

# **Polymorphisms within autophagy-related genes as susceptibility biomarkers for pancreatic cancer: a meta-analysis of three large European cohorts and functional characterization**

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## ***1. Genotyping technology and quality control of genotyping***

DNA of PANDoRA samples was isolated from whole blood using QIAamp DNA extraction kit (Qiagen, California, USA) and distributed in 384-well plates for genotyping. For quality control, 8% of the samples were randomly replicated throughout the plates, and no-template controls were included in each plate. Genotyping was performed using KASP assay. A QuantStudio 5 instrument and QuantStudio software (Applied Biosystems) were used to detect the genotypes. After genotyping, deviation from HWE distribution was assessed in controls, considering the overall population and dividing by the country of origin of the samples.

## ***2. Imputation and quality controls of imputation***

The genotypes of PDAC cases and controls were downloaded from the database of Genotypes and Phenotypes (dbGaP; study accession nos. phs000206.v5.p3 and phs000648.v1.p1; project reference no. 12644). Genotyping procedures, quality control and data collection details of these studies have been previously described in the original publications[1-4]. After downloading the datasets, we carried out quality controls (QCs) and imputation. The QCs were performed prior to the imputation and included: removal of individuals with gender mismatches, call rate < 0.98, minimal or excessive heterozygosity (> 3 SDs from the mean) or cryptic relatedness (PI\_HAT > 0.2) and exclusion of SNPs with minor allele frequency (MAF) < 0.01, call rate < 0.98 or evidence for violations of HWE ( $p < 1 \times 10^{-6}$ ). The genotypes were phased using SHAPEIT v2 software and the imputation was performed, separately for each dataset, using the Michigan Imputation Server (<https://imputationserver.sph.umich.edu>), and the Haplotype Reference Consortium (HRC) as reference, and merged using PLINK 2.0 software<sup>51</sup>. Afterwards the SNPs with completion rate and call-rate < 98%, a minor allele frequency (MAF) < 0.01, evidence for violations of HWE ( $p < 1 \times 10^{-5}$ )

and low-quality imputation score (INFO score < 0.7) were discarded, leaving 7,509,345 SNPs in the final dataset.

### ***3. PANDoRA ethnic composition***

The PANDoRA consortium has been extensively described elsewhere[5]. Briefly, it consists of a multicentric study conducted in 10 European countries (Italy, Greece, Germany, Netherlands, Denmark, Czech Republic, Hungary, Poland, Lithuania and United Kingdom), and Brazil. Cases had a confirmed diagnosis of PDAC and data on age at diagnosis, sex and country of origin was retrospectively collected for each patient. Controls were selected from blood donors, the general population and hospitalised subjects without oncological diseases. In addition to PANDoRA subjects, the genotypes of British and Dutch controls from the European Prospective Investigation into Cancer and Nutrition (EPIC)[6, 7], a prospective cohort study with 519,978 participants (aged 35–70 years) from ten European countries, and German controls from Epidemiologische Studie zu Chancen der Verhütung, Früherkennung und optimierten THERapie chronischer ERkrankungen in der älteren Bevölkerung (ESTHER)[8], a cohort study that includes 9,961 German people aged between 50 and 74 years, were included.

