Supplementary Material

Improvement of differential diagnosis of lung cancer by use of multiple protein tumor marker combinations

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Supplementary Table 1 Power of discrimination of different groups by single tumor markers and combinations.

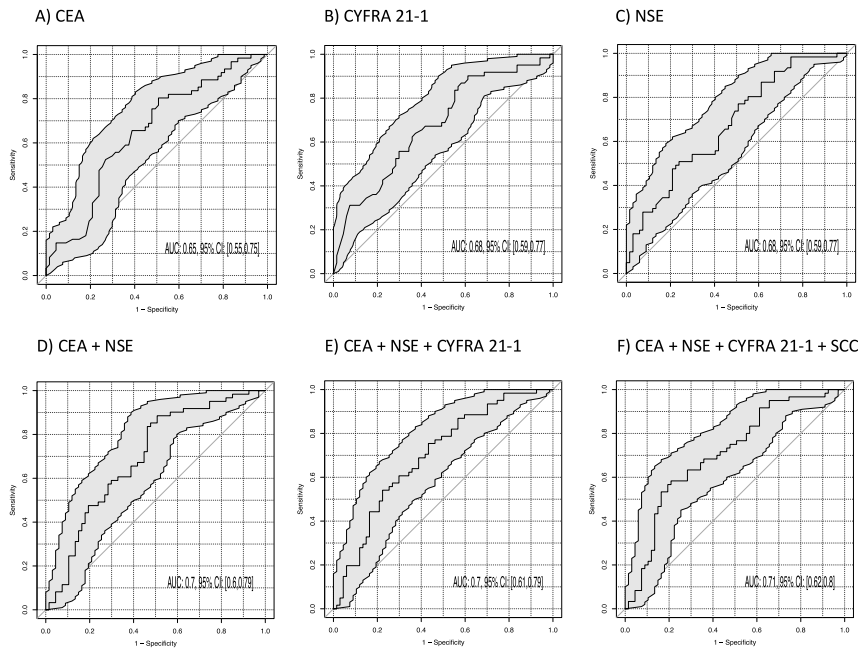|		CEA	CYFRA	NSE	PROGRP	SCC	CA 125	CA 15-3	CA 19-9	CA 72-4	Best double marker combi	Best triple marker combi	Best four marker combi
											SCC+ CA15-3	SCC+ CA15-3+ CA125	SCC+ CA15-3+ CA125+ CEA
NSCLC_ADC* vs NSCLC_SQC	AUC	0.69**	0.58	0.49**	0.5	0.75	0.71**	0.75**	0.6**	0.68**	0.86	0.88	0.89
	95% CI	0.62-0.76	0.5 – 0.66	0.41 – 0.57	0.42 – 0.57	0.69 – 0.82	0.64 – 0.78	0.69 – 0.82	0.53 – 0.75	0.61 – 0.75	0.81 - 0.91	0.83 - 0.93	0.84 - 0.93
	Sens90	17%	16%	9%	9%	37%	24%	24%	13%	16%	60%	61%	66%
	95% CI	0.08 - 0.31	0.05 - 0.29	0.03 - 0.19	0 -	0.28 - 0.59	0.15 - 0.42	0.16 - 0.43	0.06 - 0.24	0.1 - 0.32	0.41 - 0.77	0.49 - 0.80	0.52 - 0.81
	p-value	<0.001	<0.05	0.84	0.9	<0.001	<0.001	<0.001	<0.05	<0.001			
											NSE+ CYFRA	NSE+ CYFRA+ CEA	NSE+ CYFRA+ CEA+ ProGRP
NSCLC loc vs met*	AUC	0.58	0.61	0.63	0.56	0.49	0.58	0.57	0.55	0.54	0.63	0.64	0.63
	95% CI	0.51 – 0.64	0.54 – 0.67	0.56 – 0.69	0.5 – 0.63	0.42 – 0.55	0.51 – 0.64	0.51 – 0.62	0.48 – 0.62	0.47 – 0.61	0.56 - 0.69	0.57 - 0.7	0.57 - 0.7
	Sens90	15%	19%	27%	11%	13%	12%	24%	18%	12%	29%	22%	21%
	95% CI	0.08 - 0.27	0.08 - 0.28	0.13 - 0.38	0.04 - 0.22	0.06 - 0.2	0.06 - 0.25	0.12 - 0.31	0.07 - 0.28	0.05 - 0.22	0.09 - 0.41	0.09 - 0.35	0.09 - 0.38
	p-value	<0.05	<0.01	<0.001	0.06	0.68	<0.05	<0.05	0.15	0.25			
											CEA+ NSE	CEA+ NSE+ CYFRA	CEA+ NSE+ CYFRA+ SCC
SCLC lim vs ext	AUC	0.65**	0.68**	0.68**	0.57**	0.57	0.53**	0.58**	0.56**	0.53	0.70	0.70	0.71
	95% CI	0.55 – 0.75	0.59 – 0.77	0.59 – 0.77	0.47 – 0.67	0.47 – 0.67	0.43 – 0.63	0.48 – 0.68	0.46 – 0.66	0.42 – 0.63	0.60 - 0.79	0.61 - 0.79	0.62 - 0.80
	Sens90	15%	31%	28%	10%	13%	15%	2%	5%	18%	12%	20%	20%
	95% CI	0.07 - 0.26	0.15 - 0.44	0.13 - 0.42	0 - 0.28	0.02 - 0.31	0.05 - 0.32	0 - 0.13	0 - 0.17	0 - 0.28	0.03 - 0.48	0.07 - 0.46	0.03 - 0.57
	p-value	<0.01	<0.001	<0.001	0.19	0.19	0.54	0.1	0.24	0.61			

