

# High-resolution respiratory inductive plethysmography in rats: validation in anesthetized conditions.

T Flénet, J Fontecave-Jallon, P-Y Guméry, C Eynard, F Boucher, P Baconnier, S Tanguy

Univ. Grenoble Alpes, CNRS, TIMC-IMAG Laboratory UMR 5525, PRETA team, F-38041 Grenoble, France

E-mail: [timothe.flenet@univ-grenoble-alpes.fr](mailto:timothe.flenet@univ-grenoble-alpes.fr)

## Abstract

Respiratory monitoring is often required in experimental physiological and pharmacological studies in rodents. Currently, the mostly used techniques are direct measurement of airflow on intubated animals and whole body plethysmography. Although the reliability of these methods has been broadly demonstrated, they also have several drawbacks such as invasiveness, high cost of use or confinement of the animals. Respiratory Inductive Plethysmography (RIP) is a non-invasive technique already used in medium-sized mammals that has not yet been evaluated in small rodents. The implementation of inductive plethysmography in rats represents an instrumental challenge because of the small inductances that are expected. A rodent-specific RIP apparatus has been developed and compared to direct airflow measurement provided by a pneumotachograph (PNT) considered as the invasive gold standard for respiratory monitoring. The experiments were carried out on anesthetized rats artificially ventilated at different levels of tidal volumes ( $V_T$ ) covering the whole physiological range. Based on the Euclidian distance between signals, this study shows that after calibration, signals from RIP fit at 93% with PNT values. The Bland & Altman plot evidences differences between RIP and PNT lower than 20% and the values obtained are highly correlated ( $R = 0.98$ ,  $p < 0.001$ ). This study demonstrates that it is possible to design RIP systems suitable for measurement of tidal volumes and airflow in anesthetized rats. Further studies will now be focused on the validation in extended physiological conditions.

Keywords: non-invasive, respiratory monitoring, inductive plethysmography, rats, tidal volume, airflow, pneumotachograph, preclinical studies.

## 1 Introduction

Functional pulmonary parameters are commonly used in preclinical research either as experimental parameters or as vital sign indicators. Respiratory diseases are one of the 5 leading causes of death in the world (World Health Organization - WHO 2014) and the development of new treatments targeting these diseases largely relies on pathophysiology and efficacy preclinical studies. As a vital function, assessment of the respiratory system is a mandatory endpoint in the preclinical safety evaluation of new pharmaceutical compounds (ICH Expert Working Group 2000).

Rodents are the most common animal models in preclinical research. Among them, rats represent a significant part (European commission 2013). Existing non-invasive respiratory monitoring standard techniques in rats are derivatives of plethysmography chambers such as Unrestrained Whole Body Plethysmography (UWBP), Double Chamber Plethysmography (DCP) and Head Out Plethysmography (HOP) (Hoymann 2012). These techniques enable measurement of flow- and volume-derived parameters in more physiological conditions than invasive methods. The main drawbacks are the previous animal acclimation-time to reduce the stress of being confined in a box (Hoymann 2007) and the long thermal stabilization period to achieve proper precision. In addition, the hermetic compartment does not allow access to the animal during the experiment.

Non-invasive methods have been demonstrated to allow proper monitoring of the respiratory system and are widely used in rodents particularly in safety pharmacology and toxicology (Hoymann 2012). As recommended by the Safety Pharmacology Society, achieving lower invasiveness and impact on the animals is desirable as it contributes to improving sensitivity of experiments (Leishman *et al* 2012). For instance, avoiding surgery and minimizing the stress induced by the protocol help phenotype preservation of fragile animals. Non-invasive approaches may also be beneficial regarding simplification of experimental setup as illustrated by the works of Cleary *et al* on non-invasive respiratory monitoring in neurology (Cleary *et al* 2012). Finally, the advantage of reducing invasiveness is not only to increase the relevance of the data collected but also to reduce ethical concerns.

In medium-sized mammals such as dogs, monkeys, pigs, sheep or rabbits, another technique known as Respiratory Inductive Plethysmography (RIP) is frequently used (Murphy *et al* 2010, Maucotel *et al* 2010, Brazelton III *et al* 2001, Hepponstall *et al* 2012). RIP is already embedded in commercial garments dedicated to clinical and preclinical research. It relies on reconstruction of chest volumes from abdomen and thorax cross-sectional area-changes during respiratory cycles. This non-invasive and electrode-free method is convenient for use on animals with fur but to our knowledge it has not yet been applied to rats. Our purpose is to evaluate RIP potential as a non-invasive monitoring method in rats. Considering the physiological range of respiratory tidal volumes in rats, a scale gap precludes direct use of existing RIP systems in these animals. With tidal volumes being approximately 200 times smaller (2.5 to 3.5 ml) than in humans (500 to 1000 ml), higher instrumental performances are required to implement inductive plethysmography in rats. Because existing RIP systems are not adapted to properly measure rat respiratory volume variations, a dedicated inductive plethysmographic device has been developed (DECRO) including an instrumented plethysmographic jacket and a high-resolution inductance analog to digital converter. The present work aims at demonstrating interchangeability of the RIP with a reference method of quantitative respiratory monitoring. Pneumotachograph (PNT) has been chosen as it allows a direct and calibrated measurement of airflow (Hoymann 2012, 2007). The validation protocol consists in simultaneously monitoring respiratory function with PNT and RIP on anesthetized and artificially ventilated rats.

