


The Prognostic Role of Cyclin D1 in Multiple Myeloma: A Systematic Review and Meta-Analysis

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Abstract

Purpose: Cyclin D1 has been identified as a proto-oncogene associated with the uncontrolled proliferation of tumor cells. This systematic review and meta-analysis aims to estimate the prognostic significance of cyclin D1 in multiple myeloma (MM) patients. **Method:** We searched for qualified data in PubMed, Embase, and Web of Science up to February 2020. Data quality was assessed by the Newcastle-Ottawa scale (NOS). Hazard ratios (HRs) and 95% confidence intervals (95% CIs) were used to evaluate the relationship between cyclin D1 expression and overall survival (OS), progression-free survival (PFS)/event-free survival (EFS) in patients with MM. **Result:** A total of 13 studies involving 961 patients were included. Overall, pooled analysis revealed significant heterogeneity between cyclin D1 expression and the prognosis of MM (OS, HR = 1.08, 95% CI: 0.71–1.64, $I^2 = 67.9%$; PFS/EFS, HR = 0.97, 95% CI: 0.49–1.93, $I^2 = 85.8%$). Subgroup analysis revealed that the prolongation of OS was relevant to increased expression of cyclin D1 in MM patients in the relapsed and refractory group (OS, HR = 0.46, 95% CI: 0.24–0.90). Another subgroup assessment of OS established that MM patients with *CCND1* overexpression in the bortezomib group had longer survival time (HR = 0.30, 95% CI: 0.11–0.82), whereas, those overexpressing *CCND1* in the conventional chemotherapy group had poor prognosis (HR = 2.19, 95% CI: 1.18–4.08). We also found that increased cyclin D1 expression correlated favorably with PFS in the autologous stem cell transplantation (ASCT) (HR = 0.45, 95% CI: 0.28–0.73) or reverse transcription-polymerase chain reaction (RT-PCR) group (HR = 0.41, 95% CI: 0.26–0.64). **Conclusion:** The result of this meta-analysis suggested that *CCND1* overexpression might be a predictive biomarker for MM patients when treated with bortezomib, receiving ASCT, or in relapsed and refractory period.

Keywords

multiple myeloma, prognosis, bortezomib, cyclin D1, meta-analysis

Abbreviations

OS, Overall survival; PFS, Progression-free survival; EFS, Event-free survival; NR, Not report; ND, Newly diagnosed; RR, Relapsed/refractory; NOS, Newcastle-Ottawa Scale; NF- κ B, Nuclear factor κ -B; EIF4E, Eukaryotic initiation factor 4E; NOXA,

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A BH3-only member of Bcl-2; AKT, Protein kinase B; PERK, PKR-like ER kinase; CHOP, C/EBP-homologous protein; UPR, Unfolded protein response; ER, Endoplasmic reticulum; Rb, Retinoblastoma gene; E2F, The E2F family of DNA-binding transcription factors; P, Phosphoric acid.

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Introduction

Multiple myeloma (MM) is the second most widespread tumor in the hematological system, and its main feature is the clonal proliferation of malignant plasma cells in the bone marrow.¹ In recent years, with the advent of proteasome inhibitors (PIs), immunomodulatory drugs (ImiDs), and monoclonal antibodies (Mab), the 5-year survival rate of MM patients has increased to 40%.² However, overall, their long-term prognosis remains unsatisfactory. The soluble IL-6 receptor (sIL-6R), β 2-microglobulin (β 2-MG), and *TP53* mutation have all been shown to be valuable to predict survival of MM patients.³⁻⁵ Hematologists remain committed to discovering new prognostic indicators to guide clinical individualized treatment more accurately.

The double allele named *CCND1*, mostly hyperdiploid, is expressed in 40% of MM patients.⁶ Cyclin D1, which is expressed by the *CCND1* gene, belongs to the G1 cyclin family that promotes progression from the G1 phase to the S phase by regulating transcription factor E2F-1 and participates in tumorigenesis and development.⁷ *CCND1* is located at the second core of the 11q13q14 amplicon and acts as the “promotor” of 11q13q14 amplification. The frequent co-amplification of the 11q13 gene in cancer suggests the presence of a group of genes in the amplicons associated with tumor growth and metastasis dominance, which may be related to double-strand DNA breaks in fragile chromosomal sites.^{8,9} The most common chromosomal translocation involving *CCND1* is t(11;14)(q13;q32), which juxtaposes *CCND1* with immunoglobulin heavy chain enhancer elements, resulting in cyclin D1 protein expression.¹⁰ These reactions and interactions suggest that cyclin D1 may play a major role in predicting prognosis and making a clinical decision.

So far, there has been evidence from meta-analyses that *CCND1* overexpression has prognostic value in renal cancer, bladder cancer, oral squamous cell carcinoma, breast cancer, and melanoma.¹¹⁻¹⁵ However, the relationship between *CCND1* overexpression and the prognosis of MM patients remains controversial. While Sewify et al¹⁶ and Tasidou et al¹⁷ found that expression of *CCND1* increased with the progression of MM, suggesting that MM patients overexpressing *CCND1* have a poor prognosis, Soverini et al¹⁸ and Cook et al¹⁹ discovered that cyclin D1 overexpression was linked to better prognosis of MM patients. Due to the complex nature of the regulatory mechanism of cyclin D1 in the cell cycle, there are insufficient evidence-based analyses on the prognostic significance of cyclin D1 expression in MM. Therefore, in this systematic review and meta-analysis, we have analyzed the

survival data extracted from publications to provide a basis for finding new targets for MM treatment.

Materials and Methods

The systematic review and meta-analysis has been registered at INPLASY (ID: INPLASY 202190019). Our study protocol was designed and implemented in accordance with PRISMA reporting guidelines²⁰ and Cochrane Collaboration criterion.²¹

Retrieval Strategy

We searched for studies on the relationship between cyclin D1 and the prognosis of MM published up to February 2020 in PubMed, Web of Science, and EMBASE. We used the method of subject word + free word search to search the words “myeloma” or “multiple myeloma” and “cyclin D1 or *CCND1*” and “result” or “prognosis” or “survival” without geographical and linguistic restrictions. To further expand the scope of retrieval, we also manually searched unretrieved original literature and relevant references.

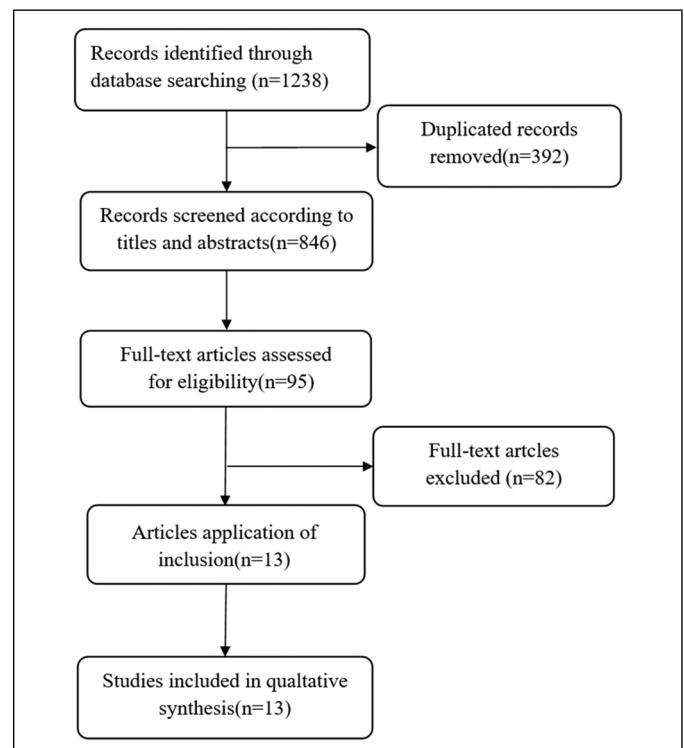


Figure 1. Flowchart of database search and study inclusion.

Table 1. Detailed Characteristics of Included Studies.

