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Targeting plasma cells: Are we any closer to a Panacea for diseases of antibody secreting cells?

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Running title

Therapeutic targeting of plasma cells

Summary

Antibody secreting cells are critical for a functional and effective adaptive immune system. In a number of illnesses, however, these same cells contribute to the underlying disease state leading to significant morbidity and mortality. While therapeutic targeting of antibody secreting cells has progressed significantly over the last two decades, a number of these conditions remain major health problems. In this review we will discuss current and potential therapeutic targeting of antibody secreting cells in the context of the known biology of these cells.

Keywords:

Antibody-secreting cells, Plasma cells, Therapeutics

Antibody secreting cells (ASCs) or plasma cells (PC), are effector cells in the humoral arm of the adaptive immune system, responsible for production of antibodies of varying avidity for the purpose of eliminating foreign antigens while conserving the body's own tissues(1). In a normal, effective immune system, PC and their resultant antibodies are critical to both resolution and subsequent long-term protection against foreign pathogens. These attributes have been exploited by the scientific and medical communities with 22 of 24 licensed vaccines functioning directly through antibody production(2). Despite such clear contributions of humoral immunity to human health, there are diseases in which ASC play a critical role in pathogenesis and as such, direct targeting of pathological ASCs may therefore be a route for therapeutic intervention. In this article we explore the mechanisms by which ASCs are produced, the transcriptional program that dictates their differentiation and ASC

survival pathways with a view to the efficacy of current ASC-focused therapeutics and possible future directions.

Development of Antibody-secreting cells

ASCs differentiate from mature B lymphocytes via three broad pathways (Figure 1). First, a small subset of cells (B1 and marginal zone B cells) can differentiate spontaneously into the ASC that are responsible for so-called natural immunoglobulin(3). This pathway does not require exogenous antigen and, in normal circumstances, is responsible for a minority of the overall ASC population(3).

The majority of ASCs are generated in response to exogenous antigen stimulation of B cells expressing the cognate B cell receptor (BCR)(1). In these immune responses, the activation of B cells, and thus the generation of ASCs, can be compartmentalised into two pathways based on attributes of the antigen, namely whether it requires the presence of T cells to elicit antibody production. T-cell independent (TI) antigens, which are further partitioned into Type 1 (antigen is intrinsically mitogenic for B-cells) and Type 2 (not intrinsically mitogenic), predominantly arise from B1 and marginal zone B cells and lead to rapid formation of foci of PC and plasmablasts, located outside the B-cell follicles and at the border of the red and white pulp in the spleen or along the medullary cords of lymph nodes(4, 5). These extra-follicular ASCs arise early in the immune response, and primarily secrete low affinity IgM, although class switch recombination can occur(4). Typically also, TI immune responses are considered transient with limited persistence of antibody or PC. This is not, however, always the case and persistent TI responses and TI derived PC in the spleen and BM have been described(6, 7).

Antigen responses that are T-cell dependent, which arise from the follicular B cell and marginal zone B cell populations, provide the majority of antibody in circulation and are typically considered the origin of long-lived and affinity matured ASC (Figure 1). TD responses comprise both extra-follicular ASC foci as well as ASC that arise from the intra-follicular germinal centres (GC). Both these pathways rely on signals from T cells, previously activated by dendritic cells (DC). Interactions between T and

B cells drive the migration, proliferation and subsequent differentiation of the B cells, both in the foci of ASC and in the GC. The CD4⁺ T-helper cells express the co-stimulatory molecules CD40 ligand (CD40L, or CD154), ICOS, members of the SLAM family and cytokines that promote B-cell proliferation and differentiation, all of which are required for GC formation(8).

GCs are transient structures that typically last a few weeks after antigen exposure before contracting. By light microscopy they are comprised of juxtaposed light and dark zones in which the different functions of the GC occur. In current models, the dark zone is comprised of rapidly dividing centroblasts, which are also undergoing Activation-Induced Cytidine Deaminase (AID) driven somatic hypermutation (SHM), while the light zone is comprised of non-proliferating centrocytes, interspersed with a dense follicular dendritic cell (FDC) network(9). It is further proposed that deposits of antigen on the FDC network provide the basis for selective expansion of high affinity variants within the GC. In these models B-cells compete for access to, and acquisition of, the antigen from the FDC, which they subsequently process and present to the specialised follicular CD4⁺ T helper cells (Tfh). The Tfh provide proliferation and differentiation signals to the successful GC B cells(10), which ultimately results in the dominance of high affinity clones within the GC and amongst its product populations, the memory B-cells and long-lived ASC. B cells that fail to receive signals from Tfh are thought to undergo apoptosis and thus are eliminated from the reaction(10, 11).

Beyond the germinal centre, B cells have two possible fates, namely ASCs or memory B cells, both of which have the potential for extraordinary longevity. Memory B cells, compared to naïve B lymphocytes involved in the initial response, are able to rapidly replicate and give rise to ASCs of enhanced affinity when rechallenged with cognate antigen(12). The mechanism of memory B-cell survival remains something of a mystery in so far as no specific factors or gene products, either soluble or membrane bound, intrinsic or extrinsic, have been identified as being crucial for their persistence. Equally, the signals within GC that promote memory B-cell formation or ASC differentiation over re-cycling into the dark zone, remain something of an

enigma, although Tfh interaction seems to be a major determinant(13). The differentiation of GC B-cells into ASC and their subsequent persistence is somewhat better understood in terms of transcription factors, migration and survival mechanism. The probability of differentiation seems to be influenced by the affinity of the B cell for antigen, with high affinity GC B-cells preferentially undergoing differentiation, and the resultant ASC guided through the action of chemokines towards their long-term survival niches in bone marrow where they receive the signals necessary to sustain expression of their key survival gene, Mcl-1(14). Knowledge of the factors controlling the various stages of this process provides exciting opportunities for the development of therapeutics that may treat antibody-mediated diseases.

Transcriptional control of plasma cell development

Studies into transcription factors have revealed the genetic program that guide development towards different cell types. There appears to be interplay between ASC promoting transcription factors including Interferon regulatory factor 4 (IRF4) and B-lymphocyte induced maturation protein 1 (Blimp-1) and B cell promoting factors including Paired box protein 5 (PAX5), B cell lymphoma 6 (BCL6) and Interferon regulatory factor 8 (IRF8) (Figure 2)(15). As the transcription factors associated with ASC differentiation have been review extensively elsewhere(16), here we focus on the few that may be useful therapeutically, providing a brief account of their role in this process.

Factors promoting ASC fate:

IRF4 is critical for ASC differentiation and is now thought to be the primary initiator of the ASC transcriptional program(17-19). In mature B cells IRF4 is kept at low levels by microphthalmia-associated transcription factor (Mitf) with the loss of Mitf leading to spontaneous differentiation to ASCs(20). Low amounts of IRF4 promote germinal centre activity via modulation of AID(21), while higher levels repress BCL6 and activate Blimp-1 to promote plasma cell differentiation(21, 22). Other studies

suggest IRF4 may also be a crucial regulator of ASC persistence, although the mechanism for this remains uncertain(23).

Blimp-1 is a key transcriptional repressor for plasma cell development. It is encoded by the *Prdm1* gene, expressed in numerous haematopoietic cells and is critical for terminal B and T cell development(24)(25)(26). Blimp1 is thought to function by down-regulating B cell genes that promote and maintain B cell characteristics including PAX5, Spi-B and BCL6, thereby allowing the ASC program to unfold including Xbp-1(25).

X-box binding protein-1 (XBP1) was initially thought to have a key role in ASC generation(27), however, further studies revealed that ASCs could form without XBP-1 but lack normal ASC function and morphology leading to reduced antibody secretion(28, 29). XBP1 has a role in the unfolded protein response that occurs when the endoplasmic reticulum is placed under stress by large amounts of unfolded protein(30, 31). Given the large amounts of immunoglobulin that ASCs produce, inability to deal with unfolded proteins such as in XBP-1 deletion was thought to induce apoptosis(32), however, the persistence of XBP-1 deficient plasma cells casts doubt over this association(28).

Factors that promote memory B cell fate

PAX5 is the master regulator of the B cell program, required by lymphoid progenitors for commitment to the B cell lineage and maintenance of B-cell identity(33, 34).

PAX5 activity is normally down regulated at the time of commitment to ASC differentiation(35). PAX5 functions by regulating B cell associated genes including components of the B cell receptor (IgH, Blnk and Ig α), immune receptors (CD19, CD21 and CD23) and transcription factors such as IRF4, IRF8 and BACH2.

Simultaneously PAX5 suppresses a number of genes including those that promote ASCs(15).

BCL6 was discovered due to its role in B cell lymphomagenesis(36, 37), but is critical for germinal centre development as its absence results in a complete lack of GCs(38-

40). BCL6 is considered critical for the germinal centre reaction due to its role in promoting cell proliferation and for suppressing DNA damage responses thus allowing SHM(41, 42). BCL6 is regulated, in part by IRF8 within germinal centres(43).

IRF8 is now thought to be critical in promoting differentiation towards memory B cells by suppressing the ASC program. IRF8 in conjunction with PU.1 limits terminal differentiation with double knockout of these two proteins leading to heightened class switching and ASC differentiation(18). Given that IRF8 and IRF4 are similar and both require PU.1 coupling to strengthen their DNA binding, a logical hypothesis is that IRF8 and IRF4 may directly compete for the same binding sites to dictate alternate maturation programs(18).

Antibody secreting cell survival

Once ASCs are generated, their persistence requires them to migrate to survival niches normally in the bone marrow, spleen, and mucosal surfaces. ASCs generated in vitro or studied ex vivo survive only a day or so before they die(7, 44), indicating the necessity for external signals to prolong longevity. Normally, ASCs comprise around 0.1-1% of cells within the bone marrow in both humans and mice with this percentage not changing for the majority of life(45). Despite the fixed number of ASCs, new infections are able to generate antigen specific, long lived ASCs, with excess ASCs lost during a phase of contraction(46)(47). This indicates that turnover of existing ASC does occur, although whether it is caused by a 'push factor' of newly created ASC remains to be determined (44). External factors that have been reported to be involved in the ASC survival niche include Interleukin-6 (IL-6), APRIL, CXCL12, CD44 ligand, granulocyte-macrophage colony stimulating factor and Tumour Necrosis Factor α (7, 48). A variety of cells may provide these external survival signals including eosinophils, megakaryocytes, monocyte precursors, basophils, and bone marrow stromal cells(49). *In vivo* validation of the roles of individual factors on established ASC is rare, although targeting APRIL or its receptor, BCMA, has been shown to effect ongoing survival(14, 50).

