

2.2. Mouse dobutamine stress survival assay

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A. OBJECTIVE

To evaluate the ability of mice to survive dobutamine stress on heart.

B. CAUTIONS

- Dobutamine is sensitive to light. It is recommended to make it fresh for using in mice. When diluted to the desired concentration, you may store it at 2-8°C, protected from light.
- Dobutamine is administered at a 5 µg/g body weight dose. It is important to measure the mouse body weight prior to the surgery procedure.
- Dobutamine administration is done in mice under anesthesia (usually mouse is attached to a ventilator following tracheostomy). It is important to closely monitor the heart rate and left ventricular pressure at all times during the procedure.
- Mouse heart rate is dependent on the body temperature and under anesthesia, body temperature can decline. It is very important to monitor and maintain the body temperature at 36-38°C. You may use a heating pad and a heat lamp to maintain the temperature.

C. MATERIALS

- Dobutamine Hydrochloride (Sigma Aldrich Cat. No D0676)
- 0.9% Saline
- 0.5 CC insulin syringe
- Isoflurane anesthesia system and a Rodent ventilator
- Oxygen tank
- Surgical tools and sutures for the surgery

RECIPES:

1µg/µL dobutamine solution:
1.0 mg dobutamine hydrochloride
1000 µL of 0.9% NaCl

D. METHODS

1. Weigh each mouse and prepare the appropriate amount of dobutamine.
Dobutamine is administered intraperitoneally at a 5 µg/g BW dosage.
Example: A mouse weighing 30g get a 150µL of 1µg/µL dobutamine
2. Allow the mouse to stabilize under anesthesia. Measure any baseline data when the mouse reaches stability (e.g. LV pressure/ volume etc).

3. Administer Dobutamine intraperitoneally using a 0.5cc insulin syringe. Make sure the needle is not inserted to any of the organs.
4. Start timing immediately after dobutamine administration. Set the timer to 5 mins. You will observe the mouse heart rate increase both within first 30-45 seconds. Record the tracing (ECG or PV-loop) during the whole time.
5. At the end of the 5 mins, set the timer for additional 10 mins and continue to monitor the mouse and ECG or PV-loop recording.
6. The death is diagnosed if heart rate reaches ≤ 250 bpm and the systolic pressure reaches ≤ 30 mmHg.

E. EVALUATION AND INTERPRETATION OF RESULTS

1. Record the amount of time between dobutamine administration and death for each mouse.

F. REFERENCES

1. Bostick B, *et al.* (2009) Cardiac expression of a mini-dystrophin that normalizes skeletal muscle force only partially restores heart function in aged Mdx mice. *Molecular Therapy: The Journal of the American Society of Gene Therapy* 17(2):253-261.
2. Bostick B, Yue Y, & Duan D (2011) Phenotyping cardiac gene therapy in mice. *Methods in Molecular Biology (Clifton, N.J.)* 709:91-104.