Biology of the Transition of Monoclonal Gammopathy of Undetermined Significance (MGUS) to Multiple Myeloma

John A. Lust, MD, PhD, and Kathleen A. Donovan, PhD

Changes in clonal plasma cells that occur in the clinical progression from monoclonal gammopathy to myeloma are reviewed.

Background: Approximately 25% of patients with monoclonal gammopathy of undetermined significance (MGUS) eventually develop multiple myeloma (MM) or a related plasma cell disorder that is universally fatal. In this report, we examine the changes that occur in the clonal plasma cell that are likely to be important in the progression of MGUS to active myeloma.

Methods: Studies that investigate the mechanisms involved in the multistep pathogenesis of monoclonal gammapathies are reviewed. Cytokines such as IL-6 and IL-1, adhesion molecules, viruses, and oncogenes including ras, bcl-2, Rb, and p53 are discussed.

Results: IL-1 is produced by plasma cells from virtually all MM patients but is undetectable in most MGUS patients. IL-1 has potent osteoclast activating factor activity, can increase the expression of adhesion molecules, and can induce paracrine IL-6 production. The increased production of adhesion molecules could explain why myeloma cells are found predominantly in the bone marrow. Subsequently, these "fixed" monoclonal plasma cells could now stimulate osteoclasts through the production of IL-1 and paracrine generation of IL-6 resulting in osteolytic disease. With continued progression of the myeloma, the monoclonal plasma cells may later acquire the ability to produce IL-6 in an autocrine fashion that will be manifested clinically by an elevated labeling index.

Conclusions: A better understanding of the progression of MGUS to myeloma may lead to novel therapeutic strategies to prevent the development of MM.

Introduction

A wealth of information is currently available in the literature that postulates a role for cytokines, oncogenes and, more recently, viruses in the pathogenesis of monoclonal gammapathies. In this article, much of these data will be reviewed in the context of the clinical progression of monoclonal gammapathy of undetermined significance (MGUS) to active myeloma. A better understanding of the biology of monoclonal gammapathies may provide a rational basis for future research and the development of novel biologic therapies to treat myeloma in the future.

Clinical Features of the Transition of MGUS to Multiple Myeloma

Multiple myeloma (MM) is recognized clinically by the proliferation of malignant plasma cells in the bone marrow, the detection of a serum or urine monoclonal protein, anemia, hypercalcemia, renal insufficiency, and lytic bone lesions. MGUS is characterized by a monoclonal protein (M protein) in the serum or urine without other clinical features of MM.1,2 MM patients are asymptomatic and have stable serum M-protein measurements.3 MM is more common than myeloma occurring in 1% of the population over age 50 and 3% over age 70.2 It is of great clinical importance to distinguish between patients with MM from individuals with MGUS because MGUS patients may be safely observed off chemotherapy. Unnecessary treatment can lead to acute leukemia2 or morbidity/mortality from chemotherapy.

As shown in Table 1, patients with MGUS usually have less than 10% marrow plasma cells, a serum monoclonal protein <3 g/dL, no urinary Bence-Jones protein, and no anemia, renal failure, lytic bone lesions, or hypercalcemia. In contrast, patients with active myeloma will present with a marrow plasmacytosis of >10%, a serum monoclonal protein of >3 g/dL, a 24-hour urine monoclonal protein of >1 g, and lytic bone lesions. Patients with smoldering MM (SMM) have a marrow plasmacytosis of >10% and/or a serum monoclonal protein of >3 g/dL, but lytic bone lesions are absent and they have stable disease. These are general guidelines; the total clinical and laboratory picture should be assessed by clinicians experienced in the field. Many patients with MM have a history of a prior MGUS. In one Mayo Clinic study, 58% had prior MGUS or plasmacytoma.4 During long-term follow-up of 241 patients with MGUS, 59 patients (24.5%) went on to develop MM or a related plasma cell proliferative disorder.3