Although the mechanisms allowing ASC longevity have not been elicited, evidence suggests that the intrinsic apoptotic pathway is critical. Our laboratory has demonstrated that the anti-apoptotic protein Mcl-1 is indispensable for ASC survival. In vivo induced deletion of Mcl-1 in the B-cell lineage resulted in almost complete abolition of the ASC population in spleen and bone(14), suggesting that the critical pro-survival factors for ASC feed into a common end-product, Mcl-1 expression. While Mcl-1 is indispensable for plasma cell survival, suppression or deletion of other Bcl-2 pro-survival members can affect the plasma cell population. Treatment with the BH3-mimetic ABT-737, an antagonist of Bcl-2, Bcl-xL and Bcl-w mediated cell survival, specifically inhibited establishment but not persistence of the bone marrow ASC population with no obvious affect on ASC in the spleen(55). These findings suggest different survival strategies during both development and/or localisation of ASC.

Nature of the antibody-secreting cell compartment

Within the ASC compartment, phenotypically different cells have been identified. Plasma cells are characterised by the ability to produce antibodies and are considered terminally differentiated with an inability to replicate. In contrast, so-called plasmablasts, are thought of as an intermediate between B cells and plasma cells as they express B cell markers (such as CD79, CD20, CD22, and CXCR5), replicate yet secrete antibody and express some plasma cell marker such as CD38 and CD138(51). Using the Blimp-1^{Bfp} reporter (in which cells expressing Blimp-1 also express a proportional amount of green fluorescence protein(35), ASC have been divided into two populations identified as Blimp-1^{int} and Blimp-1^{high} with the difference in expression being about 5 fold(35). Without deliberate stimulation, mice have around 50% of each group in the spleen while in the bone marrow the majority of ASC are Blimp-1^{high}. On stimulation with either T-independent or T-dependent antigens there is a rapid increase in Blimp-1^{int} ASC with little or no change in the proportion of Blimp-1^{high} antibody secreting cells(35, 47). Additionally Blimp-1^{int} cells turn over rapidly while the Blimp-1^{high} ASC population turn over much more slowly. These results suggest that the Blimp-1^{int} population comprises intermediate plasmablasts and short-lived ASC while the Blimp-1^{high} population comprises

terminally differentiated plasma cells(35). Interestingly, only Blimp-1^{int} cells are detectable in blood(35), which is consistent with the content of PBMC from human samples in the days post vaccine boost(47, 52, 53). It is therefore tempting to presume that Blimp-1^{int} cells eventually differentiate into Blimp-1^{high} plasma cells however this conversion is still to be definitively proven and it is possible that these two groups represent independent entities.

Heterogeneity has been identified also when investigating ASC survival. For example, BCMA knockout mice lack the ability to signal via APRIL and lose a significant proportion of bone marrow plasma cells yet splenic plasma cells remain unaffected(14, 54). Additionally, BCMA knockout mice have reduced expression of Mcl-1 in their remaining bone marrow ASCs but again splenic ASCs are unaffected(14). These findings suggest that APRIL regulates Mcl-1 in some ASC populations, but is either redundant or has no effect on other ASC populations. Again, the different impact of treatment with ABT-737 on ASC numbers in bone marrow and spleen support this proposal(14, 55, 56).

Lastly, human studies have identified heterogeneity within the bone marrow ASC compartment, adding to that already identified between the spleen and bone marrow populations(57). Analysing surface expression of CD138, CD38 and CD19 on BM ASC revealed discrete subpopulations. Importantly, the CD19⁻, CD138⁺, CD38^{hi} population had a distinct RNA signature and appeared to contain ASCs producing antibodies against antigens that the patients had no known exposure to for over 40 years, suggesting that this phenotype identifies particularly long lived ASCs(58). This proposal is supported by a parallel analysis of the same population(59). Heterogeneity within the ASC compartment has important implications for therapeutic interventions, as subpopulations may be more or less susceptible to specific therapies depending on their underlying phenotype or survival pathways. Interestingly, the absence of CD19 on plasma cells is often used in conjunction with other surface markers for identification of abnormal plasma cells in myeloma(60).

Antibody secreting cells in pathology

Plasma cell neoplasms

The most obvious human pathology involving ASCs are the disorders collectively termed plasma cell neoplasms (PCN). PCNs are characterised by the neoplastic proliferation of ASCs and represent a spectrum of disease from indolent monoclonal gammopathy of undetermined significance (MGUS) to plasma cell myeloma that includes in rare instances highly aggressive plasma cell leukaemia(61). While early stages are asymptomatic, advanced disease is characterised by end organ damage including skeletal lytic lesions, renal impairment, anaemia, hypercalcaemia and immunodeficiency. Despite improving understanding and treatment options, with the exception of some patients treated with an allogeneic bone marrow transplant, the PCNs are still considered incurable conditions(62). The classic clinical pattern is one of disease response followed by multiple relapses until the disease becomes insensitive to therapy.

The first clinically identifiable stage of PCN is MGUS, defined as the presence of a monoclonal protein (M-protein), with less than 10% plasma cells in the bone marrow and no evidence of end organ damage(62). This definition is arbitrary in its boundaries as patients with >10% plasma cells or an M-protein of >30g/L without end organ damage are classified as smouldering/asymptomatic myeloma and are for the most part managed with close monitoring similar to MGUS. The aetiology of MGUS is unknown although risk factors include African descent, older age, male, exposure to pesticides and family history(63). Population based studies have shown that almost all cases of myeloma are preceded by MGUS(64, 65). Furthermore, MGUS is relatively common in the general population, affecting approximately 2-3% of all people aged 50 years or older with 1% progressing to myeloma each year(66, 67). The M-protein in MGUS can be of any class of immunoglobulin with IgG being the most prevalent, followed by IgA, biclonal and the rare IgD and IgE MGUS(66). Although IgM myeloma has also been reported (68), M-proteins of IgM class are classically associated with Waldenström's macroglobulinaemia, a low grade lymphoproliferative disorder of differing pathology typically with activating mutation in MYD88(62, 69). PCN plasma cells can also secrete incomplete immunoglobulin and

classically this involves the light chains with an absence of heavy chain. It is also possible to detect a 'light chain MGUS' that presumably precedes light chain myeloma, which comprises approximately 20% of all new myeloma diagnoses(70).

Several groups have attempted to identify and characterise the so-called myeloma initiating cells (MIC). It is presumed that these cells give rise and comprise the first clonal ASCs that, over time, accumulate further mutations and progress to malignant plasma cell myeloma. It is also hypothesised that these cells, with an ability to self-replicate, are a major reason why cure of plasma cell neoplasms remains elusive. For the most part, MICs have been investigated by taking a fraction of malignant plasma cells from patients with myeloma, and attempting to transplant them into immunocompromised murine models to determine their ability to reconstitute disease in a new host(71). A number of surface markers are thought to differentiate the sub-population of plasma cells that have this ability and therefore represent MICs. These markers include absent CD138, absent CD19, low CD45 and high CD38. Whether these markers can reliably differentiate MICs remains contentious(71). Other attempts to identify MIC's include correlation with a normal counterpart. Analysis of the V region of the heavy chain in 48 myeloma patients by PCR and sequencing indicates that myeloma has mostly undergone SHM with the distribution of resultant amino acid substitutions indicating antigenic selection had occurred. These findings indicate that myeloma plasma cells are germinal centre derived as opposed to extra-follicular in origin(72). Furthermore, there is no evidence that further SHM occurs within different clones of myeloma, suggesting that clones are derived from the same post germinal centre cell(73). Overall, our understanding of what comprises the MICs remains rudimentary with further research required to unlock the biological changes that allow a plasma cell to replicate.

Symptomatic plasma cell myeloma is thought to result from the expansion of altered clones that are already present within the precursor MGUS(74, 75), however, the mechanisms behind this progression remain unclear and are likely to be through branching pathways typically associated with the evolution of species(76). Current understanding of the genetic changes that lead to myeloma first developed from

cytogenetic analyses. Karyotype abnormalities observed in PCN's constitute two broad groups of "early" cytogenetic abnormalities, which are thought to be early events in the evolution to myelomas because they are also found in the majority of patients with MGUS(77, 78). The first group is hyperdiploidy (near double the number of normal chromosomes) with a specific genotype involving trisomies of chromosomes 3,5,7,9,11,19 and 21. The second group is non-hyperdiploid, but with translocations involving the IgH locus at chromosome 14q(79, 80). What triggers these initiating events remains unclear although dysregulation of the cyclin D genes on chromosomes 11 (CCND1) or 6 (CCND3) has been postulated as an early unifying event(81, 82). Classically the translocations involving Chromosome 14 bring partner oncogenes under the influence of the IgH promotor region resulting in upregulation of the juxtaposed gene(83).

These early abnormalities are thought to allow accumulation of further mutations such as MYC translocations (Chromosome 8q42) leading to increased cell cycling and proliferation, the loss of apoptotic pathways and cell cycle arrest signals including p53 (deletion 17p) and CDKN2 (deletion 1p) and genetic aberrations with less well understood consequences such as deletion 13q and deletion 1q, which correlates with deletion of the gene FAM46C whose function remains unknown(79, 80, 83, 84).

Recent analyses of the genetics of plasma cell myeloma have used whole exome and whole genome sequencing, comparing myeloma plasma cell samples with other blood cells from the same patients. One study of 38 patients tried to identify genes of interest by comparing those with higher mutation rates, identical mutations in the same genes or 'sets' of genes that are thought to modify a specific pathway(85). The genes identified comprise several categories. Cancer associated genes already implicated in multiple myeloma including TP53, CCND1 and proteins involved in the Ras-Raf pathway such as KRAS, NRAS and BRAF. Genes with mutations normally associated with plasma cell ontogeny including IRF4, PRDM1 and XBP-1. There were also a number of mutations in genes that encode proteins with less clear roles including DIS3 and FAM46C that are thought to have roles in regulation of protein translation. A larger data set that included the initially reported 38 patients was

reported and included a further 203 myeloma patients(86). In this study marked heterogeneity was noted with only 11 genes showing a statistical significant increase in mutation rates. These genes included many of those identified in the previous study although the authors were unable to identify any association between patient outcomes or characteristics and these mutations. Of particular note is the idea that most patients identified had subclones, with some carrying differing mutations to the dominant clone(86).

