SUPPLEMENTARY MATERIAL

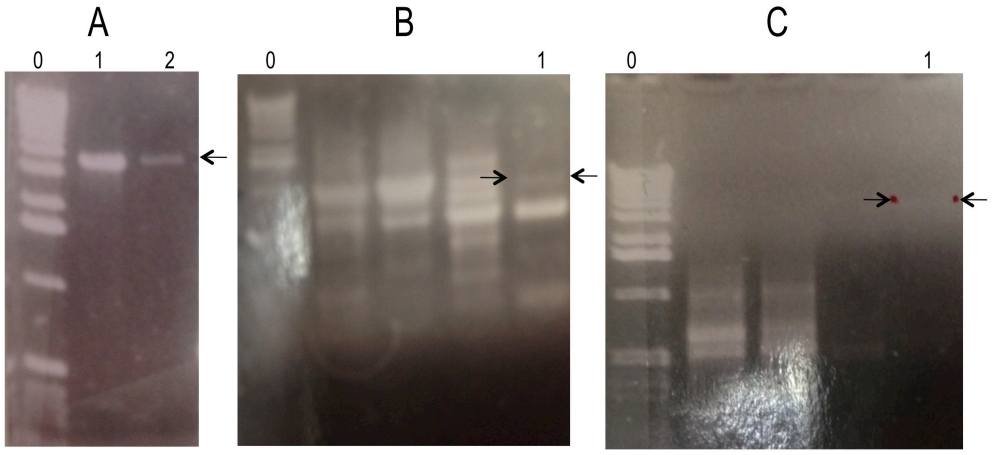
Primers for NBCn1 Splice Variants starting with MERF

For kidney and skeletal muscle, in the first round of PCR, the forward primer designated "OUTF17N1" was CATCATCAGAAACAAGG that is 32-bp upstream of the methionine start codon in the 5'-untranslated region (UTR), and the reverse primer designated "ROUT17N1" was CGCACACATTACATATG that is 37-bp downstream of the stop codon in the 3'-UTR. Using the first-round products (not visible on agarose gels) as template in the second round of PCR, a gene-specific, forward primer "FN1XMAHU" was TCCCCCCGGGGCCACCATGGAAAGATTTCGTCT GG that contains the coding sequence for the first residues of the MERF splice variant, and the gene-specific reverse primer "RN1HINDHU" was CCCAAGCTTCTATAATGAAGTTTCAGC that contains the coding sequence for the last resides of the MERF splice variant. For liver, in the first round of PCR, the forward primer was FN1XMAHU, and the reverse was the RN1HINDHU primer. In the second round, to identify NBCn1 clones, the forward primer was either AP1 that came with the kit or "450" that is upstream of cassette II, and the reverse was "1450" that is downstream of cassette II.

Primers for NBCn1 Splice Variants starting with MEAD

For skeletal muscle, in the first round of PCR, the forward primer "OUTPN1HU" was GGCAGACCGCGCGGGGCACTACCG that is 54-bp upstream of the methionine start in the 5'-UTR of the exon encoding for MEAD, and the reverse primer "N1HU3721R" was CTATATGTATAATGCCTCTTGGTTC that is downstream of the stop codon in the 3'-UTR. Using the first round products as template in the second round of PCR, to confirm PCR products with exon 7, the gene specifc forward primer "N145FHU" was GAAGCTGTTGTGGATCTTGGCAAAA CTAGC that contains the MEAD sequence, and the reverse primer "N1HU3567R" was TTCAAATGGAAGGTGCACAGTATC. Similar PCR products were obtained from skeletal muscle using the forward primer "PUTR130N1" GCGCGCACACCTCGCGCACGGACG in the 5'-UTR of the exon encoding for MEAD with the reverse primer "N1HU3780R" GTGACATGATACGCACACATTACATATG, with second rounds using gene-specific forward primer "PUTR775N1" CGACTCGAGCTGGCCGGCCGGCCATGGAGC and reverse "N13670RHU" CATCCACGTATTTCTTTCTTGGTTCATCTTC. For kidney, in the first round, the forward primer "PUTR1N1" was GCCGGTTCGCTCAGTTCTAGCTTCAGG, and the reverse "N1HU3841R" was GTCTCCACGGTGCTCATTACAAACTCCAGACAC. In the second round, the forward primer was PUTR130N1 that is in the 5'-UTR, and the reverse was N1HU3780R in the 3'-UTR. For liver, in the first round, the forward primer was OUTPN1HU and the reverse was N1HU3780R. PCR products containing exon 7 were detected using the gene-specific forward primer "N145FHU" GAAGCTGTTGTGGATCTTGGCAAAACTAGC or "N1HU450F" GGAAAGAAACT GCTAGATGG, and the gene-specific reverse "N1HU1450R" CCTGGTTGGAACAGGGACCTC or N1HU3780R.

Additional nested-PCR amplification of NBCn1 with Exon 7 from kidney, liver and skeletal muscle cDNA libraries



A. Repeat Experiment showing the 3.7-kb products in **FIG 3A**: lane 0: 1-kb DNA marker; lane 1: nested-PCR product from skeletal muscle; nested-PCR product from kidney. The relative intensities may reflect the relative abundance differences of NBCn1-Exon 7 in these tissues.
B. NBCn1-Exon7 starting with MERF in liver: lane 0: 1-kB DNA marker; lane 1: nested-PCR product using gene-specific primers against Exon 7 itself, yielding a minor 1-kb product.
C. NBCn1-Exon7 starting with MEAD in kidney: lane 0: 1-kB DNA marker; lane 1: nested-PCR product using primers in the UTR region yields a minor ~3.7-bp band.