AIRWAY MECHANICS AND LUNG TISSUE VISCOELASTICITY: EFFECTS OF ALTERED BLOOD HEMATOCRIT IN THE PULMONARY CIRCULATION

Ferenc Peták[1], Gergely H. Fodor[1], Barna Babik[2] Walid Habre[3, 4]

[1] Department of Medical Physics and Informatics, University of Szeged, Szeged, Hungary

[2] Department of Anesthesiology and Intensive Therapy, University of Szeged, Szeged, Hungary

[3] Pathophysiological Experimental Platform, Department of Anesthesiology, Pharmacology and Intensive Care, University of Geneva, Geneva, Switzerland

[4] Pediatric Anesthesia Unit, Geneva Children's Hospital, Geneva, Switzerland

Running title: Hematocrit and lung tissue viscoelasticity

Address for correspondence:

Ferenc Peták

Department of Medical Physics and Informatics

University of Szeged

9, Korányi fasor H-6720, Szeged, Hungary

Email: petak.ferenc@med.u-szeged.hu

Phone/Fax: +36 62 545077

ABSTRACT

The contribution of the hematocrit (Hct) of the blood in the pulmonary vasculature to the overall lung mechanics has not been characterized. We therefore set out to establish how changes of the Hct level in the pulmonary circulation affect the airway and lung tissue viscoelastic properties. The Hct level of the blood in an isolated perfused rat lung model was randomly altered. Intermediate (26.5%), followed by low (6.6%) or normal Hct (43.7%) were set in 2 consecutive sequences. The pulmonary capillary pressure was maintained constant throughout the experiment and the pulmonary hemodynamic parameters were monitored continuously. The airway resistance (Raw), the viscous (G) and elastic (H) parameters and the hysteresivity (η=G/H) of the lung tissues were obtained from measurements of forced oscillatory input impedance data. Raw was not affected by the alterations of the Hct levels. As concerns the lung tissues, the decrease of Hct to intermediate or low levels resulted in close to proportional decreases in the viscoelastic parameters G (16.5±7.7%, 12.1±9.5%, $p<0.005$) and H (13.2±8.6%, 10.8±4.7%, $p<0.001$). No significant changes in η were detected in a wide range of Hct, which indicates that coupled processes cause alterations in the resistive and elastic properties of the lungs following Hct changes in the pulmonary circulation. The diminishment of the viscous and elastic parameters of the pulmonary parenchyma following a reduction of blood Hct demonstrate the significant contribution of the red blood cells to the overall lung viscoelasticity.

Key words: lung mechanics, pulmonary vasculature, cardiopulmonary interactions, lung tissue resistance, lung tissue elastance

New & Noteworthy

Additional insight into the participation of the pulmonary vascular capillary network in the elastic and dissipative properties of the lung tissue was revealed in the present study performed in isolated perfused rat lungs. Decreasing hematocrit in the pulmonary circulation below physiological diminished the viscous and elastic parameters of the pulmonary tissues, therefore demonstrating that the energy dissipation and storage displayed by the red blood cells are significant contributing factors in the total lung viscoelasticity.

INTRODUCTION

The mechanical properties of the pulmonary system are determined by the individual contributions of and the complex interactions between its main constituents, the tracheobronchial tree, the lung parenchymal matrix, and the pulmonary vasculature. While the airway properties can be well described by resistive and inertive properties (13), the involvement of the lung tissues in the overall lung mechanics is more complex: the importance of the complex interactions between the extravascular proteins, fibers, cells, surface film layer and interstitial fluids has been well documented (3, 7, 23).

There is increasing evidence of the key role of cardiopulmonary interactions in a wide variety of lung disorders, such as congenital malfunctions (2, 12), pulmonary hypertension (1, 15) and airway hyperresponsiveness (19, 24). The participation of the pulmonary vascular capillary network in the gross elastic and dissipative properties of the lung, however, is less well understood. There is a particular lack of knowledge as to how the rheological properties of the blood affect the lung tissue viscoelasticity, despite the clearly-established fact that the blood composition determines the mechanical properties of the pulmonary vasculature (13, 14, 21). Higher levels of hematocrit (Hct) at a given shear rate result in an elevated viscosity of whole blood (26), thereby raising the vascular resistance in an exponential fashion, and altering the shape of the pulmonary vascular resistance-blood flow curve (13, 14, 20). Furthermore, the viscoelastic properties of the blood depend greatly on the level of Hct (22). Indeed, our earlier data suggest that hemodilution and/or hemoconcentration may alter the bronchial tone and lung tissue viscoelasticity (8). However, no data are available that characterize the link between the physical properties of the blood at different Hct levels and the mechanical properties of the lungs. A clearer understanding of such cardiopulmonary interactions would facilitate a more complete description of the basic contributors to the lung tissue viscoelasticity. Further, a more complete picture would be of major importance in

clarifying the lung mechanical consequences of clinical situations involving hemodilution (i.e. fluid replacement therapy), hemoconcentration (i.e. polycythemia, dehydration and chronic hypoxia) or the weaning of the patient from the ventilator.

The aim of the present experiments was therefore to characterize how alterations in the blood hematocrit in the pulmonary circulation affect the airway mechanics and the lung tissue viscoelasticity. We hypothesized that the airway parameters would not be affected by alterations of Hct, however Hct would affect the lung tissue viscoelasticity via modifying parameters related to both the energy loss (damping) and storage (elastance).

METHODS

Animal preparations

The experimental protocol was approved by the Experimental Ethics Committee of the University of Geneva and the Animal Welfare Committee of the Canton of Geneva, Switzerland (No. GE/171/14).

