

Review Article

Cyclin E: a potential treatment target to reverse cancer chemoresistance by regulating the cell cycle

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Abstract: The cyclin family plays important roles in regulating the proliferative cycle of mammalian cells. Among the members of this family, cyclin E regulates multiple downstream molecules, such as the retinoblastoma susceptibility gene (RB1) and the transcription factor E2F, by interacting with cyclin-dependent kinases (CDKs) and plays an important role in the cell cycle transition from G1 to S phase. Over the years, studies have shown that cyclin E is closely related to the chemotherapy resistance of tumor cells and that its expression in tumor cells is closely related to prognosis. The dysregulated expression of cyclin E has a definite effect not only on the cell cycle regulation of tumor cells but also on the presence of low-molecular-weight cyclin E (LMW-E) and other cyclins that render tumor cells resistant. In addition, many studies in recent years have confirmed that chemotherapy resistance mediated by cyclin E can be reversed. For example, the combination of a cyclin-dependent kinase inhibitor (CKI) with anticancer drugs or the therapeutic targeting of related genes improves chemotherapy resistance by reducing the level or activity of cyclin E in tumor cells. This review summarizes the specific processes by which cyclin E regulates the cell cycle, its relationship to chemotherapy resistance in cancer, and its potential as a clinical therapeutic target to reverse chemotherapy resistance.

Keywords: Cyclin E, cell cycle, cancer, chemotherapy resistance

Introduction

Chemotherapy drugs can be subdivided according to their mechanism of action into alkylating agents, antimetabolites, antitumor antibiotics, plant anticancer drugs, hormones, and immunological preparations. Among them, antitumor antibiotics, plant anticancer drugs and hormone drugs can inhibit tumor growth by regulating cell mitosis and blocking the cell cycle. These drugs are frequently used in chemotherapy regimens for cancers such as breast cancer, ovarian cancer, stomach cancer, and liver cancer.

However, at the same time, the effects of these drugs in some diseases, such as hepatocellular carcinoma, are unknown, and the advantages of monotherapy are unclear. Although the effects of combination regimens have been studied, the results have been disappointing [1], as reported in ovarian cancer following

chemotherapy [2]. This may be related to the abnormal expression of certain proteins in tumor cells, which decreases the efficacy of chemotherapy drugs, leading to tumor recurrence and chemotherapy resistance.

Cyclins are a class of proteins whose expression rises and falls in tandem with the cell cycle in eukaryotic cells. Cyclin B, the first cyclin to be isolated, was identified in sea urchin embryos. Other cyclins were subsequently discovered by sequence similarity or functional homology [3]. The cell cycle can be artificially divided into G1 phase, G0 phase, S phase, G2 phase and M phase. The cyclin family consists of 11 members, among which cyclin D and cyclin E mainly regulate the G1/S phase transition, and cyclin A is mainly responsible for regulating S phase. Cyclin B is mainly related to the completion of M phase. Cyclin-dependent kinases (CDKs), which control kinase activity and substrate specificity, contain a serine/threonine-specific

catalytic core and partner with cyclins [4]. These cyclins bind to CDKs to form complexes, such as cyclin D/CDK4 and cyclin E/CDK2, which help to promote and regulate the cell cycle.

As a member of the cyclin family, cyclin E plays an important role in regulating the cell cycle [5]. The cyclin E gene encodes several polypeptides whose molecular weights range from 39 to 52 kDa. Full-length cyclin E (FL-E) contains a domain at a specific position in the polypeptide chain, known as the “cyclin box”, whose sequence is relatively conserved among cyclins [6]. All forms of cyclin E with an intact cyclin box are capable of binding to and activating the catalytic subunit of cyclin E, CDK2. Since most substrates of cyclin E/CDK2 are involved in processes that regulate chromatin, such as transcription or DNA replication, and antibody staining shows a predominantly nuclear localization, cyclin E is present in the nucleus [5, 6]. In addition, cyclin E is abnormally detected in various high-grade malignant cells, suggesting that it plays an important role in tumor chemotherapy resistance [7-11].

This article aimed to introduce the key role of cyclin E in the chemoresistance of tumor cells, including the specific mechanism by which cyclin E mediates the cell cycle, and its expression and regulatory mechanisms in drug-resistant cells. We also present a potential program for drug resistance reversal and introduce cyclin E as a new target for tumor resistance testing.

Cell cycle regulation by cyclin E

Retinoblastoma protein (pRb), which belongs to the pocket protein family and is a transcription product of the RB1, negatively regulates cell proliferation. In normal cells, the pRb is present in mitotic and quiescent cells in an active hypophosphorylated form that binds to the LxCxE motifs in many chromatin-associated proteins and transcription factors using a conserved pocket structure found in members of the E2F family [12]. The E2F family is a large, general family of transcription factors with one or more conserved DNA-binding domains that bind to the promoters of target genes and regulate their expression [13].

The pRb negatively regulates the expression of E2F target genes, many of which are required for entry into S phase, by recruiting various repressive chromatin regulatory complexes and histone-modifying enzymes or by blocking the transactivation function of E2F proteins [12]. In mammalian cells, mitotic stimulation signals first induce the synthesis of cyclin D, leading to the formation and nuclear localization of cyclin D/CDK4 and cyclin D/CDK6 complexes, which phosphorylate pRb early in G1 phase; the level of cyclin E is then increased, and cyclin E associates with CDK2 to further phosphorylate pRb in preparation for progression into S phase [13]. In addition, both the correct assembly of the cyclin E/CDK2 complex and the interaction of the cyclin E/CDK2 complex with cyclin-dependent kinase inhibitors (CKIs), such as p27 or p21, are critical for cyclin E to function. The cyclin E/CDK2 complex binds to CKIs to form a ternary complex that inhibits kinase activity. Similarly, the pocket protein p107, a substrate and inhibitor of cyclin/CDK complexes, has been shown to modulate the activity of cyclin E/CDK2 via a p21-like domain [14-16]. Moreover, cyclin E kinase activity is critical in cells lacking pRb. There are additional critical substrates of the CDK2/cyclin E complex, indicating that cyclin E may have important direct functions [17]. For example, cyclin E can induce S phase-specific genes, such as thymidine kinase B-Myb or cyclin A, to induce S phase entry and initiate DNA replication [6].

