Title: A controlled randomized, open-label, multi-centre study evaluating if a steroid-free immunosuppressive protocol, based on ATG- induction, low tacrolimus-dose and therapeutic drug monitoring of mycophenolate mofetil, reduces the incidence of new onset diabetes after transplantation, in comparison with a standard steroid-based protocol with low-dose tacrolimus.

Short Title: SAILOR-study (trial of Steroid AvoIdance and LOw-dose cni by atg-induction in Renal transplantation)

Investigational product: Thymoglobulin (Genzyme) and Simulect (Novartis)

Indication: Kidney transplantation

Sponsor: Transplant Institute, Sahlgrenska University Hospital

SE-413 45 Gothenburg, Sweden

Protocol: SUGBG-012011

EudraCT number: 2012-000451-13

Phase of development: Phase IV

Study initiation date: 2013-02-19 (first subject first visit)

Study completion date: 2019-05-01 (last subject last visit)

Principal investigator: Per Lindnér, Transplant Institute, Sahlgrenska University Hospital

Sponsor signatory: Per Lindnér, phone: +46705548400, fax: +4631823290

Statement: This study was performed in accordance with Good Clinical Practice (GCP), including the archiving of essential documents.

SYNOPSIS

Name of Sponsor: Transplant Institute (Sahlgrenska University Hospital)
Investigational Product: Thymoglobuline® (Sanofi AB, test) and Simulect® (Novartis, comparator)
Active Ingredient: Anti-thymocyte immunoglobulin, rabbit (Thymoglobuline®, test) and Basiliximab (Simulect®, comparator)
Title of Study: A controlled randomized, open-label, multi-centre study evaluating if a steroid-free immunosuppressive protocol, based on ATG- induction, low tacrolimus-dose and therapeutic drug monitoring of mycophenolate mofetil, reduces the incidence of new onset diabetes after transplantation, in comparison with a standard steroid-based protocol with low-dose tacrolimus. (SAILOR-study)
Principal investigator: Per Lindnér
Study centers: Transplant Institute (Sahlgrenska University Hospital, Gothenburg, Sweden); Department of Transplantation, Skåne University Hospital (Malmö, Sweden); Department of Nephrology, Aarhus University Hospital (Skejby, Denmark)
Publications:

- Ekberg J, Ekberg H, Jespersen B, Källen R, Skov K, Olausson M, Mjörnstedt L, Lindnér P. An in-progress, open-label, multi-centre study (SAILOR) evaluating whether a steroid-free immunosuppressive protocol, based on ATG induction and a low tacrolimus dose, reduces the incidence of new onset diabetes after transplantation. *Transplant Res.* 2014 Jun 13;3:12. doi: 10.1186/2047-1440-3-12. PMID: 24959347; PMCID: PMC4067097.
- Jana Ekberg, Seema Baid-Agrawal, Bente Jespersen, Ragnar Källen, Ehab Rafael, Karin Skov and Per Lindnér, MD, SAILOR study – steroid avoidance is safe in immunologically low-risk kidney transplant recipients: A randomized controlled trial with two-years follow-up. Submitted to *Kidney International*.

Phase of development: IV
Study period: February 2013 to May 2019
Study objective(s): To evaluate whether the use of a steroid-free and a low tacrolimus-dose regimen, based on a high dose of anti-thymocyte globuline (ATG) induction and mycophenolate mofetil (MMF) dosed by therapeutic drug monitoring, reduces the cumulative incidence of new onset diabetes after transplantation, in comparison with a steroid-based protocol, based on Tacrolimus (Tac), steroids, MMF and IL-2-receptor antibodies (IL-2 ab).
Primary objective: To evaluate the cumulative incidence of NODAT (new onset of diabetes) after transplantation 12 months after transplantation, defined as of one of the following:

- ≥2 FPG ≥7,0 mmol/l ≥ 30 days apart
- 2-h Plasma Glucose ≥11,1 mmol/l in the OGTT≥ 30 days apart
- Oral hypoglycemic ≥30 consecutive days
- Insulin ≥30 consecutive days

Secondary objective(s): To evaluate the composite measure of freedom from acute rejection (AR), graft survival, and patient survival at 12 and 24 months after transplantation. Other endpoints include:

- The cumulative incidence of NODAT 3 and 12 months after transplantation as defined above, per protocol population.

- Renal function evaluated by measured glomerular filtration rate (mGFR), by Iohexol or Cr-EDTA clearance, at 12 and 24 months.
- Cumulative incidence of major adverse cardiovascular events (MACE), defined as 1. myocardial infarction/PTCA/CABG; 2. stroke; 3. hospitalisation for heart failure; 4. cardiovascular death.
- Use of antidiabetic medication at 3, 6, 12, and 24 months.
- Cumulative incidence of biopsy proven rejection using the Banff Classification of Renal Allograft Nephropathy (Roufosse 2018) and of HLA-donor specific antibodies.
- Incidence of chronic changes, analysed by protocol biopsies at transplantation and after 12 months, evaluated by the Banff Classification of Renal Allograft Pathology (Roufosse 2018).
- Incidence of hypertension measured in a standardized way at 3, 12 and 24 months
- Number and type of antihypertensive drugs at 3, 12 and 24 months
- Number and type of lipid lowering drugs at 3, 12 and 24 months
- Incidence of malignancies
- Incidence of infections
- Safety and tolerability assessment of insulin resistance at 3 and 12 months.

Design and Methodology: Prospective, multi-center controlled, randomized, parallel group, open-label study in kidney transplant patients. Follow-up was performed for 24 months after transplantation with visits performed as per clinical practice according to each center's follow-up scheme.

Number of subjects (planned and analyzed): 222 subjects were planned to be enrolled in total (3 sites); 224 were actually randomized and 222 received a transplant as well as at least one study medication and attended at least one follow-up visit.

Inclusion criteria: 1) Patients receiving a first or second single kidney transplant from a deceased or a living donor; 2) Female or male aged above 18 years; 3) Patients considered for a standard immunosuppressive protocol; 4) Patients capable of giving written informed consent for participation in the study for 24 months.

Exclusion criteria: 1) Patients with known diabetes mellitus or plasma glucose >11.1 mmol/l at admission; 2) Patients receiving steroids at the time of transplantation or likely to need steroids after transplantation; 3) Recipients of multiorgan transplants, and or previously transplanted with any other organ than kidney; 4) Patients with CDC-PRA > 25 % in most recent test or for any other reason considered to be of a high risk for rejection which requires an enhanced immunosuppression; 5) Patients receiving a renal transplant from a HLA-identical sibling; 6) Patients with hypersensitivity to, or other reasons to not be able to take the immunosuppressive drugs used in the study; 7) Patients who are recipients of ABO-incompatible transplants; 8) Patients who are unlikely to comply with the study requirements; 9) Patients, and/or those receiving organs from donors, who are positive for HIV, Hepatitis B surface antigen or Hepatitis C virus; 10) Females of childbearing potential, who are, or are planning to be, pregnant, and/or are unwilling to use effective means of contraception.

