Aus der

Abteilung für Thoraxchirurgie

Klinikum der Ludwig-Maximilians-Universität München

Lung transplantation in times of Covid-19 – Immune response and clinical management of recent lung allograft recipients infected with SARS-CoV 2

Dissertation

zum Erwerb des Doktorgrades der Medizin

an der Medizinischen Fakultät

der Ludwig-Maximilians-Universität München

vorgelegt von

Olaf Michael Glück

aus

Ulm

Jahr

2025

Mit Genehmigung der Medizinischen Fakultät der
Ludwig-Maximilians-Universität München

Erstes Gutachten: Priv. Doz. Dr. Teresa Kauke

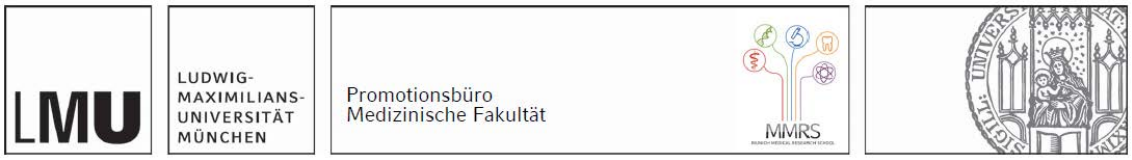
Zweites Gutachten: Prof. Dr. Sebastian Michel

Drittes Gutachten: Priv. Doz. Dr. Christof Geldmacher

Dekan: Prof. Dr. med. Thomas Gudermann

Tag der mündlichen Prüfung: 16.04.2025

1. Affidavit



Eidesstattliche Versicherung

Glück, Olaf Michael

Ich erkläre hiermit an Eides statt, dass ich die vorliegende Dissertation mit dem Titel:

“Lung transplantation in times of Covid-19 – Immune response and clinical management of recent lung allograft recipients infected with SARS-CoV 2”

selbständig verfasst, mich außer der angegebenen keiner weiteren Hilfsmittel bedient und alle Erkenntnisse, die aus dem Schrifttum ganz oder annähernd übernommen sind, als solche kenntlich gemacht und nach ihrer Herkunft unter Bezeichnung der Fundstelle einzeln nachgewiesen habe.

Ich erkläre des Weiteren, dass die hier vorgelegte Dissertation nicht in gleicher oder in ähnlicher Form bei einer anderen Stelle zur Erlangung eines akademischen Grades eingereicht wurde.

München, 28.04.2025	Olaf Michael Glück
Ort, Datum	Unterschrift Doktorand

2. Inhaltsverzeichnis

3. Abkürzungsverzeichnis

SARS-CoV-2 – severe acute respiratory syndrome coronavirus 2

LuTx – lung transplantation

LTR – lung transplant recipient(s)

MLTG – Munich lung transplant group

COPD – chronic obstructive pulmonary disease

ILD – interstitial lung disease

CF – cystic fibrosis

PAH – pulmonary arterial hypertension

CLAD – chronic lung allograft dysfunction

BOS – bronchiolitis obliterans syndrome

NK – natural killer (cells)

APC – antigen presenting cells

MHC – major histocompatibility complex

TCR – T-cell receptor

BCR – B-cell receptor

NF-kB – nuclear factor kappa B

IL – interleukin

ATG – antithymocyte globulin

NFAT – nuclear factor of activated T-cells

TNFα – tumor necrosis factor alpha

IFN-γ – interferon gamma

MMF – Mycophenolate mofetil

DNA – deoxyribonucleic Acid

IMPDH – inosine-monophosphate-dehydrogenase

DC – dendritic cells

RV – respiratory virus

WHO – World Health Organization

ARDS – acute respiratory distress syndrome

VOC – variant of concern

SOT – solid organ transplantation

ACR – acute cellular rejection

AMR – antibody mediated rejection

4. Publikationsliste

Glueck OM, Ehrl D, Hatz RA, Fertmann JM.

Mind the gap! Interdisciplinary approach to anterior chest wall reconstruction after total sternectomy.

J Cardiothorac Surg. 2024 Apr 30;19(1):269. doi: 10.1186/s13019-024-02743-6. PMID 38689350

O. M. Glueck, J.Kovacs, S. Corradini, J. M. Fertmann, W. G. Sienel, T. Kauke, R. Hatz, S. Michel, M. Irlbeck, N. Kneidinger, C. Schneider

Brachytherapy Treatment for Bronchial Anastomosis Narrowing After Invasive Aspergillosis

Posterpräsentation, 43. Jahreskongress International Society for Heart and Lung Transplantation, 2023 Denver, USA

Glueck OM, Fertmann JM, Hatz R

Großflächige rigide Thoraxwandstabilisierung und Lungenfunktion

Vortrag, 62. Jahreskongress Deutsche Gesellschaft für Pneumologie 2023, Düsseldorf

Yavuz G, Heck S, Sienel W, Irlbeck M, Kneidinger N, Michel S, Forbrig R, Walter J, Zimmermann J, Kovács J, **Glück OM**, Pan M, Schneider C, Fertmann JM, Hatz RA, Kauke T.

Posterior reversible encephalopathy syndrome after lung transplantation: Risk factors and management.

Clin Transplant. 2023 Jan;37(1):e14850. doi: 10.1111/ctr.14850. Epub 2022 Dec 7. PMID: 36398875.

Zimmermann J, **Glueck OM**, Fertmann JM, Sienel WG, Yavuz G, Damirov F, Kovács JR, Tufman A, Irlbeck M, Kneidinger N, Michel S, Kauke T, Hatz RA, Schneider CP

COVID-19 in Recent Lung Transplant Recipients: Clinical Outcomes and Management Strategies.

Transplant Proc. 2022 Jul-Aug;54(6):1504-1516. doi: 10.1016/j.transproceed.2021.12.014

Glück O, Fertmann JM, Zimmermann J, Hatz R

Penetrating chest trauma by fall on large-diameter umbrella stand - a case report and analysis of institutional traumatic hemothorax experience

Vortrag, Jahreskongress Deutsche Gesellschaft für Chirurgie 2021

Haider T, Simader E**, Glück O**, Ankersmit HJ, Heinz T, Hajdu S, Negrin LL

Systemic release of heat-shock protein 27 and 70 following severe trauma

Sci Rep. 2019 Jul 3;9(1):9595. doi: 10.1038/s41598-019-46034-w.

Glueck OM: Current Cases, Lung Transplant Coordination, LAS

Vortrag auf der Vienna Lung Transplant Academy, Nov. 2017

Benazzo A, Schwarz S, **Glueck O**, Schweiger T, Hoetzenecker K, Moser B, Matilla Sigüenza J, Lang G, Taghavi S, Klepetko W

Pediatric Retransplantation: a single center experience

Posterpräsentation, 25. ESTS Meeting, 2017, Innsbruck, Österreich

Lang C, Hrdliczka E, Schweiger T, **Glueck O**, Lewik G, Schwarz S, Benazzo A, Lang G, Klepetko W, Hoetzenecker K.

Impact of cyclooxygenase-2 and prostaglandin-E2 expression on clinical outcome after pulmonary metastasectomy.

J Thorac Dis. 2017 Mar;9(3):621-635. doi: 10.21037/jtd.2017.02.83.

Schweiger T, Berghoff AS, Glogner C, **Glueck O**, Rajky O, Traxler D, Birner P, Preusser M, Klepetko W, Hoetzenecker K.

Tumor-infiltrating lymphocyte subsets and tertiary lymphoid structures in pulmonary metastases from colorectal cancer.

Clin Exp Metastasis. 2016 Oct;33(7):727-39. doi: 10.1007/s10585-016-9813-y. Epub 2016 Jul 23

Schweiger T, Starkl V, Glogner C, **Glueck O**, Jedamzik J, Birner P, Klepetko W, Hoetzenecker K

Clinical Impact of c-met expression and mutational status in patients with primary colorectal cancer lung metastases

Eur J Cardiothorac Surg. 2016 Apr;49(4):1103-11; discussion 1111. doi: 10.1093/ejcts/ezv323. Epub 2015 Oct 24.

Schweiger T, Nikolowsky C, Graeter T, Seebacher G, Laufer J, **Glueck O**, Glogner C, Birner P, Lang G, Klepetko W, Ankersmit HJ, Hoetzenecker K

Increased lymphangiogenesis in lung metastases from colorectal cancer is associated with early lymph node recurrence and decreased overall survival

Clin Exp Metastasis. 2016 Feb; 33(2):133-41. doi

Ghanim B, Schweiger T, Jedamzik J, **Glueck O**, Glogner C, Lang G, Klepetko W, Hoetzenecker K.

Elevated inflammatory parameters and inflammation scores are associated with poor prognosis in patients undergoing pulmonary metastasectomy for colorectal cancer

Interact Cardiovasc Thorac Surg. 2015 Nov;21(5):616-23. doi: 10.1093/icvts/ivv206. Epub 2015 Aug 4.

O. Glueck, C. Glogner, C. Nikolowsky, T. Schweiger, G. Lang, W. Klepetko, K. Hoetzenecker

5-year-results of a prospective, single-center Lung Metastasis registry

Posterpräsentation; Jahreskongress österreichische Gesellschaft für Pneumologie 2015, Graz Österreich

5. Ihr Beitrag zu den Veröffentlichungen

5.1 Beitrag zu Paper I

Paper No. I: "Impaired immune responses and prolonged viral replication in lung allograft recipients infected with SARS-CoV-2 in the early phase after transplantation", was conceived by me and set up in depth together with PD Dr. M. Muenchhoff from the Pettenkofer Institute of Virology. I was primarily responsible for recruiting the participating patients into the study and I provided the follow-up in clinical as well as scientific matter during the course of the study. I was significantly involved in clinical care and solely responsible for continuing sample acquisition of the patient cohort. In addition, I obtained ethical clearance through application at the CorKUM cluster after existing vote on ethical safety within the CorKUM cluster. Furthermore, I co-designed the composition of the paper and wrote the introduction, as well as the sections on clinical management and demographics alongside large parts of the discussion. Forth following, I was primarily responsible for the preparation and editing of the final manuscript version and subsequent submission of the paper. Accordingly, I also incorporated the required major revisions, revised the paper in depth and was ultimately responsible for resubmitting and acquiring the final acceptance for publication of the paper.

6. Introduction

Lung transplantation (LuTx) has been demonstrated over the last decades to be a safe treatment option in end stage lung disease in selected patients. Through thorough patient selection alongside advanced immunosuppressive therapy and monitoring, survival has continuously been improved. On the other hand, significant immunosuppression leaves lung allograft recipients vulnerable to various diseases, in lung transplantation recipients (LTR) especially of airborne origin. Therefore, risking significantly increased morbidity and mortality after infection. Since the Munich Lung Transplant Group (MLTG) represents one of the largest lung transplant centers in Europe, we naturally struggled with the rapid development and pathogenicity of severe acute respiratory syndrome coronavirus 2 (Sars-CoV 2). It did not only interfere with already severe organ shortage, but also put our allograft recipients at grave risk for increased morbidity and mortality before the wide availability and efficacy of vaccinations and disease-specific therapy. In the beginning of the pandemic, we had to rely on rudimentary preventive measures like isolation and face masks alongside mostly empiric treatment strategies. As a result, we delved deeper into the topic of immune response in LTR with high-dose immunosuppression early after transplantation and with Sars-CoV 2 infection. Due to the volatility of the disease, various different courses of immune response and of disease severity were observed.

