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Supporting information

Nicotinic receptor subunits atlas in the adult human lung

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1. Supporting information Tables

Table S1. Percentages of detection of each nAChR subunit in whole lung tissue transcriptomes.

	Non-Smokers (n=42)		Smokers (n=111)		
CHRN	**Detection (%)**	**SD**	**Detection (%)**	**SD**	**p**
A1	31.11	11.48	40.17	15.96	***
A2	29.43	18.56	42.70	15.97	***
A3	19.64	35.77	4.65	19.64	*
A4	21.93	39.78	5.04	21.21	*
A5	45.39	27.60	47.80	15.67	ns
A6	50.95	30.07	57.04	16.94	ns
A7	0.00	0.0	25.53	34.62	NA
A9	55.71	19.48	31.49	26.00	***
A10	71.02	17.65	68.19	15.47	ns
B1	80.99	11.40	81.80	9.33	ns
B2	20.37	37.01	4.61	19.40	*
B3	53.87	31.47	64.87	18.65	*
B4	26.84	18.74	46.92	26.15	***
D	65.00	13.14	40.45	31.39	***
E	76.07	13.68	75.21	8.35	ns
G	21.05	38.22	4.73	19.88	*

ns, non-significate; SD, standard deviation ; NA, not applicable. Coloured subunits indicate upregulation (green) and downregulation (red) in both groups when statistically significate.

Table S2. Repartitions of the gene expressions of each nAChR subunit in whole lung tissue transcriptomes.

	Non-Smokers (n=42)		Smokers (n=111)		
CHRN	**Expression (%)**	**SD**	**Expression (%)**	**SD**	**p**
A1	7.09	2.36	9.20	3.44	***
A2	4.20	2.59	6.32	2.37	***
A3	2.17	3.94	0.51	2.14	***
A4	2.05	3.72	0.47	1.97	***
A5	7.00	4.17	7.59	2.40	ns
A6	6.38	3.76	7.31	2.06	ns

A7	0.00	0.00	2.80	3.81	NA
A9	5.13	2.12	2.84	2.42	***
A10	14.79	3.23	14.34	3.00	ns
B1	15.29	1.72	15.55	1.618	ns
B2	2.66	4.83	0.60	2.51	***
B3	6.33	3.68	7.83	2.17	**
B4	3.47	2.35	6.33	3.64	***
D	8.15	1.79	5.02	3.91	***
E	12.76	1.85	12.73	1.22	ns
G	2.53	4.58	0.56	2.37	***

ns, non-significate; SD, standard deviation ; NA, not applicable. Coloured subunits indicate upregulation (green) and downregulation (red) in both groups when statistically significate.

Table S3. Percentages of detection of each nAChR subunit in LAEC transcriptomes.

	Non-Smokers (n=5)		Smokers (n=5)		
CHRN	**Detection (%)**	**SD**	**Detection (%)**	**SD**	**p**
A1	0.00	0.00	0.00	0.00	NA
A2	0.00	0.00	0.00	0.00	NA
A3	20.00	44.72	6.48	14.50	ns
A4	0.00	0.00	0.00	0.00	NA
A5	79.80	33.01	45.57	15.82	*
A6	20.00	44.72	13.57	30.33	ns
A7	74.13	23.32	47.17	30.96	ns
A9	49.00	35.99	32.56	28.86	ns
A10	59.49	25.23	28.97	18.51	*
B1	0.00	0.00	34.44	48.17	NA
B2	8.88	19.88	28.50	44.00	ns
B3	0.00	0.00	0.00	0.00	NA
B4	0.00	0.00	20.00	44.72	NA
D	0.00	0.00	0.00	0.00	NA
E	70.50	23.07	39.02	15.03	*
G	13.71	30.65	25.86	43.34	ns

ns, non-significate; SD, standard deviation ; NA, not applicable. Coloured subunits indicate upregulation (green) and downregulation (red) in both groups when statistically significate.

Table S4. Repartitions of the gene expressions of each nAChR subunit in LAEC transcriptomes.

	Non-Smokers (n=5)		Smokers (n=5)		
CHRN	**Expression (%)**	**SD**	**Expression (%)**	**SD**	**p**
A1	0.00	0.00	0.00	0.00	ns
A2	0.00	0.00	0.00	0.00	ns
A3	1.01	2.26	0.50	1.13	ns
A4	0.00	0.00	0.00	0.00	ns
A5	17.98	9.69	19.73	18.14	ns

A6	0.73	1.63	0.70	1.56	ns
A7	33.29	12.50	24.63	16.38	ns
A9	22.82	14.14	20.23	16.04	ns
A10	18.00	2.99	11.26	7.32	ns
B1	0.00	0.00	1.81	2.63	NA
B2	0.60	1.33	11.02	21.25	NA
B3	0.00	0.00	0.00	0.00	ns
B4	0.00	0.00	0.61	1.37	NA
D	0.00	0.00	0.00	0.00	ns
E	2.99	3.21	6.38	6.25	ns
G	2.58	5.77	3.13	5.15	ns

ns, non-significate; SD, standard deviation ; NA, not applicable.

Table S5. Percentages of detection of each nAChR subunit in SAEC transcriptomes.

