#### Supporting Material S1: Additional Method Details

#### **1** Sequence processing

The SOLiD reads were obtained in dinucleotide color space [1]. Adapter removal and mapping were done in color space as well. The first color of all reads, which encodes the transition from the last primer base to the first sequenced nucleotide, was removed from all reads since the mapping software cannot use it. Since SOLiD reads (35 nt) were longer than the expected length of mature miRNAs (20–23 nt), miRNA reads contained part of the adapter sequence. The start position of the adapter sequence within the read was located using our own software, which implements a free-end-gap (semiglobal) sequence alignment, allowing at most 12 % errors over the length of the matching part of the adapter. Reads were truncated at the color preceding the adapter sequence since that color encodes the transition from the actual sequence into the adapter and therefore cannot be used during mapping. The procedure is illustrated below. Colors are represented by numbers between 0 and 3.

adapter sequence: 330201030313112312 original read: T30002321001012222223330201030313112 trimmed read: 000232100101222222

The distribution of trimmed read lengths in nucleotide space is plotted in Fig. 1A (main text), and detailed statistics per patient are shown in S2. Computation of full alignments requires quadratic time, but since the sequences were short, adapter removal took less than one minute per million reads.

#### 2 Mapping processed reads

Only processed reads of at least 12 colors (13 nt) were mapped since shorter reads are unlikely to be uniquely mappable to the human genome. We allowed two errors for read lengths 12–14 and three errors (the maximum possible with MAQ) for longer reads. MAQ only detects ungapped alignments (no insertions or deletions), and maps non-unique matches randomly to one of the matching locations. For reproducibility, the seed of the random number generator was set to a constant value for all calls to the program. This random assignment of reads, however, does not affect miRNA read counts as non-unique reads are discarded for expression statistics (see next paragraph for details). Mature miRNA records from miRBase with identical sequences were merged into single entries prior to mapping. For short reference sequences, a number of 'N' characters (which do not match any nucleotide) were added before and after the sequence to work around a bug in MAQ that prevents it from reporting negative coordinates for read start locations. Since MAQ can only work with files containing reads all of the same length, files were split appropriately, then further split into jobs of approximately one million reads each. The results of the finished mapping jobs were merged and the "mag csmap2nt" command was used to convert from color space to nucleotide space. This command decodes a color space read of length n (which contains information about n+1 nucleotides) to the most likely nucleotide sequence, guided by the reference sequence to which the read was mapped. The command retains only those nucleotides for which two colors are available, that is, the resulting read has a length of n-1.

**Obtaining microRNA read counts.** After mapping all reads against mature miRNA sequences, mapped reads are postprocessed in the following way. First, our software readds the flanking nucleotides lost during "csmap2nt". Since the additional nucleotides are not copied from the reference sequence, but computed based on the color read and

converted nucleotide read, this may introduce additional mismatches. The number of errors is therefore recomputed.

Next, a read is discarded if it was mapped non-uniquely, or if it is is shorter than 15 nt, or if it contains more than two nucleotide mismatches, or if its reverse complement was mapped instead of the forward strand. Aligned reads usually cover the entire miRNA, but reads starting at an offset greater than two within the reference are also discarded.

Each read that is mapped to a specific mature miRNA and that was not discarded increases the expression count for that miRNA by one.

The confidence in a correct assignment of reads to mature miRNAs was increased by discarding reads not uniquely mappable and by the error-correcting properties of color space. Color space encoded mapping bears the advantage of removing technical sequencing errors during conversion from color space to nucleotide space, since two colors need to be changed if one nucleotide is altered.

Detailed mapping statistics, also for mapping against other references, are shown in S3.

**Estimating the sequencing error rate.** Among the reads uniquely mapped to mature miRNAs, 6.3% of all positions in SOLiD color space did not match the reference sequence. This fraction includes sequencing errors as well as SNPs and post-transcriptional editing events. To estimate the technical sequencing error, we used the Phred quality values supplied by the SOLiD device. Let q be a Phred quality score of a color. The error probability at that position in color space is then given by  $p = 10^{-q/10}$ . We computed the average error probability for each position in mature miRNAs; it varied between 0.6% near the 5' end and 3.8% near the 3' end (shown in Fig. 3D, blue line and crosses for positions -15 to -1, where -1 corresponds to the 3' end). The error probability in nucleotide space is lower because of the error correcting capabilities of color space. Positions -15 to -9 (Fig. 3D) give an indication: Even though the color space error rate is estimated at approximately 2.0%, almost no nucleotide differences are observed. We may therefore assume that most observed differences are due to SNPs and/or editing.

**Handling terminal additions.** Even when 'N' characters are appended at the end of the miRNA reference sequences, as described above, the MAQ software does not map reads that overlap the reference at the right end. This is a useful property as it prevents to (parts of) pre-miRNA sequences to be wrongly counted as mature miRNAs. To analyze terminal additions, however, we had to work around this behavior. We remapped all reads against modified reference sequences where the 'N' characters at the right side were replaced by 'T' characters. In a postprocessing steps, the 'T' characters were handled like 'N' characters and the number of mismatches was recomputed.

#### 3 Normalization of read counts

Quantile normalization, which is appropriate for some microarray platforms, was considered to be inappropriate for these data as there is no evidence to assume equal count distributions. We normalized raw read counts using pure scaling transformations to remove a potential bias in miRNA expression across the datasets. The scaling factors were obtained from qq-plots (S4A) by computing the median of differences of corresponding quantile values of a dataset and the reference dataset. This qq-scale transformation was shown to be superior to scaling to a given constant (such as 1,000,000 raw counts), as the latter was incapable of managing outliers (S4B). Statistical analysis was performed using the open-source software package R, version 2.9.1.

## 4 Unbiased analysis of mature miRNAs discriminating favorable from unfavorable NB

Fig. 3D shows that the p-value distribution was not uniform, and that low p-values were more common than expected by chance. The interpretation of this figure can be aided by applying multiple testing correction procedures. Using the approach proposed by Benjamini and Hochberg [2, 3] and setting the false discovery rate to  $\leq 0.33$  resulted in 92 differentially expressed miRNAs. Only one miRNA, miR-181a-2\*, significantly (p = 0.027) discriminated between the two groups when the more stringent Bonferroni correction was applied.

#### 5 Prediction of putative new miRNAs

For performance reasons, the script excise\_candidate.pl from the miRDeep software package was reimplemented in Python with different data structures, avoiding quadratic time behavior. As required by miRDeep, the RNA secondary structure prediction tool RNAfold from the Vienna package (version 1.8.2) was run on the excised candidate regions. Subsequently, auto\_blast.pl from the miRDeep package was run with parameter "-b", and then the actual prediction procedure in the mirdeep.pl script was called (parameters: -v '-50'). Score histograms (see S8) were consistent with the published reference histograms [4]. A clustering of the set of all predicted mature miRNA sequences and all mature miRNA sequences from miRBase was done using blastclust (BLAST package version 2.2.20) with parameters "-p F -L 0.8 -b F -S 90 -W 11". A search in miRBase using blastall was performed for all sequences. All clusters with at least one sequence having an E-value better than 0.1 were discarded.