Test product, dose, and mode of administration, batch number: Thymoglobuline® induction (2.5 mg/kg, pre-/peroperatively day 0, and day 1); intraventricular use; batch number not registered as it is an approved drug used over a period of 5 years

Duration of treatment: 2 days
Reference therapy, dose and mode of administration, batch number: Simulect® induction (20 mg, day 0 and day 4); intravenous/intravenous bolus use; batch number not registered as it is an approved drug used over a period of 5 years
Criteria for evaluations:

- **Efficacy:** Incidence of NODAT as defined as any of the following, ≥2 FPG ≥7,0 mmol/l ≥ 30 days apart; 2-h Plasma Glucose ≥11,1 mmol/l in the OGTT≥ 30 days apart; Oral hypoglycemic ≥30 consecutive days; Insulin ≥30 consecutive days
- **Safety:** Adverse events and serious adverse events including acute rejection and death, renal function.

Statistical methods: Demographics and baseline characteristics were evaluated descriptively. For comparison between groups, dichotomous variables were evaluated using Fischer's exact test, categorical variables using Mantel-Haenszel Chi-squared test, and continuous variables using Fischer's non-parametric permutation test. Additionally, Kaplan-Meier methods were applied to evaluated time to reach PTDM, AR, graft loss, and death, while independent predictors of PTDM were selected using Cox proportional hazard regression analyses. Statistical significance was set at a p-value of 0.05.
Summary – Results
Efficacy: No significant difference between the two arms was observed in PTDM at 12 months after transplantation (12.4% arm A versus 18.3% arm B; p=0.3)
Safety: No significant differences were observed between the two arms regarding incidence of BPR, kidney function, adverse events, or other outcomes at 24 months post-transplantation.
Conclusions: Avoidance of oral steroids is safe and feasible in combination with the low-dose Tac/MMF maintenance regimen in the study population, albeit without a reduction in incidence in PTDM.

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4 ABBREVIATIONS AND DEFINTION OF TERMS

4.1 LIST OF ABBREVIATIONS

Abbreviations	Description of abbreviations
ADA	American Diabetes Association
AE	Adverse Event
AR	Acute Rejection
ATG	Anti-Thymocyte Globuline
AUC	Area Under the Curve
CABG	Coronary Artery Bypass Grafting
CRF	Case Report Form
ESKD	End-Stage Kidney Disease
ET	Early Termination
FPG	Fasting Plasma Glucose
GCP	Good Clinical Practice
ICF	Informed Consent Form
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IRB	Institutional Review Board
ITT	Intension To Treat
MACE	Major Adverse Cardiovascular Events
mGFR	Measured Glomerular Filtration Rate
MMF	Mycophenolate mofetil
NODAT	New onset diabetes after transplantation
OGTT	Oral Glucose Tolerance Test
PP	Per Protocol
PTCA	Percutaneous Transluminal Coronary Angioplasty
PTDM	Post-Transplantation Diabetes Mellitus
RCTs	Randomized Clinical Trials
SA	Steroid Avoidance
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SM	Steroid Maintenance
SW	Steroid Withdrawal

4.2 LIST OF KEY STUDY TERMS

Terms	Definition of terms
Baseline	1) Observed values/findings, which are regarded as calibrated zero status in the present study, 2) Time when 'Baseline' is observed.
Investigational period	Period of time where major interests of protocol objectives related to defined endpoints are observed, and usually where the test drug or comparative drug (sometimes without randomization) is given to a subject, and continues until the last observation after completing administration of the test drug or comparative drug.
Investigator	A physician or dentist responsible for the conduct of the clinical trial at a trial site. If a team of individuals at a trial site conducts a trial, the investigator is the responsible leader of the team.
Randomization	Action to allocate a subject to the treatment group or treatment cohort. Depending on the type of rules for handling for study drugs, 'Randomization' is usually executed just before entering the 'investigational period'
Randomization/ Treatment number	Number assigned to each subject who has completed ALL screening assessments successfully at baseline and is willing to take the study drug.
Randomized subject/ Subjects given the test drugs	Subjects randomized to the treatment group (test drug group) or control group, and those received open label study treatment.
Source data	All information in original records and certified copies of original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial. Source data are contained in source documents (original records or certified copies).
Source documents	Original documents, data, and records including source data.
Subject	An individual who participates in a clinical trial, either as a recipient of the investigational product(s) or as a control.
Withdrawal	Subject enrolled but did not complete the study for any reason.

5 ETHICS

5.1 INDEPENDENT ETHICS COMMITTEE OR INSTITUTIONAL REVIEW BOARD

The study protocol and all amendments were reviewed by the Independent Ethics Committee (IEC) or Institutional Review Board (IRB) for each country (Sweden, Denmark), as listed in Appendix 16.1.3.

5.2 ETHICAL CONDUCT OF THE STUDY

The study was conducted according to the ethical principles of the Declaration of Helsinki, Good Clinical Practice (GCP), ICH Guidelines, and the applicable laws and regulations.

5.3 PATIENT INFORMATION AND CONSENT

Signed and dated informed consent was obtained in writing from each patient prior to start of study-related activities. Samples of the written information given to each patient and the consent form are presented in Appendix 16.1.3.

6 INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

The study was performed at three centers (two in Sweden: Gothenburg and Malmö; and one in Denmark: Aarhus) and included a data monitoring committee. The administrative structure of the study is described in detail in Appendix 16.1.4, including a list of investigators, their affiliations, and their qualifications, plus that of other important staff.

The signatures of the principal investigators and important staff are provided in Appendix 16.1.5.

7 INTRODUCTION

Kidney transplantation is today the best option for eligible patients with kidney failure. However, although short-term results have yielded declining acute rejection (AR) rates, long-term results have not improved considerably due to premature death mainly related to complications of immunosuppressive treatment such as cardiovascular complications, infections and malignancies (1), as well as chronic rejection (2). Kidney transplantation can also be followed by post-transplantation diabetes mellitus (PTDM), which is an independent predictor of graft failure, major cardiovascular events (3), and mortality (4,5). Different immunosuppressive regimens and a lack of standard PTDM diagnostic criteria results in variable incidence of PTDM in the literature. However, one study looking at two randomized controlled trials (RCTs) using a definition of PTDM based on criteria from the American Diabetes Association (ADA), observed a one-year PTDM incidence of 30-37% using an immunosuppressive protocol with standard doses of tacrolimus, mycophenolate mofetil (MMF), and steroids (6,7). Together with demographic factors such as age, ethnicity, and weight, both tacrolimus and steroids are considered to be risk factors for PTDM, as steroids are believed to result in insulin resistance while insulin secretion is impaired in a dose-related manner by tacrolimus (8).