	Non-Smokers (n=63)		**Smokers (n=72)**		
CHRN	**Detection (%)**	**SD**	**Detection (%)**	**SD**	**p**
A1	19.76	18.19	17.94	16.64	ns
A2	40.37	19.26	38.47	18.26	ns
A3	28.09	18.87	29.69	21.35	ns
A4	26.91	21.26	26.16	20.95	ns
A5	19.34	12.45	25.44	19.05	*
A6	44.07	22.46	49.31	21.30	ns
A7	33.33	17.59	42.75	23.26	**
A9	42.73	22.00	39.41	17.81	ns
A10	35.52	17.83	35.41	16.35	ns
B1	39.45	21.82	42.64	20.75	ns
B2	35.57	17.80	48.21	20.14	***
B3	29.03	20.59	37.20	21.75	*
B4	15.82	13.51	20.19	20.67	ns
D	21.04	18.37	20.74	16.82	ns
E	26.68	17.79	32.67	23.12	ns
G	29.29	20.95	30.43	23.50	ns

ns, non-significate; SD, standard deviation. Coloured subunits indicate upregulation (green) and downregulation (red) in both groups when statistically significate.

Table S6. Repartitions of the gene expressions of each nAChR subunit in SAEC transcriptomes.

	Non-Smokers (n=63)		**Smokers (n=72)**		
CHRN	**Expression (%)**	**SD**	**Expression (%)**	**SD**	**p**
A1	2.41	1.70	2.03	1.68	ns
A2	9.43	3.86	8.13	3.77	*
A3	4.23	2.37	3.95	2.46	ns
A4	3.37	2.22	3.04	2.33	ns

A5	2.83	1.56	3.24	1.81	ns
A6	4.95	2.00	5.11	1.99	ns
A7	8.61	3.46	9.93	4.61	*
A9	13.88	5.53	11.99	4.86	*
A10	13.76	5.08	12.62	4.55	ns
B1	6.87	3.07	6.78	2.81	ns
B2	13.89	4.98	16.81	5.17	**
B3	3.41	1.83	4.09	2.00	*
B4	2.14	1.35	2.46	2.04	ns
D	2.14	1.66	1.86	1.38	ns
E	3.41	2.34	3.62	2.57	ns
G	4.67	2.65	4.34	2.67	ns

ns, non-significate; SD, standard deviation. Coloured subunits indicate upregulation (green) and downregulation (red) in both groups when statistically significate.

Table S7. List of CHRN antibodies.

Antibodies	Species	Reference	Companies	Concentrations
α1	Rabbit	HPA071554	Sigma-Aldrich	1:100
α2	Mouse	NBP2-61667	Novus Biological	1:50
α3	Rabbit	HPA029430	Sigma-Aldrich	1:100
α4	Mouse	NBP2-61674	Novus Biological	1:100
α5	Rabbit	HPA054381	Sigma-Aldrich	1:50
α6	Mouse	NBP2-61679	Novus Biological	1:100
α7	Mouse	NBP2-61738	Novus Biological	1:100
α9	Rabbit	26025-1-AP	Proteintech	1:100
α10	Mouse	NBP2-61666	Novus Biological	1:50
β1	Rabbit	HPA005822	Sigma-Aldrich	1:100
β2	Rabbit	17844-1-AP	Proteintech	1:50
β3	Rabbit	APrEST84413	Novus Biological	1:100
β4	Mouse	NBP2-61742	Novus Biological	1:100
δ	Rabbit	HPA056404	Sigma-Aldrich	1:100
ε	Rabbit	NBP1-79951	Novus Biological	1:100
γ	Rabbit	NBP1-79952	Novus Biological	1:100

Table S8. List of recognition antigens of CHRN antibodies and their percentages of identity.

