# THE IMPORTANCE OF EFFECTIVE PLATELET AGGREGATION INHIBITION AFTER ORGAN TRANSPLANTATION

**Doctoral (PhD) Thesis**

**Adam Varga M.D.**

Supervisor: Dr. Peter Szakaly PhD., Med. habil.

Program leader: Prof. Ors Peter Horvath PhD., Sc.D.

Head of Doctoral School: Prof. Gabor L. Kovacs PhD., Sc.D.


Department of Surgery

University of Pecs

2015

# 1. INTRODUCTION

The history of organ transplantation, and of transferring organs and body parts from one man to another, or from an animal to a human being date back thousands of years. One of the earliest and perhaps most well-known story is about Saints Cosmas and Damian, who replaced a severed leg of a verger with a leg from a dead man in the 2nd Century. The story has been portrayed in many paintings; in fact the two are still respected patron saints of today's physicians and surgeons.

In the history of kidney transplantation in the 20th century, a researcher and physician from Pecs played a role as well. Imre Ullmann (Emerich Ullmann) performed successful kidney transplants in a number of animal experiments, but his attempt in 1902, in which he transplanted a goat kidney into a woman, ended unsuccessfully.

Successful and safe kidney transplantation only began in the 1960s and 1970s. The first successful kidney transplant is attributed to Murray et al., who carried out a transplant between identical twin siblings on December 23, 1954 in Boston. In order for organ transplantation to further progress the development of detailed vascular suture techniques and experimental methods of kidney removal and implantation were needed. A solution had to be found for immunological questions with the legal and ethical background of the intervention being ensured as well.

The development of immunosuppression further boosted the progression of transplantation. Initially total body irradiation and high-dose steroid monotherapy were applied with supplementary azathioprine from 1962; however the real breakthrough was brought on by the clinical introduction of cyclosporine.

In this day and age surgical techniques, immunosuppression as well as additional tests leading to a successful surgery (i.e. immunological typing) have evolved to such a level, where long term success of the transplantation is not primarily determined by these factors. Leading causes of post-transplant complications are cardiovascular diseases, infections and cancer. It is thus clear that the improvement of long-term results is based on the prevention of these diseases.

Thanks to advanced immunosuppression, it is possible to improve patient and graft survival compared to what we achieve at present. However, the average age of patients that are operated on keeps growing, thus the long-term survival is expected decline in the future. The paradoxical dichotomy between demand and reality will determine the future of organ transplantation greatly.

## 2. OBJECTIVES

In my research I examined the leading cardiovascular complications after kidney transplantations and simultaneous pancreas and kidney transplantations, the frequency and preventive opportunities of mortality.

**1.** I provide an overview of the complications and long-term results of kidney and simultaneous pancreas-kidney transplantation. My objective was to assess the risk factors among the patients of our clinic, and to compare them with the average population values.

**2.** I review the possibilities of platelet aggregation inhibition, which is one of the cornerstones of cardiovascular prevention. I introduce tests for assessing the effectiveness of the therapy, with special emphasis on optical aggregometry which was used in my own research as well. The phenomenon of resistance against platelet aggregation inhibitors is well-known, so in my research I assess resistance to acetyl-salicylic acid (ASA) and clopidogrel among transplant patients. I compare the results to the results of a group of patients with a history of cardiovascular diseases.

**3.** On top of general factors resulting in resistance known from scientific literature, I analyse the causes of the development of resistance using special variables of our patient material.

**4.** According to the literary references, in case of the emergence of resistance we change preventive treatment, and then check its effectiveness with a re-test.

**5.** I compare cardiovascular mortality and complications from before the beginning of the research to data found in scientific literature, and to the time period lasting from the first aggregometric measurements to now. The results are compared to a control group's data, which has previous positive cardiovascular history.

# 3. ABOUT KIDNEY AND SIMULTANEOUS PANCREAS-KIDNEY TRANSPLANTATIONS AND COMPLICATIONS

## 3.1. The long-term complications of kidney transplantation

A number of factors play a role in the progression of chronic kidney failure: diabetes mellitus, hypertension, smoking and dyslipidaemia. Despite the fact that kidney replacement therapy (haemodialysis, peritoneal dialysis) keeps developing day by day, there is evidence that in terms of patients' quality of life and survival, and also cost-effectiveness, kidney transplantation is the best treatment.
Based on data of a 17-year retrospective study, 1, 5 and 10 year survival rates were significantly higher among transplant patients, compared to those, who were undergoing haemodialysis: 95.2, 88.0 and 78.8% vs. 90.6, 62.7 and 39.8%.

Because of the well-proven surgical technique of kidney transplantation, today we can rarely expect complications to occur. The vast majority of complications after surgery can be attributed to diseases from before the operation or flare ups of immune reactions after the transplantation, and immunosuppression. The long-term results are basically determined by the origin of the organ donor (cadaver vs living donors), surgery, complications developed during the postoperative stage, and aftercare.

It would seem obvious that most commonly occurring side effects of immunosuppressive therapy after transplantation are infections and tumours. But this concept is not correct. The study of Matas et al., which examined 2,202 renal transplant recipients for more than 10 years, revealed the following main causes of death:

- Cardiovascular causes: 35% and 38% (Living and cadaver donors)
- Cancer: 29% and 22%
- Infection: 13% and 9%.

### 3.1.1. Cardiovascular complications

*Cardiovascular risk factors in general*

The main risk factors such as smoking, diabetes mellitus, hypertension, dyslipidaemia, obesity, and certain non-modifiable factors such as age, sex, genetic predisposition play a significant role in this patient group too.

Specific and general cardiovascular risk factors of patients suffering from kidney disease and having undergone kidney transplantation are listed in Table 1 based on medical literature.

| | |
|---|---|
| hypertension (donor and recipient) | hyperuricemia |
| diabetes mellitus | calcium-phosphorus homeostasis disorder |
| hyperlipidaemia | inflammatory diseases |
| obesity | hyperhomocysteinemia |
| increasing age (donor and recipient) | increased trombopoesis |
| sex | poliglobulia |
| genetic predisposition | increased blood clotting |
| microalbuminuria | reduced fibrinolytic activity |
| uraemia | increased haemoglobin levels |
| anti-platelet therapy is less effective | increased blood viscosity |
| graft artery stenosis | higher haematocrit |
| immunosuppression | acute rejection |
| high CRP levels | chronic allograft nephropathy |
| cytomegalovirus infection | metabolic syndrome |

**Table 1:** Cardiovascular risk factors

### 3.2. The characteristics of simultaneous pancreas-kidney transplantation

Type 1 diabetes mellitus is one of the most common chronic diseases among children that have a key role in determining life expectancy. Appropriate treatment of the disease is especially important, otherwise serious complications can occur: microvascular damage, such as retinopathy, neuropathy and nephropathy, and macrovascular damage which affects the cerebrovascular system, peripheral vascular and coronary system as well.

