Early Sirolimus-Based Immunosuppression is Safe for Lung Transplantation Patients: Retrospective, Single Arm, Exploratory Study

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Background: Sirolimus, a mechanistic target of sirolimus inhibitor, is an immunosuppression medication for patients undergoing heart and abdominal transplantation. Sirolimus-based immunosuppression administered de novo post-lung transplantation is associated with bronchial Anastomosis healing-related complications. We hypothesized that sirolimus administration within the first postoperative month in selected lung transplant recipients is safe and may be associated with favorable short-term and long-term outcomes due to its anti-proliferative properties and minimal adverse side effects.

Material/Methods: Thirteen patients (13.3%; mean age, 46.8±11.9 years) received early sirolimus-based immunosuppression along with cyclosporine and prednisone; 10 patients received single-lung transplantation, 3 received double-lung transplantation, and all received induction immunosuppressants. Patients received early sirolimus-based immunosuppression after an uncomplicated postoperative course and detailed bronchoscopic assessment.

Results: Sirolimus was begun on a mean of 20.6±4.7 days postoperatively (range, 14–32 days). The in-hospital and 30-day mortality rate was 0%. At long-term follow-up, 5 patients died (due to bacterial infection in 4 patients and pneumocystis jiroveci pneumonia in 1 patient). The mean overall survival was 4.4±2.53 (range, 0.8–10.0) years, 1-year survival was 92%, and 5-year survival was 62%. In 4 patients (30.8%), sirolimus was stopped due to infection in 3 patients and re-transplantation in 1 patient. Only one of the 13 patients developed bronchiolitis obliterans syndrome. In patients still taking sirolimus, renal function, systolic blood pressure, and lipid profile were within normal ranges; however, these patients required statin therapy.

Conclusions: In selected lung transplant recipients, early sirolimus-based immunosuppression is safe and associated with beneficial short-term and long-term outcomes.

MeSH Keywords: Immunosuppression • Lung Transplantation • Organ Transplantation • Sirolimus • TOR Serine-Threonine Kinases

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Background

The early and late results of lung transplantation (LTx) are constantly improving. Early acute rejection (AR) episodes are prognostic and have been implicated in the development of chronic rejection, which is termed chronic lung allograft dysfunction (BOS) [1]. The current immunosuppression regimen involves calcineurin inhibitors; however, they have a serious side effect profile that includes chronic renal insufficiency (with rates of 20–30%) that leads to worse long-term survival in LT recipients [2]. Sirolimus has an established immunosuppressive role in heart and abdominal transplantation patients [3,4]. Studies of other organs showed that calcineurin inhibitor use can be significantly reduced and patients can even be weaned off them with good results when m-tor inhibitors are used for treatment [5]. Importantly, sirolimus, when administered de novo after LTx, has been associated with an increased number of bronchial healing complications that lead to increased early mortality. Therefore, the US Food and Drug Administration (FDA) warns that sirolimus should not be administered de novo after LTx [5,6]. Regardless of its bronchial complications, sirolimus has been shown to reduce the number of AR episodes when it is administered de novo after LTx [5]. Reducing the number of AR episodes and eliminating medications that cause and contribute to long-term morbidity may significantly improve the long-term survival of LT recipients. We believe that sirolimus, when begun within the first postoperative month in selected LT recipients according to the standardized inclusion criteria, is safe and associated with favorable short-term and long-term outcomes due to its anti-proliferative properties and minimal adverse side effects [7].

Immunosuppression with sirolimus as the primary immunosuppressant and implementation within the first month after LTx should be achieved with the least possible nephrotoxicity; should have antiviral, antitumor, and antifungal properties; and should have properties that may inhibit the development of bronchiolitis obliterans (BOS). We hypothesized that these 3 targets can be achieved by the early (within the first month) administration of sirolimus-based immunosuppression in appropriately selected LT recipients. Therefore, this study aimed to evaluate the safety of using sirolimus early in the postoperative period. The study was retrospective, single arm, exploratory in selected group of patients.

