Thalidomide as a novel therapeutic agent: new uses for an old product

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Thalidomide and its immunomodulatory analogues have numerous effects on the body's immune system, including potential anti-cancer and anti-inflammatory activities. Thalidomide is currently used experimentally to treat various cancers, dermatological, neurological and inflammatory diseases. This drug is approved in the USA for cutaneous manifestations of lepromatous leprosy and is in Phase III trials for multiple myeloma. Thalidomide and its analogues modulate the immune system in various ways. Some of these immunomodulatory activities, together with the anti-angiogenic, anti-proliferative and pro-apoptotic properties, are believed to mediate anti-tumor responses as observed in multiple myeloma and some solid tumors. The analogue lenalidomide has shown potential in treating the bone marrow disorders multiple myeloma and myelodysplastic syndrome, and is presently in Phase II and III trials, respectively.

In August 1998, thalidomide (Figure 1) was approved for sale in the USA for the chronic treatment of erythema nodosum leprosum (ENL), a painful inflammatory dermatological reaction of lepromatous leprosy [1]. This marked the approval of the world's most controversial drug after it was withdrawn from Europe and other countries over 40 years ago. Thalidomide was never approved in the USA (see Ref. [2] for a more comprehensive history of thalidomide). Thalidomide was first synthesized in 1954 from the glutamic acid derivative α-phthaloylisoglutamine. Soon thereafter, limited animal studies showed it to be non-toxic. It was erroneously concluded then that the purported structural resemblance to the then widely-used barbiturates could indicate its potential as a 'safe' sedative [2]. A single questionable study in mice was performed to show the sedative hypnotic effects of thalidomide [2]. Based on this study, human trials were initiated in Germany under the then lax pharmaceutical regulatory environment without comprehensive animal toxicology studies. These should have included reproductive toxicology in a non-rodent species such as rabbits or monkeys because studies in rodents have shown limited sensitivity to the teratogenic effects of thalidomide *in utero* [3]. Thalidomide was found to be an effective sedative and sleep-inducing agent in humans with less potential for death due to apnea from an overdose when compared with the potential for death from overdosing on barbiturates. Thalidomide was approved in Germany in 1957 and subsequently in other countries including the UK, Canada and Australia under brand names such as Contergan, Distaval, Talimol and Kevadon. Thalidomide was also found to be an effective anti-emetic in pregnancy and its use in this group of patients subsequently increased. The error in this presumption of good sedative and anti-emetic efficacy with limited toxicity became apparent when reports of deformed babies started appearing after 1956. By the time it was withdrawn in 1961, ~5000–12 000 deformed babies (and an unknown number of aborted fetuses) from 46 countries were

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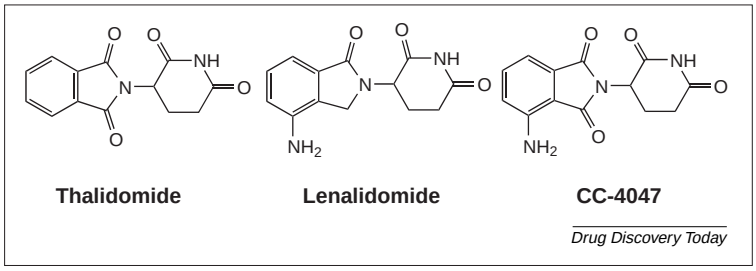

FIGURE 1

Structures of thalidomide and immunomodulatory analogues. Using the inhibition of the pro-inflammatory cytokine TNF-α as basis for comparison, the analogues are at least 2000 times more potent than thalidomide.
Abbreviation: TNF-α, tumor necrosis factor-α.

already born [4]. Thalidomide was not approved in the USA because of the diligence of the Food and Drug Administration (FDA) reviewer Frances Kelsey who requested more information from the petitioning company on the reported peripheral neuritis [5]. The company was not forthcoming and the application was subsequently withdrawn. This would have been the end of the drug were it not for subsequent reports of its effectiveness in treating various inflammatory and dermatological conditions such as ENL. In 1964, Jacob Sheskin, an Israeli doctor, experimentally prescribed thalidomide to an advanced ENL patient wracked with pain and concomitant insomnia [6]. The results were dramatic. The patient slept soundly with no subsequent hangover, and his ENL-related pain and fever resolved entirely and the cutaneous sores healed within days. The ENL symptoms were kept at bay as long as the patient was taking thalidomide. The therapeutic effect was confirmed with other ENL patients (Figure 2). Thalidomide was found to be so effective in treating ENL that it is now the World Health Organization's recommended drug for this form of leprosy [7].

