

Chen et al Supplemental Figure S1

Fig. S1: Cloning of porcine ADAM9, ADAM10 and ADAM17/TACE cDNA fragments. cDNA fragments of porcine ADAM9 (930 bp), ADAM10 (725 bp), and ADAM17/TACE (943 bp) were amplified from total RNA isolated from the porcine renal epithelial cell line, LLCPKcl4, by RT-PCR and run on 1% agarose gels as detailed in *Methods and Materials* in the principal text. The specific RT-PCR products were cut out and purified from the gels, followed by cloning into pCRII-TOPO vector using the TOPO T-A Cloning Kit (Invitrogen), respectively. These cloned cDNA fragments were sequenced at the DNA Core at Vanderbilt University, and aligned with the cDNA sequences of ADAM9, ADAM10 and ADAM17/TACE of human and mouse for comparison (Accession numbers were NM_003816, NM_001110, and NM_003183 for human; NM_007404, NM_007399, and NM_009615 for mouse, respectively). The 930-bp ADAM9, 725-bp ADAM10, and 943-bp ADAM17/TACE of porcine origin are 92%, 95%, and 89% homologous to the corresponding human ADAMs and 85%, 91%, and 86% homologous to the corresponding mouse ADAMs. Also noted are the three additional nucleotides, AGT, which are present only in the mouse ADAM10 but absent in both porcine and human ADAM10 as indicated by dashed line “---”. The siRNA sequences used in the present study are indicated by bold type in red. The nucleotide differences within the siRNA sequences between porcine and human or mouse are underlined.

The cDNA sequence of ADAM9 cloned from the porcine renal epithelial cell line, LLCPKcl4 cells:

Human	CACGTCGGAGACATGACAGTGCACAGCTAGTTCTAAAGAAAGGTTTTGGTGGAACTGCAGGAATGGCATTGTG
Mouse	CTCGTCGGAGACACGACAGTGCACAGTTGGTTTTGAAGAAAGGCTTTGGTGGAACTGCAGGAATGGCGTTTGTA
Porcine	CACGTCGGAGACATGACAGTGCACAGCTAGTTCTGAAGAAAGGGTTTTGGTGGGACTGCAGGAATGGCGTTTGTG
Human	GGAACAGTGTGTTCAAGGAGCCACGCAGGCGGGATTAATGTGTTTGGACAAA TCACTGTGGAGACATTTC TTC
Mouse	GGAACAGTATGTTCAAGGAGCCACGCAGGTGGGATCAATGTGTTTGGGCAA TCACTGTGGAGACATTTC ATC
Porcine	GGAACCGTGTGTTCAAGGAGTCATGCAGGCGGGATTAATGTGTTTGGGCGAA TCACAGTGGAGAC <u>CGTTTC</u> ATC
Human	CATTGTTGCTCATGAATTGGGTCATAATCTTGAATGAATCACGATGATGGGAGAGATTGTTCTGTGGAGCAA
Mouse	CATTGTTGCTCATGAATTGGGGCATAACCTTGAATGAATCATGATGATGGGAGAGAGTGTCTGTGGAGCAA
Porcine	CATCGTTGCTCATGAGTTGGGTCATAACCTTGAATGAATCATGATGATGGCAGAGACTGTTACTGTGGAGCAA
Human	AGAGCTGCATCATGAATTCAGGAGCATCGGGTTCCAGAACTTTAGCAGTTGCAGTGCAGAGGACTTTGAGAAG

Mouse AGAGCTGTATCATGAATTCAGGAGCATCCGGGTCAGAACTTTAGCAGTTGCAGTGCAGGAGGACTTTGAGAAG
 Porcine AGAGCTGCATCATGAATTCAGGAGCATCTGGTTCCAGGAACTTTAGCACTTGCAGTGCAGAAGACTTTGAGAAG

 Human TTAACCTTTAAATAAAGGAGGAACTGCCTTCTTAATATTCCAAAGCCTGATGAAGCCTATAGTGCTCCCTCCTG
 Mouse TTAACGTTGAATAAGGGAGGAAGCTGCCTGCTTAACATCCCGAAGCCTGACGAAGCCTACAGCGCGCCCTCCTG
 Porcine TTAACCTTTAAATAAAGGTGGAACTGCCTTCTGAATATTCCAAAGCCTGATGAGGCCTATAGTGCTCCTTTCTG