Adult male Sprague-Dawley rats (n=18, weighing 424-514 g) were anesthetized with isoflurane (3-4% induction dose). Tracheotomy was performed with a 14-gauge polyethylene cannula (B|Braun, Melsungen, Germany), and volume-controlled positive pressure mechanical ventilation was applied with a tidal volume of 7 ml/kg, a frequency of 70-80/min and a positive end-expiratory pressure of 2.5 cmH_2O (model 683, Harvard Apparatus Co. Inc., South Natick, MA) while the inspired fraction of oxygen was maintained at 0.5. During the surgical preparation, anesthesia was maintained with 1.4% isoflurane. A femoral artery and vein were catheterized for blood sampling and continuous arterial blood pressure monitoring. Blood gases from arterial blood samples were analyzed regularly (Ultima^{TM}, Datex/Instrumentarium, Helsinki, Finland). Airway pressure was measured (Validyne DP 45, Northridge, CA) and monitored continuously (Biopac, Santa Barbara, CA). The rats were

fully anticoagulated (heparin, 1.5 IU/g iv) and 20-25 ml blood was then gently withdrawn via the arterial cannula, the collected blood being continuously replaced by the iv administration of colloid solution (6% hydroxyethyl-starch). This procedure resulted in the collection of blood with a Hct of around 25-30% from each animal.

Preparation of isolated lungs

Harvesting and perfusion of the isolated rat lungs were carried out in 9 rats as detailed previously (17), while the other 9 rats served as blood donors to allow Hct level elevation with a sufficient volume priming the perfusion circuit. The blood collected from all rats was centrifuged (3000 g for 10 min). The supernatant plasma was extracted from the centrifuged blood of the donor rats to obtain concentrated blood with a Hct of ~50%, thereby allowing perfusions with higher Hct levels. This supernatant plasma was used to dilute the perfusate to achieve low Hct levels of ~5-10%. In the rats involved in the main study group, the centrifuged blood was re-concentrated to attain Hct of ~30%; this was used to perfuse the lungs initially. A median sternotomy was performed in these animals and the main pulmonary artery was cannulated via the right ventricular outflow track (14-gauge, B|Braun, Melsungen, Germany). Another catheter was placed in the left ventricle through the left ventriculotomy, in which a Combifix®-Adapter (B|Braun, Melsungen, Germany) was tightly fixed and connected to medical-grade silicone tubing. A third catheter was placed directly in the left atrium (polyethylene tubing, ID 0.88 mm, Portex, Hythe, GB). The lungs and the heart were excised and extracted in a single block, dissected free of adjacent tissue.

Lung perfusion

The setup used for the lung perfusion has been detailed previously (17). The heart-lung block was placed in a humidified box (590 ml) and volume-controlled positive pressure mechanical

ventilation was applied in the same manner as in the intact animal, except that 5% CO_2 was added to the inspired air to avoid hypocapnia. In the absence systemic oxygen consumption, this ventilation provided adequate oxygenation of the lung tissue with maintaining normocapnia. After the initiation of mechanical ventilation, a series of two hyperinflations (peak pressure 25-30 cmH_2O) was applied to eliminate the atelectatic areas. Lung perfusion was then started with the blood with an intermediate Hct level (~30%). The initial perfusion parameters were set to achieve a pulmonary blood flow (Qp) of 7-15 ml/min, with approach to a pulmonary arterial pressure (Ppa) of 25 mmHg and a left atrial pressure (Pla) of 10 mmHg. These target values were attained by placing the container supporting the pulmonary artery and the distal extremity of the left ventricular outflow cannula at appropriate heights. Both Pla and Ppa were greater than the mean airway pressure (Paw), and the lungs were therefore maintained under West's zone 3 conditions (Ppa > Pla > mean (Paw)). The blood dripping from the left ventricular outflow cannula was collected in a cylinder, and aspirated from this reservoir with polyethylene tubing passing through a roller pump (Ismatec Pump, Glattburg, Zurich, Switzerland). The Ppa and Pla values were recorded (Honeywell, model 156-PC 06-GW2), and Qp was measured continuously with a transit-time flowmeter (T-201 CDS, Transonic Systems Inc., Ithaca, NY) situated between the perfusion reservoir and the catheter cannulating the main pulmonary artery. The pulmonary vascular resistance (Rv) was calculated by dividing the pressure difference between Ppa and Pla by Qp. Pulmonary capillary pressure (Pc) was estimated by applying the Gaar equation (Pc = Pla + 0.44 x [Ppa - Pla]) (11). Airway pressure, Ppa, Pla, Pc, Qp, Rv and the lung weight obtained by using an isometric force displacement transducer (Grass FT03, Quincy, MA, USA) were displayed continuously and stored on a microcomputer at a sampling rate of 100 Hz via an analog/digital interface converter (Biopac, Santa Barbara, CA). The weight gain (WG) as an on-line edema index was calculated as the relative change in the weight of the lung-heart

block per unit time.

Lung mechanical measurements

The mechanical parameters were estimated from the input impedance (ZL) measured by forced oscillation with the wave tube technique, as described previously (18). Briefly, at end-expiration, the mechanical ventilation was paused; during these apneic periods, a loudspeaker and the tracheal cannula were connected through a polyethylene wave tube (length=102 cm, ID=0.2 cm), and a small-amplitude (1 cmH_2O peak to peak) forcing signal was delivered into the trachea. The loudspeaker was driven by a computer-generated pseudorandom signal containing 23 components ranging from 0.5 to 21 Hz, which is a suitable frequency range to obtain both tissue and airway parameters. Lateral pressures were measured at both ends of the wave tube with miniature side-arm transducers (ICS 33NA00D). These pressure signals were low-pass filtered (< 25Hz) and digitized at a sampling frequency of 128 Hz. The pressure transfer function was created by fast Fourier transformation from the 8-s recording. The ZL was computed from the pressure transfer function, as the load impedance of the wave tube, by using the transmission line theory (9).

A model containing an airway and a constant-phase tissue compartment was fitted to the averaged ZL data in each condition by minimizing the squared sum of weighted differences between the measured and the modeled ZL data. The airways were characterized by a frequency-independent airway resistance (Raw) and inertance (Iaw), while the tissue compartment was described by the parenchymal damping (G) and elastance (H) (13). The lung tissue hysteresivity (η) was calculated as G/H (10). The airway parameters were corrected for the resistance and inertance of the ET tube.

