ABSTRACTS

POSTER PRESENTATIONS

RENALE AND TRANSPLANT PATHOLOGY AND LAB MEDICINE

1. Comparison of LCR- MFI between Cadaver and Live donor renal in Lysate based DSA testing by Luminex ® technology
   Dhital R

2. Recurrent Diseases After Kidney Transplantation: A 5- Year Single Centre Experience
   Agarwal K

3. Denovo Diseases In Renal Transplantation- A Single Centre Five Year Experience
   Baishya P

4. C4d Negative Antibody Mediated Rejection And Renal Allograft Dysfunction: Single Centre Experience
   Nigam LSC

5. Post Renal Transplant Lymphoproliferative Disorder: A Case Report
   Agarwal K

6. De Novo Focal and Segmental Glomerulosclerosis in renal allograft: A single centre five year experience
   Baishya P

7. Invasive Fungal Infections In Renal Transplant Patients: A Single Centre Study
   Patel MH

8. Role Of Pre-Transplant Biopsy Evaluation In Single Versus Dual Kidney Allocation Of Deceased Donor Kidney Transplantation: Single Center Experience
   Suthar KS

   Krishnan RG

10. Comparison Of Two Isoagglutinin Titre Methods For Anti-ABO Antibody Estimation In ABO Incompatible Kidney Transplants
    Chandak SA
Comparison of LCR- MFI between Cadaver and Live donor renal in Lysate based DSA testing by Luminex ® technology.

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Abstract:
Introduction: Lysate control Reagent (LCR), a mixture of biotinylated monoclonal antibodies that are specific for and bind to HLA- class I and II, is run in parallel to test sera in order to assure an attachment of HLA- I and II present in the lysate to the capture beads. Low Mean Fluorescence Intensity (MFI) of LCR (LCR-MFI) signifies mal-optimum attachment of HLA antigens to the detecting beads. We aim to compare LCR- MFI between lymphocyte lysates of cadaver and live donors.

Materials and Method: Sixty live donor and 10 cadaver donor renal transplant recipients, at 3 months of transplantation or at the time of rejection, were tested for the presence of DSA against their respective donor’s lymphocyte lysate by Luminex ® technology (Immucor Gamma, Immucor, USA). For each test lysate, one well was assigned for the LCR and was run in parallel with the test using 8 µl of donors lymphocyte lysate and 5 µl of capture beads. Further processing was done as per manufacturer’s protocol. Twenty standard lysate controls were run in parallel for comparison.

Result: All mentioned values are mean of MFI ± standard error of mean (Table 1) LCR- MFI of donor lymphocyte lysate for HLA- class II was significantly lower than that of standard control. There was a marked difference in LCR- MFI for HLA- class II between live donor and cadaver donor, lower in the later suggesting less concentration of HLA- class II antigen in the lysate. With these LCR-MFI, corresponding test MFI (taking >1000 as a significant MFI) had a diagnostic accuracy of 60%. A correction study was performed to check if this LCR-MFI could be increased by increasing initial volume of donors lysate in serial fold from 8 µl to 16 µl, 24 µl, 32 µl, 40 µl and 48 µl. At 40 µl volume, the initially low LCR-MFI for HLA- class II of cadaver donor increased to the level which was similar to that of live donor (p= 0.6838) for which the diagnostic accuracy was increased to 80%.
Conclusion: The concentration of HLA-class II antigen in lysate prepared from lymphocyte pool of cadaver donor is lower than that of live donor leading to low MFI of LCR. This problem can be overcome by increasing the initial volume of lymphocyte lysate.

2: Recurrent Diseases after Kidney Transplantation: A 5 Year Single Centre Experience

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Abstract:
Background: Recurrence of disease following renal transplantation (RT) is a well-known cause of allograft failure. The impact of recurrence on graft survival varies widely with different diseases. This study aims to find out the incidence of recurrent diseases and their impact on long-term graft survival.

Material and method: This was a retrospective study of renal allograft biopsies of a single center between January,’11 to October,’15. Primary glomerular, metabolic and parenchymal diseases were included. Formalin fixed biopsies were evaluated under light microscopy using standard stains and C4d by immunohistochemistry. Immunofluorescence was studied using anti-human IgG, IgA, IgM, C3, C1q, albumin and fibrinogen antisera whenever indicated. Diagnosis was made as per modified Banff guidelines.