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>MM</th>
<th>SMM</th>
<th>MGUS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Marrow plasma cells</td>
<td>&gt;10%</td>
<td>&gt;10%</td>
<td>&lt;10%</td>
</tr>
<tr>
<td>Serum M-spike</td>
<td>&gt;3 g/dL</td>
<td>&gt;3 g/dL</td>
<td>&lt;3 g/dL</td>
</tr>
<tr>
<td>Bence-Jones protein</td>
<td>&gt;1 g/24 h</td>
<td>&lt;1 g/24 h</td>
<td>&lt;1 g/24 h</td>
</tr>
<tr>
<td>Anemia</td>
<td>usually present</td>
<td>may be present</td>
<td>absent</td>
</tr>
<tr>
<td>Hypercalcemia, renal insufficiency</td>
<td>usually present</td>
<td>absent</td>
<td>absent</td>
</tr>
<tr>
<td>Lytic bone lesions</td>
<td>usually present</td>
<td>absent</td>
<td>absent</td>
</tr>
</tbody>
</table>

MM = multiple myeloma
SMM = smoldering multiple myeloma
Closer examination of those individuals who developed MM or a related plasmacytoid disorder revealed that the majority of patients remained stable for an extended period of time and then subsequently progressed (developed myeloma) over a relatively short period of time. Of the 59 patients who progressed, 39 went on to develop MM. Of these 39 patients, 18 had undergone serial serum studies (Table 2). In both groups, the M-protein remained stable for a median of eight years and then increased slowly over one to four years or rapidly in less than one year with the development of myeloma. Based on these clinical observations, it is likely that differences exist between MGUS and myeloma in which additional changes arise in the monoclonal plasma cells leading to overt myeloma. These changes could lead to aberrant expression of cytokines, adhesion molecules, or other cellular factors that may be responsible for the transition from MGUS to SMM to active MM.

### Table 2.-- Clinical Progression of MGUS to MM (39 Patients)

<table>
<thead>
<tr>
<th>Mprotein Stable Over</th>
<th>Mprotein Increased Over</th>
<th>Number of Patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>4-18 years (median 8)</td>
<td>4-18 years (median 8)</td>
<td>1</td>
</tr>
<tr>
<td>4-18 years (median 8)</td>
<td>1-4 years (median 3)</td>
<td>11</td>
</tr>
<tr>
<td>4-18 years (median 8)</td>
<td>&lt;1 year</td>
<td>7</td>
</tr>
</tbody>
</table>

### Role of Cytokines in the Progression of MM: Importance of IL-6, sIL-6R, and IL-1

In normal B-cell ontogeny, interleukin-6 (IL-6) is an important growth factor in the terminal differentiation of B cells into immunoglobulin-secreting plasma cells. Normal B cells produce antibody in response to IL-6 but do not proliferate. In contrast, IL-6 has been shown to be a central growth factor for myeloma cells. The fact that myeloma cells proliferate in response to IL-6 is a major difference that distinguishes malignant from normal plasma cells and is of critical importance in the pathogenesis of the disease.

Early work by Potter et al demonstrated that paraffin oil or pristane induced plasmacytomas when injected into BALB/c mice. The generation of the plasmacytomas was dependent on factors produced by the inflammatory cells. These cells were subsequently shown to produce IL-6, a potent growth factor for plasmacytomas.

More recently, transgenic mice (C57BL/6) carrying the human IL-6 gene fused to a human immunoglobulin heavy chain enhancer developed a massive lethal plasmacytosis. 

Kishimoto and colleagues have demonstrated that IL-6 is an autocrine growth factor for human myeloma cells. They have shown that myeloma cells freshly isolated from patients produce IL-6 and express its receptor. Exogenous IL-6 augments the in vitro growth of myeloma cells, and anti-IL-6 antibody inhibits their growth. Schwab et al have demonstrated that a myeloma cell line U266 expresses mRNA for both IL-6 and sIL-6R. The proliferation of this cell line can be inhibited using anti-IL-6 antibody or antisense IL-6 oligonucleotides further supporting the critical role of IL-6 in the growth of these cells. Significantly elevated serum IL-6 levels have been detected in 3% of MGUS/SMM patients, in 35% of overt myeloma patients, and in 100% of a plasma cell leukemia group. Using an antibromodeoxyuridine monoclonal antibody to specifically count myeloma cells in S-phase (i.e., the labeling index), the IL-6 responsiveness of myeloma cells in vitro correlates with their labeling index in vivo and hence to the severity of the disease. Most importantly, an antibody to IL-6 administered in vivo has been shown to dramatically decrease the labeling index of the tumor cells in five patients with aggressive MM.

