

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ (ط)

emalik

Platelet Dysfunction in Children with Chronic Kidney Disease Stage 5 on Hemodialysis

Dr Salwa H. Peracha

Dr Nisar Ahmed

Haematology And Transfusion

Medicine Division CH&ICH

INTRODUCTION

- Patients with CKD stage 5 i.e. ESRD suffer from a bleeding diathesis as well as a prothrombotic state. Platelet dysfunction contributes to both of these abnormalities.
- Platelet aggregation studies have been done to understand the platelet dysfunction in CKD 5.
- Variable results have been obtained in different studies while using adenosine diphosphate (ADP) and Ristocetin as platelet agonists.

OBJECTIVE

To compare the mean maximal platelet aggregation percentage in children with CKD 5 on hemodialysis and healthy children, using ADP and ristocetin as agonists.

STUDY DESIGN

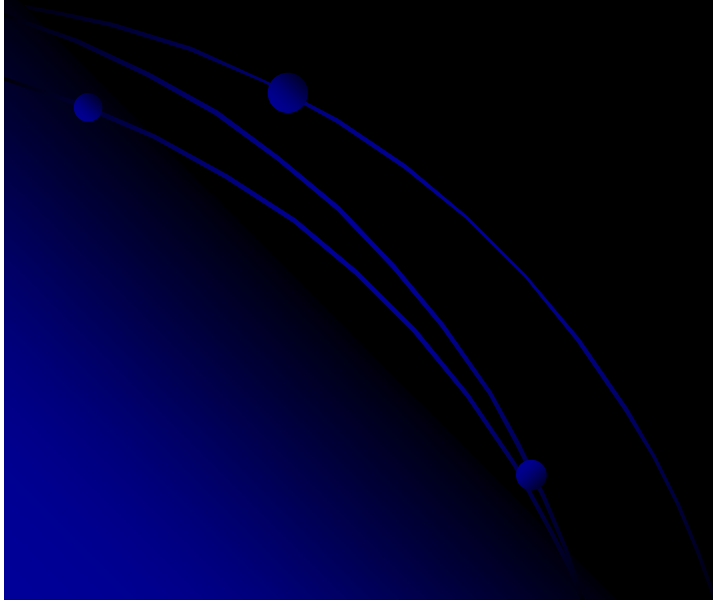
- A cross-sectional study.

DURATION

January 1, 2009 to August 31, 2009.

SUBJECTS AND METHODS

17 CKD 5 patients on hemodialysis and healthy children (normal controls) were included in the study. Non-probability purposive sampling was done.



SUBJECTS AND METHODS

The etiologies of CKD 5 included:

- Renal calculi n=3
- Reflux nephropathy n=3
- Uremic medullary cystic disease n=3
- Nephrocalcinosis n=2
- Membranoproliferative glomerulonephritis n=2
- Congenital renal hypoplasia n=1
- Neurogenic bladder n=1
- Renal parenchymal disease n=1
- Unknown n=1

INCLUSION CRITERIA

FOR PATIENTS:

Children:

1. of either gender aged 1 to 16 years.
2. diagnosed with CKD 5, on regular hemodialysis for 3 or more months.
3. having platelet count equal to or greater than $150 \times 10^9 /L$.

FOR CONTROLS:

Normal healthy children having the same inclusion criteria as above except for criteria 2.

EXCLUSION CRITERIA

All children who:

1. were diagnosed cases of acquired (other than CKD 5 for patients only) or hereditary platelet aggregation defect.
2. Had been transfused within 15 days prior to platelet aggregation studies.
3. Had taken any medication known to effect platelet aggregation within 15 days or 48 hours of the study according to the duration of effect of that drug. Exception of Ca antagonists and β -blockers in the case of CKD 5 on HD.

