release drug in a first-order process controlled by diffusion and polymer degradation (Soppimath et al 2001). Nanoparticulate systems administered by the intravenous route can also be used to facilitate drug targeting to specific tissues and cell types. Active targeting can be achieved through surface modifications, such as conjugation of ligands e.g. biotin to poly(D,L-lactide-co-glycolide) nanoparticles to deliver paclitaxel to tumors (Patil et al 2009).

Restrictions are placed on the size of particulates that can be injected into the circulatory system so that controlled release systems designed for intravenous use are typically within colloidal range with diameters less than 500 nm (Gaumet et al 2008). With the increasing research into particulate delivery systems, the understanding of the tissue distribution of colloidal particles following administration into the blood has advanced. Moghimi et al. (Moghimi et al 2001), in a review of long-circulating and target-specific nanoparticles, reported that passive tissue distribution patterns of intravenously administered particles depended on size and deformability of the particles as well as their surface chemistry. Slack et al. (Slack et al 1981) showed polystyrene-divinylbenzene microparticles with diameters 7.4 and 11.6 μm were deposited mainly in the lungs, while particles with diameter 3.4 μm deposited in the liver and spleen following intravenous administration. Smaller nanoparticles (less than 150 nm) appear to be distributed more widely than larger colloids with particles seen in the liver, spleen, bone marrow, bone, heart, kidney and stomach (Hobbs et al 1998; Banerjee et al 2002; Gaumet et al 2008). At tumour sites and sites of tissue inflammation, the spaces between endothelial cells lining blood capillaries are enlarged so that correctly sized nanoparticles can escape the vasculature into underlying tissue in these sites (Jain 1987; Hobbs et al 1998; Tarner & Müller-Ladner 2008). This mechanism is exploited in the targeting of cytotoxic drugs to tumors (Duncan 1999; Torchilin 2000) or anti-inflammatory drugs to sites of tissue inflammation (Tarner & Müller-Ladner 2008). Tumours at different sites of the body, however, appear to have different level of microvasculature porosity, which may affect the accumulation of particulate delivery systems (Hobbs et al 1998). Pore sizes ranging from 200 nm to 1.2 μm were reported with experimental subcutaneous tumours, but lower pore size was reported in brain tumours less than 550 nm. The microvascular pore size varied depending on the tumour cell line (Hobbs et al 1998). At even smaller sizes, particles may be eliminated from the body by filtration through the kidney. Choi et al. suggested that particles less than 5 nm diameter were freely filtered in the kidneys, while those with diameter greater than 5 nm were retained in the body (Choi et al 2007). This suggests that there is also a lower particle size limit (5 nm) as well as the higher one (500 nm) for particulate systems administered via the intravenous route.

# Intramuscular route

The intramuscular site is reached by injection through the hypodermis into the underlying skeletal muscle. The structure of musculature is such that vasculature extends into the muscle and each muscle fibre is surrounded by a number of capillaries lying parallel to each fibre with transverse vessels between muscle fibres (Maried 1995). Thus, muscle tissue is typically highly perfused with blood for the delivery of oxygen and nutrients to muscle cells and for the removal of waste material and so can be utilized for the systemic delivery of therapeutics. Extracellular fluid in skeletal muscle is reported to have a pH of 7.1 at rest but decreases with exercise to 6.8 due to lactate accumulation (Sullivan et al 1994). This slightly acidic pH may influence release and absorption properties of weakly acidic or basic drugs. Lymphatic vessels are also present within the connective tissue that surround the muscle fibres and bundles, however the lymph system is more extensive in the subcutaneous site compared to the intramuscular site (Zuidema et al 1994). Drug characteristics that promote absorption into the lymphatic system are discussed later.

