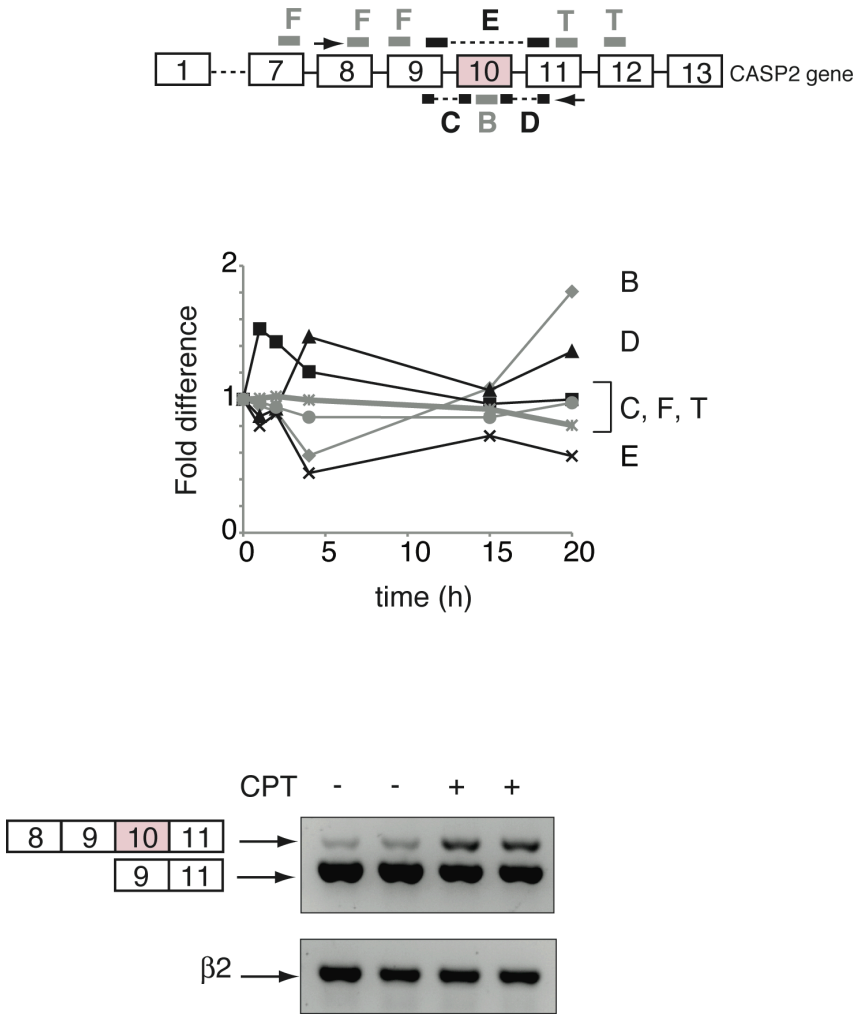
Genome-wide analysis of novel splice variants induced by topoisomerase I poisoning shows preferential occurrence in genes encoding splicing factors

Stéphanie Solier[1], Jennifer Barb[2], Barry R. Zeeberg[1], Sudhir Varma[1], Mike C Ryan[1], Kurt W. Kohn[1], John N. Weinstein[1], Peter J. Munson[2] and Yves Pommier[1]*

[1]*Laboratory of Molecular Pharmacology, Center for Cancer Research, National Cancer Institute, Bethesda, Maryland.*

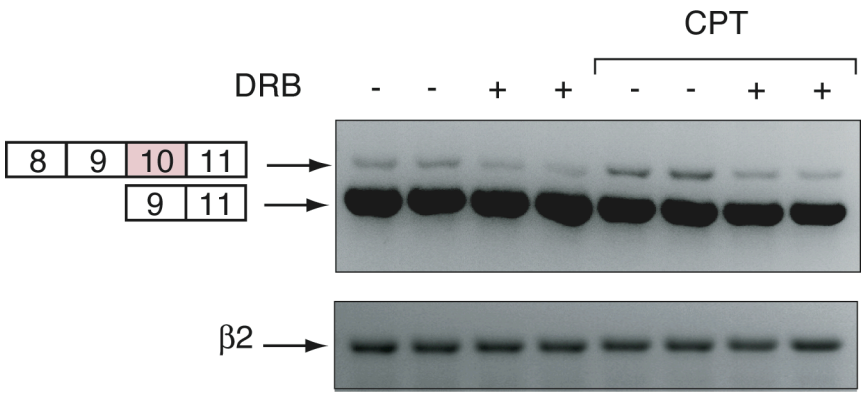
[2]*Mathematical and Statistical Computing Laboratory, Division of Computational Bioscience, Center for Information Technology, National Institutes of Health, Bethesda, Maryland.*

Supplementary Material



Supplemental Figure 1. Validation of the ExonHit results in HCT116 cells treated with CPT (10 μM, 20 h): example of the caspase-2 gene.

