# Supporting Information

# Dynamic Origins of Differential RNA Binding Function in Two dsRBDs from the miRNA "Microprocessor" Complex

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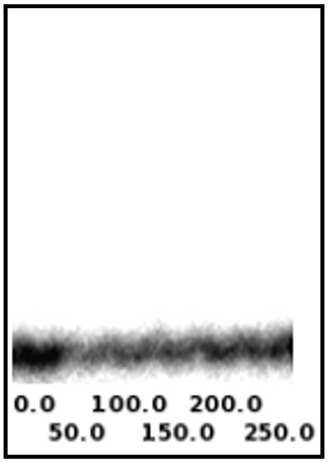

**Figure S1:** EMSA gel of Drosha-dsRBD that demonstrates Drosha-dsRBD does not bind pri-miR-16-1. Protein concentration (μM) is shown below the lanes.

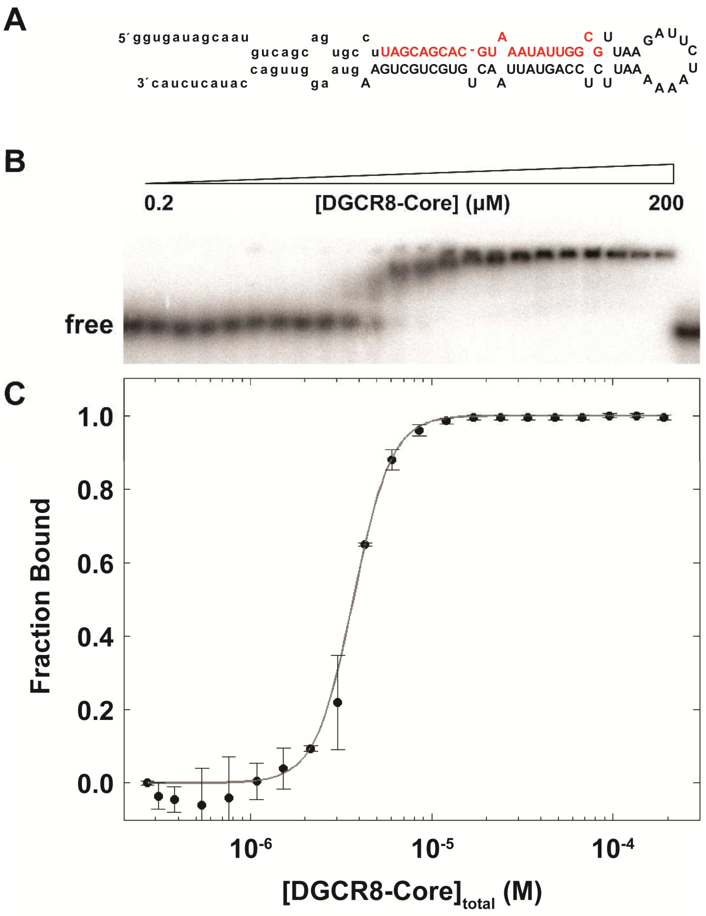

**Figure S2:** EMSA assay of pri-miR-16-1 binding to DGCR8-Core. (A) Predicted secondary structure of pri-miR-16-1 with the sequence of the mature miRNA shown in red and the region removed by Drosha cleavage indicated through lower-case letters. (B) Representative gel showing addition of 2-200 µM DGCR8-Core to 0.25 nM pri-miR-16-1. (C) Fitted EMSA fraction bound as a function of DGCR8-Core concentration with data points and uncertainties represented by filled circles and the best fit to the data shown in grey.