The most common muscles into which injections are made in humans are the gluteus maximus, vastus lateralis and deltoid. Blood flow in these muscles is reported to be fastest in the deltoid, and slowest in the gluteus maximus giving rise to potential differences in drug absorption rates at different sites of administration (Evans et al 1975). Differences in muscle perfusion can be expected to have the greatest effect on drug absorption when uptake into injection site capillaries is the rate limiting step in drug absorption. For example, more rapid absorption of diazepam from injections into the deltoid muscle compared with the vastus lateralis has been reported (Korttila & Linnoila 1975). Additionally, when muscle perfusion is the rate-limiting step for drug absorption, activities that increase local blood flow such as exercise, and local muscle massage may be expected to increase the rate of drug absorption following intramuscular injection (Dandona et al 1978). Increased absorption has been reported for both intramuscular administration of penicillin and diazepam with exercise (Ylitalo 1991). However, when drug release from the depot is slow, then the effects of increased muscle perfusion may not be great. Soni et al. showed no significant effects of injection site, massage or muscle activity on plasma levels from a depot injection fluphenzine decanoate (Soni et al 1988). Muscle activity may also have an effect on the surface area available for drug release for low to intermediate viscosity systems as exercise could affect the spreadability of the extended release depot within the muscle tissue. For example, absorption of the protein aprotinin (Bjerregaard et al 2001) was found to be greater from formulations with a lower viscosity that spread more extensively within the muscle compared to the formulations with higher viscosity, 30% and 60% w/w emulsions, respectively (**Figure 4**).

The amount of fat associated with the muscle can also modulate absorption from intramuscular injections and has been given as an explanation for the slower rate of drug absorption following injection into the gluteus maximus in females compared to males (Buxton 2006). Cockshott et al. have shown that injections intended for intramuscular administration into the gluteal muscle may not reach the muscle and may indeed be made most of the time into the fat surrounding the muscle (Cockshott et al 1982). Their study of 63 men and 60 women indicated that at any given weight, the skin to muscle distance in the gluteal region was approximately 2.5 cm greater in women than in men. If a standard 3.5 mm length needle was used, then under 5% of women would receive the intramuscular injection at the desired depth of at least 0.5 cm into the muscle (Cockshott et al 1982). Hence, the reported slower absorption following intramuscular injection into the gluteus maximus site may be due to injection into fat overlying the muscle rather than muscle itself.

# Subcutaneous route

The subcutaneous tissue, or hypodermis is situated directly below the dermis layer of the skin and exterior to the muscle layer (Washington et al 2001). The characteristic feature of this site is the storage of dietary adipose within loose connective tissue in the interstitial space (Washington et al 2001). The interstitial space comprises a collagen network embedded in a gel of glycosaminoglycans, salts and proteins (Schmid-Schönbein 1990) and has a pH of 7.3 (Kuntz et al 2000). The accessibility of the subcutaneous site and relative ease of injection with short, fine needles contribute to its popularity for self-administration of medications such as insulin. Implants, particulates or *in situ* gelling systems can sit comfortably within the connective tissue of the subcutaneous site and release drug that is then absorbed into surrounding blood or lymph capillaries.

Extended release injections containing contraceptive hormones have been in use since the late 1960s, initially with intramuscular Depo Provera®, a long acting aqueous suspension of medroxyprogesterone acetate administered once every three months (Jeppsson 1972) and more recently a lower dose subcutaneous form, depo-subQ Provera (Jain et al 2004). The duration of effective contraceptive action was further extended with the introduction of levonorgesterel containing polydimethylsiloxane implants. The first of these were the Norplant® implants of the 1980s which gave slow release over five years following subcutaneous administration. Six rod shaped implants were needed for treatment with Norplant, and re-design of this product allowed the number of implants to be reduced to two (Sivin et al 1998). More recently a single rod implant for subcutaneous administration has been developed incorporating etonogestrel in ethylene vinyl acetate (Implanon™) to provide contraceptive effects over a period of three years (Funk et al 2005).