Study protocol

Figure 1 outlines the scheme of the experimental protocol. After the establishment of steady-state pulmonary hemodynamic conditions while the lungs were perfused with blood with intermediate Hct, a recruitment maneuver was performed by inflating the lungs 3 times to a transpulmonary pressure (Ptp) of 30 cmH_2O. A 15-min-long stabilization interval was then allowed, and a hyperinflation maneuver was made to a Ptp of 25 cmH_2O to standardize the volume history. This was achieved by occluding the expiratory port of the ventilator for one respiratory cycle 1 min prior to the impedance measurements. A set of ZL data was next recorded including 4-6 data epochs in 1-min intervals. The blood in the perfusion circuit was then changed to a perfusate with either normal or low Hct level in random sequence. A 3-min equilibration period with the new perfusate was subsequently allowed, during which Ppa and Pla were adjusted to maintain the same Pc, if necessary. A hyperinflation maneuver was made and another set of ZL data was collected. The blood was then changed to the opposite Hct level, and the same experimental procedure was repeated. The recruitment maneuver was next performed as detailed above, and ZL was measured again during lung perfusion with blood with an intermediate Hct level after a 15-min equilibration period and a recruitment maneuver. If this second baseline measurement proved to be identical to that obtained initially, another sequence was applied with the intermediate first, and a low or normal Hct level in a random order. Since exact reproduction of the Hct levels between the two parts of the experiment within an individual lung was not possible, the data triplets obtained in each phase were analyzed separately. The whole protocol including 6 Hct settings was completed in 4 isolated lungs; technical problems with the perfusion precluded reliable data collection in the last (6^{th}) stage of the experiment in the other 5 lungs. In 2 out of these 5 lungs, the hemodynamical monitoring was not complete due to technical problems with the blood flow sensor.

Statistical analyses

Scatters in the parameters were expressed in SD values. The Kolmogorov-Smirnov test was used to test data for normality. One-way repeated measures analysis of variances (ANOVA) was applied to compare the mechanical parameters obtained during perfusions with different Hct levels. Pairwise comparisons were performed by using the Dunnett method. The strength of correlations between variables was assessed by using Pearson statistical analyses. The statistical tests were performed with a significance level of $p<0.05$.

RESULTS

The lung perfusion parameters obtained at the different Hct levels are demonstrated in Fig. 2. Increases of Hct decreased Qp markedly ($p<0.0001$), while Pc remained constant throughout the study protocol ($p=0.34$). These changes resulted in obvious elevations in Rv ($p<0.0001$). There were no statistically detectable changes in WG ($p=0.37$), indicating the lack of edema formation during the experiments.

The effects of altered Hct on the airway mechanics are demonstrated in Fig. 3. Changes in Hct did not cause statistically significant alterations in Raw ($p=0.25$).

Figure 4 illustrates the influence of the altered Hct on the viscoelastic parameters of the lung tissue. Lowering Hct to intermediate and low levels led to statistically significant decreases in both the G and the H parameters of the pulmonary parenchyma ($p<0.001$ for all) with no significant differences between the lower levels. These reasonably proportional decreases in the viscoelastic parameters did not result in detectable alterations in η ($p=0.61$).

The relationships between the resistive properties of the pulmonary vasculature (Rv) and the viscoelastic parameters of the lung parenchyma (G and H) are demonstrated in Fig. 5. Rv exhibited a significant correlation with G ($r=0.52$, $p<0.0005$), and a somewhat lower, but still statistically highly significant associations with H ($r=0.48$, $p<0.002$).

DISCUSSION

In the present study, the airway mechanics and the changes in the viscoelastic mechanical parameters of the lung parenchyma were characterized following alterations in the rheological properties of the blood consequent to changes of the Hct level in isolated perfused rat lungs. While the airway properties were not affected by a wide range (6-fold) of alterations of Hct, the viscoelastic parameters of the lung parenchyma exhibited differential responses. At lower than physiological Hct values, alterations in the viscous and elastic properties of the lung tissue were observed. Moreover, the changes in the resistive properties of the pulmonary vasculature following the alterations of Hct correlated with lung mechanical indices reflecting the energy loss (damping) and storage (elastance) of the pulmonary parenchyma.

Methodological considerations

The present study involved the use of a well-validated model to investigate the effects of pulmonary vasculature alterations on the lung mechanics without the interference of neuro-humoral control (17, 19). This model was earlier shown to offer stable conditions over time and hence to allow repeated lung impedance measurements without the confounding influence of potential lung congestion (17, 19). In agreement with these earlier studies, the preparation was stable in the vast majority of the predefined measurement conditions (49/54 cases). The failure of data collection in the remaining 5 cases was due to irreversible increases in Rv. Low intraindividual variability was noted in the present study, with no WG (Fig. 2), suggesting a

stable lung function and structure without biasing factors such as edema development or pulmonary hemodynamic instability. The present experiments revealed that the perfusion parameters reflecting the pulmonary vascular pressures and blood flow were in accord with previously established values in isolated perfused rat lungs (17, 19). Furthermore, the airway and lung tissue mechanical parameters obtained in the present study proved to be in excellent agreement with those reported previously in similar experimental settings (17, 19).

An important feature of the current experimental model is the use of positive airway pressure to keep the lungs inflated rather than the physiological negative pressure around the pleural space. While lung inflation performed with negative pleural or positive airway pressure affected markedly the pulmonary vascular changes, earlier results revealed that the mode of inflation does not influence the pressure-dependent changes in the airway and parenchymal mechanics (16). Therefore, it does not seem likely that our results are significantly biased by this factor, and the findings are most probably valid for intact chest conditions.

