Substance Administration in Laboratory Animals: Routes, Volumes, and Needle Sizes

1. Purpose

The purpose of this guideline is to provide minimum standards for common administration routes and recommended volumes in rodents.

1. Scope

This guideline applies to all personnel administering substances to animals used in biomedical research.

1. Abbreviations/Definitions
   1. Intramuscular (IM)
   2. Intravenous (IV)
   3. Intradermal (ID)
   4. Intraperitoneal (IP)
   5. Subcutaneous (SC or SQ)
   6. Retro-orbital (RO)
   7. Parenteral – administered elsewhere in the body than through the mouth or alimentary canal (i.e., IM, SC or SQ, IV, ID, IP)
   8. Per os, by way of mouth (PO)
   9. Intratracheal (IT)
   10. Intranasal (IN)
2. Guidance
   1. General Administration Guidance

This guideline applies to healthy adult animals. If using immature, aged, debilitated, or otherwise compromised animals, the Attending Veterinarian must be consulted regarding the best options for substance administration**.**

* + 1. The substance and the associated vehicle must be appropriate for the species, route of administration, and purpose of the experiment.
    2. The volume depends on the administration route and the size of the animal. Excessive volume can be harmful, therefore always use the smallest volume possible. The rate of absorption depends on the substance’s solubility and the route of delivery. In general, the absorption rate per route is arranged as follows: IV > IP > IM > SC > PO.
    3. Substances should be room temperature or body temperature prior to administration, especially when large volumes are to be delivered
    4. The smallest possible needle size (highest gauge) should be used considering the dose volume, species, route of administration, viscosity, and speed of injection. The use of the smallest needle possible will limit the amount of trauma to the surrounding tissue. Flexible gavage needles are a recommended animal welfare refinement over rigid metal tubes. Needles and syringes used for parenteral injection must be from a sterile source and limited to one needle use per animal except as indicated herein:

a. Use of a single, initially sterile needle for IP, SQ, or IM injections of multiple animals is strongly discouraged due to cross-contamination concerns. Use of a single needle for up to 5 rodents from the same cage may be permitted only if specifically described and scientifically justified in the IACUC protocol. For each of these cases:

* + - * The same product is used for all injections
      * Any damaged or blunted needles will be discarded
      * If any blood or other fluids is aspirated into the needle, it will be discarded
    1. Syringes should be of the locking type in order to prevent accidental dislodgement which may result in autoinoculation or back spray.
    2. Used needles and syringes must be disposed of properly.
    3. Prior to parenteral injection, the injection site should be free of visible contamination. The intended injection site may require preparation including shaving and skin disinfection.
    4. When using animals for training exercises, it is recommended to administer the minimal volume necessary for each procedure.
  1. Recommended Needle Size and Administration Volumes/Day

The table below provides guidance on injection volumes. If the volumes presented below must be exceeded, consult the AV for additional guidance. Dose volumes must be IACUC approved. For rodents a needle size of 25 gauge to 30 gauge must be used.

| **Route of Administration** | **Mouse** | **Rat** | **Guinea Pig** |
| --- | --- | --- | --- |
| **IM volume** | 0.05 - 0.1 ml/site | 0.1 - 0.2 ml/site | 0.3 ml/site |
|  |  |  |  |
| **ID volume** | 0.1 ml/site | 0.1 ml/site | 0.1 ml/site |
|  |  |  |  |
| **IP volume** | 20-80 ml/kg | 10-20 ml/kg | 20 ml/kg |
|  |  |  |  |
| **IV bolus**  (~1 min)  **IV infusion**  (max rate 3 ml/min) | 5 ml/kg  25ml/kg | 5 ml/kg  25 ml/kg | 1 ml/kg  5 ml/kg |
|  |  |  |  |
| **PO volume** | 10-50 ml/kg | 10-40 ml/kg | 20 ml/kg |
| gavage needle size | 18 G if mouse weighs greater than 30 grams  20-30 G if mouse weighs less than 30 grams | 18-20 G | 13-30 G or  5-6 Fr |
| **SC volume** | 10-40 ml/kg | 5-10 ml/kg | 10 ml/kg |
|  |  |  |  |
| **RO injection volu**me | 150 ul (max one/eye) | n/a | n/a |
| Facial Vein Injection | 50 ul(max) | n/a | n/a |
| IT | 100 ul(max) | 100 ul(max) | See AV |
| IN | 0.03- 0.05 ml/inj. | 0.03- 0.05 ml./inj. | See AV |

* + 1. Diehl KH, Hull R, Morton D et al: A Good Practice Guide to the Administration of Substances and Removal of Blood, Including Routes and Volumes. J Appl Toxicology 21: 15-23, 2001.
    2. Turner PV, Brabb T, Pekow C, Vasbinder MA. Administration of substances to laboratory animals: routes of administration and factors to consider. J Am Assoc Lab Anim Sci. 2011 Sep;50(5):600-13. PMID: 22330705; PMCID: PMC3189662.