Management of multiple myeloma in the era of novel drugs

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Multiple Myeloma (MM) is a malignancy of terminally differentiated plasma cells and is the second most common hematological neoplasm to Non-Hodgkin's Lymphoma. Generally, it is disease of older patients. Our knowledge about the underlying biological and cytogenetic abnormalities leading to MM is rapidly increasing. Similarly our ability is improving to treat this complex disease. A number of new treatments have been introduced into our armamentarium in the past 10-15 years. Until recently, high rates of complete responses (CR) and other major responses were seen only in patients undergoing treatment with high dose chemotherapy with autologous stem cell support (HD+ASCT). However new regimens, incorporating new agents (thalidomide, lenalidomide, bortezomib) are now offering similar response rates and lower toxicity than HD+ASCT. The new agents seem to combine well with classical chemotherapy agents (melphalan, cyclophosphamide), modern chemotherapy (pegylated liposomal doxorubicin) and steroids (dexamethasone, prednisone). In addition, the novel agents show significant activity when combined with each other in patients with newly diagnosed as well as relapsed/refractory MM patients. Although this is still considered an incurable disease, the life expectancy and quality of life of MM patients is continuously improving. Our hope is that progress in this area of research will continue with the advent of new treatment options and will lead to the ultimate goal: a cure.

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Introduction

Multiple myeloma (MM) results from the clonal proliferation of plasma cells arising in the lymph nodes and "homing" to the bone marrow where these cells localize and proliferate. It represents the second most common hematological malignancy. Multiple myeloma is a neoplastic disorder of plasma cells that accounts for 10% of all hematologic cancers in Caucasians and 20% in African Americans (1). Annually, this malignant disease causes over 19,000 deaths in Europe. Approximately 19,920 new cases of MM

were diagnosed in the United States in 2008, with 10,690 deaths, representing almost 2% of all cancer deaths (2). The median survival of patients with multiple myeloma is 3 to 5 years (3). Persons affected by MM are often elderly, with a median age at diagnosis of 65 years; 80% of patients are older than 60 years and less than 3% are younger than 40 years (4, 5). The disease is twice as common in African Americans as in Caucasians. MM is one of the leading causes of cancer death in African Americans. MM is one of three cancers that show increased mortality rates for both men and women in the 1990's (5.6% and 3.8%, respectively) (6).

The disease is characterized by overproduction of a patient-specific intact monoclonal immunoglobulin (Ig) heavy and/or light chain (paraprotein or M-protein). IgG is detected in about 53% of MM cases and IgA in about 25%; 40% of these IgG and IgA patients also have Bence Jones proteinuria. Light chain MM is found in 15 to 20% of patients; their plasma cells secrete only free monoclonal light chains (κ or λ) that can be detected by the Serum Freelite Assay or as Bence Jones proteins; serum M-components are usually absent on electrophoresis. IgD MM accounts for about 1% of cases. IgM and IgE as well as non-secretory MM are rare (4, 7, 8). Aside from the serum and urine M-proteins, other features in MM patients include anemia (80% of patients), bone pain (70% of patients) due to lytic lesions, renal dysfunction (25% of patients), hypercalcemia, increased susceptibility to infection, and constitutional symptoms (4). Other less common complications include spinal cord compression due to medullary and extramedullary plasmacytomas or vertebral collapse, peripheral neuropathy, amyloidosis, and hyperviscosity syndrome (4, 7, 8).

Durie and Salmon developed a clinical staging system for MM based on a combination of factors that correlate with myeloma cell mass (9). Most patients (40 - 60%) present with advanced (Stage III) disease (4). Other alternative staging systems have been proposed (10). Recently, an International Staging System using two simple blood tests (beta-2-microglobulin and albumin levels) was developed, based on data from 11,171 patients (11). These staging methods do not take into account newer diagnostic studies such as Serum Freelite Assay, cytogenetics of focal lesions versus bone marrow, or gene expression profiling (GEP), all of which could have potential prognostic significance (12-15). A high plasma cell labeling index (PCLI) and elevated B2-microglobulin predict poor prognosis in an untreated patient, but may not be as important in a previously treated patient. If both markers are low, the median survival is about six years in untreated patients. The correlation between response and survival has been evaluated in multiple studies. Although there is no absolute agreement among all, it seems that achieving complete response (CR) or very good partial response (VGPR) are important prognostic factors for long-term survival (16-20). In the South Western Oncology Group (SWOG) large retrospective analysis of 1,555 MM patients treated with standard-dose chemotherapy, the magnitude of response, as a single variable, did not predict survival duration. However, the best indicator of survival was time to first progression (21). Important prognostic variables for response to therapy and survival include the patient's age, stage, immunoglobulin type, ß-2-microglobulin level, PCLI, and the presence of circulating plasma cells. A poor outcome is associated with chromosome 13 deletion or hypodiploidy on conventional karyotyping, deletion of 17p – or immunoglobulin heavy chain translocation t (4:14) or t (14:16) on molecular genetic studies and plasma cell labeling index of 3% or higher (15, 22).

MM is almost always preceded by monoclonal gammopathy of undetermined significance (MGUS), an asymptomatic phase

characterized by a relatively small burden of clonal cells and low levels of monoclonal protein (23). The diagnosis of active symptomatic MM requiring therapy should be based on the end-organ effects of the disease (elevation of ***C***alcium, ***R***enal insufficiency, ***A***nemia or ***B***one disease).

Therapeutic approach

The role of high-dose therapy and peripheral stem cell transplant (HD+ASCT) continues to be controversial, with overall survival (OS) only minimally improved if any (24, 25). In a recent review and meta-analysis of 9 randomized controlled studies involving 2,411 patients, single HD+ASCT was compared with conventional chemotherapy and was found to benefit progression free survival (PFS), but not OS (26). The risk of treatment-related mortality (TRM) was increased three-fold with HD+ASCT. It is worth mentioning that in this analysis HD+ASCT was compared with classical chemotherapy, not new agents or their combinations. Patients with progressive disease can achieve a 50-75% response rate to salvage regimens such as vincristine, doxorubicin, and dexamethasone (VAD) (27, 28); however, these responses are often short-lived. Even in transplant-eligible and willing patients initial therapy has undergone a sea change in the past decade (29). Before the advent of drugs such as thalidomide and lenalidomide (IMiDs) or bortezomib, single agent dexamethasone (dex) and VAD were the most commonly used treatments (28,30,31). From 2005 to 2009 numerous Phase 2 and 3 trials compared different combinations of drugs used as induction therapies prior to HD+ASCT (32). These clinical trials showed the clear advantage of newer agents over classical chemotherapy in improving PFS and response rates, but a mixed picture regarding OS, particularly in clinical trials incorporating high dose melphalan as a consolidated approach (33-35). The first study ever to show the survival advantage of a chemotherapy combination over HD+ASCT was a French study (IFM 99-06) using a combination of a new agent (thalidomide) with classical melphalan and prednisone (MTD), published in 2007 by Thierri Facon and collaborators (36). In this study MM patients treated with MTD had longer OS when compared with Mel-100 HD+ASCT. It is worth mentioning that Mel-100 HD+ASCT is not considered standard care for MM patients by most transplant physicians and could be inferior to standard Mel-200. Despite this caveat, IFM 99-06 opened the door for very intensive investigation into the role of new agents as a primary treatment or cytoreductive therapy prior to HD+ASCT.

