Consensus Virtual Screening Identified [1,2,4]Triazolo[1,5-b] isoquinolines As MELK Inhibitor Chemotypes

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Table of contents

Table S1. Screening scores and single-point inhibition results of the selected virtual hits........3
Figure S1. Core chemotypes of the 64 virtual hits .................................................................4
Figure S2. Dendrogram of the MELK structures .................................................................4
Figure S3. ROC curves of the shape and pharmacophore screening steps ......................5
Table S2. Purities and lab codes of the compounds .........................................................5
Table S3. ChEMBL IDs of the active compounds of the retrospective training set .........6
HRMS spectra .........................................................................................................................7
1H NMR spectra .....................................................................................................................28
13C NMR spectra .................................................................................................................49
Table S1. Screening scores and single-point inhibition results of the selected virtual hits at 10μM concentration. Members of the [1,2,4]triazolo[1,5-b]isoquinoline series are highlighted in bold. Other compounds are color-coded to match the respective core chemotypes in Figure S1 with at least two occurrences in this list (the rest of the compounds are singletons). Inhibition values above 40% (compounds promoted to \(IC_{50}\) determination) are highlighted in green. Score values above the respective thresholds (promoting the compounds as virtual hits) are highlighted in green.

| ID  | Inh. % | Docking score (geom. mean) | Phase score | Shape similarity score | ID  | Inh. % | Docking score (geom. mean) | Phase score | Shape similarity score |
|-----|--------|-----------------------------|-------------|------------------------|-----|--------|-----------------------------|-------------|------------------------|
| 52  | 3      | 5.992                       | 1.891       | 0.601                  | 1384| -3     | 6.315                       | 1.663       | 0.57                   |
| 97  | 21     | 6.483                       | 1.629       | 0.65                   | 1418| 8      | 5.699                       | 1.959       | 0.766                  |
| 229 | 10     | 6.683                       | 1.883       | 0.574                  | 1442| 9      | 6.467                       | 1.869       | 0.537                  |
| 235 | 3      | 6.625                       | 1.881       | 0.569                  | 1458| 22     | 6.624                       | 2.029       | 0.682                  |
| 244 | 8      | 7.052                       | 1.91        | 0.552                  | 1462| 45     | 5.521                       | 1.877       | 0.608                  |
| 246 | 45     | 6.785                       | 1.467       | 0.563                  | 1480| 20     | 6.98                        | 1.401       | 0.676                  |
| 248 | 13     | 6.978                       | 1.948       | 0.573                  | 1492| 5      | 6.486                       | 1.48        | 0.579                  |
| 304 | 13     | 6.607                       | 1.871       | 0.468                  | 1496| 9      | 7.037                       | 1.894       | 0.468                  |
| 342 | -2     | 6.534                       | 1.903       | 0.496                  | 1514| 1      | 5.754                       | 1.868       | 0.642                  |
| 496 | 4      | 6.055                       | 1.96        | 0.766                  | 1569| 35     | 6.665                       | 1.932       | 0.533                  |
| 542 | 32     | 7.003                       | 1.452       | 0.632                  | 1696| 3      | 6.211                       | 1.941       | 0.489                  |
| 572 | 12     | 6.65                        | 1.623       | 0.567                  | 1711| 15     | 6.3                          | 1.87        | 0.467                  |
| 581 | 19     | 6.178                       | 1.793       | 0.691                  | 1731| 17     | 6.727                       | 1.87        | 0.511                  |
| 595 | 25     | 7.086                       | 1.858       | 0.866                  | 1746| -4     | 6.147                       | 1.87        | 0.44                   |
| 638 | 15     | 7.357                       | 1.858       | 0.701                  | 1795| 13     | 5.837                       | 1.923       | 0.698                  |
| 639 | 5      | 6.39                        | 1.871       | 0.555                  | 1805| 12     | 5.281                       | 1.911       | 0.577                  |
| 657 | -1     | 6.399                       | 1.863       | 0.585                  | 1828| 13     | 5.745                       | 1.912       | 0.603                  |
| 669 | 16     | 6.132                       | 1.891       | 0.658                  | 1830| 6      | 5.428                       | 1.907       | 0.586                  |
| 741 | 21     | 6.997                       | 1.934       | 0.563                  | 1855| 51     | 7.039                       | 1.442       | 0.647                  |
| 742 | 7      | 5.566                       | 1.885       | 0.595                  | 1883| 9      | 6.682                       | 1.723       | 0.675                  |
| 760 | 17     | 6.117                       | 1.871       | 0.595                  | 1939| 56     | 7.185                       | 1.865       | 0.735                  |
| 761 | 5      | 6.861                       | 1.951       | 0.578                  | 2125| 16     | 7.032                       | 1.942       | 0.689                  |
| 763 | 8      | 6.759                       | 1.951       | 0.569                  | 2126| 51     | 6.91                        | 1.892       | 0.695                  |
| 779 | 54     | 7.071                       | 1.842       | 0.689                  | 2127| 48     | 6.924                       | 1.904       | 0.695                  |
| 939 | 9      | 6.299                       | 1.858       | 0.684                  | 2128| 39     | 6.988                       | 1.923       | 0.695                  |
| 980 | 14     | 6.042                       | 1.948       | 0.579                  | 2130| 20     | 7.354                       | 1.937       | 0.695                  |
| 983 | 5      | 6.196                       | 1.901       | 0.481                  | 2131| 45     | 7.312                       | 1.892       | 0.695                  |
| 1007| 17     | 4.621                       | 1.858       | 0.663                  | 2136| 35     | 6.916                       | 1.872       | 0.694                  |
| 1078| 19     | 4.561                       | 1.899       | 0.611                  | 2137| 18     | 6.939                       | 1.955       | 0.694                  |
| 1099| 20     | 5.967                       | 1.898       | 0.623                  | 2142| 46     | 7.029                       | 1.897       | 0.73                   |
| 1250| 5      | 6.555                       | 1.979       | 0.506                  | 2145| 15     | 6.933                       | 1.92        | 0.654                  |
| 1304| 12     | 5.833                       | 1.889       | 0.568                  | 2147| 33     | 6.968                       | 1.912       | 0.654                  |
**Figure S1.** Core chemotypes (Murcko scaffolds) of the 64 virtual hits with at least two occurrences in the hit-list (the rest of the virtual hits are singletons). Box colors match the color codes in Table S1.