Material and Methods

Ethical considerations

The approval of the local ethics committee was obtained for this study (KNW/0022/KB229/17, the Bioethical Committee, Silesian University of Katowice). Due to the retrospective nature of this study, the need to obtain informed consent was waived.

Patients

We reviewed cases of patients who received early sirolimus-based immunosuppression. We did not perform a comparative study because of major heterogeneity between the eventually studied groups (patients with uncomplicated follow-up versus those with complications during follow-up). From December 2004 to November 2014, 98 LTx procedures were performed at the Zabrze Lung Transplant Program at the Silesian Centre for Heart Disease in Zabrze, Poland. Thirteen patients (13.3%) with a mean age of 46.8±11.9 years were selected to undergo an early sirolimus-based immunosuppression regimen.

Early sirolimus administration was defined as administration within the first 30 days post-transplantation. Ten patients received single LTx, 3 patients received double LTx, and all patients received induction immunosuppression. Patients were selected to receive early sirolimus if they had an uncomplicated postoperative course and were decisively selected after undergoing a detailed bronchoscopic assessment.

Inclusion and exclusion criteria

Patients were administered early sirolimus if they had: uncomplicated healing of bronchial anastomosis after a detailed bronchoscopic assessment until the 14th postoperative day (POD), an uncomplicated intraoperative and early postoperative course, and no primary graft dysfunction.

The exclusion criterion for early sirolimus administration was the presence of any healing complications other than bronchial (e.g., prolonged air leaks or wound healing complications).

Drug administration

Each consecutive patient with LTx with an uncomplicated intraoperative course was assessed 72 hours postoperatively to see if early sirolimus could be administered. At our institution, the standard protocol is to perform a bronchoscopic examination of bronchial anastomosis daily and before extubation. When early anastomotic healing is appropriate, patients received single LTx, 3 patients received double LTx, and all patients received induction immunosuppression. Patients were selected to receive early sirolimus if they had an uncomplicated postoperative course and were decisively selected after undergoing a detailed bronchoscopic assessment.

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Drug administration

Each consecutive patient with LTx with an uncomplicated intraoperative course was assessed 72 hours postoperatively to see if early sirolimus could be administered. At our institution, the standard protocol is to perform a bronchoscopic examination of bronchial anastomosis daily and before extubation. When early anastomotic healing is appropriate, patients are flagged as candidates for early sirolimus-based immunosuppression. Then, on approximately days 10–14 and before days 28–30, a decisional bronchoscopic examination is performed to determine if an LTx patient should receive early sirolimus-based immunosuppression. Our bronchoscopic examination consists of assessing the bronchial anastomosis using the Couraud Grading System. Only patients with grade 1 anastomosis (complete, circumferential, primary mucosal healing) were accepted as candidates for sirolimus [8]. When an LTx patient is indicated to undergo sirolimus-based immunosuppression, sirolimus is begun at 1 mg per day; the trough levels are checked daily. Then, the dose is augmented twice daily.
When sirolimus was begun, mycophenolate mofetil was immediately discontinued. A low dose of daily prednisolone was administered (5 mg/day). If the patient did not experience the side effects of this drug, our aforementioned regimen was maintained until 6 months postoperatively, after which our protocol calls for a slow reduction of cyclosporine to a level of approximately 150 ng/mL at the end of the first year after transplantation. Simultaneously, the sirolimus dose was raised to a target level of 10–14 ng/mL. After the first year, cyclosporine was slowly reduced over a period of 3 to 6 months to possibly discontinue the drug and start mycophenolate mofetil/azathioprine. Thus, the target long-term therapy in this regimen was combined sirolimus, mycophenolate mofetil, and prednisolone. Depending on the number of AR episodes and infections, further treatment was individualized. The appearance of nephrotoxicity at any given time resulted in a very rapid and large (50%) reduction in cyclosporine doses (100–150 ng in the first year after transplantation), with a mild increase in the target sirolimus level of 10–14 ng/mL.