This review highlights and presents some of the therapeutic activities observed with thalidomide and its immunomodulatory analogues (IMID®s) (Figure 1), including the compounds' surprising anti-cancer activity in hematological cancers such as myelodysplastic syndrome and multiple myeloma, and in various solid tumor cancers. Their activities in various inflammatory diseases are described, in addition to a discussion of the role of immunomodulatory and non-immunomodulatory mechanisms for the efficacy.

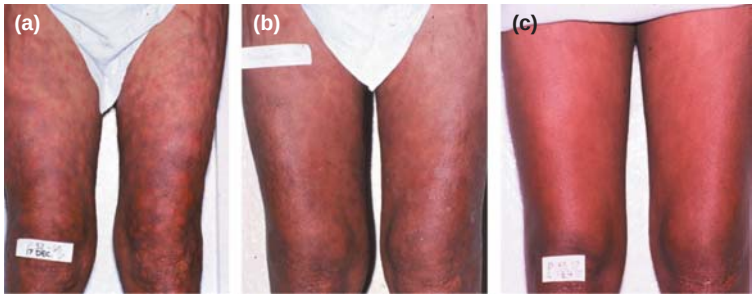

FIGURE 2

Efficacy of thalidomide in a patient with erythema nodosum leprosum. Thalidomide is now the World Health Organization's recommended drug for ENL and was approved in the USA for this indication in 1998. **(a)** Before treatment with thalidomide. **(b)** After one week treatment with thalidomide (dosage of 200 mg day^{-1}). **(c)** After six weeks treatment with thalidomide (dosage of 200 mg day^{-1}).
Abbreviation: ENL, erythema nodosum leprosum.

Current experimental uses of thalidomide

Apart from its approved use in ENL, thalidomide is prescribed experimentally, mostly co-administered with standard therapies, and is used in >150 clinical trials in the USA for various oncological, dermatological and inflammatory conditions [8–11]. Therapeutic doses are 200–400 mg day^{-1} (4–8 mg kg^{-1} day^{-1}) for oncological conditions, and 100–400 mg day^{-1} (2–8 mg kg^{-1} day^{-1}) for dermatological and inflammatory diseases. Adverse events associated with use of thalidomide, some of which can be dose-related, include somnolence, constipation, rash, peripheral neuropathy and deep vein thrombosis [12]. Whereas reducing or stopping thalidomide treatment can alleviate most of these adverse events, the peripheral neuropathy can be permanent.

Anti-cancer activity

Potential activity has been observed in clinical trials for various hematological and solid tumor cancers including relapsed and/or refractory multiple myeloma [13,14], myelodysplastic syndrome [15], mantle cell lymphoma [16], glioma [17,18], renal cell carcinoma [19], metastatic melanoma [20,21], pancreatic cancer [22] and androgen-independent prostate cancer [23,24]. Confirmatory studies are in progress for some of these cancers. The FDA is currently considering approval of thalidomide for newly diagnosed multiple myeloma. Multiple myeloma is an incurable B or plasma cell malignancy of the bone marrow accounting for 1–2% of all cancer and 10% of new hematological malignancies in the USA. It is diagnosed in ~15 000 patients annually in the USA and is characterized by the secretion of monoclonal proteins or immunoglobulins (M protein or paraprotein). Most patients die within five years of diagnosis because of the limited treatment options and the relapsing and refractory nature of this disease [25]. Thalidomide in heavily pre-treated relapsed multiple myeloma patients showed a total response rate ranging from 32% [13] to 49% [14]. Thalidomide has an apparent synergistic activity when used in combination with dexamethasone in newly diagnosed and relapsed and/or refractory multiple myeloma, and could even reduce the median response time when compared with the response time from thalidomide alone [26,27]. This combination has been shown to be significantly better with three-year survival at 60% for thalidomide–dexamethasone versus 26% for non-thalidomide conventional chemotherapy, and is being used as the first salvage regimen in relapsed and/or refractory patients [28]. Thalidomide use with

melphalan and prednisone in newly diagnosed patients has also showed encouraging results [29].