Proteosome inhibitors

The ubiquitin-proteasome pathway is the principal pathway for intracellular protein degradation (37, 39) (Figure 1).

This pathway selectively degrades an extensive number of short-lived regulatory proteins involved in the control of normal cellular processes. In order to be degraded, proteins targeted by the ubiquitin-proteasome pathway are covalently tagged by polyubiquitination, via a three-step enzymatic process, which ultimately leads to their recognition and degradation, by the 26S proteasome in a highly specific and regulated manner. This process is accomplished by the sequential action of three enzymes: an ATP-dependent ubiquitin-activating enzyme (E1), an ubiquitin-conjugating enzyme (E2) and an ubiquitin-protein ligase (E3) (39). This cascade covalently links the C terminus of ubiquitin to a free amino group on the target protein, usually the ε-amino of a lysine residue.

The 26S proteasome is comprised of a catalytic proteolytic core (20S) and an activator (19S) (Figure 2).

It plays a vital role in degrading regulatory proteins that govern many signaling pathways, including the cell cycle, transcription factor activation, apoptosis, and pathways that regulate the expression of proteins, which direct angiogenesis, cell trafficking,

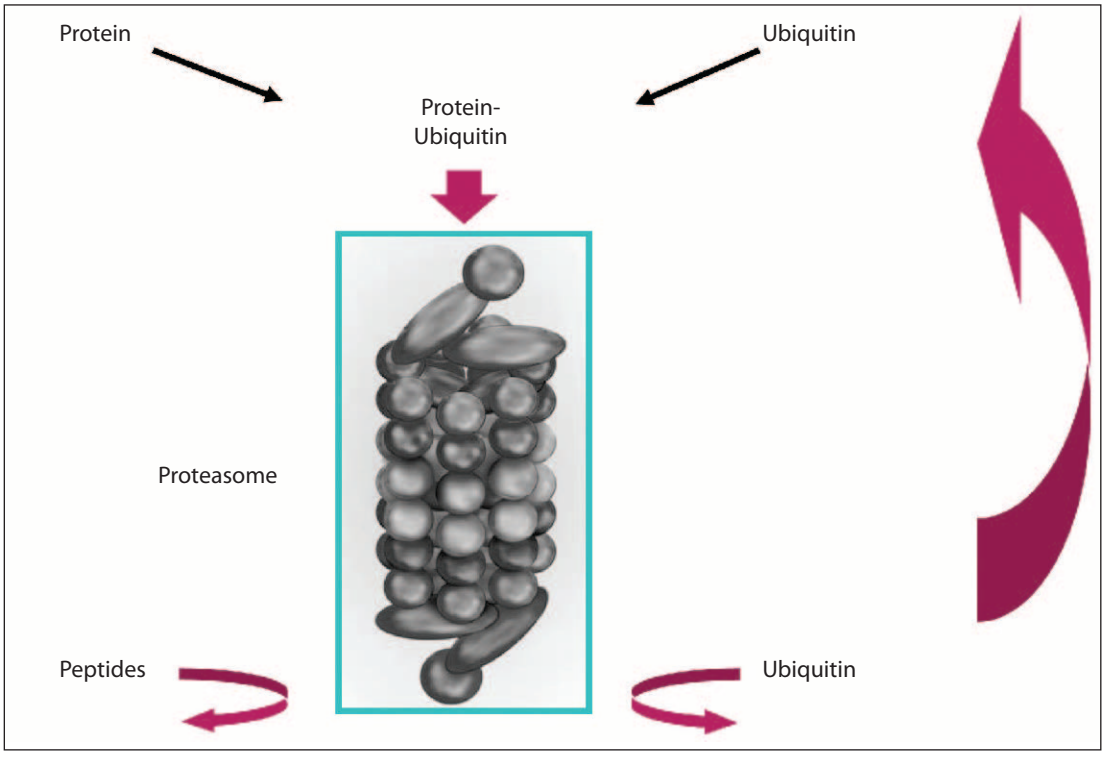

Figure 1 The Ubiquitin-Proteasome degradation pathway

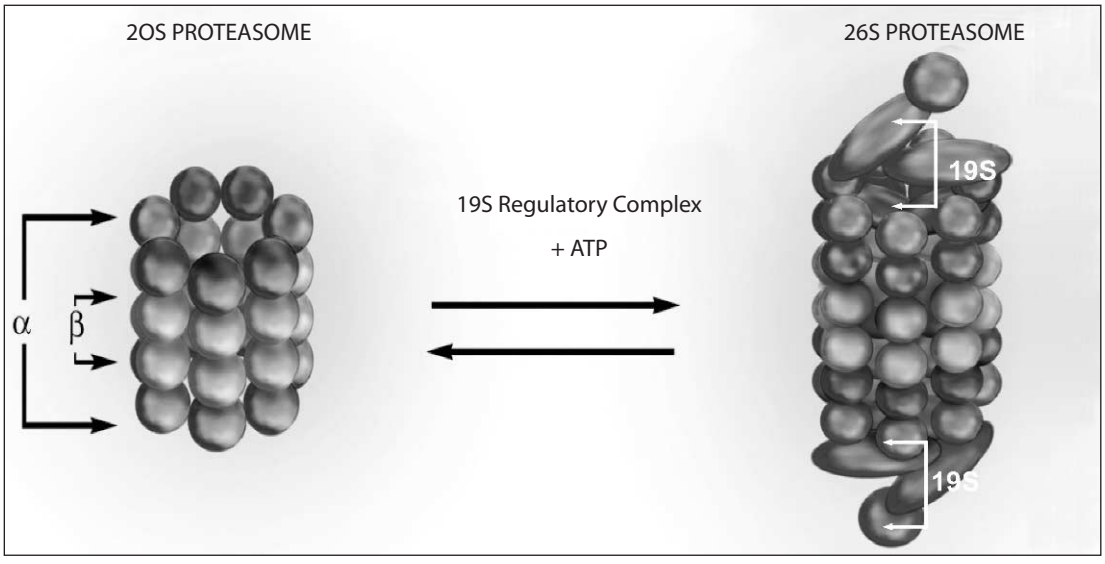

Figure 2 The structure of the proteasome. The 20S core (left) is capped by two 19S regulatory units to form the 26 proteasome (right)