The cell cycle is also regulated by an important positive feedback amplification loop involving cyclin E, which can be induced by proteins of the E2F transcription factor family. The expression of cyclin E varies during the cell cycle and peaks at the G1/S phase boundary [6]. When the cyclin E/CDK2 complex regulates the entry of cells into S phase and increases E2F activity by phosphorylating E2F, it further amplifies the transcription of cyclin E. Therefore, positive feedback amplification is also an important part of G1/S control [6, 12, 18] (**Figure 1**).

Based on the above findings, determining which factors regulate cyclin E expression is also important for understanding the regulation of the cell cycle. In terms of transcription, the 5' regulatory region of cyclin E, also known as

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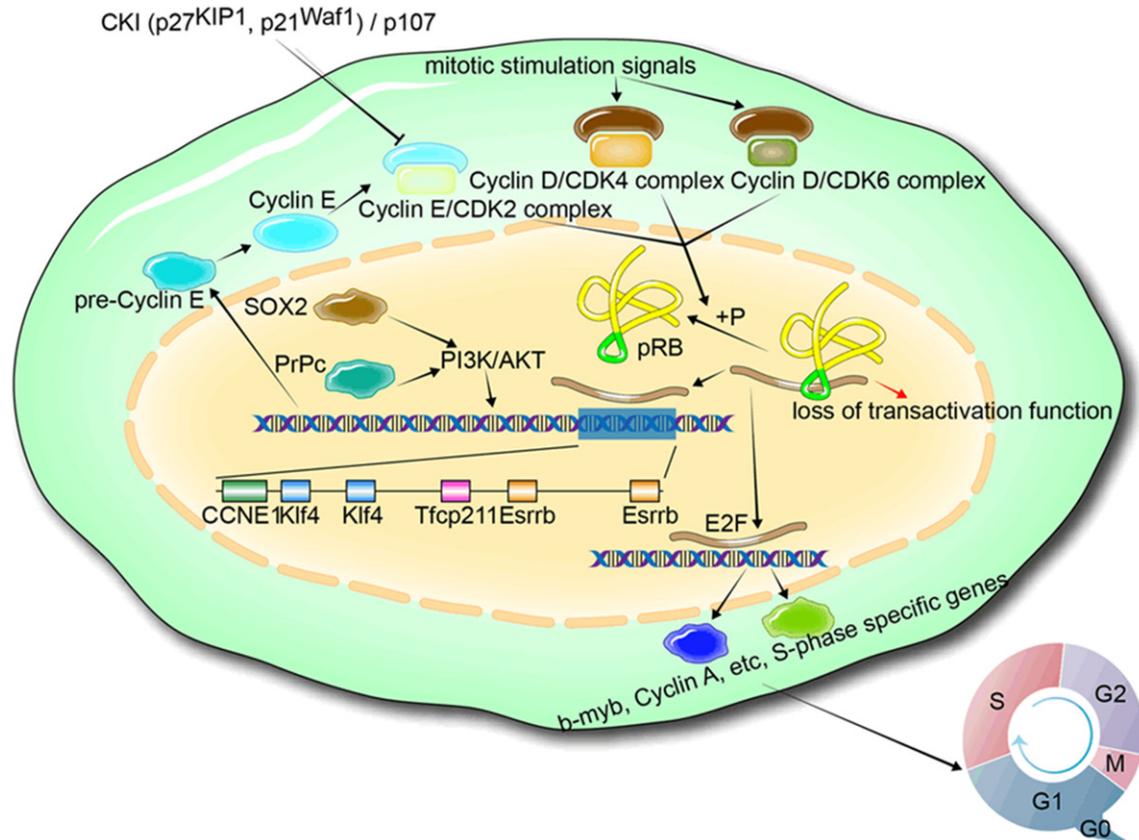


Figure 1. Cell cycle regulation through cyclin E. pRB exerts an inhibitory effect on cell proliferation. It binds to the transcription factor E2F in its hypophosphorylated form to inhibit the binding of E2F to its target genes. A large proportion of these target genes are necessary for cells to enter S phase. When mitotic stimulation signals induce the formation of complexes such as cyclin E-CDK2 and cyclin D-CDK4/6 through the PI3-AKT pathway, which can be activated by SOX2 and PrPc, these complexes phosphorylate pRB in the early G1 phase, and phosphorylated pRB dissociates from E2F, allowing E2F to regulate gene expression normally. Correct assembly of the cyclin E/CDK2 complex and the interaction of the cyclin E/CDK2 complex with CKIs, such as p27 or p21, are critical for cyclin E to function. The cell cycle is also regulated by an important positive feedback amplification loop involving cyclin E that can be induced by proteins of the E2F transcription factor family. When the cyclin E-CDK2 complex regulates the entry of cells into S phase and increases E2F activity by phosphorylating E2F, it further amplifies the transcription of cyclin E.