s.u.	Antigenic sequences	Position	Identity*
α1	MKLGTWTYDGSVVAINPESDQPDLSNFMESGEWVIKESRGWKHSVTYSCCPDTPYLDITYH F	189-250	α2/3/4/6
α2	EEAKRPPPRAPGDPLSSPSPTALPQGGSHTETEDRLFKHLFRGYNRWARPVPNTSDVVIVRFG LSIAQLIDVDEKNQMMTTNVWLKQEWSDYKLRWNPTDFGNITSLRVPSEMIWIPDIVLYNN ADGEFAVTHMTKAHLFSTGTVHWVPPAIYKSSCSIDVTFFPFDQQNCKMKFGSWTYDKAKI DLEQMEQTVDLKDYWESGEWAIVNATGTYNSKKYDCCAEIYPDVTYAFVIRRL	27-264	α4 α3/5/6 β3
α3	RTPTTHTMPSWVKTVFLNLLPRVMFMTRPTSNEGNAQKPRPLYGAELSNLNCFSRAESKGC KEGYPCQDGMCGYCHHRRIKISNFSANLTRSSSSESVDAVLSLSALSPEIKEAIQSVKYIAENM KAQNEAKEIQDDWKYVAM	331-473	α1/2/6 β2/4
α4	HVETRAHAEERLLKKLFSGYNKWSRPVANISDVVLVRFGLSIAQLIDVDEKNQMMTTNVW VKQEWHDYKLRWDPADYENVTSIRIPSELIWRPDIVLYNNADGDFAVTHLTKAHLFHDGR VQWTPPAIYKSSCSIDVTFFPFDQQNCTMKFGSWTYDKAKIDLVNMHSRVDQLDFWESGE WVIVDAVGTYNTRKYECCAEIYPDITYAFVIRRL	29-242	α2 α3/5/6 β3
α5	AQRGLSEPSSIAKHEDSLLKDLFQDYERWVRPVEHLNDK	33-71	β3 α4
α6	KGCVGCATEERLFHKLFSHYNQFIRPVENVSDPVTVHFEVAITQLANVDEVNQIMETNLWL RHIWNDYKLRWDPMEYDGIETLRVPADKIWKPDIVLYNNAVGDFQVEGKTKALLKYNGMI TWTPPAIFKSSCPMDITFFPFDHQNCSLKFGSWTYDKAEIDLLIIGSKVDMNDFWENSEWEII DASGYKHDIKYNCCEEIYTDITYSFYIRRL	26-239	α3 α2/4/5 β3
α7	LYKELVKNYNPLERPVANDSQPLTVYFSLSLLQIMDVDEKNQVLTTNIWLQMSWTDHYLQ WNVSEYPGVKTVRFPDGQIWKPDILLYNSADE	58-149	α2/3/4/10
α9	HFCGAEARPVPHWARVVILKYMSRVLFVYDVGESCLSPHHSRERDHLTKVYSKLPESNLKA ARNKDLSRKKDMNKRLKNDLGCQGKNPQEAESYCAQYKVLTRNIEYIAKCLKDHKATNS KGSEWKKVAKVIDRFFMWIFFIMVFVMTILIIARAD	324-479	α2/4/6/10
α10	AEGRLALKLFRDLFANYTSALRPVADTDQTLNVTLEVTLSQIIDMDERNQVLTLYLWIRQEW TDAYLRWDPNAYGGLDAIRIPSSLVWRPDIVLYNKADAQPPGSASTNVVLRHDGAVRWDA PAITRSSCRVDVAAFPFDAQHCGLTFGSWTHGGHQLDVRPRGAAASLADFVENVEWRVLG MPARRRVLTYGCCSEPYPDVTFTLLLRRRAA	25-237	α9 α3/7
β1	LSVVVLNLHHRSPHTHQMPLWVRQIFIHKLPLYLRLKRPKPERDLMPEPPHCSSPGSGWGR GTDEYFIRKPPSDFLFPKPNRFQPELSAPDLRRFIDGPNRAVALLPELREVVSSISYIARQLQEQ EDHDALKEDWQF	325-462	α3 β2/4
β2	LLRLCSGVWGTDTEERLVEHLLDPSRYNKLIRPATNGSELVTVQLMVSLAQLISVHEREQIMT TNVWLTQEWEDYRLTWKPEEFDNMKKVRLPSKHIWLPDVVLYNNADGMYEVSFYSNAVV SYDGSIFWLPPAIYKSACKIEVKHFPFDQQNCTMKFRSWTYDRTEIDLVLKSEVASLDDFTPS GEWDIVALPGRRNENPDDSTYVDITYD	16-227	β4 α2

β3	TGFNSIAENEDALLRHLFQGYQKWVRPVLHSNDTI	20-54	α5 α2/4
β4	CRVANAEEKLMDDLLNKTRYNNLIRPATSSSQLISIKLQLSLAQLISVNEREQIMTTNVWLKQ EWTDYRLTWNSSRYEGVNILRIPAKRIWLPDIVLYNNADGTYEVSVYTNLIVRSNGSVLWLP PAIYKSACKIEVKYFPFDQQNCTLKFRSWTYDHTEIDMVLMTPTASMDDFTPSGEWDIVALP GRRTVNPQDPSYVDVTYDFIIKRKPLFYT	21-236	β2 α3 β3
δ	LVRRSSSLGYISKAEEYFLLKSRSDLMFEKQSERHGLARRLTTARRPPASSEQAQQELFNELKP AVDGANFIVNHMRDQNNYNEEKDSWNR	374-464	β2
ε	GLLGRGVGKNEELRLYHHLFNNYDPGSRPVREPEDTVTISLKVTLTNLIS	13-62	α6, γ
γ	NYDPNLRPAERDSDVVNVSLKLTLTNLISLNEREEALTTNVWIEMQWCDY	36-85	δ α7, β2/4

