Histo-morphometric analysis of kidneys harvested from deceased donors. Important factor for the future outcome

Introduction: Kidney transplantation is recognized as best treatment option for patients with chronic kidney disease stage 5. Long-term outcome of transplanted kidney may be determined among others by graft morphology.

Objectives: The aim of this study was to perform a preimplantation, histo-morphometric analysis of glomeruli obtained by needle biopsies of kidneys harvested from deceased donors to assess the relationships between kidney weight, glomerular volume (GV) and glomerular density (GD).

Patients and methods: Of the 28 adult kidney donors, 56 biopsied and weighted at time of transplantation kidneys were included into the analysis. Total number of complete glomeruli, as well as percentage of normal (Norm), globally sclerosed (GS), segmentally sclerosed (SS), hyperperfused (Hyp) and ischemic (Isch) glomeruli were calculated. Glomerular passenger cells (neutrophils, monocytes/macrophages and CD-20 positive cells) number per glomerulus were measured. Peritubular capillary density (PTCD) was expressed as the number of capillary profiles per mm² of cortical area. Mean glomerular volumes (MGV) were estimated from the maximal glomerular profile area according to the formula: 

\[ GV = \frac{4}{3} \pi r^3 (\mu m^3) \]

and glomerular density (GD) was expressed as the number of non-globally sclerosed glomeruli per mm² of cortical area.

Results: Significant negative correlation was found between MGV and GD (r = -0.31; p = 0.017). A significant positive correlations between donor age and kidney weight (r = 0.390; p = 0.001) and kidney weight and MGV (r = 0.258; p = 0.044) were observed. Significant negative correlations have been found between donor age and glomerular density as well as between kidney weight and glomerular density (r = -0.306; p = 0.016 and r = -0.394; p = 0.0016, respectively). Additionally, a significant positive correlation was found between kidney weight and percentage of segmental glomerulosclerosis (r = 0.261; p = 0.042).

Conclusions: 1. Mean glomerular volume in kidney biopsy may serve as surrogate marker of glomerular number. 2. Negative relationships between kidney weight and both glomerular density, and percentage of glomerulosclerosis, as well as, between mean glomerular volume and glomerular density suggest that higher kidney weight in adults is mainly related to kidney hypertrophy. 3. These pretransplantation histo-morphometric findings may influence the future outcome of transplanted kidney.

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Introduction: Kidney transplantation is recognized as best treatment option for patients with chronic kidney disease stage 5. Both short- and long-term kidney allograft outcomes are influenced by multiple host versus graft interactions which are immune and non-immune in their nature. It was postulated by Seron et al. [1] and Tisoni et al. [2] that subtle morphological changes in primary transplantation protocol biopsies can be useful in predicting long term graft outcome. Other studies [3,4] suggested, that identification of morphological changes detectable at the very early stages after transplantation or even in pre-implantation biopsies, before chronic changes superimpose could even be more useful to assess organ quality and predict long term graft outcome. Donor biopsies, usually do not display acute inflammatory lesions and the range of chronic lesions is even narrower than in protocol biopsies [5]. For these reason, some have proposed to evaluate donor biopsies with a view to further preimplantation analysis in order to differentiate in this range of lesions [6,7], while others have proposed to employ a morphometric methods [8,9]. Although more laborious and time consuming than conventional histologic evaluation, morphometric biopsy analysis reduces the subjectivity and variability in interpretation. Therefore, due to its laboriousity, it is important to know if histological changes available at transplantation provide only information concerning donor – derived organ injury or which lesions in pre-implantation biopsies could eventually allow the best prediction on outcomes and whether the evaluation of damage in all renal compartments allow a better prediction of outcome.

Results of some quantitative histopathological studies underscore the role of morphological changes detectable at the very early stages after transplantation or even in pre-implantation biopsies, before chronic changes superimpose could even be more useful to assess organ quality and predict long term graft outcome. Donor biopsies, usually do not display acute inflammatory lesions and the range of chronic lesions is even narrower than in protocol biopsies [5]. For these reason, some have proposed to evaluate donor biopsies with a view to further preimplantation analysis in order to differentiate in this range of lesions [6,7], while others have proposed to employ a morphometric methods [8,9]. Although more laborious and time consuming than conventional histologic evaluation, morphometric biopsy analysis reduces the subjectivity and variability in interpretation. Therefore, due to its laboriousity, it is important to know if histological changes available at transplantation provide only information concerning donor – derived organ injury or which lesions in pre-implantation biopsies could eventually allow the best prediction on outcomes and whether the evaluation of damage in all renal compartments allow a better prediction of outcome.

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Korelacja między wagą nerki a gęstością kłębuszków nerkowych w grupie dawców.

Figure 1
Correlation between kidney mass and glomerular density in donors group.
Korelacja między wagą nerki a gęstością kłębuszków nerkowych w grupie dawców.

Glomerular density (GD) was calculated in the section containing the highest number of complete glomeruli and defined as the number of non-globally sclerotic glomeruli per mm² of renal cortical area of needle biopsy.

Statistical analysis Results were expressed as the median and 95% confidence interval (CI). The significance of correlation between variables was assessed using the method of Spearman. A p value < 0.05 was considered as statistically significant.

Results
Median of age and 95% CI of the whole donors group was 44.7 (40.9 – 47.2) years (range 18-64). Last serum creatinine concentration (median and 95% CI) were 1.47mg% (1.14 – 1.81) and weight of kidneys between donors ranged two fold, from 248kg to 504g. In 16 cases intracranial hemorrhage was cause of death. Remaining twelve donors died due to brain trauma. Significant positive correlation was observed between donor age and kidney weight (r = 0.390; p = 0.001). Significant negative correlations have been found between donor age and glomerular density as well as between kidney weight and glomerular density (Fig. 1) and donor age or kidney weight respectively.