SUBJECTS AND METHODS

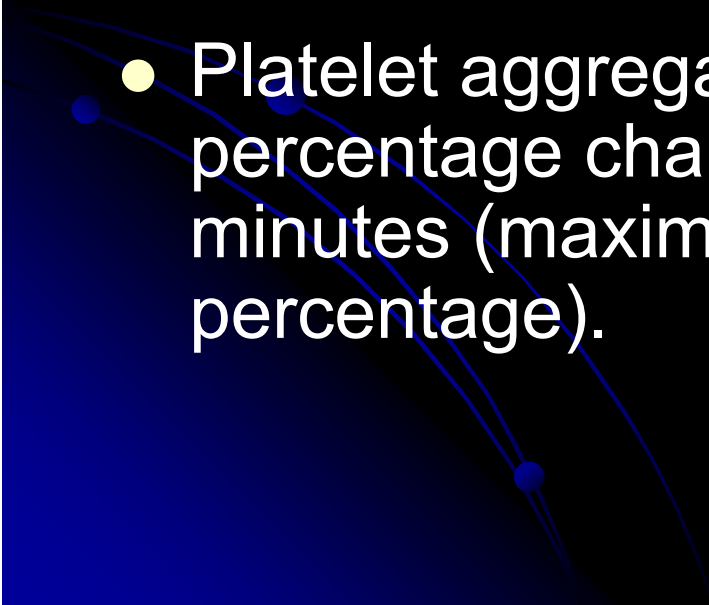
PATIENT CHARACTERISTICS

	Controls	CKD 5
Age	8 ± 3	11 ± 2
Gender M/F	5/12	8/9
Duration of HD (months)	0	6 ± 4
Erythropoietin	0	17
Ca antagonists	0	06
β-blockers	0	03

SUBJECTS AND METHODS

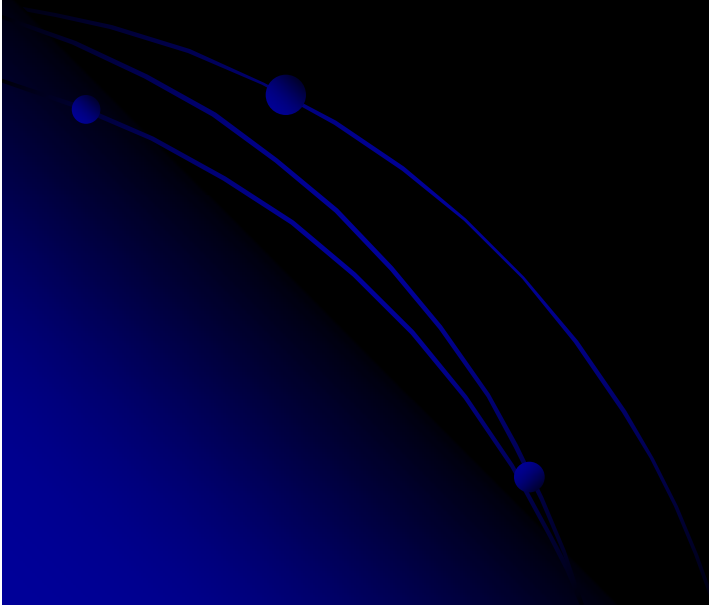
- For platelet aggregation studies predialysis blood samples were drawn directly from the dialysis tubing, immediately after insertion of the needle into the vascular access and prior to administration of heparin. In the case of control subjects blood was drawn from a forearm venipuncture with minimal venous occlusion.
- The citrated blood was centrifuged to obtain PRP. Platelet count of PRP was adjusted to between $200-400 \times 10^9/L$.

SUBJECTS AND METHODS

- Platelet aggregation was performed by turbidometric technique on Chronolog aggregometer by adding **10 μm** ADP and **1.25 mg/ml** Ristocetin in 250 μL PRP in separate cuvettes.
 - Platelet aggregation was recorded as the percentage change in light transmission after 3 minutes (maximal platelet aggregation percentage).
- 

STATISTICAL ANALYSIS

All results were expressed as mean values \pm SD. Paired Student's t test was used for comparison between two groups and a value where $p < 0.05$ was considered statistically significant.



