

Supplemental Figure S1. ADAMTSL2 conservation and predicted significance of R221C.

(A) Alignment of ADAMTSL2 orthologs from Ensembl. The mutated arginine (Arg221; red overlay) is among a highly conserved stretch of residues. Sequence similarity among the orthologs listed was 89.7 ± 1.9%. A probable artifact insertion (Ala587) in the Ensembl annotation was revealed by RT-PCR (green overlay). (B) Analysis with SIFT [1], a program that assesses the theoretical impact of polymorphisms, predicted R221C was ‘probably damaging’ (PSIC >2) [2].

A

Dog	MDGQRQPSRWVWALLAVVLLAGGVASTGATDNSPTSNSLEGGTAYWWGEWTKWTACS	60
Cow	MEGRWQSSRWAWPLLAVALVTGSVSSTGTTDNSPTSNNLEVGPDATAYWWGEWTKWTACS	60
Horse	MDGRQWP SHVWVSLLAVALLVAGSAASTGATDNSPTSNSLEGGTAYWWGEWTKWTACS	60
Man	MDGRWQCSCWAWFLLVAVVAGDVTSTGSTDNSPTSNSLEGGTAYFWWGEWTKWTACS	60
Mouse	MDGRRQHPHWAWSLLAVALVAGGAAPTEASDNSPTSNSLEGGADTAYWWGEWTKWTACS	60
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Dog	RSCGAGVTSQERHCLQQRRTSVMGAGNRTCTGTSKRYQLCRVQECPPDGRSFREEQCISF	120
Cow	RSCGAGVTSQERHCLQQRKTSVTGAGNRTCTGTSKRYQLCRVQECPPDGRSFREEQCISF	120
Horse	RSCGAGVTSQERHCLQQRRTSVTGTGNRTCTGTSKRYQLCRVQECPPDGRSFREEQCVSF	120
Man	RSCGGGVTSQERHCLQQRKSVPGPNRTCTGTSKRYQLCRVQECPPDGRSFREEQCVSF	120
Mouse	RSCGGGVTSQERHCLQQRKSVPGTGNRTCVGTSKRYQLCRVQECPPDGRSFREEQCVSF	120
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Dog	NSRVYDGRTHQWKPLYPDDYVHISSKPCDLHCTTVDGQRQLMVPARDGTSCKLTDLRGVC	180
Cow	NSRVYNGRRHQWKPLYPDDYVHISSKPCDLHCTTVDGQRQLMVPARDGTSCKLADLRGVC	180
Horse	NSHVYNGRTHQWKPLYPDDYVHISSKPCDLHCTTVDGQRQLMVPARDGTSCKLTDLRGVC	180
Man	NSHVYNGRTHQWKPLYPDDYVHISSKPCDLHCTTVDGQRQLMVPARDGTSCKLTDLRGVC	180
Mouse	NSRVYDGRAYQWKPLYPDDYVHISSKPCDLHCSTVDGQRQLTVPARDGTSCKLTDLRGVC	180
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Dog	VSGKCEPIGCDGVLFSTHTLDKCGVCQGDGSSCTHVTGNYRKGNAHLGYSLVTHIPAGAR	240
Cow	VSGKCEPIGCDGVLFSTHTLDKCGVCQGDGSSCTHVTGNYRKGNTHLGYSLVTHIPAGAR	240
Horse	VSGKCEPIGCDGVLFSTHTLDKCGVCQGDGSSCTHVTGNYRKGNTHLGYSLVTHIPAGAR	240
Man	VSGKCEPIGCDGVLFSTHTLDKCGVCQGDGSSCTHVTGNYRKGNAHLGYSLVTHIPAGAR	240
Mouse	VSGKCEPIGCDGVLFSTHTLDKCGVCQGDGSSCTHVTGNYRKGNNHLGYSLVTHIPAGAR	240
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Dog	DIQIVERKKSADVLALADEAGYFFNGNFKVDSPKNFNIAGTVVKYRRPMDVYETGIEYI	300
Cow	DIQIVERKKSADVLALADEAGYFFNGNFKVDSPKNFNIAGTVVKYRRPMDVYETGIEYI	300
Horse	DIQIVERKKSADVLALADEAGYFFNGNFKVDSPKNFNIAGTVVKYRRPMDIYETGIEYI	300
Man	DIQIVERKKSADVLALADEAGYFFNGNYKVDSPKNFNIAGTVVKYRRPMDVYETGIEYI	300
Mouse	DIQIVERKKSADVLALADEAGFYFFNGNYKVDSPKNFNIAGTVVKYRRPMDVYETGIEYI	300
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Dog	VAQGPTNOGLNVMVWNQNGKSPSITFEYTLLOPPHTRIQPVYYSFSEPTSESAES---Q	357
Cow	VAQGPTNOGLNVMVWNQNGKSPSITFEYTLLOPPHTRLQPIYYFSFSDPTSDSAES---Q	357
Horse	VAQGPTNOGLNVMVWNQNGKSPSITFEYTLLOPPHARPOPLYYSFSEPAPESTES---Q	357
Man	VAQGPTNOGLNVMVWNQNGKSPSITFEYTLLOPPHESRPQPIYYGFS----ESAES---Q	353
Mouse	VAQGPTNOGLNVMVWNQNGKSPSITFEYTLLOSPHMHLPPVYYSFSEAASQSTESTERQ	360
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Amino Acid Alignment of Mammalian ADAMTS-Like 2 Proteins