## 2 Materials and protocol

### 2.1 Respiratory inductive plethysmography (RIP) system—DECRO

#### 2.1.1 Measurement principle and instrumental challenge in rats

RIP is a broadly used technique in humans when non-invasive respiratory monitoring is desirable. RIP systems generally consist of two transducers placed on the thorax and the abdomen that enable monitoring of cross-sectional changes of these compartments (Konno and Mead 1967, Bloch *et al* 1998). Volumes of the chest are reconstructed from linear combination of thoracic *THX* and abdominal *ABD* signals.

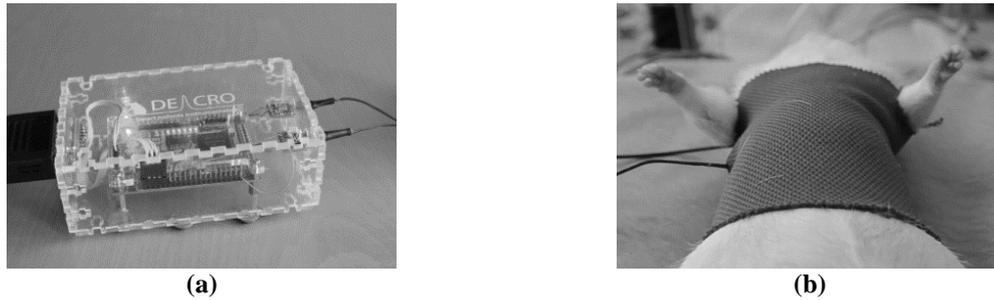
The underlying physical principle is that for a uniform and fixed orientation of the electromagnetic field the self-inductance of a conductor ( $L_{coil}$ ) linearly depends on the surface enclosed ( $S_{coil}$ ):  $L_{coil} = M * S_{coil}$  (Brüllmann *et al* 2009). Considering a coil enclosing the chest, self-inductance variations mainly stem from the fluid exchanges caused by respiratory activity. The coils integrated in RIP systems are generally made up of an insulated wire sewed in a sinusoidal pattern onto an elastic fabric. In several RIP systems, inductance measurement relies on a LC oscillator whose resonant frequency is determined by the value of the coil inductance (Zhang *et al* 2012). LC oscillators are known to be governed by the following relation:  $F_{osc} = 1/(2\pi\sqrt{L_{coil}C})$ . The oscillation frequency ( $F_{osc}$ ) depends on the equivalent resonant constant capacitor ( $C$ ) in parallel with the inductance of the coil ( $L_{coil}$ ). Little variations of  $L_{coil}$  occur because of the chest section variations ( $S_{coil}$ ) due to ventilation and provoke shift in  $F_{osc}$ . In numerical RIP systems  $F_{osc}$  (or its variations) is digitized, and allows computing of  $S_{coil}$  variations.

The design of the LC oscillator and the digitizer becomes increasingly complex when scaling down. Indeed, the self-inductance of the coil is proportional to the area enclosed, which mainly depends on the size of the lungs. In humans, typical total lung capacity is 6 liters compared to 12 ml for an adult rat (Krinke *et al* 2000). Thus, the chest volume of the rat is approximately 500 times smaller than in humans. As a consequence, a drop in the transducer inductance is observed from a few microhenries for human jacket coils to a few hundred nanohenries in rats. These small inductance values complicate oscillator design because of their lower quality factor (Tooley 2006, Zhang *et al* 2012). Considering the amplitude of the respiratory signals in rats, volume variations are expected to be approximately 200 times smaller (2.5 to 3.5 ml) than in humans (500 to 1000 ml). As a consequence, the amplitude of frequency variations  $dF_{OSC}$  will be smaller, as well as section variations  $dS_{coil}$ . Proper acquisition of this signal requires a digitizer with higher resolution. Additionally, respiratory frequencies in rats (around 1 Hz) are five times higher than in humans (0.2 Hz). Then the bandwidth setup and the sampling frequency of systems dedicated to humans (electronics, signal processing, etc.) are not suitable for proper monitoring in rats and have to be accordingly adapted.

#### 2.1.2 Developed device

The high-resolution RIP device developed consists of a plethysmographic jacket and an acquisition system (Fig.1). The acquisition system implements the function of an inductance to digital converter (LDC). Commercial inductance measurement integrated circuits such as LDC family (Texas Instruments, Dallas, USA) offer resolution up to 24 bits, but they are unsuitable with such small values of inductance. A high-resolution analog to digital inductance converter (HR-LDC) has therefore been designed. The HR-LDC is composed of an oscillator optimized for small inductance and a 32bit digital ultra-high resolution frequency measurement system with 1ppm resolution. The jacket is made of breathable and elastic fabric that guarantees constant fit of the coils to the trunk of the rat. It has a cylindrical shape and two holes for the front paws to improve reproducible placement of the sensors on the animals. Two coils made up of highly flexible insulated wire are sewed in a sinusoidal pattern onto the fabric. Sensor location is defined to enclose thoracic and abdominal sections. These transducers are

connected to the HR-LDC. Data are fetched through a RS232 serial bus by an acquisition program executed on a computer. The system is powered through the USB port of the computer.