Since IL-6 is a central growth factor for myeloma cells, sIL-6R may modulate IL-6 activity. Soluble receptors have been shown to be potent immunomodulators of their respective ligands. We have previously reported a novel IL-6R mRNA from myeloma cells that exhibits a 94-nt deletion of the entire transmembrane domain from codons 356 (G-TG) to 387 (AG-G). The transmembrane domain deletion results in a shift in the translational reading frame with the insertion of 10 new amino acids followed by a stop codon. Sequence analysis shows the ligand-binding domain of the sIL-6R to be identical to that of the membrane-bound IL-6R up to the transmembrane domain deletion. The sIL-6R cDNA was expressed and supernates were collected from mock or sIL-6R transfected PA-1 cells after 48 hours and assayed for their ability to stimulate or suppress the growth of an IL-6-dependent cell line, ANBL-6. Soluble IL-6R alone had no effect on the growth of the ANBL-6 cells. However, the growth of ANBL-6 cells by sIL-6R was potentiated in the presence of IL-6 and could be blocked by anti-IL-6 antibody. The above results suggest that, in the presence of IL-6, sIL-6R associates with gp130 leading to signal transduction and cell growth.

Among 30 healthy individuals, 32 patients with MGUS, and 74 with myeloma, sIL-6R levels were increased similarly in MGUS and MM -- 51% and 44%, respectively. An elevated sIL-6R level correlated with a poor survival and was independent of the plasma-cell labeling index and \( M \). Soluble IL-6R plays an important role in the pathogenesis of MM by potentiating IL-6 activity.

Although it is clear that IL-6 expression plays a fundamental role in the growth of MM cells, the source of IL-6 expression is controversial. Klein et al reported that the high levels of IL-6 found in the bone marrow of patients with progressive MM is confined to the adherent cells of the bone marrow environment and that IL-6 is not expressed by myeloma cells. In addition, bone marrow monocytic and myeloid cells, but not myeloma cells, have been reported to express IL-6 mRNA. Together, these results suggest a paracrine rather than an autocrine mechanism of myeloma cell growth by IL-6. As discussed above, Kishimoto and colleagues have demonstrated that IL-6 is an autocrine growth factor for human myeloma cells. Hata et al detected IL-6 mRNA by RT/PCR in purified plasma cells from myeloma patients as well, and they also demonstrated that these CD38+ myeloma cells expressed intermediate levels of CD45. Our data demonstrate that monoclonal plasma cells from the majority of myeloma patients with active disease manifested by a high labeling index can express IL-6 mRNA in an autocrine fashion. Furthermore, we have reported three new myeloma cell lines isolated from patients with very high labeling indices that produce IL-6 in an autocrine fashion, two of which are CD38+/CD45+.

The apparent discrepancy between our data and those of Klein et al is likely due to the differences in sensitivities of the techniques used to detect IL-6 expression; we utilized polymerase chain reaction, which is more sensitive than Northern analysis. However, it is likely that both autocrine and paracrine sources of IL-6 production play a role in the pathogenesis of myeloma.

Production of IL-1 by myeloma cells may be responsible for the paracrine generation of IL-6 by marrow stromal cells. It has been shown that IL-1 can induce expression of the genes for IL-6, colony-stimulating factors, and adhesion proteins. In vitro fibroblasts, macrophages, T lymphocytes, and other marrow stromal cells are all capable of responding to IL-1 by expressing one or more IL-1-inducible genes. Carter et al have found that human myeloma cells are able to induce IL-6 production in marrow stromal cells. The stimulatory activity of the myeloma cells appears to be mediated through endogenously released IL-1 and antibodies to IL-1 completely abrogate the IL-6 production.

Normal plasma cells do not produce IL-1; however, abnormal IL-1 production by myeloma cells has been detected at both the mRNA and protein levels by several different investigators. Using fresh myeloma cells, Lichtenstein and colleagues detected IL-1 at the protein level, and Klein et al found strong IL-1 gene expression by in situ hybridization. Cozzolino et al have shown that culture supernatants of plasma cells, isolated by a rosetting procedure, from 12 of 12 patients with MGUS/SMM expressed IL-1.

MGUS = monoclonal gammopathy of undetermined significance
with MM contained high amounts of IL-1β. In contrast, plasma cells from 9 of 9 patients with MGUS showed undetectable levels of IL-1β. Using flow cytometric sorting to enrich for plasma cells and RTPCR for cytokine expression, we have found that IL-1 mRNA is expressed by plasma cells from virtually all MM patients but is not detectable in the plasma cells of most MGUS patients. Future studies will determine whether the detection of IL-1 expression will differentiate between patients with MGUS or MM.