The simultaneous pancreas and kidney transplantation is indicated in patients, with type 1 diabetes mellitus and uraemia or pre-uraemia (serum creatinine is twice as much as the upper limit of the normal ranges). The results of combined transplantation are significantly better than that of dialysis, or kidney transplantation alone.

It is important to take into account post-operative complications, since with their possible prevention and treatment results can be further improved. Rogers et al analysed the results of 156 transplants focusing on two main things: what were the causes of pancreatic and renal graft loss. In case of pancreatic graft loss most were due to early and late thrombosis, but organs were lost because of infections and rejection as well. The leading cause of graft loss after kidney transplantation was chronic allograft nephropathy but infections, graft thrombosis and acute rejection played an important role as well.

A study by Sollinger et al of 1000 post-transplant patients found that 5, 10 and 20 year survival rates were 89, 80, and 58% respectively.

Main causes of mortality after simultaneous transplantation in patients with functioning grafts are cardiovascular diseases. The incidence of cerebrovascular, cardiovascular and peripheral arterial diseases within the first five years after transplantation are 33, 41 and 41% respectively based on a study of a group of patients by Biesenbach. These results further deteriorated (41, 50, 50% respectively) after a period of ten years.

# 4. ANALYSIS OF OUR PATIENT POOL IN LIGHT OF RISK FACTORS

## 4.1. Objectives

The assessment of causes of mortality in patients that underwent transplantation. The assessment of cardiovascular risk factors in patients that underwent transplantation after the commencement of the study. The analysis of data pertaining to kidney or simultaneous kidney-pancreas transplant recipients, and the assessment of cardiovascular risk factors.

## 4.2. Patient pool and methods

Between 3 September 1993 and 28 February 2009 based on our electronic database, the assessment of mortality causes of 586 patients was carried out who underwent transplantation in our facility.

After which all patients that received organ transplants and some form of antiplatelet therapy after the surgery between March 2009 and December 2013 were added to the trial in the Department of Surgery of University of Pecs. Their date are summarised in Table 2.

| **Kidney transplant recipients** | |
|---|---|
| Total | 254 |
| Man | 154 |
| Woman | 100 |
| Average age | 53,87±13,0 years |
| **Simultaneous pancreas-kidney transplant recipients** | |
| Total | 32 |
| Man | 22 |
| Woman | 10 |
| Average age | 47,35±8,57 years |

**Table 2:** Data of study population

In order to assess cardiovascular risk factors the following parameters were determined by the Department of Laboratory Medicine University of Pecs, renal function, fasting glucose, HbA1c, total cholesterol, triglycerides, haemoglobin concentration, haematocrit, platelet count, urinalysis.

At our transplant outpatient clinic blood pressure, weight and height were measured, from BMI values were calculated. The proportion of smokers was determined based on personal interviews. We kept track of their medication and comorbidities on their individual registration sheet. Their data were compared to laboratory reference ranges and average values of the Hungarian population.

### 4.3. Results

In a period stretching from 1993 to 2009 the death of patients having undergone organ transplantation could be traced back to cardiovascular diseases in most cases. They were followed by infections and malignant diseases.

| Mortality cause | Number of patients |
|---|---|
| Cardiovascular disease | 65 (11.09%) |
| Infection | 36 (6.14%) |
| Cancer | 12 (2.04%) |
| Other causes (accident, suicide, surgical complications, etc.). | 9 (1.53%) |

**Table 3:** Causes of mortality in transplant patients (1993-2009)

The data of patients with renal transplant and combined surgeries are summarised separately in the table below. (Table 4 and 5)

| | ***Kidney*** | ***SPK*** | ***Average population*** |
|---|---|---|---|
| **Diabetes mellitus (%)** | 6.3 | N.A. | 7.47 |
| **BMI (kg / m2)** | 26.37 | 28.24 | - |
| **Smoking (%)** | 7.48 | 3.13 | 19 |
| **Increased triglycerides (%)** | 54.33 | 28.13 | 40 |
| **Increased cholesterol levels (%)** | 46.46 | 28.13 | 66 |
| **Antilipid treatment (%)** | 28.35 | 18.75 | ~6 |

**Table 4:** The incidence of cardiovascular risk factors in transplant patients compared to the general population

| | ***Kidney*** | ***SPK*** | ***Local normal lab values*** |
|---|---|---|---|
| **blood glucose level (mmol/L)** | 6.07 | 5.88 | 3.9-5.6 |
| **platelet count** | 239.28 | 283.28 | 150-300 |
| **haematocrit** | 0.385 | 0.394 | 0.37-0.51 |
| **haemoglobin** | 124.91 | 125.59 | 120-180 |

**Table 5:** The incidence of cardiovascular risk factors in transplant patients compared to normal ranges of the local laboratory

**4.4. Discussion**

According to an extensive survey conducted in 2008 the incidence of type I diabetes mellitus was 7.47% in the Hungarian population between the ages of 20 and 69. Even though almost a quarter of patients that underwent kidney transplantation develop diabetes, within our test group the incidence of diabetes requiring treatment was only 6.3%. It is evident that patients undergoing simultaneous transplantation have a 100% incidence of diabetes since the indication of the procedure is having type 1 diabetes. Mean blood glucose values in renal, and simultaneous pancreas- kidney transplant recipients were 6.07 ±1.33 and 5.88±1.89 mmol/L respectively. According to recommendations fasting glucose should be kept below 6.0 mmol/L and postprandial values below 7.5 mmol/L. In both patient groups mean values were kept within recommended ranges. Physiological insulin secretion and long lasting normal insulin levels, which are the real advantages of simultaneous pancreas- kidney transplantation, are best reflected by HbA1c values, which on average were 5.57±0.86%. In diabetic patients a target value below 7.0% is recommended according to guidelines.

40% of the Hungarian population is overweight and 20% exceeds a BMI of 30kg/$m^2$ thus they fall under the 'obese' category. For patients suffering from chronic kidney diseases a value below 25kg/$m^2$ should be targeted whilst patients without cardiovascular symptomes but at great risk thereof should stay below 27kg/$m^2$. Our kidney transplant patients' BMI averages 26.37kg/$m^2$ which falls between the two aforementioned values, and our simultaneous pancreas- kidney transplant patient average 28.24kg/$m^2$ which isn't a significant deviation ($p>0.5$) from the reference value.

Based on a survey conducted in 2013, 19% of the Hungarian population smokes on a daily basis. According to the interviews with our patients only 7.48 and 3.13% of them smoke.

According to a 9000 patient study, 66% of the adult population have cholesterol levels above the upper limit of the normal range which is 5.2 mmol/L. 25% of this group has serum cholesterol levels exceeding 6.5 and 7-8% above 7.8 mmol/L. Mean serum cholesterol was 5.7 mmol/L. Trigliceride levels were above the recommended 2.3 mmol/L(it has since changed) limit in 18% of the population. Recently however the reference value has dropped to 1.7 mmol/L thus the prevalence of trigliceridemia has reached around 40%. Hypercholesterolemia (>5.2 mmol/L) was found in 46.46% and 28.13% of our patients, while hypertrigliceridemia (1.7 mmol/L) was found in 54.33% and 28.13%. 28.35 and 18.75% of our patients recieve antilipid therapy. Comparing these results with our own we can conclude that in Hungary about 6% of the population is on lipid lowering medications (2/3 of which are statins), but it is estimated that every 5th person needs therapy. It is clear that lipid metabolic disorders are common amongst our patients but in their case it is also more often recognised and appropriately treated as opposed to the general population.

