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Analysis

Causal relationships between four types of lipids and breast cancer risk with potential mediators: evidence from Mendelian randomization study and bioinformatics analysis

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Abstract

Background and purpose Breast cancer (BC) is the primary cause of cancer-related deaths among women worldwide, with increasing evidence pointing to the effect of metabolic factors, particularly lipid levels, in its pathogenesis. In this research, Mendelian randomization (MR) was employed to explore the causality between four plasma lipid traits—total cholesterol (TC), triglycerides (TG), high-density lipoprotein cholesterol (HDL-C) and low-density lipoprotein cholesterol (LDL-C)—and the risk of BC. Additionally, we explored the potential mediating effects of coronary artery disease (CAD), total testosterone (TT) on these associations and possible mechanisms through bioinformatics analyses.

Methods Data of genome-wide association study (GWAS) on lipids, CAD, TT and BC were obtained from public sources and websites as part of a genome-wide association research. The inference of causality was primarily assessed through the inverse variance weighting (IVW) approach, with supplementary tests for horizontal pleiotropy and heterogeneity. To verify the directionality of causal relationships, the MR Steiger test was applied. Additionally, reverse causality was evaluated by regarding BC as the exposure. To adjust for confounders, multivariate MR (MVMR) was performed, followed by a two-step mediation analysis to investigate the mediating roles of CAD in the lipid-BC association, and of TT in the CAD-BC relationship. The intersecting SNP (rs11556924) between causal pathways was established through a Venn diagram and its associated gene (Zinc Finger C3HC-Type Containing 1, *ZC3HC1*) was identified through the g:Profiler database. The expression of *ZC3HC1* was further explored using the TIMER, GEPIA2 and HPA database. Finally, enrichment analyses of Gene Ontology (GO), Kyoto Encyclopedia of Genes and Genomes (KEGG) and protein–protein interactions (PPI) network analysis were conducted on *ZC3HC1* and its related genes.

Results The random-effects IVW analysis yielded the following results: HDL-C on CAD (OR = 0.843, 95% CI 0.771–0.921, $P < 0.001$), CAD on BC (OR = 0.935, 95% CI 0.892–0.980, $P = 0.005$), HDL-C on BC (OR = 1.127, 95% CI 1.059–1.199, $P < 0.001$), CAD on TT (OR = 0.987, 95% CI 0.975–0.998, $P = 0.020$) and TT on BC (OR = 1.354, 95% CI 1.148–1.598, $P < 0.001$). The MR Steiger test results support the validity of the inferred causal direction ($P < 0.001$). There were no discernible causal relationships between BC and HDL-C/CAD according to reverse MR analysis ($P > 0.05$). Following MVMR adjustment, the causal effects of HDL-C, CAD, and TT on BC were still statistically significant ($P < 0.05$). Besides, the two-step mediation analysis

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indicated that CAD mediated 7.8% of the causal effect of HDL-C on BC, whereas TT mediated 6.1% of the causal effect between CAD and BC. The expression of *ZC3HC1* showed no significant expression difference between normal and BC tissues ($P > 0.05$), which might indicate a carcinogenic effect independent of expression levels but driven by functional alterations induced by variants (C > T). Functional network analysis suggested that *ZC3HC1* was associated with multiple signal pathways in cancers, such as PI3K-Akt and MAPK signal pathways.

Conclusions From a genetic perspective, our study reveals that there is causality between HDL-C levels and BC risk, with CAD and TT acting as partial mediators in this relationship. Moreover, our study firstly establishes a potential link between CAD-associated SNP (rs11556924), the corresponding gene (*ZC3HC1*) functional dysregulation, and the initiation of BC. These findings shed light on the biological links between lipids and BC, potentially contributing to future prevention and treatment strategies.

Keywords Breast cancer · Lipids · High-density lipoprotein cholesterol · Total testosterone · Coronary artery disease · Mendelian randomization · *ZC3HC1*

1 Introduction

Breast cancer (BC) is the most prevalent malignant tumor among women globally and remains a major contributor to cancer-associated deaths in female populations [1, 2]. Projections from the GLOBOCAN Cancer Tomorrow tool estimate that the incidence of BC will rise by over 54% by 2050. At the same time, despite advances in early diagnosis and therapy strategies, the prognosis of BC varies substantially, necessitating further study into novel biomarkers and possible therapeutic targets [3]. The etiology of BC is multifactorial, encompassing genetic predispositions, environmental exposures, and lifestyle influences [4, 5]. There is mounting evidence that some metabolic disorders (obesity, insulin resistance and dyslipidemia) are potential risk factors for BC because these conditions can further influence hormone levels and inflammatory pathways, contributing to BC's development and progression [6, 7]. Numerous studies have indicated a positive association between dyslipidemia and BC risk [8, 9]. However, most of the current studies on the association between dyslipidemia and BC incidence are observational studies, making it impossible to draw causal inference conclusions.

Cholesterol is essential for a variety of physiological processes, including the formation/function of cell membranes and cellular homeostasis regulation [10, 11]. Importantly, total cholesterol (TC) levels are not only correlated with the incidence of coronary artery disease (CAD) [11] but also with adiposity and diabetes, all of which have been suggested as potential risk factors for BC [12, 13]. However, the causal relationship between cholesterol BC susceptibility remains unclear, as studies have reported conflicting findings, including positive, inverse or absent associations between lipid levels and BC risk [9, 14, 15]. In addition, triglycerides (TG) have complicated functions in cardiovascular health, and their significance in CAD has been extensively investigated and discussed [16]. Concurrently, TG can also serve as an independent reservoir of fatty acid oxidation, which can further drive cell proliferation and cancer development [17], suggesting the possible carcinogenicity of TG. However, the effect of TG on BC remains unidentified, with some studies suggesting that high TG levels increase BC risk [18], whereas others have reported an inverse association [15]. Low-density lipoprotein cholesterol (LDL-C) is broadly acknowledged as a significant risk factor for CAD. Some studies also indicated that lower LDL-C levels might increase the occurrence of cancer [19]. However, in BC, the association appears to be the opposite [20]. Moreover, high-density lipoprotein cholesterol (HDL-C) has gained prominence as a protective factor for CAD [21]. Previous studies have also demonstrated HDL-C as a powerful univariate predictor of CAD [22]. Moreover, multiple investigations have suggested a negative correlation between HDL-C levels and tumor risk [23, 24]. But for BC, observational studies have yielded conflicting results. Some studies concluded that HDL-C were positively linked to the risk of BC [25, 26], whereas other studies found a negative association [9, 27] or no association [28]. These contradictory findings suggest that HDL-C may exert a regulatory function in BC development, but its actual direction and mechanisms have not yet been clarified. In summary, there seems to be a connection between the risk of BC and these four categories of plasma lipids. But the research findings are inconclusive or contradictory, and the distinct causal relationships between these plasma lipid levels and BC have not been fully elucidated.

For CAD, it is the leading cause of mortality in affluent nations, imposing a significant economic burden [29]. Several studies have identified an association between CAD and cancer prevalence [30, 31], including BC [32], which prompts that these two diseases may have shared risk factors and biological mechanisms. Nonetheless, the current research indicates an absence of randomized controlled trials (RCTs) exploring the connection between CAD and

BC and conducting such trials in the future presents significant ethical challenges. Thus, the probable causative link between CAD and BC has largely remained unexplored.

Mendelian Randomization (MR) is a genetics-based approach that employs variations in the genome as instrumental variables (IVs) to reveal the causality between exposures and outcomes. It offers the advantages of reducing confounding bias, avoiding reverse causality and enabling the evaluation of exposures'long-term effects [33]. Given the existing evidence indicating inconsistencies in the correlation between lipid levels, CAD and BC, as well as the uncertainty surrounding causality, we conducted two-sample MR (TSMR) analyses to clarify the relationship between genetically elevated levels of four lipid traits and BC, as well as the causal association between CAD and BC. Moreover, to better elucidate the mechanism through which CAD may influence the development of BC, we further selected total testosterone (TT) levels as a bridging variable to connect CAD and BC, owing to its distinct associations with both CAD [34] and BC [35, 36] based on current evidence. To adjust for confounding variables that could influence causal estimations [37], we also employed Multivariable Mendelian Randomization (MVMR). In addition, the first mediation analysis was performed to evaluate the possible mediating function of CAD in the HDL-C-BC causative link [38]. Subsequently, we conducted a further mediation analysis to clarify TT's function in the causal pathway between CAD and BC to offer a deeper insight into the underlying biological mechanisms. Finally, we aimed to identify key SNPs and associated genes implicated in causal relationships through comprehensive bioinformatics analyses. We also integrated findings both from our study and existing studies to elucidate potential mechanisms at both the gene expression level and from a functional perspective. Our exploration, from the perspective of genetics, may provide new perspectives on the processes that drive the incidence and progression of BC, as well as identify novel targets for metabolic therapeutic approaches.

2 Materials and methods

2.1 Study design and data sources

To evaluate the causative correlation between lipids and BC and determine if this relationship was mediated by CAD, different MR and mediation analyses were employed. Additionally, the effect of CAD on BC was further investigated by examining TT as a potential mediator. Briefly, we first explored the overall causal associations between four common lipid molecules (TC, TG, LDL-C, HDL-C) and BC through TSMR. Next, for positive associations, mediation effect analyses were conducted to quantify the mediating role of CAD and to assess its proportion in the overall causal relationship. Subsequently, we explored the function of TT (as a mediator) in the interaction between CAD and BC. The MVMR was employed to assess the mediating effect and proportion mediated by CAD and TT. The publicly accessible GWAS dataset served as the basis for this MR investigation as well, with all participants having submitted written informed consent. Concurrently, this study was conducted in keeping with the *Strengthening the Reporting of Observational Studies in Epidemiology using MR* (STROBE-MR) checklist (Additional file 1) [39]. The study design schematic and used MR analyses are also illustrated in Fig. 1.

Data for the two exposures, LDL-C (n = 70,814, European descent) and HDL-C (n = 77,409, European descent), were both extracted from the Family GWAS consortium data summarized in the IEU database, a comprehensive GWAS collecting resource (https://gwas.mrcieu.ac.uk/). Data on TC, TG and TT was originated from GWAS conducted by Barton AR et al. (n = 437,878, European ancestry) [40], Richardson TG et al. (n = 115,082, European ancestry) [41] and Ruth KS et al. (n = 425,097, European ancestry) [42]. Genetic data on CAD as a mediator were available from a comprehensive meta-analysis focusing on GWAS, which included 42,096 cases and 361 controls of European ancestry [43]. Moreover, genetic summary data on BC risk were acquired from the Breast Cancer Association Consortium (BCAC) [https://bcac.ccge.medschl.cam.ac.uk/ (accessed on 20 May 2024)] website, which was a multidisciplinary dynamic consortium formed in April 2005 and recruited over 100 groups to collect data from over 420,000 people. Sample overlap is minimal as data for exposures and outcomes come from completely different GWAS studies and databases. Additional introductions for diseases and lipids can be obtained from the IEU database, original studies [40–43] and the BCAC website. More detailed information for each GWAS summary data is shown in Table 1.

