Published in final edited form as:

Best Pract Res Clin Haematol. 2007 December ; 20(4): 797–816. doi:10.1016/j.beha.2007.09.008.

From the bench to the bedside: emerging new treatments in multiple myeloma

Constantine S. Mitsiades, Patrick J. Hayden, Kenneth C. Anderson, and Paul G. Richardson*

Dana Farber Cancer Institute, 44 Binney St, Dana 1B02, Boston, MA 02115, USA

Abstract

Within the last decade, several novel classes of anti-myeloma therapeutics have become available. The clinical successes achieved by thalidomide, lenalidomide, and the proteasome inhibitor bortezomib, and in particular the ability of these agents to lead to major clinical responses in patients resistant to conventional or high-dose chemotherapy, have highlighted the importance of expanding even further the spectrum of classes of agents utilized for the treatment of myeloma. Herein, we review the current state of the field of development of novel anti-myeloma agents, with emphasis on classes of therapeutics which have already translated into clinical trials or those in advanced stages of preclinical development. These include second-generation proteasome inhibitors (NPI-0052 and PR-171), heat shock protein 90 (hsp90) inhibitors, 2-methoxyestradiol, histone deacetylase (HDAC) inhibitors (e.g. SAHA, tubacin and LBH589), fibroblast growth factor receptor 3 (FGF-R3) inhibitors, insulin-like growth factor 1 receptor (IGF-1R) inhibitors, mTOR inhibitors, monoclonal antibodies, and agents targeting the tumor microenvironment, including defibrotide.

Keywords

myeloma; targeted therapy; immunomodulatory; thalidomide; lenalidomide; bortezomib; hsp90 inhibitor

Recent advances in the understanding of the pathobiology of multiple myeloma (MM) have provided the basis for a more comprehensive effort to develop novel therapies for this disease. The two major drug classes which have been approved by the US Food and Drug Administration (FDA) for the treatment of MM in the last decade – the immunomodulatory drugs of the thalidomide class (i.e. thalidomide itself and lenalidomide) and proteasome inhibitors – represent clinical advances associated, at least in part, with their ability to target both the neoplastic clone and its local microenvironment in the bone marrow (BM).

The complex interactions between MM cells and their surrounding microenvironment in the bone marrow are now recognized as central to the biology of the disease and its propensity for development of drug resistance.(1) MM cells interact with diverse partners, including BM stromal cells (BMSCs), which activate in MM cells a series of signaling pathways that in turn (1) support MM-cell proliferation and survival; (2) confer resistance of MM cells to

© 2007 Elsevier Ltd. All rights reserved.

*Corresponding author. Tel.: +1-617-632-2104; Fax: +1-617-632-4301. paul_richardson@dfci.harvard.edu.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

conventional therapies, such as corticosteroids or alkylating agents; and (3) trigger MM cells to produce cytokines and growth factors which enhance the increased bone resorption observed in MM and/or the recruitment of new blood vessels to the BM.(1–3)

The Road to Clinical Successes of Novel Anti-Mm Agents and How they Influence Ongoing Drug Development Efforts in MM

The anti-angiogenic activity of thalidomide in non-neoplastic models(4) provided the rationale for its initial clinical assessment in patients with advanced MM.(5) The encouraging clinical results of thalidomide monotherapy in relapsed or refractory MM – overall response rate (ORR) of 29.4% and median overall survival (OS) of 14 months, according to systematic review of 42 communications on a total of 1674 patients(6) – were also recapitulated (with more favorable depth and duration of responses) when thalidomide was combined with other conventional anti-MM agents(7–10) including phase-III data about the superiority of thalidomide– dexamethasone (Thal-Dex) combination over pulsed high-dose Dex alone in newly diagnosed disease.(11)

The realization that nuclear factor κ B (NF- κ B), a key transcription factor for MM-cell proliferation, survival and drug resistance,(12–14) is regulated by 20S chymotryptic-like activity of the proteasome(15) which is, in turn, targeted by the small-molecule inhibitor bortezomib (also known as PS-341 or Velcade™),(16–18) provided the framework for preclinical(19) and clinical studies of this agent in MM,(20,21) leading to its FDA approval for treatment of advanced MM.(22)

The search for agents that capitalize on the anti-MM properties of thalidomide but exhibit more pronounced clinical activity with fewer side-effects led to the preclinical(23–25) and clinical (26–28) development of lenalidomide (also known as CC-5013, IMiD-3 or Revlimid™). Lenalidomide is not only active in cases of patients resistant to conventional or other novel anti-MM agents,(26,27) it also has a profile of manageable side-effects that do not include key toxicities associated with thalidomide use (e.g. neuropathy, somnolence, and constipation).

Thalidomide, bortezomib and lenalidomide appear to share some intriguing common denominators which may explain, at least in part, why these classes of drugs are active even in patients with chemorefractory and/or steroid-refractory MM: all three agents trigger MM-cell death through mechanisms and molecular targets distinct from those of conventional chemotherapy and glucocorticoids. Furthermore, all these agents are capable of targeting MM cells not only by induction of MM-cell death (through these agents themselves and/or their in-vivo metabolites, in the case of thalidomide and perhaps lenalidomide) but also via effects on key aspects of tumor– host interactions, including: (1) disruption of tumor-associated neo-angiogenesis;(4,29,30) (2) attenuation of MM-cell adhesion to stromal cells, resulting in suppression of ensuing cytokine production;(19,25,29) and (3) modulation of the activity of the host immune system against MM cells.(24,31) In addition, there are data to support a role of proteasome inhibitors and lenalidomide in suppressing bone resorption(32,33) and an effect of proteasome inhibition in enhancing the activity of osteoblasts.(32) The aforementioned features of these classes of novel anti-MM agents can perhaps account for a cardinal translational feature that sets them apart from conventional chemotherapy and steroids: unlike these latter two drug classes, proteasome inhibitors, thalidomide and lenalidomide are capable of leading to MM-cell death despite the presence of BMSCs. Indeed, while BMSCs significantly attenuate the anti-MM effect of conventional chemotherapy and glucocorticoids, proteasome inhibitors and immunomodulatory agents of the thalidomide family can overcome this protective effect.

The progress achieved in the therapeutic management of MM with the introduction of these three novel agents has led to the realization that perhaps more progress can be achieved through two distinct, but potentially synergistic, approaches: one is to develop combination regimens where recently developed novel anti-MM agents function as a therapeutic ‘backbone’, around which other conventional and/or novel agents can be added. The second direction involves the attempt to develop more drug classes that share the aforementioned functional features of thalidomide, lenalidomide and bortezomib, i.e., the ability to target both the MM tumor cell and its microenvironment and to overcome its protective effects, through molecular mechanisms/targets not previously addressed by existing drugs. In this review, we highlight the progress in these two key aspects of developmental therapeutics for MM and discuss some of the novel classes of anti-MM agents currently being tested in phase-I and/or -II clinical trials in MM, as well as on agents which are currently in advanced stages of their preclinical development and are anticipated to be tested clinically in the near future.

Combinations of Novel Agents

Preclinical studies have shown that bortezomib triggers apoptosis of MM cells in a dual caspase-8- and -9-mediated manner. In terms of the direct pro-apoptotic activity of lenalidomide and other IMiDs, this is caspase-8. Both agents can suppress NF- κ B activity, but with different kinetics and likely acting at different levels of the molecular regulation of NF- κ B transcriptional activity (with bortezomib leading to accumulation of I κ B, an inhibitor of NF- κ B activity, while lenalidomide is proposed to act at a more upstream molecular level or levels. The observation that these two agents operate at these distinct functional levels, which converge to mutually enhance the anti-MM activity of these agents, provided the rationale for preclinical studies which indeed confirmed that the combination of a proteasome inhibitor, such as bortezomib, with an immunomodulatory thalidomide derivative, such as lenalidomide or CC-4047 (ActimidTM), can lead to synergistic anti-MM effects.(23) These preclinical studies led to a phase-I study of this two-drug combination in heavily pretreated patients.(34) A series of dose-escalation steps established that administration of lenalidomide at 15 mg daily and bortezomib at 1.0 mg/m² (at the standard regimen of twice weekly administration for the first 2 weeks of 3-week cycles) offered the optimal balance between efficacy and side-effects. In 36 evaluable patients, the ORR was 58% after a median of six cycles, with a very favorable side-effect profile and remarkable durability of response. A phase-II ‘Rev-Vel’ study is currently accruing patients, and studies of the ‘Rev-Vel-Dex’ combination in the upfront setting are also showing great promise.(35)

Concurrently with the preclinical and clinical development of the bortezomib–lenalidomide-based combinations, the combination of bortezomib (at standard dose), thalidomide (100 mg daily increasing to a maximum of 200 mg per day) and dexamethasone (three 4-day blocks of Dex 20 mg/m² each cycle) (VTD) was initiated and was observed to lead to an overall response rate (ORR) of 87% with a complete response (CR) rate of 16%. (36) Other permutations of the same concept – i.e. use of a proteasome inhibitor and an immunomodulatory agent as the ‘backbone’ agents of a multi-agent combination – include a phase-III trial in Europe currently comparing VTD with Thal-Dex as induction therapy and consolidation therapy before and after double autologous stem-cell transplantation for younger patients with newly diagnosed MM; a study of bortezomib, polyethylene glycolated (PEGylated) liposomal doxorubicin and Thal (VDT), which showed a 56% response rate (CR rate of 22%) among 18 heavily pretreated patients,(37) and a trial (n = 30 patients with relapsed/refractory MM) of the combination of bortezomib, melphalan, prednisone, and Thal (VMPT), which led to partial response (PR) rate of 67%(10) – including a PR rate of 79% and immunofixation-negative CR rate of 36% – when the VMPT combination was administered as second-line treatment.