The idea that myeloma is comprised of multiple clones and subclones each with varying genetic mutations has been corroborated in a study of 28 patients whose myeloma underwent genomic analysis at different time points during therapy(87). This analysis showed that within single patients, there were several different 'clones' at any single time point detected by copy number abnormalities. The authors were able to demonstrate that different clones were dominant at different time points in therapy and hypothesised that clones were selected by pressure induced with therapy. This so called intra-clonal heterogeneity has been validated in a number of further studies(88). At a clinical level, intra-clonal heterogeneity would be expected to have a significant impact on the efficacy of myeloma therapies. While gene mutations are well documented, the hypothesis of accumulated exon mutations alone in progressive PCN's is too simplistic. Rather the transformation from MGUS to plasma cell myeloma involves an increase in copy number abnormalities and copy number neutral loss of heterozygosity(89), DNA hypomethylation and gene-specific DNA hypermethylation(90). These "late" abnormalities commonly correlate with progressive disease. This theory of accumulated damage fits clinically with the majority of myeloma having a relatively indolent course but becoming progressively more aggressive and resistant to therapy, usually over a period of years.

In addition to abnormalities within the ASC population of PCNs, the microenvironment of neoplastic plasma cells is critical to survival and therefore malignant potential(91, 92). IL-6 is an important stimulator of plasma cell growth and is upregulated by NF- κ B stimulation in surrounding stromal cells(83, 91, 93). IL-6 stimulates several pathways within neoplastic plasma cells including the MAPK

pathway, PI3K-AKT pathway (upstream of mTOR) and the JAK-STAT pathway. IGF-1 is also secreted by bone marrow stromal cells and osteoblasts and can trigger the same pathways(94). Documented mutations and upregulation of each of the MAPK, PI3K-AKT and JAK-STAT pathways, as well as mutations in genes that regulate the NF- κ B pathway(95, 96), have been demonstrated in myeloma samples and may lead to upregulation of anti-apoptotic molecules including Mcl-1 and Bcl-2 and proliferative signals(91).

Autoimmune conditions and Transplantation medicine

A number of autoimmune conditions are characterised by the production of antibodies to self antigen which can affect almost any organ in the body including the thyroid gland (Grave's disease, Hashimoto's thyroiditis), pancreas (Type 1 Diabetes Mellitus) and neuronal synapses (myasthenia gravis). Additionally, autoimmune disease can be caused by the deposition of antibody in tissues as seen in patients with systemic lupus erythematosus (SLE)(97).

The underlying pathophysiology for the majority of autoimmune conditions remains unclear with a combination of genetic and environmental factors likely to play a role. For example, SLE, a quintessential multisystem autoimmune condition, is more common in females of African-American, Hispanic and Asian decent indicating a role for genetic factors. Despite this, monozygotic twins have only a 34% concordance indicating factors outside of genetics play a critical role in disease development(98). Numerous immune defects have been described in patients with SLE including abnormalities in dendritic cells, T cells and B cells(98). A loss of tolerance leads to production of multiple antibodies targeted against self-antigen including double stranded-DNA, anti-nuclear antigens and extractable nuclear antigens. Deposition of antibody-antigen complexes and surrounding inflammatory cell invasion is a characteristic of organ histology in patients with SLE(97). Therefore, despite disease resulting from incompletely understood mechanisms, targeting plasma cells and the production of antibody is a logical and appealing therapeutic target for SLE and a number of other autoimmune conditions.

Transplantation of organs such as the heart, lung, liver and kidneys to replace irreparably damaged recipient organs has become common over the last century(99). A major complication of solid organ transplantation, however, remains antibody-mediated rejection in which the recipients' immune system recognises epitopes on the transplanted tissues and mounts an appropriate, but often devastating, immune response against this foreign material culminating in the production of antibodies specific for the transplanted organ(100). This response can lead to failure and loss of the transplanted organs. We will not specifically discuss antibody-mediated rejection further within this review, however ASCs in this condition are an attractive therapeutic target much like in autoimmune diseases. We would expect that effective ASC directed therapies in autoimmune conditions would also have clinical utility in patients with antibody-mediated rejection.

Therapeutic targeting of antibody-secreting cells

Direct targeting of ASCs is the most obvious approach in the treatment of the plasma cells neoplasms (Figure 3). Outcomes for patients with myeloma have seen a dramatic improvement over the last 2 decades(101). Currently, standard front-line therapy combines chemotherapy in conjunction with corticosteroids and one or more of the so-called 'novel biological agents' which encompass thalidomide analogues (so called immunomodulatory drugs or IMiDs: thalidomide, lenalidomide, pomalidomide) and proteasome inhibitors.

Thalidomide analogues are critical to the treatment of PCN's and have been widely studied. Thalidomide, first used as an anti-emetic, resulted in disastrous outcomes secondary to congenital malformations(102). Three decades later it was approved for use in multiple myeloma. Its efficacy in myeloma has been extensively studied in an attempt to separate its anti-myeloma effects from its teratogenicity. Thalidomide and its analogues are thought to disrupt angiogenesis, intracellular pathways (including PTEN and mTOR), the myeloma microenvironment and to activate T and NK cells against the malignant plasma cells(103). More recently thalidomide was shown to bind to Cereblon, part of the ubiquitin ligase complex thought to mediate the teratogenesis of thalidomide(104). Further studies demonstrated that

lenalidomide and pomalidomide promoted Cereblon degradation of Ikaros and Aiolos which was critical for the anti-myeloma effect(105-107). A number of ongoing questions remain regarding the mechanism of action of the thalidomide analogues. Whether alterations in chemical structure can separate the anti-myeloma effects of the thalidomide analogues from the teratogenic effects remains to be seen. Whilst Aiolos has been reported to be required for high affinity bone marrow plasma cells, the role of Ikaros in plasma cells remains to be determined(108). Whether these transcription factors can be specifically targeted in myeloma therapy rather than through an intermediate in Cereblon also remains to be investigated. To our knowledge thalidomide analogues have not been reported to be therapeutically useful in depletion of plasma cells outside of the PCNs. It is likely that their mechanisms of action are therefore specific to hijacked pathways in plasma cell myeloma and not generic to all ASCs.

Proteasome inhibitors are the second class of agents used in myeloma that have significantly improved clinical outcomes. As their name suggests proteasome inhibitors inhibit degradation of intracellular proteins via the proteasome which is responsible for the degradation of 80-90% of intracellular proteins(109, 110). It is thought that blockade of the proteasome causes an imbalance in intracellular proteins that can result in cell cycle arrest and apoptosis(111). Bortezomib's inhibitory effects on NF κ B were initially thought to be critical to its effects in myeloma, however evidence suggests that it can also activate the canonical NF κ B pathway indicating that the exact mechanisms by which proteasome inhibitors work remain elusive(112, 113). Most recently, it has been proposed that one proteasome inhibitor may indirectly inhibit proteasomes in malignant plasma cells through post-translational modifications; although the mechanism underlying this action is poorly understood(114).

Two proteasome inhibitors are currently licensed. Bortezomib is a reversible inhibitor of the proteasome with limited single drug efficacy in myeloma (115) but combining it with other anti-myeloma therapies has produced marked improvements in responsiveness(116-119). Mechanistically these observations are

logical, as bortezomib may block proteasome degradation of the apoptotic signals generated by other therapies leading to cellular death. Carfilzomib is the second proteasome inhibitor to be licensed and binds irreversibly to the proteasome. Carfilzomib is thought to be more specific with less off target effects than bortezomib(120). Interestingly, carfilzomib is able to induce responses in patients that have become refractory to bortezomib, albeit only in the minority of those treated(121).

Proteasome inhibitors have been used in settings outside of the PCNs as ASC, compared to T and B lymphocytes, appear particularly vulnerable to proteasome inhibition(122). It is presumed that the production of large amounts of immunoglobulin and the resultant endoplasmic stress in ASC makes them susceptible to the imbalances in cellular proteins that result from proteasome inhibition and the unfolded protein response. Successful depletion of ASCs and clinical improvements with bortezomib therapy has been reported in a number of autoimmune conditions including thrombotic thrombocytopenic purpura(123, 124), SLE(125) and myasthenia gravis(126). These case reports suggest that proteasome inhibition is effective in targeting ASCs in the non-malignant setting as well as in PCNs.

Whilst the above therapies have significantly improved outcomes in patients with PCNs, and are showing promise in autoimmune conditions, additional therapies targeting plasma cells are still warranted as PCNs remain incurable for the vast majority of patients. Additionally, improved plasma cell therapies in autoimmune conditions are likely to translate into improved patient outcomes. Below we will discuss other therapeutic targets for plasma cells, some already available and in trials, others being more theoretical but promising future targets.

Targeting Antibody-secreting cell production/differentiation

Targeting of plasma cell production is unlikely to be effective in the PCNs as they are thought to have evolved the ability to self-replicate with no convincing evidence of clonotypical B cells being a source for additional malignant plasma cells(71, 127).

Nevertheless, Rituximab, a monoclonal anti-CD20 antibody that causes apoptosis and antibody dependent cell-mediated cytotoxicity (ADCC) of B cells has been trialled in plasma cell myeloma patients with partial response reported in the minority of patients(128). These results should be interpreted with some caution as a minority of patients with PCN's have malignant plasma cells that aberrantly express CD20(129). Hence, the effects of Rituximab may represent direct targeting of these malignant cells rather than evidence that targeting the B cell compartment is effective in myeloma patients.

Contrary to PCN's, targeting of B cells and therefore plasma cell differentiation, mechanistically warrants investigation in autoimmune conditions. Evidence suggests that ASCs that produce self-reactive antibodies in autoimmune disorders are not entirely comprised of long-lived plasma cells. Instead, ASC populations in flares of SLE and Sjogren's syndrome have significant contributions from the B cell compartment(130, 131). Therefore targeting the B cell reservoir of potentially auto-reactive plasma cells may be beneficial. Given its critical role in B cell biology, the BCR and its downstream signalling pathway is a particularly attractive therapeutic target in B cells. Bruton's Tyrosine Kinase (BTK) is a critical intermediate in the BCR signalling cascade. Overexpression of BTK in transgenic mice leads to a SLE-like phenotype with production of auto-antibodies and an autoimmune-like disease in the kidneys, lungs and salivary glands(132). Additionally BTK inhibition abrogated glomerulonephritis in a mouse model of SLE(133). Ibrutinib is the only currently FDA approved BTK inhibitor and has shown efficacy in lymphoproliferative disorders including CLL and mantle cell lymphoma(134, 135). At this time, we are unaware of reports on Ibrutinib's efficacy in patients with autoimmune conditions, but investigation may be warranted.