CCNE1, has been studied, and binding sites for transcription factors of the naive pluripotency network, including Esrrb, Klf4, and Tfcp2l1, have been identified within 1 kilobase upstream of the transcription start site. The knock-down of Esrrb, Klf4, and Tfcp2l1 reduced cyclin E expression, whereas the overexpression of Esrrb and Klf4 increased cyclin E expression, indicating a strong correlation between the expression levels of these factors and cyclin E [19]. Moreover, cyclin E is modified via ubiquitination by the multiprotein complex SCF, which contains the proteins Skp-1, CDC53/Cullin, Rbx1/ROC1 and CDC34 and a variable F-box protein (e.g., FBW7, hCDC4, p45Skp2 or Cullin 3) [20, 21]. The function of SCF, the substrate

recognition component of the SCF ubiquitin ligase, is known, and the F-box protein FBW7/hCDC4 also plays an important role in the regulation of cyclin E. Thus, it is also important to determine which factors regulate FBW7. A previous study identified the ubiquitous miRNA miR-27a as a major suppressor of FBW7; in other words, miR-27 prohibits the function of FBW7, downregulating the ubiquitylation and turnover of cyclin E [22].

Cyclin E-related cancer chemotherapy resistance

There are many cyclin E/CDK2-related chemotherapy resistance mechanisms involved in the

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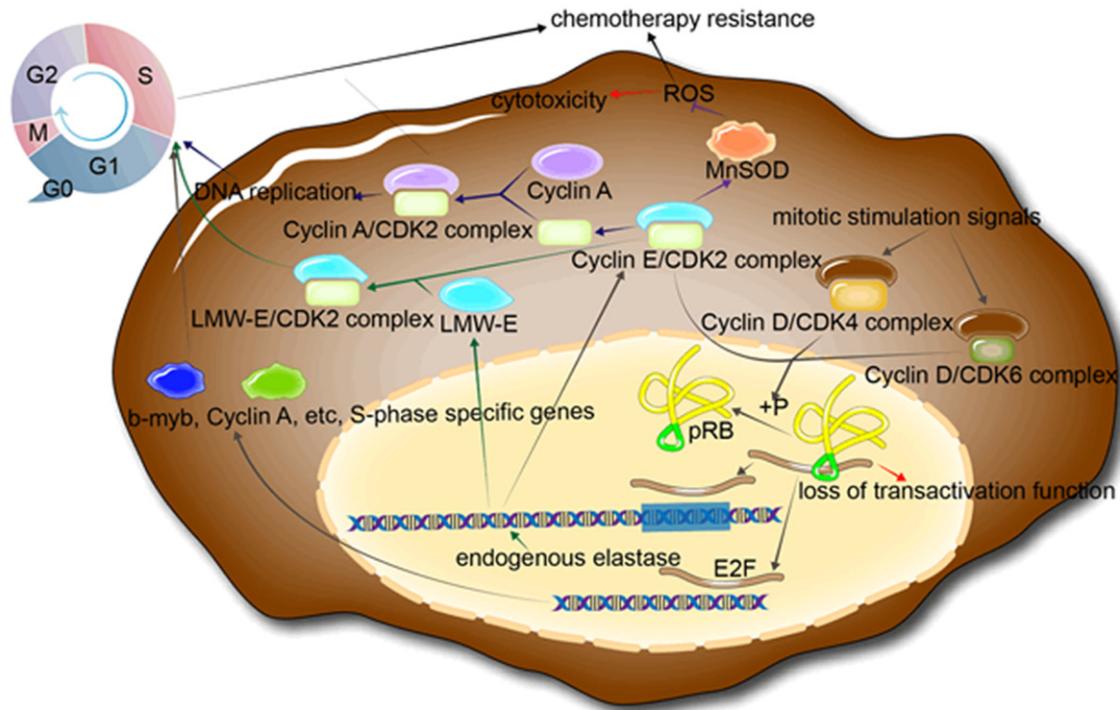


Figure 2. Chemotherapeutic resistance is related to cyclin E. There are three main types of chemotherapeutic resistance in tumor cells related to cyclin E: 1. The overexpression of cyclin E not only accelerates the proliferation and division of tumor cells by promoting cell cycle transformation but also induces the expression of related products, leading to chemotherapy resistance. The increased expression of cyclin E in adriamycin-resistant cells leads to the increased expression of MnSOD, which increases the ability of tumor cells to scavenge ROS, decreasing doxorubicin-induced cytotoxicity. 2. Cyclin A replaces the function of cyclin E and binds to CDK2 to promote cell cycle entry into the S phase. 3. Special forms of cyclin E/LMW-E escape attack by antitumor drugs and bind to CDK2 to promote cell cycle entry into the S phase.

treatment of cancer with chemotherapeutic drugs, and the results of research on corresponding mechanisms to reduce or reverse transformation resistance are also quite critical (Figure 2).

Overexpression of cyclin E and related substances

The general effect of a variety of chemotherapeutic drugs is blockade of the cell cycle, but in tumor cells overexpressing cyclin E, due to induction of the cell cycle by cyclin E/CDK2, the G1/S phase transition is promoted, causing chemotherapy resistance. Studies have confirmed that in a variety of tumors, cyclin E is overexpressed, leading to chemotherapy resistance (Table 1).

Ahn MJ investigated the expression of the cyclin E protein in 84 gastric cancer (GC) tissues

and the relevance of cyclin expression to clinical outcomes. Cyclin E overexpression was noted in 40.5% of GC tissues [23]. Additionally, in another study, specimens from 89 patients with GC treated with "curative" intent were evaluated for cyclin E expression using immunohistochemistry. Cyclin E overexpression was observed in 35 tumor specimens. The incidence of cyclin E overexpression was significantly high in deeply invasive cancers, cancers with lymph node metastasis, and cancers at an advanced stage [24].