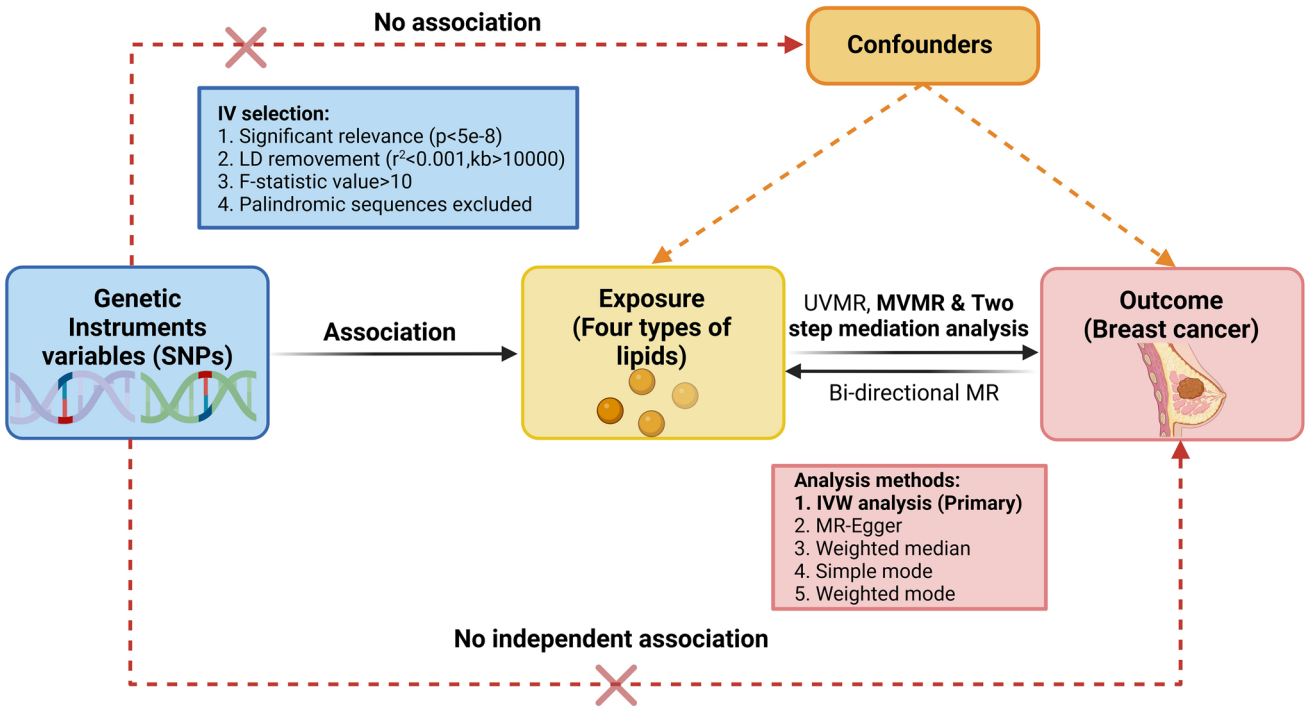


Fig. 1 The flowchart of this MR study including the basic assumptions. The purpose of the UVMR, MVMR and bidirectional MR analyses was to investigate the potential genetic correlations between lipids and BC risk. Mediation analysis was also employed to validate the effects of the mediating factors

Table 1 A detailed summary of Genome-Wide Association Studies (GWAS) included in the Mendelian randomization analysis

Exposures/ Outcomes	GWAS ID	Consortium	Ethnicity	Sample sizes	Number of SNPs	Year
TC	ebi-a-GCST90025953	NA	European	437,878	4,232,052	2021
TG	ebi-a-GCST90092992	NA	European	115,082	11,590,399	2022
LDL-C	ieu-b-4846	Family GWAS consortium	European	70,814	7,892,997	2022
HDL-C	ieu-b-4844	Family GWAS consortium	European	77,409	7,892,377	2022
TT	ebi-a-GCST90012114	NA	European	425,097	16,132,861	2020
CAD	ebi-a-GCST003116	NA	European	141,217	8,597,751	2015
BC	ieu-a-1129	BCAC	European	106,776	10,680,257	2017

TC: Total cholesterol levels; TG: Triglycerides levels; LDL-C: Low-density lipoprotein cholesterol; HDL-C: High-density lipoprotein cholesterol; TT: Total testosterone levels; CAD: Coronary artery disease; BC: Breast cancer

2.2 Selection of genetic IVs

We identified single nucleotide polymorphisms (SNPs) significantly related to exposure using a threshold of $P < 5 \times 10^{-8}$. To minimize bias from linkage disequilibrium (LD), a standard ($r^2 < 0.001$, kb > 10,000) was set to filter IVs and to ensure that they were distributed independently [44]. Palindromic sequences with moderate allele frequencies (0.42–0.58) were eliminated during the harmonization step. In addition, SNPs associated with the results (*P* value for BC risk $< 5 \times 10^{-8}$) were removed from the MR analysis to reduce potential confounding effects. The F-statistic value was calculated as well using the formula: $F = [(N-k-1)/k] \times [R^2/(1-R^2)]$ (N—the sample size in the GWAS analysis, k—the number of IVs, R^2—the proportion of exposure variance explained by these variables). And R^2 was calculated by $2 \times (1-MAF) \times MAF \times \beta^2$ (MAF—minor allele frequency, β—effect size of SNP on exposure). SNPs receiving an F value ≥ 10 were thought to be powerful IVs [45].

2.3 Mendelian randomization and evaluations of statistics

The "TwoSample MR" package and R (version 4.3.2) were primarily used to conduct the TSMR analysis. To elucidate the causation, we chose the random effects model within the inverse variance weighting (IVW) framework [46]. Also, MR-Egger [47], weighted median [48], simple mode, and weighted mode [49] were performed as additional supporting analytic approaches. The risks of BC were interpreted through the odds ratio (OR) and 95% confidence interval (CI). MVMR was also implemented using the "MendelianRandomization" and "TwoSample MR" packages. Additionally, the validity of the causative direction was evaluated by conducting the MR Steiger test [50]. $P < 0.05$ indicated that a significant causal link was identified. Besides, IVW method results are the main elements presented in this article and the results of other methods for the main causal pathways were included in the Additional file 2 (Tables S1-5).

2.4 Sensitivity analysis

Three approaches for sensitivity analyses were chosen: heterogeneity test, horizontal pleiotropy test and leave-one-out method. In addition, the IVs'heterogeneity was evaluated through the Cochran Q test. Significant heterogeneity was indicated by a *P* value of Q less than 0.05 and IVW with random effects would be performed to get more accurate estimations. Furthermore, horizontal pleiotropy—the genetic variants might influence multiple phenotypic traits independently—was evaluated by performing the MR-Egger intercept method. If $P > 0.05$ for the intercept, then the possibility of horizontal pleiotropy of IVs was considered negligible [51]. Finally, the leave-one-out strategy evaluates the overall impact of the remaining SNPs by sequentially removing each SNP and observing the changes in results.

2.5 Mediation analyses

The effect values for HDL-C on CAD (βA), CAD on BC (βB), HDL-C on BC (βC), CAD on TT (βD) and TT on BC (βE) were sequentially calculated using the TSMR analysis firstly to test whether the mechanism could be passed through. Then MVMR method was used to demonstrate the relationship between CAD/TT and BC by adjusting for HDL-C (βB') or CAD (βE'). The calculation of the mediating effect was performed as βA*βB'(CAD as mediator) and βD*βE'(TT as mediator). The ratio of the mediating effect to the overall effect was established through the formulas: $R = \beta A*\beta B'/\beta C*100\%$ and $\beta D*\beta E'/\beta B*100\%$. After controlling for confounding factors, the impact of exposure was regarded as a direct effect (βC – βA*βB', βB – βD*βE').

2.6 MR Data visualization

To illustrate the influences of sequentially excluded single SNP on the research results, the leave-one-out method was conducted. Scatter plots were graphical depictions of causal effect estimates, with the x-axis displaying the effect of SNPs associated with exposure and the y-axis indicating the effect of SNPs associated with outcome. Forest plots were utilized to evaluate the effect estimates of genetic variations. Furthermore, funnel plots were utilized to investigate the possibility of significant heterogeneity among individual genetic variants. R software (version 4.3.2) was used to generate all the visualization graphs.

2.7 Identification of the common SNP and related gene expression analysis

The intersecting SNPs in different causal relationships were analyzed using the "VennDiagram" R package. In addition, the most relevant coding gene (*ZC3HC1*) for the common SNP was identified from the g:SNPense module in the g:Profiler database (https://biit.cs.ut.ee/gprofiler/) [52]. The g:Profiler is a public database for characterizing and manipulating gene lists and is g:SNPense a tool that allows mapping of human SNPs to gene names, chromosomal locations, and variant consequence terms in the sequence ontology [52]. Subsequently, pan-cancer expression analyses of the relevant genes were performed via The Tumor IMmune Estimation Resource (TIMER) (https://cistrome.shinyapps.io/timer/) database [53]. The *ZC3HC1* mRNA and protein expression levels were analyzed through The Gene

Expression Profiling Interactive Analysis 2 (GEPIA2) (http://gepia2.cancer-pku.cn/#index) [54] and Human Protein Atlas (HPA) (https://www.proteinatlas.org/) [55] databases.

2.8 Analysis of ZC3HC1 co-expressed genes in BRCA

The co-expressed genes of *ZC3HC1* in BRCA were explored utilizing the LinkedOmics (http://www.linkedomics.org/) [56] database that contains multi-omics data from various cancers. The results of all co-expressed genes were analyzed using Spearman's correlation test and illustrated as heat maps. The "clusterProfiler" R package was employed to analyze and visualize the Gene Ontology (GO) categories, including cellular components (CC), biological processes (BP), and molecular functions (MF), as well as the Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways associated with the co-expressed genes.

2.9 Protein–protein interactions network analysis of ZC3HC1

STRING (https://cn.string-db.org/) is a comprehensive database that compiles both known and predicted interactions between proteins [57]. The Protein–Protein Interaction (PPI) network of *ZC3HC1* co-expressed proteins in BRCA was constructed and analyzed using the STRING database. The "clusterProfiler" R package was also applied to conduct GO term and KEGG pathway analyses.

3 Results

3.1 Selection of IVs

SNPs were evaluated for independence after LD was eliminated ($r^2 = 0.001$, kb = 10,000) and using the threshold ($P < 5 \times 10^{-8}$). After removing palindromic sequences with intermediate allele frequencies, the following numbers of IVs were identified to investigate the causal relationships between lipids, TT and BC: TC-CAD/BC: 185, 181; TG-CAD/BC: 62, 62; LDL-C-CAD/BC: 42, 42; HDL-C-CAD/BC: 67, 63; CAD-BC/TT: 37, 40; TT-BC: 164; BC-HDL-C: 51. In addition, following the computation, the strength of all IVs was deemed robust (F statistics > 10). Detailed IV data for relationships of HDL-C/CAD/BC and CAD/TT/BC are included in the Additional file 2 (Tables S6-10).