**Figure 1 DECRO RIP system: (a) High-Resolution inductance to digital converter to which the coils are connected. (b) An anesthetized rat wearing the plethysmographic jacket.**

## 2.2 Experimental setup and recorded signals

### Animal Preparation

Experiments were conducted in accordance with the Directive L276-33 2010/63/EU on the protection of animals used for scientific purposes and the recommendations from the Declaration of Helsinki. The protocol received the agreement APAFIS#5099-2016041916128833 v3.

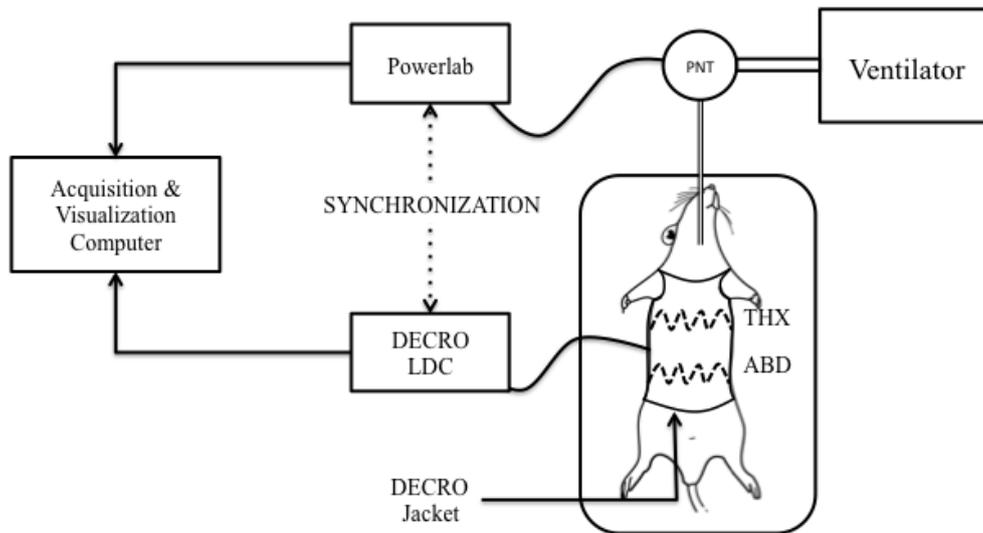
Nine healthy 4-month-old male Wistar rats (Charles River) weighing 350–400g were anesthetized with an intraperitoneal injection of a mixture of ketamine (50 to 75 mg/kg) and xylazine (10 to 15 mg/kg). Animals were dressed with DECRO plethysmographic jacket as shown in Fig.1.b before intubation via tracheostomy. Mechanical ventilation was carried out with a Harvard Model 683 Small Animal Ventilator (Harvard Apparatus, Holliston, Massachusetts USA). Anesthesia level during the experiment was maintained with a mix of air (78%), O<sub>2</sub> (20%) and Isoflurane (2%).

### Physiological signal measurement

Cross sectional changes of *THX* and *ABD* were recorded at 100 Hz with the RIP apparatus developed in the laboratory (DECRO) as illustrated in Fig1.a

Reference measurement of the respiratory flow  $\dot{V}(t)$  was performed with a pneumotachograph (Zephyr flowmeter—Model HAFBLF0750CAAX5, Honeywell, USA) inserted between the ventilator and the endotracheal cannula and mentioned as PNT in Fig.2. This sensor is especially designed for medical applications such as ventilators and anesthesia machines. The sensor has been soldered on a self-made printed circuit board mounted into a custom Plexiglas box to protect it. It was powered with a 5 V DC supply from a standard laboratory supply (Model 72–10480, Tenma). The Zephyr provides an analog  $\pm 750$ scm (standard cubic centimeters per minute) full-scale calibrated and temperature compensated flow signal. The Zephyr resolution is 12 bits, corresponding to 0.37 ml/min and the response time is 1 ms. The flow signal is acquired at 100 Hz with a PowerLab acquisition system and LabChart Software V7 (ADIstruments Pty Ltd).

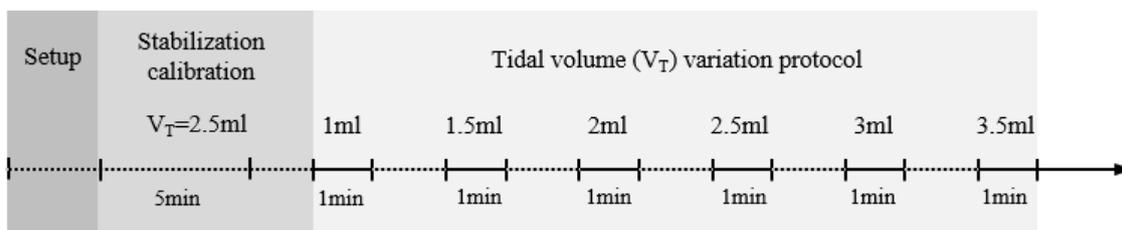
As indicated in Fig.2, a trigger signal connected to one of the inputs of the PowerLab ensured synchronization between the PowerLab and the RIP system.



**Figure 2 Experimental setup.** Pneumotachograph (PNT) is the flowmeter inserted between the ventilator and the tracheal cannula. Flow signal is acquired with a PowerLab (ADInstruments Pty Ltd). Inductive plethysmography thoracic (THX) and abdominal (ABD) cross sectional variations are recorded using DECRO RIP system.