Blood perfusion in the subcutaneous tissue is recognised to be lower than in the intramuscular site, which translates into comparatively slower absorption, lower maximum plasma concentrations and longer times to maximum plasma concentration as illustrated for the antibiotic cefotaxime in sheep (**Figure 5**). A further delay in the appearance of drug within the systemic circulation results if drug is absorbed into subcutaneous lymphatic capillaries because time is required for the drug to transverse the lymphatic system and enter the blood circulation. The lymphatic system has unidirectional flow and maintains interstitial pressure through the collection of fluid and proteins from the interstitial fluid through a series of draining lymph nodes and is eventually returned to systemic circulation (Swartz 2001). Lymph capillaries are more permeable than blood capillaries because they are lined with a single layer of endothelial cells that have an incomplete basal layer and lack coherent tight junctions between adjacent endothelial cells (Schmid-Schönbein 1990). The composition of protein in the lymph is similar to that in blood plasma and lymph flow rate is 100-500 times slower than the flow in blood vessels (Swartz 2001). The importance of the lymphatic system for absorption of drugs is increasingly recognised for large molecular weight protein drugs that do not partition well into blood capillaries (Porter & Charman 2000). Small drug molecules (< 1 kDa) are predominantly absorbed into blood capillaries as their small size means they can partition relatively easily across the capillary endothelium and their diffusion through the interstitial fluid is not restricted (McLennan et al 2005; Larsen et al 2009). For larger peptide and protein compounds, absorption into blood vessels is limited primarily by their poor permeability across the capillary endothelial cell wall due to their large size. Consequently, these remain in the extracellular fluid until taken up by the lymphatic system. Porter and Charman have shown that the fraction of a dose absorbed into the lymphatic system is directly proportional to molecular weight (Porter & Charman 2000). Compounds with a molecular weight greater than 30 kD have been shown to be predominantly absorbed from into the lymphatic capillaries (McLennan et al 2005). Colloidal particles may also be absorbed into the lymphatic system, with an optimal size for uptake reported to be 10-100 nm (Swartz 2001) and larger particles taking longer to be absorbed (Porter et al 2001). Whilst absorption into the lymphatic vessels appears not to be selective, the rate of diffusion of the solute through the extracellular matrix of the interstitial fluid is determined by size, charge and hydrophilicity of the drug and so will influence lymphatic uptake (Porter & Charman 2000; Swartz 2001).

The spread of formulations within the subcutaneous tissue is another potentially important variable affecting *in vivo* drug release as it will have an effect on the available surface area across which drug can escape from the delivery system. The influence of depot spreadability on drug absorption can be demonstrated by the effects of co-administration of formulations with the enzyme hyaluronidase. This enzyme degrades hyaluronic acid within the subcutaneous interstitial matrix and basement membrane, reducing the resistance to flow of injected material through the subcutaneous tissue (Dunn et al 2010). Bookbinder et al. (Bookbinder et al 2006) described the effects on subcutaneous injection spread on co-administration of a recombinant form of hyaluronidase with typan blue dye. The area of spread of the trypan blue dye was dose dependant over 0.05 to 5.0 units hyaluronidase per injection and because the enzyme acts only on the hyaluronic acid and not the collagen network, a significant structural matrix still existed. This enzyme also allowed the spread of small particulates (less than 200 nm) within the subcutaneous tissue (Bookbinder et al 2006).

# Effects of the tissue response on extended release parenteral systems

For many extended release parenteral systems, extensive *in vitro* characterisation is carried out and understanding of the effects of formulation and processing variables on the release characteristics are determined. *In vitro* release methods to study the release of drugs from extended release parenteral delivery systems make only small attempts to simulate the *in vivo* environment and methods used may be categorised as: sample and separate, continuous flow and dialysis based methods as recently reviewed by Larsen et al. (Larsen et al 2009). These methods mimic the poorly-stirred, limited fluid conditions that are expected in intramuscular and subcutaneous sites and little is done to account for the effect of the tissue inflammatory reaction on release characteristics.