Exposure	Outcome	SNPs(n)	IVW method results	OR (95%CI)	P value
TC	CAD	185		1.554 (1.389-1.738)	<0.001***
TC	BC	181		1.035 (0.977-1.096)	0.238
TG	CAD	62		1.125 (0.979-1.293)	0.098
TG	BC	62		0.922 (0.866-0.983)	0.012*
LDL-C	CAD	42		1.668 (1.429-1.946)	<0.001***
LDL-C	BC	42		1.027 (0.973-1.084)	0.331
HDL-C	CAD	67		0.843 (0.771-0.921)	<0.001***
HDL-C	BC	63		1.127 (1.059-1.199)	<0.001***
CAD	BC	37		0.935 (0.892-0.980)	0.005**
CAD	TT	40		0.987 (0.975-0.998)	0.020*
TT	BC	164		1.354 (1.148-1.598)	<0.001***
BC	HDL-C	51		1.000 (0.979-1.022)	0.968[a]
BC	CAD	59		0.964 (0.903-1.030)	0.279[a]

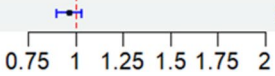


Fig. 2 Results utilizing the IVW method as the primary analytical method for two-sample MR analysis between lipids, CAD, TT and BC. TC, total cholesterol; TG, triglycerides; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; CAD, coronary artery disease; TT, total testosterone. *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$. [a]Significance for reverse Mendelian randomization between BC and HDL-C/CAD. $P > 0.05$ indicating the absence of reverse causal associations

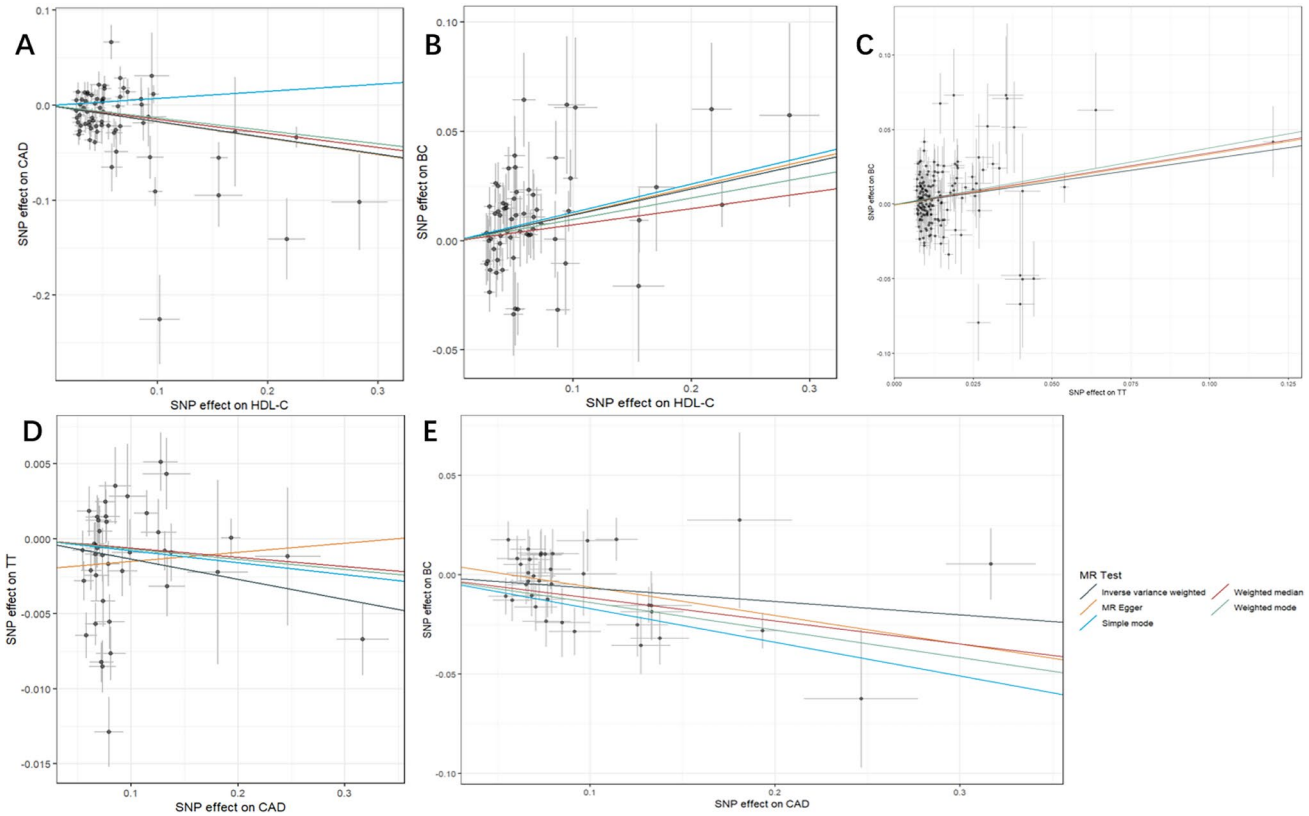


Fig. 3 Scatter plots illustrating the genetic correlation among HDL-C, CAD, TT and BC using various MR analysis methods. **A**, **B** HDL-C on CAD/BC. **C** TT on BC. **D**, **E** CAD on TT/BC

Table 2 Sensitivity analysis for two-sample MR results: heterogeneity and horizontal pleiotropy tests

Exposure	Outcome	Heterogeneity test (IVW)			Horizontal pleiotropy test (MR−Egger)		
		Cochrane's Q	Q_df	*P* value	Egger_Intercept	SE	*P* value
HDL-C	CAD	223.848	66	< 0.001	< 0.001	0.005	0.985
HDL-C	BC	113.876	62	< 0.001	< − 0.001	0.004	0.904
CAD	BC	54.131	36	0.027	0.008	0.005	0.133
CAD	TT	183.208	39	< 0.001	− 0.002	0.001	0.110
TT	BC	308.828	163	< 0.001	− 0.001	0.002	0.783
BC	HDL-C	92.063	50	< 0.001	< 0.001	0.002	0.797
BC	CAD	278.416	58	< 0.001	− 0.010	0.007	0.180

3.2 Two-sample MR analysis between lipids, CAD, TT and BC

The MR results obtained using the IVW approach are shown in Fig. 2. The results mainly revealed the negative causal effect of HDL-C on CAD (OR = 0.843, 95% CI [0.771, 0.921], $P < 0.001$); CAD on BC (OR = 0.935, 95% CI [0.892, 0.980], $P = 0.005$); CAD on TT (OR = 0.987, 95% CI [0.975, 0.998], $P = 0.02$). Also, significant positive causal effect of HDL-C on BC (OR = 1.127, 95% CI [1.059, 1.199], $P < 0.001$); TT on BC (OR = 1.354, 95% CI [1.148, 1.598], $P < 0.001$) and modest inverse causality between TG and BC (OR = 0.922, 95% CI [0.866, 0.983], $P = 0.012$) were observed. Moreover, scatter plots demonstrate that genetically driven elevation of HDL-C is related to a higher risk of developing BC and a lower risk of developing CAD (Fig. 3A, B). An elevated level of TT is associated with an increased BC risk (Fig. 3C). However, as the incidence of CAD increases, both the TT levels and the risk of BC decrease (Fig. 3D, E).

The results of the heterogeneity and horizontal pleiotropy tests are presented in Table 2. Heterogeneity was observed to varying degrees in all MR analysis results (Cochrane's Q: 54.131–308.828, *P*: 0.027- < 0.001). Thus, IVW

Table 3 Multivariate MR results of the relationship between HDL-C/CAD/TT and BC

Exposure	Outcome	SNPs (n)	MVMR-IVW		
			Beta (95%CI)	OR (95%CI)	P value
CAD as a mediating factor (HDL-C-CAD-BC)					
HDL-C	BC	58	0.119 (0.058, 0.180)	1.126 (1.059, 1.198)	< 0.001
CAD	BC	30	− 0.054 (− 0.106, − 0.003)	0.947 (0.899, 0.997)	0.039
TT as mediating factor (CAD-TT-BC)					
TT	BC	141	0.304 (0.136, 0.472)	1.355 (1.146–1.604)	< 0.001
CAD	BC	24	− 0.070 (− 0.126, − 0.015)	0.932 (0.882, 0.985)	0.013

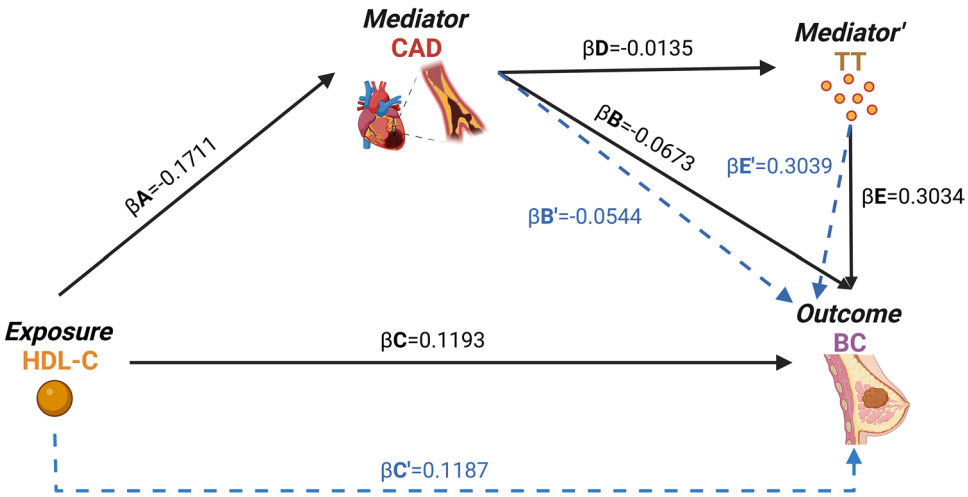


Fig. 4 Diagram of effect values for causal pathways in this study. Univariable MR results: β**A** effect of HDL-C on CAD; β**B** effect of CAD on BC; β**C** effect of HDL-C on BC; β**D** effect of CAD on TT. β**E** effect of TT on BC. Multivariable MR results: β B′, effect of CAD on BC adjusted HDL-C; β**C**′, effect of HDL-C on BC adjusted CAD; β**E**′, effect of TT on BC adjusted CAD

with random effects analysis for all results were performed. In addition, our analysis did not demonstrate significant horizontal pleiotropy, indicating that the IVs were unlikely to substantially influence the outcomes through pathways unrelated to the exposures (Table 2). Forest and funnel plots, which are consistent with the heterogeneity test results, are included in Additional file 3 (Figs. S1–10). Furthermore, the robustness of the MR analysis results was confirmed by the leave-one-out approach, which revealed a minor change in the error line (Additional file 3, Figs. S11-15). Besides, as evidenced by the MR-Steiger directionality test findings, our assessment of causal direction was accurate (both *P* for HDL-C/CAD as exposure, BC as outcome: < 0.001).

3.3 Reverse two-sample MR analysis between BC, HDL-C and CAD

In the TSMR analysis between BC and HDL-C/CAD, a total of 51 and 59 SNPs were identified respectively (Fig. 2). The IVW results demonstrated no causal relationships between BC and HDL-C ($P = 0.968$) or between BC and CAD ($P = 0.279$). Similarly, owing to the studies' heterogeneity, we performed a random effects model for IVW methods and did not observe significant horizontal pleiotropy in these studies as well (Table 2).

3.4 Multivariate MR analysis and mediation analysis

After adjusting for CAD, HDL-C still had a substantial causative influence on BC (OR = 1.126, 95% CI [1.059, 1.198], $P < 0.001$), as indicated in Table 3. Similarly, after adjusting for HDL-C levels, CAD was still significantly inversely associated with BC risk (OR = 0.947, 95% CI [0.899, 0.997], $P = 0.039$). When adjusting for each other's presence, TT levels and CAD still had substantial causal influences on the risk of BC respectively. The aforementioned results suggested that HDL-C levels, CAD, and TT levels might collectively contribute to the initiation and progression of BC.

Mediation studies were conducted to investigate if CAD mediated the causal effect of HDL-C on BC and whether the effect of CAD on BC was mediated by TT (Fig. 4). The results indicated that with HDL-C as the exposure and CAD as the mediator, the total effect was 0.1193 (Beta C) and the mediation effect was 0.0093 (Beta A * Beta B′, 95% CI: 0.0004–0.0210), accounting for 7.8% of the total effect. Furthermore, when the exposure was CAD and the mediator was TT, the total

effect was −0.0673 (Beta B) and the mediation effect was − 0.0041 (Beta D * Beta E', 95% CI: 0.0001–0.0023). The percentage of mediating effect was 6.1%. In addition, the direct effect was 0.11 and − 0.0632, respectively. The results above indicate that CAD might mediate the causal effect of HDL-C on BC, and TT could act as a mediator between CAD and BC.

