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Identification of six new susceptibility loci for invasive epithelial ovarian cancer

Genome-wide association studies (GWAS) have identified 12 epithelial ovarian cancer (EOC) susceptibility alleles. The pattern of association at these loci is consistent in *BRCA1* and *BRCA2* mutation carriers who are at high risk of EOC. After imputation to 1000 Genomes Project data, we assessed associations of 11 million genetic variants with EOC risk from 15,437 cases unselected for family history and 30,845 controls and from 15,252 *BRCA1* mutation carriers and 8,211 *BRCA2* mutation carriers (3,096 with ovarian cancer), and we combined the results in a meta-analysis. This new study design yielded increased statistical power, leading to the discovery of six new EOC susceptibility loci. Variants at 1p36 (nearest gene, *WNT4*), 4q26 (*SYNPO2*), 9q34.2 (*ABO*) and 17q11.2 (*ATAD5*) were associated with EOC risk, and at 1p34.3 (*RSPO1*) and 6p22.1 (*GPX6*) variants were specifically associated with the serous EOC subtype, all with $P < 5 \times 10^{-8}$. Incorporating these variants into risk assessment tools will improve clinical risk predictions for *BRCA1* and *BRCA2* mutation carriers.

The risk of developing invasive EOC is higher than the population average for relatives of women diagnosed with the disease[1,2], indicating the importance of genetic factors in disease susceptibility. Approximately 25% of the familial aggregation of EOC is explained by rare, high-penetrance alleles of *BRCA1* and *BRCA2* (ref. 3). Furthermore, population-based GWAS have identified common variants associated with invasive EOC at 11 loci[4–9], but only 6 have also been evaluated in *BRCA1* and/or *BRCA2* mutation carriers. All loci analyzed displayed associations in mutation carriers that were consistent with the associations observed in the general population[10–12]. In addition, the 4q32.3 locus is associated with EOC risk for *BRCA1* mutation carriers only[13]. However, the common genetic variants identified explain less than 3.1% of the excess familial risk of EOC, so additional susceptibility loci are likely to exist.

Women diagnosed with EOC and unaffected women from the general population ascertained through the Ovarian Cancer Association Consortium (OCAC)[14] and *BRCA1* and *BRCA2* mutation carriers from the Consortium of Investigators of Modifiers of *BRCA1/2* (CIMBA)[15] were genotyped as part of the Collaborative Oncological Gene-environment Study (COGS) using the iCOGS custom array. In addition, data were available for cases and controls from three EOC GWAS. We first evaluated whether the EOC susceptibility loci at 8q21.13, 10p12.31, 17q12, 5p15.33 and 17q21.31 recently identified by OCAC[7–9] also showed evidence of association in *BRCA1* and *BRCA2* mutation carriers. Using data from >200,000 genotyped SNPs[7,13,16], we performed imputation of common variants from 1000 Genomes Project data[17] and evaluated the associations of these SNPs with invasive EOC risk in OCAC samples and in *BRCA1* and *BRCA2* mutation carriers from CIMBA. Given the strong evidence for a significant overlap in loci predisposing to EOC in the general population and those associated with risk in *BRCA1* and *BRCA2* mutation carriers, we carried out a meta-analysis of the EOC risk associations to identify new EOC susceptibility loci.

Genotype data were available for imputation on 15,252 *BRCA1* mutation carriers and 8,211 *BRCA2* mutation carriers, of whom 2,462 and 631, respectively, were affected with EOC[13,16]. For OCAC samples, genotyping data were available from 15,437 women with invasive EOC (including 9,627 with serous EOC) and 30,845 controls from the general population[7]. Imputation was performed separately for *BRCA1* mutation carriers, *BRCA2* mutation carriers, OCAC-COGS samples and samples included in the three OCAC GWAS (**Supplementary Figs. 1** and **2**, and **Supplementary Tables 1** and **2**). The meta-analysis was based on data for 11,403,952 SNPs (**Supplementary Fig. 3**).

Of the five EOC susceptibility loci that had not yet been evaluated in mutation carriers, two were associated with EOC risk for both *BRCA1* and *BRCA2* mutation carriers at $P < 0.05$ (10p12.31 and 17q21.31) (**Supplementary Table 3**). Overall, 7 of the 12 known EOC susceptibility loci provided evidence of association in *BRCA1* mutation carriers and 6 were associated in *BRCA2* mutation carriers. With the exception of 5p15.33 (*TERT*), all loci had hazard ratio (HR) estimates in *BRCA1* and *BRCA2* mutation carriers that were in the same direction as the odds ratio (OR) estimates for the serous subtype EOC samples in OCAC (**Fig. 1**). Analyzing the associations jointly in *BRCA1* and *BRCA2* mutation carriers and serous EOC cases in OCAC provided stronger evidence of association, with smaller *P* values for eight of the susceptibility variants in comparison to the analysis in OCAC samples alone.

Using the imputed genotypes, we observed no new associations at $P < 5 \times 10^{-8}$ in the analysis of associations in *BRCA1* and *BRCA2* mutation carriers separately. However, we identified seven previously unreported associations ($P < 5 \times 10^{-8}$) in OCAC samples alone, in the meta-analysis of EOC associations in *BRCA1* and *BRCA2* mutation carriers and OCAC samples, or in the meta-analysis in *BRCA1* and *BRCA2* mutation carriers and serous EOC cases from OCAC (**Supplementary Fig. 4** and **Supplementary Tables 4** and **5**). SNPs in six of these loci remained genome-wide statistically significant after we reimputed genotypes with imputation parameters set to

A full list of authors and affiliations appears at the end of the paper.

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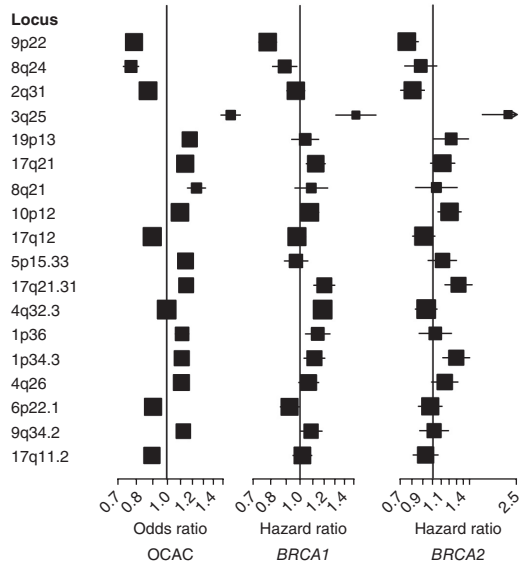

Figure 1 HR estimates for association with EOC of 12 previously reported EOC susceptibility variants and the 6 new susceptibility variants for OCAC samples, *BRCA1* mutation carriers and *BRCA2* mutation carriers. Error bars indicate 95% confidence intervals. The arrow indicates that the confidence interval extends beyond the scale of the *x* axis.

maximize accuracy (**Fig. 1** and **Table 1**). We found SNPs at 17q11.2 (near *ATAD5*) to be associated with invasive EOC in the OCAC samples ($P < 5 \times 10^{-8}$) (**Table 1**). For the lead SNP, chr17:29181220:I, the estimated HR value for *BRCA1* mutation carriers was significantly different from the estimate in OCAC samples ($P = 0.005$); the association for *BRCA2* mutation carriers was consistent with the OCAC OR estimate (*BRCA2*-OCAC meta-analysis $P = 2.6 \times 10^{-9}$). SNPs at four loci were associated at $P < 5 \times 10^{-8}$ with risk of all invasive EOC subtypes in the meta-analysis (**Supplementary Fig. 5**): 1p36, 1p34.3, 4q26 and 9q34.2. At 1p34.3, the most strongly associated SNP, rs58722170, displayed stronger associations in the meta-analysis of serous EOC cases from OCAC ($P = 2.7 \times 10^{-12}$). In addition, SNPs at 6p22.1 were associated at a genome-wide significance level in the meta-analysis of associations with serous EOC ($P = 3.0 \times 10^{-8}$) but not in the meta-analysis of all invasive EOC associations ($P = 6.8 \times 10^{-6}$).

The most significantly associated SNP at each of the six new loci had high imputation accuracy ($r^2 \geq 0.83$). At the 1p34.3, 1p36 and 6p22.1 loci, there was at least one genome-wide significant genotyped SNP correlated with the lead SNP (pairwise $r^2 \geq 0.73$) (**Supplementary Fig. 5**, **Supplementary Table 6** and **Supplementary Note**). We genotyped the leading (imputed) SNPs of the three other loci in a subset of the samples using iPLEX technology (**Supplementary Note**). Correlations between the expected allele dosages from imputation and the observed genotypes for the variants at 4q26 and 9q34.2 ($r^2 = 0.90$ and 0.84, respectively) were consistent with the estimated imputation accuracy scores (0.93 and 0.83 for CIMBA samples). The lead SNP at 17q11.2 failed iPLEX design. However, the risk-associated allele was highly correlated with the AA haplotype of two genotyped variants on the iCOGS array (rs9910051 and rs3764419). This haplotype was strongly associated with ovarian cancer risk in the subset of samples genotyped using the iCOGS array (*BRCA2*-OCAC meta-analysis $P = 8.6 \times 10^{-8}$ for this haplotype and $P = 1.8 \times 10^{-8}$ for chr17:29181220:I) (**Supplementary Table 7**).

None of the regions contained additional SNPs that displayed EOC associations at $P < 1 \times 10^{-4}$ in OCAC samples, *BRCA1* mutation carriers or *BRCA2* mutation carriers in multi-variable analyses adjusted

Table 1 Association test results for loci associated at $P < 5 \times 10^{-8}$ in the second imputation stage

Locus	Nearest gene	rs ID	Ref[g]	Eff[g]	EAF[h]	OCAC all histologies			OCAC serous		*BRCA1* mutation carriers			*BRCA2* mutation carriers			Meta-analysis all histologies[b]	Meta-analysis serous[c]
						$r^{2\,a}$	OR (95% CI)	P	OR (95% CI)	P	$r^{2\,a}$	HR (95% CI)	P	$r^{2\,a}$	HR (95% CI)	P	P	P
1p36	*WNT4*	rs56318008	C	T	0.15	0.98	1.11 (1.07–1.16)	3.9×10^{-7}	1.12 (1.07–1.18)	$\mathbf{3.1 \times 10^{-6}}$	0.98	1.15 (1.05–1.26)	3.1×10^{-3}	0.98	1.03 (0.86–1.23)	0.74	7.6×10^{-9}	5.7×10^{-8}
1p34.3	*RSPO1*	rs58722170	G	C	0.23	0.85	1.08 (1.04–1.12)	9.7×10^{-5}	1.12 (1.08–1.18)	1.1×10^{-7}	0.83	1.14 (1.05–1.23)	1.5×10^{-3}	0.83	1.35 (1.17–1.57)	5.2×10^{-5}	1.6×10^{-8}	2.7×10^{-12}
4q26	*SYNPO2*	rs17329882	A	C	0.24	0.95	1.09 (1.06–1.13)	5.9×10^{-7}	1.11 (1.07–1.16)	6.4×10^{-7}	0.93	1.08 (1.00–1.17)	0.042	0.93	1.15 (1.00–1.33)	0.06	1.4×10^{-8}	1.6×10^{-8}
6p22.1	*GPX6*	rs116133110[e]	T	C	0.31	0.99	0.93 (0.91–0.97)	9.0×10^{-5}	0.91 (0.87–0.94)	2.6×10^{-7}	0.99	0.92 (0.86–0.99)	0.023	0.99	0.97 (0.85–1.10)	0.64	6.8×10^{-6}	3.0×10^{-8}
9q34.2	*ABO*	rs635634	C	T	0.19	0.85	1.11 (1.07–1.16)	1.1×10^{-7}	1.12 (1.08–1.18)	1.0×10^{-6}	0.83	1.11 (1.02–1.21)	0.012	0.83	1.05 (0.89–1.23)	0.55	4.4×10^{-9}	4.2×10^{-8}
17q11.2	*ATAD5*	chr17:29181220:I[f]	A	AT	0.28	0.95	0.91 (0.88–0.94)	5.4×10^{-9}	0.91 (0.87–0.94)	8.1×10^{-7}	0.94	1.01 (0.94–1.08)	0.88	0.93	0.92 (0.80–1.05)	0.23	$2.6 \times 10^{-9\,d}$	$3.9 \times 10^{-7\,d}$

Results are reported for ovarian cancer in *BRCA1* and *BRCA2* mutation carriers, for ovarian cancer as well as the serous subtype of ovarian cancer in OCAC, for the meta-analysis for ovarian cancer, and for the meta-analysis for all tumor histologies in *BRCA1* and *BRCA2* mutation carriers and serous ovarian cancer cases in OCAC. The SNP with the smallest *P* value is reported for each locus.
[a]Imputation accuracy r^2 estimate. [b]*P* value from the meta-analysis association test for ovarian cancer in OCAC samples and *BRCA1* and *BRCA2* mutation carriers. [c]*P* value from the meta-analysis association test for ovarian cancer in *BRCA1* and *BRCA2* mutation carriers and serous ovarian cancer in OCAC samples. [d]Meta-analysis of ovarian cancer associations in *BRCA2* mutation carriers and OCAC samples only. [e]rs116133110 is listed as rs6456822 in dbSNP. [f]chr17:29181220:I is listed as rs199661266 in dbSNP. [g]Reference and effect alleles. [h]Effect allele frequency.