Investigational Agents

Several novel classes of drugs are currently in advanced preclinical development or early phase-I/II trials in the clinic. The most clinical experience has been amassed on new proteasome inhibitors, hsp90 inhibitors, 2-methoxyestradiol, histone deacetylase (HDAC) inhibitors, arsenic trioxide, perifosine, atiprimid, and vascular endothelial growth factor receptor (VEGF-R) inhibitors. Fibroblast growth factor receptor 3 (FGF-R3) inhibitors and monoclonal antibody against insulin-like growth factor 1 receptor (IGF-1R) are also under way, while IGF-1R kinase inhibitors, mTOR inhibitors, and I κ B kinase (IKK) inhibitors are also among the classes of drugs which have generated promising results in preclinical studies in vitro and/or in vivo, representing interesting candidates for phase-I studies (Figure 1).

Proteasome inhibitors

The excellent clinical results obtained with bortezomib have validated the proteasome as a therapeutic target in MM, and have therefore ignited an intense effort to identify ‘second-in-class’ proteasome inhibitors that would hopefully exhibit more potency, fewer side-effects, and more convenient routes of administration compared to bortezomib. The first such new proteasome inhibitors to be studied preclinically in MM was NPI-0052 (salinosporamide A). This small molecule was derived from a fermentation by *Salinospora*, a new marine gram-positive actinomycete, and was shown in 2003 to be a potent proteasome inhibitor.(38) Preclinical studies in MM models showed that NPI-0052 induces apoptosis of MM cells, including cells resistant to conventional anti-MM therapies as well as cells isolated from patients who had developed resistance to bortezomib-based therapies.(39) NPI-0052 is distinct from bortezomib in terms of chemical structure and effects on individual proteolytic activities of the proteasome activities. For instance, while bortezomib inhibits exclusively the chymotrypsin-like activity of the 20S proteasome, NPI-0052 inhibits all three protease activities in the proteasome: the chymotrypsin-like (CT-L), trypsin-like (T-L), and caspase-like (C-L) activities.(39) Another key difference is that NPI-0052 is orally bioavailable, while bortezomib in its current formulation is not amenable to oral administration. In animal model studies of subcutaneous plasmacytomas, NPI-0052 was well tolerated and prolonged survival, with significantly reduced tumor volume of the plasmacytomas. Interestingly, synergistic in-vitro activity was seen when NPI-0052 was tested in combination with bortezomib. The oral bioavailability of NPI-0052 and the promising results from the animal tumor models in MM have led to ongoing clinical trials of this agent.

Another new proteasome inhibitor recently developed is PR-171 (carfilzomib), a novel epoxyketone-based irreversible proteasome inhibitor.(40–42) In preclinical studies in MM models, carfilzomib was found to potently bind and specifically inhibit the chymotrypsin-like proteasome and immunoproteasome activities, resulting in accumulation of ubiquitinated substrates. It induced a dose- and time-dependent inhibition of MM-cell proliferation, with ultimate induction of MM-cell apoptosis, which was associated with JNK activation, mitochondrial membrane depolarization, release of cytochrome c, and activation of both intrinsic and extrinsic caspase pathways, similarly to bortezomib. Interestingly, however, carfilzomib was active against bortezomib-resistant MM cell lines, as well as samples from MM patients with clinical resistance to bortezomib. Carfilzomib also overcame resistance to other conventional agents, and acted synergistically with dexamethasone to enhance cell death.(42) Taken together, these data provided a rationale for the clinical evaluation of carfilzomib in MM. So far, a phase-I trial is testing two different dose schedules of carfilzomib: 2-week cycle, daily treatment for 5 days (QDx5) with 9 days’ rest versus 4-week cycle, daily treatment for 2 days (QDx2) every week for 3 weeks with 12 days’ rest. This trial has enrolled patients with MM and other B-cell neoplasias, and a report at the recent 2007 International Myeloma Workshop(43) indicated clinical responses in

some of the MM patients enrolled in that trial: one PR and one minimal response (MR) among patients treated on the QDx5 schedule, as well as three PRs and two MRs among 11 patients treated with the QDx2 protocol). Dose-limiting toxicities (DLTs) have included febrile neutropenia at 20 mg/m² in the QDx5 protocol, and in the QDx2 schedule, grade 4 anemia and grade 3 hypoxia at 27 mg/m². No painful peripheral neuropathy has been observed in that trial so far.(43)

Hsp90 inhibitors

Heat shock protein 90 (hsp90) is a molecular chaperone which interacts intracellularly with a broad range of client proteins and functions to preserve their three-dimensional conformation in a functionally competent state, as well as facilitating their intracellular trafficking. Compared to many other heat shock proteins, hsp90 interacts with a more restricted set of client proteins (e.g. cell-surface receptors for diverse cytokines and growth factors, intracellular kinases and kinase targets, as well as other effectors of signal transduction cascades) which tend to promote cell proliferation, survival and drug resistance. Work from diverse centers, including our group, has shown that indeed small-molecule inhibitors which competitively inhibit the ATP-binding domain of hsp90, such as the ansamycin geldanamycin and its analogs – e.g. 17-allylamino-17-demethoxygeldanamycin (17-AAG) or 17-(dimethylaminoethylamino)-17-demethoxygeldanamycin (DMAG) – can suppress the chaperoning function of hsp90 and therefore perturb the stability and function of its client proteins, leading to anti-proliferative pro-apoptotic effects in various solid tumor models and hematologic neoplasias. In particular, our work in MM models has shown that MM cells are responsive to hsp90 inhibitors in vitro (at pharmacologically achievable concentrations) and in clinically relevant orthotopic in vivo xenograft models,(44) and can also function to sensitize MM cells to other pro-apoptotic agents.(45) These original preclinical observations have since been confirmed in multiple MM experimental settings, both in vitro(46–49) and in vivo,(47) using diverse members of the ansamycin class, as well as different drug formulations.

The aggregate preclinical experience with hsp90 inhibitors indicates that the mechanistic basis of their anti-MM effects is related to the pleiotropic nature of their molecular sequelae. (44) Indeed, hsp90 inhibitors abrogate signaling cascades induced by cytokines (e.g. IGF, IL-6) at multiple molecular levels, including suppression of cell-surface expression of IGF-1R and IL-6R and inhibition of downstream signaling (via PI-3K/Akt/mTOR, Ras/Raf/MAPK, IKK/NF-κB), via molecular events which include the suppression of expression and/or function of Akt, Raf, KK-α, and p70^{S6K}. These events lead to multiple downstream pro-apoptotic sequelae, including increased nuclear translocation of pro-apoptotic members of the forkhead family of transcription factors; suppressed expression of intracellular inhibitors of apoptosis (e.g. FLIP, XIAP, cIAP-2), as well as decreased constitutive and IGF-induced activity of NF-κB, telomerase, HIF-1α and 20S proteasome. These molecular events not only contribute to decreased MM proliferation, but can also increase their chemo-/Dex-sensitivity (e.g. through NF-κB inhibition), suppress the long-term replicative potential of MM cells (e.g. through inhibition of telomerase function), or blunt pro-angiogenic effects (e.g. via suppression of HIF-1α transcriptional activity). These pleiotropic anti-proliferative/pro-apoptotic events allow hsp90 inhibitors to abrogate BMSC-derived protection on MM tumor cells, and sensitize them to other anticancer agents, including cytotoxic chemotherapy. Furthermore, because MM cells exposed to the proteasome inhibitor bortezomib up-regulate the expression of heat shock proteins, including hsp90, as a stress response to counteract the intracellular accumulation of misfolded proteins, hsp90 inhibitors can sensitize MM cells to the anti-MM effects of bortezomib.

These studies provided the rationale for ongoing clinical trials of tanespimycin (17-AAG in the KOS-953 cremophor-based formulation) either as a single agent(50) or in combination

with bortezomib(51) in patients with relapsed or refractory MM. In these trials, tanespimycin has shown a manageable profile of side-effects (without significant cardiotoxicity, peripheral neuropathy or deep vein thrombosis), durable disease stabilization, and minor responses with single-agent treatment in relapsed and refractory MM patients, as well as encouraging anti-MM activity by the combination of tanespimycin with bortezomib. This experience, coupled with the lack of additive toxicity or pharmacokinetic interactions in the tanespimycin + bortezomib combination, provides a platform for future phase-III trials of this regimen. Hsp90 inhibitors therefore represent an emerging class of anti-tumor agents with a specific target, but pleiotropic and versatile anti-proliferative/pro-apoptotic properties, which can allow them to function as possible sensitizers that can enhance the response to existing anti-MM therapies or investigational agents.

2-Methoxyestradiol

2-Methoxyestradiol (2ME2) is an estrogen derivative which has been shown to inhibit growth and induce apoptosis in MM cell lines and patient cells. It has also been demonstrated to slow tumor proliferation, prolong survival, and decrease angiogenesis in a murine MM model.(52) In early clinical studies, 2ME2 had a favorable safety profile and achieved durable disease control in some patients who had previously relapsed or were refractory to conventional therapies.(53)