Could aberrant IL-1 production be contributing to the progression from MGUS to myeloma? The development of osteolytic lesions is an important clinical finding that clearly distinguishes MGUS from myeloma. Since IL-1 has potent osteoclast-activating factor (OAF) activity, it may be responsible for the presence of bone lesions. Initially, two different groups had shown that the bone resorbing activity in supernatants of myeloma cell cultures was likely due to IL-1 and not to IL-1 tumor necrosis factor, or lymphotoxin. More recently, Torcia and colleagues have shown a critical role for IL-1 in the pathogenesis of bone disease. Using the fetal rat long-bone tissue culture assay, they demonstrated that the OAF activity of culture supernatants from unfractured bone marrow cells from myeloma patients correlated with the IL-1 content (r = 0.949). Furthermore, the OAF activity could be completely abolished by IL-1ra, sIL-1R type I or II, or neutralizing anti-IL-1 antibodies but not anti-IL-6 antibodies. These results demonstrate that the OAF activity of myeloma cells from patients is predominantly, if not solely, related to IL-1

Hawley and colleagues developed a mouse model of myeloma that demonstrates the importance of IL-1 expression in inducing pathology that mimics human disease. They introduced an IL-1 cDNA into an IL-6 dependent murine B-cell line by retroviral-mediated gene transfer. After injection of these IL-1 producing B-cells into syngeneic mice, these cells were shown to “home” to the bone marrow and produce metastatic bone lesions. By comparison, intravenous injection of autonomously growing B-cell lines generated in vitro by retroviral insertion of an IL-6 cDNA rarely resulted in bone marrow or bone metastases. Subsequent work has shown that aberrant expression of IL-1 can alter adhesion molecules such as ICAM and CD44 on the surface of mouse plasmacytoma cells. A similar mechanism may occur in human myeloma in which aberrant expression of IL-1 induces increased expression of adhesion molecules such as VLA-4, CD44, CD54, CD56, and other surface molecules on the monoclonal plasma cells.

**Adhesion Molecules in the Biology of Myeloma**

A central issue in the differentiation process of normal B cells concerns their homing ability to various tissues regulated by surface adhesion molecules. A striking clinical feature of myeloma cells relates to their tendency to remain in the bone marrow environment until the very end-stage of the disease. Both the differentiation stage of the B cell as well as aberrant cytokine production potentially contribute to inducing osteolytic disease through increased production of various adhesion molecules. Several investigators have identified the presence of various adhesion molecules on myeloma cells such as NCAM (CD56), ICAM (CD54), HCAM (CD44), VLA-4 (CD49d), LFA-3 (CD58), and others.

Recent studies on ICAM suggest that the IL-1-induced up-regulation of adhesion molecule may be crucial in the pathogenesis of myeloma. Tumor cells from virtually all myeloma patients strongly express ICAM-1 (CD54). Although ICAM is detectable on normal plasma cells, CD54 can be further up-regulated by cytokines such as IL-1 through cytokine-inducible enhancers identified in the ICAM promoter region. As discussed above, transduction of an IL-1 cDNA into a murine B-cell line resulted in increased expression of ICAM and “homing” of the IL-1 transduced plasmacytoma cells to the bone marrow. Finally, SCID mice that have been injected intravenously with ARH cells are protected from developing hind limb paralysis and lytic bone lesions by pretreatment with anti-CD54 antibodies. These observations underscore the importance of adhesion molecules in the “homing” of myeloma cells to the bone marrow and in the development of lytic lesions and spinal cord paralysis, both frequently seen in patients with myeloma.

It has been hypothesized that the acquisition of NCAM expression in myeloma is a malignancy-related phenomenon. CD56 (NCAM), a cell-adhesion molecule, is strongly expressed on myeloma plasma cells but is not found on normal plasma cells. CD56 expression in high density was present in 43 of 57 patients with untreated MM but in none of 23 patients with MGUS. In another study, normal plasma cells from various tissues were all CD19+CD56-, whereas mature myeloma cells from 12 of 20 cases were CD19-CD56+. Both CD19+CD56- and CD19-CD56+ plasma cells were found in all five cases of MGUS tested, suggesting that MGUS consists of phenotypically normal plasma cells and myeloma cells. These results have recently been confirmed by another investigator.