 Human TGGTAATAAGTTGGTGGACGCTGGGGAAGAGTGTGACTGTGGTACTCCAAAGGAATGTGAATTGGACCCTTGCT
 Mouse TGGTAATAAGCTGGTGGACCCTGGAGAGGAGTGTGACTGCGGCACAGCGAAGGAGTGTGAGGTGGACCCATGCT
 Porcine TGGTAATAAACTGGTGGACCCAGGGGAAGAGTGTGACTGTGGTACTCCAAAGGAGTGTGAATTGGACCCTTGTT

 Human GCGAAGGAAGTACCTGTAAGCTTAAATCATTGCTGAGTGTGCATATGGTACTGTTGTAAAGACTGTCGGTTC
 Mouse GTGAAGGAAGCACTTGTAAAGCTCAAGTCAATGCTGAGTGTGCATATGGCGACTGTTGTAAAGATTGCCAGTTC
 Porcine GTGAAGGAAGTACATGTAAACTTAAATCATCTGCTGAGTGTGCATATGGCGACTGTTGTCAAGACTGTTGGTTC

 Human CTTCCAGGAGGTACTTTATGCCGAGGAAAAACCAGTGAGTGTGATGTTCCAGAGTACTGCAATGGTTCTTCTCA
 Mouse CTTCCAGGAGGCTCCATGTGCAGAGGGAAAGACCAGTGAGTGTGATGTTCCCTGAGTACTGCAACGGTTCTTCTCA
 Porcine CTTCCAGGAGGTACTTTGTGCCGAGGAAAAACCAATGAATGCGACGTTCCAGAGTATTGCAATGGTTCTTCTCA

 Human GTTCTGTCCAGCCAGATGTTTTTATTGAGAAATGGATATCCTTGCCAGAATAACAAAGCCTATTGCTACAACGGCA
 Mouse GTTCTGCCCGCCAGATGTCTTCATTGAGAAATGGATATCCTTGCCAGAACAGCAAAGCCTACTGCTACAATGGCA
 Porcine GTTTTGCCAGCCAGATGTTTTTATTGAGAAATGGATATCCTTGCCAGAATAACAAAGCCTATTGTTACAATGGCA

 Human TGTGCCAGTATTATGATGCTCAATGTCAAGTCATCTTTGGCTCAAAGCCAAAGGCTGCCCCAAAGATTGTTTC
 Mouse TGTGCCAATATTATGACGCGCAGTGTCAAGTCATCTTTGGTTCAAAGGCTAAGGCTGCCCCAAGAGATTGCTTC
 Porcine TGTGCCAGTATTATGATGCTCAGTGTCAAGTCATCTTTGGCTCAAAGCTAAGGCTGCCCCAAGAGATTGTTTC

 Human ATTGAAGTGAATTCTAAAGGTGACAGATTTGGCAATTGTGGTTTTCTCTGGCAATGAATACAAGAAGTGTGCCAC
 Mouse ATTGAAGTCAATTCTAAAGGTGACAGATTTGGCAACTGTGGTTTTCTCCGGCAGTGTGATACAAGAAGTGTGCCAC
 Porcine ATTGATGTGAATTCCAAAGGTGACCGATTTGGCAATTGTGGTTTTCTCTGGCAATGAATATAAGAAGTGTGCCAC

 Human TGGGAATGCTTTGTGTGGAAAGCTTCAGTGTGAGAATGTACAAGAGATACCTGTATTTGGAATTGTGCCTGCTA
 Mouse TGGGAACGCGCTGTGTGGAAAGCTTCAATGCGAGAATGTACAGGACATGCCGGTGTGTTGGAATAGTACCAGCTA
 Porcine TGGGAATGCTTTGTGTGGAAAGCTTCAGTGTGAGAATGTACAAGAAATGCCTGTATTTGGAATTGTGCCTGCTA