and metastasis (38, 40). Polyubiquinated proteins cannot be degraded directly by the active catalytic proteolytic core (20S). Rather, proteolysis requires another protein, known alternatively as PA700, ball, 19 S cap or μ-particle (39). It is the 700,000 dalton, 20-subunit complex that binds to one or both of the terminal rings of the proteasome in a cooperative manner. This integral role of the 26S proteasome in cellular signal transduction has provided a new target for exploring the therapeutic potential of proteasome inhibition in neoplastic diseases. It is known that several key regulatory proteins relevant to cancer initiation and progression are known to be temporally degraded during the cell cycle by the ubiquitin-proteasome pathway. Ordered ubiquitination and degradation of regulatory proteins is required for the cell to progress through the cell cycle, undergo mitosis and proliferate. Similarly, the proper function of specific ubiquitin ligases responsible for the ubiquitination of these same proteins is required for key cell cycle transitions. Aberrant degradation of cell cycle control proteins can result in accelerated and uncontrolled cell division, thereby promoting cancer growth. Recent evidence from studies reveals that expression of the ubiquitin-proteasome pathway enzymes is elevated in tumor samples. The cyclin and the cyclin-dependent kinase inhibitors p21 cip1 and p27 kip1 are an example of growth regulatory proteins degraded by proteasome-dependent proteolysis (41, 42). Both p21 cip1 and p27 kip1 can induce cell cycle arrest through functional inhibition of cyclin D-, E-, and A-dependent kinases (42). In addition, the p53 tumor suppressor required for cell cycle control and initiation of apoptosis induced by cellular damage, including ionizing radiation and chemotherapy, is also a substrate of the the ubiquitin-proteasome pathway (42, 43). Hence, proteasome inhibition has the potential to arrest the cell cycle in cancer cells through the disruption of a large number of growth regulatory pathways.

The ubiquitin-proteasome pathway also plays an important role in the regulation of many transcriptional responses. On the other hand, proteasome function in the cell can be regulated by altering levels of proteasome, proteasome regulatory proteins, or proteins of the ubiquitin conjugation system.

The relationship between proteasome function, gene transcription and potential cancer therapy is best understood for the transcription factor nuclear factor-kappa B (NF-κB) (Figure 3).

NF-κB activation is regulated by 26S proteasome-mediated degradation of the inhibitor protein I-κB (44, 45). NF-κB activation is integral to many aspects of tumorigenesis, such as tissue invasion and metastasis, angiogenesis, evasion of apoptosis, cell growth, and survival (46). Activation of NF-κB can proceed through multiple mechanisms, including autocrine or paracrine extracellular cytokine signaling, upstream oncogenic signaling mutations in NF-κB and/or I-κB, and in response to DNA damage. Cell adhesion molecules such as E-selectin, ICAM-1, and VCAM-1, as well as IL-8, vascular endothelial growth factor (VEGF), and matrix metalloproteinases (MMPs) are regulated by NF-κB and have been implicated in tumor metastasis and angiogenesis in vivo (45, 47). Furthermore, NF- κB is required in numerous cell types to maintain and control cell viability via the production of anti-apoptotic survival proteins such as cellular inhibitors of apoptosis (cIAPs), and the B-cell lymphoma-2 (Bcl-2) family of proteins. NF-κB also plays a role in cell proliferation by activating target genes of the cell cycle such as D1-cyclin, and growth factors such as interleukin-6 (IL-6) (46). It has been demonstrated that blocking NF-κB activation by stabilizing its inhibitor, I-κB, sensitizes cells to environmental stressors and cytotoxic agents, ultimately leading to apoptosis (48,

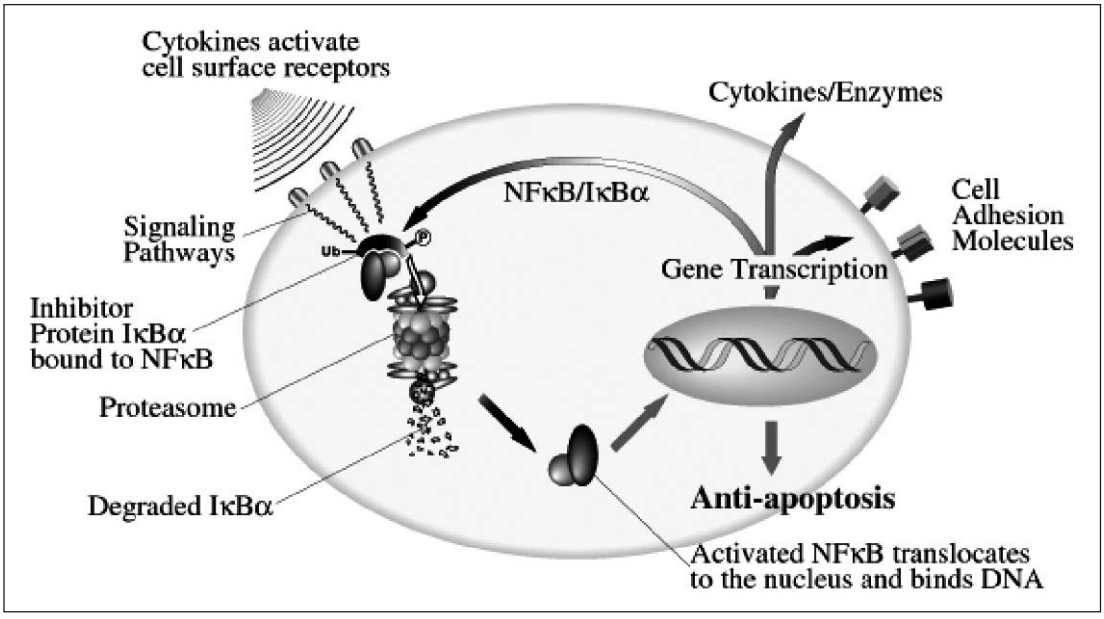

Figure 3 Nuclear factor κB (NF-κB) and its role in the cell.

49). Hence, regulation of NF-κB dependent transcriptional regulation and activation through proteasome inhibition can impact several cancer virulence mechanisms.

