LEUKEMIA & LYMPHOMA SOCIETY®

HEALTHCARE PROFESSIONALS

No. 5 in a series
providing the latest information

Facts About
Measurable Residual Disease (MRD)

Introduction

Patients who achieve complete hematologic remission after treatment for blood cancer often harbor residual cancer cells in the bone marrow or peripheral blood that can result in relapse. These cells can be present at levels so low they are undetectable by conventional cytomorphology. The ability to detect low levels of residual cells, referred to as measurable residual disease (sometimes called minimal residual disease, or MRD) has vastly improved in recent years. Technology available today can detect the presence of 1 cancer cell in 10,000–1,000,000 nucleated cells, compared to 1 cancer cell in 20 nucleated cells for conventional cytomorphology. Due to advancing technology in MRD assessment, "remission" in acute lymphoblastic leukemia (ALL) has been redefined and new response categories in acute myeloid leukemia (AML) and multiple myeloma (MM) have been determined.

It is important for clinicians to understand the different methods available to assess MRD, how samples should be obtained, and how to interpret the results to best inform risk assessment and make treatment decisions. This Fact Sheet will explain the methods currently used for MRD assessment, how and when testing should occur for different hematologic malignancies, and how the resulting information can inform prognosis and decisions about care.

Highlights

- Methods used for MRD assessment include multiparameter flow cytometry (MFC), real-time quantitative polymerase chain reaction (RQ-PCR) and next-generation sequencing (NGS)-based assays.
- MFC can detect 1 cancer cell in 10,000–100,000 nucleated cells. It relies on the detection of antigens on neoplastic cells as compared to normal cells.
- RQ-PCR is able to detect 1 cancer cell in 100,000 nucleated cells. It is widely used for patients harboring well-defined genetic aberrations, like the *BCR-ABL1* fusion gene.
- Some newer NGS-based assays developed to address some of the limitations of RQ-PCR can detect 1 cancer cell in 1 million nucleated cells.
- The ClonoSEQ® assay is an NGS-based assay that is FDA-cleared for MRD analysis in B-cell ALL [B-ALL], chronic lymphocytic leukemia (CLL) and multiple myeloma (MM).
- MRD assessment is standard clinical practice in both adult and pediatric ALL to predict outcomes and guide therapy.
- MRD monitoring is used in AML as a prognostic indicator, to identify impending relapse and allow for robust post-transplant surveillance.
- In MM, MRD is used to measure depth of response at each stage of treatment to inform prognosis.
- MRD analysis in chronic myeloid leukemia (CML) is used to gauge response to therapy, inform prognosis and identify patients in deep remission who might discontinue therapy.

MRD Basics: Sensitivity and Methods of Assessment

The sensitivity, or detection thresholds, of different MRD techniques can be expressed in several ways, reflecting the number of cancer cells that can be detected in a sample per the number nucleated cells. This is shown in **Table 1.**

Table 1. MRD Detection Sensitivity Thresholds

Maximum Sensitivity (no. cancer cells per no. nucleated cells)	Percentage	Sensitivity Threshold
1 in 20	5%	
1 in 1,000	0.1%	10^{-3}
1 in 10,000	0.01%	10^{-4}
1 in 100,000	0.001%	10^{-5}
1 in 1,000,000	0.0001%	10^{-6}

The modalities used for MRD assessment in hematologic malignancies include multiparameter flow cytometry (MFC) and molecular methods, including real-time quantitative polymerase chain reaction (RQ-PCR) and next-generation sequencing (NGS)-based assays. These modalities differ in their sensitivity thresholds and clinical applications.

Sampling for MRD Assessment

For MRD assessment of bone marrow (BM) samples, the quality of the sample is very important. In order to avoid false negative results that can be caused by hemodilution, a sample of 2–5 mL resulting from the first pull is recommended. Dividing a large-volume pull is not appropriate for MRD assessment. If an interventional radiologist will be involved in the aspiration, it is important to communicate that the sample is for MRD analysis so that appropriate guidelines can be followed.[1-5]

Multiparameter Flow Cytometry (MFC)

MFC relies on the detection of the expression of antigens on neoplastic cells compared to normal cells. MFC involves labeling cells in suspension with fluorochrome-linked antibodies specific to cancer cell antigens.[6]

There are 2 MFC-based methods for quantitating MRD[7]:

1. **Leukemia-associated immunophenotypes (LAIPs).** This method relies on identification of the immunophenotype of leukemia blasts at the time of diagnosis that can then be followed over time. "MRD positive" refers to the presence of cells with the identified immunophenotype after treatment.

2. **"Different from Normal" (DfN).** This approach relies on the differences between the immunophenotype of cells in the MRD sample compared to a stereotypical "normal" sample. A diagnostic sample is not necessarily needed for the DfN approach.

