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# Exploring the Potential Mechanisms of Compound Banmao Capsule Against Ovarian Cancer Through Network Pharmacology and Bioinformatics

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**Abstract:** The objective of this study was to characterize the potential mechanisms underlying the suppressive effects of Compound Banmao Capsule (CBC) on ovarian cancer (OC) using network pharmacology combined with bioinformatics. CBC-associated targets and OC-related targets were retrieved from public databases, and overlapping targets were identified and analyzed using a PPI network. GO and KEGG enrichment analyses were performed to identify relevant biological functions and signal pathways. Gene expression and clinical associations were further investigated using the GEPIA database. Fourteen overlapping targets were identified: AKR1B1, GJA1, BCL2, ESR1, CCL2, COL3A1, MMP1, DPP4, TNF, IL1B, F3, PTGS2, IL6, and VCAM1. GO enrichment analysis indicated that these targets were associated with multiple biological processes, particularly inflammatory responses, immune regulation, and the regulation of nitric oxide biosynthesis and interleukin-8 production. KEGG enrichment analysis highlighted several cancer-related and immune-associated pathways, including the AGE-RAGE, IL-17, TNF, NF-κB, NOD-like receptor, and C-type lectin receptor signaling pathways. GEPIA analysis further revealed differences in the expression of several candidate targets between OC and normal ovarian tissues and associations between selected genes and clinical characteristics. Taken together, this study provides a systematic overview of the potential targets and signaling pathways associated with CBC in OC, offering bioinformatics-based insights for further investigation of its underlying mechanisms.

**Keywords:** Compound Banmao capsule; Ovarian cancer; Network pharmacology; Bioinformatics

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## 1. Introduction

Ovarian cancer (OC) remains a major challenge in women's health because of its aggressive biological

behavior, frequently advanced stage at diagnosis, and substantial risk of recurrence [1]. For the majority of patients, cytoreductive surgery in conjunction with platinum and taxane-based chemotherapy is the standard of care; nonetheless, the long-term clinical results are still unsatisfactory, especially for those with advanced-stage disease [2,3]. Acquired resistance to platinum-based agents further limits treatment efficacy and contributes to disease progression and relapse [3,4]. These challenges highlight the need to explore additional therapeutic strategies and identify the molecular mechanisms underlying ovarian cancer development and treatment response.

The use of traditional Chinese medicine (TCM) as a supplemental strategy for the treatment of cancer has grown in popularity [5]. Owing to the complex composition of herbal formulations, Chinese herbal medicine may exert biological effects through multiple compounds and interconnected molecular pathways [6,7]. However, this complexity also presents considerable challenges in elucidating the mechanisms responsible for their therapeutic effects [7]. Therefore, systematic approaches capable of integrating compound-target relationships with disease-associated molecular information may help clarify the potential pharmacological actions of Chinese herbal medicine formulations and provide a basis for further experimental investigation.

Compound Banmao Capsule (CBC), a TCM formulation, has been used in clinical practice for the management of various malignancies [8,9]. According to the pharmacopoeial formulation, CBC consists of 11 medicinal ingredients [10]: Mylabris (Banmao, BM), Ginseng *Radix et rhizoma* (Renshen, RS), *Acanthopanacis senticosi Radix et rhizoma seu caulis* (Ciwujia, CWJ), *Astragali radix* (Huangqi, HQ), *Curcumae rhizoma* (Ezhu, EZ), *Sparganii rhizoma* (Sanleng, SL), *Scutellariae barbatae* Herba (Banzhilian, BZL), Corni Fructus (Shanzhuyu, SZY), *Ligustri lucidi fructus* (Nvzhenzi, NZZ), Bear bile powder (Xiongdanfen, XDF), *Glycyrrhizae Radix et rhizoma* (Gancao, GC). Traditionally, CBC is associated with the actions of resolving blood stasis and detoxifying, and it has been investigated for its potential antitumor effects [10]. Previous studies have suggested that CBC may influence several cancer-associated biological processes, including apoptosis, protein phosphorylation, and immune regulation [8,10]. Nevertheless, the molecular basis of its potential activity against ovarian cancer remains insufficiently characterized, and the key targets and signaling pathways involved require further investigation.