Author	Year	Country	Mean Age(year)	Sample Size	Follow-up period(months)	ND/RR	Stage	Detection methods	Outcome	NOS	Study design
Marković O	2004	Serbia	63	59	29-36	59/0	DS I-III	immunohistochemistry	OS	6	Prospective cohort study
Kelley TW	2008	USA	59	94	NR	49/45	SWOG stage I-IV DS I-III	immunohistochemistry	OS/PFS	7	Prospective cohort study
Soverini S	2003	Italy	53	74	9-64,34	74/0	DS I-III	RT-PCR	EFS	7	Prospective cohort study
Cook JR	2006	USA	58	44	NR	44/0	NR	immunohistochemistry	OS/PFS	9	Prospective cohort study
Ngo BT	2010	Germany	67	20	NR	0/20	DS I-III	RT-PCR	OS/PFS	7	Retrospective cohort study
Pruneri G	2008	Italy	64	128	27	NR	DS I-III	immunohistochemistry	OS/PFS	7	Retrospective cohort study
Dawson MA	2009	Australia	61	89	1-20,11	0/89	NR	immunohistochemistry	OS	8	Prospective cohort study
Maillet D	2012	Mexico	59	24	1-54	24/0	NR	immunohistochemistry	OS	6	Prospective cohort study
Tasidou A	2012	Greece	68	130	22	130/0	ISS I-III	immunohistochemistry	OS	8	Prospective cohort study
Sewify EM	2014	Egypt	59	50	NR	NR	ISS I-III	FISH	OS/PFS	7	Retrospective cohort study
Rasmussen T	2001	Denmark	62	76	NR	76/0	DS I-III	RT-PCR	OS	6	Prospective cohort study
Hoechtlen-Vollmar	2000	Germany	63	50	14	50/0	DS I-III	immunohistochemistry	OS	7	Prospective cohort study
Inagaki A	2013	Japan	68	123	24	NR	DS I-III	RT-PCR	OS/PFS	7	Prospective cohort study

Abbreviations: OS, overall survival; PFS, progression-free survival; EFS, event-free survival; NR, not report; ND, newly diagnosed; RR, relapsed/refractory; NOS, Newcastle-Ottawa scale.

Table 2. Main Results of Pooled HRs in the Meta-Analysis.

Comparisons	HRs	95% CIs	Heterogeneity, I^2 , %	<i>P</i> -value
OS	1.08	0.71-1.64	67.9	0.000
Detection method				
IHC	1.19	0.74-1.90	65.1	0.003
RT-PCR	0.57	0.24-1.35	59.3	0.061
Cut-off value				
10%	0.73	0.17-3.17	76.6	0.039
20%	1.69	0.61-4.68	83.5	0.002
30%	1.32	0.77-2.27	0	0.629
50%	0.87	0.44-1.71	0	0.321
Cohort studies				
Prospective	0.98	0.61-1.58	67.9	0.001
Retrospective	1.45	0.42-5.04	76	0.015
Disease phase				
Relapsed and refractory	0.46*	0.24-0.9	0	0.523
Newly diagnosed	1.24	0.73-2.09	73.5	0.000
Treatment				
Bortezomib	0.3*	0.11-0.82	0	0.853
Conventional chemotherapy	2.19*	1.18-4.08	59	0.062
PFS	0.97	0.49-1.93	85.8	0.000
Detection method				
IHC	1.13	0.75-1.72	35.5	0.199
RT-PCR	0.41*	0.26-0.64	0	0.481
Whether or not ASCT				
Non-ASCT	1.28	0.43-3.83	88.2	0.000
ASCT	0.45*	0.28-0.73	0	0.825
Cohort studies				
Prospective studies	0.78	0.44-1.39	67.4	0.015
Retrospective studies	1.31	0.22-7.72	93.1	0.000

Abbreviations: OS, overall survival; PFS, progression-free survival; ASCT, autologous stem cell transplantation; RT-PCR, transcription-polymerase chain reaction; IHC, immunohistochemistry. * means results were statistically significant.

Inclusion and Exclusion Criteria

Inclusion criteria: (1) studies that detected cyclin D1 expression/*CCND1* amplification in MM cells via immunohistochemistry (IHC), reverse transcription-polymerase chain reaction (RT-PCR) or fluorescence in situ hybridization (FISH); (2) studies that reported the relationship between cyclin D1 expression and survival parameters, such as overall survival (OS), progression-free survival (PFS), and event-free survival (EFS); (3) studies that divided *CCND1* expression into positive and negative groups; (4) studies that designed a prospective or retrospective cohort study; (5) studies that provided available hazard ratios (HRs) and 95% confidence intervals (95% CIs) for survival analysis.

Exclusion criteria: (1) reviews, case reports, and animal studies; (2) studies with no available data or no full text related to the topic; (3) repeated articles. If the selected study overlapped with data from other published literature, we selected the latest and largest sample study.

Data Extraction

Two researchers independently extracted the data from predetermined included studies: the first author, mean age, year of

publication, sample size, country, detection methods, treatment methods, clinical-stage, disease phase, follow-up period, prognostic indicators (OS, PFS, EFS, HRs, and 95% CIs) and study design. If the data information in any study was inadequate, we requested the full data we needed from the corresponding author. Any differences of opinion between the two independent researchers were settled by consulting a neutral researcher.

Quality Evaluation

The quality of selected studies was assessed by the Newcastle-Ottawa scale (NOS). We graded the selected articles based on the choice of the study, comparability between groups, and determination of the results. Scoring ranged from 0 to 9, with 0 being the minimum score, and 9 being the maximum score. Studies with NOS ≥ 6 considered to be of high quality and low biased risk. All differences between the two observers involved in the process were settled through dialogue.

Statistical Analysis

The meta-analysis was conducted via the STATA software (version 16.0). OS and PFS/EFS were used to determine the prognosis. HRs and 95% CIs were used to evaluate the

