

SUPPLEMENTARY MATERIAL

Supplementary Figure 1. Immunohistochemical analysis of TAGLN, TAGLN2, and PPIA expression in pulmonary adenocarcinoma (patient LA11). Photomicrographs of normal lung tissue (A) and lung adenocarcinoma (B) stained with hematoxylin-eosin. Protein expression of TAGLN (C, D), TAGLN2 (E, F), and PPIA (G, H) in lung tissue (left, normal; right, adenocarcinoma). Original magnifications: 640×.

Supplementary Figure 2. Immunohistochemical analysis of TAGLN, TAGLN2, and PPIA expression in pulmonary adenocarcinoma (patient LA12). Photomicrographs of normal lung tissue (A) and lung adenocarcinoma (B) stained with hematoxylin-eosin. Protein expression of TAGLN (C, D), TAGLN2 (E, F), and PPIA (G, H) in lung tissue (left, normal; right, adenocarcinoma). Original magnifications: 640×.

Supplementary Figure 3. Immunohistochemical analysis of TAGLN, TAGLN2, and PPIA expression in pulmonary adenocarcinoma (patient LA10). Protein expression of TAGLN (A, B), TAGLN2 (C, D), and PPIA (E, F) in lung tissue (left, normal; right, adenocarcinoma). Note the physiologic expression of TAGLN in smooth muscle cells of small blood vessels in normal lung (A, arrowheads). In this case, PPIA is expressed in few scattered cancer cells (F, arrowheads). Original magnifications: 400×.

Supplementary Figure 4. Immunohistochemical analysis of TAGLN, TAGLN2, and PPIA expression in pulmonary adenocarcinoma (patient LA14). Protein expression of TAGLN (A,

B), TAGLN2 (C, D), and PPIA (E, F) in lung tissue (left, normal; right, adenocarcinoma). Original magnifications: 400×.

Supplementary Figure 5. Expression of TAGLN in pulmonary squamous cell carcinomas.

(A, B) Representative immunohistochemical demonstration of TAGLN overexpression in myofibroblastic stroma surrounding invasive islands of squamous cell carcinoma (patient LS3). Arrowheads indicate examples of two smaller nests of carcinoma (A). Original magnifications: 100× (A), 400× (B). (C) Western blots of tissue protein extracts from four patients (LS1-4) with moderately differentiated pulmonary squamous cell carcinomas after heparin affinity enrichment.

Supplementary Table 1. Detailed information on the MS/MS sequencing of the three identified protein spots