3.5 Common SNP in the causal pathways and related gene expression

Next, after performing intersection analyses among two causal pathways (HDL-CAD-BC, CAD-TT-BC), one overlapping SNP (rs11556924, effect allele = T) was identified in the causal relationships between CAD and BC (Fig. 5A). Then we identified the most relevant coding gene—*ZC3HC1*, for this SNP through the g:SNPense online tool in the g:Profiler database. Compared to normal controls, the expression of *ZC3HC1* was significantly upregulated (all $P < 0.05$) in several cancer types including bladder, bile duct, colon, esophageal cancer, glioblastoma and other malignancies, as shown in Fig. 5B. However, there was no significant difference in the expression of *ZC3HC1* in Breast invasive carcinoma (BRCA) and its subtypes compared to normal tissue (Fig. 5B). Meanwhile, the *ZC3HC1* mRNA expression of BRCA in the GEPIA database was used as evidence for validation (Fig. 5C). Additionally, immunohistochemistry (IHC) results from the HPA database did not demonstrate notable differences in *ZC3HC1* protein expression (Fig. 5D). This implies that it is the functional alterations in the protein encoded by the gene, rather than its expression level, that may be more pivotal in the causal relationship between CAD and BC.

3.6 Functional enrichment of ZC3HC1 co-expressed genes and PPI network analysis of ZC3HC1 co-expressed proteins

To further elucidate the association network of this gene, the co-expression profiling of *ZC3HC1*-associated genes was performed on BRCA mRNA sequencing data utilizing the functional module of Linkedomics. The heat map displays the top 50 genes that are positively (Fig. 6A) or negatively (Fig. 6B) correlated with *ZC3HC1* respectively. GO analysis of *ZC3HC1*co-expressed genes indicates that they were mainly implicated in small GTPase mediated cellular signal transduction (Fig. 6C). The CC comprised neuronal cell body, cell-substrate junction and basal plasma membrane and the primary MF is characterized by activities including DNA-binding transcription activator activity and transcription factor binding (Fig. 6C). KEGG pathway analysis identified significant enrichment in the PI3 K-Akt and MAPK signaling pathways, with additional notable pathways including the breast cancer signaling pathway (Fig. 6D). In conclusion, the GO and KEGG analyses of co-expressed genes suggest that *ZC3HC1* plays a pivotal role in intracellular signal transduction pathways. And the correlation analysis of the top 10 co-expressed genes related to *ZC3HC1* was shown in Additional file 3 (Fig. S16).

The PPI network was established using STRING, encompassing 11 highly interconnected proteins (Fig. 6E). GO analysis indicated that these proteins are mainly localized in the Cul7-RING/SCF ubiquitin ligase complex (Additional file 3, Fig. S17 A) and are primarily involved in the SCF-dependent proteasomal ubiquitin-mediated protein catabolic process (Fig. S17B). Reactome and KEGG pathway enrichment analysis from STRING database revealed that these proteins participate in nuclear processes activated by ALK signaling in cancer, as well as in cell cycle regulation and cancer-associated pathways (Fig. S17 C, D).

4 Discussion

In this TSMR study, we demonstrated a causal association between genetically determined HDL-C levels and BC within the European population. MVMR and mediation analysis indicated that CAD contributes a mediating function (proportion of mediating effect: 7.8%) in the causal connection between HDL-C and BC. Further mediation study also emphasized the significant role of TT as a mediator (Proportion of the mediating effect: 6.1%) in the causal pathway between CAD and BC. Finally, bioinformatics analyses identified a putative key SNP (rs11556924) implicated in the causal association, along with its corresponding coding gene, *ZC3HC1*. Notably, no significant differences in *ZC3HC1* expression were observed between normal and cancerous tissues, suggesting that alterations in the function of its encoded protein caused by genetic variation may play an essential role in the causal correlation between CAD and BC.

Evidence from previous meta-analyses and observational research suggested that there was a negative relationship between HDL-C/TC levels and BC risk [9, 58], whereas TG/LDL-C levels were positively associated with BC [59, 60]. Yet, TC/HDL-C levels have been found to positively correlate with BC in certain studies [61], while TG/LDL-C levels were inversely correlated with BC risk [15, 58]. In addition, multiple studies together showed that none of these four lipids appeared

Fig. 5 Identification of the hub SNP in two causal pathways and the expression of associated coding gene (*ZC3HC1*) in BC. **A** The common SNP between causal pathways (A-B-C, SNPs associated with causal links of HDL-C on CAD, CAD on BC and HDL-C on BC; D-E**,** SNPs associated with causal links of CAD on TT and TT on BC). **B** A comprehensive pan-cancer analysis of the common SNP-associated gene (*ZC3HC1*) expression using the TIMER database. The expression data in BC are displayed within the area highlighted by the red frame. Besides, red and blue denote tumor and normal tissue samples, respectively. **C** *ZC3HC1* mRNA expression in BRCA tissues in the GEPIA2 database. **D** *ZC3HC1* protein expression in both normal breast tissue and tumor, as summarized in the HPA database. *ZC3HC1*, Zinc Finger C3HC-Type Containing 1, also named NIPA; BRCA: breast invasive carcinoma

to significantly correlate with BC [9, 59, 62]. In contrast to the aforementioned studies, our MR analyses efficiently clarify these contradictory results and indicate probable directions of effect, which is typically impossible to accomplish in other observational studies given the influences of reverse causation and the existence of confounding factors. Additionally, according to Nowak and colleagues' MR study, genetically elevated LDL-C levels might increase BC risk, while HDL-C and TG levels were not related to BC development [63], which contradicted the findings of this study. This may be explained by the fact that they used a rigorous pruning procedure to separate the influence of different lipid traits, which would diminish statistical power owing to the resilient genetic interrelation between these traits. The multivariate method employed in this research is a different approach for accessing exposure effects while adjusting for other relevant exposures. Moreover, MR studies from Johnson et al. and Beeghly-Fadiel et al. suggested that higher HDL-C levels were correlated to an increased risk of BC, which aligned with our primary finding [20, 64]. However, they also noted a positive relationship between LDL-C levels and BC risk, which might stem from the fact that they used genetic data for a much smaller defined range (Core LDL pathway genetic instrument). Besides, as also indicated in our analysis, their investigation discovered a previously unreported inverse connection between TG and BC (OR = 0.94, 95% CI [0.90, 0.98], $P = 2.60 \times 10^{-3}$), although this might be attributed to the correlation of TG with HDL-C rather than the independent effect of TG. Moreover, our findings broadly align with those of the recent MR research on lipids and BC by Beeghly-Fadiel et al. [64]. Multiple MR analyses were performed in both studies and a positive association of HDL-C with BC was observed as well as other lipid traits (TC/TG/LDL-C) did not appear to be related to the incidence of BC. However, our study goes a step further by analyzing the possible mediating role of CAD in the HDL-C-BC causal relationship, as well as the role of TT as a mediator between CAD and BC, providing deeper insights into the superficial causal relationships.

HDL, representing approximately 25–30% of the circulating lipoproteins, plays a crucial role in reverse cholesterol transport [65]. HDL-C is an important indicator of HDL. Many previous studies have discussed HDL-C as the protective factor for CAD, citing its antithrombotic, anti-inflammatory, reverse cholesterol transport and antioxidant properties as possible mechanisms [66, 67]. But recent growing explorations also suggested a complicated relationship between genetically determined HDL-C levels and BC risk [64, 68]. More importantly, the specific mechanisms through which HDL-C influences the development and progression of BC remain largely unelucidated. According to current research, HDL-C tends to promote the proliferation of BC cells [69, 70]. Furthermore, the association between elevated HDL-C levels and BC risks seems to be prompted by the heterogeneous populations of HDL-C particles that may have vital functions beyond their levels, such as the capabilities and status of different HDL particles [71]. It appears that HDL-C levels alone may not sufficiently explain the biological mechanisms underlying its causal association with BC. Instead, the status and function of HDL—the particle's functional core, likely play a more crucial role. For instance, oxidized HDL and diabetic HDL are more capable of facilitating the tumorigenic processes of distinct BC cells [69, 72]. More specifically, different HDL particles could stimulate BC cells to synthesize and release more matrix metalloproteinase (MMP)−2/9 and vascular endothelial growth factor-C (VEGF-C) [72]. Additionally, they likely orchestrated BC cell migration/invasion via the p38 mitogen-activated protein kinase (MAPK) pathway, as well as the extracellular signal-regulated kinase (ERK) and Akt pathways [72]. In addition, some HDL particles significantly stimulate the expression of cell surface integrins and protein kinase C (PKC) to augment metastatic potential and invasive capability [69]. Furthermore, studies have reported that the receptor for HDL, Scavenger receptor class B type I (SR-BI), might play a pivotal role by facilitating the selective uptake of HDL-cholesteryl ester (HDL-CE, the primary components of HDL-C) and activating oncogenic signaling pathways [73, 74]. To be specific, SR-BI interacts with PDZK1 to trigger PI3K, c-Src, and Erk1/2 signaling, promoting BC cell proliferation and migration [75–77]. Elevated SR-BI expression in BC cell lines [75, 78], tissues [78, 79] and mouse models [80] further underscores its oncogenic potential. Therefore, in light of evidence suggesting a positive correlation between HDL-C levels and the number of HDL particles [81], it is sensible to conclude that elevated HDL-C levels may indirectly impact the occurrence of BC through the aforementioned mechanisms. Besides, HDL-C particles can directly shift from anti-inflammatory to pro-inflammatory under oxidative stress and might promote cancer initiation and progression [82, 83]. Likewise, cholesterol and its oxysterol metabolites, whether circulating or within the tumor microenvironment, may directly influence the proliferation of mammary tissue, thereby promoting breast tumorigenesis [84, 85]. In conclusion,