Furthermore, when cyclosporine was discontinued, in a significant percentage of cases with long-term follow-up, mycophenolate mofetil was not restarted and patients were essentially maintained on 2-drug immunosuppression to have the fewest number of other-organ complications as possible. If a major surgical procedure was needed, patients were admitted as inpatients and transitioned to cyclosporine or tacrolimus. All elective surgical procedures were deferred for at least 3 months. After this time, patients could be transitioned back to sirolimus.

Because each patient undergoing LTx was considered a potential candidate for sirolimus therapy, the calcineurin inhibitor of choice was cyclosporine. We think that the combination of sirolimus and tacrolimus, which affect the same target protein (the FK 506 binding protein) might be unfavorable because of the competitive mechanism of these drugs.

Statistical analyses

The statistical analyses were performed with Microsoft® Office Excel 2000 (Microsoft Corporation) and Statistica 6.0 (StatSoft, Inc.). The distribution of the analyzed data was assessed using the Shapiro-Wilk test for normality. The differences between means were analyzed with Student’s t-test, and in other cases, the non-parametric Mann-Whitney U significance test was used; P values of ≤0.05 were considered statistically significant. Data are presented as the mean ± standard deviation or median ± quartile deviation.

Results

Patients

The patient characteristics are summarized in Table 1. Sirolimus was begun on a mean POD of 20.6±4.7 (range, 14–32 days). The detailed results of all sirolimus levels that were achieved in patients who were treated with this drug are presented in Table 2. The in-hospital and 30-day mortality rate was 0%. No anastomosis dehiscence was observed in the study cohort. The detailed results regarding the cyclosporine levels are shown in Table 3.

On long-term follow-up, 5 patients died. The reasons for death included bacterial infection in 4 patients and pneumocystis jiroveci (PJP) infection in 1 patient. The post-transplantation results of patients who were treated with sirolimus are presented in Table 4.

The details of the deceased patients were as follows:

Patient number 2, Table 4

This patient died 0.82 years after undergoing LTx. She had a very smooth and uncomplicated postoperative and hospital course and was discharged home in very good condition.
At home, she ceased preventive cotrimoxazol treatment due to nausea without conveying this information to the transplantation team. Eight months after transplantation, she was admitted on an emergency basis with dyspnea. Transbronchial biopsy was performed and showed a massive number of PJP. Despite undergoing maximal therapy, the patient died.

**Patient number 3, Table 4**

This patient died 5.42 years after undergoing LTx. After having an uncomplicated follow-up course, this patient had some social status problems as well as poor compliance and problems that were reported too late. The patient suffered from

| Patient No. | 1 | 2 | 3 | 4 | 5 | 6 |
|-------------|---|---|---|---|---|---|
| Age* (years) | 54.7 | 47.5 | 44.5 | 58.9 | 46.6 | 29.5 |
| Sex | M | F | M | M | M | F |
| BMI* (kg/m²) | 27.7 | 21 | 20.4 | 21.2 | 16.0 | 19 |
| Diagnosis | IPF | IPF | IPF | COPD | ILD | BO |
| Days of being on the waiting list | 1 | 161 | 297 | 633 | 407 | 393 |
| Diabetes before LTx | Yes | No | No | No | No | No |
| Hypertension before LTx | No | Yes | No | No | Yes | No |
| Cr* (µmol/L) | 79 | 50 | 64 | 79 | 32 | 90 |
| GFR* (mL/min/1.73 m²) | 112.4 | 108 | 101.4 | 93.5 | 273.8 | 68.2 |
| Hypercholesterolemia before LTx | No | Yes | No | Yes | No | Yes |
| LTx type | SLT | SLT | SLT | SLT | SLT | SLT+reLT |
| Duration of mechanical ventilation (hours) | 54 | 11 | 33 | 27 | 22 | 35 |
| CPB time (min) | 136 | N/A | 312 | N/A | N/A | N/A |
| ICU length of stay (days) | 4 | 10 | 7 | 10 | 5 | 4 |
| Induction agent | ATG | ATG | ATG | ATG | ATG | ATG |
| Hospital length of stay (days) | 77 | 38 | 35 | 33 | 66 | 59 |
| Other immunosuppressive drugs | CSA, MMF, Prednisolone | MMF, Prednisolone | CSA | CSA, MMF, Prednisolone | CSA, MMF, Prednisolone | CSA, MMF, Prednisolone |
| Postoperative day of starting sirolimus | 17 | 16 | 14 | 18 | 26 | 32 |
| Postoperative day of stopping sirolimus and reason | Still taking from 41 to 58 days because of groin healing complications | 254 days; Acute rejection, PCP | 1,581 days | Took until the end of life | 335 days; general surgery procedure | 150 days; reLT, pneumonia due to immunosuppression |
Table 1 continued. Characteristics of sirolimus-administered patients.

