



# **Specialist information**

**from the Committee for Animal Welfare Officers  
(GV-SOLAS) and Working Group 4 in the TVT**

## **Recommendation for administration of substances to laboratory animals**

**Status July 2017**

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## 1. Basic principles

This recommendation was compiled for persons conducting or planning animal procedures, animal welfare officers and authorities. It is intended to serve as a reference guide for working in line with animal welfare needs and for standardization of the procedures applied today. If the volumes specified in the table are complied with, the substance administration procedure will generally not exceed a “mild” degree of severity – relative to the volume administered.

The values given in the table outline recommended volumes which may be exceeded if specific experimental demands occur, and higher volumes are approved by authorities. This results in severity levels higher than “mild” and must be considered in the severity assessment.

This recommendation is a recasting of several previous recommendations on administration volumes. The table has been supplemented with additional information and some of the values given here deviate substantially in some respects from those in the previous versions. This is explained by the fact that numerous new findings have been obtained in recent times with regard to administration volumes and their tolerability for animals. The publishers have taken this into account.

The volumes indicated apply to the use of readily absorbable, aqueous solutions (see also the following remarks); they do not apply to immunizations, where it is necessary to work with much lower volumes, e.g., when using Freund’s adjuvant.

## 2. Injection solutions

Requirements for injection solutions:

- isotonic.
- body temperature
- pH neutral (pH 7,0 - 7,3); may possibly be within different pH limits depending on the route of administration.

The **tolerance to the pH value** falls in the following sequence:

***oral > intravenous > intramuscular > subcutaneous***

The concentration, chemical composition and physical properties of the injection solution should be such that no systemic harm or local irritation occurs. Hypertonic and hypotonic solutions or solutions within a non-physiological pH range, e.g., in the case of perivascular injection, result in considerable pain and destruction of tissue as well as erythrocyte damage (haemolysis).

## 3. Routes of administration

While different routes of administration are possible, depending on the objective of the experiment, it is the method which is most protective of the animal that should be selected. With all methods involving injection, prior aspiration is necessary.

**Oral** doses may be administered in the feed or drinking water, by oropharyngeal administration or by gavage.

**Subcutaneous** injections are advisable. Exception: substances that cause local irritation, which must only be administered intravenously.

**Intramuscular** injections are basically painful for most animals. The injection volume should therefore be kept as small as possible, or the injected volume distributed over several injection sites. The substance should be injected slowly, considering the anatomical circumstances (nerve structures!).

**Intravenous** injections should be preferred with all substances that cause local irritation. Care must be taken here to ensure that no air is injected into the vessel (risk of embolism!). Barring a few exceptions, emulsions and particulate substances must not be administered intravenously.

**Paravenous** injections must be categorically avoided, because they lead to very painful venous inflammation/obliteration, as well as necrosis of the surrounding tissue. As a precaution, therefore, the cannula should be inserted far enough into the vessel and its correct position verified by aspiration. If metal needles are used, the vein may be damaged by a sudden movement of the animals, so solutions that cause irritation should be administered via a venous catheter to be on the safe side.

**Intraperitoneal** injections carry the risk of accidental puncture of the abdominal organs (e.g., bladder, intestine, liver or spleen), which can be associated with considerable pain. Puncture injuries of the parenchymatous organs, however, are rare and easily recognizable based on clinical findings. Less obvious are partial or total erroneous injections into the subcutis, the bladder or the gastrointestinal tract, here in particular the caecum. Since the caecum in most rats lies on the left side, the injections should be made on the right side. Peritonitis, adhesions, or foreign body granulomas may occur, especially with repeated i.p. administration or when vehicles are used that cause mild irritation.

**Intradermal** injections must be delivered slowly, because otherwise the objective of distributing the entire fluid in the epidermis cannot be achieved. During injection, the needle must be moved slowly; the epidermis should not be penetrated. As a rule, this type of injection is painful and only very small volumes can be administered.