Upper panel: schematic gene representation and position of the ExonHit probes (B, C, D, E, F, T) and of the primers used for the RT-PCR (black arrows). Middle panel: fold difference for each of the probes depending on CPT treatment normalized to the untreated controls. Lower panel: RT-PCR showing the effect of CPT on exon 10 inclusion. Caspase-2 mRNA was analyzed by RT-PCR using CASP2 E8s and CASP2 E11as primers. Controls cells received vehicle (DMSO) alone. β2 microglobulin (β2) mRNA was used as a standardizing control.



Supplemental Figure 2. The CDK inhibitor, 5, 6-Dichloro-β-D ribofuranosyl benzimidazole (DRB) suppresses the effect of CPT on the alternative splicing of caspase-2.

HCT116 cells were pretreated with DRB (100 μM, 1 h) prior treatment by CPT (10 μM, 2 h). Caspase-2 splicing was analyzed by RT-PCR using CASP2 E8s and CASP2 E11as primers. Controls cells received vehicle (DMSO) alone. β2 microglobulin (β2) mRNA was used as a standardizing control.

Supplemental Table 1. List of primers

oligonucleotide name	sequence (5' to 3')
AASDHPPT E4s	AAAAGCCATTGGTGTTGGAC
AASDHPPT E6as	CTGGGTTGGTTTGGAATCAT
APTX E3s	CAGCGAATCAGACTTCCACA
APTX E6as	TGGGGTCCTGCATAGAAATC
BAT1 E2s	CTCTGGCTTTCGTGACTTCC
BAT1 E6as	GGGCAGTTCTTCTTCAGCAC
CASP2 E8s	GTTACCTGCACACCGAGTCACG
CASP2 E11as	GCGTGGTTCTTTCCATCTTGTTGGTCA
CCT2 E6s	GTTGGAGAGAAGCCACGAAG
CCT2 E8as	GCATTTTCAATTCGTTTTGG
DDX17 E7s	CCCAGATTCGAGACTTGGAA
DDX17 E9as	ATGTTGTGGTTGGCACTCAA
EIF2S2 E2s	CCCAGCCTTCAGAAACAAAA
EIF2S2 E6as	TTCTCCCCAGCAACCATATC
PNN E4s	AGAAGAGAATCACGCCAGGA
PNN E7as	CCGCTTTTGCCTTTCAGTAG
PPFIA1 E9s	CAGAGCAAAAGCTGCAACAG
PPFIA1 E11as	TCTTCCAAAGCAGCCATTCT
PSMD12 E1s	CGTCAAGATGGAGGTGGACT
PSMD12 E6as	GCCTTCGGTAACCATTCGTA
RBM8A E2s	GATTATGACAGCGTGGAGCA
RBM8A E4as	GTCTCCGGTCTGGACTTCTG
RIOK1 E7s	CACAGCAAATGGAGAGAGCA
RIOK1 E8-9as	CAGGCATGTCATCTTTACCG
RTN4 E13s	CATTTCACTCTTCAGTGTTCCTG
RTN4 E15as	CTGACCCTCCCCCGTATAAT
SF1 E3s	ATTCCCCCTGGACTTACTCG
SF1 E6as	ACTCACACGTGTTGCTGGAG
TCP1 E3s	GGTGCAACCATCCTGAAGTT
TCP1 E8as	GGCAAGCAATTTTTGCATTT
WHSC1 E23s	CACACGAGAACGACATCACC
WHSC1 E25as	CTATCCCCGAGGAATCCACT
ZRANB2 E3s	GAGCCGAGGCCTATTTAGTG
ZRANB2 E6as	GACCAACTGCTTTCCCTCTG

Supplemental Table 2. HTGM parameters

Parameter	Value
GoMiner Build	312
GO Database Build	September, 2009
Data Source	UniProtKB
Organism	*H. sapiens*
LookupSettings	none enabled
Evidence Codes	Level 1
FDR Randomizations	100
FDR & p-value Threshold	0.10
Root Category	GO:0008150 biological_process
Min Category Size	5

HTGM served a dual purpose. (1) First, to provide quality assurance we validate that the majority of the candidate alternatively spliced genes coherently focus into a small number of specific GO categories. We have higher confidence in a group of candidate alternatively spliced genes that all map to the same significant category in contrast to an "isolated" candidate alternatively spliced gene. We can readily assess those alternatively spliced candidate genes meeting the quality assurance criteria *vis a vis* the GO categories that they map to. (2) The second purpose is to select the significant categories and the genes of interest mapping to them.

Supplemental Table 3. List of the genes with a significance index (SI) of 6 or greater.