### Protocol

As shown on Fig.3, after animal preparation, standard basal ventilation conditions (2.5 ml—60 cycles per min - cpm) were maintained for 5 minutes to ensure proper stabilization. This step was used for RIP calibration. Six levels of tidal volume ranging from 1 ml to 3.5 ml by 0.5 ml increments were then imposed. Respiratory rate was constant throughout the protocol. Each step was maintained for one minute. Between each increment a stabilization time was added to allow proper volume ventilator adjustments. Volume was controlled by both the PNT and the visual indicator of the ventilator. Volume levels applied during the protocol have been deliberately defined on a range slightly exceeding normal physiological conditions to ensure validation over the whole range of tidal volumes.



**Figure 3 Experimental protocol.** Solid horizontal lines represent measurement times.

## 3 Methodology

Signal processing and data analysis were performed using *MATLAB* (MathWorks, Natick, MA, USA). The following notation convention was adopted:  $V$  referred to volumes and  $\dot{V}$  to flows.

### 3.1 Signal processing

A reference respiratory volume signal  $V_{PNT}(t)$  was integrated from the respiratory flow  $\dot{V}_{PNT}(t)$  given by the pneumotachograph.

Respiratory *ABD* and *THX* signals were first filtered with a numeric low pass Finite Impulse Response (FIR) filter (Cut-off frequency  $F_C = 10$  Hz, Order = 100). Instant trunk volume variations  $V_{RIP}(t)$  were estimated from a linear combination of two compartments *ABD* and *THX* as defined by Konno and Mead (Konno and Mead 1967).

$$V_{RIP}(t) = \alpha ABD + \tau THX$$

$\alpha$  and  $\tau$  were estimated using a least squares optimization on the Euclidian distance separating the reference  $\dot{V}_{PNT}(t)$  and  $\dot{V}_{RIP}(t)$  signals using the method proposed by Loveridge *et al.* (Loveridge *et al* 1983). The RIP flow  $\dot{V}_{RIP}(t)$  was calculated from the derivative of  $V_{RIP}(t)$  filtered with a low pass FIR filter ( $F_C = 25$  Hz, Order = 100). As detailed by Eberhard *et al.* (Eberhard *et al* 2001), optimization was calculated over a 30 second standard stationary condition period ( $V_T = 2,5$  ml — RR (Respiratory rate) = 60 cpm) taken at the end of the stabilization period (marked as Stabilization calibration on Fig. 3, see also the paragraph “Protocol” of chapter 2.2).

### 3.2 Comparison between RIP and PNT data

#### Goodness of fit between RIP and PNT flows

The distance between the flows given by the two measurements was calculated in order to assess flow signal reconstruction quality. The Euclidian distance  $\rho$  between  $\dot{V}_{RIP}(t)$  and  $\dot{V}_{PNT}(t)$  was used as a fit goodness indicator as in similar studies in humans (Eberhard *et al* 2001):

$$\rho = 1 - \frac{\sum_{i=1}^N (\dot{V}_{RIP}(t_i) - \dot{V}_{PNT}(t_i))^2}{\sum_{i=1}^N (\dot{V}_{PNT}(t_i) - \overline{\dot{V}_{PNT}})^2}$$

$\overline{\dot{V}_{PNT}}$  is the average of  $\dot{V}_{PNT}(t_i)$  over the whole recording, N corresponds to the total number of samples of the datasets and  $t_i$  refers to the current sample.

#### Estimation of tidal volumes

Tidal volumes from RIP ( $V_{T,RIP}$ ) and PNT ( $V_{T,PNT}$ ) were calculated by computing the differences between consecutive maxima and minima respectively on  $V_{RIP}(t)$  and  $V_{PNT}(t)$ . For each step of the protocol (times marked by solid lines in Fig.3), an averaged value was computed per animal on a timeframe of 20 seconds. RR (Respiratory rate) was also calculated by measuring the inverse of the period that separates two consecutive maxima on  $V_{RIP}(t)$  and  $V_{PNT}(t)$ . RR is expected to be invariant as frequency of the ventilator was maintained constant and is not used for analysis.

