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Myeloma minimal residual disease testing in the United States: Evidence of improved standardization

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To the Editor

An increasingly important question for treating hematologists/oncologists managing patients diagnosed with multiple myeloma is how to assess response to therapy beyond clinical response. Therefore, minimal residual disease (MRD) testing became increasingly important in assessing treatment response in patients with myeloma. MRD negativity by flow cytometry (FC) is consistently associated with improved progression-free and overall survival. [1] This has prompted the Food and Drug Administration (FDA) to state that FC-MRD, if appropriately validated and standardized, could be used as a surrogate end point biomarker in clinical trials evaluating novel therapies.[2] Variability in FC-MRD methodology and sensitivity, however, remains a challenge. In a survey from 2013, 11/26 U.S. medical institutions confirmed performing myeloma FC-MRD, with enormous variability in methodology, and a striking 100-fold difference in the sensitivity (limit of detection; LOD).[3] Subsequently, an international consensus process was undertaken to provide validated standardized methods for myeloma FC-MRD testing. [4] We undertook a repeat survey to assess the current state of myeloma FC-MRD testing.

A 17 question survey was e-mailed to the 26 institutions participating in the previous survey and responses compared to those received in 2013. The survey evaluated progress in FC methodology and the developing role of molecular MRD (M-MRD) testing for myeloma. Ninety-six percent (25/26) of institutions responded (table I). The number of institutions performing myeloma FC-MRD testing was increased (56%) compared to the previous survey (42.3%). All laboratories performing FC-MRD set a defined number of acquisition events for MRD detection (in contrast to only 9/11 in 2013). International consensus on myeloma FC-MRD detection stipulates a minimal acceptable cell acquisition number of 2,000,000 events (3,000,000-5,000,000 acquired events recommended).[5, 6] The number of laboratories acquiring 2,000,000 events increased tremendously (42.9%) compared to 2013 (9.1%). On the other hand, fewer laboratories are acquiring <2,000,000 events in this survey (57.1%) compared to the earlier survey (72.7%). As the LOD in FC-MRD testing is highly dependent upon the number of events acquired, improved LOD was demonstrated, with 6/14 (42.9%) laboratories achieving an LOD of 0.001% or better, in contrast to only 2/11 (18.2%) in 2013. Greater sensitivity is attained with technology utilizing a higher number of parameters/colors as it enhances the ability to differentiate normal from abnormal plasma cells. Three laboratories (21.4%) currently utilize 10 color FC (none did in 2013) and only 1/14 (7.1%) laboratories uses less than 8 color FC, compared to 6/11 (54.5%) laboratories in 2013. Specimens should be concentrated before staining to maximize cell yield for acquisition and to decrease antibody cost; therefore, erythrocytes lysing before staining (“pre-lyse”), and subsequent concentration of leukocytes is the preferred method. [5] However, most laboratories performing FC-MRD (9/14, 64.3%) continue to stain unconcentrated cells followed by red cell lysis (“stain then lyse”), similar to the findings of the 2013 survey (7/11, 63.6%). Molecular methods, mainly based on next generation sequencing (NGS), was reported by 5 laboratories. Per the new IMWG response criteria, [7] NGS is part of the approved testing platforms to determine MRD status. Based on ongoing development in the field, in the near future, it seems reasonable to believe that the use of NGS will continue to increase for the purpose of MRD testing. Indeed, NGS has fewer limitations and practical hurdles when it comes to standardization than FC-MRD has. Currently, a major limitation of NGS-based MRD assays is the restricted availability. In the near future, as new products become available, this is anticipated to be resolved.

This survey shows that more institutions are implementing myeloma FC-MRD testing, with improved sensitivity/LOD. Areas for improvement include 1) defining a minimal acceptable cell acquisition number, and 2) implementation of the recommended “pre-lyse” staining method. However, there are still areas for improvement to be done as only 6/14 laboratories acquire the minimal acceptable number of cells and only 5/14 utilize the optimal “pre-lyse” staining method. Ideally, a more standardized approach to myeloma MRD testing should hopefully be implemented. Standardization of FC-MRD in myeloma was recently included in the updated International Myeloma Working Group (IMWG) response criteria, [7] allowing MRD to serve as an endpoint in clinical trials. This is important as it facilitates the development of MRD as a future surrogate regulatory endpoint for accelerated FDA approval of novel agents in myeloma treatment.

