With respect to the dissociation between the risk of deep venous thrombosis and pulmonary embolism, we very much agree with D.P. Potaczek and A. Undas that this suggests that the activation of haemostasis in our population is not due to prolonged immobilisation and hospitalisations, but is the result of inflammation in the airways *per se*. Of course, our findings have to be replicated in larger cohorts and more studies are needed to disentangle the underlying mechanisms, in particular with respect to asthma severity, type of airway inflammation and the use of corticosteroids.

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***Post hoc* analysis in subgroups, stratified for atopic and non-atopic asthma, reveals no differences in risk of VTE** http://ow.ly/mK7BW

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Is the sensitivity of primary ciliary dyskinesia detection by ciliary function analysis 100%?

To the Editor:

We read with interest the article by Pifferi *et al.* [1] in the April issue of the *European Respiratory Journal*, describing a technique of soft computing analysis to increase the diagnostic accuracy of air liquid interface cultures for the diagnosis of primary ciliary dyskinesia (PCD).

The diagnosis of PCD is indeed difficult, time-consuming and expensive [2, 3]. Extensive efforts have been made to increase the diagnostic accuracy of the available tests.

We worry that the authors miscalculated the statistical parameters sensitivity and specificity. Sensitivity reflects the accuracy of a new test to detect an abnormal result in a disease state (true positives), compared to the results of the gold standard to diagnose the disease state [4]. It is calculated by the formula a/a+b, in which a is the number of true positives and b the number of false positives (table 1). Specificity, on the other hand, reflects the accuracy of the new test to diagnose a normal value, compared to the gold standard (true negatives) [4]; it is calculated by the formula d/c+d, in which d is the number of true negatives and c the number of false negatives (table 1). In this study, the gold standard for diagnosis of PCD is defined as

TABLE 1 Definition of sensitivity and specificity for a new diagnostic test, compared to the gold standard

	Gold standard	
	Disease	**Health**
New test		
Abnormal test	a (true positive)	c (false negative)
Normal test	b (false positive)	d (true negative)

absence of coordinated ciliary motility after ciliogenesis, and thus the newly evaluated parameters should be presented as absence of rotation of spheroids, absence of migration, *etc.* For the parameter absence of rotation of the spheroids, a=35, b=1, c=17, d=64; sensitivity amounts to 35/35+1=97.2% and specificity 64/64+17=79.0%, rather than the reverse. Sensitivity and specificity, similarly, PPV and NPV have to be exchanged for all the parameters in table 2 of the mentioned article.

With this new technique of soft computing analysis, the authors thus reach a high sensitivity (97–100%) and a poor to acceptable specificity (7–79%) for the diagnosis of PCD.

Generally, the main reason to perform cell culture is to exclude a false diagnosis of PCD due to secondary changes and thus to increase the specificity [5]. However, it is proven in the mentioned paper that ciliary motility before ciliogenesis was completely normal in one PCD patient. Therefore, the diagnosis would have been missed without cell culture. This means that cell culture can increase sensitivity as well as specificity. This finding is especially important for patients with PCD and normal ultrastructure, because in these cases transmission electron microscopy (TEM) has no added value.

Since 1990, we have systematically performed ciliogenesis in all biopsy samples with a sequential monolayer-suspension culture, as described previously [6]. The success rate of the culture was 75%, which is higher than the aforementioned 50% of the air liquid interface culture technique [1]. Based on the absence of ciliary coordination (with or without absence of ciliary motility) after ciliogenesis in culture, we detected 206 patients with PCD. However, initial evaluation of the ciliary coordination and ciliary beat frequency in the biopsy was normal in 21 persons (10.2%) with a final diagnosis of PCD. 10 of them had specific abnormalities on TEM (three outer dynein arm deficiency, four absence of the central pair, two eccentric central pair and inner dynein arm deficiency, and one excessive absence of a peripheral microtubular pair), which confirmed PCD. Normal ultrastructure was seen in 11 patients. These patients all had typical clinical signs and symptoms of PCD (upper and lower respiratory tract infections, neonatal respiratory symptoms, ear problems and random situs) and the diagnosis was confirmed in a repeat biopsy with ciliogenesis in nine out of 11 patients, which makes abnormalities due to the culture technique or due to misinterpretation unlikely. Moreover, biallelic mutations in DNAH11 were detected in four out of nine examined patients with normal ultrastructure, which is even more than the reported 22% of detected DNAH11 mutations in a PCD population with normal ultrastructure in the literature [7]. Nasal nitric oxide values were abnormal in these 11 patients, the median value was 62 ppb (range 11–337 ppb). In this way, the diagnosis of PCD in these patients is robust, although it would have been missed when only evaluation of ciliary motility was performed on the biopsy. In conclusion, we would like to state that the sensitivity of the evaluation of ciliary motility in the biopsy to detect PCD in our cohort was only 89% (and only 84% for those with normal ultrastructure) and this is not acceptable for a rare disease. It suggests that evaluation of ciliary motility in the biopsy specimen only can lead to a considerable amount of missed PCD diagnoses.

Although we have made extensive efforts to publish the data on our large cohort of PCD patients with normal ultrastructure, we have not succeeded yet. The paper of Pifferi *et al.* [1] gives us the ideal opportunity to emphasise that ciliogenesis in culture remains the best technique (concerning specificity and sensitivity) to make the diagnosis of PCD with normal ultrastructure.

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Is the detection of primary ciliary dyskinesia via ciliary function analysis really 100% sensitive?
http://ow.ly/neGBi

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From the authors:

We wish to thank M. Boon and co-workers for their careful reading of our manuscript and to apologise to them and the readers for our mistake in the preparation of table 2 of our original article [1]. We erroneously reversed the sensitivity and specificity values, as well as the negative predictive value and positive predictive value for the first four parameters (rotation of spheroids, migration of spheroids, ability of cilia to remove debris, and normal ciliary beat pattern). The correct version of table 2 is given below. The correct version improves the true positive rate of ciliogenesis in culture for the identification of the percentage of sick people who are correctly identified as having primary ciliary dyskinesia (PCD). We want to make clear that the subject with PCD associated with rotation of the spheroids (on the 10th day of observation) similar to that described in secondary ciliary dyskinesia, did show a nonflexible and hyperkinetic ciliary motion pattern. We agree with the diagnostic approach of M. Boon and co-workers, and would be very happy to share our artificial neural network-based model with them, and anyone else, attempting to make the diagnosis of PCD.

TABLE 2 Sensitivity, specificity, positive predictive values (PPVs), and negative predictive values (NPVs) of the different parameters used for ciliary activity evaluation in suspension cell culture for the diagnosis of primary ciliary dyskinesia

Suspension cell cultures parameters	Sensitivity %	Specificity %	PPV %	NPV %
Rotation of the spheroids	97.2	79.0	67.3	98.5
Migration of the spheroids	100	17.3	34.9	100
Ability of cilia to remove debris	100	7.4	32.4	100
Normal ciliary beat pattern	100	64.2	55.4	100
Pathological ciliary beat pattern	33.3	100	100	77.1

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The detection of primary ciliary dyskinesia via ciliary function analysis has sensitivity close to 100%
http://ow.ly/nf5C4

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