List of Abbreviations

ALL: Acute lymphoblastic leukemia
AML: Acute myeloid leukemia
ASO: Allele-specific oligonucleotide
BiTE: Bi-specific T-cell engager
BM: Bone marrow
B-ALL: B-cell ALL
CLL: Chronic lymphocytic leukemia
CML: Chronic myeloid leukemia
CR: Complete remission
DfN: "Different from Normal"
ELN: European LeukemiaNet
HSCT: Hematopoietic stem-cell transplantation
Ig: Immunoglobulin
IMWG: International Myeloma Working Group
IS: International Scale
LAIP: Leukemia-associated immunophenotype
MFC: Multiparameter flow cytometry
MM: Multiple myeloma
MMR: Major molecular response
MRD: Measurable (or minimal) residual disease
MRI: Magnetic resonance imaging
NGF: Next-generation flow cytometry
NGS: Next-generation sequencing
PB: Peripheral blood
PET: Positron emission tomography
PET-CT: PET-computed tomography
Ph+: Philadelphia chromosome positive
RQ-PCR: Real-time quantitative polymerase chain reaction
TCR: T-cell receptor
T-ALL: T-cell ALL
WBC: White blood cell

MFC is widely available and has a sensitivity down to 10^{-4}, or 10^{-5} with next-generation technology. The sensitivity increases with increased numbers of cellular events captured by the flow cytometer. MFC is applicable to nearly 100% of patients with AML, ALL, CLL and MM. Most laboratories use at least 6-color flow cytometry assays for MRD assessment, which can provide a detection sensitivity down to 10^{-4}.[2]

Sampling for MFC

A fresh sample is required for MRD analysis by MFC. As mentioned above, the detection threshold depends on the number of events captured by the flow cytometer. In order to reach a detection sensitivity of 10^{-4}, 200,000–500,000 cells must be sorted. A sensitivity of 10^{-5} requires that 2–5 million cells are sorted, which is difficult to obtain.[1]

It is important for clinicians to communicate with the laboratory that an MRD assessment is being performed to ensure enough events are run and that the proper antibody panel is used. Without such guidance, clinical laboratories can fail to capture enough events to assure high sensitivity MRD assessments.[1] Because immunotherapy can affect the immunophenotype of the cells being monitored, it is important to inform the laboratory performing MRD analysis if the patient has received immunotherapy.[2]

In order to accurately interpret MRD assessments by MFC, clinicians should be aware of the antibody panels being used and the number of cellular events captured in the analysis. There is also a degree of operator subjectivity in the interpretation of MFC results, which should be considered when comparing analyses at different timepoints across different labs and pathologists

Molecular Techniques

Genetic approaches to MRD assessment in hematologic malignancies rely on the following observations[3]:

1. More than 90% of lymphoid malignancies contain clonally rearranged immunoglobulin (Ig) and/or T-cell receptor (TCR) genes ("VDJ recombination")

2. In 25%–35% of cases there are well-defined chromosome aberrations or mutations

Due to the clonal expansion of cancerous cells, the sequences of VDJ regions can be used to quantify disease.[3] The most commonly used methods for genetic MRD assessment are (RQ-PCR)- and (NGS)-based assays.[2]

RQ-PCR

PCR is widely used for patients harboring well-defined genetic aberrations, like the *BCR-ABL1* fusion gene in Philadelphia chromosome positive (Ph+) ALL and chronic myeloid leukemia (CML).[2,7,11,12] RQPCR to quantitate MRD for these patients is simple, inexpensive, and widely applicable.[7,12,13] RQ-PCR has a sensitivity threshold of approximately 10^{-5}.[14]

Allele-specific oligonucleotide (ASO)-based RQ-PCR can be used in malignancies that do not harbor welldefined genetic aberrations. It involves quantifying Ig or TCR rearrangements using primers and patient-specific PCR probes. In this approach, leukemia-specific Ig/TCR rearrangements must first be identified for each patient by sequencing these regions in a diagnostic sample. Patientspecific primers must then be generated so that MRD can be quantitated through RQ-PCR in a post-treatment sample.[14] In patients with ALL, ASO-based RQ-PCR is a feasible approach in 90%–95% of cases.[7,13] For multiple myeloma (MM), it is applicable in 60%–70% of cases.[15,16]

Because ASO-based RQ-PCR is labor intensive, it is also expensive. While used widely in Europe (the EuroMRD Consortium has developed guidelines for the interpretation of data), it is not typically used in the US.

Sampling for RQ-PCR

PCR approaches do not require a fresh sample—both fresh and stored material can be used. If an ASO-based RQ-PCR approach is used, a baseline sample with detectable disease is required in order to subsequently characterize the clones that will be analyzed.[4]

Next Generation Sequencing (NGS)

In order to address some of the limitations of RQ-PCR, MRD assessment technologies that combine PCR and NGS approaches have been developed. Instead of relying on unique, patient-specific PCR primers and probes, this technology relies on PCR "consensus primers" that allow the amplification of the complete set of Ig or TCR gene sequences in a patient sample.[18]

Once amplified, the collective samples are immobilized on a glass chip and sequenced simultaneously using NGS technology. The frequencies of different clonotypes in the baseline sample are determined, which can then be followed over time to measure disease burden.[18] MRD is quantified using bioinformatic analysis.[19]

A high disease burden sample is required in order to identify the dominant clone.[1] There may be multiple MRD-relevant clones that can be identified through sequencing. NGS has a sensitivity threshold >10^{-6} and has been shown to be highly concordant with MFC and PCR techniques.[7] This approach is available commercially as the ClonoSEQ® Assay, which has received FDA-clearance for MRD assessment in patients with B-ALL, CLL and MM.