#### 6 Cluster analysis

Heatmaps and cluster dendrograms were produced using the "heatmap" function with "Canberra" distances and complete linkage clustering in R. The Canberra distance between two vectors x, y of length n is defined as  $d(x, y) := \sum_{i=1}^{n} |x_i - y_i|/(|x_i| + |y_i|)$ . It relates differences to absolute values, which is required when values on different orders of magnitude are compared. Therefore Canberra distance allows for data analysis without prior normalisation steps.

#### References

- [1] A theoretical understanding of 2 base color codes and its application to annotation, error detection, and error correction. Applied Biosystems, White Paper.
- [2] Benjamini Y, Hochberg Y (1995) Controlling the false discovery rate: A practical and powerful approach to multiple testing. *Journal of the Royal Statistical Society Series* B 57:289–300.
- [3] Storey JD (2002) A direct approach to false discovery rates. Journal of the Royal Statistical Society Series B 64:479–498.
- [4] Friedländer MR, et al. (2008) Discovering microRNAs from deep sequencing data using miRDeep. Nat Biotechnol 26:407–415.

Read length distribution for each patient. The top row identifies the dataset. The leftmost column identifies the read length in color space (0 to 34; add 1 to obtain read length in nucleotide space). Numbers in the table specify the number of reads of each length in each dataset after adapter removal.

Length	552	553	554	555	556	557	558	559	560	561
0	5474	7791	13491	7190	229247	19794	7940	10481	13947	39344
1	393	485	791	558	16097	1476	605	699	908	2440
2	262	277	416	308	9689	846	341	362	556	1367
3	384	525	739	605	17649	1666	574	724	942	2509
4	1192	1627	2403	1743	46862	4096	1563	2074	2541	6953
5	7480	8983	12460	10637	295850	28115	9261	12597	16119	44419
6	2205	4006	2771	1552	50182	6108	5253	3895	4418	10809
7	4213	7575	5742	2505	81149	8356	16261	8068	7479	17274
8	12270	21867	18098	7109	212224	22269	48830	21574	20195	47816
9	35993	64667	52105	14657	490924	42236	114633	49249	53119	127217
10	177213	295879	174718	39628	2008522	138061	309643	168661	255503	613781
11	494129	752817	408924	86542	4734437	299117	578971	364413	575791	1506585
12	214703	291384	159972	33981	1123327	124038	299007	109713	149572	374278
13	87523	100680	78191	22617	588743	81056	238417	98171	63768	138494
14	36984	55477	54539	20132	476489	57483	224205	91457	53937	103618
15	37347	57269	68456	24278	603527	69688	225922	102721	62273	125571
16	114697	175640	553997	207280	10922227	614804	466764	457986	588076	1894714
17	58860	116462	125786	46418	711820	133210	271538	113902	129314	192641
18	68324	103208	130765	46202	870524	94322	277835	134729	93796	218636
19	107727	148192	227457	79999	1326241	138336	326782	172220	138281	367496
20	426693	389837	741518	305646	7174400	598103	590973	531172	617455	1848766
21	225652	507296	571654	262880	1701694	280493	527823	423713	329748	715240
22	231328	530976	559356	250886	1029122	252631	608351	482809	363142	525440
23	89648	208148	186171	82564	537195	95644	338039	212414	99940	198849
24	46663	85995	62529	37918	381935	73508	257461	122250	67676	92904
25	11550	25705	17358	5392	88278	25331	106904	50734	33331	18583
26	31913	101039	35752	27242	132727	257342	164825	122817	227887	61003
27	92283	150105	58254	157932	379835	237041	250231	164335	232052	165102
28	58381	92912	44765	95979	314573	159682	205203	104370	181337	99513
29	32573	48055	32465	42644	255498	86452	191390	81934	152809	79084
30	29023	23289	23696	60536	217371	72898	121431	74922	97763	65927
31	15225	11279	11394	70790	376825	110341	69414	39891	74642	49635
32	4387	3860	5006	2505	42476	3602	8089	7533	3119	5489
33	3680	2674	4294	1081	14019	2141	5071	8261	2007	2990
34	3536722	5358319	5033016	4162738	52616393	5827816	9314564	6298502	5747355	9959034
Sum	6303094	9754300	9479049	6220674	90078071	9968102	16184114	10649353	10460798	19723521

Statistics after mapping processed NGS reads to the Human Genome (RefSeq Hg18) and miRBase. Rows represent datasets 552-561, divided into favorable (EFS) and unfavorable (DoD) NBs. Row "Sum/Average" shows the total number of reads and unweighted average statistics. Column "#Reads" refers to the total number of reads for each dataset from the SOLiD run. Column "hg" shows the percentages of reads mappable to the human genome reference sequence Hg18. Column "rna" shows the percentages of reads mappable to annotated RNA sequences in Hg18. Column "pre" shows the percentages of reads mappable to known pre-miRNAs in miRBase. Column "mat" shows the percentages of reads mappable to known mature miRNAs in miRBase. Ratios "mat/pre" and "mat/hg" are shown for convenience. On average, 5% of all sequences (range 2.6-12.1%) were identified as mature miRNAs and 6.1% as pre-miRNAs. The final column nomap lists the percentages of reads that could not be mapped.

Dataset (Class)	#Reads	hg	rna	pre	mat	<i>mat/pre</i>	mat/hg	nomap
552 (EFS)	6,303,094	43.5 %	28.6 %	12.7 %	8.1 %	0.64	0.19	55.7 %
553 (EFS)	9,754,300	38.6 %	27.4 %	17.9%	12.1%	0.68	0.31	60.8 %
554 (EFS)	9,479,049	45.8 %	31.2 %	20.5 %	12.1%	0.59	0.26	53.4 %
555 (EFS)	6,220,674	50.2 %	35.4 %	10.7%	7.4 %	0.69	0.15	49.3 %
556 (EFS)	90,078,071	33.5 %	16.4%	9.73%	2.6 %	0.27	0.08	66.0 %
557 (DoD)	9,968,102	56.2%	35.4 %	8.9%	4.9%	0.55	0.09	42.8 %
558 (DoD)	16,184,114	45.5 %	28.0 %	10.1%	5.6%	0.55	0.12	48.9 %
559 (DoD)	10,649,353	53.7 %	36.7 %	11.6%	6.3%	0.54	0.12	45.2 %
560 (DoD)	10,460,798	48.9 %	31.2 %	10.7%	5.8%	0.54	0.12	50.5%
561 (DoD)	19,723,521	41.2 %	22.1%	11.6%	6.5 %	0.56	0.16	58.4 %
Sum / Average	188,821,076	40.3 %	29.2 %	11.2%	5.1%	0.46	0.13	53.1%

(A) Quantile-quantile plots (black circles) of raw miRNA counts (number of mapped reads) in each patient vs. dataset 552. Blue lines: main diagonal. Red lines: Best diagonal fit of present qq-plot. The vertical shift between black and blue line equals the logarithm of the linear scaling factor to normalize each dataset to dataset 552. (B) Box-plot comparison of the miRNA count distribution: before normalization (left, raw counts), after qq-scale normalization as described in Materials and Methods and S1 (middle), and after a simple normalization method scaling the total read count in each dataset to 1,000,000 (right). Patient datasets are designated 552-561.



