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**Autor:** Herholz, C. / Tschudi, P. / Gerber, H.

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*Division of equine internal medicine, Clinic for horses and farm animals, University of Berne*

# Ultrasound spirometry in the horse: a preliminary report on the method and the effects of xylazin and lobelin hydrochloride medication

*C. Herholz, P. Tschudi, H. Gerber, Y. Moens, R. Straub*

## Summary

A new computerised ultrasound-based spirometry system according to Buess et al. (1995) modified by a double flow measurement facility was used to study pulmonary function in healthy horses and horses affected with subclinical and manifest chronic bronchiolitis (CB).
The horses were first evaluated at rest without any medication.
On another occasion all horses were tested following i.v. administration of xylazine (0.4 mg/kg) and following i.v. administration of lobelin hydrochloride (l.hy.; 0.2 mg/kg) to evaluate the effect of xylazine and l.hy. on different spirometric variables.
Ultrasound-based spirometry proved to be an easily applicable method for lung function testing, even in difficult horses. However, there existed a pronounced physiological variation for all measured lung function parameters and no significant differences between healthy horses and horses with chronic bronchiolitis could be found except for the expiratory tidal volume (VTE, $p < 0.05$).
Individually, a marked decrease of variability from breath to breath following either xylazin and l.hy. administration could be observed for all parameters, except the flow-time-ratio (Tpef./Texp.) and the flow-volume-ratio (Vpef./Vexp).

***Key words:*** **ultrasound spirometry – horses – chronic bronchiolitis – xylazin – lobelin hydrochloride**

## Ultraschall-Spirometrie beim Pferd: ein vorläufiger Bericht über die Methode und die Effekte von Xylazin- und Lobelinum-hydrochloricum-Medikation

Zur Lungenfunktionsprüfung bei gesunden, subklinisch und klinisch an chronischer Bronchiolitis (CB) erkrankten Pferden wurde das Ultraschall-Spirometrie-System mit entsprechender Computersoftware nach Buess et al. (1995) eingesetzt, welches durch eine bilaterale Flowmessung modifiziert wurde.
Alle Pferde wurden zunächst in Ruhe, ohne Medikation spirometriert. Des weiteren folgte die Untersuchung der Tiere nach intravenöser Gabe von Xylazin (0.4 mg/kg ) und nach intravenöser Gabe von Lobelinum hydrochloricum (L.hy.; 0.2 mg/kg), um die Auswirkungen der jeweiligen Medikation auf die spirometrischen Resultate zu prüfen.
Die Ultraschall-Spirometrie erwies sich auch bei schwierigen Pferden als einfach anzuwendende Methode zur Lungenfunktionsprüfung. Es ergaben sich bei den Ergebnissen der Messungen in Ruhe beträchtliche physiologische Schwankungen, so dass keine signifikanten Unterschiede zwischen gesunden und subklinisch bzw. klinisch an CB erkrankten Tieren ermittelt werden konnten, mit Ausnahme des exspiratorischen Atemzugvolumens (VTE, $p < 0.05$).
Von Atemzug zu Atemzug war die Variabilität der Resultate nach Injektion von Xylazin respektive L.hy, bei allen Lungenfunktionsparametern mit Ausnahme des Fluss-Zeit-Quotienten (Tpef./Texsp.) und des Fluss-Volumen-Quotienten (Vpef./Vexsp.) deutlich geringer.

***Schlüsselwörter:*** **Ultraschall-Spirometrie – Pferde – chronische Bronchiolitis – Xylazin – Lobelinum hydrochloricum**