6.1 Current practice in LuTx

Until present, more than 70.000 worldwide adult LuTx have been reported, of which bilateral LuTx account for approximately 80%. (1) Main indications for LuTx are Chronic obstructive pulmonary disease (COPD), Interstitial lung disease (ILD) or fibrosis, Cystic fibrosis (CF), Non-cystic fibrosis bronchiectasis, Pulmonary arterial hypertension (PAH) and others. (2) The overall median survival after LuTx ranges around 6-7 years, with an increase to around 9 years for recipients that survive the first year post-transplantation. (1, 3-5)

Long-term survival is mostly limited by the development of chronic lung allograft dysfunction (CLAD). In most cases a form of airway fibrosis, bronchiolitis obliterans syndrome (BOS) develops. (6-8) CLAD is diagnosed by a decrease in clinical status and loss of lung capacity displayed in spirometry. (9, 10)

By international standards, LTR receive combined triple immunosuppressive therapy with a calcineurin inhibitor (Tacrolimus, previously cyclosporine), corticosteroids (prednisone) and antimetabolites (mycophenolate-mofetile, previously azathioprine) (see chapter 6.2.2). Around 80% of LTR receive similar therapy regimens globally. (11)

6.2 Immunology in lung transplantation

6.2.1 Immunological basics

The first rapid immune response is carried out by innate immune system effectors shortly following pathogen recognition. This recognition and the consecutive response are triggered by structures of the invading pathogen, like bacterial cell wall components or viral nucleic acid patterns. Corresponding to recognizable patterns from exogenous threats, there are patterns for endogenous threats. They can be found after cell stress and/or death. (12) Their recognition triggers not only direct phagocytic activity of effector cells, but also a release of cytokines and chemokines, enhancing immediate immune response as well as inducing the adaptive immune system. (12) The main effector cells in innate immune response are neutrophils (granulocytes), monocytes, macrophages, and natural killer (NK) cells. They attack the intruding pathogen via phagocytosis or apoptosis induction as well as activation of the adaptive immune system by presenting antigens through Antigen-presenting cells (APC) to the adaptive effector cells, that are T- and B-Lymphocytes. (13)

The adaptive immune response works through allorecognition, antigen detection and presentation via antigen presenting cells through the major histocompatibility complexes I and II (MHC). This results in T-cell (mainly CD4+ and CD8+ lymphocyte) and B-cell activation and

consecutive cell or pathogen degradation through different pathways and effectors, alongside antibody-mediated immune response. (14, 15)

The best-known receptor cascade in T-cell activation follows the trigger of T-cell receptors (TCR). Here, an interaction between innate and adaptive immune system comes into play, as peptides bound to the MHC complex and presented by APC are recognized by the TCR alongside co-stimulatory signals. (16) This triggers several secondary intracellular messaging cascades, resulting in transcriptional activation in the nucleus, thus triggering measurable immune response through lymphocyte activation (see figure 1).

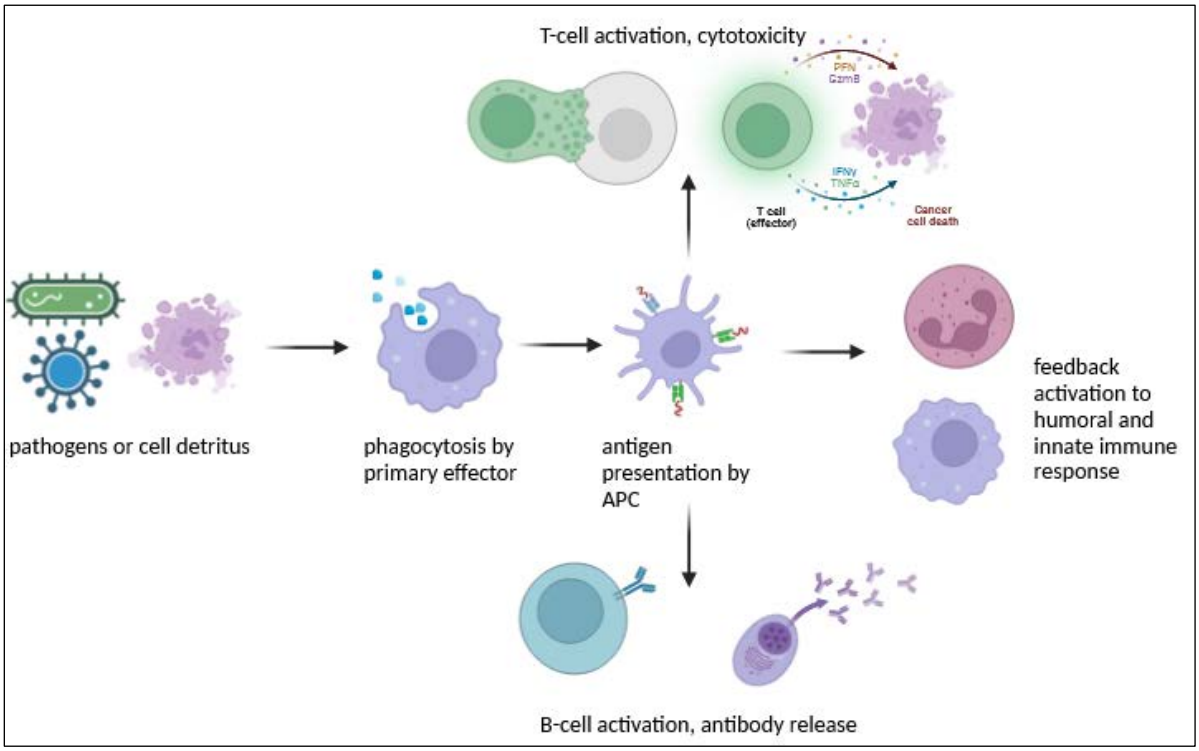


Figure 1: activation of adaptive immune system. See effector cells T-cell activation and B-cell activation, measurable as described in the publication. (Figure created with BioRender.com)

T-Lymphocyte activation is followed by clonal expansion of antigen-specific differentiated effector T-cells. The activation of CD4+ T-cell subsets results in production of effector cytokines like interferons or different interleukins (like interleukin 2, IL2), that can trigger further immune response through activation of other immune cells (B-lymphocytes) or through

feedback loops to the humoral immune response. CD8+ T-cells execute cell death through cytotoxicity. (17, 18)

The other main effector in cellular to humoral immune response is the B-lymphocyte. After activation through B-cell receptors (BCR), the B-lymphocyte can present the antigen through MHC II. (12) For full function, costimulation by activated CD4+ T-cells is needed, resulting in clonal expansion and differentiation of the activated B-cells. The most important effector of activated B-cells (effector = plasma cell) is immunoglobulins like IgM or IgG, directly impacting the immune response through agglutination, activation of complement cascades or opsonization (marking) of pathogens.(12, 19)

6.2.2 Immune response and immunosuppression in LuTx

In case of lung transplantation or transplantation in general, the immune system works through various pathways to “defend” the recipient against the newly implanted organ.

The innate immune system can recognize necrotic cellular fragments, for example from cell necrosis through stress after reperfusion injury. Following in short, there is intracellular activation of different signaling molecules (e.g. nuclear factor kappa B; NF-kB), which induce cytokine release and effector cell activation. (20) The result is inflammasome formation and activation of interleukins (IL1ß, IL18). This leads to apoptosis, and induces a cell death cascade through multiplication of fragments from depleted allograft cells. (21)

On the level of cellular immune response, the pathways are as described above. Following the presentation of antigens like “foreign” cellular fragments, activation of the T- and B-lymphocyte pathways ensues, resulting in measurable clonal expansion of effector T-cells or immunoglobulin production.

Therefore, the immunosuppressive treatment focuses on different levels of the recipient’s immune reaction. In the following, the most common combination of immunosuppressive drugs and their interaction with immune response will be discussed. Some centers use induction immunosuppressive therapy, aiming to minimalize T-lymphocyte activation before LuTx. There

are different agents in use today. The most commonly used are anti-thymocyte globulin (ATG), Alemtuzumab and Basiliximab. (1, 22) ATG targets T-lymphocyte surface molecules, leading to depletion through apoptosis. (23) Alemtuzumab targets CD52, a surface protein not only on T- and B-lymphocytes but also in other mononuclear immune system effectors, leading to depletion of these cells. (23, 24) Basiliximab is an antibody against interleukin-2 receptor on T-lymphocytes and therefore inhibits intercellular signaling and activity, while not depleting the cells. (23, 25)

The most commonly used drugs for maintenance immunosuppression fall into three categories. (11, 26) Most centers typically use a triple combination of the therapeutics described below, even though there is no standard protocol. (27)

One cornerstone is calcineurin inhibitors like Cyclosporine or Tacrolimus. Both of the above predominantly target T-lymphocytes. They inhibit the intracellular dephosphorylation process of NFAT (nuclear factor of activated T-cells) through blockade of calcineurin formation, resulting in non-translocation of this factor into the nucleus and the arrest of production of pro-inflammatory agents like interleukins (mainly interleukin 2), tumor necrosis factor alpha (TNFα) or interferon gamma (IFN-γ). In short, this leads to inhibition of further activation and inhibited function of already activated T-cells. (23, 25, 26)

The second cornerstone is antimetabolites. Mostly used is Mycophenolic acid in its formulation Mycophenolate mofetil (MMF), Azathioprine has no significance in our center's maintenance immunosuppression. MMF works through interference with DNA synthesis by inhibition of Inosine-Monophosphate-Dehydrogenase (IMPDH) and hence blocks de-novo guanine synthesis. This is also lymphocyte-specific and reduces clonal expansion as well as function of activated lymphocytes. Furthermore, it limits migration of monocytes and antigen presentation by dendritic cells (DC). (23, 25, 26, 28)

The third cornerstone is Corticosteroids. Most centers use prednisone for maintenance immunosuppression. Acute rejection is treated by high-dose corticosteroid shock therapy. Steroids affect all immune cells. (29) They bind to intracellular receptors that are thereby

activated and migrate into the nucleus. There, they activate the transcription of glucocorticoid-sensitive genes, but also transcription factors like NF-κB. As a result, immune effector molecules such as interleukins, TNFα and IFN-γ are reduced. Thereby preventing further activation of lymphocytes and inducing apoptosis. Also, corticosteroids inhibit the production of other inflammatory molecules like prostaglandins. (25-27, 29)

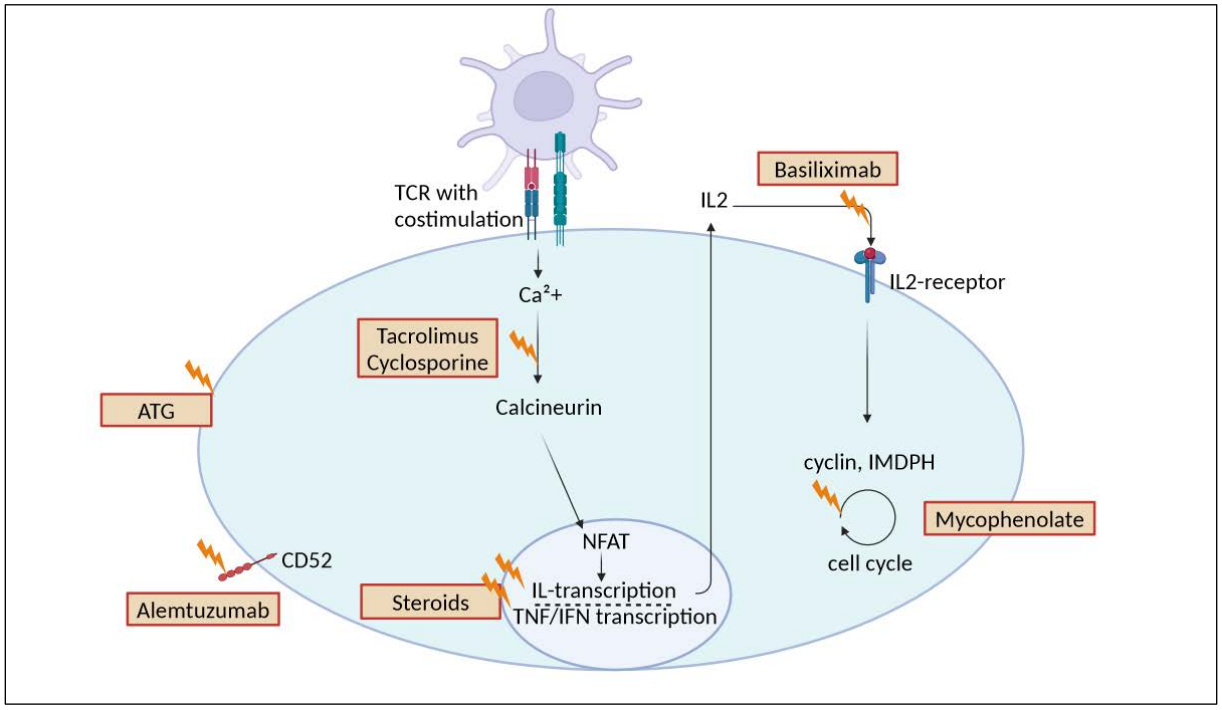


Figure 2: Immunosuppressive pathways (cellular level) used in solid organ transplantation. Adapted from (23), created with BioRender.com

6.2.3 Implications of immunosuppression in LuTx

Even though use of advanced immunosuppressive agents has reduced graft failure in solid organ transplantation, it also puts the recipients at elevated risk for infectious diseases. Due to the need for high levels of immunosuppression and therefore intensely reduced immune competence in the early phase (< 12 months) after transplantation, the risk for bacterial, fungal or viral infection is particularly elevated. (30-32) This is amplified by comorbidities such as cardiac, renal or metabolic disease as well as reduced strength and mobility that are often present in patients requiring solid organ transplantation. In heart and lung transplantations,

infections are the leading factor in mortality in the first year post-transplantation. (33, 34) As the respiratory tract is specifically exposed to airborne pathogens, the risk of respiratory infection is high across all solid organ transplantations particularly with respiratory viruses (RV). (35-38) This leads to very high susceptibility for airborne viral infection of the respiratory system in LTR, as they often present with a combination of the previously mentioned risk factors. (39-43) As a conclusion, one must assume a high susceptibility also for Sars-CoV 2 in lung transplant recipients, especially in the early phase after transplantation.