Scatter plots of miRNA expression from NGS vs. RT-qPCR Ct values for each dataset. Each figure shows a scatter plot of (logarithmized) normalized miRNA expression values determined from NGS vs. negative Ct values determined by RT-qPCR. Note that Ct (cycle threshold) is defined as the number of cycles required for the fluorescent signal to exceed the background level. Therefore, Ct levels are inversely proportional to the logarithmized amount of target nucleic acid in the sample. Transformation of the Ct values by multiplication with (-1) results in negative Ct values which should be proportional to the logarithmized amount of target nucleic acid. Each point represents one of 204 miRNAs. Strong correlation can be observed between NGS and negative Ct values, indicating good agreement between both techniques.



Heat map and cluster dendrogram using all 465 expressed miRNAs. The EFS (552-556) and DoD (557-561) classes are separated. Clustering is based on the Canberra distance and single-linkage clustering. Blue: low expression, yellow: high expression.



(A) MiRNA 3'-variation by dataset, aggregated over all miRNAs. The x-axis indicates the position in each miRNA (-1 corresponds to the most 3' position). The per-dataset variation corresponds to the global picture (see main text).
(B) Empirical p-value distribution for differential 3'-editing. Each plot represents one of the six most 3' miRNA positions, and displays a histogram of empirical p-values for differential 3'-editing. There is no indication of global differential editing because the distribution follows the uniform distribution that is expected for random data.



Histograms of miRDeep scores. Each plot shows a histogram of miRDeep prediction scores for one patient; only the scores above 1.0 are generally considered to be interesting predictions for further examination (shown in red). The number of these predicted miRNAs that are not known pre-miRNAs in miRBase are given. These may be either true new miRNAs, false predictions or similar to known miRNAs.



Putative new miRNAs predicted by miRDeep that met the following criteria: (1) they were independently predicted from at least three different datasets and (2) the best BLAST hit against a known miRNA from miRBase had an E-value >0.1. The leftmost column contains a running number; the next column shows the nucleotide sequence; "#P" provides the number of patients in which predicted miRNAs were observed; "Score" contains the best miRDeep score among these predictions. "Coordinates" shows the location within the Hg18 genome reference, and "RefSeq // RNA" contains the UCSC genome browser RefSeq Genes and RNA Genes annotation of these coordinates. "miRBase" provides the closest BLAST match in miRBase and the respective E-value; (RC) denotes a hit to the reverse complement.

No.	Sequence	#P	Score	Coordinates	RefSeq. // RNA	miRBase
1	AGAAGTGATGAATTGATCAGATA- -GACGAGGCCGG	8	54.2	chr1: 109,444,682–109,444,715	SCARNA2 (NR_003023)	miR-485-5p (E=0.41)
2	ACCTATGATGATGACTGGTGGCG- -TATGAGTCAT	9	1167.7	chr14: 100,485,926–100,485,958	SNORD114-1 (NR_003193)	miR-1259 (E=1.6)
3	CAACCCTAGGAGAGGGTGCCATT	9	5.6	chrX: 109,185,233–109,185,255	TMEM164 (NM_032227)	miR-30c-1* (E=0.81)
4	AGGTAGATAGAACAGGTCTT	8	319.1	chr15: 81,221,816–81,221,835	SCARNA15 (NR_003011) // snoRNA (ACA45)	miR-1266 (E=0.75)
5	ATGCGCCGCCCACTGCCCCGCG	4	1.6	chr3: 50,687,566–50,687,587	-	miR-1229 (E=0.88)
6	GGTATCCGTTTGGGGATGGT	6	1.4	chr15: 74,666,043–74,666,062	SCAPER (NM_020843)	miR-23a* (E=0.75)
7	TATACAGGGGGAGACTCTTA	7	72.5	chr14: 100,579,120–100,579,139	_	miR-300 (E=0.19)
8	TCCATGATGATTTCAAGTTATCC- -CTGTCTGAAG	6	462.3	chr17: 72,068,791–72,068,823	SNORD1B (NR_004396) // snoRNA (R38b)	miR-1208 (RC) (E=0.41)
9	GCTCAGTCCTAGAAGGTCTCT	6	1.1	chr15: 73,291,354–73,291,374	C15orf39 (NM_015492)	miR-484 (E=0.75)
10	ACCTTGCGCTACTCAGGTCTG	5	17.3	chr22: 29,457,598–29,457,618	OSBP2 (NM_030758)	miR-510 (E=3.2)
11	GCACGGCACTGGGGACACGTG	4	1.4	chr16: 1,725,041–1,725,061	MAPK8IP3 (NM₋001040439)	miR-139-5p (RC) (E=0.81)
12	GAAGGCAGCAGTGCTCCCCTGT	3	1	chr3: 16,949,692–16,949,713	PLCL2 (NM_001144382)	let-7d* (RC) (E=0.22)
13	GTCCCACCCCCACTCCTGTTT	3	3.9	chr2: 64,421,409–64,421,430	_	miR-1225-5p (RC) (E=0.75)

# Supporting Material S10: Expression ratios

This table shows the ratio of expression values comparing -3p miRNAs to their -5p form and comparing star miRNAs to their nonstar form. All -3p/-5p and all star/nonstar miRNA pairs in which both miRNAs are expressed in our dataset are included (117 pairs). These are the same pairs that are shown in Fig. 3B (3p/5p) and Fig. 3C (star/nonstar). For each pair and each patient, the ratio of the normalized expression values was computed (first miRNA divided by second miRNA). Raw *p*-values resulting from t-tests and *p*-values adjusted for multiple testing (according to Benjamini-Hochberg), using the function rawp2adjp of the R library multtest are also shown.