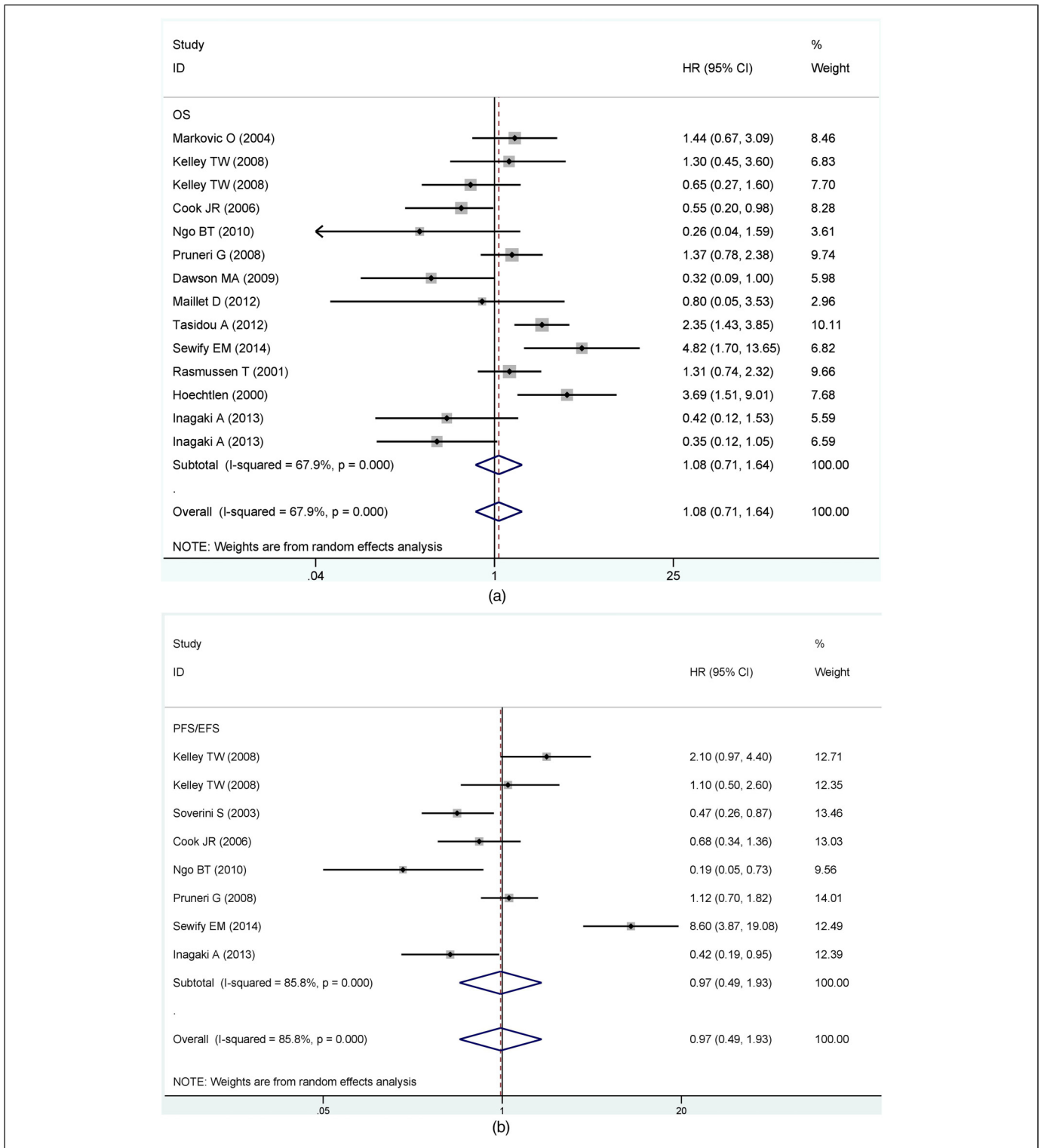
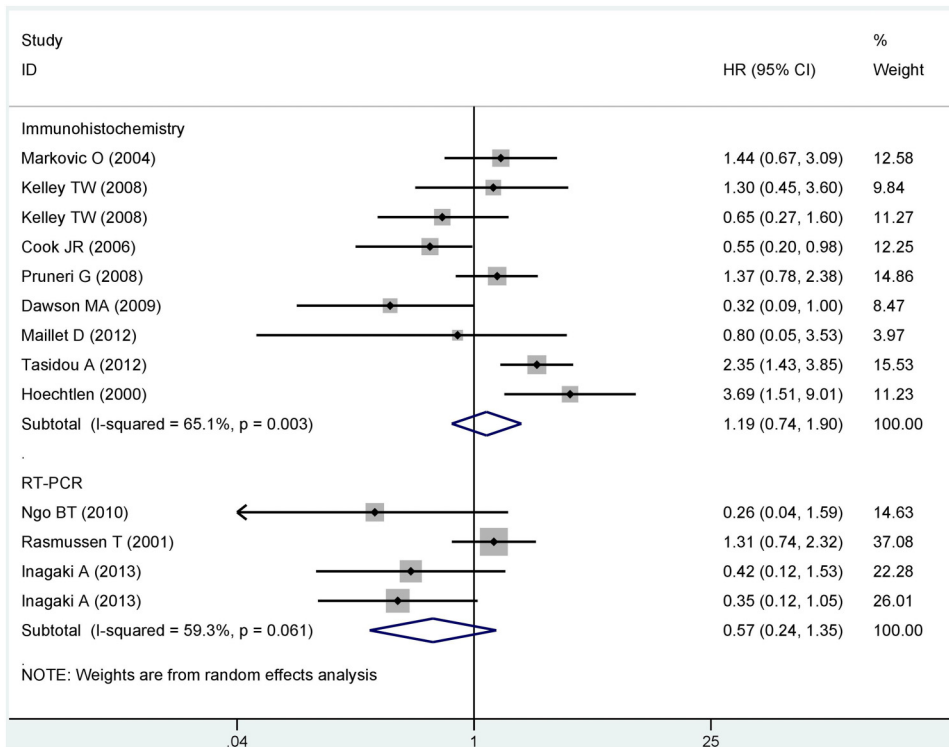


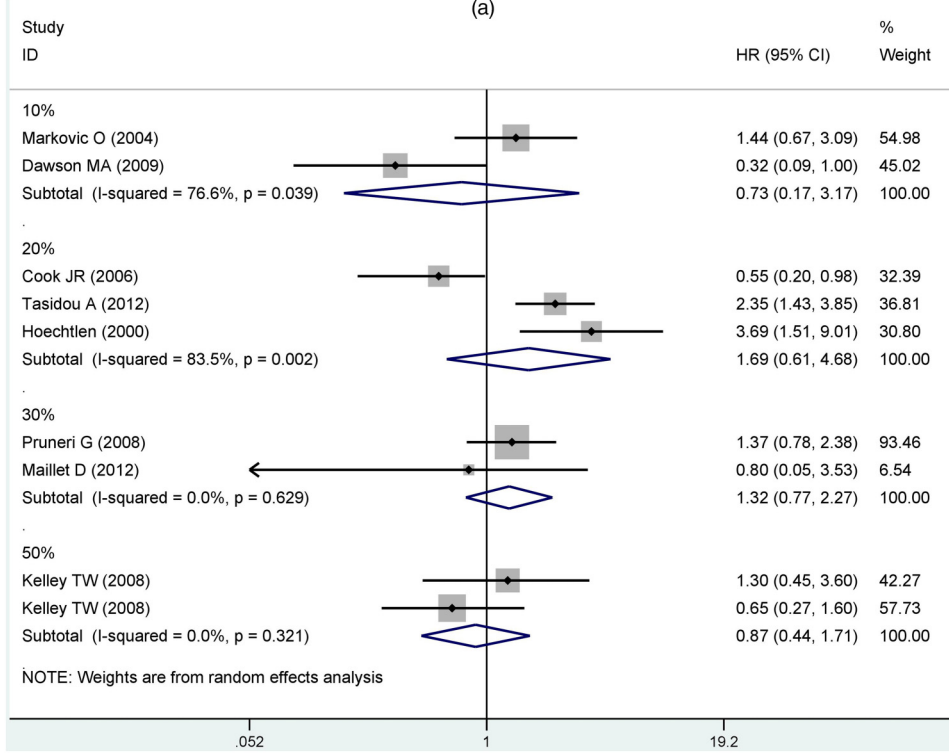
Figure 2. Forest plots for the association between cyclin D1 and (a) OS and (b) PFS/EFS of MM patients. Abbreviations: OS, overall survival; PFS/EFS, progression-free survival/event-free survival.

relationship between *CCND1* expression and the prognosis of MM. For articles that did not directly report HRs and 95% CIs but provided Kaplan-Meier (KM) curves, we used Engauge Digitizer4.1 to extract survival data from the KM

curves and then calculated the HRs and 95% CIs. An HR > 1 signified a negative result for *CCND1* expression, which is associated with a poor prognosis. Conversely, an HR < 1 denoted a positive result. Heterogeneity among included



(a)



(b)

Figure 3. Subgroup analysis of overall survival (OS) by (a) different detection methods; (b) cut-off value; (c) cohort studies; (d) disease phases; (e) treatment. (continued)