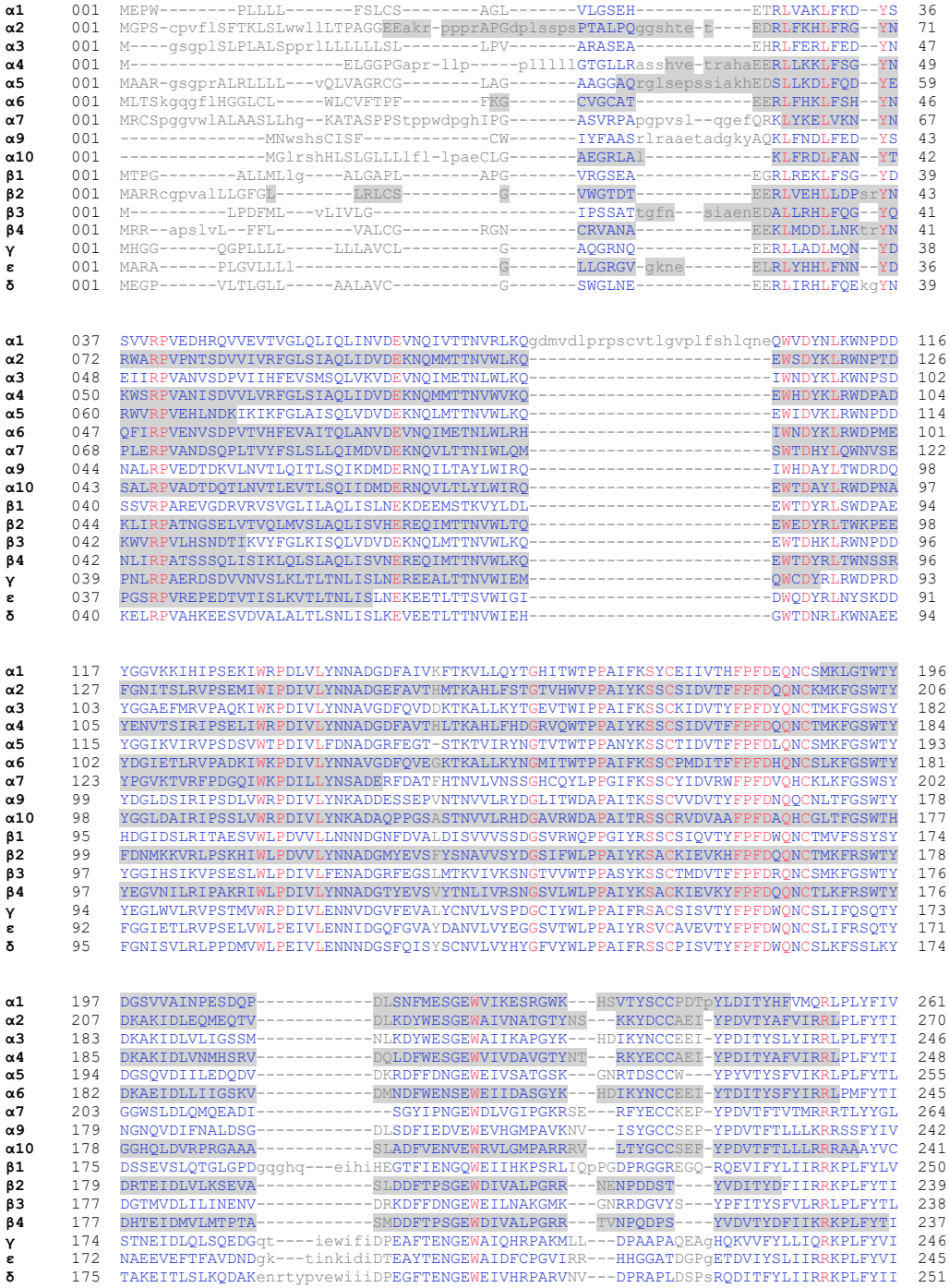
* Range of the percentage of identity obtained from blastp: yellow, 65-80%; light green, 50-65%; dark green, <50%.

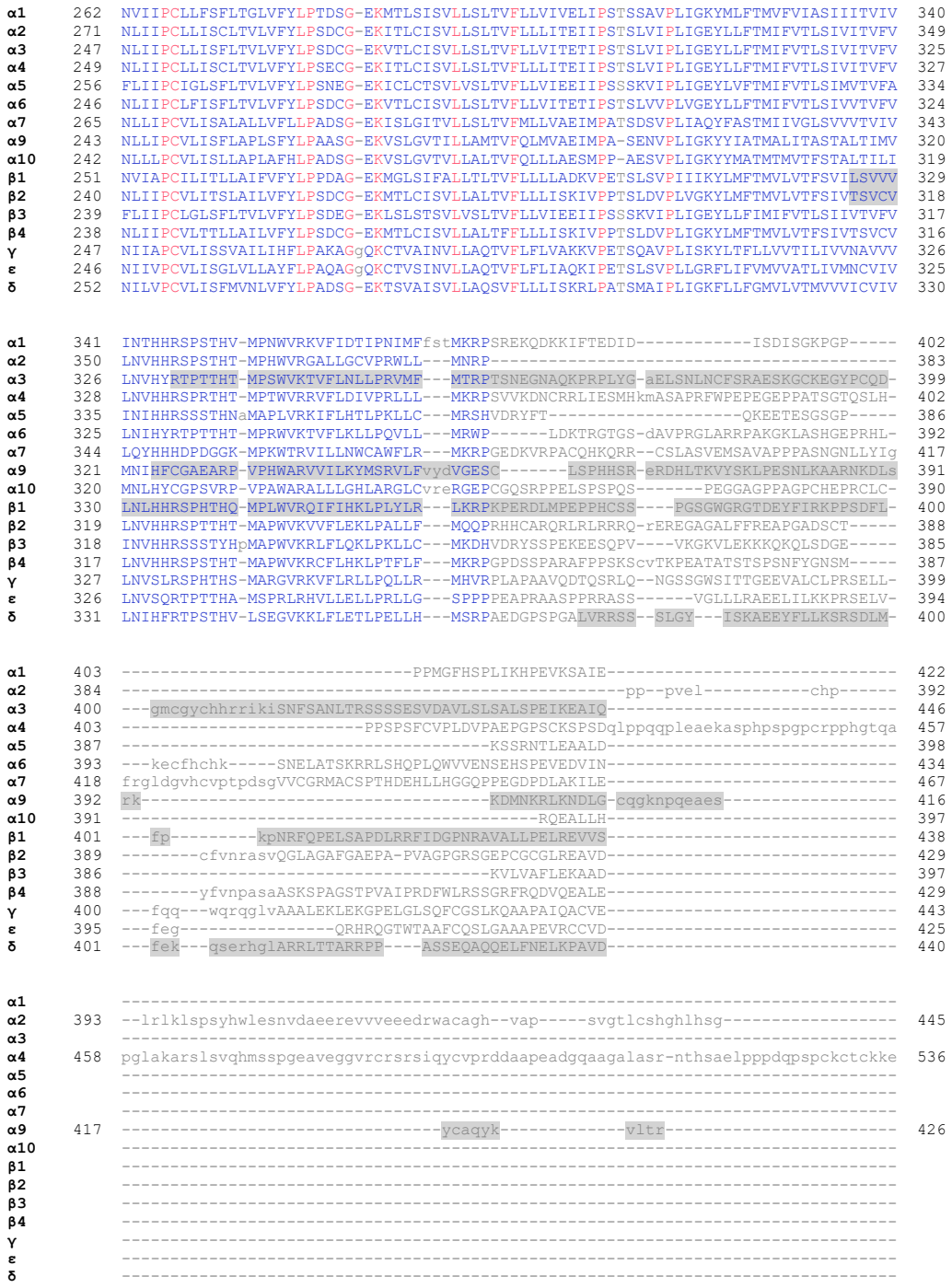
Table S9. List of primers.