Results: Out of 1586 biopsies, recurrence was noted in 16 (1.01%) biopsies. Mean patient age was 28 ± 9.01 years with males constituting 68.75%. Fourteen (87.5%) biopsies belonged to living donor RT and 2 (12.5%) were from cadaver donor transplants. Oxalosis was the commonest (25%) followed by diabetic nephropathy (18.75%), lupus nephritis (12.5%), membranous glomerulonephritis (12.5%) and one (6.25%) each of membranoproliferative glomerulonephritis, crescentic glomerulonephritis, benign nephrosclerosis, thrombotic microangiopathy and tubulointerstitial nephritis. Among the cadaver donor transplantation, recurrence was noted in
membranoproliferative glomerulonephritis and tubulointerstitial nephritis. Ten year graft and patient survival were 52.1% and 66.9% respectively.

**Conclusion:** Recurrence of metabolic diseases and tubulointerstitial nephritis along with glomerulonephritis is an important determinant of long-term graft outcome. Research on pathophysiology of primary and recurrent diseases will help to evolve new strategies for their prevention thereby improving long term graft survival.

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**3: Denovo Diseases in Renal Transplantation: A Single Centre Five Year Experience**

**Baishya P, Vanikar AV, Patel RD, Kanodia KV, Suthar KS, Nigam LSC, Agarwal K.**

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**Abstract:**

**Background:** De novo diseases are the diseases occurring in renal allograft unrelated to original disease, affecting graft function and even survival. This study was undertaken to evaluate the incidence of de novo diseases in renal allograft biopsies, and to study their impact on graft function.

**Material and method:** This was a retrospective single center study conducted on biopsies performed between January,’11 and October,’15. Renal allograft biopsies were evaluated by standard techniques and diagnosed as per modified Banff guidelines.

**Results** : Out of 1586 biopsies, 57(3.59%) revealed de novo diseases, at mean 2.4 ± 2.50 years post-transplant, with mean serum creatinine (Scr) of 2.47 ±1.21 mg/dl. Mean patient age was 34.50 ± 11.34 years, majority belonging to males (92.9%); 91.2% being living donor and 8.7% were cadaver donor transplants. Most common lesions were focal segmental glomerulosclerosis (FSGS) (63.1%), crescentic glomerulonephritis (CrGN) (10.5%), benign nephrosclerosis (BN) and oxalosis (5.2% each), cryoglobulinemic glomerulonephritis, diabetic nephropathy (DN) and membranous glomerulonephritis (3.5% each). Ten year graft and patient survival were 46.8% and 71.8% respectively with mean Scr of 2.10 ± 1.22 mg/dL. Five year graft and patient survival were 76.2 % and 57.6% with FSGS, 53.3% and 100% with CrGN, and 0% and 100%, each with
thrombotic microangiopathy (TMA) and BN respectively. Other diseases had no significant impact on survival.

**Conclusion:** De novo diseases like TMA FSGS, CrGN and BN can adversely affect graft function and survival. Research, early diagnosis and management can help in improving their outcome after transplantation.

**Key Words:** de novo, renal transplantation, graft survival, serum creatinine.

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### 4: C4d Negative Antibody Mediated Rejection And Renal Allograft Dysfunction: Single Centre Experience

**Nigam LSC, Patel RD, Kanodia KV, Suthar KS, Vanikar AV**

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**Abstract:**

**Background:** Banff’13 update included C4d negative antibody mediated rejection (ABMR) as a separate entity responsible for graft dysfunction with limited clinical/ prognostic implications. We carried out a prospective study of renal allograft biopsies to determine the incidence and outcome of C4d negative ABMR.

**Material and Method:** Biopsies from Jan’13 to October’15 were subjected to light microscopy using standard stains and C4d immunohistochemistry on paraffin sections. Only adequate biopsies with immunological injuries were included in study. Graft function was measured in terms of serum creatinine (SCr) (mg/dl). Qualitative donor specific antibody (DSA) evaluation was performed in a subset of patients.

**Results:** Totally 404 of 987 biopsies were included. Results are displayed in table.
Groups (Total: 404)  
<table>
<thead>
<tr>
<th>Group – 1 (ABMR) (n=113)</th>
<th>Group – 2 (ABMR + CR) (n=245)</th>
<th>3 (CR) (n=46)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1a (C4d Positive) (n=92)</td>
<td>2a (C4d Positive) (n=199)</td>
<td>1.61±0.66</td>
</tr>
<tr>
<td>1b (C4d Negative) (n=21)</td>
<td>2b (C4d Negative) (n=46)</td>
<td></td>
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</tbody>
</table>