Significant activity has been seen in another bone marrow disease called myelodysplastic syndrome (MDS) affecting ~13 000 new patients each year in the USA. In MDS, bone marrow function is abnormal with defects in the maturation of various hematological cells. Anemia that might require blood transfusion is the most common disease manifestation. MDS could eventually change into acute myeloid leukemia [30]. A 2001 study showed that single-agent thalidomide could induce hematological improvement, in particular, increasing hemoglobin levels in some refractory patients and making them transfusion independent [15]. Of 51 patients that completed 12 weeks of thalidomide, 16 developed hematological improvement of which ten became transfusion-independent. In patients with glioblastoma multiforme, a thalidomide–temozolomide combination was found to have longer (103 weeks) survival compared with that from thalidomide alone (63 weeks) [18]. In addition, results from a study using thalidomide–cyclophosphamide combination also showed encouraging activity with one out of 11 patients demonstrating a complete response and one each with partial and stable disease [17]. This thalidomide–temozolomide combination was also determined to be promising in metastatic malignant melanoma with median survival of 7.3 months compared with 5.3 months for temozolomide alone [21]. In a review of thalidomide use in metastatic renal cell carcinoma, thalidomide in combination with other chemotherapeutics [interleukin 2 (IL-2), interferon γ (IFN-γ), capecitabine] had anti-tumor activity [19]. Thalidomide in combination with gemcitabine could also have potential benefit in advanced pancreatic cancer. The time to disease progression was greater than historical data from the single agent gemcitabine [22]. Nine and four out of 27 evaluable patients in the combination regimen survived for more than six and twelve months, respectively, compared with a median survival of four to six months with gemcitabine alone. A Phase II study of thalidomide in combination with docetaxel in androgen-independent prostate cancer had an 18-month survival of 68% versus 43% for docetaxel alone [23]. Up to 53% of patients in the combination regimen had >50% decline in the prostrate-specific antigen (PSA) compared with 37% in the docetaxel group. All these anti-cancer activities are could be mediated by immunomodulatory and non-immunomodulatory mechanisms.

Dermatological and anti-inflammatory activities

Thalidomide has activity in various dermatological conditions [10,31] and was tried on other dermatological conditions after its beneficial effects on cutaneous manifestations of ENL were discovered. The activity of thalidomide has been determined in Behcet's disease, prurigo nodularis, aphthous ulcers and actinic prurigo who have unsuccessfully tried other therapies. In addition, thalidomide has activity in discoid lupus and the skin manifestations of systemic lupus erythematosus. A recent study showed that low-dose thalidomide induced complete resolution of the cutaneous erythema in 17 out of 23 (78%) refractory erythematosus patients [32]. Activity has also been reported in refractory Crohn's disease [33]. Studies have shown remissions and reduction and/or discontinuation of steroid use in some patients. It is not known exactly how thalidomide produces these dermatological and anti-inflammatory activities, but it is believed to involve immunomodulatory mechanisms.

Pain

A surprising thalidomide effect is in complex regional pain syndrome (CRPS), otherwise known as reflex sympathetic dystrophy, which is characterized by neuropathic pain, allodynia, edema, autonomic dysfunction, disordered movements, dystrophy and atrophy [34]. Thalidomide was serendipitously found to have a dramatic effect on CRPS in a multiple myeloma patient [35] that was later confirmed in a subsequent study of 42 patients who had failed numerous prior treatments [36]. Thirteen of these patients (31%) reported either a significant or modest pain relief, and some were even able to reduce their pain medication. Hence, further studies are in progress. The mechanism of this novel activity is unclear, but the inhibitory activity of thalidomide on tumor necrosis factor α (TNF-α) and other cytokines such as IL-6 have been suggested.

Current clinical trials of thalidomide analogues

Various IMID®s have been synthesized and screened for anti-cancer and anti-inflammatory activities because of their numerous effects on the human immune system. Lenalidomide (REVLIMID™; CC-5013) and CC-4047 (ACTIMID™) are second-generation IMID®s currently in clinical trials. Both analogues have potent immunomodulatory activities with lenalidomide in Phase II and III clinical trials [37]. In *in vitro* studies, thalidomide has been shown to inhibit production of TNF-α from human monocytes [38].

