

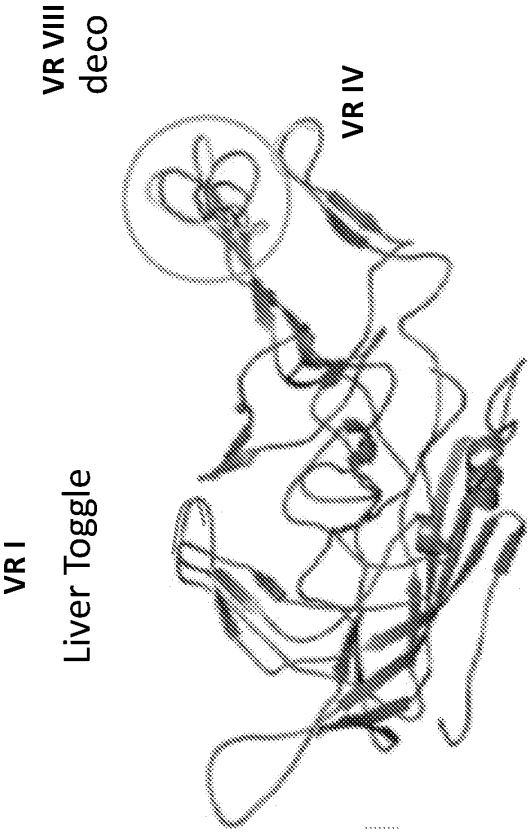


FIG. 1

[illegible]

FIG. 2A

AAV2 (AAC03780)	ORLNNWGFREKRLNFKLENIQVKEVTQNDGTTIANNLTSTVQVFTDSEYQLPYVLGSAHQGLPPFPADVFWVQYGYLTILN--NGSQAVGRSFFYC	394
AAV1 (AAD27757)	T..V.....S.....I.....	395
AAV6 (AAB95450)	T..V.....S.....I.....	395
AAV3 (AAC55049)	K.S.....RG.....	394
AAV LK03	K.S.....	394
AAV7 (AF513851_2)	K.R.....T..V.....I..S.....I.....	396
AAV8 (AF513852_2)	S.....E..K.....I.....	397
AAV hu.37 (AAS99285)	S.....E..K.....I.....	397
AAV zh.10 (AAO88201)	E..K.....I.....	397
AAV9 (AAS99264)	D.N.VK.....D.....E.VK.....I.....-D.....	396
AAV hu.68	D.N.VK.....D.....E.....-D.....	396
AAV10 (AAT46337)	S.....E..K.....I.....	397
AAV5 (AAD13756)	Y.....RS.RV.I.....VQ.S.....DD.....V.NGTE.....A.PQ..TL.....A...RD.TENPTE....F..	387
AAV2 (AAC03780)	LEVFPQMLRTGNFTSYTEDEVPHSSVIAHSOSLDRLMNLPLIDQVLYLSRN--TPSGTTTOSRLQFSQAGASDIRDOSRNWLPGEVYRQVRKTSIA	493
AAV1 (AAD27757)	E.....N..Q-NQ..SAQNKD.L..RGSPAGMSV.PK.....	494
AAV6 (AAB95450)	N..Q-NQ..SAQNKD.L..RGSPAGMSV.PK.....	494
AAV3 (AAC55049)	Q.....Q.....N..Q-NQ..SAQNKD.L..RGSPAGMSV.PK.....	494
AAV LK03	Q.....Q.....N..Q-NQ..SAQNKD.L..RGSPAGMSV.PK.....	494
AAV7 (AF513851_2)	E..S.....A..QSN.G..AGNEP...Y.G.P.TWAE.AK.....F.....LD	496
AAV8 (AF513852_2)	Q.T.....Q..TG..ANTQT.G..G.PNTWAN.AK.....T.TG	496
AAV hu.37 (AAS99285)	E.....Q..STG..QGTQQ.L.....PANKSA.AK.....T.LS	496
AAV zh.10 (AAO88201)	E..Q.....Q..STG..QGTQQ.L.....PNNKSA.AK.....T.LS	496
AAV9 (AAS99264)	Q.....E..N.....K.I-NG..-QN.QT.K.V..P.NNAV.G..YI...S.....T.VI	494
AAV hu.68	Q.....E..N.....K.I-NG..-QN.QT.K.V..P.NNAV.G..YI...S.....T.VI	494
AAV10 (AAT46337)	E.....Q..STG..QGTQQ.L.....PANKSA.AK.....T.LS	496
AAV5 (AAD13756)	K.....E.T.N.E.....F.P.N.FK.A..V.....RFVS..-NTG..-V..NKSL.GRYANTYK..F...MG.T.GWNLG.G	480
AAV2 (AAC03780)	DNNSEYWTGATKYHLNGRDSLVPNGPAMASHKDDDEKFTFQSGVLITGKSGEKTNDI---EKWITDBEEIRTNVATEQYGSVSTNLQKGRQA	590
AAV1 (AAD27757)	NFT.....S..N...E.II...T.....D..M...M...ESAGAS.TAL---DN.....KA.....RF.T.AV.F.SSSTDP	591
AAV6 (AAB95450)	NFT.....S..N...E.II...T.....D..M...M...ESAGAS.TAL---DN.....KA.....RF.T.AV...SSSTDP	591
AAV3 (AAC55049)	NFP..A.S.....MH.N...E.ITAS.AEL---DN.....T.AN..SS.TAP	591
AAV LK03	NFP..A.S.....MH.N...E.ITAS.AEL---DN.....T.AN..SS.TAP	591
AAV7 (AF513851_2)	NFA.....N.....V..T.....DR..S.....T.A-TNKITL---N.LM.N...P.....E..I..S...AA.TA.	592
AAV8 (AF513852_2)	NFA.....N.A..I..T.....R..SN.I.....NAARD.A.Y---SD.L.S..K.....E..I..AD..QO.TAP	593
AAV hu.37 (AAS99285)	NFA.....V..T.....R..S..M.....AGRD..Y---SS.L.S..K.....V.AD..QT.TGP	593
AAV zh.10 (AAO88201)	NFA.....V..T.....R..S..M.....AG.D..Y---SS.L.S..K.....V.AD..QO.PAP	593
AAV9 (AAS99264)	FA.P..SSWA...N..M.....EG.DR..L.S.....TGRD..A---D.....N..K.....S..Q.A..H.SACA...	591
AAV hu.68	FA.P..SSWA...N..M.....EG.DR..L.S.....TGRD..A---D.....N..K.....S..Q.A..H.SACA...	591
AAV10 (AAT46337)	NFA.....V..T.....R..S..M.....AGRD..Y---SS.L.S..K.....V.AD..QO.TGP	593
AAV5 (AAD13756)	V.PA.VSAFATTNME.E.ASVQ.P.Q.NGMTNNLQGSNTVALENTM..NS.PANPCTTATYLEGNWL..S.S.TQPV.R..YNYVG.QMA..N.ESSITAP	580

FIG. 2B

AAV2 (AAC03780)	ATADVNTQGVLFQWVWQDRDVTYLOQPIWAKIEHTDGHFHSPLMGGRGLKHPPPOILLIKNTVPANPSTTIFSAAKFASFIITQYSTQVSVIEIWELOKEN	690
AAV1 (AAD27757)	..G..HAM.A.....N.....PAE...T.....	691
AAV6 (AAB95450)	..G..HVM.A.....PAE...T.....	691
AAV3 (AAC5049)	T.GT..H..A.....M.....P...P.....	691
AAV LK03	T.RT..D..A.....M.....P...P.....	691
AAV7 (AF513851_2)	Q.QV..N..A.....N.....FEV.TP.....	692
AAV8 (AF513852_2)	QIGT..S..A.....N.....D.P...NQS.LN.....	693
AAV hu.37 (AAS99285)	IVGN..S..A.....N.....D.P...Q...L.....	693
AAV zh.10 (AA088201)	IVGA..S..A.....N.....D.P...Q...L.....	693
AAV9 (AAS99264)	Q.GW.QN..I.....M.....D.P.A.NKD.LN.....	691
AAV hu.68	Q.GW.QN..I.....M.....D.P.A.NKD.LN.....	691
AAV10 (AAT46337)	IVGN..S..A.....N.....D.P...Q...L.....	693
AAV5 (AAD13756)	..GTY..L.EIV..S..ME.....E.GA.....A.....MM.....G..I..S...DVPVS.....T..M...K...	679
AAV2 (AAC03780)	SKRNWPEIQYTSNKNKSVNVDTVDINGVISEPRPIGTRYLTRLN-	735
AAV1 (AAD27757)	V.....A..A.....N..L.T.....P.-	736
AAV6 (AAB95450)	V.....A..A.....N..L.T.....P.-	736
AAV3 (AAC5049)	P.....	736
AAV LK03	P.*	737
AAV7 (AF513851_2)	FE.QTG..A..SQ.....	737
AAV8 (AF513852_2)	Y..TS...A.N.E.....	738
AAV hu.37 (AAS99285)	Y..T...A.N.E.T.....	738
AAV zh.10 (AA088201)	Y..T...A.N.D.T.....	738
AAV9 (AAS99264)	Y..N..E.A.N.E.....	736
AAV hu.68	Y..N..E.A.N.E.....*	737
AAV10 (AAT46337)	Y..T...A.N.E.T.....	738
AAV5 (AAD13756)	N...DPQF...AP..ST.E.RTT.....P.-	724

FIG. 2C

AAV2 (AAC03780)	MADGVLPDWLBDTISBGIQWKLKPGPPPKPAERKDDSGVLVPGHYLGPFGNLKGBFVNEDAAALEHDKAYDQLDSSGDPYLKYNHADADEF	100
Anc80	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc80L1	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc80L44	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc80L27	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc80L36	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc80L33	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc80L59	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc80L60	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc80L62	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc80L65 (AKU89595)	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc81 (AKU89596)	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc82 (AKU89597)	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc82DI	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc83 (AKU89598)	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc84 (AKU89599)	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc94	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc110 (AKU89600)	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc113 (AKU89601)	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc126 (AKU89602)	N.....E.D...A.K.ANQKQ.G.....A.....Q.KA.....R.....	100
Anc127 (AKU89603)	N.....E.D...A.Q.ANQQ.Q.G.....Q.KA.....	100
168		
AAV2 (AAC03780)	QBELKEDTSFGNLRGAVFOAKKRVLBFLGLVBEFVKTAFCRKRVVHSFY-BFDSSSGTGKAGQOPARKLNFGOTGHADSVDFPQPLGQFPAPSGLG	199
Anc80	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc80L1	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc80L44	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc80L27	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc80L36	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc80L33	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc80L59	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc80L60	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc80L62	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc80L65 (AKU89595)	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc81 (AKU89596)	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc82 (AKU89597)	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc82DI	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc83 (AKU89598)	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc84 (AKU89599)	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc94	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc110 (AKU89600)	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc113 (AKU89601)	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc126 (AKU89602)	Q.....GA.....Q.Q-.....I.K...K.....SE.....E.....V.	199
Anc127 (AKU89603)	Q.....AA.....Q.Q-.....I.S...S.....SE.....E.....V.	199

FIG. 3A

AAV2 (AAC03780)	FFSQMLTGNNTFTSYFEDVPFHSSVAHSQSLDLNMLPLDQYLYLSRTN-TPSGTTQSRQLQFSQAGSDIRQSNRWLPQPCYQQRVSKTSADNN	496
Anc80	Q.....Q-T..AGNRE.....P.SWAN.AK.....TNQ..	497
Anc80L1	E.....Q-T..AGNRT.....P.SWAN.AK.....ANO..	497
Anc80L44	Q.....Q-T..AGNRE.....P.SWAN.AK.....TNQ..	497
Anc80L27	E.....Q-T..AGNRT.....P.SWAN.AK.....ANO..	497
Anc80L36	E.....Q-T..AGNRT.....P.SWAN.AK.....ANO..	497
Anc80L33	E.....Q-T..AGNRT.....P.SWAN.AK.....ANO..	497
Anc80L59	Q.....Q-T..AGNRE.....P.SWAN.AK.....TNQ..	497
Anc80L60	E.....Q-T..AGNRE.....P.SWAN.AK.....TNQ..	497
Anc80L62	E.....Q-T..AGNRE.....P.SWAN.AK.....TNQ..	497
Anc80L65 (AKU89595)	Q.....Q-T..AGNRT.....P.SWAN.AK.....TNQ..	497
Anc81 (AKU89596)	E.....Q-TG..AGNRT.....P.SWAN.AK.....TNQ..	498
Anc82 (AKU89597)	Q.....Q-TG..AGTQT.....P.SWAN.AK.....T.TNQ..	499
Anc82DI	Q.....Q-TG..AGTQT.....P.SWAN.A..V.....T.TNQ..	499
Anc83 (AKU89598)	Q.....Q-TG..AGTQT.....P.SWAN.AK.....T.TSQ..	499
Anc84 (AKU89599)	E.....Q-STG..AGTQQ.L.....P.NWSA.AK.....T.LSQ..	499
Anc94	E.....Q-STG..AGTQQ.L.....P.NWSA.AK.....T.LSQ..	499
Anc110 (AKU89600)	Q.....Q-T..AGTQT.....P.SWAN.AK.....T.TNQ..	497
Anc113 (AKU89601)	E.....A.OS.TG..AGNRE..Y..P.TWAE.AK.....LDQ..	499
Anc126 (AKU89602)	E.....Q-T..AGNRE.....P.SWAN.AK.....AN..	496
Anc127 (AKU89603)	Q.....Q-T..Q.....P.SNAQ.AK.....AN..	496
AAV2 (AAC03780)	NSEYSWTGATKVHLNGRDSLVPGEAMASHKDEBKFFPSGYLIFGKQSEKTNVDIEKVMITDBEEIRTNFVATEQYGSVSTNLFGFRQAATADVN	596
Anc80	..NFA.....T...D...M.....AGNS...LDN...S...K.....E..T.A...SS.IAP..GT..	597
Anc80L1	..NFA.....T...D...M.....AGNS...LDN...S...K.....T.A...SS.IAP..GT..	597
Anc80L44	..NFA.....T...D...M.....AGNS...LDN...N...K.....T.A...SA.IAP..GT..	597
Anc80L27	..NFA.....T...D...M.....AGNS...LDN...N...K.....T.A...SA.IAP..GT..	597
Anc80L36	..NFA.....T...D...M.....AGNS...LDN...S...K.....E..T.A...SS.IAP..GT..	597
Anc80L33	..NFA.....T...D...M.....AGNS...LDN...S...K.....T.A...SS.IAP..GT..	597
Anc80L59	..NFA.....T...D...M.....AGNS...LDN...N...K.....E..T.A...SA.IAP..GT..	597
Anc80L60	..NFA.....T...D...M.....AGNS...LDN...S...K.....E..T.A...SS.IAP..GT..	597
Anc80L62	..NFA.....T...D...M.....AGNS...LDN...S...K.....E..T.A...SA.IAP..GT..	597
Anc80L65 (AKU89595)	..NFA.....T...D...M.....AGNS...LDN...N...K.....E..T.A...SA.IAP..GT..	597
Anc81 (AKU89596)	..NFA.....V..T...DR..S.....AGND..LDN...S...K.....E..V.A...SA.IAPQ.GT..	598
Anc82 (AKU89597)	..NFA.....V..T...DR..S.....AGND..YSN...S...K.....E..V.A...SA.IAPQ.GT..	599
Anc82DI	..NFA.....M..V.....DR..S.....AGND..YSN...S...K.....E..V.A..H.SA.IT.Q.GT.Q	599
Anc83 (AKU89598)	..NFA.....V..T...DR..S..I.....AG.D..YSN..L.S...K.....E..V.AD..QQ.IAPQIGT..	599
Anc84 (AKU89599)	..NFA.....V..T...DR..S..I.M.....AG.D..YSN..L.S...K.....V.AD..QQ.IAPQIGT..	599
Anc94	..NFA.....V..T...R..S.....M.....AG.D..YSS..L.S...K.....V.AD..QQ.IAPQIGT..	599
Anc110 (AKU89600)	..NFA.....M..V.....DR..S.....AGND..YSQ...N...K.....E..A..N.SA.IT.Q.GL.H	597
Anc113 (AKU89601)	..NFA.....N.....V..T...DR..S.....T.AAKMTT-L.N.LM.N...K.....E..V..S...SA.IAPQ.OT..	598
Anc126 (AKU89602)	..NFA.....D...M.....AGAS...LDN...K.....T.A...SS.IAP..GT..	596
Anc127 (AKU89603)	..NFA.....MH.....TGAS...LDN...K.....T.A...SS.IAP..GT..	596

VR VIII site for targeting peptide

FIG. 3C

AAV2 (AAC03780)	TGCVLPGMWQDRDNYVYLGQPIWAKIPHTDGHFPLMGSGFGUKHPDPQILLIKNTFVPANPSTTSSAAKFASTIQTGTGQSVSEIENELQKENSKEWNP	696
Anc80	S..A.....	697
Anc80L1	S..A.....P..P.....	697
Anc80L44	S..A.....N.....	697
Anc80L27	S..A.....P..P.....	697
Anc80L36	S..A.....P..P.....	697
Anc80L33	S..A.....N.....	697
Anc80L59	S..A.....P..P.....	697
Anc80L60	S..A.....N.....	697
Anc80L62	S..A.....E.....	697
Anc80L65 (AKU89595)	S..A.....	697
Anc81 (AKU89596)	S..A.....P..P.....	697
Anc82 (AKU89597)	S..A.....N.....	698
Anc82DI	S..A.....N.....D.P..NQ..LN.....	699
Anc83 (AKU89598)	N.I.....N.....N.....D.P..NQ..LN.....	699
Anc84 (AKU89599)	S..A.....N.....N.....D.P..NQ..LN.....	699
Anc94	S..A.....N.....N.....D.P..NQ..LN.....	699
Anc110 (AKU89600)	S..A.....N.....N.....D.P..Q..L.....	699
Anc113 (AKU89601)	S..A.....N.....N.....D.P..NQ..LN.....	697
Anc126 (AKU89602)	S..A.....PEV..TP.....	698
Anc127 (AKU89603)	S..A.....P..P.....	696
AAV2 (AAC03780)	EIQXTSNKSNVNDFTVDINGVSEPRPIGTRYLTRLN-	735
Anc80	T....A.....*	737
Anc80L1	T....A.....	736
Anc80L44	T....A.....	736
Anc80L27	T....A.....	736
Anc80L36	T....A.....	736
Anc80L33	T....A.....	736
Anc80L59	T....A.....	736
Anc80L60	T....A.....	736
Anc80L62	T....A.....	736
Anc80L65 (AKU89595)	T....A.....	736
Anc81 (AKU89596)	T....A...E.....	737
Anc82 (AKU89597)	Y..T....A.N.E.....	738
Anc82DI	Y..T....A.N.E.....	738
Anc83 (AKU89598)	Y..T....A.N.E.....	738
Anc84 (AKU89599)	Y..T....A.N.E.....	738
Anc94	Y..T....A.N.E..T.....	738
Anc110 (AKU89600)	Y..T....A.N.E.....	736
Anc113 (AKU89601)	D..T....A..SE.....	737
Anc126 (AKU89602)	A.....	735
Anc127 (AKU89603)		735

FIG. 3D

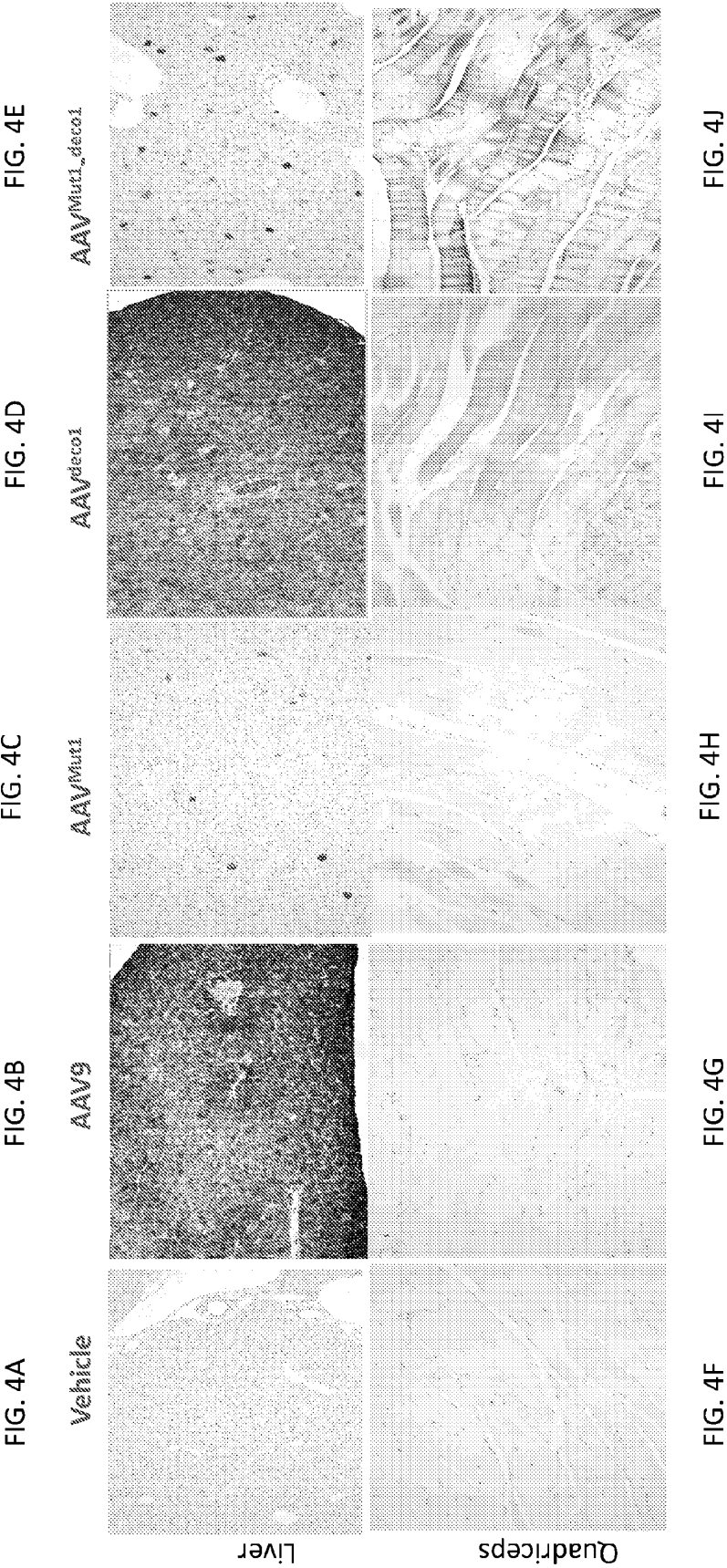


FIG. 4A

FIG. 4B

FIG. 4C

FIG. 4D

FIG. 4E

FIG. 4F

FIG. 4G

FIG. 4H

FIG. 4I

FIG. 4J

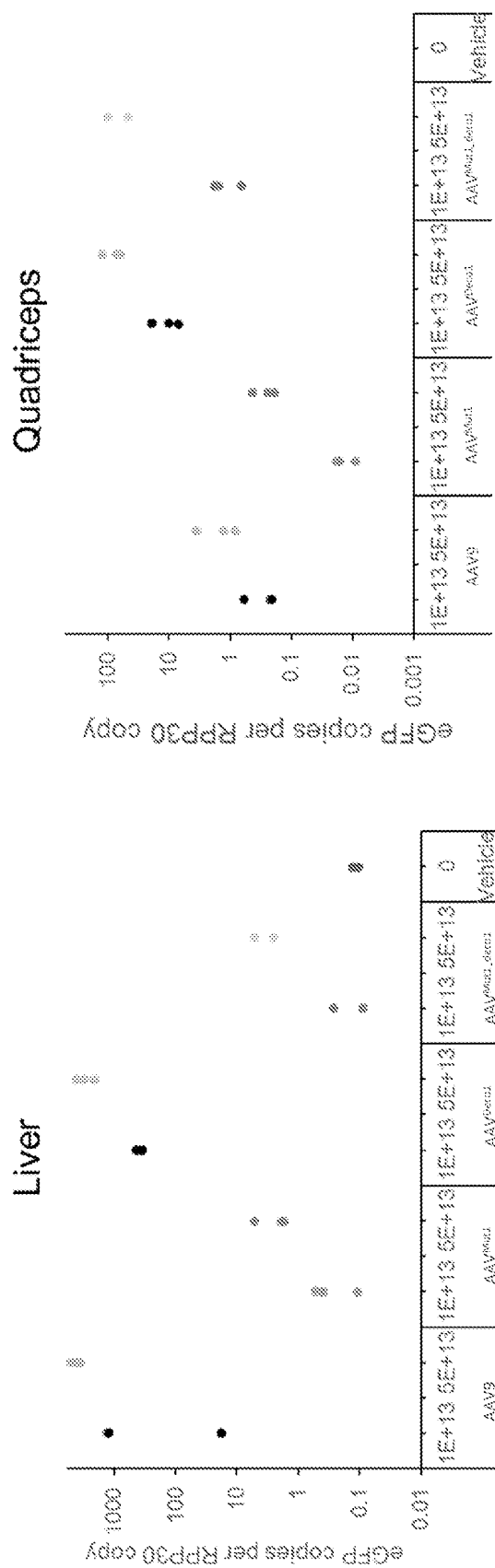


FIG. 5A

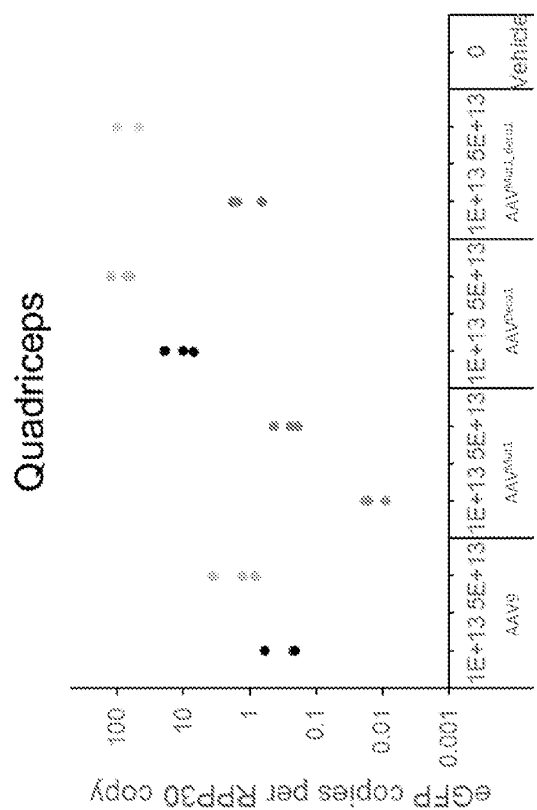
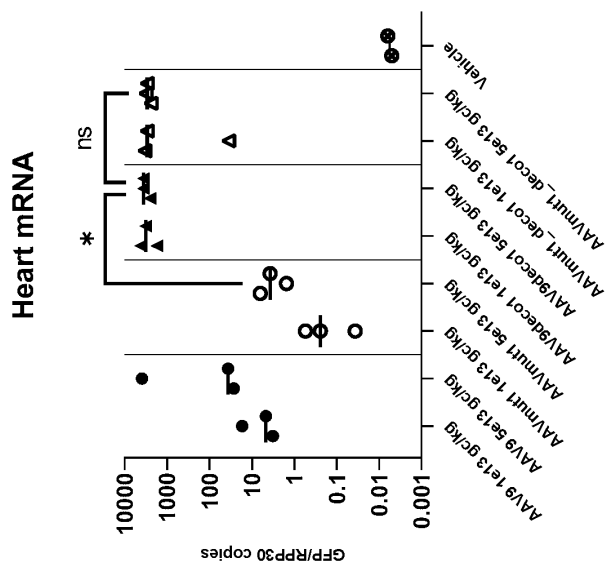
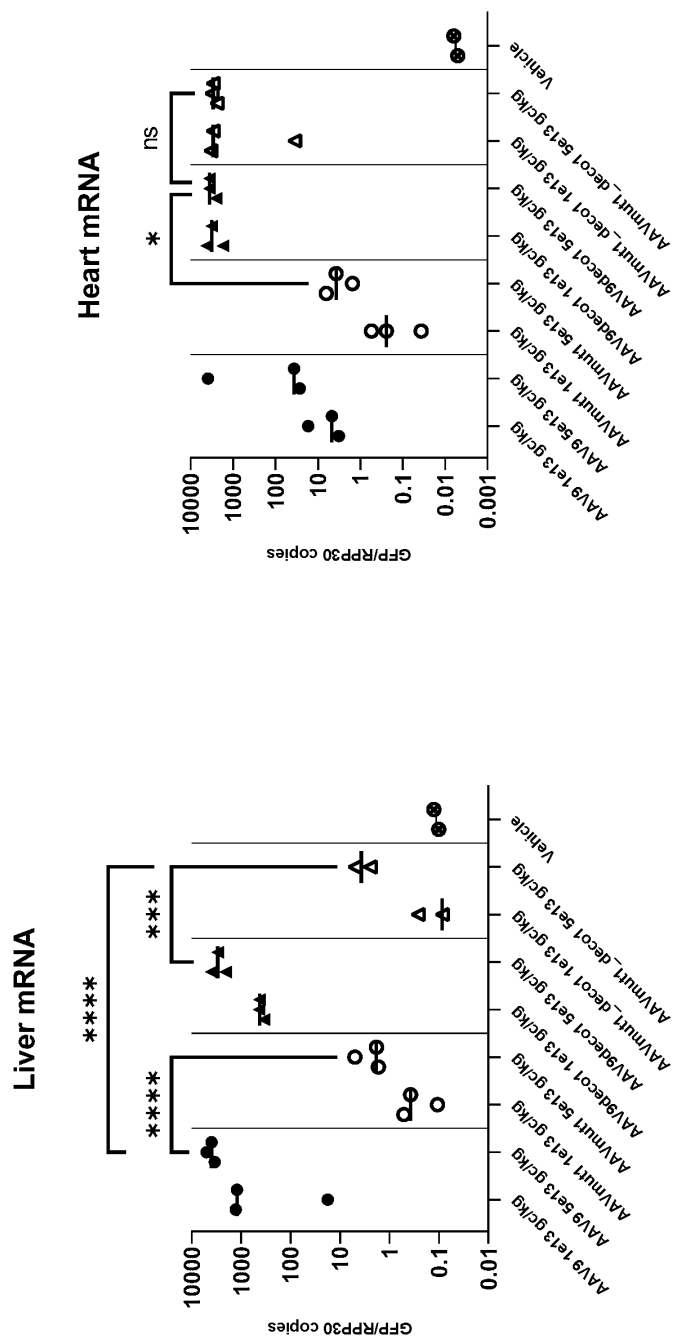
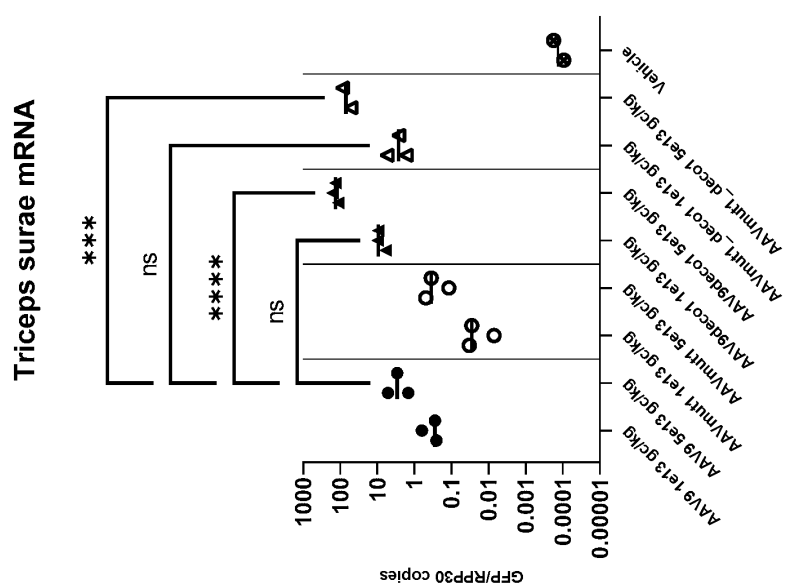
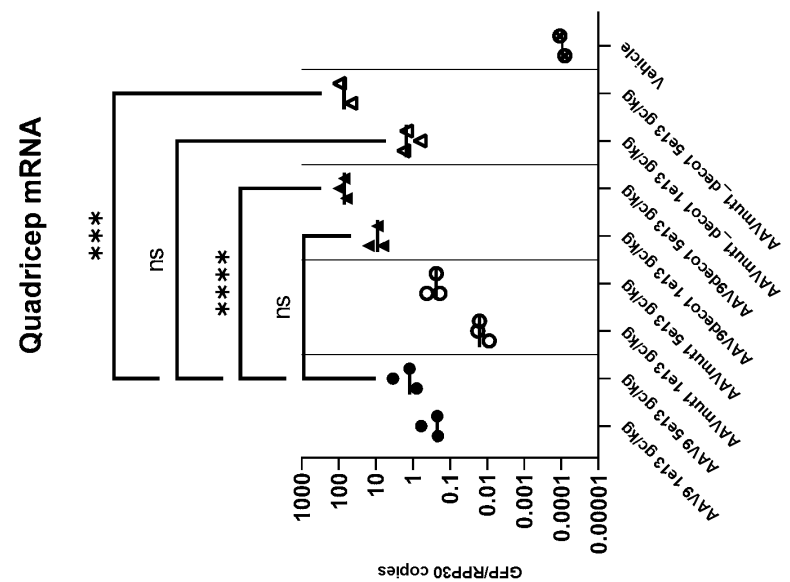