Discussion
A wide diversity of histological damage has been found in studies of subjects older than 50 years without history of renal disease or arterial hypertension and among organ donors with major comorbidities and risk factors [22]. Therefore, histological assessment of pre-implantation biopsies not only permits single recording of chronic lesions but also could help to assess organ quality, properly allocate harvested organs and predict short- and long-term outcomes of renal allografts [23]. It has been shown, that chronic injury observed in pre-implantation biopsies of kidneys from deceased brain-dead, older and extended criteria donors, comprised interstitial [8], vascular [24] and glomerular [24] compartmental cell combinations of these lesions and correlated with the incidence of DGF, short- and long-term kidney function and survival.

Due to its simplicity for evaluation, global glomerulosclerosis is the most frequent glomerular change used in clinical studies. However its meaning and interpretation is worth of comment. Glomerulosclerosis is not very advanced process in most kidneys harvested from younger donors without significant comorbidities and risk factors. Global glomerulosclerosis is not an active lesion but should be interpreted as a consequence of aging process or pre-existed donor kidney disease, which has no impact
Correlation coefficient $r=0.261$, $p=0.0421$

Figure 3
Correlation between kidney mass and percentage of segment glomerulosclerosis in donors group.

Korelacja między wagą nerki a odsetkiem nerkowych z osobami ukadowanymi w grupie dawców.

on kidney functional reserve at the time of transplantation. Additionally, most of the studies to date uses wedge biopsies to obtain kidney tissue samples for histological assessment. However, superficial wedge biopsies overestimate arterial- and glomerular sclerotic lesions in the cortico-medullary junction of the kidney [27]. Furthermore, the sample size of renal biopsies is an important determinant of accurate assessment of the percentage of glomerulosclerosis in the kidney [28]. So, it seems reasonable, that the quality of kidneys for transplantation and prediction of its long-term outcome should not be assessed based exclusively on glomerulosclerosis index.

The Banff 2007 classification recommends routine scoring of zero-time needle biopsies similar to biopsies performed after kidney transplantation, which help to interpret the lesions as a continuum, enables obtaining both cortical and medullary parts of the kidney and allow evaluation particular compartments at different levels of the kidney parenchyma [29]. However, for the reasons mentioned earlier, the importance of glomerulosclerosis examined in needle biopsies to predict quality and long-term outcome of transplanted kidney harvested from younger donors, in which advanced chronic changes are scarce, remain very controversial. Therefore, it seems valuable to study early objective histological markers/changes which could serve as better predictors of both, the quality of transplanted kidney and long-term graft function.

Results of some quantitative histological studies underscore the role of neprhon number and glomerular size as well as its variability in the pathogenesis of arterial hypertension and kidney failure [11,10]. It has been found that patients with essential hypertension are characterized by lower neprhion number and higher glomerular volume (GV) in comparison to normotensive patients [12]. Results of autopsy studies [12,13] revealed an inverse relationships between segmental glomerulosclerosis and glomerular volume as well as between glomerular volume and glomerular (neprhion) number, so the GV could serve as surrogate of total glomerular number in clinical studies [13,14].

In the present study, median and 95% CI of complete glomeruli obtained in preimplantation kidney biopsies was 17.35 (14.95-19.74) which is similar to the values obtained in other studies [10,34]. However, according to Corwin et al. [28] these numbers are somewhat too low for accurate assessment of the percentage glomerulosclerosis in the kidney but completely enough for a reliable estimate of glomerular size by the MPA method [20]. Index of globally sclerotic glomeruli in the studied population of donors was 3.84% (1.97–5.71) median nad 95% CI, which is close to the results published by Hoy WE et al. [11].

For the measurement of the GV, glomerulus was defined as an area inside the intact Bowman’s capsule containing tuft, as the strong correlation between glomerular capsular area and glomerular tuft area has been found [20]. Due to strong correlation between the method of profile area (MPA) with the Cavaliert method considered the gold standard [20], maximal glomerular volume was estimated from the maximal glomerular profile area. The mean glomerular volume measured for the whole group of donors in preimplantation biopsies was $5.15 \times 10^5 \mu m^3$ (3.86–6.44). The variability in mean glomerular volumes in whole group of donors ranged 3.74. Mean GV correlated positively with kidney weight in the whole group of donors.

Significant negative correlations have been found between donor age and glomerular density, as well as between kidney weight and glomerular density in the whole group of donors. These results confirm age-dependent glomerular number decrease observed by others and underscore, that higher kidney mass in adults is not always related to higher glomerular number. Additionally, significant negative correlation was found between GD and maxGV in the whole group of donors. This correlation is in line with the previous observations, which also noted an inverse relationship between both parameters and considered GV not only as a surrogate measure of glomerular number/density [13,14] but also as poor predictor of long-term graft outcome [33].

A significant positive correlation observed between kidney weight and percentage of segmental glomerulosclerosis in the whole donors group additionally confirms, that the consequence of the lower glomerular number/density are structural glomerular disturbances like glomerular enlargement and segmental glomerulosclerosis.

In conclusion the presented study revealed that: 1. Mean glomerular volume in kidney biopsy may serve as surrogate marker of glomerular number. 2. Negative relationships between kidney weight and both glomerular density, and percentage of glomerulosclerosis as well as between mean glomerular volume and glomerular density suggest that higher kidney weight is mainly related to kidney hypertrophy. 3. These pretransplantation histo-morphometric findings may influence the future outcome of transplanted kidney.

References


