

Abnormal vocal cord movement in patients with and without airway obstruction and asthma symptoms

K. Low^{1,2,*}, L. Ruane^{1,2,*}, N. Uddin¹, P. Finlay¹, K. K. Lau³, K. Hamza⁴, P. W. Holmes¹, G. Hamilton^{1,2} and P. G. Bardin^{1,2}

¹Monash Lung and Sleep, Monash University and Hospital, and ²Hudson Institute, ³Diagnostic Imaging, and ⁴Statistical Services, Monash University and Hospital, Melbourne, Vic., Australia

Clinical & Experimental Allergy

Summary

Background Abnormal vocal cord movements can cause laryngeal extrathoracic airway obstruction (often called vocal cord dysfunction – VCD) leading to asthma-like symptoms. These aberrant movements are characteristically present during inspiration and termed paradoxical vocal cord movement (PVC). We have reported PVC in up to 40% of severe asthmatics, but it is not known if PVC is detectable in all patients with asthma-like symptoms and if the condition is more often associated with abnormal lung function.

Objective We hypothesized that PVC is frequently associated with asthma symptoms accompanied by airflow limitation. Studies examined whether PVC is solely linked to experiencing asthma symptoms, or if PVC is related to airflow limitation and/or other disease characteristics.

Methods Patients with asthma symptoms were recruited from general practice and severe asthma clinics ($n = 155$). Pulmonary function measurements were conducted, asthma control and Nijmegen (dysfunctional breathing) questionnaires were administered and skin prick testing was carried out. PVC was quantified using dynamic 320-slice computerized tomography of the larynx. Groups were divided into patients with $FEV_1 \geq 80\%$ predicted or $FEV_1 < 80\%$ predicted and $FEV_1/FVC < 0.7$. ATS/ERS definitions of severity were also applied and evaluated. Detection of PVC in the groups was compared and analyses performed to identify features associated with PVC.

Results Overall ($n = 155$), PVC was detected in 42 cases (27.1%). Patients with $FEV_1 < 80\%$ predicted had PVC more often (25/68, 36.8%) than individuals with normal spirometry (17/87, 19.5%; $P = 0.016$). PVC was associated with older age ($P = 0.003$) and with Nijmegen scores > 20 ($P = 0.04$). Patients with $FEV_1 < 80\%$ predicted plus Nijmegen scores > 20 were more likely to have PVC (OR = 9.3, $P = 0.02$).

Conclusions and Clinical Relevance Paradoxical vocal cord movement is more often associated with asthma symptoms accompanied by airflow limitation and dysfunctional breathing. Further studies are needed to determine whether PVC is induced by dysfunctional breathing practices and/or airway obstruction. How PVC links with symptomatic asthma and VCD also requires evaluation.

Keywords airflow obstruction, asthma, dysfunctional breathing, paradoxical vocal cord movement, vocal cord dysfunction

Submitted 22 March 2016; revised 24 August 2016; accepted 29 August 2016

Correspondence:

Prof. Philip G. Bardin, Lung and Sleep, Monash Medical Centre, 246 Clayton Road, Clayton, Melbourne, Vic 3168, Australia.

E-mail: philip.bardin@monash.edu

Cite this as: K. Low, L. Ruane, N.

Uddin, P. Finlay, K. K. Lau, K. Hamza,

P. W. Holmes, G. Hamilton and P. G.

Bardin, *Clinical & Experimental*

Allergy, 2017 (47) 200–207.

Introduction

Abnormal vocal cord movement can cause extrathoracic airway obstruction and asthma-like symptoms [1]. The best-known condition causing obstruction at the

level of the larynx is vocal cord dysfunction (VCD) [2]. First described in 1983, VCD was considered to mimic asthma [3] but subsequent studies have revealed that asthma and VCD frequently coexist [4–7]. It has raised the prospect that VCD could intensify already existing asthma symptoms [8]. Consequently, VCD has long been suspected as a cause of treatment-resistant asthma as

*These authors contributed equally to the study.

current asthma treatments are effective to control disease manifestations in a majority of patients [2].

Vocal cord dysfunction is characterized by abnormal vocal cord movement with inspiratory closure of the vocal cords [3] termed paradoxical vocal cord movement (PVC) [5]. During PVC, the true vocal cords close anteriorly with formation of a posterior-narrowed diamond-shaped 'chink'. The false cords are not affected except in severe cases [3]. It is not known how severe, sustained and frequent PVC has to be for symptoms of VCD to occur. This is a diagnostic challenge given a continuum of abnormality and distinguishing movement abnormality (PVC – as evaluated in the current studies) from disease expression with symptoms (VCD) is difficult. However, PVC can be a valuable proxy measure of VCD providing a quantitative criterion for research. Using dynamic 320-slice CT larynx, we have developed a technique to quantify PVC and our previous studies have shown that the abnormality can be detected during inspiration in more than 40% of difficult-to-treat asthmatics [6]. Our findings suggested that PVC may occur frequently in severe asthma, but it is not known whether PVC may be an integral component of asthma itself and whether it is linked to any other particular clinical features of asthma. Additionally, recent reports have suggested that dysfunctional breathing techniques used by some asthmatics may lead to disproportionate asthma symptoms that resemble VCD-like symptoms [9, 10].

Given the strong association of VCD with asthma [6, 7], we hypothesized that PVC may occur more frequently in patients with asthma symptoms accompanied by airflow limitation. Studies therefore examined whether detection of PVC is linked to abnormal lung function and other patient characteristics.

Methods

Studies were conducted at Monash Medical Centre, a tertiary care hospital in Melbourne, Australia. Study protocols were reviewed and approved by Monash Health Human Research Ethics Committee (09254A, 12158A and 15156Q), and the written informed consent was obtained from the patient.

Study subjects

Overall, 155 patients were recruited in collaboration with their general practitioner from general practice or from hospital asthma clinics using similar criteria for asthma as employed in the ISAAC phase III survey [11]. Asthma was diagnosed based on the following: (i) a typical past history of asthma symptoms, (ii) favourable clinical responses to treatment with beta-agonists and/or inhaled corticosteroids (ICSs) and (iii) a diagnosis of

asthma by their general practitioner. At the time of study, all patients were in possession of at least a beta-agonist inhaler (see Supporting information for further detail and motivation of recruitment strategy). Patients reported in our previous studies [6] were not included.

Demographic characteristics of the study group are shown in Table 1. Overall, 71 of 155 patients had bronchodilator responses $\geq 12\%$ (30 patients with $FEV_1 > 80\%$ predicted and 41 patients with $FEV_1 < 80\%$ predicted). An additional 27 patients had $FEV_1 < 80\%$ predicted with $FEV_1/FVC < 70\%$ in the absence of smoking (group with airway obstruction: 41 + 27; $n = 68$). The remaining 57 patients all had $FEV_1 \geq 80\%$ predicted (group without airway obstruction: 30 + 57; $n = 87$) with a history of asthma symptoms and were enrolled as per the ISAAC criteria [11]. Although bronchial provocation testing has considerable utility in asthma diagnosis, this was not performed as tests may have limited specificity in very mild asthmatics with normal FEV_1 [12]. All patients were non-smokers or had < 10 -pack-year smoking history.

To extend the analyses, patients were also grouped using the definition of severe asthma promulgated by the ATS/ERS [2]. Other ATS/ERS definitions were also used to further group severity [2] using asthma control test (ACT) scores < 16 and asthma control questionnaire-5 (ACQ5) scores > 1.5 [13, 14]. As recent reports implicated dysfunctional breathing in asthma [9, 10], we examined putative mechanisms of PVC in the final 30 patients recruited to the study using the Nijmegen questionnaire. Nijmegen scores > 20 are considered indicative of the presence of dysfunctional breathing [15] although the instrument can be considered imprecise and relatively non-specific. This group's characteristics were similar to the overall group (Table S1).

