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A rebreathing method for measuring lung volume, diffusing capacity and cardiac output in conscious small animals

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Abstract

We developed a multiple gas rebreathing technique for measuring lung diffusing capacity (DL_{CO}), lung volume (V_L) and cardiac output simultaneously in conscious spontaneously breathing small animals. Lung volume was measured from the dilution of methane (CH_4) or sulfur hexafluoride (SF_6) and verified independently by a helium washout technique. Cardiac output and DL_{CO} were estimated from the uptake of acetylene and carbon monoxide, respectively. We tested guinea pigs at two levels of alveolar oxygen tension in order to estimate membrane diffusing capacity and pulmonary capillary blood volume by the Roughton–Forster technique. Results show that measured DL_{CO} are consistent with reported values in anesthetized guinea pigs as well as with allometric comparison across species. Lung volume estimated from SF_6 dilution agreed closely with that estimated independently from helium washout; however, lung volume estimated from CH_4 dilution was systematically lower due to the addition of endogenously produced CH_4 to the rebreathing system. We conclude that this technique can be used to measure resting lung function in conscious unsedated small animals.

Keywords: Guinea pig; Lung volume; Membrane diffusing capacity; Pulmonary capillary blood volume; Cardiac output; Allometry

1. Introduction

With the increasing use of small animal models for studying pulmonary physiology and pathophysiology, there is a growing need for new techniques to quantify lung function in these species. Physiologic lung diffusing capacity for carbon monoxide (DL_{CO}) has been measured in anesthetized tracheostomized small animals by single breath (Takezawa et al., 1980) and rebreathing (Johanson and Pierce, 1973) techniques. However, up till now it has not been possible to measure DL_{CO} in conscious spontaneously breathing small animals. In addition, DL_{CO} varies directly with lung volume and cardiac output, and the ability to simultaneously measure all of these parameters greatly enhances data interpretation. Our laboratory has previously reported the non-invasive measurement of DL_{CO} and its components, membrane diffusing capacity (Dm_{CO}) and pulmonary capillary blood volume (V_C), with

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simultaneous measurement of cardiac output and lung volume using a multiple gas rebreathing technique in human subjects (Hsia et al., 1995; Niranjan et al., 1997) and in dogs (Carlin et al., 1988; Hsia et al., 1994) at rest and during exercise. We have now adapted this technique to study lung function in conscious spontaneously breathing small animals, and report here the use of this rebreathing system in guinea pigs at two levels of alveolar oxygen tension at rest in order to estimate membrane diffusing capacity (Dm_{CO}) and pulmonary capillary blood volume (V_C) by the Roughton–Forster technique (Roughton and Forster, 1957). We verified the measurement of lung volume by this technique against that obtained independently by a helium washout technique. Combining our results with resting DL_{CO} measurements that have been reported in the literature, we also describe the allometric relationship between resting DL_{CO} and body weight across a wide range of species.

2. Methods

2.1. Animals

The Institutional Advisory Committee on Use of Animals at University of Texas Southwestern Medical Center approved the protocol. Studies were performed on seven male weanling Hartley guinea pigs (Harlan Industries, Indianapolis, IN) (average weight 1167 ± 103 (S.D.) g, range: 1040–1310 g, age 18 months).

2.2. Apparatus

The apparatus is shown schematically in Fig. 1. The animal was placed inside a cylindrical chamber with a pneumatic sealing cuff around the neck (Respiromax^{TM}, Columbus Instruments, Columbus, OH). The head was snugly enclosed in a customized cone-shaped plexiglass mask attached to a one-way breathing valve (Model 2384A, Hans Rudolph, Kansas City, MO), a stopcock and a small latex balloon (∼10 ml). The inspiratory port connected to a reservoir bag. The expiratory port connected via a metallic cooling tube to a pneumotachometer (Hans Rudolph, Series 8311) and an anesthetic bag for timed collections of expired gas. The pneumotachometer signal was amplified (Hans Rudolph Amplifier 1, Series 1100), and acquired by a data acquisition card (PCM-DAS08, Computer Boards, Middleboro, MA) on a PC laptop (Pentium III, Dell Inspiron 8000) running Labview 5.0 (National Instruments, Austin, TX) and Universal Library (ComputerBoards) acquisition software for real time mea-

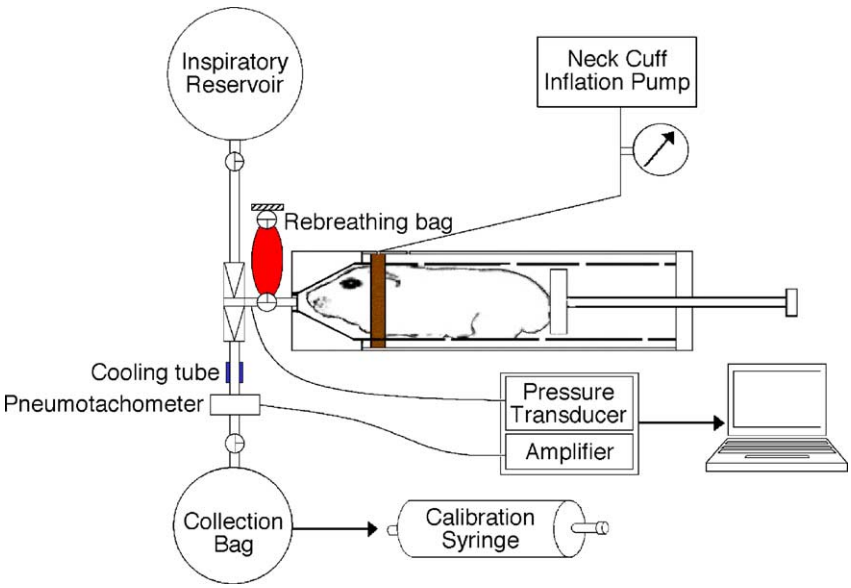

Fig. 1. Schematic diagram of the rebreathing apparatus.