Table 2 Associations with ovarian cancer subtypes in OCAC samples for loci associated with ovarian cancer at $P < 5 \times 10^{-8}$ in the meta-analysis

		All histologies		Serous		Endometrioid		Clear cell		Mucinous		
Locus	rs ID	OR (95% CI)	*P*	OR (95% CI)	*P*	OR (95% CI)	*P*	OR (95% CI)	*P*	OR (95% CI)	*P*	P_{het}[a]
1p36	rs56318008	1.11 (1.06–1.15)	8×10^{-7}	1.12 (1.06–1.17)	6×10^{-6}	1.09 (1.00–1.19)	0.05	1.24 (1.10–1.39	5×10^{-4}	1.03 (0.91–1.17)	0.65	0.22
1p34.3	rs58722170	1.07 (1.03–1.11)	2×10^{-4}	1.12 (1.07–1.17)	4×10^{-7}	0.94 (0.87–1.02)	0.16	1.00 (0.89–1.12)	0.98	1.08 (0.97–1.21)	0.17	0.001
4q26	rs17329882	1.09 (1.06–1.13)	3×10^{-7}	1.11 (1.07–1.16)	3×10^{-7}	1.09 (1.01–1.18)	0.020	1.06 (0.96–1.18)	0.26	1.11 (0.99–1.23)	0.06	0.88
6p22.1	rs116133110	0.94 (0.91–0.97)	9×10^{-5}	0.91 (0.87–0.94)	3×10^{-7}	0.95 (0.89–1.02)	0.16	1.05 (0.95–1.15)	0.34	1.03 (0.94–1.14)	0.53	0.008
9q34.2	rs635634	1.12 (1.08–1.16)	9×10^{-9}	1.13 (1.08–1.18)	2×10^{-7}	1.12 (1.03–1.21)	0.007	1.03 (0.92–1.16)	0.58	1.23 (1.10–1.38)	3×10^{-4}	0.23
17q11.2	chr17:29181 220:I	0.90 (0.87–0.93)	1×10^{-9}	0.90 (0.87–0.94)	2×10^{-7}	0.88 (0.82–0.95)	5×10^{-4}	0.88 (0.80–0.98)	0.020	1.01 (0.91–1.12)	0.84	0.18

[a]*P* value for the heterogeneity in associations with different tumor subtypes.

for the lead SNP in each region, indicating that they each contain only one independent set of correlated, highly associated variants (iCHAVs). Relative to 1000 Genomes Project data, we had genotyped or imputed data covering 91% of the genetic variation at 1p36, 84% of the variation at 1p34.3 and 83% of the variation at 4q26. The other three new loci had coverage of less than 80% (**Supplementary Note**). There was evidence for heterogeneity at $P < 0.05$ in the associations with histological subtype in OCAC samples for the lead SNPs at 1p34.4 and 6p22.1 but not for the lead SNPs at 1p36, 4q26, 9q34.2 and 17q11.2 (**Table 2**).

We carried out a competing risks association analysis in *BRCA1* and *BRCA2* mutation carriers to investigate whether these loci were also associated with breast cancer risk for mutation carriers (**Supplementary Note**). We used the most strongly associated genotyped SNPs for this purpose because the statistical method required actual genotypes[18]. The HR estimates for EOC were consistent with the estimates from the main analysis for all SNPs (**Supplementary Table 8**). None of the SNPs displayed associations with breast cancer risk at $P < 0.05$.

At each of the six loci, we identified a set of SNPs with odds of less than 100 to 1 against them being the causal variant; most were in non-coding DNA regions (**Supplementary Table 9**). None were predicted to have likely deleterious functional effects, although some were in or near chromatin biofeatures in fallopian tube and ovarian epithelial cells, which might represent the functional regulatory targets of the risk-associated SNPs (**Table 3** and **Supplementary Table 10**). We also evaluated the protein-coding genes in each region for their role in EOC development and as candidate susceptibility gene targets. Molecular profiling data from 496 high-grade serous ovarian cancers (HGSOCs) collected by The Cancer Genome Atlas (TCGA) indicated frequent loss or deletion at 4 risk loci (1p36, 4q26, 9q34.2 and 17q11.2) (**Supplementary Table 11**). Consistent with this observation, the expression of *WNT4*, *SYNPO2* and *ABO* was significantly downregulated in ovarian tumors, whereas *ATAD5* expression was upregulated ($P < 6 \times 10^{-5}$, HuEx platform). Somatic coding-sequence mutations in the six genes nearest the index SNPs were rare. We performed expression quantitative trait locus (eQTL) analysis in a series of 59 normal ovarian tissues (**Supplementary Table 12**) to evaluate the gene nearest the top ranked SNP at each locus. For the five genes expressed in normal cells, we found no statistically significant eQTL associations for any of the putative causal SNPs at each locus; neither did we find any significant tumor-eQTL associations for these genes based on data from TCGA (**Supplementary Table 12**). At the 1p36 locus, the most strongly associated variant, rs56318008, was located

Table 3 Summary of data on SNPs, closest gene and all genes in a 1-Mb region for each locus

Loci	Position of top SNP	Number of putatively causal SNPs	Genes in window of putatively causal SNP	Number of SNPs aligned with biofeatures	Normal eQTL closest gene	Tumor DNA copy number	Significant expression difference in tumor versus normal[c]	Known role of gene in cancer	Number of genes in 1-Mb region	Other known cancer genes in 1-Mb region
1p36	Promoter region of *WNT4*	39	*WNT4*, *CDC42*, *LINC00339*	11	NS	Loss	Down	Yes	11	*RAP1GAP*, *CDC42*
1p34.3	Intron 3 of *RSPO1*	15	*RSPO1*	0	NS	Gain		Yes	22	*C1orf109*, *FHL3*
4q26	Intron 3 of *SYNPO2*	4	*SYNPO2*	2	NS[b]	Loss	Down	Yes	12	None
6p22.1	Intron 1 of *GPX6*	22	*GPX6*, *GPX5*	1	NA	Gain			23	*ZKSCAN3*, *TRIM27*
9q34.2	4.3 kb upstream of *ABO*	18	*ABO*, *SLC2A6*[a]	1	NS	Loss	Down	Yes	32	*TSC1*, *RALGDS*, *RPL7A*, *VAV2*
17q11.2	Intron 6 of *ATAD5*	16	*ATAD5*, *TEFM*, *ADAP2*, *CRLF3*, *SUZ12P1*	0	NS	Loss	Up	Yes	17	*NF1*

Proximal promoter regions were defined as the regions 1 kb upstream of the transcription start site. NA indicates no expression of *GPX6* in normal tissues. NS, not significant. Biofeatures are defined as open chromatin H3K4me3 or H3K27ac marks detected in normal ovarian and/or fallopian tube cells.
[a]There are 16 genes in this region—*ABO*, *SURF6*, *MED22*, *RPL7A*, *SNORD24*, *SNORD36B*, *SNORD36A*, *SNORD36C*, *SURF1*, *SURF2*, *SURF4*, *C9orf96*, *REXO4*, *ADAMTS13*, *CACFD1* and *SLC2A6*; however, all SNPs are within or upstream of *ABO* or upstream of *SLC2A6*. [b]Trend $P = 0.067$. [c]$P < 6 \times 10^{-5}$ with the HuEx platform.

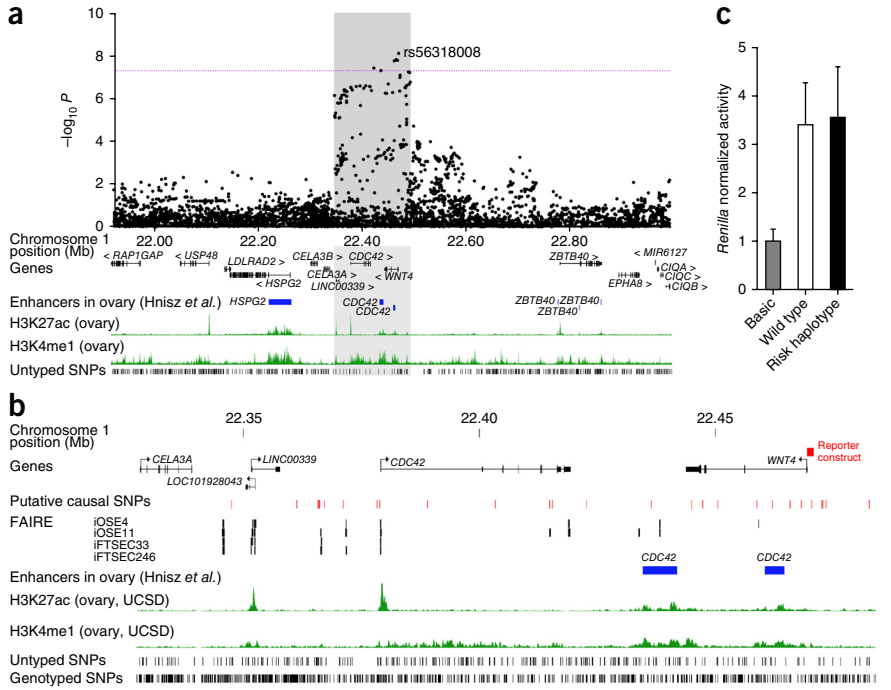

Figure 2 The 1p36 EOC susceptibility locus. (**a**) The Manhattan plot depicts the strength of association between all imputed and genotyped SNPs across the region bound by hg19 coordinates chr. 1: 21,922,893–22,991,643. The dotted line represents the genome-wide significance level of 5×10^{-8}. Additional tracks show genes and enhancers in the ovary as described in Hnisz *et al.*[38]. Positions of SNPs for which imputation $r^2 < 0.3$ and/or minor allele frequency (MAF) < 0.005 are shown in the bottom track as 'untyped' SNPs. H3K27ac, acetylation of histone H3 at lysine 27; H3K4me1, monomethylation of histone H3 at lysine 4. (**b**) The shaded iCHAV region from **a** is shown, depicting the genes and the location of the *WNT4* promoter construct as a red box. Red ticks show the positions of the putative causal variants following likelihood ratio testing. Signals from formaldehyde-assisted regulatory element sequencing (FAIRE-seq) data derived from ovarian cells are represented by black marks, and the locations of predicted *CDC42* enhancers[38] are represented by blue boxes. The positions of genotyped SNPs and those that were neither genotyped nor well imputed ('untyped') are shown. (**c**) Normalized luciferase reporter activity following triplicate transfections of wild-type and risk haplotype *WNT4* promoter constructs in iOSE4 cells. Error bars represent the s.e.m. from three independent experiments.

in the promoter region of *WNT4*, which encodes a ligand in the WNT signal transduction pathway, critical for cell proliferation and differentiation. Using a luciferase reporter assay, we found no effect of these putatively causal SNPs on *WNT4* transcription in iOSE4 normal ovarian cells (**Fig. 2**). Some of the putative causal SNPs at 1p36 were located in *CDC42* and *LINC00339*, and several were in putative regulatory domains in ovarian tissues (**Fig. 2** and **Supplementary Table 10**). *CDC42* is known to have a role in migration and signaling in ovarian and breast cancers[19,20]. SNPs at 1p36 are also associated with increased risk of endometriosis, and *WNT4*, *CDC42* and *LINC00339* have all been implicated in endometriosis[21], a known risk factor for endometrioid and clear cell EOCs[22].