Sampling for NGS

For NGS, a fresh sample is often preferred, but it may not always be possible. While it can be successfully performed on fewer than 1 million cells, higher numbers result in improved assay sensitivity.[15] For ALL and MM, suitable tissues for NGS determination of MRD include[1,15,16]:

- Fresh or frozen bone marrow
- Fresh or frozen peripheral blood (PB)
- Fresh or frozen tissue
- Bone marrow aspirate slides can be scraped to obtain material for NGS analysis
- Formalin-fixed paraffin-embedded bone marrow clot sections and tissue

While all of these sources are potentially suitable, their usability in a particular case may be limited by the fixative or the amount of DNA or cells obtained.

Decalcified bone marrow core biopsy material cannot be used. A summary comparing these 3 MRD assessment methodologies is found in **Table 2.**

Table 2. Comparison of MRD Assessment Methods[5–7,10,15,16,20]

	Multiparameter Flow Cytometry (MFC)	Molecular methods	
		RQ-PCR*	NGS (VDJ sequencing)
Availability	Widely available	Widely available	One platform/company available for MRD analysis (ClonoSEQ®)
Sensitivity	10^{-4} to 10^{-5}	~10^{-5}	~10^{-6}
Applicability	Nearly 100%	Only in cases with well-defined translocation or mutation *(e.g., BCR-ABL1)*	FDA-cleared for use in ALL, MM and CLL
Sampling	Requires fresh sample (viable cells), analyzed within 24–48h of sampling	Both fresh and stored samples can be used	Both fresh and stored samples can be used
Diagnostic sample needed?	Yes for LAIP approach, not necessarily needed for DfN	No, but does require knowledge of the genetic aberration being tracked	Yes, requires sample with detectable disease to characterize clones for analysis
Other information provided?	Assessment of BM sample can get information about WBC subsets and distribution	No other information available	Provides information about Ig/TCR gene repertoire
Standardized?	No	For some malignancies	FDA-cleared NGS assay for MRD analysis in B-cell ALL [B-ALL], MM, and chronic lymphocytic leukemia (CLL)
Results available	Within a few hours	Usually within 1 week	Usually 2 weeks
Potential limitations	• Lack of standardization • Significant technical expertise required • Risk of immunophenotypic shift can lead to false negatives in LAIP approach	• Limited to patients with well-defined genetic aberrations	• Expensive

* Information provided here applies to standard RQ-PCR (not ASO-RQ-PCR).

DfN: "Different from normal"; **LAIP:** leukemia-associated immunophenotype

Putting MRD Assessment into Context

Because the various methods of MRD assessment differ in sensitivity, a status of "MRD negative" is not universal. Communication of MRD status should be qualified using sensitivity thresholds (e.g., "MRD less than 10"[5]), particularly as more sensitive methods of MRD quantification reach routine clinical practice.[1] The method and timing of MRD assessment are also important factors to consider.

It is generally accepted that a finding of "MRD negativity" with a detection sensitivity of 10^{-4} accurately predicts outcomes in leukemia.[5,21,22] However, when available, the most sensitive assay available (e.g. NGS-based MRD in B-cell ALL) is usually preferred in order to provide the most precise assessment and quantification of MRD.

For multiple myeloma, current methods of MRD assessment provide a sensitivity of at least 10^{-5}, and are fast approaching 10^{-6}. The clinical utility of assessing MRD to a threshold of 10^{-6} is still being investigated, although several studies have shown the utility of this approach.[41,42,43,44] The ability to detect 1 cancer cell in a million nucleated cells may allow for earlier detection of impending relapse, permitting salvage therapy before hematologic relapse is evident. It may also permit the use of PB for MRD assessment.[1,16]

While MRD assessment to inform prognosis and treatment decisions has become incorporated into practice guidelines for some malignancies, its use is still being standardized for clinical decision making in others. Evaluation of the use of MRD as a surrogate endpoint for accelerated approval of new therapies is ongoing.

It is important to note that achievement of MRD negativity is not tantamount to a cure. Patients who achieve MRD negativity even by highly sensitive MRD methods can still relapse, due to persistence of disease below the level of detection.

MRD in ALL

After standard chemotherapy treatment, adults with ALL have rates of complete remission (CR) of nearly 90%, but relapses occur commonly. Up to 50% of ALL patients who achieve CR have measurable residual leukemic cells.[7] In both pediatric and adult ALL patients (including both B-ALL and T-cell ALL [T-ALL]), across all subtypes, achieving MRD negativity at a threshold of 10^{-4} has been shown to be the single best predictor of outcomes.[7,14,23,24]

MFC is commonly used in Philadelphia chromosomenegative B-ALL and T-ALL, whereas RQ-PCR for *BCR::ABL1* is commonly used for Ph+ ALL and is superior to MFC in this subtype. NGS-based MRD can be used in any ALL subtype and may identify low levels of MRD that is not detectable with MFC or PCR for *BCR::ABL1*.