There are no significant differences between favorable and unfavorable neuroblastoma.

		favorable neuroblastoma					unfavorable neuroblastoma						
first miRNA	second miRNA	552	553	554	555	556	557	558	559	560	561	p-value	adjusted $p$ -value
miR-744	miR-744*	29.8	14.6	16.5	18.3	12.6	5.6	5.5	6.2	7.3	2.6	0.011	0.682
miR-296-5p	miR-296-3p	31.4	27.8	24.9	16.5	11.4	4.1	0.8	6.6	10.3	18.5	0.018	0.682
let-7i	let-7i*	56.7	75.5	54.1	23.8	17.9	17.9	20.0	19.9	14.7	4.6	0.047	0.682
miR-500	miR-500*	0.5	0.6	1.3	0.7	0.7	1.8	1.4	2.6	0.7	1.4	0.056	0.682
miR-432	miR-432*	62.6	97.8	140.0	52.6	24.0	20.0	51.4	4.2	34.6	7.8	0.059	0.682
miR-876-5p	miR-876-3p	1.2	1.9	1.3	1.0	2.4	1.7	3.4	2.5	2.5	1.9	0.064	0.682
miR-324-5p	miR-324-3p	12.3	0.3	19.2	44.0	48.3	2.0	9.3	4.5	7.9	10.3	0.086	0.682
miR-100-5p	miR-100-5p	202.5	0.0 709.4	470.7	1005 5	204.2	0.9	1.4	1.2	166 1	225.7	0.069	0.062
miR_330-5n	miR_330_3n	20	3 2	419.1	1095.5	204.3	440.7	123.5	93.0 71.5	41 5	223.1	0.092	0.082
miR-26b	miR-26b*	159.0	60.1	138.4	51.3	56.0	19.2	43.9	41.9	65.9	56.7	0.11	0.682
miR-1224-5p	miR-1224-3p	0.5	0.2	1.0	1.5	0.4	0.2	0.2	0.0	0.2	0.6	0.116	0.682
miR-376a	miR-376a <sup>*</sup>	592.5	203.5	338.1	76.4	454.3	200.5	61.7	7.7	211.3	249.7	0.117	0.682
miR-15a	miR-15a*	32.1	527.2	100.4	541.6	84.2	38.7	11.3	25.3	26.9	69.9	0.122	0.682
miR-154	miR-154*	2.2	1.9	1.9	1.2	0.8	1.5	1.0	0.4	1.6	0.7	0.136	0.682
miR-488	miR-488*	0.6	0.7	0.5	0.1	0.5	1.0	1.9	0.9	0.2	1.0	0.147	0.682
miR-485-5p	miR-485-3p	1.0	1.8	2.1	0.2	0.5	0.1	0.3	0.7	0.9	0.2	0.148	0.682
miR-30b	miR-30b*	40.7	46.4	33.5	149.8	25.3	126.2	54.9	211.7	98.4	85.5	0.15	0.682
miR-589	miR-589*	3.1	2.1	0.8	0.9	0.6	1.5	0.3	0.1	0.6	0.5	0.154	0.682
miR-20-5p	miR-20-3p	10.9	14.0	24 5	20.1	U.7 5 2	3.2	3.4 14 0	1.2	1.1	0.7	0.150	0.062
miR_400_5n	miR_400_3n	3.6	6.2	16.6	10.1	50.2	2.8	14.2	7.5	35	9.0 5.7	0.150	0.082
miR-29c	miR-29c*	47.4	20.7	19.6	19.0	31	22.0	33.4	26.7	67.6	27.6	0.183	0.682
miR-17	miR-17*	13.8	7.1	9.3	39.8	9.5	85.3	40.3	25.6	20.2	14.8	0.183	0.682
miR-144	miR-144*	0.1	0.1	0.2	0.2	0.1	0.1	0.1	0.1	1.0	0.9	0.185	0.682
miR-505	miR-505*	4.0	1.0	5.4	3.7	2.6	1.1	5.2	22.1	3.3	25.5	0.19	0.682
miR-323-5p	miR-323-3p	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.193	0.682
miR-337-5p	miR-337-3p	0.1	0.1	0.0	0.3	0.1	0.3	1.3	0.2	0.4	0.1	0.195	0.682
miR-126	miR-126*	7.5	3.4	4.5	1.7	1.0	2.4	1.5	1.1	2.2	2.0	0.199	0.682
let-7e	let-7e*	65.2	72.4	60.7	33.1	100.1	204.0	100.7	100.9	28.1	118.6	0.201	0.682
miR-8//	miR-8//*	1.0	4.1	0.0	1.5	0.7	0.6	0.3	0.2	0.4	0.7	0.213	0.682
miR-151-5p	miR-151-3p miP 1026*	02.4	11.4	29.3	23.9	10.9	18.5	9.I 10.2	11.3	15.2	15.3	0.22	0.682
miR_0	miR_0*	44.2	9.0 20.1	14.0	9.0	3.7	51	77	11.2	4.2	10.0	0.220	0.082
miR-625	miR-625*	1.0	0.3	0.1	0.1	0.1	0.1	0.1	0.0	0.1	0.0	0.24	0.682
miR-222	miR-222*	27.3	24.5	126.7	1.0	3.8	8.5	12.6	0.7	5.2	1.4	0.252	0.682
miR-331-5p	miR-331-3p	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.253	0.682
miR-130b	miR-130b*	7.3	11.8	16.4	8.2	5.1	6.1	24.2	14.7	40.3	6.2	0.262	0.682
miR-342-5p	miR-342-3p	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.267	0.682
miR-542-5p	miR-542-3p	0.9	0.7	0.9	0.9	1.1	0.4	1.0	0.9	0.8	0.6	0.269	0.682
miR-22	miR-22*	1.4	1.2	1.8	8.2	2.6	7.5	17.2	2.1	3.2	3.9	0.27	0.682
miR-23a	miR-23a*	237.5	1259.7	896.2	758.1	449.3	539.7	215.3	335.5	603.7	725.4	0.282	0.682
miR-106b	miR-106b*	2.7	3.0	0.9	0.3	0.3	26.3	2.5	3.2	1.2	3.7	0.283	0.682
mIR-431 mIP 101	miR-431* miP 101*	20.0	0.9	3.0	2.0	0.7	0.8	22.0	20.2	0.0 20.5	1.7	0.285	0.682
miR-181a	miR_181a*	30.0	21.9	23.5	1009.9	90.4 64.8	24.4	27.5	20.2	29.5	20.4	0.298	0.082
miR-136	miR-136*	107.8	14 5	39.0	47	15	17.9	11.0	83	6.4	93	0.307	0.682
miR-493	miR-493*	0.2	0.2	0.2	0.1	0.1	0.4	3.9	0.1	0.6	0.0	0.316	0.682
miR-135a	miR-135a*	34.1	20.6	45.7	49.3	26.8	29.2	147.9	73.4	2.6	64.8	0.319	0.682
miR-214	miR-214*	55.4	222.6	34.2	20.7	62.9	44.9	20.7	16.7	49.7	53.3	0.319	0.682
miR-34a	miR-34a*	114.2	58.5	237.0	464.8	16.1	164.7	82.6	86.6	67.3	22.3	0.32	0.682
miR-378	miR-378*	1.3	1.8	0.5	0.3	0.6	1.1	234.3	0.9	2.7	23.2	0.322	0.682
miR-193a-5p	miR-193a-3p	5.2	2.1	1.4	4.0	4.2	2.2	2.2	0.7	5.6	0.0	0.324	0.682
miR-497	miR-49/*	29.2	2.5	12.4	23.9	13.0	488.5	10.6	39.5	46.2	9.2	0.331	0.682
miR-424 miP 105	miR-424* miP 105*	4.7	1.0	12.0	528.8	51.3	10.4	2.8	3.8 10.1	12.0	1.9	0.331	0.082
miR-15b	miR-15b*	22.1	1.0	24.8	10.1	53.0	13.1	0.8	38.6	13.0	12.7	0.334	0.082
miR-25	miR-25*	156.3	233.0	352.7	118.7	213.1	13632.3	713.7	152.6	112.0	696.9	0.343	0.682
miR-369-5p	miR-369-3p	2.9	2.1	4.1	5.1	2.7	7.3	6.1	1.7	12.3	0.8	0.344	0.682
miR-409-5p	miR-409-3p	0.2	0.1	0.2	0.1	2.5	0.1	0.1	0.2	0.1	0.1	0.352	0.682
miR-423-5p	miR-423-3p	2.3	0.7	0.3	1.8	0.8	3.7	0.7	0.6	0.4	22.9	0.362	0.682
miR-377	miR-377*	0.2	0.0	0.0	0.0	0.1	0.1	0.5	0.1	0.0	0.1	0.367	0.682
miR-486-5p	miR-486-3p	27.3	25.0	39.5	63.6	37.5	688.6	30.7	26.2	19.4	82.0	0.372	0.682
miR-127-5p	miR-127-3p	0.1	0.1	0.1	0.0	0.1	0.2	0.1	38.9	0.0	0.0	0.373	0.682
miR-199a-5p	miR-199a-3p	0.7	1.0	1.1	2.6	1.4	1.2	0.6	151.9	0.9	1.3	0.379	0.683
miR-142-5p	miR-142-3p	(.6	20.5	10.5	12.9	21.3	0.5	6.3	3.7	23.4	12.8	0.398	0.691
mik-654-5p	mik-054-3p	U.3	0.3	0.2	0.0	1.1 10.7	64.0	10.1	31.U 2F 7	20.6	0.2	0.399	0.691
miR_532_5p	miR-532-35	10	99.0 5.0	49.0	2.U 8.0	1 7	04.9 २.२	402.4 R 1	∠0.1 4 ∩	29.0 6 3	21.2	0.407	0.091
miR_23h	miR_23h*	166.9	358.3	441 5	1632.8	476 5	361.0	3400 6	749.3	569.7	680.7	0 421	0 703
miR-181c	miR-181c*	2.8	1.3	2.6	4.1	2.3	4.2	1.7	3.9	4.1	2.0	0.437	0.706
miR-590-5p	miR-590-3p	10.8	10.6	7.3	3.3	2.3	9.1	19.6	2.2	5.1	13.0	0.438	0.706
miR-380	miR-380 <sup>*</sup>	14.7	17.8	6.5	5.1	7.8	27.1	6.7	3.3	17.9	16.9	0.445	0.706
miR-30a	miR-30a*	1.5	1.2	2.5	196.1	10.4	10.1	4.6	28.6	6.4	2.0	0.455	0.706
miR-93	miR-93*	38.1	9.4	8.6	14.1	3.9	25.0	22.8	25.6	7.2	21.1	0.456	0.706
miR-20a	miR-20a*	198.2	508.6	60.1	43.3	14.5	85.0	168.9	69.0	52.8	68.0	0.459	0.706
miR-99b	miR-99b*	903.9	18.0	16.7	13.9	12.4	13.4	7.2	164.3	3.6	52.7	0.465	0.706
miR-664	miR-664*	1.0 DE 4	9.3	14.9	39.1	17.0	5.9	1.9	27.6	12.5	12.0	0.472	0.708
miR-20b	miR-200*	25.4	ŏŏ.4	41.5	41.4	15.5	/ 84.8	5.1	5.7	3.1	2.5	0.497	0.727

