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

March 2017

Authors:
Dr. André Dülsner, Dr. Rüdiger Hack, Dr. Christine Krüger,
Dr. Marina Pils, Dr. Kira Scherer, Dr. Barthel Schmelting,
Dr. Matthias Schmidt, Heike Weinert (GV-SOLAS)
and
Dr. Thomas Jourdan (TVT)
Contents

Basic principles ............................................................................................................................................ 3
Injection solutions ......................................................................................................................................... 3
Routes of administration ............................................................................................................................ 3
Rate of distribution ....................................................................................................................................... 4
Recommended volumes for the administration of substances ................................................................. 4
Literature ...................................................................................................................................................... 5
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.

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 (see TVT fact sheet no. 4).

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.

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).

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, taking into account 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 on the basis of 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.

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*

Recommended volumes for the administration of substances (see table p. 6)
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.
Literature

BVAAWF/FRAME/RSPCA/UFAW Joint Working Group on Refinement (2001) Laboratory birds: refinements in husbandry and procedures. Laboratory Animals 35 (S1)


Diehl KH et al. (2001) A good practice guide to the administration of substances and removal of blood, including routes and volumes. J Appl Toxicol 21: 15-23


Güttner J (1975) Komplikationen und Folgen der intraperitonealen Applikation im Tierexperiment, Die Pharmazie 30: 129-133


Morton DB et al. (2001) Refining procedures for the administration of substances. Laboratory Animals 35: 1-41


### Recommended volumes for the administration of substances

**(volume / cannula size given in each case)**

<table>
<thead>
<tr>
<th>Animal species</th>
<th>ml/kg b.w.</th>
<th>ml per injection site</th>
<th>µl per animal</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Subcutaneous¹</td>
<td>Oral bolus</td>
<td>Intraperitoneal²</td>
</tr>
<tr>
<td></td>
<td>G</td>
<td>G</td>
<td>G</td>
</tr>
<tr>
<td>Hamster (<em>Mesocricetus auratus</em>)</td>
<td>5</td>
<td>23-25</td>
<td>10</td>
</tr>
<tr>
<td>Chicken</td>
<td>10</td>
<td>22</td>
<td>10</td>
</tr>
<tr>
<td>Dog (beagle)</td>
<td>1</td>
<td>21-23</td>
<td>5</td>
</tr>
<tr>
<td>Rabbit</td>
<td>2.5</td>
<td>23-25</td>
<td>10</td>
</tr>
<tr>
<td>Cat</td>
<td>2</td>
<td>23</td>
<td>10</td>
</tr>
<tr>
<td>Marmoset (<em>Callithrix jacchus</em>)</td>
<td>2</td>
<td>23-25</td>
<td>10</td>
</tr>
<tr>
<td>Macaque</td>
<td>2</td>
<td>21-25</td>
<td>5</td>
</tr>
<tr>
<td>Mouse</td>
<td>10</td>
<td>25</td>
<td>10⁶</td>
</tr>
<tr>
<td>Guinea pig</td>
<td>5</td>
<td>23-25</td>
<td>10⁶</td>
</tr>
<tr>
<td>Gerbil (<em>Meriones unguiculatus</em>)</td>
<td>5</td>
<td>23</td>
<td>10</td>
</tr>
<tr>
<td>Mini-pig</td>
<td>1</td>
<td>20</td>
<td>10</td>
</tr>
<tr>
<td>Rat</td>
<td>10</td>
<td>25</td>
<td>10⁶</td>
</tr>
<tr>
<td>Sheep</td>
<td>1</td>
<td>20-23</td>
<td>10</td>
</tr>
<tr>
<td>Bird (zebra finch)</td>
<td>10</td>
<td>27</td>
<td>10</td>
</tr>
</tbody>
</table>

¹ The injection volume depends on skin mobility and may have to be distributed over several injection sites.
² 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!
³ The injection time for an i.v. bolus should be at least 1 minute and can run to 2.5 minutes.
⁴ With infusions over 2 hours, the maximum volume should be <10% of blood volume.
⁵ Glass capillaries or 22G cannula. Pay attention to the especially slow rate of injection (2 – 3 min in mouse and rat)!
⁶ 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.
⁷ This type of administration is not recommended because of the minimal muscle mass.
⁸ The guinea pig has a very small palatal ostium, which can be easily damaged. Administration by gavage is not recommended.
⁹ Preferably i.m. administration: contract muscle before injection and relax after injection. With large volumes, possibly s.c. into the fold of the knee.
Disclaimer

Any use of GV-SOLAS publications (specialist information, statements, booklets, recommendations, etc.) and application of the information contained therein are at the express risk of the user. Neither GV-SOLAS nor also the authors can accept liability for any accidents or damages of any kind arising from the use of a publication (e.g. resulting from the absence of safety instructions), irrespective of legal grounds. Liability claims against GV-SOLAS and the author for damages of a material or non-material nature caused by the use or non-use of the information or by the use of erroneous and/or incomplete information are in principle excluded. Legal claims and claims for damages are therefore excluded. The work, including all content, was compiled with utmost care. However, GV-SOLAS and the authors assume no responsibility and no liability for the currentness, correctness, completeness or quality of the information provided or for printing errors. GV-SOLAS and the authors accept no legal responsibility or liability in any form for incorrect statements and consequences arising therefrom. Responsibility for the content of the internet pages printed in these publications lies solely with the owner of the websites concerned. GV-SOLAS and the authors have no influence on the design and content of third-party websites and therefore distance themselves from all third-party content. Responsibility within the meaning of press legislation lies with the board of GV-SOLAS.