Over the last twenty years, risks and benefits of steroid-avoidance (SA) and steroid-withdrawal (SW) immunosuppressive regimens have been evaluated worldwide in various RCTs. In a systematic Cochrane review in 2016 (9), steroid-sparing strategies were observed to be associated with increased acute rejection (AR) rate, but not increased graft loss in adult kidney transplant recipients. Clear beneficial effects (e.g., mortality reduction or PTDM within five years after transplantation) were not demonstrated.

The standard of care immunosuppressive regiment in kidney transplant patients involves induction with monoclonal interleukin-2-receptor antibody and maintenance with low-dose tacrolimus/MMF/steroids (10). Few studies have investigated the safety and

efficacy of steroid-sparing protocols with the currently used standard of care maintenance regimen (11,12). The HARMONY study, a recent multicenter RCT in Germany, compared basiliximab induction with standard of care/steroid-maintenance (SM) therapy (arm A) or rapid steroid-withdrawal (SW) therapy (arm B) toanti-thymocyte globulin (ATG) induction with standard of care/rapid SM therapy (arm C). The primary endpoint of the study was biopsy-proven AR and although induction with ATG did not show superiority over induction with basiliximab (arm C vs arm B), a statistically significant reduction in incidence of PTDM was observed in both arms with rapid SM after one year (11).

This RCT, the SAILOR study, was designed to evaluate whether a steroid-avoidance protocol may be used in a non-diabetic population to reduce the incidence of PTDM with good safety and efficacy. The protocol is to be compared with the standard of care regimen. ATG was considered more effective in preventing AR and thus was chosen as the induction therapy in this study rather than basiliximab (13,14).

8 STUDY OBJECTIVES AND DESIGN

8.1 PRIMARY OBJECTIVE

To evaluate if steroid-free immunosuppressive protocol, based on thymoglobulin induction, reduces the incidence of new onset diabetes after transplantation (NODAT), at 12 months after transplantation, defined as adopted from the ADA criteria (2012). Primary endpoints include cumulative incidence of one of the following:

- ≥2 FPG ≥7, 0 mmol/l ≥ 30 days apart
- 2-h Plasma Glucose ≥11,1 mmol/l in the oral glucose tolerance test (OGTT) ≥ 30 days apart
- Oral hypoglycemic ≥30 consecutive days
- Insulin ≥30 consecutive days

8.2 SECONDARY OBJECTIVES

To evaluate the composite measure of freedom from AR, graft survival, and patient survival at 12 and 24 months after transplantation. Other endpoints include:

- The cumulative incidence of NODAT 3 and 12 months after transplantation as defined above, per protocol population.
- Renal function evaluated by measured glomerular filtration rate (mGFR), by Iohexol or Cr-EDTA clearance, at 12 and 24 months.
- Cumulative incidence of major adverse cardiovascular events (MACE), defined as 1. myocardial infarction/percutaneous transluminal coronary angioplasty (PTCA)/coronary artery bypass grafting (CABG); 2. stroke; 3. hospitalisation for heart failure; 4. cardiovascular death.
- Use of antidiabetic medication at 3, 6, 12, and 24 months.
- Cumulative incidence of biopsy proven rejection using the Banff Classification of Renal Allograft Nephropathy (Roufosse 2018) and of HLA-donor specific antibodies.

- Incidence of chronic changes, analysed by protocol biopsies at transplantation and after 12 months, evaluated by the Banff Classification of Renal Allograft Pathology (Roufosse 2018).
- Incidence of hypertension measured in a standardized way at 3, 12 and 24 months
- Number and type of antihypertensive drugs at 3, 12 and 24 months
- Number and type of lipid lowering drugs at 3, 12 and 24 months
- Incidence of malignancies
- Incidence of infections
- Safety and tolerability assessment of insulin resistance at 3 and 12 months.

9 INVESTIGATIONAL PLAN

9.1 OVERALL STUDY DESIGN AND PLAN: DESCRIPTION

This was a prospective, multicenter, controlled, randomized, parallel group, open-label study in kidney transplant patients. After signing informed consent and prior to transplantation, a total of 224 patients from three centers were enrolled and randomized to either of two treatment groups called arm A (SA arm) and arm B (SM arm, standard of care), described below. A total of 222 received transplantation as well as at least one study medication and attended at least one follow-up visit (arm A, n=113 and arm B, n=109). Follow-up was performed for 24 months and follow-up visits were performed as per standard clinical practice for transplantation follow-up; see

Figure 1.

- **Arm A (SA arm):** Induction with ATG (Thymoglobuline®; Sanofi AB) at 2.5 mg/kg peroperatively before perfusion at day 0, and day 1; methylprednisolone bolus (Solu-Medrol®; Pfizer) 250 mg before the first ATG dose and 50 mg before the second ATG dose, and maintenance treatment based on prolonged-release low-dose tacrolimus (Advagraf®; Astellas Pharma), starting dose 0.2 mg/kg once daily with target trough levels 5-10 ng/ml within first three months and thereafter 4-7 ng/ml, and MMF 1g twice a day controlled by a single area under the curve (AUC) measurement on day 10±5 with target AUC 40-60 mg*h/L.
- **Arm B (SM arm, standard of care):** Induction with basiliximab (Simulect®; Novartis) at 20 mg on day 0 and day 4; methylprednisolone 250-500 mg day 0 before reperfusion, according to the local center practice, and maintenance treatment as in SA-arm plus prednisolone in doses by local center practice, but not less than the final dose of 5 mg daily.

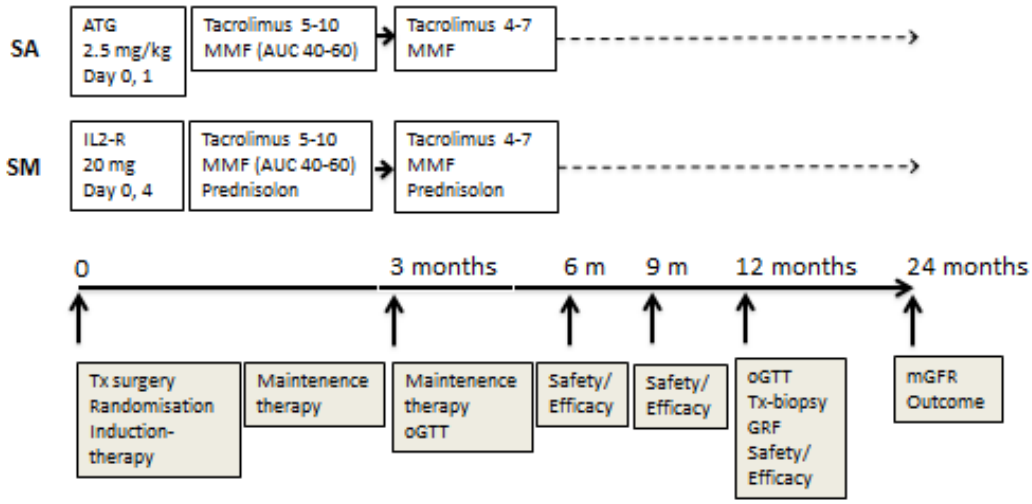

Figure 1. Study flowchart

For full details of study design please refer to the protocol in Appendix 16.1.1.

