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Minimal Residual Disease in Myeloma in 2024: **Where We are Today**

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Introduction

Minimal residual disease (MRD) refers to a small population of cancer cells that persists in the body after treatment. Often undetectable using traditional diagnostic methods, these cells can eventually cause relapse in patients who appear to have achieved a complete response (CR) to treatment. For that reason, MRD has become a vital parameter in evaluating the effectiveness of cancer therapies, particularly in hematological malignancies, such as multiple myeloma (MM), and certain solid tumours.^{1,2}

Detection of MRD represents a challenge, as the disease may not cause symptoms or be detected through traditional methods (i.e., visible under a microscope). Nevertheless, these cells are often responsible for disease relapse; alternatively, sustained absence of these cells may portend a prolonged remission and presumably be required for disease cure. Therefore, monitoring and detecting MRD are increasingly recognized as essential for long-term patient care and treatment planning.^{3,4}

Importance of MRD Detection and Monitoring

MRD detection and monitoring play a critical role in the following:

- **Assessing the depth of treatment response:** by measuring how much residual disease remains after treatment, physicians can gauge the true effectiveness of therapy.
- **Predicting relapse:** MRD-positive patients are at a higher risk of relapse. Continuous monitoring can help identify early signs of recurrence, even before clinical symptoms arise.

- **Tailoring treatment plans:** MRD detection allows personalized treatment approaches, such as intensifying or de-escalating therapy based on a patient's MRD status.

In the realm of MM, achieving MRD-negative status—meaning no residual disease is detected—is increasingly viewed as the gold standard for treatment success. The absence of detectable MRD correlates strongly with improved outcomes, such as progression-free survival (PFS) and overall survival (OS).³⁻⁶

Methods for Detecting MRD

Several advanced techniques have been developed for detecting MRD, each offering varying degrees of sensitivity and specificity:

1. **Real-time quantitative polymerase chain reaction (RQ-PCR):** this method detects residual disease by measuring specific genetic abnormalities, such as fusion genes, overexpressed genes, or mutations, that are unique to cancer cells. Although highly sensitive, it is limited by the requirement for specific primers and probes designed to target individual tumour characteristics.^{2,7,8}
2. **Multiparametric flow cytometry (MFC):** this approach uses antibodies tagged with fluorescent markers to identify cancer cells based on their surface proteins. A laser beam analyzes these cells, making it possible to detect multiple markers simultaneously. MFC can detect one cancer cell among 10,000 to 100,000 normal cells (10^{-4} to 10^{-5} sensitivity), and a more advanced version, next-generation flow cytometry (NGF), offers even higher sensitivity.^{2,4,9}

3. Next-generation sequencing (NGS): NGS examines thousands of genes simultaneously to detect residual disease with extremely high sensitivity (10^{-6} to 10^{-7}). This method is highly specific and has been increasingly adopted for monitoring MRD in various cancers, including MM.^{2,10}

MRD in MM

MM is a cancer of plasma cells that primarily affects the bone marrow. MRD testing has become critical in evaluating treatment outcomes in MM, especially as newer therapies result in deeper responses. Traditionally, treatment responses in MM were measured by evaluating monoclonal protein levels in the blood and urine or assessing bone marrow plasma cell involvement. However, the introduction of highly effective agents like proteasome inhibitors, immunomodulatory drugs, and monoclonal antibodies has increased the frequency of CRs, necessitating more sensitive methods to track MRD.^{1,2}

Therapeutic Advances and MRD in MM

Over the last two decades, MM treatment has significantly advanced with the approval of drugs like:

- **Proteasome inhibitors** (e.g., bortezomib, carfilzomib, ixazomib)
- **Immunomodulatory drugs** (e.g., lenalidomide, pomalidomide)
- **Monoclonal antibodies** (e.g., daratumumab, isatuximab)

The use of daratumumab combined with carfilzomib, lenalidomide, and dexamethasone (Dara-KRd) has led to deeper treatment responses, with CR rates as high as 95% in newly diagnosed patients.¹¹

The increasing depth of response induced by these novel therapies has made MRD testing more crucial than ever for determining long-term outcomes. Studies have demonstrated that MRD-negative patients have significantly longer PFS and OS compared to those who remain MRD-positive, even if they achieve CR by conventional measures.^{3,4}

MRD Testing: NGF vs. NGS

In MM, MRD-negative status is defined by the absence of detectable cancer cells, typically using highly sensitive methods such as Next-Generation Flow Cytometry (NGF) or NGS (**Table 1**).

- 1. NGF:** This method is capable of detecting MRD with a sensitivity of 10^{-6} and is increasingly used in clinical practice to monitor residual disease in patients with MM. NGF does not require a baseline sample, making it particularly useful in clinical settings.
- 2. NGS:** This method uses primers to amplify immunoglobulin gene segments, allowing for the detection of clonal plasma cells with high sensitivity. NGS requires a baseline sample to track the cancer clone but offers superior sensitivity, detecting one cancer cell among a million normal cells (10^{-6} to 10^{-7}).