			favoral	ole neurobl	astoma			unfavoral	ole neurob	olastoma			
first miRNA	second miRNA	552	553	554	555	556	557	558	559	560	561	p-value	adjusted
miR-340	miR-340*	0.9	0.9	0.3	0.3	0.7	0.3	0.4	1.1	0.3	0.3	0.497	0.727
miR-361-5p	miR-361-3p	7.1	15.6	26.6	17.0	15.2	5.4	7.6	24.4	17.7	10.6	0.519	0.749
miR-379	miR-379*	9.8	9.3	11.1	15.5	9.3	21.5	11.6	5.3	12.6	12.7	0.556	0.776
miR-10b	miR-10b*	123.0	172.3	108.7	113.7	188.3	216.5	50.5	171.5	139.5	245.1	0.557	0.776
miR-454	miR-454*	1.0	0.2	1.0	1.0	0.3	0.2	1.0	1.0	0.2	0.2	0.557	0.776
miR-145	miR-145*	1027.7	921.1	1951.0	2265.0	2517.8	1994.9	942.1	413.2	1783.3	2166.2	0.57	0.784
miR-140-5p	miR-140-3p	0.9	0.4	1.5	0.8	1.2	0.4	0.7	1.6	0.8	0.5	0.582	0.784
miR-629	miR-629 <sup>*</sup>	12.0	2.2	1.9	0.3	0.7	1.2	2.8	3.7	1.2	1.8	0.583	0.784
miR-200b	miR-200b*	1.0	1.0	1.0	4.2	1.0	1.0	1.0	1.0	2.3	1.0	0.597	0.794
miR-148b	miR-148b*	52.2	39.5	11.8	11.0	33.2	218.5	12.0	15.3	9.1	7.7	0.614	0.807
miR-30e	miR-30e*	1.1	0.3	0.7	11.6	1.5	5.5	1.4	15.1	0.8	0.9	0.634	0.824
let-7b	let-7b*	42.6	217.1	40.9	7.9	18.8	145.5	78.5	105.2	53.9	38.5	0.676	0.87
miR-338-5p	miR-338-3p	0.8	0.3	0.5	1.4	1.0	0.9	0.8	0.3	0.7	2.2	0.69	0.877
miR-425	miR-425 <sup>*</sup>	11.0	0.9	7.7	31.3	3.3	19.0	8.9	6.6	5.3	26.6	0.729	0.888
miR-192	miR-192*	13.8	18.6	13.2	23.4	14.2	18.6	7.4	6.7	31.7	29.0	0.729	0.888
miR-769-5p	miR-769-3p	2.7	2.2	1.6	1.7	1.6	0.1	3.2	5.8	2.3	0.3	0.729	0.888
miR-27a	miR-27a*	10.1	160.1	200.7	102.4	380.7	131.1	186.5	99.3	53.2	258.5	0.735	0.888
miR-125a-5p	miR-125a-3p	448.1	307.5	418.2	781.1	357.4	594.6	127.6	313.2	211.9	800.8	0.736	0.888
miR-411	miR-411*	0.3	0.3	0.3	0.6	0.3	0.6	0.0	0.2	0.2	1.1	0.751	0.896
miR-21	miR-21*	500.8	134.7	633.8	2240.1	23.3	1755.2	50.9	360.5	387.5	186.3	0.762	0.896
miR-129-5p	miR-129-3p	0.9	1.0	0.3	0.4	0.1	0.3	0.8	0.4	0.8	0.0	0.768	0.896
miR-483-5p	miR-483-3p	2.2	11.3	7.3	0.7	1.1	8.9	2.9	0.5	5.4	1.1	0.774	0.896
miR-105	miR-105 <sup>*</sup>	20.1	4.9	14.4	4.0	2.4	2.6	4.0	2.0	2.5	26.3	0.781	0.896
miR-132	miR-132*	1.1	3.2	2.2	1.4	2.9	1.1	1.0	1.0	1.5	5.2	0.818	0.925
miR-550	miR-550*	0.5	0.8	1.6	1.3	1.3	0.5	1.1	1.0	2.4	1.0	0.837	0.925
miR-335	miR-335*	7.4	2.9	1.9	4.7	26.0	10.5	14.2	7.6	4.2	1.0	0.838	0.925
miR-545	miR-545*	0.4	0.2	0.2	0.4	0.4	1.0	0.1	0.4	0.1	0.1	0.842	0.925
miR-299-5p	miR-299-3p	6.1	6.5	9.5	13.2	10.5	15.2	8.2	7.8	5.3	7.2	0.846	0.925
miR-671-5p	miR-671-3p	0.1	0.5	4.1	0.3	0.3	2.2	1.2	0.3	0.1	0.8	0.859	0.93
miR-574-5p	miR-574-3p	0.4	0.4	0.2	0.2	0.3	0.3	0.4	0.2	0.2	0.4	0.87	0.934
miR-148a	miR-148a <sup>*</sup>	11.1	25.6	24.2	49.7	15.6	20.2	0.4	68.4	30.4	0.0	0.927	0.961
miR-628-5p	miR-628-3p	1.5	8.9	12.6	15.3	3.7	1.0	1.6	2.3	31.0	3.1	0.928	0.961
miR-33a	miR-33a <sup>*</sup>	0.3	0.4	0.4	0.3	0.0	0.1	0.3	0.4	0.1	0.7	0.941	0.961
miR-224	miR-224*	1.0	1.7	0.2	1.9	1.2	2.2	0.3	0.7	0.7	1.8	0.945	0.961
miR-183	miR-183*	0.4	5.0	1.0	3.2	0.3	8.5	0.2	0.4	0.1	1.2	0.947	0.961
miR-27b	miR-27b*	48.3	38.2	20.7	14.5	25.5	15.2	23.6	38.9	37.5	34.5	0.947	0.961
miR-124	miR-124*	1.5	8.3	3.1	4.2	3.4	0.4	3.7	6.3	1.4	9.3	0.953	0.961
miR-362-5p	miR-362-3p	2.9	5.3	5.3	2.0	2.8	2.3	4.2	2.4	2.7	6.6	0.98	0.98