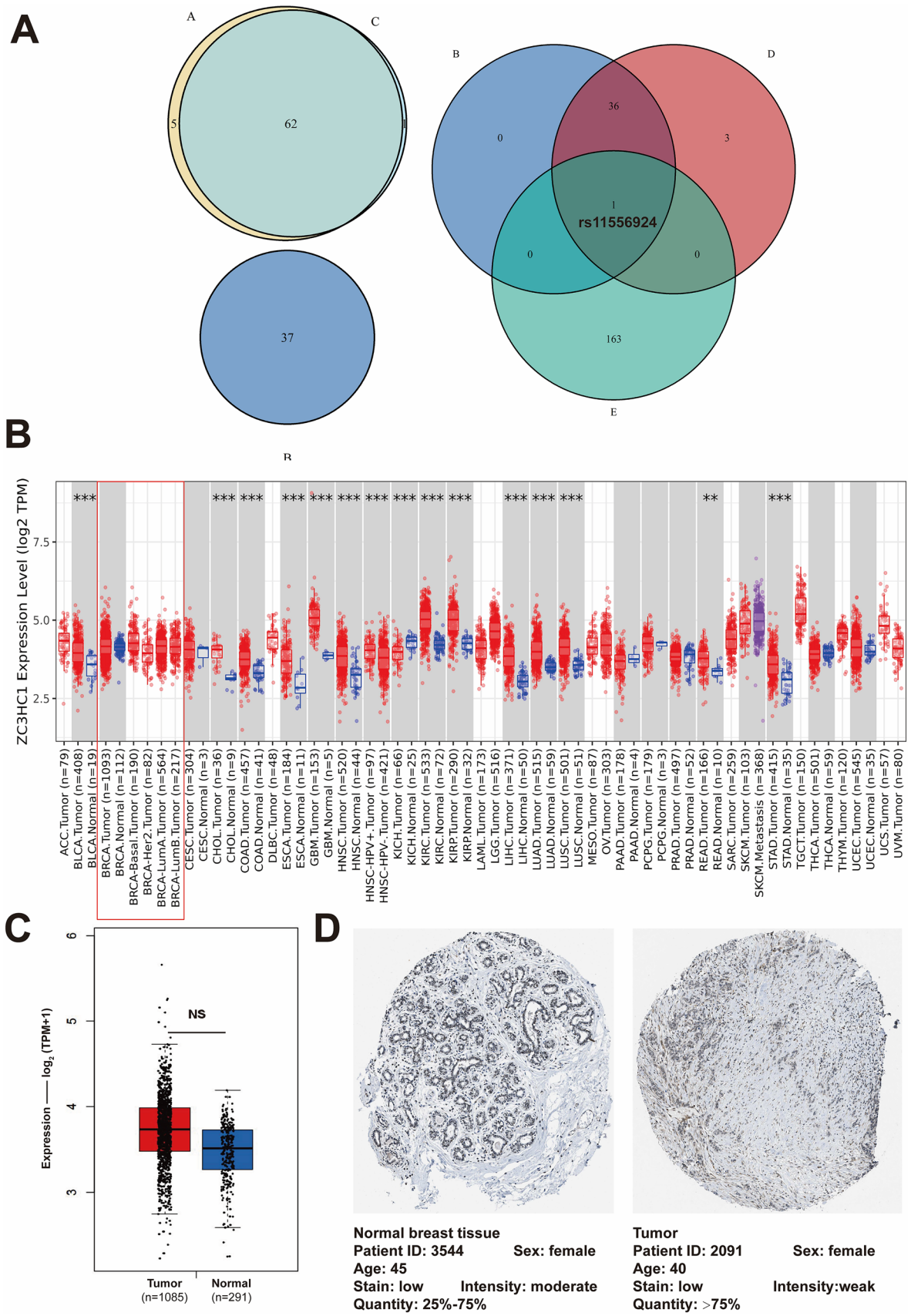

A
A
C
B
D
62
5
0
36
3
1
rs11556924
0
0
37
163
E
B
ZC3HC1 Expression Level (log2 TPM)
7.5
5.0
2.5
ACC.Tumor (n=79)
BLCA.Tumor (n=408)
BLCA.Normal (n=19)
BRCA.Tumor (n=1093)
BRCA.Normal (n=112)
BRCA-Basal.Tumor (n=190)
BRCA-Her2.Tumor (n=82)
BRCA-LumA.Tumor (n=564)
BRCA-LumB.Tumor (n=217)
CESC.Tumor (n=304)
CESC.Normal (n=3)
CHOL.Tumor (n=36)
CHOL.Normal (n=9)
COAD.Tumor (n=457)
COAD.Normal (n=41)
DLBC.Tumor (n=48)
ESCA.Tumor (n=184)
ESCA.Normal (n=11)
GBM.Tumor (n=153)
GBM.Normal (n=5)
HNSC.Tumor (n=520)
HNSC.Normal (n=44)
HNSC-HPV+.Tumor (n=97)
HNSC-HPV-.Tumor (n=421)
KICH.Tumor (n=66)
KICH.Normal (n=25)
KIRC.Tumor (n=533)
KIRC.Normal (n=72)
KIRP.Tumor (n=290)
KIRP.Normal (n=32)
LAML.Tumor (n=173)
LGG.Tumor (n=516)
LIHC.Tumor (n=371)
LIHC.Normal (n=50)
LUAD.Tumor (n=515)
LUAD.Normal (n=59)
LUSC.Tumor (n=501)
LUSC.Normal (n=51)
MESO.Tumor (n=87)
OV.Tumor (n=303)
PAAD.Tumor (n=178)
PAAD.Normal (n=4)
PCPG.Tumor (n=179)
PCPG.Normal (n=3)
PRAD.Tumor (n=497)
PRAD.Normal (n=52)
READ.Tumor (n=166)
READ.Normal (n=10)
SARC.Tumor (n=259)
SKCM.Tumor (n=103)
SKCM.Metastasis (n=368)
STAD.Tumor (n=415)
STAD.Normal (n=35)
TGCT.Tumor (n=150)
THCA.Tumor (n=501)
THCA.Normal (n=59)
THYM.Tumor (n=120)
UCEC.Tumor (n=545)
UCEC.Normal (n=35)
UCS.Tumor (n=57)
UVM.Tumor (n=80)
C
Expression —— log2 (TPM+1)
6
5
4
3
NS
Tumor
(n=1085)
Normal
(n=291)
D
Normal breast tissue
Patient ID: 3544
Sex: female
Age: 45
Stain: low
Intensity: moderate
Quantity: 25%-75%
Tumor
Patient ID: 2091
Sex: female
Age: 40
Stain: low
Intensity:weak
Quantity: >75%

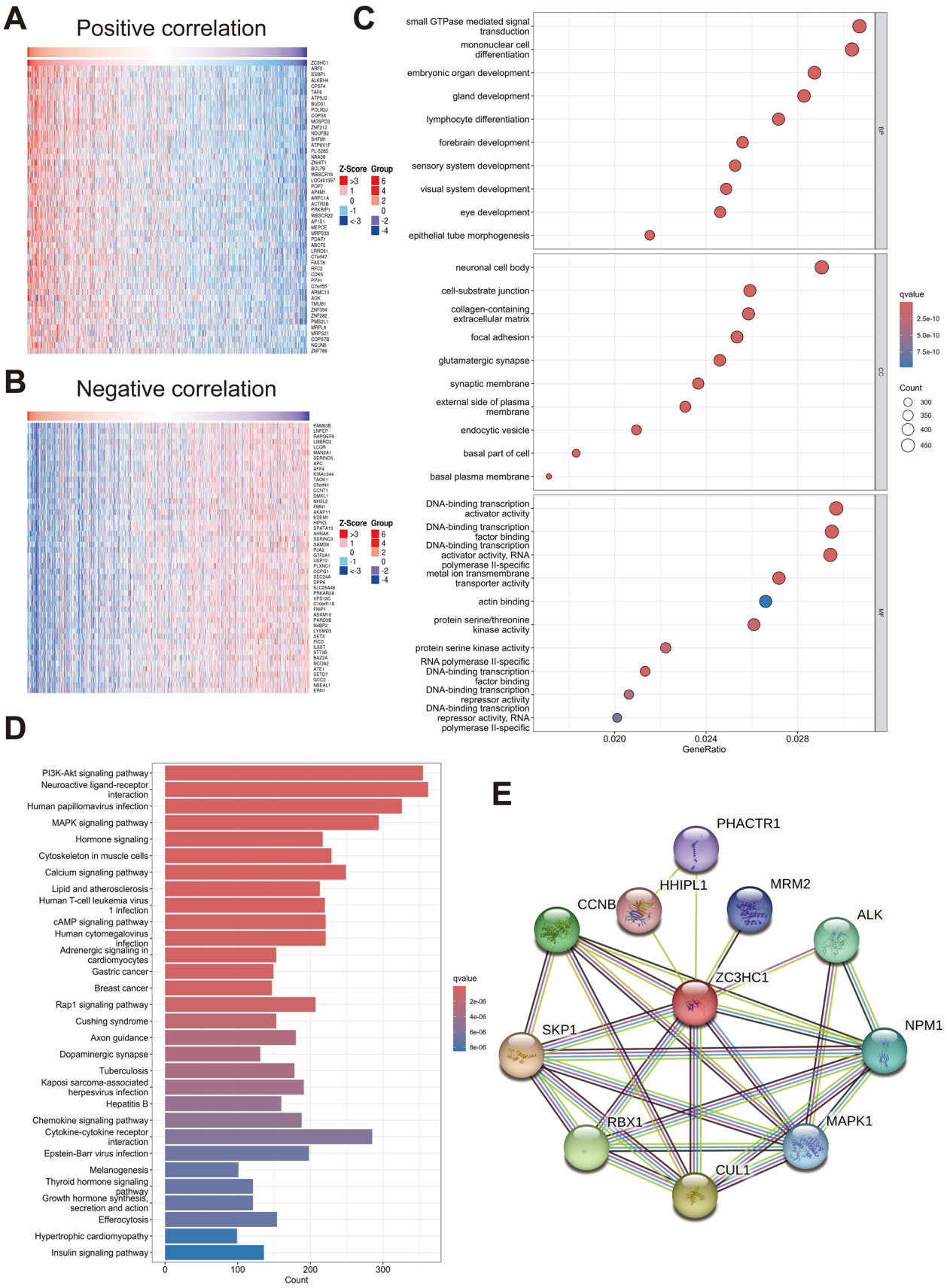
Fig. 6 Analysis of co-expressed genes of *ZC3HC1* in BRCA and *ZC3HC1* co-expressed proteins. **A**, **B** Heat maps depict the top 50 genes exhibiting positive (**A**) and negative (**B**) correlations with *ZC3HC1* in BRCA. Red: positive correlation, blue: negative correlation. **C** GO enrichment analysis identified the most significantly enriched biological processes, cellular components, and molecular functions of the co-expressed genes. **D** KEGG pathways enriched by co-expressed genes. **E** PPI network of *ZC3HC1* co-expressed proteins. The color of the lines represents the different interaction information

the aforementioned studies provide potential biological mechanisms linking elevated HDL-C levels with the higher risk of BC. But further investigations are necessary to comprehend the detailed mechanisms by exploring not only the change of HDL-C levels, but also the functionality of diverse key apolipoprotein components within HDL-C.

Although the causal association between CAD and BC and specific mechanisms behind the causality receive limited attention, our study explores the causal relationship between CAD and BC by using TSMR and determines that CAD mediates the causation between HDL-C and BC through MVMR and mediation analyses. The sensitivity analyses also confirm that our findings are consistent and robust. Notably, for better understanding the mechanism behind the effect of CAD on BC risk, we identified TT as a genetically plausible mediator in the causality through further mediation analysis as observational studies have demonstrated an inverse relationship between CAD and TT levels among female populations [86, 87], MR study (OR = 1.14, 95% CI [1.09, 1.21], $P = 4.07 \times 10^{-7}$) and another study (OR = 1.14, 95% CI [1.08, 1.20], $P = 1.60 \times 10^{-6}$) have indicated that high TT levels were linked to the risk of BC [35, 42]. It therefore seems reasonable to believe there is a possibility that TT mediates the causal association between CAD and BC on the basis of these findings. Moreover, their studies revealed that this relationship was statistically significant in the estrogen receptor-positive (ER+) BC subtype. The correlation with ER+ neoplasms involving testosterone might be interpreted by the following proposed mechanisms. The first possible answer is that high levels of testosterone are converted into more estradiol [88], which in turn binds to ER and activates the transcription of pro-growth genes while suppressing the expression of growth-inhibitory regulators, thus driving the proliferation of BC cells [89]. An alternative explanation is that the expression of the androgen receptor (AR) is positively correlated with ER expression in tumors, implying that ER expression could serve as a possible indicator of AR expression and potential interactions of androgen-estrogen signaling pathway in cancer development [90, 91]. This is further evidenced by the discovery that AR expression is present in only 20–30% of ER– BC cells [92]. In order to unravel the mechanism by which testosterone functions in BC, further detailed research focusing on BC subtypes stratified by the expression status of AR/ER and the specific effects of fluctuations in testosterone levels on key components of estrogen-related signaling pathways may be helpful.

