In pediatric ALL, intensification of consolidation based on MRD testing is considered standard of care. In adult ALL, the NCCN Guidelines recommend MRD testing because of its clinical utility.\(^1\)

**ADDITIONAL PATIENTS CAPTURED WITH CLONOSEQ MRD DETECTION**

**clonoSEQ MRD testing identifies additional MRD+ patients who were MRD- by flow cytometry**

NGS based MRD non-inferior to flow cytometry
A study of bone marrow samples from over 550 pediatric ALL patients that evaluated MRD at pre-treatment and end of induction (Day 29) showed that sequencing-based MRD detection was non-inferior to flow cytometry.\(^2\)

**Additional 55 MRD+ patients captured**
Sequencing-based MRD detection identified an additional 55 patients who were MRD+ by the clonoSEQ Assay and MRD- by flow cytometry.\(^2\)

**Worse outcomes for patients who were MRD- by flow cytometry and MRD+ by NGS**
A study evaluating MRD in 550 pediatric ALL patients, using bone marrow from pre-treatment and end of induction, demonstrated that 55 patients who were MRD- by flow cytometry and MRD+ by the clonoSEQ Assay had a worse event free survival than those who were MRD- by the clonoSEQ Assay (\(p=0.036\)).\(^2\)

### Table 1: Comparison between the clonoSEQ Assay and Flow Cytometry (both assessed at 1/10,000) \(^2\)

<table>
<thead>
<tr>
<th>Flow Cytometry MRD Status</th>
<th>clonoSEQ MRD Status</th>
<th>Number of Patients</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Negative</td>
<td>Positive</td>
<td>55</td>
<td>0.036</td>
</tr>
<tr>
<td>Negative</td>
<td>Negative</td>
<td>409</td>
<td></td>
</tr>
<tr>
<td>Positive</td>
<td>Positive</td>
<td>87</td>
<td>0.61</td>
</tr>
<tr>
<td>Positive</td>
<td>Negative</td>
<td>17</td>
<td></td>
</tr>
</tbody>
</table>

**THE CLONOSEQ ASSAY OFFERS IMPROVED MRD ASSESSMENT**

- The clonoSEQ Assay can detect additional patients with residual disease who were initially classified to be MRD- by flow cytometry.\(^2\)
- The clonoSEQ Assay demonstrated concordance with traditional methods for measurable residual disease (MRD) detection and offers increased sensitivity.\(^2\)
- The clonoSEQ Assay has been shown to be superior to flow cytometry in predicting posttreatment relapse and survival.\(^4\)
- The clonoSEQ Assay has been shown to have prognostic value in the post-transplant setting.\(^4\)
CONCORDANCE

The clonoSEQ Assay is highly concordant with traditional MRD detection methods in ALL

In a study of more than 100 pediatric ALL patients, the clonoSEQ Assay showed quantitative concordance with both flow cytometry and allele-specific oligonucleotide PCR (ASO-PCR; Figure 2).  

Increased sensitivity
The clonoSEQ Assay was able to detect additional patients with disease present below the detection limits of flow cytometry and ASO-PCR, respectively (Figure 2, red boxes).  

![Figure 2: Comparison between sequencing and flow cytometry and ASO-PCR](image)

SUPERIOR PREDICTIVE POWER

clonoSEQ MRD testing pre-transplant predicts relapse and overall survival better than flow cytometry in pediatric ALL

Analysis of pre-transplant bone marrow samples from 40 pediatric patients with ALL found that MRD detection predicted relapse and overall survival post-allogeneic transplant significantly better than 6-color flow cytometry (Table 2).  

<table>
<thead>
<tr>
<th></th>
<th>2-Year Relapse Probability</th>
<th>2-Year Overall Survival Probability</th>
</tr>
</thead>
<tbody>
<tr>
<td>clonoSEQ MRD-Negative</td>
<td>0%</td>
<td>96%</td>
</tr>
<tr>
<td>Flow MRD-Negative</td>
<td>16%</td>
<td>77%</td>
</tr>
<tr>
<td>P-value</td>
<td>0.02</td>
<td>0.003</td>
</tr>
</tbody>
</table>

The numbers of concordance measurements are shown in the lower left and upper right. The number of discordant measurements are shown in the upper left and lower right.

Boxed numbers highlight increased sensitivity provided by sequencing-based MRD detection over other methods.
Clinical Data

**PROGNOSTIC VALUE**

*The clonoSEQ MRD test has demonstrated prognostic value post-transplant in the pediatric and adult ALL settings*

Analysis of bone marrow samples from 53 pediatric patients analyzed post-allogeneic transplant showed sequencing-based MRD detection was better at predicting relapse than 6-color flow cytometry.4

**Superior relapse probability**

One month after transplant, flow cytometry was unable to distinguish between patients who ultimately relapsed and those who did not (p=0.91; Figure 3). Sequencing-based MRD showed an estimated relapse probability of 67% in MRD-positive patients vs. 25% in MRD-negative patients (p=0.01).4

![Figure 3: Flow cytometry (MRD) vs. sequencing-based (NGS) MRD +30 days post-transplant](image1)

**Long range predictive power**

Better predictive power of post-transplant sequencing-based MRD detection vs. flow cytometry continued at day 100 and 8 months post-transplant.4

**Relapse prediction in the first 100 days**

A study of peripheral blood samples from 29 adult patients who had undergone allogeneic hematopoietic stem cell transplantation found that MRD positivity (10^-6) in the first 100 days post-transplant using sequencing based MRD detection, was highly predictive of relapse (Figure 4).5

![Figure 4: MRD positivity (≥10^-6) at any time through day +100 post-transplant predicted subsequent relapse](image2)
3 month clinical relapse lead time
Sequencing-based MRD detection in peripheral blood was shown to provide 3 month lead-time before clinical relapse (range 0-207 days), which could offer an opportunity to apply additional therapeutic maneuvers while disease burden is low (Figure 5).5

Figure 5: The estimate of time from molecular progression to clinical relapse and death for patients who relapsed. The median lead-time between sequencing-based MRD detection and clinical relapse was 89 days.

Conclusions
• The clonoSEQ Assay can detect additional patients with residual disease who were initially classified to be MRD- by flow cytometry.2

• The clonoSEQ Assay demonstrated concordance with traditional methods for measurable residual disease (MRD) detection and offers increased sensitivity.3

• The clonoSEQ Assay has been shown to be superior to flow cytometry in predicting posttreatment relapse and survival.4

• The clonoSEQ Assay has been shown to have prognostic value in the post-transplant setting.4

REFERENCES