MRD assessment is now standard clinical practice in the care of patients with ALL, as reflected in the *National Comprehensive Care Network (NCCN) Clinical Practice Guidelines*, available here (please choose Treatment by Cancer Type in the Guidelines drop down menu in upper right corner). MRD assessments in ALL are carried out on bone marrow samples, and techniques used for quantitation include MFC, RQ-PCR and NGS.[6]

MRD assessment in patients with ALL is used to:

Predict Outcomes

- Achievement of MRD negativity at a threshold of 10^{-4} is a good predictor of overall survival and leukemia-free survival in patients with ALL, although even lower levels of MRD (ranging from $10^{\wedge-4}$ to $10^{\wedge-6}$) are prognostic.[14]
- MRD negativity very early in induction therapy predicts an excellent outcome in both pediatric and adult ALL.[6]
- In both children and adults, MRD positivity both pre- and post-hematopoietic stem-cell transplantation (HSCT) is predictive of posttreatment relapse.[7]

Guide Therapy

- Blinatumomab, a bispecific T-cell engager (BiTE), is approved for use in B-ALL patients in first or second remission with MRD ≥0.1% (10^{-3}).[25]
- MRD status can be used to optimize timing for HSCT and the type of conditioning regimen used.[21]

Recommendations for the timing of MRD assessment in adult ALL patients were published in a 2019 consensus document, and are shown in **Table 3**[5].

Table 3. When to Assess MRD in Adult ALL (in BM Samples)[5]

For those undergoing frontline treatment	After end of induction	In early consolidation (after ~ 3 mo of therapy)	~Every 3 mo for at least 3 y (or 5 y for patients with Ph+ ALL who do not undergo HSCT in first remission)
For those undergoing frontline treatment	Immediately prior to HSCT	~Every 3 months following HSCT	
For those with relapsed or refractory ALL receiving salvage therapy	At morphological remission	At the end of treatment	

MRD in AML

AML is a heterogeneous, complex malignancy typically diagnosed in older adults. Standard induction chemotherapy results in CR in 50%–70% of patients, but relapse rates are high.[26] The percentage of patients surviving 5 years from 2010–2016 was 28.7%.[27]

Measuring residual disease can be complicated in AML because leukemic clones are not stable over time—immunophenotypes or mutational profiles identified at diagnosis may not be those found at relapse.[28] MRD assessment is, however, critically important for prognosis. It is well established that MRD positivity after CR in a patient with AML is associated with a higher risk of relapse and shorter survival.[29,30]

MRD in AML can be assessed using MFC and PCR approaches. PCR is applicable to the approximately 40% of AML patients harboring well-defined mutations. More recently, high sensitivity NGS-based assays for *NPM1* and *FLT3*-ITD have been developed, with sensitivity of 10^-5 or better. Early data suggest that patients who achieve MRD negativity by these high-sensitivity assays can have excellent outcomes. Studies are ongoing to determine how this information can inform decisions around allogeneic stem cell transplantation.

European LeukemiaNet Recommendations

In 2017 the European LeukemiaNet (ELN) recommended a new response category for AML based on MRD status: "CR without measurable residual disease, or CR^{MRD-}."[31] This is defined as complete morphological remission accompanied by 2 successive MRD negative samples (with a sensitivity of at least 0.1%) obtained within an interval of ≥4 weeks.[30]

While MRD assessment for AML may not firmly guide treatment decisions as in the case for ALL, the ELN makes the following recommendations[29,30]:

- MRD monitoring should be considered standard of care.
- MRD should be assessed both before and after HSCT as a prognostic indicator. MRD positivity may impact monitoring decisions or prompt a recommendation to proceed to clinical trial. MRD positivity by MFC or PCR is predictive of inferior transplant outcomes.[32]
- For MFC, 500,000 to 1 million nucleated cells should be analyzed. MRD positivity by MFC is highly prognostic.
- PCR should be used for patients harboring well-known mutations like *RUNX1-RUNX1T1, CBFBMYH11, PML-RARA* and mutations in *NPM1*.
- Mutations in *NRAS, KRAS, DNMT3A, ASXL1, IDH1, IDH2, MLL-PTD,* and expression levels of *EVI1* should not be used as single markers of MRD. They may be useful when combined with a second MRD marker, however.

ELN recommendations for the timing of MRD assessment in AML are found in **Table 4**.[30]

Table 4. When to Assess MRD in AML[30]

Timing recommendations for MRD assessment in AML	At diagnosis	After 2 cycles of chemotherapy	At end of treatment (including pre-HSCT)	During follow up (only for molecular MRD), at 3 mo intervals to 24 mo

MRD assessment in AML can inform prognosis, identify impending relapse to enable early intervention and allow for robust post-HSCT surveillance.[30] In the case of patients with *FLT3*-ITD-mutated AML undergoing allogeneic HSCT, detection of MRD before or after HSCT is an indicatioj for post-HSCT maintenance with a FLT3 inhibitor. MRD assessment can also help to inform the decision of whether to recommend allogeneic stem cell transplantation in first remission, when considered along with a full assessment of patient- and disease-related factors. Given the poor outcomes associated with MRD-positive AML, enrollment of these patients into MRD-directed clinical trials is imperative.

