

Figure 1. Box-Whisker-Plots of the four biomarkers in the tubulointerstitium. Boxplots show the median and the 1.5 interquartile range of the $\log _{2}$ (relative expression) measured in the qRT-PCR experiment. A value of zero equates same expression level like in the reference RNA.


Figure 2. ROC curves: Discrimination for DGF after transplantation using donor age (solid blue line), expression features (dashed red line), or the combination of both (dashed green line).

## Table 1

Demographic data of transplant donors and recipients stratified by treatment assignment. Continuous data are provided as median ( $1^{\text {st }}$ quartile, $3^{\text {rd }}$ quartile), categorical data are given as counts.

|  | PF group | DGF group | p-value |
| :--- | :--- | :--- | :--- |
| Number of donors |  | 24 | na |
| Number of donor organs | 17 | 17 | na |
| Donor age (years) | $44.0(39,47)$ | $50.0(45,60)$ | 0.005 |
| Donor sex (f/m) | $9 / 8$ | $4 / 13$ | 0.078 |
| Last creatinine of donor (mg/dl) | $0.82(0.65,1.01)$ | $1.34(0.92,1.57)$ | 0.011 |
| Vasopressors used (n/y) | $2 / 15$ | $5 / 12$ | 0.203 |
| Multiorgan donors (n/y) | $14 / 3$ | $13 / 4$ | $1.000^{\star}$ |
| Cause of death (trauma $/$ intracranial <br> hemorrhage ) | $5 / 12$ | $2 / 15$ | $0.398^{\star}$ |
| Number of recipients | 17 | 17 | na |
| Recipient age (years) | $47.7(28.82,56.71)$ | $51.6(42.87,59.82)$ | 0.302 |
| Recipient sex (f/m) | $5 / 12$ | $7 / 10$ | 0.473 |
| Transplant number (1/2/3) | $15 / 2 / 0$ | $13 / 31$ | $0.656^{\star}$ |
| Cold ischemic time (hours) | $18.6(15.26,23.10)$ | $19.3(16.24,22.35)$ | 0.757 |
| PRA latest (\%) | $0.0(0.0,0.0)$ | $0.0(0.0,5.0)$ | 0.521 |
| Sum of HLA mismatches (0/1/2/3/4/5/6) | $1 / 0 / 4 / 9 / 3 / 0$ | $0 / 1 / 2 / 8 / 4 / 2$ | $0.560^{*}$ |
| Number of dialysis treatment (0/1/2/3/4/5) | $17 / 0 / 0 / 0 / 0 / 0$ | $0 / 0 / 10 / 4 / 1 / 2$ | $<0.001^{*}$ |
| Immunosuppression (CNI/else) | $17 / 0$ | $17 / 0$ | na |
| Induction therapy (none/antiCD25/ATG) | $10 / 6 / 1$ | $12 / 5 / 0$ | 0.721 |

na ... not applicable, * Fisher's exact test

## Table 2

Multivariable logistic regression model. The discriminative power of this model is indicated by a c-statistics of 0.83 (AUC). Given is the odds ratio (OR), the confidence interval, and the $p$-values.

| Predictors | OR | $95 \% \mathrm{Cl}$ |  | p-value |
| :--- | :---: | :--- | :---: | :---: |
| Donor age (per year) | 1.108 | 1.005 | 1.221 | 0.040 |
| LCN2 (AU) | 1.269 | 0.851 | 1.890 | 0.242 |
| HAVCR1 (AU) | 1.443 | 0.943 | 2.208 | 0.091 |

AU: arbitrary units (log2-ratio of the amount of mRNA as found in the sample with respect to the concentration of the respective mRNA given by the universal reference mRNA used in the qRT-PCR experiments).

## Table 3

Discrimination of the models and optimism (3A) derived from the re-sampling procedure ( 34 -fold cross validation). Calibration of the DGF prediction model by the Hosmer-Lemeshow goodness of fit test (3B). The expected to observed number of cases in each of the deciles of patients were not statistically different suggesting good calibration ( $p=0.76$, chi-square test).