Network pharmacology provides a systems-level framework for exploring the relationships among the chemical constituents of complex herbal formulations, their potential molecular targets, and disease-associated pathways [9,11,12]. When integrated with bioinformatics analysis, this approach can help identify candidate therapeutic targets and prioritize molecular mechanisms for subsequent validation [11-13]. In this study, network pharmacology and bioinformatics analyses were combined to investigate the potential mechanisms of CBC in ovarian cancer. Potential CBC-associated targets were integrated with OC-related targets to identify overlapping genes, followed by GO functional annotation and KEGG pathway enrichment analyses. Key candidate genes were subsequently evaluated using the GEPIA database to investigate their expression patterns and clinical relevance. This integrated analysis was designed to gain mechanistic insights into the potential effects of CBC on ovarian cancer and to generate testable hypotheses for future experimental studies.

## 2. Materials and methods

### 2.1. Prediction of related targets of CBC

TCMSP database (https://old.tcmsp-e.com/tcmsp.php) and the TCEM (http://www.nrc.ac.cn:9090/ETCM/) database were utilized to search for compounds in 11 TCMs of CBC. The compounds with OB ≥ 30% and

DL ≥ 0.18 were selected as candidate compounds for the current study. Then, we used the TCMSP database to obtain target data for these candidate compounds in CBC. The ingredients shared with OC targets were regarded as active components, and the gathered components with targets were thought to constitute the possible active components of CBC.

## 2.2. Target standardization

The target information of herbal ingredients retrieved from the TCMSP database is integrated from multiple data sources, leading to non-standardized nomenclature for collected target entries. Accordingly, UniProtKB (http://www.uniprot.org/), a core repository that compiles protein-related functional annotations, was adopted to retrieve standard official gene symbols. Target identifiers were imported into the database, and "Homo sapiens" was set as the organism for extracting corresponding official symbols.

## 2.3. OC data collection

We used "ovarian cancer" as the search term to screen ovarian-cancer-related datasets from two public repositories: the GEO (https://www.ncbi.nlm.nih.gov/geo/) and GeneCards https://www.genecards.org/). In GEO, the species was selected as Homo sapiens, the study type was "Expression profiling by array", and the appropriate dataset was selected. Serving as the discovery set, the GSE16708 dataset was generated on the GPL6947 platform. It contains ovarian tissues from 24 OV cells and 9 normal ovarian surface epithelium cells in GEO. Targets obtained by overlapping OC-related candidates from the GEO dataset and GeneCards were defined as OC tumor targets.

## 2.4. Acquisition of hub targets

Venn diagram was generated using the online tool (https://www.omicstudio.cn/tool/6) to screen overlapping targets between ovarian cancer-associated targets and CBC-related targets, which were defined as hub targets.

## 2.5. PPI network construction and hub targets screening

Hub targets were imported into the STRING 12.0 database (https://cn.string-db.org/), a repository compiling protein-protein association information, for the reconstruction of PPI networks. A threshold of minimum interaction score ≥ 0.4 was set as the filtering criterion for building the PPI network.

## 2.6. Enrichment analysis

To further characterize the biological roles of hub genes, we performed GO functional annotation and KEGG pathway enrichment for these core targets via DAVID (https://david.ncifcrf.gov/) and omicstudio (https://www.omicstudio.cn/), followed by result visualization. Enrichment terms with $P$-value < 0.05 were considered statistically significant.

## 2.7. Network construction

For clearer interpretation and visual presentation of the interrelationships linking diseases, drugs, genes, and biological processes, a regulatory network was established and visualized with Cytoscape software (v.3.10.1). Nodes within this network stand for candidate compounds, diseases, molecular targets, and KEGG pathways. The connections between nodes carry distinct biological implications, which are determined by the construction strategy of the network.

## 2.8. Overall Survival (OS) analysis, mRNA expression level analysis of hub targets

GEPIA (http://gepia.cancer-pku.cn/index.html) was used to verify OS of hub targets and mRNA expression levels in normal and OC human tissues.