MRD in MM

Over the past 15 years, refinement of multi-drug regimens for MM has resulted in greatly improved patient outcomes. Approximately 80% of patients achieve CR with approaches including immunomodulatory agents, protea some inhibitors and monoclonal antibodies combined with autologous HSCT, post-transplant consolidation and prolonged maintenance therapy.[15,20]

While the prognostic value of morphologic CR in MM is established, these improved outcomes have necessitated a more refined definition of CR. It has been shown that MRD negativity by MFC, NGS, magnetic resonance imaging (MRI) and positron emission tomography (PET) is highly prognostic for both progression-free survival and overall survival in MM.[33] The International Myeloma Working Group (IMWG) has defined new response categories of MRD negativity, with or without imaging-based absence of extramedullary disease.[15] MRD in MM should be assessed first in the bone marrow by MFC or NGS.[15,20]

Advanced next-generation flow (NGF) using 8-color 2-tube or 10-color 1-tube assays are widely used for MRD assessment in MM, as they have been shown to have superior sensitivity and prognostic value compared to conventional MFC.[20] NGF has a sensitivity of 10^{-5} if at least 2 million events are captured.

Because myeloma cells have Ig gene rearrangements that are stable over time, NGS can be used for MRD assessment.[34] NGS has high prognostic value in MM and shows high concordance with MFC and RQ-PCR.[20] In MM, the increased sensitivity provided by NGS (>106) appears to have clinical implications—patients achieving MRD negativity $<10^{-5}$ have better outcomes compared to those achieving MRD negative status $\geq 10^{-5}$.[35]

Because MM is a hematologic malignancy with solid tumor features, imaging can be used to detect MRD outside the BM. Residual disease detected by MRI and positron emission tomography and computed tomography (PETCT) has prognostic significance.[16,33,36]

MRD negativity in MM is the strongest prognostic indicator, and has become a factor that can overcome negative prognostic implications of high-risk genetic markers (it is better to be high risk and achieve MRD negativity than have standard risk with MRD positivity).[38]

Measuring depth of response by MRD status is recommended at each stage of treatment to inform prognosis.[37]

Whether to stop therapy in patients with sustained MRD negativity remains an unresolved question.

MRD in CML

MRD assessment in CML is made easier by the fact that CML is driven by a well-characterized genetic abnormality. Patients with CML harbor a translocation between chromosomes 9 and 22 which generates the Philadelphia (Ph) chromosome, leading to the production of the BCRABL1 oncoprotein.

Treatment with targeted tyrosine kinase inhibitors directed toward BCR-ABL1 results in hematologic and molecular remission in 80%–90% of patients. MRD monitoring is necessary to gauge response to treatment, inform prognosis, and identify patients in deep remission who might be able to safely discontinue therapy.[12]

The gold standard for MRD monitoring in CML remains the quantitation of *BCR-ABL1* transcripts by RQ-PCR.[12] An International Scale (IS) has been defined as the ratio of *BCR-ABL1* transcripts to a control transcript (e.g., *ABL1*). A "major molecular response" (MMR) is defined as a 1000-fold reduction in the *BCR-ABL1* transcript level compared to baseline.

MRD monitoring in CML can be done on PB or BM samples. NCCN Guidelines, available here, recommend the timing outlined in **Table 5**[40].

Table 5. When to Assess MRD in CML[40]

Timing recommendations for MRD assessment in CML (in PB or BM samples)				
	At diagnosis	At 3 mo intervals after initiating treatment	After MMR is achieved, at 3 mo intervals for 2 years, every 3-6 mo therafter	If there is a 1-log increase in *BCR-ABL1* transcript levels with MMR, repeat RQ-PCR in 1-3 months

MRD Laboratories

Flow cytometry is widely available in hospital hematopathology labs. A list of Clinical Laboratory Improvement Amendments (CLIA)-certified laboratories that conduct MRD testing can be found here.

Patient Education

The Leukemia & Lymphoma Society offers free educational material for your patients about MRD. It can be viewed and downloaded here.

Acknowledgements

LLS gratefully acknowledges

Nicholas J. Short, MD
Associate Professor, Department of Leukemia
The University of Texas MD Anderson Cancer Center

Rafael Fonseca, MD
Chief Innovation Officer
Getz Family Professor of Cancer
Mayo Clinic in Arizona

For their review of *Facts About Measurable Residual Disease* and their important contributions to the material presented in this publication.

We're Here to Help

LLS is the world's largest voluntary health organization dedicated to funding blood cancer research, education and patient services. LLS has regions throughout the United States and Canada. To find the region nearest to you, visit our website at www.LLS.org/ChapterFind or contact

The Leukemia & Lymphoma Society
1201 15th Street N.W., Suite 410,
Washington, D.C. 20005
Phone Number: (800) 955-4572
(M-F, 9 a.m. to 9 p.m. ET)
Website: www.LLS.org

LLS offers free information and services for patients and families touched by blood cancers as well as for healthcare professionals. The resources listed below are available to you and your patients and are meant to be a compliment to the HCP team and an additional source of support.