The use of Rituximab to deplete B cells has been trialled in patients with SLE but with disappointing results; no detectable improvement in clinical outcomes(136, 137). A number of limitations in these studies, including the enrolment of patients with mild to moderate disease rather than severe SLE, have meant that a role for Rituximab in SLE remains controversial(138). Evidence from non-randomised data sets suggest

that Rituximab may have efficacy as a salvage treatment in patients with refractory SLE(139, 140). The soluble BAFF inhibitor, Belimumab, has had more clinical success as an agent that depletes B cells in patients with SLE. Belimumab has been FDA approved and showed efficacy particularly in patients with mucocutaneous or musculoskeletal manifestations(141). The difference in effectiveness of Rituximab compared to Belimumab is likely to be multifactorial. Unlike treatment with Rituximab, in which patients are able to maintain a splenic plasma cell population, there is evidence that Belimumab can directly suppress ASC numbers in addition to B cells(142, 143). This is, however, unlikely to be the only reason as Atacicept, an alternative inhibitor of both BAFF and APRIL, failed to improve outcomes in patients with SLE(144). More importantly, BAFF and APRIL are involved in the extrinsic survival signals for ASCs as demonstrated in BCMA knockout mice(14). A plethora of alternative B cell targeting therapies are currently being trialled in patients with autoimmune conditions, the outcomes of which we await with anticipation (Reviewed in (145)).

Targeting Antibody-secreting cell transcription factors and epigenetic modulation

A number of transcription factors are known to be critical for ASCs and are therefore appealing targets for eliminating ASCs. Transcription factors are traditionally difficult to directly target due to an absence of deep pockets (unlike kinases) and their predominant positioning inside the nucleus requiring any therapeutic to penetrate the nuclear membrane(146). Nevertheless, suppression of transcription factors has been attempted. One of the most advanced is inhibition of STAT3, which is associated with a number of malignancies. Using oligo-deoxynucleotide decoys that inhibit transcriptional activities of STAT-3, promising results have been shown in several cancer cell lines(147, 148).

IRF4 is an ideal target for ASCs. Mice lacking IRF4 survive to a mature age with the predominant phenotype seen in the immune system secondary to impaired B and T cell maturation(149). Induced deletion of IRF4 in the B cell compartment leads to a loss of germinal centres and to a near complete loss of normal ASCs(19).

Additionally, human myeloma cells have been shown to rely on IRF4 in that shRNA

suppression was toxic in all myeloma lines tested(23). A number of downstream targets of IRF4 in myeloma cell lines were identified including MYC, which is a critical regulator of cell proliferation(23). IRF4 is therefore an ideal target for plasma cells, both malignant and non-malignant, as suppression would be expected to reduce plasma cell numbers with minimal side effects. One should remain cautious, however, as recent discoveries have unveiled far reaching roles of IRF4 in T cells including regulation of fat metabolism, hinting at possible complications should successful targeting of IRF4 be developed(150).

Thalidomide analogues, lenalidomide and pomalidomide are known to suppress IRF4. The current paradigm is that IRF4 suppression is due to thalidomide analogues' actions on Cereblon leading to degradation of Ikaros and Aiolos, which then leads to a reduction in IRF4(105-107). Whether Ikaros and Aiolos directly suppress IRF4 remains to be determined. Interestingly, forced expression of IRF4 did not protect myeloma cell lines from lenalidomide treatment suggesting that other factors may also be affected(105). Bortezomib treatment has also been shown to suppress IRF4 in human myeloma cell lines(151). More direct targeting of IRF4 has been attempted with microRNA's. MicroRNA's are small, non coding RNA's which regulate gene expression by targeting messenger RNA's for cleavage or repression of translation(152). By screening microRNA's against IRF4, one group has demonstrated that microRNA-125b is able to suppress IRF4 expression in human myeloma cell lines leading to autophagy and apoptosis. Additionally, using a humanised mouse model, the same group showed that microRNA-125b was also effective in vivo, suggesting IRF4 may become targetable in the near future(153).

Other transcriptional factors that may be good candidates to target in ASC include Blimp-1 and XBP1. Blimp-1 is critical for maturation of ASCs and the absence of Blimp-1 has been shown to reduce antibody secretion(25).Blimp-1 mutations have been reported in myeloma patients although their functional consequences remain unclear(85). Blimp-1 deletions have been more commonly reported in diffuse large B cell lymphoma, particularly in those of the activated B cell (ABC) phenotype, where it has been postulated to act as a tumour suppressor(154). Blimp-1 repression has

been shown to lead to cell death and the reduction of anti-apoptotic proteins in some human myeloma cell lines(151). As Blimp-1 is activated by IRF4 in normal plasma cells, therapeutics that target IRF4 would be expected to also suppress Blimp-1. Whether specific targeting of Blimp-1 would enhance the targeted reduction of IRF4 remains to be determined(15).

Similar to Blimp-1, XBP1 appears to be a reasonable transcriptional target in plasma cells. ASCs can survive XBP1 deletion but they lose their ability to secrete large amounts of antibody(28). Worryingly, suppression of XBP1 by shRNA's induced bortezomib resistance in human myeloma cell lines(155). Raised and reduced XBP1 amounts have been shown subsequently to correlate with good or poor responses to bortezomib therapy respectively(156, 157). Interestingly, increasing and decreasing XBP1 amounts in cell lines only partially altered sensitivity to bortezomib suggesting that loss of XBP1 may be a marker of resistance rather the specific mechanism(156). Regardless, the association between XBP1 and bortezomib responsiveness highlights concerns regarding the uncertain effects of modulating transcription factors. Whether these changes are solely applicable to patients with plasma cell myeloma where the survival program of the plasma cells has already been corrupted, or whether they would also occur in patients with autoimmune disease remains unclear.

In addition to directly targeting the transcriptional profile of ASCs, improved understanding of epigenetics has led to the use of drugs in PCNs whose primary function is believed to be modulating gene expression through epigenetic pathways. The most advanced of these are the histone de-acetylase (H-DAC) inhibitors. Two pan H-DAC inhibitors have reached phase 3 human trials in PCNs. The first, Vorinostat, showed a mild improvement in progression free survival when combined with bortezomib compared to bortezomib and placebo, although the clinical relevance of this improvement remains unclear(158). The second, Panobinostat, has recently shown an improvement in number of refractory/relapsed myeloma patients achieving complete or near complete response and progression free survival when combined with bortezomib and dexamethasone(159). The optimal integration of H-

DAC inhibitors into the clinic against PCNs remains to be determined, however, they do appear to have some efficacy. Importantly, recent evidence suggest that the pan H-DAC inhibitors can also reduce germinal centre and ASC's populations in two mouse models of SLE suggesting possibly utility in autoimmune conditions(160).

Targeting Antibody-secreting cell survival

ASCs have both intrinsic pathways and extrinsic survival signals that can be targeted to reduce their survival. As described previously, a number of external factors are thought to be critical in promoting plasma cell survival. IL-6 is reported to be an important survival signal with several myeloma cell lines requiring exogenous IL-6 to survive(161). Two drugs targeting IL-6 have reached human trials. Siltuximab, a monoclonal antibody against IL-6, has been used in two randomised phase 2 trials in conjunction with either bortezomib or melphalan in PCN patients. Unfortunately in both regimens, Siltuximab did not improve patient outcomes over the control regimen(162, 163). Tocilizumab, a humanised monoclonal antibody targeting the IL-6 receptor, has never been formally reported in targeting ASCs in the PCNs, however it has been approved for therapy of Rheumatoid arthritis(164). While Tocilizumab's effectiveness in Rheumatoid arthritis may be due in part to blockade of IL6 signalling on ASCs, its clinical effectiveness is more likely due to its effects on the myriad other cells that IL-6 stimulates(164). Although blockade of IL-6 has had disappointing results in the PCNs, blockade of other external survival signals may, in time, prove to be a useful therapeutic approach. We feel that further research into the PC survival niche will identify new targets for therapy, individual and combined, and that the potential efficacy of this approach is foreshadowed by that of Mcl-1 deletion(14).

Mcl-1 is an attractive target for eliminating the ASC population. Whilst partial deletion of Mcl-1, as demonstrated by heterozygote deletion, resulted in a partial reduction in ASC numbers, this decrease was modest(14). Therefore, if Mcl-1 suppression were to be therapeutically effective, amounts would need to be reduced to below those seen in the heterozygote state(14). Adding to Mcl-1's attractiveness as a target, it appears to be the dominant survival protein in the PCNs(165, 166). Mcl-1 inhibitors are in development and are at a pre-clinical stage of

testing(167). For the most part, small molecule inhibitors, which bind and inhibit Mcl-1 interactions with its downstream partners including Bim, have been investigated(167). Other methods of reducing Mcl-1 are also being tested including microRNA's(168). Concerns remain about whether a safe therapeutic window can be found for Mcl-1 inhibitors as complete Mcl-1 deletion is lethal at the blastocyst stage in mice(149). Additionally, Mcl-1 has been shown to be critical for a number of normal tissues including liver,(169) neurons,(170) and cardiac muscle(171). If Mcl-1 inhibition cannot be delivered safely, then identification and targeting of the upstream modulators of Mcl-1 may provide alternatives to targeting internal survival pathways in ASCs.

Alternatively, partial Mcl-1 inhibition in combination with other agents may be therapeutically viable. Our laboratory has demonstrated that other Bcl-2 family members contribute to survival of ASCs as they migrate to the bone marrow(55, 56). Therefore combining inhibitors of Mcl-1 with those of other Bcl-2 family members may have synergistic effects. Inhibitors of Bcl-xL and Bcl-2 are in pre-clinical and clinical trials respectively. As with Mcl-1, Bcl-xL is critical for a number of other cell types, especially platelets, with thrombocytopenia limiting therapeutic use of the BH3 mimetic ABT-737(172). The resultant development of ABT-199, a Bcl-2 selective inhibitor, has abrogated this undesirable side effect. ABT-199, has been safely used in patients with chronic lymphocytic leukaemia with excellent efficacy(173, 174). ABT-199 treatment alone would not be expected to have a major effect on normal ASC populations, although in vitro data suggest that Bcl-2 inhibition may effect a sub-group of myeloma with translocations involving chromosome 11 and 14 that appear to have a stronger dependence on Bcl-2 than on Mcl-1(175). Combining Mcl-1 inhibitors with other therapies such as thalidomide analogues or proteasome inhibitors to target multiple critical pathways in antibody secreting cells may also be of use.

