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LUNG TRANSPLANTATION

Helicobacter pylori Infection Does Not Impact on Lung Transplant Outcome

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Abstract

Background *Helicobacter pylori* (HP) is a spiral, gram-negative, microaerophilic bacterium that colonises the human gastric mucosa and is associated with gastrointestinal and extragastrointestinal disorders. Since no data are yet available on HP infection in lung transplant patients, we evaluated the prevalence and impact of HP infection in a population of such patients.
Methods Sixty-seven lung transplant patients were enrolled in the study (35 females and 32 males, age 48.4 ± 13.3 years), 54 underwent bilateral and 13 single lung transplant. Serum antibodies against HP and CagA were assayed in all subjects.
Results The prevalence of HP infection in lung transplant patients was similar to that in the general population (49.25% vs. 51.4%), whereas HP-positive patients showed lower CagA positivity (9% vs. 50.2%, $p < 0.0001$). There was a higher prevalence of HP infection in patients who underwent lung transplant because of pulmonary fibrosis ($p = 0.049$), and a lower prevalence in COPD patients ($p = 0.011$). No correlation was found between HP infection in lung transplant patients and graft outcome. No differences in primary graft dysfunction, acute rejection or bronchiolitis obliterans syndrome-free survival were found. However, more patients who required three or more post-transplant re-hospitalisations were observed among HP-positive patients.
Conclusions The prevalence of HP infection in lung transplant patients was comparable to that of the general population and to that reported in heart and kidney transplant recipients. It did not seem to impact short-, mid- or long-term lung allograft outcome. *H. pylori* infection did not prove to be clinically relevant in lung transplant patients.

Keywords *Helicobacter pylori* · Lung transplantation · Epidemiology

Introduction

Helicobacter pylori (HP) is a spiral, gram-negative, microaerophilic bacterium that colonises the human gastric mucosa [1]. The prevalence of HP infection, which has ubiquitous distribution, varies with age, socioeconomic status and country. It is estimated that about 50% of the world human population are carriers of the microorganism, with a higher rate in developing countries [2].

There are two major virulence factors of HP, associated with the severity of disease progression: vacuolating cytotoxin A (VacA) and cytotoxin-associated gene A (CagA) protein. VacA is secreted directly by the bacteria and causes cell damage by vacuolation [1]. The CagA causes mucosal damage by inducing local and systemic, humoral and cellular inflammatory response [2, 3]. Persistent HP infection is associated with gastrointestinal disorders, such as chronic gastritis, peptic ulcer, gastric mucosa-associated lymphoid tissue lymphoma and gastric adenocarcinoma [4, 5]. Several studies have reported a link between HP infection and a variety of extragastroduodenal diseases, such as cardiovascular, neurological, dermatological, haematological and respiratory disorders [6–17]. There is an absence of robust data in the literature on the possible effects of HP infection upon solid organ transplant outcome. The prevalence of HP

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infection in heart and kidney transplant patients is reported to be substantially similar to that of the general population and seems to have low impact on outcome [18–20]. Since no data are yet available on HP infection in lung transplant patients, we evaluated the prevalence and impact of HP infection in a population of such patients.

Methods

Sixty-seven patients who underwent lung transplant in our hospital were included in the study. They were 35 females and 32 males, age 48.4 ± 13.3 years, 54 undergoing bilateral and 13 single lung transplant. The study was conducted at the Regional Referral Centre for Sarcoidosis and other Interstitial Lung Diseases of the Respiratory Diseases and Lung Transplant Unit, Department of Medical, Surgical and Neurological Sciences, University Hospital of Siena, Italy. The local ethics committee approved the study. All patients provided written informed consent to participation.

The following data were collected from all population: underlying respiratory diagnosis, comorbidities, BMI, tobacco exposure, time on the waiting list, unilateral/bilateral transplant, need for extracorporeal membrane oxygenation (ECMO) bridge to LTX, total graft ischemia time, duration of invasive mechanical ventilation, need for tracheostomy, acute kidney injury (all stages) prevalence, grade of primary graft dysfunction (PGD), duration of ICU stay, total in-hospital stay, acute rejection (AR) episodes in the first year after transplant, re-hospitalisations and bronchiolitis obliterans syndrome (BOS)-free survival after 1 and 3 years.

Serology for HP infection was performed for screening purposes and not because of gastrointestinal or other symptoms suggesting HP infection. Blood samples were taken from all patients on the day of transplant. Serum samples were stored at − 70 °C until assay. Serum antibodies against HP and CagA were assayed with commercial kits. Serum IgG antibodies against HP were analysed in patients and controls by ELISA having a sensitivity and specificity of 96% (Diesse Diagnostica Senese, Monteriggioni, Italy) (cut-off 6.2 IU/ml); serum anti-CagA IgG was determined by ELISA with a sensitivity of 95% and a specificity of 90% (CagA–IgG; Genesis Diagnostics Ltd., Cambridgeshire, UK) (cut-off 5.5 IU/ml).