RESULTS

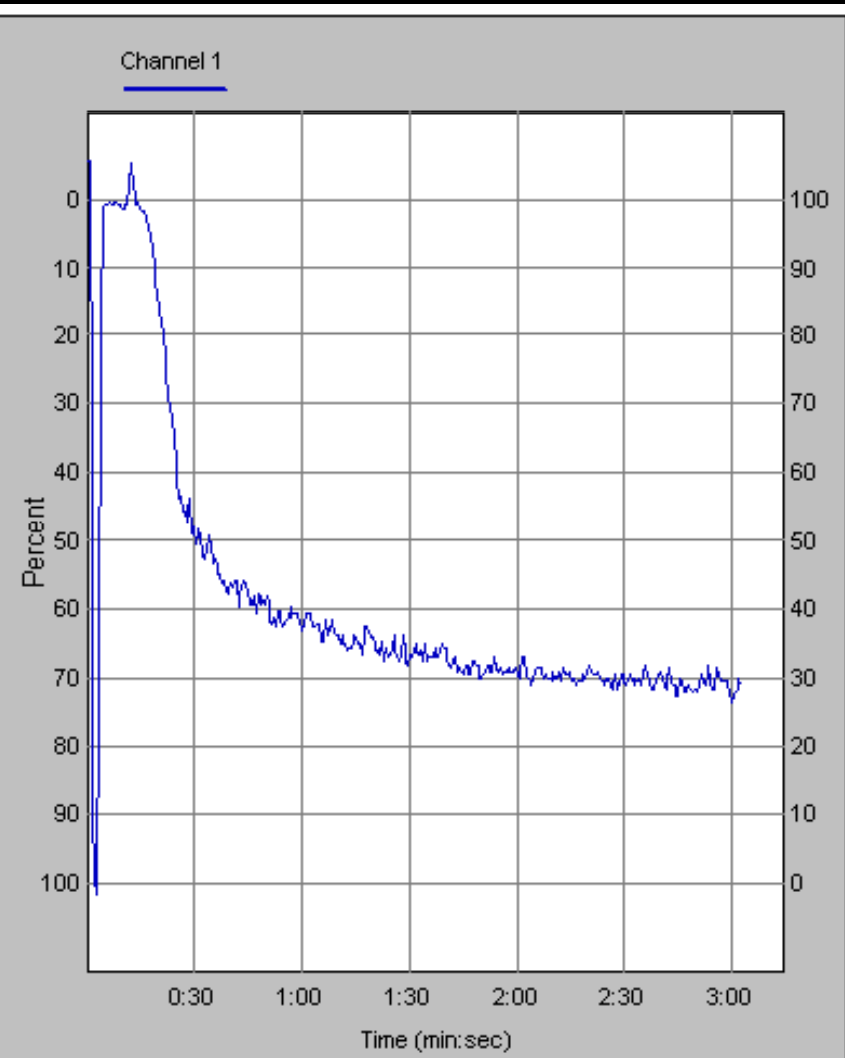
	Normal	CKD 5
ADP(%)	68 ± 6	57 ± 12*
Ristocetin(%)	80 ± 6	70 ± 13*

Data are means ± SD

*p= < 0.05 Significance was estimated using student's t-tests.