Anti tumor activity of proteasome inhibitors

A number of investigators have shown that inhibitors of proteasome, including aldehydes and lactacystin, are growth inhibitory and cytotoxic for cells in culture (50). Boronate proteasome inhibitors have been shown to kill tumor cells in culture as demonstrated in NCI tumor cell line screen (37). The NCI utilizes an *in vitro* screen comprised of 60 human tumor cell lines derived from 9 different cancer types (leukemia, lung, brain, colon, melanoma, ovarian, prostate, renal and breast). Data from the NCI screen showed that proteasome inhibitors have a mechanism of cytotoxicity unlike any other compound in the NCI database of 60000 compounds. Among the large number of proteasome inhibitors, bortezomib was selected for intensive study based on its selectivity and chemical and biological characteristics (37). Bortezomib specifically, selectively and reversibly inhibits the proteasome by tightly binding to the chymotrypsin-like site of the 20S core of the enzyme. By inhibiting a single molecular target, the 26S proteasome, bortezomib has the potential to affect multiple signaling pathways. The anti-neoplastic effects of bortezomib likely involve several distinct cell regulatory mechanisms as discussed above, including inhibition of cell growth and survival pathways, induction of apoptosis, and inhibition of gene expression integral to cellular adhesion, migration, and angiogenesis. Thus, the mechanisms by which bortezomib elicits its anti-tumor activity may vary among tumor types, and the extent to which each affected pathway is critical to the inhibition of tumor growth could also differ.

It has been demonstrated that bortezomib has a novel pattern of cytotoxicity in NCI in vitro and in vivo assays (37) and displays cytotoxic activity in a variety of xenograft tumor models, both as a single agent

and in combination with other chemotherapeutic agents and radiation (37, 51-55).

Numerous published reports show that cancer cells are more sensitive to the pro-apoptotic effects of proteasome inhibition than nontransformed cells (56-62). For example, the toxicity of bortezomib for multiple myeloma (MM) cells was more than 100-fold greater when compared to peripheral blood leukocytes and normal hematopoietic cells (56). Bortezomib has shown direct cytotoxic activity against a variety of MM cell lines and in freshly isolated cells from patients (56, 57). Significantly, these studies have included myeloma cells that are highly resistant to other chemotherapeutic agents. Time-dependent exposure to bortezomib programs MM cells to commit to apoptosis (63). It was shown that this drug directly inhibits proliferation and induces apoptosis of human MM cell lines and freshly isolated patient MM cells, inhibits mitogen-activated protein kinase growth signaling in MM cells, induces apoptosis despite induction of p21 and p27 in both p53 wild-type and p53 mutant MM cells, overcomes drug resistance, adds to the anti-MM activity of dex and overcomes the resistance to apoptosis in MM cells conferred by Interleukine-6 (IL-6) (63). Bortezomib also inhibits the paracrine growth of human MM cells by decreasing their adherence to bone marrow stromal cells and related NF-κB-dependent induction of IL-6 secretion in bone marrow stromal cells, as well as inhibiting proliferation and growth signaling of residual adherent MM cells.

Clinical studies with bortezomib

Bortezomib (Velcade®) is the first proteosome inhibitor approved by FDA for clinical use. Initially, in 2003 it was approved as a third line therapy for MM patients. The present indication is for second line MM as a single agent, first line MM in combination with melphalan-prednisone (MPV) and for patients with relapsed mantle cell lymphoma.

In the fall of 1998, the first human trial with bortezomib was initiated at M.D. Anderson Cancer Center in Houston, Texas. In May 2003 bortezomib was approved by FDA as Velcade for Injection for the treatment of MM patients who had received at least 2 prior therapies and had demonstrated disease progression on the last therapy. Conditional approval was based mostly on the results of the Phase II study of bortezomib in patients with relapsed, refractory MM (SUMMIT) (64). In this study 202 patients were enrolled and 193 could be evaluated. Most (84%) had IgG or IgA MM and advanced disease at diagnosis. Eighty percent had symptoms of peripheral neuropathy at enrollment. Of the 193 patients, 178 (92%) had previously been treated with three or more of the major classes of agents for myeloma. Patients received 1.3 mg of bortezomib per square meter of body-surface area twice weekly for 2 weeks, followed by 1 week without treatment, for up to 8 cycles (24 weeks). In patients with a suboptimal response, oral dexamethasone (20 mg daily on the day of and the day after bortezomib administration) was added to the regimen. Of the 193 patients with measurable disease, 67 (35%) had CR, PR or minimal response (MR) to bortezomib alone. Nineteen patients had CR or near-complete response (NCR). This was first study showing significant complete responses to a single, non-chemotherapeutic antimyeloma agent. The median time to a first response was 1.3 months. The median time to progression of disease among all 202 patients while they were receiving bortezomib alone was 7 months, as compared with 3 months during the last treatment before enrollment. According to a landmark analysis, achievement of CR or PR to bortezomib alone after 2 cycles was associated with significantly longer survival than in other patients ($p = 0.007$). The most common

adverse events (AE) were gastrointestinal symptoms, fatigue, thrombocytopenia and sensory neuropathy. Drug related AE led to discontinuation of bortezomib in 36 patients (18%). The most clinically significant AE was cumulative, dose-related peripheral sensory neuropathy. Overall incidence of clinically relevant neuropathy (Grade 3) was 12 percent. However, complete resolution or improvement of peripheral neuropathy was observed in the majority of patients during the follow-up period. A second study (CREST) compared two different dosages of bortezomib (1 mg/m^2 vs. 1.3 mg/m^2) in relapsed/refractory MM patients. Responses were 33% and 50%, respectively. Median time to progression (TTP) was 10 and 10.9 months (65). Addition of dexamethasone to bortezomib in patients who failed to respond or who relapsed after treatment with bortezomib alone improved responses in 18% of patients in SUMMIT and 33% in CREST trial.

An international, randomized, multicentar phase 3 trial comparing bortezomib with high-dose dexamethasone (APEX) enrolled 669 patients with relapsed/refractory MM (66). Patients treated with bortezomib had significantly higher response rates (38% vs. 18%) and CR (6% vs. < 1%), longer TTP (6.2 vs. 3.5 months) than patients treated with dexamethasone. The one-year OS rate was 80% among bortezomib treated patients and 66% for dexamethasone treated ones (p = 0.003) with hazard ratio (HR) of 0.57. A recent update with extended follow up (22 months) showed again superior OS and overall response rates (ORR) as well as CR in the bortezomib treated group, despite substantial crossover from dexamethasone to bortezomib (67).

Bortezomib is now very successfully combined with other effective therapies for MM. In an international randomized Phase III study Orlowski and collaborators compared a combination of pegylated liposomal doxorubicin and bortezomib with single agent bortezomib in 646 MM patients with relapsed/refractory disease (68). Combination therapy was associated with longer TTP (9.3 vs. 6.5 months, p = 0.000004) and longer 15-months OS (76% vs. 65%, p = 0.03) when compared to bortezomib alone. Grade 3/4 AE were more frequent in the combination arm (80% vs. 64%). The most common side effects were neutropenia, thrombocytopenia asthenia, fatigue, diarrhea and hand-foot syndrome. The significance of this unique drug combination is that steroids were not part of the therapy, allowing use by the treating physician in older patients with intolerance to steroids as well as in hard to control diabetics.