The strongest associated variant at 1q34, rs58722170, was located in *RSPO1*, which encodes R-spondin 1, a protein involved in cell proliferation (**Supplementary Fig. 6**). RSPO1 is important in tumorigenesis and early ovarian development[23,24], and it regulates *WNT4* expression in the ovaries[25]. *SYNPO2* at 4q26 encodes myopodin, which is involved in cell motility and growth[26] and has a reported tumor-suppressor role[27–30]. rs635634 is located upstream of the *ABO* gene (**Supplementary Fig. 7**). A moderately correlated variant (rs505922; $r^2 = 0.52$) determines ABO blood group and is associated with increased risk of pancreatic cancer[31,32]. Previous studies in OCAC also showed a modestly increased risk of EOC for individuals with the A blood group[33]. The moderate correlation between rs635634 and rs505922 and the considerably weaker EOC association of rs505922 ($P = 1.2 \times 10^{-5}$) suggest that the association with blood group is probably not driving the risk association. The indel chr17:29181220:I at 17q11.2 is located in *ATAD5*, which acts as a tumor-suppressor gene[34–36] (**Supplementary Fig. 8**). ATAD5 protein modulates the interaction between RAD9A and BCL2 to induce DNA damage–related apoptosis. Finally, rs116133110, at 6p22.1, lies in *GPX6*, which has no known role in cancer.

The 6 new loci reported in this study increase the number of genome-wide significant common variant loci so far identified for EOC to 18. Taken together, these loci explain approximately 3.9% of the excess familial relative risk of EOC in the general population and account for approximately 5.2% of the polygenic modifying variance for EOC in *BRCA1* mutation carriers and 9.3% of the variance in *BRCA2* mutation carriers. The similarity in the magnitude of the associations between *BRCA1* and *BRCA2* mutation carriers and cases from population-based studies suggests a general model of susceptibility whereby *BRCA1* and *BRCA2* mutations and common alleles interact multiplicatively on the relative risk scale for EOC[37]. This model predicts large differences in absolute EOC risk between individuals carrying many risk-associated alleles and individuals carrying few alleles for EOC susceptibility in *BRCA1* and *BRCA2* mutation carriers[13,16]. Incorporating EOC susceptibility variants into risk assessment tools will improve risk prediction and might be particularly useful for *BRCA1* and *BRCA2* mutation carriers.

URLs. Nature Publishing Group, *Nature Genetics*–iCOGS, http://www.nature.com/icogs/; The Cancer Genome Atlas (TCGA) Project, http://cancergenome.nih.gov/; cBio Cancer Genomics Portal, http://www.cbioportal.org/; Pupasuite 3.1, http://pupasuite.bioinfo.cipf.es/; CIMBA quality control guidelines, http://ccge.medschl.cam.ac.uk/consortia/cimba/members/data%20management/CIMBA%20and%20BCAC%20Quality%20Control%20November%202008%20v2.doc; R software, http://www.r-project.org/.

METHODS

Methods and any associated references are available in the online version of the paper.

Note: Any Supplementary Information and Source Data files are available in the online version of the paper.

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AUTHOR CONTRIBUTIONS

Writing group: K.B.K., A.C.A., G.C.-T., S.J.R., J. Beesley, P.P.P., S.G. Performed statistical analyses for CIMBA: K.B.K. Performed statistical analyses for OCAC: J.T. Performed the meta-analyses: K.B.K. CIMBA database management: L.M. and D.B. Supervised CIMBA statistical analyses, meta-analyses and CIMBA data management: A.C.A. Supervised OCAC statistical analyses: P.P.P. Initiated and coordinated CIMBA: G.C.-T. Coordinated OCAC: A. Berchuck and P.P.P. Conceived and coordinated the synthesis of the iCOGS array: D.F.E. Coordinated iCOGS genotyping: J.S., K. Offit, F.J.C. iCOGS genotyping, calling and quality control: J.M. Cunningham, J.D., P.S., D.F.E., K.B.K., J.T., P.P.P., A.C.A., G.C.-T. Programming support: A. Lee. Provided DNA samples and/or phenotypic data: S.J.R., J.T., A. Lee, H.C.S., K.L., S. Healey, J.M.L., T.J.S., Y.G.L., T.P., Y.B., Q.L., S.C., D.H., A. Miron, M. Southey, M.B.T., D.E.G., S.S.B., R.J., C.M.D., E.J.v.R., S.L.N., Y.C.D., T.V.O.H., L.J., A.-M.G., B.E., J.D., J. Benitez, A.O., M.J.G., I. Komenaka, J.N.W., P.G., P.P., L. Bernard, A.V., B.B., B.P., S. Manoukian, P.R., L.P., L.O., F.F., I. Konstantopoulou, J. Garber, D.F., J. Perkins, R.P., S.E., EMBRACE, A.K.G., R.K.S., A. Meindl, C.E., C.S., O.M.S., GEMO, F.D., S. Mazoyer, D.S.-L., K. Claes, K.D.L., J. Kirk, G.C.R., M. Piedmonte, D.M.O., M.d.l.H., T.C., K.A., H. Nevanlinna, J.M. Collée, M.A. Rookus, J.C.O., F.B.L.H., HEBON, E.O., O.D., I.B., J. Brunet, C.L., M.A.P., A. Jakubowska, J. Gronwald, J. Lubinski, G.S., R.B.B., M. Plante, J.S., P.S., M.M., S. Tognazzo, M.R.T., KConFab, V.S.P., X. Wang, N.L., C.I.S., N.K., J.V., C.A.A., G.P., A. Berger, C.F.S., M.-K.T., C.M.P., M.H.G., P.L.M., G.R., A.M.M., S. Tchatchou, I.L.A., G.G., A.E.T., U.B.J., T.A.K., M. Thomassen, A. Bojesen, J.Z., E.F., Y.L., M. Soller, A. Liljegren, B.A., Z.E., M.S.-A., O.I.O., R.L.N., T.R.R., K.L.N., S.M.D., K.H.L., B.Y.K., C.W., J. Lester, Australian Cancer Society, Australian Ovarian Cancer Study Group, A.H., A.B.E., M.W.B., P.A.F., D. Lambrechts, E.V.N., I.V., S. Lambrechts, E.D., J.A.D., K.G.W., M.A. Rossing, A.R., J.C.-C., S.W.-G., U.E., K.B.M., K. Odunsi, L.S., S. Lele, L.R.W., M.T.G., P.J.T., Y.B.S., I.B.R., M.D., P. Hillemanns, T.D., N.A., N.B., A. Leminen, L.M.P., R.B., F.M., J.L.K., R.P.E., R.B.N., A.d.B., F.H., I.S., P. Harter, K.M., S. Hosono, S.O., A. Jensen, S.K.K., E.H., H.N.H., M.A.N.A., S.-H.T., Y.-L.W., B.L.F., E.L.G., J.M. Cunningham, R.A.V., F.B., G.G.G., D. Liang, M.A.T.H., X. Wu, D.A.L., M.B., A. Berchuck, E.S.I., J.M.S., P.C., R.P.W., D.W.C., K.L.T., E.M.P., S.S.T., E.V.B., I.O., S.H.O., C.K., H.B.S., I.L.T., L. Bjorge, A.M.v.A., K.K.H.A., L.A.K., L.F.A.G.M., M.K., A.B.-W., L.E.K., L.S.C., N.D.L., C.C., H.Y., J. Lissowska, L.A.B., N.W., C.H., L.L., L.N., H.B., H.S., D.E., I.G.C., I.M., J. Paul, K. Carty, N.S., R.G., A.S.W., J.H.R., V.M., W.S., B.-T.J., W.Z., X.-O.S., Y.-T.G., B.R., H.A.R., J.R.M., S.A.N., A.N.M., A.C., H.-Y.L., J.P.-W., T.A.S., Y.-Y.T., Z.C., A.Z., H.A.-C., A.G.-M., U.M., P. Harrington, A.W.L., A.H.W., C.L.P., G.C., M.C.P., A.D.-M., A.T., I.K.R., J. Kupryjanczyk, M.F., H. Noushmehr, L.T., N.T., U.H., C.I., M. Tischkowitz, E.N.I., M.A.C., D.F.E., K. Offit, F.J.C., S.G., P.P.P., A.C.A., G.C.-T. All authors read and approved the final manuscript.