Thrombotic events occure more often in patients with higher hematocrit, hemoglobin levels and elevated platelet count. All three parameters were found to be in normal range in both patients groups. The frequent incidence of high blood pressure is not considered significant neither was diabetes in kidney transplant recipients since most patients alredy suffer from these conditions when they first see a transplant surgeon and they rarely cease after transplantation.

All in all we can conclude that according to our assessment apart from trigiceridaemia in kidney transplant recipients no conventional risk factor was found which is clearly linked to the increased cardiovascular mortality of transplanted population, and the increased mortality compared to the general population. We assume that the vascular damage in the pre-transplant uremic period plays an important role in the process and so does the post transplant immunosupression.

# 5. PLATELET AGGREGATION INHIBITION AND DEVELOPMENT OF RESISTANCE

## 5.1. Introduction

Cardiovascular prevention after kidney transplantation is dealt with in detail in the 2009 KDIGO guidelines. They recommend a daily dose of 65-100 mg acetylsalicylic acid if there are no contraindications against it as part of the prevention. This guideline also refers to a retrospective study where low doses of ASA significantly improved graft survival. Grotz et al found that both graft survival and function improved with ASA therapy in kidney transplant patients.

## 5.2. Objectives

In the following section the possibilities of modern platelet aggregation inhibition and also methods capable of determining effectiveness are summarised. After which the definition of ASA and clopidogrel resistance are dealt with. In this part of the study ASA and clopidogrel resistance are assessed using optical aggregometry in our patient group. Data obtained were compared to data found in other scientific literature.

I tried to find answers to the following questions:

- what is the incidence of resistance after transplantation
- and compared to a control group
- and does it contribute to the cardiovascular risk explained in the previous chapter

## 5.3. Platelet aggregation inhibition today

The discovery of the coagulation cascade process dates back to the 1960s, when Davie, Ratnoff and Macfarlane through their experiments found the role of interacting factors and enzymes. Platelets have a key role in the coagulation process, through their adhesion, activation and aggregation as part of the cascade thrombi develop. This process, associated to bleeding, does not only occur with injuries. Platelet activation primarily occurs when normal, linear blood flow changes to turbulent (e.g. sclerotic, narrowed vasculature, valves) and after the erosion and rupture of atherosclerotic plaques. The clots formed this way may cause ischemic damage (e.g.: stroke, acute myocardial infarction).

We intervene in a number of ways to prevent aggregation, with the available therapeutic possibilities (acetylsalicylic acid, clopidogrel, prasugrel, ticagrelor, glycoprotein IIb-IIIa antagonists).

## 5.4. Examination of platelet functions, monitoring the efficiency of aggregation inhibition

There is many therapeutic interventions and methods used to assess the effectiveness of therapy (bleeding time, platelet function analyser, flow cytometry, urine thromboxane test, tromboelastometry / graphy, Verify Now). The gold standard method at the present is platelet aggregometry, which was used in my research as well.

### 5.5. Acetyl salicylic acid and clopidogrel resistance

Resistance to platelet aggregation inhibitors became the centre of attention of clinical trials in the early 2000s.

There are also several positions regarding the definition:

- The laboratory definition of acetylsalicylic acid and clopidogrel resistance is when different platelet function tests show that aggregation inhibition does not correspond with our expectations and the frequency of cardiovascular events is also greater.

- A clinical definition is much simpler: resistance can be established retrospectively if a thrombotic event occurs despite the use of anti-aggregation treatment.

- Recently more and more publications use the concept of residual platelet reactivity (high on-treatment platelet reactivity). Objective criteria are necessary for definition. The goal is to numerically determine the activity of target receptors before and after treatment. The usability of the term presents a problem, since the device used for the measurement is still not standardised and neither is the time thereof.

The degree of resistance to both ASA and clopidogrel was the subject of several studies. In most studies, different methods and therapeutic regimens with different drug doses used are compared, however, in both cases drug resistance can be as much as 40-50%.

### 5.6. Patients and methods

Patients' data are summarized in Table 2.

214 of the 254 renal transplant patients was given a daily dose of 100 mg acetylsalicylic acid while 40 received for other cardiovascular indications 75 mg clopidogrel daily. All the 32 simultaneous pancreas- kidney transplant recipients were given ASA. Patients taking either products were required to do so for a minimum of 3 months.

A blood sample needed for measuring platelet aggregation was drawn from the cubital vein in the morning on empty stomach into 3.8% Na citrate Vacutainer tubes. Antiplatelet drugs were taken a day before blood collection. The test was performed at 1st Department of Internal Medicine, University of Pecs. Following multiple courses of centrifugation first platelet rich (PRP) then platelet poor plasma (PPP) was obtained; platelet aggregation was induced with ADP (concentration: 5mM and 10 mM), and with epinephrine (10mM). The test was performed using a Carat TX-4 platelet aggregometer (Carat Diagnosztika Kft., Budapest) which works on the principle of turbidimetry, and calculates the degree of aggregation relying on the difference between PPP-PRP optical densities. We use epinephrine to examine the effect of ASA and ADP to assess clopidogrel effectivity. A decrease in maximum aggregation is considered to be the consequence of therapy, if it is not in target range of the normal population (mean±2SD), otherwise therapy is deemed ineffective. ASA treatment is considered effective as long as the degree of induced aggregation is not over 40%, or 50% with clopidogrel.

In order to get even more accurate results treatment was considered ineffective if two consecutive tests also confirmed inappropriate aggregation inhibition.

Comparison with resistance data of other patient groups was carried out using two sample t test with the help of IBM SPSS software (IBM Corporation, Armonk, New York, United States), Version 21.0.

### 5.7. Results

| | | |
|---|---|---|
| **Renal transplant recipients** | 254 | |
| ASA resistant | 86 | **40.18%** |
| Not resistant ASA | 128 | 59.81 |
| Clopidogrel resistant | 24 | **60%** |
| Not resistant to clopidogrel | 16 | 40% |
| **Simultaneous pancreas- kidney transplant recipients** | 32 | |
| ASA resistant | 13 | **40,63%** |
| Not resistant to ASA | 19 | 59.37% |
| **Total number of transplant recipients** | 286 | |
| Resistant | 123 | **43%** |
| Not resistant | 163 | 57% |

**Table 6:** transplant patients acetylsalicylic acid and clopidogrel resistance

Based on the overall results of the transplanted population we examined we see that in 43% (123 patients) therapy was ineffective.

Comparing our patients' resistance values with data found in scientific literature it is evident that the incidence of resistance after transplantation is significantly higher ($p<0.05$) than in other examined patient groups.