6.3 Sars-CoV 2 – effects of an airborne pandemic on LuTx

6.3.1 Sars-CoV 2 – a short overview

Following the first reports of a novel viral infectious disease with impact on the pulmonary tract from China in December 2019, Sars-CoV 2 was declared a pandemic in March 2020. (44-46) After multiple waves of infections along changing virus variants, around 773 million cases have been reported, with more than 6.99 million deaths associated with the disease as of December 2023. (47) On May 5th 2023, the WHO declared that Covid-19 was no longer a “public health emergency of international concern”. (48, 49)

SARS-CoV 2 is transmitted human-to-human through fluids, aerosols and contaminated surfaces. (50, 51) Infection with SARS-CoV 2 can exhibit various manifestations, and while most patients develop only mild symptoms or stay asymptomatic, severe cases with need for intensive care and respirator can be associated with a high case-fatality rate. (52-54) Common symptoms reported from the beginning of the pandemic are fever, fatigue and of respiratory nature like dyspnea and dry cough. (52, 55-57) Accompanying clusters of gastrointestinal symptoms like nausea or diarrhea have also been described. (56-58) The severity of symptoms and course of the disease with development of ARDS (acute respiratory distress syndrome) and consecutive high fatality rates seem to be closely linked to older age and comorbidities of cardiovascular or pulmonary nature or obesity. (59-62) Through mutation, Sars-CoV 2 has developed different variants of concern (VOC) like Alpha, Beta, Gamma, Delta, and lately

Omicron among others. (62-64) Since early 2022, Omicron is the dominating variant with changing subvariants. (64-66) Therapeutics evolved from non-standardized treatment consensus. They range from high dose corticosteroids (in order to dampen “cytokine storm”-overreaction of the immune system) antivirals like remdesivir and antibody treatments by reconvalescent plasma infusion (plasma with high levels of immunoglobulins against Sars-CoV 2) to recombinant antibodies like Bamlanivimab. (67-71) Current treatment options furthermore include molecular targeting drugs in combination with further antiviral therapeutics. (71) Fortunately, efficient vaccinations have been available from late 2020, and a great number of the population is now immunized either by vaccination, prior infection or both. (72-74)

6.3.2 Sars-CoV 2 in solid organ transplantation - focus on LuTx

The emergence and quick global spread of Sars-CoV 2 had a significant impact on solid organ transplantation (SOT). Reports describe increased infection rates, hospitalization, and mortality in all SOT recipients, regardless of transplanted organ. (75-78) These reports mostly described patients under maintenance immunosuppression long-term post-transplantation. (75) SOT recipients usually present with multiple comorbidities that have been described as unfavorable for Sars-CoV 2 outcome, an additional increase in risk is therefore to be expected from the combination with high-dose immunosuppression. (75, 79, 80) Interestingly, later reports have shown increased risk for infection with, but similar mortality rates from Sars-CoV 2 in SOT recipients compared to general population. (77, 81)

According to medical-logical considerations, LTR should be particularly at risk for a severe course after infection with Sars-CoV 2, as they have been transplanted the target organ of the respiratory virus.

Most groups report on patients under maintenance immunosuppression, while reports on early infection after LuTx are scarce. (82-86) From an immunological view, these cases are considerably more interesting. LTR receive significantly higher doses of triple immunosuppression in the early phase post-Tx in comparison to non-LTR, leading to the assumption of an even higher degree of non-functionality of the immune system. Due to high-

dose immunosuppressive therapy, initial symptoms for infection can be mild, masked or mistaken for other causes. This may be related to reduced lymphocyte activity, antibody and cytokine production, and hence lower disease severity, which was found to be triggered by a strong immunological reaction or “hyperinflammation”. (87-90)

Overall, LTR have increased risk for hospitalization with Sars-CoV 2 compared to non-LTR. (91) LTR are also at elevated risk for fungal co-infection and present overall higher risk for severe course as well as higher mortality rates when infected with Sars-CoV 2. (80, 91, 92) When infected, LTR present with similar symptoms as immunocompetent patients. These are flu-like symptoms such as cough, fever or dyspnea which can culminate in pneumonia with increased supplemental oxygen requirements and ultimately the need for mechanical ventilation. (86, 93)

Most reports show the management of Sars-CoV 2 infection in LTR as a combination of means. Isolation was the most effective preventive means before vaccinations. (83) Today, vaccination regimes have taken hold and have shown effectiveness through risk reduction for severe course of Sars-CoV 2 in LTR as well as other SOT recipients. (92) As expected, SOT recipients mostly show inadequate seroconversion after standard vaccination doses. (94, 95) Therefore, usually multiple doses and early boosters are recommended and applied in SOT. (96, 97) Lately, LTR with inadequate seroconversion optionally receive pre-exposure prophylaxis with neutralizing monoclonal antibodies (tixagevimab-cilgavimab) with mixed results, probably from waning effects over time. (98, 99) For therapy, there is reduction in immunosuppression, mostly of mycophenolate, (83, 86, 87) intended to allow lymphocyte function for antiviral defense. This is usually combined with the currently available therapy, as described above. Interestingly, there has no difference been reported in lung allograft rejection, neither of acute cellular rejection (ACR), antibody mediated rejection (AMR) or CLAD in LTR with or without Sars-CoV 2 infection. (86, 87)

7. Zusammenfassung:

Nach einer langjährigen Entwicklung mit späten Erfolgen beim Verständnis der immunologischen Hintergründe der Abstoßung hat sich die Lungentransplantation (LuTx) heute als wirksame Therapie für Lungenkrankheiten im Endstadium etabliert, und die weltweit steigenden Zahlen bestätigen den Therapieerfolg. Dennoch ist eine hochdosierte Immunsuppression erforderlich, um Abstoßungsreaktionen zu verhindern. Durch verschiedene Immunsuppressiva werden viele Ebenen der Immunantwort deutlich reduziert oder blockiert. Effektorzellen wie T- oder B-Lymphozyten können dann nicht mehr angemessen auf Immunauslöser wie die Präsentation von Krankheitserregern, geschädigten Zellen oder Molekülen reagieren. Durch die Verhinderung der Abstoßung besteht daher bei Lungentransplantat-Empfängern eine erhöhte Anfälligkeit für Infektionen, insbesondere durch aerogene oder respiratorische Erreger. Dies ist besonders wichtig, da Infektionen für eine erhebliche Einschränkung der Überlebensrate im ersten Jahr nach LuTx verantwortlich sind. Das Auftreten des severe acute respiratory syndrom coronavirus 2 (Sars-CoV 2) hat die weltweite Praxis der Lungentransplantation erheblich beeinträchtigt. Sars-CoV 2 ist ein hoch ansteckender, über die Luft übertragener Erreger, der in erster Linie die Atemwege befällt und sich schnell zu einer Pandemie mit ständig wechselnden Varianten entwickelt hat. Wenn sie infiziert sind, zeigen LTR die gleichen Symptome wie nicht-transplantierte Patienten. Da der Schweregrad der Erkrankung jedoch durch eine akute Hyperinflammation beschrieben wurde, treten die schweren Symptome bei LTR später auf und können anfangs maskiert oder mit anderen Atemwegserkrankungen verwechselt werden. Daher ist davon auszugehen, dass das Immunsystem von LTR nach einer Infektion mit Sars-CoV 2 keine ausreichende Reaktion in Form von Antikörpern oder T-Zell-Reaktivität hervorbringt. In der vorliegenden Publikation untersuchten wir die Wirkung einer hochdosierten Immunsuppression bei Patienten im Frühstadium nach LuTx und die Auswirkung auf die Immunantwort nach einer Sars-CoV 2-Infektion durch Messung der Anti-Sars-CoV 2-Antikörperbildung sowie der T-Zell-Aktivierung und -Reaktivität im Vergleich zu nicht transplantierten, nicht-immunsupprimierten Kontrollen.

8. Abstract (English):

Following long term development with late success in understanding the immunological background of rejection, lung transplantation (LuTx) is today established as effective therapy for end-stage lung diseases and growing worldwide numbers confirm the feat of the method. Nevertheless, high doses of immunosuppression are needed to prevent rejection. Through different immunosuppressive agents, many levels of the immune response are significantly reduced or blocked. Effector cells like T- or B-lymphocytes can then no longer provide adequate responses to immune triggers like presentation of pathogens, damaged cells or molecules. Therefore, through prevention of rejection, there is increased vulnerability for infections, in lung transplant recipients (LTR) particularly of airborne or respiratory pathogens. This is especially important, as infection is responsible for significant decrease of first-year survival in LTR. As a result, the emergence of severe acute respiratory syndrome coronavirus 2 (Sars-CoV 2) significantly impacted the worldwide practice of lung transplantation. A highly contagious airborne pathogen, primarily affecting the respiratory system, Sars-CoV 2 quickly evolved to a pandemic with ever changing variants. When infected, LTR showed the same symptoms as non-transplanted patients. But since disease severity was described through acute hyperinflammation, severe symptoms in LTR emerge later and in the beginning can be masked or mistaken for other respiratory diseases. Therefore one has to assume that LTRs immune system will not produce sufficient response in form of antibodies or T-cell reactivity following infection with Sars-CoV 2. In the presented publication, we investigated the effect of high-dose immunosuppression in patients early after LuTx and the effect on immune response following Sars-CoV 2 infection through measurement of anti-Sars-CoV 2 antibody formation as well as T-cell activation and reactivity in comparison to non-transplant, non-immunocompromised controls.

9. Paper I

The publication “Impaired immune responses and prolonged viral replication in lung allograft recipients infected with SARS-CoV-2 in the early phase after transplantation” can be found embedded in the following pages.

Infection (2024) 52:847–855
https://doi.org/10.1007/s15010-023-02116-6

RESEARCH

Impaired immune responses and prolonged viral replication in lung allograft recipients infected with SARS-CoV-2 in the early phase after transplantation

Olaf M. Glueck[1] · Xiaoling Liang[2] · Irina Badell[2] · Paul R. Wratil[2,3] · Alexander Graf[4] · Stefan Krebs[4] · Helmut Blum[4] · Johannes C. Hellmuth[5] · Clemens Scherer[6] · Alexandra Hollaus[5,7] · Patricia M. Spaeth[2] · Burak Karakoc[2] · Thimo Fuchs[2] · Julia Zimmermann[1] · Teresa Kauke[1] · Andreas Moosmann[5,3,7,8] · Oliver T. Keppler[2,3] · Christian Schneider[1] · Maximilian Muenchhoff[2,3]

Received: 26 July 2023 / Accepted: 12 October 2023 / Published online: 3 November 2023

Abstract

Purpose Lung transplant recipients are at increased risk of severe disease following infection with severe acute respiratory syndrome coronavirus type 2 (SARS-CoV-2) due to high-dose immunosuppressive drugs and the lung is the main organ affected by Coronavirus disease 2019 (COVID-19). Several studies have confirmed increased SARS-CoV-2-related mortality and morbidity in patients living with lung allografts; however, detailed immunological studies of patients with SARS-CoV-2 infection in the early phase following transplantation remain scarce.

Methods We investigated patients who were infected with SARS-CoV-2 in the early phase (18–103 days) after receiving double-lung allografts ($n=4$, LuTx) in comparison to immunocompetent patients who had not received solid organ transplants ($n=88$, noTx). We analyzed SARS-CoV-2-specific antibody responses against the SARS-CoV-2 spike and nucleocapsid proteins using enzyme-linked immunosorbent assays (ELISA), chemiluminescence immunoassays (CLIA), and immunoblot assays. T cell responses were investigated using Elispot assays.

Results One LuTx patient suffered from persistent infection with fatal outcome 122 days post-infection despite multiple interventions including remdesivir, convalescent plasma, and the monoclonal antibody bamlanivimab. Two patients experienced clinically mild disease with prolonged viral shedding (47 and 79 days), and one patient remained asymptomatic. Antibody and T cell responses were significantly reduced or undetectable in all LuTx patients compared to noTx patients.