Consult with an Information Specialist. Information Specialists are highly trained social workers and nurses who assist through treatment, financial, and social challenges. They offer up-to-date disease and treatment information. Language services are available. For more information, please:

- Call: (800) 955-4572 (M-F, 9 a.m. to 9 p.m. ET)
- Visit: www.LLS.org/IRC
- Email or Live chat: www.LLS.org/InformationSpecialists

Clinical Trials Support Center (CTSC). Work one-on-one with an LLS clinical trial nurse navigator who will personally assist throughout the entire clinical trial process. A nurse navigator will help identify potential clinical trials and overcome the barriers to enrollment (navigators help HCPs and patients). For more information about this free service, please:

- Call an Information Specialist: (800) 955-4572 to be referred to the CTSC
- Visit: www.LLS.org/CTSC
- Complete a referral form for your patient at: www.LLS.org/CTSCreferral

Nutrition Consultations. Nutrition Education Services Center (NESC) provides one-on-one *free* nutrition education and consultations to patients and caregivers of all cancer types with registered dietitians who have expertise in oncology nutrition.

- Visit: www.LLSnutrition.org

Free Information Booklets. LLS offers free education and support publications that can either be read online or downloaded. Free print versions can be ordered. For more information, please:

- Visit: www.LLS.org/booklets

Información en Español. (LLS information in Spanish) Para mayor información por favor:

- Visite: www.LLS.org/espanol

LLS Community. LLS Community is an online social network and registry for patients, caregivers, and healthcare professionals. It is a place to ask questions, get informed, share your experience, and connect with others. To join:

- Visit: www.LLS.org/community

LLS Regions. LLS offers community support and services in the United States and Canada including the *Patti Robinson Kaufmann First Connection Program* (a peer-to-peer support program), in-person support groups, and other great resources. For more information about these programs or to contact your region, please:

- Call: (800) 955-4572
- Visit: www.LLS.org/ChapterFind

Patti Robinson Kaufmann First Connection® Program. A free peer-to-peer support program that connects patients and their loved ones to a trained peer volunteer who has gone through a similar experience.

- www.LLS.org/FirstConnection

Resources for Healthcare Professionals: Webinars, Podcasts, In-person Education Programs, Videos, and Fact Sheets:

- www.LLS.org/CE (free accreditation)
- www.LLS.org/HCPpodcast
- www.LLS.org/HCPvideos
- www.LLS.org/HCPbooklets

Resources for your Patients:

- www.LLS.org/programs
- www.LLS.org/EducationVideos
- www.LLS.org/podcast

Additional Resources

The National Cancer Institute (NCI)

www.cancer.gov
(800) 422-6237

The National Cancer Institute, part of the National Institutes of Health, is a national resource center for information and education about all forms of cancer. The NCI also provides a clinical trial search feature, the PDQ® Cancer Clinical Trials Registry, at www.cancer.gov/clinicaltrials, where healthcare professionals and patients can look for clinical trials.

References

1. Logan AC. Minimal Residual Disease in Hematologic Malignancies: Testing considerations and challenges: Overview of the latest guidelines in hematological malignancies: acute lymphoblastic leukemia. HMP|CME Oncology Learning Network. Available at: https://www.naccme.com/program/7335-2?utm_campaign=OLN_7335-2&utm_medium=email&_hsmi=96638114&_hsenc=p2ANqtz-8nnPkHN0bgHKbvdRa_rs2TAsPm-CFu0rB1iL9nFGs-VsWq9yInzGcNaHQGnIAbmOmw4d5OWkxym-MioDxwgtLzFlxheXOHg&utm_content=95233077&utm_source=hs_email

2. Acute Lymphoblastic Leukemia. NCCN Clinical Practice Guidelines in Oncology. Version 2.2020. https://www.nccn.org/guidelines/category_1

3. van Dongen JJM, van der Velden VHJ, Brüggemann M, et al. Minimal residual disease diagnostics in acute lymphoblastic leukemia: need for sensitive, fast, and standardized technologies. *Blood.* 2015;125:3996-4009. doi:10.1182/blood-2015-03-580027

4. Romano A, Palumbo GA, Parrinello NL, et al. Minimal residual disease assessment within the bone marrow of multiple myeloma: a review of caveats, clinical significance and future perspectives. *Front Oncol.* 2019;9:699. doi:10.3389/fonc.2019.00699

5. Short NJ, Jabbour E, Albitar M, et al. Recommendations for the assessment and management of measurable residual disease in adults with acute lymphoblastic leukemia: A consensus of North American experts. *Am J Hematol.* 2019;94(2):257-265. doi:10.1002/ajh.25338

6. Kim I-S. Minimal residual disease in acute lymphoblastic leukemia: technical aspects and implications for clinical interpretation. *Blood Res.* 2020;55(S1):S19-S26. doi:10.5045/br.2020.S004

7. Dalle IA, Jabbour E, Short NJ. Evaluation and management of measurable residual disease in acute lymphoblastic leukemia. *Ther Adv Hematol.* 2020;11: 1-13.