## Supporting Material S11: miRNA Editing and Terminal Additions

# **A Editing Statistics**

The following table shows editing events in reads that could uniquely be mapped onto mature miRNA sequences. All rows are normalized to 100%.

		sequenced										
		A	U	G	С							
e	А	98.510 %	0.372 %	0.712 %	0.406 %							
ene	U	1.073%	96.997 %	0.688 %	1.242%							
sfer	G	1.559%	0.379 %	97.509 %	0.554 %							
Ψ.	С	0.492 %	0.387 %	0.287 %	98.834 %							

#### B Editing of miR-376a/c

For each dataset, the fraction of  $A \rightarrow G$  editing events at position 6 of miR-376a/c is shown.

#### hsa-miR-376a (reference: AUCAUAGAGGAAAAUCCACGU)

		fa	vorable N	В	unfavorable NB					
	552	553	554	555	556	557	558	559	560	561
$A \rightarrow G$ at position 6	79.0%	70.1 %	69.6 %	54.1 %	71.7%	73.6%	50.0%	81.6 %	43.0 %	73.8 %
rest	21.0%	29.9%	30.4 %	45.9%	28.3 %	26.4 %	50.0%	18.4%	57.0%	26.2%

#### hsa-miR-376c (reference: AACAUAGAGGAAAUUCCACGU)

		fa	vorable N	В	unfavorable NB					
	552	553	554	555	556	557	558	559	560	561
$A \rightarrow G$ at position 6	12.8 %	4.8 %	9.1 %	3.7 %	12.0 %	8.0 %	4.9%	7.2 %	1.7%	15.6%
rest	87.2%	95.2 %	90.9%	96.3%	88.0%	92.0%	95.1%	92.8%	98.3%	84.4 %

#### C Terminal additions to mature miRNAs



# Supporting Material S12: MIQE checklist

Table 1. MIQE checklist for authors, reviewers and editors. All essential information (E) must be submitted with the manuscript. Desirable information (D) should be submitted if available. If using primers obtained from RTPrimerDB, information on qPCR target, oligonucleotides, protocols and validation is available from that source.