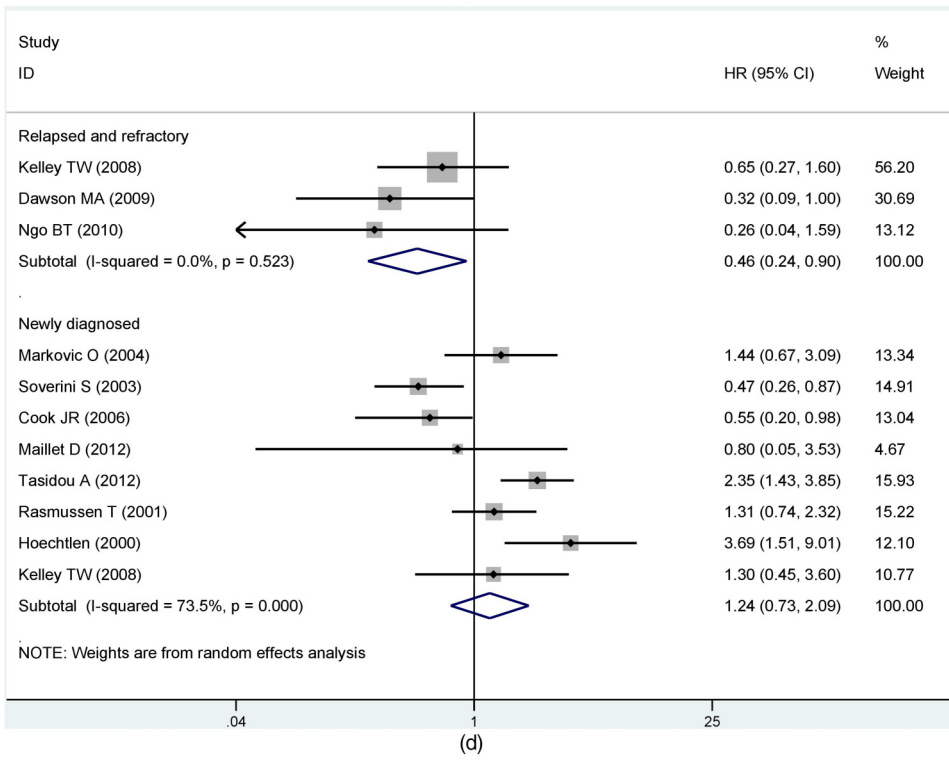
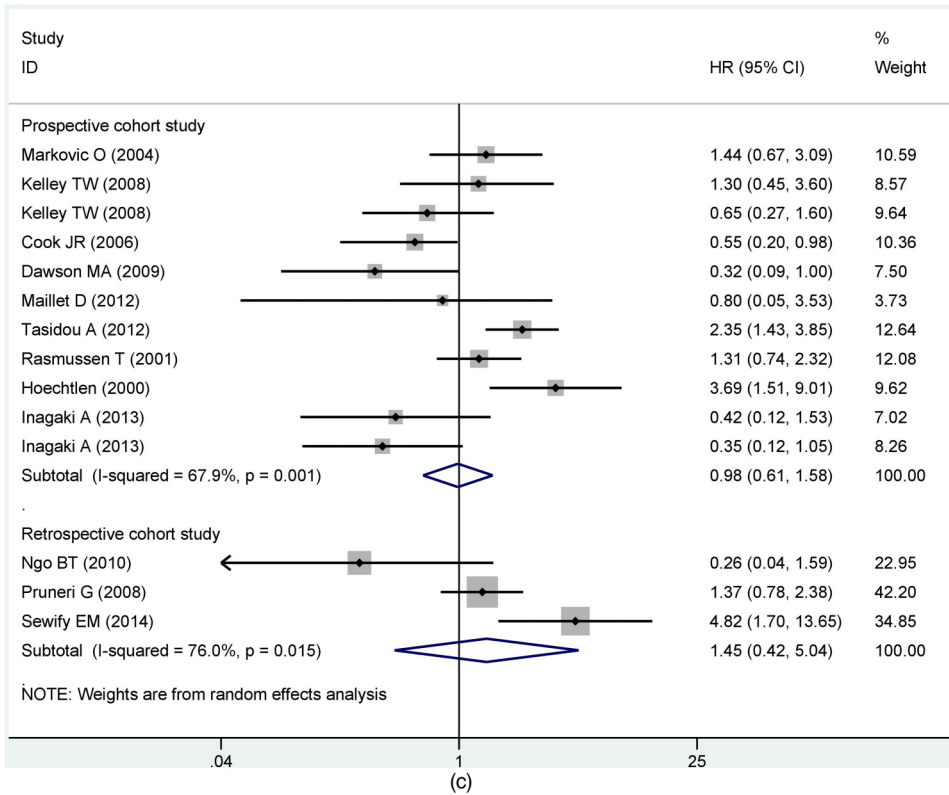
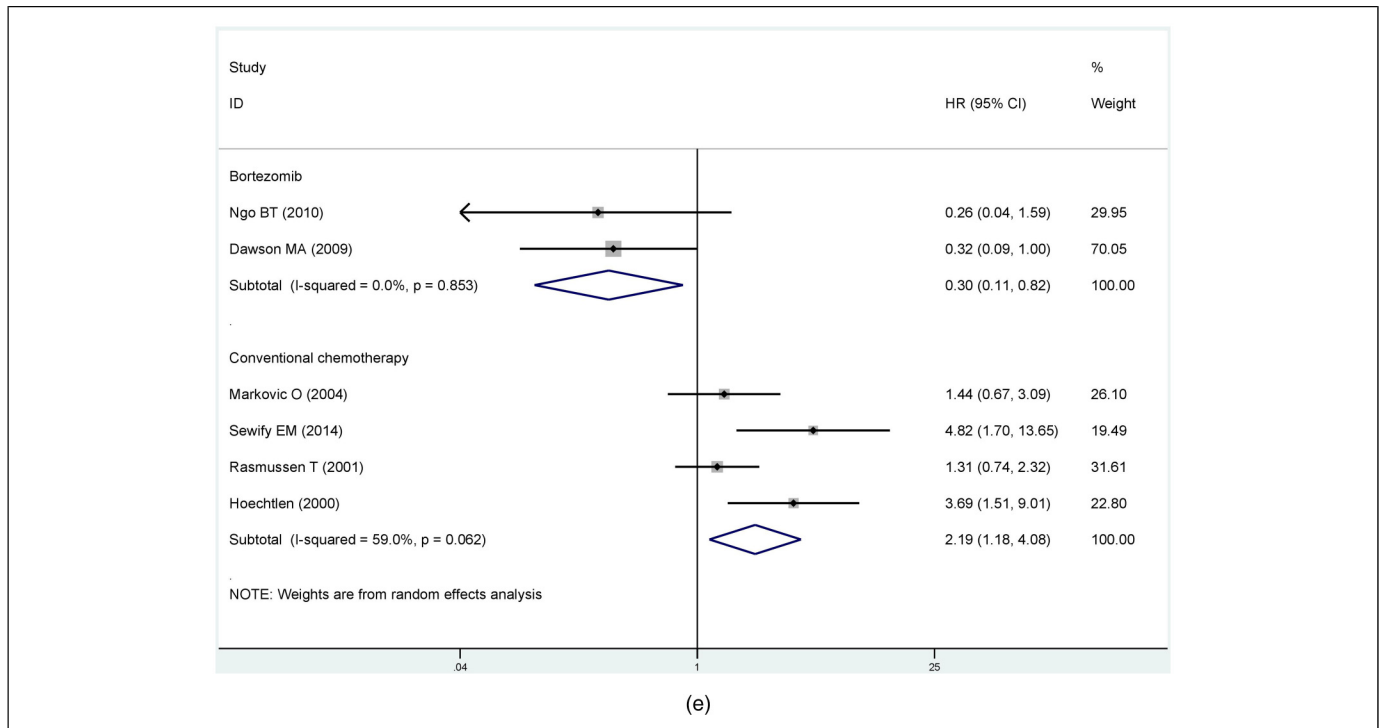


Figure 3. Continued.



studies was assessed by I^2 statistics. For $I^2 \geq 50\%$, a random-effects model was applied through inverse variance according to the Cochrane Collaboration guidelines.²¹ Otherwise, a fixed-effect model was adopted. We used Begg's and Egger's tests to find the symmetry on the funnel chart to evaluate the publication bias on *CCND1* expression.²² Results were considered statistically significant if the 95% CI did not include 1. All the P values exploited in this paper are bilateral, with $P < 0.05$ considered statistically significant. In the event of heterogeneity, we used the subgroup analysis method to analyze the potential factors.

Result

Search Result

Based on a previously determined retrieval strategy, the researchers retrieved 1238 records from the database of which 392 duplicate articles were excluded, and 95 full-text articles were retrieved by screening titles and abstracts. Of these 95 full-text articles, 82 were subsequently rejected because of insufficient data. In the end, 13 cohort studies that reported the relationship between cyclin D1 expression and survival of MM patients were included in our meta-analysis.^{16-19,23-31} The flow-chart of the specific literature search strategy is shown in Figure 1.

Research Characteristics and Quality Evaluation

The literature included in this meta-analysis was published from 2000 to 2014, with average follow-up periods between 11 and

36 months. Of the 13 studies evaluated, 10 were prospective cohort studies and 3 were retrospective cohort studies. Four studies directly provided HRs and 95% CIs; for the other 9 studies, we derived these values from the given survival curve. Most of the MM patients were newly diagnosed, and a few of them were in refractory and recurrent conditions. The clinical features of the selected studies are shown in Table 1. The quality of the articles was evaluated per the NOS standard: 3 were of high quality (8-9 points), and 10 were of medium quality (6-7 points). Overall, the quality of this meta-analysis was good.