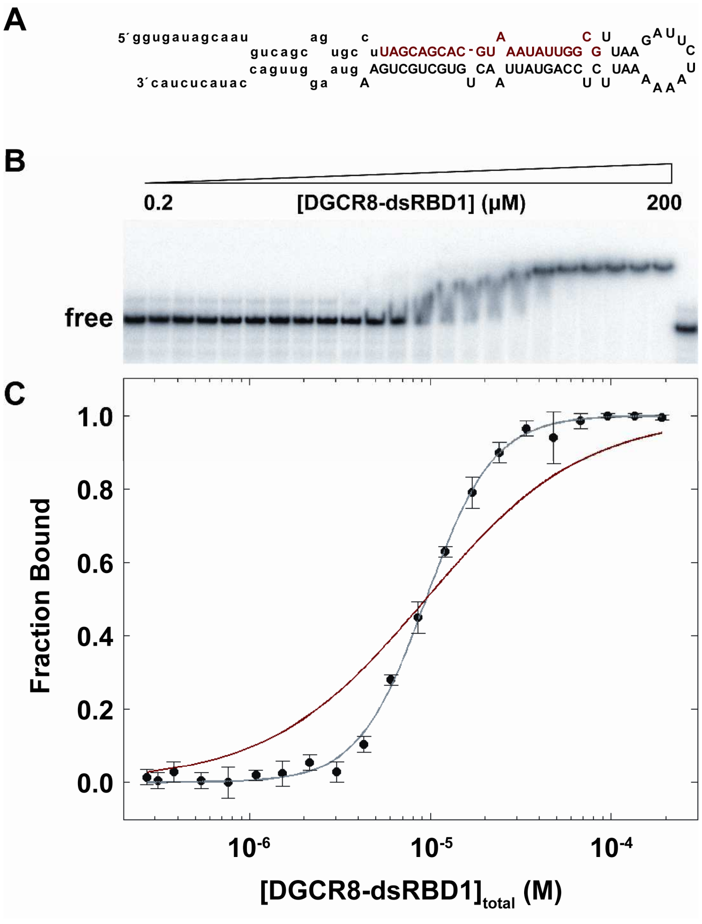

**Figure S3:** EMSA assay of pri-miR-16-1 binding to DGCR8-dsRBD1. (A) Predicted secondary structure of pri-miR-16-1 with the sequence of the mature miRNA shown in red and the region removed by Drosha cleavage indicated through lower-case letters. (B) Representative gel showing addition of 2-200 μM DGCR8-dsRBD1 to 0.25 nM pri-miR-16-1. (C) Fitted EMSA fraction bound as a function of DGCR8-dsRBD1 concentration with data points and uncertainties represented by filled circles. The N-independent and identical sites best fit to the data is shown in red and the fully cooperative best fit to the data is shown in grey.