## Introduction

Chronic respiratory disease is a common problem in horses. Several tests have long been used to evaluate lung function in the horse using e.g. the pneumotachograph as described by Fleisch (1925), or multiple breath nitrogen washout (Spörri and Lehmann, 1964; Spörri and Denac, 1967, 1970; Gillespie and Tyler, 1969; Sasse, 1971; Muylle and Oyaert, 1973; Robinson et al., 1975; Speirs, 1977; Willoughby and McDonell, 1979; Derksen et al., 1982). They have, however, never played an important role in the routine diagnostic procedure for bronchopulmonary diseases on their own, whereas the combination with direct intrapleural pressure measurements provided lung function parameters of some diagnostic value (Spörri and Leemann, 1964; Spörri and Zerobin, 1964; Spörri and Denac, 1967). Disadvantages, such as the rather complicated equipment and the time-consuming elaboration of the results, remained.
An easier method to apply is the indirect intrapleural pressure measurement using an oesophageal balloon-tipped catheter. The sensitivity of this method to detect bronchopulmonary disease is limited for reasons of inherent inaccuracy of the measurement method and, more important, the high variability of the results (Spörri and Leemann, 1964; Banchero et al., 1967; Derksen and Robinson, 1980; Hegner, 1994).
Ultrasound-based spirometry relies on the precise measurement of absolute sound travelling times through the respired gas to determine flow velocity and flow volumes (Buess et al., 1995). This technique has opened new possibilities for lung function testing by coupling to a high accuracy and computer-aided calculation of the measurements (Buess, 1988; Buess et al., 1993; Harnoncourt, 1995). Actual software offers the possibility to measure and calculate several consecutive breath cycles, eg. 25 breaths.
It was the aim of the present preliminary study to evaluate the clinical application and the sensitivity of the ultrasound spirometry system. We wanted to see if indices of lung function measured with this method differed in sound horses from animals with subclinical or manifest CB. Furthermore, in the same horses we studied the effect of xylazine and lobelin hydrochloride (l.hy.) on ultrasound spirometric parameters in an attempt to reduce the well documented breath to breath variability of lung function indices.

## Animals, material and methods

Fifteen adult horses were used (age 4 to 23 years), all of different warmblood breeds except three who were Freibergers, a native light draught breed. The horses were divided into three groups depending on their state of pulmonary health according to Herholz (1993) and Kleiber (1996), based on clinical examination including auscultation, arterial blood gas analysis, tracheobronchoscopy, cytology of tracheobronchial aspirates and bronchoalveolar lavage samples and radiographic findings. Group I (n = 4): horses without evidence of respiratory disease; group II (n = 5): horses with subclinical CB; group III (n = 6): horses with clinically manifest CB.
The mean age, body weight (kg), height at the withers (cm) and thorax circumference (cm) of group I was 6 years, 533 kg, 162 cm and 198 cm, of group II 12 years, 538 kg, 162 cm and 200 cm and of group III 7 years, 529 kg, 162 cm and 192 cm.
Pulmonary function tests were done using ultrasound-based spirometry as described by Buess et al. (1993). The

*Table 1 : Spirometric variables, abbreviations, units and references*

| variable | abbreviation | unit | references |
|---|---|---|---|
| inspiratory tidal volume | VTI | l | Spörri and Leemann (1964)<br>Spörri and Denac (1967, 1970)<br>Robinson et al. (1975)<br>Willoughby and McDonell (1979) |
| expiratory tidal volume | VTE | l | |
| respiratory frequency | fR | 1/min | |
| minute volume | VE | l/min | |
| peak inspiratory flow | PIF | l/s | Spörri and Leemann (1964)<br>Spörri und Denac (1967)<br>Speirs (1977)<br>Comroe (1968)<br>Harnoncourt (1995)<br>Morris et al. (1995) |
| peak expiratory flow | PEF | l/s | |
| ratio of expiratory to inspiratory time | Texp./Tinsp. | | Spörri und Denac (1967)<br>Gillespie and Tyler (1969)<br>Sasse (1971)<br>Muylle und Oyaert (1973) |
| flow-time-ratio | Tpef./Texp. | | Carlsen and Lødrup (1994) |
| flow-volume-ratio | Vpef./Vexp. | | Harnoncourt (1995) |