Targeting cell surface markers

A number of antibodies against cell surface markers on antibody secreting cells have been developed. For the most part they have been trialled in the plasma cell

neoplasms, but if effective, may be useful in other diseases involving plasma cells. The aim of these antibodies is to mark the plasma cells for killing much like Rituximab marks B cells and has revolutionised treatment for B cell lymphomas(176). The bound antibody results in the death of its target through several possible mechanisms including ADCC, complement dependent lysis and triggering intracellular signalling cascades that alter cellular outcomes including apoptosis(177). More recently, attempts have been made to deliver cytotoxic therapies directly to cells using antibodies against cellular surface molecules.

At least three antibodies against ASCs are currently in human trials. The most advanced is Elotuzumab, a monoclonal antibody against SLAMF7, which is highly expressed on activated T cells, activated B cells, NK cells, antibody secreting cells and very highly expressed in PCNs.(178, 179) SLAMF7 is a member of the SLAM family, a group of Src homology 2 domain adaptors(180). Elotuzumab appears to have minimal direct effects on myeloma cells(181), but rather appears to cause ADCC of myeloma cells and interferes with their ability to interact with the bone marrow stroma(182, 183). A recent randomised, controlled trial has shown that Elotuzumab in conjunction with lenalidomide and dexamethasone improved outcomes in patients with relapsed/refractory myeloma compared to lenalidomide and dexamethasone alone(184). Currently no data exists for Elotuzumab efficacy in patients outside of plasma cell myeloma however its promising results in the malignant setting suggest it also may be useful in non-malignant settings.

We are aware of two other antibodies in clinical trial currently. Daratumumab is a monoclonal antibody directed against CD38 that shows overall response rates of 75% in patients with relapsed/refractory myeloma in conjunction with lenalidomide and dexamethasone(185, 186). Impressively Daratumumab is relatively well tolerated when combined with a number of currently used regimens(185, 187). The second antibody is Indatuximab Ravtansine, a chimerised monoclonal antibody against CD138 coupled to the cytotoxic agent maytansinoid DM4. Overall response rates in relapsed/refractory plasma cell myeloma patients are reported at 78% when combined with lenalidomide and dexamethasone(188). As with Elotuzumab, no data

are currently available for Daratumumab or Indatuximab Ravidansine outside of the plasma cell myeloma setting, however in time they may prove to be useful agents in autoimmune conditions.

Looking to the future

Despite ongoing breakthroughs in understanding the biology of ASCs, both normal and malignant, the plasma cell neoplasms and autoimmune conditions remain significant health problems. While new therapies are on the horizon and will hopefully soon be available to improve patient outcomes (Figure 4); we believe that understanding normal plasma cell biology will be critical for identifying novel targets. In an ideal world, therapeutics would not only be specific to the ASC population but also be critical to their survival and function. Recently, our laboratory and collaborators have analysed the entire ASC transcriptome(189). This revealed that ASCs have a transcriptome distinctive from all other B cell populations and it identified a number of novel targets specific to ASCs such as Tribbles-1 and Creb3l2(189). Correlation and validation of a number of these proteins in human ASC populations are currently underway. If confirmed, the protein products of these transcripts would warrant further investigation into their functional roles in ASCs. It is hoped that identification of these new targets in conjunction with improved methods of inhibiting known targets will lead to effective therapies for all patients with diseases mediated by ASCs.

Figure Legends

Figure 1. Pathways of differentiation for antibody secreting cells

Antibody secreting cells (ASC) or Plasma cells (PC) can differentiate through three pathways. Firstly B1 and marginal zone B cells can differentiate without antigenic stimulation (top). Secondly, follicular and marginal zone B cells, on binding of cognate antigen, can develop in ASC via an extrafollicular pathway (middle). Lastly, follicular and marginal zone B cells, on binding with cognate antigen can enter the germinal center, which can generate memory B cells and ASCs (bottom). Memory B cells are able to rapidly differentiate into ASCs on rechallenge (bottom right). After

differentiation, PC are required to migrate to their survival niches for persistence (far right)

Figure 2. Transcriptional program that controls antibody secreting cell differentiation
Studies into transcription factors have revealed the interplay between pro-ASC factors (coloured red on the left) and pro-B cell factors (coloured blue on the right). IRF4 is postulated to be the critical initiating factor of the antibody secreting cell (ASC) program. It is regulated within the germinal centre by MITF(20). IRF4 and IRF8 are thought to compete to drive differentiation towards ASC or B cell fates respectively(18). IRF4 directly binds and upregulates Blimp-1(21) which in turn is thought to suppress a number of B cell genes such as BCL6 and PAX5(25). The suppression of PAX5 is then thought to allow for upregulation of XBP1(15). Conversely, B cell promoting factors such as PAX5 and BACH2 suppress the ASC transcriptional program(15).

Figure 3. Available drugs and cell populations targeted

A number of available therapies have been trialled to reduce antibody secreting cell (ASC) numbers, targeting different steps within the pathway to ASC production. The arrows (inclusive) represent the cell populations believed to be targeted by specific classes of drugs. Some classes such as BTK inhibitors and Rituximab specifically target the B cell pool(143). Others such as proteasome inhibitors and thalidomide analogues appear to specifically target activated B cells and ASC populations(103, 122). BAFF inhibitors and pan-HDAC inhibitors appear to target parts of both the B cells and ASC compartments(142, 160).

Figure 4. Targetable sites for drugs against antibody secreting cells

There are multiple sites that Antibody secreting cells (ASC) can be targeted. Overall we identified five major sites on, within or around ASC that can be targeted: External survival signals and bone marrow niche, intracellular survival pathways, intracellular signalling pathways, cell surface markers and transcription factors (specific examples of each are shown). At our current understanding of ASC biology, we believe that

these five broad sites are the most likely to be successfully targeted for future therapeutics against ASC.

REFERENCES

1. Delves PJ, Roitt IM. The immune system. First of two parts. *N Engl J Med*.2000;343:37-49.
2. Siegrist C. Vaccine Immunology. In: Plotkin SA, Orenstein W, Offit PA, eds. *Vaccines*. 6th ed. Philadelphia: Saunders Elsevier, 2012:14-32.
3. Savage HP, Baumgarth N. Characteristics of natural antibody-secreting cells. *Ann N Y Acad Sci*.2015.
4. MacLennan IC, et al. Extrafollicular antibody responses. *Immunol Rev*.2003;194:8-18.
5. Fairfax KA, Kallies A, Nutt SL, Tarlinton DM. Plasma cell development: from B-cell subsets to long-term survival niches. *Semin Immunol*.2008 20:49-58.
6. Bortnick A, Allman D. What is and what should always have been: long-lived plasma cells induced by T cell-independent antigens. *J Immunol*.2013;190:5913-5918.
7. Cassese G, et al. Plasma cell survival is mediated by synergistic effects of cytokines and adhesion-dependent signals. *J Immunol*.2003;171:1684-1690.
8. Crotty S. Follicular helper CD4 T cells (TFH). *Annu Rev Immunol*.2011;29:621-663.
9. Victora GD, Nussenzweig MC. Germinal centers. *Annu Rev Immunol*.2012;;30.
10. Shulman Z, et al. T follicular helper cell dynamics in germinal centers. *Science*.2013;341:673-677.
11. Shlomchick MJ, Weisel F. Germinal center selection and the development of memory B and plasma cells. *Immunol Rev*.2012;247:52-63.
12. Ahmed R, Gray D. Immunological memory and protective immunity: understanding their relation. *Science*.1996;272:54-60.
13. Gitlin AD, Shulman Z, Nussenzweig MC. Clonal selection in the germinal centre by regulated proliferation and hypermutation. *Nature*.2014;509:637-640.

14. Peperzak V, et al. Mcl-1 is essential for the survival of plasma cells. *Nat Immunol.*2013;14:290-297.
15. Nutt SL, Taubenheim N, Hasbold J, Corcoran LM, Hodgkin PD. The genetic network controlling plasma cell differentiation. *Semin Immunol.*2011;23:341-349.
16. Nutt SL, Hodgkin PD, Tarlinton DM, Corcoran LM. The generation of antibody-secreting plasma cells. *Nat Rev Immunol.*2015;15:160-171.
17. Shaffer AL, Emre NC, Romesser PB, Staudt LM. IRF4: Immunity. Malignancy! Therapy? *Clin Cancer Res.*2009;15:2954-2961.
18. Carotta S, et al. The transcription factors IRF8 and PU.1 negatively regulate plasma cell differentiation. *J Exp Med* 2014;211:2169-2181.
19. Willis SN, et al. Transcription factor IRF4 regulates germinal center cell formation through a B cell-intrinsic mechanism. *J Immunol.*2014;192:3200-3206.
20. Lin L, Gerth AJ, SL. P. Active inhibition of plasma cell development in resting B cells by microphthalmia-associated transcription factor. *J Exp Med.*2004;200:115-122.
21. Sciammas R, Shaffer AL, Schatz JH, Zhao H, Staudt LM, Singh H. Graded expression of interferon regulatory factor-4 coordinates isotype switching with plasma cell differentiation. *Immunity.*2006;25:225-236.
22. Kwon H, et al. Analysis of interleukin-21-induced Prdm1 gene regulation reveals functional cooperation of STAT3 and IRF4 transcription factors. *Immunity.*2009;31:941-952.
23. Shaffer AL, et al. IRF4 addiction in multiple myeloma. *Nature.*2008;454:226-231.
24. Nutt SL, Fairfax KA, Kallies A. BLIMP1 guides the fate of effector B and T cells. *Nat Rev Immunol.*2007;7:923-927.
25. Kallies A, et al. Initiation of plasma-cell differentiation is independent of the transcription factor Blimp-1. *Immunity.*2007;26:555-566.
26. Shapiro-Shelef M, Lin KI, McHeyzer-Williams LJ, Liao J, McHeyzer-Williams MG, Calame K. Blimp-1 is required for the formation of immunoglobulin secreting plasma cells and pre-plasma memory B cells. *Immunity.*2003;19:607-620.

