Anesthesia Types in Rodents

- Injectable (e.g. ketamine cocktails)
- Volatile gas (e.g. isoflurane)
- Physical (e.g. hypothermia)

Injectable or Inhalation Anesthesia?

Injectable Anesthetics - Advantages
- Simple
- Cheap
- Minimal equipment
- Many routes (IM, SQ, IP, IV)

Injectable Anesthetics - Disadvantages
- Higher morbidity & mortality when compared to inhalants
- Easier to overdose
- Slow and variable absorption
- Longer recovery times
- Variable effects from a single dose
- Strain and gender differences

Mice Strain & Male/Female Sleep Times

<table>
<thead>
<tr>
<th>Strain</th>
<th>Male</th>
<th>Female</th>
</tr>
</thead>
<tbody>
<tr>
<td>CE</td>
<td>90</td>
<td>110</td>
</tr>
<tr>
<td>AKR</td>
<td>80</td>
<td>100</td>
</tr>
<tr>
<td>C57BL</td>
<td>120</td>
<td>140</td>
</tr>
<tr>
<td>DBA/1</td>
<td>220</td>
<td>240</td>
</tr>
</tbody>
</table>

Pentobarbital 60 mg/kg
Inhalant Anesthetics - Advantages

• Usually suitable as sole agents to reach ‘balanced’ anesthesia
• Unconsciousness
• Absence of pain perception
• Blunted reflexes
• Immobility

Inhalant Anesthetics - Disadvantages

• Anesthetic delivery system - $ but eventually cheaper
• Requires a scavenging system
• Requires yearly calibration (unless otherwise specified by manufacturer)

Volatile Anesthetics

• Ether
• Halothane
• Methoxyflurane
• Enflurane
• Isoflurane
• Desflurane
• Sevoflurane
Isoflurane

- Decreases rate of production of cerebrospinal fluid (CSF) while increasing the rate of its absorption
- Most commonly used gas anesthesia in rodents

Isoflurane - Advantages

- Rapid induction & recovery
- Can manipulate depth of anesthesia easily & rapidly
- Non-irritant, non-explosive & non-flammable
- Main advantage: Nearly 100% eliminated in exhaled air... minimal interference in drug metabolism
- Inexpensive... eventually

Isoflurane - Disadvantages

- Some respiratory depression but cardiovascular (CV) depression is minimal
- Pungent odor may lead to breath holding in rabbits, but does not appear to be a problem in other spp
- Decreases CSF production / increases CSF absorption - A factor in central nervous system (CNS) studies???

Sevoflurane - Advantages

- Rapid induction & recovery
- Major advantage: Nonirritating to airways and well accepted, lending itself to mask or chamber inductions
- Becoming increasingly more popular in rodents

Sevoflurane - Disadvantages

- Undergoes slightly greater hepatic metabolism than isoflurane
- Cost has been higher than isoflurane... but price coming down now that it is off patent (generics)

Anesthetic Gas Recovery as Metabolite

- Methoxyflurane: 50%
- Halothane: 20-25%
- Sevoflurane: 2-5%
- Enflurane: 2.4%
- Isoflurane: 0.17%
- Desflurane: 0.02-0.2%

1Humans
Somnosuite (Kent Scientific)

- Pre-warmed isoflurane gas
- Tissue oxygen-hemoglobin concentration (SpO₂)
- Heart rate
- Respiration rate
- Ventilator
- Superior data
- Warming gas

The Savings

<table>
<thead>
<tr>
<th>Somnosuite</th>
<th>Traditional</th>
</tr>
</thead>
<tbody>
<tr>
<td>Isoflurane</td>
<td>×</td>
</tr>
<tr>
<td>Waste Gas Canister</td>
<td>×</td>
</tr>
</tbody>
</table>

Physiosuite (Kent Scientific)

- Homeothermic (warms rodent at exact temp through far infrared warming that goes beyond cutaneous warming by heating deep into rodent’s body with temp feedback)
- Pulse Oximeter and Heart Rate
- Automatic Ventilator (ventilate animals from 3g to 500g, enter weight and press run)
- End Tidal CO₂ Monitor

Recovery times (min) in rats after 1 hour of anesthesia - Eger and Johnson, 1987.
Open Drop Method of Isoflurane Administration

- Although current practice is to vent out (e.g., fume hood), 2009 publication has shown this method may be safe on tabletop w/o scavenging, [http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2694708/](http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2694708/)
  - Check with your Safety Department
- Used for short term anesthesia (vs. calibrated vaporizer)
- Prolonged use = deaths

Open Drop

- Pathology cassette with isoflurane soaked material
- Centrifuge tube
- Depth of anesthesia can be controlled by moving the nose cone closer or farther away from the nostrils
- Wire or plastic grate
- Absorbent material

