SEL24/MEN1703 provides PIM/FLT3 Downstream Pathway Inhibition in Acute Myeloid Leukemia (AML) Blast Cells: Results of the Pharmacodynamic (PD) Assay in the Dose Escalation Part of First-in-Human DIAMOND-01 Trial

A. M. TOMIROTTI1, D. BELLAROSA1, S. SOLOMON2, A. NAZHA3, S. STRICKLAND4, R. WALTER5, F. RAVANDI6, K. BRZOZKA7, M. MAZAN7, S. BALDINI8, M. SALERNO1, M. BINASCHI9, D. LAURENT10, A. PELLACANI11

1Experimental and Translational Oncology Department, Menarini Ricerche Spa, Pomezia, Italy; 2Northside Hospital Cancer Institute, Atlanta; 3Cleveland Clinic, Cleveland; 4Vanderbilt University Medical Center, Nashville; 5Fred Hutchinson Cancer Research Center, Seattle; 6MD Anderson Cancer Center, Houston, United States; 7Ryvu Therapeutics, Krakow, Poland; 8Clinical Sciences, Menarini Ricerche Spa, Firenze; 9Preclinical and Translational Sciences, Menarini Ricerche Spa, Pomezia, Italy; 10Menarini Ricerche/BERLIN-CHEMIE AG, Berlin, Germany; 11Menarini Ricerche Spa, Firenze, Italy.
Conflict of Interest Disclosure

- A.M. Tomirotti, D. Bellarosa, M. Binaschi, M. Salerno, S. Baldini, A. Pellacani are or were Menarini Ricerche employees
- D. Laurent is Menarini Ricerche/BerlinChemie AG, Berlin, Germany employee
- K. Brzozka is Ryvu Therapeutics, Krakow, Poland employee, board member and shareholder, Ardigen, Krakow, Poland supervisory board member and shareholder, Nodthera Ltd. Cambridge, UK shareholder
- M. Mazan is Ryvu Therapeutics, Krakow, Poland employee
- For other authors there are no relationships to disclose
Background

- SEL24/MEN1703 is a first-in-class, orally available, dual PIM/FLT3 kinase inhibitor investigated in unselected AML patients in the First-in-Human, Dose Escalation (DE) and Cohort Expansion CLI24-001 (DIAMOND-01) trial\(^1\)

- The recently completed DE part showed an acceptable safety profile up to the recommended dose (RD), with initial evidence of single agent efficacy\(^2\)

- **AIM:** To assess the target engagement by evaluating phosphorylation of S6 (pS6), a downstream effector of the PIM/FLT3 pathway, and its preliminary correlation with the anti-leukemic effect of SEL24/MEN1703

---

1: Clinicaltrials.gov identifier: NCT03008187
2: Solomon *et al.*, EHA 2020
Methods

• S6 phosphorylation has been longitudinally monitored in the DIAMOND-01 study through a flow cytometric assay both on peripheral blood (PB) and bone marrow (BM) samples

• Percentage of pS6 inhibition was calculated considering the data from screening to the last day of dosing in the first treatment cycle (C1D14)

• A total of N=9 PB and N=7 BM samples, collected from patients belonging to 100 mg and 125 mg (RD) dose levels, were analyzed

• Blast counts were monitored to assess their possible correlation with target engagement
In vitro pS6 Inhibition vs Cytotoxic Activity

- In vitro studies with 26 AML cell lines bearing various genetic alterations showed a direct correlation between SEL24/MEN1703 activity and pS6 inhibition.

- pS6 inhibition levels ranges from 50% to 100% in cells showing higher growth inhibition degree ($\text{GI}_{50} < 0.5 \ \mu M$, green box).
At screening, PB and BM samples showed heterogenous levels of pS6+ blast cells (range: 1-53%), consistent with the unselected AML patient population recruited in the DIAMOND-01 trial.
% of pS6 inhibition assessed at C1D14

Following SEL24/MEN1703 treatment, blast pS6 inhibition was observed at the end of Cycle 1 in:

- 7 out of 10 PB samples (70%), range: 70-94%
- 4 out of 7 BM samples (57%), range: 26-76%

<table>
<thead>
<tr>
<th>Patient #</th>
<th>Dose level (mg)</th>
<th>PB % pS6 inhibition*</th>
<th>BM % pS6 inhibition*</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>100</td>
<td>94</td>
<td>-64</td>
</tr>
<tr>
<td>2</td>
<td>100</td>
<td>70</td>
<td>77</td>
</tr>
<tr>
<td>3</td>
<td>100</td>
<td>83</td>
<td>26</td>
</tr>
<tr>
<td>4</td>
<td>100</td>
<td>82</td>
<td>-122</td>
</tr>
<tr>
<td>5</td>
<td>125</td>
<td>78</td>
<td>76</td>
</tr>
<tr>
<td>6</td>
<td>125</td>
<td>-211</td>
<td>N/A</td>
</tr>
<tr>
<td>7</td>
<td>125</td>
<td>89</td>
<td>40</td>
</tr>
<tr>
<td>8</td>
<td>125</td>
<td>-415</td>
<td>-234</td>
</tr>
<tr>
<td>9</td>
<td>125</td>
<td>75</td>
<td>N/A</td>
</tr>
<tr>
<td>10</td>
<td>125</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td>11</td>
<td>125</td>
<td>-23</td>
<td>N/A</td>
</tr>
</tbody>
</table>

*pS6 inhibition (%) = 100 - (\frac{pS6_{test}}{pS6_{screening}}) \times 100
pS6 levels at Screening and C1D14 in PB

pS6 levels (determined as % of pS6 blast cells × MFI) were measured in PB blasts at screening and after the first cycle of dosing in patients treated at 100 and 125 mg.

MFI: Median Fluorescence Intensity
Patient 5 (125 mg cohort, >40-50% pS6+ blasts) showed direct correlation between inhibition of pS6 and blast count reduction both in PB (left) and BM (right).
Conclusions

• The longitudinal pharmacodynamic (PD) study, performed through the assessment of S6 phosphorylation status by flow cytometry, confirmed that target engagement of patients treated with SEL24/MEN1703 at 100 and 125 mg was achieved, both in PB and BM

• Preliminary data suggest that the FLT3/PIM pathway inhibition might be associated with blast count reduction, particularly in patients with high pS6 values at baseline

• Longitudinal monitoring of PD will continue in the CE part of the DIAMOND-01 trial