| Patient No. | 7 | 8 | 9 | 10 | 11 | 12 | 13 |
|-------------|---|---|---|----|----|----|----|
| **Age** (years) | 24.8 | 45.1 | 59.5 | 57.1 | 41.6 | 17.9 | 49.3 |
| **Sex** | F | M | F | M | M | M | M |
| **BMI** (kg/m$^2$) | 17.7 | 20.0 | 26.3 | 23.2 | 26.7 | 17.8 | 17.7 |
| **Diagnosis** | iPAH | COPD | LAM | COPD | COPD | CF | CF |
| **Days of being on the waiting list** | 290 | 582 | 932 | 96 | 1 | 64 | 489 |
| **Diabetes before LTx** | No | No | No | No | No | Yes | Yes |
| **Hypertension before LTx** | No | No | Yes | Yes | No | No | No |
| **Cr** (µmol/L) | 82 | 93 | 67 | 95 | 97 | 44 | 71 |
| **GFR** (mL/min/1.73 m$^2$) | 78.7 | 81.35 | 82.50 | 75.8 | 105.0 | 232.5 | 109.0 |
| **Hypercholesterolemia before LTx** | No | No | Yes | No | Yes | No | No |
| **LTx type** | SLT | DLT | SLT | SLT | SLT | DLT | DLT |
| **Duration of mechanical ventilation (hours)** | 49 | 20 | 12 | 20 | 11 | 10 | 16 |
| **CPB time (min)** | 190 | N/A | N/A | N/A | N/A | N/A | N/A |
| **ICU length of stay (days)** | 17 | 8 | 5 | 3 | 5 | 4 | 2 |
| **Induction agent** | ATG | ATG | ATG | Basiliximab | Basiliximab | Basiliximab | ATG |
| **Hospital length of stay (days)** | 74 | 100 | 50 | 42 | 48 | 41 | 44 |
| **Other immunosuppressive drugs** | CSA, MMF | CSA, MMF, Prednisolone | MMF, Prednisolone | MMF, Prednisolone | CSA, MMF | CSA, Prednisolone | CSA, MMF |
| **Postoperative day of starting sirolimus** | 21 | 19 | 24 | 18 | 21 | 20 | 22 |
| **Postoperative day of stopping sirolimus and reason** | Still taking | Still taking | Still taking | 947 days; Operation: volume lung reduction surgery – native lung hyperinflation syndrome | Still taking | Still taking | Still taking |

* At LTx day; CBP – cardiopulmonary bypass; ICU – intensive care unit; IPF – idiopathic pulmonary fibrosis; LAM – lymphangioleiomyomatosis; ILD – interstitial lung disease; iPAH – idiopathic pulmonary arterial hypertension; COPD – chronic obstructive pulmonary disease; BO – bronchiolitis obliterans; CF – cystic fibrosis; SLT – single lung transplant; DLT – double lung transplant; reLT – lung retransplantation; N/A – not applicable; ATG – anti-thymocyte globulin; POD – postoperative day; CSA – cyclosporin; MMF – mycophenolate mofetil.
Table 2. Average sirolimus levels obtained from all measurements in all sirolimus-administered patients after transplantation.