Conclusion Patients in the early phase following lung allograft transplantation are vulnerable to infection with SARS-CoV-2 due to impaired immune responses. This patient population should be vaccinated before LuTx, protected from infection post–LuTx, and in case of infection treated generously with currently available interventions.

Keywords COVID-19 · Solid organ transplantation · Immune responses · Immunosuppression · Lung transplant recipients

✉ Maximilian Muenchhoff
muenchhoff@mvp.lmu.de

[1] Division of Thoracic Surgery, LMU University Hospital, LMU Munich, Munich, Germany

[2] Max Von Pettenkofer Institute and Gene Center, Virology, National Reference Center for Retroviruses, Ludwig Maximilian University of Munich, Pettenkoferstr. 9a, 80336 Munich, Germany

[3] German Center for Infection Research (DZIF), Partner Site, Munich, Germany

[4] Laboratory for Functional Genome Analysis, Gene Center, Ludwig Maximilian University of Munich, Munich, Germany

[5] Department of Medicine III, LMU University Hospital, LMU Munich, Munich, Germany

[6] Department of Medicine I, LMU University Hospital, LMU Munich, Munich, Germany

[7] Helmholtz Munich, Munich, Germany

[8] German Cancer Consortium (DKTK), Partner Site Munich, Munich, Germany

Introduction

Transplant recipients (TRs) have a higher risk of contracting infectious diseases due to the use of immunosuppressive agents during the early phase after transplant (< 12 months) [1, 2]. In fact, infections are the leading cause for increased mortality in the first year post-transplantation [3–6]. Bacterial and viral pneumonia are common in Lung Transplant Recipients (LTRs) since the allograft is directly exposed to the pathogen. It is therefore important to find the suitable balance between immunosuppressive rejection prophylaxis and preservation of protective immune responses. While total cell counts for CD4 and CD8 T cells in LTRs seem to remain stable, their function measured as cytokine secretion is significantly impaired [7, 8]. Conversely, humoral immunity is impaired with lower IgG titers especially in the early phase post-solid organ transplantation (SOT) [9, 10]. This is confirmed in recent studies of immune responses to SARS-CoV-2 vaccination, even after a longer time post-transplantation and especially for infection with later variants of concern (VOCs) like omicron [11–13].

LTRs therefore have a high susceptibility for airborne viral infections of the respiratory system such as SARS-CoV-2 [14]. Several studies have confirmed increased morbidity and mortality of LTRs due to COVID-19 in comparison to the general population [13, 15–18]. Risk factors for severe disease include advanced age, male sex, impaired kidney function, and time point of infection following SOT with patients infected at a later time following SOT showing fewer clinical effects than patients with early-onset infections.

Since the outbreak of the COVID-19 pandemic, only few cases of early-onset infection with SARS-CoV-2 have been described in LTRs as clinical case reports without further investigation of their antiviral immune responses. The aim of this study is therefore to investigate the clinical course and SARS-CoV-2-specific immune responses of four LTRs with early-onset infection (LuTx) in comparison to COVID-19 patients who have not received an allograft (noTx).

Methods

Study design and subjects

This study is a retrospective monocenter case–control analysis of early-phase lung transplant recipients (LuTx) and non-transplant patients with SARS-CoV-2 infection (noTx). From the day of the WHO declaration classifying COVID-19 as a global pandemic on March 16, 2020 until March 3, 2021, 87 patients underwent a lung transplant in the Munich Lung Transplant Group (MLTG). Infection with SARS-CoV-2 was detected in six of these patients in the early phase after transplant (< 6 months, 22–103 days). Four of these patients, subsequently referred to as LuTx A-D, gave written informed consent for participation in this study.

Patients were recruited in the COVID-19 Registry of the LMU University Hospital Munich (CORKUM, WHO trial ID DRKS00021225). Patient data were anonymized for analysis, and this study was approved by the local ethics committee (Institutional Review Board) (No: 20-245).

For comparison of humoral and cellular immune responses, we selected 88 non-transplant patients (noTx) without documented medical conditions associated with significant immunodeficiency. These patients were part of the CORKUM study and cryopreserved samples were used retrospectively for analysis.

SARS-CoV-2 RNA detection and quantification

Nasopharyngeal swab samples (ESwab, Copan Diagnostics, Murrieta, USA) were collected twice weekly for patients on the normal ward and transported to the accredited routine diagnostics laboratory of the Max von Pettenkofer Institute. PCR tests were performed using the Roche Cobas SARS-CoV-2 assays on the Cobas 6800 system. Viral load results were calculated as copies per ml of transport medium for the E-gene reaction as described previously [19].

SARS-CoV-2 whole genome sequencing

Amplicon pools covering the SARS-CoV-2 genome were prepared according to the ARTIC network nCoV-2019 sequencing protocol v2 and analyzed utilizing the ARTIC bioinformatics protocol as described previously [20]. The consensus sequences and associated sample metadata were uploaded to the GISAID repository.

Antibody detection assays

The commercial recomLine SARS-CoV-2 IgG line immunoassay (Mikrogen, Neuried, Germany) was used to analyze IgG antibodies against the SARS-CoV-2 spike receptor binding domain (RBD) and nucleocapsid (N). Quantitative results were obtained by analyzing test strips with the recomScan software. According to the manufacturer's guidelines, the "fold cut-off" value was determined by subtracting the signal of interest with that of the internal cut-off band. IgG antibodies against the spike S1 subunit were quantified using the commercial ELISA by Euroimmun (Lübeck, Germany). Nucleocapsid-specific IgG was analyzed using the

Abbott SARS-CoV-2 IgG assay (Abbott Diagnostics, Abbott Park, Illinois, United States). All these tests were performed following the manufacturer's instructions on cryopreserved serum samples collected at the indicated time points.

ELISPOT analysis

IFN-gamma ELISPOT assays were performed with cryopreserved patients' PBMCs according to the manufacturer's recommendations (Mabtech, Nacka, Sweden; Bio-Rad, Puchheim, Germany). Frozen PBMCs were thawed and incubated at 2.5×10^5 cells/well with SARS-CoV-2 peptide pools (Wuhan-Hu-1 PepMix, JPT, Berlin, Germany), consisting of 15mer peptides with 11 amino acid overlap, at a final concentration of 0.5 µg/ml per peptide for 14–18 h. A stimulation cocktail of phorbol 12-myristate 13-acetate (PMA) and ionomycin (0.5X, Thermo Fisher Scientific, Waltham, United States) was used as positive control. Conditions without peptide stimulation serve as negative control and were subtracted from the sample values. Due to limited sample availability, each condition was tested in a single reaction.

Statistical analysis

Statistical analyses were performed using GraphPad Prism version 9.5 software. Groupwise comparisons were done using Mann–Whitney test. For summary visualization of serological results of noTx patients, locally estimated scatterplot smoothing (LOESS) was performed using the ggplot package in RStudio version 1.2.5033 with the geom_smooth function.

Results

Clinical course of SARS-CoV-2 infection in recent lung transplant recipients

We investigated four lung allograft recipients who experienced infection with SARS-CoV-2 in the early phase post-transplantation (18–103 days after transplantation). Clinical characteristics of these patients are summarized in Table 1 and described more detailed in supplementary information as well as in a previously published paper by Zimmermann et al. who investigated the clinical course of patient A and patient B of this study [18].

Remarkably, the spectrum of SARS-CoV-2-induced disease ranged from asymptomatic infection with rapid clearance (patient A) to persistent infection with ultimately fatal outcome (patient B). The clinical course and viral load results from respiratory samples in relation to the time point of transplantation are illustrated in Fig. 1a, b.

Patient A cleared the infection with negative PCR results from 31 DPI and never developed COVID-19-related symptoms.

Despite multiple therapeutic approaches including remdesivir, transfusion of convalescent plasma [three consecutive infusion regimens with 2×200 ml administration of COVID convalescent plasma (CCP)], and application of the monoclonal antibody bamlanivimab, patient B remained PCR positive. He developed pulmonary and gastrointestinal symptoms including diarrhea. He was transferred to the intensive care unit 95 days after infection. Forth following, he developed hepatopathy with subsequent liver failure alongside intermittent renal failure with renal replacement therapy for 7 days, while experiencing worsening lung affection showing consolidations matching viral pneumonia with bacterial superinfection. Respiratory failure required intubation and mechanical ventilation from day 102 to death. (Fig. 1b). Patient C presented with mild symptoms (elevated temperature but no fever, mild dyspnea with oxygen therapy, and only marginal infiltrates in chest CT). Due to extended therapy for cytomegalovirus (CMV) reactivation (see supplementary material), he developed leukopenia. As no viral clearance could be achieved, he received remdesivir therapy on DPI 72–78 and tested negative after 79 DPI. Patient D developed progredient bilateral pulmonary ground-glass lesions as well as mild disease symptoms. He also developed acute renal failure (max. creatinine levels 4.1 mg/dl, minimum GFR 14) without need for renal replacement therapy. As he experienced increasing symptoms (dyspnea, need for oxygen therapy, fatigue) alongside pulmonary affection in CT scans, he received dexamethasone alongside two therapy regimens of remdesivir (DPIs 2–6 and 28–32). He cleared the infection on 47 DPI.

All three survivors are being followed up closely by our LuTx program. They do not show signs of long COVID. All received at least three vaccinations against SARS-CoV-2 alongside tixagevimab/cilgavimab (Evusheld) for pre-exposure prophylaxis. None experienced another infection with SARS-CoV-2. The latest anti-SARS-CoV-2 S antibodies were 8.7 for patient A, 2.9 for patient C, and 8.5 for patient D (U/ml; IgG; Euroimmun; cut-off < 0.8).

Patient D developed CLAD type BOS [humorally triggered by donor-specific antibodies (DSAs)]. Therapy with intravenous immunoglobulins (IVIG) was not successful in clearing the DSA, so that he is currently under extracorporeal photopheresis therapy (ECP, currently 22 treatment regimens). Patients A and C did not develop CLAD.

Survival and graft function along with current treatment and further clinical data are also summarized in Table 1. No patient in our cohort underwent induction therapy prior to LuTx.

As mentioned above, in a previous detailed report by Zimmermann et al. [18] focusing on clinical course of

Table 1 Patient characteristics and follow-up

	Patient A	Patient B	Patient C	Patient D
General characteristics				
Age (y)	66	59	58	68
Gender	f	m	m	m
BMI (kg/m^2)	28	27,7	28	26
Smoking history (PY)	0	100	5	0
Survival	Yes	No	Yes	Yes
Survival (months post-LuTx)	33	4	34	36
Survival SARS-CoV-2 free (months)	32	NA	28	31
Vaccination against SARS-CoV-2	4x	NA	3x	4x
Pre-exposition prophylaxis	Evusheld 1x	NA	Evusheld 1x (booster scheduled)	Evusheld 1x
Underlying disease	EAA (Hypersensitivity pneumonitis)	UIP	ILD	IPF
Comorbidities	HP gastritis coronary atherosclerosis	Alcohol abuse Nicotine abuse	Arterial hypertension coronary atherosclerosis	Arterial hypertension atrial fibrillation
Type LuTx	double	double	double	double
Immunosuppression	Tacrolimus, mycophenolate mofetil, prednisolone	Tacrolimus, mycophenolate mofetil, prednisolone	Tacrolimus, mycophenolate mofetil, prednisolone	Tacrolimus, switch to cyclosporine A, mycophenolate mofetil, prednisolone
Graft function				
Humoral rejection (HLA-DSA)	NA	NA	NA	15.09.2020 A24, -B8, -Cw9 MFI 3800
Cellular rejection	NA	28.10.2021 A1, B0	NA	NA
Treatment of rejection	NA	high-dose corticosteroids	NA	IVIG ECP (ongoing)
Latest FEV1 (L/% of best)	1.78 (99)	NA	2.07 (100)	1.69 (84%)
Time after Tx to SARS-CoV-2-positive PCR (d)	18	22	94	103
COVID severity and mortality risk factors				
Renal function (minimal GFR during SARS-CoV-2)	20	12	39	14
(maximum serum creatinine mg/dl)	2.4	4.9	1.9	4.1
D-dimer (µg/ml)	0.6	2.9	1	0.5
Obesity (BMI > 25)	Yes	Yes	Yes	Yes
Diabetes	No	No	No	No
Hypertension	No	No	Yes	Yes

BMI body mass index, *y* years, *py* pack years, *UIP* unspecified interstitial pneumonia, *EAA* extrinsic allergic alveolitis/hypersensitivity pneumonitis, *ILD* interstitial lung disease, *IPF* idiopathic pulmonary fibrosis, *LuTx* lung transplantation, *FEV1* Forced expiratory volume in 1 s

disease, more in-depth information on the same patients has been described. This includes further data on comorbidities, transplant specifics like HLA typing, and overall clinical course of SARS-CoV-2 and post-transplantation period, but limited data on immune responses reporting SARS-CoV-2-specific antibodies without distinguishing between nucleocapsid- and spike-specific responses and quantity.

