Flow Cytometry as a Diagnostic Tool in Monoclonal Gammopathy of Renal Significance

Dear Editor,

Renal diseases associated with monoclonal gammopathy without symptomatic multiple myeloma (MM), Waldenstrom's macroglobulinemia (WM), or chronic lymphocytic leukemia (CLL) are increasingly known.[1] Many of these patients have a small clonal population of plasma cells (PCs) or B cells. The International Kidney and Monoclonal Gammopathy Research group (IKMG) introduced the term monoclonal gammopathy of renal significance (MGRS) in 2012.[1] The MGRS includes monoclonal gammopathy of uncertain significance (MGUS), smoldering MM, smoldering WM, monoclonal B-cell lymphocytosis (MBL), CLL, and low-grade B-NHL associated with renal involvement.[1,2] The diagnosis of MGRS is based on renal biopsy and monoclonal protein identification. B-cell or PC clone identification is paramount for a clone-directed therapy for long-term hematologic response.[2] As these clones are small, a highly sensitive technique like flow cytometry (FCM) should be used to identify clonality.[3] It is important to identify MGRS as these patients do not respond well to immunosuppressive therapy, have a high rate of recurrence post renal transplantation, and can progress to corresponding hematological malignancy.[4]

We are describing two cases of MGRS where we could confirm the presence of a small clonal PC population using FCM. The case characteristics are listed in Table 1.

Table 1: Clinical characteristics of the two cases of MGRS

	Case 1	Case 2
Age in years/sex	55/Female	49/Male
Renal biopsy	C3 glomerulopathy	Monoclonal immunoglobulin deposition disease (IgG lambda)
Clinical symptoms	Bilateral lower limb swelling, periorbital swelling, hematuria and hypertension, transfusion-dependant anemia	Periorbital swelling, hypertension, progressive renal dysfunction requiring dialysis
Duration of symptoms	1 year	1.2 years
Hb (g/dL)	6.2	8.3
TLC ($\times 10^6/\mu L$)	5.2	7.3
Platelets ($\times 10^6/\mu L$)	246	187
Peripheral blood smear	Normocytic normochromic anemia, mild rouleaux formation	Normocytic normochromic anemia
Creatinine (mg/dL)	4.5	10.18
24-h urine protein (g/24 h)	1.5	1.8
SPEP (M-spike) (g/dL)/IFE	0.8, IgG kappa	0.2, IgG lambda
sFLC (kappa: lambda)	4.9	0.2
Imaging (whole-body CT scan/skeletal survey)	No skeletal lesions	No skeletal lesions
Renal biopsy (MGRS-related lesion)	C3 glomerulopathy	MIDD
Plasma cell % on bone marrow aspirate and biopsy	6%; 15% binucleate forms and Dutcher bodies seen	9%; 9%
IHC	Polyclonal pattern	Polyclonal pattern
Flow cytometry		
% Abnormal plasma cells in viable nucleated cells Abnormal to total plasma cell ratio	0.1% 0.5	0.8% 0.8
FISH panel for del 13q14.3, del 17p13, t (4;14), t (11;4), t (14;16)	Inadequate sample	del 13q14.3 was found in 13% of plasma cells
Therapy	Received three cycles of VCD	Completed two cycles of VCD
Response	Normal serum free light chain ratio, M spike- 0.35 g/dL, Hb- 8 g/dL with infrequent transfusion requirement	Reduced frequency of dialysis from twice a month before diagnosis to once in last 2 months post initiation of therapy

CT=computed tomography, FISH=fluorescence *in situ* hybridization, Hb=hemoglobin, IHC=immunohistochemistry, MGRS=monoclonal gammopathy of renal significance, VCD=bortezomib, dexamethasone, and cyclophosphamide, SPEP=serum protein electrophoresis, IFE=Immunofixation electrophoresis, sFLC=serum free light chain, MIDD=Monoclonal immune deposit disease