Dog ELDGAALLGFLOHNTSFYQASSERLGLDNRLFHGLGPGIELGLSKGQETNEVCEQAGGR 417
Cow ELDGAGLAGFLOHNASLFGQASSERLGLDNRLFHGRGPGIELGLSRGQESNEVCAQASGR 417
Horse ELDGADLVGFLOHNGSLYQASSERLGLDNRLFHGLGPGMELGLPKGQETNEVCEQASGG 417
Man GLDGAGLMGFVPHNGSLYQASSERLGLDNRLFHHPGLDMELGPSQGQETNEVCEQAGGG 413
Mouse ELDSARLLGFMQHNGSLYRQTSSERLGLNSQLFQPPAPEVELGPSRGQESNEVCKQASGG 420
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Dog ACEGPPRGKGF RDGNATGTALTGDKDEQQVDKRLTSQRLLSANVISDQLL GAGSDSRELP 477
Cow ACEGPPRGKGF RDGNATLTALVGD PDDREADARFASQELLSANAISDQLL GAGSDSRELS 477
Horse ACEGPPRGKGF RDGNATGTALAGDQDDHEADVRFASQELLSANAISGQLLGTGSDSKELA 477
Man ACEGPPRGKGF RDNRVTGTPLTGDKDDEEVDTHFASQEFFSANAISDQLL GAGSDLKDFT 473
Mouse VCEGPPRGKGF QDHNATGRAFSADK DREISAHFTSHELLSANTISDQLLGTGSESEEF 480
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Dog FNETGNSIFAQ GAPRRSPAESLYVDYEENEEAGAYLLNGSYLELSSDRVTNSSSEAPFPN 537
Cow LNETVNSIFSQ GAPRGSPAESLYLDYEESEGAGAYLLNGSYLELSSDRLANTSSEAPFPN 537
Horse LNETVNSIFAQ GAPRSSPAETFYVDYEENEGAGAYLLNGSYLELSSDRVANTSSEAPFPN 537
Man LNETVNSIFAQ GAPRSSLAESFFVDYEENEGAGPYLLNGSYLELSSDRVANSSEAPFPN 533
Mouse LNETMNSIFAQ GAPRSSPAESLYVDYEENEGPAAYLINGSYLELSSDRIN-TSSEAPFPN 539
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Dog ASSSLPALAGNRTHKARTRPK-TRKQGVSPADMYRWKLS SHEPCSATCTTAGVMSTYAMC 595
Cow ASASLPALAGNRTO KARTRPK-SRKQGVSPADMYRWKLS SHEPCSSTCTT-GVLSTYAMC 595
Horse ISASLP TTAGNRTHKARTRPKGAASKGVSPADMYRWKLS SHEPCSATCTT-GVMSTYAMC 596
Man VSTSLLT SAGNRTHKARTRPK-ARKQGVSPADMYRWKLS SHEPCSATCTT-GVMSAYAMC 591
Mouse TSASPPNLAGNRTHKARTRPK-ARKQGVSPADMYRWKLS SHEPCSATCTT-GVMSTYAMC 597
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Dog VRYDGI EVDSDYCDALTRPEPVQEF CAGRECQPRWETSSWSECSRTC GEGYQFRIVRCWK 655
Cow VRYDGI EVDSDYCDALTRPEPVQEF CAGRECQPRWETSSWSECSRTC GEGYQFRIVRCWK 655
Horse VRYDGI EVDSDYCDALTRPEPVQEF CAGRECQPRWETSSWSECSRTC GEGYQFRIVRCWK 656
Man VRYDGI EVDSDYCDALTRPEPVHEF CAGRECQPRWETSSWSECSRTC GEGYQFRVRCWK 651
Mouse VRYDGI EVDSDYCDALTRPEPVHEF CAGRECQPRWETSSWSECSRTC GEGH QFRIVRCWK 657
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Dog MLSPGF DSSVYSDLCEAAEAVRPEEHKTCRNPACGPQWEMSEWSECTAKCGERSVVTRDI 715
Cow MLSPGF DSSVYSDLFEAAEAVRPEERKTCRNPACGPQWEMSQWSECTAKCGERSVVTRDI 715
Horse MLSPGF DSSVYSDLCEAAEAVRPEERKTCRNPACGPQWEMSEWSECTAKCGERSVVTRDI 716
Man MLSPGF DSSVYSDLCEAAEAVRPEERKTCRNPACGPQWEMSEWSECTAKCGERSVVTRDI 711
Mouse MLSPGF DSSVYSDLCEATEAVRPEERKTCRNPACGPQWEMSEWSECTAKCGERSVVTRDI 717
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Dog RCSEDEKLCDPNTRPVGEKDCTGPPCDRQWTVSDWGPCSGSCGQGRMIRHVYCKTSDGRV 775
Cow RCSEDEKLCDPNTRPVGEKDCTGPPCDRQWTVSDWGPCSGSCGQGRMIRHVYCKTSDGRV 775
Horse RCSEDEKLCDPNTRPVGEKDCTGPPCDRQWTVSDWGPCSGSCGQGRMIRHVYCKTSDGRV 776
Man RCSEDEKLCDPNTRPVGEKNCTGPPCDRQWTVSDWGPCSGSCGQGR TIRHVYCKTSDGRV 771
Mouse RCSEDEKLCDPSTKPVGEKNCTGPPCDRQWTVSDWGPCSGSCGQGR TIRHVYCKTSDGRV 777
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Amino Acid Alignment of Mammalian ADAMTS-Like 2 Proteins