 Human TTATTCAAACGCCTAGTCGAGGCACCAAATGTTGGGGTGTGG
 Mouse TCATTGAGACACCCAGTCGAGGCACCAAATGCTGGGGTGTGG
 Porcine TTATTGAGACTCCTGGGAGAGGCACCAAATGTTGGGGTGTGG

The cDNA sequence of ADAM10 cloned from the porcine renal epithelial cell line, LLCPKcl4 cells

Human GTTAATTCTGCTCCTCTCCTGGGCGGCGGGGATGGGAGGTCAGTATGGAAATCCTTTAAATAAAATATATC**AGAC**
 Mouse GTTAATTCTGCTCCTCTCCTGGGCGGCGGGGCTGGGAGGTCAGTATGGAAATCCTTTAAATAAAATATATT**AGAC**
 Porcine GTTAATTCTGCTCCTCTCCTGGGCGGCGGGGCTGGGAGGTCAGTATGGAAATCCTTTAAATAAAATACATT**AGAC**

 Human **ATTATGAAGGATTAT**CTTACAATGTGGATTCATTACACCAAAAACACCAGCGTGCCAAAAGAGCAGTCTCACAT
 Mouse **ATTATGAAGGATTAT**CTTACAATGTGGATTCATTACACCAAAAACACCAGCGTGCCAAAAGAGCAGTCTCACAT
 Porcine **ATTATGAAGGATTGT**CTTATGATGTGGATTCATTACACCAAAAACACCAGCGTGCCAAAAGAGCAGTTTCACAT

 Human GAAGACCAATTTTTACGTCTAGATTTCCATGCCCATGGAAGACATTTCAACCTACGAATGAAGAGGGACACTTC
 Mouse GAGGACCAGTTTTTACTTCTAGATTTCCATGCTCATGGAAGACAGTTCAACCTACGAATGAAGAGGGACACTTC
 Porcine GAGGACCAGTTTTTACGTCTAAATTTCCATGCTCATGGAAGACATTTCAACCTACGAATGAAGAGGGACACTTC

 Human CCTTTTTAGTGATGAATTTAAAGTAGAAACATCAAATAAAGTACTTGATTATGATACCTCTCATATTTTACTG
 Mouse CCTTTTTAGTGATGAATTTAAAGTAGAAACATCAAATAAAGTACTTGATTATGATACCTCTCATATTTTACTG
 Porcine CCTTTTTAGTGATGAATTTAGGGTGGAAACATCAAATAAAGTACTTGATTATGATACCTCTCATATTTTACTG

Human GACATATTTATGGTGAAGAAGGAAGTTTTAGCCATGGGTCTGTTATTGATGGAAGATTTGAAGGATTCATCCAG
 Mouse GACATATTTATGGTGAAGAAGGAAGTTTTAGTCATGGGTCTGTCATTGATGGAAGATTTGAAGTTTTATCAAG
 Porcine GACATATTTATGGTGAAGAAGGAAGTTTTAGCCATGGGTCTGTTATTGATGGAAGATTTGAAGGATTCATTCAG

Human ACTCGTGGTGGCACCATTTTTATGTTGAGCCAGCAGAGAGATATATTAAGACCGAACTCTGCCATTTCACTCTGT
 Mouse ACTCGTGGTGGCAGCTTTTTACATTGAGCCAGCAGAGAGATACATTAAGATCGAATCCTGCCATTTCACTCTGT
 Porcine ACTCATGGCGGCACATTTTTATATTGAGCCAGCAGAGAGATATATTAAGACCGAACTCTGCCATTTCACTCTGT

Human CATTTATCATGAAGATGATATTAACATATCCCCATAAAATACGGTCCTCAGGGGGGGCTGTGCAGATCATTCACTAT
 Mouse CATTTATCATGAAGATGATATTAACATATCCCCATAAAATACGGCCACAGGGGGGGCTGTGCAGATCACTCCGTTT
 Porcine CATTTATCATGAAGATGATATTAACATATCCCCATAAAATATGGTCCACAGGGGGGGCTGTGCAGATCATTCACTAT