320-slice CT larynx

CT larynx was carried out as detailed previously [6] (see Supporting information). Briefly, studies were performed using a Toshiba Aquilion One CT (Toshiba Medical Systems, Tokyo, Japan) with 320-slice detector. Radiation doses are modest and generally < 1.5 mSv. Integrated CT software programs were used to obtain continuous dynamic axial, sagittal and coronal multiplanar images and lateral diameter at the level of the vocal cords and first tracheal ring was measured on axial views. The ratio of vocal cord lateral diameter to tracheal diameter (RATIO) was obtained and an in-house computerized program (HAMZACT) automatically constructed a curve based on the RATIO generated every 0.35 s. A predicted mean normal curve with a second curve depicting the lower limit of normal (LLN) had previously been generated in studies conducted in

	All patients (<i>n</i> = 155)	FEV ₁ ≥ 80% (<i>n</i> = 87)	FEV ₁ < 80% (<i>n</i> = 68)	<i>P</i> -value
Age (m ± SD)	53.6 ± 16.2	52.6 ± 15.6	54.8 ± 17.1	0.38
Gender (M/F)	42/113	26/61	16/52	0.47
BMI (kg/m ²)	30.5 ± 7.5	30.1 ± 7.0	31.1 ± 8.2	0.38
FEV ₁ (% predicted)	81.9 ± 25.1	100.2 ± 13.7	58.6 ± 15.1	< 0.0001
FEV ₁ /FVC (%)	70.33 ± 11.89	77.00 ± 6.71	61.78 ± 11.59	< 0.0001
TLco	81.78 ± 14.75	85.90 ± 11.81	76.38 ± 16.31	0.001
BD response (% change)	7.1 ± 10.3	4.1 ± 7.2	12.0 ± 11.7	< 0.0001
ACT score*	15.3 ± 5.8	17.2 ± 6.0	13.1 ± 4.5	0.005
ACQ5 score*	2.0 ± 1.3	1.3 ± 1.1	2.7 ± 1.2	< 0.0001
Nijmegen score†	24.1 ± 9.3	22.1 ± 9.3	25.8 ± 9.1	0.25
Medications				
SABA only	47/155	47/87	0/68	< 0.0001
ICS only	16/155	10/87	6/68	0.08
ICS and LABA	92/155	30/87	62/68	< 0.0001
Other‡	5/155	4/87	1/68	0.38

PVCM, paradoxical vocal cord movement; BMI, body mass index; FEV₁, forced expiratory volume–1 s; FVC, forced vital capacity; TLco, transfer factor for carbon monoxide; BD, bronchodilator; SABA, short-acting beta-agonist; ICS, inhaled corticosteroid; LABA, long-acting beta-agonist.

*Total 91 subjects, 48 had normal FEV₁, 43 had FEV₁ < 80% predicted.

†Total 30 subjects, 14 had normal FEV₁, 16 had FEV₁ < 80% predicted.

‡One nedocromil, two montelukast, one cromoglycate, one tiotropium.

healthy individuals [6]. For an abnormal curve to occur, the patient curve has to be below the LLN curve (for an example see Fig. S1) during inspiration. Severity of PVCM was assessed by measurement of the area of the patient's RATIO < LLN curve and expressed as % of overall area < LLN.

The level of agreement between PVCM detected on a first CT larynx was evaluated in 37 patients who consented to have repeat CT larynx within a 24-month period (see Fig. S2). Patient characteristics were similar to the overall group (not shown). Cohen's kappa coefficient for categorical data was used to measure agreement [16].

Lung function testing

Lung function testing (see Supporting information) was carried out according to American Thoracic Society guidelines [17] using predictive equations published by Knudson *et al.* [18].

Skin prick testing

This was carried out using a standard panel of allergens (*n* = 9, Supporting information) and according to established guidelines. A positive skin test reaction was distinguished if wheal size for response to at least one allergen was ≥ 5 mm compared to the control [19, 20].

Table 1. Characteristics of subjects enrolled in studies of PVCM

Statistical analysis

Primary outcome of the study was detection of PVCM in asthmatics with FEV₁ < 80% predicted compared to subjects with FEV₁ ≥ 80% predicted. Based on our previous studies that detected PVCM during inspiration in approximately 40% of difficult-to-treat asthmatics, we postulated that PVCM in subjects with FEV₁ ≥ 80% predicted would be half, that is 20% (0.4 vs. 0.2). For power 0.8, *P* < 0.05 (Fisher's exact test) and allocation ratio of one, a sample size of 146 patients was required (73 per group).

Secondary outcomes evaluated detection of PVCM in patient groups based on the treatment requirements (ICS plus long-acting beta-agonist for severe asthma in ATS/ERS definition). Additional analyses were conducted using ACT < 16 or ACQ5 > 1.5 [2]. Factors putatively associated with PVCM (age, gender, BMI, Nijmegen scores, bronchodilator responses) were assessed using regression analyses. Patients with/without PVCM were compared based on the positive-or-negative skin prick test reactions or Nijmegen scores > 20 using Fisher's exact test. As FEV₁ < 80% predicted ACQ5 scores and Nijmegen scores were significantly linked to detection of PVCM, we combined FEV₁ < 80% plus ACQ5 scores > 1.5 or FEV₁ < 80% plus Nijmegen scores > 20. Fisher's exact test and logistic regression were used to examine associations with PVCM and to calculate odds ratios (ORs).

All analyses were carried out using SPSS (SPSS Inc, Chicago, IL, USA), and $P \leq 0.05$ was considered significant.

Results

Characteristics of subjects

Baseline demographic details of patient groups with $FEV_1 < 80\%$ predicted or $FEV_1 \geq 80\%$ predicted are shown in Table 1. Both ACT and ACQ5 scores reflected poorer asthma control in patients with $FEV_1 < 80\%$ predicted, but Nijmegen scores were not significantly different. Asthma medications used by the group with $FEV_1 < 80\%$ predicted were reflective of more severe disease.

Paradoxical vocal cord movement in asthma with and without airflow limitation

Overall PVCMM during inspiration was detected in 42 of 155 asthmatics (27.1%) and the % area $< LLN$ was $16.8 \pm 12.5\%$ (mean $\pm$ SD; minimum 2% and maximum 65%). The condition was more often present in asthma with $FEV_1 < 80\%$ predicted (25/68, 36.8%) than in the group with normal spirometry (17/87, 19.5%, $P = 0.016$; Fig. 1). Adjusting for age did not alter this finding. Comparisons of the patients who achieved evidence on spirometry of asthma vs. those who did not was identical to the overall findings. Similarly, occurrence of PVCMM in the $FEV_1 \geq 80\%$ group with ($n = 30$) and without ($n = 57$) bronchodilator responses $\geq 12\%$ was the same (19%). Finally, there were 15 patients with expiratory PVCMM only and hence $42 + 15 = 57$ cases (36.7%) with seven of 15 cases having $FEV_1 < 80\%$ predicted. If inspiratory and expiratory

PVCMM are combined, the overall results were unchanged (not shown).

Comparison of PVCMM in patients without and with airflow limitation was performed (Table S2). There were no differences linked to detection of PVCMM although a trend was noted for patients with $FEV_1 < 80\%$ and detectable PVCMM to have higher Nijmegen scores.

If the definition of asthma severity promulgated by the ATS/ERS guidelines [2] was applied to the cohort (GINA step 4–5 recommended treatment: $n = 95$, compared to GINA step 1–3 treatment: $n = 60$), there were no differences in the rate of PVCMM (29/95 vs. 13/60; $P = 0.26$). If ACT scores < 16 and ACQ scores > 1.5 were used to assess asthma severity and control as proposed [2], there were again no differences noted in the occurrence of PVCMM (not shown).

Analyses of repeat CT found a significant association between the first and second CT ($P = 0.025$) with agreement in 27 of 37 cases (73%, Fig. S2). Cohen's kappa coefficient denoted moderate agreement ($\kappa = 0.42$).

Patient and asthma characteristics associated with paradoxical vocal cord movement

The association of patient and asthma characteristics with PVCMM was examined (Table S3). Increased age was associated with PVCMM ($P = 0.003$), but none of the other features were linked to PVCMM including elevated BMI and female gender. Mean Nijmegen scores were higher in PVCMM but did not reach significance, most likely because asthma by itself often yields higher scores [21] (Fig. S3).

Clinical and diagnostic cut-offs were employed to categorize patients (Table 2). BMI $> 30 \text{ kg/m}^2$, $FEV_1 < 80\%$ predicted, Nijmegen scores > 20 , ACT scores < 16 , ACQ5 scores > 1.5 , BD responses $\geq 12\%$ and presence/absence of atopy were used to group patients. Asthmatics with PVCMM had greater airflow limitation manifest by $FEV_1 < 80\%$ predicted ($P = 0.02$) and ACQ5 scores > 1.5 more often ($P = 0.01$). Notably, Nijmegen scores > 20 were more frequently associated with demonstrable PVCMM (four of 11 if scores ≤ 20 vs. seven of 11 if scores > 20 ; $P = 0.04$; see also right panel Fig. S3). Using a threshold score of 23 did not alter the findings. Combining $FEV_1 < 80\%$ predicted plus ACQ5 scores > 1.5 was associated with PVCMM ($P = 0.02$ with OR = 3.03). $FEV_1 < 80\%$ predicted plus Nijmegen scores > 20 were also significantly associated with PVCMM ($P = 0.02$ with OR = 9.3, Table 2).