COMPETING FINANCIAL INTERESTS

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1. Auranen, A. *et al.* Cancer incidence in the first-degree relatives of ovarian cancer patients. *Br. J. Cancer* **74**, 280–284 (1996).
2. Stratton, J.F., Pharoah, P., Smith, S.K., Easton, D. & Ponder, B.A. A systematic review and meta-analysis of family history and risk of ovarian cancer. *Br. J. Obstet. Gynaecol.* **105**, 493–499 (1998).
3. Jervis, S. *et al.* Ovarian cancer familial relative risks by tumour subtypes and by known ovarian cancer genetic susceptibility variants. *J. Med. Genet.* **51**, 108–113 (2014).
4. Bolton, K.L. *et al.* Common variants at 19p13 are associated with susceptibility to ovarian cancer. *Nat. Genet.* **42**, 880–884 (2010).
5. Goode, E.L. *et al.* A genome-wide association study identifies susceptibility loci for ovarian cancer at 2q31 and 8q24. *Nat. Genet.* **42**, 874–879 (2010).
6. Song, H. *et al.* A genome-wide association study identifies a new ovarian cancer susceptibility locus on 9p22.2. *Nat. Genet.* **41**, 996–1000 (2009).
7. Pharoah, P.D. *et al.* GWAS meta-analysis and replication identifies three new susceptibility loci for ovarian cancer. *Nat. Genet.* **45**, 362–370 (2013).
8. Permuth-Wey, J. *et al.* Identification and molecular characterization of a new ovarian cancer susceptibility locus at 17q21.31. *Nat. Commun.* **4**, 1627 (2013).
9. Bojesen, S.E. *et al.* Multiple independent variants at the *TERT* locus are associated with telomere length and risks of breast and ovarian cancer. *Nat. Genet.* **45**, 371–384 (2013).
10. Couch, F.J. *et al.* Common variants at the 19p13.1 and *ZNF365* loci are associated with ER subtypes of breast cancer and ovarian cancer risk in *BRCA1* and *BRCA2* mutation carriers. *Cancer Epidemiol. Biomarkers Prev.* **21**, 645–657 (2012).
11. Ramus, S.J. *et al.* Ovarian cancer susceptibility alleles and risk of ovarian cancer in *BRCA1* and *BRCA2* mutation carriers. *Hum. Mutat.* **33**, 690–702 (2012).
12. Ramus, S.J. *et al.* Genetic variation at 9p22.2 and ovarian cancer risk for *BRCA1* and *BRCA2* mutation carriers. *J. Natl. Cancer Inst.* **103**, 105–116 (2011).
13. Couch, F.J. *et al.* Genome-wide association study in *BRCA1* mutation carriers identifies novel loci associated with breast and ovarian cancer risk. *PLoS Genet.* **9**, e1003212 (2013).
14. Bolton, K.L., Ganda, C., Berchuck, A., Pharaoh, P.D. & Gayther, S.A. Role of common genetic variants in ovarian cancer susceptibility and outcome: progress to date from the Ovarian Cancer Association Consortium (OCAC). *J. Intern. Med.* **271**, 366–378 (2012).
15. Chenevix-Trench, G. *et al.* An international initiative to identify genetic modifiers of cancer risk in *BRCA1* and *BRCA2* mutation carriers: the Consortium of Investigators of Modifiers of BRCA1 and BRCA2 (CIMBA). *Breast Cancer Res.* **9**, 104 (2007).
16. Gaudet, M.M. *et al.* Identification of a *BRCA2*-specific modifier locus at 6p24 related to breast cancer risk. *PLoS Genet.* **9**, e1003173 (2013).
17. 1000 Genomes Project Consortium. An integrated map of genetic variation from 1,092 human genomes. *Nature* **491**, 56–65 (2012).
18. Barnes, D.R. *et al.* Evaluation of association methods for analysing modifiers of disease risk in carriers of high-risk mutations. *Genet. Epidemiol.* **36**, 274–291 (2012).
19. Bourguignon, L.Y., Gilad, E., Rothman, K. & Peyrollier, K. Hyaluronan-CD44 interaction with IQGAP1 promotes Cdc42 and ERK signaling, leading to actin binding, Elk-1/estrogen receptor transcriptional activation, and ovarian cancer progression. *J. Biol. Chem.* **280**, 11961–11972 (2005).
20. Zuo, Y., Wu, Y. & Chakraborty, C. Cdc42 negatively regulates intrinsic migration of highly aggressive breast cancer cells. *J. Cell. Physiol.* **227**, 1399–1407 (2012).
21. Pagliardini, L. *et al.* An Italian association study and meta-analysis with previous GWAS confirm *WNT4*, *CDKN2BAS* and *FN1* as the first identified susceptibility loci for endometriosis. *J. Med. Genet.* **50**, 43–46 (2013).
22. Pearce, C.L. *et al.* Association between endometriosis and risk of histological subtypes of ovarian cancer: a pooled analysis of case-control studies. *Lancet Oncol.* **13**, 385–394 (2012).
23. Tomaselli, S. *et al.* Human *RSPO1*/R-spondin1 is expressed during early ovary development and augments β-catenin signaling. *PLoS ONE* **6**, e16366 (2011).
24. Parma, P. *et al.* R-spondin1 is essential in sex determination, skin differentiation and malignancy. *Nat. Genet.* **38**, 1304–1309 (2006).
25. Tomizuka, K. *et al.* R-spondin1 plays an essential role in ovarian development through positively regulating Wnt-4 signaling. *Hum. Mol. Genet.* **17**, 1278–1291 (2008).
26. De Ganck, A. *et al.* Multiple isoforms of the tumor suppressor myopodin are simultaneously transcribed in cancer cells. *Biochem. Biophys. Res. Commun.* **370**, 269–273 (2008).
27. Jing, L. *et al.* Expression of myopodin induces suppression of tumor growth and metastasis. *Am. J. Pathol.* **164**, 1799–1806 (2004).
28. Lin, F. *et al.* Myopodin, a synaptopodin homologue, is frequently deleted in invasive prostate cancers. *Am. J. Pathol.* **159**, 1603–1612 (2001).
29. Sanchez-Carbayo, M., Schwarz, K., Charytonowicz, E., Cordon-Cardo, C. & Mundel, P. Tumor suppressor role for myopodin in bladder cancer: loss of nuclear expression of myopodin is cell-cycle dependent and predicts clinical outcome. *Oncogene* **22**, 5298–5305 (2003).
30. Yu, Y.P. & Luo, J.H. Myopodin-mediated suppression of prostate cancer cell migration involves interaction with zyxin. *Cancer Res.* **66**, 7414–7419 (2006).
31. Amundadottir, L. *et al.* Genome-wide association study identifies variants in the *ABO* locus associated with susceptibility to pancreatic cancer. *Nat. Genet.* **41**, 986–990 (2009).
32. Rummel, S., Shriver, C.D. & Ellsworth, R.E. Relationships between the *ABO* blood group SNP rs505922 and breast cancer phenotypes: a genotype-phenotype correlation study. *BMC Med. Genet.* **13**, 41 (2012).
33. Poole, E.M. *et al.* ABO blood group and risk of epithelial ovarian cancer within the Ovarian Cancer Association Consortium. *Cancer Causes Control* **23**, 1805–1810 (2012).
34. Sikdar, N. *et al.* DNA damage responses by human ELG1 in S phase are important to maintain genomic integrity. *Cell Cycle* **8**, 3199–3207 (2009).

35. Bell, D.W. *et al.* Predisposition to cancer caused by genetic and functional defects of mammalian Atad5. *PLoS Genet.* **7**, e1002245 (2011).
36. Lee, K.Y. *et al.* Human ELG1 regulates the level of ubiquitinated proliferating cell nuclear antigen (PCNA) through Its interactions with PCNA and USP1. *J. Biol. Chem.* **285**, 10362–10369 (2010).
37. Wacholder, S., Han, S.S. & Weinberg, C.R. Inference from a multiplicative model of joint genetic effects for ovarian cancer risk. *J. Natl. Cancer Inst.* **103**, 82–83 (2011).
38. Hnisz, D. *et al.* Super-enhancers in the control of cell identity and disease. *Cell* **155**, 934–947 (2013).

Karoline B Kuchenbaecker[1,203], Susan J Ramus[2,203], Jonathan Tyrer[3,203], Andrew Lee[1], Howard C Shen[2], Jonathan Beesley[4], Kate Lawrenson[2], Lesley McGuffog[1], Sue Healey[4], Janet M Lee[2], Tassja J Spindler[2], Yvonne G Lin[5], Tanja Pejovic[6,7], Yukie Bean[6,7], Qiyuan Li[8], Simon Coetzee[9–11], Dennis Hazelett[2,12], Alexander Miron[13], Melissa Southey[14], Mary Beth Terry[15], David E Goldgar[16], Saundra S Buys[17], Ramunas Janavicius[18,19], Cecilia M Dorfling[20], Elizabeth J van Rensburg[20], Susan L Neuhausen[21], Yuan Chun Ding[21], Thomas V O Hansen[22], Lars Jønson[22], Anne-Marie Gerdes[23], Bent Ejlertsen[24], Daniel Barrowdale[1], Joe Dennis[1,3], Javier Benitez[25–27], Ana Osorio[25,27], Maria Jose Garcia[25,27], Ian Komenaka[28], Jeffrey N Weitzel[29], Pamela Ganschow[30], Paolo Peterlongo[31], Loris Bernard[32,33], Alessandra Viel[34], Bernardo Bonanni[35], Bernard Peissel[36], Siranoush Manoukian[36], Paolo Radice[37], Laura Papi[38], Laura Ottini[39], Florentia Fostira[40], Irene Konstantopoulou[40], Judy Garber[41], Debra Frost[1], Jo Perkins[1], Radka Platte[1], Steve Ellis[1], EMBRACE[42], Andrew K Godwin[43], Rita Katharina Schmutzler[44–47], Alfons Meindl[48], Christoph Engel[49], Christian Sutter[50], Olga M Sinilnikova[51,52], GEMO Study Collaborators[42], Francesca Damiola[51], Sylvie Mazoyer[51], Dominique Stoppa-Lyonnet[53–55], Kathleen Claes[56], Kim De Leeneer[56], Judy Kirk[57], Gustavo C Rodriguez[58], Marion Piedmonte[59], David M O'Malley[60], Miguel de la Hoya[61], Trinidad Caldes[61], Kristiina Aittomäki[62], Heli Nevanlinna[63], J Margriet Collée[64], Matti A Rookus[65], Jan C Oosterwijk[66], Breast Cancer Family Registry[42], Laima Tihomirova[67], Nadine Tung[68], Ute Hamann[69], Claudine Isaccs[70], Marc Tischkowitz[71], Evgeny N Imyanitov[72], Maria A Caligo[73], Ian G Campbell[74], Frans B L Hogervorst[75], HEBON[42], Edith Olah[76], Orland Diez[77], Ignacio Blanco[78], Joan Brunet[79], Conxi Lazaro[80], Miquel Angel Pujana[80], Anna Jakubowska[81], Jacek Gronwald[81], Jan Lubinski[81], Grzegorz Sukiennicki[81], Rosa B Barkardottir[82], Marie Plante[83], Jacques Simard[84], Penny Soucy[84], Marco Montagna[85], Silvia Tognazzo[85], Manuel R Teixeira[86,87], KConFab Investigators[42], Vernon S Pankratz[88], Xianshu Wang[89], Noralane Lindor[88], Csilla I Szabo[90], Noah Kauff[91], Joseph Vijai[91], Carol A Aghajanian[91], Georg Pfeiler[92], Andreas Berger[92], Christian F Singer[92], Muy-Kheng Tea[92], Catherine M Phelan[93], Mark H Greene[94], Phuong L Mai[94], Gad Rennert[95], Anna Marie Mulligan[96,97], Sandrine Tchatchou[98], Irene L Andrulis[96,99], Gord Glendon[98], Amanda Ewart Toland[100], Uffe Birk Jensen[101], Torben A Kruse[102], Mads Thomassen[102], Anders Bojesen[103], Jamal Zidan[104], Eitan Friedman[105], Yael Laitman[105], Maria Soller[106], Annelie Liljegren[107], Brita Arver[107], Zakaria Einbeigi[108], Marie Stenmark-Askmalm[109], Olufunmilayo I Olopade[110], Robert L Nussbaum[111], Timothy R Rebbeck[112], Katherine L Nathanson[112], Susan M Domchek[112], Karen H Lu[113], Beth Y Karlan[114], Christine Walsh[114], Jenny Lester[114], Australian Cancer Study (Ovarian Cancer Investigators)[42], Australian Ovarian Cancer Study Group[42], Alexander Hein[115], Arif B Ekici[116], Matthias W Beckmann[115], Peter A Fasching[115,117], Diether Lambrechts[118,119], Els Van Nieuwenhuysen[120], Ignace Vergote[120], Sandrina Lambrechts[120], Ed Dicks[3], Jennifer A Doherty[121], Kristine G Wicklund[122], Mary Anne Rossing[122,123], Anja Rudolph[124], Jenny Chang-Claude[124], Shan Wang-Gohrke[125], Ursula Eilber[124], Kirsten B Moysich[126], Kunle Odunsi[127], Lara Sucheston[126], Shashi Lele[126], Lynne R Wilkens[128], Marc T Goodman[129,130], Pamela J Thompson[129,130], Yurii B Shvetsov[128], Ingo B Runnebaum[131], Matthias Dürst[131], Peter Hillemanns[132], Thilo Dörk[133], Natalia Antonenkova[134], Natalia Bogdanova[133], Arto Leminen[63], Liisa M Pelttari[63], Ralf Butzow[63,135], Francesmary Modugno[136–139], Joseph L Kelley[137], Robert P Edwards[137,138], Roberta B Ness[140], Andreas du Bois[141,142], Florian Heitz[141,142], Ira Schwaab[143], Philipp Harter[141,142], Keitaro Matsuo[144], Satoyo Hosono[145], Sandra Orsulic[114], Allan Jensen[146], Susanne Kruger Kjaer[146,147], Estrid Hogdall[146,148], Hanis Nazihah Hasmad[149], Mat Adenan Noor Azmi[150], Soo-Hwang Teo[149,151], Yin-Ling Woo[150,151], Brooke L Fridley[152], Ellen L Goode[88], Julie M Cunningham[89], Robert A Vierkant[153], Fiona Bruinsma[154], Graham G Giles[154], Dong Liang[155], Michelle A T Hildebrandt[156], Xifeng Wu[156], Douglas A Levine[157], Maria Bisogna[157], Andrew Berchuck[158], Edwin S Iversen[159], Joellen M Schildkraut[160,161], Patrick Concannon[162,163], Rachel Palmieri Weber[161], Daniel W Cramer[164,165], Kathryn L Terry[164,165], Elizabeth M Poole[166,167], Shelley S Tworoger[166,167], Elisa V Bandera[168], Irene Orlow[169], Sara H Olson[169], Camilla Krakstad[170,171], Helga B Salvesen[170,171], Ingvild L Tangen[170,171], Line Bjorge[170,171], Anne M van Altena[172], Katja K H Aben[173,174], Lambertus A Kiemeney[174,175], Leon F A G Massuger[172],