Three levels of Hct were set in the present study to characterize how the pulmonary blood rheological characteristics affect the lung mechanics. The intermediate level of Hct (26.5% as group average) was the perfusate obtained from the animals with an attempt to establish pulmonary hemodynamic conditions encountered *in vivo* (17, 19). The lower level of Hct (6.6%) was selected so as to achieve conditions where the influence of the erythrocytes is minimal, while the oncotic and osmotic pressures in the pulmonary capillaries are maintained. The rationale for the highest Hct as normal (43.7%) was to establish near-physiological pulmonary capillary pressure without resulting in extreme increases in Rv.

The present study clearly confirmed that the blood viscoelasticity changes resulting from the

adjustment of Hct in the pulmonary vessels ultimately modifies the pulmonary hemodynamics (Fig. 2). The acute alterations in the pulmonary hemodynamics subsequently influence the mechanical properties of the lungs (17). These changes, however, are governed primarily by the level of Pc, with minor effects of Qp in the range encountered in our experiments (17). The level of Pc in the present study was therefore adjusted to avoid the potential confounding biasing effects of an altered Pc, while the inevitable alterations in Qp *per se* were not likely to bias the findings.

Effects of the altered Hct on the airway mechanics

The main lung mechanical parameter reflecting the flow resistance of the conducting airways was not affected by the wide range of alterations of the Hct (Fig. 3). This finding indicates the lack of influence of blood Hct on the airway properties. However, it should be born in mind that the bronchial circulation was not perfused in the present experimental model. While our primary aim was to characterize the impact of the pulmonary circulation on the lung mechanics, the potential airway effects of an altered blood viscoelasticity in the bronchial circulation cannot be excluded.

Effects of the altered Hct on the lung tissue viscoelasticity

The main finding of the present study was the observation of consistent changes in the viscous and elastic parameters of the lung tissue when Hct was lowered below the physiological level (Fig. 4). The changes in these mechanical parameters imply that alterations in Hct affect commonly observed variables reflecting lung mechanics in clinical practice, such as total pulmonary resistance (via its component, G) or dynamic lung compliance (related to the reciprocal of H). Furthermore, the work of breathing required to overcome the frictional dissipative resistance (reflected by G) and against the elastic recoil (represented by H) are also affected by the level of Hct in the blood. This contribution of the blood to the overall lung

tissue viscoelasticity can be attributed to various factors. Firstly, since the erythrocytes themselves exert viscoelastic properties (6, 22), their lower amount in the fluid in the pulmonary capillaries following an Hct decrease is expected to diminish the dissipative and elastic properties of the lung tissue embedding these structures. Secondly, the red blood cells are less likely to aggregate under the low Hct conditions, thereby limiting cell-to-cell interactions (4, 5, 25). The resulting elevated Qp, as observed in the present study, may further lessen red cell deformation and adhesion, and in this way restraining the viscous and elastic properties of the lung parenchyma. Thirdly, a potential contributory factor may be related to mechanical interdependence of the lung parenchyma and the embedded pulmonary vasculature. This participation of the pulmonary vasculature in the overall lung parenchymal viscoelasticity can be anticipated from the correlations between Rv and the lung tissue damping and elastance (Fig. 5). Regardless of the potential mechanisms, it is noteworthy that η proved to be invariable to a wide range of changes of Hct (Fig. 4). This suggests a coupling between the processes causing alterations in the resistive and elastic properties in the blood in the pulmonary circulation following the changes of Hct.

Summary and conclusions

The results of the present study have provided additional insight into the participation of the pulmonary vascular capillary network in the elastic and dissipative properties of the lung tissue. The decreases in the mechanical parameters reflecting the viscous and elastic properties of the pulmonary tissues in response to a lowered Hct in the lung vessels demonstrate that the energy dissipation and storage displayed by the red cells are significant contributing factors in the total lung viscoelasticity. This implies that the lung mechanics may be affected by changes in the rheological properties of the intrapulmonary blood following treatments involving hemodilution when Hct is lowered from the normal range (e.g. fluid

resuscitation). These findings should be taken into consideration in the treatment strategies to optimize the lung function in the presence of disorders involving altered Hct, such as hypervolemia or polycythemia.

ACKNOWLEDGMENTS

The authors thank Xavier Belin and Jean-Pierre Giliberto for their excellent technical assistance.

This research was supported by the Swiss National Science Foundation Grant (32003B-143331), a Hungarian Basic Research Council Grant (OTKA K115253) and a Research Grant obtained from the Faculty of Medicine, University of Szeged, Hungary. Gergely H Fodor was supported by an Endre Szirmai research fellowship grant.

REFERENCES

1. **Albu G, Petak F, Fontao F, Biton C, Pache JC, and Habre W**. Mechanisms of airway hyper-responsiveness after coronary ischemia. *Respir Physiol Neurobiol* 162: 176-183, 2008.
2. **Bancalari E, Jesse MJ, Gelband H, and Garcia O**. Lung mechanics in congenital heart disease with increased and decreased pulmonary blood flow. *J Pediatr* 90: 192-195, 1977.
3. **Bates JH, Davis GS, Majumdar A, Butnor KJ, and Suki B**. Linking parenchymal disease progression to changes in lung mechanical function by percolation. *Am J Respir Crit Care Med* 176: 617-623, 2007.
4. **Brooks DE, Goodwin JW, and Seaman GV**. Interactions among erythrocytes under shear. *J Appl Physiol* 28: 172-177, 1970.
5. **Drasler WJ, Smith CM, 2nd, and Keller KH**. Viscoelasticity of packed erythrocyte suspensions subjected to low amplitude oscillatory deformation. *Biophys J* 52: 357-365, 1987.
6. **Evans EA**. Structure and deformation properties of red blood cells: concepts and quantitative methods. *Methods Enzymol* 173: 3-35, 1989.
7. **Faffe DS, and Zin WA**. Lung parenchymal mechanics in health and disease. *Physiol Rev* 89: 759-775, 2009.
8. **Fodor GH, Babik B, Czovek D, Doras C, Balogh AL, Bayat S, Habre W, and Petak F**. Fluid replacement and respiratory function: comparison of whole blood with colloid and crystalloid: A randomised animal study. *Eur J Anaesthesiol* 33: 34-41, 2016.
9. **Franken H, Clement J, Cauberghs M, and Van de Woestijne KP**. Oscillating flow of a viscous compressible fluid through a rigid tube: a theoretical model. *IEEE Trans Biomed Eng* 28: 416-420, 1981.
10. **Fredberg JJ, and Stamenovic D**. On the imperfect elasticity of lung tissue. *J Appl Physiol (1985)* 67: 2408-2419, 1989.
11. **Gaar KA, Jr., Taylor AE, Owens LJ, and Guyton AC**. Pulmonary capillary pressure and filtration coefficient in the isolated perfused lung. *Am J Physiol* 213: 910-914, 1967.
12. **Habre W, Schutz N, Pellegrini M, Beghetti M, Sly PD, Hantos Z, and Petak F**. Preoperative pulmonary hemodynamics determines changes in airway and tissue mechanics following surgical repair of congenital heart diseases. *Pediatr Pulmonol* 38: 470-476, 2004.