27. Reimold AM, et al. Plasma cell differentiation requires the transcription factor XBP-1. *Nature*.2001;412:300-307.
28. Taubenheim N, Tarlinton DM, Crawford S, Corcoran LM, Hodgkin PD, Nutt SL. High rate of antibody secretion is not integral to plasma cell differentiation as revealed by XBP-1 deficiency. *J Immunol*.2012;189:3328-3338.
29. Todd DJ, et al. XBP1 governs late events in plasma cell differentiation and is not required for antigen-specific memory B cell development. *J Exp Med*.2009;206:2151-2159.
30. Todd DJ, Lee AH, Glimcher LH. The endoplasmic reticulum stress response in immunity and autoimmunity. *Nat Rev Immunol*.2008;8:663-674.
31. Yoshida H, Matsui T, Yamamoto A, Okada T, Mori K. XBP1 mRNA is induced by ATF6 and spliced by IRE1 in response to ER stress to produce a highly active transcription factor. *Cell*.2001;107:881-891.
32. Walter P, Ron D. The unfolded protein response: from stress pathway to homeostatic regulation. *Science*.2011;334:1081-1086.
33. Cobaleda C, Schebesta A, Delogu A, Busslinger M. Pax5: the guardian of B cell identity and function. *Nat Immunol*.2007;8:463-470.
34. Nutt SL, Heavey B, Rolink AG, Busslinger M. Commitment to the B-lymphoid lineage depends on the transcription factor Pax5. *Nature*.1999;401:556-562.
35. Kallies A, et al. Plasma cell ontogeny defined by quantitative changes in blimp-1 expression. *J Exp Med*.2004 200:967-977.
36. Kerckaert JP, Deweindt C, Tilly H, Quief S, Lecocq G, Bastard C. LAZ3, a novel zinc-finger encoding gene, is disrupted by recurring chromosome 3q27 translocations in human lymphomas. *Nat Genet*.1993;5:66-70.
37. Ye BH, Rao PH, Chaganti RS, Dalla-Favera R. Cloning of bcl-6, the locus involved in chromosome translocations affecting band 3q27 in B-cell lymphoma. *Cancer Res*.1993;53:2732-2735.
38. Ye BH, et al. The BCL-6 proto-oncogene controls germinal-centre formation and Th2-type inflammation. *Nat Genet*.1997;16:161-170.
39. Dent AL, Shaffer AL, Yu X, Allman D, Staudt LM. Control of inflammation, cytokine expression, and germinal center formation by BCL-6. *Science*.1997;276:589-592.

40. Fukuda T, et al. Disruption of the Bcl6 gene results in an impaired germinal center formation. *J Exp Med*.1997;186:439-448.
41. Basso K, Dalla-Favera R. BCL6: master regulator of the germinal center reaction and key oncogene in B cell lymphomagenesis. *Adv Immunol*.2010;105:193-210.
42. Calame KL, Lin KI, Tunyaplin C. Regulatory mechanisms that determine the development and function of plasma cells. *Annu Rev Immunol*.2003;;21:205-230.
43. Lee CH, et al. Regulation of the germinal center gene program by interferon (IFN) regulatory factor 8/IFN consensus sequence-binding protein. *J Exp Med*.2006;203:63-72.
44. Radbruch A, et al. Competence and competition: the challenge of becoming a long-lived plasma cell. *Nat Rev Immunol*.2006;6:741-750.
45. Terstappen LW, Johnsen S, Segers-Nolten IM, Loken MR. Identification and characterization of plasma cells in normal human bone marrow by high-resolution flow cytometry. *Blood*.1990;76:1739-1747.
46. Sze DM, Toellner KM, García de Vinuesa C, Taylor DR, MacLennan IC. Intrinsic constraint on plasmablast growth and extrinsic limits of plasma cell survival. *J Exp Med*.2000;192:813-821.
47. Blink EJ, Light A, Kallies A, Nutt SL, Hodgkin PD, Tarlinton DM. Early appearance of germinal center-derived memory B cells and plasma cells in blood after primary immunization. *J Exp Med*.2005;201:545-554.
48. Minges Wols HA, Underhill GH, Kansas GS, Witte PL. The role of bone marrow-derived stromal cells in the maintenance of plasma cell longevity. *J Immunol*.2002;169:4213-4221.
49. Kometani K, Kurosaki T. Differentiation and maintenance of long-lived plasma cells. *Curr Opin Immunol*.2015;33:64-69.
50. Vincent FB, Saulep-Easton D, Figgett WA, Fairfax KA, Mackay F. The BAFF/APRIL system: emerging functions beyond B cell biology and autoimmunity. *Cytokine Growth Factor Rev*.2013;24:203-215.
51. Oracki SA, Walker JA, Hibbs ML, Corcoran LM, Tarlinton DM. Plasma cell development and survival. *Immunol Rev*.2010;237:140-159.

52. Odendahl M, et al. Generation of migratory antigen-specific plasma blasts and mobilization of resident plasma cells in a secondary immune response. *Blood*.2005;105:1614-1621.
53. Wrammert J, et al. Rapid and massive virus-specific plasmablast responses during acute dengue virus infection in humans. *Journal of Virology*.2012;86:2911-2918.
54. O'Connor BP, et al. BCMA is essential for the survival of long-lived bone marrow plasma cells. *J Exp Med*.2004;199:91-98.
55. Carrington EM, et al. BH3 mimetics antagonizing restricted prosurvival Bcl-2 proteins represent another class of selective immune modulatory drugs. *Proc Natl Acad Sci U S A*.2010;107:10967-10971.
56. Vikstrom I, et al. Mcl-1 is essential for germinal center formation and B cell memory. *Science*.2010;330:1095-1099.
57. Ellyard JI, Avery DT, Phan TG, Hare NJ, Hodgkin PD, Tangye SG. Antigen-selected, immunoglobulin-secreting cells persist in human spleen and bone marrow. *Blood*.2004;103:3805-3812.
58. Halliley JL, et al. Long-Lived Plasma Cells Are Contained within the CD19(-)CD38(hi)CD138(+) Subset in Human Bone Marrow. *Immunity*.2015;43:132-145.
59. Mei HE, et al. A unique population of IgG-expressing plasma cells lacking CD19 is enriched in human bone marrow. *Blood*.2015;125:1739-1748.
60. Flores-Montero J, et al. Immunophenotype of normal vs. myeloma plasma cells: Toward antibody panel specifications for MRD detection in multiple myeloma. *Cytometry B Clin Cytom*.2015.
61. Swerdlow SH, et al. WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues, Fourth Edition Lyon: IARC, 2008.
62. Swerdlow S, et al. The International Agency for Research on Cancer: WHO Classification of Tumours of Haematopoietic and Lymphoid Tissue (IARC WHO Classification of Tumours). 4th Edition ed. Lyon: International Agency for Research on Cancer, 2008.
63. Korde N, Kristinsson SY, Landgren O. Monoclonal gammopathy of undetermined significance (MGUS) and smoldering multiple myeloma (SMM):

novel biological insights and development of early treatment strategies.

Blood.2011;117:5573-5581.

64. Landgren O, et al. Monoclonal gammopathy of undetermined significance (MGUS) consistently precedes multiple myeloma: a prospective study.

Blood.2009;113:5412-5417.

65. Abadie J, Verma P, Howard RS, Kuehl WM. A monoclonal gammopathy precedes multiple myeloma in most patients.

Weiss BM, . Blood.2009;113:5418-5422.

66. Kyle RA, et al. Prevalence of monoclonal gammopathy of undetermined significance. N Engl J Med.2006;354:1362-1369.

67. Wadhera RK, Rajkumar SV. Prevalence of monoclonal gammopathy of undetermined significance: a systematic review. Mayo Clin Proc.2010;85:933-942.

68. King RL, Howard MT, Hodnefield JM, Morice WG. IgM multiple myeloma: pathologic evaluation of a rare entity. Am J Clin Pathol.2013;140:519-524.

69. Hunter ZR, et al. The genomic landscape of Waldenstrom macroglobulinemia is characterized by highly recurring MYD88 and WHIM-like CXCR4 mutations, and small somatic deletions associated with B-cell lymphomagenesis. Blood 2014;123:1637-1646.

70. Dispenzieri A, et al. Prevalence and risk of progression of light-chain monoclonal gammopathy of undetermined significance: a retrospective population-based cohort study. Lancet.2010;375:1721-1728.

71. Hajek R, Okubote SA, Svachova H. Myeloma stem cell concepts, heterogeneity and plasticity of multiple myeloma. Br J Haematol.2013;163:551-564.

72. Vescio RA, et al. Myeloma Ig heavy chain V region sequences reveal prior antigenic selection and marked somatic mutation but no intracloal diversity. J Immunol.1995;155:2487-2497.

73. Bakkus MH, Heirman C, Van Riet I, Van Camp B, Thielemans K. Evidence that multiple myeloma Ig heavy chain VDJ genes contain somatic mutations but show no intracloal variation. Blood.1992;80:2326-2335.

74. Hallek M, Bergsagel PL, Anderson KC. Multiple myeloma: increasing evidence for a multistep transformation process. Blood.1998;91:3-21.

75. Agarwal A, Ghobrial IM. Monoclonal gammopathy of undetermined significance and smoldering multiple myeloma: a review of the current understanding of epidemiology, biology, risk stratification, and management of myeloma precursor disease. *Clin Cancer Res.*2013;19:985-994.
76. Morgan GJ, Kaiser MF. How to use new biology to guide therapy in multiple myeloma. *Hematology Am Soc Hematol Educ Program.*2012;2012:342-349.
77. Fonseca R, et al. Genomic abnormalities in monoclonal gammopathy of undetermined significance. *Blood.*2002;100:1417-1424.
78. Chiecchio L, et al. Timing of acquisition of deletion 13 in plasma cell dyscrasias is dependent on genetic context. *Haematologica.*2009;94:1708-1713.
79. Sawyer JR. The prognostic significance of cytogenetics and molecular profiling in multiple myeloma. *Cancer Genet.*2011;204:3-12.
80. Liebisch P, Döhner H. Cytogenetics and molecular cytogenetics in multiple myeloma. *Eur J Cancer.*2006;42:1520-1529.
81. Zhan F, et al. The molecular classification of multiple myeloma. *Blood.*2006 108:2020-2028.
82. Bergsagel PL, Kuehl WM, Zhan F, Sawyer J, Barlogie B, Shaughnessy J Jr. Cyclin D dysregulation: an early and unifying pathogenic event in multiple myeloma. *Blood.*2005 106:296-303.
83. Boyd KD, Pawlyn C, Morgan GJ, Davies FE. Understanding the molecular biology of myeloma and its therapeutic implications. *Expert Rev Hematol.*2012 5:603-617.
84. Kuehl WM, PL. B. Multiple myeloma: evolving genetic events and host interactions. *Nat Rev Cancer.*2002;2:175-187.
85. Chapman MA, et al. Initial genome sequencing and analysis of multiple myeloma. *Nature.*2011;471:467-472.
86. Lohr JG, et al. Widespread genetic heterogeneity in multiple myeloma: implications for targeted therapy. *Cancer Cell.*2014;25:91-101.
87. Keats JJ, et al. Clonal competition with alternating dominance in multiple myeloma. *Blood.*2012;120:1067-1076.
88. Brioli A, Melchor L, Cavo M, Morgan GJ. The impact of intra-clonal heterogeneity on the treatment of multiple myeloma. *Br J Haematol.*2014.

