

# Relative strength of 5' splice-site strength defines functions of SRSF2 and SRSF6 in alternative splicing of Bcl-x pre-mRNA

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## Running title:

5' splice site determines splicing of Bcl-x

## Key words:

SRSF2, SRSF6, 5' splice-site, Bcl-x, alternative splicing

**Supplementary Figure 1. Multiple potential binding motifs of SRSF2 and SRSF6 are predicted in the Bcl-x minigene.**

Sequences of Bcl-x minigene are shown. Exon is shown with upper case. Introns are shown with lower case. Potential binding sites of SRSF2 and SRSF6 are shown with red and blue colors separately. Overlapped binding sites of SRSF2 and SRSF6 are shown in orange. 5' splice-sites show shown with slashes. Sequences analyzed in Figure 3 for potential SRSF2 and SRSF6 sites are shown with “SRSF2” and “SRSF6”.

CTCTTTCTCTCCCTTCAGAAATCTTATCTTGGCTTTGGATCTTGAAGAGAATCACTAACCCAGAGACGAGACTCAGTGAGTGAG  
CAGGTGTTTTGGACAATGGACTGGTTGAGCCATCCCTATTATAAAAAATGTCTCAGAGCAACCGGGAGCTGGTGGTTGACTTT  
CTCTCTACAAGCTTCCAGAAAAGGATACAGCTGGAGTCAGTTTAGTGATGTGGAAGAGAACAGGACTGAGGCCCCAGAAG  
GGACTGAATCGGAGATGGAGACCCCAAGTGCCATCAATGGCAACCCATCCTGGCACCTGGCAGACAGCCCCGCGGTGAATGG  
AGCCACTGGCCACAGCAGCAGTTTGGATGCCCGGGAGGTGATCCCCATGGCAGCAGTAAAGCAAGCGCTGAGGGAGGCAGG  
CGACGAGTTTGAACGCGGTACCGGCGGGCATTCACTGACCTGACATCCAGCTCCACATCACCCAGGGACAGCATATCAG  
AGCTTTGAAACAG /5'(S) GTAGTGAATGAACTCTCCGGGATGGGGTAAACTGGGGT

SRSF6: [CGCATGTGGCCTTTTCTCTTCGGCGGGCACTGTGCGTGAAAGCG]

TAGACAAAGGAGATGCAGGTATTGGTGAGTCGGATCGCAGCTTGGATGGCCACTTACCTGAATGACCACCTAGAGCCTTGGAT

SRSF2: [CCAGGAGAACGGCGGCTGG /5'(L) gtaagaaccaa]

gcccctgtgtgccctttttggcctctgfcagagatccccacagcccttctctgtatctctttctgtgtgtgggtgattgttgagacggtgatagtgaggaaacctgactggcctcattaccacaa  
gaggttaactcttgaaatacaaatgacctgtgcttcagagatcaggcatcttaccctgtctctacctgtttctcagcagtaacatggttgattatcaccaatggagggtcaagaagagaccacc  
ttagccctttacctggccccagatgggcacagatggctacagttttcacatcaaccaagccatagcatgcagatggccccttccccacccacctacatcactctctgaggactaaaactgcacctctct  
ctctccctgcag /3'

GATACTTTGTGAACTCTATGGGAACAATGCAGCAGCCGAGAGCCGAAAAGGGCCAGGAACGCTTCAACCGCTGGTTCCTGA  
CGGGCATGACTGTGGCCGGCGTGGTTCTGCTGGGCTCACTCTTCAGTCGGAAA

Supplementary Table 1. Primer list.