The case report form (CRF) can be found in Appendix 16.1.2.

9.2 DISCUSSION OF STUDY DESIGN, INCLUDING THE CHOICE OF CONTROL GROUPS

The comparator arm (arm B, SM arm) consisted of the standard of care immunosuppressive regimen in kidney transplant patients, which involves induction with monoclonal interleukin-2-receptor antibody and maintenance with low-dose tacrolimus/MMF/steroids.

The test arm (arm A, SA arm) involved the use of ATG, which was chosen as an indication therapy rather than basilizimab as it was considered more effective in preventing AR based on existing literature.

The study was designed to take advantage of standard clinical routine for follow-up visits and all follow-up visits were performed as per the standard schedule for follow-up after transplantation at each center.

Selection bias was reduced by employing randomization. Although this study was open and thus did not employ blinding, patient identity and treatment assignment were concealed to the Primary Endpoint Committee, two independent nephrologists who assessed the accuracy of the PTDM diagnosis, and to two pathologists, who centrally evaluated all transplant biopsies.

9.3 STUDY POPULATION

9.3.1 INCLUSION CRITERIA

Subjects were eligible for the study if all of the following applied:

1. Patients receiving a first or second single kidney transplant from a deceased or a living donor.
2. Female or male aged above 18 years

3. Patients considered for a standard immunosuppressive protocol.
4. Patients capable of giving written informed consent for participation in the study for 24 months

9.3.2 EXCLUSION CRITERIA

Subjects were excluded from participation if any of the following applied:

1. Patients with known diabetes mellitus or plasma glucose >11.1 mmol/l at admission
2. Patients receiving steroids at the time of transplantation or likely to need steroids after transplantation
3. Recipients of multiorgan transplants, and or previously transplanted with any other organ than kidney
4. Patients with CDC-PRA > 25 % in most recent test or for any other reason considered to be of a high risk for rejection which requires an enhanced immunosuppression.
5. Patients receiving a renal transplant from a HLA-identical sibling
6. Patients with hypersensitivity to, or other reasons to not be able to take the immunosuppressive drugs used in the study
7. Patients who are recipients of ABO-incompatible transplants
8. Patients who are unlikely to comply with the study requirements
9. Patients, and/or those receiving organs from donors, who are positive for HIV, Hepatitis B surface antigen or Hepatitis C virus.
10. Females of childbearing potential, who are, or are planning to be, pregnant, and/or are unwilling to use effective means of contraception.

9.3.3 REMOVAL OF PATIENTS FROM THERAPY OR ASSESSMENT

Study termination for individual subjects was applied if any one of the following termination criteria occurred:

- Withdrawal of consent
- Graft loss
- Death

Furthermore, subjects enrolled in the study and for whom study treatment was discontinued prematurely for some reason including if clinical condition warranted it, were also discontinued. These individuals remained in follow-up and reasons for discontinuation were recorded in the CRF and Visit 6 (End of Study Visit or Early Termination (ET)) was performed.

9.4 STUDY TREATMENTS

All medications used in this study were handled as per normal routine at the study centers.

9.4.1 TREATMENTS ADMINISTERED

The following treatments were administered for treatment arm A:

- Thymoglobuline® induction, intraventricular (2.5 mg/kg, pre-/peroperatively day 0; 2.5 mg/kg day 1)
- Advagraf® 0.2 mg/kg/day p.o. in one dose (conc.: 5-10 ng/ml, after 3 months 4-7), with the initial dose (day 0) 0.1 mg/kg given preoperatively.
- MMF 1gx2 started preoperatively, controlled by a single AUC measurement day 10±5 days, with a target AUC between 40 and 60 mg.h/L
- Steroids day 0 (250 mg methylprednisolone i.v. before start of Thymoglobuline infusion) and day 1 (50 mg methylprednisolone i.v. before start of Thymoglobuline® infusion)

The following treatments were administered for treatment arm B:

- Simulect® induction, i.v. (20mg day 0 and day 4)
- Advagraf® 0.2 mg/kg/day p.o. in one dose (conc.: 5-10 ng/ml, after 3 months 4-7). The initial dose (day 0) 0.1 mg/kg was given preoperatively.
- MMF 1gx2, started preoperatively (controlled by a single AUC measurement day 10±5 days, with a target AUC between 40 and 60 mg.h/L)
- Steroids according to hospital practice but not less than 5 mg prednisolone daily after 6 months.

9.4.2 IDENTITY OF INVESTIGATIONAL PRODUCT(S)

The investigational drug, Thymoglobuline®, at 2.5 mg/kg, marketing authorization number PL 12375/0021, was used in treatment arm A in comparison with standard of care (treatment arm B) including Simulect®, 20 mg, EU number EU/1/98/084, NDC code 0078-0331-84. Batch numbers were not registered, as both Thymoglobulin (study arm) and Simulect (control arm) are approved drugs.

9.4.3 METHOD OF ASSIGNING PATIENTS TO TREATMENT GROUPS

Patients were randomized before transplantation and after providing written informed consent, to one of the two treatment arms using a central web-based computerized system. Assignment of subjects to treatment groups was stratified by donor status (living versus deceased) and by center.

For randomization, the subject was assigned a patient identifier study number with four digits and the treatment assigned. A paper with this information was printed out from the computer screen and stored.

The first digit identified the center and the following three was the number assigned to the patient. The first patient randomized at each the center got number 001 and then it increased with one for each patient.

Please refer to Appendix 16.1.7 for randomization scheme and codes.

9.4.4 SELECTION OF DOSES IN THE STUDY

The doses used in the study were selected based on prior experience in humans (15,16). The two doses of Thymoglobuline were given day 0 during surgery and day 1 in order to minimize side effects.

9.4.5 SELECTION AND TIMING OF DOSE PER EACH PATIENT

Patients were randomized to selected fixed drug and dose regimens. For treatment arm B, steroids were provided according to hospital practice, but not less than 5 mg daily after 6 months.

9.4.6 BLINDING

Blinding is not relevant as this was an open study. However, patient identity and treatment assignment were concealed to the Primary Endpoint Committee, two independent nephrologists who assessed the accuracy of the PTDM diagnosis, and to two pathologists, who centrally evaluated all transplant biopsies.

9.4.7 PRIOR AND CONCOMITANT THERAPY

Patients were instructed to notify the study center about any new medications he/she took after start of the study drug. All medications administered after the patient was enrolled into the study were required to be recorded in the CRF.

9.4.8 TREATMENT COMPLIANCE

Treatment compliance was ensured and documented by the study nurse responsible for the patient in the patient's medical journal.

9.5 EFFICACY AND SAFETY ASSESSMENTS

9.5.1 Efficacy and safety measurements assessed

The specific efficacy and safety variables assessed are described in detail in Appendix 16.1.1 – section 8.3.

9.5.2 Appropriateness of measurements

Assessments described in the protocol are standard assessments for this indication and patient population.