Mean Time posttransplant (months)±SD  
| 1.32±0.28 | 1.51±0.27 | 1.98±0.30 | 1.33±0.15 |

Mean SCr (mg/dl)±SD at the time of biopsy  
| 2.74±0.35 | 1.88±0.29 | 2.39±0.46 | 2.34±0.21 |

Current Mean follow-up (years)±SD  
| 2.3±2.58 | 1.69±1.35 | 3.08±2.54 | 2.04±1.91 |

Current Mean SCr (mg/dl)±SD  
| 2.57±1.09 | 3.52±2.72 | 3.2±0.49  | 3.28±1.66 |

Patient Loss (%)  
| 6.52%  | 9.52%  | 3.02%  | 8.70%  | Nil |

Graft loss  
| 5.43%  | 2.08%  | 3.02%  | Nil    | Nil |

Conclusion: Graft loss is higher with C4d+ve ABMR, however C4d negative rejections cannot be ignored.

5: Post Renal Transplant Lymphoproliferative Disorder: A Case Report

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Abstract:
Background: Post-transplant lymphoproliferative disease (PTLD) are heterogeneous group of diseases characterised by abnormal lymphoid proliferation occurring after solid organ
transplantation (SOT). We report PTLD in transplanted kidney in a 30 year old male occurring 5 years after transplant.

**Material and method:** Formalin fixed biopsies was evaluated under light microscopy using standard stains and C4d by immunohistochemistry. Immunofluorescence was studied using anti-human IgG, IgA, IgM, C3, C1q, albumin, fibrinogen and C4d antisera.

**Case history:** A 30 year old male renal transplant patient who received mother’s kidney in 2010 (HLA match 3/6) presented with rise in serum creatinine of 2.78 mg/dl (baseline 2.4 mg/dl) on cyclosporine based triple immunosuppression. He had acute borderline T-cell mediated rejection in 2010 and acute T + B-cell mediated rejection in 2012. Graft biopsy revealed 26 glomeruli with mesangial prominence and fairly open capillary lumina lined by membranes of normal thickness. Tubules were mildly degenerated. Interstitium was markedly prominent for diffuse mononuclear cellular infiltration with presence of large nodular aggregates and sheets of monomorphic large round cells, positive for CD20 and negative for CD4, SV40 and CD138, with hyperchromatic coarse nuclei and prominent nucleoli, and occasional atypical mitotic figures. Blood vessels were unremarkable. Immunofluorescent microscopy revealed no significant immunofluorescence with anti-human IgG, IgA, IgM, C3, C1q, fibrinogen, albumin and C4d antisera. Thus he was diagnosed as monomorphic B-cell lymphoma/PTLD (B-cell type). PCR for EBV DNA was negative.

**Conclusion:** PTLD represents a serious problem after SOT requiring further research, early diagnosis and preventive measures.

**Key words:** PTLD, SOT, EBV.
6: De Novo Focal And Segmental Glomerulosclerosis In Renal Allograft: A Single Centre Five Year Experience


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Abstract:

Background: De Novo Focal Segmental Glomerulosclerosis (FSGS) often presents as nephrotic syndrome post-transplant, due to nephron loss/ glomerular hyperfiltration / calcineurin inhibitor(CNI) induced toxicity. We carried out this study to evaluate the natural history of de novo FSGS in renal allograft biopsies, and evaluate their impact on graft function.

Material and method: This was a retrospective single center study performed between January,’11 and October,’15. Renal allograft biopsies were evaluated by standard techniques and diagnosed as per modified Banff guidelines. Associated urinary proteins, serum creatinine (SCr) and immunosuppression were studied.

Results: Out of 1586 biopsies, 36(2.26%) revealed de novo FSGS, at mean 1.73 ±1.70 years post-transplant with mean SCr of 2.22 ± 1.09 mg/dl. Mean patient age was 30.17 ± 8.25 years, males (91.6%) outnumbering females; and majority were living donor transplants (91.6%) on CNI based immunosuppression. Mean 24 hours urinary proteins were 3.84 ±2.90 gms. Histopathology revealed classical (47.2%), collapsing (44.4%), cellular (5.5%) and perihilar (2.7%) variants. Five and 10 year patient survival was 57.6%. Five year graft survival was 76.2%, all grafts were lost at 10 years. Mean SCr (mg/dl) at 5 and 9 years was 1.86 ± 0.40 and 1.93 ±0.40 with 0.18 ± 0.26 gms 24 hour urinary protein loss at 9 years.