Melissa Kellar[6,7], Angela Brooks-Wilson[176,177], Linda E Kelemen[178], Linda S Cook[179], Nhu D Le[180], Cezary Cybulski[181], Hannah Yang[182], Jolanta Lissowska[183], Louise A Brinton[182], Nicolas Wentzensen[182], Claus Hogdall[147], Lene Lundvall[147], Lotte Nedergaard[184], Helen Baker[3], Honglin Song[3], Diana Eccles[185], Ian McNeish[186], James Paul[187], Karen Carty[187], Nadeem Siddiqui[188], Rosalind Glasspool[187], Alice S Whittemore[189], Joseph H Rothstein[189], Valerie McGuire[189], Weiva Sieh[189], Bu-Tian Ji[182], Wei Zheng[190], Xiao-Ou Shu[190], Yu-Tang Gao[191], Barry Rosen[192,193], Harvey A Risch[194], John R McLaughlin[195], Steven A Narod[196], Alvaro N Monteiro[93], Ann Chen[197], Hui-Yi Lin[197], Jenny Permuth-Wey[93], Thomas A Sellers[93], Ya-Yu Tsai[93], Zhihua Chen[197], Argyrios Ziogas[198], Hoda Anton-Culver[198], Aleksandra Gentry-Maharaj[199], Usha Menon[199], Patricia Harrington[3], Alice W Lee[2], Anna H Wu[2], Celeste L Pearce[2], Gerry Coetzee[2,12], Malcolm C Pike[2,200], Agnieszka Dansonka-Mieszkowska[201], Agnieszka Timorek[202], Iwona K Rzepecka[201], Jolanta Kupryjanczyk[201], Matt Freedman[8], Houtan Noushmehr[9], Douglas F Easton[1], Kenneth Offit[91], Fergus J Couch[88,89], Simon Gayther[2], Paul P Pharoah[3], Antonis C Antoniou[1,204] & Georgia Chenevix-Trench[4,204] for the Consortium of Investigators of Modifiers of *BRCA1* and *BRCA2*[42]

[1]Centre for Cancer Genetic Epidemiology, Department of Public Health and Primary Care, University of Cambridge, Cambridge, UK. [2]Department of Preventive Medicine, Keck School of Medicine, University of Southern California Norris Comprehensive Cancer Center, Los Angeles, California, USA. [3]Department of Oncology, University of Cambridge, Strangeways Research Laboratory, Cambridge, UK. [4]Cancer Division, QIMR Berghofer Medical Research Institute, Brisbane, Queensland, Australia. [5]Department of Obstetrics and Gynecology, University of Southern California, Los Angeles, California, USA. [6]Department of Obstetrics and Gynecology, Oregon Health and Science University, Portland, Oregon, USA. [7]Knight Cancer Institute, Portland, Oregon, USA. [8]Department of Medical Oncology, Center for Functional Cancer Epigenetics, Dana-Farber Cancer Institute, Boston, Massachusetts, USA. [9]Department of Genetics, Ribeirão Preto Medical School, University of São Paulo, Ribeirão Preto, Brazil. [10]Center for Cell-Based Therapy, Monte Alegre, Ribeirão Preto, Brazil. [11]Center for Integrative Systems Biology, Monte Alegre, Ribeirão Preto, Brazil. [12]Department of Urology, University of Southern California, Los Angeles, California, USA. [13]Department of Genomics and Genome Sciences, Case Western Reserve University Medical School, Cleveland, Ohio, USA. [14]Genetic Epidemiology Laboratory, Department of Pathology, University of Melbourne, Parkville, Victoria, Australia. [15]Department of Epidemiology, Columbia University, New York, New York, USA. [16]Department of Dermatology, University of Utah School of Medicine, Salt Lake City, Utah, USA. [17]Department of Medicine, Huntsman Cancer Institute, University of Utah School of Medicine, Salt Lake City, Utah, USA. [18]Department of Molecular and Regenerative Medicine, Hematology, Oncology and Transfusion Medical Center, Vilinius University Hospital Santariskiu Clinics, Vilnius, Lithuania, Vilnius University Hospital Santariskiu Clinics, Hematology, Oncology and Transfusion Medicine Center, Department of Molecular and Regenerative Medicine, Vilnius, Lithuania. [19]State Research Institute Centre for Innovative Medicine, Vilnius, Lithuania. [20]Department of Genetics, University of Pretoria, Pretoria, South Africa. [21]Department of Population Sciences, Beckman Research Institute of City of Hope, Duarte, California, USA. [22]Center for Genomic Medicine, Rigshospitalet, Copenhagen University Hospital, Copenhagen, Denmark. [23]Department of Clinical Genetics, Rigshospitalet, Copenhagen University Hospital, Copenhagen, Denmark. [24]Department of Oncology, Rigshospitalet, Copenhagen University Hospital, Copenhagen, Denmark. [25]Human Genetics Group, Human Cancer Genetics Program, Spanish National Cancer Centre (CNIO), Madrid, Spain. [26]Human Genotyping Unit (CEGEN), Human Cancer Genetics Program, Spanish National Cancer Centre (CNIO), Madrid, Spain. [27]Biomedical Network on Rare Diseases (CIBERER), Madrid, Spain. [28]Maricopa Medical Center, care of City of Hope Clinical Cancer Genetics Community Research Network, Duarte, California, USA. [29]Clinical Cancer Genetics, for the City of Hope Clinical Cancer Genetics Community Research Network, Duarte, California, USA. [30]Cook County Health and Hospital System, care of City of Hope Clinical Cancer Genetics Community Research Network, Duarte, California, USA. [31]Fondazione Istituto FIRC (Italian Foundation for Cancer Research) di Oncologia Molecolare (IFOM), Milan, Italy. [32]Department of Experimental Oncology, Istituto Europeo di Oncologia, Milan, Italy. [33]Cogentech Cancer Genetic Test Laboratory, Milan, Italy. [34]Division of Experimental Oncology, CRO (Centro di Riferimento Oncologico) Aviano National Cancer Institute, Aviano, Italy. [35]Division of Cancer Prevention and Genetics, Istituto Europeo di Oncologia, Milan, Italy. [36]Unit of Medical Genetics, Department of Preventive and Predictive Medicine, Fondazione IRCCS (Istituto di Ricovero e Cura a Carattere Scientifico (Italian Research Hospital)) Istituto Nazionale Tumori (INT), Milan, Italy. [37]Unit of Molecular Bases of Genetic Risk and Genetic Testing, Department of Preventive and Predictive Medicine, Fondazione IRCCS Istituto Nazionale Tumori (INT), Milan, Italy. [38]Unit of Medical Genetics, Department of Biomedical, Experimental and Clinical Sciences, University of Florence, Florence, Italy. [39]Department of Molecular Medicine, University La Sapienza, Rome, Italy. [40]Molecular Diagnostics Laboratory, INRASTES (Institute of Nuclear and Radiological Sciences and Technology), National Centre for Scientific Research 'Demokritos', Aghia Paraskevi Attikis, Athens, Greece. [41]Cancer Risk and Prevention Clinic, Dana-Farber Cancer Institute, Boston, Massachusetts, USA. [42]A full list of members appears in the **Supplementary Note**. [43]Department of Pathology and Laboratory Medicine, University of Kansas Medical Center, Kansas City, Kansas, USA. [44]Center for Hereditary Breast and Ovarian Cancer, Medical Faculty, University Hospital Cologne, Cologne, Germany. [45]Center for Integrated Oncology (CIO), Medical Faculty, University Hospital Cologne, Cologne, Germany. [46]Center for Molecular Medicine Cologne (CMMC), University of Cologne, Cologne, Germany. [47]On behalf of the German Consortium of Hereditary Breast and Ovarian Cancer (GC-HBOC). [48]Department of Gynaecology and Obstetrics, Division of Tumor Genetics, Klinikum Rechts der Isar, Technical University of Munich, Munich, Germany. [49]Institute for Medical Informatics, Statistics and Epidemiology, University of Leipzig, Leipzig, Germany. [50]Institute of Human Genetics, University Heidelberg, Heidelberg, Germany. [51]INSERM U1052, CNRS UMR 5286, Université Lyon, Centre de Recherche en Cancérologie de Lyon, Lyon, France. [52]Unité Mixte de Génétique Constitutionnelle des Cancers Fréquents, Hospices Civils de Lyon–Centre Léon Bérard, Lyon, France. [53]Institut Curie, Department of Tumour Biology, Paris, France. [54]Institut Curie, INSERM U830, Paris, France. [55]Université Paris Descartes, Sorbonne Paris Cité, Paris, France. [56]Center for Medical Genetics, Ghent University, Ghent, Belgium. [57]Australia New Zealand Gynecologic Oncology Group (ANZGOG) and Familial Cancer Service, Westmead Hosptial, Sydney, New South Wales, Australia. [58]Division of Gynecologic Oncology, NorthShore University HealthSystem, Evanston, Illinois, USA. [59]Gynecologic Oncology Group, Statistical and Data Center, Roswell Park Cancer Institute, Buffalo, New York, USA. [60]James Cancer Center, Ohio State University, Columbus, Ohio, USA. [61]Molecular Oncology Laboratory, Hospital Clinico San Carlos, IdISSC (Instituto de Investigación Sanitaria del Hospital Clínico San Carlos), Madrid, Spain. [62]Department of Clinical Genetics, Helsinki University Central Hospital, Helsinki, Finland. [63]Department of Obstetrics and Gynecology, University of Helsinki and Helsinki University Central Hospital, Helsinki, Finland. [64]Department of Clinical Genetics, Family Cancer Clinic, Erasmus University Medical Center, Rotterdam, the Netherlands. [65]Department of Epidemiology, Netherlands Cancer Institute, Amsterdam, the Netherlands. [66]Department of Genetics, University Medical Center, Groningen University, Groningen, the Netherlands. [67]Latvian Biomedical Research and Study Centre, Riga, Latvia. [68]Department of Medical Oncology, Beth Israel Deaconess Medical Center, Boston, Massachusetts, USA. [69]Molecular Genetics of Breast Cancer, Deutsches Krebsforschungszentrum (DKFZ), Heidelberg, Germany. [70]Lombardi Comprehensive Cancer Center, Georgetown University, Washington, DC, USA. [71]Program in Cancer Genetics, McGill University, Montreal, Quebec, Canada. [72]N.N. Petrov Institute of Oncology, St. Petersburg, Russia. [73]Section of Genetic Oncology, Department of Laboratory Medicine, University of Pisa and University Hospital of Pisa, Pisa, Italy. [74]VBCRC (Victorian Breast Cancer Research Consortium) Cancer Genetics Laboratory, Peter MacCallum Cancer Centre, Melbourne, Victoria, Australia. [75]Family Cancer Clinic, Netherlands Cancer Institute, Amsterdam, the Netherlands. [76]Department of Molecular Genetics, National Institute of Oncology, Budapest, Hungary. [77]Oncogenetics Group, University Hospital Vall d'Hebron, Vall d'Hebron Institute of Oncology (VHIO) and Universitat Autònoma de Barcelona, Barcelona, Spain. [78]Genetic Counseling Unit, Hereditary Cancer Program, IDIBELL (Bellvitge Biomedical Research Institute)–Catalan Institute of Oncology, Barcelona, Spain. [79]Genetic Counseling Unit, Hereditary Cancer Program, IDIBGI (Institut d'Investigació Biomèdica de Girona)–Catalan Institute of Oncology, Girona, Spain. [80]Molecular Diagnostic Unit, Hereditary Cancer Program, IDIBELL–Catalan Institute of Oncology, Barcelona, Spain. [81]Department of Genetics and Pathology, Pomeranian Medical University, Szczecin, Poland. [82]Department of Pathology, Landspitali University Hospital and Biomedical Centre (BMC), Faculty of Medicine, University of Iceland, Reykjavik, Iceland. [83]Gynaecologic Oncology Service, Centre Hospitalier Universitaire de Québec (CHUQ), Quebec City, Quebec, Canada. [84]Centre Hospitalier Universitaire de Québec (CHUQ) Research Center, Laval University, Quebec City, Quebec, Canada. [85]Immunology and Molecular