| Patient No. | Mean/median | SD/quartille deviation | Minimum | Maximum |
|-------------|-------------|-------------------------|---------|---------|
| 1           | 11*         | 5.22                    | 1       | 28.7    |
| 2           | 12.3*       | 8.75                    | 2.9     | 30      |
| 3           | 10.7*       | 3.30                    | 3.6     | 27.5    |
| 4           | 13.6        | 6.48                    | 2.5     | 28.7    |
| 5           | 12.9*       | 6.23                    | 2.5     | 30      |
| 6           | 14.7*       | 3.75                    | 2.5     | 30      |
| 7           | 12.9*       | 4.43                    | 2.9     | 46.6    |
| 8           | 30.0*       | 2.35                    | 6.1     | 31.7    |
| 9           | 13.6        | 4.46                    | 6.1     | 25.3    |
| 10          | 14.1        | 14.00                   | 3.3     | 27.5    |
| 11          | 10.0        | 4.06                    | 2       | 17.8    |
| Average     | 13.6        | 3.3                     |         | 27.9    |
| SD          | 5.24        | 1.47                    |         | 7.32    |

SD – standard deviation. * Median ± quartile deviation.

Table 3. Average Cyclosporin levels obtained from all measurements during the simultaneous administration of sirolimus after transplantation.

| Patient No. | Mean/median | SD/quartille deviation | Minimum | Maximum |
|-------------|-------------|-------------------------|---------|---------|
| 1           | 122.5*      | 45.74                   | 37.4    | 308.9   |
| 2           | 250.3       | 92.16                   | 110.2   | 415.7   |
| 3           | 163.65*     | 62.34                   | 22.7    | 331     |
| 4           | 232.1*      | 128.90                  | 37.7    | 855.4   |
| 5           | 290.9*      | 84.86                   | 118     | 809     |
| 6           | 267.4*      | 114.55                  | 25.3    | 798.6   |
| 7           | 200.6*      | 119.13                  | 23.9    | 1570    |
| 8           | 262.2*      | 88.14                   | 89.4    | 847.2   |
| 9           | 174.3*      | 49.80                   | 5.3     | 396.6   |
| 10          | 247.5*      | 91.03                   | 21.7    | 320.2   |
| 11          | 172.7       | 88.59                   | 50.1    | 349.9   |
| 12          | 156.7*      | 51.83                   | 20.7    | 531.9   |
| 13          | 202.4       | 117.38                  | 14.5    | 470.6   |
| Average     | 211.0       | 44.4                    |         | 615.8   |
| SD          | 51.26       | 37.25                   |         | 358.44  |

SD – standard deviation. * Median ± quartile deviation.
Table 4. Post-transplantation results (based on the last visit or last measurement during sirolimus administration) of sirolimus-administered patients.