Discussion

This study demonstrates for the first time that PVCMM during inspiration occurs in approximately 37% of patients with asthma symptoms accompanied by

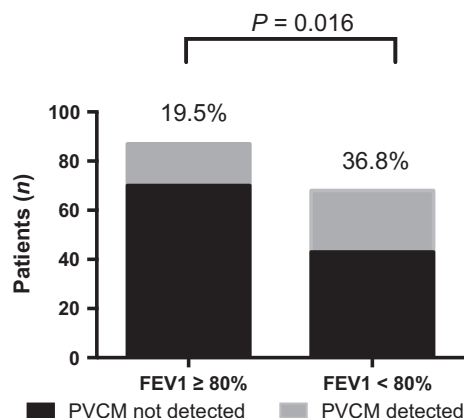


Fig. 1. Paradoxical vocal cord movement (PVCMM) can be detected more frequently in asthmatics with $FEV_1 < 80\%$ (% predicted). PVCMM was identified in 25 of 68 (36.8%) of this group vs. 17 of 87 (19.5%) of patients who had $FEV_1 \geq 80\%$ predicted ($P = 0.016$).

Table 2. Association of patient and asthma disease features with PVCMM

	PVCM not detected (<i>n</i> = 113)	PVCM detected (<i>n</i> = 42)	OR	<i>P</i> -value
Gender (Female)	84/113	29/42	0.77	0.54
BMI > 30 kg/m ²	50/113	20/42	1.14	0.72
FEV ₁ < 80% (predicted)	43/113	25/42	2.3	0.02
FEV ₁ ≥ 80% (predicted)	70/113	17/42	—	—
Nijmegen score > 20 (<i>n</i> = 11)*	4/19	7/11	6.5	0.04
Nijmegen score ≤ 20 (<i>n</i> = 19)	15/19	4/11	—	—
ACT score < 16 (<i>n</i> = 45; overall <i>n</i> = 91) [†]	29/65	12/26	1.0	0.81
ACQ5 score > 1.5 (<i>n</i> = 48; overall <i>n</i> = 91) [†]	13/65	13/26	4.0	0.01
BD response ≥ 12%	34/113	9/42	0.63	0.31
Atopy (SPT+)	23/113	12/42	1.57	0.28
FEV ₁ < 80% predicted plus ACQ > 1.5 (overall <i>n</i> = 91)	15/62	14/29	3.03	0.02
FEV ₁ < 80% predicted plus Nijmegen score > 20 (overall <i>n</i> = 30)	6/19	9/11	9.30	0.02

PVCM, paradoxical vocal cord movement; BMI, body mass index; ACT, Asthma Control Test; FEV₁, forced expiratory volume–1 s; FVC, forced vital capacity; BD, bronchodilator; SPT, skin prick testing.

*Obtained in 30 patients overall; *n* = 11 had PVCMM.

[†]Obtained in 91 patients overall; *n* = 26 had PVCMM.

significant airflow limitation. The condition can be detected about half as often in individuals with asthma symptoms who have normal lung function. Our findings also indicate that PVCMM may occur more commonly in asthmatics who have airway obstruction plus associated dysfunctional breathing responses.

The importance and role of upper/middle airway dysfunction leading to extrathoracic airway obstruction as a mimic of asthma symptoms remains controversial [8]. Uncertainty and ambiguity remain about exact links between abnormal movements of the vocal cords and the generation of asthma-like symptoms. Moreover, PVCMM/VCD has inadvertently become the collective term for a variety of clinical presentations due to disorders of the larynx. There is uncertainty surrounding nomenclature [8], diagnostic criteria and optimal treatment modalities [5]. Although individual case reports [22] and our recent study [23] demonstrated improvements in asthma control using botulinum toxin, it is not known how severe, sustained and frequent PVCMM has to be for clinical symptoms to occur and for VCD to be clinically significant. Using PVCMM as a proxy for VCD, our previous studies have detected inspiratory PVCMM in approximately 40% of a cohort of difficult-to-treat severe asthmatics with variable impairment of lung function [6]. However, the occurrence of PVCMM in mild asthma was not investigated, and severity was not based on objective criteria – such as measurements of lung function. The patient population for the current study was therefore chosen to answer a key question raised in our initial studies: Is PVCMM chiefly associated with asthma characterized by abnormal lung function or is it a widespread phenomenon in all individuals who experience typical asthma symptoms, even in the absence of airflow limitation? The current studies

represent a pragmatic approach that permitted comparisons in patients with asthma symptoms attributable to asthma and possibly associated PVCMM (see Supporting information). However, it is important to appreciate that PVCMM was not assessed in patients who uniformly had verified asthma and that this may have influenced the rate of detection.

Asthma characterized by an objective measurement (FEV₁ < 80% predicted and FEV₁/FVC < 0.7) was compared as a primary outcome to asthma without evidence of airflow limitation. We found that PVCMM was detectable in 27.1% overall and approximately twice as often in the group with airflow limitation (36.8%) compared to patients with 'milder' disease (19.5%, *P* = 0.016). If severity was defined by requirement for GINA step 4–5 medications (ATS/ERS criteria), PVCMM was detected at similar rates in severe and non-severe asthma. This is not entirely surprising as medication requirements can be inaccurate and identify patients as severe asthmatics who are high symptom perceivers. This finding also pertained to ACT and ACQ5 when these measures were used to group patients. Importantly, the association of PVCMM with airflow limitation demonstrated here has never been reported and hints at the possibility that intrinsic disease factors may elicit aberrant extrathoracic responses.

There is uncertainty surrounding the importance of PVCMM during expiration [24–26]. Vocal cord narrowing during expiration is physiological, but the degree is uncertain [24–28] and PVCMM during inspiration was employed in the original descriptions of VCD [3]. For the current studies, PVCMM was therefore defined as excessive narrowing during inspiration. A related question is how persistent PVCMM may be such that CT (or endoscopy) at a random time when symptoms may be

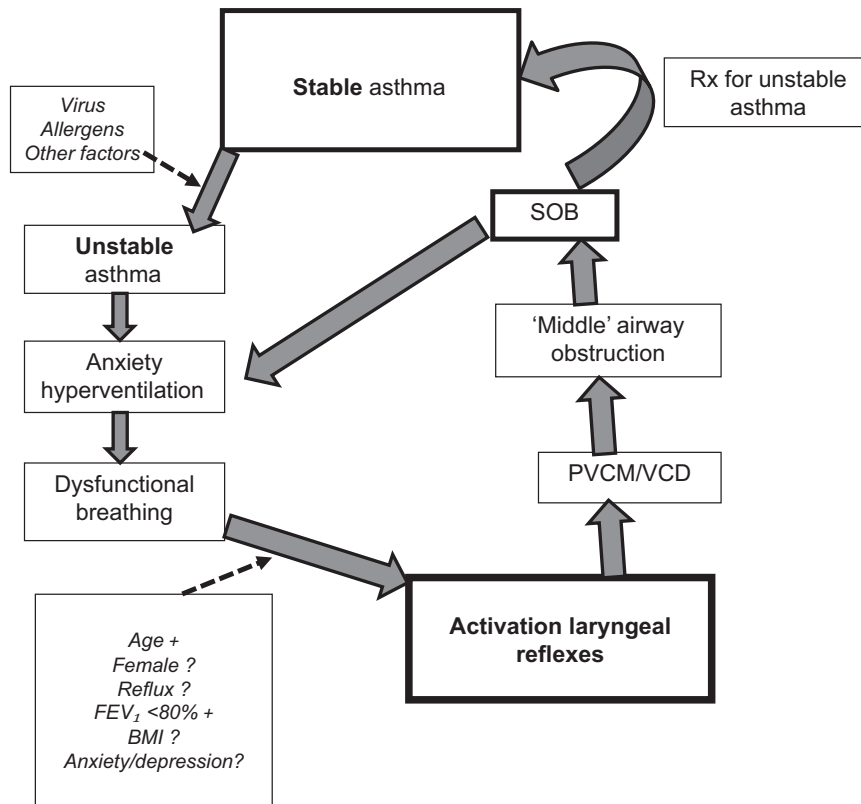


Fig. 2. Pathways postulated to lead to development of paradoxical vocal cord movement (PVC) and vocal cord dysfunction (VCD). Unstable asthma may induce dysfunctional breathing patterns leading to activation of laryngeal reflexes – followed by PVC and VCD. If severe PVC occurs, breathlessness may ensue leading to augmented dysfunctional breathing and worsening of PVC and VCD. Improved asthma symptoms and control may reduce or resolve PVC and VCD. SOB; shortness of breath; '?' denotes proposed risk factor; '+' denotes risk factor noted in current study.