8. Kalina T, Flores-Montero J, van der Velden VHJ, et al. EuroFlow standardization of flow cytometer instrument settings and immunophenotyping protocols. *Leukemia.* 2012;26:1986-2010. doi: 10.1038/leu.2012.122

9. Theunissen P, Mejstrikova E, Sedek L, et al. Standardized flow cytometry for highly sensitive MRD measurements in B-cell acute lymphoblastic leukemia. *Blood.* 2017;129:347-357. doi:10.1182/blood-2016-07-726307

10. Flores-Montero J, Sanoja-Flores L, Paiva B, et al. Next generation flow for highlysensitive and standardized detection of minimal residual disease in multiplemyeloma. *Leukemia.* 2017;31:2094-2103. doi:10.1038/leu.2017.29

11. Jones D, Kamel-Reid S, Bahler D, et al. Laboratory practice guidelines fordetecting and reporting BCR-ABL drug resistance mutations in chronicmyelogenous leukemia and acute lymphoblastic leukemia. A report of the Association for Molecular Pathology. *J Mol Diagn.* 2009;11(1):4-11. doi:10.2353/jmoldx.2009.080095

12. Cumbo C, Anelli L, Specchia G, et al. Monitoring of minimal residual disease (MRD) in chronic myeloid leukemia (CML). *Cancer Manag Res.* 2020;12:3175-3189. doi:10.2147/CMAR.S232752

13. Branford S, Hughes T. Diagnosis and monitoring of chronic myeloid leukemia by qualitative and quantitative RT-PCR. *Methods Mol Med.* 2006;125:69-92.

14. Akabane H, Logan AC. Clinical significance and management of MRD in adults with acute lymphoblastic leukemia. *Clin Adv Hematol Oncol.* 2020;18:413-22.

15. Kumar S, Paiva B, Anderson KC, et al. International Myeloma Working Group consensus criteria for response and minimal residual disease assessment in multiple myeloma. *Lancet Oncol.* 2016;17(8):e328-e346. doi:10.1016/S1470-2045(16)30206-6

16. Kapoor P. Minimal Residual Disease in Hematologic Malignacies: Testing considerations and challenges: HMP|CME Oncology Learning Network. Available at: https://www.naccme.com/program/7335-2?utm_campaign=OLN_7335-2&utm_medium=email&_hsmi=96638114&_hsenc=p2ANqtz-8nnPkHN0bgHKbvdRa_rs2TAsPmCFu0rB1iL9nFGs-VsWq9y-InzGcNaHQGnIAbmOmw4d5OWkxymMioDxwgtLzFlxheXOHg&utm_content=95233077&utm_source=hs_email

17. Romano A, Palumbo GA, Parrinello NL. Minimal residual disease assessment within the bone marrow of multiple myeloma: a review of caveats, clinical significance and future perspectives. *Front Oncol.* 2019;9:699. doi:10.3389/fonc.2019.00699

18. ClonoSEQ Technical Information. Adaptive Biotechnologies.

19. Logan AC, Zhang B, Narasimhan B, et al. Minimal residual disease quantification using consensus primers and high-throughput IGH sequencing predicts posttransplant relapse in chronic lymphocytic leukemia. *Leukemia.* 2013;27:1659-1665. doi:10.1038/leu.2013.52

20. Mina R, Oliva S, Boccadoro M. Minimal residual disease in multiple myeloma: state of the art and future perspectives. *J Clin Med.* 2020;9:20142; doi:10.3390/jcm907214.

21. Campana D. Minimal residual disease in acute lymphoblastic leukemia. *Semin Hematol.* 2009;46:100-6. doi:10.1053/j.seminhematol.2008.09.001

22. Brüggemann M, Raff T, Florh T, et al. Clinical significance of minimal residual disease quantification in adult patients with standard-risk acute lymphoblastic leukemia. *Blood.* 2006;107:1116-23. doi:10.1182/blood-2005-07-2708

23. Berry DA, Zhou S, Higley H, et al. Association of minimal residual disease with clinical outcome in pediatric and adult acute lymphoblastic leukemia. A metaanalysis. *JAMA Oncol.* 2017;3:e170580. doi:10.1001/jamaoncol.2017.0580

24. Dix C, Lo T-H, Clark G, Abadir E. Measurable residual disease in acute myeloid leukemia using flow cytometry: a review of where we are and where we're going. *J Clin Med.* 2020;9(6):1714. doi.10.3390/jcm906714

25. Blincyto [prescribing information]. Thousand Oaks, CA. Amgen. 2020.

26. Medeiros BC, Chan SM, Daver NG, et al. Optimizing survival outcomes with post-remission therapy in acute myeloid leukemia. *Am J Hematol.* 2019;94:803-11. doi:10.1002/ajh.25484

27. Cancer Stat Facts: Leukemia–Acute Myeloid Leukemia (AML). National Cancer Institute Website. https://seer.cancer.gov/statfacts/html/amyl.html. Accessed November 12, 2020.