Human TTGAAAGAATGAGGAAATACCAGATGACTGGTGTAGAGGAAGTAACACAGATACCTCAAGAAGAATGCTGCT
 Mouse TTGAAAGGATGAGGAAGTACCAAATGACTGGAGTAGAGGAAGGAGCCCGGGCACATCCAGAGAAGCATGCTGCT
 Porcine TTGAAAGGATGAGGAAGTACCAGATGACTGGTGTAGAAGAAGTAACACAGACACCTCAAGAAAAGCATGCTAAT

Human AAT---GGTCCAGAACTTCTGAGGAAAAACGTACAACCTTCAGCTGAAAAAATACTTGTGCTGCTTTTATATCCA
 Mouse AGTAGTGGTCCCTGAGCTCCTGAGGAAAAACGCACAACCTCTGGCTGAAAGAAAATACTTGTGCTGCTTATATCCA
 Porcine AAT---GGTCCAGAACTTCTGAGGAAAAACGTACAACCTTCAGCTGAAAAAATACTTGTGCTGCTTTTATATCCA

Human GACTGATCATTTGTTCTTTAAATATTACGGAACACGAGAAGCTGTGATTGCCAGATATCCA
 Mouse GACAGATCACCTGTTCTTTAAATACTATGGAACACGAGAAGCTGTGATTGCTCAGATATCCA
 Porcine GACTGATCATCTGTTCTTTAAATATTATGGAACACGAGAAGCTGTGATTGCCAGATATCCA

The cDNA sequence of ADAM17 cloned from the porcine renal epithelial cell line, LLCPKcl4 cells

Human GCAGTCTCTCCTATTCCTGACCAGCGTGGTTTCCTTTTCGTGCTGGCGCCGCGACCTCCGGATGACCCGGGCTTCG
 Mouse GCAGTCTCTCCTCATCCTGACCACCTTTGGTGCCTTTTCGTGCTGGCACCCCGACCTCCGGAGGAAGCAGGCTCTG
 Porcine GCAGTCTCTCCTATTCCTGACCAGCTTGGTTTCCTATCGTGCTGGCGCCGCGACCCCGGACGAGCCGGGCTTCG

Human GCCCCCACCAGAGACTC**GAGAAGCTTGATTCTTTGC**TCTCAGACTACGATATTCTCTCTTTATCTAATATCCAG
 Mouse GCTCCCATCCGCGACTT**GAGAAGCTTGATTCTTTGC**TCTCAGACTACGACATCCTCTCCTTAGCTAATATCCAG
 Porcine GCTCCCCTCAGCGACTC**GAAAAGCTTGATTCTTTGC**TCTCAGACTACGACATCCTCTCTTTATCCAGCATTCCG

Human CAGCATTCCGGTAAGAAAAAGAGATCTACAGACTTCAACACATGTAGAAAACACTACTAACTTTTTTCAGCTTTGAA
 Mouse CAGCACTCCATAAGGAAAAGGGATCTACAGTCTGCGACACACTTAGAAAACATTACTAACTTTTTTCAGCTTTGAA
 Porcine CAGCACTCCGTAAGGAAAAGGGATCTGCAGGCCTCAACACACCTAGAGACACTACTAACTTTTTTCAGCCTTGAA

Human AAGGCATTTTTAAATTATACCTGACATCAAGTACTGAACGTTTTTTCACAAAATTTCAAGGTCGTGGTGGTGGATG
 Mouse AAGACATTTTTAAATTATACCTGACATCAAGTACCGAACGTTTTTTCACAAAATTTGAGAGTCGTGGTGGTGGACG
 Porcine CAGGCATTTTTAAATTATACCTGACATCAAGTACTGAACGTTCTCCAGAATTTCAAGTCTGTGGTGGTGGATG

Human GTAAAAACGAAAGCGAGTACACTGTAAAATGGCAGGACTTCTTCACTGGACACGTGGTTGGTGGAGCCTGACTCT
 Mouse GGAAAGAAGAAAGCGAGTACAGCGTGAAGTGGCAGAACTTCTTCACTGGTGGTGGTGGAGCCTGACTCT
 Porcine GGGAAAGATGAAAGTGAGTACCCCGTCAAGTGGCAGGACTTCTTCACTGGACACGTGGTTGGTGGAACTGACTCT