Zhou Q detected cyclin E overexpression in 45 hepatocellular carcinoma specimens and 45 intraoperative cancer tissues by the immunohistochemical SABC method [25]. Peng SY examined the RNA in paired hepatocellular carcinoma and liver tissues obtained from 71 patients who were followed for more than 4 years after tumor resection using reverse tran-

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Table 1. Cyclin E expression in different types of cancer

| Type of carcinoma | Total number of cases | Number of cases of cyclin E overexpression | Rate of cyclin E overexpression | Significance and conclusion of the study | Reference |
|---|-----------------------|--|---------------------------------|---|-----------|
| Gastric cancer | 84 | 34 | 40.5% | Cyclin E overexpression was related to poor survival rates; it may useful as a prognostic measure in gastric cancer. | [23] |
| Knockout of INMAP | 89 | 35 | 39.3% | Cyclin E overexpression was an important prognostic factor with regard to survival. Patients with low cyclin E expression had higher five-year survival rates than patients with high expression. | [24] |
| Hepatocellular cancer and intraoperative cancer | 90 | 45 | 50% | Cyclin E overexpression was associated with the formation of tumor thrombi, tumor invasion and metastatic potential. | [25] |
| Pancreatic ductal adenocarcinoma | 118 | 39 | 33.1% | Cyclin E overexpression was identified as an independent predictor of poor outcomes in pancreatic cancer patients. | [26] |
| Metastatic colon cancer | 114 | 32 | 28% | Cyclin E overexpression in cancer was related to an increased risk of tumor recurrence and poor outcomes. | [27] |
| Rectal cancer | 360 | 104 | 29% | Evaluation of cyclin E expression may provide useful prognostic information for rectal cancer patients. | [28] |
| Non-small cell lung cancer | 156 | 64 | 41% | Cyclin E expression was highly variable and determined to play a role in tumor grade and differentiation in NSCLC. | [31] |

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scription-polymerase chain reaction supplemented with Northern blotting and immunohistochemistry. Skalicky DA assessed the expression of cyclin E and p27 by immunohistochemistry using tissue microarrays of tumor samples from 118 patients with pancreatic ductal adenocarcinoma (75 resections and 43 biopsies). High cyclin E expression was identified in 39 of the 118 patients [26].

In the tumors and mucinous tissues of 114 patients with metastatic colon tumors analyzed by Corin I, FL-E was found in 29 (25.4%) tumors and only three (2.6%) mucosa samples; the corresponding values for the low-molecular-weight (LMW) isoform were 80 and 67, respectively [27]. In a study by Zhou YJ, cyclin E expression in 360 rectal cancer patients with stage I to III disease who underwent resection was assessed by Western blotting. A total of 19%, 24% and 29% of patient tumors exhibited elevated levels of FL cyclin E, low-molecular-weight cyclin E (LMW-E) and FL-E, respectively. Cyclin E expression was also significantly correlated with blood vessel invasion, deep invasion, histology grade and lymph node metastasis [28].

The expression of cyclin E is also abnormal in certain endocrine-related cancers. Brzeziński J et al. found cyclin E overexpression in thyroid papillary carcinoma cells [29]. Jordan S et al. studied 95 pituitary cancer specimens and found that cyclin E was sparsely expressed in these specimens but highly expressed in the corticosteroid tumors of patients with Cushing's disease (CD) [30].

Cyclin E is also overexpressed in non-small cell lung cancer (NSCLC). Yamanouchi H found that among 156 previously studied patients with NSCLC, 64 overexpressed cyclin E. Furthermore, cyclin E overexpression was more frequently observed in squamous cell carcinoma than in adenocarcinoma [31].

Among the cancers common among women, Kato N et al. found that both proliferative and secretory endometria and endometrial hyperplasia expressed negligible cyclin E. Cyclin E expression was significantly lower in normal and hyperplastic endometria than in endometrial adenocarcinomas [32]. Among the endometrial specimens, cyclin E immunoreactivity was higher in endometrial cancer samples

than in normal proliferative and hyperplastic endometrial samples [33]. Tsuda H et al. found that between clear cell ovarian carcinoma (CC) and serous ovarian carcinoma (SC), relative cyclin E mRNA expression was significantly higher in CC than in SC. The percentage of cells exhibiting positive nuclear staining for cyclin E was significantly higher in CC than in SC [34]. Farley J et al. studied 139 specimens and observed high cyclin E protein expression in 62 (45%) patients with advanced, suboptimally debulked ovarian cancer [35]. Recently, Zi-Ming Zhao et al. proved that cyclin E1 amplification may confer resistance to chemotherapy and is associated with poor overall survival in patients with triple negative breast cancer (TNBC) [36].

Cyclin E overexpression not only accelerates the proliferation and division of tumor cells by promoting cell cycle transformation but also induces the expression of related products, leading to chemotherapy resistance. In a study of doxorubicin, it was found that doxorubicin induces the formation of reactive oxygen species (ROS), which contributes to its cytotoxicity [37]. The expression of MnSOD, an antioxidant enzyme, especially in cells exposed to doxorubicin, predicts increased resistance to different types of oxidative stress [38]. However, it was found that the increased expression of cyclin E in adriamycin-resistant cells led to the increased expression of MnSOD, which increased the ability of tumor cells to scavenge ROS, thus decreasing doxorubicin-induced cytotoxicity [39, 40]. In 1,25-dihydroxyvitamin D3-resistant sublines of promyelocytic leukemia cells, the protein levels of cyclins E, D1, D2 and D3 are elevated; in contrast, in these sublines, the protein levels of CDK2, CDK4 and CDK6 are not altered, but these CDKs exhibit significantly higher activity. More direct evidence of resistance is the increased ratio of cyclin E to p27 in resistant cells, leading to the activation of CDK2 and termination of G1/S transition blockade induced by 1,25-dihydroxyvitamin D3 [41].