9.5.3 Primary efficacy variable(s)

Cumulative incidence of new onset of diabetes after transplantation

The cumulative incidence of NODAT was defined as adapted from the ADA criteria (7):

- ≥2 Fasting plasma glucose (FPG) ≥7,0 mmol/l ≥ 30 consecutive days apart
- 2-h Plasma Glucose ≥11,1 mmol/l in the OGTT≥ 30 days apart
- Oral hypoglycemic ≥30 consecutive days
- Insulin ≥30 consecutive days

9.6 DATA QUALITY ASSURANCE

The sponsor was responsible for ensuring maintenance of data quality, including that the study was conducted and generated, documented, and reported in compliance with the protocol, GCP, and applicable regulatory requirements.

The study was monitored by an independent monitor at each site. All data in the CRF was monitored in every fifth patient at each site and selected key parameters including serious adverse events (SAEs) were monitored in all patients. Monitors verified that data in the CRF matched source documents and any inconsistencies identified on collected CRFs were clarified using data clarification forms submitted to the monitor or investigator directly.

All lab analyses were performed at the local hospitals per standard protocol and no inter-laboratory standardization methods were employed. The study was monitored to ensure quality and monitoring reports are included in Appendix 16.1.10.

9.7 STATISTICAL METHODOLOGY AND DETERMINATION OF SAMPLE SIZE

Statistical methodology is described in detail in the Statistical Analysis Plan (SAP). Please see section 9.8 for information concerning changes in planned analyses.

9.7.1 DETERMINATION OF SAMPLE SIZE

A total of 222 participants were planned based on the following assumptions, to achieve 80% power for the superiority comparison (Fisher's exact test) on the intention to treet (ITT) population of the primary endpoint between the two treatment groups, with a 2-sided type I error of 5% and allowing for a 5 % drop-out rate:

- Incidence of NODAT was 36% in two phase III studies with Advagraf, MMF, and steroids (17)
- Considered reasonable to assumed that an SA regimen can reduce incidence of NODAT with 50%
- Expected that percentage of subjects who reach the endpoint of NODAT after 12 months will be 18% in the test arm and 36% in the comparator arm.

9.8 CHANGES IN THE CONDUCT OF THE STUDY OR PLANNED ANALYSES

The study protocol was amended once in Sweden and not amended in Denmark; please see Appendix 16.1.1. The key features of this amendment in Sweden were:

- Addition of long-term follow-up visit after 5 years ± 2 years
- A retrospective substudy comparing the incidence of delayed graft function to be conducted

No changes in the planned analyses were made.

10 STUDY PATIENTS

10.1 DISPOSITION OF PATIENTS

Disposition of patients per center is shown in Table 1.

Table 1 Disposition of patients per center and in total.

	Gothenburg		Malmö		Aarhus		Total	
Treatment arm	**SA**	**SM**	**SA**	**SM**	**SA**	**SM**	**SA**	**SM**
Enrolled and randomized	66	65	8	9	40	35	114	110
Treatment failure							1	1
Received intervention (ITT)	65	66	8	9	40	34	113	109
Premature termination		4			5	5	5	9
- Death		2			1		1	2
- Graft loss		1			2		2	1
- Graft loss + death		1					0	1
- Consent withdrawal					2	5	2	5
Completed study (24-months)	65	62	8	9	35	29	108	100
Protocol deviations	38	3	2		1		41	3
Completed study with allocated treatment (PP)	27	59	6	9	34	29	67	97

10.2 PROTOCOL DEVIATIONS

Protocol deviations for group A consisted of addition of steroids (n=40, of which 39 were addition of prednisolone and 1 involved more than three doses of methylprednisolone) or addition of everolimus (n=1).

Protocol deviations for group B involved addition of everolimus (n=3).

Please refer to Appendix 16.2.2 for details regarding protocol deviations.

11 EFFICACY EVALUATION

11.1 DATA SETS ANALYZED

The analysis sets defined as per the protocol and SAP were:

- **Intention To Treat (ITT) population**, consisting of all subjects who were randomized into the study, who received at least one study treatment and have at least one follow-up visit with measurements. Patients were analyzed for efficacy according to their randomized treatment.
- **Per Protocol (PP) population**, consisting of all subjects who were randomized into the study, who received at least one study treatment, who had no major protocol violations and completed the study at 12 months and at 24 months. The PP population was used to assess the robustness of the primary analysis result.

Patient disposition and data sets analyzed are shown below in Table 2.

Table 2. Patient disposition and data sets analyzed

Variable	Arm A (SA arm) (n=113)	Arm B (SM, standard of care) (n=109)
Safety	113 (100.0%)	109 (100.0%)
ITT	113 (100.0%)	109 (100.0%)
PP 3m	89 (78.8%)	108 (99.1%)
PP 6m	82 (72.6%)	107 (98.2%)
PP 12m	77 (68.1%)	104 (95.4%)
PP 24m	67 (59.3%)	97 (89.0%)
For categorical variables n (%) is presented.		

Please see Appendix 16.2.3 for a listing of all patients, visits, and observations excluded from efficacy analyses.

11.2 DEMOGRAPHIC AND OTHER BASELINE CHARACTERISTICS

Demographic and other baseline characteristics are presented in Table 3. Individual patient demographic and baseline data are presented in Appendix 16.2.4.

Table 3 Demographics and baseline characteristics

Characteristic	SA (n=113)	SM (n=109)
Age	52.1 (13.9)	49.2 (14.5)
Age>60	33 (29.2)	28 (25.7)
Females	30 (26.5)	31 (28.4)
BMI	25.9 (3.9)	26.2 (4.0)
Waist-hip ratio	0.98 (0.1)	0.98 (0.1)
Plasma glucose baseline	5.4 (0.7)	5.4 (0.8)
Blood pressure systolic	143.8 (18.3)	143.5 (18.9)
Blood pressure diastolic	85.1 (10.4)	84.9 (11.0)
Cause of End-Stage Kidney Disease (ESKD)		
polycystic kidney disease	38 (33.6)	32 (29.4)
glomerulonephritis	34 (30.1)	32 (29.4)
other defined causes	28 (24.7)	26 (23.9)
undefined cause	13 (11.5)	19 (17.4)
Second transplant	3 (2.7)	0
Deceased donor	63 (55.8)	68 (62.4)
HLA antigen mismatch A; B; DR (mean)	1.1; 1.3; 1.2	1.1; 1.4; 1.1
SA, steroid avoidance; SM, steroid maintenance; Data are n (%) or mean (SD). Between-group differences for demographic and clinical characteristics were not statistically significant, calculated with Fischer's exact test or t-test.		

11.3 MEASUREMENT OF TREATMENT COMPLIANCE

Treatment compliance was documented in medical charts by study nurses. Please refer to Appendix 16.2.5 for details regarding tacrolimus concentration per center.