By using inhibition of the pro-inflammatory cytokine TNF-α as a basis for comparison, lenalidomide and CC-4047 are 2000 and 20 000 times, respectively, more potent than thalidomide [39]. Lenalidomide at 5–50 mg day^{-1} can overcome conventional drug resistance in relapsed multiple myeloma patients with no thalidomide-like side-effects (sedation, constipation, peripheral neuropathy) [40]. Myelosuppression characterized by reductions of white blood cell and platelet counts however was observed at a dosage of thalidomide at 50 mg day^{-1}, requiring dose reduction or a 'drug holiday'. Seventeen out of 24 patients (71%) responded positively to treatment with >25% reduction in paraprotein levels. Lenalidomide is currently in Phase III trials for relapsed multiple myeloma. Preliminary studies showed that lenalidomide at dosages of 10 or 25 mg day^{-1} or 10 mg day^{-1} for 21 days with a seven-day treatment-free period produced significant

responses in patients with low and intermediate risk MDS. Erythroid response was observed in 21 out of 33 patients (64%) with a major response (transfusion-independence) seen in 19 patients [41]. Significant cytogenetic response was seen in a subset of MDS patients exhibiting a 5q chromosomal deletion of a single chromosome. Lenalidomide is in Phase II trials for MDS and in Phase II trials for CRPS. CC-4047 at a dosage of 1 mg day^{-1} has shown promising activity in patients with metastatic hormone-refractory prostate cancer [42], where six out of 13 patients had a significant decrease in the level of prostate-specific antigen. However, adverse effects included constipation, nausea and fatigue. Expanded trials based on the CC-4047 dosage of 2 mg day^{-1} are ongoing.

Immunomodulatory and non-immunomodulatory properties

The precise mechanism(s) of thalidomide and the IMID®s efficacious activities are unknown. This class of compounds, however, has numerous immunomodulatory and non-immunomodulatory properties, which are probably working in concert to produce the observed efficacy.

Immunomodulatory properties

Inhibition and stimulation of cytokines

Cytokines are soluble glycoproteins released by cells of the immune system, which act non-enzymatically through specific receptors to regulate immune responses. TNF-α is a pro-inflammatory cytokine produced by monocytes, macrophages, lymphocytes and natural killer (NK) cells [43]. This cytokine plays an important role in host immune and inflammatory response to viral, parasitic, fungal and bacterial infections. TNF-α has been implicated in the pathophysiology of infections and autoimmune diseases. Elevated TNF-α levels are associated with various inflammatory and autoimmune diseases such as rheumatoid arthritis, Crohn's disease, tuberculosis, cancer cachexia and ENL. The ameliorative effects of thalidomide on ENL are particularly striking (Figure 2). Thalidomide and its analogs are potent inhibitors of TNF-α produced by lipopolysaccharide (LPS)-stimulated human monocytes [43]. This inhibition is a result of the increased degradation of TNF-α mRNA [44]. Levels of other cytokines, IL-1β, IL-6 and granulocyte–macrophage colony-stimulating factor (GM–CSF), are also inhibited by thalidomide, whereas IL-10 is stimulated [45]. Lenalidomide and CC-4047 had similar effects on these cytokines although with varying degrees of potency when compared with those by thalidomide. The effects of these findings on various diseases are still being investigated.

Co-stimulation of primary human T cells

Thalidomide co-stimulates primary human T cells, inducing their proliferation, cytokine production and cytotoxic activity [46]. Co-stimulation involves the delivery of a second signal to naïve T cells to produce an antigen-specific response. The immunological adjuvant action of thalidomide therefore stimulates the otherwise ineffectual immune response, for example, to tumor antigens enhancing an anti-cancer response. This thalidomide action is dependent on the type of immune cell that is activated and the type of stimulus the cell receives. In *in vitro* studies, thalidomide induced an IL-2-mediated primary T-cell proliferation through the T-cell receptor (TCR) complex with a concomitant increase in IFN-γ production [45]. This proliferation is greater for the cytotoxic, rather than helper, T-cell subset, which is supported by observations in thalidomide-treated HIV seropositive patients where it increased the population of cytotoxic T cells and plasma levels of IL-2 receptor, a marker of T-cell activation. The stimulatory property could partially explain thalidomide's anti-inflammatory effects in inflammatory bowel disease in which the activity of cytotoxic T cells is diminished. The effects of thalidomide depend on the disease and immunological status: the co-stimulatory activity explains the unexpected increase in TNF-α production in certain diseases [47]. IMID®s are more potent than thalidomide in co-stimulating T cells that have been partially activated by the TCR. For example, the co-stimulatory action of CC-4047 is thought to produce the prolonged anti-tumor response seen in mice implanted with colorectal cancer cells [47]. Protection is thought to be mediated by T helper cell 1 (Th1) cellular immunity.

