Supplementary Figure 1. Alignment of the deduced amino acid sequences of mammalian NBCe1-A orthologs. Protein sequence alignments were generated using Clustal W (http://www.ebi.ac.uk/Tools/msa/clustalw2/). The GenBank protein accession numbers for the sequences used in this alignment are AAC51645 (human), ADM25849 (mouse), AAC40034 (rat), and AFS49951 (rabbit). Amino acid residues that are conserved among human, mouse, rat, and rabbit sequences are marked with asterisks. Amino acid residues that are predicted to comprise NBCe1-A domains are marked with an N (cytosolic amino-terminus), T (putative transmembrane spans), L (predicted cytosolic or extracellular loops that join putative transmembrane spans), or C (cytosolic carboxy terminus). Residues that are colored in red are poorly conserved among orthologs. Residues that are highlighted in yellow are not conserved between human and rabbit NBCe1-A. DIDS-binding motifs at the extracellular end of transmembrane span 5 are highlighted in gray. The three putative glycosylation sites in the third extracellular loop are highlighted in green. The four conserved cysteines in the third extracellular loop are highlighted in cyan.

```
mouse       MSTENVEGKPNNLGERGRARSSTFLRVFQPMFNHSIFTSAVSPAAERIRFILGEEDDSPA 60
rat         MSTENVEGKPNNLGERGRARSSTFLRVFQPMFNHSIFTSAVSPAAERIRFILGEEDDSPA 60
human       MSTENVEGKPSNLGERGRARSSTFLRVVQPMFNHSIFTSAVSPAAERIRFILGEEDDSPA 60
rabbit      MSTENVEGKPNNLGERGRARSYTFLRVVQPMFNLSIFTSAVSPAAERIRFILGEEDDSPA 60
            ********** ********** ***** ***** **************************
            NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNN

mouse       PPQLFTELDELLAVDGQEMEWKETARWIKFEEKVEQGGERWSKPHVATLSLHSLFELRTC 120
rat         PPQLFTELDELLAVDGQEMEWKETARWIKFEEKVEQGGERWSKPHVATLSLHSLFELRTC 120
human       PPQLFTELDELLAVDGQEMEWKETARWIKFEEKVEQGGERWSKPHVATLSLHSLFELRTC 120
rabbit      PPQLFTELDELLAVDGQEMEWKETARWIKFEEKVEQGGERWSKPHVATLSLHSLFELRTC 120
            ************************************************************
            NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNN

mouse       MEKGSIMLDREASSLPQLVEMIADHQIETGLLKPDLKDKVTYTLLRKHRHQTKKSNLRSL 180
rat         MEKGSIMLDREASSLPQLVEMIADHQIETGLLKPDLKDKVTYTLLRKHRHQTKKSNLRSL 180
human       MEKGSIMLDREASSLPQLVEMIVDHQIETGLLKPELKDKVTYTLLRKHRHQTKKSNLRSL 180
rabbit      MEKGSIMLDREATSLPQLVEMIVDHQIETGLLKPDLKDKVTYTLLRKHRHQTKKSNLRSL 180
            ************ ********* *********** *************************
            NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNN

mouse       ADIGKTVSSASRMFSNPDNGSPAMTHRNLTSSSLNDISDKPEKDQLKNKFMKKLPRDAEA 240
rat         ADIGKTVSSASRMFSNPDNGSPAMTHRNLTSSSLNDISDKPEKDQLKNKFMKKLPRDAEA 240
human       ADIGKTVSSASRMFTNPDNGSPAMTHRNLTSSSLNDISDKPEKDQLKNKFMKKLPRDAEA 240
rabbit      ADIGKTVSSASRMFTSPDNGSPAMTHRNLTSSSLNDISDKPEKDQLKNKFMKKLPRDAEA 240
            **************  ********************************************
            NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNN

mouse       SNVLVGEVDFLDTPFIAFVRLQQAVMLGALTEVPVPTRFLFILLGPKGKAKSYHEIGRAI 300