The Relationship Between Expression of Cyclin D1 and Prognosis

The main results of pooled HRs in the meta-analysis were presented in Table 2. In general, there was a significant degree of heterogeneity between *CCND1* expression and prognosis (OS, $I^2 = 67.9\%$, $P = 0.000$; PFS/EFS, $I^2 = 85.8\%$, $P = 0.000$). Under the premise, the joint HRs of *CCND1* expression were calculated using the random effect. A comprehensive analysis revealed no significant correlation between *CCND1* expression and survival (OS, HR = 1.08, 95% CI: 0.71-1.64; PFS/EFS, HR = 0.97, 95% CI: 0.49-1.93) (Figure 2a and b), but the heterogeneity in two groups was significant.

Based on results with great heterogeneity, we categorized the OS group with sufficient sample data into subgroups and analyzed them. First, we performed subgroup analysis by detection of cyclin D1/*CCND1*. The results showed that differences between detection methods had no statistically significant

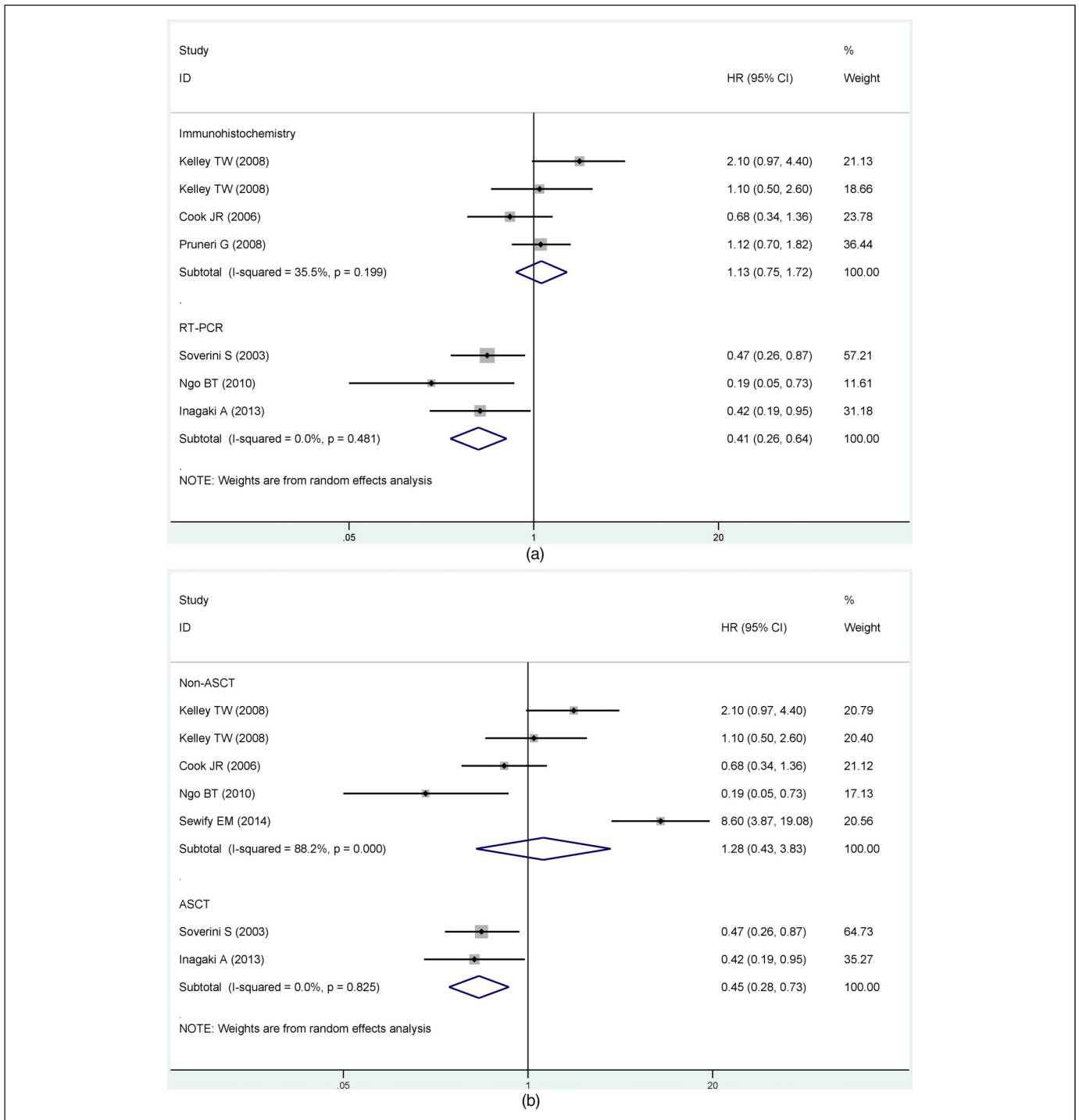


Figure 4. Subgroup analysis of PFS/EFS based on (a) different detection methods; (b) whether to perform ASCT; (c) cohort studies. Abbreviations: PFS/EFS, progression-free survival/event-free survival; ASCT, autologous stem cell transplantation. (continued)

effect on the prognosis (Immunohistochemistry, HR = 1.19, 95% CI: 0.74-1.90; RT-PCR, HR = 0.57, 95% CI: 0.24-1.35) (Figure 3a). Grouped immunohistochemistry analyses established no correlation between cyclin D1 expression and the prognosis of MM in difference of the critical value (10%, HR = 0.73, 95% CI: 0.17-3.17; 20%, HR = 1.69, 95% CI: 0.61-4.68; 30%, HR = 1.32, 95% CI: 0.77-2.27; 50%, HR =

0.87, 95% CI: 0.44-1.71). Obviously, there was no clear heterogeneity for cut-off values between 30% and 50% (30%, $I^2 = 0\%$, $P = 0.629$; 50%, $I^2 = 0\%$, $P = 0.321$). Conversely, heterogeneity was substantial for critical values between 10% and 20% (10%, $I^2 = 76.6\%$, $P = 0.039$; 20%, $I^2 = 83.5\%$, $P = 0.002$) (Figure 3b). Another subgroup assessment revealed less heterogeneity in prospective cohort studies than in

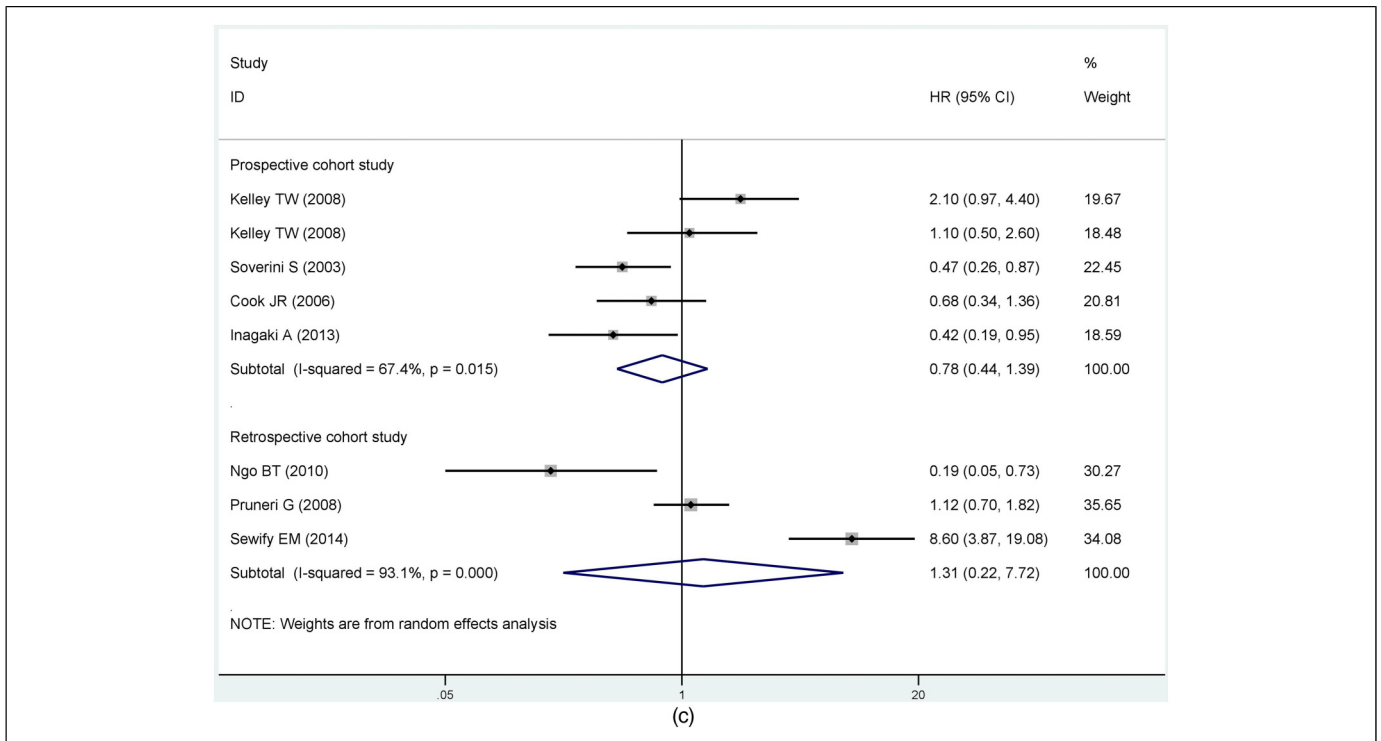


Figure 4. Continued.