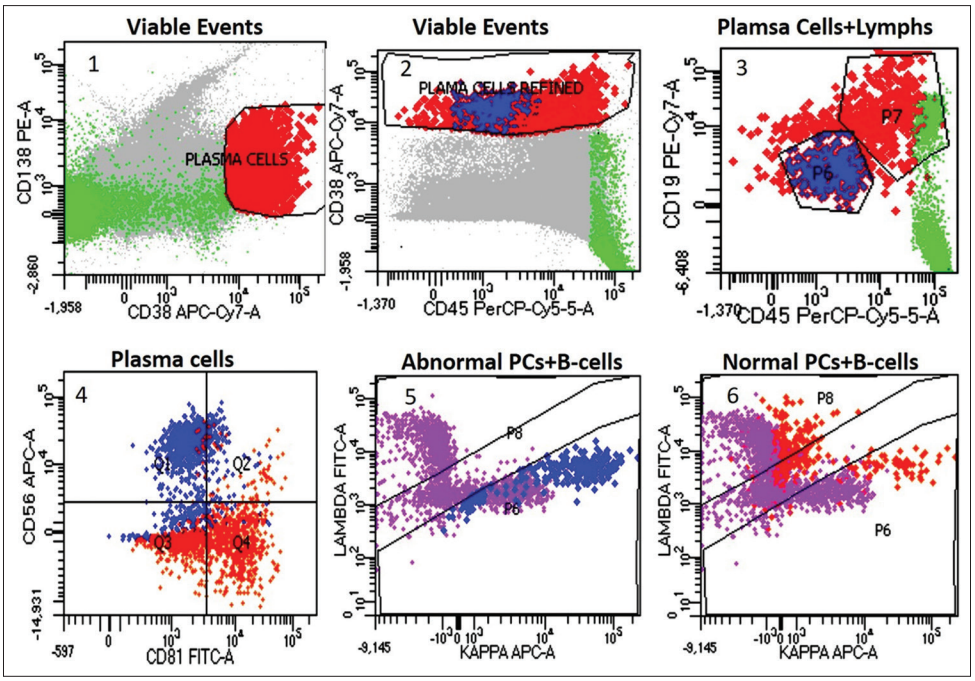

Figure 1: Plasma cell immunophenotyping in case 1: plasma cell gating on CD38/CD138 (plot 1), plasma cell gate refinement on CD38/CD45 (plot 2), abnormal PCs (blue), normal PCs (orange), and mature lymphocytes (green) on CD19/CD45 (plot 3), CD56 expression with CD81 loss in abnormal PCs, while the normal PCs express CD81 and are negative for CD56 (plot 4), κ-restricted abnormal PCs and polyclonal B cells (purple) (plot 5), polyclonal normal PCs (orange) and B cells (purple) (plot 6). PC = plasma cell

FCM for PCs was performed on the bone marrow (BM) sample collected in ethylenediaminetetraacetic acid (EDTA). The sample was lysed and staining done using a panel of antibodies against CD38-APC-Cy7, CD138-PE, CD45-PerCP-Cy5.5, CD19-PE-Cy7, CD27-FITC, CD81-FITC, CD56-APC, CD117-APC, intracellular anti-kappa-APC, and anti-lambda-FITC. Specimens were acquired using three-laser BD FACS Canto-II (BD Biosciences, San Jose, CA, USA) and analyzed on BD FACS Diva software version 8.0.1.

Case 1 showed 0.2% PCs on CD38, CD138, CD45, and side scatter (SSC) gating. Of these, half, that is, 0.1% PCs, showed an abnormal immunophenotype (CD56+/CD19−/CD81−/CD45) with a κ-restriction [Figure 1]. Case 2 showed 0.9% PCs on CD38, CD138, CD45, and SSC gating, including 0.8% λ-clonal PCs with an abnormal immunophenotype (CD56+/CD19−/CD27−/CD45 partial loss).

In both cases, clonality in BM could be proven on FCM, whereas immunohistochemistry showed a polyclonal population. Clonal identification is important as same renal lesions can be found in different hematological disorders and treatment varies depending upon the type of clone (B cell/PC).[2] Immunohistochemistry could be useful only when a major PC clone is present and polyclonal population is lacking.[2] However, immunohistochemistry has low sensitivity when less number of abnormal PCs are admixed with polyclonal population.[3] FCM has the advantage of studying a large number of cells and simultaneous measurement of multiple antigenic expressions. Sensitive FCM can detect monoclonal PCs at a sensitivity of 10^{-4}–10^{-6} and can discriminate between MGUS and MM. The number of residual polyclonal PCs is a useful discriminating marker between MGUS and MM.[5] MGUS usually has more than 5% normal plasma cells (NPCs) within total BM PCs (both our cases showed NPCs of 50% and 20%, respectively).[5]

To conclude, characterization and clonality identification of PCs or B cells in BM by FCM is a must in cases of MGRS as it is highly sensitive and guides in appropriate decision-making to guide correct therapy.

Acknowledgments

We would like to acknowledge the staff of Department of Hematology, AIIMS, New Delhi.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

Asish Rath, Jasmita Dass, Ganesh Kumar Viswanathan, Rishi Dhawan, Mukul Aggarwal, Manoranjan Mahapatra

Department of Hematology, All India Institute of Medical Sciences, New Delhi, India

Address for correspondence:
Dr. Jasmita Dass,
Department of Hematology, All India Institute of Medical Sciences, Room No. 206, AIIMS, New Delhi, India.
E-mail: jasmita@aiims.edu, drjasmita@gmail.com

References

1. Leung N, Bridoux F, Batuman V, Chaidos A, Cockwell P, D'Agati VD, *et al.* The evaluation of monoclonal gammopathy of renal significance: A consensus report of the International Kidney and Monoclonal Gammopathy Research Group. Nat Rev Nephrol 2019;15:45-59.
2. Jain A, Haynes R, Kothari J, Khera A, Soares M, Ramasamy K. Pathophysiology and management of monoclonal gammopathy of renal significance. Blood Adv 2019;3:2409-23.
3. Kumar S, Kimlinger T, Morice W. Immunophenotyping in multiple myeloma and related plasma cell disorders. Best Pract Res Clin Haematol 2010;23:433-51.
4. Leung N, Bridoux F, Nasr SH. Monoclonal gammopathy of renal significance. N Engl J Med 2021;384:1931-41.
5. Pérez-Persona E, Vidriales MB, Mateo G, García-Sanz R, Mateos MV, de Coca AG, *et al.* New criteria to identify risk of progression in monoclonal gammopathy of uncertain significance and smoldering multiple myeloma based on multiparameter flow cytometry analysis of bone marrow plasma cells. Blood 2007;110:2586-92.

Access this article online	
Quick Response Code:	Website: https://journals.lww.com/ijon
	DOI: 10.4103/ijn.ijn_335_22

How to cite this article: Rath A, Dass J, Viswanathan GK, Dhawan R, Aggarwal M, Mahapatra M. Flow cytometry as a diagnostic tool in monoclonal gammopathy of renal significance. Indian J Nephrol 2023;33:321-3.

Received: 10-10-2022, **Accepted:** 15-11-2022, **Published:** 08-03-2023