ENES	GenBank	Forward sequence	Reverse sequence	Amplicon size (b)
CHRNA1	NM_001039523.2	5′- GTCCACACAAGCTCCGGTA-3′	5′- CAGACGGGTCTCATGTTCG-3′	104
CHRNA2	NM_000742.3	5′- CTGTGGTGGCTCCTTCTGA-3′	5′- GGGAGAGGAGAGTGGGTCTC-3′	87
CHRNA3	NM_000743.4	5′- TGAAATGGAACCCCTCTGAC-3′	5′- GAAATCCCCAACAGCATTGT-3′	107
CHRNA4	NM_000744.6	5′- GCCGGACATCGTCCTCTAC-3′	5′- TGCAGGAGCTCTTGTAAATGG-3′	125
CHRNA5	NM_000745.3	5′- GACAACAAACGTCTGGTTGAAA-3′	5′- ACAGAGTCTGAAGGAACACGTATAAC-3′	105
CHRNA6	NM_004198.3	5′- TTCATGGGGGCTTGTGTC-3′	5′- GAGCCTCTCCTCAGTTGCAC-3′	83
CHRNA7	NM_000746.5	5′- CAATGACTCGCAACCACTCA-3′	5′- GTGATCTGTCCAAGACATTTGC-3′	121
CHRNA9	NM_017581.3	5′-TCAGAAAATGTGCCCCTGAT-3′	5′- GGCCCCACAGAAGTGGATA-3′	108
CHRNA10	NM_020402.3	5′- CCCAGATCATCGACATGGA-3′	5′- CCCATCGTAGGTAGGCATCT-3′	90
CHRNB1	NM_000747.2	5′- CACAAAGGTGTACTTAGACCTGGA-3′	5′- TTCAGTAGCACCACGTCAGG-3′	129
CHRNB2	NM_000748.2	5′- CTGGCCCAGCTCATCAGT-3′	5′- TCCAGGTGAGGCGATAATCT-3′	94
CHRNB3	NM_000749.4	5′- GGTCCGCCCTGTATTACATTC-3′	5′- TCAGCTGATTCTTTTCATCCAC-3′	95
CHRNB4	NM_000750.4	5′- TGACGATGAAGACCAGAGTGTC-3′	5′- GGACGCACACAAACATGAAC-3′	95
CHRND	NM_000751.3	5′- GGGACCAGAACAATTACAATGAG-3′	5′- GCAGGAAGATCCAGGCTGT-3′	113
CHRNE	NM_000080.4	5′- CGACACAGAGGCCTATACTGAG-3′	5′- GCGGATGATGAGCGAGTAG-3′	93
CHRNG	NM_005199.4	5′- AGCAGAGTCACTTTGACAATGG-3′	5′- GTAGTGGGCCATGAGGAAGA-3′	131

2. Supporting information Figures

Figure S1. Constraint-based Multiple Alignment of CHRN antibodies. COBALT alignment is shown for the 16 subunits and antigen sequences of corresponding antibodies are highlighted in grey. Red amino acids are conserved for all subunits.