Gene	SI
AASDHPPT	17.959539
AASS	6.2724678
ABCA8	11.2226934
ABCC9	9.09602522
ABCF3	8.59826277
ABCG2	6.05348611
ABHD12B	7.57391441
ABHD3	6.21554399
ABI3BP	10.5771143
ABTB2	6.47074745
ACACA	6.28951569
ACACB	6.37658455
ACAT2	6.91444144
ACIN1	7.34676388
ACLY	8.89176111
ACSL5	7.31508938
ACTR3B	6.08714405
ADAM19	10.286135
ADAMTS14	8.64715723
ADAMTS17	8.45209472
ADAMTS6	6.61984895
ADCY6	10.8122553
ADD2	9.10765289
AFAP1	7.16055966
AFF4	7.92680845
AGPAT1	6.34941811
AHR	9.10507706
AKR1A1	9.47850168
ALDH1A3	7.43065612
ALDH7A1	6.08144241
ALS2	6.75848169
ALS2CR7	12.0299352
AMBP	10.1401724
AMOT	9.65360758
AMPD3	7.32663081
ANAPC11	8.03063519
ANK3	11.6946264
ANKFN1	7.94663138
ANKRD11	8.00572247
ANKRD5	12.5518866

ANXA1	14.4190682
ANXA2	9.69705406
ANXA6	6.2217634
AP1G2	9.85953433
AP1GBP1	7.51839295
AP2A2	7.46020499
AP4M1	7.24173122
APAF1	6.70574833
APEX1	12.2394718
APITD1	7.70315353
APOB48R	7.37051027
APPL1	8.61695445
APTX	18.2554202
AQP7	10.2212222
AQP9	13.4015721
ARAF	6.70339157
ARHGEF19	13.7866532
ARHGEF5	10.7742084
ARID1B	16.2893232
ARID3B	6.64415975
ARID4A	6.80276338
ARMC3	8.84266045
ARMC9	6.73923896
ARMCX3	6.40464553
ARNT2	6.96153413
ARRB1	6.27989526
ASAH2	6.407854
ASCC2	6.56724365
ASL	6.57218178
ASS1	6.48177633
ASTL	8.87137478
ATG9A	10.9048997
ATM	10.9399978
ATP10D	6.20831774
ATP5A1	6.35257871
ATP6V0A2	10.9166983
ATP8B2	8.08795663
ATPBD4	7.44380604
ATXN2L	7.94624854
AUP1	6.27590402
B3GALNT1	11.7963957
B4GALNT1	7.24034342
B4GALT3	6.27990296
BARX2	6.83895792
BAT1	6.43187807

BAT2	9.15986858
BAZ2A	7.6266616
BCCIP	17.0881438
BCL2L12	6.57163026
BCL2L13	6.88031815
BCL9	8.25284027
BET1	6.76570711
BIN1	12.2759009
BMS1	13.4092797
BNIP3	6.66066412
BPIL2	6.13679792
BRAP	7.01244487
BRD3	7.74113156
BRD8	6.89153611
BTBD10	10.5506637
BUB1B	7.04737555
BXDC2	9.65138104
C10orf118	8.64524433
C10orf137	12.790085
C10orf64	12.126907
C10orf91	6.67742341
C10orf92	6.05338786
C11orf48	8.66988185
C11orf75	6.23394441
C12orf48	6.48352428
C14orf133	7.02262679
C14orf43	6.94864347
C14orf68	7.51179304
C17orf55	6.14719364
C17orf63	8.67692699
C17orf68	6.49043059
C19orf22	7.98234794
C1orf103	6.68817513
C1orf112	9.10160418
C1orf125	7.45790571
C1orf19	7.12280751
C1orf52	12.9135781
C1orf77	14.2905983
C20orf117	7.53928234
C20orf74	16.536651
C22orf30	6.43127335
C2orf50	7.96756257
C3orf19	7.96315407
C4orf20	6.61451004
C4orf28	6.32411419

C4orf29	7.94332361
C5orf5	18.9016595
C6orf107	6.14842852
CABIN1	9.93803087
CAD	11.0527865
CALB2	6.30472455
CAMK2G	8.05351082
CAP2	6.35762724
CARM1	6.70747593
CARS	6.39747509
CASC3	10.6031649
CASP7	7.79082832
CBLC	12.7575358
CBWD5	7.55822038
CC2D1B	9.73817879
CCAR1	6.67331965
CCDC110	17.3736632
CCDC144B	8.00046373
CCDC46	11.0858302
CCDC47	15.2051217
CCDC74A	6.03091474
CCDC75	6.13745566
CCDC77	17.1989042
CCDC84	7.67107374
CCM2	6.2851178
CCNB2	7.18076426
CCNG1	8.48692429
CCNJ	6.84580197
CCNK	9.13714533
CCR7	6.84968476
CCRK	6.17290803
CCT2	17.538741
CCT8	8.03550978
CD109	6.05852761
CD207	7.21660262
CD276	6.60756171
CD3D	6.00527146
CD53	9.0574579
CD97	9.06690312
CDC123	8.60797978
CDC14A	8.7780135
CDC20	6.48857509
CDC20B	6.25386104
CDC2L1	8.0364282
CDH3	6.92272429