absent is representative of overall vocal fold function – or dysfunction. The solitary endoscopic study of patients with symptomatic PVC found that all subjects ($n = 50$) had PVC detectable on repeat endoscopy when they were asymptomatic [29]. We evaluated detection of PVC on a subsequent CT larynx and found that agreement between the first and second CT larynx was moderate (27/37; $\kappa = 0.42$). Interestingly, PVC was not detectable on the first CT but detectable on the second CT in 7/26 (27%) cases and meant that 18 of 37 (48%) of the overall group had evidence of PVC after two CTs. This non-invasive and convenient modality is yet to gain widespread application, and it seems likely that accurate and sensitive diagnosis of VCD will ultimately be made up of several diagnostic components that may include questionnaires, endoscopy and CT larynx [30]. Taken together, our findings raise the possibility that VCD (at least in asthma) may not be intermittent but exist as a continuum of laryngeal dysfunction that can contribute to breathlessness and may be detectable – if PVC intensifies. Further research is needed to determine whether CT-based detection of PVC can be used as a reliable and

practicable 'biomarker' for VCD. Finally, no evidence of PVC manifesting as flattening of inspiratory flow–volume curves was detectable on spirometry and this accords with other studies in patients with verified PVC and VCD [5–7].

Individual patient and asthma features such as age, BMI, gender, atopy, asthma control scores and dysfunctional breathing patterns may be linked to the occurrence of PVC. Increased age but not asthma duration was associated with detection of PVC. As the history of asthma duration is often not accurate [31], age may be a better surrogate measure of duration and we speculate that PVC could be a 'complication' linked to duration of asthma, much as diabetic renal complications increase over time. Elevated BMI and/or female gender have been thought to be overrepresented in PVC and VCD [7], but this association was not observed.

Dysfunctional breathing occurs more commonly in asthmatics compared to healthy individuals [21, 32], but we also found an independent association of PVC with dysfunctional breathing as measured using the Nijmegen score. If airflow limitation was coupled to high dysfunctional breathing scores, the association

with PVCMM was particularly strong (OR = 9.3; Table 2). This finding requires confirmation in larger studies but suggests that there may be a link between PVCMM and how individuals regulate breathing patterns, perhaps simply because they have asthma or have asthma that is accompanied by airflow limitation. Collectively, these observations hint at the possibility that PVCMM may be initiated as a part of normal breathing adaptations (e.g. hyperventilation) occurring in asthmatic individuals in response to airflow limitation [33–35]. Hyperventilation and/or other forms of dysfunctional breathing may in turn lead to activation of laryngeal reflexes [36–38] with development of PVCMM (Fig. 2).

In conclusion, this study provides evidence that PVCMM (and therefore by implication VCD) can be detected more often in asthma with airway obstruction and raises the possibility that the abnormality may be interrelated with asthma itself. Dysfunction of the 'middle' airway merits greater attention in asthma [8], and research is needed to ascertain to what extent extrathoracic airway obstruction, PVCMM and VCD contribute to asthma symptoms. More information is needed to understand these conditions as effective treatment of intermittent middle airway obstruction may help to reduce disproportionate airway symptoms and improve seemingly poor asthma control.

References

- Bucca C, Rolla G, Brussino L, De Rose V, Bugiani M. Are asthma-like symptoms due to bronchial or extrathoracic airway dysfunction? *Lancet* 1995; **346**:791–5.
- Chung KF, Wenzel SE, Brozek JL *et al*. International ERS/ATS guidelines on definition, evaluation and treatment of severe asthma. *Eur Respir J* 2014; **43**:343–73.
- Christopher KL, Wood RP II, Eckert RC, Blager FB, Raney RA, Souhrada JF. Vocal-cord dysfunction presenting as asthma. *N Engl J Med* 1983; **308**:1566–70.
- Holmes PW, Lau KK, Crossett M *et al*. Diagnosis of vocal cord dysfunction in asthma with high resolution dynamic volume computerized tomography of the larynx. *Respirology* 2009; **14**:1106–13.
- Ibrahim WH, Gheriani HA, Almohamed AA, Raza T. Paradoxical vocal cord motion disorder: past, present and future. *Postgrad Med J* 2007; **83**:164–72.
- Low K, Lau KK, Holmes P *et al*. Abnormal vocal cord function in difficult-to-treat asthma. *Am J Respir Crit Care Med* 2011; **184**:50–6.
- Newman KB, Mason UG, Schmalzing KB. Clinical features of vocal cord dysfunction. *Am J Respir Crit Care Med* 1995; **152**:1382–6.
- Bardin PG, Johnston SL, Hamilton G. Middle airway obstruction—it may be happening under our noses. *Thorax* 2013; **68**:396–8.
- Agache I, Ciobanu C, Paul G, Rogozia L. Dysfunctional breathing phenotype in adults with asthma – incidence and risk factors. *Clin Transl Allergy* 2012; **2**:18.
- Jones M, Harvey A, Marston L, O'Connell NE. Breathing exercises for dysfunctional breathing/hyperventilation syndrome in adults. *Cochrane Database Syst Rev* 2013; **5**:CD009041.
- Ellwood P, Asher MI, Beasley R, Clayton TO, Stewart AW, Committee IS. The international study of asthma and allergies in childhood (ISAAC): phase three rationale and methods. *Int J Tuberc Lung Dis* 2005; **9**:10–6.
- Cockcroft D, Davis B. Direct and indirect challenges in the clinical assessment of asthma. *Ann Allergy Asthma Immunol* 2009; **103**:363–9.
- Juniper EF, O'Byrne PM, Guyatt GH, Ferrie PJ, King DR. Development and validation of a questionnaire to measure asthma control. *Eur Respir J* 1999; **14**:902–7.
- Schatz M, Sorkness CA, Li JT *et al*. Asthma control test: reliability, validity, and responsiveness in patients not previously followed by asthma specialists. *J Allergy Clin Immunol* 2006; **117**:549–56.
- van Dixhoorn J, Duivenvoorden HJ. Efficacy of Nijmegen Questionnaire in recognition of the hyperventilation syndrome. *J Psychosom Res* 1985; **29**:199–206.
- Cohen J. A coefficient of agreement for nominal scales. *Educ Psychol Meas* 1960; **20**:37–46.
- Lung function testing: selection of reference values and interpretative strategies. American Thoracic Society. *Am Rev Respir Dis* 1991; **144**:1202–18.
- Knudson RJ, Slatin RC, Lebowitz MD, Burrows B. The maximal expiratory flow-volume curve. Normal standards, variability, and effects of age. *Am Rev Respir Dis* 1976; **113**:587–600.

Acknowledgements

We thank Belinda Thomas, Jason Barker, Ine Andersen, Marcus Crossett and Joanne McKenzie for assistance with patient recruitment and CT imaging.

Author contributions

KL, PWH, GH and PGB conceived studies and designed the protocol, KL, LR, NU, PF and KL recruited subjects and conducted studies, KH performed statistical analyses and all authors participated in data analyses and writing of the manuscript.

Funding

The study was funded from Monash Lung & Sleep Institute and Monash Health, Melbourne, Australia.

Conflict of interest

None of the authors have any conflict of interest pertaining to the work detailed in this manuscript.