**Figure S2.** Dendrogram of the MELK structures (23) resulting from the hierarchical clustering of their binding site residues (Euclidean distance, complete linkage).
**Figure S3.** ROC curves of the shape (A) and pharmacophore screening steps (B). The 4-point pharmacophore model selected for screening is shown as an inset of panel B, overlaid on a reference ligand (it features, from left to right, an H-bond donor, two aromatic rings and a hydrophobic group).

**Table S2.** Purities and lab codes of the compounds. Purities were determined with analytical HPLC/MS measurements on a Shimadzu LC-20AD liquid chromatograph pump system with a Shimadzu SPD-M20A photodiode array detector. A Shimadzu LCMS-2020 system was used for MS detection. Purities are reported as the percentage peak area of the compounds by UV detection at 254 nm.

| #  | ID   | Purity (%) | #  | ID   | Purity (%) |
|----|------|------------|----|------|------------|
| 1  | 779  | 98.6       | 12 | 2135 | 95.4       |
| 2  | 1939 | 98.4       | 13 | 2136 | 99.3       |
| 3  | 2125 | 98.3       | 14 | 2137 | 65.1       |
| 4  | 2126 | 87.6       | 15 | 2138 | 99.1       |
| 5  | 2127 | 99.5       | 16 | 2139 | 98.7       |
| 6  | 2128 | 98.5       | 17 | 2140 | 97.6       |
| 7  | 2130 | 99.0       | 18 | 2141 | 97.5       |
| 8  | 2131 | 99.3       | 19 | 2142 | 99.1       |
| 9  | 2132 | 99.6       | 20 | 2145 | 99.1       |
| 10 | 2133 | 92.2       | 21 | 2147 | 81.2       |
| 11 | 2134 | 93.0       |
| CHEMBL ID | CHEMBL ID | CHEMBL ID | CHEMBL ID | CHEMBL ID |
|-----------|-----------|-----------|-----------|-----------|
| CHEMBL3674298 | CHEMBL3674348 | CHEMBL3684195 | CHEMBL3688361 |   |
| CHEMBL3674302 | CHEMBL3679179 | CHEMBL3684219 | CHEMBL3688369 |   |
| CHEMBL3679182 | CHEMBL3684227 | CHEMBL3692801 |   |   |
| CHEMBL3679224 | CHEMBL3684246 | CHEMBL3692813 |   |   |
| CHEMBL3679226 | CHEMBL3684252 | CHEMBL3692837 |   |   |
| CHEMBL3679238 | CHEMBL3684257 | CHEMBL3822876 |   |   |
| CHEMBL3679269 | CHEMBL3684285 | CHEMBL3823597 |   |   |
| CHEMBL3679341 | CHEMBL3684286 | CHEMBL3823975 |   |   |
| CHEMBL3684156 | CHEMBL3688345 | CHEMBL4083922 |   |   |
| CHEMBL3684159 | CHEMBL3688359 | CHEMBL4088246 |   |   |
| Formula     | Calculated mass | Error / mDa | Error / ppm | DBE |
|------------|-----------------|-------------|-------------|-----|
| C17 H14 N3 | 260.1187        | -0.5725     | -2.2012     | 12.5 |
| Formula | Calculated mass | Error / mDa | Error / ppm | DBE |
|---------|----------------|-------------|-------------|-----|
| C19 H18 N3 | 288.15 | -0.2727 | -0.9465 | 12.5 |
Formula | Calculated mass | Error / mDa | Error / ppm | DBE
--- | --- | --- | --- | ---
C17H13N3Cl | 294.0798 | -0.7002 | -2.3812 | 12.5
| Formula       | Calculated mass | Error / mDa | Error / ppm | DBE |
|---------------|-----------------|-------------|-------------|-----|
| C₁₈ H₁₆ N₃ O | 290.1293        | -0.7373     | -2.5413     | 12.5|
| Formula  | Calculated mass | Error / mDa | Error / ppm | DBE  |
|----------|----------------|-------------|-------------|------|
| C18 H16 N3 O | 290.1293 | -0.3373 | -1.1626 | 12.5 |
| Formula   | Calculated mass | Error / mDa | Error / ppm | DBE |
