SUPPLEMENTARY MATERIALS
to the article V.A. Markelov L.Z. Akhmadishina, T.R. Nasibullin, Y.G. Aznabaeva, O.V. Kochetova, N.N. Khusnutdinova, S.M. Izmailova, N.Sh. Zagidullin, G.F. Korytina "The contribution of FOXO family transcription factor genes (*FOXO1*, *FOXO3*) to chronic obstructive pulmonary disease"

Table S1. Baseline demographics and clinical characteristics of the COPD patients and the control group

Parameters	Patients ($N = 710$)	Health ($N = 655$)	*p*-value
Male, *n* (%) Female, *n* (%)	634 (89.29) 76 (10.71)	582 (88.85) 73 (11.15)	0.259
Age (M ± SD)	63.04 ± 12.02	60.67 ± 11.31	0.191
BMI (M ± SD)	26.32 ± 4.75	25.12 ± 4.01	0.07
Smokers, *n* (%) ($N = 1125$) Non-smokers, *n* (%) ($N = 240$)	590 (83.09) 120 (16.91)	535 (81.67) 120 (18.33)	0.74
Pack/years for smokers (M ± SD)	43.08 ± 25.75	39.75 ± 25.87	0.08
Baseline pulmonary function test (M ± SD): FEV_1 (%) FEV_1/FVC (%) FVC % VC (%)	 41.99 ± 19.0 62.29 ± 20.98 56.65 ± 22.71 57.94 ± 15.4	 100.5 ± 21.32 87.82 ± 12.47 103.52 ± 9.14 92.9 ± 21.83	 0.0001
Exacerbation history "rare exacerbator" COPD phenotype "frequent exacerbator" COPD phenotype	 336 (47.32) 374 (52.68)	–	–
GOLD status (http://goldcopd.org) *n* (%) Stage 2 (GOLD II) – moderate, FEV_1 ≥80%, Stage 3 (GOLD III) – severe, 30% ≤ FEV_1 <50% Stage 4 (GOLD IV) – very severe, FEV_1 <30%.	 185 (26.06) 208 (29.30) 317 (44.64)	–	–

Abbreviations: BMI, body mass index; FEV1, forced expiratory volume in 1 s; FVC, forced vital capacity; Mean ± SD: mean values and standard deviation; Pack/years = (number of cigarettes per day × number of years smoked)/20.

Table S2. Functional analysis of the SNPs selected for the study

Gene RefSNP HGVS Names	Chromosomal position	Regulatory		Promotor histone marks	Enhance histone marks	DNAse	Regulatory proteins	Motifs	TFBS	Expression QTLs (Haplo Reg, GTEx portal
		rank	score							
FOXO1 rs12585277 g.20922 G>A (Intron Variant)	13q14.11 (chr13:40650676)	1f	0.55436		12 tissues	Liver	–	Nr2e3, Pou3f4, Pou4f3	–	Brain
FOXO1 rs9549240 g.41186644 T>G (Intron Variant)	13q14.11 (chr13:40612507)	1f	0.22271	Blood	15 tissues	Blood, vessels	–	7 motifs HIF1, Hmx_1, Mtf1_1)	–	6 tissues
FOXO3A rs3800231 g.108998266 A>G (Intron Variant)	6q21 (chr6:108677063	1f	0.55436		17 tissues	–	–	NRSF	–	7 tissues
FOXO3A rs2253310 g.108567390 C>G (Intron Variant)	6q21 (chr6:108567390	1f	0.55436	Blood, brain	19 tissues	–	–	LBP-9, TAL1	–	Blood, pancreas, brain, fibroblast cell cultures

Note. RefSNP according to the NCBI database (https://www.ncbi.nlm.nih.gov); the functions of SNPs were predicted based on RegulomeDB Version 1.1 (https://regulomedb.org), SNPinfo Web Server (https://snpinfo.niehs.nih.gov), HaploReg v3 (https://pubs.broadinstitute.org/mammals/haploreg/haploreg.php), GTEx (https://www.gtexportal.org). DNAse, DNAse hypersensitive regions; Motifs, altered regulatory motifs for binding to transcription regulators; TFBS, Transcription Factor Binding Sites; Regulatory proteins, binding sites to regulatory proteins; Expression QTLs, expression quantitative trait loci.

Table S3. Association of *FOXO1* and *FOXO3A* gene polymorphic loci haplotypes with COPD

Haplotype	Frequency of COPD/control	OR (95%CI)	P_{adj}
FOXO1 rs12585277 G>A – *FOXO1* rs9549240 T>G (*N* = 1365)			
G-G	0.5084/0.5394	1.00	–
A-G	0.2426/0.1881	1.33 (1.07–1.67)	0.011
A-T	0.1887/0.2157	0.97 (0.78–1.19)	0.74
G-T	0.0603/0.0569	1.08 (0.74–1.57)	0.7
P-value of haplotype frequency distribution between groups			0.045
FOXO3A rs3800231 A>G – *FOXO3A* rs2253310 C>G (*N* = 1365)			
G-G	0.494/0.4962	1.00	–
A-G	0.4027/0.3256	1.25 (1.02–1.53)	0.03
A-C	0.0694/0.111	0.65 (0.48–0.89)	0.0076
G-C	0.034/0.0672	0.51 (0.31–0.83)	0.0072
P-value of haplotype frequency distribution between groups			0.00001