Platelet Aggregation

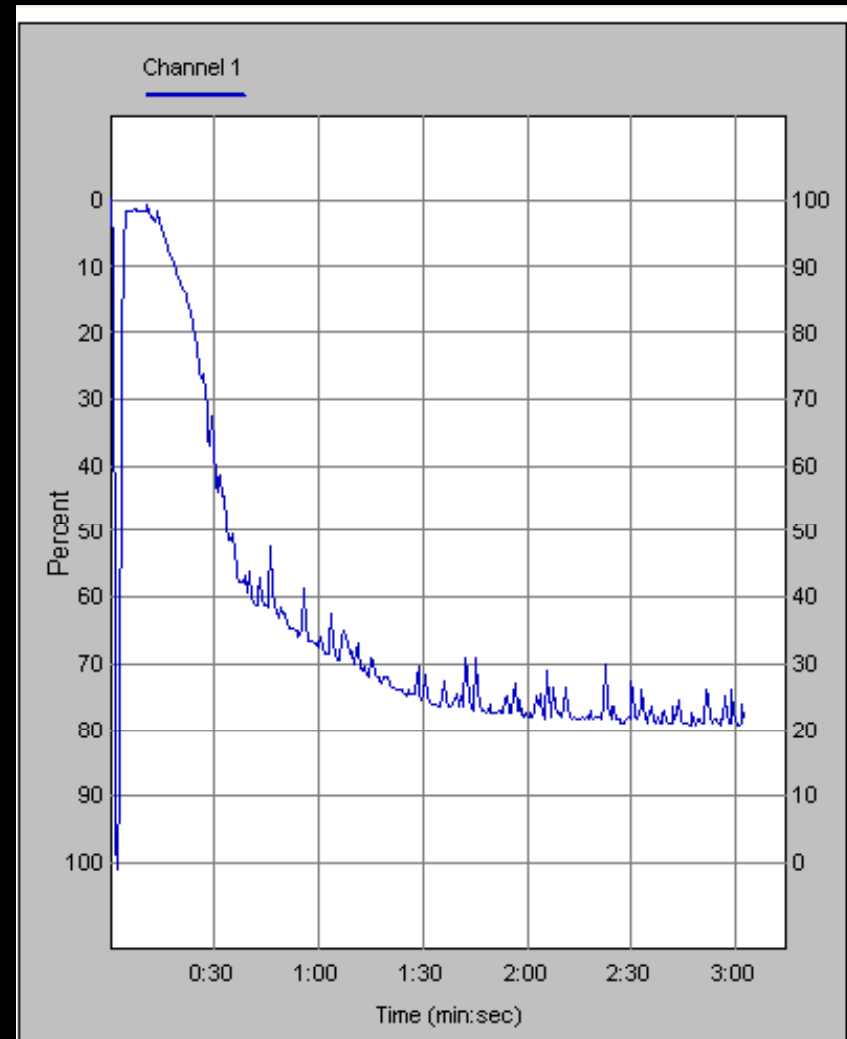
Platelet aggregation with
ADP in control.
Mean maximal platelet
aggregation percentage is
70%.



Platelet Aggregation

Platelet aggregation with Ristocetin in control.