# 3. Results

## 3.1. Construction of targets network map of CBC

A total of 178 candidate compounds were collected from the TCMSP and TCEM databases after removing duplicate entries, including 175 compounds from TCMSP and three additional compounds from TCEM. Based on the available compound-target information, 161 potential targets associated with these compounds were identified. Cytoscape software was subsequently used to construct a compound-target network illustrating the relationships between the candidate compounds and their associated targets (**Figure 1**). Among them, some compounds are found in a variety of Chinese herbal medicines (**Table 1**).

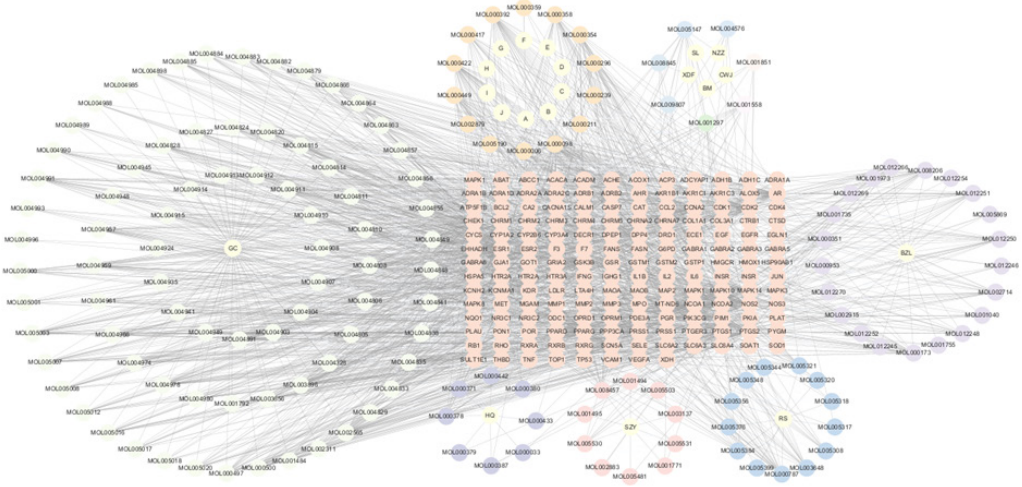


**Figure 1.** The interaction network between CBC bioactive constituents and molecular targets.

**Table 1.** Shared compounds among different TCM

| Number | Compound ID | Compound Name | Chinese herbal medicine |
|---|---|---|---|
| A | MOL000006 | Luteolin | BZL、NZZ |
| A | MOL005190 | Eriodictyol | BZL、NZZ |
| B | MOL000098 | Quercetin | BZL、GC |
| C | MOL000211 | Mairin | GC、HQ |
| C | MOL000239 | Jaranol | GC、HQ |
| C | MOL000417 | Calycosin | GC、HQ |
| C | MOL000354 | Isorhamnetin | GC、HQ |
| D | MOL000296 | Hederagenin | EZ、HQ |
| E | MOL000358 | Beta-sitosterol | SZY、BZL |
| F | MOL000359 | Sitosterol | SZY、BZL |
| G | MOL000392 | Formononetin | GC、HQ |
| H | MOL000422 | Kaempferol | GC、HQ |
| I | MOL000449 | Stigmasterol | SZY、BZL |
| J | MOL002879 | Diop | SZY、RS |

## 3.2. Screening of OC-related targets

3231 OC targets were obtained from the GeneCards database. In addition, GSE16708 was accessed from the GEO database. The microarray dataset GSE16708, derived from the GPL6947 platform, contains gene expression profiles associated with OC. The dataset included 24 OC cells and 9 normal ovarian surface epithelium cells. After the integration and de-duplication of relevant target genes, a total of 756 genes were screened out, including 454 upregulated and 302 downregulated genes. The distribution of these DEGs is presented in the volcano plot (**Figure 2A**). By integrating the GeneCards-derived targets with the DEGs identified from GSE16708, 296 overlapping OC-related targets were obtained (**Figure 2B**).