retrospective studies (prospective cohort study, $I^2 = 67.9\%$, $P = 0.001$; retrospective cohort study, $I^2 = 76\%$, $P = 0.015$) although there was no significant association between *CCND1* expression and prognosis in both groups (prospective cohort study, HR = 0.98, 95% CI: 0.61-1.58; retrospective cohort study, HR = 1.45, 95% CI: 0.42-5.04) (Figure 3c).

To further explore the source of heterogeneity in the OS group, we conducted subgroup analysis based on the disease phases and treatment. Our findings based on different disease phases suggested that *CCND1* overexpression in the relapsed and refractory group was associated with longer survival periods for MM patients (OS, HR = 0.46, 95% CI: 0.24-0.90), and there was no significant heterogeneity in this group ($I^2 = 0.0\%$, $P = 0.523$). In the newly diagnosed group, cyclin D1 expression was not substantially linked to prognosis (OS, HR = 1.24, 95% CI: 0.73-2.09, $I^2 = 73.5\%$, $P = 0.000$) (Figure 3d). Per the results based on the mode of treatment, high cyclin D1 expression in the traditional chemotherapy group was linked to poor prognosis (OS, HR = 2.19, 95% CI: 1.18-4.08), and the heterogeneity in this group was considerably smaller than before ($I^2 = 59\%$, $P = 0.062$). In contrast, *CCND1* overexpression in the bortezomib group was related to better prognosis with no heterogeneity (OS, HR = 0.30, 95% CI: 0.11-0.82, $I^2 = 0.0\%$, $P = 0.853$) (Figure 3e).

Similarly, the PFS/EFS group was subdivided by different *CCND1* assays. The forest plot displayed that heterogeneity of the result was smaller (immunohistochemistry, $I^2 = 35.5\%$, $P = 0.199$; RT-PCR, $I^2 = 0.0\%$, $P = 0.481$) and MM patients with *CCND1* overexpression had a better prognosis in the RT-PCR group (immunohistochemistry, HR = 1.13, 95% CI:

0.75-1.72; RT-PCR, HR = 0.41, 95% CI: 0.26-0.64) (Figure 4a). Then we conducted subgroup analyses of the PFS/EFS group based on whether autologous stem cell transplantation (ASCT) was performed. We found that the prognosis of MM patients with *CCND1* overexpression in the ASCT group was better (HR = 0.45, 95% CI: 0.28-0.73) and there was no observable heterogeneity ($I^2 = 0\%$, $P = 0.825$). The non-ASCT group had no reference meaning because of its remarkable degree of heterogeneity (HR = 1.28, 95% CI: 0.43-3.83, $I^2 = 88.2\%$, $P = 0.000$) (Figure 4b). Results were moderately heterogeneous in the prospective cohort study and highly heterogeneous in the retrospective cohort study (prospective cohort study, $I^2 = 67.4\%$, $P = 0.015$; retrospective cohort study, $I^2 = 93.1\%$, $P = 0.000$), although no statistical significance existed in the results of both study types (Figure 4c).

Sensitivity Analysis and Publication Bias

In our sensitivity analysis, we eliminated the included studies one after another and merged HRs to examine the degree of difference between re-merged HRs and original changes (Figure 5). We found that re-evaluated HRs did not differ from those obtained from the first evaluation. As a result of the enormous heterogeneity among our meta-analyses, a random-effects model was chosen for statistical analysis. We used Egger's test and Begg's test in the STATA 16.0 software to evaluate publication bias. The assessment of publication bias in the study displayed that shape of the figure obtained was symmetrical. Egger's test and Begg's test

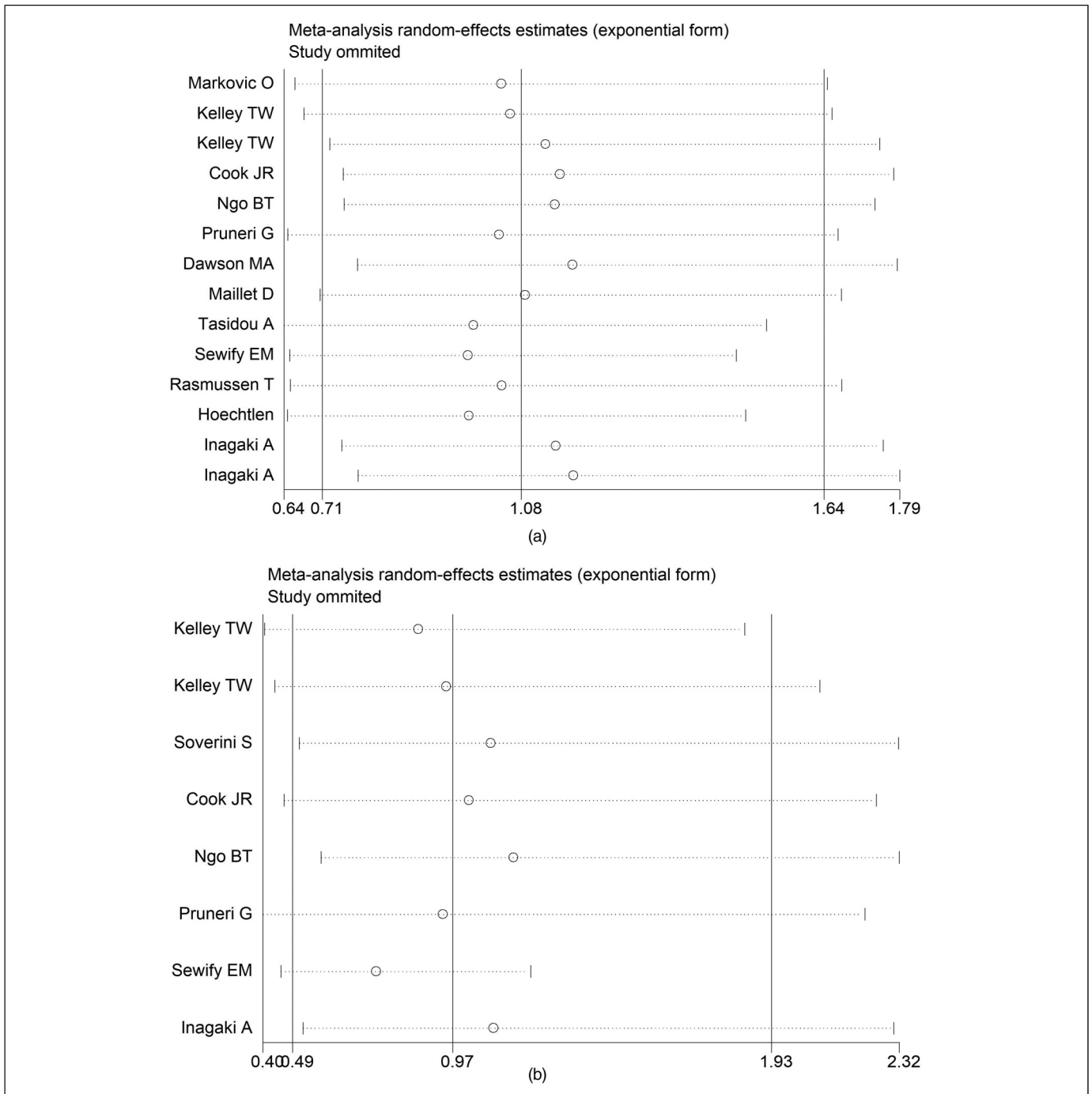


Figure 5. (a) Sensitivity analysis of OS; (b) Sensitivity analysis of PFS/EFS. Abbreviations: OS, overall survival; PFS/EFS, progression-free survival/event-free survival.

revealed no publication bias on the OS and PFS/EFS of patients, a nod to the robustness of our findings (Figure 6).