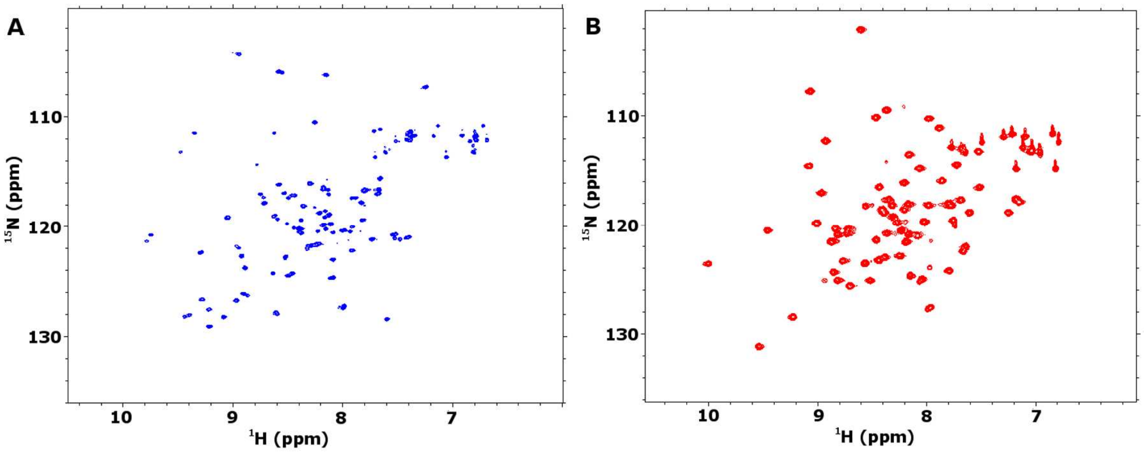

**Figure S4:** The representative $^{1}$H,$^{15}$N-HSQC spectra of (A) Drosha-dsRBD and (B) DGCR8-dsRBD1.

**Table S1:** Chemical shifts (ppm) from the backbone assignment of Drosha-dsRBD.

| Residue | N | H | CO | CA | CB |
|---|---|---|---|---|---|
| G1258 | - | - | 173.7 | 45.27 | - |
| N1259 | 119.4 | 7.82 | 176.2 | 54.98 | 34.04 |
| D1260 | 122.8 | 8.51 | 174.8 | 54.61 | 29.14 |
| P1261 | - | - | 178.0 | 65.41 | 32.80 |
| K1262 | 117.0 | 7.68 | 180.6 | 60.41 | 31.86 |
| S1263 | 119.0 | 8.12 | 176.8 | 61.38 | 62.25 |
| Q1264 | 121.9 | 8.32 | 178.5 | 58.76 | 28.87 |
| L1265 | 120.3 | 8.44 | 176.9 | 58.27 | 41.06 |
| Q1266 | 116.7 | 7.67 | 177.8 | 58.42 | 28.19 |
| Q1267 | 116.8 | 8.13 | 179.0 | 58.72 | 27.97 |
| C1268 | 199.3 | 8.16 | 177.5 | 58.37 | 39.24 |
| C1269 | 119.9 | 8.50 | 177.6 | 62.02 | 33.332 |
| L1270 | 120.2 | 7.92 | 179.0 | 57.56 | 41.62 |
| T1271 | 111.4 | 7.71 | 175.4 | 64.23 | 69.66 |
| L1272 | 121.1 | 7.40 | 176.9 | 54.82 | 41.61 |
| R1273 | 120.8 | 7.52 | 176.0 | 56.38 | 30.65 |
| T1274 | 119.1 | 8.61 | 174.2 | 61.44 | 70.29 |
| E1275 | 124.3 | 8.63 | 177.1 | 57.60 | 29.52 |
| G1276 | 111.5 | 8.62 | 173.7 | 45.17 | - |
| K1277 | 118.3 | 8.26 | 174.4 | 52.77 | 39.02 |
| E1278 | 119.9 | 8.17 | 174.1 | 52.05 | 40.88 |
| P1279 | - | - | 176.2 | 62.90 | 32.04 |
| D1280 | 121.7 | 8.28 | 174.6 | 54.79 | 43.25 |
| I1281 | 116.5 | 8.17 | 174.0 | 58.06 | 40.14 |
| P1282 | - | - | 174.8 | 63.17 | 32.05 |
| L1283 | 124.6 | 8.49 | 174.8 | 53.70 | 45.13 |
| Y1284 | 123.8 | 8.88 | 176.2 | 57.78 | 39.77 |
| K1285 | 122.4 | 9.29 | 175.0 | 55.08 | 36.87 |
| T1286 | 122.7 | 8.91 | 174.8 | 63.21 | 68.08 |
| L1287 | 129.1 | 9.22 | 177.2 | 55.44 | 43.28 |
| Q1288 | 115.7 | 7.66 | 173.9 | 55.49 | 32.05 |
| T1289 | 119.5 | 8.38 | 173.3 | 61.87 | 70.65 |
| V1290 | 121.6 | 8.22 | 174.8 | 60.29 | 35.49 |
| G1291 | 110.6 | 8.25 | 172.1 | 43.76 | - |
| P1292 | - | - | 177.7 | 62.12 | 32.37 |

| | | | | | |
|---|---|---|---|---|---|
| S1293 | 114.4 | 8.78 | 177.7 | 62.06 | 62.84 |
| H1294 | - | - | 174.8 | 56.04 | 30.36 |
| A1295 | 128.5 | 7.59 | 174.3 | 51.61 | 18.03 |
| R1296 | 123.0 | 8.08 | 177.0 | 55.34 | 32.15 |
| T1297 | 116.2 | 8.57 | 173.5 | 62.39 | 70.65 |
| Y1298 | 127.6 | 9.22 | 174.9 | 57.32 | 40.67 |
| T1299 | 117.1 | 8.75 | 173.9 | 61.78 | 70.15 |
| V1300 | 128.2 | 9.40 | 172.7 | 59.98 | 35.94 |
| A1301 | 126.3 | 8.87 | 174.5 | 49.56 | 23.77 |
| V1302 | 118.7 | 8.16 | 173.0 | 57.62 | 34.53 |
| Y1303 | 126.7 | 9.28 | 175.2 | 56.32 | 42.90 |
| F1304 | 119.3 | 9.04 | 175.1 | 56.66 | 42.83 |
| K1305 | 128.3 | 9.08 | 176.3 | 57.58 | 29.49 |
| G1306 | 104.3 | 8.94 | 173.6 | 45.16 | - |
| E1307 | 121.2 | 7.73 | 175.2 | 54.28 | 31.76 |
| R1308 | 126.7 | 8.96 | 176.7 | 56.95 | 30.06 |
| I1309 | 121.9 | 8.95 | 174.8 | 61.37 | 39.81 |
| G1310 | 107.4 | 7.24 | 172.2 | 45.43 | - |
| C1311 | 120.8 | 9.75 | 172.3 | 57.04 | 36.41 |
| G1312 | 111.5 | 9.35 | 170.5 | 44.58 | - |
| K1313 | 117.1 | 8.52 | 176.7 | 53.96 | 37.15 |
| G1314 | 106.0 | 8.58 | 171.0 | 45.99 | - |
| P1315 | - | - | 174.7 | 63.70 | 31.81 |
| S1316 | 111.5 | 7.38 | 173.7 | 56.11 | 66.6 |
| I1317 | 124.4 | 8.45 | 177.4 | 65.23 | 37.08 |
| Q1318 | 118.1 | 8.36 | 178.8 | 59.57 | 27.88 |
| Q1319 | 116.8 | 7.81 | 178.8 | 58.92 | 29.18 |
| A1320 | 124.7 | 8.09 | 179.0 | 55.86 | 18.44 |
| E1321 | 117.9 | 8.72 | 178.7 | 59.66 | 29.83 |
| M1322 | 116.8 | 7.80 | 178.6 | 58.94 | 32.01 |
| G1323 | 106.2 | 8.15 | 176.3 | 47.27 | - |
| A1324 | 127.9 | 8.60 | 178.5 | 55.08 | 17.64 |
| A1325 | 120.3 | 8.39 | 179.5 | 55.45 | 17.45 |
| M1326 | 116.1 | 8.30 | 178.3 | 59.09 | 32.96 |
| D1327 | 120.4 | 7.99 | 177.7 | 57.78 | 43.24 |
| A1328 | 119.8 | 8.10 | 180.0 | 54.81 | 19.07 |
| L1329 | 117.1 | 8.46 | 179.3 | 58.15 | 42.25 |
| E1330 | 117.7 | 7.83 | 178.7 | 58.72 | 30.10 |
| K1331 | 117.5 | 7.88 | 177.6 | 57.74 | 33.88 |
| Y1332 | 120.5 | 8.22 | 174.8 | 59.57 | 38.38 |
| N1333 | 121.1 | 7.56 | 173.7 | 52.48 | 38.09 |
| F1334 | 122.2 | 7.92 | 173.8 | 56.92 | 38.70 |
| P1335 | - | - | 176.4 | 63.10 | 29.12 |
| Q1336 | 120.6 | 8.39 | 175.2 | 55.80 | 29.39 |
| M1337 | 127.3 | 7.99 | - | 56.94 | 33.76 |