The mechanism of cyclin E overexpression is complicated. The increased expression of cyclin E in some cancers may be due to abnormalities in gene expression. For example, studies have shown that the sex-determining region Y-box2 (Sox2) is aberrantly overexpressed in many types of cancers. Sox2 promotes esophageal carcinoma growth by regulating the

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phosphoinositide 3-kinase (PI3K)/AKT signaling pathway and activates the expression of cyclin E and other key proteins related to cell survival [42]. Sustained activation of the PI3K/AKT signaling pathway, whose key role in cells is to promote the expression of genes necessary for cell survival, confers tumor cell resistance to paclitaxel (Pac) chemotherapy [43]. Prostate cancer is a common cancer that is often treated clinically with docetaxel, a Pac derivative-based chemotherapeutic. Experiments have shown that chemotherapy resistance in prostate cancer is also associated with the expression level of Sox2. In cells with high Sox2 expression, cell proliferation is increased, cell cycle arrest induced by Pac is significantly impaired, and Sox2 expression causes tumor cells to escape Pac-induced apoptosis [44]. Since the mitotic stimulation signal in mammalian cells induces cyclin E to activate CDK2 after the synthesis of cyclin D/CDK4 and cyclin D/CDK6, the abnormal expression of the above two complexes may also lead to the abnormal expression of cyclin E [13]. Cyclin D1 is overexpressed in progesterone-treated breast cancer cells and associated with the decreased activity of cyclin E and the increased expression of p27 [45]. Palbociclib, a specific inhibitor of CDK4/6, exerts antiproliferative effects by inducing G1 phase cell cycle arrest and cell senescence in GC cells, and these effects are determined by the protein expression of cyclin E [46]. The abnormal expression of other proteins in the cell can also induce an increase in cyclin E/CDK2 levels. For example, the expression of prion protein (PrPc) is significantly increased in 5-FU-resistant colorectal cancer (CRC) cells and is related to the abnormal expression of cyclin D1 and cyclin E (relative to normal CRC cells) [47]. The results of this experiment indicated that the activation of PI3K and AKT is significantly increased in 5-FU-resistant CRC cells [47, 48]. Moreover, the activation of AKT promotes cell proliferation through multiple downstream targets (such as cyclin E) and thus regulates the cell cycle [49]. PrPc accelerates the transition from G1 phase to S phase through the PI3K/AKT/cyclin D1 axis [50]. The experiment also showed that the level of PrPc can directly affect the levels of cell cycle-associated proteins, such as cyclin E, CDK2, cyclin D1, and CDK, which in turn affect cell proliferation [47].

Combination of cyclin A and CDK2

Notably, the expression level of cyclin E/CDK2 is downregulated in some forms of protein-driven chemotherapeutic resistance, but this decreased expression promotes the binding of other cyclins to CDK2, which also promotes S phase transition. Cytidine deaminase (CDA) is a proliferation-inhibiting molecule in normal cells that inactivates more than 90% of gemcitabine by converting it into 2'-deoxy-2',2'-difluorouridine, resulting in chemotherapy resistance [51]. Moreover, when CDA is highly expressed, the level of cyclin E/CDK2 is downregulated, leading to the binding of cyclin A to CDK2. When the level of the cyclin A/CDK2 complex reaches a certain threshold, the binding of cyclin E and CDK2 is terminated, which promotes DNA replication in S phase [52]. Thus, CDA-mediated cell cycle redistribution is closely related to the cyclin E/CDK2 complex and its associated molecules [53].

LMW-E: a special form of cyclin E in tumor cells

In tumor tissues, cyclin E is expressed not only in its full-length form but also in its special LMW form. The characteristic patterns of cyclin E processing found in tumor cells and tumor tissues produce many forms of LMW-E, ranging in size from 33 to 45 kDa, and the FL-E, with a molecular weight of 50 kDa [54]. The expression of endogenous elastase causes the expression of LMW-E. The overexpression of various cyclin E isoforms results in an increase in elastase levels, thereby leading to an increase in the ratio of LMW-E to cyclin E [55]. LMW-E also promotes cell cycle transition by binding to CDK2 [56]. In addition, LMW-E has a higher affinity for CDK2 than FL-E, is resistant to inhibition by CKIs, and can cause genomic instability.

Biological significance of cyclin E in other diseases

In addition to its important role in the cell cycle regulation of cancer cells, cyclin E also has certain importance in normal cells.

One example of the importance of cyclin E in normal cells concerns the proliferation of vascular smooth muscle cells (VSMCs). VSMCs in adult arteries normally proliferate very slowly,

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remaining in the G0 phase of the cell cycle. An imbalance between growth factors and inhibitors causes VSMCs to enter a proliferative state from a quiescent state. The uncontrolled proliferation of VSMCs is also the main mechanism of diseases such as atherosclerosis [57]. Based on previous research, the G1 to S phase transition requires a synergistic increase in the intracellular Ca²⁺ concentration [58]. This is at least partly mediated by Ca²⁺/calmodulin (CaM)-dependent cyclin E/CDK2 activity [59]. In recent years, research has shown that the CaM-binding peptide reduces cyclin E/CDK2 activity and eliminates Ca²⁺-sensitive cyclin E/CDK2 activity in nuclear extracts from mouse VSMCs. These findings may indicate a new possible therapeutic approach to atherosclerosis and stenosis [60].

Cyclin E is also important in CD; it leads to hypercortisolism due to pituitary corticotroph tumor ACTH hypersecretion [61]. Past research has found that cyclin E is expressed at significantly higher levels in corticotroph tumors than in tumors associated with the pituitary, and its expression is undetectable in the normal pituitary [30]. Recent research has suggested that R-roscovitine, a 2,6,9-trisubstituted purine analog that downregulates CDK2/cyclin E activity [62], can inhibit gene expression of the corticotroph precursor hormone proopiomelanocortin by targeting the lineage-specific cyclin E/E2F1 pathway [63]. Thus, drugs such as R-roscovitine can be used as pharmacotherapeutic agents against CD.

Research directions and prospects for cyclin E

Important findings during cancer treatment

Studies have shown that many substances and genes with a clear or potential therapeutic effect on cancer affect the expression of cyclin E (Table 2).