Oncology Unit, Istituto Oncologico Veneto (IOV)-IRCCS, Padua, Italy. [86]Biomedical Sciences Institute (ICBAS), Porto University, Porto, Portugal. [87]Department of Genetics, Portuguese Oncology Institute, Porto, Portugal. [88]Department of Health Sciences Research, Mayo Clinic, Rochester, Minnesota, USA. [89]Department of Laboratory Medicine and Pathology, Mayo Clinic, Rochester, Minnesota, USA. [90]National Human Genome Research Institute, US National Institutes of Health, Bethesda, Maryland, USA. [91]Department of Medicine, Memorial Sloan-Kettering Cancer Center, New York, New York, USA. [92]Department of Obstetrics and Gynecology, Comprehensive Cancer Center, Medical University of Vienna, Vienna, Austria. [93]Department of Cancer Epidemiology, Moffitt Cancer Center, Tampa, Florida, USA. [94]Clinical Genetics Branch, Division of Cancer Epidemiology and Genetics, National Cancer Institute, US National Institutes of Health, Rockville, Maryland, USA. [95]Department of Community Medicine and Epidemiology, Carmel Medical Center, Haifa, Israel. [96]Department of Laboratory Medicine and Pathobiology, University of Toronto, Toronto, Ontario, Canada. [97]Laboratory Medicine Program, University Health Network, Toronto, Ontario, Canada. [98]Lunenfeld-Tanenbaum Research Institute of Mount Sinai Hospital, Toronto, Ontario, Canada. [99]Department of Molecular Genetics, University of Toronto, Toronto, Ontario, Canada. [100]Department of Molecular Virology, Immunology and Medical Genetics, Ohio State University, Columbus, Ohio, USA. [101]Department of Clinical Genetics, Aarhus University Hospital, Aarhus, Denmark. [102]Department of Clinical Genetics, Odense University Hospital, Odense, Denmark. [103]Department of Clinical Genetics, Vejle Hospital, Vejle, Denmark. [104]Institute of Oncology, Rivka Ziv Medical Center, Zefat, Israel. [105]Susanne Levy Gertner Oncogenetics Unit, Sheba Medical Center, Tel Aviv, Israel. [106]Department of Clinical Genetics, Lund University Hospital, Lund, Sweden. [107]Department of Oncology, Karolinska University Hospital, Stockholm, Sweden. [108]Department of Oncology, Sahlgrenska University Hospital, Gothenburg, Sweden. [109]Division of Clinical Genetics, Department of Clinical and Experimental Medicine, Linköping University, Linköping, Sweden. [110]Center for Clinical Cancer Genetics and Global Health, University of Chicago Medical Center, Chicago, Illinois, USA. [111]Department of Medicine and Genetics, University of California, San Francisco, San Francisco, California, USA. [112]Abramson Cancer Center, Perelman School of Medicine, University of Pennsylvania, Philadelphia, Pennsylvania, USA. [113]Department of Gynecologic Oncology, University of Texas MD Anderson Cancer Center, Houston, Texas, USA. [114]Women's Cancer Program at the Samuel Oschin Comprehensive Cancer Institute, Cedars-Sinai Medical Center, Los Angeles, California, USA. [115]Department of Obstetrics and Gynecology, Erlangen University Hospital, University of Erlangen-Nuremberg, Erlangen, Germany. [116]Institute of Human Genetics, University of Erlangen-Nuremberg, Erlangen, Germany. [117]Division of Hematology and Oncology, Department of Medicine, David Geffen School of Medicine, University of California, Los Angeles, Los Angeles, California, USA. [118]Vesalius Research Center, VIB, Leuven, Belgium. [119]Laboratory for Translational Genetics, Department of Oncology, University of Leuven, Leuven, Belgium. [120]Division of Gynecological Oncology, Department of Oncology, University Hospitals Leuven, Leuven, Belgium. [121]Department of Community and Family Medicine, Section of Biostatistics and Epidemiology, Geisel School of Medicine at Dartmouth, Lebanon, New Hampshire, USA. [122]Program in Epidemiology, Division of Public Health Sciences, Fred Hutchinson Cancer Research Center, Seattle, Washington, USA. [123]Department of Epidemiology, University of Washington, Seattle, Washington, USA. [124]Division of Cancer Epidemiology, German Cancer Research Center (DKFZ), Heidelberg, Germany. [125]Department of Obstetrics and Gynecology, University of Ulm, Ulm, Germany. [126]Department of Cancer Prevention and Control, Roswell Park Cancer Institute, Buffalo, New York, USA. [127]Department of Gynecologic Oncology, Roswell Park Cancer Institute, Buffalo, New York, USA. [128]Cancer Epidemiology Program, University of Hawaii Cancer Center, Honolulu, Hawaii, USA. [129]Cancer Prevention and Control, Samuel Oschin Comprehensive Cancer Institute, Cedars-Sinai Medical Center, Los Angeles, California, USA. [130]Community and Population Health Research Institute, Department of Biomedical Sciences, Cedars-Sinai Medical Center, Los Angeles, California, USA. [131]Department of Gynecology, Jena University Hospital–Friedrich Schiller University, Jena, Germany. [132]Clinics of Obstetrics and Gynaecology, Hannover Medical School, Hannover, Germany. [133]Gynaecology Research Unit, Hannover Medical School, Hannover, Germany. [134]Byelorussian Institute for Oncology and Medical Radiology Aleksandrov N.N., Minsk, Belarus. [135]Department of Pathology, Helsinki University Central Hospital, Helsinki, Finland. [136]Department of Epidemiology, Graduate School of Public Health, University of Pittsburgh, Pittsburgh, Pennsylvania, USA. [137]Department of Obstetrics, Gynecology and Reproductive Sciences, University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, USA. [138]Ovarian Cancer Center of Excellence, University of Pittsburgh, Pittsburgh, Pennsylvania, USA. [139]Women's Cancer Research Program, Magee-Women's Research Institute and University of Pittsburgh Cancer Institute, Pittsburgh, Pennsylvania, USA. [140]University of Texas School of Public Health, Houston, Texas, USA. [141]Department of Gynecology and Gynecologic Oncology, Dr. Horst Schmidt Kliniken Wiesbaden, Wiesbaden, Germany. [142]Department of Gynecology and Gynecologic Oncology, Kliniken Essen-Mitte, Essen, Germany. [143]Institut für Humangenetik Wiesbaden, Wiesbaden, Germany. [144]Department of Preventive Medicine, Kyushu University Faculty of Medical Sciences, Fukuoka, Japan. [145]Division of Epidemiology and Prevention, Aichi Cancer Center Research Institute, Nagoya, Japan. [146]Department of Virus, Lifestyle and Genes, Danish Cancer Society Research Center, Copenhagen, Denmark. [147]Department of Gynecology, Rigshospitalet, University of Copenhagen, Copenhagen, Denmark. [148]Molecular Unit, Department of Pathology, Herlev Hospital, University of Copenhagen, Copenhagen, Denmark. [149]Cancer Research Initiatives Foundation, Sime Darby Medical Centre, Subang Jaya, Malaysia. [150]Department of Obstetrics and Gynaecology, University Malaya Medical Centre, University Malaya, Kuala Lumpur, Malaysia. [151]University Malaya Cancer Research Institute, Faculty of Medicine, University Malaya Medical Centre, University Malaya, Kuala Lumpur, Malaysia. [152]Biostatistics and Informatics Shared Resource, University of Kansas Medical Center, Kansas City, Kansas, USA. [153]Department of Health Science Research, Division of Biomedical Statistics and Informatics, Mayo Clinic, Rochester, Minnesota, USA. [154]Cancer Epidemiology Centre, Cancer Council Victoria, Melbourne, Victoria, Australia. [155]College of Pharmacy and Health Sciences, Texas Southern University, Houston, Texas, USA. [156]Department of Epidemiology, University of Texas MD Anderson Cancer Center, Houston, Texas, USA. [157]Gynecology Service, Department of Surgery, Memorial Sloan-Kettering Cancer Center, New York, New York, USA. [158]Department of Obstetrics and Gynecology, Duke University Medical Center, Durham, North Carolina, USA. [159]Department of Statistical Science, Duke University, Durham, North Carolina, USA. [160]Cancer Control and Population Sciences, Duke Cancer Institute, Durham, North Carolina, USA. [161]Department of Community and Family Medicine, Duke University Medical Center, Durham, North Carolina, USA. [162]Department of Pathology, Immunology and Laboratory Medicine, University of Florida, Gainesville, Florida, USA. [163]Genetics Institute, University of Florida, Gainesville, Florida, USA. [164]Harvard School of Public Health, Boston, Massachusetts, USA. [165]Obstetrics and Gynecology Epidemiology Center, Brigham and Women's Hospital and Harvard Medical School, Boston, Massachusetts, USA. [166]Channing Division of Network Medicine, Brigham and Women's Hospital and Harvard Medical School, Boston, Massachusetts, USA. [167]Department of Epidemiology, Harvard School of Public Health, Boston, Massachusetts, USA. [168]Cancer Prevention and Control, Rutgers Cancer Institute of New Jersey, New Brunswick, New Jersey, USA. [169]Department of Epidemiology and Biostatistics, Epidemiology Service, Memorial Sloan-Kettering Cancer Center, New York, New York, USA. [170]Centre for Cancer Biomarkers, Department of Clinical Medicine, University of Bergen, Bergen, Norway. [171]Department of Gynecology and Obstetrics, Haukeland University Hospital, Bergen, Norway. [172]Department of Gynaecology, Radboud University Medical Centre, Nijmegen, the Netherlands. [173]Comprehensive Cancer Center The Netherlands, Utrecht, the Netherlands. [174]Department for Health Evidence, Radboud University Medical Centre, Nijmegen, the Netherlands. [175]Department of Urology, Radboud University Medical Centre, Nijmegen, the Netherlands. [176]Canada's Michael Smith Genome Sciences Centre, British Columbia Cancer Agency, Vancouver, British Columbia, Canada. [177]Department of Biomedical Physiology and Kinesiology, Simon Fraser University, Burnaby, British Columbia, Canada. [178]Department of Public Health Sciences, College of Medicine, Medical University of South Carolina, Charleston, South Carolina, USA. [179]Division of Epidemiology and Biostatistics, Department of Internal Medicine, University of New Mexico, Albuquerque, New Mexico, USA. [180]Cancer Control Research, British Columbia Cancer Agency, Vancouver, British Columbia, Canada. [181]International Hereditary Cancer Center, Department of Genetics and Pathology, Clinic of Opthalmology, Pomeranian Medical University, Szczecin, Poland. [182]Division of Cancer Epidemiology and Genetics, National Cancer Institute, Bethesda, Maryland, USA. [183]Department of Cancer Epidemiology and Prevention, Maria Sklodowska-Curie Memorial Cancer Center and Institute of Oncology, Warsaw, Poland. [184]Department of Pathology, Rigshospitalet, University of Copenhagen, Copenhagen, Denmark. [185]Wessex Clinical Genetics Service, Princess Anne Hospital, Southampton, UK. [186]Institute of Cancer Sciences, University of Glasgow, Wolfson Wohl Cancer Research Centre, Beatson Institute for Cancer Research, Glasgow, UK. [187]Cancer Research UK Clinical Trials Unit, Glasgow, Beatson West of Scotland Cancer Centre, Glasgow, UK. [188]Department of Gynaecological Oncology, Glasgow Royal Infirmary, Glasgow, UK. [189]Department of Health Research and Policy–Epidemiology, Stanford University School of Medicine, Stanford, California, USA. [190]Vanderbilt University School of Medicine, Nashville, Tennessee, USA. [191]Shanghai Cancer Institute, Shanghai, China. [192]Department of Obstetrics and Gynecology, Faculty of Medicine, University of Toronto, Toronto, Ontario, Canada. [193]Department of Gynecologic-Oncology, Princess Margaret Hospital, Toronto, Ontario, Canada. [194]Department of Chronic Disease Epidemiology, Yale School of Public Health, New Haven, Connecticut, USA. [195]Prosserman Centre for Health Research, Lunenfeld-Tanenbaum Research Institute, Mount Sinai Hospital, Toronto, Ontario, Canada. [196]Women's College Research Institute, University of Toronto, Toronto, Ontario, Canada. [197]Department of Biostatistics, Moffitt Cancer Center, Tampa, Florida, USA. [198]Department of Epidemiology, University of California–Irvine, Irvine, California, USA. [199]Women's Cancer, University College London Elizabeth Garrett Anderson (EGA) Institute for Women's Health, London, UK. [200]Department of Epidemiology and Biostatistics, Memorial Sloan-Kettering Cancer Center, New York, New York, USA. [201]Department of Pathology, Maria Sklodowska-Curie Memorial Cancer Center and Institute of Oncology, Warsaw, Poland. [202]Department of Obstetrics, Gynecology and Oncology, 2nd Faculty of Medicine, Warsaw Medical University and Brodnowski Hospital, Warsaw, Poland. [203]These authors contributed equally to this work. [204]These authors jointly supervised this work. Correspondence should be addressed to G.C.-T. (georgia.trench@qimrberghofer.edu.au).