13. **Hantos Z, Daroczy B, Suki B, Nagy S, and Fredberg JJ**. Input impedance and peripheral inhomogeneity of dog lungs. *J Appl Physiol (1985)* 72: 168-178, 1992.
14. **Hoffman JI**. Pulmonary vascular resistance and viscosity: the forgotten factor. *Pediatr Cardiol* 32: 557-561, 2011.
15. **Low AT, Medford AR, Millar AB, and Tulloh RM**. Lung function in pulmonary hypertension. *Respir Med* 2015.
16. **Petak F, Albu G, Lele E, Hantos Z, Morel DR, Fontao F, and Habre W**. Lung mechanical and vascular changes during positive- and negative-pressure lung inflations: importance of reference pressures in the pulmonary vasculature. *J Appl Physiol (1985)* 106: 935-942, 2009.
17. **Petak F, Habre W, Hantos Z, Sly PD, and Morel DR**. Effects of pulmonary vascular pressures and flow on airway and parenchymal mechanics in isolated rat lungs. *J Appl Physiol (1985)* 92: 169-178, 2002.
18. **Petak F, Hantos Z, Adamicza A, Asztalos T, and Sly PD**. Methacholine-induced bronchoconstriction in rats: effects of intravenous vs. aerosol delivery. *J Appl Physiol* 82: 1479-1487, 1997.
19. **Petak F, Janosi TZ, Myers C, Fontao F, and Habre W**. Impact of elevated pulmonary blood flow and capillary pressure on lung responsiveness. *J Appl Physiol (1985)* 107: 780-786, 2009.
20. **Raj JU, and Anderson J**. Comparison of dextran and hematocrit effects in the pulmonary microcirculation. *Circ Res* 68: 1108-1116, 1991.
21. **Raj JU, Ramanathan R, Chen P, and Anderson J**. Effect of hematocrit on microvascular pressures in 3- to 5-wk-old rabbit lungs. *Am J Physiol* 256: H766-771, 1989.
22. **Skalak R, and Chien S**. Rheology of blood cells as soft tissues. *Biorheology* 19: 453-461, 1982.
23. **Suki B, and Bates JH**. Lung tissue mechanics as an emergent phenomenon. *J Appl Physiol (1985)* 110: 1111-1118, 2011.
24. **Tanabe T, Rozycki HJ, Kanoh S, and Rubin BK**. Cardiac asthma: new insights into an old disease. *Expert Rev Respir Med* 6: 705-714, 2012.
25. **Thurston GB, and Henderson NM**. Effects of flow geometry on blood viscoelasticity. *Biorheology* 43: 729-746, 2006.
26. **Wells RE, Jr., and Merrill EW**. Influence of flow properties of blood upon viscosity-hematocrit relationships. *J Clin Invest* 41: 1591-1598, 1962.

FIGURE LEGENDS

Figure 1. Experimental design. RM: recruitment maneuver including 3 consecutive hyperinflations to a transpulmonary pressure of 30 cmH_2O. Sigh: single hyperinflation maneuver to a transpulmonary pressure of 25 cmH_2O. ZL: collection of a set of pulmonary input impedance data including 4 to 6 data epochs. Hct: hematocrit.

Figure 2. Pulmonary perfusion parameters obtained during lung perfusions with low (n=14), intermediate (n=17) or normal (n=15) blood hematocrit levels. Open symbols with dashed lines denote data points obtained in individual rat lungs. Closed symbols with error bars connected with solid lines represent group means and SD. Qp: pulmonary blood flow, Pc: pulmonary capillary pressure, Rv: pulmonary vascular resistance, WG: weight gain related to the respective initial weight. *: $p<0.05$.

Figure 3. Absolute value of airway resistance (left) and its changes relative to the normal hematocrit level (right) during lung perfusions with low (n=15), intermediate (n=18) or normal (n=16) levels of blood hematocrit. Open symbols with dashed lines denote data points obtained in individual rat lungs. Closed symbols with error bars connected with solid lines represent group means and SD. Sample numbers are reduced by 2 on the right panel due to lack of data at normal Hct.

Figure 4. Absolute values of viscous (G), elastic (H) and hysteresivity (η) parameters of the lung tissues (left) and their changes relative to the normal hematocrit level (right) during lung perfusions with low (n=15), intermediate (n=18) or normal (n=16) levels of blood hematocrit. Open symbols with dashed lines denote data points obtained in individual rat lungs. Closed symbols with error bars connected with solid lines represent group means and SD. *: $p<0.05$. Sample numbers are reduced by 2 on the right panels due to lack of data at normal Hct.