ІТЕМ ТО СНЕСК	IMPORTANCE	CHECKLIST
<b>EXPERIMENTAL DESIGN</b> Definition of experimental and control groups Number within each group Assay carried out by core lab or investigator's laboratory? Acknowledgement of authors' contributions	E E D D	Yes Yes Yes
SAMPLE Description Volume/mass of sample processed Microdissection or macrodissection Processing procedure If frozen, how and how quickly? If fixed, with what and how quickly? Sample storage conditions and duration (especially for FFPE sample	E D E E E E S E	Yes Yes No No No
NUCLEIC ACID EXTRACTION Procedure and/or instrumentation Name of kit and details of any modifications Source of additional reagents used Details of DNase or RNAse treatment	E E D E	Yes Yes not applicable DNAse treatment is irrelevant as stem-loop miRNA reverse transcription only ammplies to mature miRNAs (see reference 10 in manuscript)
Contamination assessment (DNA or RNA) Nucleic acid quantification Instrument and method Purity $(A_{260}/A_{280})$ Yield RNA integrity method/instrument RIN/RQI or C <sub>q</sub> of 3' and 5' transcripts Electrophoresis traces Inhibition testing (C <sub>q</sub> dilutions, spike or other)	E E D E E D E	irrelevant (see above) Yes No No Yes Yes No No
<b>REVERSE TRANSCRIPTION</b> Complete reaction conditions Amount of RNA and reaction volume Priming oligonucleotide (if using GSP) and concentration Reverse transcriptase and concentration Temperature and time Manufacturer of reagents and catalogue numbers $C_q s$ with and without reverse transcription Storage conditions of cDNA	E E E D D <sup>1</sup> D	Yes Yes No (Manufacturer does not provide sequences of stem-loop RT primers) Yes Yes No No
<b>qPCR TARGET INFORMATION</b> If multiplex, efficiency and LOD of each assay. Sequence accession number Location of amplicon Amplicon length <i>In silico</i> specificity screen (BLAST, etc) Pseudogenes, retropseudogenes or other homologs? Sequence alignment Secondary structure analysis of amplicon Location of each primer by exon or intron (if applicable) What splice variants are targeted?	E E D D D E E E	No (MIQE guidelines state that for screening studies these specifications are not required) No No No (Manufacturer does not provide this information) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (Manufacturer does not provide this information) No (Manufacturer does not provide this information)
<b>qPCR OLIGONUCLEOTIDES</b> Primer sequences RTPrimerDB Identification Number Probe sequences Location and identity of any modifications Manufacturer of oligonucleotides Purification method	E D E D D	No (Manufacturer does not provide this information) No No (Manufacturer does not provide this information) not applicable Yes No
<b>qPCR PROTOCOL</b> Complete reaction conditions Reaction volume and amount of cDNA/DNA Primer, (probe), Mg <sup>2+</sup> and dNTP concentrations Polymerase identity and concentration Buffer/kit identity and manufacturer Exact chemical constitution of the buffer Additives (SYBR Green I, DMSO, etc.) Manufacturer of plates/tubes and catalog number Complete thermocycling parameters Reaction setup (manual/robotic) Manufacturer of qPCR instrument	E E E D E D E D E E	Yes Yes No (Manufacturer does not provide this information) No (Manufacturer does not provide this information) Yes No (Manufacturer does not provide this information) not applicable Yes Yes Yes Yes
<b>qPCR VALIDATION</b> Evidence of optimisation (from gradients) Specificity (gel, sequence, melt, or digest) For SYBR Green I, $C_q$ of the NTC Standard curves with slope and $y$ intercept PCR efficiency calculated from slope Confidence interval for PCR efficiency or standard error $r^2$ of standard curve Linear dynamic range	D E E D E E	No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) not applicable No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE guidelines state that for screening studies these specifications are not required) No (MIQE specifications are not required) No (MIQE specifications are not required)

 $^{1}$ Assessing the absence of DNA using a no RT assay is essential when first extracting RNA. Once the sample has been validated as RDNA-free, inclusion

of a no-RT control is desirable, but no longer essential. <sup>2</sup>Disclosure of the probe sequence is highly desirable and strongly encouraged. However, since not all commercial pre-designed assay vendors provide this information, it cannot be an essential requirement. Use of such assays is advised against.

$C_q$ variation at lower limit	E	No (MIQE guidelines state that for screening studies these specifications are not required)
Confidence intervals throughout range	D	No (MIQE guidelines state that for screening studies these specifications are not required)
Evidence for limit of detection	E	Yes (see reference Chen et al. and Mestdagh et al. in manuscript)
If multiplex, efficiency and LOD of each assay.	Е	No (MIQE guidelines state that for screening studies these specifications are not required)
DATA ANALYSIS		
qPCR analysis program (source, version)	E	Yes
$C_q$ method determination	E	Yes
Outlier identification and disposition	E	Νο
Results of NTCs	E	No (no NTCs were included)
Justification of number and choice of reference genes	E	Yes
Description of normalisation method	E	Yes
Number and concordance of biological replicates	D	Yes
Number and stage (RT or qPCR) of technical replicates	E	Yes
Repeatability (intra-assay variation)	E	No (MIQE guidelines state that for screening studies these specifications are not required)
Reproducibility (inter-assay variation, %CV)	D	No (MIQE guidelines state that for screening studies these specifications are not required)
Power analysis	D	No
Statistical methods for result significance	E	Yes
Software (source, version)	E	Yes
$C_q$ or raw data submission using RDML	D	Yes

Chen C, Ridzon DA, Broomer AJ, Zhou Z, Lee DH, Nguyen JT, Barbisin M, Xu NL, Mahuvakar VR, Andersen MR et al: Real-time quantification of microRNAs by stem-loop RT-PCR. Nucleic Acids Res 2005, 33(20):e179. Mestdagh P, Feys T, Bernard N, Guenther S, Chen C, Speleman F, Vandesompele J: High-throughput stem-loop RT-qPCR miRNA expression profiling using minute amounts of input RNA. Nucleic Acids Res 2008, 36(21):e143.

# Supporting Material S13: RTqPCR Expression

This table shows the normalized expression values of the RTqPCR experiments on new miRNAs.

The raw data for this table is attached to this document. It can be accessed by right-clicking this text if your PDF viewer supports it.