Phenylpropanoid-based sulfonamides were synthesized, and their cytotoxic activity was evaluated against the MCF-7 cell line, which is derived from human tumors. These sulfonamides induced cell cycle arrest at the G1/S transition, probably due to their ability to reduce cyclin D1 and cyclin E expression [64]. INMAP was first identified as a spindle protein that plays important roles in cell cycle progression, and previous studies have revealed that

its abnormal expression leads to mitotic disorder and inhibits the growth of human tumor xenografts. INMAP knockout in HEK293T cells upregulated the level of cyclin B, downregulated the level of cyclin E, postponed mitotic exit and caused a spindle assembly anomaly [65].

Sun X investigated whether bufalin can act as a chemoprophylactic agent to prevent colon tumorigenesis in two murine models of colorectal cancer, namely, colitis-associated colorectal cancer and Apc germline mutation-developed colorectal cancer. The long-term administration of low-dose bufalin effectively suppressed tumorigenesis in both colorectal cancer models, accompanied by attenuated epithelial cell proliferation (reduced bromodeoxyuridine incorporation, decreased levels of cyclin E and cyclin-dependent kinases-2/4, and increased levels of p21 and p27) and promoted apoptosis [66].

Neuropilin-1 (NRP-1) is a nontyrosine kinase receptor that interacts with multiple signaling pathways, underpinning the biological behavior and fate of cancer cells. In pancreatic cancer, NRP-1 depletion inhibits cell proliferation by inducing cell cycle arrest at the G0/G1 phase by upregulating p27 and downregulating cyclin E and CDKs [67].

Ganoderiol F is purified from Ganoderma. The ganoderiol F-mediated suppression of breast cancer cell viability occurs through cell cycle arrest. Furthermore, ganoderiol F downregulates the expression of cyclin D, CDK4, CDK6, cyclin E and CDK2 and inhibits cell cycle progression, arresting cells in G1 phase. These results show that cyclin D-CDK4/CDK6 and cyclin E-CDK2 are central components of the mechanism by which ganoderiol F regulates cell cycle progression [68].

β-Cryptoxanthin has been associated with a reduced risk of some cancers. In this study, we examined the effect of β-cryptoxanthin on AMPK signaling in human GC cells. β-Cryptoxanthin treatment induced G0/G1 arrest, reduced the expression of cyclin E, cyclin D1, CDK4 and CDK6, and increased the expression of p53 and p21 in two GC cell lines [69].

Trifluoperazine (TFP), an approved antipsychotic drug, has been reported to have potential anticancer effects against several cancer

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Table 2. Cyclin E-related tumor treatment

| Drug/treatment | Function | Effect | Reference |
|------------------------------------|---|---|-----------|
| Phenylpropanoid-based sulfonamides | Downregulate the expression of cyclin D1 and cyclin E | G1/S cell cycle arrest | [64] |
| Knockout of INMAP | Upregulates the expression of cyclin B and downregulates the expression of cyclin E | Postponement of mitotic exit and spindle assembly anomaly | [65] |
| Bufalin | Downregulates the expression of cyclin E and CDKs and upregulates the levels of p21 and p27 | Tumorigenesis suppression | [66] |
| NRP-1 depletion | Downregulates the expression of cyclin E and CDKs and upregulates the levels of p21 and p27 | G0/G1 cell cycle arrest | [67] |
| Ganoderiol F | Downregulates the expression of cyclin D, CDK4, CDK6, cyclin E and CDK2 | G0/G1 cell cycle arrest | [68] |
| β-Cryptoxanthin | Reduces the expression of cyclin E, cyclin D1, and CDK4/6 and increases the expression of p53 and p21 | G0/G1 cell cycle arrest | [69] |
| Trifluoperazine (TFP) | Downregulates the expression of CDK2/4 and cyclin D1/E and upregulates the expression of p27 | G0/G1 cell cycle arrest | [70] |
| Metformin | Downregulates the expression of CCNE1/2 and CDK4/6 | Tumor proliferation inhibition | [71] |
| XRRA1 low expression/silenced | Regulates the cyclin A, cyclin E, and p21 proteins | G2/M cell cycle arrest | [72] |
| VR23 | Ubiquitinates cyclin E | Abnormal mitosis of tumor cells and cell cycle arrest | [73] |
| Fisetin:etoposide | Downregulates the expression of cyclin B1 and E1 | Cell cycle arrest | [74] |
| miR-195 | Targets cyclin E1 | Reverses TMZ resistance | [78] |
| HuR | Downregulates the expression of cyclin E | G1 cell cycle arrest and induces apoptosis | [79] |
| CDK 2 inhibitor | Indirectly interferes with cyclin E1 function | Reverses chemoresistance | [81] |
| CCNE1/PIK3CA blockade | Targets cyclin E1 and Pik3ca mutation | Reverses trastuzumab resistance | [82] |
| Tehranolide | Modulates the PI3K/AKT signaling pathway and downregulates the expression of cyclin D1 | G0/G1 cell cycle arrest G1/S cell cycle arrest | [83] |

types. In vitro studies showed that TFP induced G0/G1 cell cycle arrest to dramatically inhibit CRC cell proliferation by downregulating CDK2, CDK4, cyclin D1, and cyclin E and upregulating p27 [70].

Metformin has been reported to function as an antitumor agent by inhibiting the growth of different cancer types, including bladder cancer. Our study revealed a new potential regulatory pathway through which metformin inhibits cell proliferation via the AMPK α /Yap1/TEAD4/CCNE1/2 axis in bladder cancer (BLCA) cells. Metformin decreased the expression of CCNE1/2 and CDK4/6, providing new insights into novel molecular therapeutic targets for BLCA [71]. Wang W found that the blockade of XRR1 expression led to G2/M cell cycle arrest by regulating the cyclin A, cyclin E, and p21 proteins in CRC, and the expression of XRR1 reduced cell cycle arrest and increased cell proliferation in CRC [72].