CDK5RAP1	8.35925182
CDYL	6.27837365
CENPP	7.93520588
CENPT	8.70567326
CENTD2	6.06001504
CENTG1	7.10782373
CEP110	6.61481243
CEP250	16.225372
CEP76	8.66793934
CHD1L	6.29324642
CHD2	8.25014204
CHD6	6.21925812
CHMP6	7.17195359
CHRNA6	7.5361628
CLCN6	8.48701761
CLEC16A	10.1067823
CLK1	14.8365318
CLPTM1	9.03693247
CNOT3	6.38710545
CNTN5	6.67271829
CNTNAP3	8.30835547
CNTROB	6.62948845
COASY	7.86892514
COG1	7.27543032
COG2	7.52258405
COG4	6.88895086
COL12A1	6.44514948
COL13A1	6.76125055
COL4A5	12.6838807
COMMD3	6.09119876
COPB2	9.28166704
COPS4	6.72241586
COPS7B	7.0450535
COX5A	6.86544425
COX7A1	8.56806091
CPA1	6.19238605
CPNE1	6.95499505
CRNKL1	8.55204492
CSNK1A1	6.63844677
CSNK1G3	6.25838882
CUEDC1	8.74909389
CUTL1	6.27235385
CXorf56	9.69126919
CYCS	11.1892252
CYP20A1	13.2878012

CYP24A1	6.10114188
DACT1	6.28389113
DAPK1	7.6449489
DBN1	9.31049445
DCBLD1	12.0282004
DCTN2	10.902369
DCTN4	6.06848768
DCUN1D1	8.19649205
DDB1	7.13580971
DDB2	6.70339344
DDX1	8.02399318
DDX11	8.00862614
DDX17	17.8533125
DDX25	7.20284405
DDX27	13.3413832
DDX3Y	6.59925254
DDX47	6.44964547
DDX56	8.23498067
DENND1B	7.04805848
DENND2D	6.19945683
DENND4B	13.4637155
DENND4C	11.5068948
DGKG	10.6276607
DHX33	6.1042646
DHX37	7.24335563
DIP2A	7.64674482
DIP2B	6.47963149
DIP2C	7.85390812
DIS3L2	6.60591386
DLG3	7.91440056
DLST	7.73844971
DMXL2	7.92111347
DNAJA3	6.26160792
DNAJC19	7.38776168
DNAJC7	7.38168521
DNAJC9	6.76163024
DNMT1	10.8863226
DNMT3A	11.6734956
DOCK5	6.15379429
DPF3	6.59140146
DPP7	8.37746219
DRG1	6.61250153
DSCR3	9.5060451
DSE	6.51286462
DSG3	10.2510895

DULLARD	7.3708787
DUS2L	8.51871639
DUT	9.92605094
DYNC1H1	9.19589564
DYSF	6.50616586
E2F5	7.15074384
EAF1	7.02117158
EBNA1BP2	18.7995666
EDEM1	7.52866585
EED	17.2428409
EEF2K	6.89475907
EFTUD2	9.35200412
EGLN2	9.41067961
EHMT1	12.4776529
EI24	7.26506477
EIF2AK4	6.99523673
EIF2C2	7.13172136
EIF2S2	19.1675114
EIF2S3	11.3236711
EIF3F	7.05897972
EIF4ENIF1	6.83585645
EIF4G1	11.8057407
EIF4G3	6.8548453
ELOVL2	7.2384836
ELP4	11.1514435
EME1	7.67706174
EML4	6.12444433
EMP2	6.37691195
ENO2	7.5317092
ENPP2	9.0794635
ENPP6	8.75679368
ENTPD3	9.76827602
ENTPD7	8.85891556
EP400	10.7133295
EPB41L4A	9.9080548
EPHB4	7.03228585
ERBB2	13.6776869
ERCC1	7.0990902
ERMP1	6.66493855
EWSR1	15.2930239
EXOD1	15.8238755
F12	6.63492489
F8	7.08893695
FAM115A	7.04883418
FAM120A	7.43339089

FAM38A	7.38406669
FAM44A	8.44112618
FAM49B	10.3049799
FAM50A	6.514028
FAM62B	6.65679725
FAM65A	8.70050455
FAM73A	6.28675946
FAM83B	8.47866631
FAM8A1	6.50225201
FAM9A	6.74685926
FANCA	7.30972733
FARSA	9.41007889
FAS	7.98556146
FASN	14.7163179
FAT	7.42890846
FBL	6.06243154
FBXL10	6.20754309
FBXL11	8.35045427
FBXL17	9.70432025
FBXL2	7.93705677
FBXO11	11.5199182
FBXO22	12.832014
FDPS	8.19831947
FEZ1	11.1818974
FEZ2	10.5877505
FKBP15	6.81508017
FKBP4	6.00249998
FLJ10081	8.11059602
FLJ10781	6.12228344
FLJ11184	9.06030874
FLJ20254	9.92928502
FLJ21986	6.95282114
FLJ23356	10.2394918
FLJ25371	9.3841179
FLJ30092	15.678307
FLJ44048	6.21076129
FOXJ2	6.38730561
FOXK1	12.2605373
FOXR1	9.01881232
FRAS1	7.61513151
FTSJ3	9.78772466
FUBP1	8.44258483
FUNDC1	6.34926949
FYN	9.86101488
GAK	9.91453975

