Smoking resumption after lung transplantation: need for standardized screening and importance for long-term outcome.

D. Ruttens\textsuperscript{1}, S.E. Verleden\textsuperscript{1}, P.C. Goeminne\textsuperscript{1}, K. Poels\textsuperscript{2}, E. Vandermeulen\textsuperscript{1}, L. Godderis, D.E. Van Raemdonck\textsuperscript{1}, B.M. Vanaudenaerde\textsuperscript{1}, J. Vanoirbeek\textsuperscript{2}, R. Vos\textsuperscript{1}, G.M. Verleden\textsuperscript{1}.

\textsuperscript{1}Lung Transplant Unit, Lab of Pneumology, KU Leuven, University Hospital Gasthuisberg Leuven, Belgium

\textsuperscript{2}Laboratory for Occupational and Environmental Hygiene, Center for Environment and Health, KU Leuven, Belgium

Running title: smoking after LTx

Key words: smoking, relapse, lung transplantation, cancer, screening, cotinine.

Address for correspondence: Prof. dr. Geert M. Verleden
University Hospital Gasthuisberg and KU Leuven
Department of Respiratory Medicine
Lab of Pneumology and Lung Transplantation Unit
49 Herestraat, B-3000 Leuven, Belgium
Tel: + 32 16 346805  Fax: + 32 16 246803
E-mail: geert.verleden@uzleuven.be

Copyright 2013 by the European Respiratory Society.
Abbreviations:

CF: cystic fibrosis
COPD: chronic obstructive pulmonary disease
COT: urinary cotinine
eCO: exhaled carbon monoxide
HL: heart-lung transplantation
ILD: interstitial lung disease
LTx: lung transplantation
PAHT: pulmonary arterial hypertension
PY: pack-years
PR: prevalence risk
SLTx: single lung transplantation
SSLTx: sequential single lung transplantation
To the editors:

Worldwide, about 40% of the lung transplantations (LTx) are performed for end-stage emphysema (1). Eligible patients are enrolled on the waiting list after smoking cessation for at least 6 months (1). Although in most centers smoking behavior after LTx is not routinely monitored, resuming smoking can complicate post-transplant outcome (2-4;6).

In general, smoking relapse can be found in 12-40% of all liver, heart and renal transplant patients (3). Smoking is mostly assessed by use of a questionnaire, only the study of Botha et al (4) combined a questionnaire with urinary cotinine detection (COT). We previously reported post-LTx smoking in 11% of our LTx patients (5). Patients that regain smoking after heart (4) and liver transplantation (6) have an increased prevalence of cancer, yet, there are no data for LTx.

We assessed all living, mainly adult (98%), LTx patients (n=331, of whom 230 were also included in our previous study (7)) with a minimal follow up of 1 year, after approval by the local Ethics Committee (S51577) and informed consent. Smoking behavior was investigated by questionnaire, semi-quantitative and quantitative measurement of COT and exhaled CO (eCO) levels. The questionnaire addressed past and current smoking habits. Second-hand smoking was defined as an in-house relative who smoked. The eCO was quantified by using an electrochemical sensor (Bedfont Scientific, Kent, UK; detection limit 1 ppm), as previously described (6), a value of 10 ppm or more was considered positive.

Quantitative Cotinine analysis was performed using ultra-pressure liquid chromatography, in combination with tandem mass spectrometry, using d3-cotinine as an internal standard. Values of >7.5ng/ml were detected while a value of >75 ng/ml was considered as positive.

Semi-quantitative Cotinine measurement was assessed by gas chromatography and mass spectrometry (Thermo Scientific, Geel, Belgium) as previously described (6).
Graph prism 4.0 software (San Diego, Cal, USA) was used for statistical analysis. Mann-Whitney or chi-square tests were used for analysis of patient characteristics, where appropriate. For multi-variate analysis, stepwise regression was used (SAS 9.3; SAS Institute Inc, Cary, NC), including all clinically relevant and univariate significant variables.