| Patient No. | Acute rejection <1 year | Acute rejection >1 year | Diabetes | Hypertension | Dyslipidemic | Statin use | Cr (μmol/L) | GFR (mL/min/1.73 m²) | PJP | CMV Disease | In-hospital/30-day mortality | Mortality | Survival (year) | Best FEV1 (% of best) | BOS |
|-------------|-------------------------|-------------------------|----------|--------------|--------------|------------|-------------|----------------------|-----|--------------|-------------------------|-----------|------------------|-------------------------|-----|
| 1           | Yes                     | No                      | Yes      | Yes          | Yes          | Yes        | 128         | 75.62                | Yes | No           | No                      | No        | 10.01            | 1.85                   | 1.63 (88) |
| 2           | Yes (1)                 | Yes (1)                 | Yes      | Yes          | Yes          | No         | 118         | 66                   | Yes | No           | No                      | Yes        | 0.82             | 1.33                   | 1.3 (100) |
| 3           | No                      | No                      | Yes      | Yes          | Yes          | Yes        | 119         | 59.8                 | Yes | No           | No                      | Yes        | 5.42             | 1.75                   | 1.43 (82) |
| 4           | No                      | No                      | Yes      | Yes          | Yes          | Yes        | 193         | 32.93                | No  | Yes          | No                      | Yes        | 1.62             | 3.35                   | 0       |
| 5           | Yes                     | No                      | No       | Yes          | Yes          | Yes        | No          | 103                  | 71.73 | No           | No                      | No        | 7.00             | 1.87                   | 1.87 (100) |
| 6           | No                      | No                      | No       | No           | No           | No         | No          | 93                   | 65.75 | Yes          | No                      | No        | Yes              | 1.15                   | 0.95 (83) |
| 7           | No                      | No                      | No       | No           | Yes          | Yes        | 70          | 91.62                | No  | Yes          | No                      | No        | 6.75             | 3.55                   | 3.51 (99) |
| 8           | Yes (1)                 | No                      | No       | No           | No           | Yes        | No          | 99                   | 75    | Yes          | No                      | No        | 5.16             | 2.48                   | 1.30 (52) |
| 9           | No                      | No                      | Yes      | Yes          | Yes          | Yes        | Yes         | 95                   | 55.33 | No           | No                      | No        | 4.26             | 1.24                   | 1.19 (96) |
| 10          | No                      | N/A                     | No       | Yes          | No           | Yes        | No          | 162                  | 40.47 | No           | Yes                     | Yes        | 3.24             | 1.85                   | 1.37 (74) |
| 11          | Yes (1)                 | No                      | No       | Yes          | Yes          | Yes        | Yes         | 150                  | 47.26 | No           | No                      | No        | 2.76             | 2.89                   | 2.68 (93) |
| 12          | No                      | No                      | No       | No           | Yes          | Yes        | No          | 81                   | 132.03| No           | No                      | No        | 2.74             | 3.54                   | 3.32 (94) |
| 13          | No                      | Yes                     | No       | Yes          | No           | Yes        | Yes         | 209                  | 31.39 | Yes          | No                      | No        | 2.41             | 2.92                   | 2.82 (96) |

Cr – creatinine; GFR – glomerular filtration rate; PJP – pneumocystis jiroveci; CMV – cytomegalovirus; FEV – forced expiratory volume.

Sudden dyspnea about a month before admission to the hospital, experienced rapid graft damage, and died during the diagnostic process due to massive hemorrhage in the native lung.

**Patient number 4, Table 4**

This patient died 1.62 years after undergoing LTx. The patient had poor compliance; although he had the symptoms of advanced pneumonia, he refused any medical help. He reported his problems too late and suffered from severe bacterial pneumonia. The patient was treated in a local hospital and died from pneumonia-related respiratory insufficiency.

**Patient number 6, Table 4**

This patient died 4.53 years after undergoing LTx due to infection. Revision LTx was performed as a rescue therapy on POD 150 due to massive pneumonia with respiratory insufficiency as a result of central vascular catheter contamination.

**Survival and complications**

The mean overall probability of survival was 4.4±2.53 years (range, 0.8–10.0), the 1-year survival rate was 92%, and 5-year...
The survival rate was 62%. The Kaplan-Meier survival estimation of patients who were treated with sirolimus is presented in Figure 2. The survival of LT recipients who were treated with sirolimus did not differ from the survival of LT recipients who were treated without it.

In 4 patients (31%), sirolimus was stopped completely; 3 patients had infection and 1 patient was referred for re-transplantation. Due to our protocol, sirolimus was discontinued in 3 patients as our study target (Table 1).

The sirolimus trough level varied (minimum: 1 ng/mL and maximum: 30 ng/mL). The average value was 13.6 ng/mL in patients who continued to take cyclosporine. The average cyclosporine trough level was 211.0 ng/mL in patients who continued to take sirolimus.

Acute cellular rejection within the first year was diagnosed in 4 patients. Only 1 patient had an episode of acute cellular rejection after 1 year of observation (Table 4).

The creatinine values were statistically and significantly increased after transplantation during the study period, and the glomerular filtration rate (GFR) was decreased. There was a statistically significant difference in the creatinine level and GFR before and after transplantation during the study period: 73 μmol/L versus 125 μmol/L and 117 mL/min/1.73 m² versus 65 mL/min/1.73 m², respectively (Table 5).

The percentage of patients with diabetes increased during the studied period, from 23% to 38%, but not significantly (P>0.05). The percentage of patients with hypertension symptoms increased, from 31% to 46% (P>0.05) (Table 5).