GALNT10	6.30744625
GALNTL4	8.10094207
GANAB	9.73413468
GCAT	7.96707847
GFPT1	16.2731596
GGA3	6.97698174
GGT6	6.01870743
GLT8D1	10.426003
GLTSCR2	8.85373951
GMPPA	7.98964688
GNPDA2	8.70257895
GOLGA2L1	8.75986121
GPKOW	6.0748488
GPM6A	14.8651126
GPR125	7.37508121
GRHL1	12.0023132
GRIN2B	6.98726957
GRK6	6.66633361
GTF2H3	9.64498358
GTF3C1	6.67766245
HAVCR1	6.72251351
HCCS	6.13167736
HCFC1	6.6672746
HD	6.56655726
HDAC3	8.01896161
HDAC4	6.14042025
HDAC6	9.39128718
HDAC9	7.18906289
HDGF	6.23749208
HDGF2	11.5312979
HDPY-30	6.52132009
HEBP1	6.97051508
HEL308	7.30122819
HEPH	10.5202803
HIP1	7.14724932
HMGB2	7.33268279
HNRNPA2B1	7.35146434
HNRPDL	14.9597889
HNRPF	6.41234811
HNRPH3	10.0473516
HPS4	6.48177547
HPS5	6.72049333
HSPA4	12.2304054
HSPA4L	7.87453421
HSPA5	16.4465551

HSPBP1	6.26630034
HSPC111	17.4995733
HTATIP	7.4500206
IDUA	10.7936878
IFIH1	7.62486437
IFT20	9.52446411
IGF2R	6.82737276
IGFBP4	7.61499189
IKBKB	7.89763535
IKBKE	10.0449949
IL17RB	7.47152495
IL1RAPL1	7.47859793
IMPACT	6.40195169
INPPL1	9.99013072
INTS9	6.09539099
IQCE	7.64448031
IQGAP2	13.6977695
IQGAP3	6.87051785
IQSEC2	6.48128252
ITGB4	7.63780569
ITGB5	7.08327213
ITIH2	6.01760574
ITPR1	6.06487672
ITPR2	7.97037492
IVD	7.25668838
IWS1	6.48154031
JARID1A	6.25114284
JARID1C	6.77736477
JARID1D	6.84269687
JOSD3	16.2239606
JTB	7.0081281
KATNAL2	9.32167628
KCNG1	6.04124675
KIAA0182	6.87068609
KIAA0196	6.29447045
KIAA0280	7.65432665
KIAA0319L	6.37150679
KIAA0372	10.6732198
KIAA0515	8.87969106
KIAA0556	9.02065456
KIAA0586	9.85652643
KIAA0974	6.18675068
KIAA1109	8.77276368
KIAA1274	10.716347
KIAA1303	14.4330063

KIAA1529	14.5811454
KIAA1542	9.49700104
KIAA1627	8.22112599
KIAA1632	9.82447366
KIAA1688	6.32864848
KIAA1751	9.73249471
KIAA1797	7.56396867
KIAA1856	13.551489
KIF13B	6.83942616
KIF22	6.55724957
KIF24	6.12567243
KIRREL2	7.13035922
KLK11	6.9184311
KNG1	8.8369758
KPNB1	8.58449157
KREMEN1	7.36366176
KRI1	6.68526165
LAD1	12.0860841
LAMA2	7.09857187
LAMB2	11.4681068
LCP2	7.33213016
LDLR	7.47792693
LEMD2	7.4481769
LENG4	6.200381
LILRA3	6.19158888
LIMCH1	6.29707182
LIN54	7.79782309
LNX1	6.2715321
LOC100101267	6.84610359
LOC144097	6.25988899
LOC146909	7.34065039
LOC162632	6.07962479
LOC197322	16.4530526
LOC219854	6.63573296
LOC400506	6.13703113
LPHN1	7.65047218
LRP1B	10.7411896
LRRC37A3	8.1121459
LRRK1	11.8296463
LSDP5	8.0019745
LSM2	7.99112233
LSM3	15.3943452
LSM8	9.50107994
LTB4DH	7.58982336
LYAR	17.184239

LYZL4	7.90214614
MAGI1	7.57048419
MAP2K5	6.47014994
MAP3K3	6.09374985
MAP3K7IP1	6.91463941
MAP7D1	15.5521656
MARK2	6.11283265
MARS	8.31762364
MAST2	8.10904827
MAT2A	6.24974118
MBP	8.35856424
MEA1	6.20569891
MED12	11.3024734
MED12L	12.8606289
MED15	12.462116
MED23	7.55786068
MED8	7.79061511
MEF2C	6.40452857
METTL3	12.5333599
MFSD8	8.53575073
MGC13379	8.84676333
MGC32805	6.53751322
MGEA5	10.2542784
MGRN1	8.87562862
MIA3	7.24092748
MICAL1	8.74800602
MIST	8.40227375
MKNK1	8.95489628
MKRN2	10.8746236
MKX	6.57089975
MLL2	7.81315466
MMS19	7.93220915
MOBKL1B	10.0634048
MOCS1	6.73357331
MORC1	10.1664015
MORC2	6.38016475
MRPL12	7.27659303
MRPL2	10.5564891
MRPL52	10.7004805
MRPS10	7.67084662
MRPS18B	7.91579614
MRPS23	12.0177173
MS4A8B	7.61097588
MSL2L1	6.60767594
MST1R	7.68371804