Histone deacetylase (HDAC) inhibitors

Histone acetylation modulates gene expression, cellular differentiation, and survival, and is regulated by the opposing activities of histone acetyltransferases (HATs) and histone deacetylases (HDACs).(54,55) HDAC inhibition results in accumulation of acetylated nucleosomal histones and induces differentiation and/or apoptosis in transformed cells. Given the multiplicity of molecular pathways which regulate MM- cell proliferation, survival and drug resistance, as well as the fact that these pathways, directly or indirectly, feed into the process of transcriptional regulation, we hypothesized that HDAC inhibition could function as a master switch that could simultaneously affect multiple pathways critical for MM cells. We therefore studied the effect of suberoylanilide hydroxamic acid (SAHA), the prototype of a series of hydroxamic-acid-based HDAC inhibitors, on MM cell lines and primary MM tumor cells, and observed that SAHA (1) induced MM-cell apoptosis, with increased p21 and p53 protein levels and dephosphorylation of Rb; (2) sensitized MM cells to other anti-MM agents; and (3) inhibited the secretion of IL-6 triggered by MM-cell binding to BMSCs, suggesting that SAHA can overcome cell-adhesion-mediated drug resistance.(56) Further studies have shown that MM cells treated with SAHA exhibit a constellation of anti-proliferative and/or pro-apoptotic molecular events, including down-regulation of transcripts for members of the IGF/IGF-1 receptor (IGF-1R) and IL-6 receptor (IL-6R) signaling cascades, anti-apoptotic molecules (e.g. caspase inhibitors), oncogenic kinases, DNA synthesis/repair enzymes, and transcription factors (e.g. XBP-1, E2F-1) implicated in MM pathophysiology. Importantly, SAHA treatment suppresses the activity of the proteasome and expression of its subunits, and enhances MM-cell sensitivity to proteasome inhibition by bortezomib (PS-341). SAHA also enhances the anti-MM activity of other pro-apoptotic agents, including dexamethasone, cytotoxic chemotherapy, and thalidomide analogs.(57) Based on these preclinical studies, an open-label phase-I dose-escalation of oral SAHA (200, 250 and 300 mg p.o. b.i.d. for 5 consecutive days followed by 2 days of rest) was administered in 4-week cycles in patients with relapsed/refractory MM. Side-effects (fatigue, diarrhea, dehydration) were manageable with appropriate supportive care and, importantly, no significant myelosuppression, neuropathy or sedation (which are associated with other anti-MM agents) was seen. In seven evaluable patients with relapsed refractory MM, two MRs were observed and two patients had stable disease. The fact that SAHA, as an orally administered HDAC inhibitor, showed a manageable profile of

side-effects and preliminary evidence of anti-tumor activity in advanced MM has provided the basis for further studies of SAHA in MM.(58) An interesting area for possible improvement of the clinical activity of SAHA in MM involves the optimization of the schedule of administration. The aforementioned phase-I trial of SAHA involved twice-daily dosing, while the approval of SAHA (vorinostat) for cutaneous T-cell lymphoma was based on studies where the agent was administered once daily. It is possible that daily administration of SAHA would be associated with a more favorable profile of side-effects and therefore delivery of higher cumulative doses of the medication. This hypothesis is consistent with the clinical precedent of lenalidomide, which, when administered once daily, is better tolerated than the twice-daily administration of the same cumulative dose.(27)

The anti-MM activity of SAHA provided the impetus and framework for further studies of other HDAC inhibitors in MM models, such as the hydroxamic acid derivatives NVP-LAQ824(59) and NVP-LBH589,(60,61) as well as the HDAC-6-selective inhibitor tubacin. (62) NVP-LAQ824 and NVP-LBH589 trigger MM-cell apoptosis against cell lines resistant to conventional therapies, and overcome the effect of BMSCs on MM cells.(59–61) Interestingly, treatment of MM cells with NVPLAQ824 also led to proteasome inhibition, as determined by reduced proteasome chymotrypsin-like activity and increased levels of cellular polyubiquitin conjugates.(59) Finally, a study using NVP-LAQ824 in a preclinical murine myeloma model provides in-vivo relevance to the aforementioned in-vitro studies. (59) Concerns regarding the effects of LVP-LAQ824 on QTc interval led to increased interest in the clinical development of NPV-LBH589, which is currently being evaluated in clinical trials in MM.

Arsenic trioxide

The clinical success of arsenic-based therapies in acute promyelocytic leukemia(63) raised the possibility that arsenic trioxide might be an interesting agent to test preclinically in MM. Indeed, in-vitro studies showed that arsenic trioxide mediates anti-MM activity both directly on tumor cells and indirectly by inhibiting production of myeloma growth and survival factors in the bone marrow (BM) microenvironment. Arsenic trioxide levels that were deemed to be clinically achievable levels (2–5 μM) were observed to induce apoptosis of even drug-resistant MM cell lines, enhance the anti-MM effect of dexamethasone, inhibit the secretion of IL-6 and VEGF from MM cell-BMSC co-cultures, and overcome the protective effect of IL-6 or BMSCs on MM cells.(64) In a phase-II study of single-agent arsenic trioxide in relapsed and refractory MM, reductions in paraprotein levels of at least 25% were seen in eight (33%) of 24 patients with a median time to response of 67 days.(65) The MAC regimen comprising melphalan, arsenic trioxide and ascorbic acid was associated with an ORR of 48% when administered to 65 patients with advanced MM.(66) This steroid-free regimen was well tolerated, and the median OS of this heavily pretreated group was 19 months. Bortezomib has been combined with arsenic trioxide and ascorbic acid in the ABC protocol on which 22 patients with heavily pretreated MM were enrolled.(67) A total of 6 (27%) patients had objective responses, and the OS rate was 74% at 12 months.

Perifosine

Despite the wealth of data that support the role of the PI-3K/Akt pathway as an important therapeutic target for MM,(68–72) few, if any, agents which selectively target these molecules have translated into clinical applications, perhaps because of the difficulty of generating agents with sufficient selectivity and potency, yet acceptable safety profile. Until such agents become available, extensive efforts have been made to utilize less selective agents, which could nonetheless function to inhibit the pathway for clinical purposes. A key representative of this approach is perifosine (Keryx Biopharmaceuticals, NY), a synthetic novel alkylphospholipid, which has been shown to inhibit the phosphorylation of Akt in

MM cells in a time- and dose-dependent manner, and to induce apoptosis in M cell lines, including those resistant to other anti-MM agents.(73) The cytotoxicity of alkylphospholipids against MM cells appears also to include a novel lipid-raft-mediated mechanism, which involves the concentration of death receptors in membrane rafts with Fas/CD95 playing a major role in alkylphospholipid-mediated apoptosis.(74) The recent finding that perifosine both has anti-tumor activity and also induces myeloid hyperplasia in a murine myeloma model(75) provides an additional element in support of clinical testing of perifosine, i.e., the notion that in MM patients heavily pretreated with myelosuppressive agents, or with BM function compromised by the MM-cell infiltration, the use of perifosine may offer an approach that will, at the very least, spare the BM function from further compromise, if not help alleviate the myelosuppression of other anti-MM agents. Phase-I/II studies of perifosine in combination with low-dose Dex,(76) bortezomib(77) or lenalidomide–Dex(78) are currently under way.

Fibroblast growth factor receptor 3 inhibitors

FGF-R3 is a receptor tyrosine kinase which is dysregulated as a result of the t(4;14) chromosomal translocation that occurs in approximately 15% of MM patients.(79) The poor prognosis associated with this subtype(80,81) has made FGF-R3 an attractive target for novel drug development. CHIR-258, for example, is a small-molecule inhibitor of multiple receptor tyrosine kinases. In an orthotopic murine MM model, daily treatment with CHIR-258 inhibited tumor growth, resulting in a significant improvement in animal survival.(82) Other potential compounds under evaluation include PRO-001, a highly specific anti-FGF-R3-neutralizing antibody which has been shown to induce apoptosis in primary t(4;14) MM samples.(83)

Several key questions will have to be answered before a clearer picture emerges on the future role of FGF-R3 inhibitors in the therapeutic management of MM. The safety and efficacy profile of this class of drugs in the current clinical testing in MM will be an important factor, as with any other drug class evaluated in clinical trials. An additional factor that should be taken into consideration is whether primary MM tumor cells that express FGF-R3 still depend on this target at the time of treatment with this class of agents.

Azaspirane (Atiprimod)

Azaspirane (Atiprimod®) is an orally bioavailable cationic amphiphilic compound that significantly inhibits production of IL-6 and inflammation in rat arthritis and autoimmune animal models.(84,85) Given the role of IL-6 in MM, the experience with azaspirane in these non-neoplastic diseases has provided a strong impetus for preclinical evaluation of this compound in MM models. Azaspirane was shown to significantly inhibit MM-cell proliferation and induce caspase-mediated apoptosis in MM cell lines, as well as primary MM patient cells. BM-derived cytokines (e.g. IL-6 or IGF-1) and adherence of MM cells to BMSCs did not protect against the effect of azaspirane. Both conventional (dexamethasone, doxorubicin, melphalan) and novel (arsenic trioxide) agents augment apoptosis induced by azaspirane, which also inhibits signal transducer activator of transcription 3 (STAT3) and a PI3-K (phosphatidylinositol 3-kinase) target (Akt), but not extracellular signal-regulated kinase 1 and 2 (ERK1/2); it also demonstrates in-vivo anti-tumor activity against human MM cell growth in severe combined immunodeficient (SCID) mice.

Vascular endothelial growth factor inhibitors

Vascular endothelial growth factor (VEGF) and its receptors have been implicated the pathophysiology of a wide range of neoplasias, predominantly because of the role of VEGF in stimulating tumor-associated neo-angiogenesis. In the BM of MM patients, the microvascular density is higher compared to that in MGUS patients or normal donors, and is

higher in patients with advanced disease compared to newly diagnosed cases.(86,87) Furthermore, the experience of the clinical successes of thalidomide, an agent with anti-angiogenic properties, suggested that development of other agents targeting MM-associated neo-angiogenesis would be useful clinically. In the case of MM, the exogenous administration of VEGF triggers a modest increase in proliferation of tumor cells in vitro and a more pronounced increase in MM-cell migration in Boyden-chamber assays,(88) which depend on the regulatory role of caveolin,(89) while treatment of MM cell lines with VEGF-R kinase inhibitors (e.g. PTK787, GW654652, and pazopanib) can suppress MM-cell proliferation and migration.(90–92) In addition, pazopanib has been shown to inhibit VEGF-triggered signaling pathways in endothelial cells and lead to a decrease in growth rate of human plasmacytomas implanted subcutaneously in immunocompromised mice.(90) Interestingly, however, there have so far been three reports of clinical trials of different VEGF-R inhibitors which did not achieve clinical responses in MM.(93–95)

mTOR inhibitors

Rapamycin and two analogues, CCI-779 (Wyeth Ayerst, PA, USA) and RAD001 (Everolimus, Novartis, NJ, USA), are inhibitors of the phosphoprotein mammalian target of rapamycin (mTOR), and have shown preclinical potential as MM therapies. In an MM xenograft model, CCI-779 induced significant dose-dependent anti-tumor responses against subcutaneous growth of the cell lines 8226, OPM-2 and U266.(96) There may also be a role for combination therapy with this class in that synergy has been demonstrated when rapamycin has been used in conjunction with either Len(97) or the hsp90 inhibitor 17-AAG.(49) A phase-I/II study of intravenous CCI-779 in combination with bortezomib in patients with relapsed MM is currently accruing.