### 5.8. Discussion

High resistance measured in our patients may contribute to the more frequent incidence of cardiovascular complications, and can be a cause of increased cardiovascular mortality. That is why it is important to explore the underlying mechanisms of the phenomenon and, if resistance is present convert to a different drug.

# 6. POSSIBLE CAUSES OF ACETYLSALICYLIC ACID RESISTANCE AND CLOPIDOGREL RESISTANCE

## 6.1. Introduction

Because of the high prevalence of resistance to platelet aggregation inhibitors many research groups are looking for the causes of the phenomenon. When designing my own research one of my goals was to find a factor that may explain the increased resistance among transplant patients. First, I collected available date from scientific literature, and I completed them with the analysis of certain factors only typical for our patient group (e.g. immunosuppressive drugs).

## 6.2. Patients and methods

254 patients were selected into the study that was previously enrolled into our aggregometric examination, furthermore another 32 patients that underwent simultaneous pancreas- kidney transplantation (Table 2).

The following parameters were determined from the serum of the subjects in the Department of Laboratory Medicine, University of Pécs: creatinine, glucose, cholesterol, triglycerides, haemoglobin concentration, haematocrit, and platelet count.
In our transplant outpatient clinic patients' weight, height and BMI (calculated from the other two parameters) were measured. Smokers were identified during personal interviews, and so were medicines used in therapy that may have affected resistance according to literature. The frequency of chronic allograft nephropathy and steroid resistant acute rejection were also assessed.

A total of 24 variables were analysed which are summarized in Table 7.

The statistical analysis of the available data was carried out using the 21.0 version of the IBM SPSS software (IBM Corporation, Armonk, New York, United States) with logistic regression analysis.

| | | |
|---|---|---|
| age | mycophenolate mofetil | ACE inhibitors |
| Se creatinine level | methylprednisolone | CAN |
| diabetes mellitus | tacrolimus | SRAR |
| type of transplantation | cyclosporine | platelet count |
| BMI | mycophenolic acid | haematocrit |
| Se triglyceride level | everolimus | Se glucose |
| Se cholesterol level | sirolimus | haemoglobin |
| anti-lipid therapy | Calcium channel blockers | smoking |
| ARB | | |

**Table 7**: We investigated the possible factors associated with resistance

### 6.3. Results

Statistically significant correlation could only be detected in the group taking ASA. In patients receiving anti-lipid therapy (statin therapy) resistance occurred at lower rates. In patients taking cyclosporine containing products resistance occurred at a significantly higher rate as opposed to those not taking it (Table 8.) In the clopidogrel treatment group none of the examined factors were found to affect resistance significantly.

| | *beta coefficient* | *p value* | *OR* | *95% C.I. for OR* | |
|---|---|---|---|---|---|
| | | | | *lower* | *upper* |
| **BMI** | -,002 | ,980 | ,998 | ,865 | 1,152 |
| **hematocrit** | -,006 | ,965 | ,994 | ,772 | 1,280 |
| **platelet count** | ,000 | ,906 | 1,000 | ,995 | 1,005 |
| **type of transplant** | -,251 | ,774 | ,778 | ,140 | 4,320 |
| **methylprednisolone** | -,174 | ,616 | ,840 | ,426 | 1,658 |
| **CAN** | ,522 | ,562 | 1,685 | ,289 | 9,813 |
| **ARB** | -,311 | ,417 | ,733 | ,346 | 1,552 |
| **age** | ,010 | ,445 | 1,010 | ,985 | 1,036 |
| **tacrolimus** | ,424 | ,485 | 1,529 | ,465 | 5,031 |
| **everolimus** | ,472 | ,418 | 1,603 | ,512 | 5,022 |
| **Se triglyceride level** | ,301 | ,371 | 1,351 | ,699 | 2,609 |
| **diabetes mellitus** | -,742 | ,379 | ,476 | ,091 | 2,485 |
| **Se cholesterol level** | -,273 | ,393 | ,761 | ,406 | 1,425 |
| **Se glucose** | ,120 | ,362 | 1,128 | ,871 | 1,461 |
| **smoking** | ,592 | ,327 | 1,807 | ,553 | 5,906 |
| **hemoglobine level** | -,011 | ,272 | ,990 | ,971 | 1,008 |
| **SRAR** | -1,172 | ,232 | ,310 | ,045 | 2,115 |
| **mycophenolic acid** | ,877 | ,220 | 2,403 | ,592 | 9,760 |
| **mycophenolate mofetil** | ,198 | ,546 | 1,219 | ,641 | 2,316 |
| **ACE inhibitors** | ,436 | ,174 | 1,547 | ,824 | 2,903 |
| **sirolimus** | ,645 | ,168 | 1,907 | ,761 | 4,776 |
| **Calcium channel blockers** | -,527 | ,108 | ,590 | ,310 | 1,123 |
| **Se creatinine level** | -,004 | ,073 | ,996 | ,993 | ,999 |
| **anti-lipid therapy** | ,682 | **,050** | 1,979 | ,990 | 3,955 |
| **cyclosporine** | -1,042 | **,001** | ,353 | ,192 | ,649 |

**Table 8:** Results of multiple variable logistic regression of those receiving ASA therapy (BMI: body mass index, CAN: chronic allograft nephropathy, ARB: angiotensin receptor blockers, SRAR: steroid-resistant acute rejection)

### 6.4. Discussion

Comparing the results of my own research and data found in literature we come to the conclusion that my research failed to confirm the hypotheses raised by other research groups. The strength of our results lies in the fact that our study is based on large number of cases as opposed to studies of small groups mentioned in the introduction. No other study has been published up to this point regarding transplant patients that deals with this topic in such depths.

We can establish that receiving statin therapy in order to reduce cholesterol levels improves cardiovascular prevention in several ways, since the incidence of acetylsalicylic acid resistance is lower in this group. This is another component that can be added to the pleiotropic effects of statin therapy.

Calcineurin inhibitor cyclosporine A has a number of serious side effects as well. Its nephrotoxic, hypertension and hyperlipidaemia inducing effects are well known so is its ability to increase cardiovascular mortality. According to the result of this study it also reduces the efficacy of ASA therapy.

# 7. TREATMENT OPTIONS FOR PATIENTS WITH RESISTANCE

## 7.1. Introduction

In previous chapters the possible causes of resistance to aggregation inhibitors was described in detail furthermore why it is especially important to apply this type of therapy in populations having undergone transplantation. The treatment options for resistant patients have been examined by multiple studies. The overall conclusion is that in case of ASA resistance, according to most studies, conversion to clopidogrel appears to be a good choice or complementing ASA therapy with clopidogrel and if clopidogrel resistance occurs increasing doses or conversion to prasugrel is acceptable.

## 7.2. Objectives

My aim was to modify therapy of patients resistant to antiplatelet treatment relying on scientific literature, and to assess efficacy of the new therapy using aggregometric measurements.