FIG. 5B





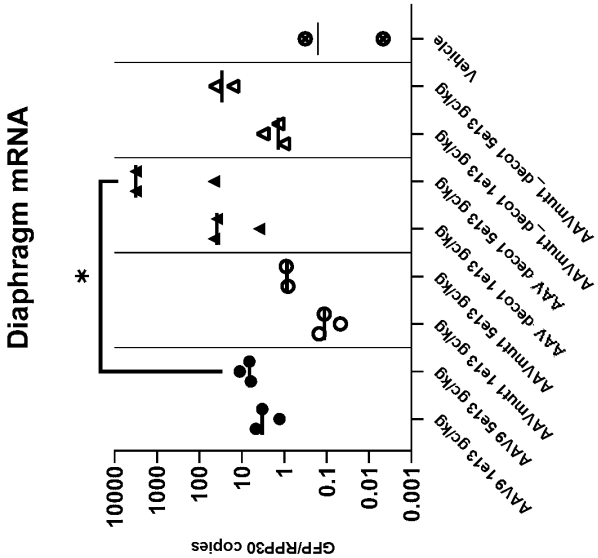


FIG. 6E

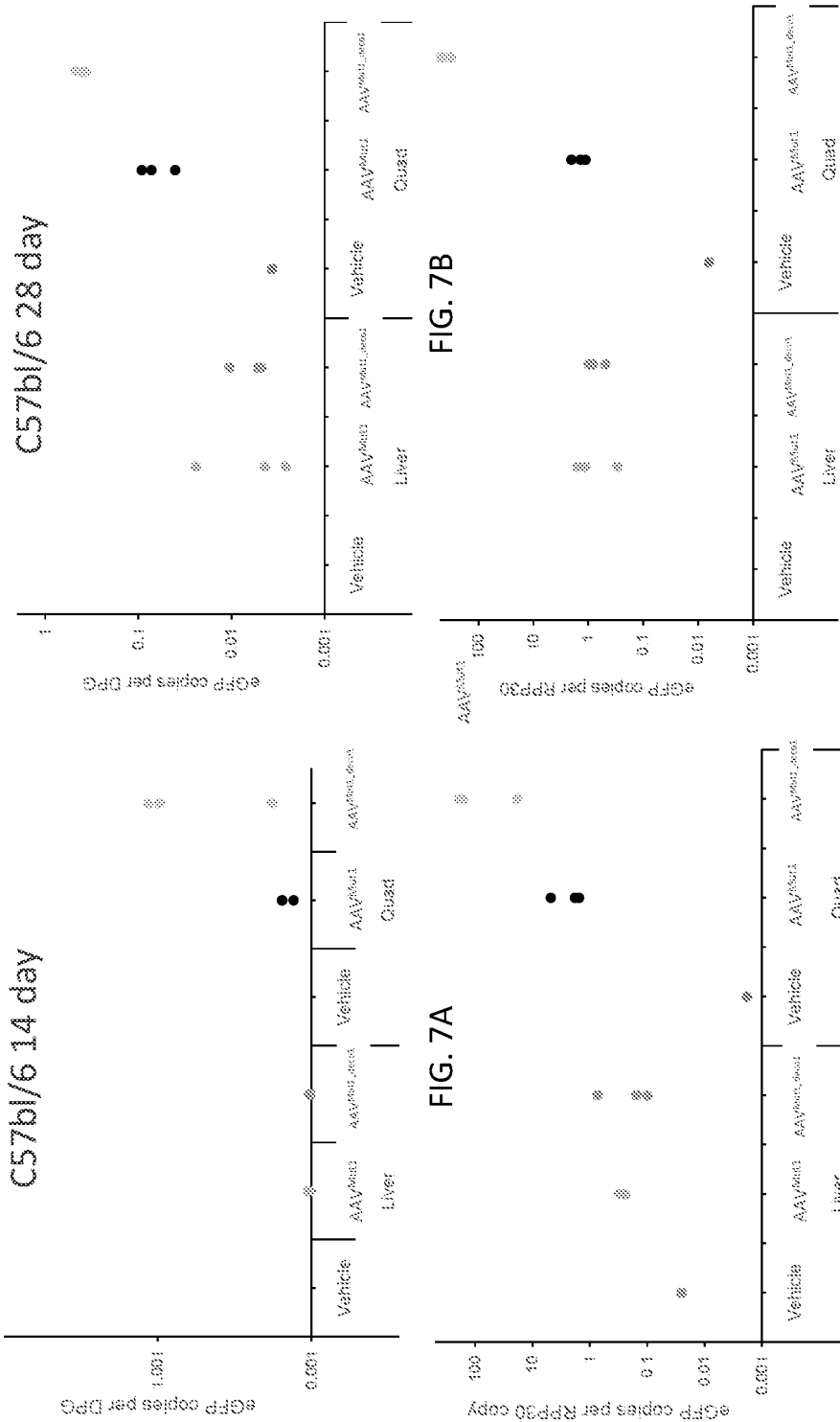


FIG. 7A

FIG. 7B

FIG. 7C

FIG. 7D

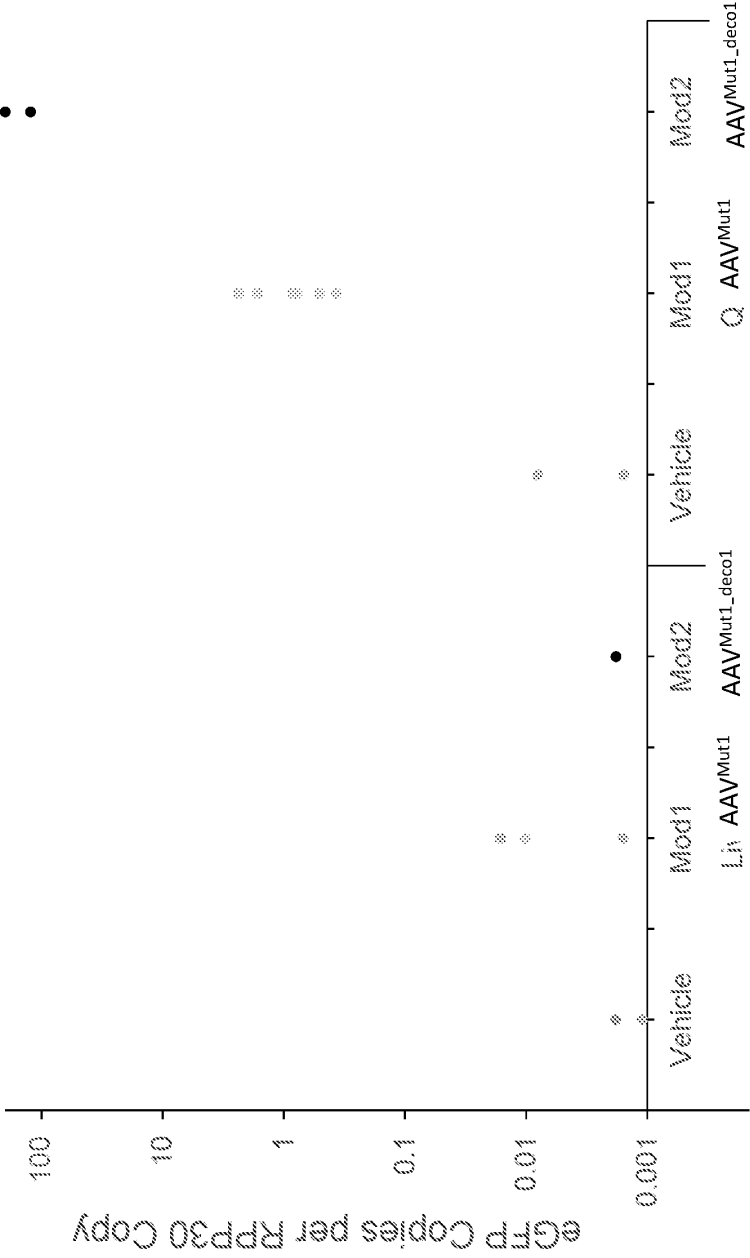


FIG. 8

Quadriceps

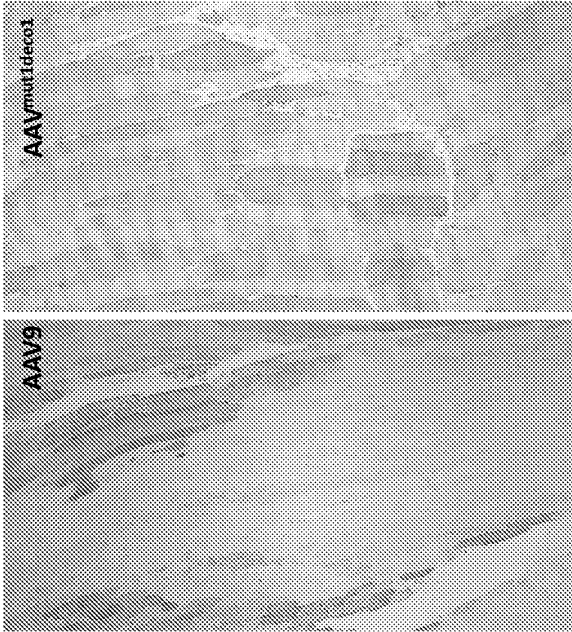


FIG. 9B

Liver

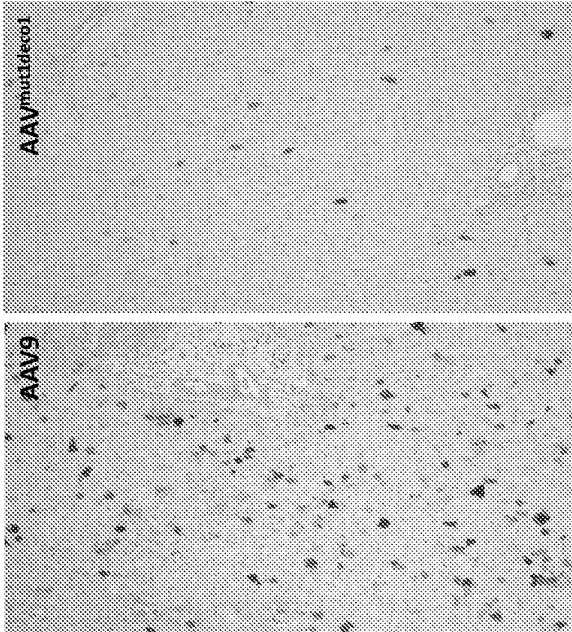


FIG. 9A

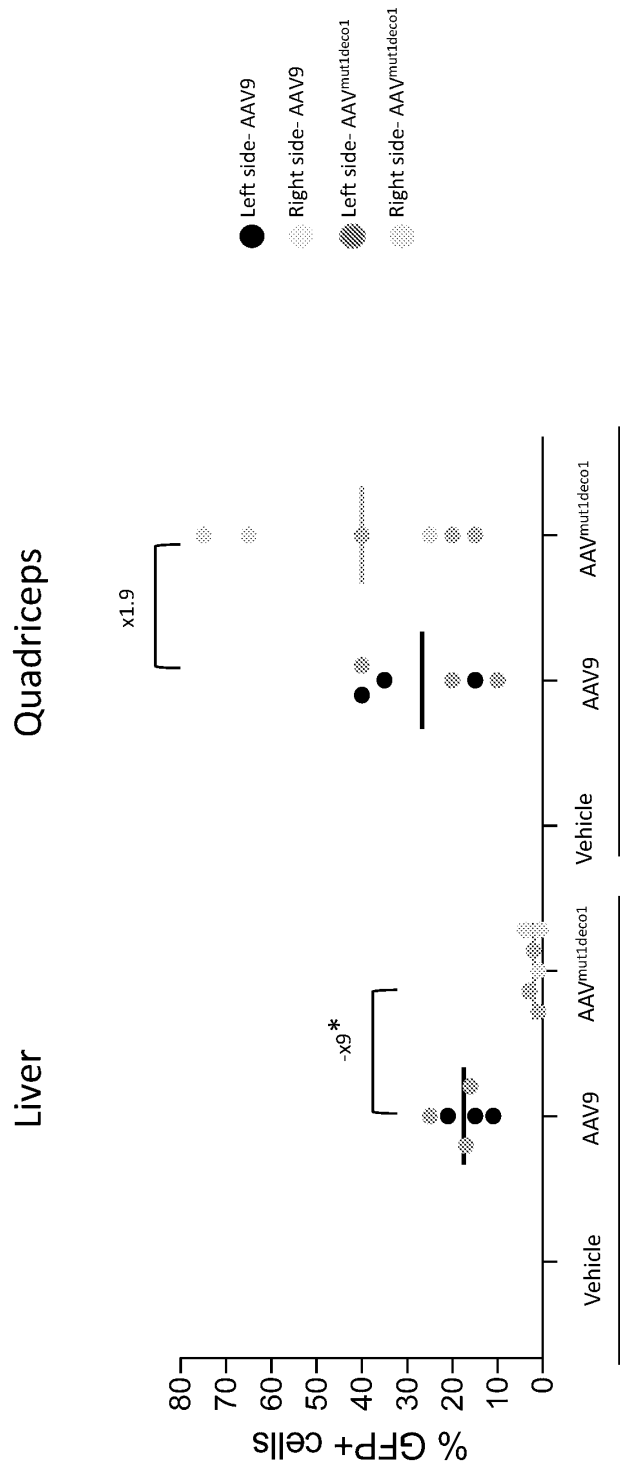


FIG. 10

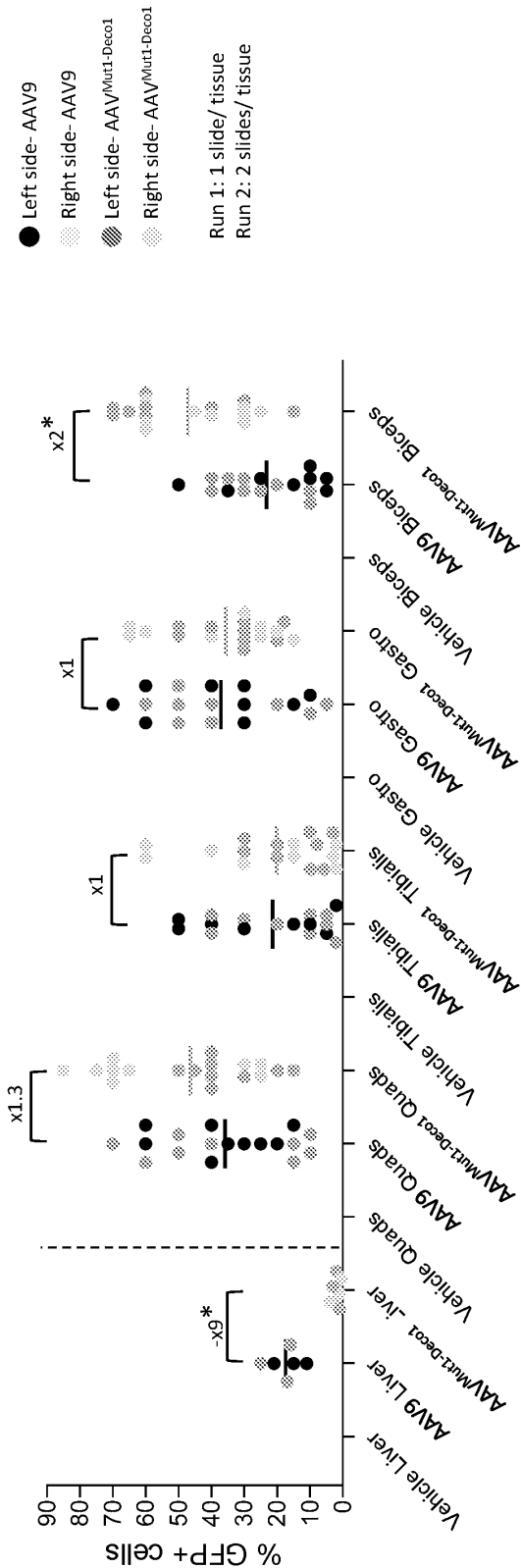


FIG. 11

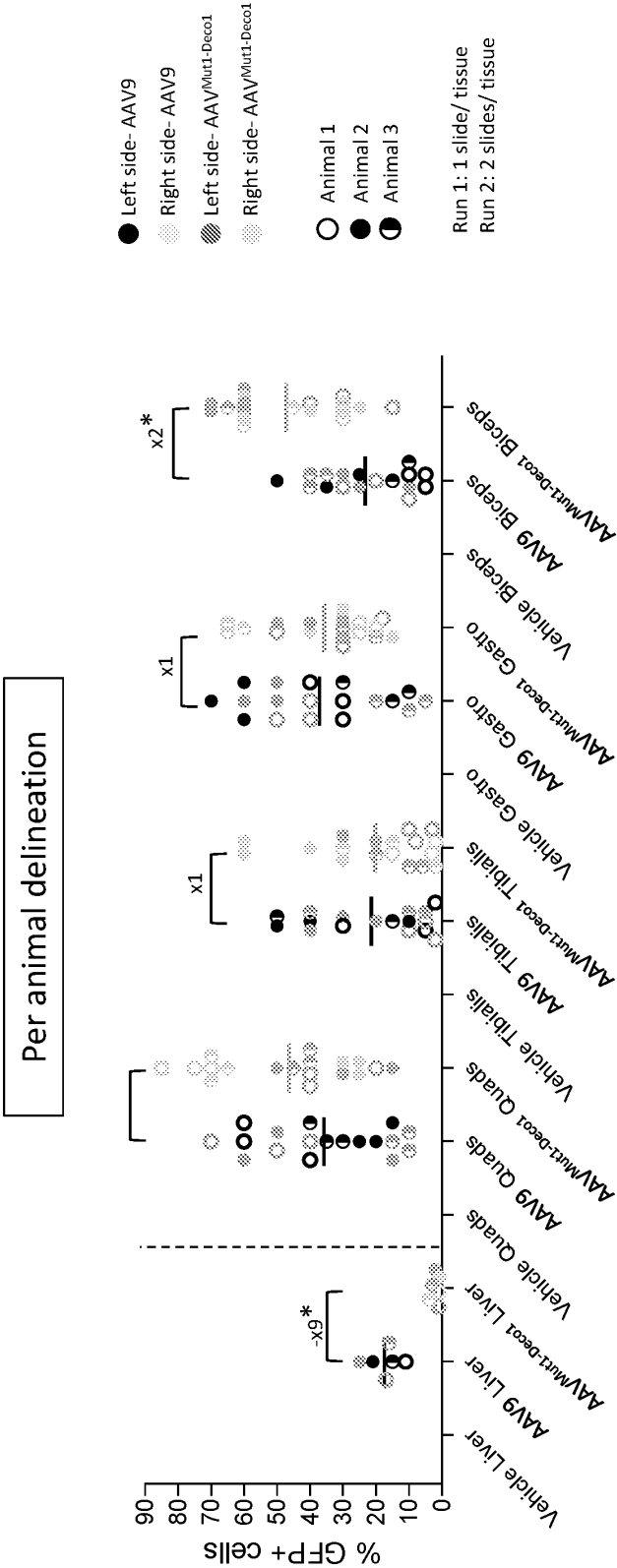


FIG. 12

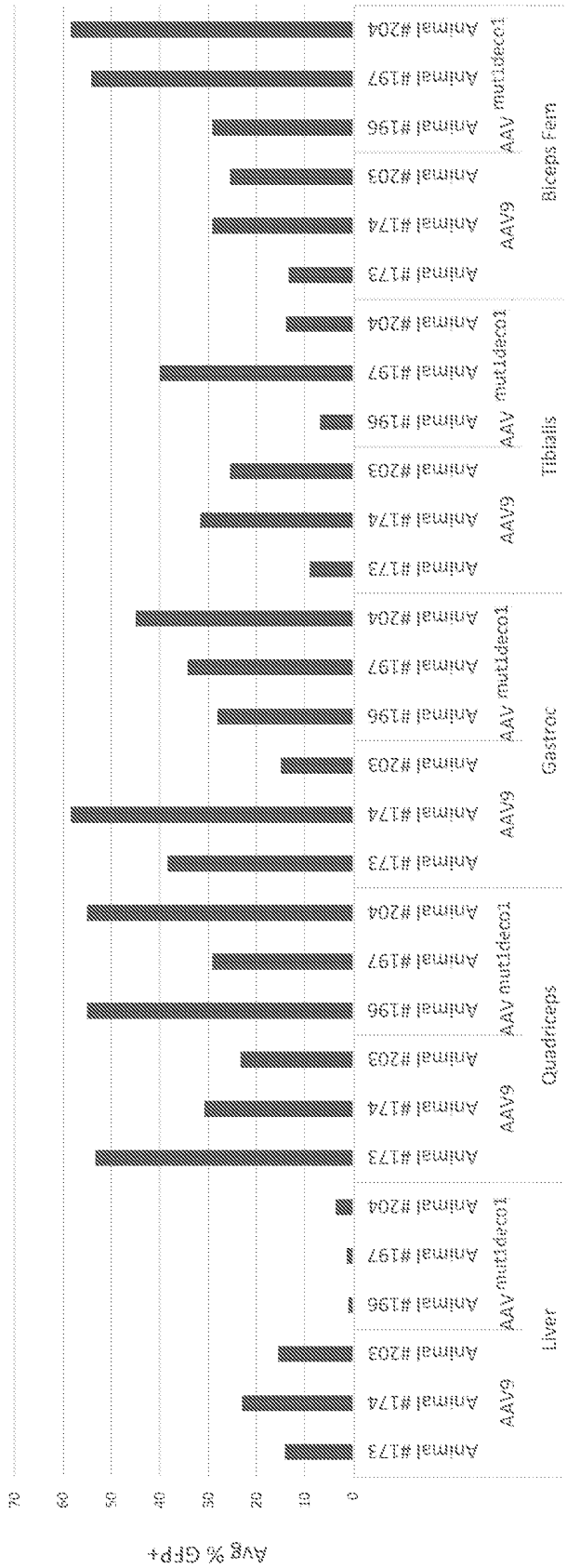


FIG. 13

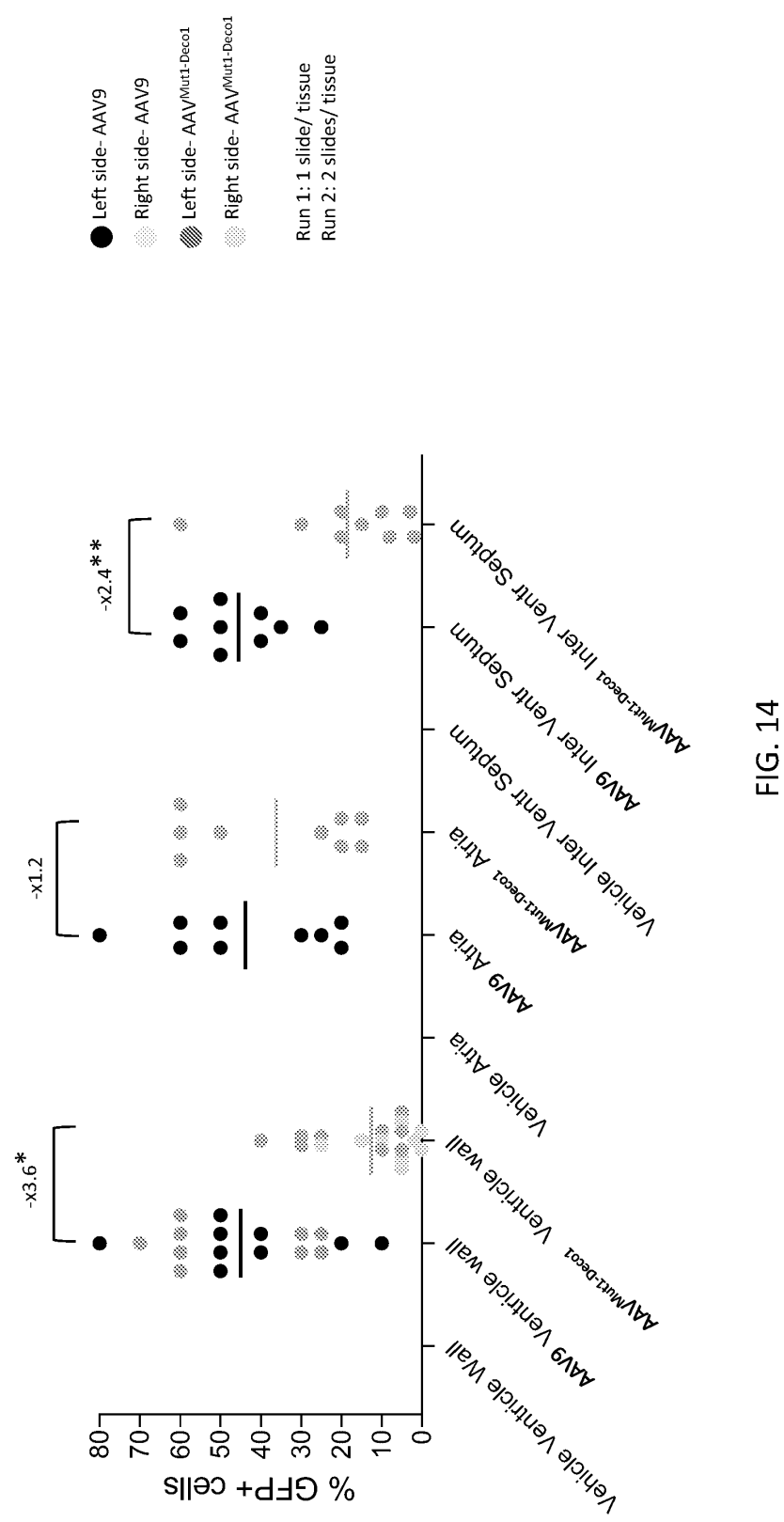


FIG. 14

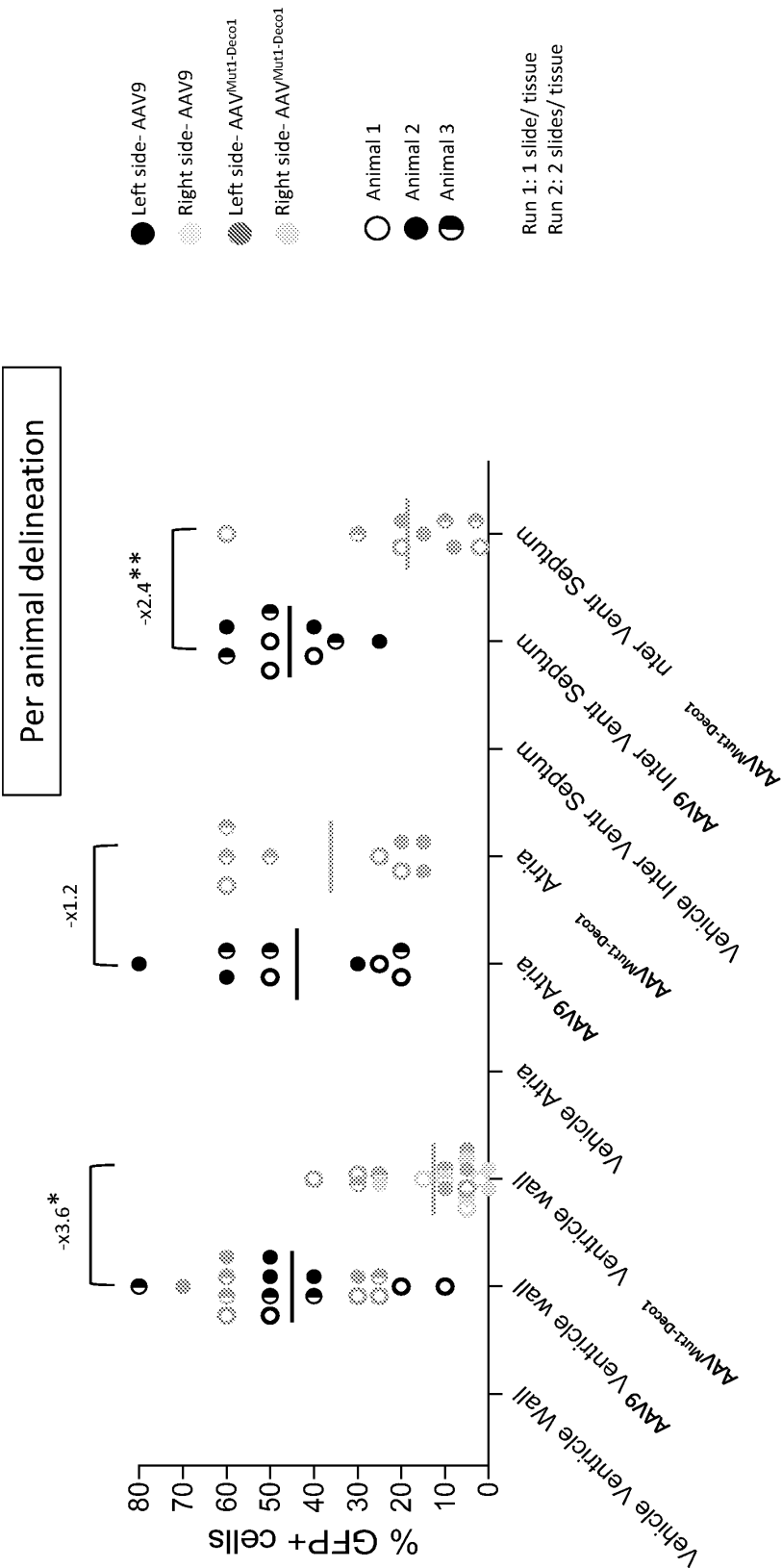


FIG. 15

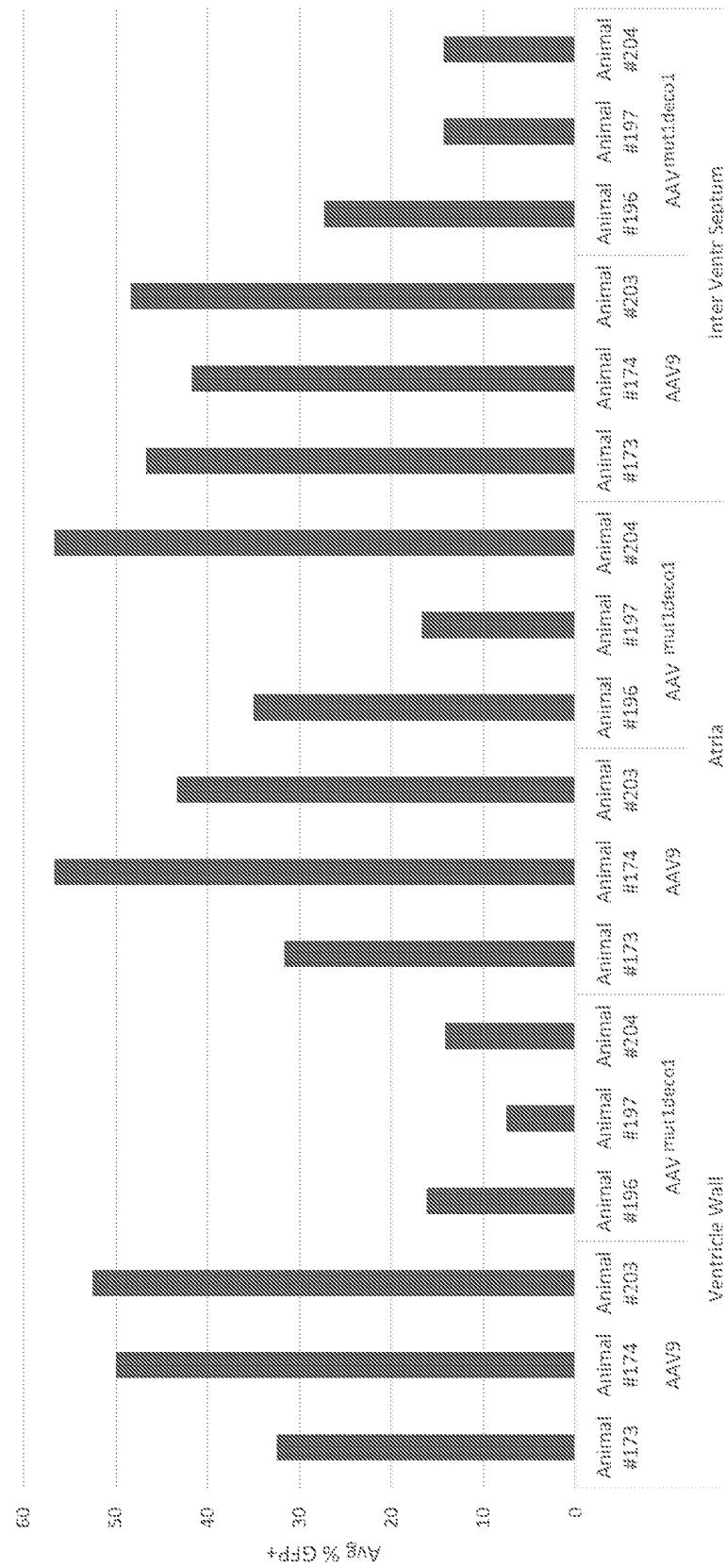


FIG. 16

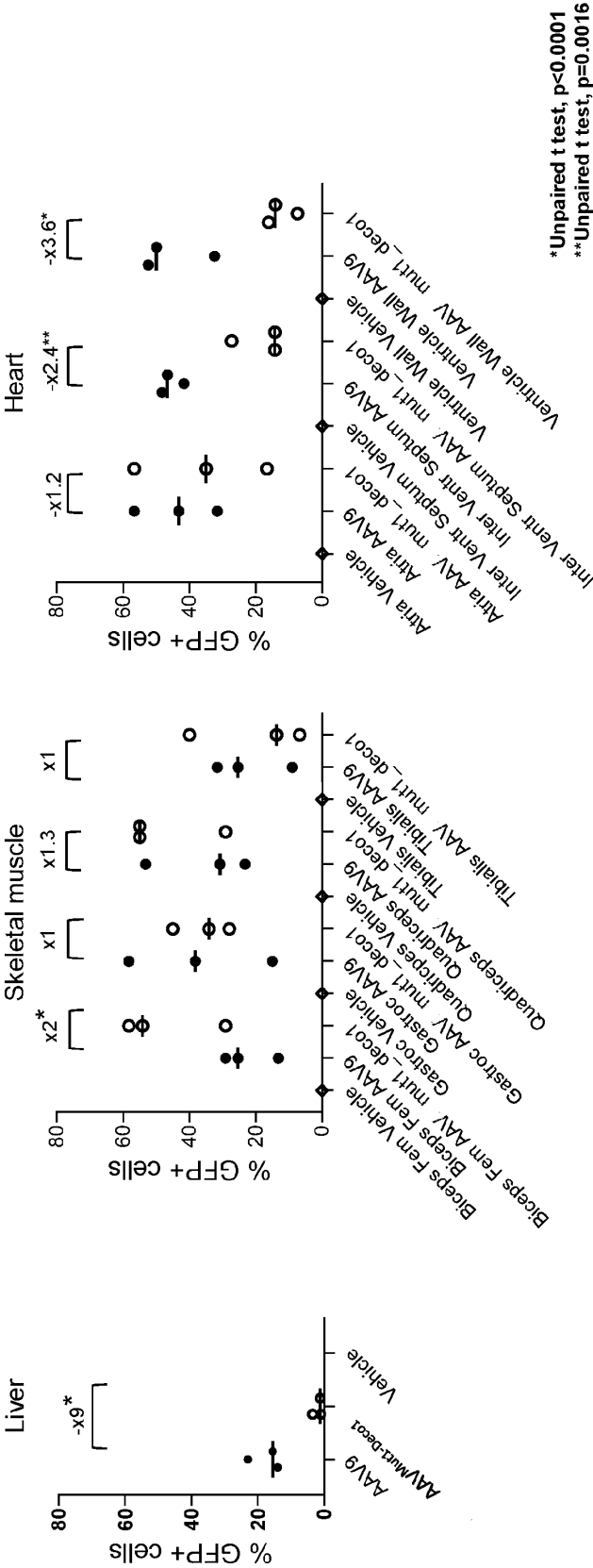


FIG. 17A

FIG. 17B

FIG. 17C

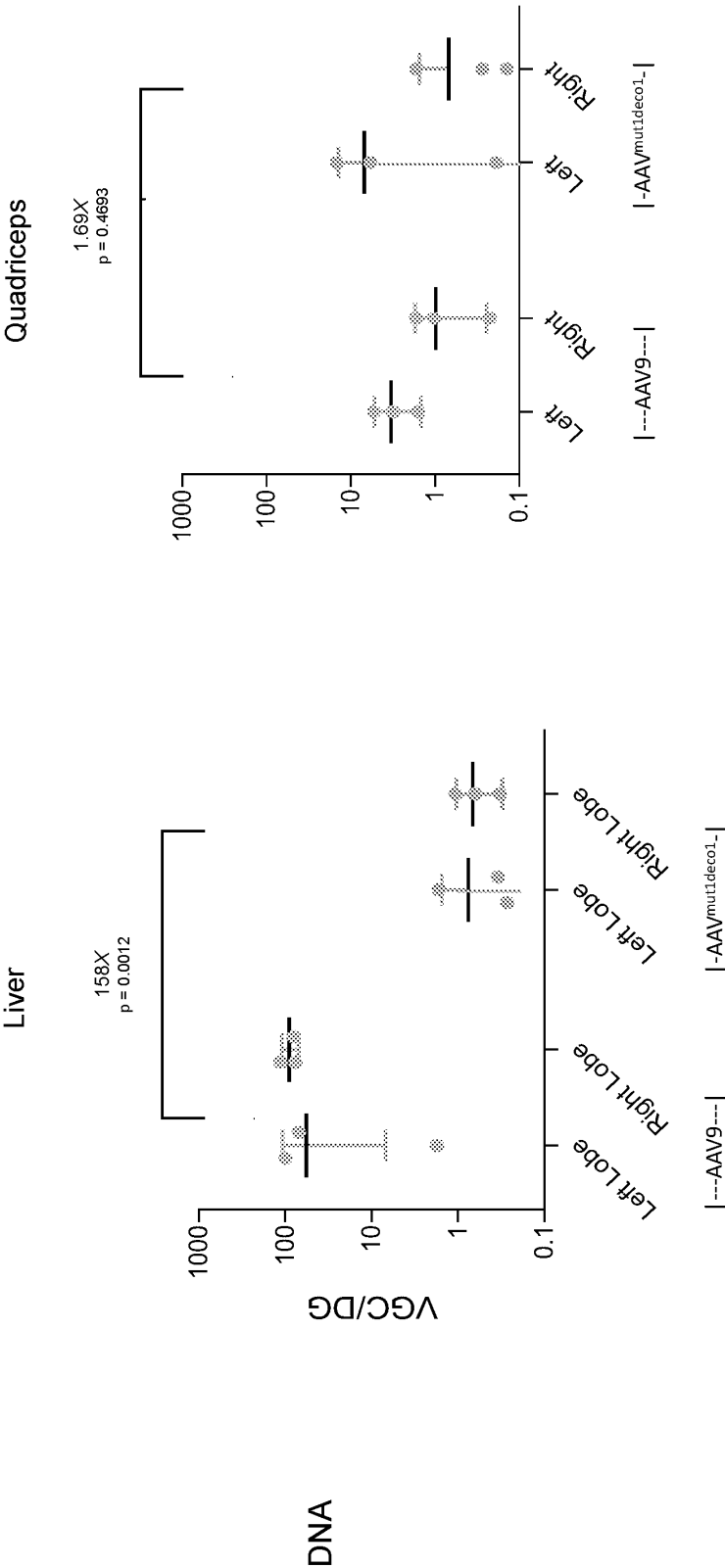


FIG. 18B

FIG. 18A

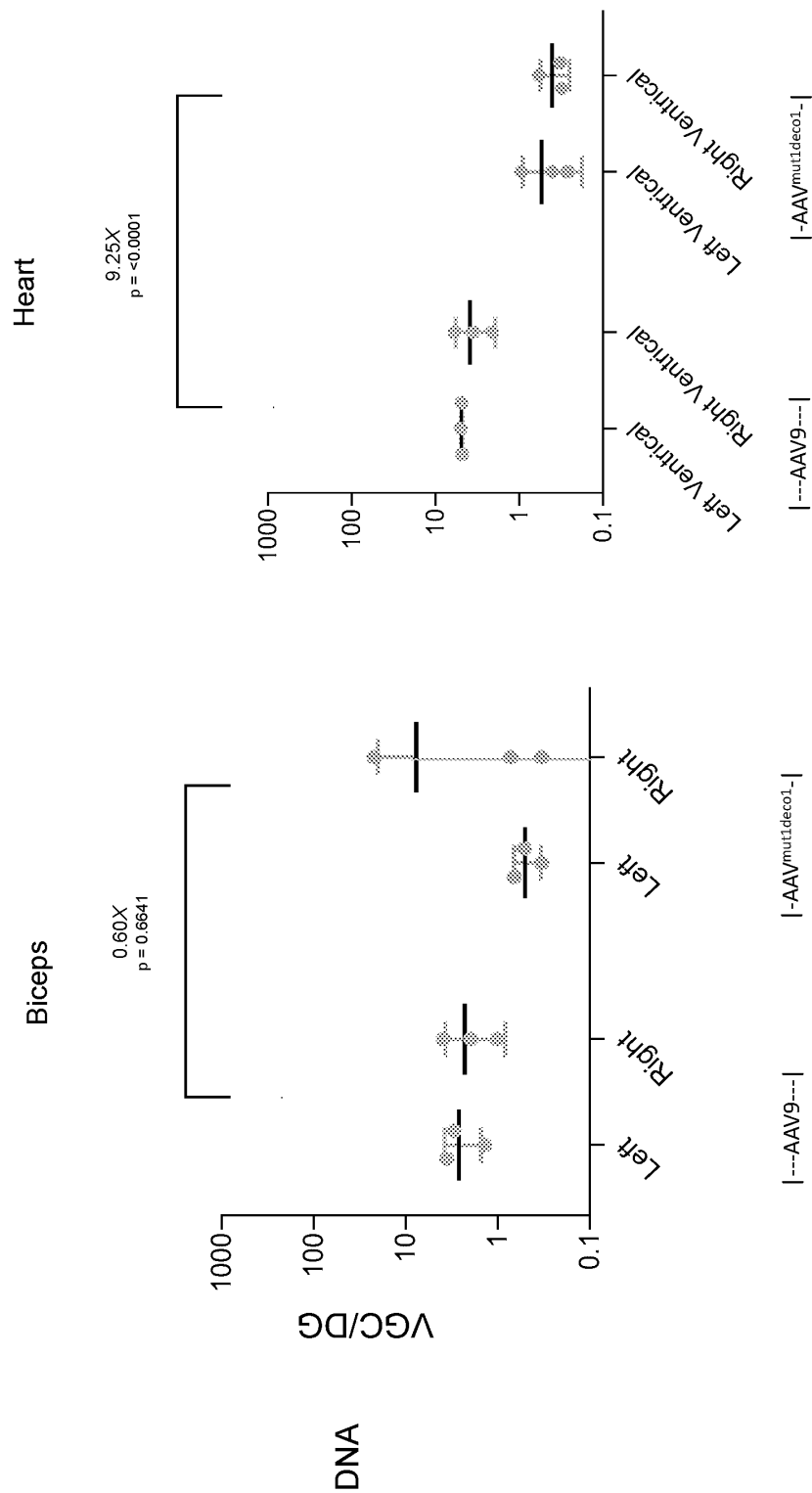


FIG. 18C

FIG. 18D

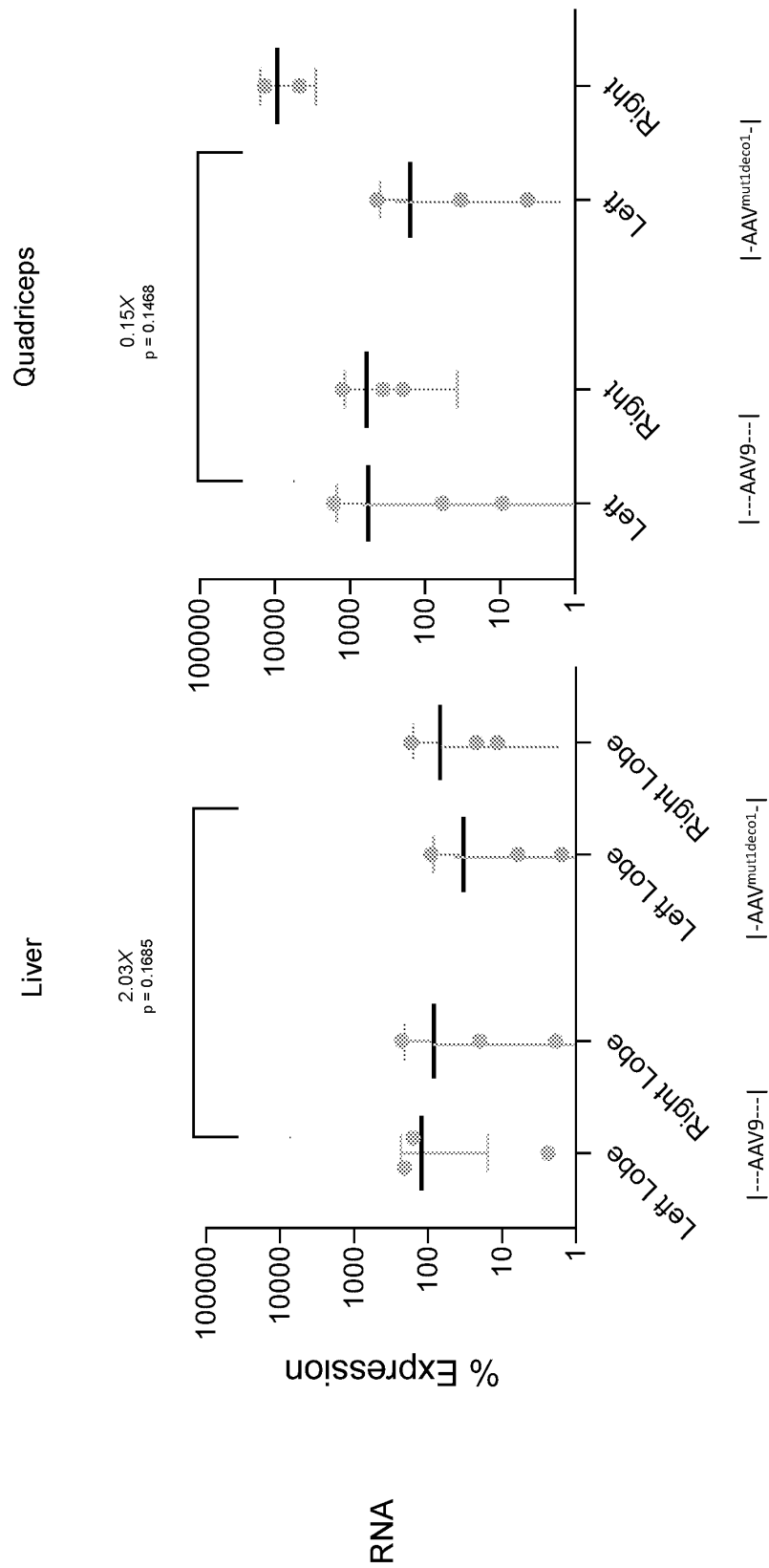
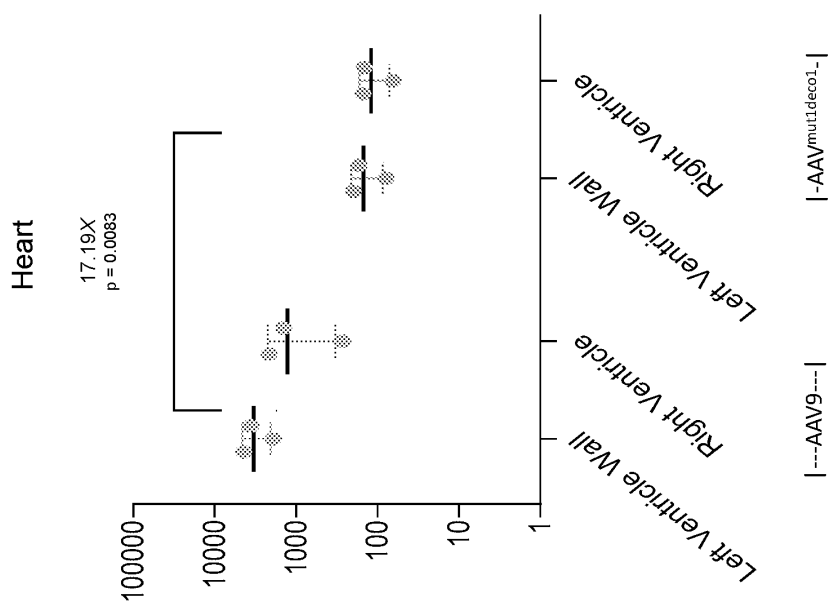
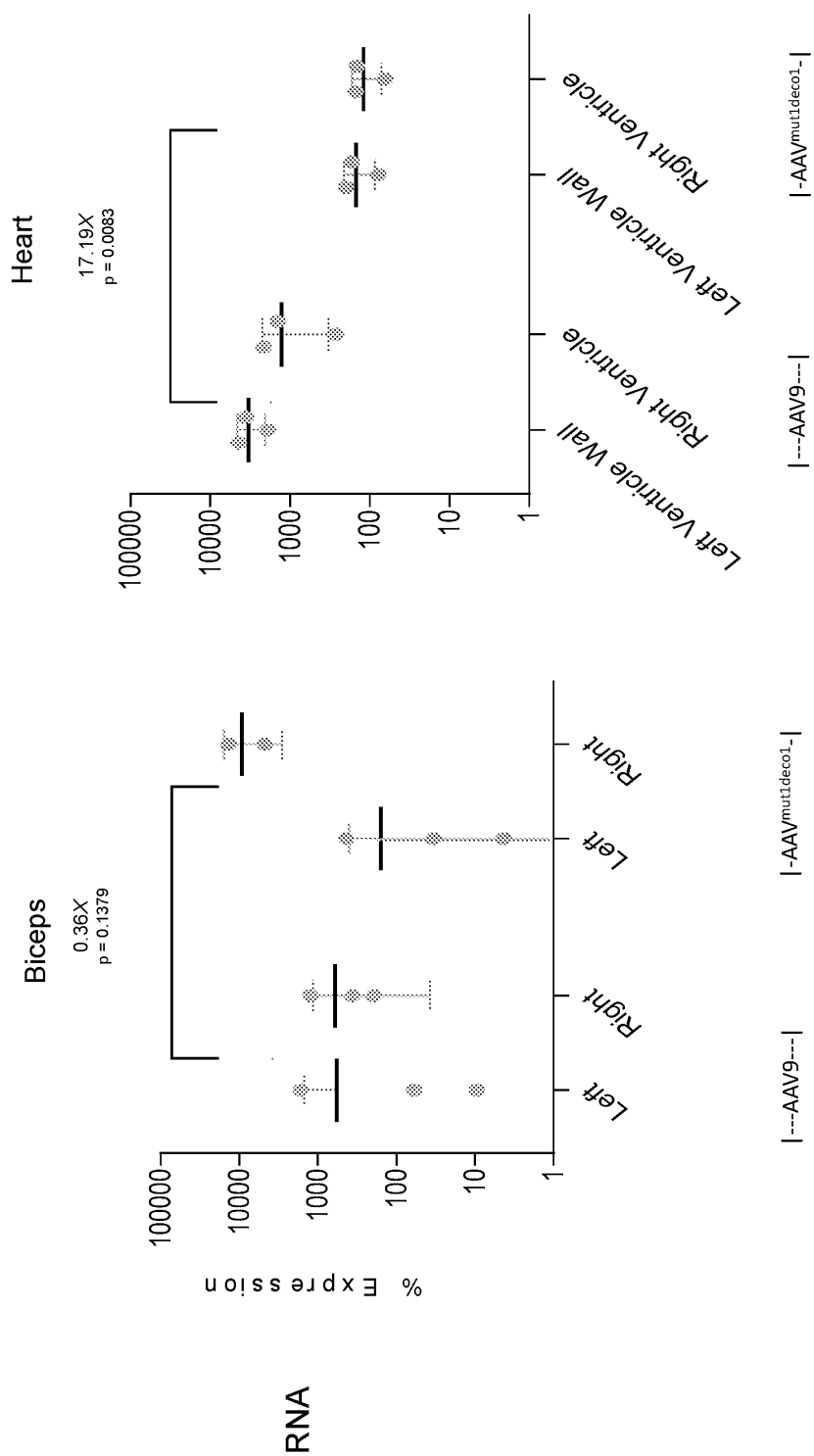


FIG. 19A

FIG. 19B



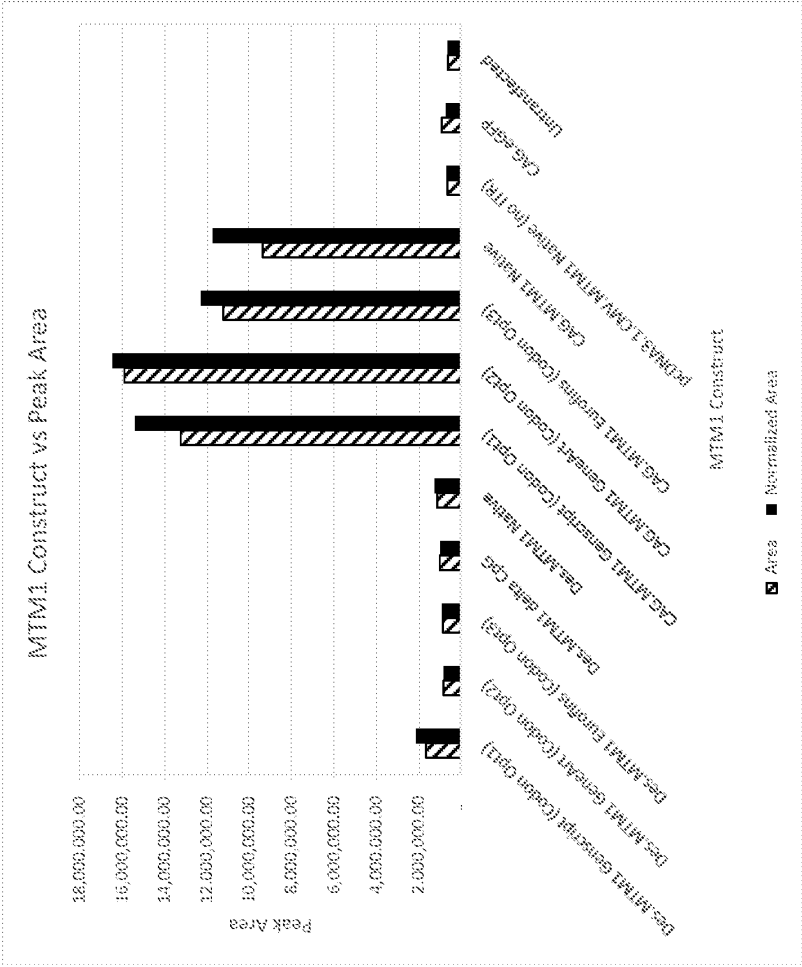


FIG. 20

MSP2 STUDY DESIGN

Purpose of Study: To access the Anc80 Library distribution in Non-Human Primates.