There was a significant difference in patients with dyslipidemia before and after transplantation during the study period: 38% versus 77%, respectively (P<0.05) (Table 5).

Pre-emptive anti-cytomegalovirus treatment (based on a positive pp65 test) was necessary in 3 patients. Cotrimoxazol was administered in 7 patients based on the results of immunofluorescence sputum or a bronchoalveolar lavage study for the presence of PJP. This treatment was implemented as a preventative protocol, even if there were no symptoms of pneumonia or infection.

Only 1 of the 13 patients developed BOS-2 symptoms. Probable BOS (0-p) was observed in 4 patients. Bronchial complications appeared in 1 of the patients about 6 months after transplantation. These complications were judged as serious, and the patients were treated with stent implantation and frequent balloon bronchoplasty procedures. These consisted of the malacia distal to the anastomosis part of the bronchi.

**Discussion**

The main purpose of the study was to prove that sirolimus administered early and with specific criteria applied after lung transplantation is safe. As we have shown, administration of sirolimus as early as POD 15 in selected patients (without

**Table 5.** Mean values of parameters assessed before and after transplantation.

| Parameter          | Mean value pre-transplantation | Mean value post-transplantation* | p-Value   |
|--------------------|--------------------------------|----------------------------------|-----------|
| Creatinine [μmol/L] | 72.5±20.6                       | 124.6±42.6                       | 0.0008^   |
| GFR [mL/min/1.73 m²]| 117.1±62.6                      | 65.0±26.9                        | 0.0049^   |
| Diabetes           | 23% (n=3)                       | 38% (n=5)                        | NS^       |
| Hypertension       | 31% (n=4)                       | 46% (n=6)                        | NS^       |
| Hypercholesterolemia| 38% (n=5)                       | 77% (n=10)                       | 0.0236^   |

GFR – glomerular filtration rate; * last visit; ^ student t-test; ^ the significance difference test between two proportions.
Sirolimus administration does not show any advantages or close to those of the general population of LT recipients [2]. Transplantation (ISHLT) data showed, these diagnoses are cellular rejection in the first year after transplantation, and Four patients in our study group experienced severe acute point of the study in terms of adhering to the sirolimus protocol. shortened exposure to calcineurin inhibitors. This was a weak expected nephroprotective effect may occur with decreased and to the introduction of immunosuppression strategies. The ex- erative effect of the protocol, possibly due to the inadequate, not dynamic enough, dose reduction of calcineurin inhibitors. During the long-term follow-up, the lipid and glucose profiles showed an unfavorable trend. This study had some limitations. We could not use a compar- ative analysis because it was impossible to randomize the patients. Patients who had to be excluded from the study group based on bronchoscopic evaluation could not serve as the control group. In addition, the number of patients was small despite the long enrolment time. Our program start- ed in December 2004 and was the only lung transplantation program in Poland. The number of transplantations per year was low at the beginning – mainly due to a very low number of patients directed to this treatment as it was very new at that time. After a few years, the situation had changed. The strict adherence to criteria which allowed us to enroll patients into early sirolimus immunosuppression was the reason only 13 patients were studied. Furthermore, in this retrospective analysis, we found that the cyclosporine tapering rate was not in accordance with the established protocol in which a lower level of the drug was postulated.

Conclusions

The study was retrospective, single arm, exploratory in a select- ed group of patients. Although the number of patients in our study was small, this is perhaps the largest group of its kind. The described protocol is safe for bronchial healing with strict adherence to the enrollment protocol, with the definition of complications postoperatively) was not associated with bron- chial dehiscence.

Considering the current knowledge about the physiology of wound healing, in this study we rigorously adhered to a proto- col to completely avoid non-healing of bronchial anastomo- sis. More specifically, we did not note any cases with bronchi- cal anastomosis dehiscence, and no patients were at a risk of having such a complication.