ONLINE METHODS

Study populations. We obtained data on *BRCA1* and *BRCA2* mutation carriers through CIMBA. Eligibility in CIMBA is restricted to females 18 years or older with pathogenic mutations in *BRCA1* or *BRCA2*. The majority of the participants were sampled through cancer genetics clinics[15], including some related participants. Fifty-four studies from 27 countries contributed data. After quality control, data were available on 15,252 *BRCA1* mutation carriers and 8,211 *BRCA2* mutation carriers, of whom 2,462 and 631, respectively, were affected with EOC (**Supplementary Table 1**).

Data were available for stage 1 of three population-based EOC GWAS. These included 2,165 cases and 2,564 controls from a GWAS from North America ('US GWAS')[39], 1,762 cases and 6,118 controls from a UK-based GWAS ('UK GWAS')[6], and 441 cases and 441 controls from the Mayo GWAS. Furthermore, 11,069 cases and 21,722 controls were genotyped using the iCOGS array ('OCAC-iCOGS' stage data). Overall, 43 studies from 11 countries provided data on 15,437 women diagnosed with invasive EOC, 9,627 of whom were diagnosed with serous EOC, and 30,845 controls from the general population.

All subjects included in this analysis were of European descent and provided written informed consent as well as data and blood samples under ethically approved protocols. Further details of the OCAC and CIMBA study populations as well as the genotyping, quality control and statistical analyses have been described elsewhere[7,13,16].

Genotype data. Genotyping and imputation details for each study are shown in **Supplementary Table 1**.

Confirmatory genotyping of imputed SNPs. To evaluate the accuracy of imputation for the SNPs we found to be associated with EOC risk, we genotyped rs17329882 (4q26) and rs635634 (9q34.2) in a subset of 3,541 subjects from CIMBA using Sequenon's iPLEX technology. The lead SNP at 17q11.2, chr17:29181220:I, failed iPLEX design. We performed quality control of the iPLEX data according to CIMBA guidelines. After quality control, we used the imputation results to generate the expected allele dosage for each genotyped sample and computed the Pearson product-moment correlation coefficient between the expected allele dosage and the observed genotype. The squared correlation coefficient was compared to the imputation accuracy as estimated from the imputation.

Quality control of GWAS and iCOGS genotyping data. We carried out quality control separately for *BRCA1* mutation carriers, *BRCA2* mutation carriers, the three OCAC GWAS and the OCAC-iCOGS samples, but quality criteria were mostly consistent across studies. We excluded samples if they were not of European ancestry, if they had a genotyping call rate of <95%, if they showed low or high heterozygosity, if they were not female or had ambiguous sex or if they were duplicates (cryptic or intended). In the OCAC studies, one individual was excluded from each pair of samples found to be first-degree relatives, and duplicate samples between the iCOGS stage and any of the GWAS were excluded from the iCOGS data. SNPs were excluded if they were monomorphic, had a call rate of < 95%, showed evidence of deviation from Hardy-Weinberg equilibrium or had low concordance between duplicate pairs. For the Mayo GWAS and the UK GWAS, we also excluded rare SNPs (MAF < 1% or allele count < 5, respectively). We visually inspected genotype cluster plots for all SNPs with association $P < 1 \times 10^{-5}$ from each of the newly identified loci. We used the R GenABEL library version 1.6.7 for quality control.

Genotype data were available for analysis from iCOGS for 199,526 SNPs in OCAC-iCOGS samples, 200,720 SNPs in *BRCA1* mutation carriers and 200,908 SNPs in *BRCA2* mutation carriers. After quality control, for the GWAS, data were available on 492,956 SNPs for the US GWAS, 543,529 SNPs for the UK GWAS and 1,587,051 SNPs for the Mayo GWAS (**Supplementary Table 2**).

Imputation. We performed imputation separately for *BRCA1* mutation carriers, *BRCA2* mutation carriers, OCAC-iCOGS samples and each of the OCAC GWAS. We imputed variants from 1000 Genomes Project data using the v3 April 2012 release[17] as the reference panel. For OCAC-iCOGS samples, the UK GWAS and the Mayo GWAS, imputation was based on the 1000 Genomes Project data with singleton sites removed. To improve computation efficiency, we initially used a two-step procedure, which involved pre-phasing in the first step and imputation of the phased data in the second step. We carried out pre-phasing using SHAPEIT software[40]. We used IMPUTE version 2 software for the subsequent imputation[41] for all studies with the exception of the US GWAS, for which the MACH algorithm implemented in Minimac software version 2012.8.15, MACH version 1.0.18, was used. To perform imputation, we divided the data into segments of approximately 5 Mb each. We excluded SNPs from the association analysis if their imputation accuracy was $r^2 < 0.3$, their MAF was <0.005 in *BRCA1* or *BRCA2* mutation carriers or their accuracy was $r^2 < 0.25$ in OCAC-iCOGS samples, the UK GWAS, the US GWAS or the Mayo GWAS.

We performed more accurate imputation for the regions around the new EOC loci from the joint analysis of the data from *BRCA1* and *BRCA2* mutation carriers and the general population (any SNP with association $P < 5 \times 10^{-8}$). The boundaries of these regions were set 500 kb away from any significantly associated SNP in the region. As in the first run, 1000 Genomes Project data v3 were used as the reference panel, and IMPUTE2 software was applied. However, for the second round of imputation, we imputed genotypes without pre-phasing to improve accuracy. To further increase imputation accuracy, we changed some of the default parameters in the imputation procedure. These included an increase in the MCMC iterations to 90 (out of which the first 15 were used as burn-in), an increase in the buffer region to 500 kb and an increase in the number of haplotypes used as templates when phasing observed genotypes to 100. These changes were applied consistently for all data sets.

Statistical analyses. *Association analyses in the unselected ovarian cancer cases and controls from OCAC.* We evaluated the association between genotype and disease using logistic regression by estimating the associations with each additional copy of the minor allele (log-additive models). The analysis was adjusted for study and for population substructure by including the eigenvectors of the first five ancestry-specific principal components as covariates in the model. We used the same approach to evaluate SNP associations with serous ovarian cancer after excluding all cases with any other or unknown tumor subtype. For imputed SNPs, we used expected dosages in the logistic regression model to estimate SNP effect sizes and *P* values. We carried out analyses separately for OCAC-iCOGS samples and the three GWAS and pooled data thereafter using a fixed-effects meta-analysis. We carried out the analysis of reimputed genotypes for putative new susceptibility loci jointly for the OCAC-iCOGS samples and the GWAS samples. All results are based on the combined data from iCOGS and the three GWAS. We used custom written software for the analysis.

Associations in BRCA1 and BRCA2 mutation carriers from CIMBA. We carried out the ovarian cancer association analyses separately for *BRCA1* and *BRCA2* mutation carriers. The primary analysis was carried out within a survival analysis framework, with time to ovarian cancer diagnosis as the endpoint. Mutation carriers were followed until the age of ovarian cancer diagnosis or risk-reducing salpingo-oophorectomy (RRSO) or to the age at last observation. Breast cancer diagnosis was not considered to be a censoring event. To account for the non-random sampling of *BRCA1* and *BRCA2* mutation carriers with respect to their disease status, we conducted the analyses by modeling the retrospective likelihood of the observed genotypes conditional on the disease phenotype[18]. We assessed the associations between genotype and risk of ovarian cancer using the 1-degree-of-freedom score test statistic based on retrospective likelihood[18,42]. To account for the non-independence among related individuals in the sample, we used an adjusted version of the score test statistic, which uses a kinship-adjusted variance of the score[43]. We evaluated associations between imputed genotypes and ovarian cancer risk using a version of the score test as described above but with the posterior genotype probabilities replacing the genotypes. All analyses were stratified by the country of origin of the samples.

We carried out retrospective likelihood analyses in CIMBA using custom written functions in Fortran and Python. The score test statistic was implemented in R version 3.0.1 (ref. 44).

We evaluated whether there was evidence for multiple independent association signals in the region around each newly identified locus by evaluating the associations of genetic variants in the region while adjusting for the SNP with the smallest meta-analysis *P* value in the

respective region. This was done separately for *BRCA1* mutation carriers, *BRCA2* mutation carriers and OCAC samples.

For one of the new associations, it was not possible to confirm the imputation accuracy of the lead SNP chr17:29181220:I at 17q11.2 through genotyping. Therefore, we inferred two-allele haplotypes for rs9910051 and rs3764419, highly correlated with the lead SNP ($r^2 = 0.95$), using an in-house program. These variants were genotyped on the iCOGS array, and this analysis was therefore restricted to 14,733 ovarian cancer cases and 9,165 controls from OCAC-COGS and 8,185 *BRCA2* mutation carriers for whom genotypes were available for both variants based on iCOGS. The association between the AA haplotype and risk was tested using logistic regression in OCAC samples and using Cox regression in *BRCA2* mutation carriers.

Meta-analysis. We conducted a meta-analysis of the EOC associations in *BRCA1* mutation carriers, *BRCA2* mutation carriers and the general population for genotyped and imputed SNPs using an inverse variance approach assuming fixed effects. We combined the logarithm of the per-allele HR estimate for the association with EOC risk in *BRCA1* and *BRCA2* mutation carriers and the logarithm of the per-allele OR estimate for the association with disease status in OCAC. For associations in *BRCA1* and *BRCA2* carriers, we used the kinship-adjusted variance estimator[43], which allows for the inclusion of related individuals in the analysis. We only used SNPs with results in OCAC and in at least one of the *BRCA1* or the *BRCA2* analyses. We carried out two separate meta-analyses, one for the associations with EOC in *BRCA1* mutation carriers, *BRCA2* mutation carriers and EOC samples in OCAC, irrespective of tumor histological subtype, and a second using only the associations with serous EOC in OCAC samples. The number of *BRCA1* and *BRCA2* mutation carriers with tumor histology information was too small to allow for subgroup analyses. However, previous studies have demonstrated that the majority of EOCs in *BRCA1* and *BRCA2* mutation carriers are high-grade serous[45–49]. Meta-analyses were carried out using Metal software, 2011-03-25 release[50].

Candidate causal SNPs in each susceptibility region. To identify a set of potentially causal variants, we excluded SNPs with a likelihood of being causal of less than 1:100, by comparing the likelihood of each SNP from the association analysis with the likelihood of the most strongly associated SNPs[51]. The remaining variants were then analyzed using Pupasuite 3.1 to identify potentially functional variants[52,53] (**Supplementary Table 9**).

Functional analysis. *Expression quantitative trait locus analysis in normal ovarian and fallopian tube cells.* Early-passage primary normal ovarian surface epithelial cells (OSECs) and fallopian tube epithelial cells were collected from disease-free ovaries and fallopian tubes. Normal ovarian epithelial cells were collected by brushing the surface of the ovary with a sterile cytobrush and were cultured in NOSE-CM[54]. Fallopian tube epithelial cells were collected by Pronase digestion as previously described[55], plated onto collagen-coated plastics (Sigma) and cultured in DMEM/F12 (Sigma-Aldrich) supplemented with 2% Ultroser G (BioSepra) and 1× penicillin-streptomycin (Lonza). By the time of RNA isolation, the fallopian tube cultures tested consisted of PAX8-positive fallopian tube secretory epithelial cells (FTSECs), consistent with previous observations that ciliated epithelial cells from the fallopian tube do not proliferate *in vitro*. Cell lines were routinely tested for mycoplasma.

For gene expression analysis, RNA was isolated from 59 early-passage samples: 54 OSECs and 5 FTSECs from cell cultures collected at ~80% confluency using the Qiagen miRNAeasy kit with on-column DNase I digestion. RNA (500 ng) was reverse transcribed using the Superscript III kit (Life Technologies). We preamplified 10 ng of cDNA using TaqMan Preamp Mastermix; the resulting product was diluted 1:60 and used to quantify gene expression with the following TaqMan gene expression probes: *WNT4*, Hs01573504_m1; *RSPO1*, Hs00543475_m1; *SYNPO2*, Hs00326493_m1; *ATAD5*, Hs00227495_m1; and *GPX6*, Hs00699698_m1. Four control genes were also included: *ACTB*, Hs00357333_g1; *GAPDH*, Hs02758991_g1; *HMBS*, Hs00609293_g1; and *HPRT1*, Hs02800695_m1 (all Life Technologies). Assays were run on an ABI 7900HT Fast Real-Time PCR system (Life Technologies).