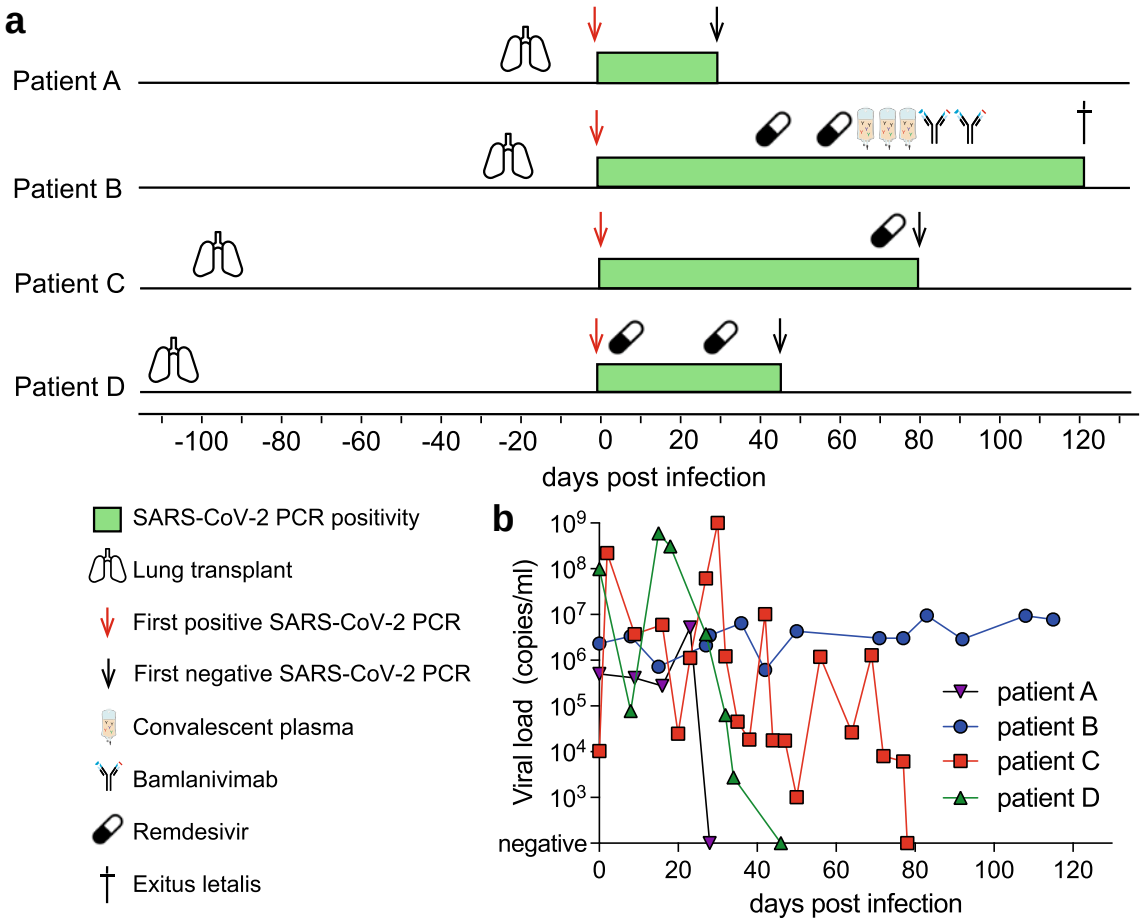


Fig. 1 **a** Clinical course of SARS-CoV-2 infection of recent lung transplant recipients. The timeline indicates the order of events in relation to the date of the first positive SARS-CoV-2 PCR result (day 0, red arrow). The symbols indicate the time points of administration of remdesivir (pills), convalescent plasma (infusion bag), and bamlanivimab (monoclonal antibody), respectively. The first negative SARS-CoV-2 PCR result (black arrow) is indicated. Patient B passed away after 121 days of infection (cross). **b** SARS-CoV-2 viral load trajectories of recent lung transplant recipients. Viral load is indicated as copy numbers of the SARS-CoV-2 ORF1ab gene per ml of transport medium of nasopharyngeal swab samples. Values are plotted in relation to the first positive PCR result (days of infection) for each of the four transplant recipients

In this study we aimed to decipher SARS-CoV-2-specific immune responses in these four LuTx cases with differential disease outcome and in comparison to noTx patients.

Limited antibody responses following SARS-CoV-2 infection in recent lung transplant recipients

To investigate the effect of iatrogenic immunosuppression in the early post-transplantation phase in lung allograft recipients on antibody responses upon SARS-CoV-2 infection, we measured IgG antibodies against spike and nucleocapsid antigens over time (Fig. 2). In comparison to the lung transplant recipients (LuTx), we tested 309 longitudinal samples from 88 patients without documented causes for immunosuppression from the local COVID-19 cohort, the COVID-19 registry of the LMU clinic (CORKUM) (noTx). Clinical characteristics of these control patients are summarized in the Supplementary Information (SI).

Compared to these control patients, LuTx showed generally lower levels of SARS-CoV-2-specific antibodies. Patient B had detectable IgG targeting the spike S1 subunit and receptor binding domain (Fig. 2a, b).

Of note, longitudinal sequence analysis of viral isolates from this patient revealed that the well-characterized antibody escape mutation at spike residue position 484 in the receptor binding domain (RBD) (E484K) had emerged as de novo mutation at 42 DPI. This may have resulted in reduced neutralizing efficacy of these autologous antibodies and the administered convalescent plasma and therapeutic monoclonal antibody, bamlanivimab, possibly contributing to viral persistence. Patient A who cleared the infection relatively quickly had detectable antibodies to the spike

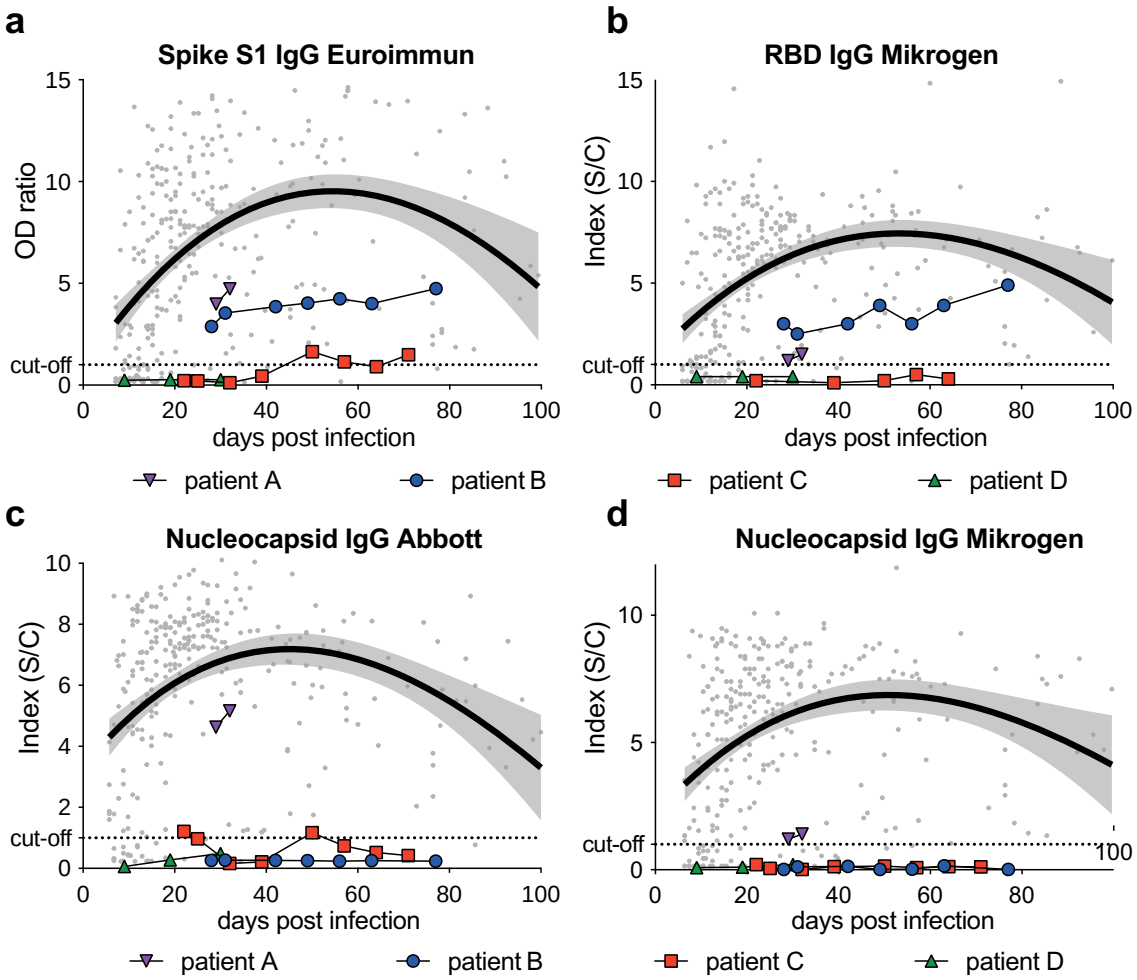


Fig. 2 Limited SARS-CoV-2-specific antibody responses in lung transplant recipients in comparison to non-immunocompromised patients. Serological results are shown for four commercially available assays for the detection of IgG antibodies against the SARS-CoV-2 spike S1 subunit (**a**), the receptor binding domain (RBD) (**b**) and nucleocapsid (**c**, **d**). Longitudinal results for the four lung transplant recipients (indicated in color) are shown in relation to the first positive SARS-CoV-2 PCR result. For comparison, longitudinal serological results of patients who have not received a solid organ transplant (noTx $n=88$) are plotted as gray dots with locally estimated scatterplot smoothing (LOESS) shown as black curve with 95% confidence interval. Results are indicated as optical density for the chemiluminescence assay (**a**), or signal to cut-off ratio for immunoblot assays (**b–d**). The dotted horizontal line represents the cut-off considered for test result positivity as provided by the manufacturer

and nucleocapsid antigens, whereas the other patients only showed negative or borderline reactivity.

Dampened T cell responses against SARS-CoV-2 in the early post-transplantation phase of lung allograft recipients

The immunosuppressive regimen used in the early phase post-transplantation consists of tacrolimus, mycophenolate mofetil, and prednisolone to prevent allograft rejection. To investigate the effect of this potent combination on T cell responses against SARS-CoV-2, we analyzed Elispot responses of PBMCs of the infected transplant recipients against peptide pools covering the S1 and S2 subunits of the SARS-CoV-2 spike protein and the nucleocapsid protein (N) of the Wuhan-Hu-1 reference strain. In comparison we tested samples of ten SARS-CoV-2-infected immunocompetent donors. Due to sample availability, we were only able to test the S1 antigen for all ten control patients and the S2 and N antigen for a subset of these ($n=8$ and $n=5$, respectively). Since the magnitude of T cell responses expands and contracts over time after antigen contact, we selected samples to match time after infection around 30 DPI (20–45 DPI for control patients). As expected, the magnitude of the T cell response in transplant recipients (LuTx) was lower compared to the control patients for all three antigens ($p=0.031$, $p=0.046$, and $p=0.039$ for S1, S2, and N, respectively) (Fig. 3a). For patients B and C we had longitudinal samples

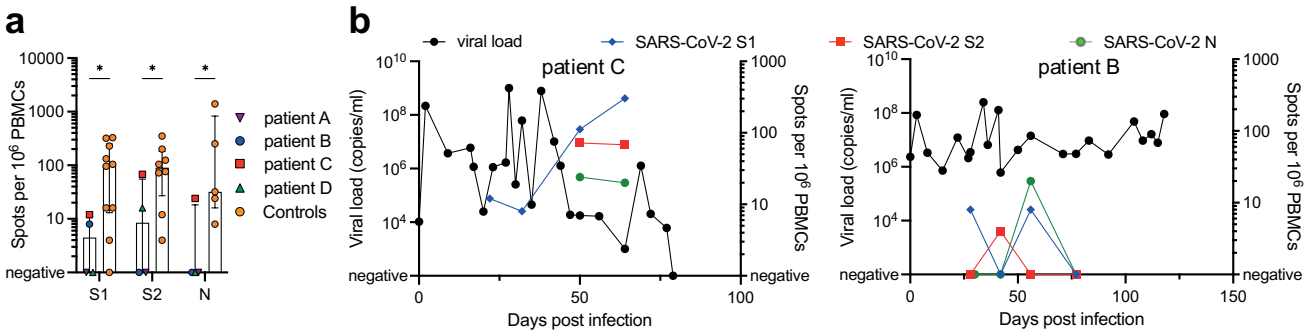


Fig. 3 Impaired T cell responses against SARS-CoV-2 in lung transplant recipients compared to non-immunocompromised controls. **a** PBMC samples of the four lung transplant recipients were stimulated with peptide pools covering the S1 and S2 subunits of the SARS-CoV-2 spike protein and the nucleocapsid protein (N). Results are shown as spots per million PBMCs for the time point closest to 30 days post-infection if multiple time points were available (patient A: 16, patient B: 28, patient C: 32 for S1 and 50 for S2 and N, patient D: 29 days post-infection). For comparison, non-immunocompromised donors were tested matched to 30 days post-infection for S1 ($n=10$), S2 ($n=8$) and N ($n=5$) Elispot responses (blue). Bar graphs represent median values with interquartile range. Groupwise comparisons were performed using the Mann–Whitney test with * indicating p-values <0.05. **b** Longitudinal T cell responses against S1 (blue), S2 (red), and N (green) peptide pools are shown for patient C and patient B in relation to viral load (black)

available for further testing. Interestingly, for patient C we detected robust T cell responses against the three antigens shortly before viral clearance, whereas for patient B we only detected transient responses at lower magnitude consistent with persistent viral replication (Fig. 3b).