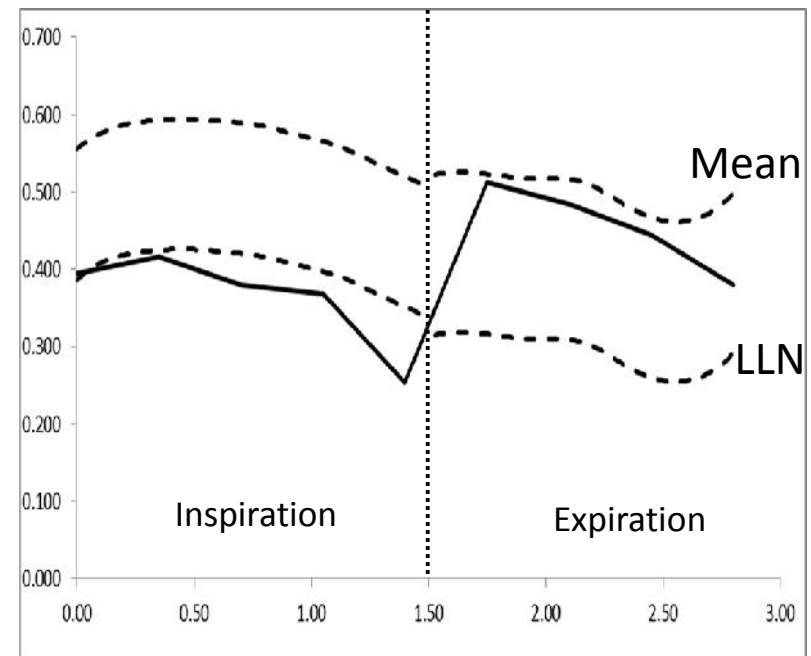
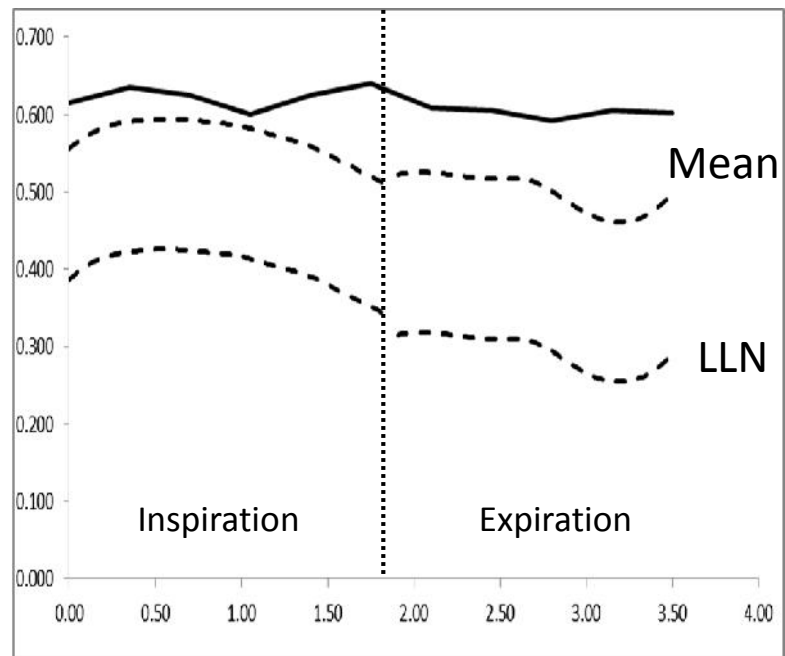
- 19 Fatteh S, Rekkerth DJ, Hadley JA. Skin prick/puncture testing in North America: a call for standards and consistency. *Allergy Asthma Clin Immunol* 2014; 10:44.
- 20 Haahtela T, Burbach GJ, Bachert C *et al.* Clinical relevance is associated with allergen-specific wheal size in skin prick testing. *Clin Exp Allergy* 2014; 44:407–16.
- 21 Thomas M, McKinley RK, Freeman E, Foy C, Price D. The prevalence of dysfunctional breathing in adults in the community with and without asthma. *Prim Care Respir J* 2005; 14:78–82.
- 22 Maillard I, Schweizer V, Broccard A, Duscher A, Liaudet L, Schaller MD. Use of botulinum toxin type A to avoid tracheal intubation or tracheostomy in severe paradoxical vocal cord movement. *Chest* 2000; 118:874–7.
- 23 Baxter M, Uddin N, Raghav S *et al.* Abnormal vocal cord movement treated with botulinum toxin in patients with asthma resistant to optimised management. *Respirology* 2014; 19:531–7.
- 24 Echternach M, Delb W, Verse T, Richter B. Does isolated expiratory vocal cord dysfunction exist? *Otolaryngol Head Neck Surg* 2008; 138:805–6.
- 25 Murakami Y, Kirchner JA. Mechanical and physiological properties of reflex laryngeal closure. *Ann Otol Rhinol Laryngol* 1972; 81:59–71.
- 26 Wood RP II, Milgrom H. Vocal cord dysfunction. *J Allergy Clin Immunol* 1996; 98:481–5.
- 27 Collett PW, Brancatisano AP, Engel LA. Upper airway dimensions and movements in bronchial asthma. *Am Rev Respir Dis* 1986; 133:1143–9.
- 28 Murakami Y, Kirchner JA. Respiratory movements of the vocal cords. An electromyographic study in the cat. *Laryngoscope* 1972; 82:454–67.
- 29 Treole K, Trudeau MD, Forrest LA. Endoscopic and stroboscopic description of adults with paradoxical vocal fold dysfunction. *J Voice* 1999; 13:143–52.
- 30 Bardin PG, Low K, Ruane LE. Vocal cord dysfunction: asking the right questions. *Clin Exp Allergy* 2015; 45:1374–5.
- 31 Ponte EV, Stelmach R, Franco R, Souza-Machado C, Souza-Machado A, Cruz AA. Age is not associated with hospital admission or uncontrolled symptoms of asthma if proper treatment is offered. *Int Arch Allergy Immunol* 2014; 165:61–7.
- 32 Thomas M, McKinley RK, Freeman E, Foy C, Prodger P, Price D. Breathing retraining for dysfunctional breathing in asthma: a randomised controlled trial. *Thorax* 2003; 58:110–5.
- 33 Gardner WN. The pathophysiology of hyperventilation disorders. *Chest* 1996; 109:516–34.
- 34 McFadden ER Jr, Zawadski DK. Vocal cord dysfunction masquerading as exercise-induced asthma. a physiologic cause for “choking” during athletic activities. *Am J Respir Crit Care Med* 1996; 153:942–7.
- 35 Rafferty GF, Evans J, Gardner WN. Control of expiratory time in conscious humans. *J Appl Physiol* 1995; 78:1910–20.
- 36 Jounieaux V, Aubert G, Dury M, Delguste P, Rodenstein DO. Effects of nasal positive-pressure hyperventilation on the glottis in normal sleeping subjects. *J Appl Physiol* 1995; 79:186–93.
- 37 Kuna ST, Insalaco G, Villeponteaux DR, Vanoye CR, Smickley JS. Effect of hypercapnia and hypoxia on arytenoideus muscle activity in normal adult humans. *J Appl Physiol* 1993; 75:1781–9.
- 38 Roy B, Samson N, Moreau-Bussiere F *et al.* Mechanisms of active laryngeal closure during noninvasive intermittent positive pressure ventilation in nonsedated lambs. *J Appl Physiol* 2008; 105:1406–12.