Modification of surface cell adhesion molecules

In response to an inflammatory stimulus, leukocytes are recruited into the injured tissue by capture, rolling, tight binding, transmigration across the endothelium and chemotaxis. Thalidomide can decrease the density of TNF-α-induced intercellular adhesion molecule-1 (ICAM-1), vascular cell adhesion molecule-1 (VCAM-1) and E-selectin on the endothelial cells of human umbilical vein [48]. L-selectin was also decreased by thalidomide *in vitro* and blocking this adhesion cascade by thalidomide is believed to mediate the anti-vasculitis effect seen with ENL. This decrease in cell adhesion molecule expression is also thought to occur with multiple myeloma: lenalidomide decreases binding of multiple myeloma cells to the endogenous bone marrow stromal cells thereby decreasing the production of vascular endothelial growth factor (VEGF) and IL-6. There is evidence that suggests modulation of adhesion molecules between these two cell types [40].

Stimulation of Th1 immunity

Thalidomide has been shown to stimulate a Th1 response in healthy humans after oral dosing, which was manifested by an increase in IFN-γ without changes in IL-2 and IL-4 levels [49]. In murine models of colorectal cancer and melanoma, CC-4047 increased the Th1 cytokines IFN-γ and IL-2 [50]. In scleroderma patients, incubation of their peripheral blood mononuclear cells (PBMC) with thalidomide produced a dose-dependent increase in IFN-γ and IL-2 levels [51]. Subsequent studies of the IMID®s showed that the increase in T-cell cytokine production is through potentiation of the transcription factor activator protein

1 [52]. The results in healthy humans and scleroderma patients strongly suggest enhancement of Th1 type immune activity by thalidomide. However, earlier work disputes this finding and suggests that thalidomide shifts the immune response from Th1 to Th2 [53]. This disagreement is probably a result of the different cell-based models used and the type of immune activation.

Induction of natural killer cells

Thalidomide and its IMID®s significantly increased the lysis of human multiple-myeloma cell lines and patient multiple-myeloma cells after incubation with IL-2-primed PBMCs [54], which was found to be mediated by NK cells. Patients with relapsed multiple myeloma had an increased number of NK cells with only responding patients showing an increase in the percentage of such cells. This increase in NK cells was accompanied by a decrease in the plasma disease biomarker paraprotein, and an increase in the levels of IL-2 and IFN-γ secretion [54]. These findings correlate with the known predominant Th1 and minor Th2 stimulatory effects of thalidomide and its IMID®s, as discussed earlier. Elevated levels of these Th1-related circulating cytokines stimulated the activity and number of NK cells, leading to the observed lysis of multiple myeloma cells.

Inhibition of nuclear factor-kappa B activity

The transcription factor nuclear factor-kappa B (NF-κB) is a key regulator of inflammatory genes including TNF-α and IL-8. In the nonstimulated state, NF-κB resides mainly in the cytoplasm where it is tightly bound to inhibitory proteins of the IκB family. After stimulation of NF-κB by TNF-α or IL-8, IκBα is phosphorylated by IκB kinase leading to the ubiquitination and subsequent degradation of IκBα by the proteasome. This is followed by nuclear translocation of NF-κB where it regulates the expression of genes involved in immune and inflammatory processes, cell growth, suppression of apoptosis and metastasis [55–58]. Thalidomide has been shown to inhibit NF-κB through suppression of IκB kinase [59] – representing another mechanism of its anti-inflammatory and anti-cancer properties. Because of the multiplicity of mechanisms of action for thalidomide and the IMID®s, it is difficult to understand which mechanism or mechanisms are responsible for therapeutic activities observed for individual diseases.