MTA2	13.5021203
MTFMT	13.1554363
MTHFD1L	8.08820274
MTMR10	8.15203721
MTMR11	6.54398248
MTRF1L	7.1313028
MTRR	7.21074261
MUM1	9.75662082
MUS81	8.93973011
MXI1	7.74473551
MYB	10.0342463
MYBBP1A	14.9150074
MYCBP	9.26968941
MYH11	7.4135926
MYH9	6.88051557
MYLC2PL	8.07744019
MYLIP	7.37872579
MYNN	7.60224567
MYO1C	14.5333104
MYO9B	8.33934876
MYRIP	12.029172
NAB2	6.25578549
NARF	6.94500794
NAT5	10.8042121
NBEA	8.92298158
NBPF7	7.63366863
NBR1	7.67794366
NCAPD3	7.88147963
NCOR1	7.80917934
NCOR2	11.6117321
NDUFA8	6.25247439
NDUFAB1	14.5654788
NDUFS7	10.7415028
NECAP1	6.89149166
NEFH	8.10510168
NEK3	6.52812036
NEK4	7.83819737
NET1	6.51890847
NF2	6.9338704
NFATC1	6.45810877
NFE2L1	7.36911239
NFIC	6.77656803
NFIX	6.10061912
NFKB2	11.911801
NHN1	14.4987875

NIP30	13.5215042
NLE1	7.90824383
NME7	6.02509471
NOC3L	8.04516843
NOL14	10.1165196
NOL5A	11.7807213
NOLC1	15.8941507
NOSIP	12.3199726
NPAS2	8.71210242
NQO1	8.035266
NRAP	8.22169318
NRD1	6.41001273
NSFL1C	6.41665915
NSUN2	13.0124323
NUDT14	7.33771414
NUDT15	8.57604955
NUMA1	13.4806948
NUP210	9.36446163
NUP214	8.56555186
NXF1	18.0536318
NY-SAR-48	6.8220779
OCIAD1	13.0054725
ODF2	7.5360303
ODF2L	6.89114018
OGDH	6.6110004
OGFOD1	6.64911443
ORAI3	6.60223847
OSBP2	7.83018769
OTC	6.17732541
OTUD3	7.63351495
OTUD5	8.2873896
OVOS2	6.7464022
OXA1L	7.58684632
P2RY6	6.6576954
PABPC1	17.0898496
PABPN1	13.882367
PAGE3	6.07532022
PAK4	6.64695125
PALM2-AKAP2	7.20925138
PAQR5	7.43031784
PARP2	7.33973575
PCF11	10.8408688
PCM1	7.49096431
PCSK7	7.02946869

PDE10A	6.11799349
PDE4DIP	7.98333044
PDE5A	6.90291176
PDGFB	6.43998108
PDIA5	6.32747449
PDILT	6.03928701
PDZD2	7.14718143
PDZRN4	6.80281147
PELP1	6.18402018
PET112L	8.785963
PGAP1	6.18544604
PGS1	10.5280826
PHF1	7.60088775
PHF15	6.20351888
PHF19	7.9113392
PHF2	9.09917983
PHKA2	6.85477086
PHLDB1	10.6102495
PI4KA	6.01170008
PIK3C2B	6.83550346
PIK3C2G	6.77662413
PILRB	9.86200814
PIP5K1C	6.76423218
PKN3	14.2710508
PLA2G1B	12.2754799
PLA2G7	6.54022845
PLAGL1	6.3412048
PLCD1	6.13618439
PLCG1	8.52239652
PLCH2	6.15849754
PLEKHG3	7.64241278
PLEKHH2	6.73811037
PLEKHM1	6.06011198
PLOD1	10.4048573
PLRG1	13.7476983
PLXNB2	10.5196063
PMS1	10.1047128
PNN	18.3309348
POLDIP2	7.73199071
POLE	15.8844233
POLG2	12.8626091
POLH	10.9427618
POLL	7.22445539
POLR1E	7.91051598
POLR2F	6.94848426

POLR3C	16.9980907
POMT1	7.94279874
POU2F1	15.2954032
PPFIA1	17.501694
PPIG	7.20334515
PPIL1	7.52806496
PPIL3	6.75937611
PPM1A	7.26128831
PPOX	8.04168672
PPP2R5C	10.1025286
PPP6C	6.78963457
PRC1	8.2226817
PRDM13	7.77576418
PRDX1	7.06950997
PREX1	6.01126638
PRKCE	12.7760772
PRMT5	9.22128049
PRNPIP	7.92222255
PRPF3	6.15161922
PRPF6	7.38591862
PSMC5	8.14745577
PSMD12	18.3880881
PSMD13	6.53079721
PSMD5	7.00772308
PSPH	6.95038163
PTAR1	6.9988995
PTCD1	7.124887
PTGES2	6.858876
PTPN11	6.20216163
PTPN9	7.83044863
PTPRB	11.6389099
PTPRU	8.78270602
PTRH1	8.3834025
PVR	6.28257219
QSOX2	8.00016205
R3HDM2	9.61804923
RAB11FIP3	7.25431007
RAB31	8.93743081
RAB40B	7.75906412
RAB5B	7.9218267
RAC1	13.4672883
RAD1	9.70348897
RAI1	10.0868275
RALGPS1	7.34589275
RANBP3	11.0990589