89. López-Corral L, et al. SNP-based mapping arrays reveal high genomic complexity in monoclonal gammopathies, from MGUS to myeloma status. *Leukemia*.2012;26:2521-2529.
90. Walker BA, Morgan GJ. Could DNA methylation become a useful measure for multiple myeloma prognoses? *Expert Rev Hematol*.2011;4:125-127.
91. Manier S, Sacco A, Leleu X, Ghobrial IM, Roccaro AM. Bone marrow microenvironment in multiple myeloma progression. *J Biomed Biotechnol*.2012;2012.
92. Kuehl WM, Bergsagel PL. Molecular pathogenesis of multiple myeloma and its premalignant precursor. *J Clin Invest*.2012;122:3456-3463.
93. Kawano M, et al. Autocrine generation and requirement of BSF-2/IL-6 for human multiple myelomas. *Nature*.1988 332:83-85.
94. Hideshima T, Mitsiades C, Tonon G, Richardson PG, Anderson KC. Understanding multiple myeloma pathogenesis in the bone marrow to identify new therapeutic targets. *Nat Rev Cancer*.2007 7:585-598.
95. Keats JJ, et al. Promiscuous mutations activate the noncanonical NF-kappaB pathway in multiple myeloma. *Cancer Cell*.2007;12:131-144.
96. Annunziata CM, et al. Frequent engagement of the classical and alternative NF-kappaB pathways by diverse genetic abnormalities in multiple myeloma. *Cancer Cell*.2007;12:115-130.
97. Cotran RS, Kumar V, T C. Robbins Pathological Basis of Disease. 6th ed. Philadelphia, Pennsylvania: Elsevier Science, 1999.
98. Squatrito D, et al. Pathogenesis and potential therapeutic targets in systemic lupus erythematosus: from bench to bedside. *Auto Immun Highlights*.2014;5:33-45.
99. Watson CJ, Dark JH. Organ transplantation: historical perspective and current practice. *Br J Anaesth*.2012;108:i29-42.
100. Jordan SC, Vo AA. Curr Opin Organ Transplant. Donor-specific antibodies in allograft recipients: etiology, impact and therapeutic approaches.2014;19:591-597.
101. Barlogie B, Mitchell A, van Rhee F, Epstein J, Morgan GJ, Crowley J. Curing myeloma at last: defining criteria and providing the evidence. *Blood*.2014;124:3043-3051.

102. Mellin GW, Katsenstein M. The saga of thalidomide. Neuropathy to embryopathy, with case reports of congenital anomalies. *New England Journal of Medicine*.1962;267.
103. Shortt J, Hsu AK, Johnstone RW. Thalidomide-analogue biology: immunological, molecular and epigenetic targets in cancer therapy. *Oncogene*.2013;5:4181-4202.
104. Ito T, et al. Identification of a primary target of thalidomide teratogenicity. *Science*.2010;327:1345-1350.
105. Lu G, et al. The myeloma drug lenalidomide promotes the cereblon-dependent destruction of Ikaros proteins. *Science*.2014;343:305-309.
106. Krönke J, et al. Lenalidomide causes selective degradation of IKZF1 and IKZF3 in multiple myeloma cells. *Science*.2014;343:301-305.
107. Gandhi AK, et al. Measuring cereblon as a biomarker of response or resistance to lenalidomide and pomalidomide requires use of standardized reagents and understanding of gene complexity. *British Journal of Haematology*.2014;164:233-244.
108. Cortés M, Georgopoulos K. Aiolos is required for the generation of high affinity bone marrow plasma cells responsible for long-term immunity. *J Exp Med*.2004;199:209-219.
109. Ciechanover A. The ubiquitin-proteasome proteolytic pathway. *Cell*.1994;79:13-21.
110. Hochstrasser M. Ubiquitin, proteasomes, and the regulation of intracellular protein degradation. *Curr Opin Cell Biol*.1995;7:215-223.
111. Dou QP, Zonder JA. Overview of proteasome inhibitor-based anti-cancer therapies: perspective on bortezomib and second generation proteasome inhibitors versus future generation inhibitors of ubiquitin-proteasome system. *Curr Cancer Drug Targets*.2014;14:518-536.
112. Orłowski RZ, et al. Phase I trial of the proteasome inhibitor PS-341 in patients with refractory hematologic malignancies. *J Clin Oncol*.2002;20:4420-4427.
113. Hideshima T, et al. Bortezomib induces canonical nuclear factor-kappaB activation in multiple myeloma cells. *Blood*.2009;114:1046-1052.

114. Pitcher DS, de Mattos-Shipley K, Tzortzis K, Auner HW, Karadimitris A, Kleijnen MF. Bortezomib Amplifies Effect on Intracellular Proteasomes by Changing Proteasome Structure. *EBioMedicine*.2015;2:642-648.
115. Jagannath S, et al. Bortezomib therapy alone and in combination with dexamethasone for previously untreated symptomatic multiple myeloma. *British Journal of Haematology*.2005;129:776-783.
116. Reeder CB, et al. Cyclophosphamide, bortezomib and dexamethasone induction for newly diagnosed multiple myeloma: high response rates in a phase II clinical trial. *Leukemia*.2009;23:1337-1341.
117. San Miguel JF, et al. Bortezomib plus melphalan and prednisone for initial treatment of multiple myeloma. *N Engl J Med*.2008;359:906-917.
118. Cavo M, et al. Bortezomib with thalidomide plus dexamethasone compared with thalidomide plus dexamethasone as induction therapy before, and consolidation therapy after, double autologous stem-cell transplantation in newly diagnosed multiple myeloma: a randomised phase 3 study. *Lancet*.2010;376:2075-2085.
119. Oakervue HE, et al. PAD combination therapy (PS-341/bortezomib, doxorubicin and dexamethasone) for previously untreated patients with multiple myeloma. *Br J Haematol*.2005;129:755-762.
120. Ruschak AM, Slassi M, Kay LE, Schimmer AD. Novel proteasome inhibitors to overcome bortezomib resistance. *J Natl Cancer Inst*.2011;103:1007-1017.
121. Jagannath S, et al. An open-label single-arm pilot phase II study (PX-171-003-A0) of low-dose, single-agent carfilzomib in patients with relapsed and refractory multiple myeloma. *Clin Lymphoma Myeloma Leuk*.2012;12:210-218.
122. Neubert K, et al. The proteasome inhibitor bortezomib depletes plasma cells and protects mice with lupus-like disease from nephritis. *Nat Med*.2008;14:748-755.
123. Shortt J, Oh DH, Opat SS. ADAMTS13 antibody depletion by bortezomib in thrombotic thrombocytopenic purpura. *N Engl J Med*.2013;368:90-92.
124. van Balen T, Schreuder MF, de Jong H, van de Kar NC. Refractory thrombotic thrombocytopenic purpura in a 16-year-old girl: successful treatment with bortezomib. *Eur J Haematol*.2014;92:80-82.

125. Alexander T, et al. The proteasome inhibitor bortezomib depletes plasma cells and ameliorates clinical manifestations of refractory systemic lupus erythematosus. *Ann Rheum Dis*.2015;74:1474-1478.
126. Gomez AM, et al. Proteasome inhibition with bortezomib depletes plasma cells and specific autoantibody production in primary thymic cell cultures from early-onset myasthenia gravis patients. *J Immunol*.2014;193:1055-1063.
127. Thiele B, et al. Next-generation sequencing of peripheral B-lineage cells pinpoints the circulating clonotypic cell pool in multiple myeloma. *Blood*.2014;123:3618-3621.
128. Kapoor P, Greipp PT, Morice WG, Rajkumar SV, Witzig TE, Greipp PR. Anti-CD20 monoclonal antibody therapy in multiple myeloma. *Br J Haematol*.2008;141:135-148.
129. Grigoriadis G, et al. Is CD20 positive plasma cell myeloma a unique clinicopathological entity? A study of 40 cases and review of the literature. *Pathology*.2012;44:552-556.
130. Al-Kindi M, Chataway TK, Colella AD, Gordon TP, eds. High-Resolution mass spectrometric sequencing of secreted autoantibody proteomes in lupus and Sjorgen's Syndrome reveals expression of public BCR clonotypes and rapid clonal turnover. 2015 4-5th June 2015; Walter and Eliza Hall Institute of Medical Research.
131. Tipton CM, et al. Diversity, cellular origin and autoreactivity of antibody-secreting cell population expansions in acute systemic lupus erythematosus. *Nat Immunol*.2015;16:755-765.
132. Kil LP, et al. Btk levels set the threshold for B-cell activation and negative selection of autoreactive B cells in mice. *Blood*.2012;119:3744-3756.
133. Mina-Osorio P, et al. Suppression of glomerulonephritis in lupus-prone NZB × NZW mice by RN486, a selective inhibitor of Bruton's tyrosine kinase. *Arthritis Rheum*.2013;65:2380-2391.
134. Byrd JC, et al. Targeting BTK with ibrutinib in relapsed chronic lymphocytic leukemia. *N Engl J Med*.2013;369:32-41.
135. Wang ML, et al. Targeting BTK with ibrutinib in relapsed or refractory mantle-cell lymphoma. *N Engl J Med*.2013;369:507-516.