Non-immunomodulatory properties

Anti-angiogenic activity

Angiogenesis is the development of new blood vessels. In cancer, angiogenesis can nurture the growth and metastasis of tumors and tumor cells, respectively. Thalidomide and the IMID®s have anti-angiogenic properties that are independent of their immunomodulatory effects [60,61], which could have a role in the apparent anti-neoplastic activity of this drug seen with various cancers. In the rat aorta assay, the IMID®s are two to three times more potent in their anti-angiogenic activity when compared with that by thalidomide. Lenalidomide, but not thalidomide and CC-4047, significantly inhibited the migration of endothelial cells. CC-4047 also inhibits VEGF, but not the expression of basic fibroblast growth factor (bFGF). The IMID®s anti-TNF-α activity had no effect on anti-angiogenic activity [61]. In multiple myeloma, the close proximity interaction between the indigenous bone marrow stromal cells and patient multiple myeloma cells significantly increased levels of the pro-angiogenic factors VEGF and IL-6 (a multiple myeloma growth and survival factor) [40,47,54,62,63]. Thalidomide and CC-4047 significantly decreased expression of these factors thereby reducing the production and growth of new blood vessels feeding the multiple myeloma cells [62]. These results underscore the importance of stromal and multiple myeloma cell interaction in the bone marrow microenvironment for the maintenance and progression of the disease, and provides another target for thalidomide and its IMID®s.

Anti-proliferative and pro-apoptotic activity

Thalidomide and its IMID®s inhibit <20% and 50% of DNA synthesis, respectively, in human multiple-myeloma cell lines and cells from patients [63]. The IMID®s also inhibited the proliferation of doxorubicin- and melphalan-resistant multiple-myeloma cells by 20–50% [40,47]. These results correlate with the anti-tumor activity seen in patients with the drug-resistant forms of the disease. The anti-proliferative mechanism of action is considered to be by inhibition of IL-6 production. IMID®s have pro-apoptotic activity in human multiple myeloma cells. They arrest cell growth at the G1 phase and trigger activation of caspase 8, enhance multiple myeloma cell sensitivity to Fas-induced apoptosis, downregulate the activity of NF-κB, and decrease the expression of apoptosis-inhibitory protein [63,64].

Cyclooxygenase-2 inhibition

Thalidomide and its IMID®s inhibited the protein expression of cyclooxygenase-2 (COX-2), but not COX-1, in LPS-, TNF-α- and IL-1β-stimulated PBMCs, and decreased the half-life of COX-2 mRNA in a dose-dependent manner. They also inhibited the synthesis of prostaglandin E2 from LPS-stimulated PBMC. Whereas anti-TNF-α or anti-IL-1β neutralizing antibodies had no effect on COX-2 expression, anti-IL-10 neutralizing antibody elevated the expression of COX-2 mRNA and protein from treated PBMC. These data suggest that the anti-inflammatory and anti-tumor effects of IMID®s could result in part to the elevation of IL-10 production and its subsequent inhibition of COX-2 expression [65].

Mechanisms of action in multiple myeloma

Unexpectedly, thalidomide was found to have anti-myeloma activity when it was thought its anti-angiogenic activity could slow the disease by inhibiting the formation of new blood vessels in this highly vascularized cancer. There is now ample evidence to show that the anti-cancer activity of thalidomide and its IMID®s in multiple myeloma is through different mechanisms and sites in the bone