α1 262 NVIIPCLLFSFLTGLVFYLPTDSG-EKMTLSISVLLSLTVFLLVIVELIPSTSSAVPLIGKYMLFTMVFVIASIIITVIV 340
α2 271 NLIIPCLLISCLTVLVFYLPSDCG-EKITLCISVLLSLTVFLLLITEIIPSTSLVIPLIGEYLLFTMIFVTLSIVITVFV 349
α3 247 NLIIPCLLISFLTVLVFYLPSDCG-EKVTLCISVLLSLTVFLLVITETIPSTSLVIPLIGEYLLFTMIFVTLSIVITVFV 325
α4 249 NLIIPCLLISCLTVLVFYLPSECG-EKITLCISVLLSLTVFLLLITEIIPSTSLVIPLIGEYLLFTMIFVTLSIVITVFV 327
α5 256 FLIIPCIGLSFLTVLVFYLPSNEG-EKICLCTSVLVSLTVFLLVIEEIIPSSSKVIPLIGEYLVFTMIFVTLSIMVTVFA 334
α6 246 NLIIPCLFISFLTVLVFYLPSDCG-EKVTLCISVLLSLTVFLLVITETIPSTSLVVPLVGEYLLFTMIFVTLSIVVTVFV 324
α7 265 NLLIPCVLISALALLVFLLPADSG-EKISLGITVLLSLTVFMLLVAEIMPATSDSVPLIAQYFASTMIIVGLSVVVTVIV 343
α9 243 NLLIPCVLISFLAPLSFYLPAASG-EKVSLGVTILLAMTVFQLMVAEIMPA-SENVPLIGKYYIATMALITASTALTIMV 320
α10 242 NLLLPCVLISLLAPLAFHLPADSG-EKVSLGVTVLLALTVFQLLLAESMPP-AESVPLIGKYYMATMTMVTFSTALTILI 319
β1 251 NVIAPCILITLLAIFVFYLPPDAG-EKMGLSIFALLTLTVFLLLLADKVPETSLSVPIIIKYLMFTMVLVTFSVILSVVV 329
β2 240 NLIIPCVLITSLAILVFYLPSDCG-EKMTLCISVLLALTVFLLLISKIVPPTSLDVPLVGKYLMFTMVLVTFSIVTSVCV 318
β3 239 FLIIPCLGLSFLTVLVFYLPSDEG-EKLSLSTSVLVSLTVFLLVIEEIIPSSSKVIPLIGEYLLFIMIFVTLSIIVTVFV 317
β4 238 NLIIPCVLTTLLAILVFYLPSDCG-EKMTLCISVLLALTFFLLLISKIVPPTSLDVPLIGKYLMFTMVLVTFSIVTSVCV 316
γ 247 NIIAPCVLISSVAILIHFLPAKAGgQKCTVAINVLLAQTVFLFLVAKKVPETSQAVPLISKYLTFLLVVTILIVVNAVVV 326
ε 246 NIIVPCVLISGLVLLAYFLPAQAGgQKCTVSINVLLAQTVFLFLIAQKIPETSLSVPLLGRFLIFVMVVATLIVMNCVIV 325
δ 252 NILVPCVLISFMVNLVFYLPADSG-EKTSVAISVLLAQSVFLLLISKRLPATSMAIPLIGKFLLFGMVLVTMVVVICVIV 330
α1 341 INTHHRSPSTHV-MPNWVRKVFIDTIPNIMFfstMKRPSREKQDKKIFTEDID----------ISDISGKPGP----- 402
α2 350 LNVHHRSPSTHT-MPHWVRGALLGCVPRWLL---MNRP---------- 383
α3 326 LNVHYRTPTTHT-MPSWVKTVFLNLLPRVMF---MTRPTSNEGNAQKPRPLYG-aELSNLNCFSRAESKGCKEGYPCQD- 399
α4 328 LNVHHRSPRTHT-MPTWVRRVFLDIVPRLLL---MKRPSVVKDNCRRLIESMHkmASAPRFWPEPEGEPPATSGTQSLH- 402
α5 335 INIHHRSSSTHNaMAPLVRKIFLHTLPKLLC---MRSHVDRYFT----------QKEETESGSGP----- 386
α6 325 LNIHYRTPTTHT-MPRWVKTVFLKLLPQVLL---MRWP------LDKTRGTGS-dAVPRGLARRPAKGKLASHGEPRHL- 392
α7 344 LQYHHHDPDGGK-MPKWTRVILLNWCAWFLR---MKRPGEDKVRPACQHKQRR--CSLASVEMSAVAPPPASNGNLLYIg 417
α9 321 MNIHFCGAEARP-VPHWARVVILKYMSRVLFvydVGESC-------LSPHHSR-eRDHLTKVYSKLPESNLKAARNKDLs 391