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Dog  VPESQCQMETKPLAIHPCGDRNCPAHWLAQDWERCNTTCGRGVKKRLVLCMELANGKPQT 835
Cow  VPESQCQMETKPLAIHPCGDKNCPAHWLAQDWERCNTTCGRGVKKRLVLCMELANGKPQT 835
Horse VPESQCQMETKPLAIHPCGDKNCPAHWLAQDWERCNTTCGRGVKKRLVLCMELANGKPQT 836
Man  VPESQCQMETKPLAIHPCGDKNCPAHWLAQDWERCNTTCGRGVKKRLVLCMELANGKPQT 831
Mouse VPESQCQMETKPLAIHPCGDKNCPAHWLAQDWERCNTTCGRGVKKRLVLCMELANGKPOI 837
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Dog  RSGPECGLAKKPPEESTCFERPCFKWYTPWSECTKTTCGVGVRMRDVKCYQGTDIVRGCD 895
Cow  RSGPECGLAKKPPEESTCFERPCFKWYTPWSECTKTTCGVGVRMRDVKCYQGTDIVRGCD 895
Horse RNGAECGLAKKPPEESTCFERPCFKWYTPWSECTKTTCGVGVRMRDVKCYQGTDIVRGCD 896
Man  RSGPECGLAKKPPEESTCFERPCFKWYTPWSECTKTTCGVGVRMRDVKCYQGTDIVRGCD 891
Mouse RSGPECGLARKPPEESTCFERPCFKWYTPWSECTKTTCGVGVRMRDVKCYQGTDIVRGCD 897
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Dog  PLVKPVGRQACDLQPCPTEPPDDSCQDQPGTNCALAIKVNLCGHWYYSKACCRSCRPPHS 955
Cow  PLVKPVGRQACDLQPCPTEPPDDSCQDQPGTNCALAIKVNLCGHWYYSKACCHSCRPPPS 955
Horse PLVKPVGRQACDLQPCPTEPPDDSCQDQPGTNCALAIKVNLCGHWYYSKACCRSCRPPHS 956
Man  PLVKPVGRQACDLQPCPTEPPDDSCQDQPGTNCALAIKVNLCGHWYYSKACCRSCRPPHS 951
Mouse PLVKPVGRQACDLQPCPTEPPDDSCQDQPGTNCALAIKVNLCGHWYYSKACCRSCRPPHS 957
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B

Predicted Not Tolerated	Position/Amino Acid	Predicted Tolerated
	215H 1.00	c w d m p g i n s v f t e H a q y L R k
y w t s r q p n m l k i h g f e d c a	216V 1.00	V
w f y h m c i r l q e d k v p	217T 1.00	g n A T S
y w v t s r q p n m l k i h f e d c a	218G 1.00	G
w m f i c l v y r h p q e k a	219N 1.00	t g d S N
d c k p n q e r g h	220Y 1.00	t s m a v i w L F Y
w c f m	221R 1.00	y i h d p v g l n e a q s k T R
w c f y m	222K 1.00	i h v p l g t n q a s e R D K
w	223G 1.00	c m f h i y p V l n d q R t G e k s A
	224N 1.00	w c m f i h y p v L r q t d N G k e s A
	225A 1.00	w c M f h i y p v l r q N d t k G e s A

R → **C**

Arginine Cysteine

Substitution at pos 221 from R to C is predicted to **AFFECT PROTEIN FUNCTION** with a score of 0.03. Median sequence conservation: 3.05 Sequences represented at this position:7

[1] Ng PC, Henikoff S (2003) SIFT: predicting amino acid changes that affect protein function. *Nucleic Acids Res.* 31:3812-3814.

[2] Sunyaev S, Ramensky V, Koch I, Lathe W 3rd, Kondrashov AS, Bork P (2001) Prediction of deleterious human alleles. *Hum Mol Genet.* 10:591-597.