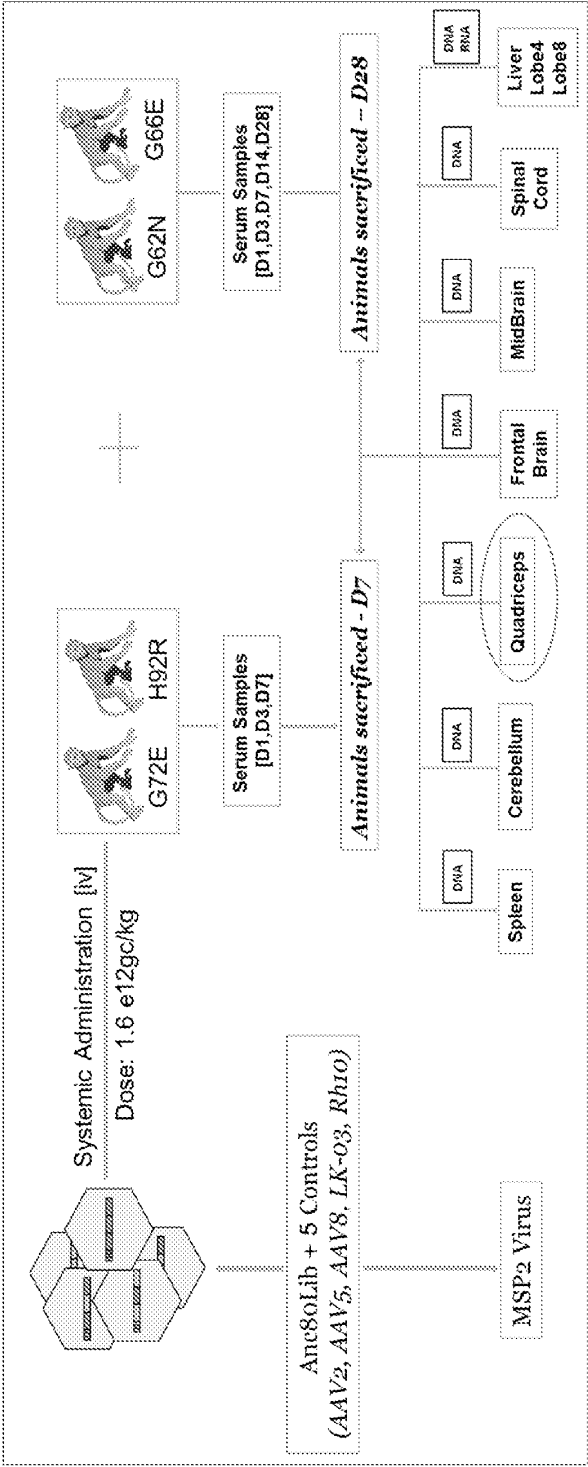


FIG. 21

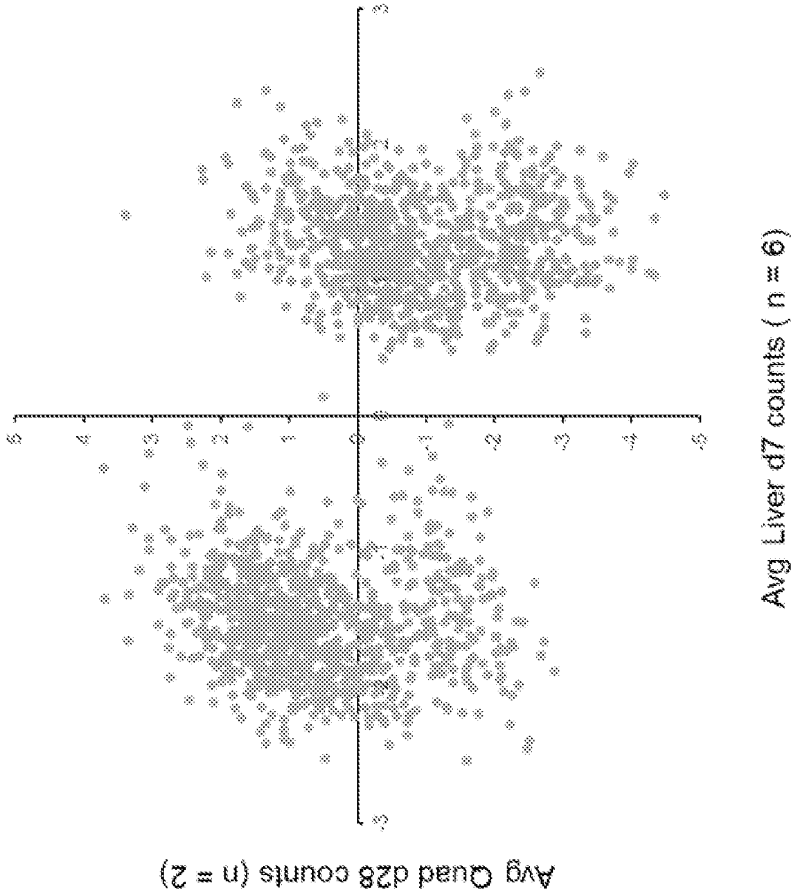


FIG. 22

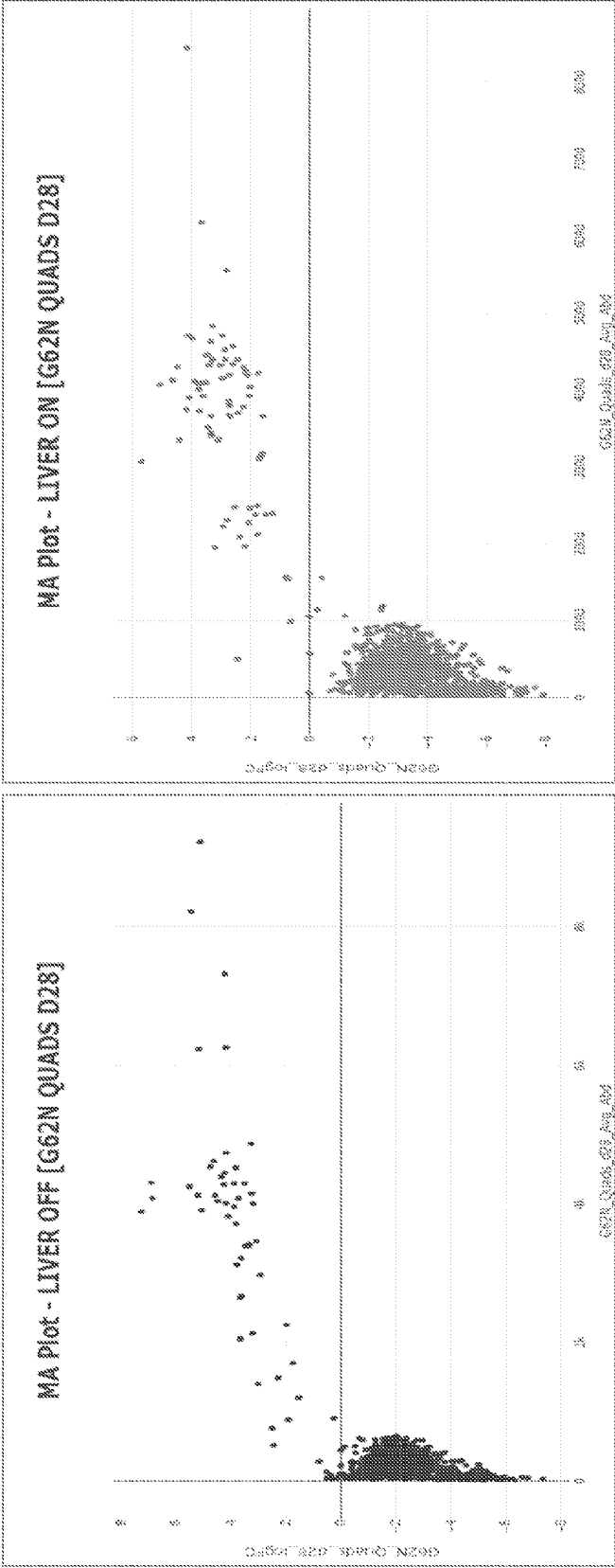


FIG. 23

Effect of P3 on Logfoldchange in G62N d28 Quads

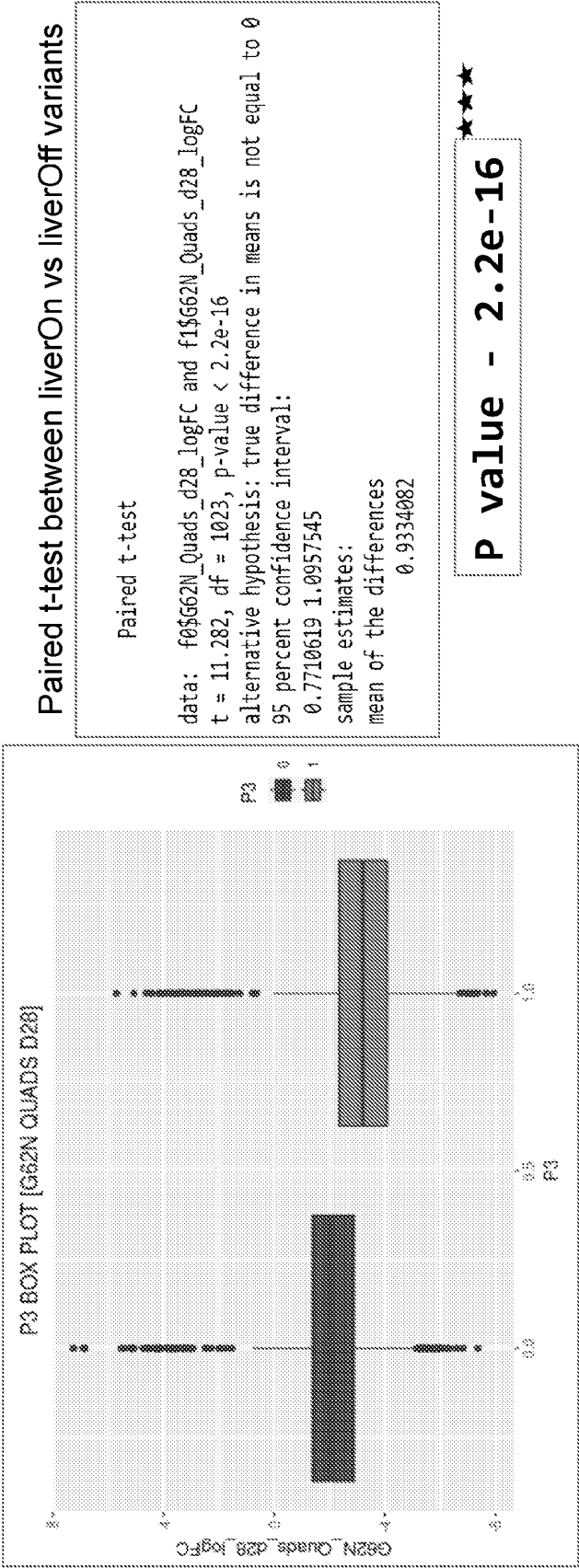


FIG. 24

Effect of P3 on Logfoldchange in G66E d28 Quads

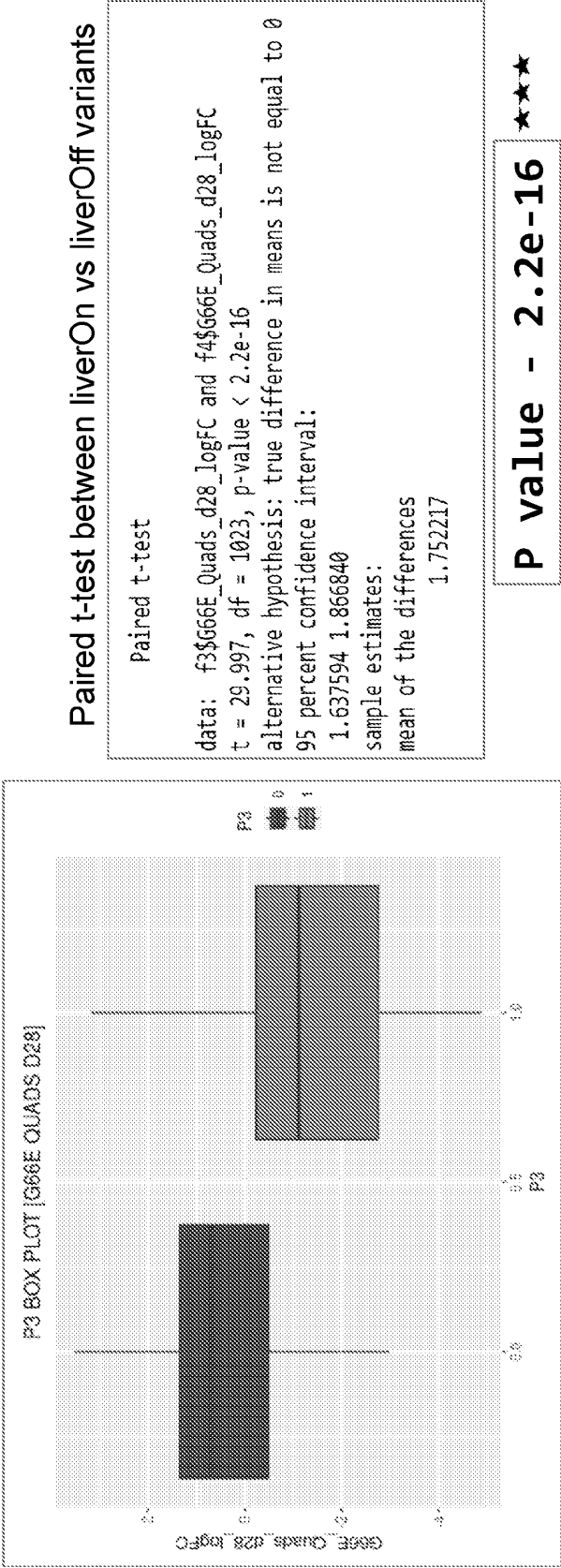


FIG. 25

Effect of P3 on Average LogFC in both d28 Quads

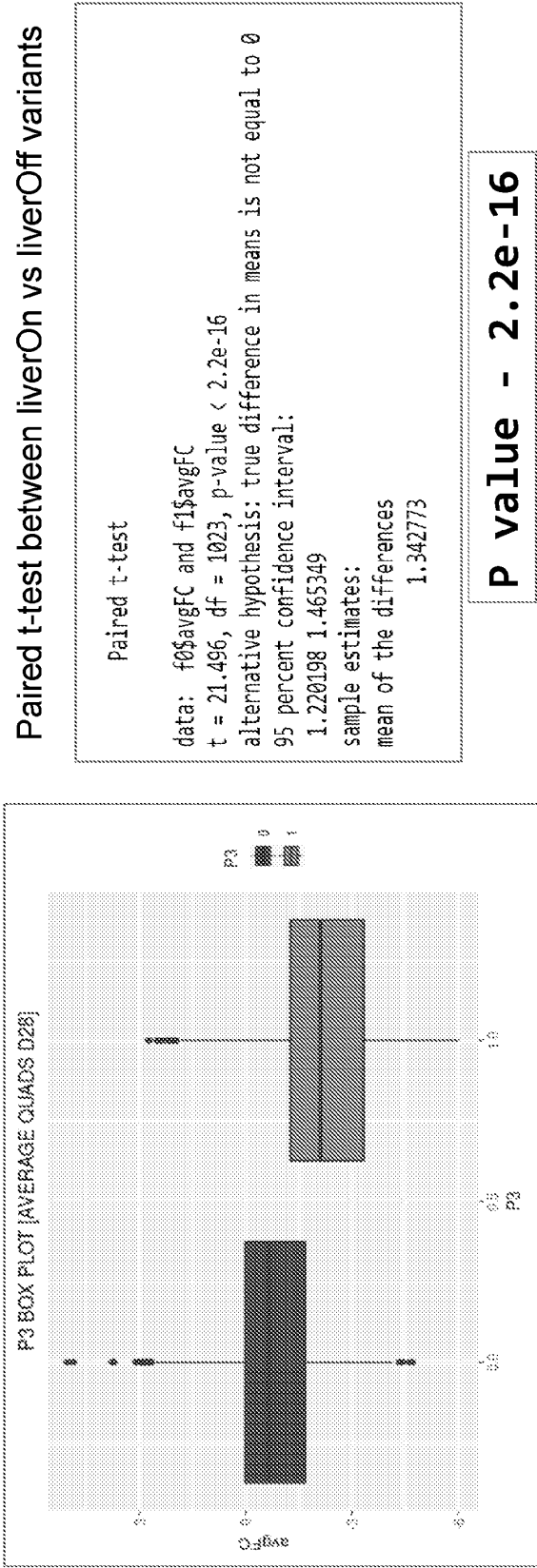


FIG. 26

Quadrant Plot - G62N d28 Quads

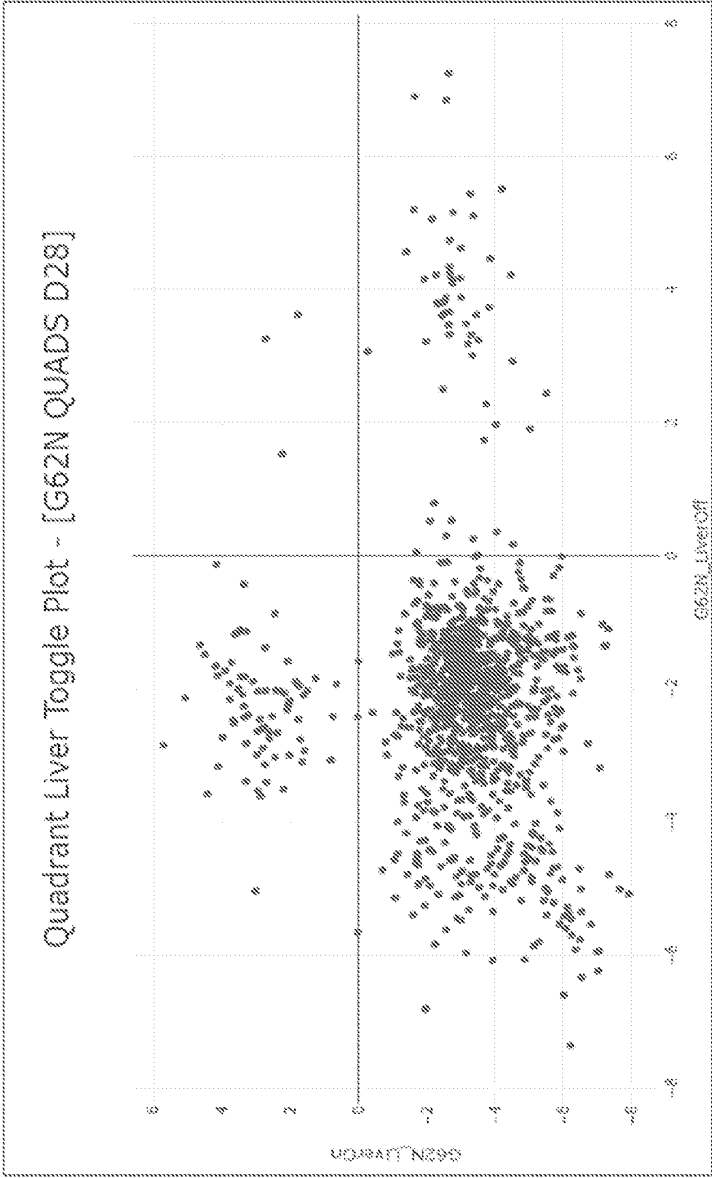


FIG. 27

Quadrant Plot - G66E d28 Quads

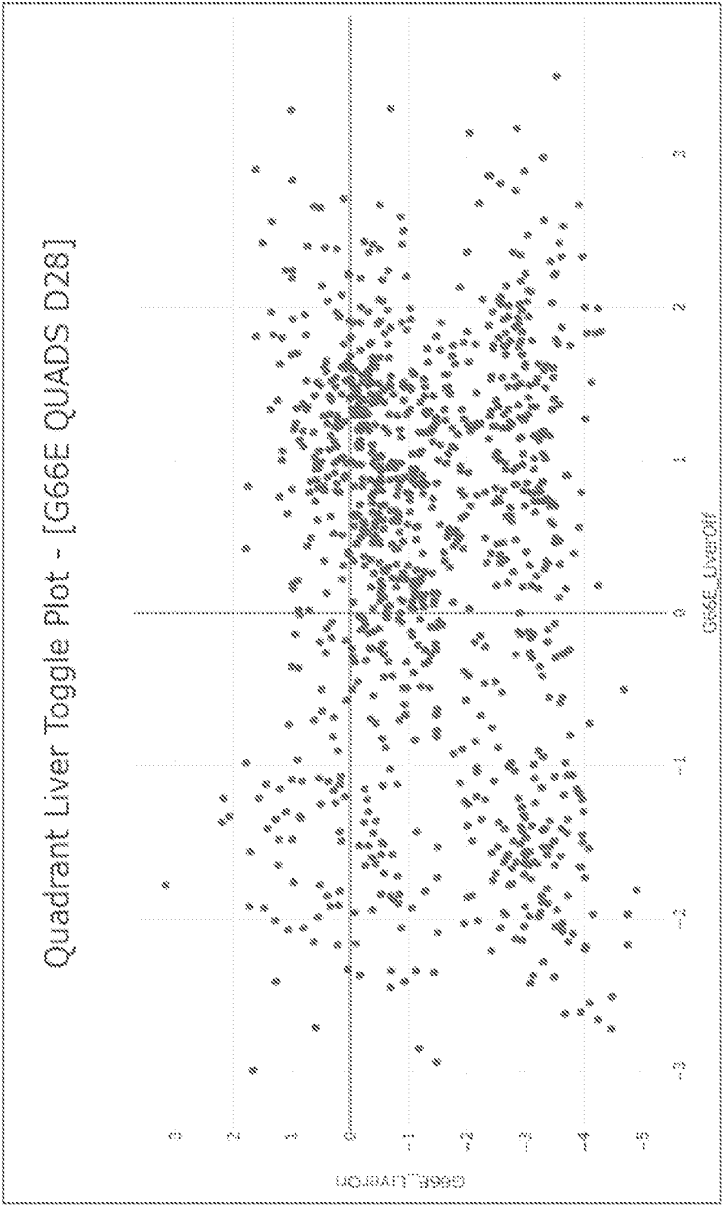


FIG. 28

UL Higher with Toggle On UR High in both LL Low in both LR Lower with Toggle On	
Plot Position	No. of Variants
UR	125
UL	66
LR	566
LL	271

Quadrant Plot – Animal Comparative Analysis

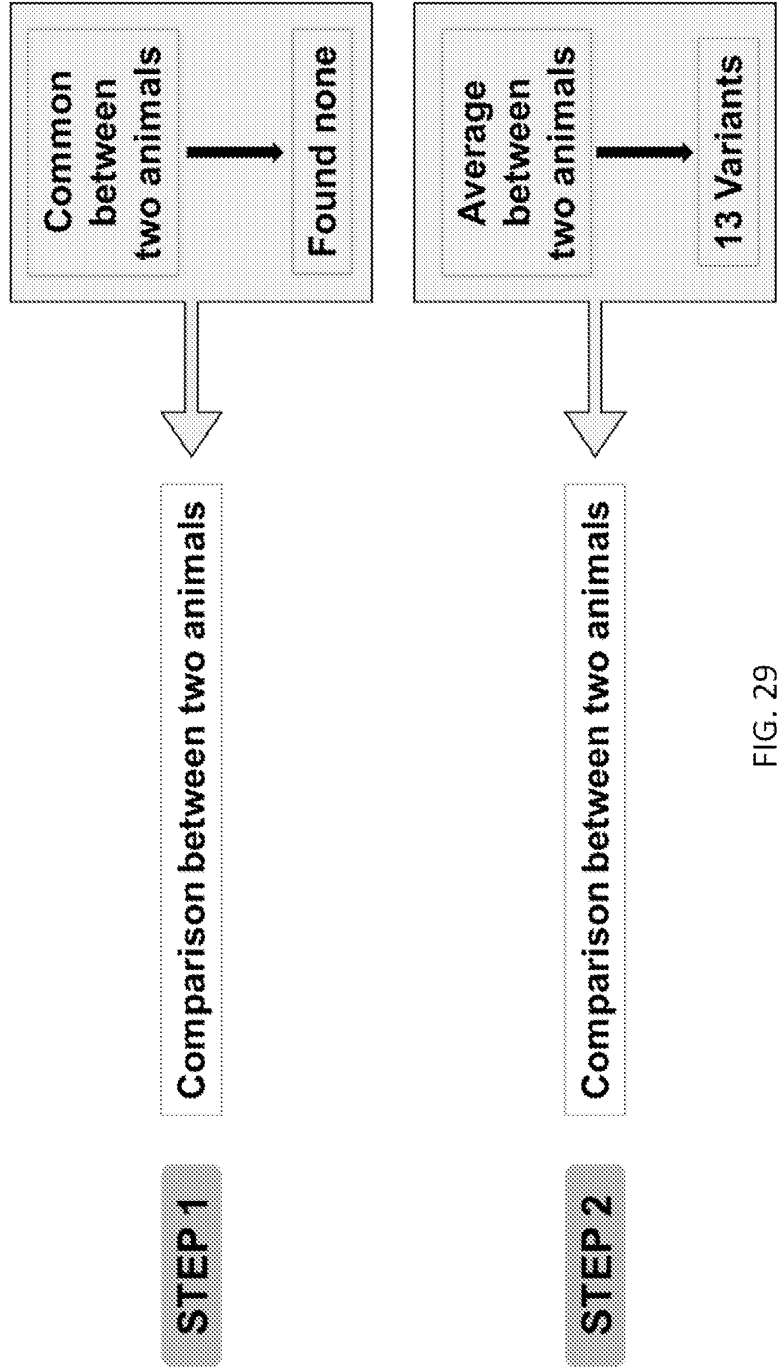


FIG. 29

Quadrant Plot – Average d28 Quads

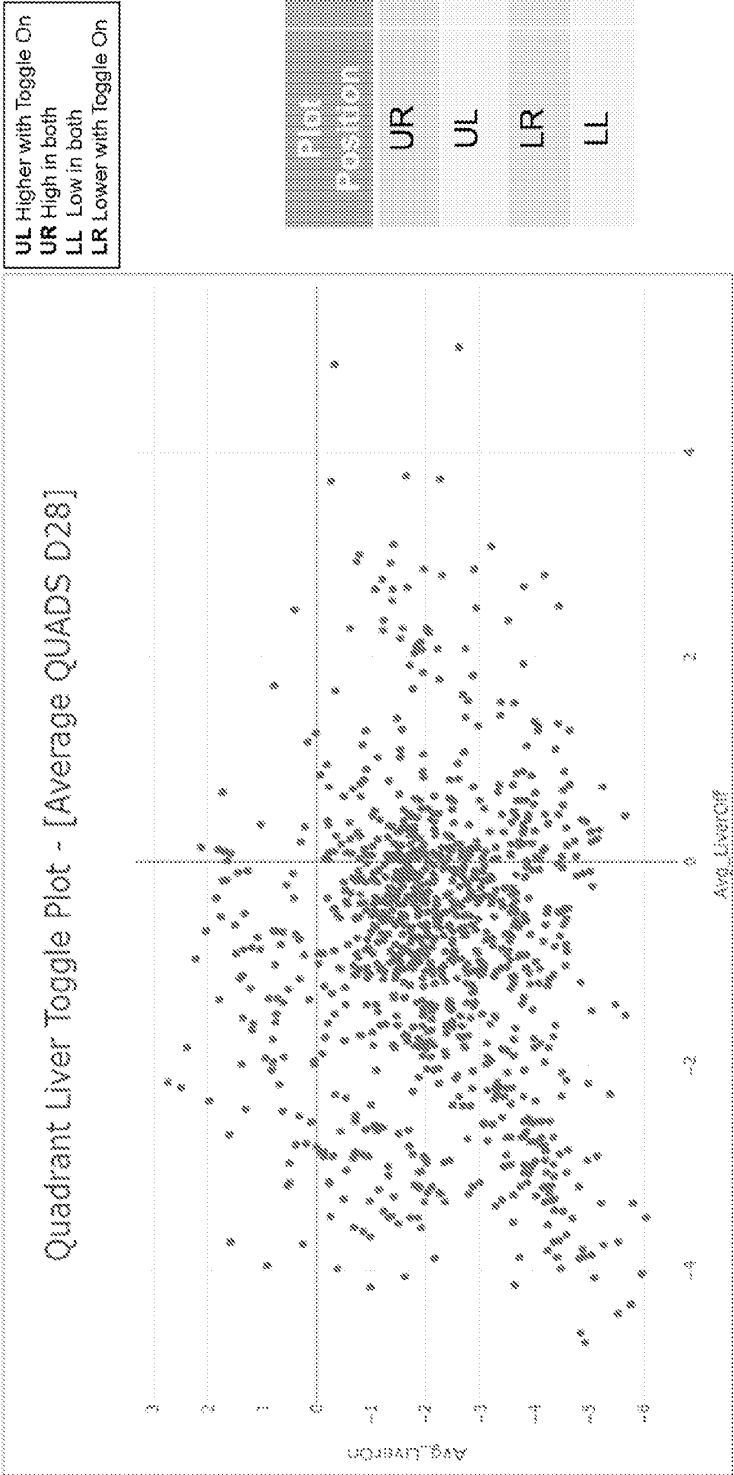
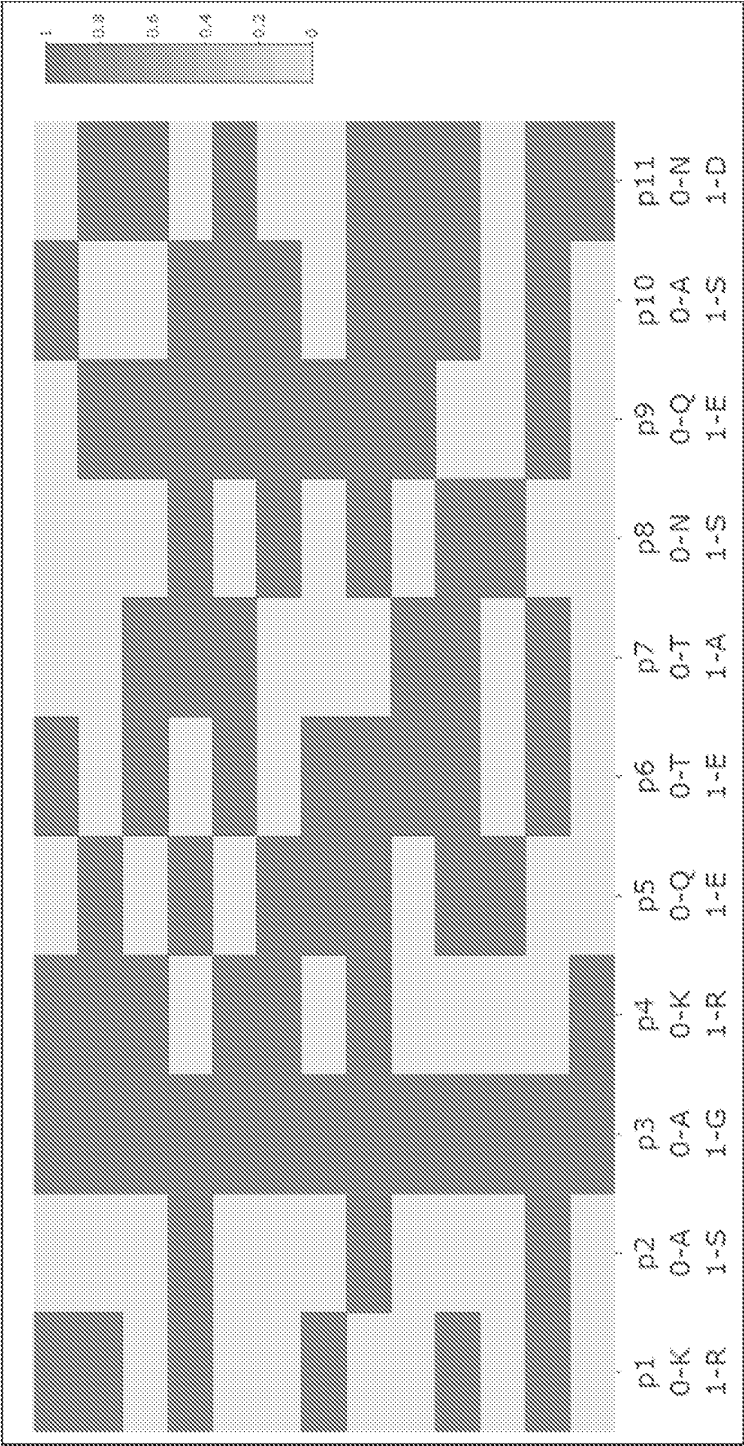


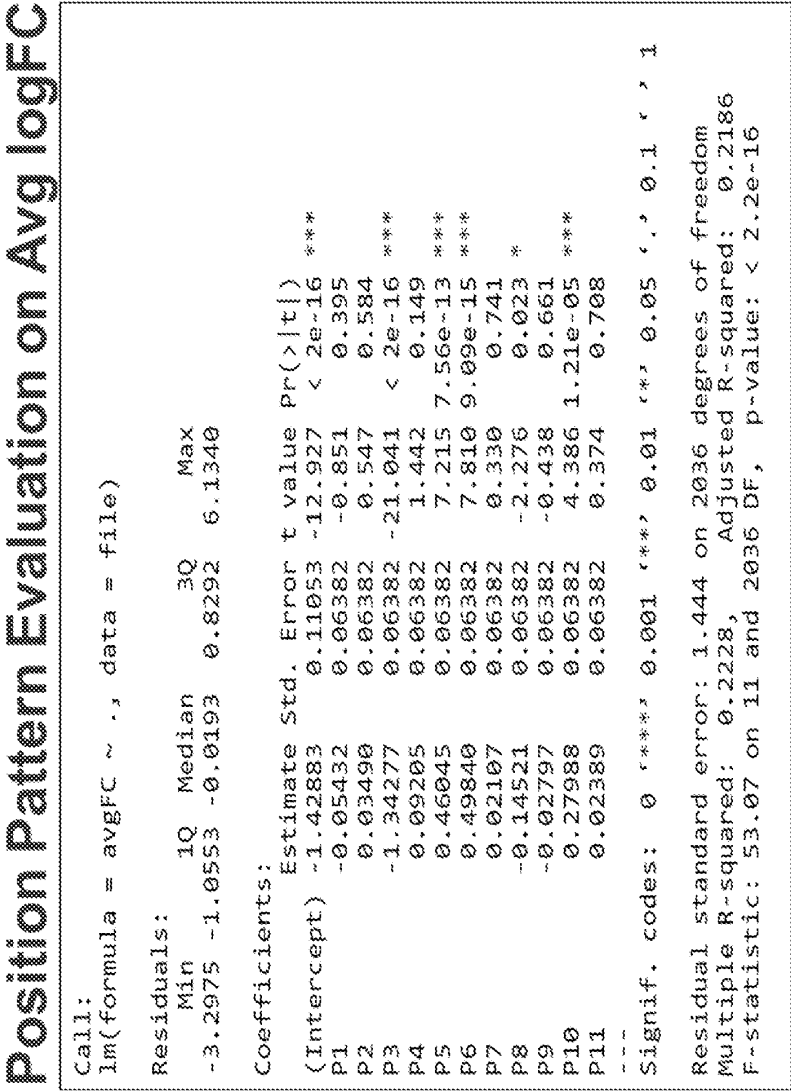
FIG. 30

FingerPrint Plot [UR]– Average d28 Quads



Variants sorted in decreasing order by Liver On foldchange

FIG. 31



Comment: P3,P5,P6 AND P10 can have some pattern impact on the average logfc of the quads tissues.