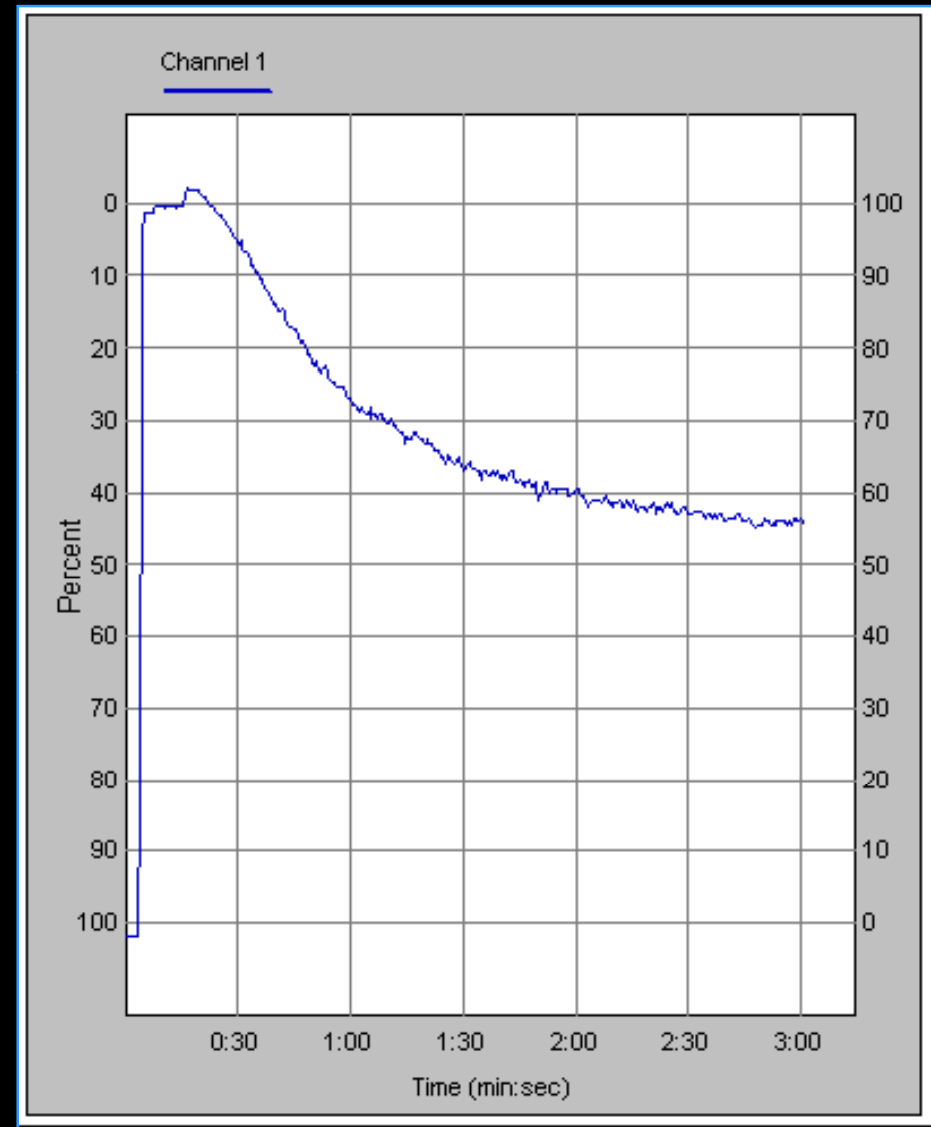
Mean maximal platelet aggregation percentage is 79%.



Platelet Aggregation

Platelet aggregation with ADP in one of the CKD 5 patients on hemodialysis.

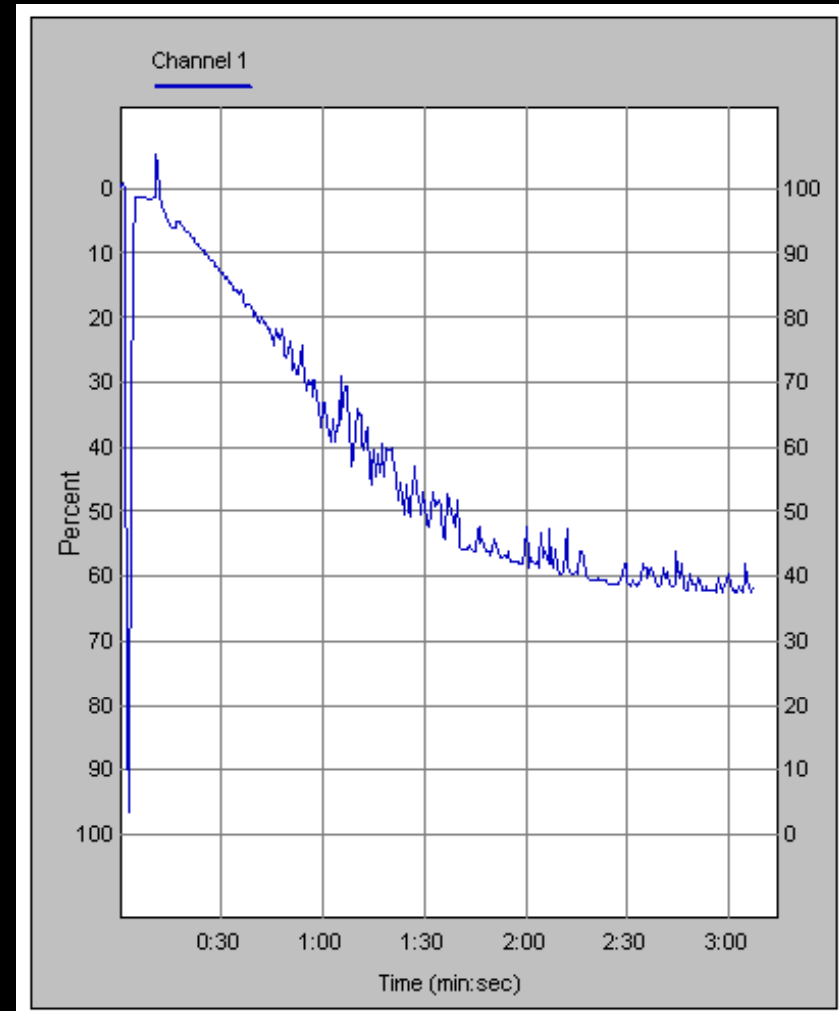
- Mean maximal platelet aggregation percentage is 43%.