***4. Supplementary Table 1 (uploaded as separated Excel file due to its large size).***

***5. Supplementary Table 2 (uploaded as separated Excel file due to its large size).***

6. Supplementary Table 3.

**Supplementary Table 3.** Cell types analysed either in whole blood or peripheral mononuclear blood cells.

Number	Name	Parent	Grandparent
1	Leukocytes_CD45P_LMI1	None	None
2	Neutrophils_LMI1	1	None
3	Monocytes_CD14P_LMI1	1	None
4	Classical_monocytes_CD14PPCD16N_LMI1	3	1
5	Intermediate_monocytes_CD14PPCD16P_LMI1	3	1
6	NonClassical_monocytes_CD14PCD16P_LMI1	3	1
7	Lymphocytes_LMI1	1	None
8	T_cells_CD3P_CD56N_LMI1	7	1
9	NK_cells_CD3N_CD56P_LMI1	7	1
10	NKT_cells_CD3P_CD56P_LMI1	7	1
11	B_cells_CD19P_LMI1	7	1
12	CD4P_T_cells_LMI1	8	7
13	CD8P_T_cells_LMI1	8	7
14	DP_CD4P_CD8P_LMI1	8	7
15	DN_CD4N_CD8N_LMI1	8	7
16	CD4P_CD25high_Treg_LMI1	12	8
17	NK_dim_CD56P_CD16P_LMI1	9	7
18	NK_bright_CD56PP_CD16N_LMI1	9	7
19	NK_CD56P_CD16N_LMI1	9	7
20	Leukocytes_CD45P_LMI2	justRef	justRef
21	T_cells_CD3P_CD56N_LMI2	justRef	justRef
22	CD4P_T_cells_LMI2	justRef	justRef
23	CD8P_T_cells_LMI2	justRef	justRef
24	CD45RON_CD45RAP_T_cells_LMI2	21	20
25	CD45ROP_CD45RAP_T_cells_LMI2	21	20
26	CD45ROP_CD45RAN_T_cells_LMI2	21	20
27	CD45RON_CD45RAN_T_cells_LMI2	21	20
28	CD4P_CD25P_CD127low_Treg_LMI2	22	21
29	CD4P_Naive_CD45RAP_CD27P_LMI2	22	21
30	CD4P_Eff_CD45RAP_CD27N_LMI2	22	21
31	CD4P_EM_CD45RAN_CD27N_LMI2	22	21
32	CD4P_CM_CD45RAN_CD27P_LMI2	22	21
33	CD4P_Naive_CD45RON_CD27P_LMI2	22	21
34	CD4P_Eff_CD45RON_CD27N_LMI2	22	21
35	CD4P_EM_CD45ROP_CD27N_LMI2	22	21
36	CD4P_CM_CD45ROP_CD27P_LMI2	22	21
37	CD8P_Naive_CD45RAP_CD27P_LMI2	23	21
38	CD8P_Eff_CD45RAP_CD27N_LMI2	23	21
39	CD8P_EM_CD45RAN_CD27N_LMI2	23	21
40	CD8P_CM_CD45RAN_CD27P_LMI2	23	21

41 CD8P_Naive_CD45RON_CD27P_LMI2	23	21
42 CD8P_Eff_CD45RON_CD27N_LMI2	23	21
43 CD8P_EM_CD45ROP_CD27N_LMI2	23	21
44 CD8P_CM_CD45ROP_CD27P_LMI2	23	21
45 Lymphocytes_LMI3	justRef	justRef
46 B_cells_CD3N_CD19P_LMI3	justRef	justRef
47 CD27N_IgMP_LMI3	justRef	justRef
48 CD19P_CD20N_Plasma_blasts_LMI3	47	46
49 CD19P_CD20P_B_cells_LMI3	47	46
50 IgDN_CD5PP_LMI3	47	46
51 IgDP_CD5PP_LMI3	47	46
52 IgDN_CD5P_LMI3	47	46
53 IgDP_CD5P_LMI3	47	46
54 CD24P_CD38P_LMI3	47	46
55 Transitional_B_cells_CD24PP_CD38PP_LMI3	47	46
56 IgDN_IgMP_LMI3	47	46
57 IgDP_IgMP_LMI3	47	46
58 IgDP_IgMN_LMI3	47	46
59 IgDN_IgMN_LMI3	47	46
60 Transitional_B_cell_CD27N_IgMP_CD24P_CD38high_LMI3	48	47
61 Mature_naive_CD24P_CD38P_LMI3	48	47
62 IgMN_LMI3	55	47
63 CD24P_CD38P_CD27P_IgMP_LMI3	55	47
64 Natural_effector_CD24P_CD38P_IgDP_IgMP_LMI3	55	47
65 Plasmablast_IgDN_IgMN_CD38PP_LMI3	60	47
66 Class_switched_memory_IgDN_IgMN_CD38P_CD27P_LMI3	60	47
67 IgDN_IgMN_CD27N_LMI3	60	47
68 Naive_B_cells_IgDP_IgMP_CD27N_LMI3	58	47
69 Memory_B_cells_IgDP_IgMP_CD27P_LMI3	58	47
70 IgDN_IgMP_CD27N_LMI3	57	47
71 IgM_only_memory_IgDN_IgMP_CD27_LMI3	57	47
72 IgMP_CD38PP_CD27P_LMI3	57+58	47
73 Class_non_switched_memory_IgMP_CD38P_CD27P_LMI3	57+58	47
74 IgMP_CD27N_LMI3	57+58	47
75 Lymphocytes_PBMC_LMI4	justRef	justRef
76 CD4P_T_cells_PBMC_LMI4	justRef	justRef
77 DP_CD4P_CD8P_PBMC_LMI4	justRef	justRef
78 DN_CD4N_CD8N_PBMC_LMI4	justRef	justRef
79 CD8P_T_cells_PBMC_LMI4	justRef	justRef
80 CD25N_CD127P_Tconv_PBMC_LMI4	justRef	justRef
81 CD4P_CD25P_CD127low_Treg_PBMC_LMI4	justRef	justRef
82 Prol_DN_CD4NCD8N_PBMC_LMI4	80	77
83 Prol_DP_CD4PCD8P_PBMC_LMI4	79	77
84 Prol_CD4P_Tconv_PBMC_LMI4	82	78
85 Prol_CD4P_Treg_PBMC_LMI4	83	78