**Table S2:** Chemical shifts (ppm) from the backbone assignment of DGCR8-dsRBD1.

| Residue | N | H | CO | CA | CB |
|---|---|---|---|---|---|
| G504 | - | - | 173.7 | 45.12 | - |
| I505 | 119.5 | 7.95 | 175.7 | 60.83 | 38.84 |
| P507 | - | - | 175.7 | 63.5 | 31.83 |
| N508 | 123.7 | 7.89 | 177.8 | 54.76 | 40.74 |
| G509 | 114.0 | 8.29 | 174.2 | 45.55 | - |
| K510 | 120.8 | 8.01 | 176.1 | 56.08 | 34.02 |
| S511 | 118.0 | 8.47 | 174.8 | 57.54 | 65.01 |
| E512 | 123.3 | 9.993 | 178.4 | 62.03 | 28.31 |
| V513 | 118.0 | 7.70 | 178.7 | 66.23 | 32.58 |
| C514 | 122.1 | 7.59 | 177.4 | 63.01 | 27.05 |
| I515 | 121.2 | 8.39 | 179.1 | 65.15 | 38.15 |
| L516 | 120.2 | 8.64 | 177.6 | 58.45 | 40.80 |
| H517 | 119.6 | 8.19 | 177.8 | 60.92 | 31.10 |
| E518 | 117.9 | 8.25 | 178.8 | 59.70 | 29.30 |
| Y519 | 120.5 | 8.29 | 177.6 | 62.10 | 38.86 |