FIG. 32

Binary codes for liver toggle variant pairs

	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11
Variant pair 1	0	0	0	1	0	0	1	1	1	0	0
	0	0	1	1	0	0	1	1	1	0	0
Variant pair 2	0	1	0	0	0	0	1	0	0	0	0
	0	1	1	0	0	0	1	0	0	0	0
Variant pair 3	1	0	0	0	0	0	1	0	1	0	1
	1	0	1	0	0	0	1	0	1	0	1

FIG. 33

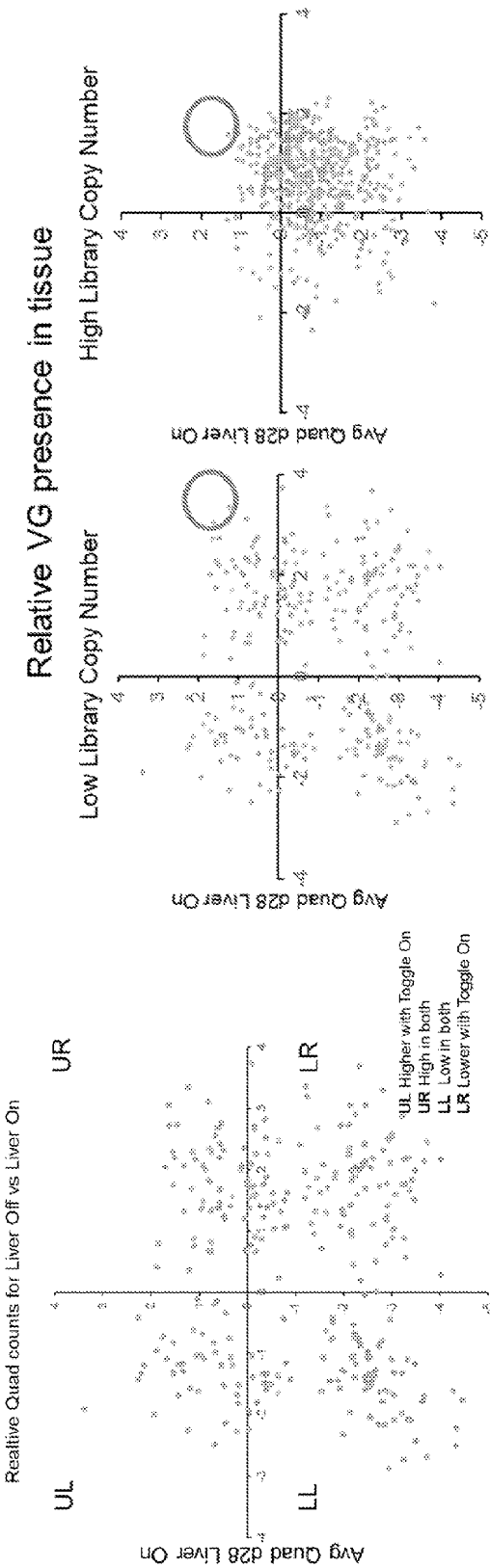


FIG. 33 (cont'd)

P4 P5 P6 P7						
A	A	A	A			
D	D	D	D			
E	E	E	E			
F	F	F	F			
G	G	G	G			
H	H	H	H			
I	I	I	I			
S	D	K	K	K	K	
		L	L	L	L	
		N	N	N	N	
		Q	Q	Q	Q	
		R	R	R	R	
		S	S	S	S	
		T	T	T	T	
		V	V	V	V	

P1 P2 P3

FIG. 34

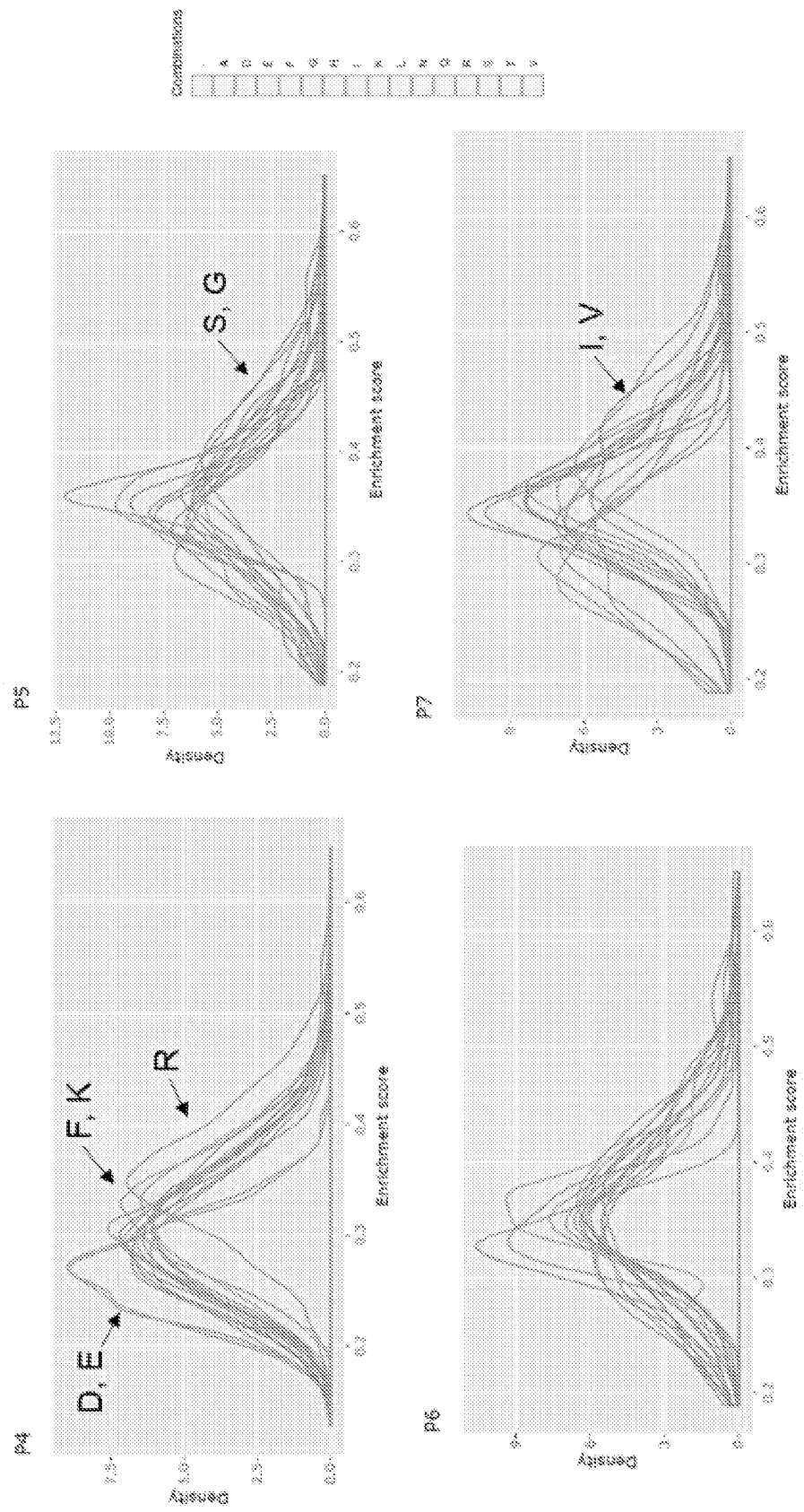


FIG. 35

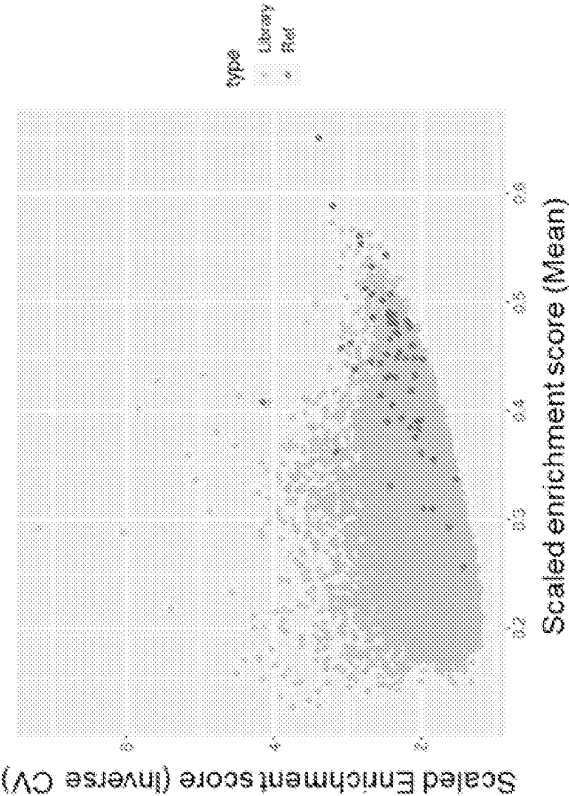


FIG. 36B

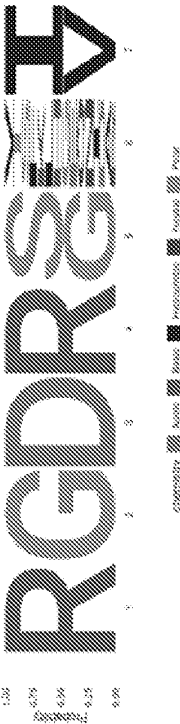


FIG. 36A

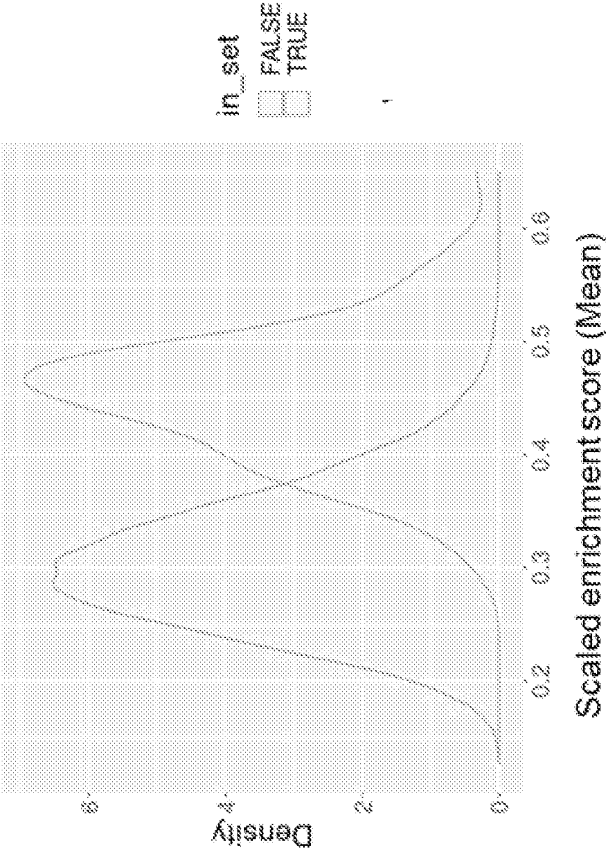


FIG. 36C

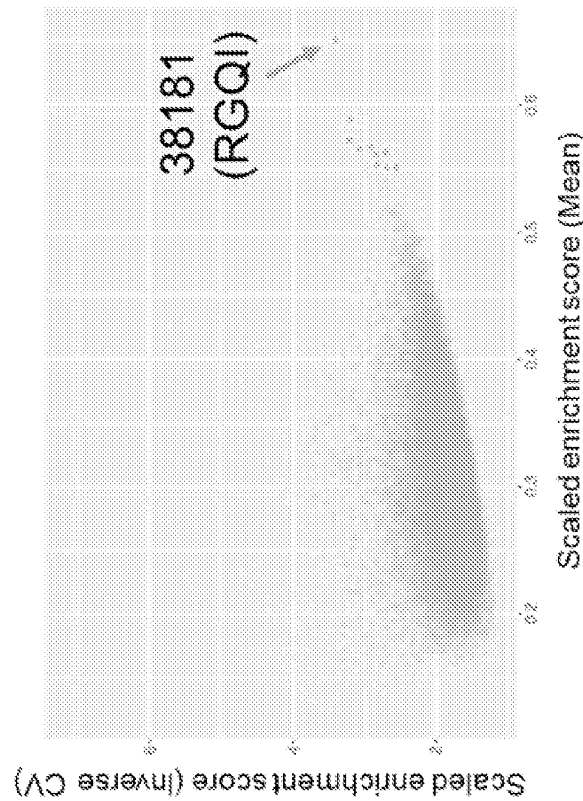


FIG. 37B

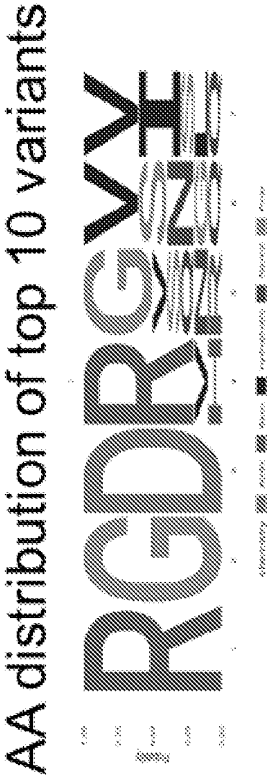
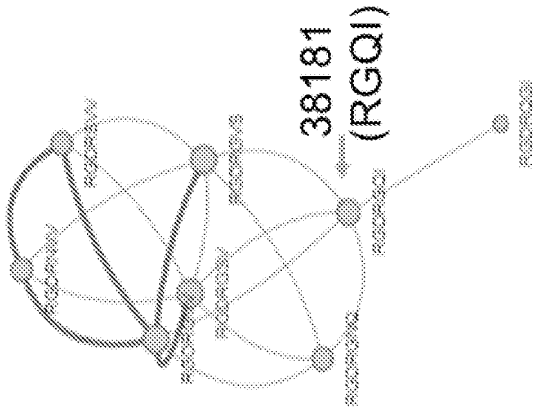


FIG. 37A

Sequence similarity network of
top 10 variants



Network edge cutoff:
reverse hamming distance ≥ 5

FIG. 37C

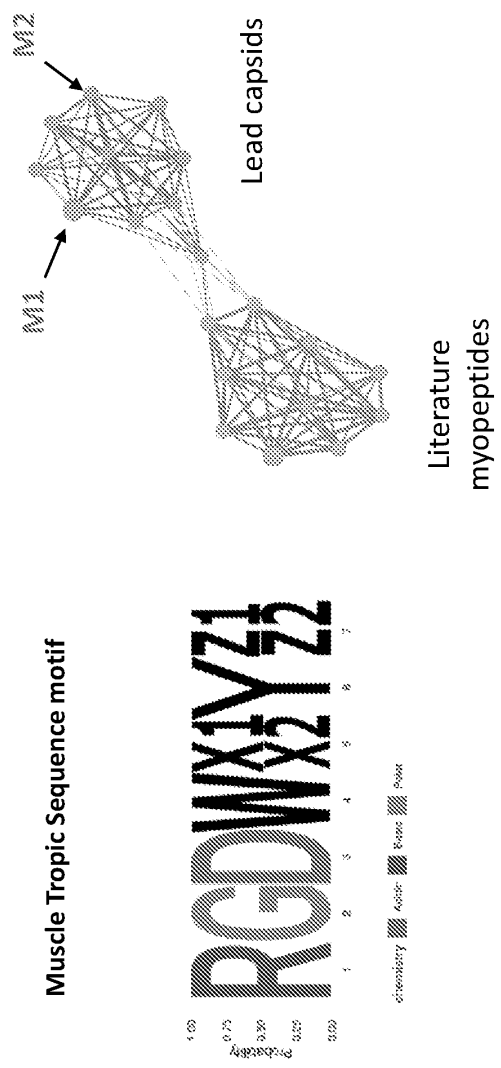


FIG. 37E

FIG. 37D

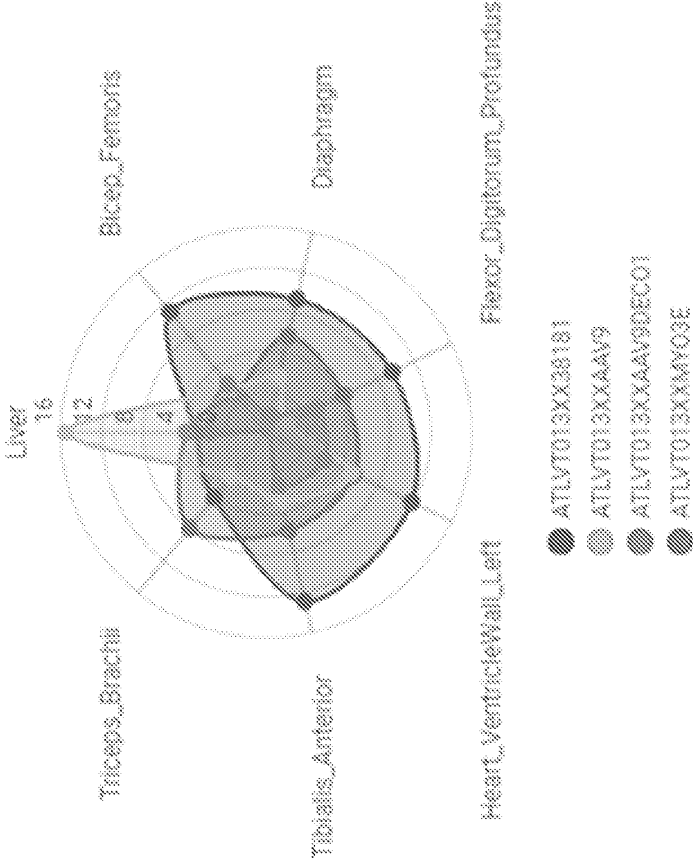
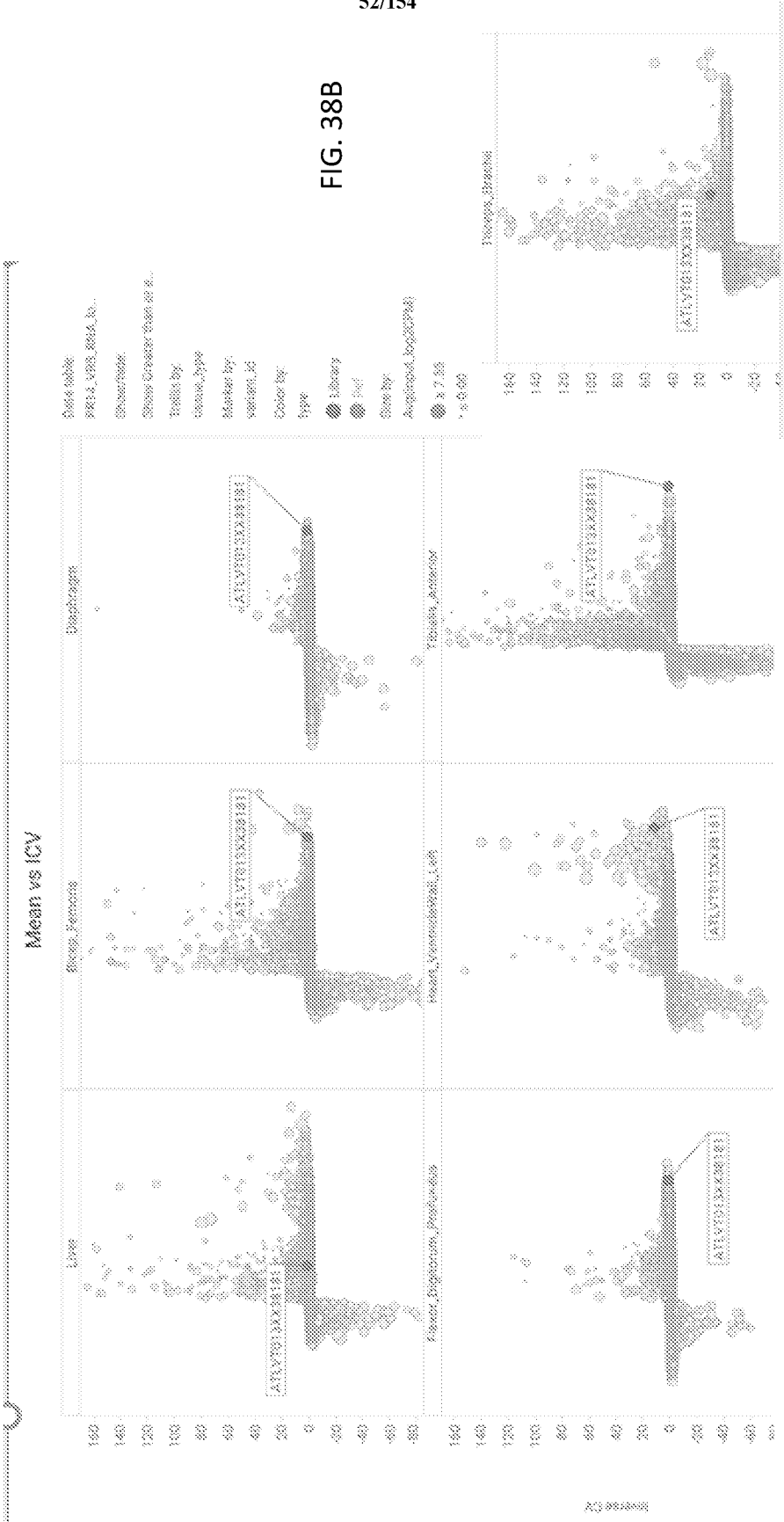


FIG. 38A



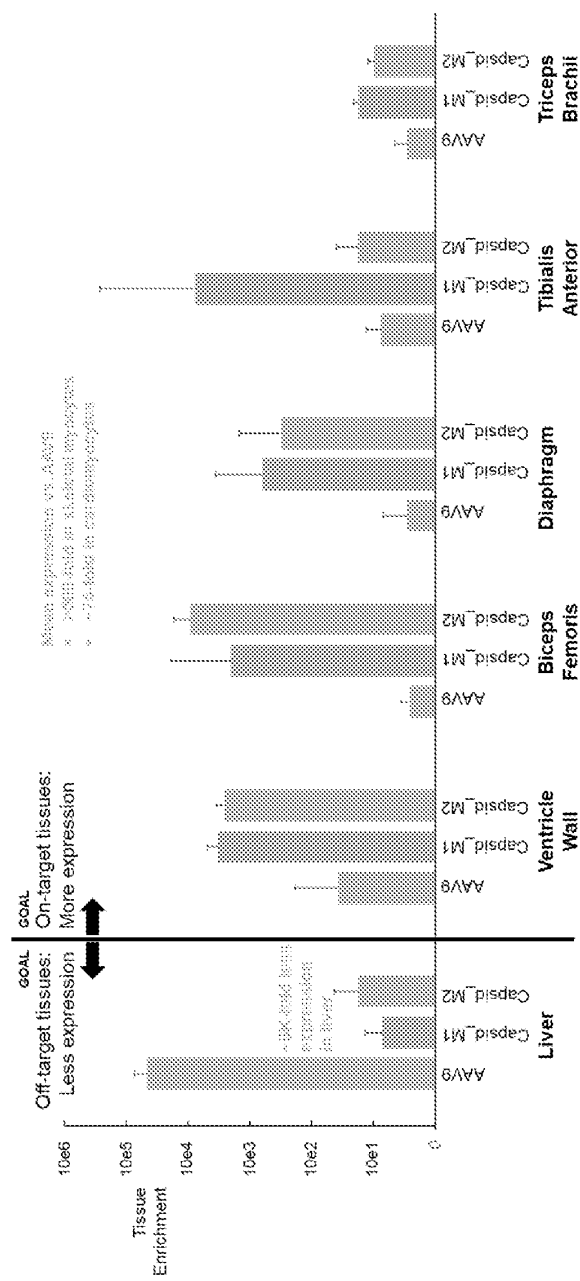
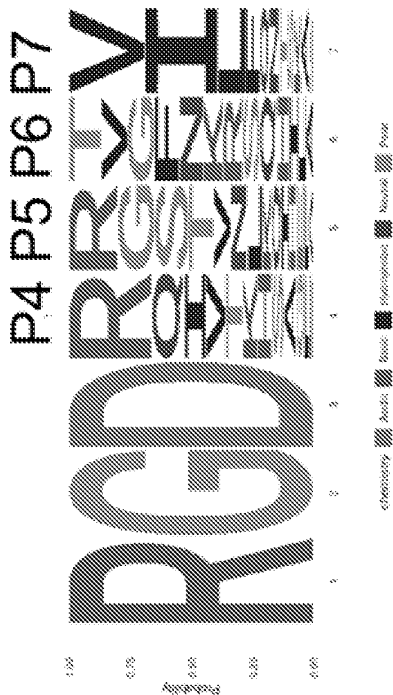


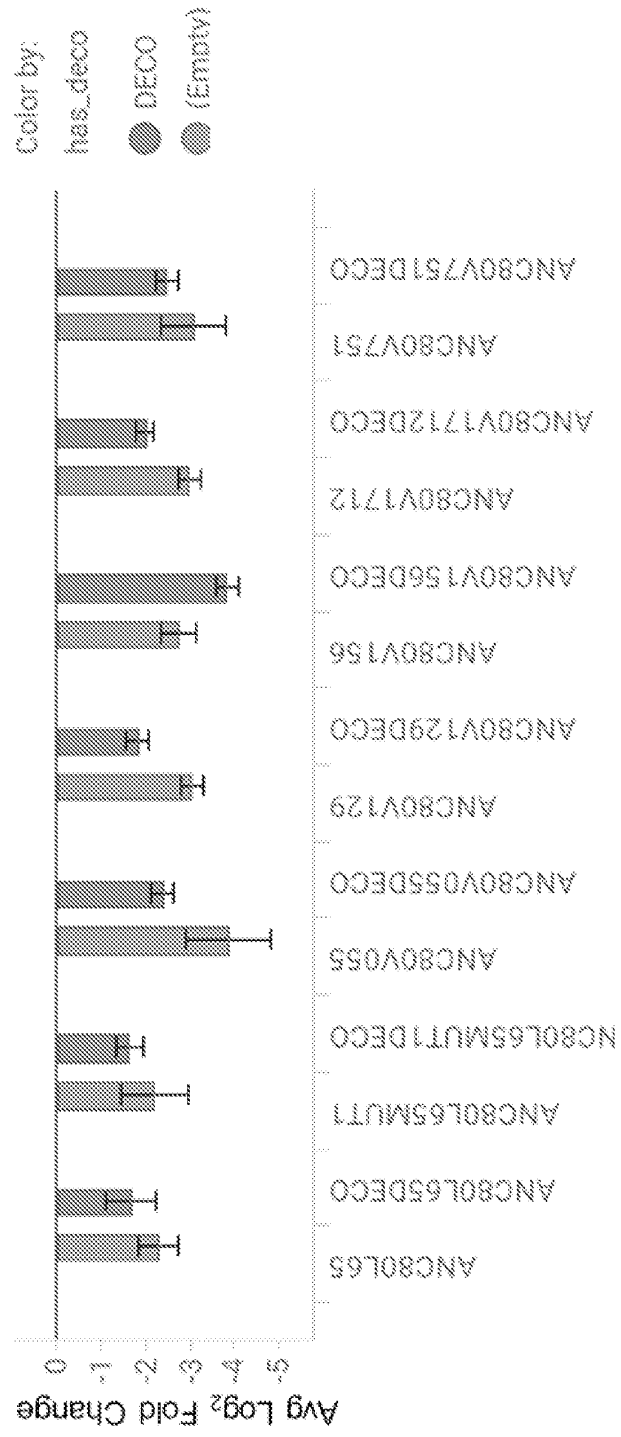
FIG. 38C

Top100 muscle variants from PR0014



- ❖ R is enriched at P4
- ❖ V, I, L are enriched at P7

FIG. 39

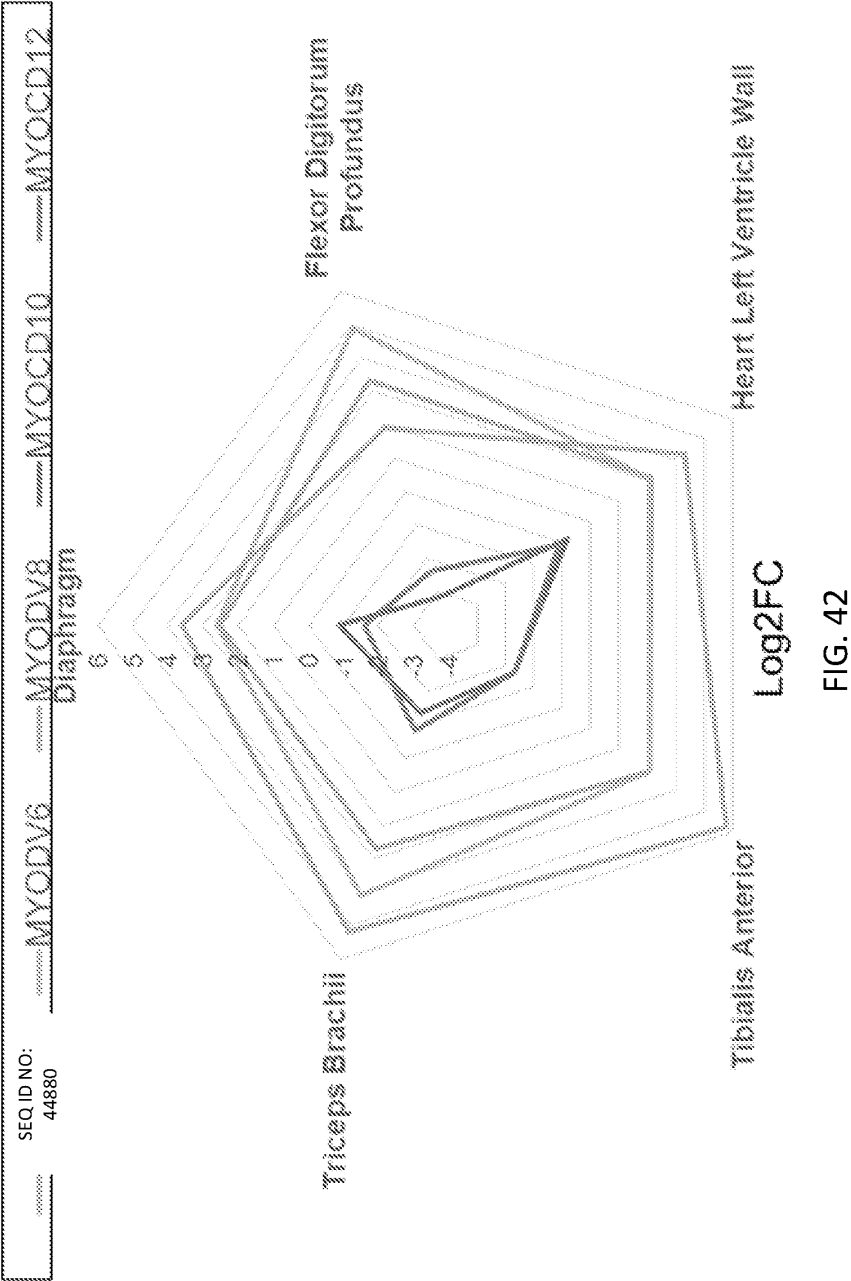


PR-0014: VR8 Library Screen (IV – Cyno)
D28, 1E14 VG/ml

FIG. 40

SEQ ID NO: 44880	Tibia		Quadriceps		Oblique		Flexor Digitorum Profundus		Heart Left Ventricle Wall		Tibia Anterior		Intercept Gracilis	
	ENR	RGD	FNNT											
MYODV1	ENR	RGD	YNSL			2.53	3.33	3.15	3.1				2.72	
MYODV2	ENR	RGD	YNNL			0.99	2.04	1.46	2.7				2.16	
MYODV3	ENR	RGD	YNNL			1.3	-0.47	1.55	2.45				1.39	
MYODV4	ENR	RGD	YNST			2.19	0.62	3.06	0.18				3.04	
MYODV5	ENR	RGD	YNNL			-1.18	-1.72	0.35	-2.37				-0.51	
MYODV6	ENR	RGD	FNSL			0.2	2.72	0.64	-0.31				0.78	
MYODV7	ENR	RGD	FNNL			2.62	4.88	3.07	3.09				4.09	
MYODV8	ENR	RGD	FNST			0.54	2.45	2.05	1.11				1.81	
MYODV9	ENR	RGD	FQNT			3.61	1.91	4.29	5.78				5.7	
MYODV10	ENR	RGD	EQNT			0.05	-3.35	0.35	-2.72				-1.73	
MYODV11	ENR	RGD	QQNT			1.3	-0.35	1.22	-0.56				1.2	
MYODV12	SNR	RGD	YNSL			0.99	-3.58	2.03	-1.19				2.13	
MYODV13	SNR	RGD	YNNL			0.77	0.44	1.37	-1.36				1.87	
MYODV14	SNR	RGD	YNST			-0.92	-7.2	-1.08	-5.53				-4.35	
MYODV15	SNR	RGD	YNNL			0.48	-3.44	-0.26	-0.82				-0.85	
MYODV16	SNR	RGD	FNSL			-1.23	-3.27	-1.06	-3.7				-1.51	
MYODV17	SNR	RGD	FNNL			-0.87	-3.17	0.18	-1.69				-1.37	
MYODV18	SNR	RGD	FNST			-2.31	5.27	-1.29	-4.26				-1.61	
MYODV19	SNR	RGD	FQNT			-1.6	-2.37	-0.11	-1.68				-0.85	
MYODV20	SNR	RGD	EQNT			0.51	-0.9	0.67	-2.25				1.45	
MYODV21	SNR	RGD	QQNT			-1.62	-0.95	-1.61	-3.92				-1.05	

FIG. 41



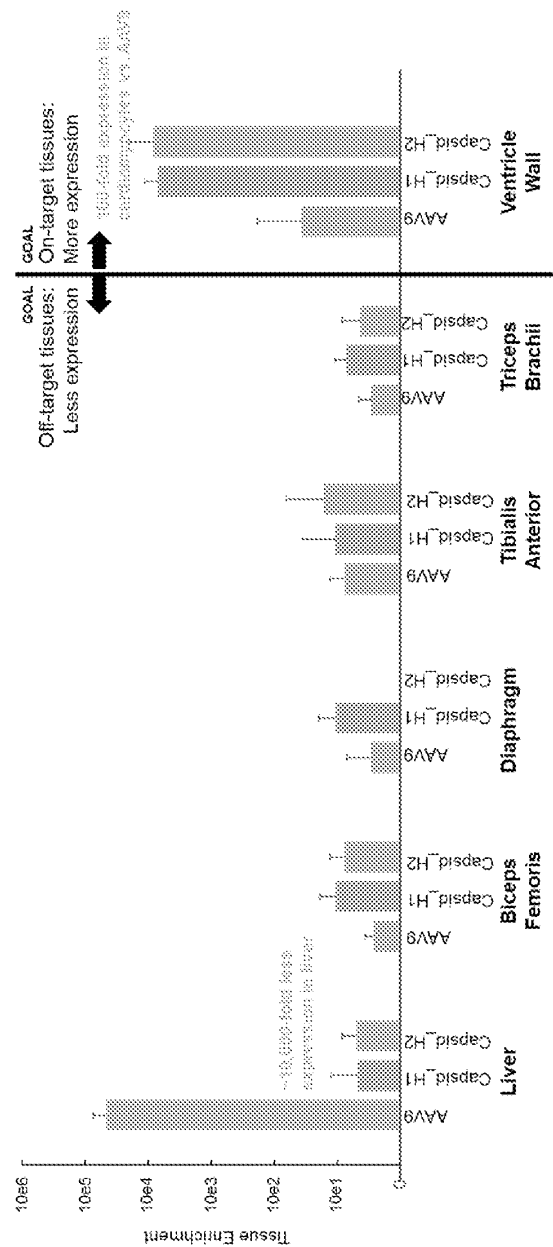


FIG. 43

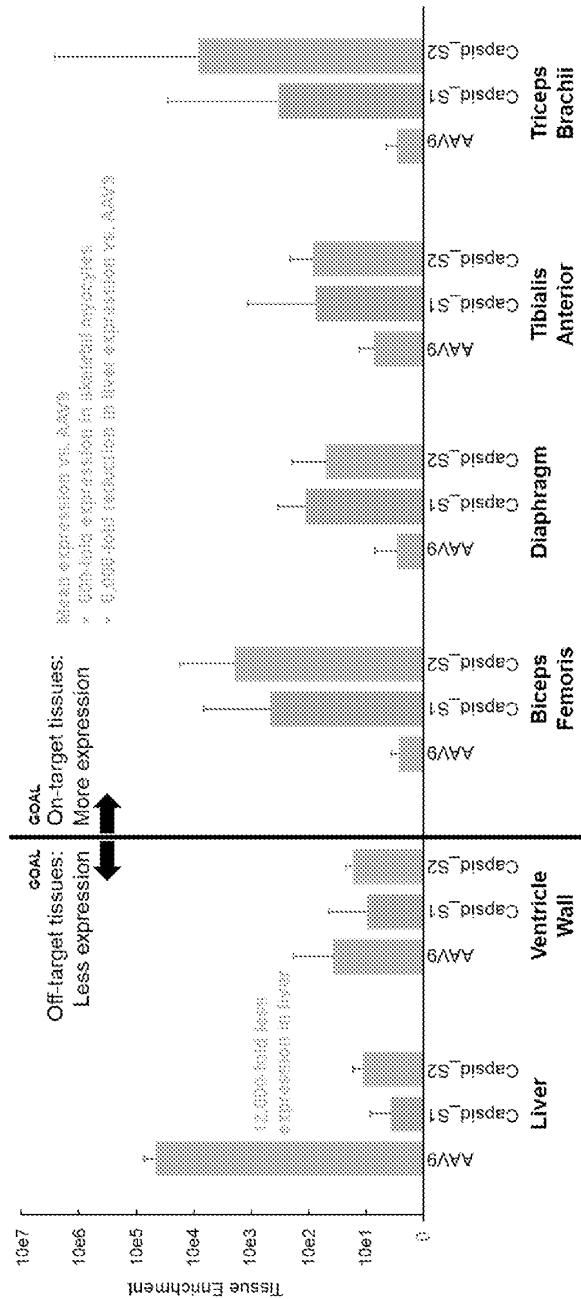


FIG. 44

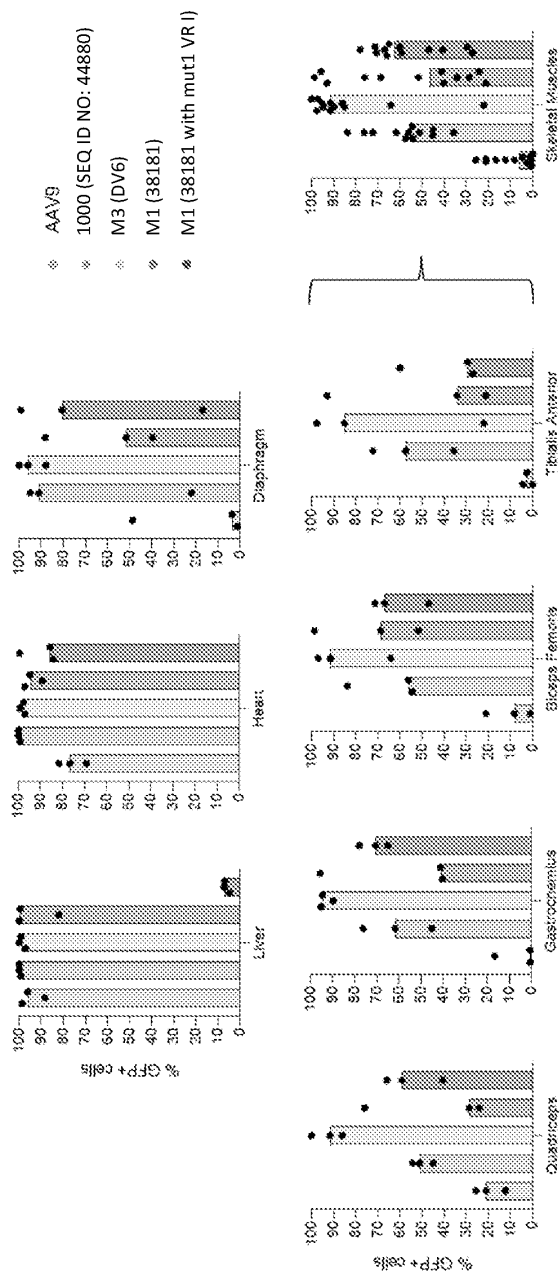


FIG. 45

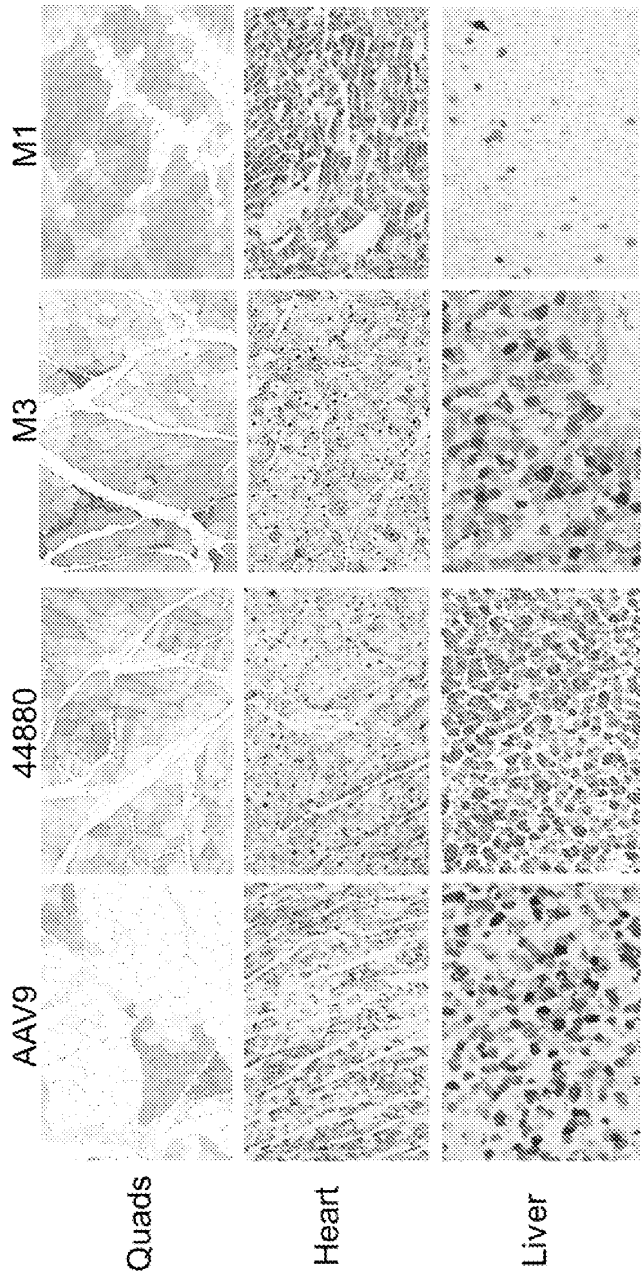


FIG. 46

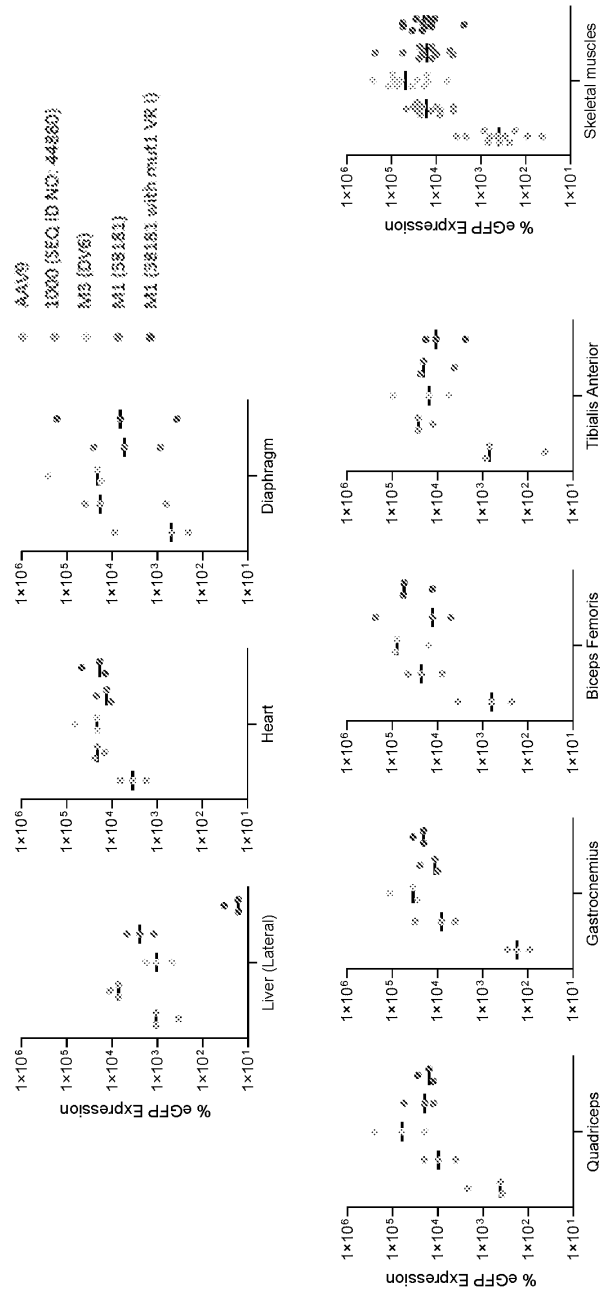


FIG. 47

		eGFP mRNA per murine RPP30 copy				Vector genomes per DPG			
		Liver	Quadriceps	Heart	Diaphragm	Liver	Quadriceps	Heart	Diaphragm
1.00E+13	WT	845.6	0.3	0.4	8.3	3.1	0.8	0.1	0.2
	mut1	0.3	0	0	0.3	0.1	0	0	0
5.00E+13	WT	4180.8	1.8	3.2	46.8	8	6.4	0.9	0.3
	mut1	2.9	0.3	0.3	3.9	0.9	0	0.4	0.1

Average of 3 mice.
5E13 VG per kg.