Insulin-like growth factor 1 receptor inhibitors

Insulin-like growth factor 1 (IGF-1) is central to the pathophysiology of multiple myeloma. A small-molecule IGF-1R tyrosine kinase inhibitor, NVP-ADW742 (Novartis Pharma AG, Switzerland), has shown significant cytotoxicity in vitro and in vivo in MM.(98) Targeting the IGF-1 receptor using picropodophyllin in the mouse 5T2MM model also showed strong anti-tumor activity and a significant OS advantage for treated mice.(99) The multiple roles of this signaling pathway were also reflected in significant inhibition of tumor-associated angiogenesis and osteolysis.

IKK inhibitors

Inhibition of activation of NF- κ B is one of several pathways through which the immunomodulatory drugs and bortezomib affect MM-cell growth. I κ B kinase (IKK) inhibitors have been employed to delineate the effects of specific blockade of this signaling cascade. MLN120B, a novel IKK β inhibitor,(100) and a pharmaceutical IKK2 inhibitor, the anilino pyrimidine derivative AS602868,(101) are promising candidates for phase-I/II trials.

Monoclonal antibodies in development for MM treatment

The major clinical success of monoclonal-antibody-based anti-CD20 therapeutic strategies (e.g. rituximab and various radionuclide-conjugated anti-CD20 monoclonal antibodies) in lymphomas have for several years now raised the question of whether MM could also be a setting for similar therapeutic advances using monoclonal antibodies. Although the plasma-cell stage of B-cell differentiation is typically not associated with CD20 expression, a sizeable minority of MM patients express CD20 on their MM plasma cells (~20%, and typically in association with t(11;14) translocation).(102) Furthermore, studies of circulating clonotypic B cells(103,104) and, in more recent years, of the proposed population of 'MM stem cells' have suggested that the in-vivo generation of the main population of CD138⁺

malignant plasma cells of MM is due to a small population of CD138⁺ CD20⁺ with stem-celllike features. These considerations have led to hypothesis that rituximab might be active in MM, due to its direct effect on the CD20⁺ subset of MM patients(105) and/or because of effects on the proposed CD20⁺ clonotypic B cells/‘MM stem cells’. However, different clinical trials of rituximab in MM have not been able to document major clinical responses, either in the CD20⁺ subset or in CD20⁺ patients.(105–107) Interestingly, in one of these studies(107) a significant decrease in peripheral B cells was observed without significant concomitant changes of BM myeloma cells. This observation was also reflected in the fact that mean paraprotein levels increased slightly during follow-up, but IgM levels dropped in all patients, indicating an effective targeting of normal, short-lived plasma cells.

While more work will be required to evaluate a possible role for CD20 targeting in MM, other targets for serotherapy in MM are also emerging. For instance, preclinical studies have shown that CD138 (syndecan-1), CD40, CD38, and CS-1 are surface antigens that merit consideration for a future role in MM therapeutics.(108–115) Expression of CD138 is a hallmark of normal and malignant plasma cells and, although its expression is not restricted to the plasma-cell lineage, the high levels of its expression in MM cells have been the basis for various efforts to develop anti-CD138 monoclonal antibodies for possible clinical applications. For instance, the murine monoclonal antibody B-B4, which targets human CD138, has been conjugated with the potent anti-microtubule agent maytansinoid DM1 (N(2′)-deacetyl-N(2′)-(3-mercapto-1-oxopropyl)-maytansine). The B-B4–DM1 immunoconjugate was found to be active against a panel of CD138⁽⁺⁾ MM cells in vitro, as well as in mouse models of subcutaneous MM-cell-line xenografts, and SCID mice implanted with human fetal bone (SCID-hu) and subsequently injected with patient MM cells. It is important to emphasize that the lack of B-B4 reactivity with mouse tissues precludes evaluation of its toxicity in these models,(108) a limitation that unfortunately applies to the large majority of monoclonal antibodies, which show no cross-reaction to the rodent version of the surface marker against which they were generated.

CD40 is expressed broadly in MM cell lines and primary MM tumor cells, which provided the impetus for preclinical studies of several different anti-human CD40 antibodies. These studies showed that a recombinant humanized monoclonal antibody against CD40 (rhuCD40 mAb) induces antibody-dependent cell-mediated cytotoxicity (ADCC) against CD40-positive MM cells.(113) Other studies with the anti-CD40 antibody SGN-40 showed that it induces modest cytotoxicity in MM cell lines and patient MM cells, and blocks sCD40L-mediated phosphatidylinositol 3′-kinase/AKT and NF-κB activation,(111) while pretreatment of MM cells with lenalidomide sensitized them to SGN-40-induced cell death.(109) CHIR-12.12 is a fully human anti-CD40 mAb which abrogates CD40L-induced growth and survival of CD40-expressing patient MM cells and triggers lysis of CD40⁺ MM cells via ADCC.(110) Finally, a recently emerging novel surface antigen for serotherapy in MM is CS1 (also known as CD2 subset 1, CRACC, or SLAMF7), a member of the CD2 family of cell-surface glycoproteins. This novel MM antigen has been studied with the use of HuLuc63, a novel humanized anti-CS1 mAb. CS-1 mRNA and protein is highly expressed in CD138-purified primary tumor cells from the majority of MM patients, while HuLuc63 induces ADCC against CS1-expressing CD138⁺ MM cells, including cells from patients with clinical resistance to various conventional and investigational anti-MM therapies.(116–118) These studies have set the stage for ongoing clinical trials of HuLuc63 in MM.

Defibrotide

Given the emerging understanding of how the interaction of the BM microenvironment with MM cells influences the pathophysiology of the disease, agents which target these interactions have become the focal point of extensive studies, not only because they may

improve the activity of already established therapies, but also because they could help, as part of combination regimens, to reduce side-effects associated with the use of conventional or other novel anti-MM agents. An example of such a microenvironment-targeting agent is defibrotide (DF), a orally bioavailable polydeoxyribonucleotide which has protective effects on endothelial cells without significant systemic anticoagulant properties and bleeding risk. Defibrotide has minimal inhibitory effect on MM cells in vitro, but targets tumor–microenvironmental interactions and potently sensitizes MM to cytotoxic chemotherapy. (119) Furthermore, it enhances the response of human MM xenografts in SCID/NOD mice to melphalan, cyclophosphamide and dexamethasone, (119) possibly by abrogating MM-cell interactions with BM stromal cells. The ability of defibrotide to protect against thrombosis while potentially enhancing the sensitivity of MM cells to other therapies has provided the rationale for an ongoing phase-I/II clinical trial to determine the efficacy and safety of melphalan, prednisone, thalidomide and defibrotide (MPTD) as salvage treatment in patients with advanced MM. (120) In this study, MPTD has provided promising evidence of anti-tumor activity in relapsed and refractory MM, with a high and durable response rate (CR +PR: 53%) and manageable toxicities. The absence of significant non-hematologic toxicity with this regimen, including only a single case of DVT (with no other prophylaxis used) and remarkably low rates of neuropathy to date, is especially encouraging.

Summary

The heterogeneity of MM biology and prognosis has become apparent over the last decade. A more personalized approach to treatment is likely to become standard as increasingly robust molecularly based prognostic information becomes available. Insights into the role of the tumor microenvironment have provided a new paradigm through which to assess the novel targeted therapies. Treatment options for younger patients eligible for ASCT now therefore include combination regimens based on novel backbone agents such as the immunoregulatory drugs thalidomide and lenalidomide, and the proteasome inhibitor bortezomib. The current focus of translational research is on how best to integrate the new targeted therapies with both conventional chemotherapy and transplant-based protocols. MPT appears to represent the standard of care for most older patients ineligible for ASCT. If, as seems likely, MPV and MPR prove to be superior to MP, three options with differing toxicities – MPT, MPV and MPR – will be available. A further feature of recent years has been a more formal attempt to base such combination treatment regimens on known molecular mechanisms, such as different apoptosis-inducing pathways or differential inhibition of key signal transduction pathways. Bortezomib and lenalidomide (Rev-Vel) is a good example of such rational targeting. Such combinations are likely to be a prominent feature of therapeutic advances in the coming years. Clinical trials continue to assess the residual role of corticosteroids and the optimal sequencing of novel agents so as to avoid the development of class-specific drug resistance and overlapping toxicities. Promising agents at the phase-I/II stage include novel proteasome inhibitors, hsp90 inhibitors, HDAC inhibitors, perifosine, mTOR inhibitors, and agents such as defibrotide which target microenvironment alone with the potential of reducing toxicity and enhancing therapeutic index in the context of combination therapy.