Proteasome inhibitors are considered a new type of treatment. Proteasomes play a critical role in the proliferation and survival of cancer cells. Therefore, the inhibition of proteolysis cannot only activate apoptosis but may also prevent angiogenesis and metastasis. Sheetal Pundir et al. reported a new type of proteasome inhibitor, 7-chloro-4-(4-(2,4-dinitrophenylsulfonyl)piperazin-1-yl)quinoline (VR23), which ubiquitinates cyclin E, leading to the abnormal mitosis of tumor cells and cell cycle arrest [73].

In the study of osteosarcoma chemotherapy resistance, José Miguel P Ferreira de Oliveira et al. proved that fisetin: etoposide combinations are effective for the inhibition of cell proliferation. These combinations result in decreased levels of cyclin B1 and E1 and exert strong anti-proliferative effects associated with cell cycle arrest in osteosarcoma [74].

MicroRNA-195 (miR-195) plays a tumor suppressor role in multiple tumors by targeting different target genes [75-77]. It has been reported that miR-195 is downregulated in temozolomide (TMZ)-resistant glioma cells and that CCNE1 is its direct target gene. This result indicates that miR-195 reverses chemoresistance to TMZ by targeting CCNE1. Therefore, miR-195 and its target gene CCNE1 could be a potential target for the treatment of glioma with TMZ resistance [78].

Human antigen (Hu) R is an RNA-binding protein that has been described as a molecule related to tumor resistance. In one study, CMLD-2, a small-molecule inhibitor directed against HuR, caused G1 phase cell cycle arrest and induced apoptosis in an NSCLC model. These effects are related to the reduction in cyclin E [79].

Consistent with these findings, another study reported that trastuzumab resistance in uterine serous carcinoma (USC) is closely related to CCNE1 amplification [80]. In recent years, another study found that the use of CDK2 inhibitors to indirectly interfere with CCNE1 function may reverse chemoresistance [81]. Recently, researchers reported that the presence of oncogenic mutations in the *Pik3ca* gene is another reason for resistance to trastuzumab, indicating that dual CCNE1/PIK3CA blockade may represent a novel therapeutic option for USC patients harboring recurrent CCNE1-amplified/PIK3CA-mutated tumors [82].

Tehranolide, a novel natural sesquiterpene lactone with an endoperoxide group, bears structural similarity to artemisinin and has been shown to inhibit cell growth. This study yielded promising results, showing for the first time that Tehranolide inhibits the growth of cancer cells. The selective inhibition of cancer cell growth, apoptosis induction via the mitochondrial pathway, and G0/G1 arrest by modulating the PI3K/AKT signaling pathway and downregulating cyclin D1 lead to the release of p27 and the association of this inhibitor with the cyclin E/CDK2 complex, ultimately preventing cell cycle progression from G1 to S phase [83].

Breakthrough point for reversing chemotherapy resistance

Some findings related to this topic are as follows: A1/BFL1 inhibits cyclin E, slows cell proliferation and promotes cell differentiation, thereby mediating chemotherapy resistance [84]; all-trans retinoic acid inhibits the expression of cyclin E and cyclin D in immortalized human bronchial epithelial cells [85]; and the Fbw7 protein, IFN- α and lamotrigine upregulate CDK2 inhibitor expression in cancer cells and reduce cyclin E expression [86-88]. The use of these substances may play a role in the treatment of chemotherapy resistance, but further research is needed. Studies have been carried

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out to explore solutions for chemotherapy resistance caused by the abnormal expression of cyclin E and its related molecules. Silencing the genes relevant to cyclin E is an important pathway for reverse transformation therapy [7].

Patients with HER2-overexpressing breast cancer often show resistance to trastuzumab after treatment [89]. Studies have shown that such resistance is associated with the upregulation of cyclin E and the increased activity of CDK2 [80]. MUC1 is a heterodimeric protein that binds to HER2 at the surface of breast cancer cells [90], and the resistance of tumor cells to trastuzumab is related to increased binding between the C-terminal fragment of MUC1-C and HER2 [9]. Current studies have demonstrated that MUC1-C contributes to HER2 activation in HER2-overexpressing breast cancer cells and thereby promotes their growth and clonogenic survival. Further studies have shown that targeting MUC1-C suppresses the activation of HER2 in trastuzumab-resistant cells, resulting in the decreased abundance of cyclin E and decreased CDK2 activity; similarly, silencing MUC1 promotes the inhibitory effects of trastuzumab on cell growth [9].

Similarly, a study on Pac resistance in the treatment of cervical cancer found that the PI3K pathway is more significantly activated in paclitaxel-resistant cells than in parental cells. Combination therapy with PTX and PI3K inhibitors inactivated cyclin A1, cyclin B1, cyclin E, and CDC2 expression, contributing to increase paclitaxel-induced S and G2M arrest in PTX-R cell sublines [10]. CARD-recruited membrane-associated protein 3 (CARMA3) is overexpressed in some human tumors and is associated with tumor cell resistance. Experiments have shown that cyclin E levels are decreased in individuals in which the CARMA3 gene is deleted and that the killing effect of cisplatin drugs on tumor cells is enhanced [11].

There is also evidence showing that statins increase the sensitivity of pancreatic cancer cells to cytostatic drugs such as gemcitabine [91], and this effect of statins has been confirmed in vivo [92]. PI3K/AKT is associated with the chemotherapy resistance of pancreatic cancer to gemcitabine [93]. A recent study showed that atorvastatin markedly reduced several PI3K/AKT signaling molecules, such as

cyclin E, which reversed drug resistance and increased the effectiveness of gemcitabine.