RAPGEF5	6.6565571
RASA3	9.21731394
RASGRP2	7.42575185
RBM10	7.18033043
RBM26	10.3802509
RBM28	7.01514171
RBM35B	6.85435688
RBM45	12.2860717
RBM8A	18.5786893
RDHE2	9.20912766
RECQL5	10.821508
RELN	6.27766919
RER1	6.82116726
RFC4	7.98538357
RFT1	7.43298783
RFTN1	7.3316778
RFX5	10.6729466
RG9MTD3	8.63827694
RGL3	12.3181335
RGPD1	15.7148564
RICH2	7.53877784
RINT1	7.26365356
RIOK1	8.88814888
RIT1	7.1086682
RNASEH2A	7.71799195
RNF10	10.2217628
RNF141	8.01581017
RNF165	7.35670062
RNF167	9.03295041
RNF19B	8.52442698
RNF213	7.84668908
ROBO1	11.479294
RP11-49G10.8	6.1433751
RP11-529I10.4	6.32408894
RP4-691N24.1	6.57180828
RPIA	6.09821603
RPL35A	11.9350152
RPL4	7.23169521
RPL9	15.4070426
RPS24	7.55291529
RPS4X	12.6996634
RRBP1	10.3809621
RRP1	8.84020963
RRP12	8.69050643
RRP9	9.02420667

RSPRY1	6.30279737
RSRC2	10.0638103
RSU1	6.70359129
RTN4	17.5648622
RUFY3	6.31305599
RWDD2B	7.53280401
RXRA	6.22963391
SAP30BP	9.5496474
SAPS1	8.29171414
SAPS2	9.16571385
SAR1A	7.16146314
SASH1	8.2822124
SBDS	8.88980078
SCIN	7.20027169
SCN8A	9.12264233
SCYL1	8.67085934
SCYL2	18.1525305
SDC2	8.85167664
SDHA	7.98168441
SDHB	14.693337
SEC16A	9.38600162
SEC23A	16.0044128
SEC23B	8.60948589
SEC61A2	7.48852803
SEC63	6.17689014
SEMA6B	10.6611887
SENP3	6.18340237
SERAC1	6.16314007
SERPINB12	10.5474896
SF1	6.20848482
SF3A1	14.5944276
SFI1	7.51157955
SFRS15	8.04512549
SFRS8	9.15397473
SH3BP1	11.1779282
SH3D19	16.7589905
SH3PXD2B	6.9275311
SH3TC2	7.37892964
SHARPIN	6.01201173
SHROOM1	11.2847411
SIGLEC9	6.19282326
SIL1	15.9735748
SIP1	13.9983818
SIPA1L3	15.1915803
SLC12A6	16.2079721

SLC17A3	8.7890919
SLC22A5	7.99912726
SLC25A15	6.51329233
SLC25A16	6.06110033
SLC25A30	6.83408095
SLC25A43	6.42177743
SLC26A6	11.3647177
SLC30A6	10.4978099
SLC30A7	7.27635997
SLC31A1	6.62616532
SLC33A1	6.55812712
SLC35B1	7.25301876
SLC35B4	6.17973242
SLC38A2	17.157503
SLC43A3	16.5483356
SLC44A1	8.79220206
SLC45A4	6.10573643
SLC5A11	6.59412971
SLC5A6	9.27231955
SLC9A8	14.847079
SLCO1A2	9.83524548
SMAD4	16.3062861
SMC1A	6.54711354
SMC3	6.43195933
SMPD4	8.99753611
SMS	7.17130972
SMUG1	7.59425309
SNAPC4	7.9235473
SNAPC5	9.04300161
SNRK	7.70645604
SOBP	9.09942132
SORL1	6.49632071
SOX13	14.1163826
SPATA20	6.26821561
SPATA6	6.09336883
SPECC1	6.03702936
SPEG	8.53753209
SPG11	6.99592122
SPOP	6.1972848
SPTBN1	8.06388525
SRA1	7.64935408
SRC	6.05852928
SRCAP	6.18408206
SRRM1	8.26726138
SSBP2	7.02086411