(1) Spot 1: TAGLN and TAGLN2

Rank	Charge	Ions	dCn	XCorr	Reference	Redundancy	Peptide
1	2	14/16	0.2503	2.3733	UPSP:TAGL_HUMAN	4	K.AAEDYGVIK.T
1	2	14/16	0.2615	3.0126	UPSP:TAGL_HUMAN	5	K.KYDEELEER.L
1	2	17/26	0.3433	3.9634	UPSP:TAGL_HUMAN	5	K.LVNSLYPDGSKPKV.V
1	3	28/96	0.2019	3.8116	UPSP:TAGL_HUMAN	5	K.LVNSLYPDGSKPKV.VPENPPSM*VFK.Q
1	2	13/18	0.1365	2.7350	UPSP:TAGL_HUMAN	6	K.QM*EQVAQFLK.A
1	2	13/18	0.0112	2.4931	UPSP:TAGL_HUMAN	6	K.QMEQVAQFLK.A
1	2	18/24	0.3753	4.6052	UPSP:TAGL_HUMAN	6	K.TDM*FQTVDLFEFGK.D
1	3	31/76	0.3029	4.0275	UPSP:TAGL_HUMAN	6	K.TDM*FQTVDLFEFGKDM*AAVQR.T
1	2	13/20	0.1069	2.6708	UPSP:TAGL_HUMAN	6	K.VPENPPSM*VFK.Q
1	2	11/14	0.0086	2.0252	UPSP:TAGL_HUMAN	5	K.YDEELEER.L
1	2	17/20	0.2336	3.1748	UPSP:TAGL_HUMAN	5	R.EFTESQLQEGK.H
1	2	10/14	0.1633	1.8580	UPSP:TAGL_HUMAN	6	R.GDPNWFWM*K.K
1	2	13/14	0.0631	2.8782	UPSP:TAGL_HUMAN	4	R.LGFQVWLK.N
1	2	17/34	0.1523	2.9128	UPSP:TAGL_HUMAN	4	R.LVEWIIVQCGPDVGRPDR.G
1	2	17/22	0.3242	3.7983	UPSP:TAGL_HUMAN	6	R.TLM*ALGSLAVTK.N
1	2	14/40	0.2070	2.1028	UPSP:TAGL2_HUMAN	5	K.DGTVLCELINALYPEGQAPVK.K
1	3	27/84	0.0695	3.0308	UPSP:TAGL2_HUMAN	5	K.DGTVLCELINALYPEGQAPVKK.I
1	2	12/16	0.0544	2.6770	UPSP:TAGL2_HUMAN	6	K.IQASTM*AFK.Q
1	2	15/20	0.1053	2.1763	UPSP:TAGL2_HUMAN	6	K.NVIGLQM*GTNR.G
1	2	21/26	0.3978	4.5299	UPSP:TAGL2_HUMAN	6	K.QM*EQISQFLQAAER.Y
1	2	19/26	0.2311	3.8568	UPSP:TAGL2_HUMAN	6	K.QMEQISQFLQAAER.Y
1	3	28/72	0.2739	4.7564	UPSP:TAGL2_HUMAN	6	K.QYDADLEQILIQWITTQCR.K
1	2	17/26	0.3401	3.9271	UPSP:TAGL2_HUMAN	6	R.DDGLFSGDPNWFVK.K
1	2	16/20	0.1198	3.0801	UPSP:TAGL2_HUMAN	6	R.NFSDNQLQEGK.N
1	2	19/22	0.3503	3.9924	UPSP:TAGL2_HUMAN	6	R.TLM*NLGGLAVAR.D
1	2	25/34	0.3975	5.8066	UPSP:TAGL2_HUMAN	6	R.YGINTTDIFQTVDLWEGK.N

UPSP:TAGL_HUMAN (Q01995, homo sapiens): Transgelin (smooth muscle protein 22-alpha) [MASS=22480]
 ANKGPSYGM S REVQSKIEK **YDEELEERLV EWIIVQCGPD VGRPDRGR LG FQVWLKNGVI LSKLVNSLYP DGSKPKVPE**
NPSPMVFKQM EQVAQFLKAA EDYGVIKTDM FQTVDLFEFGK DMAAVQRTLM ALGSLAVTKN DGHYRGDPNW FMKKAQEHKR
EFTESQLQEG KHVIGLQMG S NRGASQAGMT GYGRPRQIIS

Position	MH+	Sequence
99-107	966.0789	AAEDYGVIK
49-56	991.2211	LGFQVWLK
146-153	995.1407	GDPNWFMK
21-28	1083.0967	YDEELEER
128-139	1205.4986	TLMALGSLAVTK
20-28	1211.2708	KYDEELEER
89-98	1222.4449	QMEQVAQFLK
78-88	1245.4793	VPENPPSMVFK
161-171	1296.3764	EFTESQLQEGK
64-77	1517.7655	LVNSLYPDGSKPKV
108-120	1531.7206	TDMFQTVDLFEFGK
29-46	2053.3868	LVEWIIVQCGPDVGRPDR
108-127	2303.6102	TDMFQTVDLFEFGKDMAAVQR
64-88	2744.2216	LVNSLYPDGSKPKV.VPENPPSMVFK