Human AGGGTTCTAGCCACATAAGAGATGATGATGTTATAATCAGAATCAACACAGATGGGGCCGAATATAACATAGA
 Mouse AGGGTTCTAGCCACATAGGAGATGATGATGTTACAGTGAGAATCAACACAGATGGGGCAGAATATAACGTAGA
 Porcine AGGGTTCTCGCCACATAGGAGATGATGATATTACAGTAAGAATCAACACAGATGGGGCAGAATATAATATAGA

Human GCCACTTTGGAGATTTGTTAATGATACCAAAGACAAAAGAATGTTAGTTTATAAATCTGAAGATATCAAGAATG
 Mouse GCCACTTTGGAGGTTTGTCAATGATACTAAAGATAAACGAATGCTGGTGTATAAGTCTGAAGATATCAAGGATT
 Porcine GCCACTTTGGAGACTAATTAATGATACTAAAGACAAAAGAGTGTAGTTTATAAGTCTGAAGATATCAAGAATG

Human TTTACGTTTTGCAGTCTCCAAAAGTGTGTGGTTATTTAAAAGTGGATAATGAAGAGTTGCTCCAAAAGGGTTA
 Mouse TTTACGTTTTGCAGTCTCCAAAAGTATGTGGTTATTTAAAATGCAGATAGTGAAGAGCTGCTTCCAAAAGGGCTC
 Porcine TTTTCGTTTTGCAGTCTCCAAAAGTGTGTGGTTATATAAAGGCGGATAATGAAGAGTTGCTTCCAAAAGGGCTA

Human	GTAGACAGAGAACCACCTGAAGAGCTTGTTTCATCGAGTGAAAAGAAGAGCTGACCCAGATCCCATGAAGAACAC
Mouse	ATAGACAGAGAGCCATCTGAAGAGTTTGTTTCGTTCGAGTGAAGAGACGAGCTGAACCTAACCCCTTGAAGAATAC
Porcine	GTAGACAGAGAGCCGCCTGATGAGCTTGTTTCACCGGGTGAAGAGAAGAGCCGACCCCAATCCCCTGAGGAACAC
Human	GTGTAAATTATTGGTGGTAGCAGATCATCGCTTCTACAGATACATGGGCAGAGGGGAAGAGAGTACAACCTACAA
Mouse	TTGTAAATTACTGGTGGTAGCAGATCATCGATTTTATAAATACATGGGCCGTGGAGAAGAGAGCACCCTACAA
Porcine	GTGTAAATTATTGGTGGTGGCAGATCATCGCTTTTATAAGTACATGGGCAGAGGGGAAGAGAGCACGACCACAA
Human	ATTACTTAATAGAGCTAATTGACAGAGTTGATGACATCTATCGGAACACTTCATGGGATAATGCAGGTTTTAAA
Mouse	ATTACTTAATAGAGCTAATTGACCGAGTTGATGACATATACCGGAACACGTCGTGGGATAATGCAGGTTTTAAA
Porcine	ACTACCTGATAGAGCTAATTGACAGAGTTGATGACATCTATCGGAACACTTCATGGGACAATGCAGGTTTTAAA
Human	GGCTATGGAATACAGATAGAGCAGATTTCGATTCTCAAGTCTCCACAAGAGGTAAAACCTGGTGAAAAGCACTA
Mouse	GGGTATGGAGTGCAGATAGAGCAGATTTCGAATTCTCAAGTCTCCACAAGAGGTAAAACCTGGTGAAAAGCACTT
Porcine	GGTTATGGAATACAGATAGAGCAGATTTCGATTCTCAAGTCTCCACAAGAGGTAAAACCTGGTGAAAAGGCACTA
Human	CAACATGGCAAAAAGTTACCCAAATGAAGAAAAGGATGCTTGGGATGTGAAGATG
Mouse	CAATATGGCAAAAAGTTTCCCAAACGAAGAGAAGGATGCTTGGGATGTGAAGATG
Porcine	CAATATGGCAAAAAGTTACCCAAATGAAGAAAAGGATGCTTGGGATGTGAAGATG