Conclusion: Renal allograft recipients on CNI based immunosuppression are prone to develop de novo FSGS, commonly presenting with proteinuria at mean of 1.7 years post-transplant with common histopathological findings of classical or collapsing variants. It may be associated with any age, living/ cadaver donor and can cause long term graft/ patient loss.
Abstract:

**Background:** Timely diagnosis of invasive fungal infections (IFI) in renal transplant (RT) patients on immunosuppression is often difficult, jeopardizing their life and graft. We reported IFI and their causative fungal agents in post-RT patients.

**Materials and methods:** This was a retrospective 6 years clinical study carried out from 2010 to 2015 on 1900 RT patients. Clinical data included patient-donor demographics, time to onset of infection, risk factors and graft function in terms of serum creatinine (S.Cr). To identify IFI, we examined bronchoalveolar lavage (BAL), blood, tissue, wound swab samples by conventional mycological methods.

**Results:** IFI were diagnosed in 30 (1.56%) patients on triple immunosuppression, mainly males (n=25) with mean age of 36.57 ±11.9 years at 13.12 ±18.35 months post-RT. Aspergillus species was identified in 11 BAL, 1 tissue and 1 wound specimen each, 30.76 % of these were fatal and 15.38% caused graft loss; Candida albicans was in 9 BAL, 4 blood, 2 wound swab and 1 tissue specimens, 25% of these were fatal and 25% had graft loss and 1 mucor in BAL which was fatal. Seven patients were diabetic, 10 had superadded cytomegalovirus infection and 15 were anti-rejected.

**Conclusion:** IFI are associated with increased morbidity and mortality in RT patients. Triple immunosuppression, diabetes and superadded infection are added risks for these patients. Prevention, early diagnosis and appropriate management are necessary to improve their prognosis.

**Keywords:** Renal transplant, Invasive fungal infection, Candida albicans, Aspergillus, Mucor
8: Role Of Pre-Transplant Biopsy Evaluation In Single Versus Dual Kidney Allocation Of Deceased Donor Kidney Transplantation: Single Center Experience

Suthar KS, Vanikar AV, Patel RD, Kanodia KV, Nigam LA

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Abstract:

Background: Wedge biopsy has controversial role in deciding potential donor kidney quality versus gross appearance. We compared single versus dual-kidney deceased donor renal transplantation (DDRT) outcome and correlated baseline biopsy with radiological/gross findings.

Material and Method: Baseline wedge biopsy histopathological evaluation of frozen sections from 17 DD performed between Jan’11 to Nov’15 was correlated with graft outcome. Group-1 (9 donors, 15 recipients) included single-kidney DDRT; group-2 (N-8) included dual-kidney DDRT. Their patient-donor demography was comparable. Outcome was analyzed in terms of graft/patient loss and serum creatinine (SCr: mg/dL).

Results: All kidneys were unremarkable on gross/radiology examination with mean size of 10.6 x 5 cm. Baseline SCr was 2.02 in group-1 and 2.45 in group-2. Biopsies were adequate with mean 26 glomeruli and 2 arteries. Majority group-1 biopsies revealed unremarkable morphology; in group-2 mesangial proliferation was the commonest finding. Group-2 had significantly better outcome than group-1. Over mean follow-up of 21.8 months of group-1, 13.3% grafts, 20% patients and 6.7% combined graft +patient loss was recorded. Over mean follow-up of 10.57 months in group-2, 12.5% graft loss and 100% patient survival were observed. Two graft losses in group-1 were noted at 10 days and 3.5 months where baseline biopsies revealed extensive global sclerosis. One graft was lost in group-2 due to rejection. Mean SCr at 1 and 3 years post-transplantation were 1.4 and 2.61 in group-1 and 1.41 and 2.43 in group-2.

Conclusion: Allocation based on histopathology helps improving DDRT outcome. Dual-kidney DDRT has better outcome than single-kidney DDRT.
Study On Risk Factors & Microbiological Profile Of Post Renal Transplant Urinary Tract Infection & Its Influence On Graft Function

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Abstract:

Background - Urinary tract infection (UTI) is most common infection post renal transplant. Aims- 1. To determine incidence of post renal transplant UTI incidence 2. To determine the risk factors leading to UTI and its effect on graft function.

Method: Study design : Single centre retrospective observational study from January 2013 to June 2015.

