

## antigen (OVA) induced asthmatic animal model

### 1. Immunization and challenging

6-8 weeks old mice are sensitized with ovalbumin (OVA)/Alum (50 µg/10 mg/mouse in 200 µl PBS) intraperitoneally on 3 occasions 7 days apart. One week after the third IP immunization, mice are anesthetized with isoflurane and given 50 µl of OVA intranasally (1mg/mouse) once a day for three consecutive days.

### 2. Airway response evaluation

24 hours after third challenge, mice are anesthetized with etomidate (28 mg/kg body weight), tracheostomized and ventilated in a body plethysmograph. The mice are paralyzed with pancuronium bromide (0.1 mg/kg, i.p.), an indwelling tail vein catheter is placed, and lung resistance is measured after administration of increasing concentrations (dose range: 0.03, 0.1, 0.3, 1 and 3 µg/g body weight) of intravenous acetylcholine.

### 3. Other samples collection

Bronchoalveolar lavage fluid is collected by performing five 0.8 ml cold PBS lavages. Lungs are fixed with 10% buffered formalin instilled at an inflation pressure of 25 cmH<sub>2</sub>O.