Primer Name	Sequence (5'-3')
(N)E2aF	GCACAGCTAGCCTCTTTCTCTCCCTTCAGAATCTTATCTTG
(E)E3R	ATCTGGAATTCTTTCCGACTGAAGAGTGAGCCCAGCAG
I2F	ATCTTTCACCCTGGTCTCTACCTGTTTCTC
I2R	GAGAAACAGGTAGAGACCAGGGTGAAAGAT
bm1	TCCCAGTTCAATTACAGCTCTTA
bm2	ATTGTTCCCATAGAGTTCCAC
be1	ATGTCTCAGAGCAACCGGGA
be2	TCATTTCCGACTGAAGAGTG
SRSF <sub>2</sub> M1F	GGATCCAGGAAAACGGCGGTTGGGTAAGAA
SRSF <sub>2</sub> M1R	TTCTTACCCAACCGCCGTTTTCTGGATCC
SRSF <sub>2</sub> M2F	GATCCAGGAGTACGGCGGTTGGGTAAGAA
SRSF <sub>2</sub> M2R	TTCTTACCCAACCGCCGTAATCCTGGATC
SRSF <sub>2</sub> M3F	GATCCAGGAAAACGGCGGCT
SRSF <sub>2</sub> M3R	AGCCGCCGTTTTCTGGATC
SRSF <sub>2</sub> M4F	GAACGGCGGTTGGGTAAGAAC
SRSF <sub>2</sub> M4R	GTTCTTACCCAACCGCCGTTCC
SRSF <sub>2</sub> M5F	GGATCCAGGAAGGCAACGATTGGGTAAGAA
SRSF <sub>2</sub> M5R	TTCTTACCCAATCGTTGCCTTCTGGATCC
SRSF <sub>2</sub> M6F	GGATCCAGGAGGGCAATAGCTGGGTAAG
SRSF <sub>2</sub> M6R	CTTACCCAGCTATTGCCCTCCTGGATCC
SRSF <sub>6</sub> M1F	GGGGTCGCATTGTGATCTTTTTCTCCTTCGG
SRSF <sub>6</sub> M1R	CCGAAGGAGAAAAAGATCACAATGCGACCCC
SRSF <sub>6</sub> M2F	GGCCTTTTTCTCCCTTGGCGGGGCACTG
SRSF <sub>6</sub> M2R	CAGTGCCCCGCCAAGGGAGAAAAAGGCC
SRSF <sub>6</sub> M3F	GCGGGGCACTGCACGAGGAAAGCGTAGA
SRSF <sub>6</sub> M3R	TCTACGCTTTCCTCGTGCAGTGCCCCGC
SRSF <sub>6</sub> M4F	GATCTTTTTCTCCCTTGGCGGGGCACTG
SRSF <sub>6</sub> M4R	CAGTGCCCCGCCAAGGGAGAAAAAGATC
E2aD1F	GCAAGGCTAGCGATGCCCGGGAGGTGATCCC
E2aD2F	GCATGGCTAGCGGCAGGCGACGAGTTTGAAGTG
E2aD3F	GCTTCGCTAGCCATCCAGCTCCACATCACCC
E2bD1F	AACAGGTAGTGAATGGGGGCACTGTGCGTG
E2bD1R	CACGCACAGTGCCCCATTCACTACCTGTT
E2bD2F	TTTGAACAGGTAGTGGCTTGGATGGCCACT
E2bD2R	AGTGGCCATCCAAGCCACTACCTGTTCAA
E2bD3F	TTTTTCTCCTTCGGCCGAGCTTGGATGG
E2bD3R	CCATCCAAGCTGCGGCCGAAGGAGAAAA

E2bD4F	ATTGGTGAGTCGGATCGGCGGCTGGGTAAG
E2bD4R	CTTACCCAGCCGCCGATCCGACTCACCAAT
Inv1F	TCAGAGCTTTGAATGGGTAAGAAATGAACTCTTCC
Inv1R	GGAAGAGTTCATTTCTTACCCATTCAAAGCTCTGA
Inv2F	GAACGGCGGCCAGGTAGTGACCAAGCCC
Inv2R	GGGCTTGGTCACTACCTGGCCGCCGTTT
Cons2aF	GAACAGGTAAGTAATGAACTCTTCCGG
Cons2aR	GTTCACTACTTACCTGTTCAAAGCTC
W2aF	GCTTTGAAAGGGTAGTGAATGAACTC
W2aR	ATTCACTACCCTTTCAAAGCTCTGATA
Cons2bF	CTGGGTAAGTACCAAGCCCCTTGTG
Cons2bR	CTTGGTACTTACCCAGCCGCCGTT
W2bF	CTGGGTAAGTAACCAAGCCCCTTGTG
W2bR	CTTGGTACTTACCCAGCCGCCGTT