Combination of bortezomib with melphalan and prednisone (VMP) was compared in 682 MM patients with newly diagnosed disease with classical MP in a randomized Phase III clinical trial VISTA (69). Patients treated with VMP had significantly longer TTP (24 vs. 16.6 months; p< 0.001) and median duration of response (DOR) (19.9 vs. 13.1 months). After a median follow-up of 16.3 months, 13% of patients in the VMP and 22% in the MP group had died (HR = 0.61; p = 0.008). At the time of publication, median OS had not been reached in either group. Grade 3 AE occurred in a higher proportion of patients in the VMP than in the MP group (53% vs. 44%; p = 0.02), but there were no significant differences in grade 4 events (285 vs. 27%, respectively) or treatment-related deaths (1% and 2%). At present, VMP could be considered the standard of care therapy for newly diagnosed MM patients who are not candidates for or are refusing HD+ASCT. However, some myeloma specialists would consider other MP combinations, such as one with thalidomide (MPT) or lenalidomide (MPR) as a possible standard of care.

Tables 1 and 2 summarize response rates in selected Phase II and III studies using bortezomibe as a single agent or in combination in newly diagnosed and relapsed/refractory MM patients.

Table 1 Bortezomib combinations in newly diagnosed MM patients

Regimen	Phase	N	PR	CR	Reference
+ melphalan + prednisone	III (VISTA)	344	+ CR 71	30%	San Miguel et al. (69)
+ thalidomide + prednisone	III	128	+ CR 79%	27%	Mateos et al.(117)
+ melphalan + prednisone + thalidomide With maintenance	III	254	+ CR 86%	34%	Palumbo et al. (118)
+ Cytoxan® + dex	II/III	400	+ CR 84%	10%	Einsele et al. (119)

N = number of patients; PR = partial response; CR = complete response; dex = dexamethasone

Table 2 Bortezomib in relapsed/refractory MM patients

Regimen	Phase	N	PR	CR	Reference
Single agent	III (APEX)	333	+ CR 38%	6%	Richardson et al. (66)
+ dex	International III b	208	+ CR 51%	+ VGPR 33%	Mikhael et al. (120)
+ Doxil®	III	324	+ CR 44%	+ nCR 13%	Orlowski et al. (68)
Single agent	II (SUMMIT)	193	+ CR 27%	+ nCR 10%	Richardson et al. (64)
Single agent (1.0 and 1.3 mg/m^2)	II (CREST)	54	33-50%	not reported	Jagannath et al. (65)
+ temsirolimus	II	39	+ CR 36%	+ nCRB 10%	Ghobrial et al. (121)
+ Cytoxan® + dex - intermediate dose	II	64	+ CR 82%	16%	Kropff et al. (122)

N = number of patients; PR = partial response; CR = complete response; VGPR = very good partial response; nCR = near complete response; dex = dexamethasone

Recent studies employed bortezomib as part of cytoreductive regimens prior to autologous stem cell transplants with great success (69-72). New generation proteosome inhibitors (NPI-0052-Salinisporamide A; PX-171-carfilzomib, CEP-18770), as well as IκB inhibitors (PS-1145, MLN120B) are already going through clinical and pre-clinical studies and seem to be very promising.

Immunomodulators (IMiD's)

The IMiDs® are a group of unique, orally bio-available agents that have been refined, using thalidomide as a structural template (Figure 4).

Modification of the thalidomide structure through removal of a carbonyl on the ring formed lenalidomide (CC-5013, Revlimid®), and the addition of an amino group at the 4 position of the phthaloyl ring formed pomalidomide (CC-4047). These IMiDs® were specifically designed to enhance the immunomodulatory and anticancer properties of thalidomide with fewer side effects. Preclinical studies have shown that lenalidomide and pomalidomide are 50,000 times more potent, in vitro, than thalidomide at inhibiting tumor necrosis factor alpha (TNF-α) (73, 74). Studies have revealed that IMiDs® not only inhibit angiogenesis, but also stimulate T-cell proliferation and in-

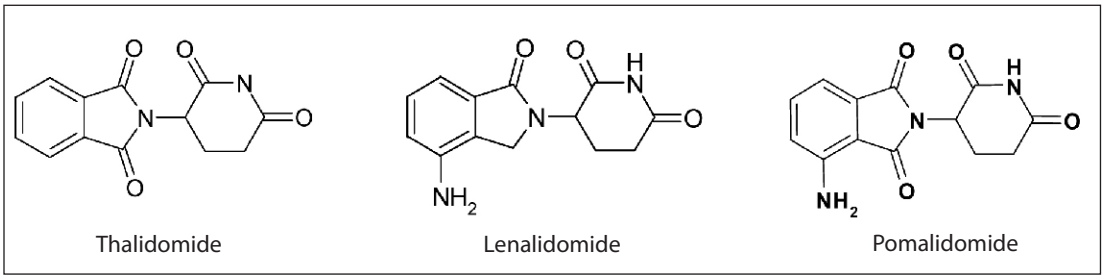

Figure 4 Molecular structure of thalidomide, lenalidomide and pomalidomide

duce apoptosis and growth arrest in resistant myeloma cells (75-77). These compounds also prevent the adhesion of myeloma cells to bone marrow stromal cells, and thereby inhibit the enhanced secretion of migratory factors, such as interleukin (IL)-6, TNF-α, and the vascular endothelial growth factor (VEGF) (78-84). Lenalidomide has more potent activity than thalidomide in the preclinical setting (73, 83), and has also demonstrated impressive clinical activity in both newly diagnosed and relapsed or refractory MM (85-88). Pomalidomide also demonstrates potent activity against TNF-α in vitro, indicating greater synergy than lenalidomide with rituximab in vivo (89). It also promotes T-cell differentiation and cytokine production via the transcription factor T-bet (90), and has demonstrated promising activity in clinical trials (91, 92).