Studies have shown high concordance between NGF and NGS, with both methods yielding similar results in over 80% of cases. However, NGS requires a baseline sample, while NGF does not, giving each method certain advantages depending on the clinical scenario. MRD detection methods like NGS and NGF are proving to be highly predictive of long-term patient outcomes, particularly in patients with newly diagnosed MM.^{9,10,12}

MRD and Patient Prognosis

MRD status has become a key factor in determining patient prognosis in MM. For example, a recent meta-analysis of clinical trials demonstrated that MRD-negative status was associated with:

- A hazard ratio (HR) of 0.33 for PFS, meaning MRD-negative patients had a 67% lower risk of disease progression or death compared to MRD-positive patients.³
- An HR of 0.45 for OS, meaning patients with MRD-negative disease had a 55% lower risk of death compared to MRD-positive patients.¹³

These findings apply across various subgroups, including patients with high-risk disease or those with relapsed MM.

	Next-generation flow cytometry (NGF)	Next-generation sequencing (NGS)
Reproducibility among centers	High	Limited Centers available
Baseline assessment	Not required	Required
Processing requirements	Fresh Samples <36 h	Fresh and stored samples
Standardization	EuroFlow Consortium	Commercial companies. (Adaptative Biotechnologies)
Quantitative	Yes	Yes
Sensitivity	1 in 10 ⁻⁵ -10 ⁻⁶	1 in 10 ⁻⁵ -10 ⁻⁶
Time to processing	<24 hours	1–2 weeks
Clonal evolution evaluation	Not evaluable	Evaluable
Cost	300 USD	700–1500 USD

Table 1. Minimal Residual Disease Assessment Techniques; adapted from Pavia et al.²⁴ and Mina et al.²⁵

Challenges and Limitations of MRD Testing

While MRD testing offers significant prognostic value, several limitations and challenges remain:

- 1. Bone marrow sampling:** MRD testing often requires bone marrow aspirates, which can be invasive and painful. Furthermore, bone marrow involvement in MM may not be uniform, leading to variability in MRD test results.¹⁴
- 2. Extramedullary disease:** MRD testing primarily focuses on the bone marrow, but MM can present as extramedullary disease (i.e., disease outside the bone marrow). For instance, some patients who are MRD-negative in the bone marrow still show signs of disease in imaging studies, such as positron emission tomography-computed tomography (PET-CT) scans. This discrepancy highlights the importance of using multiple diagnostic modalities to fully assess disease status.^{1,14}

- 3. Relapse prediction:** one of the key advantages of MRD testing is its ability to predict relapse before clinical symptoms appear. Patients who remain MRD-positive after treatment are at higher risk of relapse, often several months before biochemical or clinical indicators emerge. This raises the question of whether early intervention at the point of MRD detection could improve long-term outcomes.¹⁵
- 4. Liquid biopsies:** a less invasive alternative to bone marrow sampling is the use of liquid biopsies to detect circulating tumour DNA (ctDNA) or plasma cells in the peripheral blood. While this method is less invasive, its sensitivity is currently lower than that of bone marrow-based tests.^{16,17}
- 5. Mass spectrometry:** emerging technologies like mass spectrometry are also being explored as potential tools for detecting MRD. Mass spectrometry can measure low levels of monoclonal protein in the blood, and it has shown promise as a highly sensitive technique for identifying residual disease in patients with MM.¹⁸

MRD as a Clinical Endpoint and Surrogate Marker

MRD status is increasingly being used as a prognostic tool in clinical trials. Many trials now include MRD as an endpoint, and its presence or absence can help stratify patients based on their risk of relapse and overall prognosis.^{19,20} Guidelines from the International Myeloma Working Group (IMWG) recommend a sensitivity threshold of 10^{-5} for MRD testing. Sustained MRD negativity, defined as maintaining MRD-negative status for at least one year, is now considered the optimal endpoint in assessing long-term treatment efficacy.²

Several ongoing trials are using MRD to guide treatment decisions, with different strategies under investigation:

- 1. Intensification of therapy:** some trials are investigating whether intensifying treatment can improve outcomes for patients who remain MRD-positive after initial therapy. The AURIGA trial, for example, is evaluating the role of adding daratumumab to lenalidomide maintenance to deepen responses in patients who remain MRD-positive.^{19,21}
- 2. De-escalation of therapy:** other trials are exploring whether patients who achieve sustained MRD negativity can safely discontinue treatment. For example, the DRAMMATIC trial is investigating whether MRD-negative patients can stop maintenance therapy without compromising outcomes.²²
- 3. Early treatment of MRD relapse:** some trials, like the REMNANT study, are investigating whether treating patients at the time of MRD relapse—before biochemical or clinical relapse—can improve long-term outcomes. This approach aims to intervene at the earliest sign of disease recurrence, potentially preventing full clinical relapse.²³

Conclusion

MRD detection has become an essential tool in the management of MM and other hematological malignancies. The development of sensitive techniques like NGS and NGF has revolutionized our ability to measure disease burden, allowing the detection of even the smallest number of

remaining cancer cells. Achieving MRD-negative status is associated with significantly improved outcomes in MM, including longer PFS and OS.

Despite the remarkable advancements in MRD testing, several challenges remain, particularly in detecting extramedullary disease and developing less invasive diagnostic techniques. Nonetheless, the ongoing integration of MRD testing into clinical trials and treatment strategies provides critical insights into disease management, helping tailor therapy to individual patient needs and improve long-term survival.

As MRD testing continues to evolve, it will likely play an increasingly important role in personalized medicine, guiding treatment decisions and helping predict relapse before it occurs. The ultimate goal is to use MRD testing not only as a prognostic tool but also as a guide for real-time treatment modifications, helping to achieve the best possible outcomes for patients with MM.

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