rat         SNVLVGEVDFLDTPFIAFVRLQQAVMLGALTEVPVPTRFLFILLGPKGKAKSYHEIGRAI 300
human       SNVLVGEVDFLDTPFIAFVRLQQAVMLGALTEVPVPTRFLFILLGPKGKAKSYHEIGRAI 300
rabbit      SNVLVGEVDFLDTPFIAFVRLQQAVMLGALTEVPVPTRFLFILLGPKGKAKSYHEIGRAI 300
            ************************************************************
            NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNN

mouse       ATLMSDEVFHDIAYKAKDRHDLIAGIDEFLDEVIVLPPGEWDPTIRIEPPKSLPSSDKRK 360
rat         ATLMSDEVFHDIAYKAKDRHDLIAGIDEFLDEVIVLPPGEWDPAIRIEPPKSLPSSDKRK 360
human       ATLMSDEVFHDIAYKAKDRHDLIAGIDEFLDEVIVLPPGEWDPAIRIEPPKSLPSSDKRK 360
rabbit      ATLMSDEVFHDIAYKAKDRHDLIAGIDEFLDEVIVLPPGEWDPAIRIEPPKSLPSSDKRK 360
            ******************************************* ****************
            NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNN

mouse       NMYSGGENVQMNGDTPHDGGHGGGGHGDCEELQRTGRFCGGLIKDIKRKAPFFASDFYDA 420
rat         NMYSGGENVQMNGDTPHDGGHGGGGHGDCEELQRTGRFCGGLIKDIKRKAPFFASDFYDA 420
human       NMYSGGENVQMNGDTPHDGGHGGGGHGDCEELQRTGRFCGGLIKDIKRKAPFFASDFYDA 420
rabbit      NMYSGGENVQMNGDTPHDGGHGGGGHADCEELQRTGRFCGGLIKDLKRKAPFFASDFYDA 420
            ************************** ****************** **************
            NNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNNN

mouse       LNIQALSAILFIYLATVTNAITFGGLLGDATDNMQGVLESFLGTAVSGAIFCLFAGQPLT 480
rat         LNIQALSAILFIYLATVTNAITFGGLLGDATDNMQGVLESFLGTAVSGAIFCLFAGQPLT 480
human       LNIQALSAILFIYLATVTNAITFGGLLGDATDNMQGVLESFLGTAVSGAIFCLFAGQPLT 480
rabbit      LNIQSLSAILFIYLATVTNAITFGGLLGDATDNMQGVLESFLGTAVSGAIFCLFAGQPLT 480
            **** *******************************************************
            NNNNNNNNTTTTTTTTTTTTTTTTTTTTLLLLLLLLTTTTTTTTTTTTTTTTTTTTLLLT
```

```
mouse       ILSSTGPVLVFERLLFNFSKDHNFDYLEFRLWIGLWSAFMCLVLVATDASFLVQYFTRFT 540
rat         ILSSTGPVLVFERLLFNFSKDHSFDYLEFRLWIGLWSAFMCLILVATDASFLVQYFTRFT 540
human       ILSSTGPVLVFERLLFNFSKDNNFDYLEFRLWIGLWSAFLCLILVATDASFLVQYFTRFT 540
rabbit      ILSSTGPVLVFERLLFNFSKDHNFDYLEFRLWIGLWSAFLCLILVATDASFLVQYFTRFT 540
            *********************  **************** ** *****************
            TTTTTTTTTTTTTTTTTTLLLLLLLLLTTTTTTTTTTTTTTTTTTTTLLLLLLLLLLLLL

mouse       EEGFSSLISFIFIYDAFKKMIKLADYYPINSDFKVGYNTHFSCACLPPDPVNLSVSNDTT 600
rat         EEGFSSLISFIFIYDAFKKMIKLADYYPINSDFRVGYNTHFSCACLPPDPVNLSVSNDTT 600
human       EEGFSSLISFIFIYDAFKKMIKLADYYPINSNFKVGYNTLFSCTCVPPDPANISISNDTT 600
rabbit      EEGFSSLISFIFIYDAFKKMIKLADYYPINSDFKVGYNTFFSCACVPPDPVNISIANDTA 600
            ******************************* * ***** *** * **** * *  *** 
            LLTTTTTTTTTTTTTTTTTTTTTLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLL

mouse       LAPEDLPTISSTDMYHNVTFDWAYLSKKECVKYGGKLVGNNCDFVPDITLMSFILFLGTY 660
rat         LAPEDLPTVSSTDMYHNATFDWAYLSKKECVKFGGKLVGNNCDFVPDITLMSFILFLGTY 660
human       LAPEYLPTMSSTDMYHNTTFDWAFLSKKECSKYGGNLVGNNCNFVPDITLMSFILFLGTY 660
rabbit      PAPQELSAVSSTDMHQNATFDWAFLSKKECLRYGGKLVGNNCNFVPDVTLMSFILFLGTY 660
             **  *   *****  * ***** ******   ** ****** **** ************
            LLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLLTTTTTTTTTTTTTT

mouse       TSSMAMKKFKTSRYFPTTARKLISDFAIILSILIFCVIDALVGVDTPKLIVPSEFKPTSP 720
rat         TSSMAMKKFKTSRYFPTTARKLISDFAIILSILIFCVIDALVGVDTPKLIVPSEFKPTSP 720
human       TSSMALKKFKTSPYFPTTARKLISDFAIILSILIFCVIDALVGVDTPKLIVPSEFKPTSP 720
rabbit      TSSMALKKFKTSRYFPTTARKLISDFAIILSILIFCVIDALVGVDTPKLIVPSEFKPTSP 720
            ***** ****** ***********************************************
            TTTTTTTLLLLLLLLLLLLLLTTTTTTTTTTTTTTTTTTTTTLLLLLLLLLLLLLLLLLL

mouse       NRGWFVPPFGGNPWWVCLAAAIPALLVTILIFMDQQITAVIVNRKEHKLKKGAGYHLDLF 780
rat         HRGWFVPPFGGNPWWVCLAAAIPALLVTILIFMDQQITAVIVNRKEHKLKKGAGYHLDLF 780
human       NRGWFVPPFGENPWWVCLAAAIPALLVTILIFMDQQITAVIVNRKEHKLKKGAGYHLDLF 780
rabbit      NRGWFVPPFGGNPWWVYLAAAIPALLVTILIFMDQQITAVIVNRKEHKLKKGAGYHLDLF 780
             ********* ***** *******************************************
            LLLLLLLLLLLLLLLLTTTTTTTTTTTTTTTTTTTTTLLLLLLLLLLLLLLLLLLLLLLT

mouse       WVAILMVVCSFMALPWYVAATVISIAHIDSLKMETETSAPGEQPKFLGVREQRVTGTLVF 840
rat         WVAILMVVCSFMALPWYVAATVISIAHIDSLKMETETSAPGEQPKFLGVREQRVTGTLVF 840
human       WVAILMVICSLMALPWYVAATVISIAHIDSLKMETETSAPGEQPKFLGVREQRVTGTLVF 840
rabbit      WVAILMVVCSFMALPWYVAATVISIAHIDSLKMETETSAPGEQPKFLGVREQRVTGTLVF 840
            ******* ** *************************************************
            TTTTTTTTTTTTTTTTTTLLLLLLLLLLLLLLLLLLLLLLLLLLLLLTTTTTTTTTTTTT

mouse       ILTGLSVFMAPILKFIPMPVLYGVFLYMGVASLNGVQFMDRLKLLLMPLKHQPDFIYLRH 900
rat         ILTGLSVFMAPILKFIPMPVLYGVFLYMGVASLNGVQFMDRLKLLLMPLKHQPDFIYLRH 900
human       ILTGLSVFMAPILKFIPMPVLYGVFLYMGVASLNGVQFMDRLKLLLMPLKHQPDFIYLRH 900
rabbit      ILTGLSVFMAPILKFIPMPVLYGVFLYMGVASLNGVQFMDRLKLLLMPLKHQPDFIYLRH 900
            ************************************************************
            TTTTTTTTLLLLTTTTTTTTTTTTTTTTTTTTTLLLLLLLLLLLLLLLLLLLLLLLLLLL

mouse       VPLRRVHLFTFLQVLCLALLWILKSTVAAIIFPVMILALVAVRKGMDYLFSQHDLSFLDD 960
rat         VPLRRVHLFTSLQVLCLALLWILKSTVAAIIFPVMILALVAVRKGMDYLFSQHDLSFLDD 960
human       VPLRRVHLFTFLQVLCLALLWILKSTVAAIIFPVMILALVAVRKGMDYLFSQHDLSFLDD 960
rabbit      VPLRRVHLFTFLQVLCLALLWILKSTVAAIIFPVMILALVAVRKGMDYLFSQHDLSFLDD 960
            ********** *************************************************
            LTTTTTTTTTTTTTTTTTTTTTTLLLLLLTTTTTTTTTTTTTTCCCCCCCCCCCCCCCCC
```

```
mouse       VIPEKDKKKKEDEKKKKKKKGSLDSDNDDSDCPYSEKVPSIKIPMDIMEQQPFLSDNKPL 1020
rat         VIPEKDKKKKEDEKKKKKKKGSLDSDNDDSDCPYSEKVPSIKIPMDITEQQPFLSDNKPL 1020
human       VIPEKDKKKKEDEKKKKKKKGSLDSDNDDSDCPYSEKVPSIKIPMDIMEQQPFLSDSKPS 1020
rabbit      VIPEKDKKKKEDEKKKKKKKGSLDSDNDDSDCPYSEKVPSIKIPMDIMEQQPFLSESKPS 1020
            *********************************************** *******  ** 
            CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCC

mouse       DRERSSTFLERHTSC 1035
rat         DRERSSTFLERHTSC 1035
human       DRERSPTFLERHTSC 1035
rabbit      DREKSSTFLERHTSC 1035
            *** * *********
            CCCCCCCCCCCCCCC
```