Samples	Stage	NMYC	DoD	OS (davs)	Relapse	EFS (davs)	seqб CNRQ	seq6 SE(CNRQ)	seq12 CNRQ	seq12 SE(CNRQ)	seq2 CNRQ	seq2 SE(CNRQ)
F65	1	no	no	1406	no	1406	0.2751	0.0059	0.1991	0.1249	2.3338	0.2812
E05 E14	4s	no	no	2416	no	2416	NaN	NaN	0.2247	0.0220	2.9031	0.5761
E181	4	no	no	511	ves	367	NaN	NaN	0.2281	0.0164	2.4736	0.1504
E9	4s	no	no	1163	no	1163	NaN	NaN	0.3251	0.0050	1.1188	0.0366
E35	4	no	ves	1118	ves	532	0.8540	0.0359	0.3293	0.0140	0.0571	0.0131
E62	1	no	no	2167	no	2167	NaN	NaN	0.3788	0.1129	1.8863	0.0873
E37	4s	no	no	1750	no	1750	NaN	NaN	0.4389	0.1118	1.4687	0.0398
E58	1	no	no	2474	no	2474	NaN	NaN	0.4497	0.0181	1.0632	0.1027
E13	4	no	no	3011	no	3011	NaN	NaN	0.4731	0.1028	1.9387	0.2298
E39	4s	no	no	1839	no	1839	NaN	NaN	0.4961	0.0530	1.1677	0.0441
E176	4	yes	yes	889	yes	482	1.2866	0.0274	0.5119	0.1743	NaN	NaN
E30	2	no	no	2484	yes	485	NaN	NaN	0.5221	0.0246	1.3660	0.1076
E10	4s	no	no	2722	no	2722	NaN	NaN	0.5366	0.0641	1.9276	0.1350
E63	4	no	no	1456	no	1456	0.4885	0.0295	0.5377	0.0457	0.9600	0.0583
E34	4	no	yes	2836	yes	2016	NaN	NaN	0.5534	0.0426	0.1523	0.0178
E56	1	no	no	3745	no	3745	NaN	NaN	0.5832	0.0646	1.5899	0.2343
E21	4s	no	no	2743	no	2743	NaN	NaN	0.5907	0.0831	1.1637	0.1717
E40	4s	no	no	2900	no	2900	NaN	NaN	0.5964	0.1673	2.6716	0.4809
E24	4	no	no	3534	no	3534	NaN	NaN	0.6073	0.0803	1.4667	0.0769
E19	4	no	no	1316	no	1316	NaN	NaN	0.6154	0.0464	0.0259	0.0004
E33	1	no	no	3777	no	3777	NaN	NaN	0.6341	0.1267	4.3761	0.3316
E26	4	no	no	4110	no	4110	0.8185	0.0188	0.6448	0.2111	12.8357	0.7705
E60	1	no	no	3605	no	3605	NaN	NaN	0.6486	0.0770	1.6043	0.1252
E29	4s	no	no	1701	no	1701	NaN	NaN	0.6664	0.1503	1.5236	0.1204
E179	4	yes	yes	380	yes	369	0.9635	0.0174	0.7139	0.2204	1.4378	0.0666
E47	4	no	yes	1445	yes	497	NaN	NaN	0.7494	0.1278	0.0159	0.0005
E36	4	no	yes	329	yes	253	NaN	NaN	0.7682	0.0307	1.1725	0.0254
E27	4	yes	yes	212	yes	184	NaN	NaN	0.8231	0.1073	0.7737	0.0293
E38	4	no	yes	369	yes	344	NaN	NaN	0.8541	0.0935	0.5829	0.0111
E48	4	no	yes	770	yes	597	0.8108	0.0370	0.8828	0.0406	0.9902	0.2021
E55	1	no	no	2801	no	2801	0.0035	0.0084	0.9050	0.0649	2.0214	0.0297
E5/	4	no	yes	1397	yes	031	1.22/1	0.0473	0.9140	0.0421	0.0991	0.1086
	1	no	no	2850	no	2850	NaN	NaN	0.9310	0.2530	1.4/80	0.7829
E34 E42	1	no	no	2000	no	2000	NaN	NaN	0.9705	0.0490	2 0261	0.0305
E43 E40	1	10	no	2109	110	615	NaN	NaN	0.9949	0.2317	2.9301	0.1913
E49 E8	<del>т</del> Лс	no	no	2044	yes	2044	0 3102	0.0056	1.0274	0.0474	1 2004	0.0009
E0 F178	45 4	Ves		2044		431	0.3192	0.0000	1.0500	0.0001	0.0020	0.1720
E1/0 F7	4s	no	no	1330	no	1330	0.7328	0.0200	1.0501	0.0197	1 0913	0.0747
E7 F64	13	no	no	2260	no	2260	NaN	NaN	1 1005	0.01971	4 7326	0.6870
E01 F177	3	ves	ves	103	ves	95	3 4341	0.0687	1.1152	0.3370	0.1798	0.0061
E25	4s	no	no	3163	no	3163	0.5721	0.0210	1.1364	0.0915	4.6226	0.1808
E32	1	no	no	2135	no	2135	0.3930	0.0086	1.1478	0.1643	1.2005	0.0586
E61	4	ves	ves	207	ves	201	1.4735	0.0509	1.1598	0.1713	2.1325	0.1075
E45	4	no	no	2284	ves	1399	0.5947	0.0263	1.2143	0.1835	0.3559	0.0231
E15	4s	no	no	1491	no	1491	NaN	NaN	1.2199	0.1263	0.0440	0.0015
E59	1	no	no	2504	no	2504	NaN	NaN	1.2854	0.3339	1.1508	0.2585
E31	4s	no	no	479	no	479	NaN	NaN	1.3290	0.0826	1.0877	0.0303
E42	4	yes	yes	1375	yes	946	NaN	NaN	1.3356	0.3601	1.4472	0.1620
E53	4	no	no	663	yes	455	NaN	NaN	1.3485	0.0996	0.7573	0.0391
E174	3	yes	yes	364	yes	250	NaN	NaN	1.4212	0.1149	2.2269	0.4362
E50	3	yes	no	4553	yes	1917	NaN	NaN	1.4765	0.4053	0.2640	0.0109
E11	4	no	no	1536	no	1536	NaN	NaN	1.5225	0.3688	3.2107	0.0975
E52	4	yes	yes	1191	yes	840	NaN	NaN	1.5503	0.4819	0.8041	0.0667
E1	4	no	no	2283	no	2283	NaN	NaN	1.6274	0.5259	1.3663	0.0640
E44	4	yes	yes	539	yes	351	1.9832	0.0673	1.6934	0.1207	0.4430	0.0508
E17	4s	no	no	2358	no	2358	NaN	NaN	1.8300	0.0340	0.0145	0.0002
E175	4	yes	no	750	yes	471	1.6978	0.2476	1.8877	0.0563	0.2825	0.0223
E41	4	yes	no	547	yes	409	2.9786	0.1106	2.2420	0.0873	2.2823	0.3420
E180	4	yes	yes	203	yes	90	3.1314	0.1233	2.3000	0.0952	3.0299	0.1478

Samples	Stage	NMYC ampl.	DoD	OS (days)	Relapse	EFS (days)	seqб CNRQ	seqб SE(CNRQ)	seq12 CNRQ	seq12 SE(CNRQ)	seq2 CNRQ	seq2 SE(CNRQ)
E3	4	yes	yes	1115	yes	839	NaN	NaN	2.3956	0.2501	0.9107	0.0206
E2	3	no	no	456	no	456	NaN	NaN	2.4944	0.1117	4.3801	0.2104
E12	4s	no	no	1527	no	1527	NaN	NaN	2.5964	0.2462	4.0136	0.3738
E18	4	no	no	1349	no	1349	NaN	NaN	2.6267	0.0580	NaN	NaN
E23	4s	no	no	4844	no	4844	NaN	NaN	2.8027	0.4467	10.5369	1.4074
E20	4	no	no	3219	no	3219	2.1077	0.0594	2.8598	0.1914	2.1657	0.3396
E16	4s	no	no	1199	no	1199	NaN	NaN	2.9870	0.1305	2.4530	0.6659
E46	4	yes	yes	711	yes	433	NaN	NaN	3.4332	0.2714	0.7291	0.0426
E69	4s	no	no	2894	no	2894	1.7010	0.0607	7.6451	1.0859	2.3250	0.2153
E22	4	no	no	3670	no	3670	NaN	NaN	9.1819	2.7777	4.8695	0.8595