There was no need to discontinue sirolimus in response to the suspicious appearance of bronchial healing after switch- ing to therapy. Therefore, this finding is in contrast to the results of studies from 2 centers [5,6] that reviewed the back- ground for the FDA’s decision in which sirolimus “de novo” in lung transplantation was banned. The aforementioned pub- lications were focused on how very early sirolimus would af- fect outcomes in lung transplantation patients. These studies were unique because they initiated sirolimus on POD 1; we feel that this was the driving, most important reason behind the higher incidence of bronchial dehiscence in those studies compared to our study. Moreover, our study considered the natural process of wound healing.

The most important aspect of wound healing which sirolimus may inhibit are: fibrocytes mobilization and migration, fibrocytes turn into fibroblasts, early angiogenesis within the wound, fi- broblast production of fibrous collagen, production of required growth factors, and local healing mediators. These steps achieve their plateau around day 14. Knowing this, we decided to initi- ate sirolimus no earlier than POD 14. One of the basic principles of and expectations about the early implementation of siroli- mus was the hope of better kidney protection [9,10]. However, in the group of patients who were analyzed in this study, there was a significant deterioration in renal function. The assess- ment was based on the analysis of the GFR and serum creat-inine concentrations. At the end of the observation period, a statistically significant difference in the parameters was ob- served. After analyzing all the data, we postulated that we re- duced the cyclosporine dose very slowly and late. This was due to the introduction of immunosuppression strategies. The ex- pected nephroprotective effect may occur with decreased and shortened exposure to calcineurin inhibitors. This was a weak point of the study in terms of adhering to the sirolimus protocol.

Four patients in our study group experienced severe acute cellular rejection in the first year after transplantation, and 1 patient was diagnosed with AR beyond 1 year after trans- plantation. As the International Society for Heart and Lung Transplantation (ISHLT) data showed, these diagnoses are close to those of the general population of LT recipients [2]. Sirolimus administration does not show any advantages or disadvantages in this regard.

In our study group, there was an upward trend in the incidence of diabetes and hypertension during the long-term follow-up period. The increase in the incidence of both diabetes and hyper- tension did not differ from the data reported in the ISHLT re- ports [2] and it cannot be clearly stated that this increase was associated with the described immunosuppression protocol.

There was a significant increase in the number of patients with lipid abnormalities in our study group. However, the in- cidence of vascular complications was not increased. None of the patients had coronary artery disease or complications due to peripheral vascular disease.

The study protocol described for the use of “de novo” siroli- mus in patients undergoing LTx appears to be safe regard- ing bronchial healing. The results of long-term observation in this small group of patients indicate a favorable trend in BOS- related outcomes which can be explained by the anti-prolif- erative effect of early treatment [11]. On the other hand, we must bear in mind that the undisturbed process of bronchi- al healing seen in patients before sirolimus introduction can also be a favorable anti-BOS factor. There was no nephropro- tective effect of the protocol, possibly due to the inadequate, not dynamic enough, dose reduction of calcineurin inhibitors. During the long-term follow-up, the lipid and glucose profiles showed an unfavorable trend.

This study had some limitations. We could not use a compar-ative analysis because it was impossible to randomize the patients. Patients who had to be excluded from the study group based on bronchoscopic evaluation could not serve as the control group. In addition, the number of patients was small despite the long enrolment time. Our program start- ed in December 2004 and was the only lung transplantation program in Poland. The number of transplantations per year was low at the beginning – mainly due to a very low number of patients directed to this treatment as it was very new at that time. After a few years, the situation had changed. The strict adherence to criteria which allowed us to enroll patients into early sirolimus immunosuppression was the reason only 13 patients were studied. Furthermore, in this retrospective analysis, we found that the cyclosporine tapering rate was not in accordance with the established protocol in which a lower level of the drug was postulated.
early drug introduction being within first month. Therefore, for lung transplantation centers that are of the opinion that early modification of immunological response after transplantation may improve long-term results, especially the occurrence of BOS, our protocol could be a guide on how to identify patients who could be treated with “de novo” sirolimus. The promising BOS tendency that we report may be the impetus for a large prospective multicenter study that observes a significantly higher number of patients than that observed in our study.

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