Figure 5. Correlations between pulmonary vascular resistance (Rv) and viscous (G) and elastic (H) parameters of the lung parenchyma. Open symbols: data obtained at a low blood hematocrit level; gray symbols: data obtained at an intermediate blood hematocrit level; filled symbols: data obtained at normal blood hematocrit level. Solid lines: linear regressions.

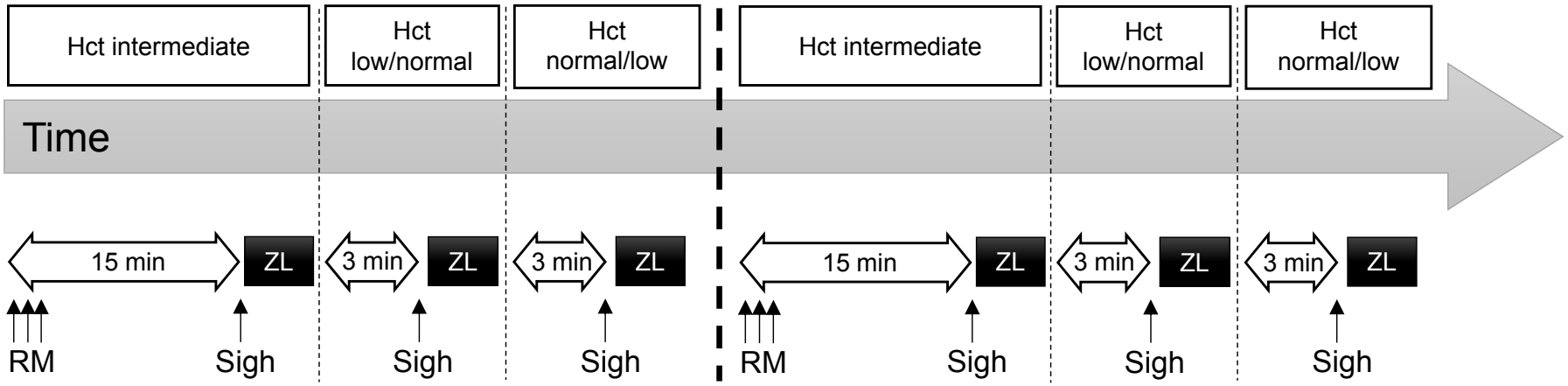
Hct intermediate
Hct low/normal
Hct normal/low
Hct intermediate
Hct low/normal
Hct normal/low
Time
15 min
ZL
3 min
ZL
3 min
ZL
15 min
ZL
3 min
ZL
3 min
ZL
RM
Sigh
Sigh
Sigh
RM
Sigh
Sigh
Sigh

Figure 1 – R1

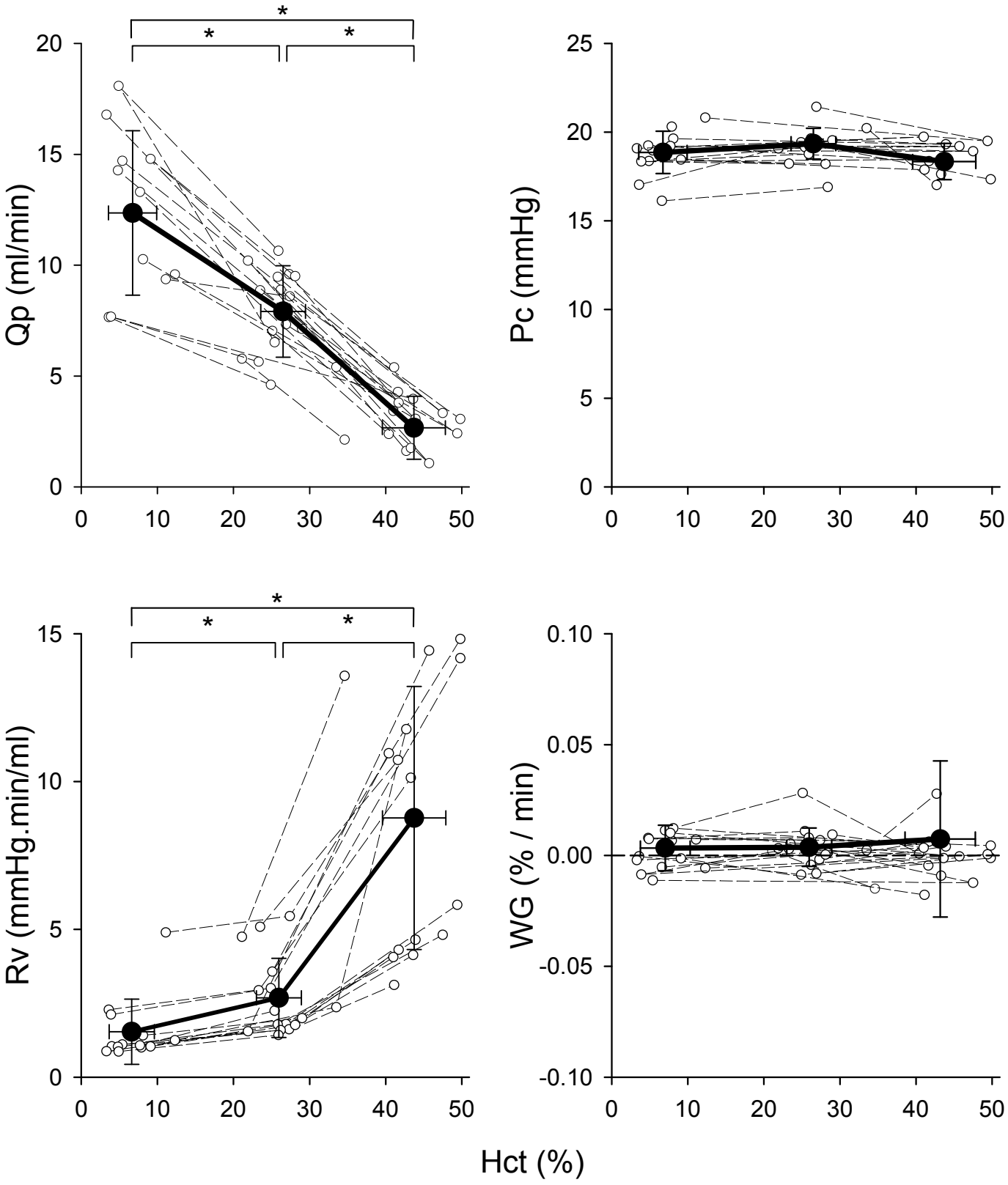
Qp (ml/min)
Pc (mmHg)
Rv (mmHg.min/ml)
WG (% / min)
Hct (%)
*
*
*
0
5
10
15
20
25
0.10
0.05
0.00
-0.05
-0.10
0
10
20
30
40
50