136. Merrill JT, et al. Efficacy and safety of rituximab in moderately-to-severely active systemic lupus erythematosus: the randomized, double-blind, phase II/III systemic lupus erythematosus evaluation of rituximab trial. *Arthritis Rheum.*2010;62:222-233.
137. Rovin BH, et al. Efficacy and safety of rituximab in patients with active proliferative lupus nephritis: the Lupus Nephritis Assessment with Rituximab study. *Arthritis Rheum.*2012;64:1215-1226.
138. Sciascia S, Talavera-Garcia E, Roccatello D, Baldovino S, Mengatti E, Cuadrado MJ. Upcoming biological therapies in systemic lupus erythematosus. *Int Immunopharmacol.*2015;15:S1567-5769.
139. Tanaka Y, et al. Efficacy and safety of rituximab in Japanese patients with systemic lupus erythematosus including lupus nephritis who are refractory to conventional therapy. *Mod Rheumatol.*2015;9:1-27.
140. Iaccarino L, et al. Efficacy and safety of off-label use of rituximab in refractory lupus: data from the Italian Multicentre Registry. *Clin Exp Rheumatol.*2015.
141. Hui-Yuen JS, Li XQ, Askanase AD. Belimumab in systemic lupus erythematosus: a perspective review. *Ther Adv Musculoskelet Dis.*2015;7:115-121.
142. Stohl W, et al. Belimumab reduces autoantibodies, normalizes low complement levels, and reduces select B cell populations in patients with systemic lupus erythematosus. *Arthritis Rheum.*2012;64:2328-2337.
143. Mahévas M, et al. Emergence of long-lived autoreactive plasma cells in the spleen of primary warm auto-immune hemolytic anemia patients treated with rituximab. *J Autoimmun.*2015;62:22-30.
144. Isenberg D, Gordon C, Licu D, Copt S, Rossi CP, Wofsy D. Efficacy and safety of atacicept for prevention of flares in patients with moderate-to-severe systemic lupus erythematosus (SLE): 52-week data (APRIL-SLE randomised trial). [Epub ahead of print]. *Ann Rheum Dis.*2014.
145. Levesque MC. Translational Mini-Review Series on B Cell-Directed Therapies: Recent advances in B cell-directed biological therapies for autoimmune disorders. *Clin Exp Immunol.*2009;157:198-208.

146. Yeh JE, Toniolo PA, Frank DA. Targeting transcription factors: promising new strategies for cancer therapy. *Curr Opin Oncol.*2013;25:652-658.
147. Ma Y, et al. STAT3 Decoy Oligodeoxynucleotides-Loaded Solid Lipid Nanoparticles Induce Cell Death and Inhibit Invasion in Ovarian Cancer Cells. *PLoS One.*2015;10:e0124924.
148. Souissi I, et al. A STAT3-decoy oligonucleotide induces cell death in a human colorectal carcinoma cell line by blocking nuclear transfer of STAT3 and STAT3-bound NF- κ B. *BMC Cell Biol.*2011;12.
149. The Jackson Laboratory. Mouse Genome Database (MGD) at the Mouse Genome Informatics website. Bar Harbor, Maine.
150. Vasanthakumar A, et al. The transcriptional regulators IRF4, BATF and IL-33 orchestrate development and maintenance of adipose tissue-resident regulatory T cells. *Nat Immunol.*2015;16:276-285.
151. Lin FR, Kuo HK, Ying HY, Yang FH, Lin KI. Induction of apoptosis in plasma cells by B lymphocyte-induced maturation protein-1 knockdown. *Cancer Res.*2007;67:1191-1123.
152. Bartel DP. MicroRNAs: genomics, biogenesis, mechanism, and function. *Cell.*2004;116:281-297.
153. Morelli E, et al. Selective targeting of IRF4 by synthetic microRNA-125b-5p mimics induces anti-multiple myeloma activity in vitro and in vivo. *Leukemia.*2015.
154. Hangaishi A, Kurokawa M. Blimp-1 is a tumor suppressor gene in lymphoid malignancies. *Int J Hematol.*2010;91:46-53.
155. Leung-Hagesteijn C, et al. Xbp1s-negative tumor B cells and pre-plasmablasts mediate therapeutic proteasome inhibitor resistance in multiple myeloma. *Cancer Cell.*2013;24:289-304.
156. Ling SC, et al. Response of myeloma to the proteasome inhibitor bortezomib is correlated with the unfolded protein response regulator XBP-1. *Haematologica.*2012;97:64-72.
157. Gambella M, et al. High XBP1 expression is a marker of better outcome in multiple myeloma patients treated with bortezomib. *Haematologica.*2014;99:e14-16.

158. Dimopoulos M, et al. Vorinostat or placebo in combination with bortezomib in patients with multiple myeloma (VANTAGE 088): a multicentre, randomised, double-blind study. *Lancet Oncol.*2013;14:1129-1140.
159. San-Miguel JF, et al. Panobinostat plus bortezomib and dexamethasone versus placebo plus bortezomib and dexamethasone in patients with relapsed or relapsed and refractory multiple myeloma: a multicentre, randomised, double-blind phase 3 trial. *Lancet Oncol.*2014;15:1195-1206.
160. Waibel M, et al. Manipulation of B-cell responses with histone deacetylase inhibitors. *Nat Commun.*2015;6.
161. Klein B, et al. Interleukin-6 is the central tumor growth factor in vitro and in vivo in multiple myeloma. *Eur Cytokine Netw.*1990;1:193-201.
162. Orlowski RZ, et al. A phase 2, randomized, double-blind, placebo-controlled study of siltuximab (anti-IL-6 mAb) and bortezomib versus bortezomib alone in patients with relapsed or refractory multiple myeloma. *Am J Hematol.*2015;90:42-49.
163. San-Miguel J, et al. Phase 2 randomized study of bortezomib-melphalan-prednisone with or without siltuximab (anti-IL-6) in multiple myeloma. *Blood.*2014;123:4136-4142.
164. Kim GW, et al. IL-6 inhibitors for treatment of rheumatoid arthritis: past, present, and future. *Arch Pharm Res.*2015;38:575-584.
165. Zhang B, Gojo I, Fenton RG. Myeloid cell factor-1 is a critical survival factor for multiple myeloma. *Blood.*2002;99:1885-1893.
166. Derenne S, et al. Antisense strategy shows that Mcl-1 rather than Bcl-2 or Bcl-x(L) is an essential survival protein of human myeloma cells. *Blood.*2002;100:194-199.
167. Levenson JD, et al. Potent and selective small-molecule MCL-1 inhibitors demonstrate on-target cancer cell killing activity as single agents and in combination with ABT-263 (navitoclax). *Cell Death Dis.*2015;6:e1590.
168. Yang Y, Li F, Saha MN, Abdi J, Qiu L, Chang H. miR-137 and miR-197 Induce Apoptosis and Suppress Tumorigenicity by Targeting MCL-1 in Multiple Myeloma. *Clin Cancer Res.*2015;21:2399-2411.

169. Vick B, et al. Knockout of myeloid cell leukemia-1 induces liver damage and increases apoptosis susceptibility of murine hepatocytes. *Hepatology*.2009;49:627-636.
170. Arbour N, et al. Mcl-1 is a key regulator of apoptosis during CNS development and after DNA damage. *J Neurosci*.2008;28:6068-6078.
171. Thomas RL, et al. Loss of MCL-1 leads to impaired autophagy and rapid development of heart failure. *Genes & Immunity Dev*.2013;27:1365-1377.
172. Mason KD, et al. Programmed anuclear cell death delimits platelet life span. *Cell*.2007;128:1173-1186.
173. Seymour JF, et al., eds. Bcl-2 Inhibitor ABT-199 (GDC-0199) Monotherapy Shows Anti-Tumor Activity Including Complete Remissions In High-Risk Relapsed/Refractory (R/R) Chronic Lymphocytic Leukemia (CLL) and Small Lymphocytic Lymphoma (SLL) 2013; Ernest N. Morial Convention Center, Atlanta. American Society of Haematology.
174. Souers AJ, et al. ABT-199, a potent and selective BCL-2 inhibitor, achieves antitumor activity while sparing platelets. *Nat Med*.2013;19:202-208.
175. Touzeau C, et al. The Bcl-2 specific BH3 mimetic ABT-199: a promising targeted therapy for t(11;14) multiple myeloma. *Leukemia*.2014;28:210-212.
176. Feugier P. A review of rituximab, the first anti-CD20 monoclonal antibody used in the treatment of B non-Hodgkin's lymphomas. *Future Oncol*.2015;11:1327-1342.
177. Weiner GJ. Building better monoclonal antibody-based therapeutics. *Nat Rev Cancer*.2015;15:361-370.
178. Hsi ED, et al. CS1, a potential new therapeutic antibody target for the treatment of multiple myeloma. *Clin Cancer Res*.2008;14:2775-2784.
179. Bouchon A, Cella M, Grierson HL, Cohen JI, Colonna M. Activation of NK cell-mediated cytotoxicity by a SAP-independent receptor of the CD2 family. *J Immunol*.2001;167:5517-5521.
180. Cannons JL, Tangye SG, Schwartzberg PL. SLAM family receptors and SAP adaptors in immunity. *Annu Rev Immunol*.2011;29:665-705.
181. Guo H, Cruz-Munoz ME, Wu N, Robbins M, Veillette A. Immune cell inhibition by SLAMF7 is mediated by a mechanism requiring src kinases, CD45,

and SHIP-1 that is defective in multiple myeloma cells. *Mol Cell Biol.*2015;35:41-51.

182. van Rhee F, et al. Combinatorial efficacy of anti-CS1 monoclonal antibody elotuzumab (HuLuc63) and bortezomib against multiple myeloma. *Mol Cancer Ther.*2009;8:2616-2624.

183. Collins SM, et al. Elotuzumab directly enhances NK cell cytotoxicity against myeloma via CS1 ligation: evidence for augmented NK cell function complementing ADCC. *Cancer Immunol Immunother.*2013;62:1841-1849.

184. Lonial S, et al. Elotuzumab Therapy for Relapsed or Refractory Multiple Myeloma. *N Engl J Med* 2015.

185. Plesner T, et al. Safety and Efficacy of Daratumumab with Lenalidomide and Dexamethasone in Relapsed or Relapsed, Refractory Multiple Myeloma. *Blood.*2014;124.

186. Lokhorst HM, et al. Targeting CD38 with Daratumumab Monotherapy in Multiple Myeloma. *N Engl J Med.*2015.

187. Moreau P, et al. An Open-Label, Multicenter, Phase 1b Study of Daratumumab in Combination with Backbone Regimens in Patients with Multiple Myeloma. *Blood.*2014;124.

188. Kelly KR, et al. Indatuximab Ravtansine (BT062) In Combination With Lenalidomide and Low-Dose Dexamethasone In Patients With Relapsed and/Or Refractory Multiple Myeloma: Clinical Activity In Len/Dex-Refractory Patients. *Blood.*2013;122.

189. Shi W, et al. Transcriptional profiling of mouse B cell terminal differentiation defines a signature for antibody-secreting plasma cells. *Nat Immunol.*2015;16:663-673.

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