References

1. Mitsiades CS, Mitsiades NS, Richardson PG, Munshi NC, Anderson KC. Multiple myeloma: A prototypic disease model for the characterization and therapeutic targeting of interactions between tumor cells and their local microenvironment. *Journal of cellular biochemistry*. 2007 Jul 1; 101(4): 950–68. [PubMed: 17546631]
2. Mitsiades CS, Mitsiades N, Munshi NC, Anderson KC. Focus on multiple myeloma. *Cancer Cell*. 2004 Nov; 6(5):439–44. [PubMed: 15542427]

3. Mitsiades CS, Mitsiades NS, Munshi NC, Richardson PG, Anderson KC. The role of the bone microenvironment in the pathophysiology and therapeutic management of multiple myeloma: interplay of growth factors, their receptors and stromal interactions. *Eur J Cancer*. 2006 Jul; 42(11): 1564–73. [PubMed: 16765041]
4. D'Amato RJ, Loughnan MS, Flynn E, Folkman J. Thalidomide is an inhibitor of angiogenesis. *Proc Natl Acad Sci U S A*. 1994 Apr 26; 91(9):4082–5. [PubMed: 7513432]
5. Singhal S, Mehta J, Desikan R, Ayers D, Roberson P, Eddlemon P, et al. Antitumor activity of thalidomide in refractory multiple myeloma. *N Engl J Med*. 1999 Nov 18; 341(21):1565–71. [PubMed: 10564685]
6. Glasmacher A, Hahn C, Hoffmann F, Naumann R, Goldschmidt H, von Lilienfeld-Toal M, et al. A systematic review of phase-II trials of thalidomide monotherapy in patients with relapsed or refractory multiple myeloma. *Br J Haematol*. 2006 Mar; 132(5):584–93. [PubMed: 16445831]
7. Wu P, Davies FE, Horton C, Jenner MW, Krishnan B, Alvares CL, et al. The combination of cyclophosphamide, thalidomide and dexamethasone is an effective alternative to cyclophosphamide - vincristine - doxorubicin - methylprednisolone as induction chemotherapy prior to autologous transplantation for multiple myeloma: a case-matched analysis. *Leuk Lymphoma*. 2006 Nov; 47(11):2335–8. [PubMed: 17107906]
8. Dimopoulos MA, Hamilos G, Zomas A, Gika D, Efstathiou E, Grigoraki V, et al. Pulsed cyclophosphamide, thalidomide and dexamethasone: an oral regimen for previously treated patients with multiple myeloma. *Hematol J*. 2004; 5(2):112–7. [PubMed: 15048060]
9. Offidani M, Brinchen S, Corvatta L, Falco P, Marconi M, Avonto I, et al. Thalidomide-dexamethasone plus pegylated liposomal doxorubicin vs. thalidomide-dexamethasone: a case-matched study in advanced multiple myeloma. *Eur J Haematol*. 2007 Apr; 78(4):297–302. [PubMed: 17286608]
10. Palumbo A, Ambrosini MT, Benevolo G, Pregno P, Pescosta N, Callea V, et al. Bortezomib, melphalan, prednisone, and thalidomide for relapsed multiple myeloma. *Blood*. 2007 Apr 1; 109(7):2767–72. [PubMed: 17148584]
11. Rajkumar SV, Blood E, Vesole D, Fonseca R, Greipp PR. Phase III clinical trial of thalidomide plus dexamethasone compared with dexamethasone alone in newly diagnosed multiple myeloma: a clinical trial coordinated by the Eastern Cooperative Oncology Group. *J Clin Oncol*. 2006 Jan 20; 24(3):431–6. [PubMed: 16365178]
12. Chauhan D, Uchiyama H, Akbarali Y, Urashima M, Yamamoto K, Libermann TA, et al. Multiple myeloma cell adhesion-induced interleukin-6 expression in bone marrow stromal cells involves activation of NF-kappa B. *Blood*. 1996 Feb 1; 87(3):1104–12. [PubMed: 8562936]
13. Feinman R, Koury J, Thames M, Barlogie B, Epstein J, Siegel DS. Role of NF-kappaB in the rescue of multiple myeloma cells from glucocorticoid-induced apoptosis by bcl-2. *Blood*. 1999 May 1; 93(9):3044–52. [PubMed: 10216101]
14. Hideshima T, Chauhan D, Richardson P, Mitsiades C, Mitsiades N, Hayashi T, et al. NF-kappa B as a therapeutic target in multiple myeloma. *J Biol Chem*. 2002 May 10; 277(19):16639–47. [PubMed: 11872748]
15. Palombella VJ, Rando OJ, Goldberg AL, Maniatis T. The ubiquitin–proteasome pathway is required for processing the NF-kappa B1 precursor protein and the activation of NF-kappa B. *Cell*. 1994 Sep 9; 78(5):773–85. [PubMed: 8087845]
16. Adams J, Palombella VJ, Sausville EA, Johnson J, Destree A, Lazarus DD, et al. Proteasome inhibitors: a novel class of potent and effective antitumor agents. *Cancer Res*. 1999 Jul 1; 59(11): 2615–22. [PubMed: 10363983]
17. Teicher BA, Ara G, Herbst R, Palombella VJ, Adams J. The proteasome inhibitor PS-341 in cancer therapy. *Clin Cancer Res*. 1999 Oct; 5(9):2638–45. [PubMed: 10499643]
18. Adams J, Palombella VJ, Elliott PJ. Proteasome inhibition: a new strategy in cancer treatment. *Invest New Drugs*. 2000 May; 18(2):109–21. [PubMed: 10857991]
19. Hideshima T, Richardson P, Chauhan D, Palombella VJ, Elliott PJ, Adams J, et al. The proteasome inhibitor PS-341 inhibits growth, induces apoptosis, and overcomes drug resistance in human multiple myeloma cells. *Cancer Res*. 2001 Apr 1; 61(7):3071–6. [PubMed: 11306489]

20. Richardson PG, Sonneveld P, Schuster MW, Irwin D, Stadtmauer EA, Facon T, et al. Bortezomib or high-dose dexamethasone for relapsed multiple myeloma. *N Engl J Med*. 2005 Jun 16; 352(24):2487–98. [PubMed: 15958804]
21. Richardson PG, Barlogie B, Berenson J, Singhal S, Jagannath S, Irwin D, et al. A phase 2 study of bortezomib in relapsed, refractory myeloma. *N Engl J Med*. 2003 Jun 26; 348(26):2609–17. [PubMed: 12826635]
22. Bross PF, Kane R, Farrell AT, Abraham S, Benson K, Brower ME, et al. Approval summary for bortezomib for injection in the treatment of multiple myeloma. *Clin Cancer Res*. 2004 Jun 15; 10(12 Pt 1):3954–64. [PubMed: 15217925]
23. Mitsiades N, Mitsiades CS, Poulaki V, Chauhan D, Richardson PG, Hideshima T, et al. Apoptotic signaling induced by immunomodulatory thalidomide analogs in human multiple myeloma cells: therapeutic implications. *Blood*. 2002 Jun 15; 99(12):4525–30. [PubMed: 12036884]
24. Davies FE, Raje N, Hideshima T, Lentzsch S, Young G, Tai YT, et al. Thalidomide and immunomodulatory derivatives augment natural killer cell cytotoxicity in multiple myeloma. *Blood*. 2001 Jul 1; 98(1):210–6. [PubMed: 11418482]
25. Hideshima T, Chauhan D, Shima Y, Raje N, Davies FE, Tai YT, et al. Thalidomide and its analogs overcome drug resistance of human multiple myeloma cells to conventional therapy. *Blood*. 2000 Nov 1; 96(9):2943–50. [PubMed: 11049970]
26. Richardson PG, Schlossman RL, Weller E, Hideshima T, Mitsiades C, Davies F, et al. Immunomodulatory drug CC-5013 overcomes drug resistance and is well tolerated in patients with relapsed multiple myeloma. *Blood*. 2002 Nov 1; 100(9):3063–7. [PubMed: 12384400]
27. Richardson PG, Blood E, Mitsiades CS, Jagannath S, Zeldenrust SR, Alsina M, et al. A randomized phase 2 study of lenalidomide therapy for patients with relapsed or relapsed and refractory multiple myeloma. *Blood*. 2006 Nov 15; 108(10):3458–64. [PubMed: 16840727]
28. Weber DM, Chen C, Niesvizky R, Wang M, Belch A, Stadtmauer Eea. Lenalidomide plus high-dose dexamethasone provides improved overall survival compared to high-dose dexamethasone alone for relapsed or refractory multiple myeloma (MM): Results of a North American phase III study (MM-009). *J Clin Oncol*. 2006; 24(18_suppl):7521.
29. Gupta D, Treon SP, Shima Y, Hideshima T, Podar K, Tai YT, et al. Adherence of multiple myeloma cells to bone marrow stromal cells upregulates vascular endothelial growth factor secretion: therapeutic applications. *Leukemia*. 2001 Dec; 15(12):1950–61. [PubMed: 11753617]
30. Roccaro AM, Hideshima T, Raje N, Kumar S, Ishitsuka K, Yasui H, et al. Bortezomib mediates antiangiogenesis in multiple myeloma via direct and indirect effects on endothelial cells. *Cancer Res*. 2006 Jan 1; 66(1):184–91. [PubMed: 16397231]
31. Spisek R, Charalambous A, Mazumder A, Vesole DH, Jagannath S, Dhodapkar MV. Bortezomib enhances dendritic cell (DC) mediated induction of immunity to human myeloma via exposure of cell surface heat shock protein 90 on dying tumor cells: therapeutic implications. *Blood*. 2007; 109:4839–4845. [PubMed: 17299090]
32. Zangari M, Esseltine D, Lee CK, Barlogie B, Elice F, Burns MJ, et al. Response to bortezomib is associated to osteoblastic activation in patients with multiple myeloma. *Br J Haematol*. 2005 Oct; 131(1):71–3. [PubMed: 16173965]
33. Breitkreutz I, Raab MS, Vallet S, Hideshima T, Raje N, Chauhan D, et al. Lenalidomide and bortezomib in multiple myeloma: Influence on osteoclast. *Haematologica-the Hematology Journal*. 2007; 92(6):131.
34. Richardson PG, Jagannath S, Avigan DE, Alsina M, Schlossman RL, Mazumder A, et al. Lenalidomide plus bortezomib (Rev-Vel) in relapsed and/or refractory multiple myeloma (MM): Final results of a multicenter phase 1 trial. *Blood*. 2006 Nov; 108(11):124A–A.
35. Richardson RG, Jagannath S, Raje NS, Ghobrial IM, Schlossman RL, Mazumder A, et al. Phase 1/2 study of upfront Rev/Vel/Dex in MM: Early results. *Haematologica-the Hematology Journal*. 2007 Jun.92(6):179.
36. Wang M, Giral S, Delasalle K, Handy B, Alexanian R. Bortezomib in combination with thalidomide-dexamethasone for previously untreated multiple myeloma. *Hematology (Amsterdam, Netherlands)*. 2007 Jun; 12(3):235–9.