Figure 2 - R1

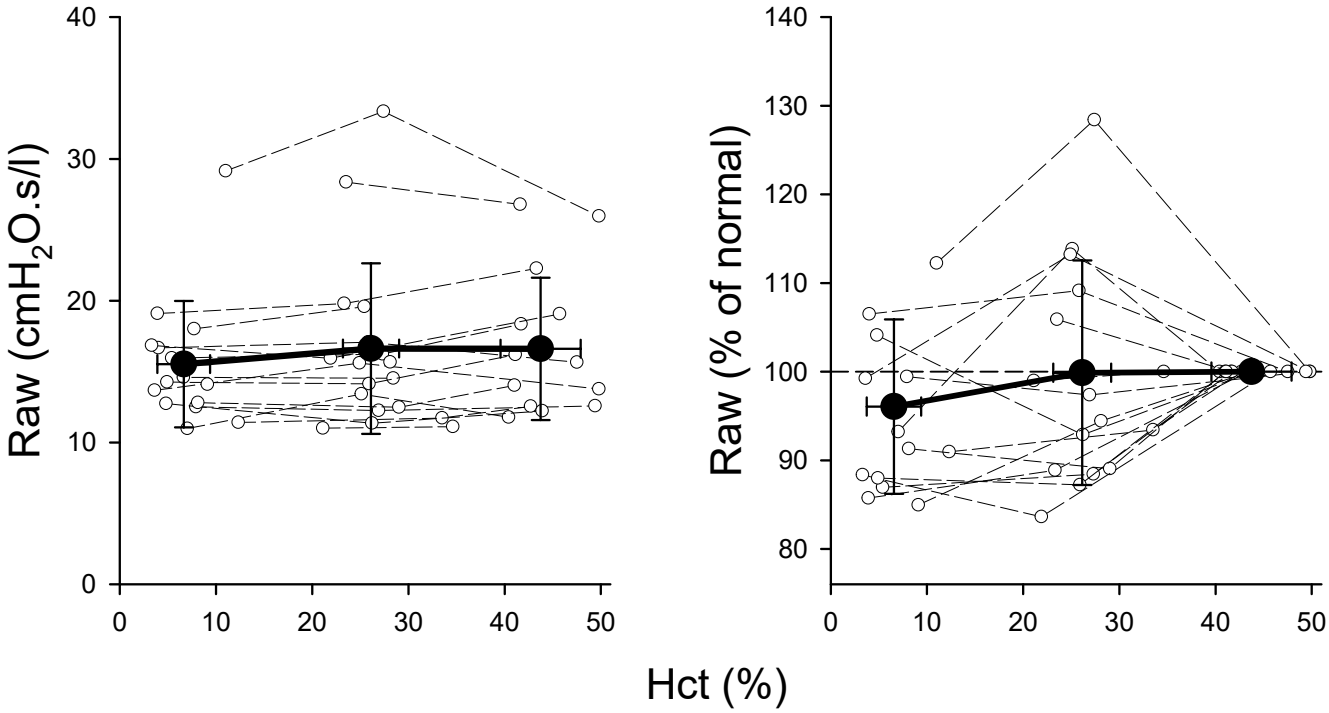
Raw ($cmH_2O.s/l$)
Raw (% of normal)
Hct (%)
0
10
20
30
40
50
80
90
100
110
120
130
140