|-----------|-----------------|-------------|-------------|-----|
| C18 H14 N3 O2 | 304.1086        | -0.1018     | -0.3349     | 13.5 |
| Formula       | Calculated mass | Error / mDa | Error / ppm | DBE |
|--------------|-----------------|-------------|-------------|-----|
| C18 H13 N3 F3 | 328.1061        | -0.5572     | -1.6985     | 12.5|
| Formula   | Calculated mass | Error / mDa | Error / ppm | DBE |
|-----------|-----------------|-------------|-------------|-----|
| C18 H13 N3 F3 | 328.1061        | -0.5572     | -1.6985     | 12.5 |
**Formula** | **Calculated mass** | **Error / mDa** | **Error / ppm** | **DBE**
--- | --- | --- | --- | ---
C16 H13 N4 | 261.114 | -0.4215 | -1.6144 | 12.5
| Formula       | Calculated mass | Error / mDa | Error / ppm | DBE |
|--------------|----------------|-------------|-------------|-----|
| C16 H12 N4 Cl | 295.075        | -0.5492     | -1.8613     | 12.5|
| Formula         | Calculated mass | Error / mDa | Error / ppm | DBE |
|-----------------|-----------------|-------------|-------------|-----|
| C16 H13 N4      | 261.114         | -0.1215     | -0.4655     | 12.5|
| C16 H12 N4 Na   | 283.0959        | 0.0337      | 0.1193      | 12.5|
| Formula | Calculated mass | Error / mDa | Error / ppm | DBE |
|---------|----------------|-------------|-------------|-----|
| C16 H13 N4 | 261.114 | -0.3215 | -1.2314 | 12.5 |
| Formula    | Calculated mass | Error / mDa | Error / ppm | DBE |
|------------|----------------|-------------|-------------|-----|
| C17 H12 N3 Cl2 | 328.0408       | 0.072       | 0.2196      | 12.5 |
**Formula**

| Formula    | Calculated mass | Error / mDa | Error / ppm | DBE |
|------------|-----------------|-------------|-------------|-----|
| C17 H12 N3 Cl2 | 328.0408        | 0.072       | 0.2196      | 12.5 |
| Formula    | Calculated mass | Error / mDa | Error / ppm | DBE |
|------------|-----------------|-------------|-------------|-----|
| C18 H15 N3 CI | 308.0954        | -0.3503     | -1.1371     | 12.5 |
| Formula    | Calculated mass | Error / mDa | Error / ppm | DBE |
|------------|-----------------|-------------|-------------|-----|
| C18 H15 N3 Cl | 308.0954        | -0.1503     | -0.488      | 12.5 |
Formula | Calculated mass | Error / mDa | Error / ppm | DBE
---|---|---|---|---
C18 H15 N3 Cl | 308.0954 | 0.2496 | 0.8102 | 12.5
| Formula   | Calculated mass | Error / mDa  | Error / ppm | DBE |
|-----------|-----------------|--------------|-------------|-----|
| C19 H18 N3 O | 304.1449        | -0.3873      | -1.2736     | 12.5 |
| Formula     | Calculated mass | Error / mDa | Error / ppm | DBE |
|-------------|-----------------|--------------|-------------|-----|
| C19 H18 N3 O | 304.1449        | 0.6126       | 2.0142      | 12.5 |
| Formula      | Calculated mass | Error / mDa | Error / ppm | DBE |
|--------------|-----------------|-------------|-------------|-----|
| C16 H11 N3 Cl | 280.0641        | -0.1501     | -0.5362     | 12.5 |
| Formula | Calculated mass | Error / mDa | Error / ppm | DBE |
|---------|----------------|-------------|-------------|-----|
| C17 H14 N3 | 260.1187 | -0.2725 | -1.0479 | 12.5 |
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