Open Drop Formula

<table>
<thead>
<tr>
<th>Volume of liquid agent/1000 ml chamber volume</th>
<th>Approximate concentration of isoflurane</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.05 ml</td>
<td>1%</td>
</tr>
<tr>
<td>0.1 ml</td>
<td>2%</td>
</tr>
<tr>
<td>0.2 ml</td>
<td>4%</td>
</tr>
<tr>
<td>0.3 ml</td>
<td>6%</td>
</tr>
</tbody>
</table>

Replenish isoflurane every 3 mice

Pentobarbital - Advantages

- Placental transfer of barbiturates occurs rapidly. However, when used in proper induction doses, excessive depression of the fetus does not occur
- Administered IV or IP
- Neither hepato- or nephrotoxic
Pentobarbital - Disadvantages

- Pharmaceutical grade very pricey
  - High cost renders it “unavailable in pharm grade” ***
- Chemical grade use must be addressed & justified
  *** NIH statement 2013

Pentobarbital - Disadvantages

- Prolonged recovery esp. if additional doses administered
- Severe CV & resp depression
  - Anesthetic & lethal doses close to each other... high mortality possible

Ketamine

- Exact mechanism of action not established
- Thought to be a specific antagonist of N-methyl-D-aspartate glutamate receptors (NMDA)
- “Cocaine-like” effect in that it inhibits uptake of catecholamines into postganglionic sympathetic nerves
- Cleared by hepatic metabolism
- Alone it is a poor anesthetic and analgesic in rodents (when used alone)... must be used with other agents (‘cocktail’)

Ketamine

- CV effects resemble sympathetic nervous stimulation - i.e. increased arterial BP, HR, CO, cardiac workload & myocardial oxygen consumption
- Such effects obtunded by prior or co-administration of tranquilizers or sedatives (‘cocktail’)
- Transient increase of norepinephrine & epinephrine in plasma

Ketamine - Advantages

- In hypovolemic patients, arterial BP is maintained with ketamine because of peripheral vasoconstriction
- No significant effect on hepatic or renal function
- Useful in ‘cocktail’ preps with sedatives & tranquilizers
- Increases myocardial contractility

Ketamine - Disadvantages

- Increased airway & salivary secretions in some species (mild in mice & rats)
- Induces epileptiform bursts in thalamus & limbic system, but w/o spread to cortical areas - may increase seizure threshold in rats & mice
- Some respiratory depression following anesthetic doses in rodents
Ketamine ‘Cocktails’
- Safer, more ‘balanced’ anesthesia than pentobarbital or ketamine alone
  - Ketamine/xyllazine
  - Ketamine/dexmedetomidine
  - Ketamine/xyllazine/acepromazine
  - Other combinations

Ketamine ‘Cocktail’ Reversal
- Antagonist atipamezole shortens recovery of ketamine/xyllazine & ketamine/dexmedetomidine
- Atipamezole (0.5-1 mg/kg SC, IP, IM, IV) partially reverses xyllazine & dexmedetomidine (not ketamine)
- Early reversal (10-20 minutes after induction) associated with undesirable behavioral disturbances due to effects of ketamine

Ketamine ‘Cocktails’
- Use of yohimbine for reversal of ketamine/xyllazine no longer recommended due to yohimbine’s reported side effects
- For cocktail doses refer to http://vpr.utsa.edu/files/larc/RodentSurgeryApplicationhandouts.pdf

Avertin
- Combination of 2,2,2 tribromethanol & tertiary amyl alcohol (amylene hydrate)
- Adequate anesthesia up to 30 min
- Repeat doses not recommended due to abdominal irritation & peritonitis reports
- Degrades in presence of heat or light - refrigerate, wrap in foil
- Non-pharmaceutical grade compound

Urethane
- Ethyl ester of carbamic acid

Urethane - Advantages
- IP Administration results in long-lasting unconsciousness of 6-10 hr
- Spinal reflexes, neural transmission and cardiopulmonary function minimally affected
- Good analgesia for surgery in rodents
- Stable CV & respiratory functions
Urethane - Disadvantages

- Mutagen/carcinogen
- Readily absorbed through skin
  - initiates pre-neoplastic changes in the skin
  - targets multiple organs
  - suppresses bone marrow
  - readily crosses the placenta
  - induces fetal tumor formation (in utero)

Urethane - Disadvantages

- Strict guidelines (gloves, mask, prepare in fume hood)
- Requires safety approval
- Non-pharmaceutical grade compound
- Not used for survival surgeries

Resources

Presentations, References & Useful Notes
http://research.utsa.edu/research-funding/laboratory-animal-resources-center/training/

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