## 7.3. Patients and methods

A follow-up study was carried out relying on groups of kidney and simultaneous pancreas-kidney transplant recipients earlier described in my thesis (Table 2).
Out of the 123 patients, resistant to either ASA or clopidogrel, a total of 118 underwent control aggregometry. 96 of them instead of the 100 mg ASA therapy that proved to be ineffective got 75 mg clopidogrel a day, and 22 of them received 150 mg clopidogrel instead of the previous dose of 75 mg.
Aggregometriy were performed as previously described.

## 7.4. Results

Conversion to ASA or clopidogrel brought the following results: in 41 patients (42.70%) aggregation inhibition was effective and in 55 patients (57.29%) the therapeutic change was ineffective (Diagram 1).

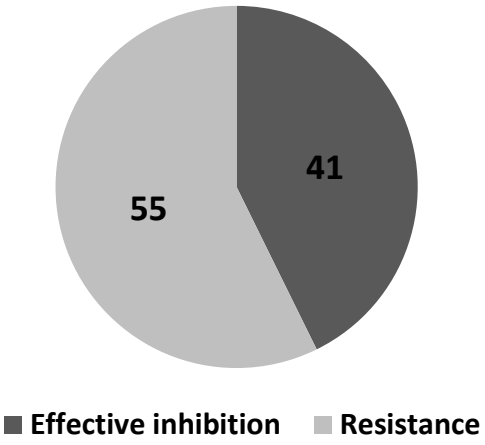


**Diagram 1:** Results of conversion ASA-clopidogrel

Increased clopidogrel doses was effective in 9 patients (40.90%), but in 13 (59.09%) patients no proper aggregation inhibition could be detected, not even with a daily dose of 150 mg (Diagram 2).

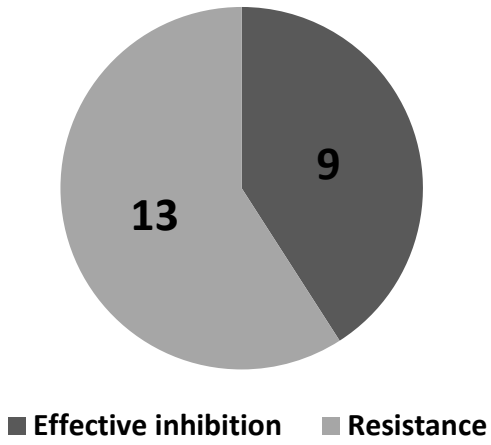


**Diagram 2:** The result of increased clopidogrel doses

**7.5. Discussion**

When deciding upon therapeutic changes of resistant patients we rely on data from literature and practical skills of the cardiology clinic performing aggregometric measurements. The results show that conversion or increasing doses was only effective in approximately 40% of the cases.

Comparing our ASA-clopidogrel conversion results with international data it is not surprising. Lev et al studied double-resistance of patient groups having had percutaneous coronary interventions and they found that 47.4% of the ASA resistant population was also resistant to clopidogrel.

Clopidogrel resistance after the increase in doses was studied by Aleil et al as well. While with a dose of 75 mg the prevalence of resistance 33.7%, it dropped to 8.6% in the group receiving 150 mg a day (58 patients). Severe bleeding occurred in neither groups, and there were no significant discrepancies between two groups in terms of minor bleedings either. In summary, the changes made in resistant patients' therapy did not always lead to efficient inhibition of aggregation. In sight of scientific literature it was not expected to happen. It is a positive result however that half of the patients that previously received ineffective therapy can have, through regular aggregometric check-ups and on-time change in therapeutic drugs, effective platelet inhibition.

The group still showing signs of resistance may benefit from switching to prasugrel or ticagrelor.

Both agents have already been through a number of multi-centre trials that have proved their efficacy and lower rates of resistance.

Due to current prescription rules we could not put our patients on the aforementioned therapy.

# 8. ASSESMENT OF CARDIOVASCULAR MORTALITY AND MORBIDITY AFTER TRANSPLANTATION

## 8.1. Introduction

Assessment of resistance to antiplatelet drugs and the consequent therapeutic modifications are implemented in order to decrease cardiovascular morbidity and mortality. We know what the most significant factors are that contribute to mortality in transplant patients, consequently we also know that cardiovascular prevention is essential in these patients. Prevention is also important because the number of implanted organs does not increase proportionally with the number of patients waiting for transplantation, thus protecting transplanted organs is extremely important.

## 8.2. Objective

The analysis morbidity and mortality rates of transplant patients and patients with a positive cardiovascular history. The comparison of cardiovascular mortality before and after our investigation, based on which we can determine the efficacy of necessary therapeutic modifications and regular aggregometric check-ups. Major cardiovascular events were summarised within the study period (stroke and acute myocardial infarction).

## 8.3. Patients and methods

The data of patients (286 simultaneous pancreas- kidney transplant recipients and renal transplant recipients) who underwent earlier aggregometric examinations were analysed based on documents from ambulatory care and the hospital computer system (MedSolution®). A gender and age suitable control group was selected from the 1st Department of Internal Medicine, University of Pecs with positive cardiovascular history (acute myocardial infarction, cerebrovascular events). The study period started in March 2009 and ended in December 2013. The comparison criteria were as follows: mortality, recent acute coronary syndrome (ACS) and the occurrence of stroke, hypertension, and diabetes. Using these criteria data of groups in Figure 1 were examined. The statistical analysis was with chi-square test using the 21.0 version IBM SPSS software (IBM Corporation, Armonk, New York, United States).

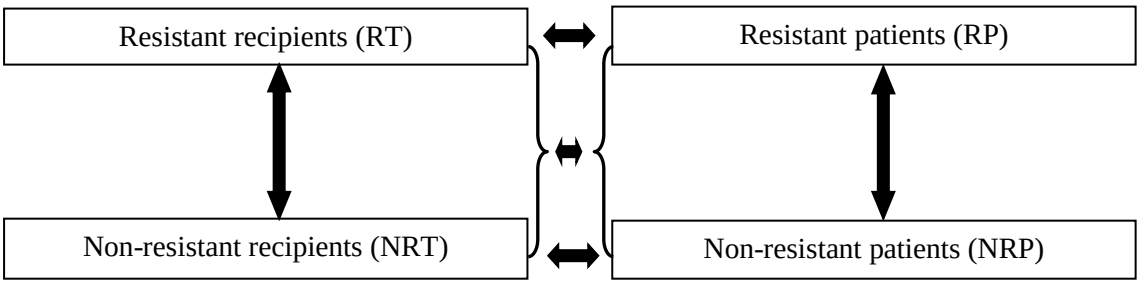


**Figure 1:** Groups compared through statistical analysis

## 8.4. Results

Transplant recipients' resistance to aggregation inhibitors is significantly higher than that of the control group ($p < 0.002$).