Data analysis. Expression levels for each gene were normalized to the average of all four control genes. Relative expression levels were calculated using the $\Delta\Delta C_t$ method. Genotyping was performed on the iCOGS chips, as described above. Where genotyping data were not available for the most risk-associated SNP, the next most significant SNP was used: rs3820282 at 1p36, rs12023270 at 1p34.3, rs752097 at 4q26, rs445870 at 6p22.1, rs505922 at 9q34.2 and rs3764419 at 17q11.2. Correlations between genotype and gene expression were calculated in R. Genotype-specific gene expression in normal tissue cell lines (eQTL analysis) was compared using the Jonckheere-Terpstra test. Data were normalized to the four control genes, and we tested for eQTL associations, grouping OSECs and FTSECs together. Second, OSECs were analyzed alone. eQTL analyses were performed using three genotype groups or two groups (with the rare homozygote samples grouped together with the heterozygote samples).

eQTL analysis in primary ovarian tumors. eQTL analysis in primary tumors was based on publicly available data from the TCGA Project, which included 489 primary HGSOCs. The methods have been described elsewhere[56]. Briefly, we determined the ancestry for each case on the basis of germline genotype data using EIGENSTRAT software with 415 HapMap genotype profiles as a control set. Only populations of northern and western European ancestry were included. We first performed a *cis*-eQTL analyses using a method we described previously, in which the association between 906,600 germline genotypes and the expression levels of mRNA or miRNA (located within 500 kb on either side of the variant) were evaluated using a linear regression model with the effects of somatic copy number and CpG methylation being deducted. (For miRNA expression, the effect of CpG methylation was not adjusted for because these data were not available.) To correct for multiple tests, we adjusted the test *P* values using the Benjamini-Hochberg method. A significant association was defined by a false discovery rate (FDR) of <0.1.

Having established genome-wide *cis*-eQTL associations in this series of tumors, we then evaluated *cis*-eQTL associations for the top risk associations between each of the six new loci and the gene in closest proximity to the risk SNP. For each risk locus, we retrieved the genotype of all SNPs in ovarian cancer cases on the basis of the Affymetrix 6.0 array. Using these genotypes and the IMPUTE2 March 2012 1000 Genomes Project Phase I integrated variant cosmopolitan reference panel of 1,092 individuals (haplotypes were phased via SHAPEIT), we imputed the genotypes of SNPs in the 1000 Genomes Project in the target regions for TCGA samples[57]. For each risk locus where data for the most risk-associated variant were not available, we retrieved the imputed variants tightly correlated with the most risk-associated variant. We then tested for association between imputed SNPs and gene expression using the linear regression algorithm described above, where each imputed SNP was coded as an expected allele count. Again, significant associations were defined by an FDR of <0.1.

Regulatory profiling of normal ovarian cancer precursor tissues. We performed genome-wide FAIRE and chromatin immunoprecipitation with sequencing (ChIP-seq) for H3K27ac and H3K4me in two normal OSECs, two normal FTSECs and two HGSOC cell lines (UWB1.289 and CAOV3) (S.C., H.S., D.H., K.L. and K.B.K. *et al.*, unpublished data). Cell lines were routinely tested for mycoplasma. These data sets annotate the epigenetic signatures of open chromatin and collectively indicate transcriptional enhancer regions. We analyzed the FAIRE-seq and ChIP-seq data sets and publically available genomic data on promoter and UTR domains, intron-exon boundaries and the positions of noncoding RNA transcripts to identify SNPs from the 100:1 likely causal set that aligned with biofeatures that might provide evidence of SNP functionality.

Candidate gene analysis using genome-wide profiling of primary ovarian cancers. *Data sets: the TCGA Project and COSMIC data sets.* TCGA has performed extensive genomic analysis of tumors from a large number of tissue types, including almost 500 high-grade serous ovarian tumors. These data include somatic mutations, DNA copy number, mRNA and miRNA expression, and DNA methylation. COSMIC is the catalog of somatic mutations in cancer that collates information on mutations in tumors from the published literature[58]. They have also identified the Cancer Gene Census, which is a list of genes known to be involved in cancer. Data are available on a large number of tissue types, including 2,809 epithelial ovarian tumors.

Somatic coding sequence mutations. We analyzed all genes for coding somatic sequence mutations generated from either whole-exome or whole-genome sequencing. In TCGA, whole-exome sequencing data were available for 316 high-grade serous EOC cases. In addition, we determined whether mutations had been reported in COSMIC[58] and whether the gene was a known cancer gene in the Sanger Cancer Gene Census.

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mRNA expression in tumor and normal tissue. Normalized and gene expression values (level 3) from gene expression profiling data were obtained from the TCGA data portal for three different platforms (Agilent, Affymetrix HuEx and Affymetrix U133A). We analyzed only the 489 primary serous ovarian tumor samples included in the final clustering analysis[57] and 8 normal fallopian tube samples. The boxplot function in R was used to compare ovarian tumor samples to the fallopian tube samples for 91 coding genes with expression data on any platform within a 1-Mb region around the most significant SNP at the 6 loci. A difference in relative expression between EOC samples and normal tissue was analyzed using the Wilcoxon rank-sum test.

DNA copy number analysis. Serous EOC samples for 481 tumors with $\log_2$ copy number data were analyzed using the cBio Portal for the analysis of TCGA data[59,60]. For each gene in a region, the classes of copy number; homozygous deletion, heterozygous loss, diploid, gain and amplification were queried individually using the advanced onco query language (OQL) option. At a region, the frequency of gain and amplification were combined as 'gain', and homozygous deletion and heterozygous loss were combined as 'loss'.

Analysis of copy number versus mRNA expression. Serous EOC samples for 316 complete tumors (those with CNA, mRNA and sequencing data) were analyzed. Graphs were generated using the cBio Portal for the analysis of TCGA data, and the settings were mRNA expression data *z* score (all genes) with a *z*-score threshold of 2 (default setting) and putative CNAs (GISTIC). The *z* score was the number of s.d. away from the mean of expression in the reference population. GISTIC is an algorithm that attempts to identify significantly altered regions of amplification or deletion across sets of patients.

Luciferase reporter assays. The putative causal SNPs at the 1p36 locus lie in the *WNT4* promoter, and we therefore tested their effect on transcription in a luciferase reporter assay (**Fig. 2d**). Wild-type and risk haplotype (comprising five correlated variants) sequences corresponding to the region bound by hg19 coordinates chr. 1: 22,469,416–22,470,869 were generated by Custom Gene Synthesis (GenScript) and then subcloned into pGL3-basic (Promega). Equimolar amounts of luciferase constructs (800 ng) and pRL-TK *Renilla* (50 ng) were cotransfected into ~8×10^4 iOSE4 (ref. 61) normal ovarian cells in triplicate wells of 24-well plates using Lipofectamine 2000 (Life Technologies). Independent transfections were repeated three times. The Dual-Glo Luciferase Assay kit (Promega) was used to assay luciferase activity 24 h after transfection using a BioTek Synergy H4 plate reader. Statistical significance was tested by log transforming the data and performing two-way ANOVA, followed by Dunnett's multiple-comparisons test in GraphPad Prism. The iOSE4 cell line (derived by K. Lawrenson) was maintained under standard conditions; it was routinely tested for mycoplasma and underwent short tandem repeat profiling.

39. Permuth-Wey, J. *et al. LIN28B* polymorphisms influence susceptibility to epithelial ovarian cancer. *Cancer Res.* **71**, 3896–3903 (2011).
40. Delaneau, O., Marchini, J. & Zagury, J.F. A linear complexity phasing method for thousands of genomes. *Nat. Methods* **9**, 179–181 (2012).
41. Howie, B.N., Donnelly, P. & Marchini, J. A flexible and accurate genotype imputation method for the next generation of genome-wide association studies. *PLoS Genet.* **5**, e1000529 (2009).
42. Antoniou, A.C. *et al. RAD51* 135G→C modifies breast cancer risk among *BRCA2* mutation carriers: results from a combined analysis of 19 studies. *Am. J. Hum. Genet.* **81**, 1186–1200 (2007).
43. Antoniou, A.C. *et al.* A locus on 19p13 modifies risk of breast cancer in *BRCA1* mutation carriers and is associated with hormone receptor-negative breast cancer in the general population. *Nat. Genet.* **42**, 885–892 (2010).
44. *R: A Language and Environment for Statistical Computing v. 3.0.1* (R Foundation for Statistical Computing, 2013).
45. Mavaddat, N. *et al.* Pathology of breast and ovarian cancers among *BRCA1* and *BRCA2* mutation carriers: results from the Consortium of Investigators of Modifiers of BRCA1/2 (CIMBA). *Cancer Epidemiol. Biomarkers Prev.* **21**, 134–147 (2012).
46. Lakhani, S.R. *et al.* Pathology of ovarian cancers in BRCA1 and BRCA2 carriers. *Clin. Cancer Res.* **10**, 2473–2481 (2004).
47. Rubin, S.C. *et al.* Clinical and pathological features of ovarian cancer in women with germ-line mutations of *BRCA1*. *N. Engl. J. Med.* **335**, 1413–1416 (1996).
48. Maehle, L. *et al.* High risk for ovarian cancer in a prospective series is restricted to *BRCA1/2* mutation carriers. *Clin. Cancer Res.* **14**, 7569–7573 (2008).
49. Shaw, P.A. *et al.* Histopathologic features of genetically determined ovarian cancer. *Int. J. Gynecol. Pathol.* **21**, 407–411 (2002).
50. Willer, C.J., Li, Y. & Abecasis, G.R. METAL: fast and efficient meta-analysis of genomewide association scans. *Bioinformatics* **26**, 2190–2191 (2010).
51. Udler, M.S., Tyrer, J. & Easton, D.F. Evaluating the power to discriminate between highly correlated SNPs in genetic association studies. *Genet. Epidemiol.* **34**, 463–468 (2010).
52. Reumers, J. *et al.* Joint annotation of coding and non-coding single nucleotide polymorphisms and mutations in the SNPeffect and PupaSuite databases. *Nucleic Acids Res.* **36**, D825–D829 (2008).
53. Conde, L. *et al.* PupaSuite: finding functional single nucleotide polymorphisms for large-scale genotyping purposes. *Nucleic Acids Res.* **34**, W621–W625 (2006).
54. Li, N.F. *et al.* A modified medium that significantly improves the growth of human normal ovarian surface epithelial (OSE) cells *in vitro*. *Lab. Invest.* **84**, 923–931 (2004).
55. Fotheringham, S., Levanon, K. & Drapkin, R. *Ex vivo* culture of primary human fallopian tube epithelial cells. *J. Vis. Exp.* **51**, 2728 (2011).
56. Li, Q. *et al.* Expressopn QTL–based analyses reveal candidate causal genes and loci across five tumor types. *Hum. Mol. Genet.* **23**, 5294–5302 (2014).
57. Cancer Genome Atlas Research Network. Integrated genomic analyses of ovarian carcinoma. *Nature* **474**, 609–615 (2011).
58. Forbes, S.A. *et al.* The Catalogue of Somatic Mutations in Cancer (COSMIC). *Curr. Protoc. Hum. Genet.* Chapter 10, Unit 10.11 (2008).
59. Gao, J. *et al.* Integrative analysis of complex cancer genomics and clinical profiles using the cBioPortal. *Sci. Signal.* **6**, pl1 (2013).
60. Cerami, E. *et al.* The cBio cancer genomics portal: an open platform for exploring multidimensional cancer genomics data. *Cancer Discov.* **2**, 401–404 (2012).
61. Lawrenson, K. *et al.* Senescent fibroblasts promote neoplastic transformation of partially transformed ovarian epithelial cells in a three-dimensional model of early stage ovarian cancer. *Neoplasia* **12**, 317–325 (2010).