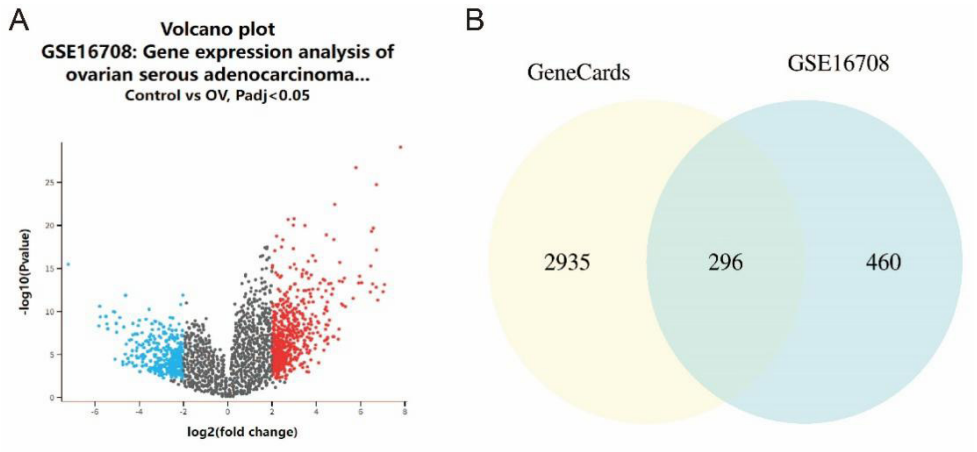



**Figure 2.** OC-related targets. (A) Volcano plot showing DEGs between OC cell samples and normal control samples from the GSE16708 dataset. (B) Venn diagram illustrating overlapping targets retrieved from GeneCards and the GSE16708 dataset.

## 3.3. Construction of PPI network

To screen core genes for regulatory network-based mechanistic exploration, overlapping entries between OC-related targets and drug-associated targets were determined using the OmicStudio cloud-based bioinformatics platform (https://www.omicstudio.cn/index). 14 hub targets were screened as candidate genes from the overlapping pool of OC targets and molecular targets of CBC active ingredients (**Figure 3A**). The STRING 12.0 database was applied to establish a PPI network comprising 14 nodes and 67 edges (**Figure 3B**), indicating that the identified targets exhibited extensive functional associations within the network. These hub targets were subsequently subjected to functional enrichment analysis to characterize their potential biological roles and associated signaling pathways.

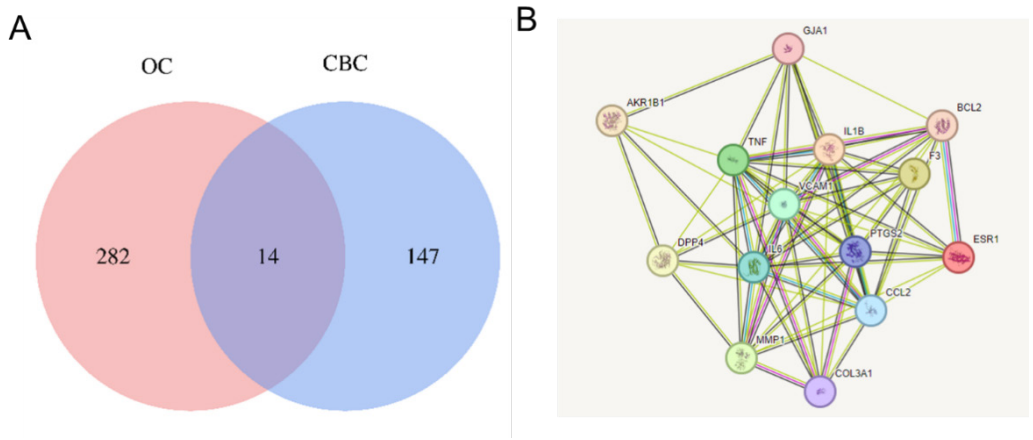



**Figure 3.** PPI network analysis of hub targets for CBC intervention against OC. (A) Venn diagram of targets appearing in OC and the targets of CBC active compounds. (B) The PPI network of hub targets.