Prevalence was compared with a population-based sample of 797 age- and sex-matched controls with a similar socioeconomic background (Siena Osteoporosis Cohort) [13].

Statistical analysis was conducted using GraphPad Pris v 4.0 for Macintosh, setting significance at $p < 0.05$. Non-parametric tests were used: differences between the two groups were analysed by Mann–Whitney test, analysis of variance was conducted by Kruskal–Wallis test and Fisher's exact test and the Chi-square test with contingency tables was used to compare prevalence. Data were expressed as mean ± standard deviation.

Results

Table 1 reports the demographic data and baseline characteristics of the 67 lung transplant patients enrolled in the study.

Serology for HP infection was positive in 33 patients (49.25%), and serum antibodies to CagA were present in only three out of 33 infected patients (9%). The prevalence of HP infection did not differ with that previously reported in the general population (51.4%) [13], whereas CagA positivity was significantly lower (9% vs. 50.2%, $p < 0.0001$).

Patients who were HP positive were significantly more frequently affected by pulmonary fibrosis ($p = 0.049$), whereas the lowest prevalence of HP positivity (6%), even

Table 1 Demographic data, baseline and perioperative characteristics of the lung transplant population in relation to positivity for *H. pylori*

	HP-negative	HP-positive	Statistics
N	34	33	
Age	50.29 ± 12.79	46.58 ± 13.80	0.33
Gender (female)	20 (58.8%)	15 (45.4%)	0.56
Smoke history	20 (58.8%)	11 (33.3%)	0.050*
BMI	23.01 ± 5.07	24.37 ± 5.06	0.35
Pre-LT diagnosis			
• Pulmonary fibrosis	10 (29.4%)	18 (54.6%)	0.049*
• Cystic fibrosis	8 (23.5%)	11 (33.4%)	1.00
• COPD	11 (32.4%)	2 (6%)	0.011*
• Other	5 (14.7%)	2 (6%)	1.00
Pre-LT comorbidities			
• Diabetes mellitus	15 (44.1%)	17 (51.5%)	0.62
• Arterial hypertension	11 (32.3%)	14 (42.4%)	0.45
• Hypercholesterolemia	15 (44.1%)	10 (30.3%)	0.31
• Osteoporosis	27 (79.4%)	24 (72.7%)	0.57
Time on the waiting list (days)	319.4 ± 271.1	442.0 ± 423.6	0.50
Procedure type (bilateral LT)	30 (88.2%)	24 (72.7%)	0.13
ECMO bridge	4 (11.7%)	4 (12.1%)	1.00
Total ischemic time (min)	346.6 ± 87.0	328.4 ± 91.4	0.66
Induction therapy			
• Basiliximab	25 (73.5%)	27 (81.8%)	0.55
• Thymoglobulin	4 (11.7%)	0	0.053*
• No induction	5 (14.7%)	6 (18.1%)	1.00
CNI therapy			
• Tacrolimus	28 (82.4%)	27 (81.8%)	1.00
• Cyclosporine	6 (17.6%)	6 (18.2%)	

BMI body mass index, *ECMO* extracorporeal membrane oxidation, *CNI* calcineurin inhibitor

*$p \leq 0.05$

lower than in the general population, was found in patients affected by COPD ($p = 0.011$). HP-positive patients were more frequently former smokers ($p = 0.05$). BMI, sex, unilateral/bilateral transplant, time on the waiting list, need for ECMO bridge to LTX, total graft ischemia time were not statistically different between HP-positive and HP-negative patients (Table 1).

Regarding induction therapy, none of the HP-positive positive patients received thymoglobulin, whereas 11.7% ($n = 4$) of HP-negative patients were thus treated ($p = 0.053$); no difference in basiliximab or no induction use was found (73.5% vs. 81.8%, $p = 0.55$ and 14.7% vs. 18.1%, $p = 1.00$). No other statistically significant differences in demographic, baseline or perioperative characteristics were found between HP-positive and HP-negative patients.

HP-positive and HP-negative patients showed similar short- and long-term transplant outcome. No difference in the duration of invasive mechanical ventilation, need for tracheostomy, acute kidney injury prevalence, duration of ICU stay and total in-hospital stay, grade of PGD, AR prevalence or BOS-free survival 1 and 3 years after transplant between HP-positive and HP-negative patients was found (Table 2); however, HP-positive patients more frequently required three or more re-hospitalisations for any cause ($p = 0.046$).

No correlations with any study variables were found in HP-CagA-positive patients.

Discussion

In the present study, we analysed seroprevalence of HP infection in a cohort of patients who underwent lung transplant in our hospital. HP infection is associated with several gastrointestinal and extragastrointestinal diseases, including cardiovascular, neurological, dermatological, haematological and respiratory disorders [4–17].

The prevalence and impact of HP infection among solid organ transplant patients has only marginally been studied. In heart transplant recipients, histological diagnosis of HP infection is reported to be widely prevalent, like in the general population [18]. In a cohort of kidney transplant patients, its prevalence was significantly lower than in a healthy control group, and the infection proved to have no impact on clinical outcome [19, 20]. Some years ago, other authors suggested that *H. pylori* was involved in the development of duodenal ulcer after adult living-donor liver transplant, particularly in patients with hypergastrinemia and high serum pepsinogen [21], but no confirmatory studies were ever performed.