Figure 3-R2

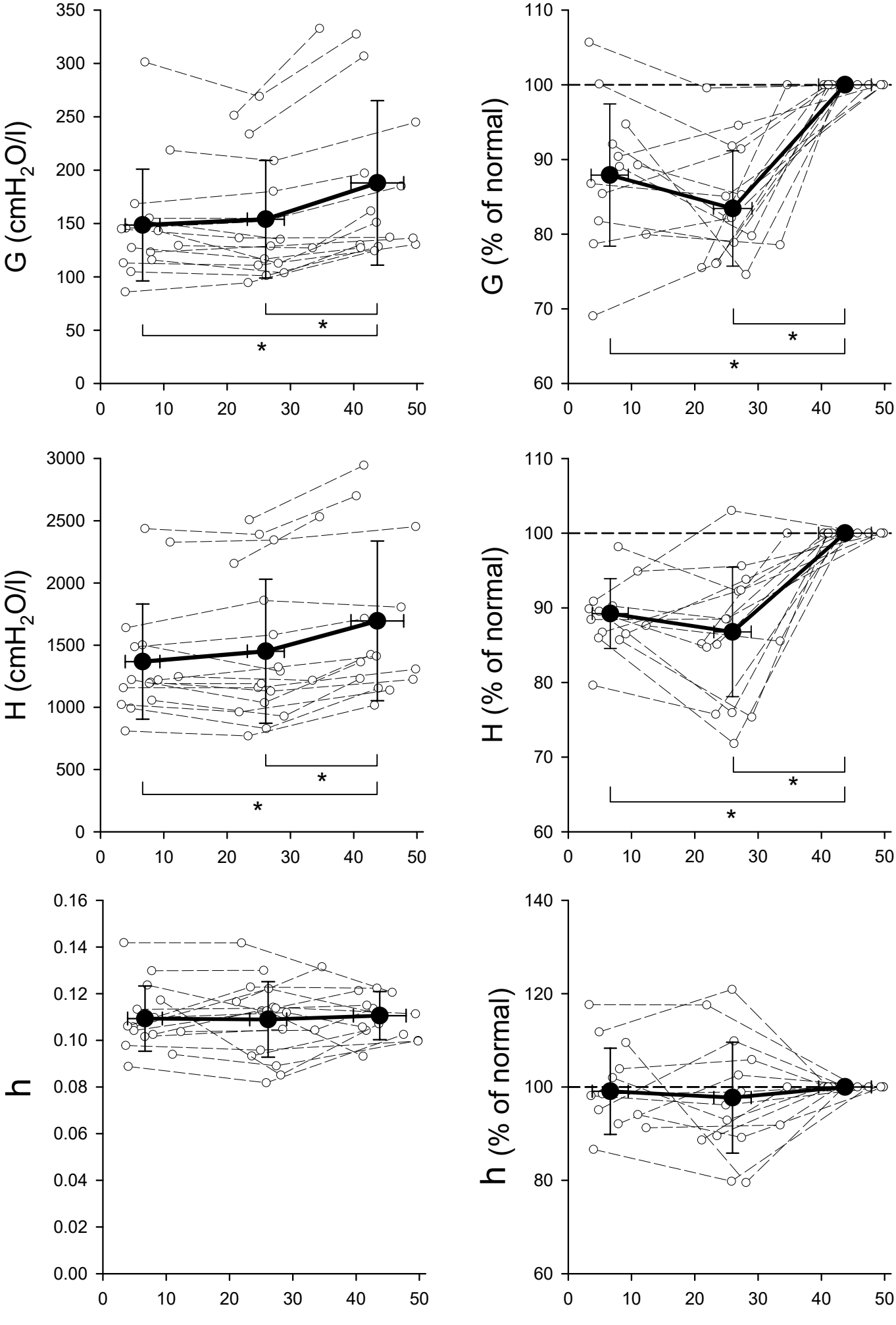

Figure 4-R2

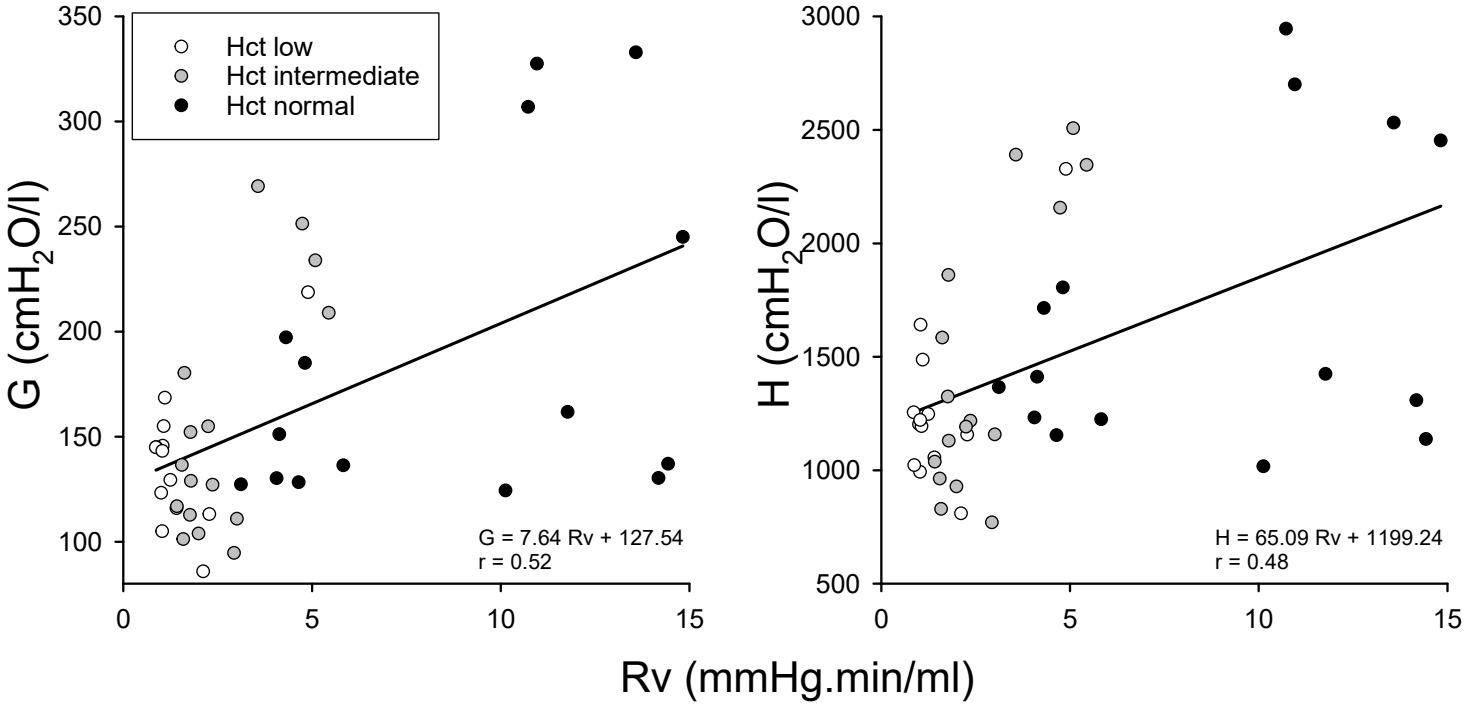
Hct low
Hct intermediate
Hct normal
G (cmH2O/l)
H (cmH2O/l)
G = 7.64 Rv + 127.54
r = 0.52
H = 65.09 Rv + 1199.24
r = 0.48
Rv (mmHg.min/ml)

Figure 5 - R2