28. Roboz GJ. Minimal Residual Disease in Hematologic Malignanci Myeloid Leukemia. HMP|CME O http://www.naccme.com/program/7335-2?utm_campaign=OLN_7335- https://www.naccme.com/program/7335-2?utm_campaign=OLN_7335- 2&utm_medium=email&_hsmi=96638114&_hsenc=p2ANqtz-8nnPkHN0b- gHKbvdRa_rs2TAsPmCFu0rB1iL9nFGs-VsWq9yInzGcNaHQGnIAbmOmw4d5OWkxymMcontent=95233077&utm_ source=hs_email

29. Short NJ, Zhou S, Fu C, et al. Association of measurable residual disease with survival outcomes in patients with acute-myeloid leukemia: a systematic review and meta-analysis. *JAMA Oncol.* 2020;6:1890-99. doi:10.1001/jamaoncol.2020.4600

30. Schuurhuis GJ, Heuser M, Freeman S, et al. Minimal/measurable residual disease in AML: a consensus document from the European LeukemiaNet MRD Working Party. *Blood.* 2018;131:1275-91. doi:10.1182/blood-2017-09-801498

31. Döhner H, Estey E, Grimwade D, et al. Diagnosis and management of AML in adults: 2017 ELN recommendations from an international expert panel. *Blood.* 2017;129:424-47. doi:10.1182/blood-2016-08-733196

32. Buckley SA, Wood BL, Othus M, et al. Minimal residual disease prior to allogeneic hematolopoietic cell transplantation in acute myeloid leukemia: a metaanalysis. *Haematologica.* 2017;102:865-73. doi:10.3324/haematol.2016.159343

33. Chari A. Minimal Residual Disease in Hematologic Malignancies: MRD in Multiple Myeloma. HMP|CME Oncology Learning Network. Available at: https://www.naccme.com/program/7335-2?utm_campaign=OLN_7335-2&utm_medium=email&_hsmi=96638114&_hsenc=p2ANqtz-8nnPkHN0b-gHKbvdRa_

34. Avet-Loiseau, H. Minimal residual disease by next-generation sequencing: pros and cons. *Am Soc Clin Oncol Educ Book.* 2016;35:3425-30. doi: 10.1200/EDBK_159088. doi:10.1182/blood-2014-01-550020

35. Martinez-Lopez J, Lahuerta JJ, Pepin F, et al. Prognostic value of deep sequencing method for minimal residual disease detection in multiple myeloma. *Blood.* 2014;123:3073-9. doi:10.1182/blood-2014-01-5500

36. Moreau P, Attal M, Caillot D, et al. Prospective evaluation of magnetic resonance imaging and [18F] fluorodeoxyglucose positron emission tomography-computed tomography at diagnosis and before maintenance therapy in symptomatic patients with multiple myeloma included in the IFM/DFCI 2009 Trial: results of the IMAJEM study. *J Clin Oncol.* 2017;35:2911-8.

37. Mikhael J, Ismaila N, Cheung MC, et al. Treatment of multiple myeloma: ASCO and CCO joint clinical practice guideline. *J Clin Oncol.* 2019;37:1228-63.doi:10.1200/JCO.18.02096

38. Perrot A, Lauwers-Cances V, Corre J, et al. Minimal residual disease negativity using deep sequencing is a major prognostic factor in multiple myeloma. *Blood.* 2016;132:2456-64. doi:10.1182/blood-2018-06-858613

39. Hochhaus A, Baccarani M, Silver RT, et al. European LeukemiaNet 2020 recommendations for treating chronic myeloid leukemia. *Leukemia.* 2020;34:966-84.

40. Deininger, MW, Shah, NP, Altman JK, et al. Chronic Myeloid Leukemia, Version 2.2021, NCCN Clinical Practice Guidelines in Oncology. *J Natl Compr Canc Netw.* 2020;18(10):1385-1415. doi:10.6004/jnccn.2020.0047

41. Short, Nicholas J et al. "High-sensitivity next-generation sequencing MRD assessment in ALL identifies patients at very low risk of relapse." *Blood Advances* vol. 6,13 (2022): 4006-4014. doi:10.1182/bloodadvances.2022007378

42. Liang, Emily C et al. "Next-generation sequencing-based MRD in adults with ALL undergoing hematopoietic cell transplantation." *Blood Advances* vol. 7,14(2023): 3395-3402. doi:10.1182/bloodadvances.2023009856

43. Thompson, Philip A et al. "Minimal residual disease undetectable by next-generation sequencing predicts improved outcome in CLL after chemoimmunotherapy." *Blood* vol. 134,22 (2019): 1951-1959. doi:10.1182/blood.2019001077

44. Perrot, Aurore et al. "Minimal residual disease negativity using deep sequencing is a major prognostic factor in multiple myeloma." *Blood* vol. 132,23 (2018): 2456-2464. doi:10.1182/blood-2018-06-858613