ZC3HC1 encodes an F-box-containing protein that serves as a crucial component of an SCF-type E3 ubiquitin ligase complex, which orchestrates cell cycle progression by modulating the initiation of mitosis [93]. The G2/M transition necessitates the formation of a regulatory complex between Cyclin-B1 and cyclin-dependent kinase 1 (CDK1). Upon activation, the ubiquitin ligase complex facilitates the proteasomal degradation of cyclin-B1, thereby preventing premature entry into mitosis and ensuring proper cell cycle control [93]. The coding SNP rs11556924 (C > T) within the *ZC3HC1* gene results in an arginine-to-histidine polymorphism at amino acid residue 363 in the Nuclear Interaction Partner of ALK (NIPA) protein [93]. It has been reported that the Arg-363 allele (C) represents the predominant variant (allele frequency = 0.62) and is linked to a 9% increase in CAD risk per allele [94], while in our study the effect allele is T. Jones et al. further demonstrated that the His-363 (T) variant exhibits a 44% increase in phosphorylation at the Ser-395 site of NIPA induced by Cyclin-B1-CDK1 compared to the Arg-363 allele, which would cause higher accumulation of Cyclin-B1 in the nucleus of these cells, thereby facilitating the progression of mitosis [95]. Although the study did not provide enough evidence that the difference in cell proliferation phenotype between two genotypes was significant, this may be explained by the compensation of cells' inherent regulatory mechanisms under normal conditions, or the small difference in proliferation time (0.2% of the entire cell cycle) being too small to allow precise detection of changes in cell number within this timescale [95]. It is important to emphasize that, within the intricate genetic landscape of BC progression, even minor alterations in cell cycle regulation can exert a profound influence on the trajectory of the disease over months or years of development.

Notably, our study is distinguished by several advantages. First, to the best of our knowledge, this study is the first MR analysis that performs a two-step mediation analysis to explore in depth the possibility of a complete causal chain between HDL-C and BC, which may provide more potential targets for intervention in the incidence of BC. Secondly, the application of MR, utilizing IVs to rigorously assess the causal association, could reduce the bias induced by confounding factors and reduce the likelihood of reverse causality relative to observational studies. Furthermore, this study features an extensive sample size and powerful IVs for each lipid trait (all F-statistics > 10), and instrument validity was also carefully assessed. In addition, data on the exposures and outcomes analyzed were sourced from large, non-overlapping


A
Positive correlation
Z-Score
Group
B
Negative correlation
C
small GTPase mediated signal transduction
mononuclear cell differentiation
embryonic organ development
gland development
lymphocyte differentiation
forebrain development
sensory system development
visual system development
eye development
epithelial tube morphogenesis
BP
neuronal cell body
cell-substrate junction
collagen-containing extracellular matrix
focal adhesion
glutamatergic synapse
synaptic membrane
external side of plasma membrane
endocytic vesicle
basal part of cell
basal plasma membrane
CC
DNA-binding transcription activator activity
DNA-binding transcription factor binding
DNA-binding transcription activator activity, RNA polymerase II-specific
metal ion transmembrane transporter activity
actin binding
protein serine/threonine kinase activity
protein serine kinase activity
RNA polymerase II-specific DNA-binding transcription factor binding
DNA-binding transcription repressor activity
DNA-binding transcription repressor activity, RNA polymerase II-specific
MF
0.020
0.024
0.028
GeneRatio
qvalue
Count
D
PI3K-Akt signaling pathway
Neuroactive ligand-receptor interaction
Human papillomavirus infection
MAPK signaling pathway
Hormone signaling
Cytoskeleton in muscle cells
Calcium signaling pathway
Lipid and atherosclerosis
Human T-cell leukemia virus 1 infection
cAMP signaling pathway
Human cytomegalovirus infection
Adrenergic signaling in cardiomyocytes
Gastric cancer
Breast cancer
Rap1 signaling pathway
Cushing syndrome
Axon guidance
Dopaminergic synapse
Tuberculosis
Kaposi sarcoma-associated herpesvirus infection
Hepatitis B
Chemokine signaling pathway
Cytokine-cytokine receptor interaction
Epstein-Barr virus infection
Melanogenesis
Thyroid hormone signaling pathway
Growth hormone synthesis, secretion and action
Efferocytosis
Hypertrophic cardiomyopathy
Insulin signaling pathway
0
100
200
300
Count
qvalue
E
PHACTR1
HHIPL1
MRM2
CCNB
ALK
ZC3HC1
SKP1
NPM1
RBX1
MAPK1
CUL1

GWAS datasets, which made the results more robust. Besides, our results remained consistent even after adjusting for potential pleiotropic effects through the application of MR-Egger regression and multivariable weighted regression. Moreover, our study represents the first indication that CAD-associated SNP (rs11556924) may be implicated in the BC risk, thereby offering new perspectives on the mechanistic pathways of BC development and potential avenues for targeted therapeutic interventions.

The association with a heightened risk of BC is unexpected given that greater HDL-C levels are typically thought to be beneficial. Although we can discuss possible mechanisms in the literature that underpin this finding, the influence of the underlying factors remains largely unclarified. Therefore, the following directions should be the focus of future studies. First, the relationship between lipids and BC may vary based on the clinical statuses of the patient's condition, including the disease's stage or grade and the presence of any coexisting metabolic disorders. Secondly, the details and exact nature of the associations between BC subtypes and lipid levels, along with the specific mechanisms underlying this relationship, remain elusive and warrant further rigorous investigation. Furthermore, as indicated by the research, if the indicators of lipid-metabolite combinations are identified as risk factors for BC as reported [96], it may become imperative to thoroughly investigate the genetically determined interactions between the levels of lipids and various metabolites. Besides, the genetically determined function of HDL may merit as much attention as HDL-C levels. Also, given the diversity of genetic backgrounds associated with lipid levels and metabolism, investigating the causal relationships across different ethnicities is essential for cross-population comparisons and validation of our findings. Notably, the paradoxical roles of HDL-C in modulating the risk of CAD and BC presents significant research challenges. Mechanistically, HDL-C exerts cardioprotective effects primarily through reverse cholesterol transport, antioxidative activity, and anti-inflammatory properties. However, HDL-C may facilitate BC development by disrupting cholesterol homeostasis, enhancing tumor cell membrane fluidity, and activating oncogenic signaling pathways. Moreover, our findings indicate that CAD and TT levels may serve as mediators in this association, reinforcing the context-dependent functionality of HDL-C and underscoring the necessity for a nuanced understanding of its role across different pathological states. Therefore, we propose that future research should aim to define the optimal range of HDL-C levels and explore the appropriate interventions (e.g. pharmacological treatments and lifestyle modifications) to maintain HDL-C within this range, thereby achieving better control for both CAD and BC. Finally, while our study primarily focuses on the genetic determinants of lipid metabolism in BC etiology, we acknowledge the necessity of integrating non-genetic influences to achieve a more comprehensive understanding. Environmental and lifestyle factors, such as diet, physical activity or hormonal exposures, may modulate lipid metabolism and, in turn, influence BC susceptibility. More importantly, these factors likely interact with genetic variants, potentially amplifying or mitigating the initial effects on cancer risk. Future investigations incorporating intricate gene-environment interactions will be crucial for refining BC risk stratification and developing targeted prevention strategies.

5 Limitations

However, there are also several important caveats to consider in our analyses. First, MR must satisfy several assumptions to make accurate causal inferences [97]. Although statistical approaches were employed to identify and correct for breaches of these assumptions, it is necessary to use additional causal inference frameworks or different sorts of verification since these approaches cannot be guaranteed to adjust for all types of confounders. Second, MR is constrained in its ability to make inferences about trait information among the population where the GWAS originated (in this study is the European population). Therefore, given the potential differences in the genetic architecture of lipid profiles [98], the applicability of our conclusions to other populations may be restricted. Moreover, it is necessary to consider that our results could be attenuated due to the discovery of BC's correlation with the usage of lipid-lowering medications [99]. This raises the possibility that the real underlying causative factor may not be restricted to lipid levels themselves, but may include another trait for which lipids serve as a proxy. Finally, our two-step mediation analyses suggest that CAD mediates partial HDL-C effects on BC, whereas TT mediates a portion of CAD effects on BC, which is certainly an exciting finding. However, the design of the two-step mediation study may introduce the possibility of more undiscovered mediators that need to be explored in more aspects or sessions. Finally, our study investigates causality exclusively from a genetic perspective and does not extend to experimental validations (in vitro or in vivo functional assays) to corroborate the observed associations. Therefore, further studies are urgently needed to complete the puzzle of lipids-BC interactions and explore the unique function of the CAD-associated SNP (rs11556924) in BC development.

6 Conclusions

To conclude, this MR analysis provided genetic evidence that higher HDL-C levels are associated with increased BC risk, with CAD mediating this effect. Further mediation analysis indicated that TT mediated the causal effect of CAD on BC. Importantly, the findings provide further evidence against the broad application of HDL-C-raising therapies in the general population while highlighting their potential value in precision medicine for identifying women at higher risk of BC based on special SNP (rs11556924) and the function of its associated gene *ZC3HC1*, which may further contribute to the mechanistic understanding about the initiation and advancement of BC as well as the potential novel metabolic targets for therapeutic intervention.

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Author contributions X.C.L. led the study by securing funding, conducting formal analysis, and supervising the project. Additionally, X.C.L. was responsible for data curation, software development, and data visualization. B.Q.W. assisted in project administration, formal analysis, and manuscript revision. Y.T. contributed to the study's conceptualization, methodology, and supervision. The final manuscript was reviewed and approved by all authors. X.C.L. assumes responsibility as the guarantor of this work.

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Data availability The lipids datasets utilized in this article are publicly accessible (https://gwas.mrcieu.ac.uk/). Additionally, data on BC is completely available at https://bcac.ccge.medschl.cam.ac.uk/. The original contributions featured in this research are provided within the corresponding article and its Supplementary information. The corresponding author can be contacted for additional information or specific queries.

Declarations

Ethics approval and consent to participate This study involving human participants did not necessitate additional ethical review or approval, as it adhered to relevant local regulations and institutional protocols. Additionally, in line with national legislation and institutional policies, supplementary written informed permission for participation was not mandated.

Consent for publication Not applicable.

Competing interests The authors declare no competing interests.

References

1. Bray F, Laversanne M, Sung H, Ferlay J, Siegel RL, Soerjomataram I, Jemal A. Global cancer statistics 2022: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 2024;74(3):229–63.
2. Giaquinto AN, Sung H, Miller KD, Kramer JL, Newman LA, Minihan A, Jemal A, Siegel RL. Breast cancer statistics, 2022. CA Cancer J Clin. 2022;72(6):524–41.
3. Harbeck N, Penault-Llorca F, Cortes J, Gnant M, Houssami N, Poortmans P, Ruddy K, Tsang J, Cardoso F. Breast cancer. Nat Rev Dis Primers. 2019;5(1):66.
4. Terry MB, Colditz GA. Epidemiology and risk factors for breast cancer: 21st Century advances, gaps to address through interdisciplinary science. Cold Spring Harbor Perspect Med. 2023;13(9): a041317.
5. Islami F, Goding Sauer A, Miller KD, Siegel RL, Fedewa SA, Jacobs EJ, McCullough ML, Patel AV, Ma J, Soerjomataram I, et al. Proportion and number of cancer cases and deaths attributable to potentially modifiable risk factors in the United States. CA a Cancer J Clin. 2018;68(1):31–54.
6. Mendonça FM, de Sousa FR, Barbosa AL, Martins SC, Araújo RL, Soares R, Abreu C. Metabolic syndrome and risk of cancer: which link? Metab Clin Exp. 2015;64(2):182–9.