No data are available on HP infection in lung transplant patients. This is therefore the first study aimed at evaluating its prevalence and impact in these patients. We found that the prevalence of HP was similar to that reported in the general population, represented by a large population-based sample of age- and sex-matched controls

Table 2 Lung transplant outcome in HP-positive and HP-negative transplant patients

	HP-negative	HP-positive	Statistics
PGD at 72 h			
• Grade 1	13 (38.2%)	9 (27.3%)	0.43
• Grade 2	10 (29.4%)	16 (48.5%)	0.13
• Grade 3	9 (26.4%)	8 (24.2%)	1.00
MV > 96 h	13 (38.2%)	15 (45.4%)	0.62
Tracheostomy	4 (11.7%)	10 (30.3%)	0.11
AKI (all stages)	7 (20.5%)	6 (18.2%)	1.00
ICU stay (days)	16.13 ± 19.60	18.52 ± 18.44	0.61
Total in-hospital stay (days)	38.04 ± 15.85	44.58 ± 26.01	0.36
AR	16 (47.0%)	12 (36.4%)	1.00
Recurring AR (> 1 episode in the 1st year)	6 (17.6%)	6 (18.2%)	1.00
Number of post LT re-hospitalisation			
• 0	13 (38.2%)	7 (21.2%)	0.11
• 1–3	11 (32.3%)	9 (27.3%)	0.60
• > 3	9 (26.5%)	18 (54.5%)	0.046*
BOS			
• BOS-free survival at 1 year post LT	83.9%	82.2%	1.00
• BOS-free survival at 3 year post LT	74.2%	59.6%	0.20

PGD primary graft dysfunction, *MV* mechanical ventilation, *AKI* acute kidney injury, *AR* acute rejection, *BOS* bronchiolitis obliterans syndrome

*$p \leq 0.05$

with a similar socioeconomic background [13]. By contrast, among HP-positive patients, CagA positivity was found to be significantly lower ($p < 0.0001$). The meaning of this finding is uncertain, and no specific variables or protective factors for HP-CagA infections were present in our population.

Interestingly, the prevalence of HP was higher in patients who underwent lung transplant because of pulmonary fibrosis. The role of HP in IPF is debated. A previous study by our research group showed similar HP seroprevalence in patients with idiopathic pulmonary fibrosis (IPF) and in the general population, but HP positivity was found to be related to poor prognosis with higher rates of mortality and lung functional decline, suggesting that the bacterium may have a role in disease progression [13]. In 2016, Kreuter at al. screened 39 lung biopsies from IPF patients for HP DNA by PCR. They failed to demonstrate the presence of bacterial genome, but the study had several limitations that advise caution in interpreting the results [22]. In our study population, we observed higher prevalence of former smokers in HP-positive patients ($p = 0.05$). Smoke is considered a risk factor for pulmonary fibrosis [13] and more than half of our pulmonary fibrosis patients were former smokers; this can in part explain our finding.

HP infection has been studied in relation to COPD and some hypotheses of direct injury and chronic inflammation via inhalation and aspiration of microorganisms have been formulated [14, 15]; however, we found a reduced prevalence of HP in our COPD patients than in other disorders and in the general population.

HP infection did not seem to play a significant role in graft outcome in our lung transplant population. A part previously discussed different prevalence of pulmonary fibrosis and COPD and rate of thymoglobulin use; no other significant difference was found in baseline and perioperative characteristics and immunosuppressive management among HP-positive and HP-negative patients.

Variables used to express short-term outcome (PGD, AR, duration of invasive mechanical ventilation, need for tracheostomy, acute kidney injury prevalence, duration of ICU stay, total in-hospital stay) were not significantly different in HP-positive and -negative patients and similarly, regarding mid- to long-term outcome, BOS-free survival was similar in the two groups. However, significantly more patients needed three or more re-hospitalisations after transplant among HP-positive patients: the reason is unclear, but due to the relative small number of subjects no clear conclusion can be drawn. Indeed, the size of the patient cohort is the main limit of our study, although it is clear that the prevalence of HP infection in our population did not differ from that of the general population and it did not have a negative effect on lung allograft outcome.

Our results substantially confirm previous observations in other solid organ transplant patients [18–21]. Although interesting hypotheses regarding Th1 inflammation due to HP infection [23] have been postulated, which could be expected to facilitate graft rejection, no impact of bacterial infection has been observed in heart, kidney and lung transplant recipients.

In conclusion, HP infection prevalence in lung transplant patients was comparable to that of the general population and to that reported in other solid organ transplants, such as heart and kidney recipients. It did not seem to impact short-, mid- or long-term lung allograft outcome. *H. pylori* infection did not prove to be clinically relevant in lung transplant patients.

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Compliance with Ethical Standards

Conflict of interest None of the authors have any competing interests to declare.

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