Note. *N* – number of individuals included in regression analysis; P_{adj} – significance of the likelihood ratio test of the log-regression model controlling for sex, age, BMI, smoking status and smoking index; OR_{adj} – odds ratio controlling for all these factors, 95 % CI – 95 % confidence interval for OR; D′ – value of normalized linkage disequilibrium coefficient (Lewontin's coefficient) between the two loci; r^2 – correlation coefficient; linkage disequilibrium between rs12585277 and rs9549240 of *FOXO1* (D′ = 0.6183, r^2 = 0.429); rs3800231 and rs2253310 of *FOXO3A* (D′ = 0.3315, r^2 = 0.1452).

Table S4. Association of *FOXO1* and *FOXO3A* gene polymorphic loci haplotypes with the COPD phenotypes

Haplotype	Frequency of COPD/control	OR (95 % CI)	P_{adj}
FOXO1 rs12585277 G>A – *FOXO1* rs9549240 T>G COPD with rare exacerbations (*N* = 991)			
G-G	0.5164/0.5394	1.00	–
A-G	0.2419/0.1881	1.29 (0.99–1.68)	0.06
A-T	0.1751/0.2157	0.88 (0.68–1.15)	0.36
G-T	0.0666/0.0569	1.14 (0.73–1.77)	0.56
P-value of haplotype frequency distribution between groups			0.12
FOXO1 rs12585277 G>A – *FOXO1* rs9549240 T>G COPD with frequent exacerbations (*N* = 1,029)			
G-G	0.5017/0.5394	1.00	–
A-G	0.2429/0.1881	1.36 (1.05–1.76)	0.022
A-T	0.2013/0.2157	1.04 (0.81–1.34)	0.76
G-T	0.0542/0.0569	1.00 (0.63–1.58)	1
P-value of haplotype frequency distribution between groups			0.12
FOXO3A rs3800231 A>G - *FOXO3A* rs2253310 C>G COPD with rare exacerbations (*N* = 991)			
G-G	0.4835/0.4962	1.00	–
A-G	0.4204/0.3256	1.31 (1.02–1.67)	0.032
A-C	0.0615/0.111	0.61 (0.40–0.94)	0.024
G-C	0.0346/0.0672	0.50 (0.25–0.98)	0.045
P-value of haplotype frequency distribution between groups			0.0001
FOXO3A rs3800231 A>G – *FOXO3A* rs2253310 C>G COPD with frequent exacerbations (*N* = 1,029)			
G-G	0.5033/0.4962	1.00	–
A-G	0.3871/0.3256	1.18 (0.92–1.51)	0.19
A-C	0.0757/0.111	0.69 (0.48–1.01)	0.0076
G-C	0.0339/0.0672	0.50 (0.27–0.93)	0.0072
P-value of haplotype frequency distribution between groups			0.00033

Note. *N* – number of individuals included in regression analysis; P_{adj} – significance of the likelihood ratio test of the log-regression model controlling for sex, age, BMI, smoking status and smoking index; OR_{adj}– odds ratio controlling for all these factors, 95 % CI – 95 % confidence interval for OR.

Table S5. Association between the studied *FOXO1* and *FOXO3A* gene polymorphic loci and quantitative phenotypes (lung function parameters and smoking index)

Gene, SNP	Genotype / model	M ± SE	P^a	β (CI 95 %)
smoking index (in pack/years) in the general smoking group (N = 1,125)				
FOXO1 rs9549240 T>G	GG+GT TT recessive	32.52 (0.83) 26.24 (1.81)	0.016	0.00 –6.27 (–11.39– –1.16)
	GG+TT GT	30.37 (0.89) 35.09 (1.49)	0.0042	0.00 4.72 (1.49–7.95)
FOXO3A rs2253310 C>G	GG GC+CC dominant	33.21 (0.9) 28.86 (1.48)	0.012	0.00 –4.35 (–7.75– –0.96)
	GG+CC GC	32.95 (0.88) 29.18 (1.57)	0.037	0.00 –3.77 (–7.29– –0.24)
	log-additive	–	0.0098	–3.87 (–6.80– –0.94)
VC (vital capacity) in %				
FOXO1 rs9549240 T>G	GG GT+TT dominant	57.19 (1.23) 62.46 (1.54)	0.0071	0.00 5.27 (1.45–9.09)
	log-additive	–	0.0044	4.41 (1.39–7.42)
FVC (forced vital capacity) in %				
FOXO1 rs12585277 G>A	GG GA+AA dominant	60.57 (2.00) 55.48 (1.40)	0.04	0.00 –3.92 (–7.76– –0.08)

Note. Mean ± SD – mean values and standard deviation; P^a – significance level for the regression equation; β (95 % CI) – the regression coefficient and 95 % confidence interval for the coefficient. Smoking index in pack/years, (number of cigarettes per day × number of years smoked)/20.