For intravenous nanoparticulate systems, protein adsorption to the particulate surface and uptake by the reticuloendothelial system reduce the circulation time within the blood. Much recent work has shown that these effects can be reduced by surface pegylation so that residence time of nanoparticulate systems in the circulation are now significantly extended (Drummond et al 1999). This technology has had wide application in the formulation of particulate targeted delivery systems. Ideally, the drug remains within the particulate system while it is circulating in the blood, but is released following deposition in the target tissue (e.g. tumour). *In vitro* evaluation of particle size and retention of the drug within the nanoparticle are important studies to characterise these delivery systems prior to *in vivo* use. However, it is the rate of *in vivo* particle oponisation, uptake by reticuloendothelial system cells and interactions with microvasculature that will ultimately determine the *in vivo* fate and effectiveness of intravenously injected nanoparticles.

Intramuscular and subcutaneous extended release systems are expected to stay at the administration site and release drug slowly over time to achieve constant plasma drug concentrations as shown in **Figure 6.** For some systems, the biological environment has a large influence on the *in vivo* release profile and understanding how drug release occurs *in vivo* may allow optimisation of *in vivo* release profiles to achieve target plasma drug concentrations. The biological environment can trigger physical change in the injected materials such as the solution to semi-solid transformation occurring for *in situ* gelling systems (Haglund et al 1996; Veyries et al 1999; Matschke et al 2002; Yu & Ding 2008). For others the opposite occurs and a solid material may be implanted, which hydrates to form a semi-solid depot at the injection site (Khan et al 1993; Walduck et al 1998). *In situ* gelling implant materials transition from liquid to solid in response to environmental changes such as temperature changes e.g. poloxomer gels (Katakam et al 1997a; Katakam et al 1997b), or loss of organic solvent into surrounding tissue e.g. polylactide-co-glycolide organic solutions (Jain 2000; Dong et al 2006; Kempe et al 2008). The tissue reaction at the implantation site can additionally influence the *in vivo* performance and may be involved in drug release. Kempe *et al.* described the *in vivo* solidification process for PLGA implants from n-methyl-2-pyrrolidone solutions as a two-stage process involving firstly surface solidification to form a shell over about 30 minutes, followed by a slower process of complete solidification over 24 hours (Kempe et al 2008). In the first stage of solidification they reported about 75% of the polymer precipitated. As well as altering the physical form of the implanted material, the biological environment may affect drug release and implant degradation and erosion rates. An early example was reported by Olanoff et al. for a tetracycline containing methacrylate tri-laminate film system (Olanoff & Anderson 1979; Olanoff et al 1979). These films released tetracycline with a zero-order profile in both *in vitro* studies (Olanoff et al 1979) and following subcutaneous implantation in rats (Olanoff & Anderson 1979). The tissue response one to two weeks after implantation was described as mild inflammatory with tissue oedema, loose granulation tissue and the beginnings of fibrous capsule formation. The fibrous capsule developed further over the following 4 to 9 weeks with dense collagen fibrils and little cellular infiltration (Olanoff & Anderson 1979). *In vivo* release rates of 123 and 158 μg/day for two film formulations were shown to be approximately equal to the tetracycline elimination rates (110 ± 32 and 153 ± 42 μg/day respectively). This resulted in reasonably steady plasma concentrations of tetracycline (0.6 – 1.0 μg/mL) over 3 to 14 days post-implantation for the films with release rate of 158 μg/day. Knowing the *in vivo* release rate (i.e. the drug input rate) allowed development of a pharmacokinetic model for controlled release tetracycline (Olanoff & Anderson 1980). Further to this, Anderson *et al.* went on to describe the diffusional barrier effects of the fibrous capsule to *in vivo* gentamicin release from silicone implants (Anderson et al 1981) and then naltrexone release from polylactide-co-glycolide beads and microparticles (Yamaguchi & Anderson 1992). They showed a well developed fibrous capsule surrounded silicone implants at four weeks. This capsule comprised three zones; one immediately surrounding the implant (approx. 2 μm thickness), then the fibrous capsule (10 – 15 μm thickness) that was surrounded by an outer zone. The outer zone contained blood capillaries but the inner two zones were avascular and therefore these inner two layers may represent a diffusional barrier (Anderson et al 1981). Around polylactide-co-glycolide beads (bead diameter ≈ 1.5 mm) a similar tissue response and fibrous layer formation was observed. In contrast, the tissue reaction to microspheres was an accumulation of inflammatory cells (monocytes, lymphocytes polymorphnuclear neutrophils and foreign body giant cells) over three days. The presence of inflammatory cells decreased by day seven but giant cell numbers increased. Interestingly, the fibrous capsule appeared to take around 2 to 4 weeks to fully develop, which means for systems which are designed to give extended release (i.e. greater than 2-4 weeks) this additional barrier may assist extension of the duration of release. For the polylactide-co-glycolide bead example in Yamaguchi and Anderson’s study, steady plasma levels of naltrexone were achieved from days 3 to 28 (Yamaguchi & Anderson 1992). For a cholesterol-based implant, another group has shown that the fibrous capsule may contribute to uptake of lipid material and hence influence release of drug (Anderson et al 1993). They reported the fibrous capsules surrounding norethisterone-cholesterol implants and interactions of cells within the fibrous capsule with the lipid material. Foam cells were observed in the inner layer of the surrounding capsule containing lipid and their involvement in an absorption mechanism was suggested. Again, the outer layers of the fibrous capsule contained blood and lymphatic capillaries (Anderson et al 1993). These studies confirm the importance of the inflammatory response and fibrous capsule not only as a potential diffusional barrier to drug release from extended release implants but as a more complex biological interface which can potentially enhance or delay drug release from implanted systems.