Discussion

Cyclin D1 is a member of the protein kinase family that regulates cell cycle progression, binding to cyclin-dependent

kinases (CDK4 or 6) to form a complex that produces activity.³² Cyclin D1 plays a key role in healthy cells’ processes through a series of phosphorylation and inactivation of tumor proteins in retinoblastoma.³³ It has been shown that the uncontrolled expression of cyclin D1 is an early and unified event in MM. Besides, *CCND1* overexpression of MM patients is possibly associated with t(11;14)(q13;q32) and polyploidy on chromosome 11.^{34,35} These findings

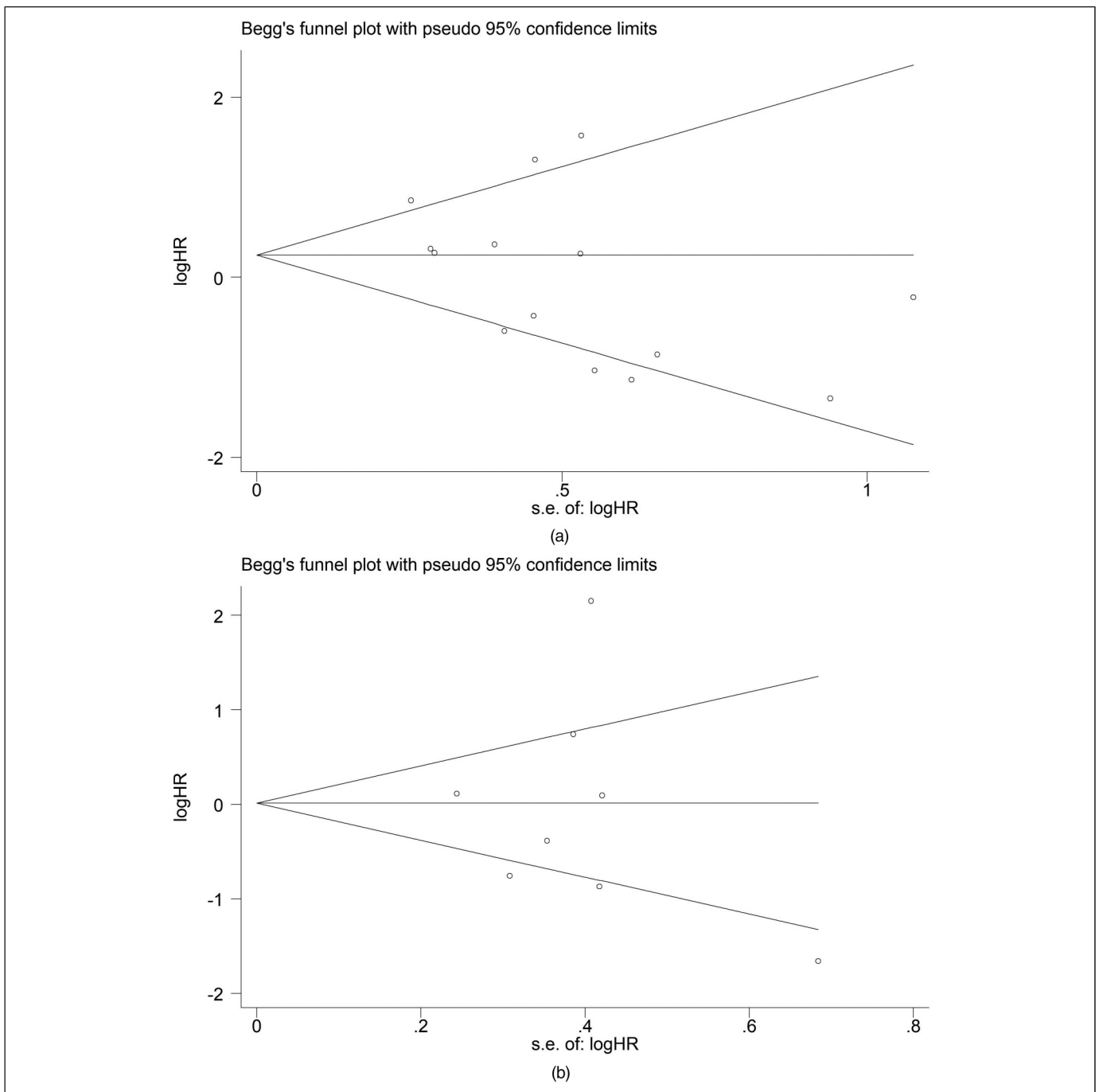


Figure 6. Begg's funnel plots for the meta-analyses of the association between cyclin D1 and (a) OS or (b) PFS/EFS of MM. Abbreviations: OS, overall survival; PFS/EFS, progression-free survival/event-free survival, MM, multiple myeloma.

suggested that hematologists could use cyclin D1 as a new prognostic indicator.

Recently, Tasidou et al¹⁷ followed up 130 patients with newly diagnosed MM for 22 months and noted that *CCND1* overexpression was associated with poor survival in MM patients. Relevant discoveries in other previous studies seemed to support the point Tasidou made. First, it was demonstrated that increased expression of cyclin D1 promotes tumor cell proliferation and interferes with the synergistic effect of

tumor suppressor genes on cell transformation.³⁶ Casimiro et al³⁷ then established that overexpression of the cyclin protein, which was linked to the proliferation and metastasis of MM cells, also affected the signal transduction process and expression of TSP-1. However, this conclusion has been questioned in recent years. Cook et al¹⁹ suggested *CCND1* overexpression benefited the prognosis of MM and increased the OS. Meanwhile, Pruneri et al²⁹ negated the above 2 inferences through a small sample study, revealing