complete integrated equipment consists of the ultrasound spirometer, a special face mask featuring two ultrasonic flow sensors «Spiroson Scientic®», a computer and appropriate software. Spirometric variables, units and references are given in table 1. The measured values represent the mean of 25 consecutive breathing cycles corrected for body temperature, pressure, and water saturation (BTPS). Before and after each test the instrument was calibrated with a 7 liter calibration pump (Hans Rudolph Inc., Kansas City, USA) and tested for linearity according to the manufacturer's recommendations.
Before spirometric measurements were done, the horses were given at least five minutes to adapt to breath through the mask. Measurements were then performed during four minutes.
Measurements were first done on unmedicated horses (study A). From the same horses measurements were also obtained 5–10 minutes following administration of xylazine at a dosage of 0.4 mg/kg body weight (study B).
In a separate experiment with a smaller number of horses (group I, n = 2; group II, n = 3; group III, n = 2) we looked at the effects of lobelin hydrochloride (Lobelin®, 0.2 mg/kg i.v.) on ultrasound spirometric results.
During the whole experimental period (5 weeks) all horses were kept on straw bedding and fed hay to keep environmental influences to a minimum. No therapeutic measures of any kind were applied during this time. All studies were performed at the same hour of the day to reduce the influence of circadian rhythms on lung function variables.
Group means, standard deviations and variation coefficients were calculated for the different spirometric parameters measured in study A and B. The significance of differences of the means between the three groups was evaluated by the Kruskal-Wallis-test for unpaired observations and for the significance of differences between two different groups the Mann-Withney U-test for unpaired observations was used. The significance of differences within groups before and after medication was evaluated by the Wilcoxon-test for paired observations.
In all cases the hypothesis of no difference was tested and the significance level set at 95% ($p < 0.05$). Correlations between VE and height at the withers, body weight, thorax circumference and age were evaluated with the Spearman-Rank-test.

## Results

The mean values and standard deviations of spirometric parameters measured in the horses of the three groups without and with sedative medication are given in table 2.
There was no significant difference between the three groups of horses for body weight, height at the withers and circumference of the thorax. Mean age of group II was significantly higher (12 years) than in group I and III.
In unmedicated horses no significant difference between the groups could be detected for the spirometric parameters except for the expiratory tidal volume (VTE, $p < 0.05$). VTE was greater in sound horses (6.5± 0.9 l) compared to sick horses in group II (5.2 ± 0.6 l, $p = 0.05$) and group III (4.6 ± 1.2 l, $p = 0.03$). The same tendency existed as expected for the inspiratory tidal volume (VTI) but did not reach statistical significance.
Xylazine exerted a pronounced effect on ventilation and provoked a more uniform breathing pattern in individual horses. Additionally, the degree of change of the different respiratory parameters provoked by xylazine was highly variable within groups and ranged from 4 to 100% change (table 2).
Significant effects on spirometric parameters within the groups before and after administration of xylazine were only detected in group III for VTE, VTI (increase), the respiratory frequency (fR, decrease) and the ratio of expiratory to inspiratory time (Texsp/Tinsp, increase), ($p < 0.05$).
Following sedation with xylazine VTE and VTI ($p < 0.05$) as well as Texsp/Tinsp ($p < 0.01$) were statistically different between the groups.

*Table 2: Mean values, standard deviations and variation coefficients in percent ( ) of the variables measured in all horses of the three groups at rest A without medication, and B after 0.4 mg/kg xylazine i.v.*