86 Prol_CD8_PBMC_LMI4	81	77
87 Treg_FOXP3P_HeliosP_PBMC_LMI4	83	78
88 Treg_FOXP3P_HeliosN_PBMC_LMI4	83	78
89 Treg_CD45RAP_PBMC_LMI4	83	78
90 Treg_CD45RAN_PBMC_LMI4	83	78
91 Treg_HLANDRP_PBMC_LMI4	83	78

**NOTE:** Cells included in LMI1-3 were measured in whole blood and those included LIM4 in PBMCs.

7. Supplementary Table 4.

**Supplementary Table 4.** Serum and plasma metabolites measured in the HFGP cohort.

Serum_markers				Plasma_markers		
Panel	Assay	Uniprot ID	Units	OlinkID	Assay	Units
	Olink INFLAMMATION(v.3021 4E-BP1	Q13541		OID00536	hsCRP	ug/mL
	Olink INFLAMMATION(v.3021 ADA	P00813		OID00560	IL18bpx	pg/mL
	Olink INFLAMMATION(v.3021 ARTN	Q5T4W7		OID00526	Resistin	ng/mL
	Olink INFLAMMATION(v.3021 AXIN1	O15169		OID00487	Leptin	ng/mL
	Olink INFLAMMATION(v.3021 Beta-NGF	P01138		OID00519	Adiponectin	ug/mL
	Olink INFLAMMATION(v.3021 CASP-8	Q14790		OID00550	AAT	mg/mL
	Olink INFLAMMATION(v.3021 CCL11	P51671		OID00505	IL-1Ra_Q (Quantikine)	pg/mL
	Olink INFLAMMATION(v.3021 CCL19	Q99731		OID00513	IL18	pg/mL
	Olink INFLAMMATION(v.3021 CCL20	P78556		OID00556	IL-1b	pg/mL
	Olink INFLAMMATION(v.3021 CCL23	P55773		OID00530	IL-6	pg/mL
	Olink INFLAMMATION(v.3021 CCL25	O15444		OID00551	VEGF-A	pg/mL
	Olink INFLAMMATION(v.3021 CCL28	Q9NRJ3		OID00539		
	Olink INFLAMMATION(v.3021 CCL3	P10147		OID00532		
	Olink INFLAMMATION(v.3021 CCL4	P13236		OID00498		
	Olink INFLAMMATION(v.3021 CD244	Q9BZW8		OID00477		
	Olink INFLAMMATION(v.3021 CD40	P25942		OID00542		
	Olink INFLAMMATION(v.3021 CD5	P06127		OID00531		
	Olink INFLAMMATION(v.3021 CD6	P30203		OID00499		
	Olink INFLAMMATION(v.3021 CD8A	P01732		OID05124		
	Olink INFLAMMATION(v.3021 CDCP1	Q9H5V8		OID00476		
	Olink INFLAMMATION(v.3021 CSF-1	P09603		OID00562		
	Olink INFLAMMATION(v.3021 CST5	P28325		OID00491		
	Olink INFLAMMATION(v.3021 CX3CL1	P78423		OID00552		
	Olink INFLAMMATION(v.3021 CXCL1	P09341		OID00496		
	Olink INFLAMMATION(v.3021 CXCL10	P02778		OID00535		
	Olink INFLAMMATION(v.3021 CXCL11	O14625		OID00486		
	Olink INFLAMMATION(v.3021 CXCL5	P42830		OID00520		
	Olink INFLAMMATION(v.3021 CXCL6	P80162		OID00534		
	Olink INFLAMMATION(v.3021 CXCL9	Q07325		OID00490		
	Olink INFLAMMATION(v.3021 DNER	Q8NFT8		OID01213		
	Olink INFLAMMATION(v.3021 EN-RAGE	P80511		OID00541		
	Olink INFLAMMATION(v.3021 FGF-19	O95750		OID00545		
	Olink INFLAMMATION(v.3021 FGF-21	Q9NSA1		OID00512		
	Olink INFLAMMATION(v.3021 FGF-23	Q9GZV9		OID00507		
	Olink INFLAMMATION(v.3021 FGF-5	P12034		OID00509		
	Olink INFLAMMATION(v.3021 Flt3L	P49771		OID00533		
	Olink INFLAMMATION(v.3021 GDNF	P39905		OID00475		
	Olink INFLAMMATION(v.3021 HGF	P14210		OID00522		
	Olink INFLAMMATION(v.3021 IFN-gamma	P01579		OID05547		
	Olink INFLAMMATION(v.3021 IL-1 alpha	P01583		OID00493		
	Olink INFLAMMATION(v.3021 IL10	P22301		OID00528		
	Olink INFLAMMATION(v.3021 IL-10RA	Q13651		OID00508		