Discussion

The findings of this study and in-depth analysis of immune responses to SARS-CoV-2 in severely immunocompromised patients early after lung transplantation emphasize the special precautions that still have to be kept in place for this vulnerable population, even though the pandemic has been declared over.

Our findings are consistent with previously published data by Hodge et al. [8], highlighting impaired immune responses to SARS-CoV-2 infection of LTRs treated with high-dose immunosuppressants through decreased T cell activity. Further groups have shown that LTR under high levels of immunosuppressive drugs can present severe COVID-19 course of disease and experience longer periods of viral shedding compared to those with lower levels of immunosuppression [21, 22]. We were able to confirm these findings by comparing our at risk group with a large cohort of immunocompetent subjects demonstrating impaired cellular and humoral immune responses.

Our patient cohort presented with several risk factor combinations affecting severity of disease as well as mortality. Consistent with literature [23–25] male gender, multiple comorbidities like diabetes or hypertension, smoking history, obesity, and acute kidney injury are in line with the differential disease severity and mortality observed in our cases, although the sample size of our study was insufficient to formally assess risk factors.

Our study has several important limitations. This study was performed during the first wave and second wave of the pandemic in Germany before SARS-CoV-2 vaccines became available, therefore precluding extrapolations of our findings to the current situation with the vast majority of the population having built up immunity to SARS-CoV-2 by vaccination or infection or most often a combination of both. Also, our study is limited to only four at risk patients due to the fact that the occurrence of SARS-CoV-2 infection in the early phase following transplantation was fortunately rare.

Only one of the investigated patients, patient B, was unable to clear the infection and passed away. In this particular case limited and transient CD8 T cell responses and potentially the emergence of the antibody immune escape mutation S:E484K may have contributed to persistent infection with an ultimately fatal outcome [26]. Of note, despite the very low to undetectable levels of anti-SARS-CoV-2 antibodies in patients C and D, both patients cleared the infection successfully. SARS-CoV-2-specific T cell responses were detected in both individuals in line with other studies that demonstrate the importance of T cell immunity for viral clearance in animal models and in patients with deficient antibody responses [27–29].

Several factors may contribute to the inability of patients with high levels of immunosuppression to clear SARS-CoV-2 infections, including the type and dosage of immunosuppressive drugs, the duration of treatment, and the presence of comorbidities. In the case of patient B, convalescent plasma therapy failed to induce viral clearance. Since convalescent plasma therapy was applied as early as available in our center, little data were available at that time on

the amount to be transfused and in what phase of infection. More recent data recommend convalescent plasma for early disease and immunosuppressed patients [30]. Of note, anti-SARS-CoV-2 antibody levels in patient B did not increase significantly after the administration of convalescent plasma potentially due to low levels of specific antibodies in the applied plasma transfusions. Further research suggests higher efficacy in dependence of the SARS-CoV-2-specific antibody content of the administered convalescent plasma therapy [31]. Therapeutic monoclonal antibodies targeting SARS-CoV-2 are another important option in the pre-exposure prophylaxis and treatment of immunocompromised patients, but it has to be considered that their neutralizing capacity may be limited against currently circulating variants as recently observed for various Omicron sublineages [32].

Overall, it is crucial to carefully monitor and case-dependent eventually isolate lung transplant recipients in the early phase after transplantation. In addition, in the case of SARS-CoV-2 infection it is vital to adjust their immunosuppressive treatment when necessary to prevent the development of severe COVID-19 disease and reduce the risk of viral persistence, as well as the emergence of new viral variants. It is crucial to strike a balance between preventing organ rejection and maintaining a robust immune response against infectious agents, particularly in the context of COVID-19.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s15010-023-02116-6.

Author contributions OMG and MM devised and designed the study OMG, MM, IB wrote the main manuscript text OMG, JZ and TK compiled Table 1 and clinical data on the study subjects MM and IB prepared Figs. 1, 2 and 3 XL, IB, PRW, AG, SK, HB, JCH, CS, AH, PMS, BK, TF, AM, CS and OTK made substantial contributions to the acquisition, analysis and interpretation of data. All authors reviewed the manuscript.

Funding Open Access funding enabled and organized by Projekt DEAL. This study was partially funded by the Free State of Bavaria under the FORCOVID (Bavarian consortium for research on the pandemic disease COVID-19) and BayVOC (Molecular Genetic SARS-CoV-2 Surveillance Network in Bavaria) research initiatives. The funding was raised by M. Muenchhoff and O.T. Keppler.

Availability of data and materials Data sets and material can be made available on request if applicable.

Declarations

Conflict of interest The authors have no competing interests to declare that are relevant to the content of this article.

Ethical approval This study was approved by the local ethics committee (Institutional Review Board) (No: 20-245). Furthermore, all patients were recruited into the COVID-19 Registry of the LMU University Hospital Munich (CORKUM) under the WHO trial ID DRKS00021225. All patients gave their written consent to participate in this study and for any resulting publication. This study was conducted following the guidelines of the declaration of Helsinki and the Rules of Good Scientific Practice (GSP) of the Ludwig Maximilian University Munich.

References

1. Fishman JA. Infection in organ transplantation. Am J Transplant. 2017;17:856–79.
2. Hernandez Mdel P, Martin P, Simkins J. Infectious complications after liver transplantation. Gastroenterol Hepatol (N Y). 2015;11:741–53.
3. Ison MG, Hirsch HH. Community-acquired respiratory viruses in transplant patients: diversity, impact, unmet clinical needs. Clin Microbiol Rev. 2019;32:e00042–19.
4. Manuel O, Estabrook M, P. American Society of Transplantation Infectious Diseases Community of. RNA respiratory viral infections in solid organ transplant recipients: Guidelines from the American Society of Transplantation Infectious Diseases Community of Practice. Clin Transplant. 2019;33: e13511.
5. Vilchez RA, et al. Influenza virus infection in adult solid organ transplant recipients. Am J Transplant. 2002;2:287–91.
6. Nam HH, Ison MG. Community-acquired respiratory viruses in solid organ transplant. Curr Opin Organ Transplant. 2019;24:483–9.
7. Hodge G, et al. Intracellular cytokines in blood T cells in lung transplant patients–a more relevant indicator of immunosuppression than drug levels. Clin Exp Immunol. 2005;139:159–64.
8. Hodge G, et al. Time post-lung transplant correlates with increasing peripheral blood T cell granzyme B and proinflammatory cytokines. Clin Exp Immunol. 2010;161:584–90.
9. van Kessel DA, et al. Long-term follow-up of humoral immune status in adult lung transplant recipients. Transplantation. 2017;101:2477–83.
10. Natori Y, et al. A double-blind, randomized trial of high-dose vs standard-dose influenza vaccine in adult solid-organ transplant recipients. Clin Infect Dis. 2018;66:1698–704.
11. Gerovasili V, et al. Impaired humoral and cellular responses to COVID-19 vaccine in heart and lung transplant recipients. Am J Respir Crit Care Med. 2022;205:1476–9.
12. Schramm R, et al. Poor humoral and T-cell response to two-dose SARS-CoV-2 messenger RNA vaccine BNT162b2 in cardiothoracic transplant recipients. Clin Res Cardiol. 2021;110:1142–9.
13. Kneidinger N, et al. Outcome of lung transplant recipients infected with SARS-CoV-2/Omicron/B.1.1.529: a Nationwide German study. Infection. 2023;51:749–57.
14. Peghin M, et al. Epidemiology and immediate indirect effects of respiratory viruses in lung transplant recipients: a 5-year prospective study. Am J Transplant. 2017;17:1304–12.

15. Kates OS, et al. Earliest cases of coronavirus disease 2019 (COVID-19) identified in solid organ transplant recipients in the United States. Am J Transplant. 2020;20:1885–90.
16. Messika J, et al. COVID-19 in lung transplant recipients. Transplantation. 2021;105:177–86.
17. Saez-Gimenez B, et al. COVID-19 in lung transplant recipients: a multicenter study. Am J Transplant. 2021;21:1816–24.
18. Zimmermann J, et al. COVID-19 in recent lung transplant recipients: clinical outcomes and management strategies. Transplant Proc. 2022;54:1504–16.
19. Osterman A, et al. Comparison of four commercial, automated antigen tests to detect SARS-CoV-2 variants of concern. Med Microbiol Immunol. 2021;210:263–75.
20. Muenchhoff M, et al. Genomic epidemiology reveals multiple introductions of SARS-CoV-2 followed by community and nosocomial spread, Germany, February to May 2020. Euro Surveill. 2021;26(43):2002066.
21. Fernandez-Ruiz M, et al. COVID-19 in solid organ transplant recipients: A single-center case series from Spain. Am J Transplant. 2020;20(7):1849–58.
22. Pereira MR, et al. COVID-19 in solid organ transplant recipients: Initial report from the US epicenter. Am J Transplant. 2020;20:1800–8.
23. Booth A, et al. Population risk factors for severe disease and mortality in COVID-19: a global systematic review and meta-analysis. PLoS ONE. 2021;16: e0247461.
24. Dessie ZG, Zewotir T. Mortality-related risk factors of COVID-19: a systematic review and meta-analysis of 42 studies and 423,117 patients. BMC Infect Dis. 2021;21:855.
25. Wolff D, et al. Risk factors for Covid-19 severity and fatality: a structured literature review. Infection. 2021;49:15–28.
26. Jangra S, et al. SARS-CoV-2 spike E484K mutation reduces antibody neutralisation. Lancet Microbe. 2021;2:e283–4.
27. Bange EM, et al. CD8(+) T cells contribute to survival in patients with COVID-19 and hematologic cancer. Nat Med. 2021;27:1280–9.
28. Moss P. The T cell immune response against SARS-CoV-2. Nat Immunol. 2022;23:186–93.
29. Zhao J, Zhao J, Perlman S. T cell responses are required for protection from clinical disease and for virus clearance in severe acute respiratory syndrome coronavirus-infected mice. J Virol. 2010;84:9318–25.
30. Tobian AAR, Cohn CS, Shaz BH. COVID-19 convalescent plasma. Blood. 2022;140:196–207.
31. Begin P, et al. Convalescent plasma for hospitalized patients with COVID-19: an open-label, randomized controlled trial. Nat Med. 2021;27:2012–24.
32. European Centre for Disease Prevention and Control. SARS-CoV-2 variant mutations conferring reducedsusceptibility to antiviral drugs and monoclonal antibodies: a non-systematic literature review for surveillance purposes—July 2023. Stockholm: ECDC; 2023.

10. Literaturverzeichnis

1. Chambers DC, Cherikh WS, Harhay MO, Hayes D, Jr., Hsich E, Khush KK, et al. The International Thoracic Organ Transplant Registry of the International Society for Heart and Lung Transplantation: Thirty-sixth adult lung and heart-lung transplantation Report-2019; Focus theme: Donor and recipient size match. J Heart Lung Transplant. 2019;38(10):1042-55.

2. Leard LE, Holm AM, Valapour M, Glanville AR, Attawar S, Aversa M, et al. Consensus document for the selection of lung transplant candidates: An update from the International Society for Heart and Lung Transplantation. J Heart Lung Transplant. 2021;40(11):1349-79.