Effect of CKIs on chemotherapy resistance

CKIs, such as p27 and p21, inhibit the activity of CDKs by forming a ternary complex with the cyclin E/CDK2 complex [94]. When the expression of CKI is dysregulated, such as the dysregulation that occurs with an increase in the ratio of cyclin E/p27, CDK2 is abnormally activated, resulting in drug resistance.

Histone acetylase inhibitor upregulates p21 and p27 in PXD101-resistant diffuse large B cell lymphoma (DLBCL) cells, increasing the correlation between CKIs and cyclin E/CDK2, and plays a key role in G1 blockade [95]. Troglitazone inhibits the expression of skp2, leading to a reduction in the ubiquitination of p27, which accumulates in human hepatoma cells; this process is one of the mechanisms of the antitumor effect of troglitazone [96]. Studies have shown that troglitazone further affects the expression of cyclin E by upregulating or downregulating the expression of p27, thereby promoting or inhibiting chemotherapy resistance, respectively. The oncogenic kinase Src regulates p27 stability through the phosphorylation of p27 at tyrosine 74 and tyrosine 88. Src-phosphorylated p27 inhibits cyclin E/CDK2 poorly in vitro, and Src transfection reduced p27/cyclin E/CDK2 complexes. Data indicate that the phosphorylation of p27 by Src impairs the inhibitory action of p27 against CDK2 and reduces its steady-state binding to cyclin E-CDK2 to facilitate cyclin E-CDK2-dependent p27 proteolysis [97]. In ovarian cancer cell lines with elevated Src activity, the combination of the Src/abl inhibitor sunitinib and the estrogen receptor antagonist Fulvestrant increased p27 expression and inhibited cyclin E/CDK2 and cell cycle progression. However, the above effects were not clear when either compound was used alone [98]. Moreover, in tamoxifen-resistant breast cancer cell lines, the inhibition of Src expression increased the level of p27 and restored sensitivity to tamoxifen [97]. In addition, the inhibition of CDK2 expression restored the sensitivity of tamoxifen-resistant cells [99].

Roscovitine is a CKI. Because LMW-E cannot bypass blockade caused by the androstenedi-

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one-induced increase in CDK2, roscovitine can be used in breast cancer patients who are unresponsive to AI treatment to overexpress LMW-E in tumor cells [100]. In HR-positive breast cancer patients, the combination of a CDK2 inhibitor and AI may also be an effective treatment [101]. Studies have shown that the inhibition of CDK2 significantly reduced tumor growth in trastuzumab-resistant xenografts in vivo [80]; therefore, the combination of trastuzumab and a CDK2 inhibitor may be suitable for HER-amplified breast cancer patients.

Cyclin E is a potential tumor marker

The effect of cyclin E on chemoresistance also makes it applicable as a cancer marker.

Cyclin E can serve as a clinical biomarker for invasive breast cancer [102]. CCNE1 mRNA has also been identified as a biomarker that is predictive of the efficacy of palbociclib [103]. A study showed that cyclin E could identify patients with the highest likelihood of recurrence among different patient cohorts with different subtypes of cancer [104].

The levels of total cyclin E and LMW-E in tumor tissues are strongly correlated with the survival rate of breast cancer patients based on Western blot analysis [105]. In one study, Kaplan-Meier survival analysis showed that among breast cancer patients who received doxorubicin treatment, those with low cyclin E expression had a better prognosis than those with high cyclin E expression [106].

Among amplicons in high-grade serous carcinomas, the CCNE1 locus, which encodes cyclin E, is the most common, occurring in approximately 36% of specimens [107], indicating that cyclin E is closely related to the pathogenesis of ovarian cancer. Among cases of serous tubal intraepithelial carcinoma (STIC), the lesions of which are believed to be precursors of most high-grade SCs [108], cyclin E nuclear staining was observed in 24 of 35 (77%) STICs, while tubal epithelial cells with a normal appearance were all negative. The above results indicate that cyclin E detection can be used to predict high-grade ovarian serous carcinoma in the early stages of development [109].

The overexpression of cyclin E is also associated with a poor prognosis in gastrointestinal

cancer and may be a prognostic marker for gastrointestinal cancer in clinical practice [110]. Cyclin E is closely related to the TNM stage and pathological differentiation in colorectal cancer tissues, median progression-free survival and prognosis. Therefore, cyclin E can also be used as a highly sensitive, specific and accurate marker for the diagnosis of colorectal cancer [111].

Conclusion

In summary, cyclin E is a key link in the drug resistance mechanism of many tumor cells. Due to this characteristic, we can use cyclin E as a target to reverse or reduce chemotherapy resistance in cancer treatment, suggesting that a variety of substances that regulate cyclin E play an important role in chemotherapy resistance.

The role of cyclin E in chemotherapy resistance cannot be ignored. However, the treatment of chemotherapy resistance with cyclin E requires more research in the laboratory and clinical experience.

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Disclosure of conflict of interest

None.

Abbreviations

BLCA, Bladder carcinoma; CaM, Ca²⁺/calmodulin; CARMA3, Membrane-associated protein 3; CC, Clear cell ovarian carcinoma; CD, Cushing's disease; CDA, Cytidine deaminase; CDKs, Cyclin-dependent kinases; CKIs, Cyclin-dependent kinase inhibitors; CRC, Colorectal cancer; FL-E, Full-length cyclin E; LMW-E, Low-molecular-weight cyclin E; NRP-1, Neuropilin-1; NSCLC, Non-small cell lung cancer; Pac, Paclitaxel; PI3K, Phosphoinositide 3-kinase;

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pRb, Retinoblastoma protein; PrPc, Prion protein; RB1, Retinoblastoma susceptibility gene; Rb, Retinoblastoma tumor suppressor protein; SC, Serous ovarian carcinoma; Sox2, Sex-determining region Y-box2; STIC, Serous tubal intraepithelial carcinoma; TFP, Trifluoperazine; VSMCs, Vascular smooth muscle cells.

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