The incidence of acute coronary syndrome is also more common ($P < 0.03$) in transplant patients. During a 16.5 year period before the start of our study out of the 586 transplants patients 65 died due to cardiovascular causes. Between March 2009 and December 2013 in a span of 4.5 years out of 286 patients participating in our study

seven died due to the same causes. If we made a mathematical comparison between the two studies there would be a significant decrease in cardiovascular mortality. But because of the different timing of the studies and differences in terms of age, therapy and cardiovascular risk factors of patients they are not scientifically comparable. Most major cardiovascular events registered in clinical trials are stroke and acute myocardial infarction. 7 of our patients had stroke, and 11 AMI.

The following tables summarize the results of the statistical analysis.

| | cases / all recipients | cases / all patients | p | significance |
|---|---|---|---|---|
| **Resistance** | 123/286 | 91/346 | 0,002 | **S** |
| **Mortality** | 7/286 | 5/346 | 0,36 | NS |
| **Recent ACS** | 11/286 | 4/346 | 0,03 | **S** |
| **Recent stroke** | 7/286 | 7/346 | 0,72 | NS |
| **Hypertension** | 270/286 | 240/346 | 0,009 | **S** |
| **Diabetes** | 48/286 | 72/346 | 0,28 | NS |

**Table 9:** Morbidity and mortality results of the analyzed population

| | cases / RT | cases / NRT | p | significance |
|---|---|---|---|---|
| **Mortality** | 3/123 | 4/163 | 0,99 | NS |
| **Recent ACS** | 8/123 | 3/163 | 0,05 | **S** |
| **Recent stroke** | 6/123 | 1/163 | 0,02 | **S** |
| **Hypertension** | 118/123 | 152/163 | 0,86 | NS |
| **Diabetes** | 20/123 | 28/163 | 0,86 | NS |

**Table 10:** Morbidity and mortality results of the transplanted patients regarding resistance

| | cases / RP | cases / NRP | p | significance |
|---|---|---|---|---|
| **Mortality** | 2/91 | 3/255 | 0,49 | NS |
| **Recent ACS** | 2/91 | 2/255 | 0,28 | NS |
| **Recent stroke** | 3/91 | 4/255 | 0,32 | NS |
| **Hypertension** | 64/91 | 176/255 | 0,92 | NS |
| **Diabetes** | 13/91 | 59/255 | 0,14 | NS |

**Table 11**: Morbidity and mortality results of the positive control population regarding resistance

| | cases / RT | cases / RP | p | significance |
|---|---|---|---|---|
| **Mortality** | 3/123 | 2/91 | 0,91 | NS |
| **Recent ACS** | 8/123 | 2/91 | 0,15 | NS |
| **Recent stroke** | 6/123 | 3/91 | 0,58 | NS |
| **Hypertension** | 118/123 | 64/91 | 0,13 | NS |
| **Diabetes** | 20/123 | 13/91 | 0,73 | NS |

**Table 12:** Morbidity and mortality results of the resistant patients

| | cases / NRT | cases / NRP | p | significance |
|---|---|---|---|---|
| **Mortality** | 4/163 | 3/255 | 0,32 | NS |
| **Recent ACS** | 3/163 | 2/255 | 0,33 | NS |
| **Recent stroke** | 1/163 | 4/255 | 0,38 | NS |
| **Hypertension** | 152/163 | 176/255 | 0,04 | **S** |
| **Diabetes** | 28/163 | 59/255 | 0,23 | NS |

**Table 13:** Morbidity and mortality results of the non-resistant patients

### 8.5. Discussion

The transplantation has a dual purpose: on the one hand it substitutes a not functioning organ with a functioning one, and secondly it provides better graft survival in the long run. There are basically two possible outcomes of renal transplantation. The recipient either loses their functioning graft, or they die with it. In the latter case the leading causes of mortality are cardiovascular diseases, as in the normal population. Our primary aim is not to reduce this mortality rate, but to delay the time of its occurrence.

The general trend is that the average age of those waiting for transplantation is getting higher year by year and also an increasing proportion of them have diabetes. A direct consequence of these factors is the increase in cardiovascular risk. It is important to maintain patients' cardiovascular status that has lost their grafts, so that they remain candidates for transplantation.

The comparison of the control and our patient groups confirms that cardiovascular diseases which are the leading cause of mortality in the general population are even more frequent in transplant recipients. The transplant group's risks are further worsened by the increased resistance to platelet aggregation inhibitors that are even greater than that of a patient group at high cardiovascular risk.

Based on the results we can see that for during the study period cardiovascular morbidity and mortality decreased compared to the period prior to our investigation. Therapeutic modification control of aggregation inhibition are not the only things that helped achieving this but also other conditions (e.g.: hypertension, hyperlipidaemia) recognised during ambulatory visits and background check and modify the therapeutic efficacy of the anti-aggregation treatment.

The result contributes to the treatment of other medical conditions detected during ambulatory care (e.g.: hypertension, hyperlipidaemia) as well. On top of this the study demonstrated the benefits of routine aggregometric measurements.

It was shown in our patients that the incidence of acute coronary syndromes is more frequent in case of ASA resistance.

The incidence of post-transplant acute myocardial infarction Lentine et al found that the incidence of myocardial infarction was 4.3% -5.6% -11.1%, 6-12-36 months after renal transplantation.

Because of atherosclerosis caused by a period of uraemia before renal transplantation, the risk of stroke was also increased in these patients. This phenomenon was also observed in in our patient group. Selinger et al found that the risk of stroke in end stage renal failure is 5-10 times higher than in patients with normal kidney function. It is therefore not surprising that cerebrovascular events are leading causes of mortality even after transplantation.

## 9. SUMMARY OF THE NEW SCIENTIFIC RESULTS

**1.** It is the first time for a research of such large patient numbers to demonstrated that the incidence of resistance to platelet aggregation inhibitors in the transplanted population is significantly higher than in the average population. This factor can be looked at as a new cardiovascular risk factor.

**2.** Our measurements have proven that the number of patients resistant to aggregation inhibitors can be decreased if we convert ASA-resistant patients from ASA to clopidogrel, or increase doses of clopidogrel in resistant patients.

**3.** According to our measurements cyclosporine plays a role in acetylsalicylic acid resistance. Increased resistance is observed in our patients when they are used simultaneously.

**4.** My study has demonstrated that statin therapy does not only reduce lipid levels, but it is also beneficial in reducing resistance.

**5.** Based on these findings regular aggregometry can be suggested in the transplant population, so as to recognize resistance to aggregation inhibitors in time, and to be able to implement therapeutic changes accordingly. Consequently, the number of cardiovascular mortality and complications can be reduced or the time of their development delayed.