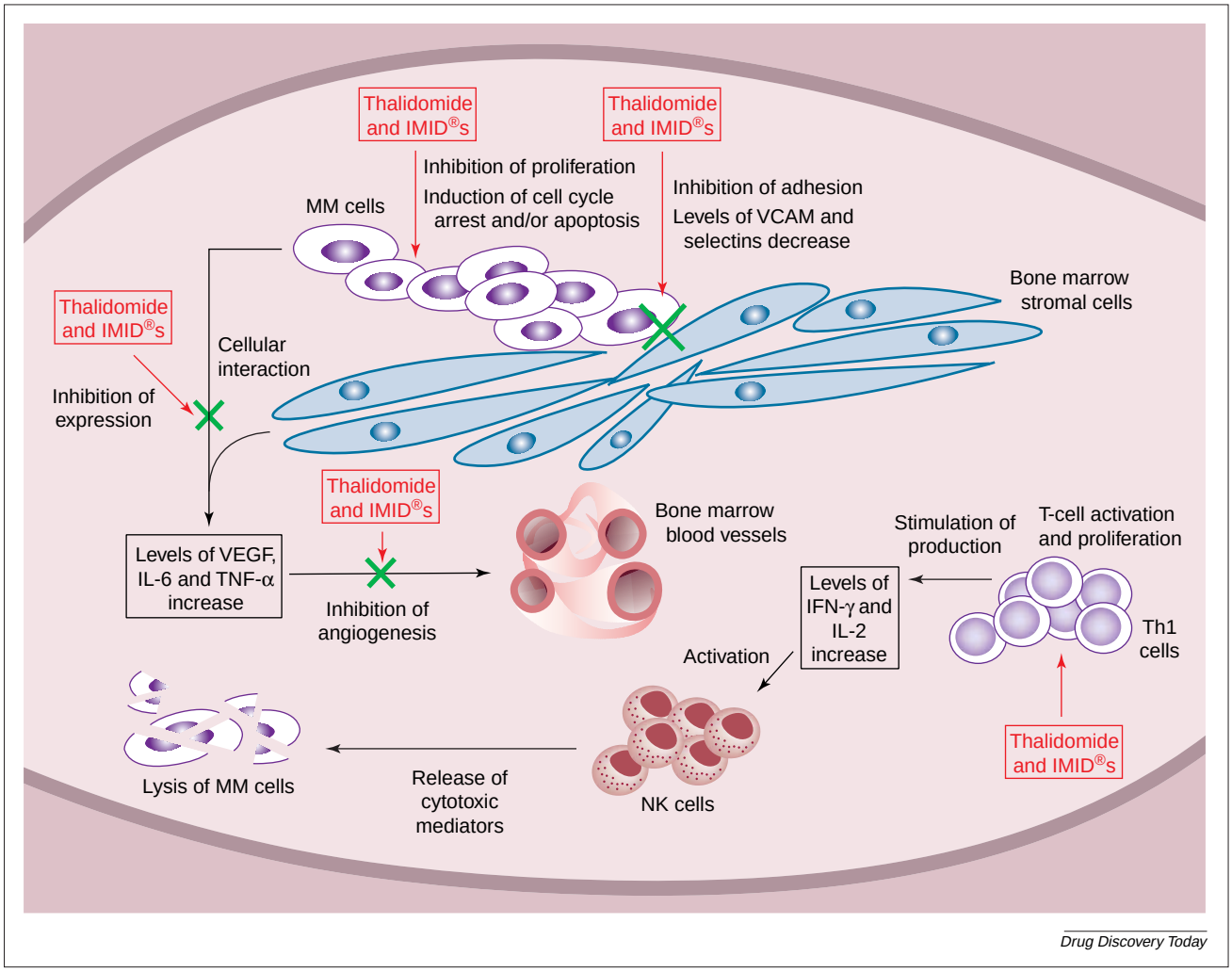

FIGURE 3

Sites of activity of thalidomide and immunomodulatory analogues in the bone marrow of multiple myeloma patients. Multiple sites and mechanisms of action are thought to be involved in the activity of these compounds in multiple myeloma. Thalidomide and the IMID®s have immunomodulatory and non-immunomodulatory properties, including: (i) T-cell activation and proliferation, resulting in lysis of MM cells by NK cells; (ii) inhibition of cell surface adhesion molecules; (iii) inhibition of proliferation and induction of apoptosis of MM cells; and (iv) anti-angiogenic activities. Abbreviations: IFN-γ, interferon-γ; IL, interleukin; IMID®s, immunomodulatory analogues; MM cells, multiple myeloma cells; NK cells, natural killer cells; VCAM, vascular cell adhesion molecule; VEGF, vascular endothelial growth factor. Figure adapted from Ref. [66].

marrow [40,47]. Figure 3 shows the bone marrow microenvironment in multiple myeloma, which contain aberrations in various cellular processes, immunology and cell interactions [66]. The immunomodulatory activities of thalidomide and its IMID®s involve the inhibition of expression of IL-6 and TNF-α by bone marrow stromal cells that in turn inhibits the growth of multiple myeloma cells. The compounds also enhance T-cell stimulation and proliferation with the activated cells then releasing IL-2 and IFN-γ. These cytokines activate NK cells causing lysis of the multiple myeloma cells [47,63]. The combination of immunomodulatory and non-immunomodulatory anti-cancer activities in the bone marrow could produce the significant anti-tumor responses observed in some multiple myeloma patients. This combination activity has significant implications for other blood and solid tumor cancers and is currently being investigated in numerous clinical trials.