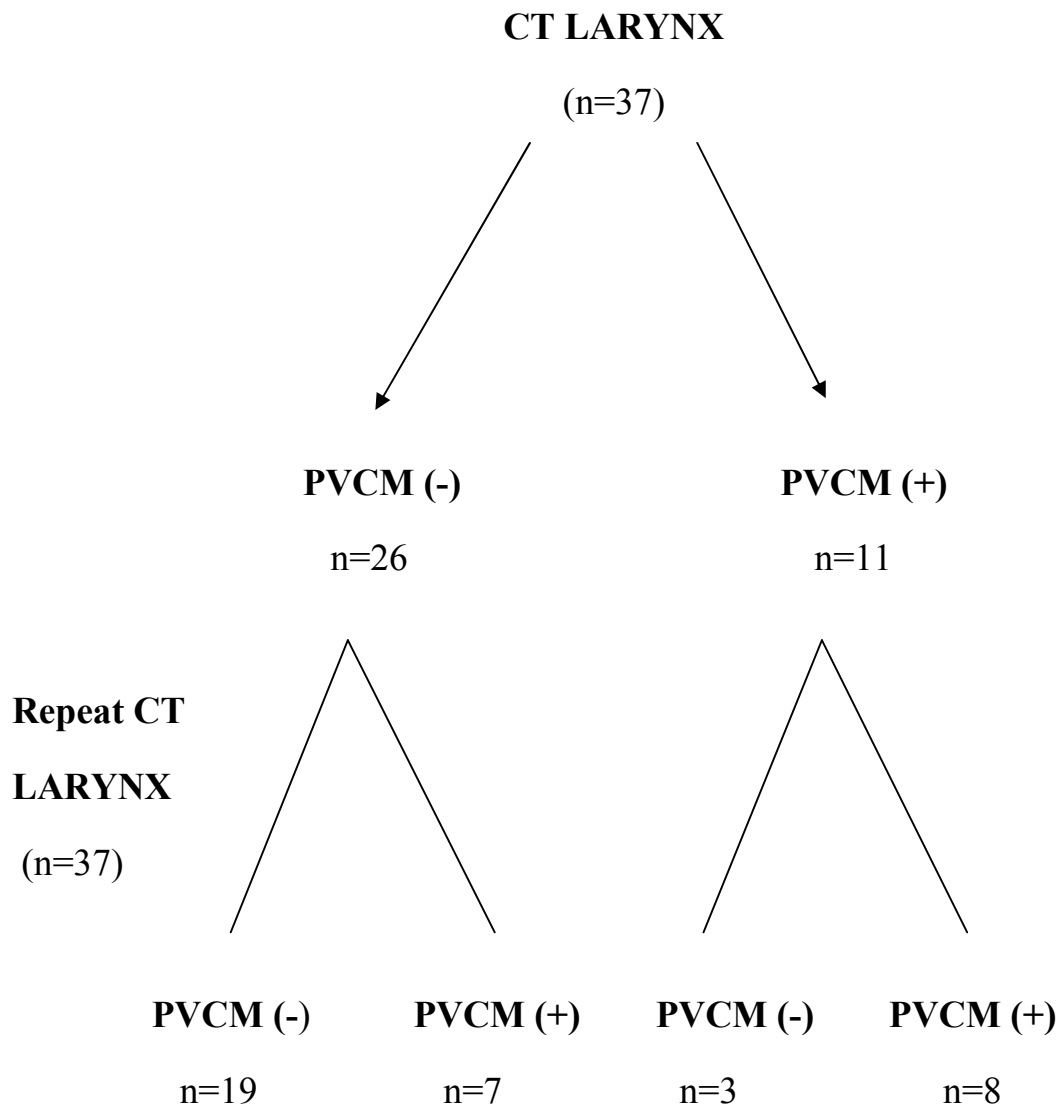
Supporting Information

Additional Supporting Information may be found online in the supporting information tab for this article:

- Figure S1. Demonstration of analyses employed to detect paradoxical vocal cord movement (PVC).
- Figure S2. Diagram illustrating results of repeat CT larynx conducted within 12 months in 37 subjects.
- Figure S3. Dysfunctional breathing (Nijmegen) scores are linked to paradoxical vocal cord movement (PVC).
- Table S1. Characteristics of thirty asthmatic patients who completed a Nijmegen questionnaire.
- Table S2. Detection of PVC in asthmatics with normal spirometry or with airflow limitation.
- Table S3. Characteristics of subjects with PVC detected or not detected by CT larynx.
- Appendix S1. Supporting information and data supplement.

Ratio





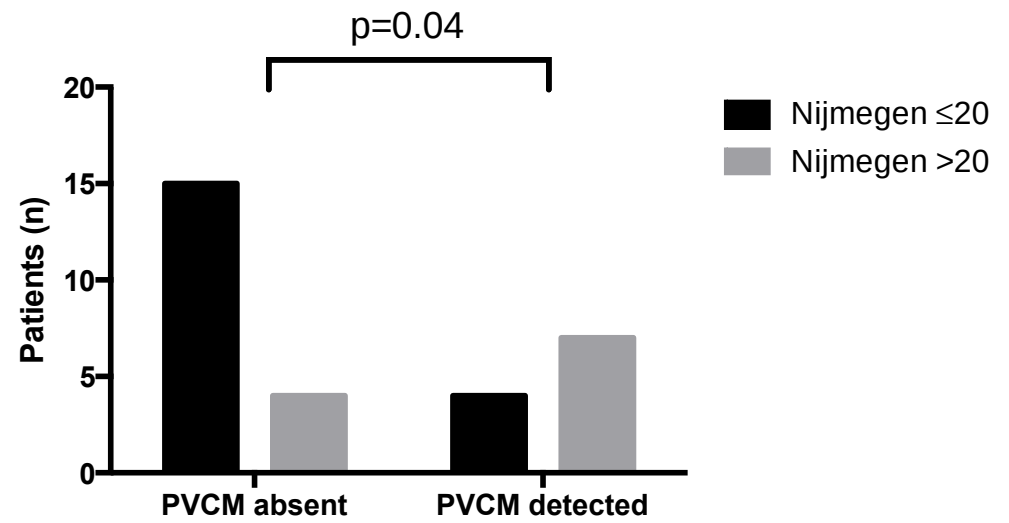
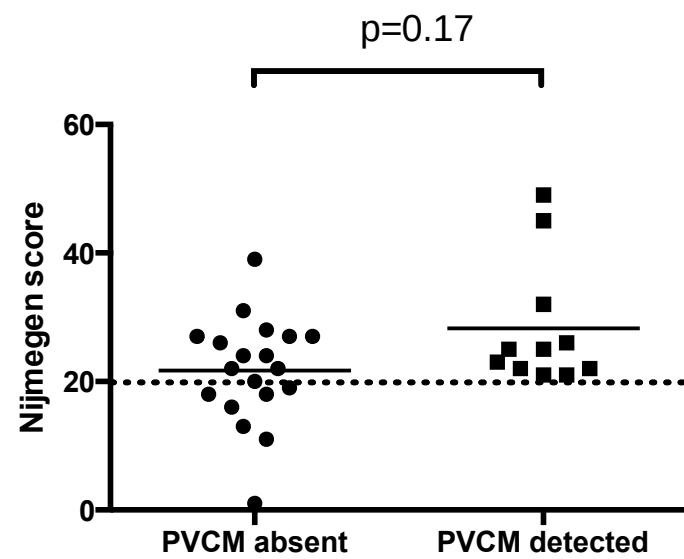


Table E1. Characteristics of thirty asthmatic patients who completed a Nijmegen questionnaire

	All patients (n=30)	PVCM not detected (n=19)	PVCM detected (n= 11)	p-value
Age (m±SD)	55.20±13.2	52.2±14.1	61.7±8.7	0.02
Gender (M/F)	11/19	7/12	4/7	0.9
BMI (kg/m ²)	33.0±8.2	34.3±7.1	31.6±10.0	0.40
FEV ₁ (% predicted)	71.4±25.0	75.7±27.6	63.8±18.3	0.20
FEV ₁ /FVC (%)	66.7±11.1	66.7±11.8	66.8±10.3	0.87
BD response (% change)	14.5±7.9	12.6±8.4	17.9±6.2	0.04
ACT score	15.0±4.9	15.9±4.8	13.7±5.0	0.23
ACQ score	2.1±1.3	1.8±1.3	2.6±1.1	0.06
Nijmegen score	24.3±9.3	21.7±8.4	28.3±9.8	0.27
Medications				
SABA only	10	8	2	
ICS only	2	1	1	
ICS and LABA	18	10	8	
Other [#]	2	1	1	

PVCM, paradoxical vocal cord movement; BMI, body-mass-index; FEV₁, forced expiratory volume–1sec; FVC, forced vital capacity; BD, bronchodilator; ACT, Asthma control test; ACQ asthma control questionnaire; SABA, short-acting beta agonist; ICS, inhaled corticosteroid; LABA, long-acting beta agonist; [#] one nedocromil, one montelukast.

Table E2. Detection of PVCMM in asthmatics with normal spirometry or with airflow limitation

	(FEV ₁ ≥80%)		(FEV ₁ <80%)		p-value
	(n=87)		(n=68)		
	PVCM - (n=70)	PVCM + (n=17)	PVCM - (n=43)	PVCM + (n=25)	
Age (m±SD)	51.73±15.11	56.18±17.53	50.40±17.47	62.60±13.75	0.23
Gender (M/F)	20/50	6 /11	9/34	7/18	0.12
BMI (kg/m ²)	30.15±7.09	29.96±7.08	30.40±7.61	32.52±9.16	0.38
FEV ₁ (% predicted)	100.10±13.76	100.40±14.02	57.67±15.47	60.20±14.87	<0.0001
BD response (% change)	4.72±7.43	1.50±6.24	14.57±12.36	7.76±9.52	<0.0001
T _{LCO}	87.37±11.94	80.25±9.78	77.85±18.45	74.37±13.04	0.001
ACT score (n=91)	16.93±6.02	18.80±6.83	12.71±4.79	13.80±4.29	0.005
ACQ5 score (n=91)	1.42±1.15	1.17±1.20	2.78±1.45	2.71±0.91	<0.0001
Nijmegen score (n=30)	20.70±9.73	23.5±2.12	22.78±7.04	29.33±10.62	0.14

PVCM, paradoxical vocal cord movement; - absent, + present; BMI, body-mass-index; BD, bronchodilator response; T_{LCO}, transfer factor for carbon monoxide; ACT, Asthma control test; ACQ, asthma control questionnaire, ACQ.

