INTRODUCTION
Phenotyping of genetic animal models in the early neonatal period has become an important part of developmental studies. Unfortunately, in many strains of transgenic mice, pups die few hours after birth due to the lack of adaptation to extra-uterine life or severe respiratory failure. In these strains, physiological and therapeutic testing cannot be carried out during the period of neonatal development.

We tested the ability of a prototype of negative pressure ventilator to deliver small tidal volume at a high respiratory rate, close-fitted to newborn mice ventilatory pattern. The first aim of this pilot study was to test the ability of this new ventilator to resuscitate newborn mice after a cardiorespiratory arrest artificially induced. We also investigated the long term consequences of this resuscitation on physiological development.

MATERIAL AND METHODS
Negative Pressure Ventilation (NPV)
The NPV is composed of two polycarbonate cylinders, thoracic and head chambers, separated by a plastic ring maintaining a latex collar (Figure 1). The collar was made of a 30 mm dental dam disc with a 5 mm central hole. The head of the animal was inserted in the collar by carefully pushing the rear of the pup. The thoracic chamber was connected to an intermittent negative pressure generator, which alternatively connects the chamber to the atmosphere and to a low-pressure vacuum source (-6 cm H2O) at 2 Hz.

Pneumotachography
The pneumotachograph is composed of a polycarbonate cylinder (ID: 1mm, OD: 3 mm, L: 15 mm) with two pressure ports (ID: 0.3 mm, D: 10 mm). The pneumotachograph was calibrated before each session using a built-in pump incorporating a gas-light micro-syringe. We used the pneumotachograph to measure forced breathing during negative pressure ventilation, by connecting the pneumotachograph to a plastic mask maintained on the snout.

Pneumotachography in Air or Isoflurane
Spontaneous and forced breathing are monitored by Head-Out Plethysmography and Pneumotachography respectively.

RESULTS
Resuscitation
In the resuscitated group, 90 % of the pups survived the experiment (45 out of 50), versus 25 % in the non-resuscitated group (12 out of 48). Survival was significantly different between the two groups (p = 0.06 x 10^-4).

Physiological testing
At P2 and P7, the resuscitated group and control groups had similar baseline breathing pattern. The first ventilation (V1) at P2 was followed by a second ventilation (V2) 15 minutes later.

Blood gases analysis
At P35, blood gases analysis showed similar levels between the resuscitated and control groups (besides).

CONCLUSION
In this study, we demonstrated that intermittent negative pressure ventilation is feasible in mice at birth, and allows resuscitation in a model of cardiorespiratory arrest. We also showed that resuscitation by negative pressure ventilation does not induce severe sequellae.

This neonatal ventilation device has numerous applications in the field of resuscitation and ventilatory assistance of murine genetic models during the neonatal period. Also, the combination of a cardiorespiratory arrest model with resuscitation opens the way to the preclinical testing of therapeutic strategies for paediatric cardiac arrest.