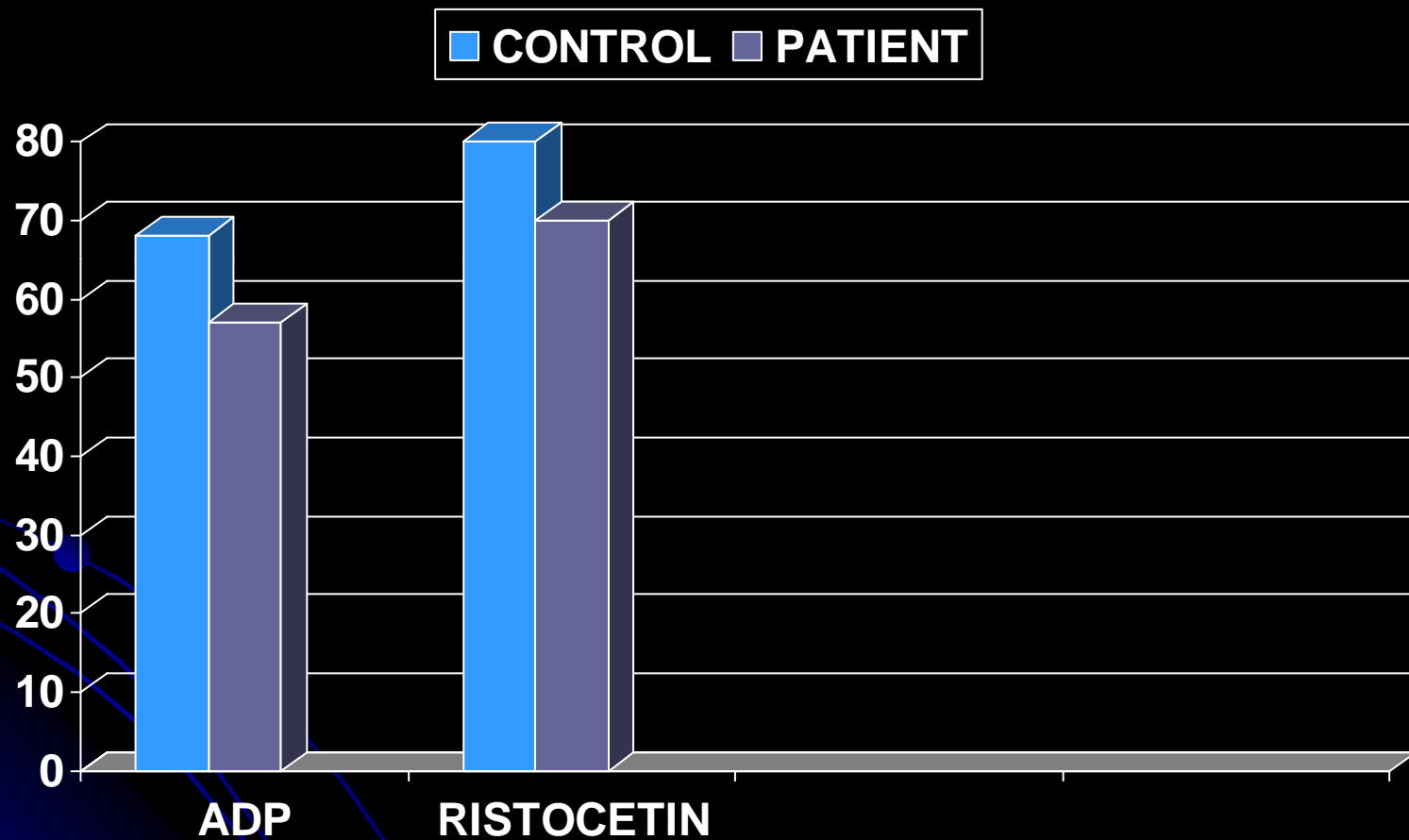
Platelet Aggregation

Platelet aggregation with Ristocetin in one of the CKD 5 patients on hemodialysis.

- Mean maximal platelet aggregation percentage is 60%.



PLATELET AGGREGATION



DISCUSSION

	Normal	CKD 5(HD)
Thekkedath et al Am J Hematol 2006; 81: 915-16	ADP (%) 100 (N=10)	52.5 ± 12.4 * (N=10)
Neiva T et al Braz J Med Biol Res 2002; 35:345-50	ADP(%) 78 ± 8.4 (N=20)	54 ± 20.5 * (N=55)
Moal V et al Nephrol Dial Transplant 2003;18:1834-41	ADP(%) 77 ± 11 Ristocetin 82 ± 8 (N=30)	76 ± 15 71 ± 9 * (N=22)

Data are means +/- SD

*p < 0.05 Significance was estimated by student's t-tests.

DISCUSSION

Ristocetin reacts with vWF and GPIb to induce platelets to clump together, while ADP causes platelets to aggregate by binding fibrinogen to GPIIb/IIIa forming fibrinogen-platelet meshes.

Integrity of GPs and a normal quantity of large molecular-weight multimeric vWF is required for normal platelet function.

DISCUSSION

My study shows platelets to be hyporesponsive to both ADP and Ristocetin.

This finding is consistent with the findings in studies in CKD 5 patients on HD which show reduced expression of GPIIb/IIIa after platelet stimulation with agonist, and reduced total GPIb.