α10 320 MNLHYCGPSVRP-VPAWARALLLGHLARGLCvreRGEPCGQSRPPELSPSPQS-------PEGGAGPPAGPCHEPRCLC- 390
β1 330 LNLHHRSPHTHQ-MPLWVRQIFIHKLPLYLR---LKRPKPERDLMPEPPHCSS----PGSGWGRGTDEYFIRKPPSDFL- 400
β2 319 LNVHHRSPTTHT-MAPWVKVVFLEKLPALLF---MQQPRHHCARQRLRLRRRQ-rEREGAGALFFREAPGADSCT----- 388
β3 318 INVHHRSSSTYHpMAPWVKRLFLQKLPKLLC---MKDHVDRYSSPEKEESQPV----VKGKVLEKKKQKQLSDGE----- 385
β4 317 LNVHHRSPSTHT-MAPWVKRCFLHKLPTFLF---MKRPGPDSSPARAFPPSKScvTKPEATATSTSPSNFYGNSM----- 387
γ 327 LNVSLRSPHTHS-MARGVRKVFLRLLPQLLR---MHVRPLAPAAVQDTQSRLQ--NGSSGWSITTGEEVALCLPRSELL- 399
ε 326 LNVSQRTPTTHA-MSPRLRHVLLELLPRLLG---SPPPPEAPRAASPPRRASS------VGLLLRAEELILKKPRSELV- 394
δ 331 LNIHFRTPSTHV-LSEGVKKLFLETLPELLH---MSRPAEDGPSPGALVRRSS--SLGY---ISKAEEYFLLKSRSDLM- 400
α1 403 ----------PPMGFHSPLIKHPEVKSAIE---------- 422
α2 384 ----------pp--pvel----------chp------ 392
α3 400 ---gmcgychhrrikiSNFSANLTRSSSSESVDAVLSLSALSPEIKEAIQ---------- 446
α4 403 ----------PPSPSFCVPLDVPAEPGPSCKSPSDqlppqqpleaekasphpspgpcrpphgtqa 457
α5 387 ----------KSSRNTLEAALD---------- 398
α6 393 ---kecfhchk-----SNELATSKRRLSHQPLQWVVENSEHSPEVEDVIN---------- 434
α7 418 frgldgvhcvptpdsgVVCGRMACSPTHDEHLLHGGQPPEGDPDLAKILE---------- 467
α9 392 rk----------KDMNKRLKNDLG-cqgknpqeaes---------- 416
α10 391 ----------RQEALLH---------- 397
β1 401 ---fp---------kpNRFQPELSAPDLRRFIDGPNRAVALLPELREVVS---------- 438
β2 389 --------cfvnrasvQGLAGAFGAEPA-PVAGPGRSGEPCGCGLREAVD---------- 429
β3 386 ----------KVLVAFLEKAAD---------- 397
β4 388 --------yfvnpasaASKSPAGSTPVAIPRDFWLRSSGRFRQDVQEALE---------- 429
γ 400 ---fqq---wqrqglvAAALEKLEKGPELGLSQFCGSLKQAAPAIQACVE---------- 443
ε 395 ---feg----------QRHRQGTWTAAFCQSLGAAAPEVRCCVD---------- 425
δ 401 ---fek---qserhglARRLTTARRPP----ASSEQAQQELFNELKPAVD---------- 440
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α2 393 --lrlklspsyhwlesnvdaeerevvveeedrwacagh--vap-----svgtlcshghlhsg---------- 445
α3
α4 458 pglakarslsvqhmsspgeaveggvrcrsrsiqycvprddaapeadgqaagalasr-nthsaelpppdqpspckctckke 536
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α6
α7
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α10
β1
β2
β3
β4
γ
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δ