The discovery that thalidomide had anti-angiogenic (93) and T-cell co-stimulatory (94) activity led to the clinical investigation of thalidomide for therapy in MM. In relapsed and refractory MM, thalidomide produced response rates of approximately 30% as a single agent (95). In newly diagnosed patients, thalidomide achieved response rates of 36% alone and 64-72% in combination with dexamethasone (96, 97). As a result, thalidomide in combination with dexamethasone received United States Food and Drug Administration (US FDA) approval for the treatment of newly diagnosed MM in 2006. In addition, recent phase III studies have investigated various thalidomide-containing regimens and reported improvements in quality of response with: thalidomide, adriamycine and dexamethasone compared to VAD (98); bortezomib, melphalan, prednisone and thalidomide (VMPT) compared to bortezomib, melphalan and prednisone (VMP) (99), melphalan, prednisone and thalidomide (MPT) compared to melphalan and prednisone (MP) (100), and bortezomib, thalidomide and dexamethasone (VTD) compared to thalidomide and dexamethasone (TD) (101). However, the encouraging effects of thalidomide are hampered by toxicity, which often compromises the dose or leads to discontinuation of therapy. Common adverse events include fatigue, somnolence, constipation, fluid retention, peripheral neuropathy, venous thromboembolism (VTE), and rash (95, 102, 103). Given the promising activity of thalidomide, synthetic analogs were developed and introduced in an effort to provide equal or greater immunomodulation, but a better tolerability profile. Clinical data indicate that the incidence of peripheral neuropathy, which is common with thalidomide, is low with lenalidomide and pomalidomide, (85, 92, 103-105).

Clinical studies with IMiD's

Studies among patients with relapsed or refractory MM have demonstrated that le-

nalidomide can overcome resistance to prior MM therapy, including thalidomide (106-108). In addition TTP and progression-free survival (PFS) are superior when lenalidomide is given at first relapse rather than later as salvage therapy (108). Two phase I trials of lenalidomide have demonstrated promising activity as well as decreased toxicity in heavily pretreated patients with relapsed or refractory MM (91, 106). These studies established 25 mg/day as the maximum tolerated dose (MTD) for lenalidomide in relapsed or refractory MM, and provided a firm foundation for continuing trials with lenalidomide, either alone or in combination with other active agents in MM.

Two large, randomized, phase III, double-blind, placebo-controlled clinical trials (North American MM-009 and European MM-010) have compared the efficacy and safety of lenalidomide plus dexamethasone (Len+Dex) with placebo plus dexamethasone in patients with relapsed or refractory MM (85, 86). In both trials, lenalidomide 25 mg/day or placebo was administered on days 1-21 of each 28-day cycle and oral dexamethasone 40 mg was administered on days 1-4, 9-12, 17-20 of each 28-day cycle. The MM-009 trial enrolled 353 patients (Len+Dex n = 177; placebo+Dex n = 176) and the MM-010 trial enrolled 351 patients (Len+Dex n = 176; placebo+Dex n = 175). The Len+Dex combination achieved a significantly ORR (MM-009: 61% vs. 20%; MM-010: 60% vs. 24%; both $p < 0.001$) and CR rate (MM-009: 14.1% vs. 0.6%; MM-010: 15.9% vs. 3.4%; both $p < 0.001$), (Figure 5).

The median TTP was significantly prolonged by the addition of lenalidomide to dexamethasone (MM-009: 11.1 months vs. 4.7 months; MM-010: 11.3 months vs. 4.7 months; both $p < 0.001$), (Figure 6) and the median OS was significantly longer in the Len+Dex arm (MM-009: 29.6 months vs. 20.2 months; $p < 0.001$; MM-010: not reached vs. 20.6; $p = 0.03$).

In the MM-009 and MM-010 studies, grade 3/4 hematologic AE were more common with Len+Dex and included neutropenia (41.2% and 29.5% vs. 4.5% and 2.3%, respectively), anemia (13.0% and 8.6% vs. 5.1% and 6.9%), thrombocytopenia (14.7% and 11.4% vs. 6.9% and 5.7%), and febrile neutropenia (3.4% vs. 0%). Other common grade 3/4 AE included infection (21.4% and 11.3% vs. 12.0% and 6.2%, respectively), and fatigue (6.2% and 6.8% vs. 6.3% and 3.4%). The incidence of VTE in the MM-009 and MM-010 studies was higher in the Len+Dex arm (14.7% and 11.4% vs. 3.4% and 4.6%, respectively); however, it was comparable to the incidence of 10% observed for the general MM population in retrospective analyses (109). On the basis of these studies, the US FDA approved lenalidomide in June 2006 and the European Medicines Agency in June 2007 for use in combination with dexamethasone in the treatment of MM in patients who have received at least one prior therapy.

Due to encouraging results in the relapsed or refractory setting, a phase II trial was undertaken to assess the efficacy and safety of the Len+Dex combination therapy in the front-line setting (104). In this phase II trial, lenalidomide (25 mg/day orally on days 1-21 of each 28-day cycle) was combined with dexamethasone (40 mg/day orally on days 1-4, 9-12, and 17-20 of each 28-day cycle) in 34 newly diagnosed, previously untreated MM patients. The ORR was 91%, with CR in 6% and very good partial response (VGPR) and near CR in 32%. Grade 3 or greater non-hematologic AE were reported in 47% of patients and included fatigue (15%), muscle weakness (6%), anxiety (6%), pneumonitis (6%), and rash (6%). Myelosuppression was minimal, most likely reflecting the preserved bone marrow reserve in this group of previously untreated patients. All patients were placed on low dose aspirin prophylaxis, based on the efficacy of

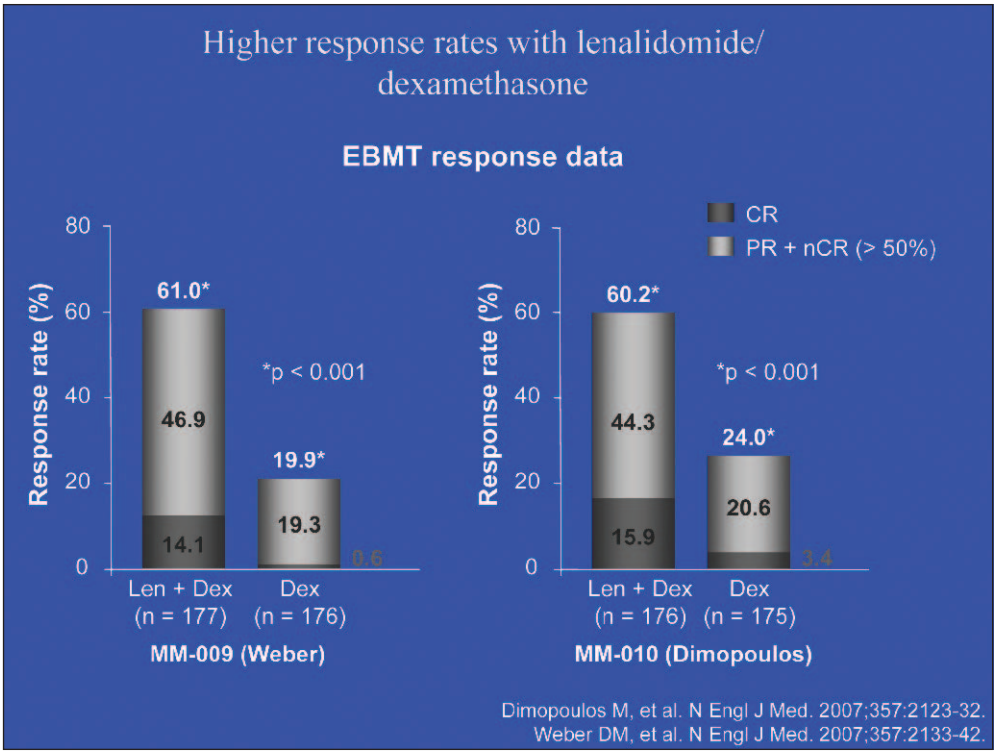

Figure 5 The European Group for Blood & Marrow Transplantation (EBMT) criteria-based response rates in MM-009 and MM-010 studies