The results of this survey are clinically relevant as they give a current update on the status of MRD monitoring in multiple myeloma in the US. Treating hematologists/oncologists

managing myeloma patients need to stay aware of the rapidly evolving field and to ensure that their own patients are monitored with up-to-date assays.

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References

1. Rawstron AC, Child JA, de Tute RM, et al. Minimal residual disease assessed by multiparameter flow cytometry in multiple myeloma: impact on outcome in the Medical Research Council Myeloma IX Study. *J Clin Oncol*. 2013; 31:2540–2547. [PubMed: 23733781]
2. Landgren O, Gormley N, Turley D, et al. Flow cytometry detection of minimal residual disease in multiple myeloma: Lessons learned at FDA-NCI roundtable symposium. *American journal of hematology*. 2014; 89:1159–1160. [PubMed: 25132630]
3. Flanders A, Stetler-Stevenson M, Landgren O. Minimal residual disease testing in multiple myeloma by flow cytometry: major heterogeneity. *Blood*. 2013; 122:1088–1089. [PubMed: 23929839]
4. Rawstron AC, Pavia B, Stetler-Stevenson M. Assessment of minimal residual disease in myeloma and the need for a consensus approach. *Cytometry Part B, Clinical cytometry*. 2015
5. Stetler-Stevenson M, Paiva B, Stoolman L, et al. Consensus guidelines for myeloma minimal residual disease sample staining and data acquisition. *Cytometry Part B, Clinical cytometry*. 2015
6. Arroz M, Came N, Lin P, et al. Consensus guidelines on plasma cell myeloma minimal residual disease analysis and reporting. *Cytometry B Clin Cytom*. 2015
7. Kumar S, Paiva B, Anderson KC, et al. International Myeloma Working Group consensus criteria for response and minimal residual disease assessment in multiple myeloma. *The Lancet Oncology*. 2016; 17:e328–346. [PubMed: 27511158]

Table I

Summary of the results obtained from the participating laboratories in 2015 survey

Institution	MRD testing		Number of FC colors		Number of antigens studied in MRD		Total acquired events in MRD testing (in millions)		Minimum number of aPCs required for MRD		LOD/ maximum possible sensitivity (%) ^a	
	2013	2015	2013	2015	2013	2015	2013	2015	2013	2015	2013	2015
1	Yes	Yes	8	8	12	12	3 - 4	3 - 4	20	20	0.0005	0.0005
2	Yes	Yes	9	9	8	8	1.8	0.5 - 0.6	50	50	0.003	0.008
3	Yes	Yes	8	8	8	8	1	2	50	20	0.005	0.001
4	Yes	Yes	7	8	6	6	0.5	0.5	25	25	0.005	0.005
5	Yes	Yes	8	8	8	8	0.5	0.5	n/a	n/a	n/a	n/a
6	Yes	Yes	6/8	8	8	8	0.3 - 0.5	> 0.1	30 - 35	n/a	0.006	n/a
7	Yes	Yes	7	8	10	10	0.25 - 0.5	5	50	50	0.01	0.001
8	Yes	Yes	6/7	8	9	9	0.1	2	20	20	0.02	0.001
9 ^b	Yes	nr	5	nr	8	nr	0.1	nr	Variable ^c	nr	n/a	nr
10	Yes	Yes	8	8	8	8	Variable	2.5	20	20	n/a	0.0008
11	Yes	Yes	5	10	8	11	n/a	0.5	30	30-40	n/a	0.006
12 ^d	No	Yes ^e	nr	6	8	8	n/a	0.1 - 0.25	nr	20-25	nr	0.008
13 ^d	No	Yes	nr	8	9	9	n/a	5	nr	50	nr	0.001
14 ^d	No	Yes ^e	nr	10	12	12	n/a	1.5	nr	n/a	nr	n/a
15 ^d	No	Yes	nr	10	18	18	n/a	0.3	nr	20	nr	0.007

MRD; minimal residual disease; FC; flow cytometry, aPCs; abnormal plasma cells, LOD; limit of detection, n/a; not applicable, nr; no result available (due to lack of participation as in case of institution 9, or no MRD studies were done as in case of institutions 12-15).

^a LOD/maximum possible sensitivity is determined by dividing the minimum number of aPCs required for MRD by the highest number of total acquired events in MRD testing and multiplying by 100. A lower percentage indicates a more sensitive test.

^b Institution 9 did not participate in the current survey.

^c The minimum number of APC was variable and pathologist dependent.

^d Institutions 12-15 were not performing MRD testing in 2013.

^e MRD is under validation.