**Intracerebroventricular** injections must be undertaken under anaesthesia (except in the case of existing implants) and particularly slowly! It is usually performed with special pumps using a stereotactic apparatus. After administration, at least 30 seconds should elapse before the injector is removed from the tissue.).

#### 4. Rate of distribution

The rate of distribution in the animal's body depends on the route of administration and decreases in the following sequence:

***intravenous > intraperitoneal > intramuscular > subcutaneous > oral***

## **5. Recommended volumes for the administration of substances (see Table p. 7)**

The values given in the table are to be seen as per animal or injection site or per kg bodyweight (b.w.). Distinctions are made according to route of administration, and recommended cannula sizes (G) are given.

For new-born and young animals, the number of injection sites or the volume used must be reduced accordingly.

Different conditions apply in the case of immunization and infusion! Further information can be found in relevant publications.

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**Table 1: Average blood volume in different animal species**

Animal Species	ml/kg b.w.							ml per injection site				µl per animal	
	Subcutaneous <sup>1</sup>		Oral bolus	Intraperitoneal <sup>2</sup>		Intravenous <sup>3,4</sup> bolus		intradermal		Intramuscular		Intracerebro-Ventricular <sup>5</sup>	
		G			G		G		G		G		G
Hamster ( <i>Mesocricetus auratus</i> )	5	23-25	10	10	23-25	5	25-27	0.02	27	0.1	24-25	3	28
Chicken	10	22	10	-		1	22	0.05	27	1	22		
Dog (Beagle)	1	21-23	5	1	21-23	2	21-25	0.1	27	3	21-23		
Rabbit	2.5	23-25	10	5	21-23	2	23-25	0.05	27	1	25	80	28
Cat	2	23	10	5	21-23	5	21-25	0.1	27	1	23	80	28
Marmoset ( <i>Callithrix jacchus</i> )	2	23-25	10	5	21-25	2.5	21-25	0.05	27	0.1	23-25		
Macaque	2	21-25	5	3	21-23	2	21-25	0.05	25	2	23-25		
Mouse	10	25	10 <sup>6</sup>	10	25-27	5	26-28	0.02	27	0.05 <sup>7</sup>	27	3	28
Guinea pig	5	23-25	10 <sup>8</sup>	10	23-25	5	25-27	0.1	27	0.1	25	5	28
Gerbil ( <i>Meriones unguiculatus</i> )	5	23	10	10	25	5	25	0.02	27	0.1	24	5	28
Mini pig	1	20	10	1	20	2	20	0.1	27	5	20		
Rat	10	25	10 <sup>6</sup>	10	23-25	5	25-27	0.05	27	0.1	23-25	5	28
Sheep	1	20-23	10	-		2	19-21	0.1	27	4	21		
Bird (zebra finch)	10	27	10	-		5		-		0.05 <sup>9</sup>	26-27		

<sup>1</sup> The injection volume depends on skin mobility and may have to be distributed over several injection sites.<sup>2</sup> In many animal species (e.g. dogs, monkeys, birds) intraperitoneal injections are uncommon. In birds, there is a risk of injecting into the air sac!<sup>3</sup> The injection time for an i.v. bolus should be at least 1 minute and can run to 2.5 minutes.<sup>4</sup> With infusions over 2 hours, the maximum volume should be <10% of blood volume.<sup>5</sup> Glass capillaries or 22G cannula. Pay attention to the especially slow rate of injection (2 – 3 min in mouse and rat)!<sup>6</sup> Maximum 5 ml per rat and 1 ml per mouse; with viscous substances (e.g. oils) not more than 4 ml per rat and 0.5 ml per mouse.<sup>7</sup> This type of administration is not recommended because of the minimal muscle mass.<sup>8</sup> The guinea pig has a very small palatal ostium, which can be easily damaged. Administration by gavage is not recommended.<sup>9</sup> Preferably i.m. administration: contract muscle before injection and relax after injection. With large volumes, possibly s.c. into the fold of the knee.

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