37. Chanan-Khan AA, Padmanabhan S, Miller KC, Musiel L, Yu JH, Bernstein ZP, et al. Final results of a phase II study of bortezomib (Velcade) in combination with liposomal doxorubicin (Doxil) and thalidomide (VDT) demonstrate a sustained high response rates in patients (pts) with relapsed (rel) or refractory (ref) multiple myeloma. *Blood*. 2006 Nov; 108(11):1010A–A.
38. Feling RH, Buchanan GO, Mincer TJ, Kauffman CA, Jensen PR, Fenical W. Salinosporamide A: a highly cytotoxic proteasome inhibitor from a novel microbial source, a marine bacterium of the new genus salinospora. *Angew Chem Int Ed Engl*. 2003 Jan 20; 42(3):355–7. [PubMed: 12548698]
39. Chauhan D, Catley L, Li G, Podar K, Hideshima T, Velankar M, et al. A novel orally active proteasome inhibitor induces apoptosis in multiple myeloma cells with mechanisms distinct from Bortezomib. *Cancer cell*. 2005 Nov; 8(5):407–19. [PubMed: 16286248]
40. Demo SD, Kirk CJ, Aujay MA, Buchholz TJ, Dajee M, Ho MN, et al. Antitumor activity of PR-171, a novel irreversible inhibitor of the proteasome. *Cancer Res*. 2007 Jul 1; 67(13):6383–91. [PubMed: 17616698]
41. Stapnes C, Doskeland AP, Hatfield K, Ersvaer E, Rynningen A, Lorens JB, et al. The proteasome inhibitors bortezomib and PR-171 have antiproliferative and proapoptotic effects on primary human acute myeloid leukaemia cells. *Br J Haematol*. 2007 Mar; 136(6):814–28. [PubMed: 17341267]
42. Kuhn DJ, Chen Q, Voorhees PM, Strader JS, Shenk KD, Sun CM, et al. Potent activity of carfilzomib, a novel, irreversible inhibitor of the ubiquitin-proteasome pathway, against pre-clinical models of multiple myeloma. *Blood*. 2007 Jun 25. in press.
43. Alsina M, O'Connor OA, Stewart AK, Trudel S, Urquilla PR, Vallone MK, et al. Clinical responses in multiple myeloma and waldenstrom's macroglobulinemia with the proteasome inhibitor carfilzomib (PR-171). *Haematologica-the Hematology Journal*. 2007 Jun.92(6):156.
44. Mitsiades CS, Mitsiades NS, McMullan CJ, Poulaki V, Kung AL, Davies FE, et al. Antimyeloma activity of heat shock protein-90 inhibition. *Blood*. 2006 Feb 1; 107(3):1092–100. [PubMed: 16234364]
45. Mitsiades N, Mitsiades CS, Poulaki V, Chauhan D, Fanourakis G, Gu X, et al. Molecular sequelae of proteasome inhibition in human multiple myeloma cells. *Proc Natl Acad Sci U S A*. 2002 Oct 29; 99(22):14374–9. [PubMed: 12391322]
46. Duus J, Bahar HI, Venkataraman G, Ozpuyan F, Izban KF, Al-Masri H, et al. Analysis of expression of heat shock protein-90 (HSP90) and the effects of HSP90 inhibitor (17-AAG) in multiple myeloma. *Leuk Lymphoma*. 2006 Jul; 47(7):1369–78. [PubMed: 16923571]
47. Sydor JR, Normant E, Pien CS, Porter JR, Ge J, Grenier L, et al. Development of 17-allylamino-17-demethoxygeldanamycin hydroquinone hydrochloride (IPI-504), an anti-cancer agent directed against Hsp90. *Proc Natl Acad Sci U S A*. 2006 Nov 14; 103(46):17408–13. [PubMed: 17090671]
48. Chatterjee M, Jain S, Stuhmer T, Andrulis M, Ungethüm U, Kuban RJ, et al. STAT3 and MAPK signaling maintain overexpression of heat shock proteins 90alpha and beta in multiple myeloma cells, which critically contribute to tumor-cell survival. *Blood*. 2007 Jan 15; 109(2):720–8. [PubMed: 17003370]
49. Francis LK, Alsayed Y, Leleu X, Jia X, Singha UK, Anderson J, et al. Combination mammalian target of rapamycin inhibitor rapamycin and HSP90 inhibitor 17-allylamino-17-demethoxygeldanamycin has synergistic activity in multiple myeloma. *Clin Cancer Res*. 2006 Nov 15; 12(22):6826–35. [PubMed: 17121904]
50. Richardson PG, Chanan-Khan AA, Alsina M, Doss D, Landrigan B, Kettner D, et al. Safety and activity of KOS-953 in patients with relapsed refractory multiple myeloma (MM): Interim results of a phase 1 trial. *Blood*. 2005; 106(11):109a.
51. Richardson P, Chanan-Khan A, Lonial S, Krishnan A, Alsina M, Carroll M, et al. A Multicenter Phase 1 Clinical Trial of Tanespimycin (KOS-953) + Bortezomib (BZ): Encouraging Activity and Manageable Toxicity in Heavily Pre-Treated Patients with Relapsed Refractory Multiple Myeloma (MM). 2006 Annual Meeting of the American Society of Hematology; 2006; Orlando FL. p. 2006

52. Chauhan D, Catley L, Hideshima T, Li G, Leblanc R, Gupta D, et al. 2-Methoxyestradiol overcomes drug resistance in multiple myeloma cells. *Blood*. 2002 Sep 15; 100(6):2187–94. [PubMed: 12200384]
53. Rajkumar SV, Richardson PG, Gertz MA, Lacy MQ, Dispenzieri A, Greipp PR, et al. Novel therapy with 2-methoxyestradiol (2ME2) for the treatment of relapsed and plateau phase multiple myeloma. *Blood*. 2003 Nov; 102(11):692A–A.
54. Marks P, Rifkind RA, Richon VM, Breslow R, Miller T, Kelly WK. Histone deacetylases and cancer: causes and therapies. *Nature Rev Cancer*. 2001 Dec; 1(3):194–202. [PubMed: 11902574]
55. Johnstone RW. Histone-deacetylase inhibitors: novel drugs for the treatment of cancer. *Nat Rev Drug Discov*. 2002 Apr; 1(4):287–99. [PubMed: 12120280]
56. Mitsiades N, Mitsiades CS, Richardson PG, McMullan C, Poulaki V, Fanourakis G, et al. Molecular sequelae of histone deacetylase inhibition in human malignant B cells. *Blood*. 2003 May 15; 101(10):4055–62. [PubMed: 12531799]
57. Mitsiades CS, Mitsiades NS, McMullan CJ, Poulaki V, Shringarpure R, Hideshima T, et al. Transcriptional signature of histone deacetylase inhibition in multiple myeloma: biological and clinical implications. *Proc Natl Acad Sci U S A*. 2004 Jan 13; 101(2):540–5. [PubMed: 14695887]
58. Richardson P, Schlossman RL, Mitsiades CS, Munshi NC, Colson K, Doss D, et al. Phase I clinical trial of oral administration of the histone deacetylase (HDAC) inhibitor suberoylanilide hydroxamic acid (SAHA) in patients with relapsed/refractory multiple myeloma. *Blood*. 2004 Nov; 104(11):420A–A. [PubMed: 15054037]
59. Catley L, Weisberg E, Tai YT, Atadja P, Remiszewski S, Hideshima T, et al. NVP-LAQ824 is a potent novel histone deacetylase inhibitor with significant activity against multiple myeloma. *Blood*. 2003 Oct 1; 102(7):2615–22. [PubMed: 12816865]
60. Catley L, Weisberg E, Kiziltepe T, Tai YT, Hideshima T, Neri P, et al. Aggresome induction by proteasome inhibitor bortezomib and alpha-tubulin hyperacetylation by tubulin deacetylase (TDAC) inhibitor LBH589 are synergistic in myeloma cells. *Blood*. 2006 Nov 15; 108(10):3441–9. [PubMed: 16728695]
61. Maiso P, Carvajal-Vergara X, Ocio EM, Lopez-Perez R, Mateo G, Gutierrez N, et al. The histone deacetylase inhibitor LBH589 is a potent antimyeloma agent that overcomes drug resistance. *Cancer Res*. 2006 Jun 1; 66(11):5781–9. [PubMed: 16740717]
62. Hideshima T, Bradner JE, Wong J, Chauhan D, Richardson P, Schreiber SL, et al. Small-molecule inhibition of proteasome and aggresome function induces synergistic antitumor activity in multiple myeloma. *Proc Natl Acad Sci U S A*. 2005 Jun 14; 102(24):8567–72. [PubMed: 15937109]
63. Soignet SL, Maslak P, Wang ZG, Jhanwar S, Calleja E, Dardashti LJ, et al. Complete remission after treatment of acute promyelocytic leukemia with arsenic trioxide. *N Engl J Med*. 1998 Nov 5; 339(19):1341–8. [PubMed: 9801394]
64. Hayashi T, Hideshima T, Akiyama M, Richardson P, Schlossman RL, Chauhan D, et al. Arsenic trioxide inhibits growth of human multiple myeloma cells in the bone marrow microenvironment. *Mol Cancer Ther*. 2002 Aug; 1(10):851–60. [PubMed: 12492118]
65. Hussein MA, Saleh M, Ravandi F, Mason J, Rifkin RM, Ellison R. Phase 2 study of arsenic trioxide in patients with relapsed or refractory multiple myeloma. *Br J Haematol*. 2004 May; 125(4):470–6. [PubMed: 15142117]
66. Berenson JR, Boccia R, Siegel D, Bozdech M, Bessudo A, Stadtmauer E, et al. Efficacy and safety of melphalan, arsenic trioxide and ascorbic acid combination therapy in patients with relapsed or refractory multiple myeloma: a prospective, multicentre, phase II, single-arm study. *Br J Haematol*. 2006 Oct; 135(2):174–83. [PubMed: 17010047]
67. Berenson JR, Matous J, Swift RA, Mapes R, Morrison B, Yeh HS. A phase I/II study of arsenic trioxide/bortezomib/ascorbic acid combination therapy for the treatment of relapsed or refractory multiple myeloma. *Clin Cancer Res*. 2007 Mar 15; 13(6):1762–8. [PubMed: 17363530]
68. Hideshima T, Nakamura N, Chauhan D, Anderson KC. Biologic sequelae of interleukin-6 induced PI3-K/Akt signaling in multiple myeloma. *Oncogene*. 2001 Sep 20; 20(42):5991–6000. [PubMed: 11593406]
69. Hsu J, Shi Y, Krajewski S, Renner S, Fisher M, Reed JC, et al. The AKT kinase is activated in multiple myeloma tumor cells. *Blood*. 2001 Nov 1; 98(9):2853–5. [PubMed: 11675360]