| VARIABLE | GROUP I (n = 4) | | GROUP II (n = 5) | | GROUP III (n = 6) | |
|---|---|---|---|---|---|---|
| | A | B | A | B | A | B |
| **VTE (l)** | 6.5 ± 0.9 (14) | 8 ± 1.3 (16) | 5.3 ± 0.6 (12) | 5.5 ± 1.2 (21) | 4.6 ± 1.2 (26) | 6.6 ± 1.1 (17) |
| **VTI (l)** | 6.4 ± 1.1 (17) | 8.2 ± 1.3 (16) | 5.3 ± 0.5 (10) | 5.5 ± 1.2 (21) | 4.7 ± 1.3 (27) | 6.5 ± 1.10 (17) |
| **fR (1/min)** | 15 ± 5 (34) | 9 ± 3 (30) | 16 ± 4 (23) | 14 ± 5 (37) | 20 ± 8 (38) | 12 ± 2 (18) |
| **VE (l/min)** | 93 ± 36 (38) | 70 ± 20 (29) | 84 ± 25 (30) | 76 ± 27 (35) | 93 ± 41 (44) | 78 ± 21 (27) |
| **PEF (l/s)** | 5.8 ± 2.1 (35) | 4.6 ± 1.7 (36) | 5.2 ± 1.3 (25) | 4.5 ± 1.8 (39) | 5.8 ± 2.4 (41) | 4.8 ± 1.9 (39) |
| **PIF(l/s)** | 5 ± 1.6 (31) | 4.2 ± 0.7 (16) | 4.3 ± 1.1 (25) | 3.9 ± 1.4 (37) | 5 ± 1.9 (37) | 4.8 ± 1.3 (26) |
| **Texp./Tinsp.** | 1.2 ± 0.2 (18) | 1.7 ± 0.4 (23) | 1 ± 0.1(10) | 1.1 ± 0.1 (12) | 1.3 ± 0.2 (17) | 1.8 ± 0.2 (11) |
| **Tpef./Texp.** | 0.19 ± 0.07 (37) | 0.11 ± 0.04 (36) | 0.12 ± 0.02 (17) | 0.22 ± 0.21 (95) | 0.23 ± 0.13 (57) | 0.17 ± 0.20 (118) |
| **Vpef./Vexp.** | 0.23 ± 0.05 (22) | 0.17 ± 0.03 (18) | 0.19 ± 0.02 (11) | 0.29 ± 0.18 (62) | 0.26 ± 0.07 (27) | 0.23 ± 0.16 (70) |

group I: sound horses
group II: horses with subclinical chronic bronchiolitis
group III: horses with manifest chronic bronchiolitis

significant differences between the three groups:
*A* without medication : VTE
*B* after 0.4 mg/kg Xylazine i.v. : VTE, VTI, fR, Texp./Tinsp.

Of all spirometric parameters only VE (l/min) showed a significant correlation with the height at the withers ($r = 0.72$).

L.hy. administration induced a marked increase of tidal volumes, respiratory frequency, minute ventilation and peak expiratory flow; e.g. mean values unmedicated (n = 5) versus l.hy. medication (n = 4) of group II : VTE 5.3 l to 20.5 l; fR 16/min 29/min; VE 84 l/min to 600 l/min and PEF 5.2 l/s to 30 l/s. However, between the three groups, l.hy. medication provoked no obvious difference in any spirometric parameter.

## Discussion

Spontaneous breathing in the horse is quite irregular and the variability of the parameters is therefore high due to both breath-to-breath variability and within day variability (Muylle and Oyaert 1973, Gallivan et al. 1990). Earlier spirometric investigations using a pneumotachograph as presented by Spörri and Leemann (1964), Spörri and Denac (1967, 1970) or Willoughby and McDonell (1979) suffer from the fact that only one particular breath was chosen to calculate respiratory parameters. This procedure idealizes to a rather high degree and might be not representative for actual pulmonary function. Computerized ultrasound spirometry on the other hand, using the means of several breathing cycles, e.g. 25, may provide a better evaluation of lung function.

In our study ultrasound spirometry using a special face mask proved to be an easily applicable method even in difficult animals. However, on the basis of the spirometric data collected in this study variabilty of the measured parameters remained high. Of all variables only VTE ($p = 0.05$) differed between sound horses and animals with subclinical or manifest CB. We expected the same result for VTI, but the difference between the three groups did not reach statistical significance ($p = 0.08$).

Morris et al. (1995) have encountered the same difficulty in man. VTE decreased significantly from group I to III. Muylle and Oyaert (1973) found no differences in inspiratory or expiratory volumes of sound and affected horses. Gillespie and Tyler (1969) described decreased volumes in «emphysematous» horses, while Sasse (1971) observed increased volumes in COPD horses and animals suffering from «interstitial pneumonia». These contradictory observations may largely be due to the pronounced methodological subjectivity of the conventional pneumotachography. In our study the mean age of the horses in the groups was different but no significant correlations between spirometric parameters and age were found. This is in contrast with the findings of Spörri and Denac (1970) who found an age dependence of VE.