Based on the questionnaire, 33/331 (10%) patients reported ever having smoked after LTx, whereas the remaining 298 patients denied smoking. Twenty-four patients concurrently had positive COT levels and 6/33 patients admitted having stopped smoking post-transplant, which was confirmed with a negative COT detection. No urine could be collected in 3/33 post-LTx smokers due to anuric renal insufficiency. In the non-smoking group (questionnaire), 6 patients had COT levels above the smoking limit and were therefore considered as active smokers. Taken together, 39/331 patients (12%) resumed smoking after LTx, of whom 33 were current smokers (10%) and 6 past smokers (2%). Post-LTx smokers resumed smoking at a median of 1.5 (0.8-3.0) year after LTx and smoked a mean of 3 (3-6) cigarettes a day. Since 33/39 post-LTx smokers admitted smoking relapse by means of the questionnaire, its sensitivity (85%) and specificity (100%) can be considered as “good”.

Quantitative determination of urinary cotinine was available in 318 patients (13 missing data: anuric renal failure (n=7), inadequate urine sample (n=6)). Cotinine values were under the detection limit in 262 patients. Cotinine levels were moderately elevated in 26 patients, with values above the detection limit (7.5ng/mL) and below smoking limit (75ng/mL); and 30 patients (10%) had strongly elevated cotinine level compatible with active smoking (> 75ng/ml). The latter 30 patients also had a positive semi-quantitative COT detection although there were 6 positive semi-quantitative COT results with a negative questionnaire, resulting in
100% sensitivity and 98% specificity of COT in our population. Median eCO levels were higher in post-LTx smokers compared to non-smokers: 8(3-13) versus 3(2-5)ppm (p<0.0001). Smoking resumption was the highest in the emphysema group (36/167 patients, 22%), while this was overall very low in non-emphysema patients (p<0.0001) (table 1). The abstinence period before LTx was shorter in patients who relapsed after LTx: 1.0 (1.0-3.0) versus 6.0 (2.0-10.5) years, respectively (p<0.0001). Smoking relapse post-LTx was not related to the pack-years (PY) pre-LTx (mean 28 PY in the post-LTx non-smoking group compared to 31 PY in the smoking group)(p=0.26).

Second-hand smoking was higher in post-LTx smokers compared with non-smokers (p<0.0001) (table 1). The exposure to second-hand smoking and a shorter period of pre-LTx smoking abstinence were independent risk factors for smoking relapse after LTx in multivariate analysis (p< 0.0001, prevalence risk (PR)=5.42 and p=0.0008, PR=1.18).

Oncological events were detected in 36 patients (10%) (13 hematological/10 lung/6 digestive tract/3 urogenital/2 brain/2 others), of whom 23 patients (7%) developed a solid tissue cancer. The prevalence of oncological events was higher in post-LTx smokers compared to non-smokers (p=0.0043) (PR=2.88). Especially, solid organ cancer was more frequently diagnosed in post-LTx smokers compared to non-smokers (p=0.0024) (PR=3.99). Particularly, lung cancer was more prevalent in post-LTx smokers compared to non-smokers (p=0.021) (PR=4.99). Time of diagnosis of the lung cancers was comparable in both groups (p=0.82) (table 1). Neither the number of pack-years (p=0.43) nor the percentage of underlying COPD patients before LTx (p=0.38) were different between the group with or without an oncological event.

The current study demonstrated a high prevalence of post-LTx smoking, as 12% of our LTx population and even 22% of the emphysema patients resumed smoking post-LTx, confirming
our previous work (6). Peer smoking is an important risk factor for smoking resumption, therefore, patients’ relatives, who most often continue smoking, should also be recommended to stop smoking. Our study also showed that shorter cessation time before LTx was associated with smoking relapse after LTx. This could imply that an abstinence period of 6 months (7) may actually be too short. However, a longer abstinence period, would be difficult to achieve in patients with for instance idiopathic pulmonary fibrosis, due to its worse prognosis.

Importantly we demonstrated an increased prevalence of oncological events in patients who resume smoking post-LTx. The oncological effects of smoking are well known, and are even strengthened by post-LTx immunosuppressive therapy (8). Patients should therefore be actively counseled concerning the possible negative health-effects of smoking resumption.

Although a questionnaire is subjective, it seemed to have a good sensitivity/specificity to detect smoking and correlated well with the urinary COT test, a more objective assessment of a patient's nicotine exposure. Besides being sensitive (100%), semi-quantitative COT determination is also very specific (98%). Because quantitative COT analyses are not available in clinical practice, we would recommend the semi-quantitative COT detection, at least several times while on the waiting list and at least once yearly after LTx or whenever smoking resumption is suspected. The results of the eCO measurements, however, showed a rather low sensitivity (48%), which is probably due to its short half-life, therefore, one should question the place of eCO determination in screening.