Protein coverage by amino acid count: 130/200 = **65.0%**
 Protein coverage by mass: 14818/22480 = **65.9%**

UPSP:TAGL2_HUMAN (P37802, homo sapiens): Transgelin-2 (sm22-alpha homolog) [MASS=22260]
 ANRGPAYGLS REVQQKLEKQ **YDADLEQILI QWITTQCRKD VGRPQPGREN FQNLKDGTV LCELINALYP EGQAPVKKIQ**
ASTMAFKQME QISQFLQAAE RYGINTTDIF QTVDLWEGKN MACVQRTLMN LGGLAVARDD GLFSGDPNWF PKKSKEENPRN
FSDNQLQEGK NVIGLQMG TN RGASQAGMTG YGMPRQIL

Position	MH+	Sequence
79-87	997.1975	IQASTMAFK
171-181	1203.4019	NVIGLQMGITNR
127-138	1216.4844	TLMNLGGLAVAR
160-170	1280.3365	NFSDNQLQEGK
139-152	1595.7081	DDGLFSGDPNWFVK
88-101	1679.8883	QMEQISQFLQAAER
102-119	2101.3202	YGINTTDFQTVDLWEGK
57-77	2231.5689	DGTVLCELINALYPEGQAPVK
20-38	2338.6402	QYDADLEQILIQWITTQCR
57-78	2359.7430	DGTVLCELINALYPEGQAPVKK

Protein coverage by amino acid count: 130/198 = **65.7%**

Protein coverage by mass: 14620/22260 = **65.7%**

(2) Spot 3: PPIA

Rank	Charge	Ions	dCn	XCorr	Reference	Redundancy	Peptide
1	2	11/20	0.1456	2.1931	UPTR:Q3KQW3_HUMAN	6	K.EGM*NIVEAM*ER.F
1	2	14/16	0.0115	3.3212	UPTR:Q3KQW3_HUMAN	4	K.FEDENFILK.H
1	3	40/104	0.2938	6.3140	UPTR:Q3KQW3_HUMAN	5	K.HTGPGILSM*ANAGPNTNGSQFFICTA K.T
1	1	12/18	0.2374	2.0223	UPTR:Q3KQW3_HUMAN	4	K.ITIADCGQLE.-
1	2	14/20	0.0544	2.5882	UPTR:Q3KQW3_HUMAN	4	K.KITIADCGQLE.-
1	2	21/28	0.2476	5.5171	UPTR:Q3KQW3_HUMAN	4	K.SIYGEKFEDENFILK.H
1	2	9/12	0.0210	1.6488	UPTR:Q3KQW3_HUMAN	7	K.TEWLDGK.H
1	2	11/24	0.1353	1.8577	UPTR:Q3KQW3_HUMAN	6	K.VKEGM*NIVEAM*ER.F
1	2	19/26	0.3914	1.6097	UPTR:Q3KQW3_HUMAN	5	R.IIPGFM*COGGDFTR.H
1	2	14/16	0.2000	3.3743	UPTR:Q3KQW3_HUMAN	4	R.VSFELFADK.V

UPTR:Q3KQW3_HUMAN (Q3kqw3, homo sapiens): Peptidylprolyl isomerase a, isoform 1 [MASS=18012] MVNPTVFFDI AVDGEPLGRV SFELFADK VPTAENFRALS TGEKGFYK SCFHR IIPGF MCQGGDFTR H NGTGGK SIYG EKFEDENFIL KHTGPGILSM ANAGPNTNGS QFFICTAKTE WLDGKHVVP G KVKEGMNIVE AMERFGSRNG KTSKKITIAD CGQLE

Position	MH+	Sequence
119-125	848.9310	TEWLDGK
20-28	1056.2036	VSFELFADK
156-165	1063.2108	ITIADCGQLE
83-91	1155.2927	FEDENFILK
155-165	1191.3849	KITIADCGQLE
134-144	1279.4689	EGMNIVEAMER
132-144	1506.7756	VKEGMNIVEAMER
56-69	1542.8109	IIPGFMCOGGDFTR
77-91	1833.0478	SIYGEKFEDENFILK
92-118	2736.0924	HTGPGILSMANAGPNTNGSQFFICTAK