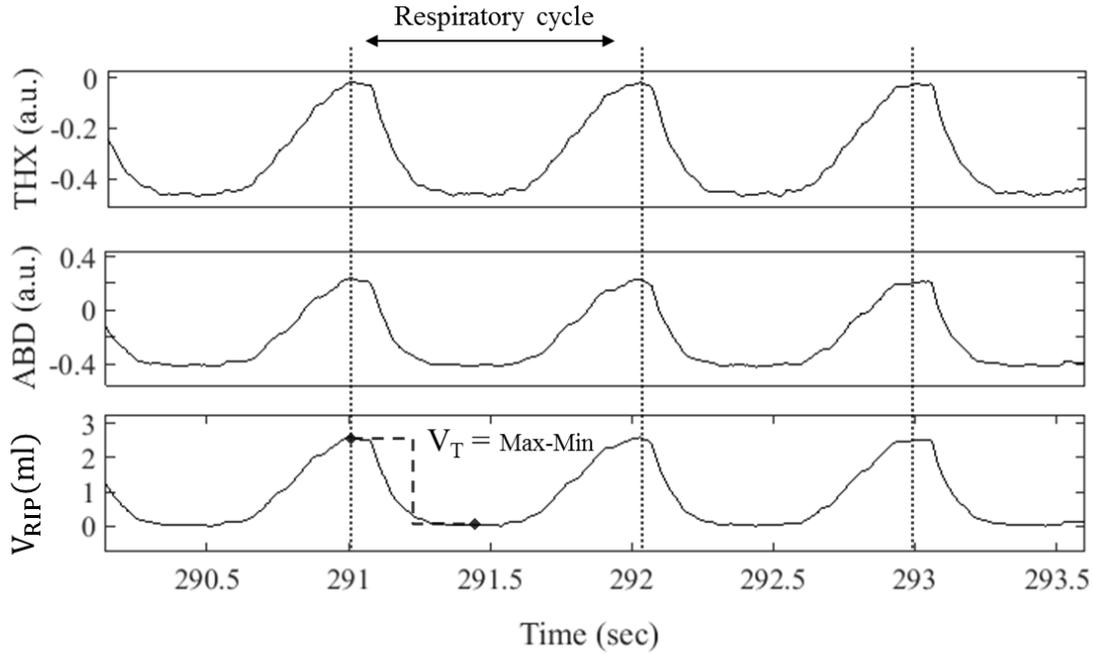
#### Statistical analysis

Results are presented as Mean $\pm$ SEM (standard error of the mean). Linearity of the measure sensitivity was quantified by applying linear regression between the two measurements. The agreement between the values given by the two sensors was finally assessed using the method described by Bland and Altman (Bland and Altman 1986).

## 4 Results

### 4.1 RIP signals

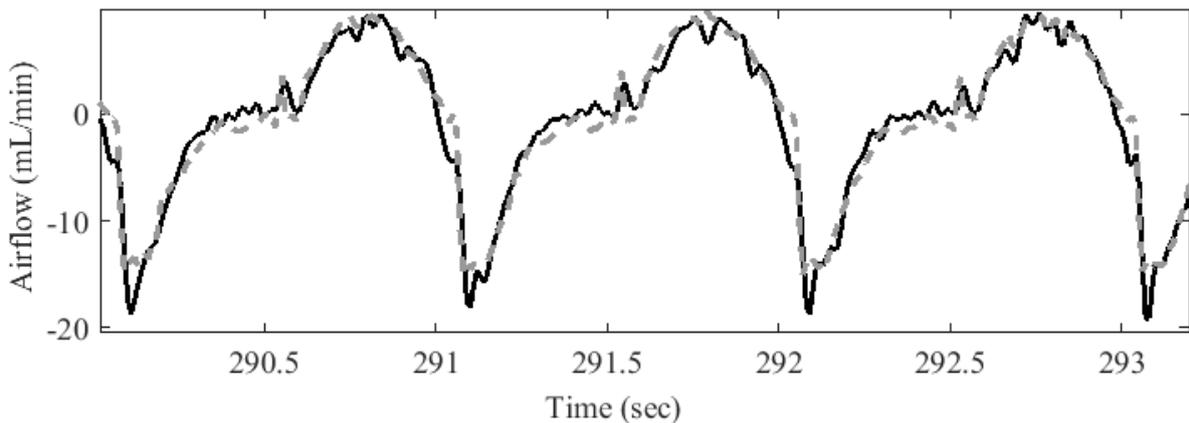
Typical respiratory signals obtained during stationary periods are shown in Fig.4. Respiratory cycles are delimited by the vertical dotted lines. An example of single tidal volume amplitude determination for a specific respiratory cycle is marked with dashed line in the lower panel of Fig.4.



**Figure 4** Instant respiratory volume signals of an artificially ventilated rat. *ABD* and *THX* are cross-sectional variations of abdomen and thorax.  $V_{RIP}$  represents calibrated respiratory trunk volume variations reconstructed from the plethysmographic signals *ABD* and *THX*. Dotted lines indicate delimitations of respiratory periods and dashed line illustrates one specific tidal volume  $V_{T,RIP}$ .

#### 4.2 Goodness of fit between PNT and RIP flows

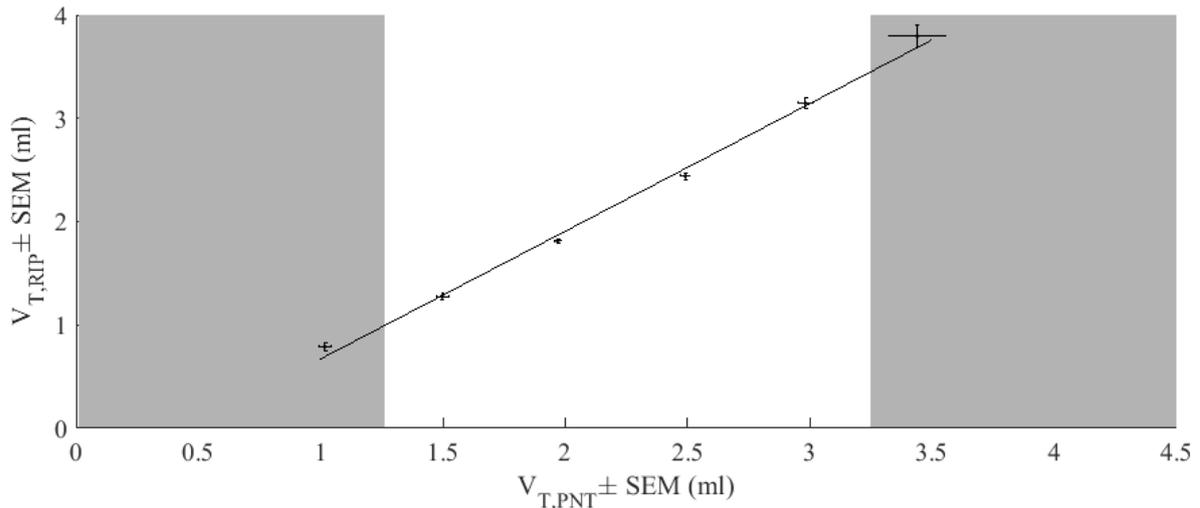
Fig.5 illustrates typical airflows obtained with RIP  $\dot{V}_{RIP}(t)$  and pneumotachograph  $\dot{V}_{PNT}(t)$ . The mean goodness of fit  $\rho$  calculated for the nine rats is  $93 \pm 0.5\%$ . Optimal  $\alpha$ ,  $\tau$  are all found with ratio equal to one and are thus set as equal. The average value of  $\alpha = \tau$  for the nine rats is  $1.8 \pm 0.1$ .