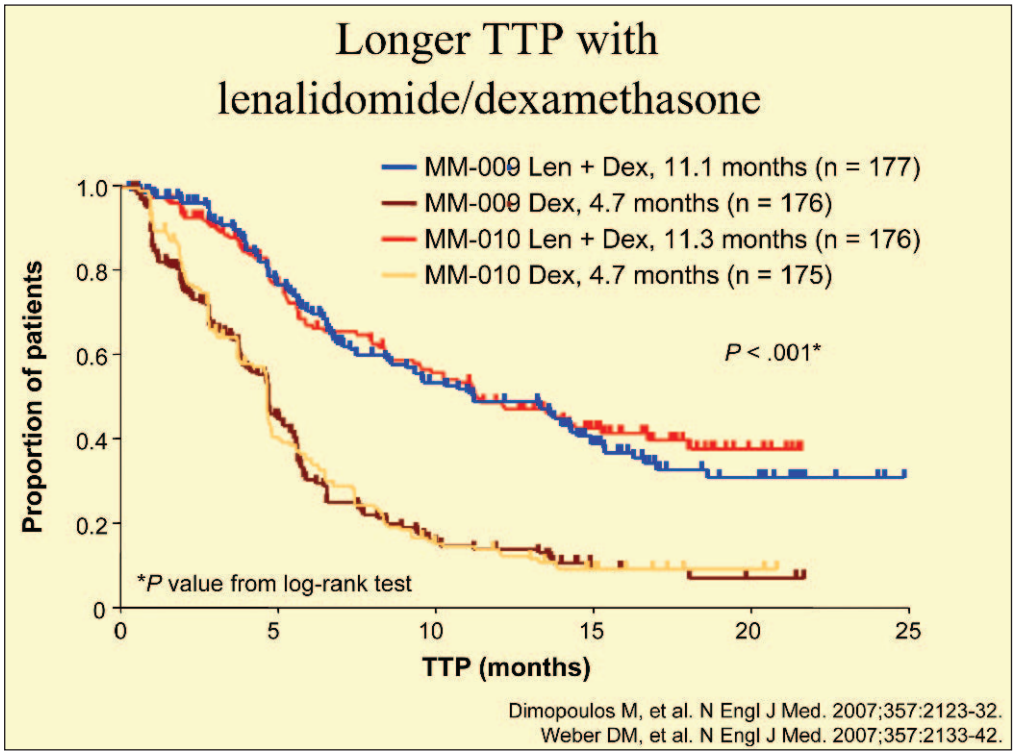

Figure 6 Time to progression (TTP) results in MM-009 and MM-010 studies

low dose aspirin in preventing VTE among patients treated on the thalidomide plus dexamethasone regimen (110), and only one patient developed a VTE. In addition, Len+Dex combination therapy appeared to be a useful pre-transplant conditioning regimen, as there was no adverse effect on stem cell mobilization among these patients.

With successful responses and better tolerability obtained from early trials, lenalidomide is rapidly being incorporated into front-line regimens. The Southwest Oncology Group (SWOG) and Eastern Cooperative Oncology Group (ECOG) performed randomized, phase III trials assessing Len+Dex as primary therapy in the front-line setting. The SWOG trial compared Len+Dex to dex alone in patients with newly diagnosed MM (87). In this study, 198 patients were randomized, 100 received lenalidomide 25 mg/day (28 of 35 days for 3 induction cycles, then 21 of 28 days as maintenance thereafter) plus dex 40 mg/day (days 1-4, 9-12, and 17-20 as induction, then days 1-4, and 15-18 as maintenance) and 98 received dex plus placebo. In 133 patients who were assessable for response, the ORR was significantly higher (85.3% vs. 51.3%; $p = 0.001$) and 1-year PFS was significantly longer (77% vs. 55%, $p = 0.002$) with Len+Dex. The 1-year OS was high and there was no difference between arms (93% vs. 91%). VTE was reported in 25% of patients treated with Len+Dex vs. 7% of patients treated with dex alone. Most patients (81%) who experienced VTE received aspirin as thromboprophylaxis, however it is to be noted that those patients received the full dose of aspirin at 325 mg daily which is known to be thrombogenic as it inhibits prostacyclin activity thus negating its anti-platelet role (111, 112). Patients in the dex arm who progressed were allowed to cross over to the Len+Dex arm. Of 40 patients who crossed over, the ORR in 23 who were assessable for response was 70.4%. These data confirm the superior efficacy with Len+Dex in newly diagnosed patients. Unfortunately, this study was prematurely closed when the results of the ECOG study E4A03 were announced and use of high-dose dex in combination with lenalidomide was no longer considered appropriate.

The ECOG trial compared lenalidomide plus standard-dose dex (RD) to lenalidomide plus low-dose dex (Rd), in an attempt to further diminish adverse events while maintaining the response rate. In this study, patients in the RD arm were treated with lenalidomide 25 mg/day on days 1-21 of each 28-day cycle and dex 40 mg/day on days 1-4, 9-12, and 17–20 of each 28-day cycle, and patients in the Rd arm received dex 40 mg on days 1, 8, 15, and 22 of each 28-day cycle (113). A total of 445 patients were randomized, 223 to RD and 222 to Rd. Grade 3 or higher AE were more common in the RD arm (49% vs. 32%; $p < 0.001$), including neutropenia (10% vs. 19%; $p=0.01$), VTE (25% vs. 9%; $p < 0.001$), and infections (16% vs. 6%; $p < 0.001$). Although response rates during the first 4 cycles were higher with RD (ORR: 82% vs. 70%; $p = 0.007$; CR + VGPR: 52% vs. 42%; $p = 0.06$), OS was significantly higher in the Rd arm, $p = 0.006$, (1-year OS: 96% vs. 88%; 2-year OS: 87% vs. 75%). The 2-year OS rate for the 102 patients who underwent stem cell transplant (94%) was comparable to the 2-year OS for patients in the Rd arm who continued primary therapy beyond 4 cycles (91%). These data demonstrated superior outcome with lenalidomide plus low-dose dex in patients with newly diagnosed MM compared to lenalidomide plus high-dose dex. The dose and schedule of dex will need to be evaluated further in light of the differences between the results of the SWOG and ECOG studies. There are probably groups of patients that would benefit from high dose dex administered according to the SWOG schedule and others that a lower dose will achieve similar disease outcome with less toxicity and mortality.