3. De Meester J, Smits JM, Persijn GG, Haverich A. Listing for lung transplantation: life expectancy and transplant effect, stratified by type of end-stage lung disease, the Eurotransplant experience. J Heart Lung Transplant. 2001;20(5):518-24.

4. Kourliouros A, Hogg R, Mehew J, Al-Aloul M, Carby M, Lordan JL, et al. Patient outcomes from time of listing for lung transplantation in the UK: are there disease-specific differences? Thorax. 2019;74(1):60-8.

5. Auraen H, Durheim MT, Dellgren G, Hammainen P, Larsson H, Geiran O, et al. Effect of Donor Age on Outcome of Lung Transplantation Stratified by Recipient Diagnosis: A Nordic Multicenter Study. Transplantation. 2019;103(4):807-14.

6. Belperio JA, Weigt SS, Fishbein MC, Lynch JP, 3rd. Chronic lung allograft rejection: mechanisms and therapy. Proc Am Thorac Soc. 2009;6(1):108-21.

7. Meyer KC, Raghu G, Verleden GM, Corris PA, Aurora P, Wilson KC, et al. An international ISHLT/ATS/ERS clinical practice guideline: diagnosis and management of bronchiolitis obliterans syndrome. Eur Respir J. 2014;44(6):1479-503.

8. Sato M, Waddell TK, Wagnetz U, Roberts HC, Hwang DM, Haroon A, et al. Restrictive allograft syndrome (RAS): a novel form of chronic lung allograft dysfunction. J Heart Lung Transplant. 2011;30(7):735-42.

9. Lama VN, Murray S, Lonigro RJ, Toews GB, Chang A, Lau C, et al. Course of FEV(1) after onset of bronchiolitis obliterans syndrome in lung transplant recipients. Am J Respir Crit Care Med. 2007;175(11):1192-8.

10. Todd JL, Neely ML, Finlen Copeland CA, Frankel CW, Reynolds JM, Palmer SM. Prognostic significance of early pulmonary function changes after onset of chronic lung allograft dysfunction. J Heart Lung Transplant. 2019;38(2):184-93.

11. Nelson J, Alvey N, Bowman L, Schulte J, Segovia MC, McDermott J, et al. Consensus recommendations for use of maintenance immunosuppression in solid organ transplantation: Endorsed by the American College of Clinical Pharmacy, American Society of Transplantation, and the International Society for Heart and Lung Transplantation. Pharmacotherapy. 2022;42(8):599-633.

12. Gadola SD. Kapitel 1 - Einführung in das Immunsystem. In: Peter H-H, Pichler W, Müller-Ladner U, editors. Klinische Immunologie (Dritte Ausgabe). Munich: Urban & Fischer; 2012. p. 3-76.

13. McDonald DR, Levy O. 3 - Innate Immunity. In: Rich RR, Fleisher TA, Shearer WT, Schroeder HW, Frew AJ, Weyand CM, editors. Clinical Immunology (Fifth Edition). London: Elsevier; 2019. p. 39-53.e1.

14. Heeger PS. T-cell allorecognition and transplant rejection: a summary and update. Am J Transplant. 2003;3(5):525-33.

15. Jaramillo A, Fernandez FG, Kuo EY, Trulock EP, Patterson GA, Mohanakumar T. Immune mechanisms in the pathogenesis of bronchiolitis obliterans syndrome after lung transplantation. Pediatr Transplant. 2005;9(1):84-93.

16. Peterson EJ, Maltzman JS. 12 - T-Cell Activation and Tolerance. In: Rich RR, Fleisher TA, Shearer WT, Schroeder HW, Frew AJ, Weyand CM, editors. Clinical Immunology (Fifth Edition). London: Elsevier; 2019. p. 183-96.e1.

17. Crotty S. T Follicular Helper Cell Biology: A Decade of Discovery and Diseases. Immunity. 2019;50(5):1132-48.

18. Zhu J, Paul WE. Peripheral CD4+ T-cell differentiation regulated by networks of cytokines and transcription factors. Immunol Rev. 2010;238(1):247-62.

19. Schroeder HW, Radbruch A, Berek C. 7 - B-Cell Development and Differentiation. In: Rich RR, Fleisher TA, Shearer WT, Schroeder HW, Frew AJ, Weyand CM, editors. Clinical Immunology (Fifth Edition). London: Elsevier; 2019. p. 107-18.e1.

20. Andrade CF, Waddell TK, Keshavjee S, Liu M. Innate immunity and organ transplantation: the potential role of toll-like receptors. Am J Transplant. 2005;5(5):969-75.

21. O'Shea JJ, Gadina M, Siegel RM. 9 - Cytokines and Cytokine Receptors. In: Rich RR, Fleisher TA, Shearer WT, Schroeder HW, Frew AJ, Weyand CM, editors. Clinical Immunology (Fifth Edition). London: Elsevier; 2019. p. 127-55.e1.

22. Furukawa M, Chan EG, Ryan JP, Hyzny EJ, Sacha LM, Coster JN, et al. Induction Strategies in Lung Transplantation: Alemtuzumab vs. Basiliximab a Single-Center Experience. Front Immunol. 2022;13:864545.

23. Wood KJ, Shankar S, Hester J, Issa F. 81 - Concepts and Challenges in Organ Transplantation: Rejection, Immunosuppression, and Tolerance. In: Rich RR, Fleisher TA, Shearer WT, Schroeder HW, Frew AJ, Weyand CM, editors. Clinical Immunology (Fifth Edition). London: Elsevier; 2019. p. 1097-114.e1.

24. van der Zwan M, Baan CC, van Gelder T, Hesselink DA. Review of the Clinical Pharmacokinetics and Pharmacodynamics of Alemtuzumab and Its Use in Kidney Transplantation. Clin Pharmacokinet. 2018;57(2):191-207.

25. Nelson J, Kincaide E, Schulte J, Hall R, Levine DJ. Immunosuppression in Lung Transplantation. In: Eisen HJ, editor. Pharmacology of Immunosuppression. Cham: Springer International Publishing; 2022. p. 139-64.

26. Kapitel 3 - Therapeutische Verfahren. In: Peter H-H, Pichler W, Müller-Ladner U, editors. Klinische Immunologie (Dritte Ausgabe). Munich: Urban & Fischer; 2012. p. 135-66.

27. Fernandez-Castillo JC, Cypel M. Immunosuppressive Therapy in Lung Transplantation. Curr Pharm Des. 2020;26(28):3385-8.

28. Staatz CE, Tett SE. Pharmacology and toxicology of mycophenolate in organ transplant recipients: an update. Arch Toxicol. 2014;88(7):1351-89.

29. Frew AJ. 86 - Glucocorticoids. In: Rich RR, Fleisher TA, Shearer WT, Schroeder HW, Frew AJ, Weyand CM, editors. Clinical Immunology (Fifth Edition). London: Elsevier; 2019. p. 1165-75.e1.

30. Hernandez Mdel P, Martin P, Simkins J. Infectious Complications After Liver Transplantation. Gastroenterol Hepatol (N Y). 2015;11(11):741-53.

31. Fishman JA. Infection in Organ Transplantation. Am J Transplant. 2017;17(4):856-79.

32. Fishman JA. Infection in solid-organ transplant recipients. N Engl J Med. 2007;357(25):2601-14.

33. Pons S, Sonneville R, Bouadma L, Styfalova L, Ruckly S, Neuville M, et al. Infectious complications following heart transplantation in the era of high-priority allocation and extracorporeal membrane oxygenation. Ann Intensive Care. 2019;9(1):17.

34. Timsit JF, Sonneville R, Kalil AC, Bassetti M, Ferrer R, Jaber S, et al. Diagnostic and therapeutic approach to infectious diseases in solid organ transplant recipients. Intensive Care Med. 2019;45(5):573-91.

35. Nam HH, Ison MG. Community-acquired respiratory viruses in solid organ transplant. Curr Opin Organ Transplant. 2019;24(4):483-9.

36. Vilchez RA, McCurry K, Dauber J, Lacono A, Griffith B, Fung J, et al. Influenza virus infection in adult solid organ transplant recipients. Am J Transplant. 2002;2(3):287-91.

37. Manuel O, Estabrook M, American Society of Transplantation Infectious Diseases Community of P. RNA respiratory viral infections in solid organ transplant recipients: Guidelines from the American Society of Transplantation Infectious Diseases Community of Practice. Clin Transplant. 2019;33(9):e13511.

38. Ison MG, Hirsch HH. Community-Acquired Respiratory Viruses in Transplant Patients: Diversity, Impact, Unmet Clinical Needs. Clin Microbiol Rev. 2019;32(4).

39. Palmer SM, Jr., Henshaw NG, Howell DN, Miller SE, Davis RD, Tapson VF. Community respiratory viral infection in adult lung transplant recipients. Chest. 1998;113(4):944-50.

40. Allyn PR, Duffy EL, Humphries RM, Injean P, Weigt SS, Saggar R, et al. Graft Loss and CLAD-Onset Is Hastened by Viral Pneumonia After Lung Transplantation. Transplantation. 2016;100(11):2424-31.

41. Gottlieb J, Schulz TF, Welte T, Fuehner T, Dierich M, Simon AR, et al. Community-acquired respiratory viral infections in lung transplant recipients: a single season cohort study. Transplantation. 2009;87(10):1530-7.

42. Vilchez R, McCurry K, Dauber J, Iacono A, Keenan R, Griffith B, et al. Influenza and parainfluenza respiratory viral infection requiring admission in adult lung transplant recipients. Transplantation. 2002;73(7):1075-8.

43. Bridevaux PO, Aubert JD, Soccal PM, Mazza-Stalder J, Berutto C, Rochat T, et al. Incidence and outcomes of respiratory viral infections in lung transplant recipients: a prospective study. Thorax. 2014;69(1):32-8.

44. WHO Director-General's opening remarks at the media briefing on COVID-19 - 11 March 2020 [press release]. 2020.

45. Wu Z, McGoogan JM. Characteristics of and Important Lessons From the Coronavirus Disease 2019 (COVID-19) Outbreak in China: Summary of a Report of 72314 Cases From the Chinese Center for Disease Control and Prevention. JAMA. 2020;323(13):1239-42.

46. Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al. A Novel Coronavirus from Patients with Pneumonia in China, 2019. N Engl J Med. 2020;382(8):727-33.

47. WHO. WHO coronavirus disease (COVID-19) dashboard. Geneva: World Health Organization; [Available from: https://covid19.who.int/.

48. WHO. Statement on the fifteenth meeting of the IHR (2005) Emergency Committee on the COVID-19 pandemic: WHO; 2023 [Available from: https://www.who.int/news/item/05-05-2023-statement-on-the-fifteenth-meeting-of-the-international-health-regulations-(2005)-emergency-committee-regarding-the-coronavirus-disease-(covid-19)-pandemic.

49. Burki T. WHO ends the COVID-19 public health emergency. Lancet Respir Med. 2023;11(7):588.

50. Li Q, Guan X, Wu P, Wang X, Zhou L, Tong Y, et al. Early Transmission Dynamics in Wuhan, China, of Novel Coronavirus-Infected Pneumonia. N Engl J Med. 2020;382(13):1199-207.

51. Guo ZD, Wang ZY, Zhang SF, Li X, Li L, Li C, et al. Aerosol and Surface Distribution of Severe Acute Respiratory Syndrome Coronavirus 2 in Hospital Wards, Wuhan, China, 2020. Emerg Infect Dis. 2020;26(7):1583-91.

52. Guan WJ, Ni ZY, Hu Y, Liang WH, Ou CQ, He JX, et al. Clinical Characteristics of Coronavirus Disease 2019 in China. N Engl J Med. 2020;382(18):1708-20.

53. Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet. 2020;395(10223):497-506.

54. Wu Z, McGoogan JM. Characteristics of and Important Lessons From the Coronavirus Disease 2019 (COVID-19) Outbreak in China: Summary of a Report of 72 314 Cases From the Chinese Center for Disease Control and Prevention. JAMA. 2020;323(13):1239-42.

55. Zhang HP, Sun YL, Wang YF, Yazici D, Azkur D, Ogulur I, et al. Recent developments in the immunopathology of COVID-19. Allergy. 2023;78(2):369-88.

56. Richardson S, Hirsch JS, Narasimhan M, Crawford JM, McGinn T, Davidson KW, et al. Presenting Characteristics, Comorbidities, and Outcomes Among 5700 Patients Hospitalized With COVID-19 in the New York City Area. JAMA. 2020;323(20):2052-9.