7. Esposito K, Chiodini P, Colao A, Lenzi A, Giugliano D. Metabolic syndrome and risk of cancer: a systematic review and meta-analysis. Diabetes Care. 2012;35(11):2402–11.
8. Peila R, Rohan TE. Circulating levels of biomarkers and risk of ductal carcinoma in situ of the breast in the UK Biobank study. Int J Cancer. 2024;154(7):1191–203.
9. Touvier M, Fassier P, His M, Norat T, Chan DS, Blacher J, Hercberg S, Galan P, Druesne-Pecollo N, Latino-Martel P. Cholesterol and breast cancer risk: a systematic review and meta-analysis of prospective studies. Br J Nutr. 2015;114(3):347–57.
10. Luo J, Yang H, Song BL. Mechanisms and regulation of cholesterol homeostasis. Nat Rev Mol Cell Biol. 2020;21(4):225–45.
11. Soliman GA. Dietary cholesterol and the lack of evidence in cardiovascular disease. Nutrients. 2018;10(6):780.
12. Rose DP, Gracheck PJ, Vona-Davis L. The interactions of obesity, inflammation and insulin resistance in breast cancer. Cancers. 2015;7(4):2147–68.
13. Sinha A, Bavishi A, Hibler EA, Yang EH, Parashar S, Okwuosa T, DeCara JM, Brown SA, Guha A, Sadler D, et al. Interconnected clinical and social risk factors in breast cancer and heart failure. Front Cardiovasc Med. 2022;9: 847975.
14. Kuzu OF, Noory MA, Robertson GP. The role of cholesterol in cancer. Can Res. 2016;76(8):2063–70.
15. Ni H, Liu H, Gao R. Serum lipids and breast cancer risk: a meta-analysis of prospective cohort studies. PLoS ONE. 2015;10(11): e0142669.
16. Asakura K, Minami Y, Nagata T, Katamine M, Muramatsu Y, Kinoshita D, Ako J. Higher triglyceride levels are associated with the higher prevalence of layered plaques in non-culprit coronary plaques. J Thromb Thrombolysis. 2024;57(1):58–66.
17. Saavedra-García P, Nichols K, Mahmud Z, Fan LY, Lam EW. Unravelling the role of fatty acid metabolism in cancer through the FOXO3-FOXM1 axis. Mol Cell Endocrinol. 2018;462(Pt B):82–92.
18. Esposito K, Chiodini P, Capuano A, Bellastella G, Maiorino MI, Rafaniello C, Giugliano D. Metabolic syndrome and postmenopausal breast cancer: systematic review and meta-analysis. Menopause. 2013;20(12):1301–9.
19. Chang Y-C, Lin C-J, Yeh T-L, Tsai M-C, Hsu L-Y, Chien K-L, Hsu H-Y. Lipid biomarkers and cancer risk - a population-based prospective cohort study in Taiwan. Lipids Health Dis. 2021;20(1):133.
20. Johnson KE, Siewert KM, Klarin D, Damrauer SM, Chang KM, Tsao PS, Assimes TL, Maxwell KN, Voight BF. The relationship between circulating lipids and breast cancer risk: a Mendelian randomization study. PLoS Med. 2020;17(9): e1003302.
21. Mahdy Ali K, Wonnerth A, Huber K, Wojta J. Cardiovascular disease risk reduction by raising HDL cholesterol–current therapies and future opportunities. Br J Pharmacol. 2012;167(6):1177–94.
22. Julve J, Escolà-Gil JC. High-density lipoproteins and cardiovascular disease: the good, the bad, and the future. Int J Mol Sci. 2021;22(14):7488.
23. Ko DT, Alter DA, Guo H, Koh M, Lau G, Austin PC, Booth GL, Hogg W, Jackevicius CA, Lee DS, et al. High-density lipoprotein cholesterol and cause-specific mortality in individuals without previous cardiovascular conditions: the CANHEART study. J Am Coll Cardiol. 2016;68(19):2073–83.
24. Ganjali S, Ricciuti B, Pirro M, Butler AE, Atkin SL, Banach M, Sahebkar A. High-density lipoprotein components and functionality in cancer: state-of-the-art. Trends Endocrinol Metab. 2019;30(1):12–24.
25. Zhou M, Henricks M, Loch V, Zhang G, Lu Y, Li X. Mendelian randomization analysis revealed potential metabolic causal factors for breast cancer. Sci Rep. 2023;13(1):14290.
26. Wang Y, Liu F, Sun L, Jia Y, Yang P, Guo D, Shi M, Wang A, Chen GC, Zhang Y, et al. Association between human blood metabolome and the risk of breast cancer. Breast Cancer Res. 2023;25(1):9.
27. Carter PR, Uppal H, Chandran S, Bainey KR, Potluri R. Algorithm for comorbidities A, length of stay, Unit MR: 3106Patients with a diagnosis of hyperlipidaemia have a reduced risk of developing breast cancer and lower mortality rates: a large retrospective longitudinal cohort study from the UK ACALM registry. Eur Heart J. 2017. https://doi.org/10.1093/eurheartj/ehx504.3106.
28. His M, Dartois L, Fagherazzi G, Boutten A, Dupré T, Mesrine S, Boutron-Ruault MC, Clavel-Chapelon F, Dossus L. Associations between serum lipids and breast cancer incidence and survival in the E3N prospective cohort study. Cancer Causes Control. 2017;28(1):77–88.
29. Duggan JP, Peters AS, Trachiotis GD, Antevil JL. Epidemiology of coronary artery disease. Surg Clin N Am. 2022;102(3):499–516.
30. Liu J, Chen S, Zhou Y, Huang H, Li Q, Liang Y, Dong S, Huang X, Chen L, Zheng X, et al. Proportion and number of incident cancer deaths in coronary artery disease. Cancer Med. 2023;12(19):20140–9.
31. Bertero E, Canepa M, Maack C, Ameri P. Linking heart failure to cancer: background evidence and research perspectives. Circulation. 2018;138(7):735–42.
32. Chen HH, Lo YC, Pan WS, Liu SJ, Yeh TL, Liu LY. Association between coronary artery disease and incident cancer risk: a systematic review and meta-analysis of cohort studies. PeerJ. 2023;11: e14922.
33. Sanderson E, Glymour MM, Holmes MV, Kang H, Morrison J, Munafò MR, Palmer T, Schooling CM, Wallace C, Zhao Q, et al. Mendelian randomization. Nat Rev Methods Primers. 2022;2(1):6.
34. Lopez DS, Taha S, Gutierrez S, Villasante-Tezanos A, Khalife WI, Alzweri L, Markides K, Baillargeon J, Tsilidis KK. Association of total and free testosterone with cardiovascular disease in a nationally representative sample of white, black, and Mexican American men. Int J Impot Res. 2024;36(4):385–93.
35. Nounu A, Kar SP, Relton CL, Richmond RC. Sex steroid hormones and risk of breast cancer: a two-sample Mendelian randomization study. Breast cancer research : BCR. 2022;24(1):66.
36. Amadou A, Freisling H, Sedlmeier AM, Bohmann P, Fontvieille E, Weber A, Konzok J, Stein MJ, Peruchet-Noray L, Jansana A, et al. Multitrait body shape phenotypes and breast cancer risk in postmenopausal women: a causal mediation analysis in the UK biobank cohort. J Epidemiol Glob Health. 2024;14(2):420–32.
37. Burgess S, Thompson SG. Multivariable Mendelian randomization: the use of pleiotropic genetic variants to estimate causal effects. Am J Epidemiol. 2015;181(4):251–60.
38. Carter AR, Sanderson E, Hammerton G, Richmond RC, Davey Smith G, Heron J, Taylor AE, Davies NM, Howe LD. Mendelian randomisation for mediation analysis: current methods and challenges for implementation. Eur J Epidemiol. 2021;36(5):465–78.
39. Skrivankova VW, Richmond RC, Woolf BAR, Davies NM, Swanson SA, VanderWeele TJ, Timpson NJ, Higgins JPT, Dimou N, Langenberg C, et al. Strengthening the reporting of observational studies in epidemiology using Mendelian randomisation (STROBE-MR): explanation and elaboration. BMJ. 2021;375: n2233.