For particulate systems, particle size influences residence time at the injection site as phagocytic cells will take up particles of appropriate size, shape and surface characteristics. Generally particles with diameters less than 0.5 μm are reported to undergo phagocytosis, however, particle shape and surface characteristics will also influence this process (Simone et al 2008; Hillaireau & Couvreur 2009). The surface properties also appear to influence the tissue response and Daugerty *et al.* have described differences in cellular infiltration between PLGA microparticles depending on their surface hydrophilicity (Daugherty et al 1997). Microparticles prepared using poly-D,L-lactic acid with hydrophobic end groups, formed clumps within subcutaneous tissue in which granulation tissue appeared to be excluded. In comparison, when poly-D,L-lactic acid with hydrophilic end groups was used and the particle surface was more hydrophilic, cellular infiltration was seen between individual microparticles. This suggests tissue structure surrounding particulates is dependant of particle surface hydrophobicity and effects on *in vivo* drug release may be important especially as granulation tissue is rich in blood capillaries.

# Concluding Remarks

Extended release delivery systems are increasingly investigated for administration by the parenteral route. For these systems the biological environment in which they are placed can be shown to affect their *in vivo* drug release characteristics. Understanding specific effects of the tissue response on drug release and delivery system degradation may allow better design of extended release parenteral delivery systems which achieve target *in vivo* release profiles which translate into optimal plasma concentration versus time profiles for therapeutic goal.

Injection/

implantation site

Implant or depot

release

kr

metabolites

bound drug

(tissue or protein)

free drug in tissue

degraded drug

absorption

ka

Blood or lymphatic capillaries

to systemic circulation

drug in plasma (Cp)

ke

drug in tissue

distribution

plasma

protein

bound drug

metabolism

and

elimination

**Figure 1.** Schematic showing pharmacokinetics for a drug administered as an extended release intramuscular or subcutaneous system.