11.4 EFFICACY RESULTS AND TABULATIONS OF INDIVIDUAL PATIENT DATA

11.4.1 Analysis of efficacy

The incidence of PTDM at 12 months after kidney transplantation was 12.4% in the SA arm vs 18.3% in the SM arm (p=0.3), as shown in Figure 2. Most PTDM events occurred within the first six months after transplantation (see Kaplan-Meier curves in Figure 3). PTDM was resolved in 40.0% of the patients in the SA arm compared to 28.5% of the patients in the SM arm (p=0.72) by 24 months after transplantation (end of study follow-up).

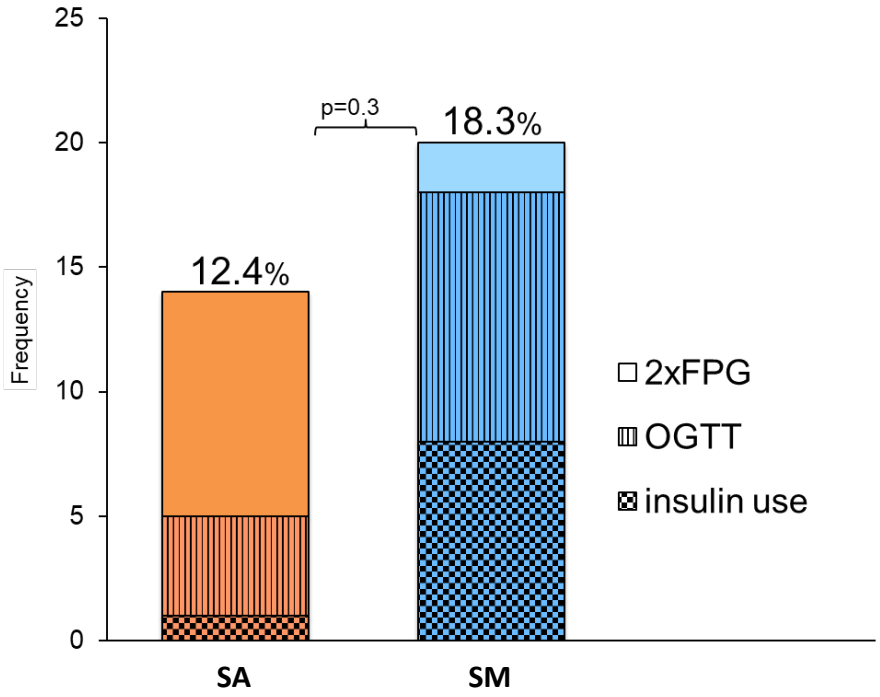

Figure 2 Incidence of PTDM at 1 year per study arm. Based on the diagnostic criteria: FPG ≥ 7,0 mmol/L ≥30 days apart; OGTT, 2-h plasma glucose ≥ 11.1 mmol/L; or insulin treatment ≥ 30 consecutive days. SA, steroid avoidance; SM, steroid maintenance

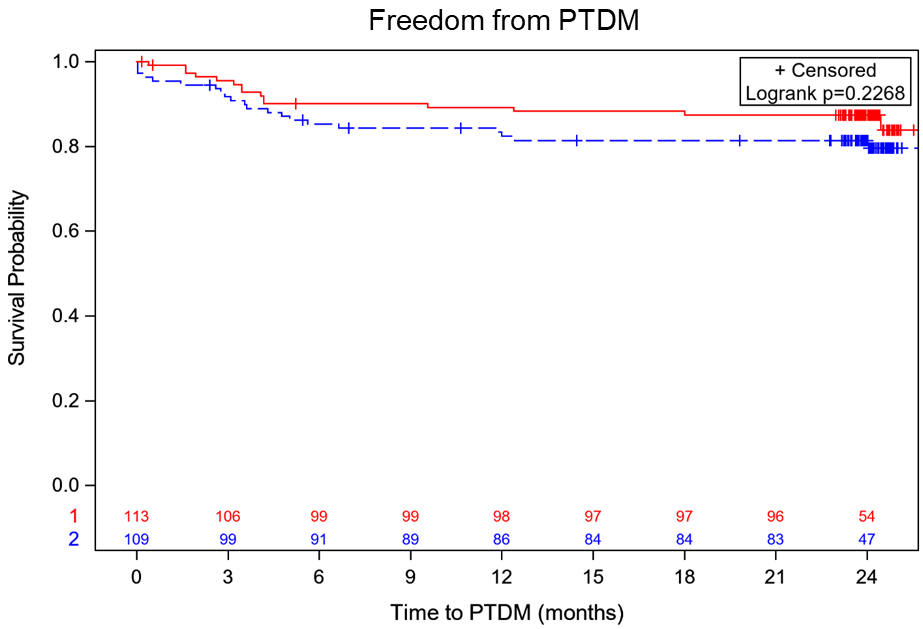

Figure 3 Kaplan-Meier curves of PTDM-free survival up to 24 months after transplantation, per each study arm. Red: SA steroid avoidance; and blue SM, steroid maintenance.

A total of 302 biopsies were performed in 92 patients per arm (184 patients in total). The for-cause:protocol biopsy ratio was 82:75 and 79:75 for the SA and SM arms, respectively (not statistically significant). Thirty-two of 33 patients who experienced rejection were proven by a biopsy; the remaining rejection could not be proven by biopsy due to risk for excessive bleeding. No significant difference was observed between the two arms in incidence of overall BPR at 12 months after transplantation (incidence of 15%, or 15.9% including non-BPR, in the SA arm versus 13.8% in the SM arm, for p-values of 0.85 and 0.70, respectively; see Figure 4). Twenty-three rejections were classified as acute and ten rejections were classified as chronic as per the Banff 2017 classification.

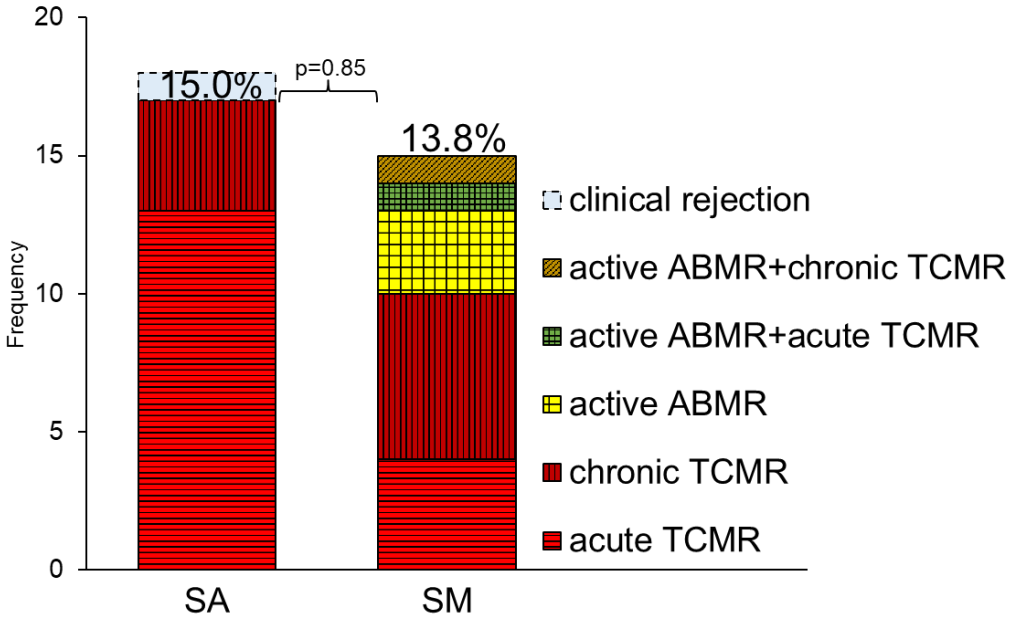

Figure 4 Incidence (%) of biopsy-proven rejection at 1 year according to study arm and type of rejection (Banff 2017 classification). TCMR: T-cell mediated rejection; ABMR: Antibody-mediated rejection. SA, steroid avoidance; SM, steroid maintenance