Areas under the curves (AUCs) of receiver operating characteristic (ROC) curves, 95% confidence intervals (95% CI) and sensitivities at 90% specificities (Sens90). Differences of values between different groups presented as p-values (Mann-Whitney U test). CEA (carcinoembryonic antigen); CYFRA (cytokeratin-19 fragment); NSE (neuron specific enolase); ProGRP (pro-gastrin-releasing peptide); SCC (squamous cell cancer antigen); CA 19-9 (carbohydrate antigen 19-9); CA 15-3 (cancer antigen 15-3); CA 125 (cancer antigen 125); CA 72-4 (cancer antigen 72-4); Small cell lung cancer (SCLC); non-small cell lung cancer (NSCLC); adenocarcinoma (ADC); squamous cell carcinoma (SQC); localized (loc); metastasized (met); limited disease (lim); extensive disease (ext). Classifier for logistic regression (*). Biomarker has negative correlation with classifier (**).
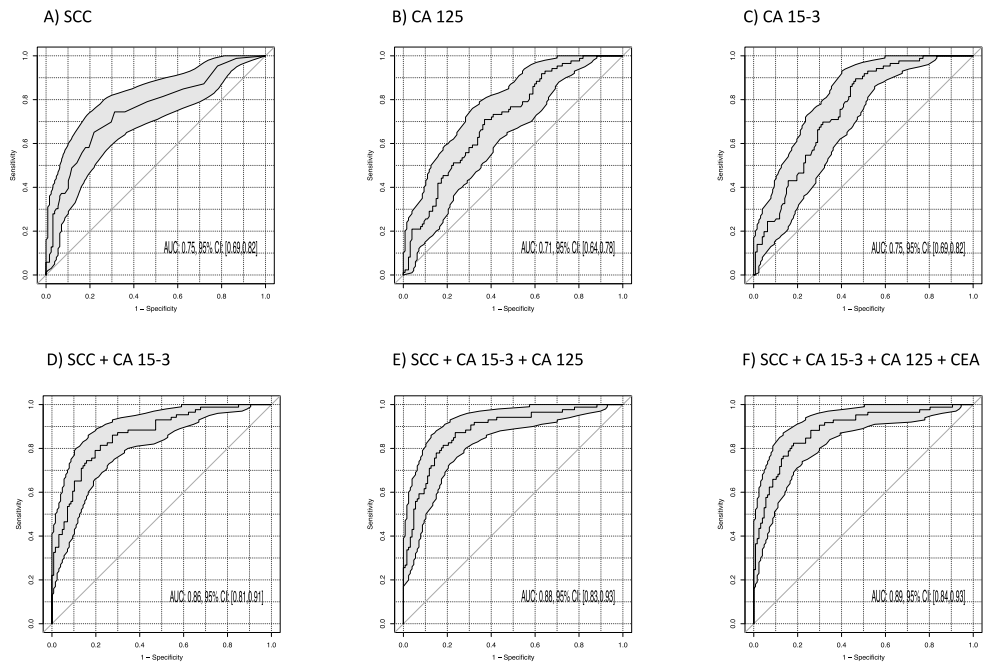
Supplementary Fig. 1 Power of discrimination between patients with adenocellular (ADC) vs squamous cell (SQC) non-small cell lung cancer (NSCLC) by use of single tumor markers and combinations illustrated by receiver operating characteristic (ROC) curves with areas under the curves (AUCs) and respective 95% confidence intervals (95% CI). CEA (carcinoembryonic antigen); SCC (squamous cell cancer antigen); CA 15-3 (cancer antigen 15-3); CA 125 (cancer antigen 125); CA 72-4 (cancer antigen 72-4).