57. Docherty AB, Harrison EM, Green CA, Hardwick HE, Pius R, Norman L, et al. Features of 20 133 UK patients in hospital with covid-19 using the ISARIC WHO Clinical Characterisation Protocol: prospective observational cohort study. BMJ. 2020;369:m1985.

58. Mao R, Qiu Y, He JS, Tan JY, Li XH, Liang J, et al. Manifestations and prognosis of gastrointestinal and liver involvement in patients with COVID-19: a systematic review and meta-analysis. Lancet Gastroenterol Hepatol. 2020;5(7):667-78.

59. Zhou F, Yu T, Du R, Fan G, Liu Y, Liu Z, et al. Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. Lancet. 2020;395(10229):1054-62.

60. Sawadogo W, Tsegaye M, Gizaw A, Adera T. Overweight and obesity as risk factors for COVID-19-associated hospitalisations and death: systematic review and meta-analysis. BMJ Nutr Prev Health. 2022;5(1):10-8.

61. Zhang JJ, Dong X, Liu GH, Gao YD. Risk and Protective Factors for COVID-19 Morbidity, Severity, and Mortality. Clin Rev Allergy Immunol. 2023;64(1):90-107.

62. Esper FP, Adhikari TM, Tu ZJ, Cheng YW, El-Haddad K, Farkas DH, et al. Alpha to Omicron: Disease Severity and Clinical Outcomes of Major SARS-CoV-2 Variants. J Infect Dis. 2023;227(3):344-52.

63. Fernandes Q, Inchakalody VP, Merhi M, Mestiri S, Taib N, Moustafa Abo El-Ella D, et al. Emerging COVID-19 variants and their impact on SARS-CoV-2 diagnosis, therapeutics and vaccines. Ann Med. 2022;54(1):524-40.

64. Nyberg T, Ferguson NM, Nash SG, Webster HH, Flaxman S, Andrews N, et al. Comparative analysis of the risks of hospitalisation and death associated with SARS-CoV-2 omicron (B.1.1.529) and delta (B.1.617.2) variants in England: a cohort study. Lancet. 2022;399(10332):1303-12.

65. Abdolreza E, Fereshteh E, Armin JM, Amir S. EG.5 (Eris) and BA.2.86 (Pirola) two new subvariants of SARS-CoV-2: a new face of old COVID-19. Infection. 2024.

66. Shrestha LB, Foster C, Rawlinson W, Tedla N, Bull RA. Evolution of the SARS-CoV-2 omicron variants BA.1 to BA.5: Implications for immune escape and transmission. Rev Med Virol. 2022;32(5):e2381.

67. Chen P, Nirula A, Heller B, Gottlieb RL, Boscia J, Morris J, et al. SARS-CoV-2 Neutralizing Antibody LY-CoV555 in Outpatients with Covid-19. N Engl J Med. 2021;384(3):229-37.

68. Group RC. Convalescent plasma in patients admitted to hospital with COVID-19 (RECOVERY): a randomised controlled, open-label, platform trial. Lancet. 2021;397(10289):2049-59.

69. Group RC, Horby P, Lim WS, Emberson JR, Mafham M, Bell JL, et al. Dexamethasone in Hospitalized Patients with Covid-19. N Engl J Med. 2021;384(8):693-704.

70. Beigel JH, Tomashek KM, Dodd LE, Mehta AK, Zingman BS, Kalil AC, et al. Remdesivir for the Treatment of Covid-19 - Final Report. N Engl J Med. 2020;383(19):1813-26.

71. Yuan Y, Jiao B, Qu L, Yang D, Liu R. The development of COVID-19 treatment. Front Immunol. 2023;14:1125246.

72. Polack FP, Thomas SJ, Kitchin N, Absalon J, Gurtman A, Lockhart S, et al. Safety and Efficacy of the BNT162b2 mRNA Covid-19 Vaccine. N Engl J Med. 2020;383(27):2603-15.

73. Hall V, Foulkes S, Insalata F, Kirwan P, Saei A, Atti A, et al. Protection against SARS-CoV-2 after Covid-19 Vaccination and Previous Infection. N Engl J Med. 2022;386(13):1207-20.

74. Fiolet T, Kherabi Y, MacDonald CJ, Ghosn J, Peiffer-Smadja N. Comparing COVID-19 vaccines for their characteristics, efficacy and effectiveness against SARS-CoV-2 and variants of concern: a narrative review. Clin Microbiol Infect. 2022;28(2):202-21.

75. Azzi Y, Bartash R, Scalea J, Loarte-Campos P, Akalin E. COVID-19 and Solid Organ Transplantation: A Review Article. Transplantation. 2021;105(1):37-55.

76. Caillard S, French Registry of Solid Organ Transplant Patients C. COVID-19 and kidney transplantation. Presse Med. 2022;51(4):104146.

77. Hadi YB, Naqvi SFZ, Kupec JT, Sofka S, Sarwari A. Outcomes of COVID-19 in Solid Organ Transplant Recipients: A Propensity-matched Analysis of a Large Research Network. Transplantation. 2021;105(6):1365-71.

78. Jering KS, McGrath MM, Mc Causland FR, Claggett B, Cunningham JW, Solomon SD. Excess mortality in solid organ transplant recipients hospitalized with COVID-19: A large-scale comparison of SOT recipients hospitalized with or without COVID-19. Clin Transplant. 2022;36(1):e14492.

79. Bartelt L, van Duin D. An overview of COVID-19 in solid organ transplantation. Clin Microbiol Infect. 2022;28(6):779-84.

80. Sahota A, Tien A, Yao J, Dong E, Herald J, Javaherifar S, et al. Incidence, Risk Factors, and Outcomes of COVID-19 Infection in a Large Cohort of Solid Organ Transplant Recipients. Transplantation. 2022;106(12):2426-34.

81. Rinaldi M, Bartoletti M, Bussini L, Pancaldi L, Pascale R, Comai G, et al. COVID-19 in solid organ transplant recipients: No difference in survival compared to general population. Transpl Infect Dis. 2021;23(1):e13421.

82. Myers CN, Scott JH, Criner GJ, Cordova FC, Mamary AJ, Marchetti N, et al. COVID-19 in lung transplant recipients. Transpl Infect Dis. 2020;22(6):e13364.

83. Donohue JK, Hyzny EJ, Clifford S, Chan EG, Coster JN, Furukawa M, et al. Immediate Postoperative COVID-19 Infection after Lung Transplantation: A Systematic Review and Case Series. J Clin Med. 2023;12(22).

84. Keller BC, Le A, Sobhanie M, Colburn N, Burcham P, Rosenheck J, et al. Early COVID-19 infection after lung transplantation. Am J Transplant. 2020;20(10):2923-7.

85. Zimmermann J, Glueck OM, Fertmann JM, Sienel WG, Yavuz G, Damirov F, et al. COVID-19 in Recent Lung Transplant Recipients: Clinical Outcomes and Management Strategies. Transplant Proc. 2022;54(6):1504-16.

86. Permpalung N, Bazemore K, Chiang TP, Mathew J, Barker L, Nematollahi S, et al. Impact of COVID-19 on Lung Allograft and Clinical Outcomes in Lung Transplant Recipients: A Case-control Study. Transplantation. 2021;105(9):2072-9.

87. Danziger-Isakov L, Blumberg EA, Manuel O, Sester M. Impact of COVID-19 in solid organ transplant recipients. Am J Transplant. 2021;21(3):925-37.

88. Gustine JN, Jones D. Immunopathology of Hyperinflammation in COVID-19. Am J Pathol. 2021;191(1):4-17.

89. Blanco-Melo D, Nilsson-Payant BE, Liu WC, Uhl S, Hoagland D, Moller R, et al. Imbalanced Host Response to SARS-CoV-2 Drives Development of COVID-19. Cell. 2020;181(5):1036-45 e9.

90. Del Valle DM, Kim-Schulze S, Huang HH, Beckmann ND, Nirenberg S, Wang B, et al. An inflammatory cytokine signature predicts COVID-19 severity and survival. Nat Med. 2020;26(10):1636-43.

91. Heldman MR, Kates OS, Safa K, Kotton CN, Georgia SJ, Steinbrink JM, et al. COVID-19 in hospitalized lung and non-lung solid organ transplant recipients: A comparative analysis from a multicenter study. Am J Transplant. 2021;21(8):2774-84.

92. Huh K, Kang M, Kim YE, Choi Y, An SJ, Seong J, et al. Risk of Severe COVID-19 and Protective Effectiveness of Vaccination Among Solid Organ Transplant Recipients. J Infect Dis. 2023.

93. Turkkan S, Beyoglu MA, Sahin MF, Yazicioglu A, Tezer Tekce Y, Yekeler E. COVID-19 in lung transplant recipients: A single-center experience. Transpl Infect Dis. 2021;23(5):e13700.

94. Lee A, Wong SY, Chai LYA, Lee SC, Lee MX, Muthiah MD, et al. Efficacy of covid-19 vaccines in immunocompromised patients: systematic review and meta-analysis. BMJ. 2022;376:e068632.

95. Haidar G, Agha M, Bilderback A, Lukanski A, Linstrum K, Troyan R, et al. Prospective Evaluation of Coronavirus Disease 2019 (COVID-19) Vaccine Responses Across a Broad Spectrum of Immunocompromising Conditions: the COVID-19 Vaccination in the Immunocompromised Study (COVICS). Clin Infect Dis. 2022;75(1):e630-e44.

96. Safa K, Kotton CN. COVID-19 Vaccines and Solid Organ Transplantation: More Doses, More Protection. Transplantation. 2023;107(1):21-2.

97. Bailey AJM, Maganti HB, Cheng W, Shorr R, Arianne Buchan C, Allan DS. Humoral and Cellular Response of Transplant Recipients to a Third Dose of mRNA SARS-CoV-2 Vaccine: A Systematic Review and Meta-analysis. Transplantation. 2023;107(1):204-15.

98. Al Jurdi A, Morena L, Cote M, Bethea E, Azzi J, Riella LV. Tixagevimab/cilgavimab pre-exposure prophylaxis is associated with lower breakthrough infection risk in vaccinated solid organ transplant recipients during the omicron wave. Am J Transplant. 2022;22(12):3130-6.

99. Demolder S, Schaevers V, Lagrou K, De Munter P, Beeckmans H, Verleden GM, et al. COVID-19 Outcomes in Lung Transplant Recipients Following Pre-Exposure Prophylaxis With Tixagevimab-Cilgavimab During the Omicron BA.5 Surge: A Single Center Analysis. Transpl Int. 2024;37:12061.

11. Danksagung

Bedanken möchte ich mich insbesondere bei meiner Partnerin Valentina. Sie steht mir mit aufmunternden Worten zur Seite, wenn ich nicht weiterweiß. Sie motiviert mich nach einem langen Arbeitstag doch noch ein paar Zeilen zu schreiben. Sie unterstützt mich nach Kräften, für unser gemeinsames Vorankommen zu kämpfen. Dafür und für noch viel mehr möchte ich mich mit dieser Arbeit bedanken.

Außerdem geht Dank an meine Familie, meine Eltern und meine Brüder.

Meinen Betreuenden, PD Dr. Teresa Kauke, PD Dr. Christian Schneider und Prof. Dr. Sebastian Michel möchte ich für ihre offenen Ohren und weiterführenden Ratschläge, ihr dauerndes Entgegenkommen und ihre Geduld danken. Sie haben mich bei dieser Arbeit maßgeblich unterstützt.

Besonderer Dank gilt auch PD Maximilian Münchhoff, ohne welchen die Realisierung der Arbeit und die Publikation nicht möglich gewesen wären. Trotz vielfältiger Arbeit und außerordentlichem Workload, welche wir alle während zweier Pandemiejahre zu schultern hatten, konnte ich auf seine Unterstützung und verzweigten Kontakte zählen, sodass die Arbeit als Team-Effort zu Ende gebracht werden konnte.

LMU LUDWIG-MAXIMILIANS-UNIVERSITÄT MÜNCHEN

Dekanat Medizinische Fakultät
Promotionsbüro

Erklärung zur Übereinstimmung der gebundenen Ausgabe der Dissertation mit der elektronischen Fassung

Glück, Olaf Michael

Name, Vorname

Hiermit erkläre ich, dass die elektronische Version der eingereichten Dissertation mit dem Titel:

Lung transplantation in times of Covid-19
—
Immune response and clinical management of recent lung allograft recipients infected with SARS-CoV 2

in Inhalt und Formatierung mit den gedruckten und gebundenen Exemplaren übereinstimmt.

München, 28.04.2025

Ort, Datum

Olaf Michael Glück

Unterschrift Olaf Michael Glück