Baz et al. combined pegylated liposomal doxorubicin, vincristine, and dex (DVd) regimen with lenalidomide (DVd-R) in a phase I/II study among patients with relapsed or refractory MM (114). The study objectives were to determine the MTD and evaluate the safety and efficacy of DVd-R. Lenalidomide was administered orally at doses of 5, 10, and 15 mg/day for 21 days of each 28-day cycle in cohorts of 3-6 patients. Patients were treated for at least 4 cycles, and a maximum of 2 cycles after best response. Maintenance therapy included continuation of lenalidomide with the addition of prednisone 50 mg every other day until disease progression. Low-dose aspirin (81 mg) was administered as VTE prophylaxis. Sixty-two patients were enrolled in the study (40 refractory to prior therapy). The MTD of lenalidomide with DVd chemotherapy was 10 mg. The ORR was 75% with CR or near CR in 29%. After a median follow-up of 7.5 months, the median PFS was 12 months and the median OS had not been reached. Grade 3/4 adverse events included neutropenia (32%), febrile neutropenia (7%), peripheral neuropathy (5%), and VTE (9%). This novel combination appears to be well tolerated, and resulted in a high response rate in the group of patients with MM, most of whom were refractory to prior therapy.

Tables 3 and 4 summarize response rates in selected Phase II and III studies using lenalidomide as a single agent or in combination in newly diagnosed and relapsed/refractory MM patients.

In addition to the ability of lenalidomide to exert effective anti-tumor activity thorough direct anti-malignant plasma cell effects, it also exerts immune modulatory effects. Lenalidomide stimulates the immune cellular system leading to a beneficial impact on infectious complications, especially those that rely on the cellular immune system. One of the major viral infections in patients with multiple myeloma is herpes zoster that occurs in 15% of multiple myeloma patients over the course of the disease. Herpes zoster has high morbidity, especially in this age group, where post herpetic neuralgia could be crippling to the patients. With lenalidomide based therapy the incidence of herpes zoster is less than 5% as compared to other regimens that include proteasome inhibitors, where the incidence ranges from 15-60% (115, 116).

The clinical activity of pomalidomide was first demonstrated in a phase I study in which 24 patients with relapsed or refractory MM were treated with pomalidomide as a single agent (91). The MTD was established at 2 mg/day. The ORR was 54%, including

Table 3 Lenalidomide (Len) in newly diagnosed MM patients

Regimen	Phase	N	PR	CR	Reference
+ standard dose dex	III	223	+ CR 82%	+ VGPR 52%	Rajkumar et al. (113)
+ low dose dex	III	222	+ CR 70%	+ VGPR 42%	Rajkumar et al.113
+ dex	III	133	+ CR 85%	+ nCR 15%	Zonder et al. (87)
+ clarithromycin + dex	II	72	+ CR 90%	+ nCR 39%	Niesvizki et al. (123)
+ melphalan + prednisone	III	153	+ CR 67%	+ VGPR 46%	Palumbo et al. (124)
+ melphalan + prednisone + len maintenance	III	152	+ CR 77%	+ VGPR 50%	Palumbo et al. (124)

N = number of patients; PR = partial response; CR = complete response; VGPR = very good partial response; nCR = near complete response; dex = dexamethasone

Table 4 Lenalidomide (Len) in relapsed/refractory MM patients

Regimen	Phase	N	PR	CR	Reference
+ dex	III	177	+ CR 61%	14%	Weber et al. (85)
+ dex	III	176	+ CR 60%	16%	Dimopoulos et al. (86)
Single agent	II	102	+ CR 17%	+ nCR 4%	Richardson et al. (105)
+ Doxil® + vincristine + dex	II	62	+ CR 75%	+ nCR 29%	Baz et al. (114)
+ melphalan + prednisone + thalidomide + maintenance Len	II	43	+ CR 91%	+ VGPR 45%	Palumbo et al. (125)

N = number of patients; PR = partial response; CR = complete response; VGPR = very good partial response; nCR = near complete response; dex = dexamethasone

CR in 17%. Four patients (17%) experienced VTE. Pomalidomide therapy was associated with significantly elevated serum IL-2 receptor and IL-12 levels, which is consistent with activation of T cells, monocytes and macrophages. Based on these results, a recent phase II study has evaluated the safety and efficacy of pomalidomide (2 mg/day) combined with low-dose dexamethasone (40 mg/day on days 1, 8, 15, and 22 of each 28-day cycle) in 37 patients with relapsed or refractory MM (92). Most patients had received prior ASCT (76%) and prior IMiD® therapy (62%). The ORR was 62%, including VGPR in 24%. Objective responses were also reported 4 of 13 patients (29%) who were refractory to lenalidomide. Grade 3 hematologic AE included neutropenia (31%), thrombocytopenia (3%), and anemia (3%). There was no grade 3 neuropathy, but grade 1/2 neuropathy was reported in 16% of patients. Due to the incidence of VTE in the phase I study, all patients received aspirin as thromboprophylaxis and there were no cases of VTE. Pomalidomide appears to be another promising agent with a role for further studies as an immunostimulatory modality of treatment among patients with relapsed or refractory MM.

New directions

Multiple new therapeutic targets in the treatment of MM have been recently identified. Agents targeting cell surface molecules, specific molecules mediating growth, survival, drug resistance and migration of MM cells, as well as signaling pathways participating in these vital functions of MM cells are already going through pre- and clinical studies. They include agents such as IkB kinase inhibitors, VEGF receptor tyrosine kinase inhibitors, FGFR3 inhibitors, farnesyltransferase inhibitors, histone deacytilase inhibitors, heath shock protein inhibitors, telomerase inhibitors, Smac mimetics, MAPK inhibitors, TGFα inhibitors, TRAIL ligands, IGF-1 receptor inhibitors, HMG-CoA reductase inhibitors, Anti CD40 and Anti CD56 agents. We hope that these new agents in combination with existing ones will lead to the ultimate result for patients and their families: the cure of multiple myeloma.

Conclusions

Treatment options for patients with multiple myeloma have increased significantly in the past 10-15 years. The length and quality of

life have improved due to the greater efficacy and lower toxicity of new treatments. A future challenge for physicians treating patients with this complicated disease is how to use available treatments in a way that best fits a particular patient looking for help, and achieve the goal of individualized therapy.

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