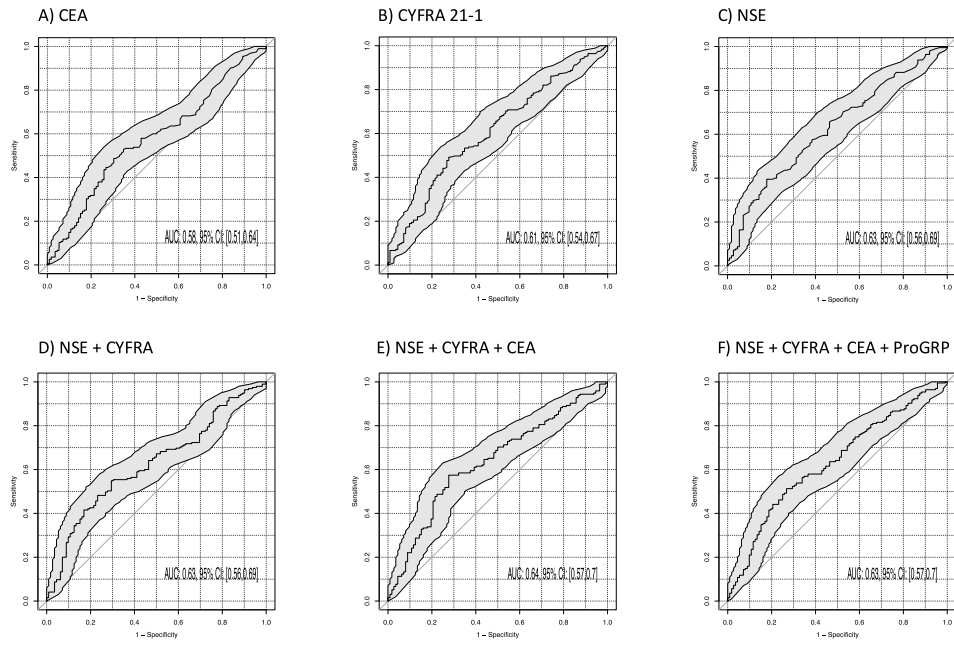
Supplementary Fig. 2 Power of discrimination between patients with localized vs metastatic non-small cell lung cancer (NSCLC) by use of single tumor markers and combinations illustrated by receiver operating characteristic (ROC) curves with areas under the curves (AUCs) and respective 95% confidence intervals (95% CI). CEA (carcinoembryonic antigen); CYFRA (cytokeratin-19 fragment); NSE (neuron specific enolase); ProGRP (pro-gastrin-releasing peptide).

A) CEA   AUC: 0.65, 95% CI: [0.55,0.75]

B) CYFRA 21-1   AUC: 0.68, 95% CI: [0.59,0.77]

C) NSE   AUC: 0.68, 95% CI: [0.59,0.77]

D) CEA + NSE   AUC: 0.7, 95% CI: [0.6,0.79]

E) CEA + NSE + CYFRA 21-1   AUC: 0.7, 95% CI: [0.61,0.79]

F) CEA + NSE + CYFRA 21-1 + SCC   AUC: 0.71, 95% CI: [0.62,0.8]

Supplementary Fig. 3 Power of discrimination between patients with limited vs extensive small cell lung cancer (SCLC) by use of single tumor markers and combinations illustrated by receiver operating characteristic (ROC) curves with areas under the curves (AUCs) and respective 95% confidence intervals (95% CI). CEA (carcinoembryonic antigen); CYFRA (cytokeratin-19 fragment); NSE (neuron specific enolase); SCC (squamous cell cancer antigen).