Protein coverage by amino acid count: 96/165 = **58.2%**

Protein coverage by mass: 10601/18012 = **58.9%**

(3) Spot 5: TAGLN2

Rank	Charge	Ions	dCn	XCorr	Reference	Redundancy	Peptide
1	2	18/40	0.3948	3.2269	UPSP:TAGL2_HUMAN	5	K.DGTVLCELINALYPEGQAPVK.K
1	3	32/84	0.3908	3.9225	UPSP:TAGL2_HUMAN	5	K.DGTVLCELINALYPEGQAPVKK.I
1	2	11/16	0.0864	1.9669	UPSP:TAGL2_HUMAN	6	K.IQASTM*AFK.Q
1	2	18/20	0.2653	3.7665	UPSP:TAGL2_HUMAN	6	K.NVIGLQM*GTNR.G
1	2	21/26	0.3265	4.4890	UPSP:TAGL2_HUMAN	6	K.QM*EQISQFLQAAER.Y
1	2	20/26	0.2154	3.8847	UPSP:TAGL2_HUMAN	6	K.QMEQISQFLQAAER.Y
1	2	20/36	0.3535	4.6966	UPSP:TAGL2_HUMAN	6	K.QYDADLEQILIQWITTQCR.K
1	2	18/26	0.3376	4.5029	UPSP:TAGL2_HUMAN	6	R.DDGLFSGDPNWFVK.K

1	2	14/28	0.3001	3.7264	UPSP:TAGL2 HUMAN	6	R.DDGLFSGDPNWFPPK.S
1	2	14/20	0.2014	3.2672	UPSP:TAGL2 HUMAN	6	R.NFSDNQLQEGK.N
1	2	20/22	0.3892	4.0798	UPSP:TAGL2 HUMAN	6	R.TLM*NLGGLAVAR.D
1	2	20/22	0.2385	4.3082	UPSP:TAGL2 HUMAN	6	R.TLMNLGGLAVAR.D
1	3	34/68	0.3539	5.7351	UPSP:TAGL2 HUMAN	6	R.YGINTTDIFQTVDLWEGK.N
UPSP:TAGL2_HUMAN (P37802 homo sapiens): Transgelin-2 (sm22-alpha homolog) [MASS=22260]							
ANRGPAYGLS REVQOKIEKQ <u>YDADLEQILI QWITTQCRK</u> D VGRPQPGREN FQNWLKDGTV <u>LCELINALYP EGQAPVKKIQ</u> <u>ASTMAFKQME QISQFLQAAE RYGINTTDIF QTVDLWEGK</u> N MACVQRTLMN <u>LGGLAVARDD GLFSGDPNWF PPKSKENPRN</u> <u>FSDNQLQEGK NVIGLQMGTN RGASQAGMTG YGMPRQIL</u>							

Position	MH+	Sequence
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171-181	1203.4019	NVIGLQMGTNR
127-138	1216.4844	TLMNLGGLAVAR
160-170	1280.3365	NFSDNQLQEGK
139-152	1595.7081	DDGLFSGDPNWFPPK
88-101	1679.8883	QMEQISQFLQAAER
139-153	1723.8822	DDGLFSGDPNWFPPK
102-119	2101.3202	YGINTTDIFQTVDLWEGK
57-77	2231.5689	DGTVLCELINALYPEGQAPVK
20-38	2338.6402	QYDADLEQILIQWITTQCR
57-78	2359.7430	DGTVLCELINALYPEGQAPVKK

Protein coverage by amino acid count: 131/198 = **66.2%**

Protein coverage by mass: 14748/22260 = **66.3%**