FIG. 48

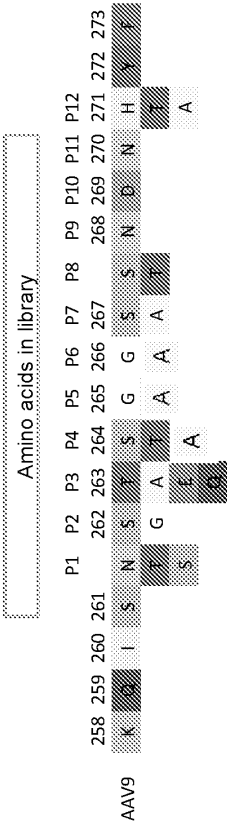


FIG. 49A

AVVR-lib1: Residues in VR1																								
	258	259	260	261	262	263	264	265	266	267	268	269	270	271	272	273								
AAV1	K	Q	I	S	-	S	A	S	T	G	A	S	N	D	N	H	Y	F						
AAV2	K	Q	I	S	-	S	Q	S	G	A	S	-	N	D	N	H	Y	F						
AAV3	K	Q	I	S	-	S	Q	S	G	A	S	-	N	D	N	H	Y	F						
AAV6	K	Q	I	S	-	S	A	S	T	G	A	S	N	D	N	H	Y	F						
AAV7	K	Q	I	S	-	S	E	T	A	G	S	T	N	D	N	T	Y	F						
AAV8	K	Q	I	S	N	G	T	S	G	G	A	T	N	D	N	T	Y	F						
AAV9	K	Q	I	S	N	S	T	S	G	G	S	S	N	D	N	A	Y	F						
AAV10	K	Q	I	S	N	G	T	S	G	G	S	T	N	D	N	T	Y	F						
AAVhu.3																								
7	K	Q	I	S	N	G	T	S	G	G	S	T	N	D	N	T	Y	F						
AAVrh.10	K	Q	I	S	N	G	T	S	G	G	S	T	N	D	N	T	Y	F						
AAVhu.6																								
8	K	Q	I	S	N	S	T	S	G	G	S	S	N	D	N	A	Y	F						
LK03	K	Q	I	S	-	S	Q	S	G	A	S	-	N	D	N	H	Y	F						

FIG. 49B

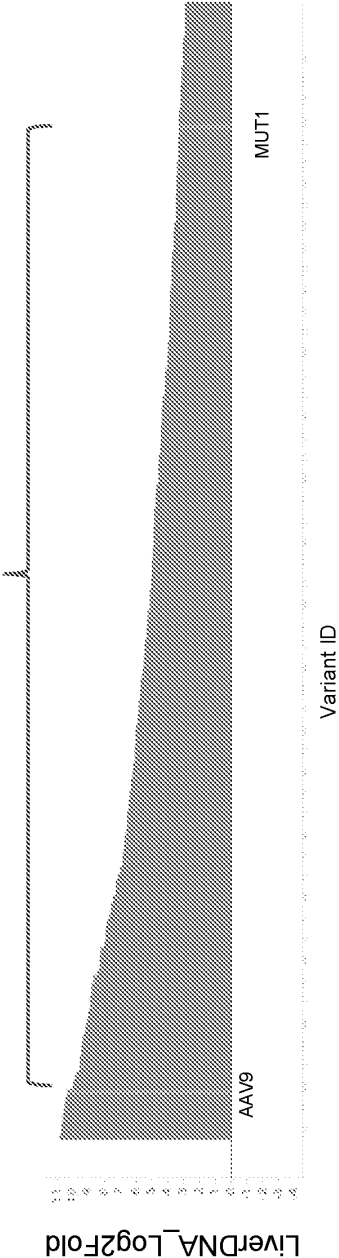


FIG. 50

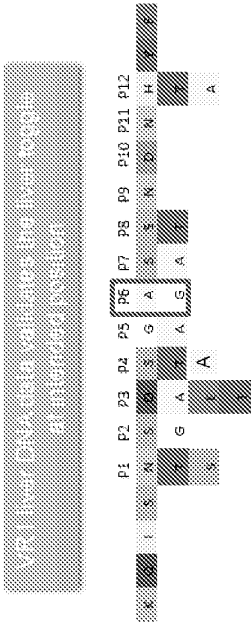


FIG. 51A

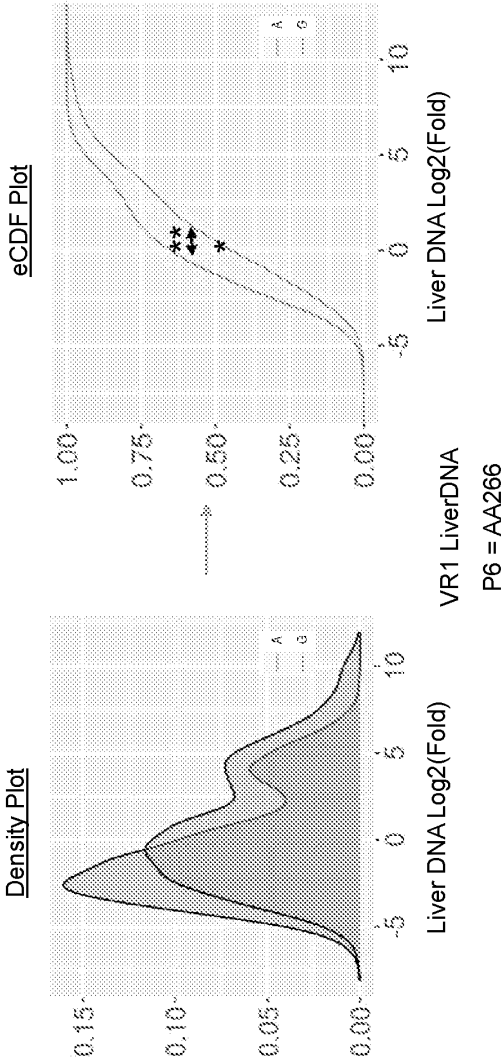


FIG. 51B

FIG. 51C

SAR analysis identified additional amino acids affecting liver targeting

$$\text{Enrichment score} = a_0 + a_1 \cdot X_1T + a_2 \cdot X_1S + a_3 \cdot X_2S + a_4 \cdot X_3E + a_5 \cdot X_3Q + a_6 \cdot X_3T + \dots + a_{13} \cdot X_{12}H + a_{14} \cdot X_{12}T$$

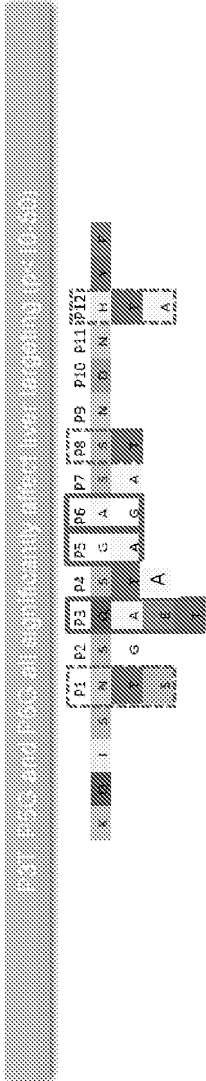


FIG. 52A

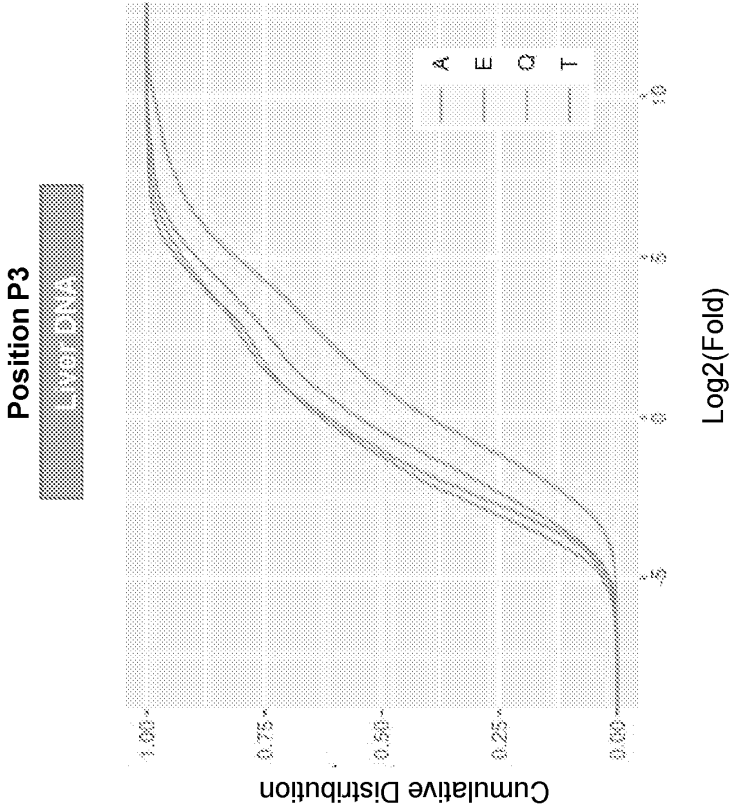


FIG. 52B

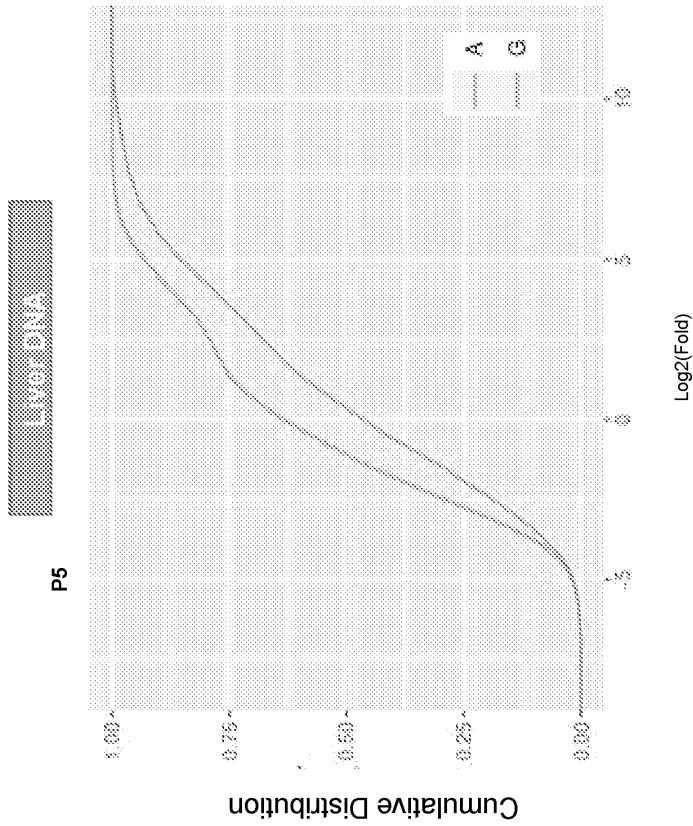


FIG. 52C

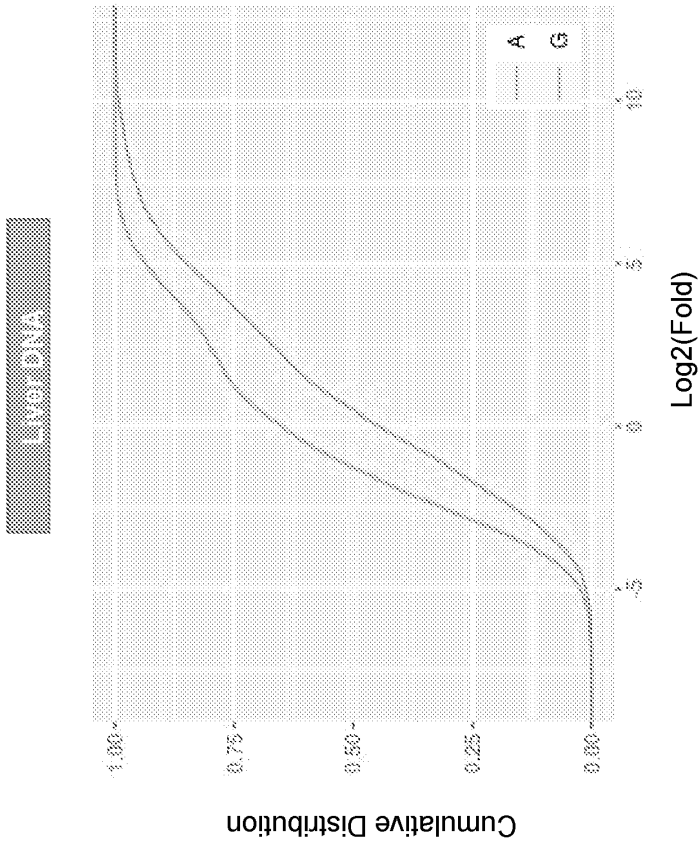


FIG. 52D

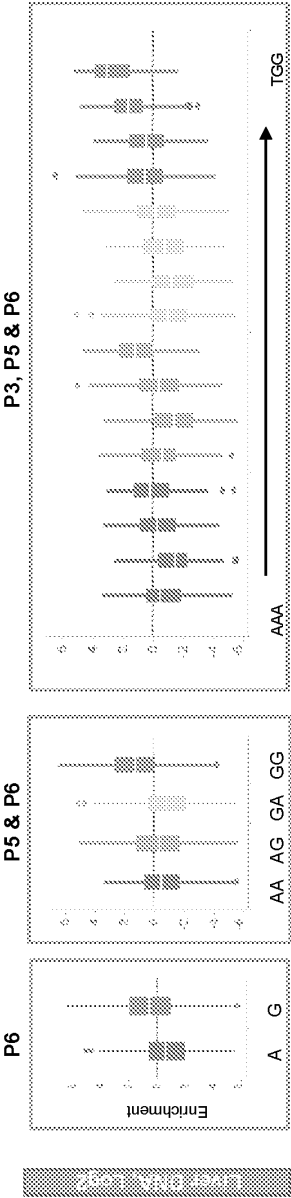


FIG. 53A

FIG. 53B

FIG. 53C

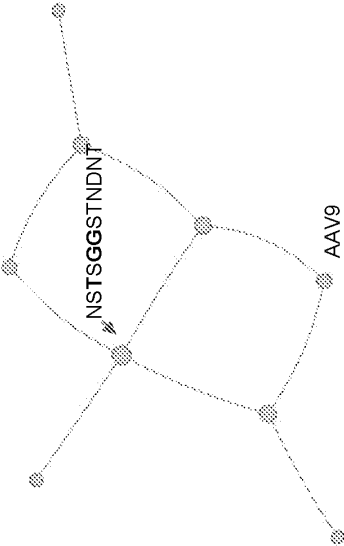


FIG. 54

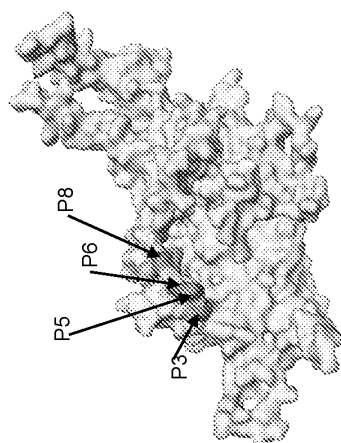


FIG. 55

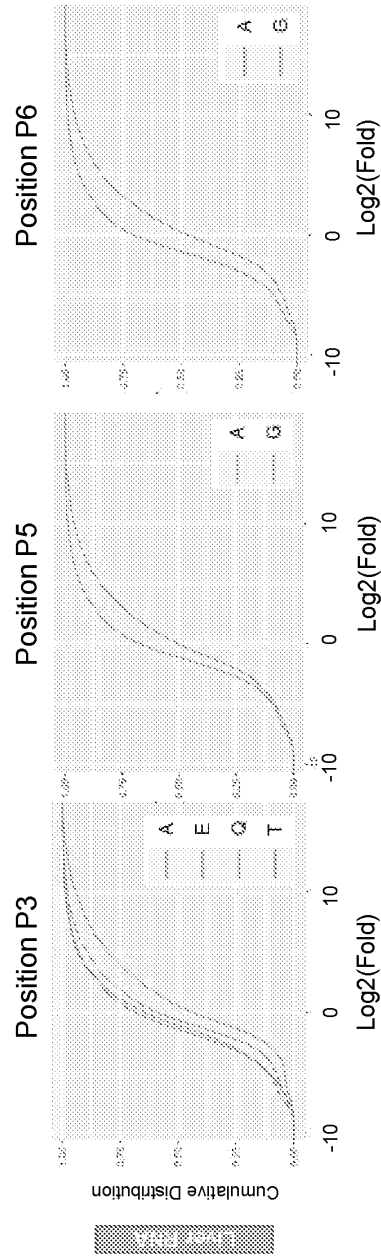


FIG. 56E

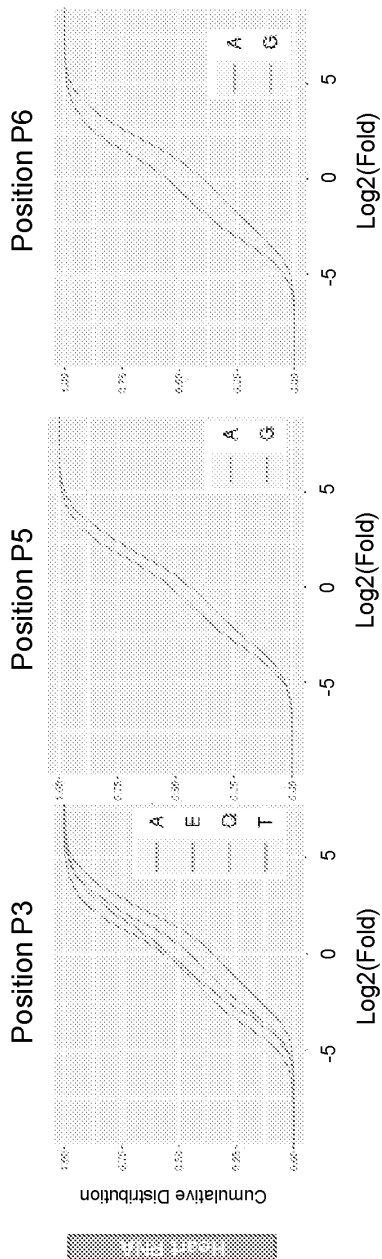


FIG. 56F

FIG. 56D

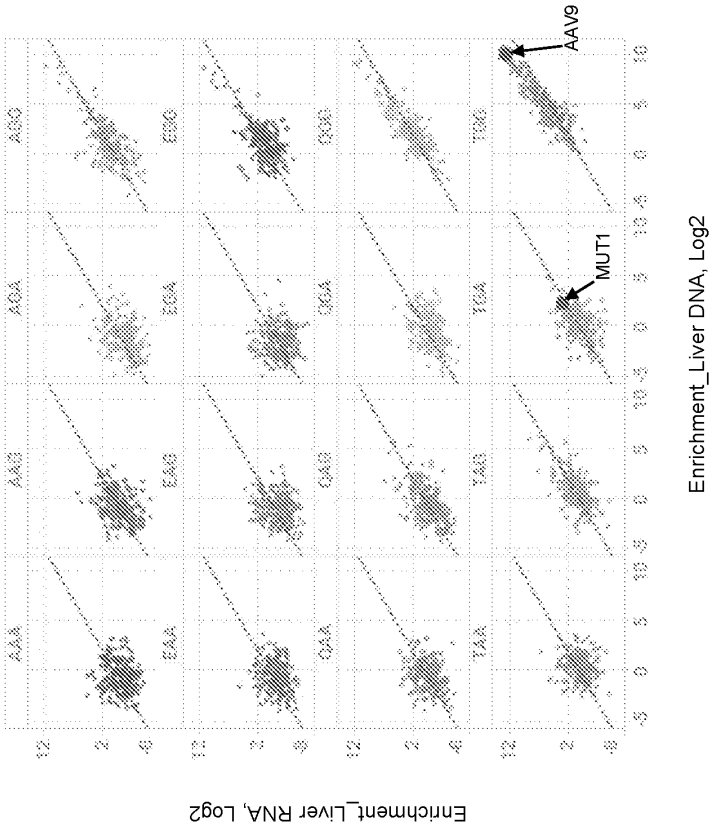
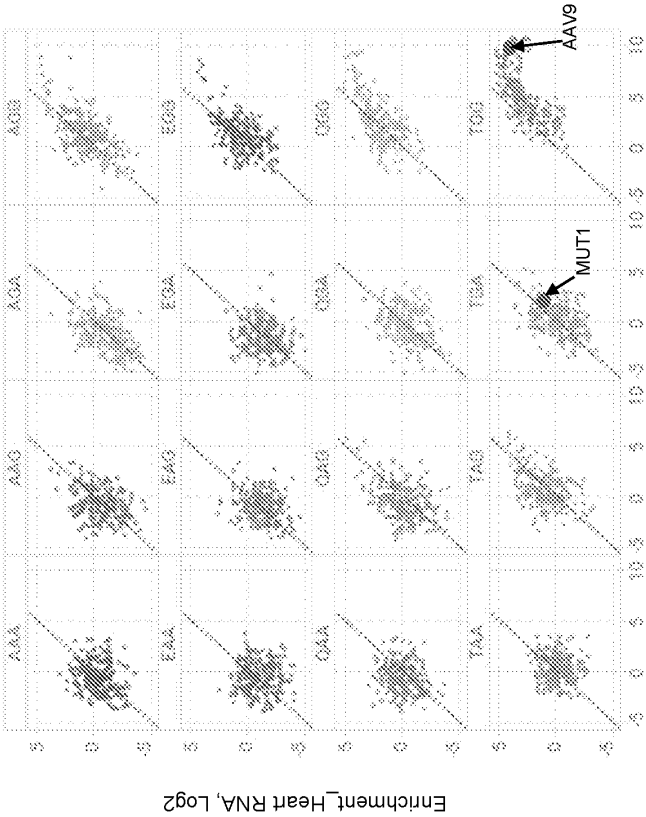


FIG. 57A



Enrichment_Liver DNA, Log2

FIG. 57B

TGG group partially uncouples liver and heart expression

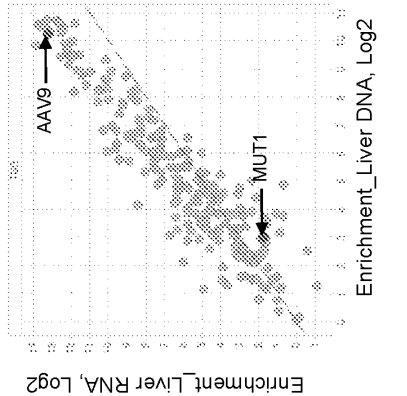


FIG. 58A

P1 position modulates TGG group effects

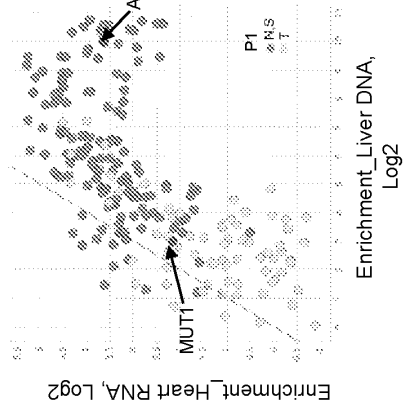


FIG. 58C

TGG group partially uncouples liver and heart expression

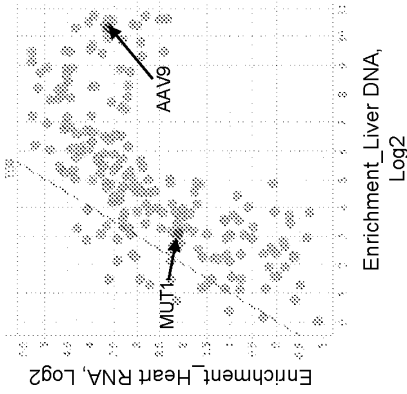
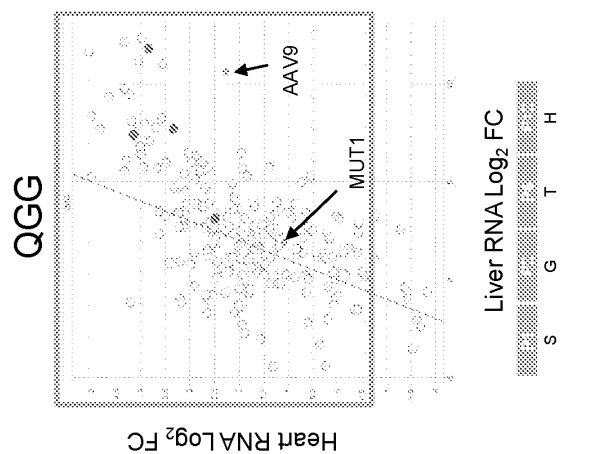
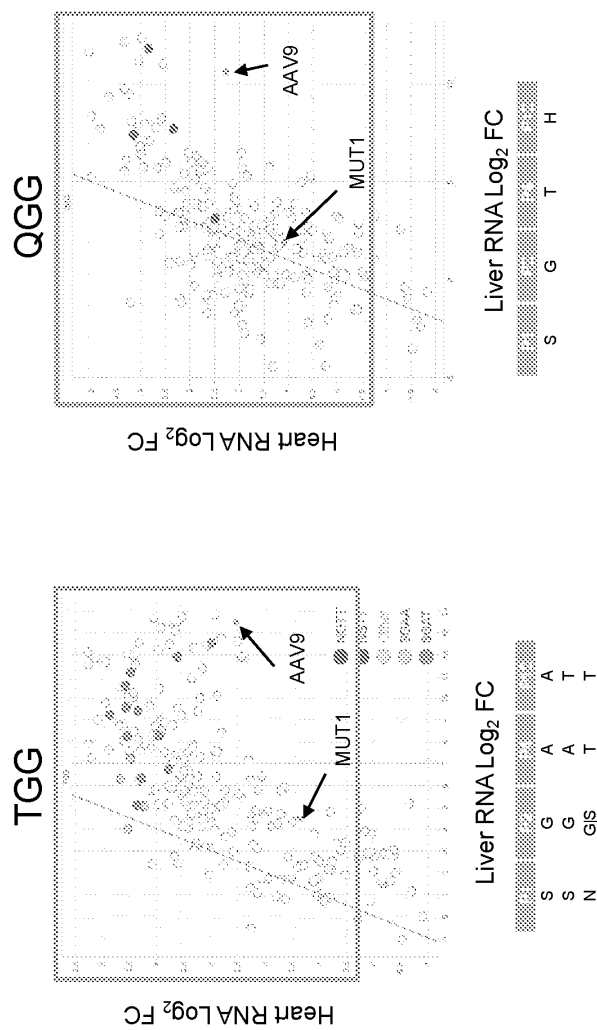
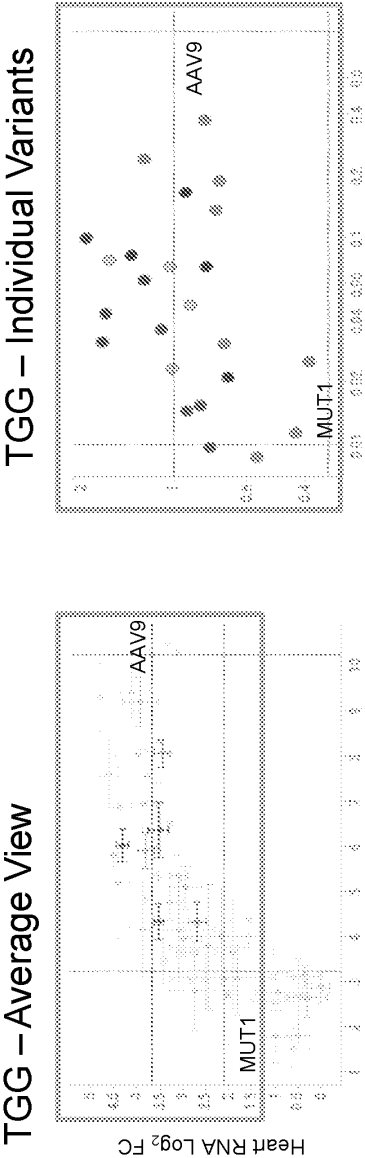


FIG. 58B



* SGAT (purple) chosen for VR8 Myo insert panel based on mid to low levels of liver tropism



Liver RNA Log₂ FC

FIG. 58F



Liver RNA Log₂ FC

FIG. 58G

TGG – VR1 + Myo Mini-Lib Variants

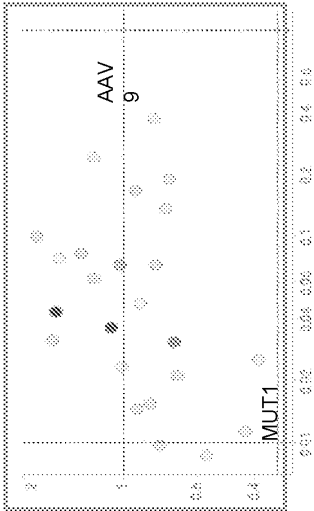


FIG. 58H

TGG – VR1 Mini-Lib Variants

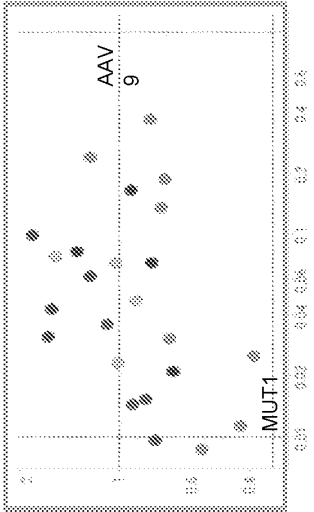


FIG. 58G

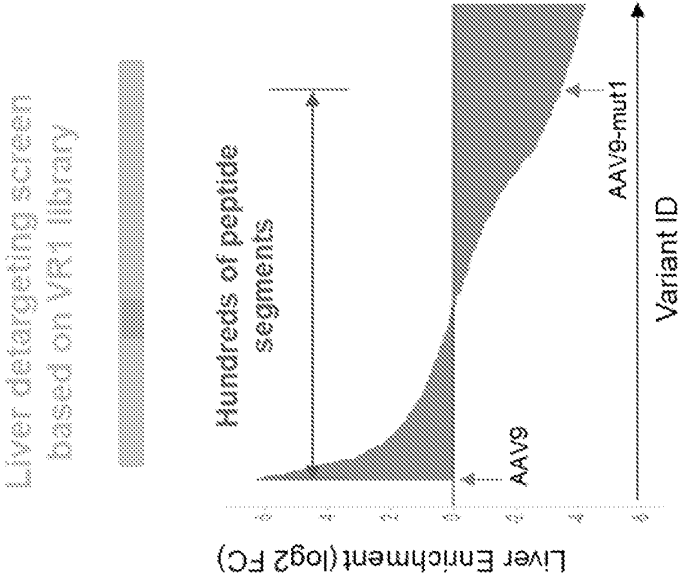


FIG. 59

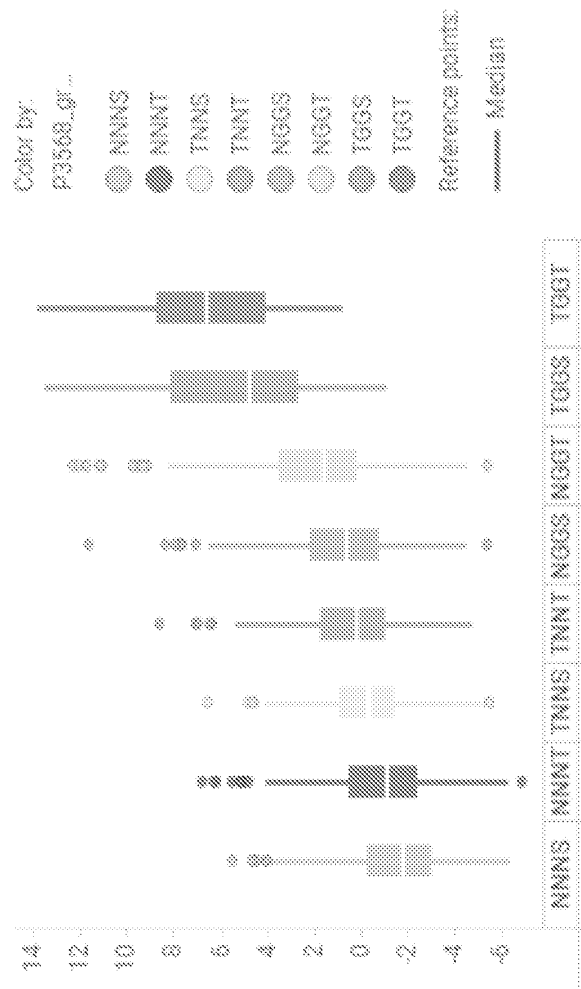


FIG. 60

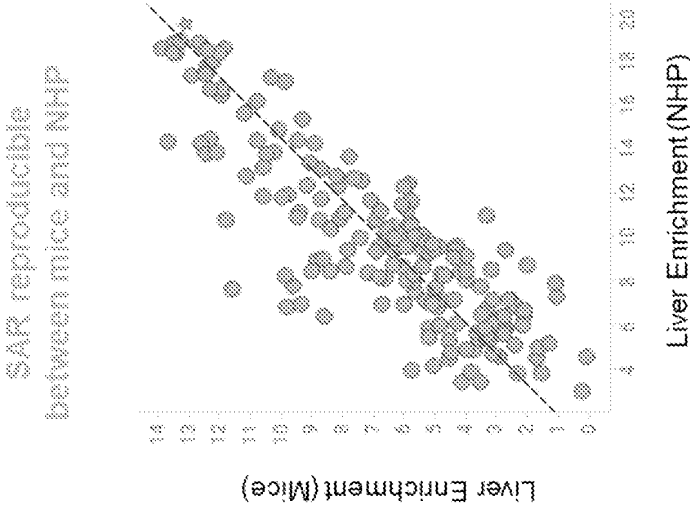


FIG. 61

Variable Triplet			
X1	X2	X3	
A	A	A	
D	D	D	
E	E	E	
N	F	F	
Q	G	G	
T	H	H	
S	I	I	
	K	K	
	L	L	
	M	M	
	N	N	
	P	P	
	Q	Q	
	R	R	
	S	S	
	T	T	
	V	V	
	W	W	
	Y	Y	

FIG. 62A

Constant
RGD

FIG. 62B

Variable Quad	
Category	Quads
(1) 4H7L	HGVL HVNL
(2) 4R7I	RGQI RQGI
(3) 4R7V	RSVV RGVV
(4) 4R7L	RDYL
(5) 4Y7L	YVGL YNSL
(6) 4Y7V	YSSV YTSV
(7) 4Y7M	YTSM
(8) 4L7T	LTVT
Muscle-related	FNNT
	FNNL
	FQNT
Heart specific variants	LIGR
	QSTL
Skeletal muscle on/heart neutral	RSQT
	LLLS

FIG. 62C

VR I Peptide segments
NSTSGASTNDNA (SEQ. ID NO.: 48390)
SGTTGGSSNDNT (SEQ. ID NO.: 46656)
SSTAGGASNDNA (SEQ. ID NO.: 47128)
SSTAGGATNDNA (SEQ. ID NO.: 47131)