Xylazine, an $\alpha_2$-agonist, has been used to induce a quiet breathing pattern prior to lung function testing (Broadstone et al., 1992; Lavoie et al., 1992, 1996). This might reduce variability in the indices and improve sensitivity of the method. In our study xylazine influenced breathing patterns profoundly: a spirogram of a horse at rest and following xylazine is shown in figure 1. This altered breathing pattern was the result of changes in tidal volumes, respiratory frequency and expiratory: inspiratory time ratio and this is in accordance with the results of Lavoie et al (1992). Xylazine influence on lung function tests is contradictorily discussed (Clarke and Hall, 1969; Kerr et al., 1972; Rohr and Schatzmann 1982; Deegen and Klein, 1985; Reitemeyer et al., 1986; Lavoie et al., 1992, 1996).

Reitemeyer et al. (1986) and Lavoie et al. (1992) have shown that xylazine alters respiratory mecanics and inspiratory/expiratory flow. The mechanism is perhaps a presynaptical inhibition of acetylcholine release in bronchial smooth muscle (Minghu et al., 1993).

In this study xylazine administration failed to diminish the mean variability of the parameters in the different groups (table 2). The degree of change of the different respiratory parameters provoked by xylazine was individually highly variable. One horse (23 years old ) in group II showed a decrease of VTE and another horse in group II (8 years old) hyperventilated on repeated occasions.

The effect of xylazine on ventilation is time - dependent (Lavoie et al., 1992), but all measurements were made in the same time period following injection of xylazine (5–10 min).

VTE, VTI, fR and Texp./Tinsp. became statistically different between groups of horses with a different state of pulmonary health.

PEF is an important parameter to detect bronchial obstruction in man, where a marked reduction in peak expiratory flow rates was observed (Morris and Lane, 1981, Morris et al., 1995). However, no significant differences in PEF between the three groups without medication or after xylazine were found and no obvious difference was detected after administration of lobelin hydrochloride (Lobelin®, 0.2 mg/kg i.v.).

It can be concluded that the new, modified Ultrasonic spirometry technique with two flowsensors mounted in a face mask is an easily applicable method for lung function tests even in difficult horses. Of all measured pulmonary indices only VTE was shown to be smaller in horses with CB than in sound horses.

For clinical use ultrasound spirometry was not helpful to detect subclinical chronic bronchiolitis since the test results remained highly variable even with the software facilities of the system.

Xylazin and l.hy. medication influenced breathing patterns profoundly in regularising breathing cycles with a marked decrease of breath to breath variability (figure 1).

However, neither xylazin nor l.hy. administration decreased the variability of test results within the three groups and provided no advantage for the diagnosis of subclinical or clinical manifest CB by ultrasound spirometry.

Even though the ease of application, the sensitivity and accuracy of spirometry are greatly enhanced by the new