## 3.4. GO and KEGG functional enrichment analysis

To uncover the underlying mechanisms by which CBC exerts therapeutic effects on OC, GO and KEGG functional enrichment analyses were carried out (**Figure 4**). Under the threshold of *P*-value < 0.05, the results demonstrated that 14 genes were significantly enriched across 115 biological processes (BPs), 11 cellular components (CCs), 7 molecular functions (MFs), and 43 KEGG pathways. Bioinformatic analysis revealed the main enriched BPs (top 10) of CBC intervening in OC (**Figure 4A**), including positive regulation of nitric oxide biosynthetic process, positive regulation of fever generation, inflammatory response, response to glucocorticoid, vascular endothelial growth factor production, humoral immune response, positive regulation of interleukin-8 production, positive regulation of peptidyl-serine phosphorylation, positive regulation of sequence-specific DNA binding transcription factor activity, and positive regulation of glial cell proliferation, which suggested that CBC against OC involves many biological processes. The enriched CCs were extracellular space, extracellular region, cell surface, macromolecular complex, membrane raft, endoplasmic reticulum lumen, endoplasmic reticulum, external side of plasma membrane, collagen trimer, and integral component of plasma membrane. The main enriched MFs included protease binding, integrin binding, serine-type endopeptidase activity, cytokine activity, receptor binding, beta-catenin binding, and protein homodimerization activity.

For KEGG enrichment analysis, the top 15 pathways with the highest statistical significance were screened (**Figure 4B**). CBC was mainly involved in the AGE-RAGE signaling pathway in diabetic complications, the IL-17 signaling pathway, the TNF signaling pathway, the NF-kappa B signaling pathway, the NOD-like receptor signaling pathway, and the C-type lectin receptor signaling pathway, indicating that CBC could treat OC by orchestrating multiple pathways. A preliminary network was generated to intuitively exhibit the regulatory interrelationships connecting CBC, candidate genes and KEGG pathways (Figure 4C). Among them, MOL000006, MOL000422, MOL000173 and MOL000098 link more than five hub genes as core components (**Table 2**), suggesting that these compounds may represent important candidates for further investigation.

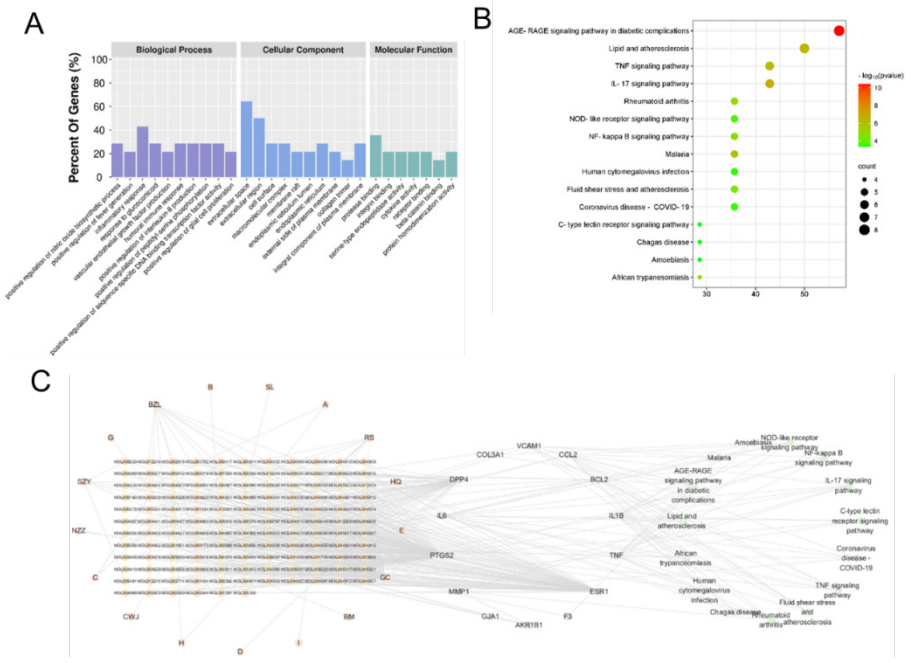



**Figure 4.** GO and KEGG enrichment analysis of CBC intervening OC hub targets.

A GO enrichment analysis (the top 10 terms of BP, CC, and MF enrichment analyses were shown, respectively). B KEGG enrichment analysis (top15 results). C Compound-target-pathway network.