70. Hsu JH, Shi Y, Hu L, Fisher M, Franke TF, Lichtenstein A. Role of the AKT kinase in expansion of multiple myeloma clones: effects on cytokine-dependent proliferative and survival responses. *Oncogene*. 2002 Feb 21; 21(9):1391–400. [PubMed: 11857082]
71. Mitsiades CS, Mitsiades N, Poulaki V, Schlossman R, Akiyama M, Chauhan D, et al. Activation of NF-kappaB and upregulation of intracellular anti-apoptotic proteins via the IGF-1/Akt signaling in human multiple myeloma cells: therapeutic implications. *Oncogene*. 2002 Aug 22; 21(37):5673–83. [PubMed: 12173037]
72. Akiyama M, Hideshima T, Hayashi T, Tai YT, Mitsiades CS, Mitsiades N, et al. Cytokines modulate telomerase activity in a human multiple myeloma cell line. *Cancer Res*. 2002 Jul 1; 62(13):3876–82. [PubMed: 12097303]
73. Hideshima T, Catley L, Yasui H, Ishitsuka K, Raje N, Mitsiades C, et al. Perifosine, an oral bioactive novel alkylphospholipid, inhibits Akt and induces in vitro and in vivo cytotoxicity in human multiple myeloma cells. *Blood*. 2006 May 15; 107(10):4053–62. [PubMed: 16418332]
74. Gajate C, Mollinedo F. Edelfosine and perifosine induce selective apoptosis in multiple myeloma by recruitment of death receptors and downstream signaling molecules into lipid rafts. *Blood*. 2007 Jan 15; 109(2):711–9. [PubMed: 17003375]
75. Kiziltepe T, Hideshima T, Ishitsuka K, Ocio EM, Raje N, Catley L, et al. JS-K, a GST-activated nitric oxide generator, induces DNA double strand breaks, activates DNA damage response pathways, and induces apoptosis in vitro and in vivo in human multiple myeloma cells. *Blood*. 2007; 110(2):709–718. [PubMed: 17384201]
76. Richardson PG, Lonial S, Jakubowiak A, Krishnan A, Wolf J, Singhal S, et al. Perifosine (KRX-0401) plus low dose dexamethasone is active in patients with relapsed and refractory multiple myeloma (MM): Perifosine MM investigator group phase II multicenter study update. *Haematologica-the Hematology Journal*. 2007 Jun; 92(6):155.
77. Richardson PG, Jakubowiak A, Wolf J, Krishnan A, Lonial S, Ghobrial I, et al. A multicenter phase I/II trial of perifosine (KRX-0401) + bortezomib in relapsed and refractory multiple myeloma patients previously treated with bortezomib: Preliminary results. *Haematologica-the Hematology Journal*. 2007 Jun; 92(6):155–6.
78. Jakubowiak A, Richardson P, Zimmerman T, Alsina M, Lonial S, Kendall T, et al. A multicenter phase I trial of perifosine (KRX-0401) in combination with lenalidomide and dexamethasone in patients with relapsed or refractory multiple myeloma: Preliminary results. Multiple myeloma research consortium (MMRC) trial. *Haematologica-the Hematology Journal*. 2007 Jun; 92(6):154–5.
79. Chesi M, Nardini E, Brents LA, Schrock E, Ried T, Kuehl WM, et al. Frequent translocation t(4;14)(p16.3;q32.3) in multiple myeloma is associated with increased expression and activating mutations of fibroblast growth factor receptor 3. *Nat Genet*. 1997 Jul; 16(3):260–4. [PubMed: 9207791]
80. Rasmussen T, Hudlebusch HR, Knudsen LM, Johnsen HE. FGFR3 dysregulation in multiple myeloma: frequency and prognostic relevance. *Br J Haematol*. 2002 Jun; 117(3):626–8. [PubMed: 12028033]
81. Keats JJ, Reiman T, Maxwell CA, Taylor BJ, Larratt LM, Mant MJ, et al. In multiple myeloma, t(4;14)(p16;q32) is an adverse prognostic factor irrespective of FGFR3 expression. *Blood*. 2003 Feb 15; 101(4):1520–9. [PubMed: 12393535]
82. Xin X, Abrams TJ, Hollenbach PW, Rendahl KG, Tang Y, Oei YA, et al. CHIR-258 is efficacious in a newly developed fibroblast growth factor receptor 3-expressing orthotopic multiple myeloma model in mice. *Clin Cancer Res*. 2006 Aug 15; 12(16):4908–15. [PubMed: 16914579]
83. Trudel S, Stewart AK, Rom E, Wei E, Li ZH, Kotzer S, et al. The inhibitory anti-FGFR3 antibody, PRO-001, is cytotoxic to t(4;14) multiple myeloma cells. *Blood*. 2006 May 15; 107(10):4039–46. [PubMed: 16467200]
84. Albrightson-Winslow CR, Brickson B, King A, Olivera D, Short B, Saunders C, et al. Beneficial effects of long-term treatment with SK&F 105685 in murine lupus nephritis. *J Pharmacol Exp Ther*. 1990 Oct; 255(1):382–7. [PubMed: 2145424]
85. Badger AM, Swift BA, Webb EF, Clark RK, Bugelski PJ, Griswold DE. Beneficial effects of SK&F 105685 in rat adjuvant arthritis: prophylactic and therapeutic effects on disease parameter progression. *Int J Immunopharmacol*. 1993 Apr; 15(3):343–52. [PubMed: 8505147]

86. Rajkumar SV, Fonseca R, Witzig TE, Gertz MA, Greipp PR. Bone marrow angiogenesis in patients achieving complete response after stem cell transplantation for multiple myeloma. *Leukemia*. 1999 Mar; 13(3):469–72. [PubMed: 10086738]
87. Vacca A, Ribatti D, Roncali L, Ranieri G, Serio G, Silvestris F, et al. Bone marrow angiogenesis and progression in multiple myeloma. *Br J Haematol*. 1994 Jul; 87(3):503–8. [PubMed: 7527645]
88. Podar K, Tai YT, Davies FE, Lentzsch S, Sattler M, Hideshima T, et al. Vascular endothelial growth factor triggers signaling cascades mediating multiple myeloma cell growth and migration. *Blood*. 2001 Jul 15; 98(2):428–35. [PubMed: 11435313]
89. Podar K, Shringarpure R, Tai YT, Simoncini M, Sattler M, Ishitsuka K, et al. Caveolin-1 is required for vascular endothelial growth factor-triggered multiple myeloma cell migration and is targeted by bortezomib. *Cancer Res*. 2004 Oct 15; 64(20):7500–6. [PubMed: 15492276]
90. Podar K, Tonon G, Sattler M, Tai YT, Legouill S, Yasui H, et al. The small-molecule VEGF receptor inhibitor pazopanib (GW786034B) targets both tumor and endothelial cells in multiple myeloma. *Proc Natl Acad Sci U S A*. 2006 Dec 19; 103(51):19478–83. [PubMed: 17164332]
91. Podar K, Catley LP, Tai YT, Shringarpure R, Carvalho P, Hayashi T, et al. GW654652, the pan-inhibitor of VEGF receptors, blocks the growth and migration of multiple myeloma cells in the bone marrow microenvironment. *Blood*. 2004 May 1; 103(9):3474–9. [PubMed: 14644994]
92. Lin B, Podar K, Gupta D, Tai YT, Li S, Weller E, et al. The vascular endothelial growth factor receptor tyrosine kinase inhibitor PTK787/ZK222584 inhibits growth and migration of multiple myeloma cells in the bone marrow microenvironment. *Cancer Res*. 2002 Sep 1; 62(17):5019–26. [PubMed: 12208756]
93. Zangari M, Anaissie E, Stopeck A, Morimoto A, Tan N, Lancet J, et al. Phase II study of SU5416, a small molecule vascular endothelial growth factor tyrosine kinase receptor inhibitor, in patients with refractory multiple myeloma. *Clin Cancer Res*. 2004 Jan 1; 10(1 Pt 1):88–95. [PubMed: 14734456]
94. Kovacs MJ, Reece DE, Marcellus D, Meyer RM, Mathews S, Dong RP, et al. A phase II study of ZD6474 (Zactima, a selective inhibitor of VEGFR and EGFR tyrosine kinase in patients with relapsed multiple myeloma--NCIC CTG IND.145. *Invest New Drugs*. 2006 Nov; 24(6):529–35. [PubMed: 16791411]
95. Prince HM, Honemann D, Spencer A, Rizzieri D, Stadtmauer EA, Roberts A, et al. VEGF-R inhibition with pazopanib (GW786034) is ineffective in pretreated myeloma. *Haematologica-the Hematology Journal*. 2007 Jun; 92(6):153.
96. Frost P, Moatamed F, Hoang B, Shi Y, Gera J, Yan H, et al. In vivo antitumor effects of the mTOR inhibitor CCI-779 against human multiple myeloma cells in a xenograft model. *Blood*. 2004 Dec 15; 104(13):4181–7. [PubMed: 15304393]
97. Raje N, Kumar S, Hideshima T, Ishitsuka K, Chauhan D, Mitsiades C, et al. Combination of the mTOR inhibitor rapamycin and CC-5013 has synergistic activity in multiple myeloma. *Blood*. 2004 Dec 15; 104(13):4188–93. [PubMed: 15319277]
98. Mitsiades CS, Mitsiades NS, McMullan CJ, Poulaki V, Shringarpure R, Akiyama M, et al. Inhibition of the insulin-like growth factor receptor-1 tyrosine kinase activity as a therapeutic strategy for multiple myeloma, other hematologic malignancies, and solid tumors. *Cancer cell*. 2004 Mar; 5(3):221–30. [PubMed: 15050914]
99. Menu E, Jernberg-Wiklund H, De Raevé H, De Leenheer E, Coulton L, Gallagher O, et al. Targeting the IGF-1R using picropodophyllin in the therapeutical 5T2MM mouse model of multiple myeloma: Beneficial effects on tumor growth, angiogenesis, bone disease and survival. *International journal of cancer*. 2007 Jun; 117:17546599.
100. Hideshima T, Neri P, Tassone P, Yasui H, Ishitsuka K, Raje N, et al. MLN120B, a novel I κ B kinase beta inhibitor, blocks multiple myeloma cell growth in vitro and in vivo. *Clin Cancer Res*. 2006 Oct 1; 12(19):5887–94. [PubMed: 17020997]
101. Jourdan M, Moreaux J, Vos JD, Hose D, Mahtouk K, Abouladze M, et al. Targeting NF- κ B pathway with an IKK2 inhibitor induces inhibition of multiple myeloma cell growth. *British journal of haematology*. 2007 Jul; 138(2):160–8. [PubMed: 17542984]