# 10. PUBLICATIONS OF THE AUTHOR

**Articles - In connection with Thesis)**

1. **Varga A**, Sandor B, Nagy KK, Viola M, Toth A, Gombos K, Toth K, Szakaly P. Acetylsalicylic Acid Resistance After Renal Transplantation. In Vivo 2015 01-02;29(1):141-144. IF: 1,148

**Varga A**, Sandor B, Kalmar Nagy K, Praksch D, Gombos K, Toth A, Toth K, Szakaly P. Clopidogrel Resistance After Renal Transplantation. In Vivo 2015 03-04;29(2):301-303. IF: 1,148

3. Sandor B, **Varga A**, Rabai M, Toth A, Papp J, Toth K, Szakaly P: Aspirin resistance as cardiovascular risk after kidney transplantation. Korea-Australia Rheology Journal 26:(2) pp. 237-241. (2014) IF: 0.632

**Presentations (In connection with Thesis)**

1. **Varga Á**, Kalmár Nagy K, Viola M, Sándor B, Tóth A, Juricskay I, Tóth K, Szakály P: A cardiovascularis betegségek és az acetil-szalicilsav rezisztencia összefüggései veseátültetés után. Magyar Transzplantációs Társaság Kongresszusa, Hajdúszoboszló, 2013. november 21-23.

2. **Varga Á**, Kalmár Nagy K, Viola M, Sándor B, Tóth A, Juricskay I, Tóth K, Szakály P. Thrombocyta aggregáció gátlásról általánosságban és transzplantáltak körében. Családorvos Kutatók Országos Szervezetének konferenciája, Pécs, 2014. március 1.

3. **Varga A**, Sandor B, Kalmar Nagy K, Toth A, Toth K, Szakaly P. The relationship between cardiovascular diseases and acetlysalicylic acid resistance after kidney transplantation. ERA-EDTA Congress, Amsterdam, 2014. május 31. - június 3.

4. Sándor B, Tóth A, Juricskay I, **Varga Á**, Tóth K, Szakály P. Vesetranszplantált páciensek acetil-szalicilsav rezisztenciája: összehasonlítás a rezisztencia populációs adataival. MST Kísérletes Sebészeti Szekció Kongresszusa, Debrecen, 2013. június 13-15.

5. Sandor B, Toth A, Juricskay I, **Varga A**, Rabai M, Papp J, Toth K, Szakaly P. Aspirin resistance after kidney transplantation. Conference of the European Society for Clinical Hemorheology and Microcirculation, Pécs, 2013. július 6-9.

6. Praksch D, Sándor B, **Varga Á**, Rábai M, Szakály P, Tóth K. Aspirin rezisztencia vizsgálata vesetranszplantáción átesett betegek körében. A Magyar Haemorheológiai Társaság, a Magyar Mikrocirkulációs és Vaszkuláris Társaság és a Magyar Szabadgyök-kutató Társaság IV. közös kongresszusa. Balatonkenese, 2014. április 4-5.

7. Praksch D, Sándor B, **Varga Á**, Rábai M, Szakály P, Tóth K: Vesetranszplantált betegek ASA-rezisztenciája: összehasonlítás a populációs rezisztencia adatokkal. Magyar Kardiológiai Társaság Kongresszusa, Balatonfüred 2014. május 14-17.

8. Praksch D, Csiszár B, Sándor B, **Varga Á**, Rábai M. Aspirin kezelés hatékonyságának felmérése vesetranszplantált betegekben. TDK Konferencia Pécs 2014. április 3-4.
9. Praksch D, Sandor B, **Varga A**, Szakaly P, Toth K. Survey of the effect of Aspirin on kidney transplant patients. The Scientific Meeting of the Japan-Hungary-Poland Surgical Society JHPSS Triangle Symposium, Tokyo, Japan, 2014. október 17-19.

**Other presentations**

1. Laszlo E, Kiss P, Szakaly P, Kovacs K, **Varga A**, Degrell P, Jancso G, Horvath G, Hashimoto H, Lubics A, Reglodi D. The role of endogenous PACAP in the kidney during ischaemia-reperfusion. The 11th International Symposium on VIP, PACAP and Related Peptides, Pécs, 2013. Augusztus 27-31.

2. László E, **Varga Á**, Degrell P, Kiss P, Szakály P, Tamás A, Reglődi D. Effect of exogenous PACAP on ischaemia-reperfusion induced kidney injury in famele rats. 10th János Szentágothai Transdisciplinary Conference and Student Competition. Pécs, 2013. november 4-5.

3. László E, **Varga Á**, Degrell P, Kiss P, Reglődi D, Tamás A, Szakály P. Az exogen PACAP hatásának vizsgálata ischaemia-reperfusio során a vesében. Magyar Transzplantációs Társaság Kongresszusa, Hajdúszoboszló, 2013. november 21-23.

4. László E, **Varga Á**, Degrell P, Kiss P, Reglődi D, Szakály P. Effect of exogenous PACAP on ischaemia-reperfusion induced kidney injury. IBRO Workshop, Debrecen 2014. január 16-17.

5. László E, **Varga Á**, Degrell P, Kiss P, Reglődi D, Tamás A, Szakály P. A PACAP hatásának vizsgálata nőstény patkányok ischaemia-reperfusiós vesekárosodásában. VI. Nemzetközi és XII. Országos Interdiszciplináris Grastyán Konferencia, Pécs, 2014. március 19-21.

6. László E, **Varga Á**, Kovács K, Degrell P, Kiss P, Szakály P, Jancsó G, Tamás A, Reglődi D. Az endogen PACAP protektív hatásának vizsgálata ischaemia-reperfusio okozta vesekárosodásban. 44. Membrán-Transzport Konferencia, Sümeg, 2014. május 20-23.

7. László E, **Varga Á**, Degrell P, Kovács K, Kiss P, Szakály P, Tamás A, Reglődi D. Protective effect of exogenous PACAP on ischaemia/reperfusion-induced kidney injury of female rats. 20th International Symposium on Regulatory Peptides Kyoto, 2014. szeptember 7-10.

8. **Varga Á**, Viola M, Kalmár Nagy K, Szakály P. Tapasztalataimk 1 évvel a teljes körű Eurotransplant csatlakozás után. Magyar Transzplantációs Társaság Kongresszusa, Zalakaros, 2014. november 20-22.

9. László E, **Varga Á**, Degrell P, Kovács K, Kiss P, Tamás A, Reglődi D, Szakály P: A PACAP védő hatásának összehasonlító vizsgálata nőstény és hím patkányok ischaemia-reperfusios vesekárosodásában. Magyar Transzplantációs Társaság Kongresszusa, Zalakaros, 2014. november 20-22.

10. **Varga Á**, Végh M, Weisz F. Segítő kezek-jelnyelvi kommunikáció az orvosi praxisban Családorvos Kutatók Országos Szervezetének Kongresszusa, Mórahalom, 2013. február 22-23.

11. Tutervai P, **Varga Á**, Végh M. Segítő kezek- jelnyelvi kommunikáció az orvosi praxisban 2. Családorvos Kutatók Országos Szervezetének Kongresszusa, Pécs, 2014. február 22-23.

12. Tutervai P, **Varga Á**, Végh M. Segítő kezek– Jelnyelvi kommunikáció az orvosi praxisban. TDK Konferencia, Pécs, 2014. április 3-4.