**Table 2.** Core active components of CBC

| Mol | Core components | Degree | Genes |
|---|---|---|---|
| MOL000006 | luteolin | 5 | MMP1, TNF, IL6, DPP4, PTGS2 |
| MOL000422 | kaempferol | 6 | PTGS2, DPP4, BCL2, MMP1, TNF, VCAM1 |
| MOL000173 | wogonin | 8 | PTGS2, ESR1, DPP4, BCL2, MMP1, TNF, IL6, CCL2 |
| MOL000098 | quercetin | 13 | PTGS2, DPP4, AKR1B1, BCL2, MMP1, TNF, VCAM1, IL6, GJA1, F3, IL1B, CCL2, COL3A1 |

## 3.5. OS Analysis, mRNA expression level analysis and pathological stage of hub targets

Expression profiles and clinical correlations of the candidate hub genes were evaluated through GEPIA. Comparisons between OC and normal ovarian tissues revealed statistically significant differences in the expression of BCL2, CCL2, ESR1, F3, IL1B, TNF, and VCAM1 (**Figure 5**). Survival analysis further indicated that MPP1 expression was significantly associated with OS in patients with OC (**Figure 6**). In addition, analysis according to pathological stage revealed differences in the expression of BCL2, IL1B, TNF, and PTGS2 between stage II and stage III-IV OC samples (**Figure 7**). These results suggest that selected candidate targets are associated with gene-expression patterns or clinical characteristics in OC and may warrant further investigation. However, these database-derived associations do not establish that the genes are direct targets of CBC or mediate its potential anti-tumor effects.