| | | | | | |
|---|---|---|---|---|---|
| M520 | 116.8 | 8.89 | 178.7 | 59.57 | 31.21 |
| Q521 | 117.9 | 8.09 | 177.3 | 58.76 | 28.79 |
| R522 | 117.5 | 7.61 | 177.3 | 58.67 | 31.03 |
| V523 | 115.9 | 8.13 | 176.7 | 64.74 | 32.69 |
| L524 | 117.7 | 7.07 | 176.1 | 55.58 | 43.50 |
| K525 | 114.6 | 7.99 | 175.4 | 56.62 | 28.91 |
| V526 | 113.0 | 6.96 | 173.8 | 59.72 | 35.62 |
| R527 | 121.3 | 8.11 | 174.3 | 52.83 | 30.88 |
| P528 | - | - | 175.3 | 62.41 | 32.41 |
| V529 | 122.7 | 8.17 | 174.9 | 61.87 | 33.56 |
| Y530 | 125.4 | 8.63 | 174.9 | 57.59 | 39.97 |
| N531 | 121.3 | 8.80 | 173.4 | 51.68 | 41.04 |
| F532 | 120.2 | 8.76 | 175.4 | 57.48 | 41.51 |
| F533 | 118.7 | 8.31 | 173.3 | 55.73 | 40.90 |
| E534 | 120.7 | 8.74 | 175.7 | 55.62 | 31.21 |
| C535 | 123.0 | 8.36 | 174.1 | 56.69 | 29.56 |
| E536 | 124.8 | 8.86 | 175.6 | 57.01 | 29.17 |
| N537 | 121.8 | 7.56 | 174.6 | 51.01 | 39.07 |
| P538 | - | - | 178.3 | 64.44 | 31.86 |
| S539 | 110.9 | 7.80 | 174.7 | 59.81 | 63.63 |
| E540 | 118.6 | 7.17 | 171.1 | 53.48 | 30.56 |
| P541 | - | - | 177.4 | 64.07 | 32.34 |
| F542 | 117.8 | 7.72 | 173.5 | 57.95 | 40.19 |
| G543 | 109.2 | 8.29 | 173.0 | 42.48 | - |
| A544 | 123.1 | 8.69 | 174.7 | 50.56 | 22.73 |
| S545 | 114.3 | 9.00 | 174.3 | 56.84 | 66.69 |
| V546 | 122.7 | 8.31 | 175.5 | 61.12 | 33.27 |
| T547 | 119.6 | 8.93 | 173.7 | 59.55 | 71.52 |
| I548 | 121.3 | 8.79 | 176.7 | 61.05 | 39.37 |
| D549 | 130.9 | 9.46 | 175.6 | 55.31 | 40.03 |
| G550 | 101.8 | 8.53 | 173.7 | 45.23 | - |
| V551 | 124.0 | 7.71 | 175.6 | 61.80 | 33.68 |
| T552 | 124.8 | 8.74 | 174.5 | 64.39 | 69.01 |
| Y553 | 128.2 | 9.15 | 174.6 | 57.60 | 39.19 |
| G554 | 110.1 | 7.90 | 171.2 | 45.07 | - |
| S555 | 113.3 | 8.08 | 172.8 | 56.84 | 66.84 |
| G556 | 107.5 | 8.99 | 170.6 | 45.59 | - |
| T557 | 110.0 | 8.38 | 173.7 | 58.84 | 71.55 |
| A558 | 120.3 | 9.38 | 175.9 | 51.55 | 23.98 |
| S559 | 112.0 | 8.85 | 173.4 | 59.79 | 63.61 |
| S560 | 113.0 | 7.45 | - | 56.15 | 66.72 |
| K561 | - | - | 177.8 | 60.53 | 32.24 |
| K562 | 118.4 | 8.13 | 179.2 | 59.70 | 32.38 |
| L563 | 118.6 | 7.53 | 179.4 | 57.33 | 42.14 |
| A564 | 124.4 | 8.08 | 178.9 | 56.12 | 17.15 |
| K565 | 117.5 | 8.27 | 178.0 | 60.47 | 32.62 |
| N566 | 116.3 | 7.44 | 177.6 | 57.04 | 38.80 |
| K567 | 120.3 | 8.16 | 179.1 | 60.14 | 33.15 |
| A568 | 123.3 | 8.49 | 180.9 | 55.29 | 18.12 |
| A569 | 124.1 | 8.78 | 179.2 | 55.69 | 17.88 |
| R570 | 120.5 | 8.65 | 177.5 | 59.89 | 29.62 |
| A571 | 119.0 | 8.24 | 180.4 | 54.56 | 18.64 |
| T572 | 115.7 | 7.78 | 175.7 | 68.16 | 67.98 |
| L573 | 124.8 | 8.44 | 177.1 | 58.72 | 41.17 |
| E574 | 114.2 | 7.65 | 177.9 | 58.74 | 29.66 |
| I575 | 117.4 | 7.10 | 177.7 | 63.93 | 39.63 |
| L576 | 118.0 | 7.91 | 177.5 | 57.22 | 44.06 |
| I577 | 118.4 | 8.33 | 173.1 | 58.50 | 38.51 |
| P578 | - | - | 177.7 | 64.46 | 31.70 |
| D579 | 116.3 | 8.36 | 176.2 | 54.43 | 39.89 |
| F580 | 120.6 | 8.09 | 176.0 | 60.16 | 39.54 |
| V581 | 119.4 | 7.67 | 175.2 | 62.13 | 32.52 |
| K582 | 124.8 | 7.97 | 175.7 | 56.30 | 32.81 |
| Q583 | 127.3 | 7.89 | 180.5 | 57.29 | 30.10 |