Study observations: A total of 96 patients were included with mean age 31 +/- 10 years and majority were males 77(80%). 58(60%) LRTRTRs & 18 (19%) DDRTRs were studied. Rejections 26% & Diabetes mellitus 45% were noted. UTI was found in 29(30%) & pyelonephritis in 28% cases. 41% were asymptomatic during the episodes. The typical symptoms were noted in 21% patients. Majority of cases occurred in 1st month post transplant. 59%(17) cases were caused by Ecoli & 38% Klebsiella. Ecoli was sensitive to cefaperazone sulbactam, amikacin & was resistant to 1st and 3rd generation cephalosporins, ampicilin and nalidixic acid. Klebsiella was found to be sensitive to imipenam. Multidrug resistant UTI was isolated in 17% (5) cases. Associated graft dysfunction was found in 19(66%) cases. Persistant graft dysfunction was noted in 31% cases. Relapse occurred in 45% cases, reinfection in 17% cases. Longer duration of foleys insertion (>13 days) and NKD like obstructive uropathy and neurogenic bladder was associated with increased risk.

Conclusion: Incidence of UTI in post renal transplant recipients is 30%. Typical symptoms of UTI was found only in 21% cases. Longer duration of Foley's catheterisation & Native kidney diseases like neurogenic bladder, obstructive uropathy with vesicoureteric reflux & stone disease were statistically associated with increased incidence of UTI. 66% patients developed graft dysfunction following UTI with persistent graft dysfunction in 31% cases.
**Abstract:**

**Background:** Anti A/B antibody titres are important to decide acceptability of ABO-incompatible (ABO-i) renal transplant (RT) donor. Tube incubation technique (TIT) for IgM antibodies and indirect antiglobulin by gel column technique (IAT-gel) for IgG antibodies are commonly used methods for anti-A/B antibody titre estimation. We carried out a prospective study for comparing these two methods in potential ABO-i RT.

**Methods:** ABO-antibody titres were estimated with corresponding donor red cells (RBCs) in 21 patients. Patient-sera were serially diluted from 1:2 to 1:1024 using normal saline (NS); 1% RBC suspension in low ionic strength saline solution in IAT-gel, and 5% washed RBC suspension in N.S in TIT.

**Results:** Blood groups were “O” in 16, “A” in 3 and “B” in 2 patients. Group “O” sera exhibited highest isoagglutinin titers in IAT-gel (IgG), with mean anti-A of 1:329 (range:1:4 to 1:1024 ) and anti-B of 1:118 (range: 1:8 to 1:256 ) vs TIT (IgM) showing 1:162 (range:1:16 to 1:512) with anti-A and 1:83 (range: 1:8 to 1:128 ) with anti-B antisera respectively. In group “B”, mean anti-A titres were 1:69 (range:1:16 to 1:128) by TIT and 1:18 (1:8 to 1:32) by IAT. With blood group “A”, anti-B titres were 1:10 (1:4 to 1:16) by IAT and 1:136 (1:16 to 1:256) by TIT. Blood group “O” exhibited highest anti-A IgG titres and blood group-A exhibited lowest IgG antibody titres.

**Conclusion:** For development of anti-AB isoagglutinins, blood group “O” patients require closer monitoring and blood group “A” require least monitoring.

**Key Words:** ABO incompatible kidney transplantation, anti-A antibody titres, IgM antibodies, IgG antibodies, anti-B antibody titres, iso-agglutinins.
Note: - The presenter has substantial contribution in this original research work. - None of the authors have any “objection” for this presentation. - None of the authors had any conflict of interest or financial interest for this presentation.

**Legends for table:** Table displaying IgG and IgM antibody titres measured by indirect anti-globulin test-gel column technique (IAT-gel) and Tube incubation technique (TIT) respectively.

<table>
<thead>
<tr>
<th>Blood group</th>
<th>O</th>
<th>A</th>
<th>B</th>
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</thead>
<tbody>
<tr>
<td>Titers (range)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anti-A IgG Ab (IAT)</td>
<td>1:329 (1: 4 to 1024)</td>
<td>-</td>
<td>1:18 (1:8 to 32)</td>
</tr>
<tr>
<td>Anti-A IgM Ab (TIT)</td>
<td>1:162 (1: 16 to 512)</td>
<td>-</td>
<td>1:69 (1:16 to 128)</td>
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<tr>
<td>Anti-B IgG Ab (IAT)</td>
<td>1:118 (1: 8 to 256)</td>
<td>1:10 (1:4 to 16)</td>
<td>-</td>
</tr>
<tr>
<td>Anti-B IgM Ab (TIT)</td>
<td>1: 83 (1: 8 to 128)</td>
<td>1:136 (1:16 to 256)</td>
<td>-</td>
</tr>
</tbody>
</table>

(Ab: antibody)