Moal V, Brunet P, Dou L et al. Impaired expression of glycoproteins on resting and stimulated platelets in uraemic patients. *Nephrol Dial Transplant* 2003; 18:1834-41.

DISCUSSION

Another study suggests presence of fibrinogen fragments in CKD 5 patient's plasma which inhibit platelet function by competitive binding to GPIIb/IIIa receptors.

Thekkedath UR, Chiranthavat T, Leypoldt JK et al. Elevated fibrinogen fragment levels in uremic plasma inhibit platelet function and expression of glycoproteins IIb/IIIa. *Am J Hematol* 2006; 81: 915-26.

DISCUSSION

One study has shown decreased levels of high molecular weight multimers of vWF which explains decreased aggregation with ristocetin.

Lopez JA, Lockhart E. Acquired disorders of platelet function. In: Hoffman R, Benz EJ, Shattil SJ, editors. Hoffman: Hematology: Basic Principles and Practice. 5th ed. Elsevier. 2008. (Online)

DISCUSSION

On the other hand chronic HD and presence of prothrombotic state in CKD 5 leads to chronic low level platelet activation and subsequent refractoriness to subsequent stimulation by agonists.

In conclusion our findings indicate that platelets in our CKD 5 patients on HD were hyporesponsive to both ADP and Ristocetin. Further studies are needed to understand the cause of platelet dysfunction in CKD 5.

THANK YOU