**Figure 5** Superposition of  $\dot{V}_{RIP}(t)$ , the reconstructed flow from respiratory inductive plethysmography (dashed light line), and  $\dot{V}_{PNT}(t)$ , the reference flow given by pneumotachograph (solid line) during the stabilization period.

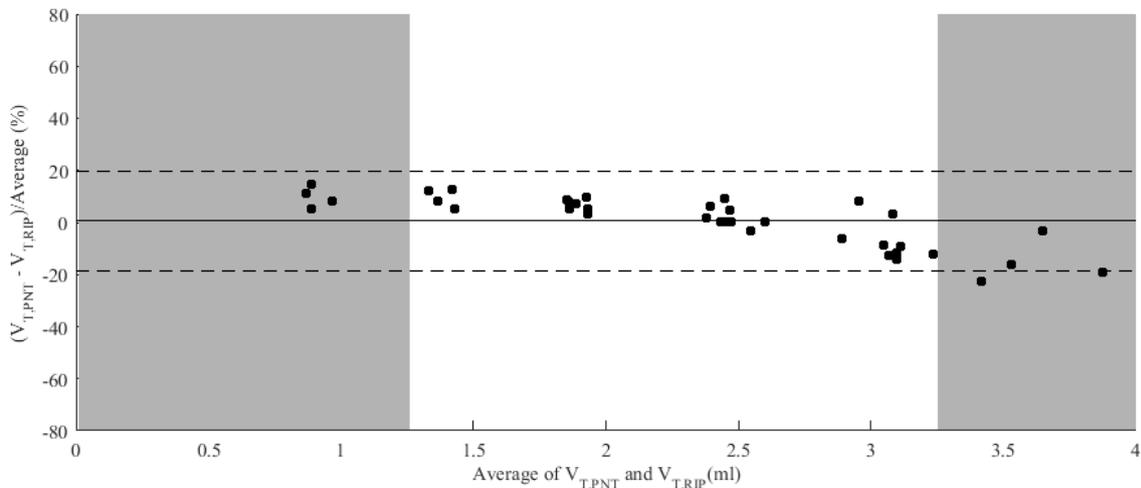
#### 4.3 Comparison of tidal volume measurements

Correlation between tidal volumes obtained from the RIP device and the reference PNT is plotted Fig.6.  $V_T$  measured with RIP appears to be well correlated to PNT volumes with a linear regression ( $p < 0.001$ ,  $R^2 = 0.975$ ,  $N=37$ ,  $V_{T,RIP} = a * V_{T,PNT} + b$ ,  $a = 1.24$ ,  $b = -0.40$ )



**Figure 6** Mean Tidal volumes (in ml) estimated from the DECRO RIP device ( $V_{T,RIP}$ ) versus reference measured with the PNT ( $V_{T,PNT}$ ). Horizontal and vertical error bars are SEM. Statistics and regression are computed on the whole dataset. Solid line represents linear least square regression. White part of the graph represents the range of physiological tidal volumes and gray areas indicate infra- and supra-physiological tidal volumes.  $N= 4-9$ .

Fig.7 shows Bland-Altman plot comparing the volumes obtained from RIP and the reference PNT. The solid line represents the mean difference between the two methods ( $12 \mu\text{L}$ ), and the two dashed lines 95% of the differences. The difference between  $V_{T,RIP}$  and  $V_{T,PNT}$  tidal volume measurements is lower than 19.6%.  $N$  is equal to 37 measurements on nine rats.



**Figure 7** Bland-Altman plot for Tidal volume ( $N=37$ ).  $V_{T,PNT}$  is the reference volume measurement acquired with a PNT in series with the airflow.  $V_{T,RIP}$  is the volume calculated from DECRO inductance plethysmography monitoring device. Dashed lines represent confidence limits ( $1.96\sigma$ ). White part of the graph represents the range of physiological tidal volumes and gray areas indicate infra- and supra-physiological tidal volumes.

## 5 Discussion

RIP is a well-known non-invasive respiratory monitoring technique that has been validated in both humans and medium-sized mammals. The present study aims at comparing a newly designed high resolution RIP apparatus with a reference measurement provided by a pneumotachograph. RIP reconstruction has been successfully performed in rats using the method based on optimization commonly used in humans. The quality of volume and airflow signals has been assessed by calculating the goodness of fit and the agreement between the two techniques over the physiological range of tidal volumes.