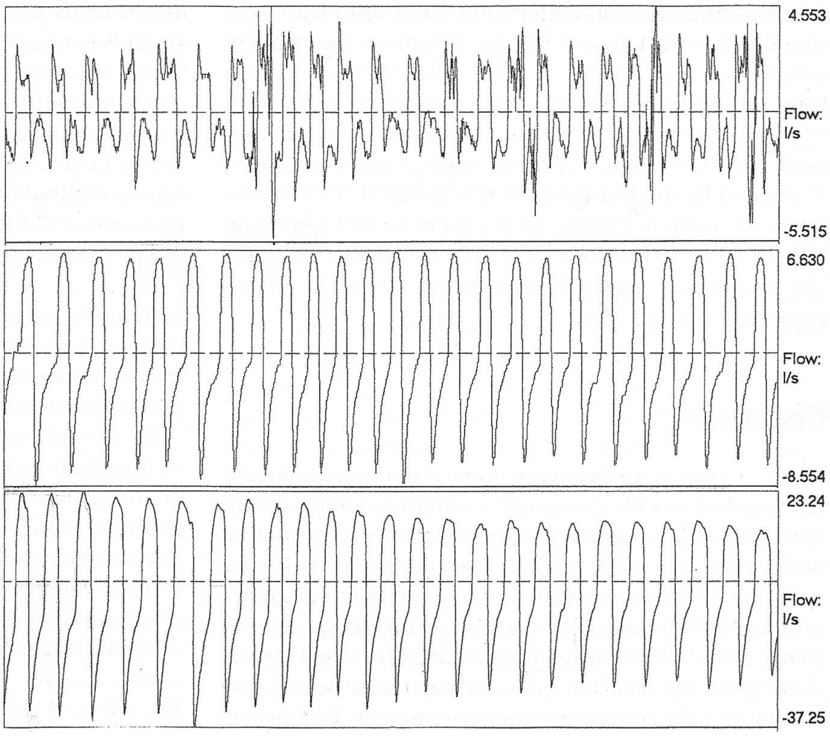


*Figure 1: Spirogramms of horse No. 12 (group III) at rest without being influenced by medication (top, fR = 20/min), following i.v. administration of 0.4 mg/kg Xylazine (middle, fR = 13/min) and after i.v. administration of 0.2 mg/kg lobelin hydrochloride (bottom, fR = 31/min).*

ultrasound method, it does not improve its clinical usefulness. Future studies combining airflow- with simultaneous carbon dioxide measurements may lead to reliable results for physiological deadspace volume and hence to an appropriate practical diagnostic method.

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## Spirométrie sonographique chez le cheval: étude préliminaire sur la méthode et l'effet de la xylazine et de l'hydrochloride de lobéline

Un nouveau système de spirométrie sonographique mis au point par Buess et al. (1995) a été utilisé pour tester la fonction pulmonaire chez des chevaux sains et des chevaux atteints de bronchiolite chronique.
En premier lieu tous les chevaux ont été évalués au repos et sans médication. L' effet de la médication sur les différents paramètres spirométriques a été étudié à différentes occasions après l'injection intraveineuse de xylazine (0.4 mg/kg) et l'injection intraveineuse d'hydrochloride de lobéline (l.hy.;0.2 mg/kg).
La spirométrie sonographique s'est avérée être une méthode d'investigation de la fonction pulmonaire particulièrement facile, même avec des chevaux à caractère difficile. A l'exception du volume tidal expiratoire - VTE - ($p < 0.05$), aucune différence significative n' a été mise en évidence entre chevaux sains et chevaux atteints de CB, à cause de la variation physiologique très importante des paramètres étudiés. L'injection de xylazine ou de l.hy était suivie d'une diminution prononcée de la variabilité entre les respirations individuelles pour tous les paramètres étudiés sauf pour le rapport temps mis jusqu'au débit expiratoire maximal/temps total de l'expiration (Tpef/Texp) et le rapport volume mis jusqu'au débit expiratoire maximal/volume total de l'expiration (Vpef./Vexp).

## Spirometria col metodo dell'ultrasuono: rapporto preliminare sul metodo e sugli effetti della medicazione con xylazina e lobelina

Per l'esame dei parametri polmonari in cavalli sani e malati di bronchiolite crinica subclinica e clinica è stato utilizzato il sistema spirometrico ad ultrasuoni secondo il metodo Buess et al. (1995) modificato tramite la misurazione dei flussi bilaterali. Tutte le misurazioni sui cavalli sono state fatte dapprima a riposo. Il test procedera quindi con l'iniezione intravenosa di Xylazina (0.4 mg/kg) e successivamente di lobelina (0.2 mg/kg) alfine di misurare l'effetto dei due medicamenti sui dati spirometrici. Il metodo della spirometria con misurazione degli ultrasuoni si è rivelato efficace per l'accertamento delle funzioni polmonari anche in cavalli difficili. Nelle misurazioni a riposo si potevano constatare grandi variazioni fisiologiche, che rendavano invano qualsiasi tentativo di intravvedere delle differenze significative fra cavalli sani e malati di bronchiolite cronica (clinica e subclinica), ad eccezione del volume espirato a riposo. (VE, $p > 0.05$). Eccetto che per i quozienti flusso-tempo e flusso-volume la variabilità dei risultati spirometrici diminuiva visibilmente col tempo sia dopo l'iniezione di xylazina che di lobelina.

*Korrespondenzadresse: Dr. C. Herholz, Klinik für Nutztiere und Pferde, Universität Bern, Länggassstrasse 124, CH-3012 Bern*

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