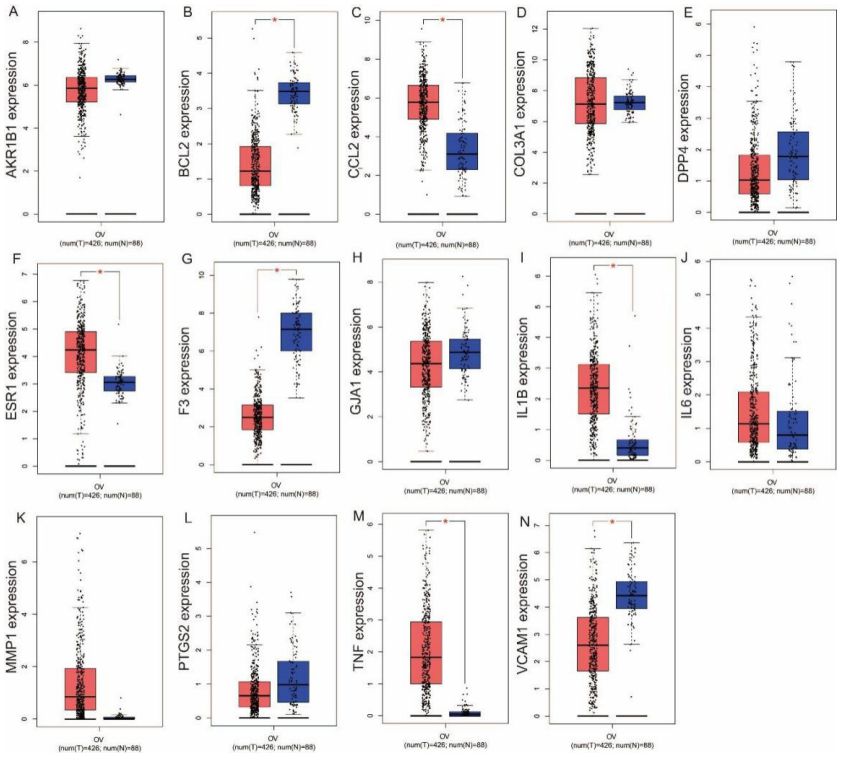



**Figure 5.** mRNA expression level in the GEPIA database.

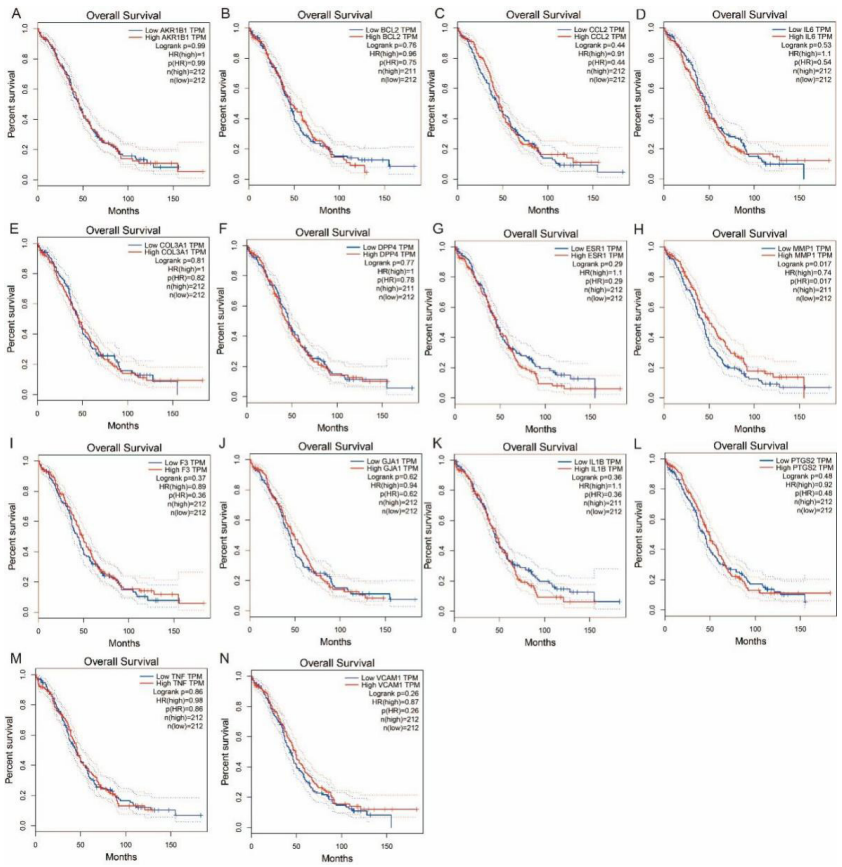



**Figure 6.** OS in the GEPIA database.

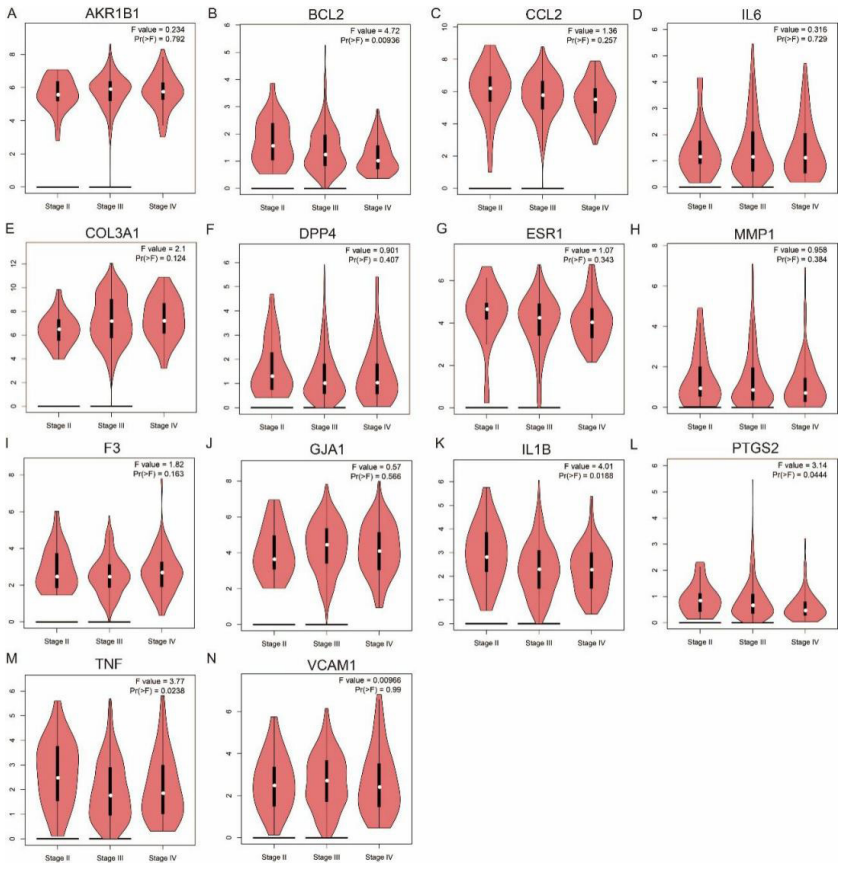



**Figure 7.** Pathological stage in the GEPIA database.

## 4. Discussion

In this work, network pharmacology combined with bioinformatic approaches was applied to decipher the underlying molecular mechanisms by which CBC exerts therapeutic effects on OC. A total of 178 candidate compounds and 161 associated targets were identified, and 14 overlapping targets were obtained by integrating CBC-associated targets with OC-related targets. GO and KEGG enrichment analyses indicated that these targets were associated with inflammatory responses, immune regulation, angiogenesis, and multiple cancer-related signaling pathways. Furthermore, GEPIA analysis revealed differential expression of several candidate genes between OC and normal ovarian tissues, as well as associations between selected genes and clinical characteristics. These outcomes lay a foundation for subsequent exploration of candidate therapeutic targets and the molecular mechanisms underlying CBC's effects against OC.

KEGG enrichment analysis highlighted the AGE-RAGE, IL-17, TNF, NF-κB, NOD-like receptor, and C-type lectin receptor signaling pathways. These biological pathways are implicated in inflammatory regulation, immune responses, and cellular signaling. In particular, NF-κB signaling participates in the regulation of inflammatory mediators and cell survival, while TNF and IL-17 signaling can influence inflammatory responses through interconnected downstream pathways. The enrichment of the NOD-like receptor pathway further suggests a potential association with innate immune regulation. However, these pathways can exert context-dependent effects in cancer, and pathway enrichment alone cannot determine whether CBC activates or inhibits them. Similarly, the identification of these pathways does not establish

NLRP3 inflammasome activation or pyroptosis. The GO results, including inflammatory response and VEGF production, also suggest potential associations with tumor-associated inflammation and angiogenesis. These predicted relationships require further experimental investigation.