### 5.1 *RIP apparatus performances and comparison to PNT*

The oscillator and the digitizer robustly provided signals on all the animals. RIP airflow showed fit at 93% with PNT. Such performance level is similar to previous human RIP assessment carried out with comparable protocol on more substantial tidal volumes (Eberhard *et al* 2001). Local differences between RIP and PNT flows can be observed in Fig.5. In particular, lower volume peaks are smaller and smoother on the RIP flow. The tidal volume corresponds to the area under the flow of one respiratory cycle, hence the area which is not detected by the RIP is very small and the resulting underestimation of the parameter is negligible. *ABD* and *THX* cross-section variation signals exhibit sensitivity, signal-to-noise ratio and resolution that are adapted for respiratory cycle detection and amplitude measurement. Compared to the RIP systems usually used in humans, the current system has a high resolution since it allows the measurement of tidal volumes 200 times smaller. Moreover, this is confirmed by results obtained on a test bench which show that this resolution is 2 microliters (Flénet *et al* 2016). Despite the very low inductance of the coils involved, inductive plethysmography can be implemented in rats and enables proper reconstruction of instantaneous respiratory airflows.

RIP shows good agreement compared with PNT for tidal volume measurement as indicated by the correlation and the Bland-Altman analysis (Fig.7). Correlation is high with a linear model ( $R^2 = 0.975$ ), differences are higher for infra and supra physiological values. Similar behavior has already been observed in humans (Eberhard *et al* 2001) and is consistent with values expected to be slightly out of the normal physiological range where lung mechanics is subjected to changes. Although linearity is still good for extrema, these observations suggest that the linear model should not be extended out of the normal physiological range. The Bland-Altman shows that tidal volume differences are lower than 20% between RIP and PNT. Limiting the Bland-Altman analysis to the physiological range shows that infra and supra physiological levels (data at 1 ml and 3.5 ml) only introduce a 2% of additional difference. The 20% variability includes both the differences between the 2 technologies and the level of access to the tidal volume measurement by each of the methods. Indeed, the use of the PNT is invasive and allows direct measurement of the inspired volumes, whereas the RIP determines these volumes indirectly, in a non-invasive manner. In a matter of comparison, in humans such levels of agreement for RIP *versus* PNT have been considered as acceptable regarding potential clinical applications such as ambulatory monitoring of patients with pulmonary disease (Brüllmann *et al* 2009).

The linear regression (Fig.6) may be interpreted as the sensitivity of RIP tidal volume measurement with respect to PNT. This contributes to lowering the agreement between the methods (Fig.7). As it is repeatable from one animal to the other, this suggests that further studies will be required to characterize the RIP operating sensitivity that will be applicable to all physiological conditions.

### 5.2 *From $V_t$ to functional respiratory monitoring with RIP*

The respiratory parameters that may be accessible by this technique are not limited to tidal volume as studied in the present protocol. Indeed, this technique is potentially relevant for estimation of all flow- and volume-derived respiratory parameters. For instance, tidal midexpiratory flow ( $EF_{50}$ ) can be measured by taking the flow given by RIP at mid expiratory tidal volume. This parameter is particularly relevant in pharmacological studies since it is a common endpoint for assessment of airway obstruction (Hoymann 2012, Glaab *et al* 2001). Another asset of RIP is that it provides information about abdomen and thorax phasing during respiration. This has been observed both in humans (Sivan and Newth 1990, Ulm *et al* 2014) and primates (Hammer *et al* 1995). Phasing measurement has been identified as a potential method for airflow limitation detection in humans (Bloch *et al* 1997). Such phasing parameters are not accessible with methods like PNT or whole body plethysmography that measure the global respiratory flow.

## 6 Conclusion

Moving from man to small animals such as rats is a prominent instrumental challenge because it drastically hardens RIP sensor design. This study is the first to validate that it is technically possible to design a RIP device for rats.

This apparatus has been assessed with respect to reference measurement of respiratory function provided by a PNT. We demonstrated that RIP can provide calibrated respiratory volume and airflow signals equivalent to PNT. Moreover, comparison of tidal volume measurement from RIP and PNT reference indicates proper agreement between the two methods. Furthermore, the protocol carried out in this study emphasized advantages of the RIP approach. In particular, the device appeared to be fast and convenient to use. This system has a minimal impact on the experimental setup and the animal remains accessible to the experimenter.

This work unveils new perspectives about RIP applicability in rats. Considering the benefits inherent to non-invasive approaches, RIP is a promising alternative for respiratory monitoring. Further studies will now be focused on the implementation of the RIP system in extended physiological conditions without ventilatory support.

### Acknowledgments

This work was partially funded by LINKSIUM - GIFT GRENOBLE ALPES (PhD thesis of Timothé Flénet's funding) and CARNOT-LSI Institute (Charles Eynard's salary).

According to the IOP ethical policy for journals / Conflicts of interest / Authors, all the authors have no conflict of interest with and have received no financial support from DECRO.