FIG. 62D

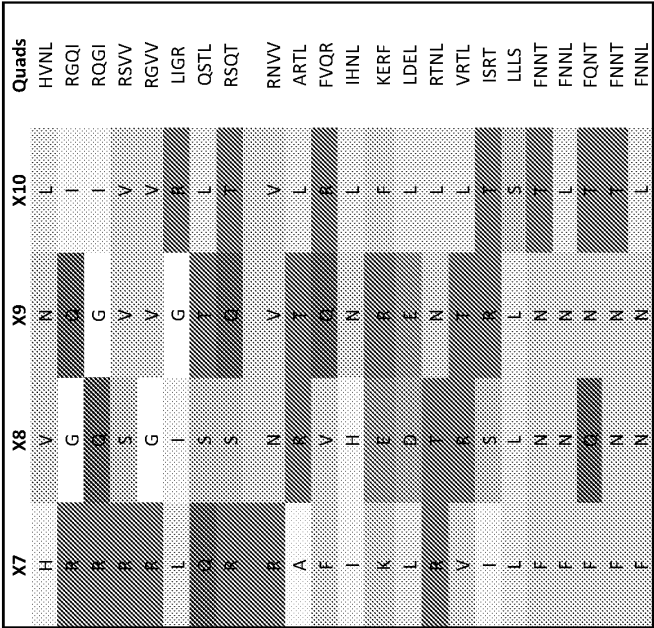


FIG. 62E

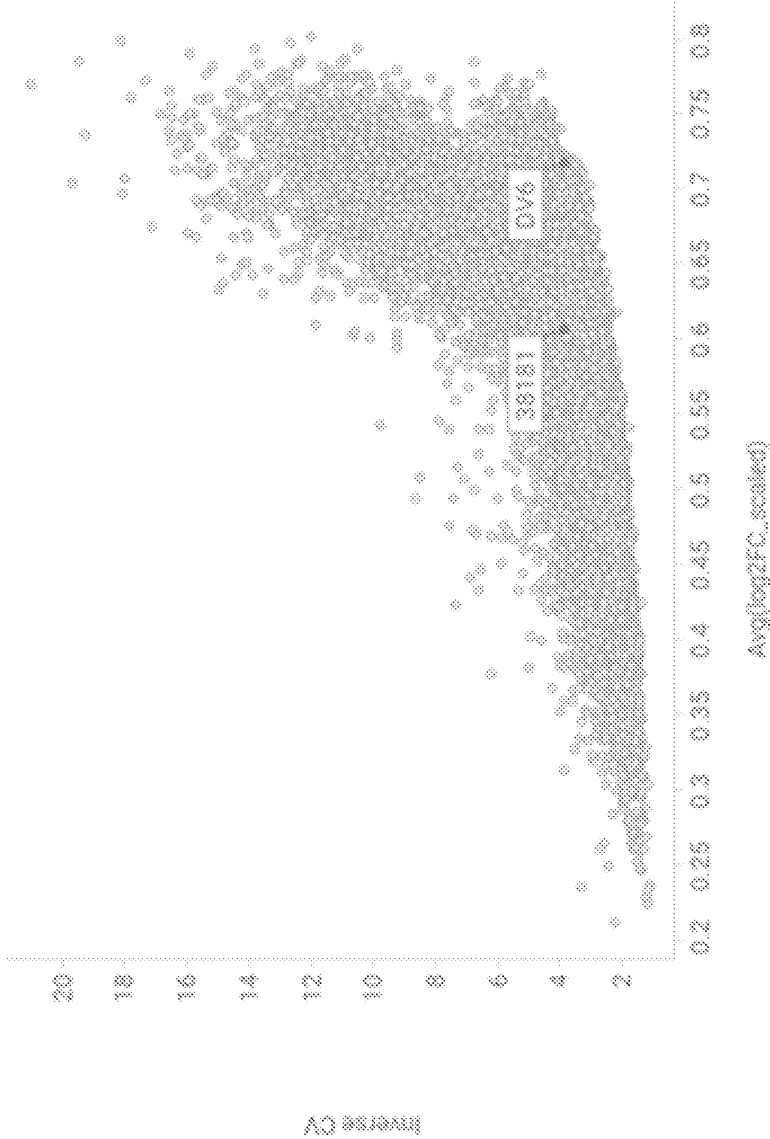


FIG. 63A

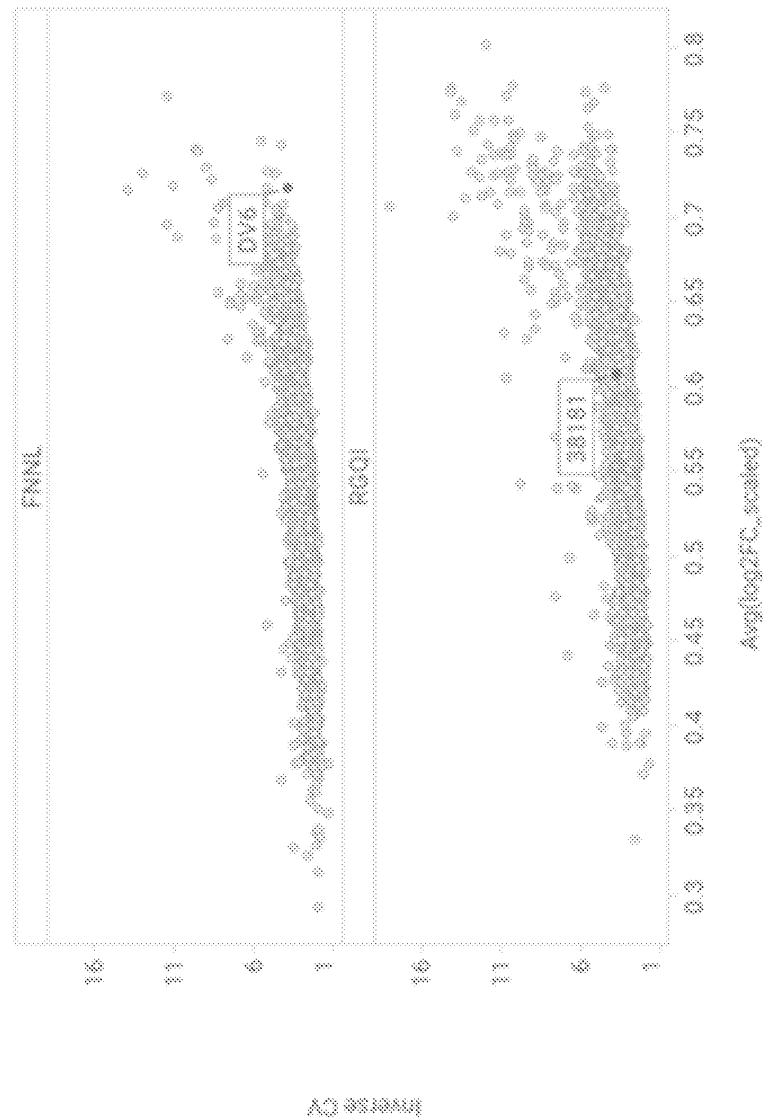


FIG. 63B

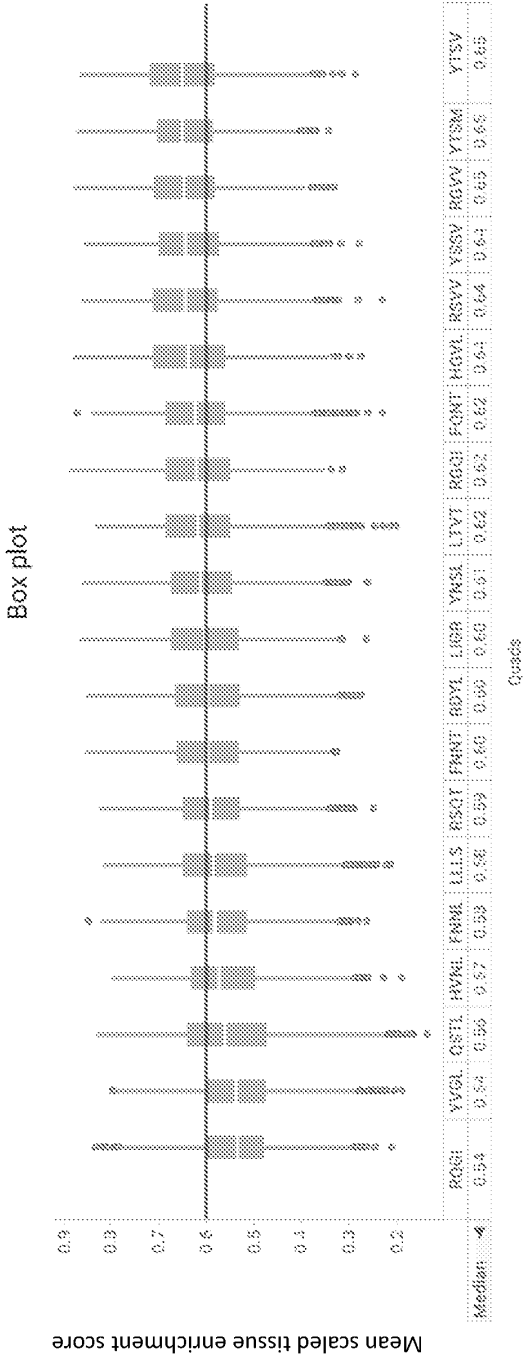


FIG. 64

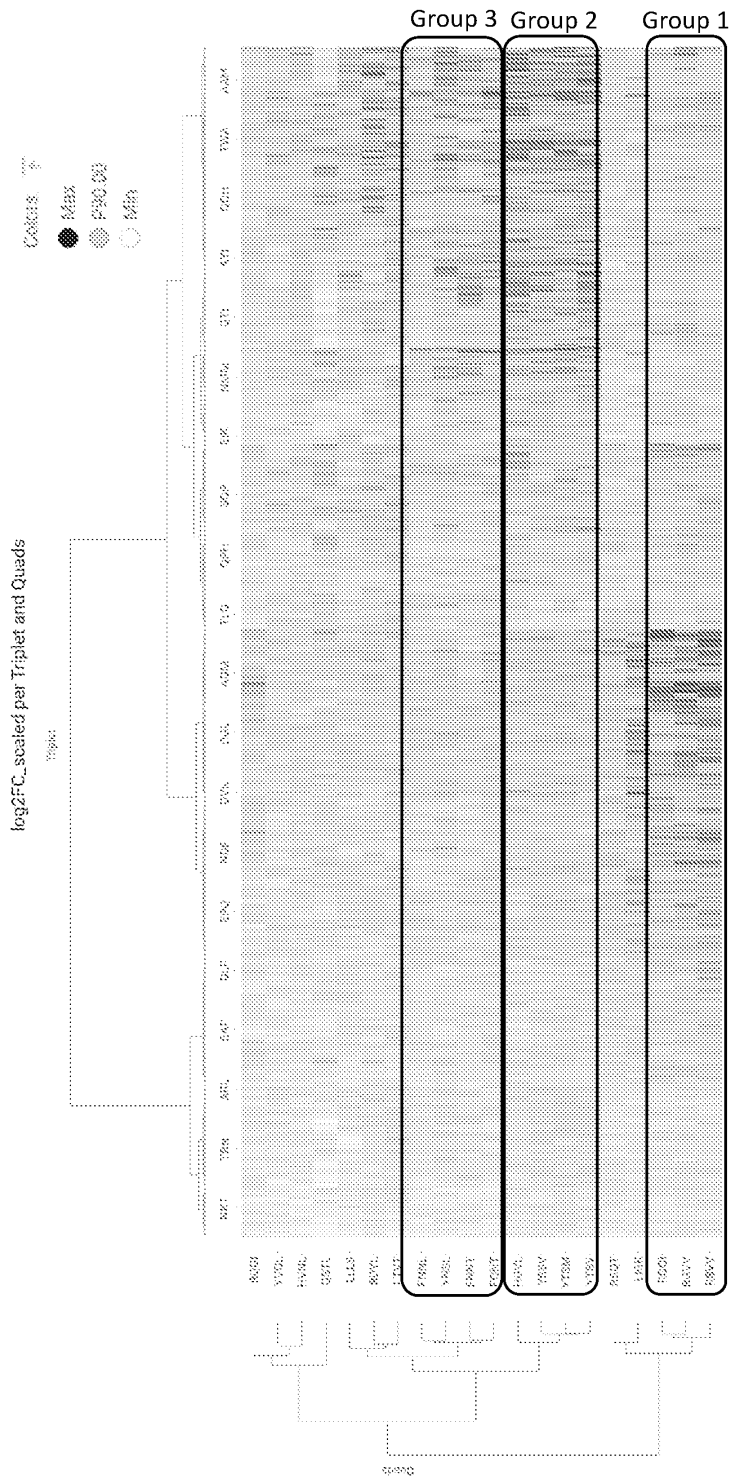


FIG. 65

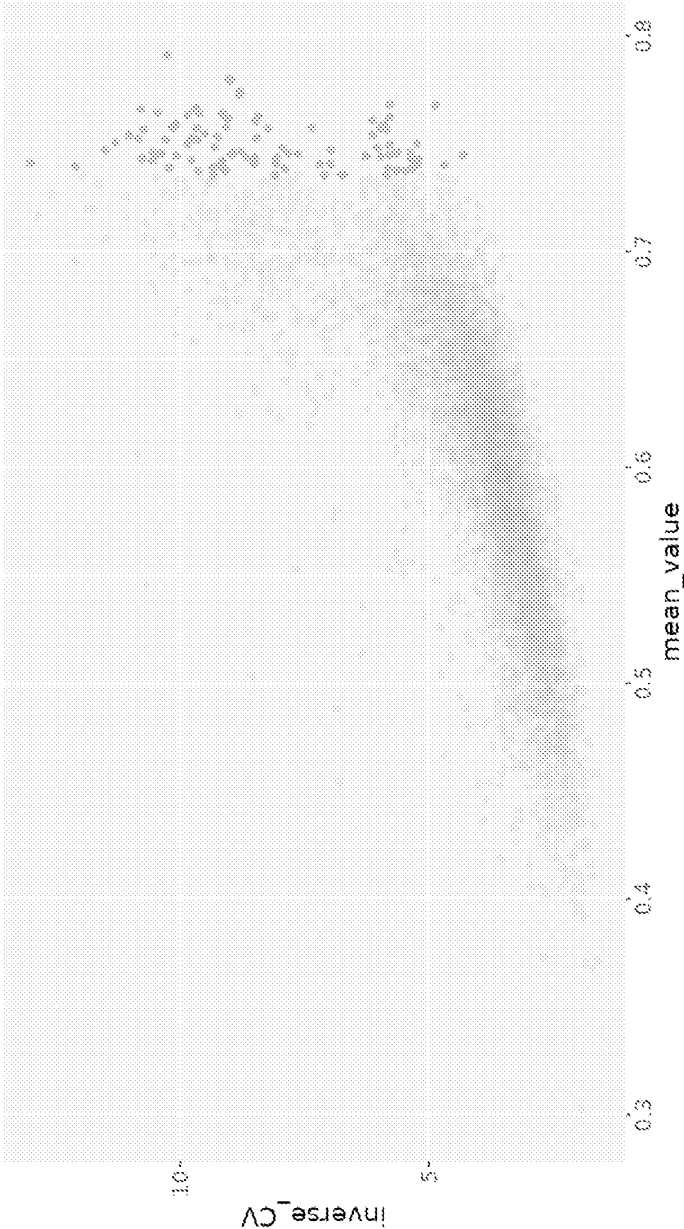


FIG. 66A

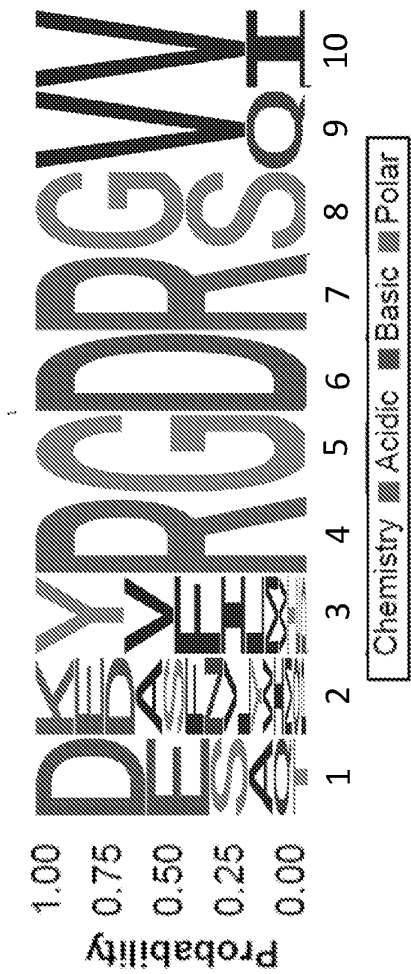


FIG. 66B

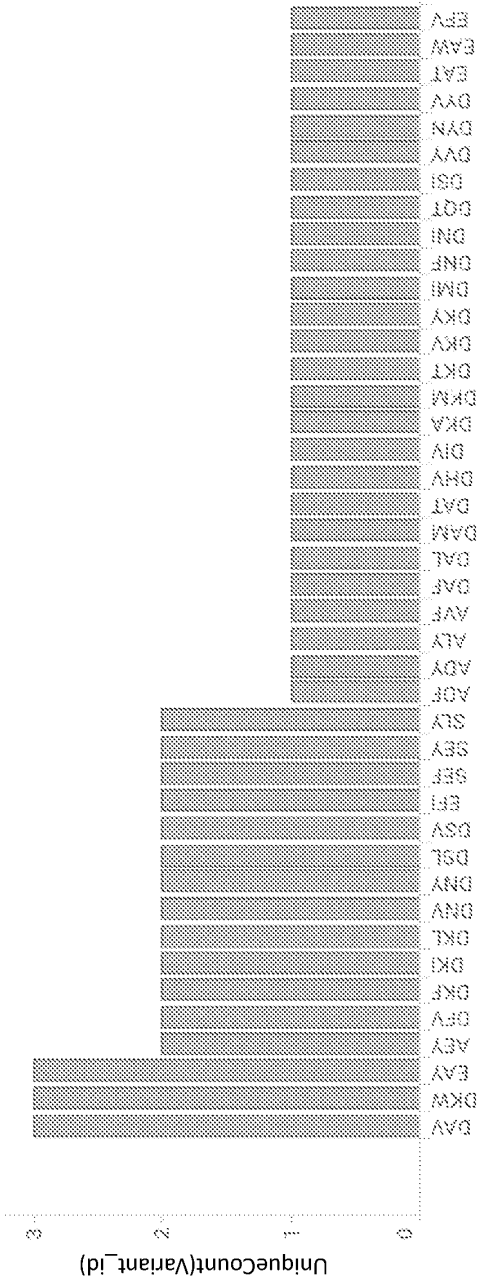
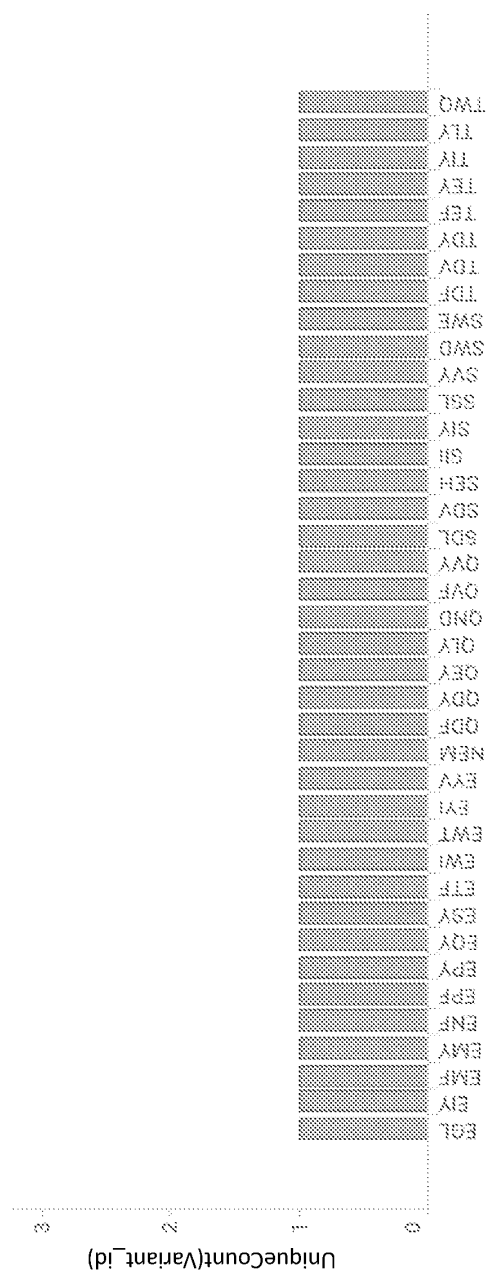


FIG. 66C



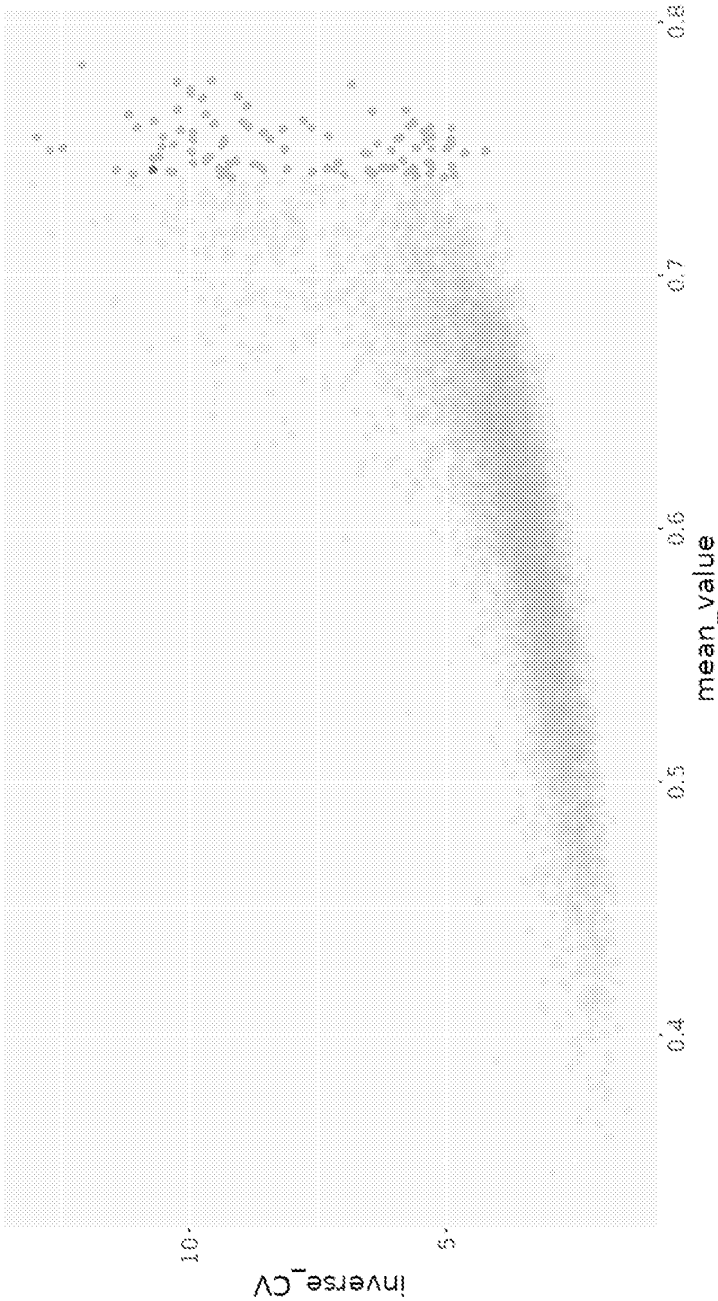


FIG. 67A

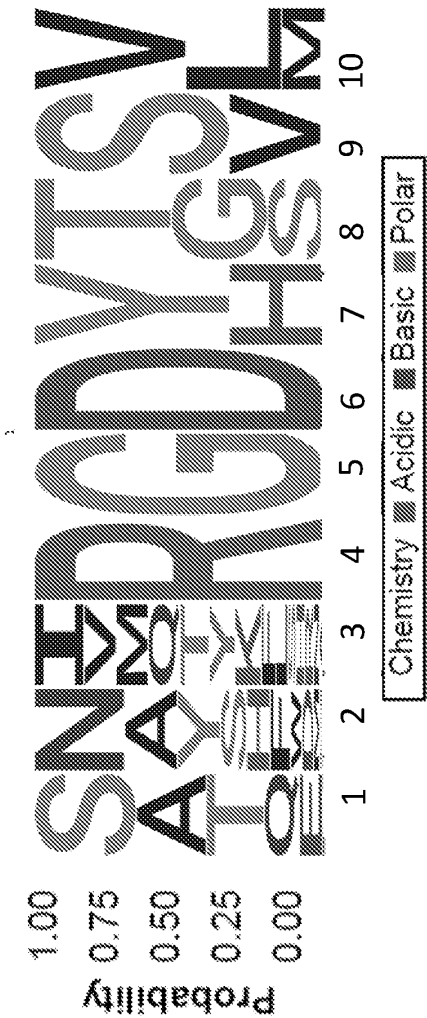
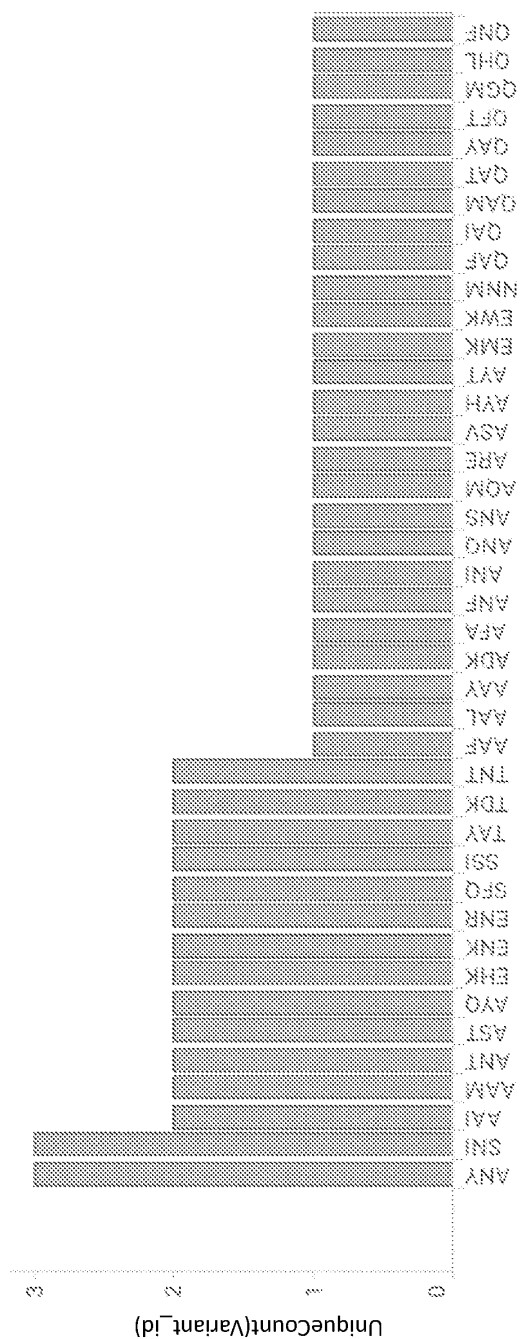


FIG. 67B



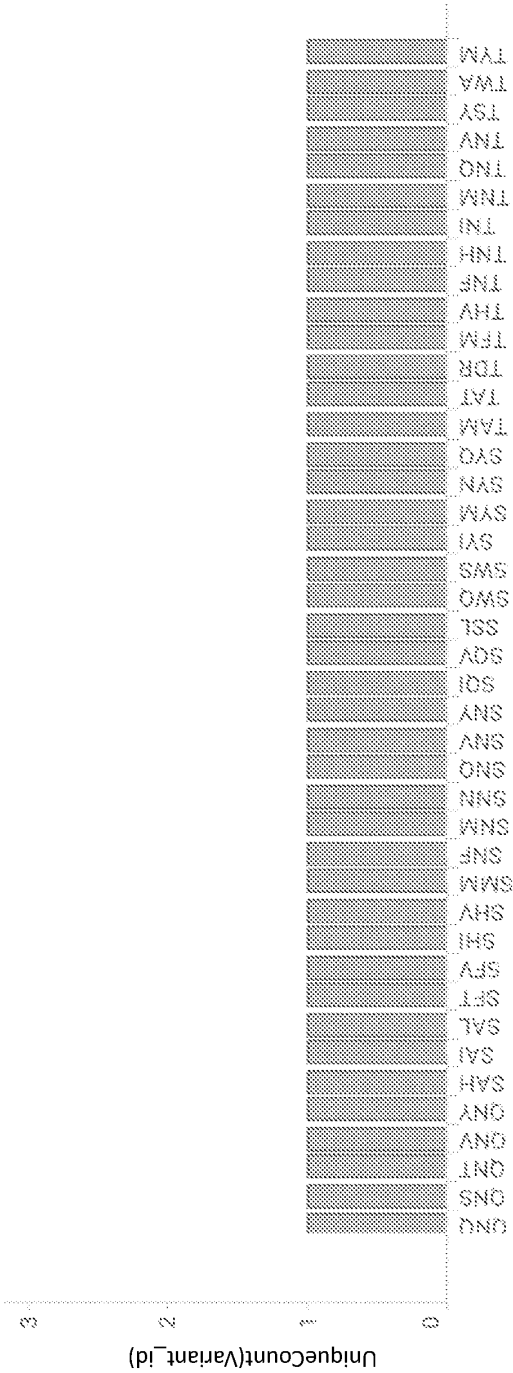


FIG. 67C (Continued)

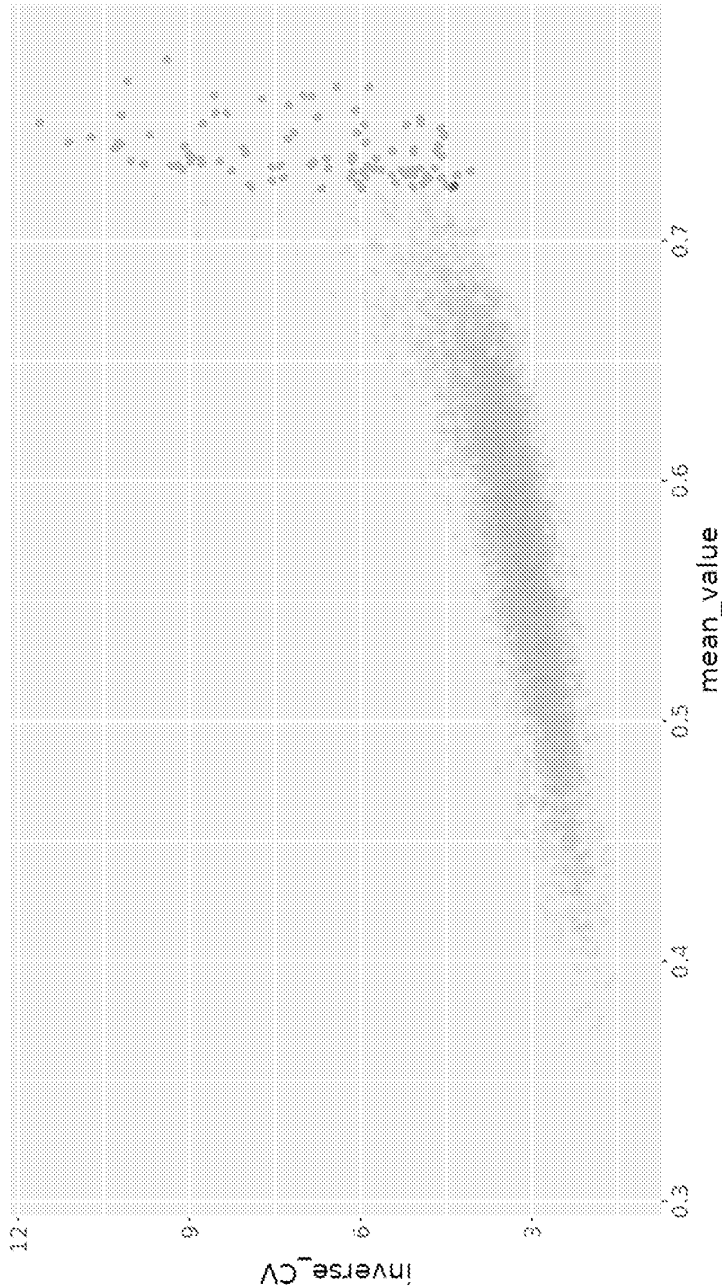


FIG. 68A

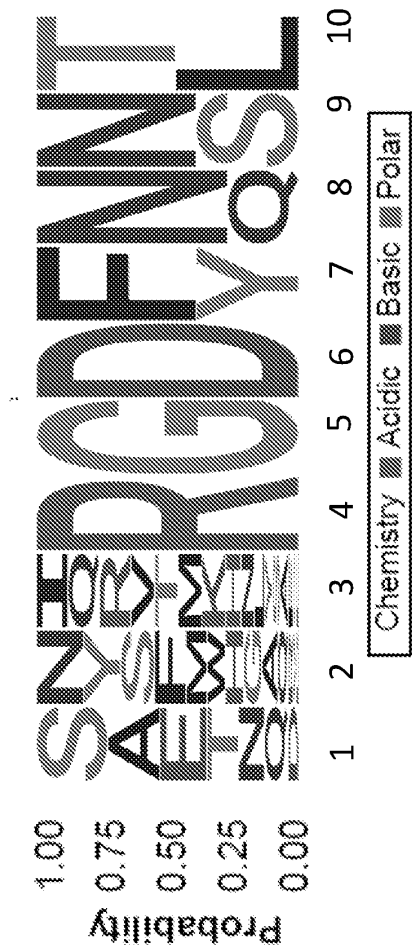
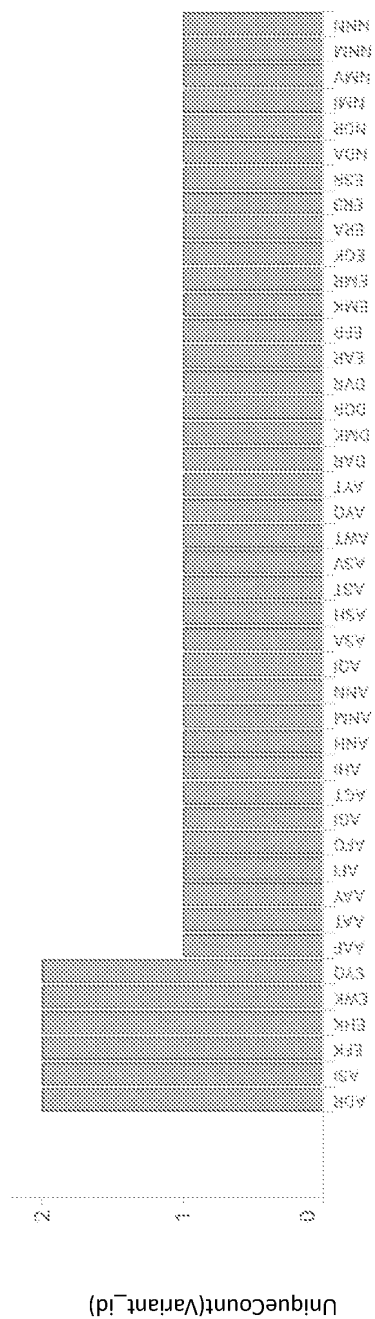


FIG. 68B





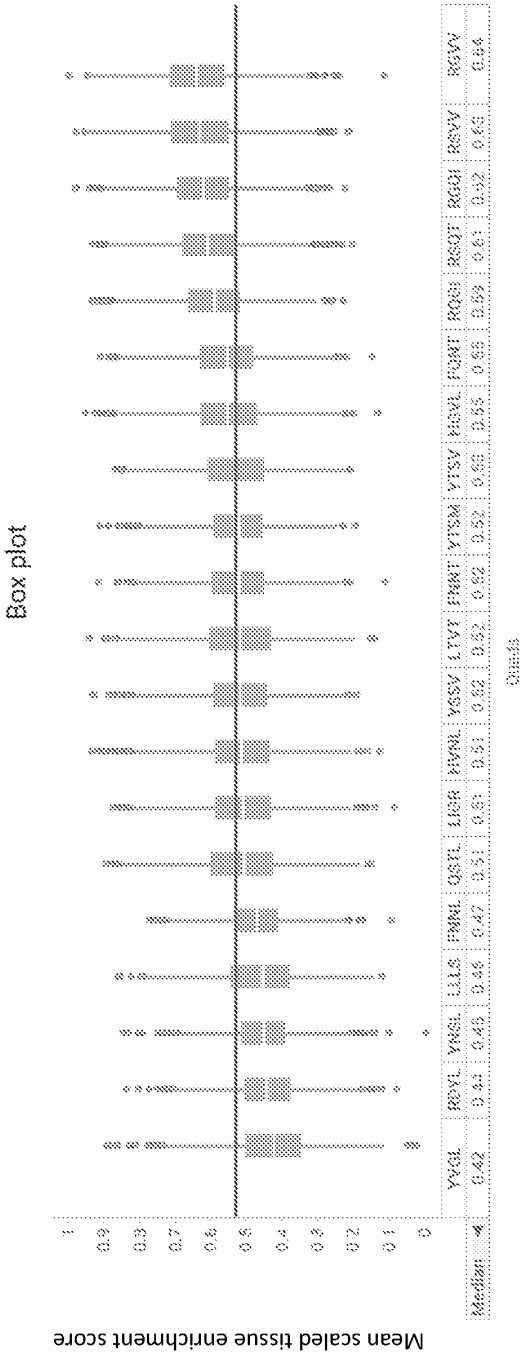


FIG. 69

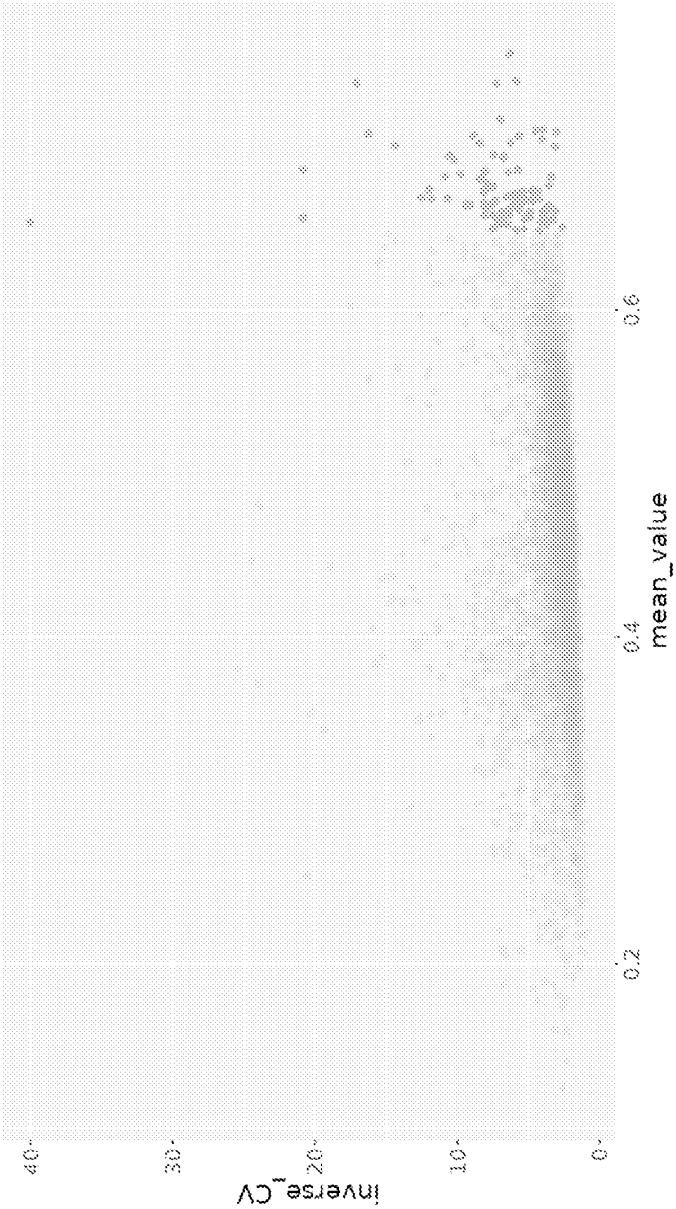


FIG. 70A

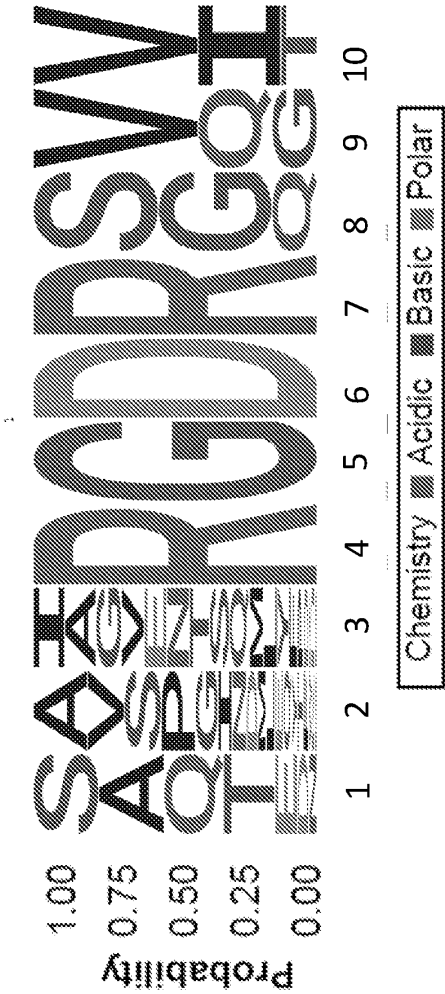


FIG. 70B

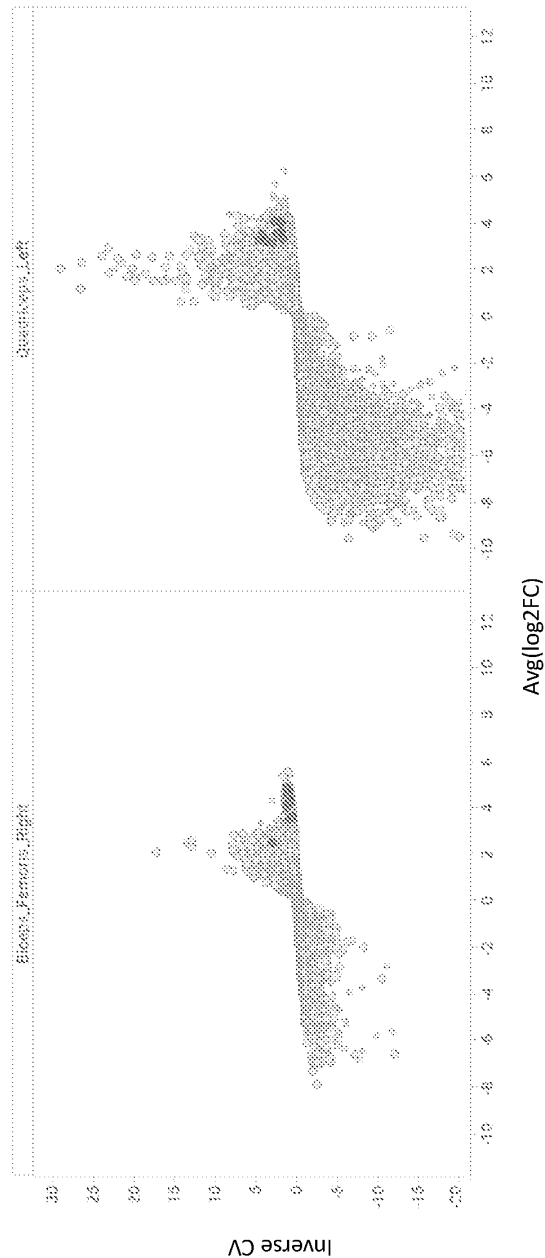


FIG. 71A

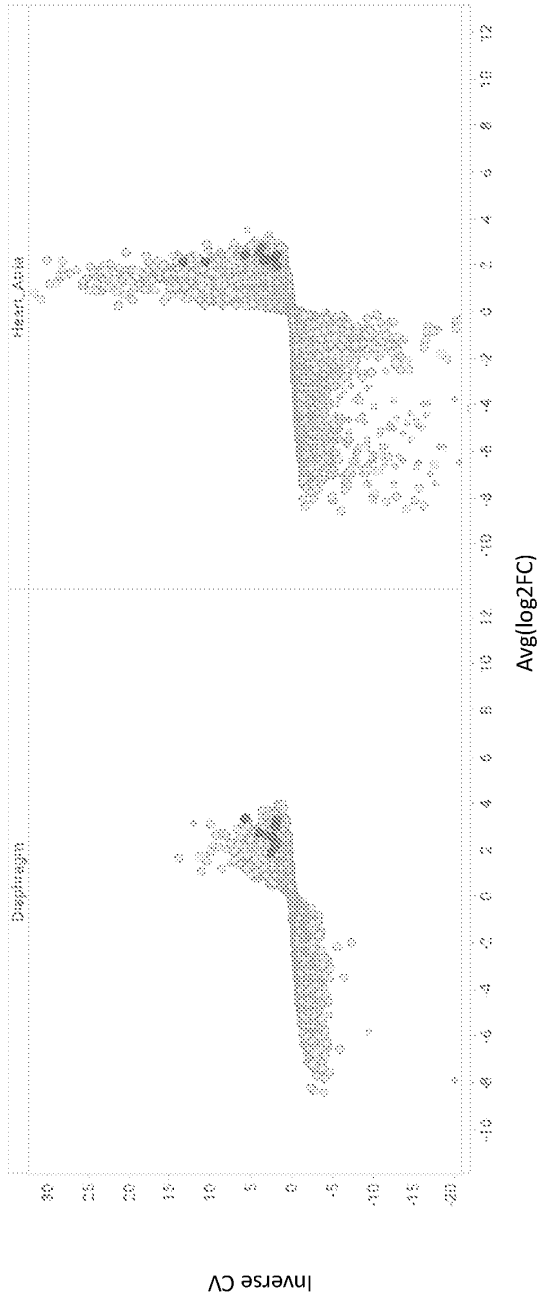


FIG. 71A (Continued)

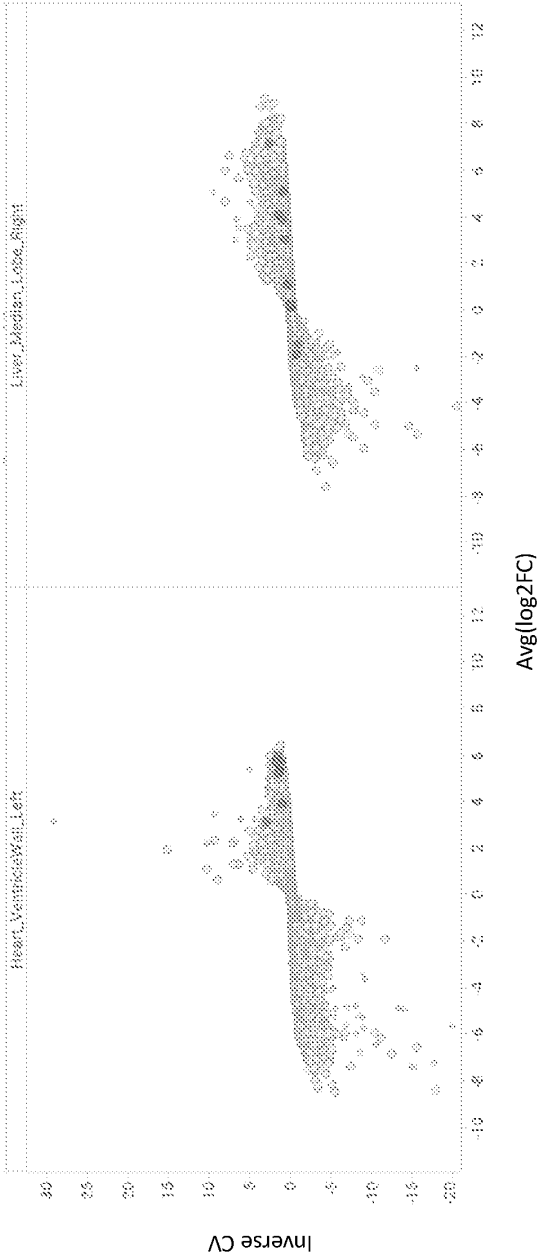


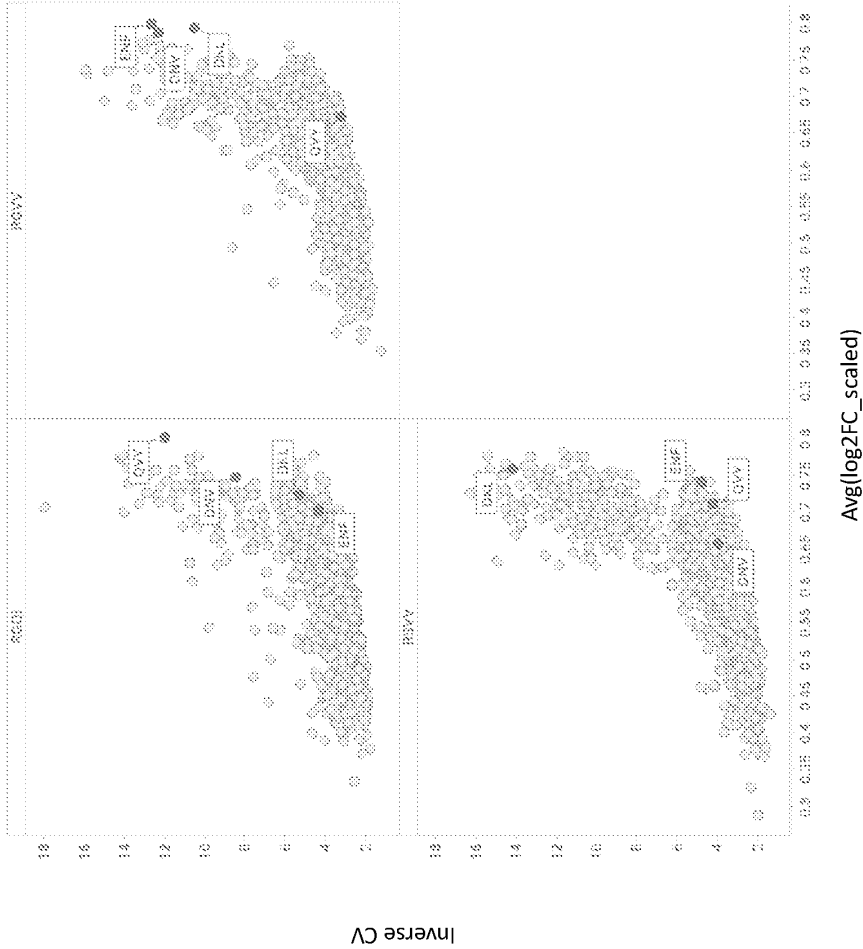
FIG. 71A (Continued)

variant_id	amino	Biceps_Femur is_Right	Quadriceps_L ent	Diaphragm	Heart_Atria	Heart_Ventric isWall_Left	Liver_Median Lobe_Right
ATLV021X847671	QWVRGDTSM	1.6	3.28	2.23	1.98	5.23	7.85
ATLV021X824406	DKL RGRGVV	1.38	2.77	2.24	2.78	5.23	5.86
ATLV021X89859	AAYRGRHGL	0.57	3.09	2.60	2.12	5.78	3.77
ATLV021X832991	EMFRGRGVV	4.18	3.19	2.49	2.46	5.78	4.87
ATLV021X826915	DWVRGRGVV	3.86	3.83	1.82	3.40	5.89	3.87
ATLV021X813226	QVYRGRROQI	6.73	3.84	3.01	2.13	5.28	1.04
ATLV021X846880	EFKRGDFQWI	3.40	3.40	2.36	3.19	5.65	0.19
ATLV021X840720	EMVRGDTSM	0.30	3.73	2.69	2.19	3.73	0.07
ATLV021X843867	DOKRDLIOR	8.73	3.89	2.71	2.38	5.74	1.94
ATLV021X844712	YFMHGDYTSV	2.37	3.84	3.12	2.83	3.79	1.52

FIG. 71B

Quads	variant_id	X ₁	X ₂	X ₃	R	G	D	X ₇	X ₈	X ₉	X ₁₀
FQNT	ATLVT021XX48600	E	F	K	R	G	D	F	Q	N	T
	ATLVT021XX8359	A	A	Y	R	G	D	H	G	V	L
	ATLVT021XX43867	D	Q	K	R	G	D	L	I	G	R
LIGR	ATLVT021XX13226	Q	V	Y	R	G	D	R	G	Q	I
RGQI	ATLVT021XX24406	D	K	L	R	G	D	R	G	V	V
	ATLVT021XX25015	D	N	V	R	G	D	R	G	V	V
	ATLVT021XX32991	E	N	F	R	G	D	R	G	V	V
YTSM	ATLVT021XX40720	E	W	K	R	G	D	Y	T	S	M
	ATLVT021XX47671	Q	N	V	R	G	D	Y	T	S	M
	ATLVT021XX44712	T	F	M	R	G	D	Y	T	S	V