102. Mateo G, Castellanos M, Rasillo A, Gutierrez NC, Montalban MA, Martin ML, et al. Genetic abnormalities and patterns of antigenic expression in multiple myeloma. *Clin Cancer Res*. 2005 May 15; 11(10):3661–7. [PubMed: 15897562]
103. Bergsagel PL, Masellis Smith A, Belch AR, Pilarski LM. The blood B-cells and bone marrow plasma cells in patients with multiple myeloma share identical IgH rearrangements. *Curr Top Microbiol Immunol*. 1995; 194:17–24. [PubMed: 7534667]
104. Bergsagel PL, Smith AM, Szczeppek A, Mant MJ, Belch AR, Pilarski LM. In multiple myeloma, clonotypic B lymphocytes are detectable among CD19+ peripheral blood cells expressing CD38, CD56, and monotypic Ig light chain. *Blood*. 1995 Jan 15; 85(2):436–47. [PubMed: 7529064]
105. Treon SP, Pilarski LM, Belch AR, Kelliher A, Preffer FI, Shima Y, et al. CD20-directed serotherapy in patients with multiple myeloma: biologic considerations and therapeutic applications. *J Immunother*. 2002 Jan; Feb; 25(1):72–81. [PubMed: 11924912]
106. Moreau P, Voillat L, Benboukher L, Mathiot C, Dumontet C, Robillard N, et al. Rituximab in CD20 positive multiple myeloma. *Leukemia*. 2007 Apr; 21(4):835–6. [PubMed: 17268523]
107. Zojer N, Kirchbacher K, Vesely M, Hubl W, Ludwig H. Rituximab treatment provides no clinical benefit in patients with pretreated advanced multiple myeloma. *Leuk Lymphoma*. 2006 Jun; 47(6):1103–9. [PubMed: 16840202]
108. Tassone P, Goldmacher VS, Neri P, Gozzini A, Shammass MA, Whiteman KR, et al. Cytotoxic activity of the maytansinoid immunoconjugate B-B4-DM1 against CD138+ multiple myeloma cells. *Blood*. 2004 Dec 1; 104(12):3688–96. [PubMed: 15292058]
109. Tai YT, Li XF, Catley L, Coffey R, Breitzkreutz I, Bae J, et al. Immunomodulatory drug lenalidomide (CC-5013, IMiD3) augments anti-CD40 SGN-40-induced cytotoxicity in human multiple myeloma: clinical implications. *Cancer Res*. 2005 Dec 15; 65(24):11712–20. [PubMed: 16357183]
110. Tai YT, Li X, Tong X, Santos D, Otsuki T, Catley L, et al. Human anti-CD40 antagonist antibody triggers significant antitumor activity against human multiple myeloma. *Cancer Res*. 2005 Jul 1; 65(13):5898–906. [PubMed: 15994968]
111. Tai YT, Catley LP, Mitsiades CS, Burger R, Podar K, Shringpaure R, et al. Mechanisms by which SGN-40, a humanized anti-CD40 antibody, induces cytotoxicity in human multiple myeloma cells: clinical implications. *Cancer Res*. 2004 Apr 15; 64(8):2846–52. [PubMed: 15087402]
112. Tai YT, Podar K, Mitsiades N, Lin B, Mitsiades C, Gupta D, et al. CD40 induces human multiple myeloma cell migration via phosphatidylinositol 3-kinase/AKT/NF-kappa B signaling. *Blood*. 2003 Apr 1; 101(7):2762–9. [PubMed: 12433678]
113. Hayashi T, Treon SP, Hideshima T, Tai YT, Akiyama M, Richardson P, et al. Recombinant humanized anti-CD40 monoclonal antibody triggers autologous antibody-dependent cell-mediated cytotoxicity against multiple myeloma cells. *Br J Haematol*. 2003 May; 121(4):592–6. [PubMed: 12752100]
114. Stevenson GT. CD38 as a therapeutic target. *Mol Med*. 2006 Nov-Dec; 12(11-12):345–6. [PubMed: 17380203]
115. Ellis JH, Barber KA, Tutt A, Hale C, Lewis AP, Glennie MJ, et al. Engineered anti-CD38 monoclonal antibodies for immunotherapy of multiple myeloma. *J Immunol*. 1995 Jul 15; 155(2):925–37. [PubMed: 7608568]
116. Hsi ED, Steinle R, Balasa B, Draksharapu A, Shum B, Huseni M, et al. CS1: A potential new therapeutic target for the treatment of multiple myeloma. *Blood*. 2006 Nov; 108(11):986A–A. [PubMed: 16621969]
117. Rice A, Dillon M, van Abbema A, Jesaitis L, Wong M, Lawson S, et al. Eradication of tumors in pre-clinical models of multiple myeloma by Anti-CS1 monoclonal antibody HuLuc63: Mechanism of action studies. *Blood*. 2006 Nov; 108(11):999A–A.
118. Tai YT, Song W, Li XF, Burger P, Lee A, Leiba M, et al. Targeting CS1, a newly identified human multiple myeloma antigen, by a novel humanized monoclonal antibody HuLuc63, blocks myeloma cell adhesion in the bone marrow microenvironment and induces potent cytotoxicity against myeloma cells even resistant to conventional and novel therapies. *Haematologica-the Hematology Journal*. 2007 Jun; 92(6):148.

119. Mitsiades CS, Rouleau C, Menon K, Teicher B, Iacobelli M, Anderson KC, et al. Defibrotide (DF) targets tumor-microenvironmental interactions and sensitizes multiple myeloma and solid tumor cells to cytotoxic chemotherapeutics. *Blood*. 2004 Nov; 104(11):85A–A.
120. Palumbo A, Larocca A, Rus C, Gay E, Rossi D, Pregno P, et al. Efficacy and safety of melphalan, prednisone, thalidomide and defibrotide in relapsed and refractory multiple myeloma patients: Results of a multicenter phase I/II trial. *Haematologica-the Hematology Journal*. 2007 Jun.92:94.

Practice points

the immunomodulatory drugs thalidomide and lenalidomide, and the proteasome inhibitor bortezomib, are now appropriate first-line agents for the treatment of MM

multi-agent regimens involving immunomodulatory drugs and proteasome inhibitors and/or conventional chemotherapeutic agents achieve high overall response rates and are appropriate choices for treatment at any stage of the disease

given recent evidence for increased morbidity and even mortality in association with the use of high-dose dexamethasone, there is greater emphasis on reduced doses or even corticosteroid-free combinations

novel targeted therapies are also increasingly being integrated into long-term transplant-based care protocols

individualized therapies based on classification systems derived from cytogenetics and gene expression profiling are likely to become more widespread in the years ahead

Research agenda

insights based on gene expression profiling and knowledge of the tumor–microenvironment interactions will continue to inform the development of novel therapies

more multi-targeted agents such as non-specific kinase inhibitors are likely to undergo preclinical evaluation

research efforts will include a more thorough evaluation of molecular targets (e.g. the kinases) to better define valid ‘druggable’ targets in MM

a further refining of molecular profiling-based risk stratification will facilitate more precise individualization of patient treatment plans

reduction of toxicity and enhancement of therapeutic index require further consideration

Figure 1.

Schematic representation of the integration of key signal transduction cascades in multiple myeloma (MM) cells and their interaction with non-malignant cells of the bone-marrow microenvironment. The molecular and cellular levels of action of some of the novel anti-MM agents in clinical or preclinical development are highlighted. Hsp90, heat shock protein 90; FGFR3, fibroblast growth factor receptor 3; IL-6(R), interleukin 6 (receptor); IGF-1(R), insulin-like growth factor 1 (receptor); VEGF(R), vascular endothelial growth factor (receptor); IKK, IκB kinase; SDF-1 α, stromal cell-derived factor 1α; BAFF, B-cell-activating factor; APRIL, a proliferation-inducing ligand; TNF α, tumor necrosis factor α; TGFβ, tumor growth factor β; NF-κB, nuclear factor κB; BMSC, bone-marrow stromal cell; MUC-1, mucin 1; LFA-1, lymphocyte function-associated antigen 1; ICAM-1, intercellular adhesion molecule 1; VLA-4, very late antigen 4; VCAM-1, vascular cell adhesion molecule 1; HGF, hepatocyte growth factor; BMEC, bone-marrow microvascular endothelial cell; bFGF, basic fibroblast growth factor; 2ME2, 2-methoxyestradiol; MIP-1 α, macrophage inflammatory protein 1α; DKK1, dickkopf-1; HDAC, histone deacetylase.