**References**

- Bland J M and Altman D G 1986 Statistical methods for assessing agreement between two methods of clinical measurement. *Lancet* **1** 307–10
- Bloch K E, Jugoon S, de Socarraz H, Manning K and Sackner M A 1998 Thoracocardiography: Noninvasive monitoring of left ventricular stroke volume *J. Crit. Care* **13** 146–57
- Bloch K E, Li Y, Sackner M A and Russi E W 1997 Breathing pattern during sleep disruptive snoring *Eur. Respir. J.* **10** 576–86
- Brazelton III T B, Watson K F, Murphy M, Al-Khadra E, Thompson J E and Arnold J H 2001 Identification of optimal lung volume during high-frequency oscillatory ventilation using respiratory inductive plethysmography *Crit. Care Med.* **29** 2349–59
- Brüllmann G, Fritsch K, Thurnheer R and Bloch K E 2009 Respiratory monitoring by inductive plethysmography in unrestrained subjects using position sensor-adjusted calibration *Respiration* **79** 112–20
- Cleary D R, Phillips R S, Wallisch M and Heinricher M M 2012 A novel, non-invasive method of respiratory monitoring for use with stereotactic procedures *J. Neurosci. Methods* **209** 337–43
- Eberhard A, Calabrese P, Baconnier P and Benchetrit G 2001 Comparison between the respiratory inductance plethysmography signal derivative and the airflow signal *Frontiers in Modeling and Control of Breathing* (Springer) pp 489–94
- European commission 2013 *Seventh Report on the Statistics on the Number of Animals used for Experimental and other Scientific Purposes in the Member States of the European Union (Part 1-5)* (Bruxelles)
- Flénet T, Fontecave-Jallon J, Tanguy S, Boucher F, Baconnier P, Guméry P-Y 2016 Non-invasive cardiac output monitoring in pharmacology: a plethysmographic solution in rats *Conf Proc IEEE Eng Med Biol Soc* 4933-4936
- Glaab T, Daser A, Braun A, Neuhaus-Steinmetz U, Fabel H, Alarie Y and Renz H 2001 Tidal midexpiratory flow as a measure of airway hyperresponsiveness in allergic mice. *Am. J. Physiol. Lung Cell. Mol. Physiol.* **280** L565-73
- Hammer J, Newth C J and Deakers T W 1995 Validation of the phase angle technique as an objective measure of upper airway obstruction. *Pediatr. Pulmonol.* **19** 167–73
- Hepponstall J M, Tingay D G, Bhatia R, Loughnan P M and Copnell B 2012 Effect of closed endotracheal tube suction method, catheter size, and post-suction recruitment during high-frequency jet ventilation in an animal model *Pediatr. Pulmonol.* **47** 749–56
- Hoymann H G 2007 Invasive and noninvasive lung function measurements in rodents *J. Pharmacol. Toxicol. Methods* **55** 16–26
- Hoymann H G 2012 Lung Function Measurements in Rodents in Safety Pharmacology Studies *Front. Pharmacol.* **3** 1–11
- ICH Expert Working Group 2000 Safety Pharmacology Studies For Human Pharmaceuticals S7A I 1–13
- Konno K and Mead J 1967 Measurement of the separate volume changes of rib cage and abdomen during breathing *J. Appl. Physiol.* **22** 407–22
- Krinke G J, Bullock G R, Bunton T and The Laboratory Rat 2000 Selected lung function parameters of different rat strains *The Laboratory Rat Handbook of Experimental Animals* (Elsevier Science) p 326
- Leishman D J, Beck T W, Dybdal N, Gallacher D J, Guth B D, Holbrook M, Roche B and Wallis R M 2012 Best practice in the conduct of key nonclinical cardiovascular assessments in drug development: Current recommendations from the Safety Pharmacology Society *J. Pharmacol. Toxicol. Methods* **65** 93–101
- Loveridge B, West P, Anthonisen N R and Kryger M H 1983 Single-position calibration of the respiratory inductance plethysmograph *J. Appl. Physiol.* **55** 1031–4
- Maucotel J, Amrani-callens F El, Lorient S, Amrani A El, Forster R and Legrand J 2010 Evaluation of respiratory function in the conscious non-restrained cynomolgus monkey using respiratory inductive plethysmography *10th Annu. Focus. issue methods safety. Pharmacol.* **68** 27005
- Murphy D J, Renninger J P and Schramek D 2010 Respiratory inductive plethysmography as a method for measuring ventilatory parameters in conscious, non-restrained dogs *J. Pharmacol. Toxicol. Methods* **62** 47–53

High-resolution respiratory inductive plethysmography in rats: validation in anesthetized conditions.

- Sivan Y and Newth J L 1990 Thoracoabdominal Asynchrony in Acute Upper Airway Obstruction in Small Children1, 2 *Am Rev Respir Dis* **142** 540–4
- Tooley M H 2006 *Electronic Circuits: Fundamentals and Applications* (Elsevier)
- Ulm L N, Hamvas A, Ferkol T W, Rodriguez O M, Cleveland C M, Linneman L A, Hoffmann J A, Sicard-Su M J and Kemp J S 2014 Sources of methodological variability in phase angles from respiratory inductance plethysmography in preterm infants. *Ann. Am. Thorac. Soc.* **11** 753–60
- World Health Organization - WHO 2014 The top 10 causes of death *Fact sheet N°310* Online: <http://www.who.int/mediacentre/factsheets/fs310/en/>
- Zhang Z, Zheng J, Wu H, Wang W, Wang B and Liu H 2012 Development of a respiratory inductive plethysmography module supporting multiple sensors for wearable systems. *Sensors (Basel)*. **12** 13167–84