In conclusion, the prevalence of post-LTx smoking is higher than generally assumed and active screening, both pre- and post-LTx, is crucial to detect smoking resumption. A standardized questionnaire and repeated COT testing is probably the best screening method to detect post-LTx smoking. Implementing a standardized smoking cessation plan after LTx, as in COPD (9) and a longer pre-LTx cessation period (especially in COPD patients) should be
considered, not only for patients, but also for smoking relatives. Finally, for the first time a
clear association between smoking resumption after LTx and an increased prevalence of
oncological events was demonstrated.
**Funding:** Funding of the research group includes the Glaxo Smith Kline (Belgium) chair in respiratory pharmacology at the KU Leuven; grants from the Research Foundation Flanders (FWO): G.0723.10, G.0679.12 and G.0705.12; and a grant from the Katholieke Universiteit Leuven: OT10/050.

The authors of this manuscript have no conflicts of interest to disclose as described by the European Respiratory Journal
### Table 1

<table>
<thead>
<tr>
<th>Patient characteristics</th>
<th>Post-LTx Non-smokers</th>
<th>Post-LTx Smokers</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total patients</td>
<td>292</td>
<td>39</td>
<td>p=0.17</td>
</tr>
<tr>
<td>Type of LTx</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SSLTx</td>
<td>232 (79%)</td>
<td>28 (72%)</td>
<td>p=0.27</td>
</tr>
<tr>
<td>SLTX</td>
<td>43 (15%)</td>
<td>10 (26%)</td>
<td>p=0.08</td>
</tr>
<tr>
<td>HL</td>
<td>17 (6%)</td>
<td>1 (2%)</td>
<td>p=0.40</td>
</tr>
<tr>
<td>Underlying disease</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Emphysema</td>
<td>131 (45%)</td>
<td>36 (91%)</td>
<td>p&lt;0.0001*</td>
</tr>
<tr>
<td>Cystic fibrosis</td>
<td>66 (22%)</td>
<td>1 (3%)</td>
<td>p=0.0034*</td>
</tr>
<tr>
<td>ILD</td>
<td>44 (15%)</td>
<td>0 (0%)</td>
<td>p=0.0092*</td>
</tr>
<tr>
<td>PAHT</td>
<td>28 (10%)</td>
<td>1 (3%)</td>
<td>p=0.15</td>
</tr>
<tr>
<td>Others</td>
<td>23 (8%)</td>
<td>1 (3%)</td>
<td>p=0.23</td>
</tr>
<tr>
<td>Age</td>
<td>52 (37-58)</td>
<td>54 (49-57)</td>
<td>p=0.062</td>
</tr>
<tr>
<td>Male sex</td>
<td>144 (49%)</td>
<td>20 (51%)</td>
<td>p=0.82</td>
</tr>
<tr>
<td>Smokers before LTx</td>
<td>178 (61%)</td>
<td>39 (100%)</td>
<td>p&lt;0.0001*</td>
</tr>
<tr>
<td>Second-hand smoking</td>
<td>92 (28%)</td>
<td>33 (75%)</td>
<td>p&lt;0.0001*</td>
</tr>
<tr>
<td>Oncologic events</td>
<td>26 (9%)</td>
<td>10 (26%)</td>
<td>p=0.0043*</td>
</tr>
<tr>
<td>Solid tissue cancer</td>
<td>15 (5%)</td>
<td>8 (20%)</td>
<td>p=0.0024*</td>
</tr>
<tr>
<td>Time to diagnose (days)</td>
<td>1460 (432-2023)</td>
<td>1900 (575-2555)</td>
<td>p=0.16</td>
</tr>
<tr>
<td>Lung cancer</td>
<td>6 (2%)</td>
<td>4 (10%)</td>
<td>p=0.021*</td>
</tr>
<tr>
<td>Time to diagnose (days)</td>
<td>2048 (1185-2516)</td>
<td>2448 (1383-2851)</td>
<td>p=0.61</td>
</tr>
</tbody>
</table>

Table 1: Patient demographics of the living patients (n=331). Results are expressed in numbers (n) with percentage or median with interquartile ranges. HL= heart-lung transplantation, ILD= interstitial lung disease, LTx= lung transplantation, PAHT= pulmonary arterial hypertension, SLTx= single lung transplantation, SSLTx= sequential single lung transplantation. P <0.05 was considered significant.