STARD8	6.98304735
STAT6	6.40165872
STEAP4	6.39831095
STIM1	6.90977121
STK36	12.4029903
STK38L	6.25136126
SULF2	11.9948041
SULT4A1	6.30423672
SUPT16H	9.58723035
SUPT6H	6.91907447
SUZ12P	6.09363845
SYNJ1	7.73339865
SYNJ2	6.17862858
SYS1	8.56668913
SYT15	6.69917439
SYT17	6.24786257
TAF15	7.3621015
TAF3	9.50126193
TAF5	6.38878748
TAF9	7.99143999
TAPBPL	6.68737295
TASP1	6.90518966
TBC1D8	9.81568895
TBC1D9B	14.4237086
TBL1Y	8.02689216
TBP	7.46602492
TCF20	6.5542597
TCF3	8.27670813
TCP1	17.4659282
TEAD3	6.58273348
TFAM	6.42240934
TFAP2D	8.44272472
TFEC	8.20812765
TGFA	6.95547675
TGFBR2	6.26714444
TGFBR3	6.43735655
TGFBRAP1	6.16420097
TGS1	6.00589981
THEM4	8.17195837
THOC5	7.49334418
THUMPD1	12.4077506
THUMPD3	8.14939385
THYN1	8.66867255
TIAM2	6.80359991
TJP3	8.15112629

TLE1	6.70917748
TLN2	8.3607757
TM2D1	7.53003233
TM9SF4	8.54380053
TMC7	10.2367433
TMEM106B	7.44360655
TMEM126B	6.80373629
TMEM135	6.6107676
TMEM5	13.0128204
TMEM51	6.79708111
TMEM67	10.6308059
TMEM69	6.1200753
TMEM91	6.4674454
TMTC1	9.21096139
TOMM40L	10.9658917
TP73	12.3064265
TPM2	7.09958819
TPR	10.6823106
TPX2	6.26210958
TRAPPC6B	7.0469533
TRDN	6.64127589
TRIP10	7.19990189
TRIP13	7.64993821
TRMT1	6.63383537
TRMT6	10.6715338
TRMU	6.82430399
TROAP	6.50639685
TRPT1	10.0370931
TRSPAP1	10.7053938
TSEN34	10.9310429
TSFM	8.14668294
TSPAN14	9.55597689
TTC12	9.97770128
TTC14	15.6507219
TTC21B	6.51832117
TUBD1	9.43268307
TUT1	7.5623266
TXNDC6	6.36546248
U2AF1	6.3415517
UBAC2	6.49470416
UBE2I	6.69056426
UBE2J2	11.0831389
UBE2Z	15.4888617
UFM1	6.95119821
ULK2	7.49734882

UNC13A	6.86972467
UNC84B	6.62649745
UNK	7.35841365
UPF3B	8.01304905
USP30	6.66580466
USP37	7.42220581
USP39	7.83972087
USP6	6.70078333
USPL1	9.51649665
UST	7.06136042
VAV1	8.97375265
VAV3	6.50744467
VEGFA	7.99690703
VEZF1	11.927135
VPS13B	7.11654175
VPS13D	6.8386904
VPS33B	7.40749726
VPS8	6.43252883
VTI1A	6.05484059
WBP11	6.41654414
WDR18	8.72441603
WDR23	6.88481791
WDR34	6.63851987
WDR4	6.17395566
WDR57	12.1778145
WDR59	6.51459094
WDR61	13.0996666
WDR69	9.25340774
WHSC1	18.2282248
WNT3	6.30649581
WNT8A	7.60090921
YEATS2	6.91610652
ZBTB11	7.19050128
ZBTB32	11.3732502
ZC3H6	7.46472595
ZCCHC2	9.61989309
ZCWPW1	6.64759385
ZFP90	9.55188854
ZKSCAN4	6.15276777
ZNF146	8.16197573
ZNF160	8.2990843
ZNF192	8.28782385
ZNF236	10.3354444
ZNF282	7.10431545
ZNF295	7.0161297

ZNF330	7.39178485
ZNF428	10.2689686
ZNF44	9.34923991
ZNF507	8.16024417
ZNF533	7.2308206
ZNF553	7.13436021
ZNF608	10.131503
ZNF704	8.17659814
ZNF706	11.6721363
ZNF76	6.81998921
ZNF783	6.71374931
ZRANB2	10.5055513
ZWILCH	8.2179599
ZZEF1	6.0143314

Supplemental Table 4. List of the genes validated by RT-PCR.

Gene	SI	Transcript E-1/E/E+1	Transcript E-1/E+1
EIF2S2	19.17	decrease	appearance
RBM8A	18.58	decrease	appearance
PSMD12	18.39	decrease	appearance
PNN	18.33	decrease	appearance
APTX	18.26	decrease	appearance
WHSC1	18.23	decrease	*no appearance*
AASDHPPT	17.96	decrease	appearance
DDX17	17.85	*weak decrease*	*no appearance*
RTN4	17.56	decrease	appearance
CCT2	17.54	decrease	appearance
PPFIA1	17.50	decrease	appearance
TCP1	17.47	decrease	appearance
ZRANB2	10.51	decrease	appearance
RIOK1	8.89	decrease	appearance
BAT1	6.43	decrease	increase
SF1	6.21	decrease	increase

The table displays the name of the gene (the splicing-related proteins are highlighted in gray), the significance index (SI), and the variation (decrease, increase or appearance) of transcript E-1/E/E+1 or transcript E-1/E+1 observed by RT-PCR. The results obtained by RT-PCR were in agreement with the ExonHit array results, except in three cases (italic).