Table S3. Characteristics of subjects with PVCMM detected or not detected by CT larynx

	PVCM not detected (n=113)	PVCM detected (n=42)	p-value
Age (m±SD)	51.2±15.9	60.0±15.5	0.003
Gender (M/F)	29/84	13/29	0.54
BMI (kg/m ²)	30.2±7.2	31.4±8.3	0.36
ACT score [*]	15.3±5.9	15.4±5.5 [*]	0.92
ACQ5 score [*]	1.72±1.1	2.1±2.2	0.07
Nijmegen score ^{**}	21.7±8.4	28.3±9.8	0.13
FEV ₁ (% predicted)	83.9±25.2	76.4±24.5	0.09
FEV ₁ /FVC (%)	83.9±14.4	83.0±15.3	0.73
BD response (% change)	8.6±10.7	5.3±8.8	0.08
Medications			
SABA only	31/113	5/42	0.05
ICS only	9/113	1/42	0.44
ICS and LABA	67/113	31/42	0.06
Other [#]	4/113	1/42	0.95

PVCM, paradoxical vocal cord movement; BMI, body-mass-index; ACT, Asthma control test; ACQ asthma control questionnaire; FEV₁, forced expiratory volume–1sec; FVC, forced vital capacity; BD, bronchodilator; ICS, inhaled corticosteroid; LABA, long-acting beta agonist; ^{*}Scores obtained from 65/113 subjects and 26/42 subjects; ^{**}Scores obtained from 19 patients who did not have PVCMM and 11 patients with PVCMM; [#] One nedocromil, two monteleukast, one cromoglycate, one tiotropium

ONLINE SUPPLEMENT

ABNORMAL VOCAL CORD MOVEMENT IN PATIENTS WITH AND WITHOUT AIRWAY OBSTRUCTION AND ASTHMA SYMPTOMS

Kathy Low FRACP*[#], Laurence Ruane BSc*[#], Nizam Uddin FRACP[#], Paul Finlay BSc[#],
Kenneth K Lau FRANZCR[^], Kais Hamza PhD⁺, Peter W Holmes FRACP[#], Garun Hamilton
FRACP[#] and Philip G Bardin FRACP[#]

[#]Monash Lung and Sleep, [^]Diagnostic Imaging and ⁺Statistics, Monash University and
Hospital, and [#]Hudson Institute, Melbourne, Australia

*These authors contributed equally to the study

Funding: Supported by Monash Lung & Sleep Institute and Monash Health

Correspondence:

Prof Phil Bardin

Lung and Sleep

Monash Medical Centre, 246 Clayton Road

CLAYTON, VIC 3168

Melbourne, Australia

Email: Philip.bardin@monash.edu

Methods

Study subjects

Overall 155 patients were recruited in collaboration with their general practitioner either from general practice or from hospital asthma clinics. Our previous studies had shown that abnormal vocal cord movement and PVCMD can be detected in approximately 40% of severe difficult-to-treat asthmatics [1] and the current studies were intended to compare a group with abnormal lung function (likely more severe) to a group with normal spirometry (likely milder asthma). We therefore recruited patients with typical asthma symptoms that conformed to the diagnostic criteria employed for the ISAAC Phase III survey [2] but who additionally had a doctor-diagnosis of asthma. Many mild asthmatics have normal spirometry and no demonstrable bronchodilator responses and asthma can be difficult to confirm on lung function in this group [3]. However, PVCMD and VCD are pertinent diagnostic considerations in these patients and their inclusion in this group permitted clinically relevant comparisons based on lung function measurements. It should be appreciated that study recruitment was not intended to mirror the distribution of asthma severity in the general population (5-20% severe asthma) [3] since the aim was to recruit approximately equal numbers in groups.

Subject characteristics

Asthma was diagnosed in all patients on the basis of: (i) a typical past history of asthma symptoms, (ii) favourable clinical responses to treatment with beta-agonists and/or inhaled corticosteroids and (iii) a doctor diagnosis of asthma. At the time of study all patients were in possession of at least a beta-agonist inhaler to be used for asthma symptoms and patients had no exacerbations (oral corticosteroids, emergency hospital visits) for at least 4 weeks. Patients with an asthma diagnosis based on a typical clinical history obtained from their general practitioner (n=57) but who did not meet spirometry benchmarks for asthma were therefore enrolled. Bronchial provocation testing is often very helpful in asthma diagnosis but was not done since tests have limited specificity in very mild asthmatics with normal FEV₁ [4, 5]. Patients were excluded if they had any other lung disease, smoking history >10 pack-years, any known vocal cord pathology, neurological disease or if pregnant. Detailed information about gastro-esophageal reflux disease (GORD) symptoms or secondary smoking was not obtained.

Study groups

Baseline demographic characteristics, relevant asthma history, history of comorbidities and asthma medications were obtained. Overall 68/155 patients had asthma symptoms with FEV₁<80% predicted whereas 87/155 patients had asthma symptoms with FEV₁≥80% predicted. For secondary comparisons asthma severity was assessed according to ATS/ERS

[3]. Asthma severity plus control were also evaluated using asthma control test (ACT) and asthma control questionnaire-5 (ACQ5) scores [8, 9].

320-slice CT larynx

No treatment for asthma was taken on the day of study. Studies were performed using a Toshiba Aquilion One CT (Toshiba Medical Systems, Tokyo, Japan) with 320-slice detector. Parameters were 80kVp, 300-350mA and gantry rotation 0.35 seconds. Patients were asked to breathe normally and to avoid swallowing. After a 'practice run' without radiation to ensure steady, normal breathing dynamic images were obtained continuously over a respiratory cycle, from beginning of inspiration to the beginning of inspiration of the next breath. Radiation doses were in a range of approximately 0.8-2 milliSievert (mSv) depending on the amount of soft tissue in the neck of a particular individual [1]. Integrated CT software programs were used to obtain continuous dynamic axial, sagittal and coronal multi-planar images of the larynx with simultaneous virtual endoscopy reconstructions. Images were reconstructed at 0.35 second/frame and included dynamic 3-dimensional laryngeal airway views.

Diagnosis of VCD

Lateral diameter at the level of the vocal cords and first tracheal ring was measured on coronal views using electronic callipers by a radiologist blinded to the study groups. The ratio of vocal cord lateral diameter to tracheal diameter (RATIO) was obtained and an in-house computerised program (HAMZACT) was then employed to automatically construct a 'curve' based on RATIO generated every 0.35 seconds. A predicted mean normal curve with a second curve depicting the lower limit of normal (LLN) had previously been generated in studies conducted in healthy individuals [1]. For an abnormal curve to occur the patient curve has to be below the LLN curve (see for example Figure E1, online supplement). VCD was only diagnosed during inspiration since criteria for diagnosis of expiratory VCD are unclear [10-12]. Severity of VCD was assessed by measurement of the area of the patient's RATIO <LLN curve and expressed as % of overall area <LLN. Endoscopy was not used to diagnose VCD and to attempt quantification since there is good concordance between orthodox endoscopy and CT virtual endoscopy [1] and because the former cannot be accurately quantified.

Lung function testing

Lung function testing was performed (MasterScreen Diffusion; Jaeger, Würzburg, Germany) on the same day immediately after CT larynx was completed. Testing comprised pre- and post-bronchodilator spirometry and measurements of transfer factor for carbon monoxide

(TLco). All testing was carried out according to American Thoracic Society guidelines [13] using predictive equations published by Knudson et al [14].

Skin prick testing

Skin prick testing was done using a standard panel of allergens and according to established guidelines [15, 16]. Allergens used were: cat, dog, horse, *Alternaria*, *Cladosporium*, *Aspergillus*, grass mix, house dust mite, and tree mix in combination with appropriate negative and positive controls.

Discussion

Patient selection

The patient population under study was chosen to answer a key question raised in our initial studies: are abnormal vocal cord movement, PVCMM and VCD chiefly associated with severe asthma defined by airflow limitation or is it a widespread phenomenon in all individuals who experience typical asthma symptoms? To meet this objective patients were recruited from general practice and as a result the majority of patients in this group (57/87) did not achieve spirometry criteria for a diagnosis of asthma. However, recruitment strategies were similar to the ISAAC Phase III survey to detect asthma (in which spirometry was not conducted) and our methodology ensured that individuals with asthma symptoms could be recruited.