Olink INFLAMMATION(v.3021 IL-10RB	Q08334	OID00515
Olink INFLAMMATION(v.3021 IL-12B	P29460	OID00523
Olink INFLAMMATION(v.3021 IL13	P35225	OID00525
Olink INFLAMMATION(v.3021 IL-15RA	Q13261	OID00514
Olink INFLAMMATION(v.3021 IL-17A	Q16552	OID00485
Olink INFLAMMATION(v.3021 IL-17C	Q9P0M4	OID00483
Olink INFLAMMATION(v.3021 IL18	Q14116	OID00501
Olink INFLAMMATION(v.3021 IL-18R1	Q13478	OID00517
Olink INFLAMMATION(v.3021 IL2	P60568	OID00495
Olink INFLAMMATION(v.3021 IL-20	Q9NYY1	OID00537
Olink INFLAMMATION(v.3021 IL-20RA	Q9UHF4	OID00489
Olink INFLAMMATION(v.3021 IL-22 RA1	Q8N6P7	OID00516
Olink INFLAMMATION(v.3021 IL-24	Q13007	OID00524
Olink INFLAMMATION(v.3021 IL-2RB	P14784	OID00492
Olink INFLAMMATION(v.3021 IL33	O95760	OID00543
Olink INFLAMMATION(v.3021 IL4	P05112	OID00546
Olink INFLAMMATION(v.3021 IL5	P05113	OID00559
Olink INFLAMMATION(v.3021 IL6	P05231	OID00482
Olink INFLAMMATION(v.3021 IL7	P13232	OID00478
Olink INFLAMMATION(v.3021 IL8	P10145	OID00471
Olink INFLAMMATION(v.3021 LAP TGF-beta-1	P01137	OID00480
Olink INFLAMMATION(v.3021 LIF	P15018	OID00547
Olink INFLAMMATION(v.3021 LIF-R	P42702	OID00511
Olink INFLAMMATION(v.3021 MCP-1	P13500	OID00484
Olink INFLAMMATION(v.3021 MCP-2	P80075	OID00549
Olink INFLAMMATION(v.3021 MCP-3	P80098	OID00474
Olink INFLAMMATION(v.3021 MCP-4	Q99616	OID00504
Olink INFLAMMATION(v.3021 MMP-1	P03956	OID00510
Olink INFLAMMATION(v.3021 MMP-10	P09238	OID00527
Olink INFLAMMATION(v.3021 NRTN	Q99748	OID00548
Olink INFLAMMATION(v.3021 NT-3	P20783	OID00554
Olink INFLAMMATION(v.3021 OPG	O00300	OID00479
Olink INFLAMMATION(v.3021 OSM	P13725	OID00494
Olink INFLAMMATION(v.3021 PD-L1	Q9NZQ7	OID00518
Olink INFLAMMATION(v.3021 SCF	P21583	OID00500
Olink INFLAMMATION(v.3021 SIRT2	Q8IXJ6	OID00538
Olink INFLAMMATION(v.3021 SLAMF1	Q13291	OID00502
Olink INFLAMMATION(v.3021 ST1A1	P50225	OID00557
Olink INFLAMMATION(v.3021 STAMBP	O95630	OID00558
Olink INFLAMMATION(v.3021 TGF-alpha	P01135	OID00503
Olink INFLAMMATION(v.3021 TNF	P01375	OID05548
Olink INFLAMMATION(v.3021 TNFB	P01374	OID00561
Olink INFLAMMATION(v.3021 TNFRSF9	Q07011	OID00553
Olink INFLAMMATION(v.3021 TNFSF14	O43557	OID00506
Olink INFLAMMATION(v.3021 TRAIL	P50591	OID00488
Olink INFLAMMATION(v.3021 TRANCE	O14788	OID00521
Olink INFLAMMATION(v.3021 TSLP	Q969D9	OID00497



Olink INFLAMMATION(v.3021 TWEAK	O43508	OID00555
Olink INFLAMMATION(v.3021 uPA	P00749	OID00481
Olink INFLAMMATION(v.3021 VEGFA	P15692	OID00472

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