FIG. 71C



	X ₁	X ₂	X ₃	R	G	D	X ₇	X ₈	X ₉	X ₁₀
RGQI	Q	V	Y	R	G	D	R	G	Q	I
RGW	D	K	L	R	G	D	R	G	V	V
	D	N	V	R	G	D	R	G	V	V
	E	N	F	R	G	D	R	G	V	V

FIG. 72B

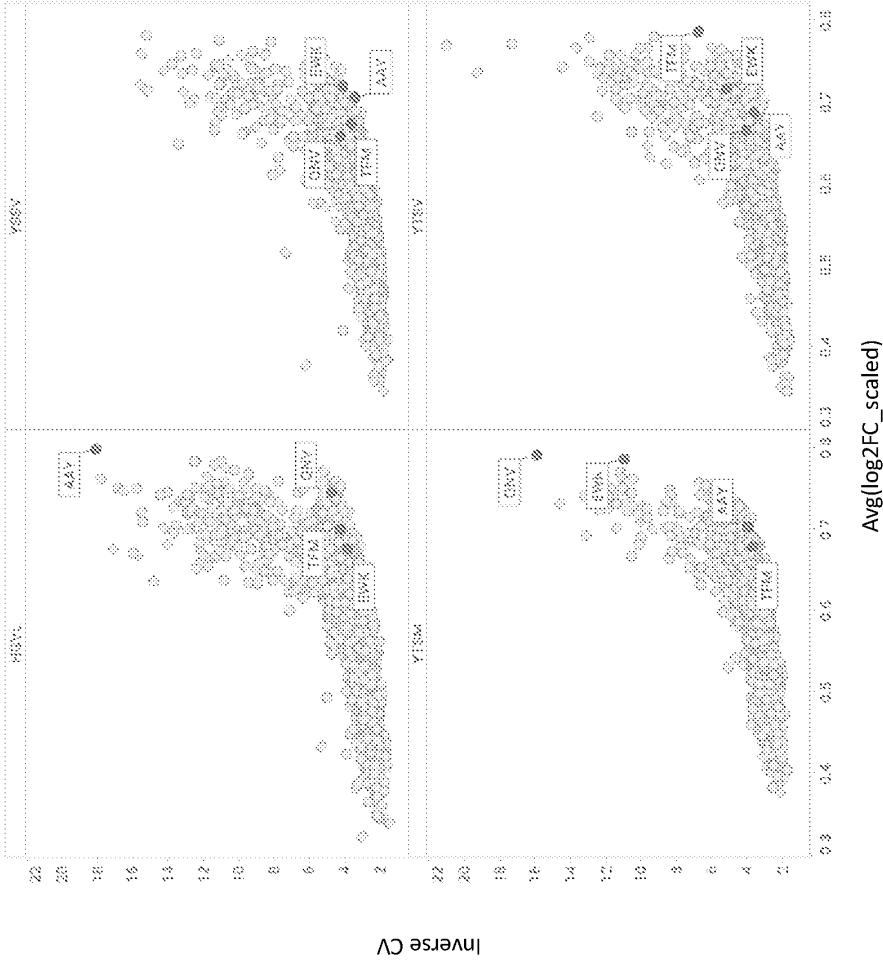
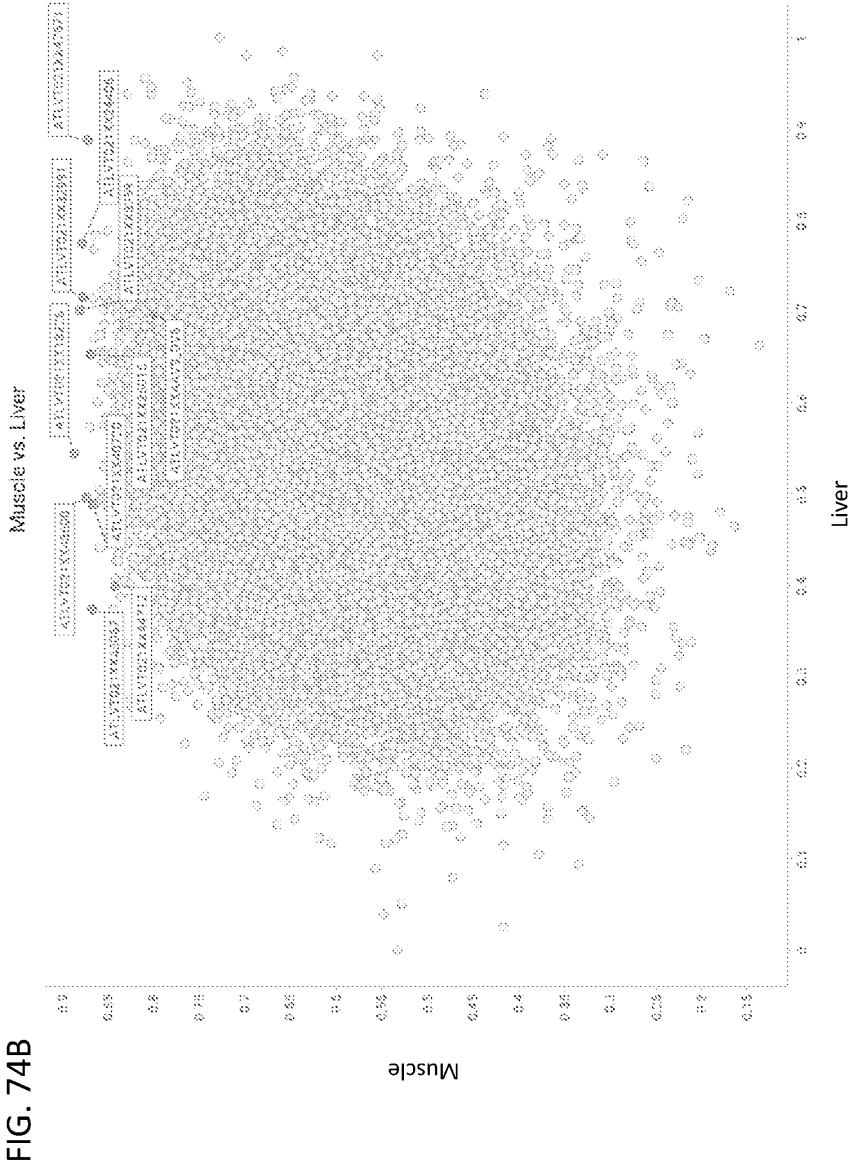


FIG. 73A

	X ₁	X ₂	X ₃	R	G	D	X ₇	X ₈	X ₉	X ₁₀
HGVL	A	A	Y	R	G	D	H	G	V	L
YTSM	E	W	K	R	G	D	Y	T	S	M
	Q	N	V	R	G	D	Y	T	S	M
YTSV	T	F	M	R	G	D	Y	T	S	V

FIG. 73B



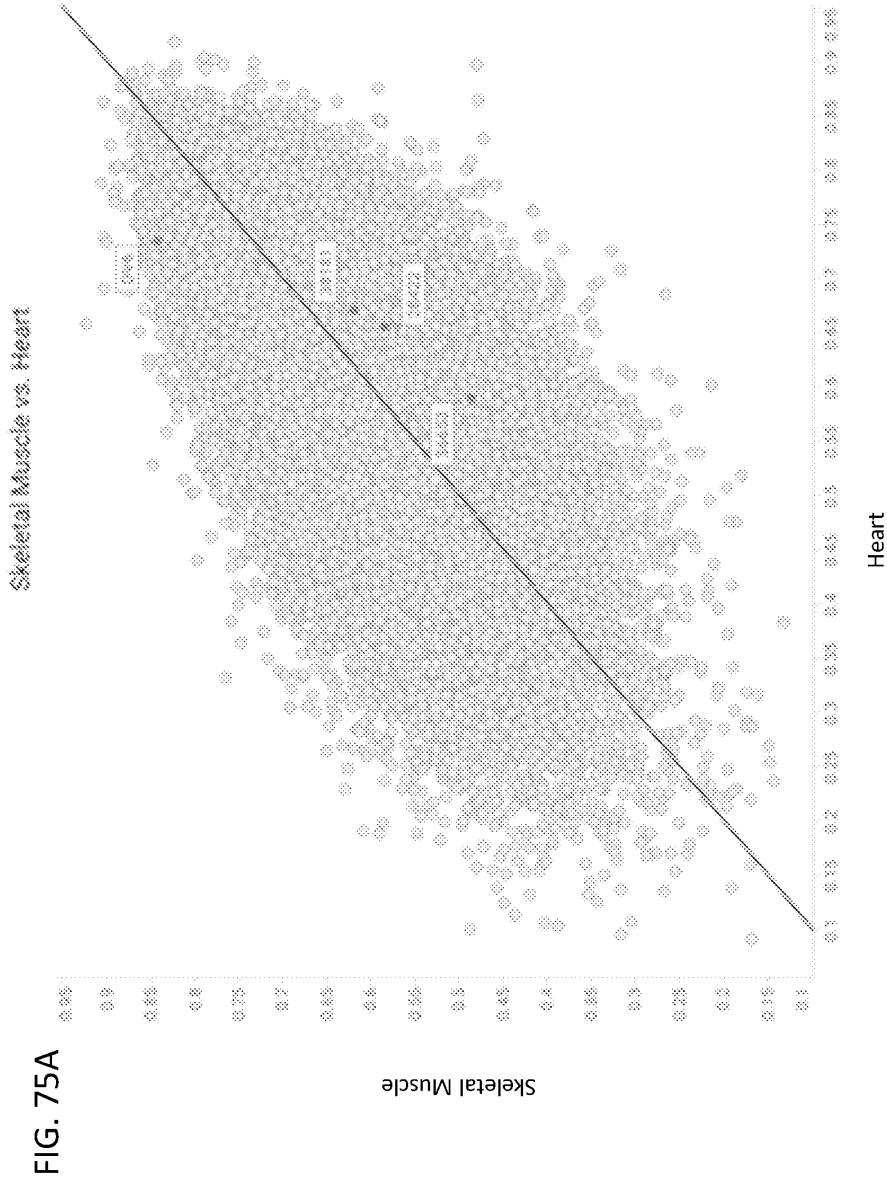


FIG. 75B

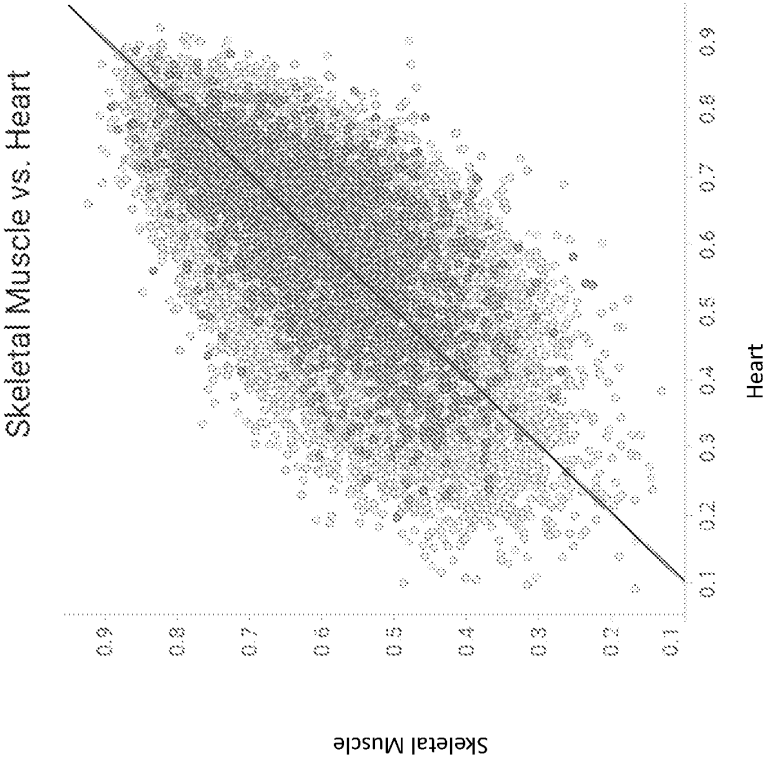


FIG. 75C

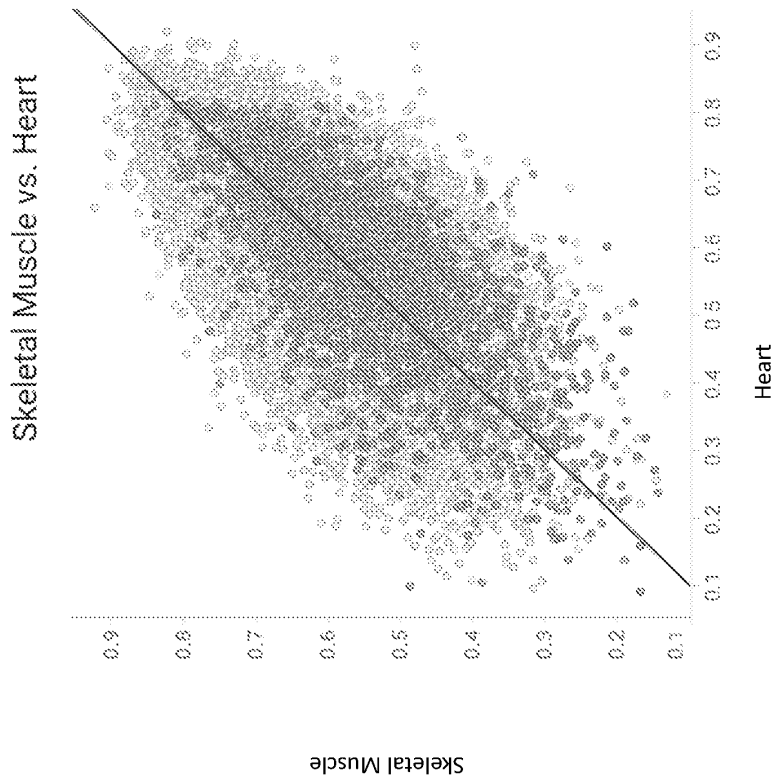


FIG. 75D

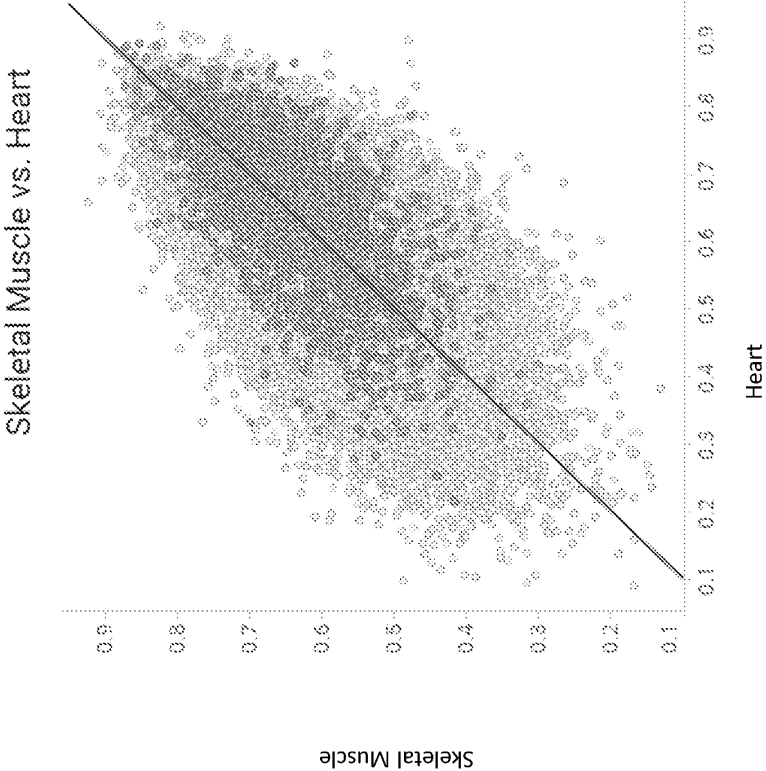
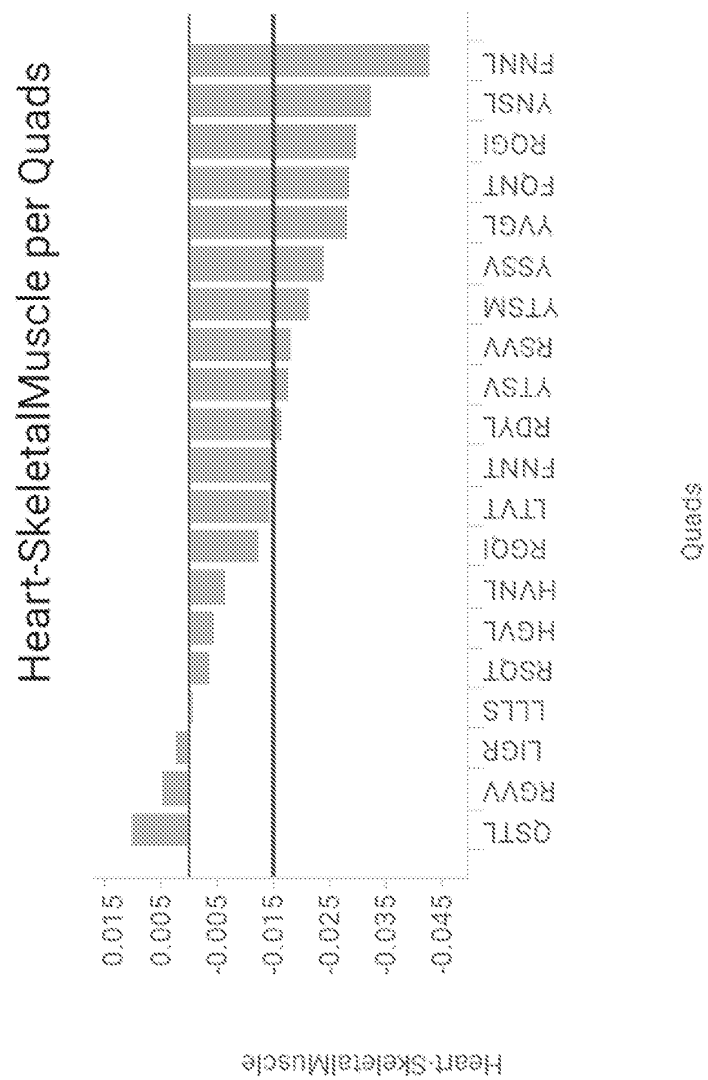


FIG. 75E



Variable Triplet			
X1	X2	X3	
A	A	A	
D	D	D	
E	E	E	
N	F	F	
Q	G	G	
T	H	H	
S	I	I	
	K	K	
	L	L	
	M	M	
	N	N	
	P	P	
	Q	Q	
	R	R	
	S	S	
	T	T	
	V	V	
	W	W	
	Y	Y	

FIG. 76A

Functional Category	VR1	VR8 Insert Name	Conserved	Quads
Overall muscle-on	mut1	20169	RGD	HVNL
Overall muscle-on	mut1	38181	RGD	RGQI
Overall muscle-on	mut1	38249	RGD	RGVV
Overall muscle-on	mut1	39441	RGD	RQGI
Overall muscle-on	mut1	40049	RGD	RSVV
Overall muscle-on	mut1	2679	RGD	ARTL
Overall muscle-on	mut1	39374	RGD	RNVV
Overall muscle-on	50	1000	RGD	FNNT
Overall muscle-on	49	DV6	RGD	FNNL
Overall muscle-on	47	DV6	RGD	FNNL
Overall muscle-on	47	DV8	RGD	FQNT
Overall muscle-on	47	1000	RGD	FNNT
Heart specific	mut1	28422	RGD	LIGR
Heart specific	mut1	36653	RGD	OSTL
Heart specific	mut1	13437	RGD	FVQR
Heart specific	mut1	21519	RGD	IHNL
Heart specific	mut1	24244	RGD	KERF
Heart specific	mut1	27264	RGD	LDEL
Heart specific	mut1	40193	RGD	RTNL
Heart specific	mut1	49928	RGD	VRTL
Skeletal muscle specific	mut1	39988	RGD	RSQT
Skeletal muscle specific	mut1	23129	RGD	ISRT
Control	mut1	Deco1	RGD	LLLS

FIG. 76B

VR I Peptide segments	
mut1	NSTSGASTNDNA (SEQ ID NO: 48390)
47	SGTTGGSSNDNT (SEQ ID NO: 46656)
49	SSTAGGASNDNA (SEQ ID NO: 47128)
50	SSTAGGATNDNA (SEQ ID NO: 47131)