The compound-target-pathway network identified several candidate constituents, including luteolin, kaempferol, quercetin, and wogonin. Previous studies have reported that luteolin may affect ovarian cancer stem-cell properties and tumor-cell growth through mechanisms involving KDM4C-associated regulation and the PPP2CA/YAP axis [14]. Kaempferol has been associated with changes in VEGF expression, cell-cycle regulation, and apoptosis in experimental cancer models [15]. Quercetin and wogonin have also been investigated for their potential anticancer activities [16-18]. These findings provide a biological rationale for further investigating the compounds identified in our network. Nevertheless, network connectivity does not directly reflect pharmacological potency, and evidence obtained using isolated compounds cannot establish their individual contributions to CBC. Their abundance, bioavailability, and interactions within the complete formulation remain to be clarified.

The GEPIA analysis showed that BCL2, CCL2, ESR1, F3, IL1B, TNF, and VCAM1 differed significantly in expression between OC and normal ovarian tissues. MPP1 expression was associated with overall survival, while BCL2, IL1B, TNF, and PTGS2 showed differences between pathological stages. These findings provide preliminary clinical context for prioritizing candidate targets, but do not establish causal relationships or demonstrate direct target engagement by CBC. Several limitations should also be acknowledged. The analyses relied on public databases and computational predictions, and the results may be influenced by database coverage, target-selection criteria, and dataset characteristics. Moreover, the relatively small number of overlapping targets warrants cautious interpretation of the enrichment results. Overall, this study provides a systematic overview of candidate compounds, targets, and pathways potentially associated with CBC in OC and offers hypotheses for subsequent experimental validation.

## 5. Conclusion

In conclusion, this study integrated network pharmacology and bioinformatics analysis to explore the potential targets and mechanisms of CBC against OC. Fourteen overlapping targets were identified, and enrichment analyses suggested that these targets were associated with inflammatory responses, immune regulation, and multiple cancer-related signaling pathways, including the TNF, NF-κB, IL-17, and NOD-like receptor signaling pathways. GEPIA analysis further provided insights into the expression patterns and clinical relevance of selected candidate targets in OC. These findings offer a systematic perspective on the potential molecular mechanisms of CBC against OC and provide a basis for further experimental validation.

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## Disclosure statement

The authors declare no conflict of interest.

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