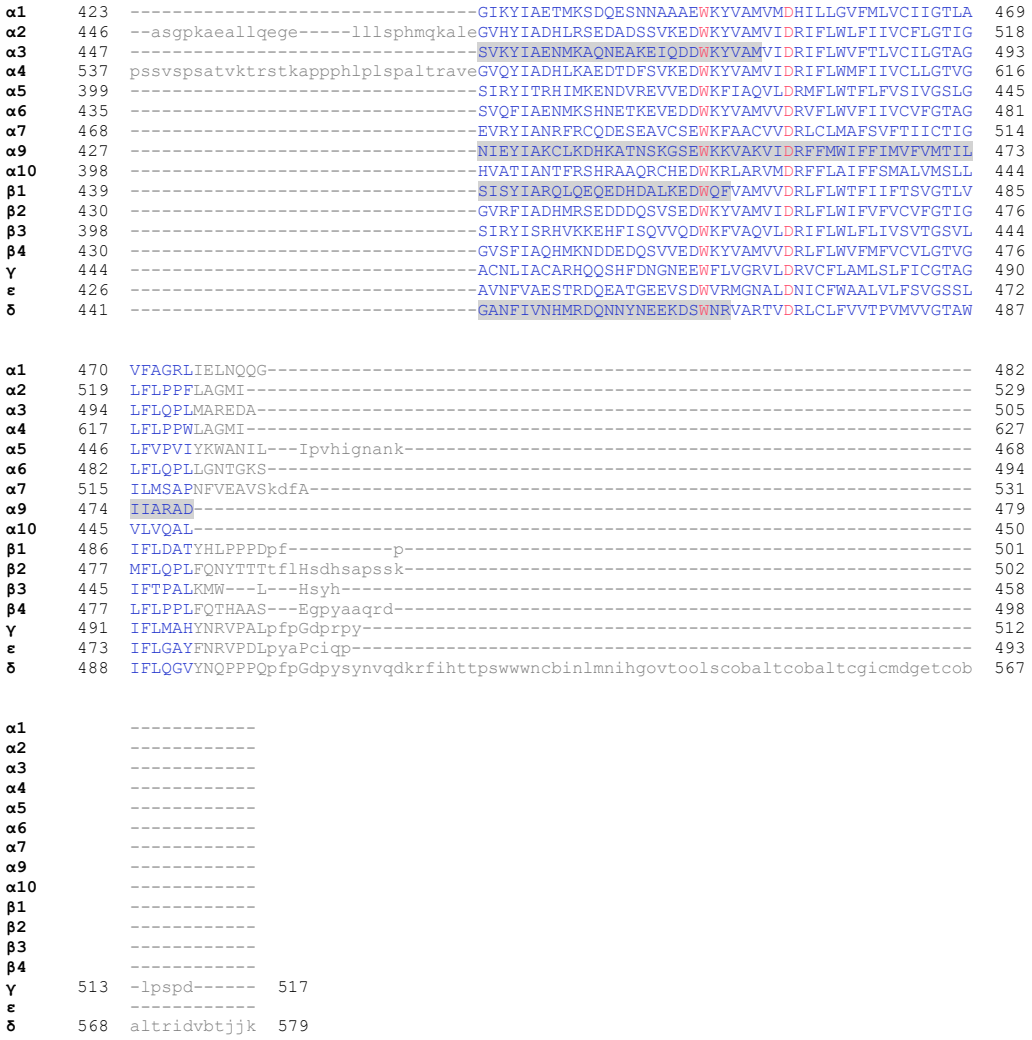
α1 423 ----------GIKYIAETMKSDQESNNAAAEWKYVAMVMDHILLGVFMLVCIIGTLA 469
α2 446 --asgpkaeallqege-----lllsphmqkaleGVHYIADHLRSEDADSSVKEDWKYVAMVIDRIFLWLFIIVCFLGTIG 518
α3 447 ----------SVKYIAENMKAQNEAKEIQDDWKYVAMVIDRIFLWVFTLVCILGTAG 493
α4 537 pssvspsatvktrstkappphlplspaltraveGVQYIADHLKAEDTDFSVKEDWKYVAMVIDRIFLWMFIIVCLLGTVG 616
α5 399 ----------SIRYITRHIMKENDVREVVEDWKFIAQVLDRMFLWTFLFVSIVGSLG 445
α6 435 ----------SVQFIAENMKSHNETKEVEDDWKYVAMVVDRVFLWVFIIVCVFGTAG 481
α7 468 ----------EVRYIANRFRCQDESEAVCSEWKFAACVVDRLCLMAFSVFTIICTIG 514
α9 427 ----------NIEYIAKCLKDHKATNSKGSEWKKVAKVIDRFFMWIFFIMVFVMTIL 473
α10 398 ----------HVATIANTFRSHRAAQRCHEDWKRLARVMDRFFLAIFFSMALVMSLL 444
β1 439 ----------SISYIARQLQEQEDHDALKEDWQFVAMVVDRLFLWTFIIFTSVGTLV 485
β2 430 ----------GVRFIADHMRSEDDDQSVSEDWKYVAMVIDRLFLWIFVFVCVFGTIG 476
β3 398 ----------SIRYISRHVKKEHFISQVVQDWKFVAQVLDRIFLWLFLIVSVTGSVL 444
β4 430 ----------GVSFIAQHMKNDDEDQSVVEDWKYVAMVVDRLFLWVFMFVCVLGTVG 476
γ 444 ----------ACNLIACARHQQSHFDNGNEEWFLVGRVLDRVCFLAMLSLFICGTAG 490
ε 426 ----------AVNFVAESTRDQEATGEEVSDWVRMGNALDNICFWAALVLFSVGSSL 472
δ 441 ----------GANFIVNHMRDQNNYNEEKDSWNRVARTVDRLCLFVVTPVMVVGTAW 487
α1 470 VFAGRLIELNQQG---------- 482
α2 519 LFLPPFLAGMI---------- 529
α3 494 LFLQPLMAREDA---------- 505
α4 617 LFLPPWLAGMI---------- 627
α5 446 LFVPVIYKWANIL---Ipvhignank---------- 468
α6 482 LFLQPLLGNTGKS---------- 494
α7 515 ILMSAPNFVEAVSkdfA---------- 531
α9 474 IIARAD---------- 479
α10 445 VLVQAL---------- 450
β1 486 IFLDATYHLPPPDpf----------p---------- 501
β2 477 MFLQPLFQNYTTTtflHsdhsapssk---------- 502
β3 445 IFTPALKMW---L---Hsyh---------- 458
β4 477 LFLPPLFQTHAAS---Egpyaaqrd---------- 498
γ 491 IFLMAHYNRVPALpfpGdprpy---------- 512
ε 473 IFLGAYFNRVPDLpyaPciqp---------- 493
δ 488 IFLQGVYNQPPPQpfpGdpysynvqdkrfihttpswwwncbinlmnihgovtoolscobaltcobaltcgicmdgetcob 567
α1 ----------
α2 ----------
α3 ----------
α4 ----------
α5 ----------
α6 ----------
α7 ----------
α9 ----------
α10 ----------
β1 ----------
β2 ----------
β3 ----------
β4 ----------
γ 513 -lpspd------ 517
ε ----------
δ 568 altridvbtjjk 579

Figure S2. Negative isotype control staining. Representative micrographs evaluating non-specific staining of the bronchial epithelia on FFPE tissues using anti-isotype matched GFP IgG antibodies (in red) from mouse (up) or rabbit (down), and DAPI (cell nuclei, blue). Magnification corresponding to the selected area is shown.

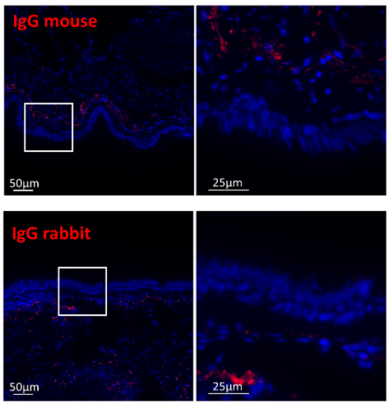

Figure S3. Localization of nAChRs on lung tissues from the Human Protein Atlas. Representative micrographs showing the bronchial epithelia on FFPE lung tissues: immunohistochemistry for $\alpha 3$, $\alpha 7$, and $\beta 1$.