**Abstracts**

1. Sándor B, Tóth A, Juricskay I, **Varga Á**, Tóth K, Szakály P. Vesetranszplantált páciensek rezisztenciája acetilszalicilsav-kezelésre: összehasonlítás a rezisztencia populációs adataival. Magyar Sebészet 66. évfolyam, 2. szám, április, 106-107., 2013.

2. Sándor B, Bíró K,Tóth A, Juricskay I, **Varga Á**, Rábai M, Papp J, Tóth K, Szakály P. Aspirin resistance after kidney transplantation. Clin Hemorheol Microcirc, 54, 139-40, 2013

IF: 2,215

3. Laszlo E, Kiss P, Szakaly P, Kovacs K, **Varga A**, Degrell P, Jancso G, Horvath G, Hashimoto H, Lubics A, Reglodi D: The role of endogenous PACAP in the kidney during ischaemia-reperfusion. Journal of Molecular Neuroscience 51: (suppl 1), 215-6, 2013 IF: 2.757

4. László E, **Varga Á**, Degrell P, Kiss P, Szakály P, Tamás A, Reglődi D. Effect of exogenous PACAP on ischaemia-reperfusion induced kidney injury in famele rats. 10th János Szentágothai Transdisciplinary Conference and Student Competition. Pécs, 2013. november 4-5. Abstract book

5. **Varga Á**, Kalmár Nagy K, Viola M, Sándor B, Tóth A, Juricskay I, Tóth K, Szakály P. A cardiovascularis betegségek és az acetil-szalicilsav rezisztencia összefüggései veseátültetés után. Magyar Transzplantációs Társaság Kongresszusa, Hajdúszoboszló, 2013. november 21-23. Absztrakt könyv

6. László E, **Varga Á**, Degrell P, Kiss P, Reglődi D, Tamás A, Szakály P. Az exogen PACAP hatásának vizsgálata ischaemia-reperfusio során a vesében. Magyar Transzplantációs Társaság Kongresszusa, Hajdúszoboszló, 2013. november 21-23. Absztrakt könyv

7. László E, **Varga Á**, Degrell P, Kiss P, Reglődi D, Szakály P. Effect of exogenous PACAP on ischaemia-reperfusion induced kidney injury. IBRO Workshop, Debrecen 2014. január 16-17. Abstract book

8. László E, **Varga Á**, Degrell P, Kiss P, Reglődi D, Tamás A, Szakály P. A PACAP hatásának vizsgálata nőstény patkányok ischaemia-reperfusiós vesekárosodásában. VI. Nemzetközi és XII. Országos Interdiszciplináris Grastyán Konferencia, Absztrakt könyv, 2014.

9. Praksch D, Sándor B, **Varga Á**, Rábai M, Szakály P, Tóth K. Aspirin rezisztencia vizsgálata vesetranszplantáción átesett betegek körében. Érbetegségek 21:(1)pp, 42-43, 2014.

10. Praksch D, Sándor B, **Varga Á**, Rábai M, Szakály P, Tóth K. Vesetranszplantált betegek ASA-rezisztenciája: összehasonlítás a populációs rezisztencia adatokkal. Cardiologica Hungarica:(Suppl E) E59, 2014.

11. **Varga Á**, Kalmár Nagy K, Viola M, Sándor B, Tóth A, Juricskay I, Tóth K, Szakály P.Thrombocyta aggregáció gátlásról általánosságban és transzplantáltak körében. Családorvos Kutatók Országos Szervezetének konferenciája, Pécs, 2014. március 1. Absztrakt könyv

12. László E, **Varga Á**, Kovács K, Degrell P, Kiss P, Szakály P, Jancsó G, Tamás A, Reglődi D. Az endogen PACAP protektív hatásának vizsgálata ischaemia-reperfusio okozta vesekárosodásban. 44. Membrán-Transzport Konferencia, Absztrakt könyv, 2014

13. **Varga Á**, Sándor B, Kalmár Nagy K, Tóth A, Tóth K, Szalály P. The relationship between cardiovascular diseases and acetlysalicylic acid resistance after kidney transplantation. Nephrology Dialysis Transplantation 29: (3 Suppl), 312, 2014 May
IF: 3,488

14. László E, **Varga Á**, Degrell P, Kovács K, Kiss P, Szakály P, Tamás A, Reglődi D. Protective effect of exogenous PACAP on ischaemia/reperfusion-induced kidney injury of female rats. 20th International Symposium on Regulatory Peptides. Kyoto, 2014. Szeptember 7-10. Abstract book

15. **Varga Á**, Viola M, Kalmár Nagy K, Szakály P: Tapasztalataink 1 évvel a teljes körű Eurtotransplant csatlakozás után. Magyar Transzplantációs Társaság kongresszusa, Absztrakt könyv, 2014

16. László E, **Varga Á**, Degrell P, Kovács K, Kiss P, Tamás A, Reglődi D, Szakály P: A PACAP védő hatásának összehasonlító vizsgálata nőstény és hím patkányok ischaemia-reperfusios vesekárosodásában. Magyar Transzplantációs Társaság kongresszusa, Absztrakt könyv, 2014

17. **Varga Á**, Végh M, Weisz F. Segítő kezek-jelnyelvi kommunikáció az orvosi praxisban Családorvos Kutatók Országos Szervezetének Kongresszusa, Mórahalom, 2013. február 22-23. Absztrakt könyv

18. Tutervai P, **Varga Á**, Végh M. Segítő kezek- jelnyelvi kommunikáció az orvosi praxisban II. Családorvos Kutatók Országos Szervezetének Kongresszusa, Pécs, 2014. február 22-23. Absztrakt könyv

## 11. ACKNOWLEDGEMENTS

I would like to express my gratitude to, Peter Szakaly MD who has been a dedicated mentor to me in my scientific and professional advancement ever since I was a medical student. He has carefully guided my work, without his help my thesis could not have been made. He played an important role in my going into the surgical profession.

The work and humanity of my program director, Prof. Ors Peter Horvath is an example for the youth, so for as well. I am grateful that I was able to complete my Ph.D. in Prof. Gabor Kovács L. Doctoral School, under the supervision of Professor Ors Peter Horvath.

I would like to thank Professor Andras Vereczkei for providing me with the opportunity to perform my scientific work beside the everyday tasks I had at the Department of Surgery. I received all the support I needed for this.

I would like to thank The Head of Department of Transplantation, Karoly Kalmar Nagy MD, head physician who has, with his wide range of experience and knowledge, helped my development. I am delighted to work with him on his ward.

I would like to thank Professor Kalman Toth, Barbara Sandor MD and the associates of the, 1st Department of Internal Medicine, Hemorheological Laboratory, University of Pecs for helping me in my work.

I am grateful to the staff of the Department of Public Health, University of Pecs, Katalin Gombos MD and Katalin Gocze MD for their help in the statistical analysis.

I am grateful for all the help rendered by the nurses working at the Department of Transplantation Surgery. Special thanks to the staff of the Transplant Outpatient Clinic and our coordinators for their precision.

I thank my parents and my brother for their patience, love, I am grateful for their support, which made it possible for me to finish my studies.