

### Dobutamine Stress Echocardiography.v1

This is a protocol for assessment of cardiac function via echocardiography with dobutamine as a stress to mimic exercise. The dose of dobutamine (2.5 mg/kg) is based on many published data ranging from 1-10 mg/kg and our testing.

#### Procedures:

1. **Perform baseline M-mode echo** following the echocardiography protocol.
2. **Inject the mouse** with a dose of 2.5 mg/kg (10  $\mu$ l/g body weight, i.p.) dobutamine working solution (0.25 mg/ml) and start a stopwatch to track time. For example, a 30 g mouse will be injected with 300  $\mu$ l of Dobutamine working solution. Hence, 0.3 ml x 0.25 mg/ml /0.03 kg = 2.5 mg/kg.
3. **Acquire an echo image every minute** for 15 minutes (15 images post dobutamine plus one baseline image).
4. Follow the echocardiography protocol to export and analyze the images.

#### Reagent Preparation:

Dobutamine stock solution (10 mg/ml)

1. **Add 1 ml dH<sub>2</sub>O** to the bottle containing 10 mg dobutamine hydrochloride powder (Sigma, D0676) to final concentration of 10 mg/ml).
2. **Mix well** by pipetting.
3. **Aliquot in 50  $\mu$ l**-aliquots (500  $\mu$ g each) in light-protected Eppendorf tubes with clear labels for the date, volume, concentration and initials of the preparer.
4. **Store the dobutamine** stock solution in -70 °C freezer. Stable for at least 6 months.

Dobutamine working solution (0.25 mg/ml)

1. Add 1950  $\mu$ L of saline to a 50  $\mu$ L-aliquot of Dobutamine stock solution to make Dobutamine working solution (0.25 mg/ml) right before use.
2. Keep the Dobutamine working solution at room temperature with aluminum foil protection between the use for each mice.

#### References:

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