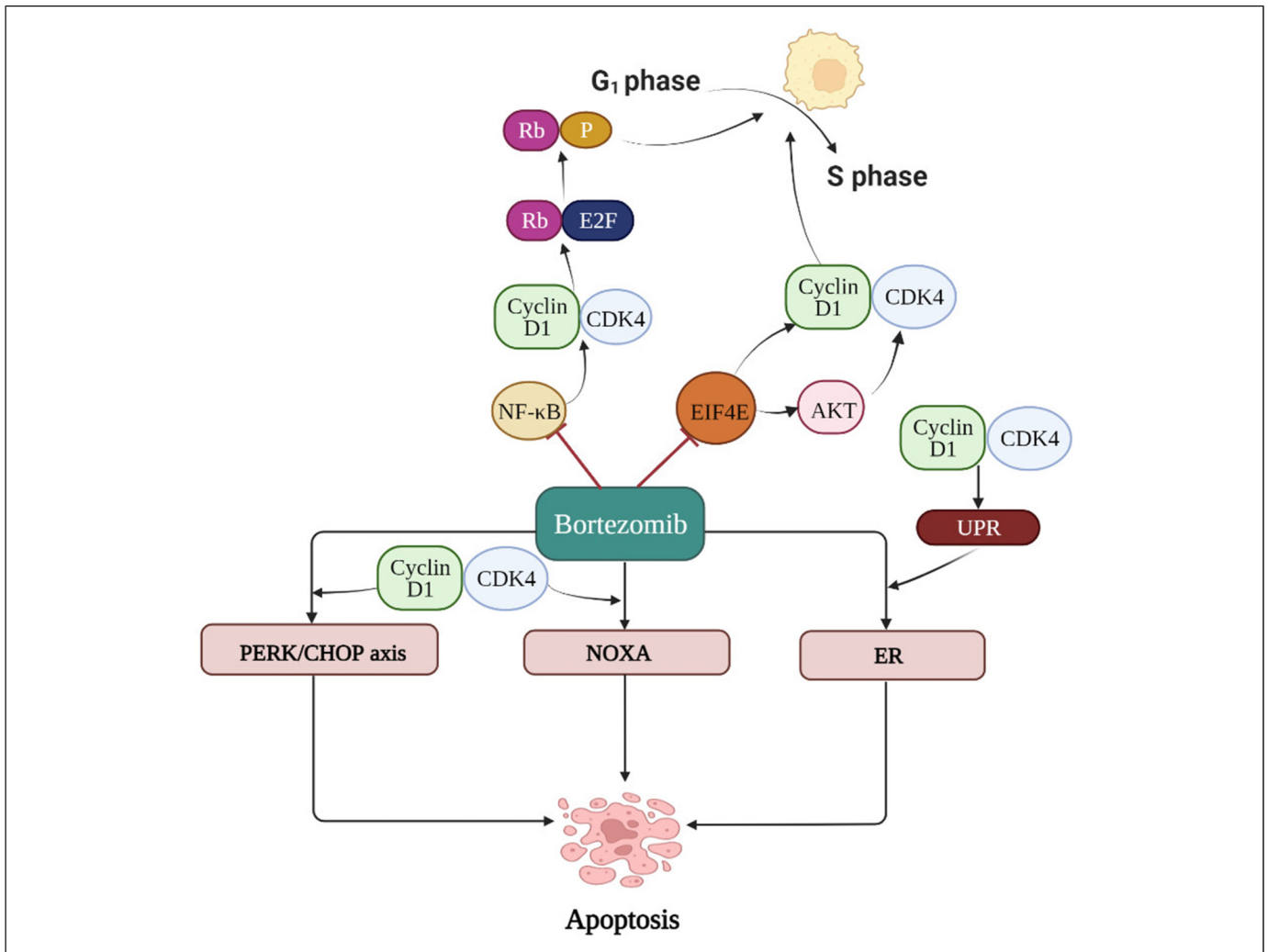


Figure 7. Mechanism diagram for interaction between bortezomib and cyclin D1 is displayed in the figure below. Bortezomib can regulate expression of cyclin D1 by inhibiting the activity of NF- κB and down-regulating expression of EIF4EmRNA to reduce expression of *CCND1*/mRNA. Cyclin D1/CDK4 activity plays an important role in the induction of NOXA protein, thus directly enhancing the anti-tumor effect of bortezomib. Besides, cyclin D1 can enhance the effect of bortezomib on PERK/CHOP axis and activate unfolded protein response (UPR) to induce the apoptosis pathway of bortezomib through endoplasmic reticulum (ER) stress.

Abbreviations: NF-κB, nuclear factor kappa-B; EIF4E, eukaryotic initiation factor 4E; NOXA, A BH3-only member of Bcl-2; AKT, protein kinase B; PERK, PKR-like ER kinase; CHOP, C/EBP-homologous protein; UPR, unfolded protein response; ER, endoplasmic reticulum; Rb, retinoblastoma gene; E2F, the E2F family of DNA-binding transcription factors; P, phosphoric acid.

rather that there was no significant correlation between the *CCND1* expression and survival time of MM patients. In short, opinions on the relationship between *CCND1* expression and the prognosis of MM differ among investigators. Now *CCND1* expression in the prognosis of MM remains uncertain, so we conducted a detailed meta-analysis to provide a statistical basis for the prognostic value of cyclin D1 expression in MM patients.

In general, our meta-analysis established no significant correlation between *CCND1* overexpression and the prognosis of MM. However, bearing in mind the heterogeneity of results, we performed subgroup analyses of the OS group and found that MM patients with *CCND1* overexpression tend to have a poor prognosis when treated with traditional chemotherapy,

which validates the findings of Sewify et al.¹⁶ In contrast, high expression of cyclin D1 acted as a favorable prognostic factor for MM patients treated with bortezomib. The possible mechanisms supporting our results are as follows: Combining *CCND1* with cyclin kinase CDK4 phosphorylates the Rb protein. The phosphorylated Rb protein is dissociated from the E2F transcription factor, and the E2F transcription factor starts to transcribe the genes that activate the cell cycle, driving the cell cycle from the G₁ phase to the S phase.³⁸ NF-κB, an important nuclear factor in the pathogenesis of MM, tends to promote *CCND1* expression. Bortezomib, now widely used to treat MM, can regulate *CCND1* expression by inhibiting the activity of NF-κB, subsequently obstructing cell proliferation.³⁹ Furthermore, studies have shown that

bortezomib down-regulates EIF4E mRNA expression to reduce *CCND1* mRNA levels, resulting in the arrest of MM cells at the G1/S phase. Therefore, MM patients with *CCND1* overexpression could be more sensitive to bortezomib and have a better prognosis.⁴⁰ The proteasome inhibitor, bortezomib, mediates cell death by facilitating the stability of the NOXA protein, one of the pro-apoptotic members of the Bcl-2 family. The NOXA protein requires the *CCND1*/CDK4 activity for effective accumulation, with this accumulation increasing the sensitivity of tumor cells to bortezomib. There is evidence that inhibiting the activity of *CCND1*/CDK4 reduces the degree of induction of the NOXA protein by bortezomib.⁴¹ Remarkably, bortezomib can mediate antitumor effects via CHOP transcription or by increasing the PERK activity, whereas *CCND1* expression can enhance the effect of bortezomib on the PERK/CHOP axis. At the same time, *CCND1* can also activate the unfolded protein response (UPR) to induce bortezomib's apoptosis pathway mediated by endoplasmic reticulum (ER) stress.⁴² The mechanisms that reflect the relationship between bortezomib and cyclin D1 are shown in Figure 7.

Remarkably, MM patients with *CCND1* overexpression had better OS in the subgroup with relapsed and refractory recurrence, possibly owing to the high utilization rate of bortezomib. We also found that MM patients with high *CCND1* levels in the ASCT group was more likely to have a longer PFS than individuals in the non-ASCT group. Based on Soverini's research, this better survival tended to be associated with transplantation itself and had no obvious correlation with treatment before and after ASCT.¹⁸

In particular, our subgroup analysis showed that results were less heterogeneous when *CCND1* was detected by RT-PCR, the immunohistochemical threshold above 30%, or prospective cohort study methods were used. Therefore, a more than 30% immunohistochemical threshold can be selected as a positive result to reduce heterogeneity. Additionally, more samples and higher-quality prospective studies can be performed to further elucidate the relationship between cyclin D1 expression and MM prognosis.

Despite our endeavor to avoid errors and improve the credibility of our statistical results, this investigation still had limitations: (1) The total number of subjects included in the study was not large enough, meaning that the assessment of MM patients was not representative enough. (2) There was no single defined method for determining *CCND1* expression, including IHC, RT-PCR, and FISH. (3) A subset of data such as follow-up time and mode of treatment, were not mentioned in certain cases, resulting in the incompleteness of the subgroups. (4) The patients' age, nationality and research methods differed and resulted in the heterogeneity of results.

Nevertheless, this is the first meta-analysis to assess the prognostic value of cyclin D1 expression in MM. As a potential prognostic factor, cyclin D1 provides a new direction to evaluate the survival status of MM patients. On the basis of the qualitative analysis of the included articles, we offer the following recommendations for future cohort studies. First, attention should be paid to the combined impact of genetics and

environmental factors on prognosis. Second, more standardized and automated procedures could be developed for specimen collection and detection. Furthermore, prospective studies with greater sample sizes and longer follow-up periods must be scrutinized.

Conclusion

Our meta-analysis showed that cyclin D1 may deserve as a valuable prognostic biomarker for MM. MM patients overexpressing *CCND1* treated with bortezomib or receiving ASCT are more likely to have better prognoses. Besides, the increased expression of *CCND1* in relapsed and refractory MM patients probably be associated with survival benefits.

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Authors' Contributions

YWJ designed the research study. YWJ, CLZ, and LL collected data and performed data analysis. YWJ drafted the manuscript and designed the figures. HYL, YJJ, and LMH contributed to the editing of the final manuscript. HMH and DG reviewed the manuscript. All authors checked the full text carefully and approved the final manuscript.

Declaration of Conflicting Interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.


Ethical Statement

Our study did not require an ethical board approval because it did not contain human or animal trials.

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