11.4.2 Statistical/analytical issues

The statistical analysis used is described in detail in Appendix 16.1.9. Briefly, for comparison between groups, dichotomous variables were evaluated using Fischer's exact test, categorical variables using Mantel-Haenszel Chi-squared test, and continuous variables using Fischer's non-parametric permutation test. Statistical significance was set at a p-value of 0.05.Additionally, Kaplan-Meier methods were applied to evaluated time to reach PTDM, AR, graft loss, and death, while independent predictors of PTDM were selected using Cox proportional hazard regression analyses. In the multivariate regression analyses, only age and FPG at baseline were identified as independent predictors or PTDM (Table 4).

Table 4 Multivariate regression analyses

	Univariate prediction		Multivariate prediction	
Variable	**HR** (98% CI)	***P value***	**HR** (98% CI)	***P value***
Age	1.05 (1.02-1.08)	0.0004	1.05 (1.02-1.08)	0.0009
Sex: Female	1.40 (0.70-2.79)	0.35		
BMI	1.09 (1.0-1.18)	0.04		
Waist-Hip Ratio	1.24 (0.79-1.93)	0.35		
Plasma glucose at baseline	1.71 (1.16-1.16)	0.007	1.56 (1.03-2.34)	0.034
Donor: Deceased	2.25 (1.06-4.79)	0.035		
Treatment group: SM	1.50 (0.77-2.91)	0.23		
Mean tacrolimus levels mo 0-3	1.00 (0.89-1.13)	1.00		
BPR at mo 24	0.74 (0.17-3.11)	0.68		
Methylprednisolone after Tx	0.00	0.99		

PTDM, posttransplantation diabetes mellitus; BMI, body mass index; BPR, biopsy-proven rejection. Intention-to-treat analysis.

Additional statistical issues:

- **Handling of drop-outs or missing data**: No imputation was performed for missing values.
- **Interim analysis and data monitoring**: Interim analyses to evaluate safety were conducted after 50 patients had been observed for 6 months post-transplantation. These analyses evaluated the composite measure of freedom from AR, graft survival, and patient survival. Continuation of the study was recommended after review of the interim analysis results by the DMC. No statistical adjustment was required after these analyses.
- **Multicenter/per-center analyses**: All analyses were performed for the entire study (rather than per center).
- **Multiple comparisons**: Not relevant.
- **Use of an "efficacy subset" of patients**: Not relevant. Patients excluded from efficacy analyses are tabulated in Appendix 16.2.3.
- **Active-control studies intended to show equivalence**: Not relevant.
- **Examination of subgroups**: Not relevant.

All tabulations of individual response data can be found in Appendix 16.2.6.

12 SAFETY EVALUATION

12.1 EXTENT OF EXPOSURE

Extent of exposure to the test and comparator drugs are specified in Appendix 16.2.5.

All participants received the study drugs as described above in section 9.4.1.

12.2 ADVERSE EVENTS

Adverse events in this study included the occurrence of infections, major adverse cardiovascular events and malignancies, summarized in Table 5. Incidence of adverse events at 4 months were comparable in the two arms. Specific events per patient are tabulated in Appendix 16.2.7.

Table 5 Occurrence of adverse events at 24 months after transplantation, per arm.

	SA arm (n=113)	SM arm (n=109)	P-value
Infection	73 (64.6)	84 (77.1)	NS
Major adverse cardiovascular event	7 (6.2)	5 (4.6)	NS
Malignancy	7 (6.2)	10 (9.2)	NS

12.3 DEATHS, OTHER SERIOUS ADVERSE EVENTS, AND OTHER SIGNIFICANT ADVERSE EVENTS

Serious adverse events were experienced by 73 patients in the SA arm (64.6%) and 69 patients in the SM arm (63.3%). Specific events per patient are tabulated in Appendix 16.2.7.

One patient died in SA-arm because of pancreas cancer, three patients died in SM-arm due to uraemia (refused dialysis), encephalitis and lung cancer. In total four graft losses, two in each arm, were observed; the causes were primary non-function, thrombosis, uraemia and recurrence of glomerulonephritis in the graft.

12.4 CLINICAL LABORATORY EVALUATION

Results of clinical laboratory evaluations are shown in Appendix 16.4 (Listing 16.2.7). Kidney function as assessed by mGFR (mean) at 12 months was 53.6 ml/min in SA-arm vs. 55.0 ml/min in SM-arm (p=0.55) without any significant deterioration at 24 months – 53.0 ml/min vs. 54.5 ml/min, resp. (p=0.58).

12.5 VITAL SIGNS, PHYSICAL FINDINGS, AND OTHER OBSERVATIONS RELATED TO SAFETY

Not relevant. Listing of vital signs shown in Appendix 16.4 (Listing 16.2.8).

12.6 SAFETY CONCLUSIONS

Although the incidence of PTDM did not significantly differ between the two arms, at up to 24 months after transplantation the SA treatment protocol was not associated with increased risk for BPR. Furthermore, incidence of AEs (infections, malignancies, and major adverse cardiovascular events), kidney function, and survival were statistically similar in the two treatment arms, suggesting that ATG induction to enable avoidance of oral steroids is safe.

13 DISCUSSION AND OVERALL CONCLUSIONS

This phase IV study aimed to evaluate the efficacy and safety of using ATG induction (SA arm) versus basiliximab (SM arm) after kidney transplantation to reduce incidence of PTDM. The study continued to completion at 24 months follow up time and no statistically significant differences were observed between the two arms regarding efficacy or safety. The SAILOR study is the first randomized study, to our knowledge, to provide evidence that complete SA is safe without an increased risk of acute/overall rejections, with no loss of efficacy and achievable in majority of immunologically low-risk kidney recipients over the first two years after transplantation even with low-dose tacrolimus/MMF maintenance regimen. The statistical power calculation in this study assumed a higher PTDM incidence in the SM arm than was observed and thus, a statistically significant difference between the arms could not be achieved, which is a limitation of the study.