FIG. 76C

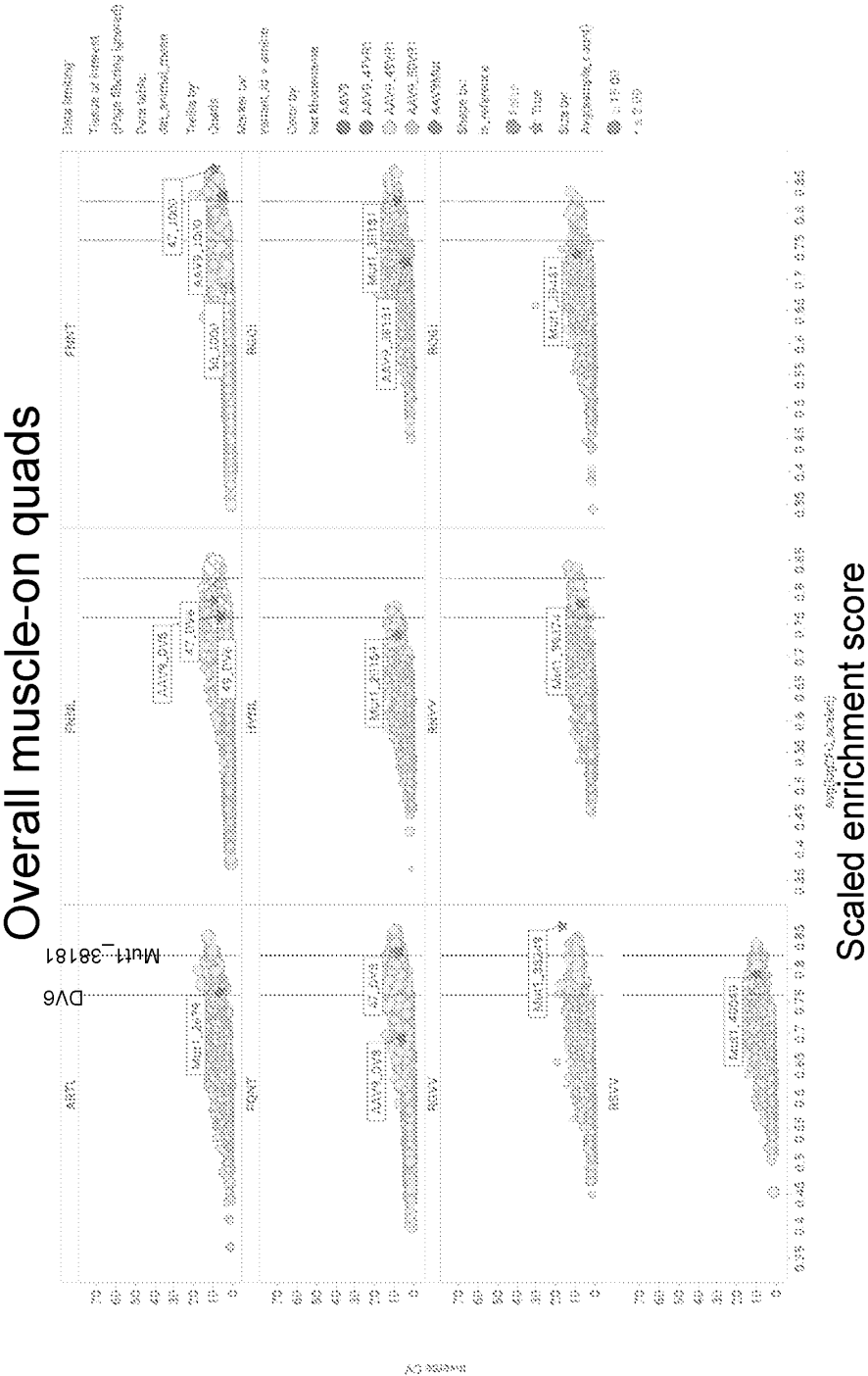


FIG. 77

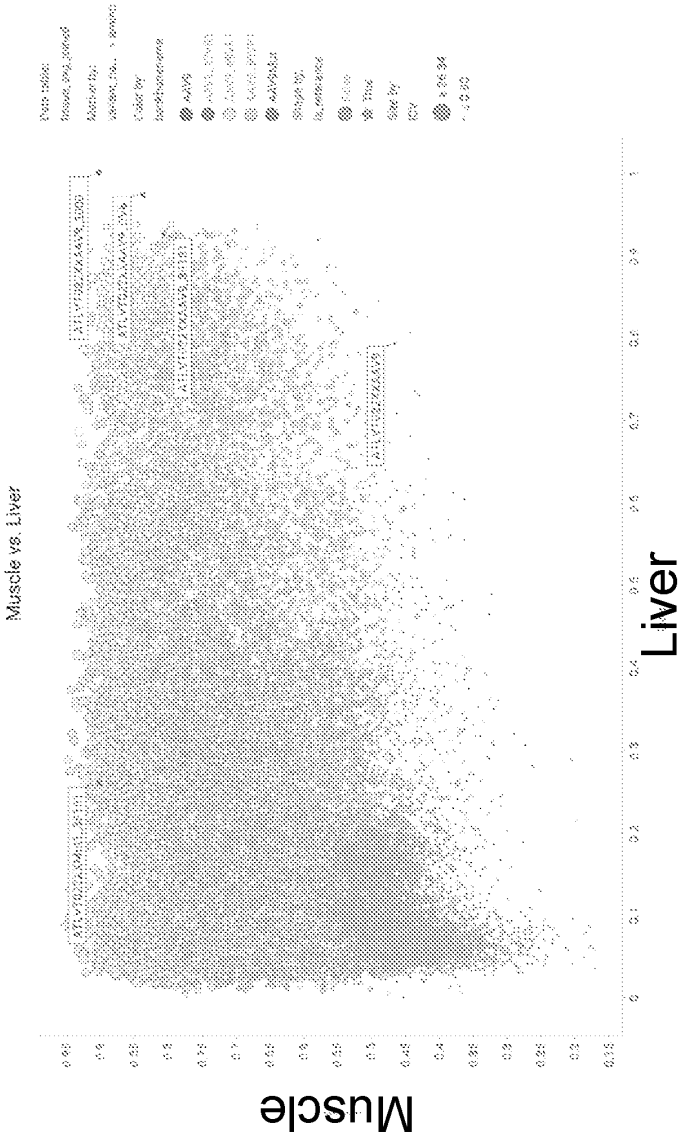


FIG. 78A

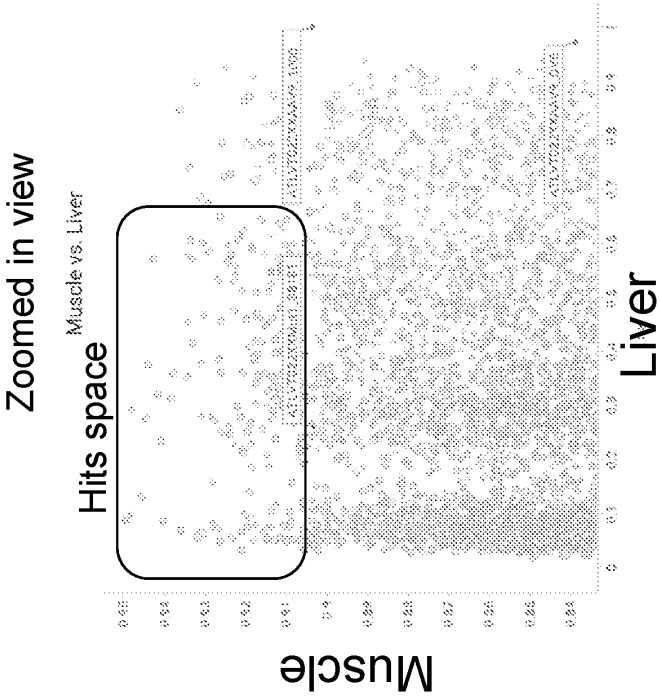


FIG. 78B

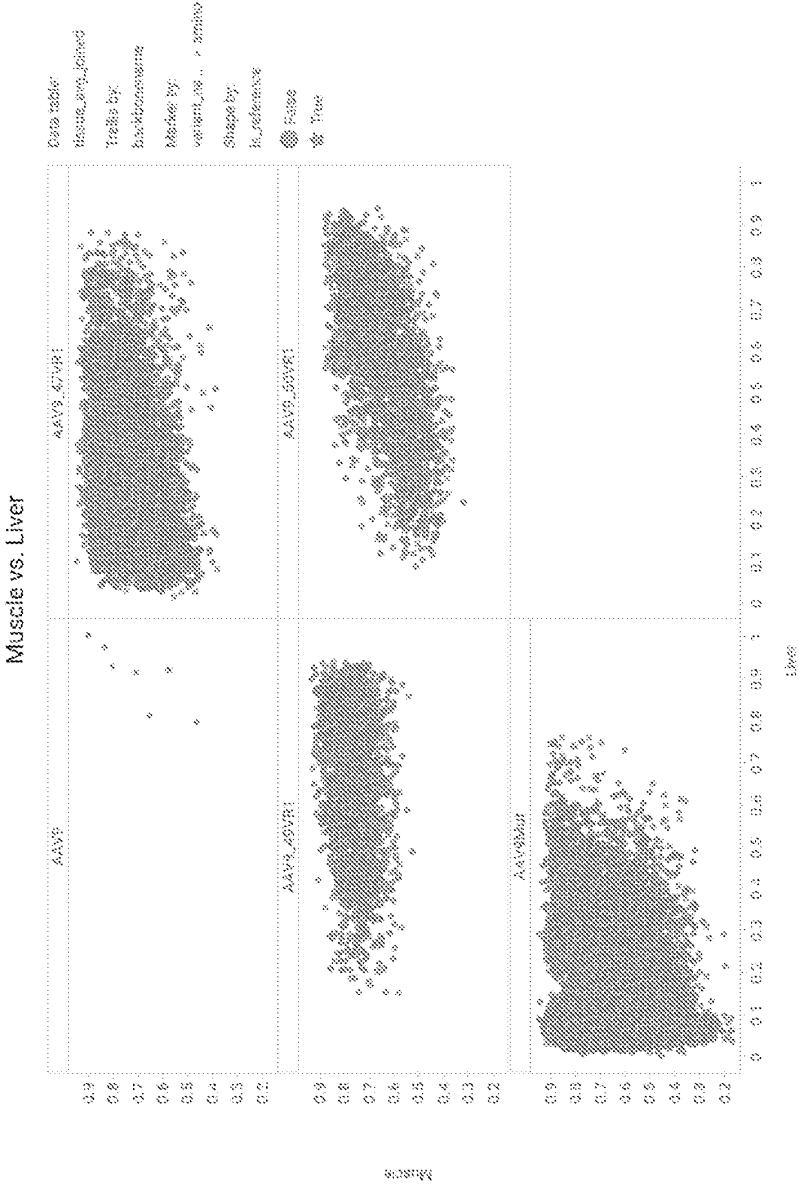


FIG. 78C

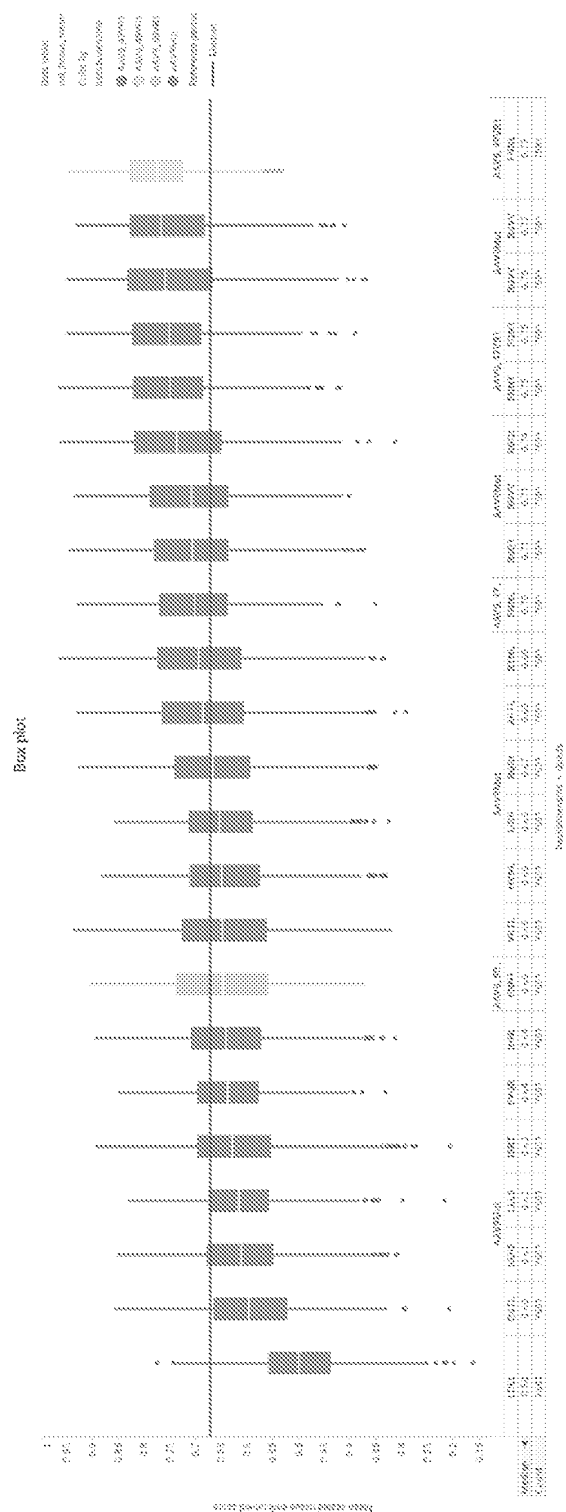


FIG. 79

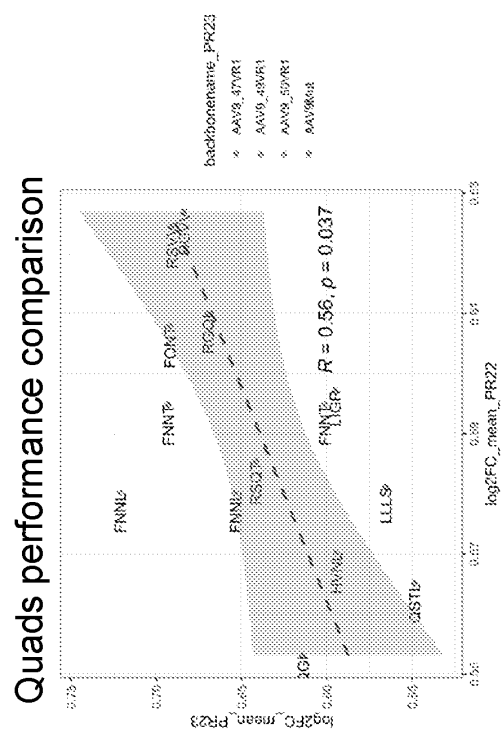


FIG. 80

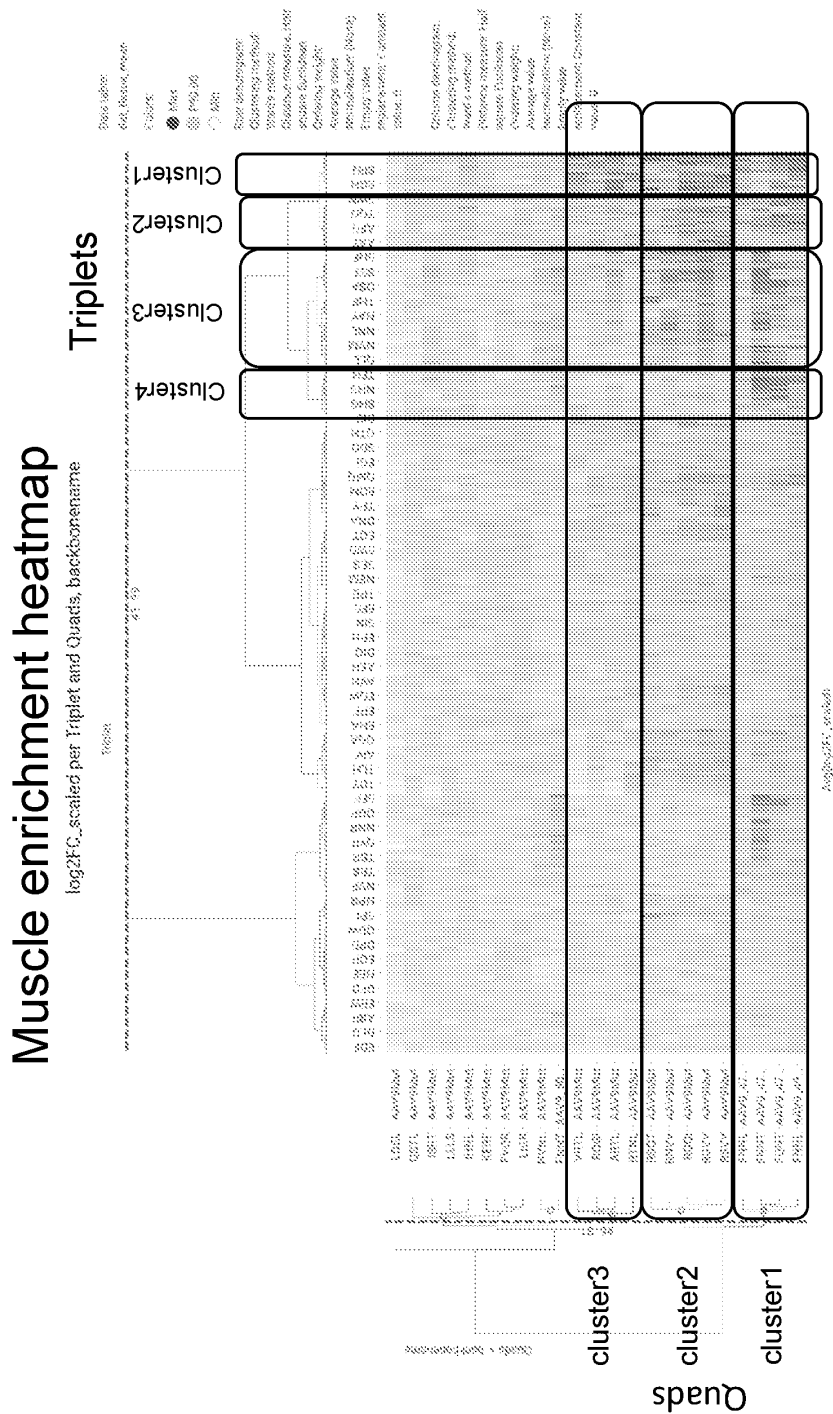


FIG. 81

Quads cluster1

Triplet cluster1

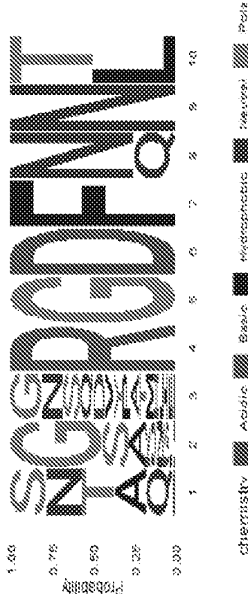


FIG. 82A

Triplet cluster3

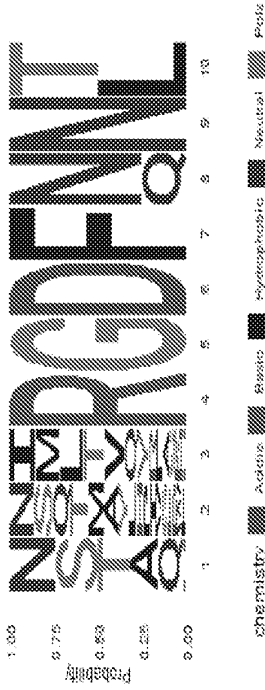


FIG. 82C

Triplet cluster2

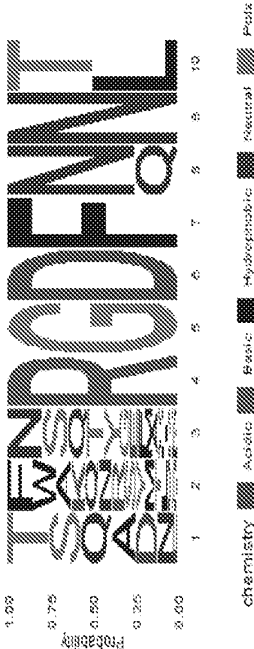


FIG. 82B

Triplet cluster4

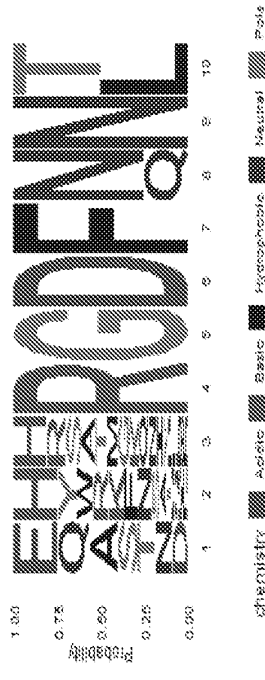


FIG. 82D

Quads cluster2

Triplet cluster1

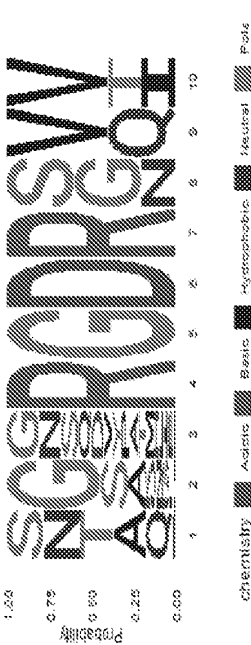


FIG. 83A

Triplet cluster2

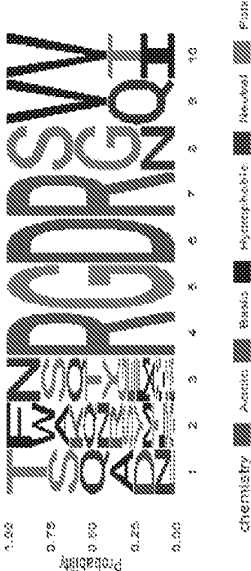


FIG. 83B

Triplet cluster3

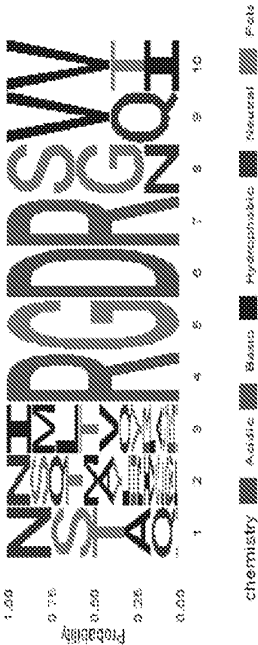


FIG. 83C

Quads cluster3

Triplet cluster1



FIG. 84

Liver detargeting by Quads

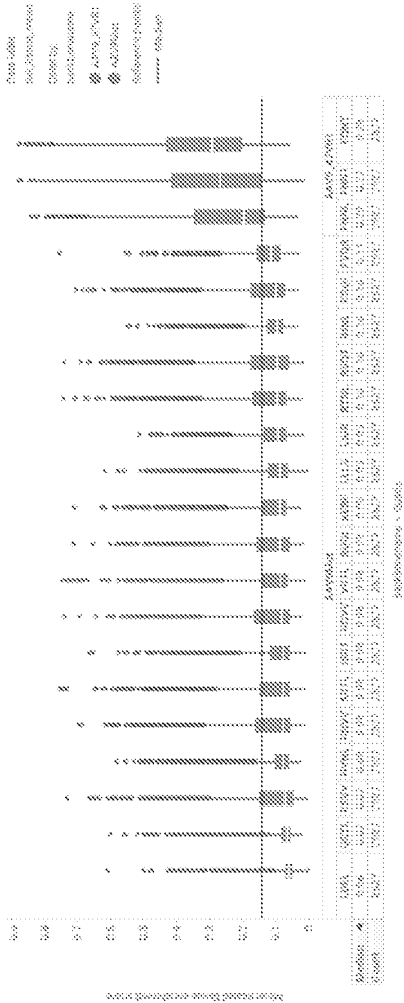


FIG. 85B

Liver enrichment heatmap
log2FC_scaled per Triplet and Quads, backbonename

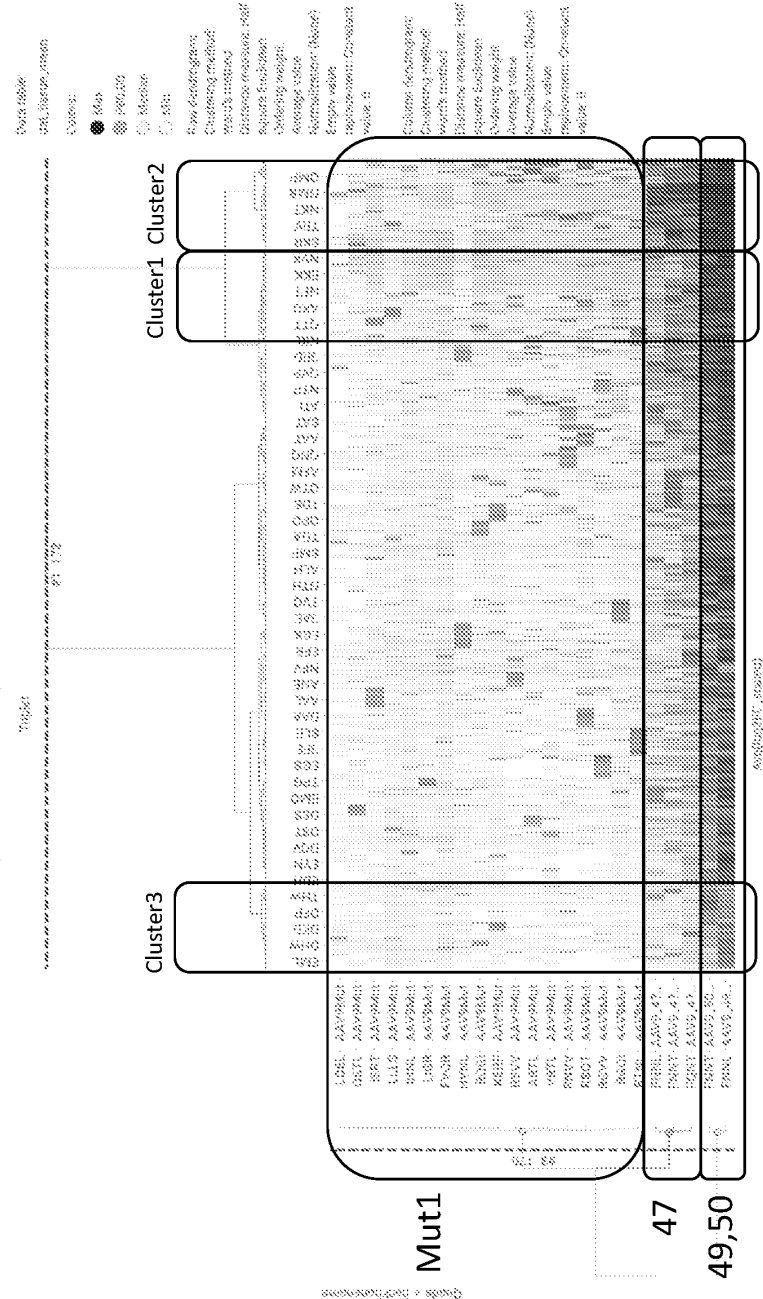


FIG. 86

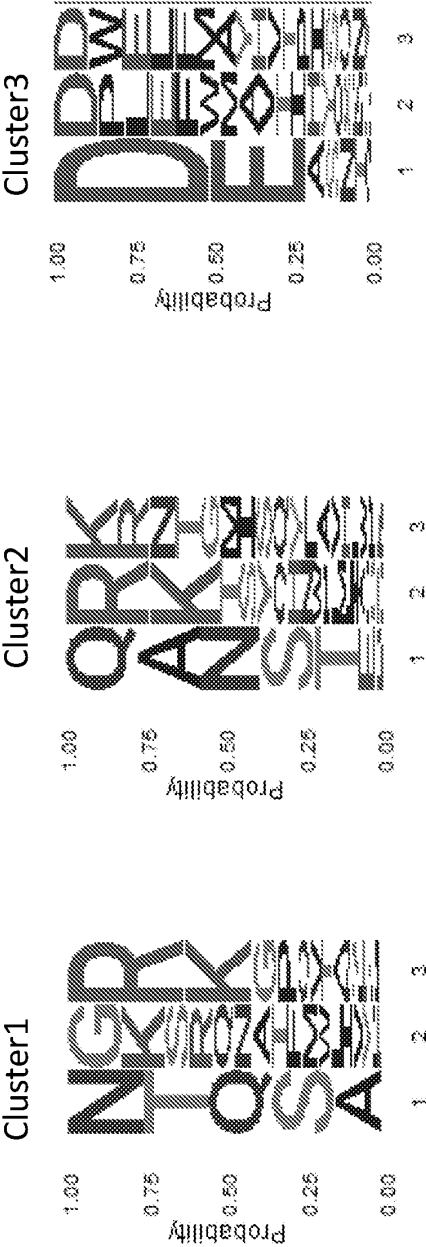


FIG. 87A

FIG. 87B

FIG. 87C

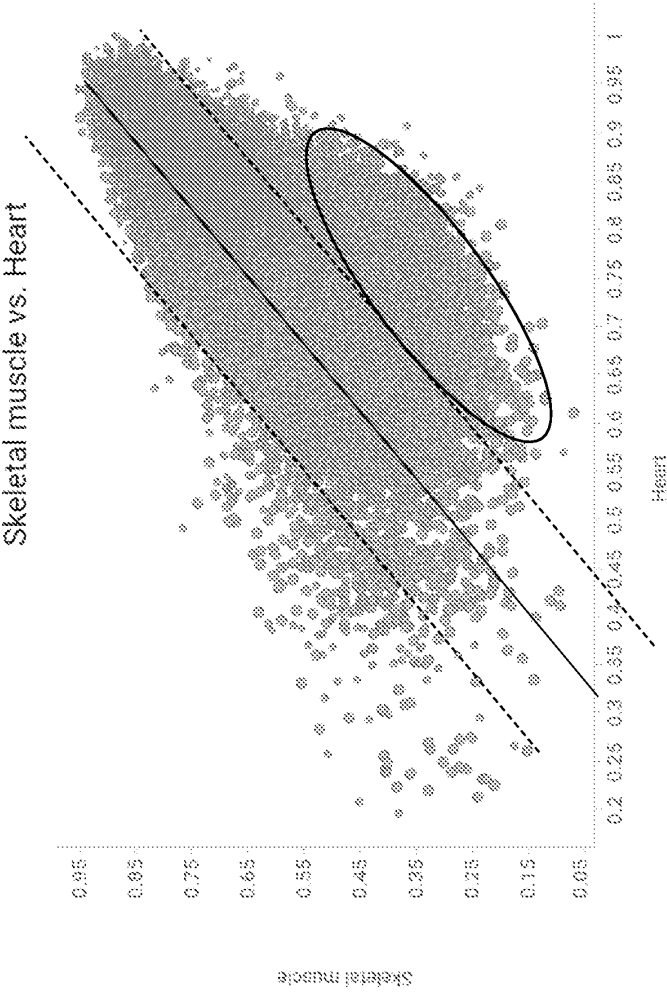


FIG. 88A

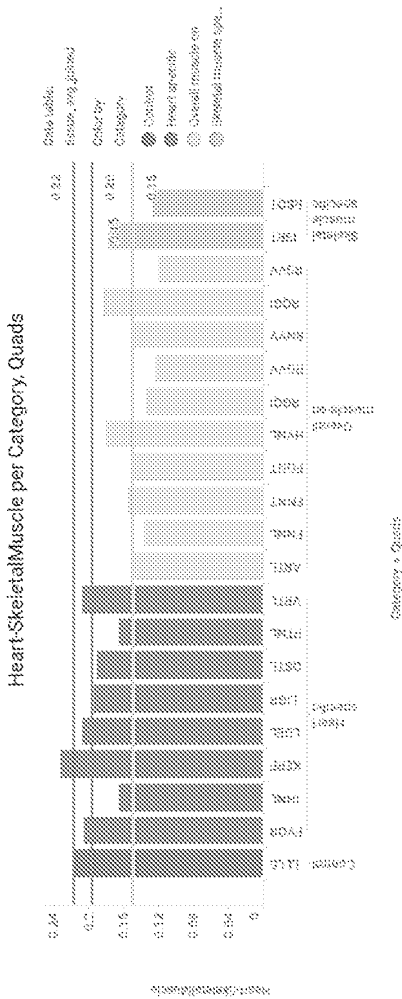


FIG. 88B

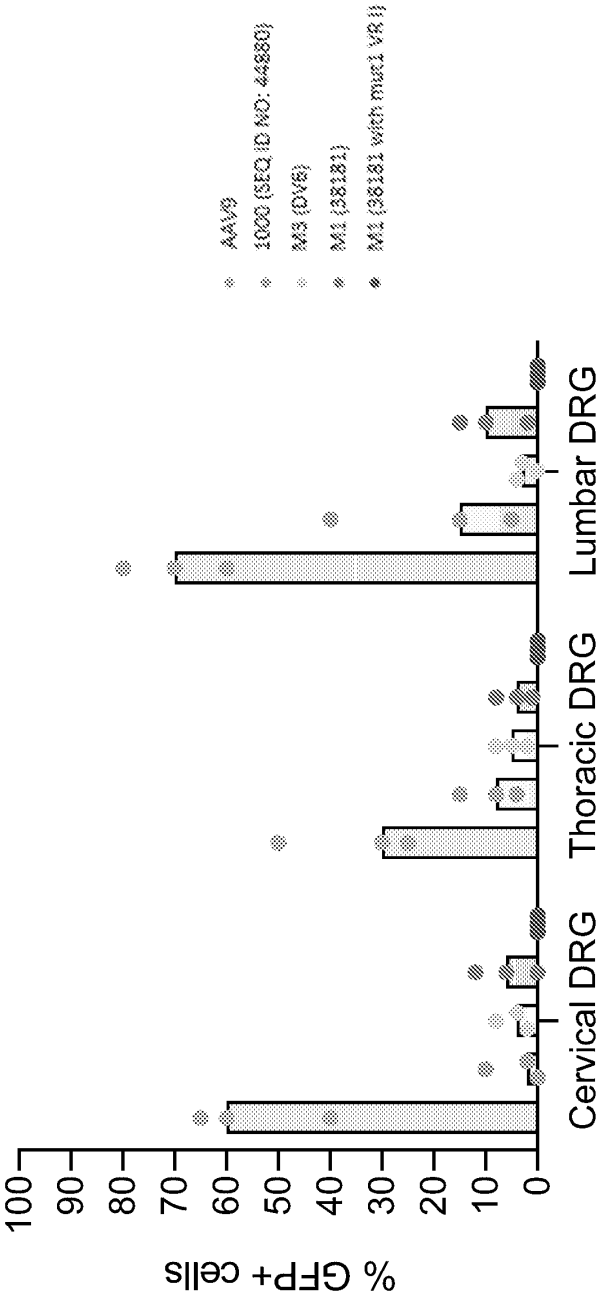


FIG. 89

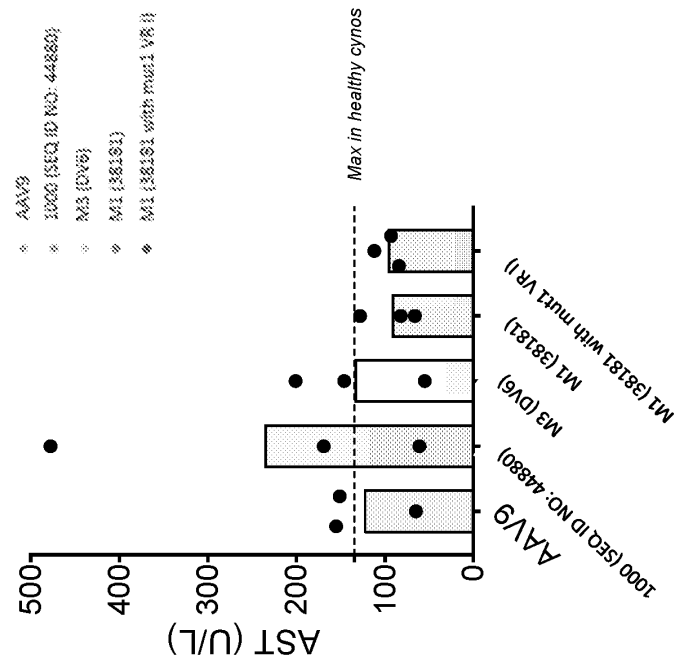


FIG. 90B

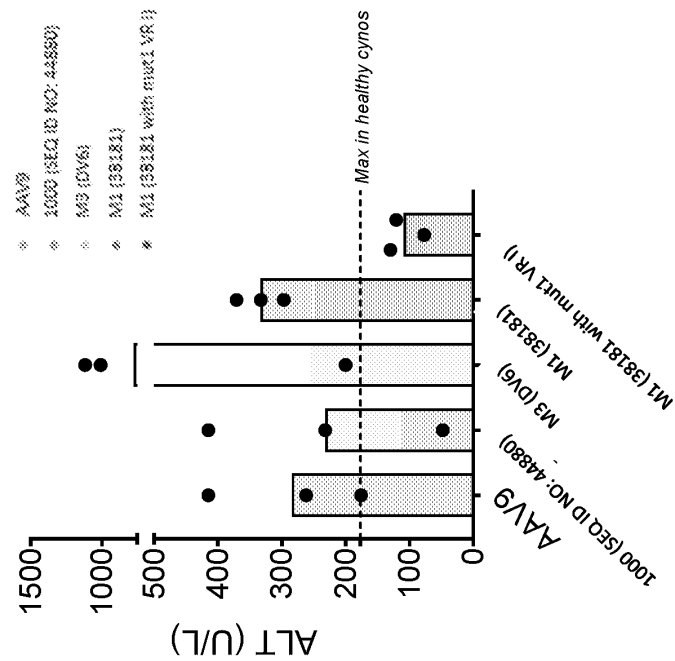


FIG. 90A

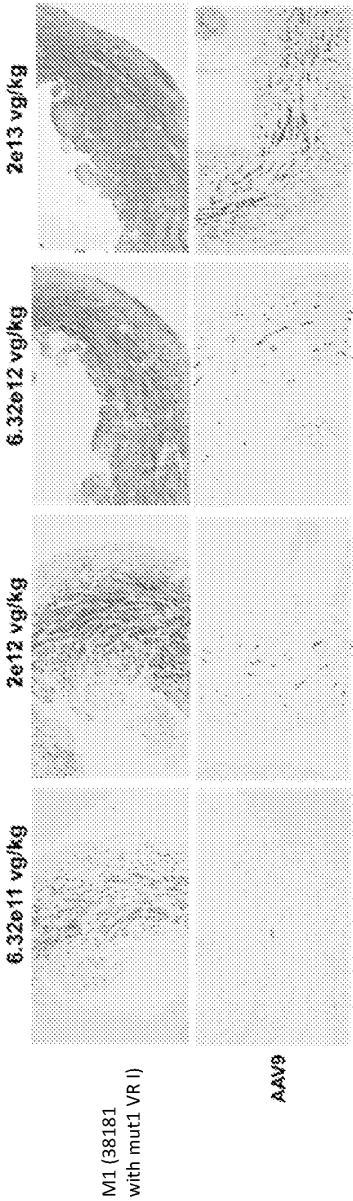


FIG. 91A

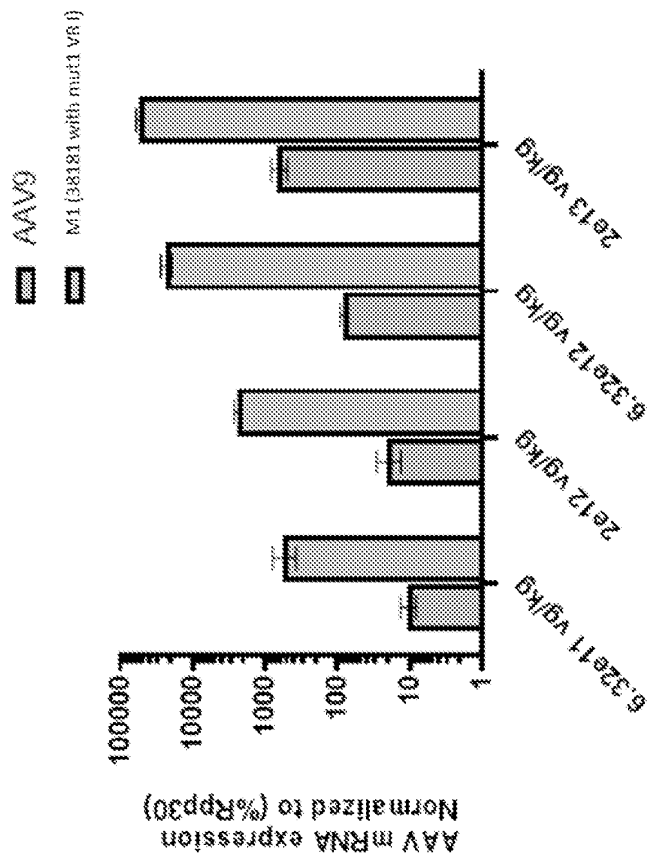


FIG. 91B

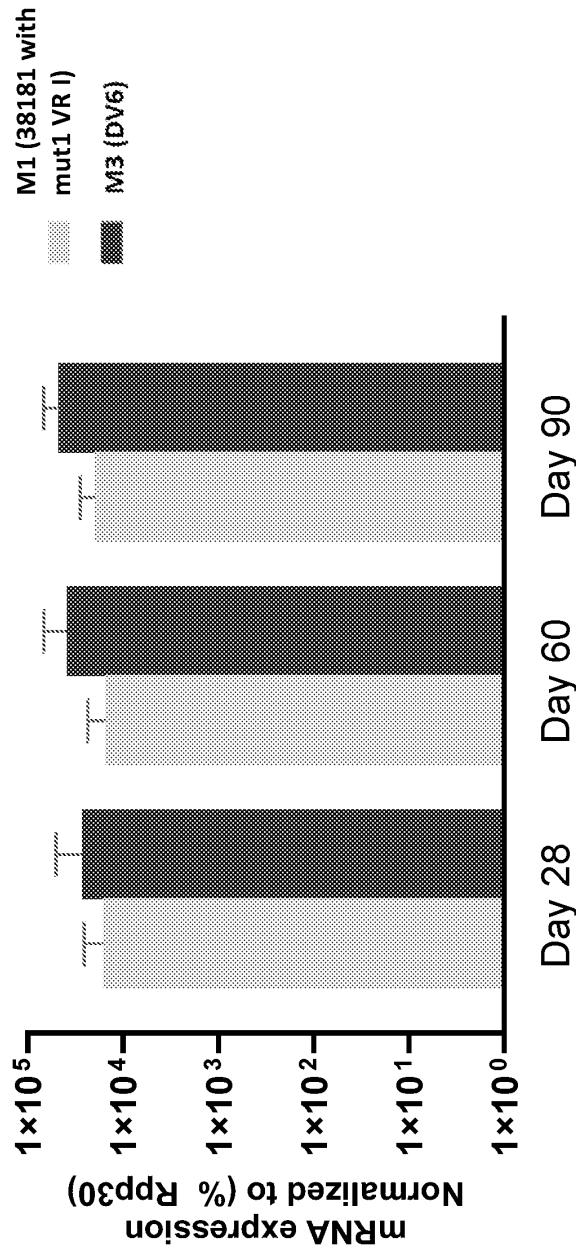


FIG. 92

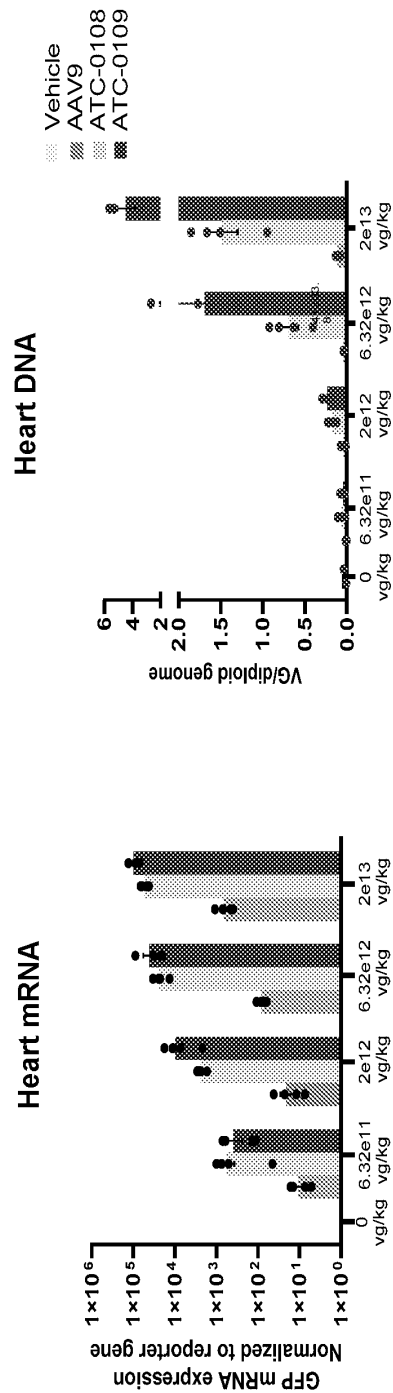


FIG. 93A

FIG. 93B

Capsid	Dose	mRNA/DNA over AAV9
M1	6.32E+11	6.7
M1	2.00E+12	21.7
M1	6.32E+12	17.4
M1	2.00E+13	6.3
M3	6.32E+11	7.4
M3	2.00E+12	69.3
M3	6.32E+12	12.4
M3	2.00E+13	3.7

FIG. 93C

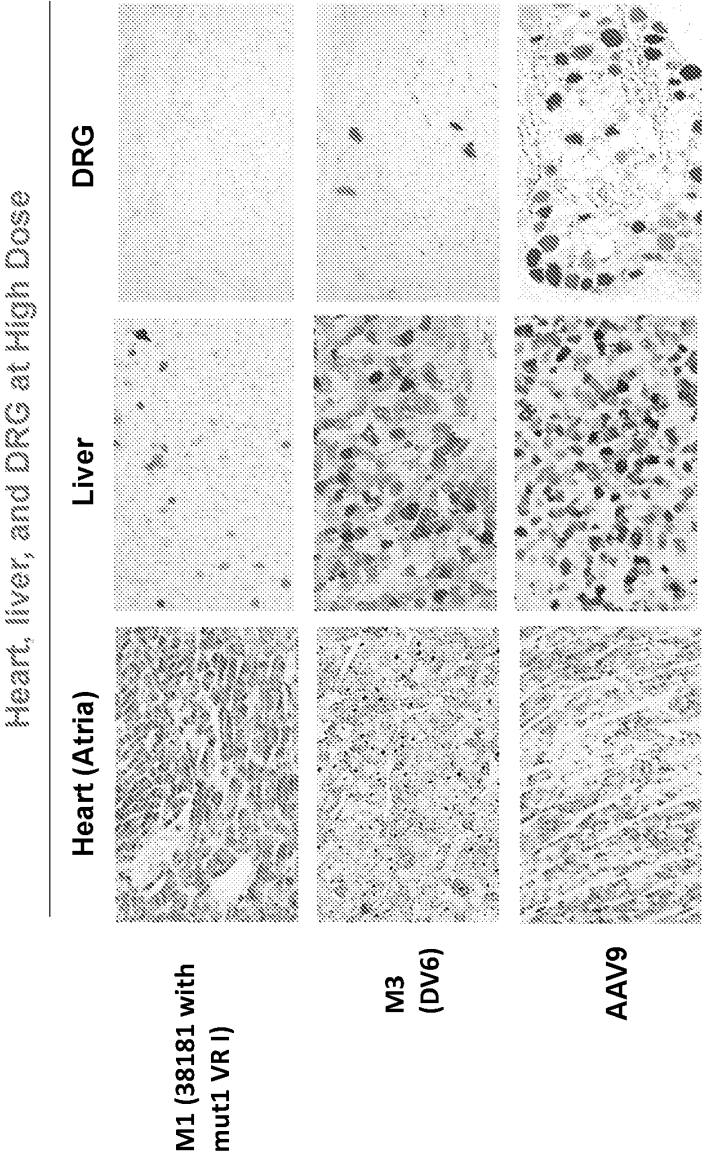


FIG. 94

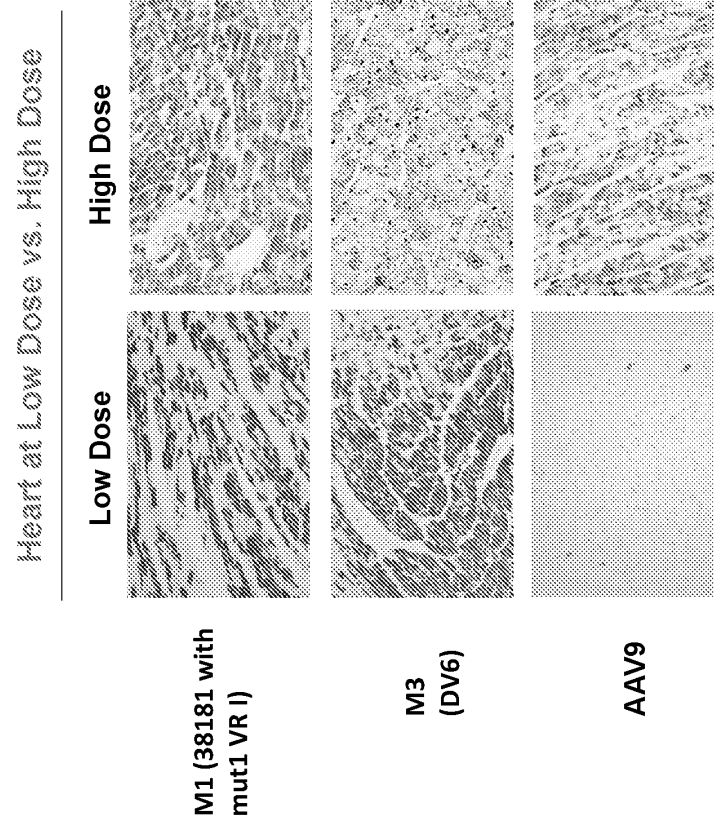


FIG. 95

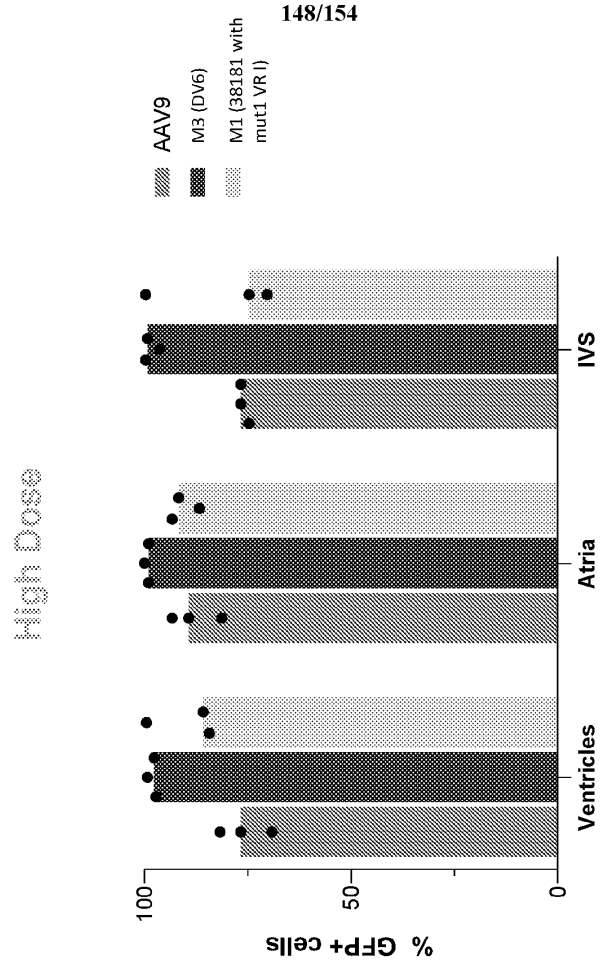


FIG. 96B

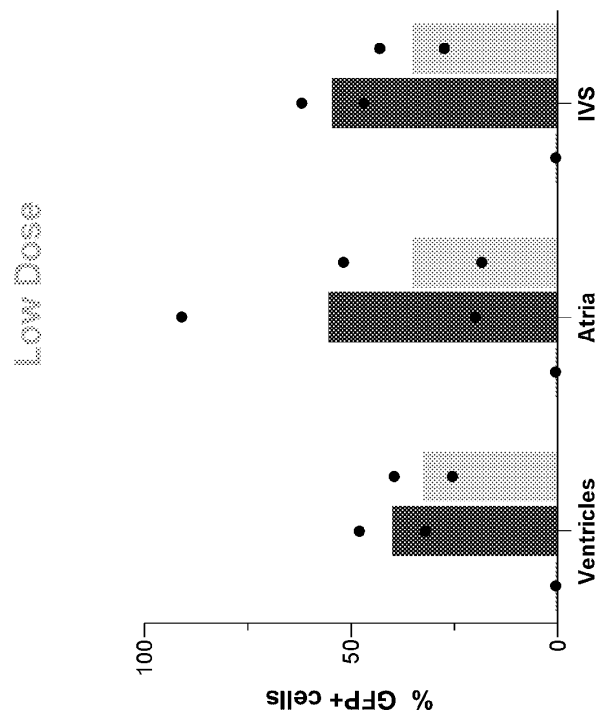


FIG. 96A

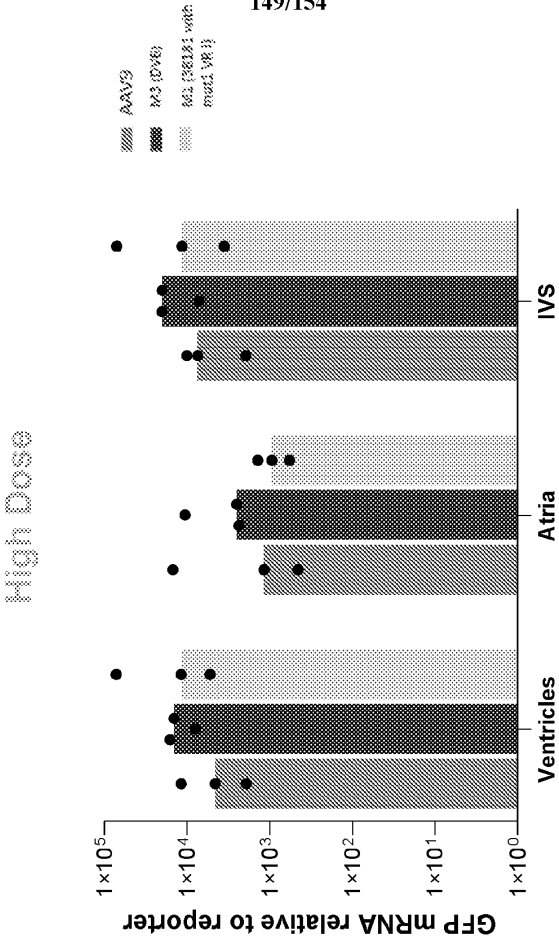


FIG. 97B

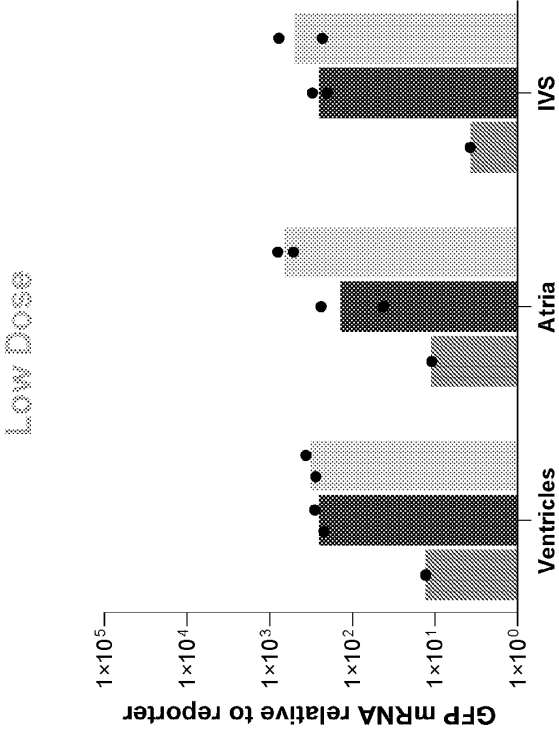


FIG. 97A

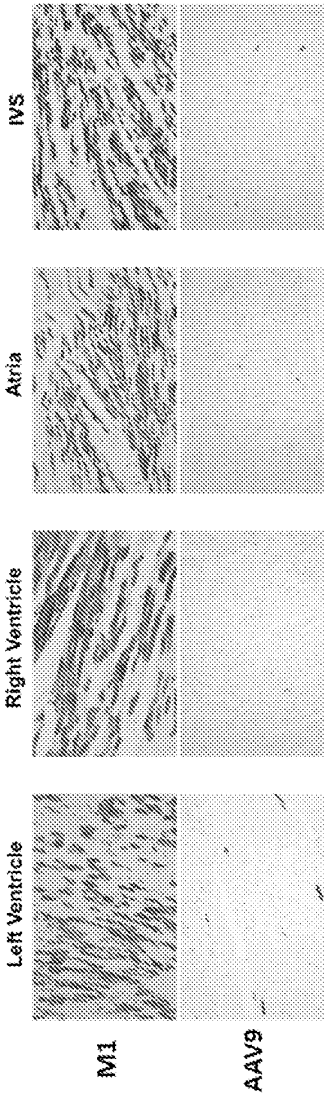


FIG. 98

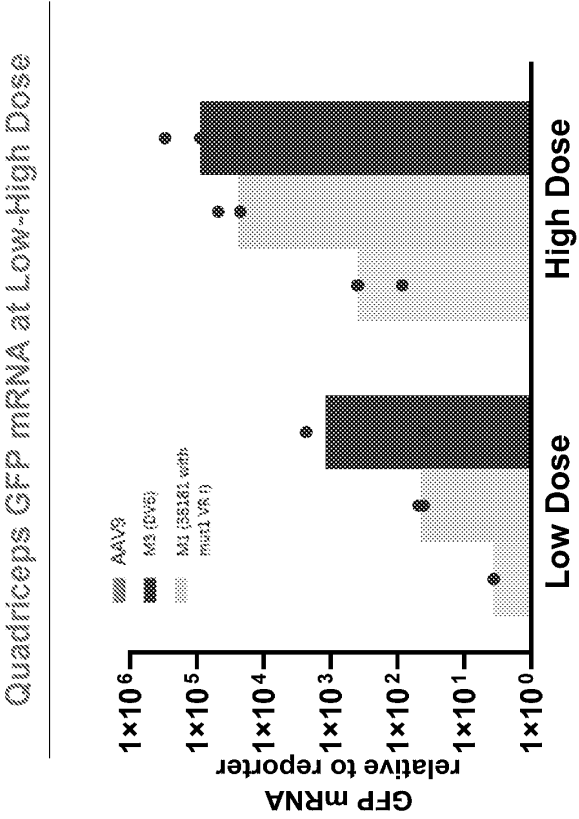


FIG. 99

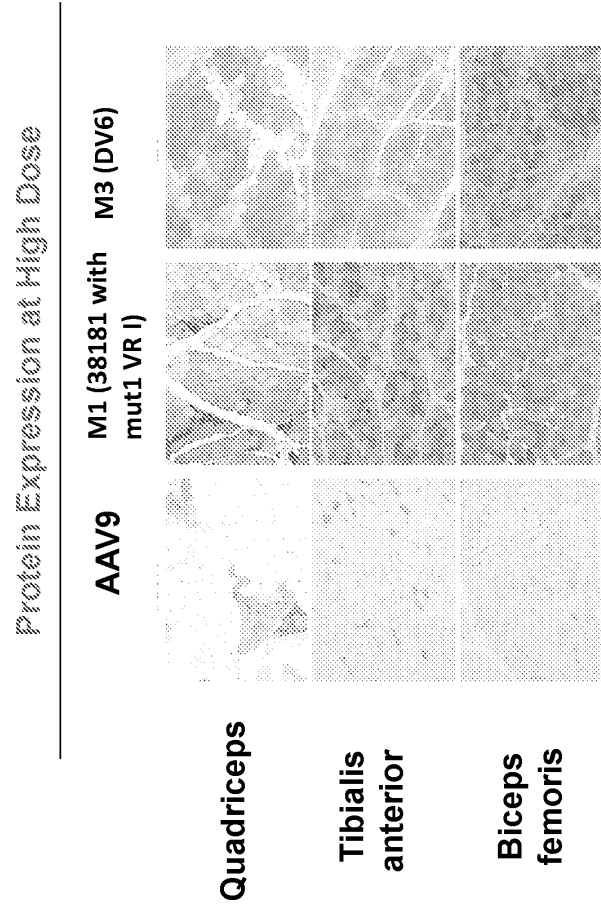


FIG. 100

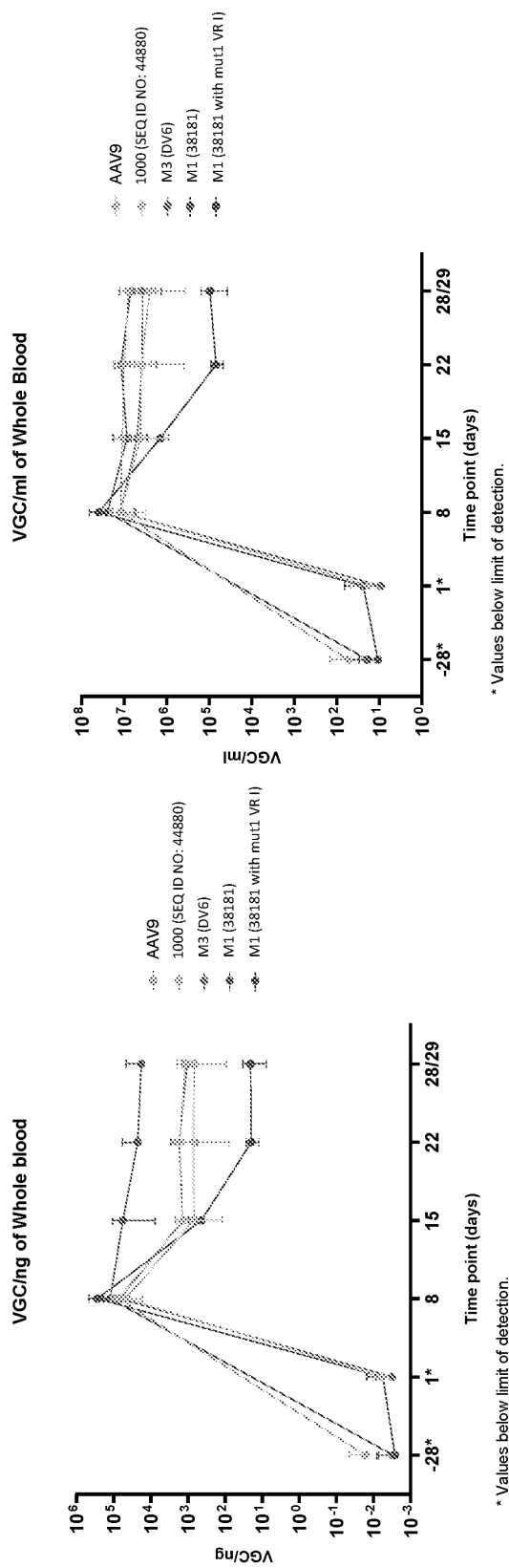


FIG. 101A

FIG. 101B

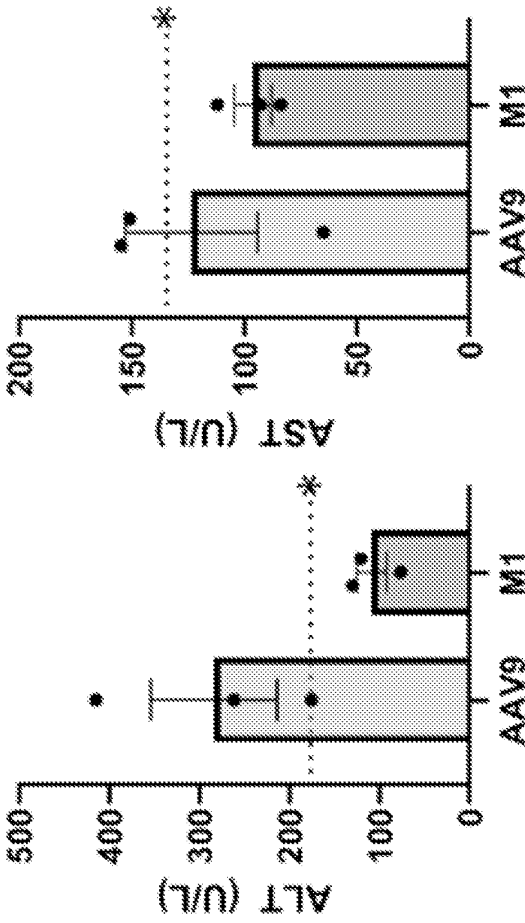


FIG. 102B

FIG. 102A