Type 1: kr < ka : device-limited PK

Device or depot

ka

kr

Type 2: kr < ka : perfusion- limited PK

Solution

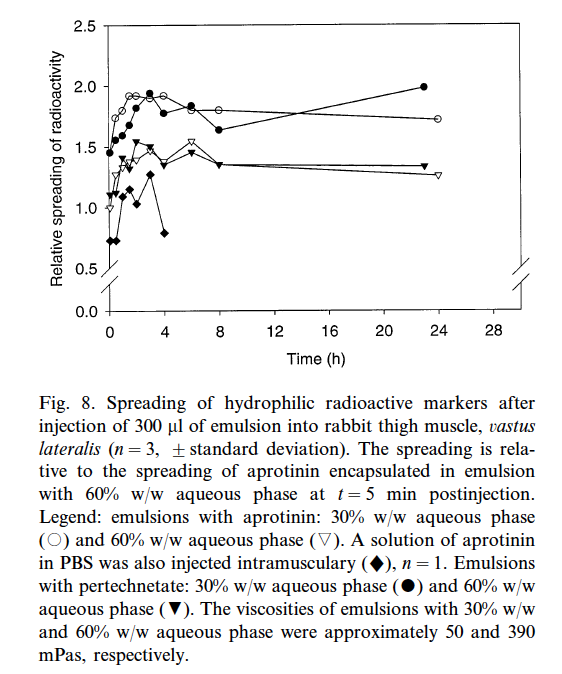
kr

ka

**Figure 2.**  Pharmacokinetic (PK) classification of intramuscular implantable delivery systems. (Modified from Washington et al 2001).



**Figure 3.** Potential sites for injection of extended release parenteral dosage forms.



**Figure 4.**  Graph of the spread following intramuscular injection of hydrophilic radioactive markers into thigh muscle of rabbit. Spreading is represented as relative to aprotinin encapsulated in an emulsion with 60% w/w aqueous phase. ○ 30% w/w aqueous phase emulsion containing aprotinin; ▽ 60% w/w aqueous phase emulsion containing aprotinin; ● 30% w/w aqueous phase emulsion containing radioactive pertechnetate; ▼ 60% w/w aqueous phase emulsion containing radioactive pertechnetate; ♦ solution of aprotinin in PBS administered intramuscularly (Figure with permission from Bjerregaard et al 2001).

**Figure 5.**  Comparative plasma concentrations following injection of an aqueous solution cefotaxime (50 mg/kg) via the (⚫) intramuscular and (⭘) subcutaneous routes in sheep *(Data with permission from Guerrini et al 1986)*



**Figure 6.**  Plasma versus time profile for an ideal controlled release delivery system.

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Figure legends

**Figure 1.** Schematic showing pharmacokinetics for a drug administered as an extended release intramuscular or subcutaneous system.

**Figure 2.**  Pharmacokinetic (PK) classification of intramuscular implantable delivery systems. (Modified from Washington et al 2001).

**Figure 3.** Potential sites for injection of extended release parenteral dosage forms.

**Figure 4.**  Graph of the spread following intramuscular injection of hydrophilic radioactive markers into thigh muscle of rabbit. Spreading is represented as relative to aprotinin encapsulated in an emulsion with 60% w/w aqueous phase. ○ 30% w/w aqueous phase emulsion containing aprotinin; ▽ 60% w/w aqueous phase emulsion containing aprotinin; ● 30% w/w aqueous phase emulsion containing radioactive pertechnetate; ▼ 60% w/w aqueous phase emulsion containing radioactive pertechnetate; ♦ solution of aprotinin in PBS administered intramuscularly (Figure with permission from Bjerregaard et al 2001).

**Figure 5.**  Comparative plasma concentrations following injection of an aqueous solution cefotaxime (50 mg/kg) via the (⚫) intramuscular and (⭘) subcutaneous routes in sheep (Data with permission from Guerrini et al 1986).

**Figure 6.**  Plasma versus time profile for an ideal controlled release delivery system.