Over one-third of the patients in the SA arm (36.1%) were prescribed oral steroids due to proven or suspected rejection, temporal reduction of MMF due to leukopenia, acute tubulointerstitial nephritis, or goat. Nonetheless, 63.9% of the patients in the SA arm remained free of oral steroids at 24 months after transplantation, suggesting that this treatment protocol is safe and feasible in the studied population. Furthermore, although a significant reduction in the incidence of PTDM was not observed with the SA regimen in this selected group at low-risk for diabetes, it may be a preferred treatment option in

those recipients who are deemed high-risk for PTDM. Our currently ongoing five-year follow-up study is expected to provide insights on the impact of complete SA on further long-term outcomes.

14 TABLES, FIGURES, AND GRAPHS REFERRED TO BUT NOT INCLUDED IN THE TEXT

The complete statistical analysis report can be found in Appendix 16.5, including, e.g.,:

- Demographic data summary figures and tables
- Efficacy data summary figures and tables
- Safety data summary figures and tables.

15 REFERENCES

1. Awan AA, Niu J, Pan JS, Erickson KF, Mandayam S, Winkelmayer WC, et al. Trends in the Causes of Death among Kidney Transplant Recipients in the United States (1996-2014). Am J Nephrol. 2018;48(6):472–81.

2. Gaston RS, Cecka JM, Kasiske BL, Fieberg AM, Leduc R, Cosio FC, et al. Evidence for antibody-mediated injury as a major determinant of late kidney allograft failure. Transplantation. 2010 Jul 15;90(1):68–74.

3. Hjelmesaeth J, Hartmann A, Leivestad T, Holdaas H, Sagedal S, Olstad M, et al. The impact of early-diagnosed new-onset post-transplantation diabetes mellitus on survival and major cardiac events. Kidney Int. 2006 Feb;69(3):588–95.

4. Kasiske BL, Snyder JJ, Gilbertson D, Matas AJ. Diabetes mellitus after kidney transplantation in the United States. Am J Transplant. 2003 Feb;3(2):178–85.

5. Valderhaug TG, Hjelmesæth J, Hartmann A, Røislien J, Bergrem HA, Leivestad T, et al. The association of early post-transplant glucose levels with long-term mortality. Diabetologia. 2011 Jun;54(6):1341–9.

6. First MR, Dhadda S, Croy R, Holman J, Fitzsimmons WE. New-onset diabetes after transplantation (NODAT): an evaluation of definitions in clinical trials. Transplantation. 2013 Jul 15;96(1):58–64.

7. Ghisdal L, Van Laecke S, Abramowicz MJ, Vanholder R, Abramowicz D. New-onset diabetes after renal transplantation: risk assessment and management. Diabetes Care. 2012 Jan;35(1):181–8.

8. van Duijnhoven EM, Boots JMM, Christiaans MHL, van Hooff JP. Metabolic aspects of tacrolimus in renal transplantation. Consequences for the choice of an immunosuppressive regimen and for the management of post-transplant diabetes mellitus. Minerva Urol Nefrol. 2003 Mar;55(1):33–42.

9. Haller MC, Royuela A, Nagler EV, Pascual J, Webster AC. Steroid avoidance or withdrawal for kidney transplant recipients. Cochrane Database Syst Rev. 2016 Aug 22;(8):CD005632.

10. Ekberg H, Tedesco-Silva H, Demirbas A, Vítko S, Nashan B, Gürkan A, et al. Reduced exposure to calcineurin inhibitors in renal transplantation. N Engl J Med. 2007 Dec 20;357(25):2562–75.

11. Thomusch O, Wiesener M, Opgenoorth M, Pascher A, Woitas RP, Witzke O, et al. Rabbit-ATG or basiliximab induction for rapid steroid withdrawal after renal transplantation (Harmony): an open-label, multicentre, randomised controlled trial. Lancet. 2016 Dec 17;388(10063):3006–16.

12. Torres A, Hernández D, Moreso F, Serón D, Burgos MD, Pallardó LM, et al. Randomized Controlled Trial Assessing the Impact of Tacrolimus Versus Cyclosporine on the Incidence of Posttransplant Diabetes Mellitus. Kidney Int Rep. 2018 Nov;3(6):1304–15.

13. Woodle ES, First MR, Pirsch J, Shihab F, Gaber AO, Van Veldhuisen P, et al. A prospective, randomized, double-blind, placebo-controlled multicenter trial comparing early (7 day) corticosteroid cessation versus long-term, low-dose corticosteroid therapy. Ann Surg. 2008 Oct;248(4):564–77.

14. Brennan DC, Daller JA, Lake KD, Cibrik D, Del Castillo D, Thymoglobulin Induction Study Group. Rabbit antithymocyte globulin versus basiliximab in renal transplantation. N Engl J Med. 2006 Nov 9;355(19):1967–77.

15. Goggins WC, Pascual MA, Powelson JA, Magee C, Tolkoff-Rubin N, Farrell ML, et al. A prospective, randomized, clinical trial of intraoperative versus postoperative Thymoglobulin in adult cadaveric renal transplant recipients. Transplantation. 2003 Sep 15;76(5):798–802.

16. Gavela Martínez E, Sancho Calabuig A, Escudero Quesada V, Avila Bernabeu AI, Beltrán Catalán S, Morales García AI, et al. Induction treatment with low-dose thymoglobulin or basiliximab in renal transplants from older donors. Transplant Proc. 2008 Nov;40(9):2900–2.

17. Dhadda S, Fitzsimmons W, Croy R, Holman J, First MR. DEFINING NEW-ONSET DIABETES AFTER TRANSPLANTATION (NODAT).: 300. Transplantation. 2010 Jul 27;90:694.

16 APPENDICES

16.1 STUDY INFORMATION

16.1.1 Protocol and protocol amendments

16.1.2 Sample case report form

16.1.3 List of IECs or IRBs

16.1.4 List and description of investigators and other important participants

16.1.5 Signatures of principal investigators and important staff

16.1.6 Listing of patients receiving investigational products from specific batches – not relevant

16.1.7 Randomization scheme and codes

16.1.8 Audit certificates – not relevant

16.1.9 Documentation of statistical methods

16.1.10 Documentation of quality assurance procedures if used

16.1.11 Publications based on the study

16.1.12 Important publications referenced in the report

16.2 PATIENT DATA LISTINGS

16.2.1 Discontinued patients

16.2.2 Protocol deviations

16.2.3 Patients excluded from the efficacy analysis

16.2.4 Demographic data

16.2.5 Compliance and/or drug concentration data

16.2.6 Individual efficacy response data

16.2.7 Adverse event listings (each patient)

16.2.8 Listing of individual laboratory measurements by patient

16.3 CASE REPORT FORMS

16.3.1 CRFs for deaths, other serious adverse events, and withdrawals for adverse events

16.3.2 Other CRFs submitted

16.4 INDIVIDUAL PATIENT DATA LISTINGS

16.5 RESULTS FROM STATISTICAL ANALYSIS REPORT