Mechanisms of action in solid tumors

While numerous Phase II trials of thalidomide have demonstrated potential activity against some solid tumors, the mechanism of action is still unclear but could involve both immunomodulatory and non-immunomodulatory activities. These tumors produce immunological suppressive factors that prevent priming and activation of $CD4^+$ and $CD8^+$ T cells of the lymph nodes [47]. Other immune cells such as NK cells and macrophages are also inhibited, and are therefore unable to respond to and destroy tumor cells [47]. Thalidomide and the IMID®s co-stimulatory action on primary human T-cells enhance the anti-tumor activity mediated by the Th1 cytokines IL-2 and IFN-γ. The co-stimulation is thought to overcome the nonresponsiveness by T cells and to prevent the release of suppressive factors thereby enabling tumor-specific cells to kill tumor cells [46,47]. Thalidomide and the IMID®s

are also thought to co-stimulate macrophages and NK cells leading to the anti-tumor activity as discussed earlier [36]. The IMID®s have enhanced anti-tumor activity when co-administered with Rituximab in a severe combined immunodeficient mouse model of non-Hodgkin's lymphoma, which could be a result of the downregulation of NF-κB in the lymphoma cells, activation of the innate immune system, cell growth arrest and induction of apoptosis [67,68]. Anti-angiogenic and pro-apoptotic activities of thalidomide and the IMID®s are hypothesized to play a role in the apparent efficacy seen in various highly vascularized solid tumors through inhibition of VEGF and induction of growth arrest, respectively [47,63].

The STEPS® program for commercial thalidomide use

With therapeutic use of thalidomide increasing in the USA, its potential for fetal toxicity is a major concern. The lowest doses and shortest treatment period, where characteristic birth defects in human fetuses have been documented, were 25 mg day^{-1} (0.5 mg kg^{-1} based on a human weighing 50 kg) for two to three days, and 50 mg day^{-1} (1 mg kg^{-1} day^{-1}) for only one day [69]. To reduce this potential, the marketing and use of thalidomide is restricted through the mandatory System for Thalidomide Education and Prescribing Safety (STEPS®) program [70]. This unique system of monitoring oversees the prescribing, dispensing and dosing of thalidomide. All patients, pharmacists and prescribing physicians must be registered in Celgene's database. In addition, a video that includes a warning from a thalidomide victim about the drug's teratogenicity is made available to first-time patients. All women of childbearing age are required to use two types of contraception. Prescriptions are only for a 28-day supply with renewals requiring a visit to the physician and completion of a questionnaire on sexual activity [70], and women of childbearing age are required to have a negative pregnancy test before prescription renewals. The STEPS® program therefore makes thalidomide the most restrictively prescribed drug ever approved. To date, >100 000 patients have been prescribed thalidomide without any instances of drug-related birth defects.

Future of thalidomide and the IMID®s

The rehabilitation of thalidomide has increased its experimental use in numerous oncological and inflammatory conditions. While it has yet to be approved for multiple myeloma in the USA and Europe, applications are pending and the drug is approved for this indication in Australia and Turkey. Recent reports of potential activity in other solid tumors have increased its experimental use. The future of this class of compounds is in the more-potent IMID®s, particularly lenalidomide, where the significantly increased immunomodulatory and anti-angiogenic potency and apparent lack of some, or a decreased amount of, thalidomide's dose-limiting side effects have made them potentially important therapeutics in cancer and inflammatory diseases. In addition, lenalidomide is not teratogenic in rabbit, a sensitive species for thalidomide-induced birth defects making lenalidomide a more desirable therapeutic than its parent. The IMID®s therefore represent second-generation small molecule compounds with novel mechanisms of anti-cancer activity and their potential as anti-cancer therapeutics is enormous should their activities be confirmed in ongoing Phase II and III trials.

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