40. Barton AR, Sherman MA, Mukamel RE, Loh PR. Whole-exome imputation within UK Biobank powers rare coding variant association and fine-mapping analyses. Nat Genet. 2021;53(8):1260–9.
41. Richardson TG, Leyden GM, Wang Q, Bell JA, Elsworth B, Davey Smith G, Holmes MV. Characterising metabolomic signatures of lipid-modifying therapies through drug target mendelian randomisation. PLoS Biol. 2022;20(2): e3001547.
42. Ruth KS, Day FR, Tyrrell J, Thompson DJ, Wood AR, Mahajan A, Beaumont RN, Wittemans L, Martin S, Busch AS, et al. Using human genetics to understand the disease impacts of testosterone in men and women. Nat Med. 2020;26(2):252–8.
43. Nikpay M, Goel A, Won HH, Hall LM, Willenborg C, Kanoni S, Saleheen D, Kyriakou T, Nelson CP, Hopewell JC, et al. A comprehensive 1,000 Genomes-based genome-wide association meta-analysis of coronary artery disease. Nat Genet. 2015;47(10):1121–30.
44. Sved JA, Hill WG. One hundred years of linkage disequilibrium. Genetics. 2018;209(3):629–36.
45. Pierce BL, Ahsan H, Vanderweele TJ. Power and instrument strength requirements for Mendelian randomization studies using multiple genetic variants. Int J Epidemiol. 2011;40(3):740–52.
46. Rees JMB, Wood AM, Dudbridge F, Burgess S. Robust methods in Mendelian randomization via penalization of heterogeneous causal estimates. PLoS ONE. 2019;14(9): e0222362.
47. Bowden J, Del Greco MF, Minelli C, Davey Smith G, Sheehan NA, Thompson JR. Assessing the suitability of summary data for two-sample Mendelian randomization analyses using MR-Egger regression: the role of the I2 statistic. Int J Epidemiol. 2016;45(6):1961–74.
48. Bowden J, Davey Smith G, Haycock PC, Burgess S. Consistent estimation in Mendelian randomization with some invalid instruments using a weighted median estimator. Genet Epidemiol. 2016;40(4):304–14.
49. Hartwig FP, Davey Smith G, Bowden J. Robust inference in summary data Mendelian randomization via the zero modal pleiotropy assumption. Int J Epidemiol. 2017;46(6):1985–98.
50. Hemani G, Tilling K, Davey Smith G. Orienting the causal relationship between imprecisely measured traits using GWAS summary data. PLoS Genet. 2017;13(11): e1007081.
51. Bowden J, Davey Smith G, Burgess S. Mendelian randomization with invalid instruments: effect estimation and bias detection through Egger regression. Int J Epidemiol. 2015;44(2):512–25.
52. Kolberg L, Raudvere U, Kuzmin I, Adler P, Vilo J, Peterson H. g:Profiler-interoperable web service for functional enrichment analysis and gene identifier mapping (2023 update). Nucleic Acids Res. 2023;51(W1):W207-w212.
53. Li T, Fan J, Wang B, Traugh N, Chen Q, Liu JS, Li B, Liu XS. TIMER: a web server for comprehensive analysis of tumor-infiltrating immune cells. Can Res. 2017;77(21):e108–10.
54. Tang Z, Kang B, Li C, Chen T, Zhang Z. GEPIA2: an enhanced web server for large-scale expression profiling and interactive analysis. Nucleic Acids Res. 2019;47(W1):W556–60.
55. Uhlén M, Fagerberg L, Hallström BM, Lindskog C, Oksvold P, Mardinoglu A, Sivertsson Å, Kampf C, Sjöstedt E, Asplund A, et al. Proteomics tissue-based map of the human proteome. Science. 2015;347(6220):1260419.
56. Vasaikar SV, Straub P, Wang J, Zhang B. LinkedOmics: analyzing multi-omics data within and across 32 cancer types. Nucleic Acids Res. 2018;46(D1):D956-d963.
57. Szklarczyk D, Kirsch R, Koutrouli M, Nastou K, Mehryary F, Hachilif R, Gable AL, Fang T, Doncheva NT, Pyysalo S, et al. The STRING database in 2023: protein-protein association networks and functional enrichment analyses for any sequenced genome of interest. Nucleic Acids Res. 2023;51(D1):D638-d646.
58. Llanos AA, Makambi KH, Tucker CA, Wallington SF, Shields PG, Adams-Campbell LL. Cholesterol, lipoproteins, and breast cancer risk in African American women. Ethn Dis. 2012;22(3):281–7.
59. Narii N, Zha L, Komatsu M, Kitamura T, Sobue T, Ogawa T. Cholesterol and breast cancer risk: a cohort study using health insurance claims and health checkup databases. Breast Cancer Res Treat. 2023;199(2):315–22.
60. Lofterød T, Mortensen ES, Nalwoga H, Wilsgaard T, Frydenberg H, Risberg T, Eggen AE, McTiernan A, Aziz S, Wist EA, et al. Impact of pre-diagnostic triglycerides and HDL-cholesterol on breast cancer recurrence and survival by breast cancer subtypes. BMC Cancer. 2018;18(1):654.
61. Garcia-Estevez L, Moreno-Bueno G. Updating the role of obesity and cholesterol in breast cancer. Breast Cancer Res. 2019;21(1):35.
62. Zeng X, Jiang S, Ruan S, Zhu L, Lian H, Liu M, Guo Z, Guo J, Ye C, Cheng Y, et al. Cardiovascular risk factors and breast cancer incidence in a large middle-aged cohort study. BMC Cancer. 2022;22(1):534.
63. Nowak C, Ärnlöv J. A Mendelian randomization study of the effects of blood lipids on breast cancer risk. Nat Commun. 2018;9(1):3957.
64. Beeghly-Fadiel A, Khankari NK, Delahanty RJ, Shu XO, Lu Y, Schmidt MK, Bolla MK, Michailidou K, Wang Q, Dennis J, et al. A Mendelian randomization analysis of circulating lipid traits and breast cancer risk. Int J Epidemiol. 2020;49(4):1117–31.
65. Sirtori CR, Corsini A, Ruscica M. The role of high-density lipoprotein cholesterol in 2022. Curr Atheroscler Rep. 2022;24(5):365–77.
66. Barter PJ, Nicholls S, Rye KA, Anantharamaiah GM, Navab M, Fogelman AM. Antiinflammatory properties of HDL. Circ Res. 2004;95(8):764–72.
67. van der Stoep M, Korporaal SJ, Van Eck M. High-density lipoprotein as a modulator of platelet and coagulation responses. Cardiovasc Res. 2014;103(3):362–71.
68. Pedersen KM, Çolak Y, Bojesen SE, Nordestgaard BG. Low high-density lipoprotein and increased risk of several cancers: 2 population-based cohort studies including 116,728 individuals. J Hematol Oncol. 2020;13(1):129.
69. Pan B, Ren H, He Y, Lv X, Ma Y, Li J, Huang L, Yu B, Kong J, Niu C, et al. HDL of patients with type 2 diabetes mellitus elevates the capability of promoting breast cancer metastasis. Clin Cancer Res. 2012;18(5):1246–56.
70. Rotheneder M, Kostner GM. Effects of low- and high-density lipoproteins on the proliferation of human breast cancer cells in vitro: differences between hormone-dependent and hormone-independent cell lines. Int J Cancer. 1989;43(5):875–9.
71. Toth PP, Barter PJ, Rosenson RS, Boden WE, Chapman MJ, Cuchel M, D'Agostino RB Sr, Davidson MH, Davidson WS, Heinecke JW, et al. High-density lipoproteins: a consensus statement from the National Lipid Association. J Clin Lipidol. 2013;7(5):484–525.
72. Pan B, Ren H, Ma Y, Liu D, Yu B, Ji L, Pan L, Li J, Yang L, Lv X, et al. High-density lipoprotein of patients with type 2 diabetes mellitus elevates the capability of promoting migration and invasion of breast cancer cells. Int J Cancer. 2012;131(1):70–82.
73. Wang D, Huang J, Gui T, Yang Y, Feng T, Tzvetkov NT, Xu T, Gai Z, Zhou Y, Zhang J, et al. SR-BI as a target of natural products and its significance in cancer. Semin Cancer Biol. 2022;80:18–38.

74. Silver DL, Jiang XC, Arai T, Bruce C, Tall AR. Receptors and lipid transfer proteins in HDL metabolism. Ann New York Acad Sci. 2000;902:103–11.
75. Danilo C, Gutierrez-Pajares JL, Mainieri MA, Mercier I, Lisanti MP, Frank PG. Scavenger receptor class B type I regulates cellular cholesterol metabolism and cell signaling associated with breast cancer development. Breast Cancer Res. 2013;15(5):R87.
76. Kimura T, Tomura H, Mogi C, Kuwabara A, Damirin A, Ishizuka T, Sekiguchi A, Ishiwara M, Im DS, Sato K, et al. Role of scavenger receptor class B type I and sphingosine 1-phosphate receptors in high density lipoprotein-induced inhibition of adhesion molecule expression in endothelial cells. J Biol Chem. 2006;281(49):37457–67.
77. Zhu W, Saddar S, Seetharam D, Chambliss KL, Longoria C, Silver DL, Yuhanna IS, Shaul PW, Mineo C. The scavenger receptor class B type I adaptor protein PDZK1 maintains endothelial monolayer integrity. Circ Res. 2008;102(4):480–7.
78. Cao WM, Murao K, Imachi H, Yu X, Abe H, Yamauchi A, Niimi M, Miyauchi A, Wong NC, Ishida T. A mutant high-density lipoprotein receptor inhibits proliferation of human breast cancer cells. Can Res. 2004;64(4):1515–21.
79. Yuan B, Wu C, Wang X, Wang D, Liu H, Guo L, Li XA, Han J, Feng H. High scavenger receptor class B type I expression is related to tumor aggressiveness and poor prognosis in breast cancer. Tumour Biol. 2016;37(3):3581–8.
80. Llaverias G, Danilo C, Mercier I, Daumer K, Capozza F, Williams TM, Sotgia F, Lisanti MP, Frank PG. Role of cholesterol in the development and progression of breast cancer. Am J Pathol. 2011;178(1):402–12.
81. Mackey RH, Greenland P, Goff DC Jr, Lloyd-Jones D, Sibley CT, Mora S. High-density lipoprotein cholesterol and particle concentrations, carotid atherosclerosis, and coronary events: MESA (multi-ethnic study of atherosclerosis). J Am Coll Cardiol. 2012;60(6):508–16.
82. Cruz-Reyes N, Radisky DC. Inflammation, infiltration, and evasion—tumor promotion in the aging breast. Cancers. 2023;15(6):1836.
83. Mazzuferi G, Bacchetti T, Islam MO, Ferretti G. High density lipoproteins and oxidative stress in breast cancer. Lipids Health Dis. 2021;20(1):143.
84. Silvente-Poirot S, Poirot M. Cancer. Cholesterol and cancer, in the balance. Science. 2014;343(6178):1445–6.
85. Nelson ER. The significance of cholesterol and its metabolite, 27-hydroxycholesterol in breast cancer. Mol Cell Endocrinol. 2018;466:73–80.
86. Debing E, Peeters E, Duquet W, Poppe K, Velkeniers B, Van den Brande P. Endogenous sex hormone levels in postmenopausal women undergoing carotid artery endarterectomy. Eur J Endocrinol. 2007;156(6):687–93.
87. Rexrode KM, Manson JE, Lee IM, Ridker PM, Sluss PM, Cook NR, Buring JE. Sex hormone levels and risk of cardiovascular events in postmenopausal women. Circulation. 2003;108(14):1688–93.
88. Simpson ER. Aromatization of androgens in women: current concepts and findings. Fertil Steril. 2002;77(Suppl 4):S6-10.
89. Frasor J, Danes JM, Komm B, Chang KC, Lyttle CR, Katzenellenbogen BS. Profiling of estrogen up- and down-regulated gene expression in human breast cancer cells: insights into gene networks and pathways underlying estrogenic control of proliferation and cell phenotype. Endocrinology. 2003;144(10):4562–74.
90. Collins LC, Cole KS, Marotti JD, Hu R, Schnitt SJ, Tamimi RM. Androgen receptor expression in breast cancer in relation to molecular phenotype: results from the Nurses' Health Study. Modern Pathol. 2011;24(7):924–31.
91. Niemeier LA, Dabbs DJ, Beriwal S, Striebel JM, Bhargava R. Androgen receptor in breast cancer: expression in estrogen receptor-positive tumors and in estrogen receptor-negative tumors with apocrine differentiation. Modern Pathol. 2010;23(2):205–12.
92. Kensler KH, Regan MM, Heng YJ, Baker GM, Pyle ME, Schnitt SJ, Hazra A, Kammler R, Thürlimann B, Colleoni M, et al. Prognostic and predictive value of androgen receptor expression in postmenopausal women with estrogen receptor-positive breast cancer: results from the Breast International Group Trial 1–98. Breast Cancer Res. 2019;21(1):30.
93. Bassermann F, von Klitzing C, Münch S, Bai RY, Kawaguchi H, Morris SW, Peschel C, Duyster J. NIPA defines an SCF-type mammalian E3 ligase that regulates mitotic entry. Cell. 2005;122(1):45–57.
94. Schunkert H, König IR, Kathiresan S, Reilly MP, Assimes TL, Holm H, Preuss M, Stewart AF, Barbalic M, Gieger C, et al. Large-scale association analysis identifies 13 new susceptibility loci for coronary artery disease. Nat Genet. 2011;43(4):333–8.
95. Jones PD, Kaiser MA, Ghaderi Najafabadi M, McVey DG, Beveridge AJ, Schofield CL, Samani NJ, Webb TR. The coronary artery disease-associated coding variant in zinc finger C3HC-type containing 1 (ZC3HC1) affects cell cycle regulation. J Biol Chem. 2016;291(31):16318–27.
96. Zhang J, Yin B, Xi Y, Bai Y. Triglyceride-glucose index is a risk factor for breast cancer in China: a cross-sectional study. Lipids Health Dis. 2024;23(1):29.
97. O'Connor LJ, Price AL. Distinguishing genetic correlation from causation across 52 diseases and complex traits. Nat Genet. 2018;50(12):1728–34.
98. Major cholesterol study reveals benefits of examining diverse populations. Nature 2021.
99. Zhong S, Zhang X, Chen L, Ma T, Tang J, Zhao J. Statin use and mortality in cancer patients: systematic review and meta-analysis of observational studies. Cancer Treat Rev. 2015;41(6):554–67.