| Model | AUC (c-statistic) | AUC (X- <br> validation) | Optimism |
| :--- | :---: | :---: | :---: |
| Donor age | 0.78 | 0.74 | 0.04 |
| LCN2, HAVCR1 | 0.80 | 0.71 | 0.09 |
| LCN2, HAVCR1 and | 0.83 | 0.75 | 0.08 |


| Group | Total <br> number of <br> patients | DGF |  | PF |  |
| :---: | :---: | :---: | :---: | :---: | :---: |
|  | 3 | 0 | 0.20 | 3 | 2.80 |
| 2 | 3 | 0 | 0.46 | 3 | 2.54 |
| 3 | 3 | 1 | 0.79 | 2 | 2.21 |
| 4 | 3 | 2 | 1.06 | 1 | 1.94 |
| 5 | 3 | 1 | 1.30 | 2 | 1.70 |
| 6 | 3 | 2 | 1.64 | 1 | 1.36 |
| 7 | 3 | 2 | 1.91 | 1 | 1.09 |
| 8 | 3 | 2 | 2.28 | 1 | 0.72 |
| 9 | 3 | 3 | 2.66 | 0 | 0.34 |
| 10 | 2 | 2 | 2.77 | 1 | 0.23 |



Figure S1. Bias Test of pre-amplification technique: Dilution series from a microdissected nephrectomy sample (T: Tubulointerstium, G: glomeruli part) were
 (Cyclophilin A) after a 10 cycle pre-amplification. All regression lines show excellent correlation between a wide dynamic range ( $125 \mathrm{ng} / \mu \mathrm{l}$ to $1 \mathrm{ng} / \mu \mathrm{l}$ cDNA concentration before pre-amplification). The regression lines of PPIA and LCN2 show similar slopes, therefore we decided to use PPIA as endogenous control gene.


Figure S2. Bias test for reverse transcription, pre-amplification and realtime PCR of the reference RNA: Correlation of fourteen different genes in the standard RNA (Stratagene Human reference RNA) is almost one, suggesting reference RNA is an excellent calibrator.

## Table S1

Multivariable logistic regression model: LCN2 and HAVCR1. The discriminative power of this model is indicated by a c-statistics of 0.80 (AUC). Given is the odds ratio (OR), the confidence interval, and the p-values.

| Predictors | OR | $95 \% \mathrm{Cl}$ |  | p-value |
| :--- | :---: | :--- | :---: | :---: |
| LCN2 (AU) | 1.328 | 0.918 | 1.920 | 0.133 |
| HAVCR1 (AU) | 1.356 | 0.948 | 1.941 | 0.096 |

AU: arbitrary units (log2-ratio of the amount of mRNA as found in the sample with respect to the concentration of the respective mRNA given by the universal reference mRNA used in the qRT-PCR experiments).

## Table S2

Multivariable logistic regression model: Donor age, cold ischemic time (CIT), panel reactive antibodies (PRA), donor last creatinine, LCN2 and HAVCR1. The discriminative power of this model is indicated by a c-statistics of 0.86 (AUC). Given is the odds ratio (OR), the confidence interval, and the p-values.

| Predictors | OR | $95 \% \mathrm{Cl}$ |  | p-value |
| :--- | :---: | :---: | :---: | :---: |
| Donor age (year) | 1.078 | 0.956 | 1.216 | 0.222 |
| CIT (hours) | 1.009 | 0.809 | 1.258 | 0.939 |
| PRA (\%) | 1.031 | 0.968 | 1.100 | 0.342 |
| Donor last creatinine (mg/dl) | 4.592 | 0.226 | 93.367 | 0.321 |
| LCN2 (AU) | 1.146 | 0.727 | 1.808 | 0.558 |
| HAVCR1 (AU) | 1.445 | 0.889 | 2.348 | 0.137 |

AU: arbitrary units (log2-ratio of the amount of mRNA as found in the sample with respect to the concentration of the respective mRNA given by the universal reference mRNA used in the qRT-PCR experiments).

## Table S3

Multivariable logistic regression model: Donor age, cold ischemic time (CIT), panel reactive antibodies (PRA) and donor last creatinine. The discriminative power of this model is indicated by a c-statistics of 0.85 (AUC). Given is the odds ratio (OR), the confidence interval, and the p-values.

| Predictors | OR | $95 \% \mathrm{Cl}$ |  | $p$-value |
| :--- | :---: | :---: | :---: | :---: |
| Donor age (year) | 1.078 | 0.967 | 1.201 | 0.175 |
| CIT (hours) | 0.961 | 0.783 | 1.180 | 0.704 |
| PRA (\%) | 1.036 | 0.975 | 1.101 | 0.255 |
| Donor last creatinine (mg/dl) | 10.797 | 0.922 | 126.451 | 0.058 |