It can be argued that individuals without evidence on spirometry did not have asthma and that as a consequence the frequency of PVCMM will logically be lower. However, the percentage of individuals with PVCMM in this group was identical to the group who had 12% or larger bronchodilator responses. On the other hand, there is a widespread belief that patients with PVCMM have ‘fictitious’ asthma [11] implying that the occurrence of PVCMM would be increased in this particular group with asthma symptoms but no objective support for a diagnosis of asthma. This was not observed and to resolve this complex issue further studies are needed to compare larger groups of mild asthmatics (who meet spirometry criteria for the diagnosis) with a symptomatic-only group as well as with patients who have severe asthma. The current studies should be viewed as a pragmatic approach that permitted a comparison of patients with asthma symptoms attributable to VCD.

Spirometry and lung function

Spirometry can indicate the presence of tracheal stenosis and other upper/middle airway conditions characterised by fixed obstruction [17]. No evidence of PVCMM manifesting as flattening of the flow-volume curves was detectable on spirometry and this accords with other studies in patients with verified PVCMM and VCD [1, 18, 19]. It may reflect poor sensitivity of a forced manoeuvre during inspiration (and expiration) to detect dynamic and variable vocal cord closure. Other techniques, such as impulse oscillometry (IOS), may be more sensitive indicators of laryngeal obstruction [17] and merit investigation.

Figure legends for online supplement

- Figure S1. Demonstration of analyses employed to detect paradoxical vocal cord movement (PVCM). X-axis depicts time during inspiration and expiration and Y-axis shows RATIO (ratio of vocal cord lateral diameter/tracheal diameter). Left panel shows normal vocal cord function with RATIO (solid black line) above both mean and lower limit of normal (LLN). Right panel demonstrates excessive vocal cord closure in a patient with inspiratory PVCM. Area under LLN is approximately 8% (calculated as % of overall area under LLN).
- Figure S2. Diagram illustrating results of repeat CT larynx conducted within 12 months in 37 subjects. Agreement between the first and second CT was noted in 19/26 patients (73%) in whom PVCM was not detected and in 8/11 (72%) cases in which PVCM was present. Overall there was moderate agreement between the two CTs with Cohen's $\kappa=0.42$.
- Figure S3. Dysfunctional breathing (Nijmegen) scores are linked to paradoxical vocal cord movement (PVCM). Left panel shows overall scores in groups with PVCM absent or detected. Right panel depicts comparison of the same groups but comparing Nijmegen scores using a threshold score of 20 (n=11 had PVCM). Seven of eleven subjects (7/11) in whom PVCM could be detected had scores >20 versus 4/11 in patients with scores ≤ 20 . If PVCM was absent 4/19 patients had a score >20 and 15/19 had a score ≤ 20 (p=0.04).

References

1. Low K, Lau KK, Holmes P, Crossett M, Vallance N, Phyland D, Hamza K, Hamilton G, Bardin PG. Abnormal vocal cord function in difficult-to-treat asthma. *Am J Respir Crit Care Med* 2011; 184(1): 50-56.
2. Ellwood P, Asher MI, Beasley R, Clayton TO, Stewart AW, Committee IS. The international study of asthma and allergies in childhood (ISAAC): phase three rationale and methods. *The international journal of tuberculosis and lung disease : the official journal of the International Union against Tuberculosis and Lung Disease* 2005; 9(1): 10-16.
3. Chung KF, Wenzel SE, Brozek JL, Bush A, Castro M, Sterk PJ, Adcock IM, Bateman ED, Bel EH, Bleecker ER, Boulet LP, Brightling C, Chanez P, Dahlen SE, Djukanovic R, Frey U, Gaga M, Gibson P, Hamid Q, Jajour NN, Mauad T, Sorkness RL, Teague WG. International ERS/ATS guidelines on definition, evaluation and treatment of severe asthma. *The European respiratory journal* 2014; 43(2): 343-373.
4. Cockcroft D, Davis B. Direct and indirect challenges in the clinical assessment of asthma. *Annals of allergy, asthma & immunology : official publication of the American College of Allergy, Asthma, & Immunology* 2009; 103(5): 363-369; quiz 369-372, 400.
5. Joos GF. Bronchial hyperresponsiveness: too complex to be useful? *Current opinion in pharmacology* 2003; 3(3): 233-238.
6. Reddel HK, Taylor DR, Bateman ED, Boulet LP, Boushey HA, Busse WW, Casale TB, Chanez P, Enright PL, Gibson PG, de Jongste JC, Kerstjens HA, Lazarus SC, Levy ML, O'Byrne PM, Partridge MR, Pavord ID, Sears MR, Sterk PJ, Stoloff SW, Sullivan SD, Szeffler SJ, Thomas MD, Wenzel SE. An official American Thoracic Society/European Respiratory Society statement: asthma control and exacerbations: standardizing endpoints for clinical asthma trials and clinical practice. *Am J Respir Crit Care Med* 2009; 180(1): 59-99.
7. Wenzel SE. Asthma: defining of the persistent adult phenotypes. *Lancet* 2006; 368(9537): 804-813.
8. Juniper EF, O'Byrne PM, Guyatt GH, Ferrie PJ, King DR. Development and validation of a questionnaire to measure asthma control. *The European respiratory journal* 1999; 14(4): 902-907.
9. Schatz M, Sorkness CA, Li JT, Marcus P, Murray JJ, Nathan RA, Kosinski M, Pendergraft TB, Jhingran P. Asthma Control Test: reliability, validity, and responsiveness in patients not previously followed by asthma specialists. *The Journal of allergy and clinical immunology* 2006; 117(3): 549-556.
10. Brugman SM, Simons SM. Vocal cord dysfunction: don't mistake it for asthma. *Phys Sportsmed* 1998; 26(5): 63-85.
11. Christopher KL, Wood RP, 2nd, Eckert RC, Blager FB, Raney RA, Souhrada JF. Vocal-cord dysfunction presenting as asthma. *N Engl J Med* 1983; 308(26): 1566-1570.
12. Echtertnach M, Delb W, Verse T, Richter B. Does isolated expiratory vocal cord dysfunction exist? *Otolaryngol Head Neck Surg* 2008; 138(6): 805-806.
13. Lung function testing: selection of reference values and interpretative strategies. American Thoracic Society. *Am Rev Respir Dis* 1991; 144(5): 1202-1218.
14. Knudson RJ, Slatin RC, Lebowitz MD, Burrows B. The maximal expiratory flow-volume curve. Normal standards, variability, and effects of age. *Am Rev Respir Dis* 1976; 113(5): 587-600.
15. Fatteh S, Rekkerth DJ, Hadley JA. Skin prick/puncture testing in North America: a call for standards and consistency. *Allergy, asthma, and clinical immunology : official journal of the Canadian Society of Allergy and Clinical Immunology* 2014; 10(1): 44.

16. Haahtela T, Burbach GJ, Bachert C, Bindslev-Jensen C, Bonini S, Bousquet J, Bousquet-Rouanet L, Bousquet PJ, Bresciani M, Bruno A, Canonica GW, Darsow U, Demoly P, Durham SR, Fokkens WJ, Giavi S, Gjomarkaj M, Gramiccioni C, Kowalski ML, Losonczy G, Orosz M, Papadopoulos NG, Stingl G, Todo-Bom A, von Mutius E, Kohli A, Wohrl S, Jarvenpaa S, Kautiainen H, Petman L, Selroos O, Zuberbier T, Heinzerling LM. Clinical relevance is associated with allergen-specific wheal size in skin prick testing. *Clinical and experimental allergy : journal of the British Society for Allergy and Clinical Immunology* 2014; 44(3): 407-416.
17. Verbanck S, de Keukeleire T, Schuermans D, Meysman M, Vincken W, Thompson B. Detecting upper airway obstruction in patients with tracheal stenosis. *Journal of applied physiology* 2010; 109(1): 47-52.
18. Ibrahim WH, Gheriani HA, Almohamed AA, Raza T. Paradoxical vocal cord motion disorder: past, present and future. *Postgrad Med J* 2007; 83(977): 164-172.
19. Newman KB, Mason UG, 3rd, Schmaling KB. Clinical features of vocal cord dysfunction. *Am J Respir Crit Care Med* 1995; 152(4 Pt 1): 1382-1386.