In contrast, NCAM expression was rarely found in acute myelogenous leukemia and was not detected on cells from patients with acute lymphoblastic leukemia, chronic lymphocytic leukemia, non-Hodgkin’s lymphoma, or hairy cell leukemia. In comparison to myeloma, bone involvement is not a major feature of these disorders. It is likely that adhesion markers play a major role in cell-to-cell contact between myeloma cells and marrow stromal cells or other myeloma cells. These interactions may be important in the homing of myeloma cells to the bone marrow and in the pathogenesis of osteolytic disease.

**IL-1 Expression and Kaposi’s Sarcoma-Associated Herpesvirus**

The etiology of acquired IL-1 expression in myeloma is unknown. However, the IL-1 gene is highly inducible, and its expression can be affected by many microbial and cellular products. A role for Kaposi’s sarcoma-associated herpesvirus (KSHV) in the pathogenesis of myeloma has been recently reported. Although it has not been demonstrated for KSHV, Epstein-Barr virus, human immunodeficiency virus-1, and respiratory syncytial virus have been shown to up-regulate IL-1 expression either directly by interacting with genomic sequences or indirectly by altering levels of transcription factors involved in IL-1 expression. If KSHV is involved in the pathogenesis of myeloma, it may play a role either directly or indirectly in the aberrant expression of the IL-1 gene.

**Circulating Myeloma Cells and Precursors**

It has been postulated that myeloma results from a genetic event that occurs in a B-cell compartment that precedes the typical cytoplasmic immunoglobulin positive plasma cell. A malignant hybrid without a normal counterpart in B-cell ontology and with coexpression of cytoplasmic μ common acute lymphoblastic leukemia antigen (CALLA), terminal deoxynucleotidyl transferase (TdT), and plasma cell antigens (PCA-1 and PC-1) were found in direct and cultured myeloma cells. These cells were found to be monoclonal by gene rearrangement studies and proliferative by labeling index experiments. Subsequent data have demonstrated that small numbers of normal plasma cells can express CALLA, thus suggesting that the association of early B-cell markers initially found on myeloma cells is a normal phenomenon and may not be associated with neoplasia. Monoclonal B cells present in both marrow and peripheral blood have been shown to be part of the clonal myeloma cell population by detection of B lymphocytes expressing the same myeloma protein idiotype/isotype on their surface or through gene rearrangement studies with immunoglobulin gene probes. B cells shown to be part of the malignant clone have been described with the morphology of lymphocytes, lymphoplasmablasts.
Circulating malignant plasma cell precursors cultured in the presence of IL-6 and IL-3 were induced to differentiate into a proliferating immunoblast-like B-cell subpopulation after three days and into a plasma-cell population after six days. These plasma cells expressed the same light- and heavy-chain produced by the bone marrow monoclonal plasma cells. The above observations would suggest that a maturation process from an early B lymphocyte to the mature plasma cell occurs in myeloma and may be similar in MGUS.

An important difference between MGUS and MM is the finding that circulating monoclonal plasma cells are increased in active myeloma. In a study of 84 patients, Witzig et al. showed that “clonal” plasma cells (plasma cells that stain monotypically for cytoplasmic light-chain of the same Ig as plasma cells in the marrow) circulate in increased numbers in patients with myeloma. Fifty-seven percent of newly diagnosed cases and 81% of relapsed MM cases had >3 x 10^6 monoclonal plasma cells per liter. All patients with MGUS had <3 x 10^6 plasma cells per liter. The number of circulating plasma cells was a better discriminator of disease activity than B-cell light-chain ratio.

Karyotypic Abnormalities

A large number of cytogenetic and molecular abnormalities that have been described in myeloma appear to contribute to the evolution and persistence of the clonal myeloma cell population. Although there is no single cytogenetic abnormality pathognomonic for myeloma, cytogenetic and flow cytometry studies have shown that aneuploid karyotypes are a common feature found in the majority of patients with myeloma. Jelinek and colleagues have characterized an IL-6-dependent myeloma cell line, ANBL-6, that exhibits a clonally rearranged immunoglobulin light locus but is composed of near-diploid and near-tetraploid subpopulations. Cytogenetic studies confirmed the existence of two aneuploid karyotypes and further revealed a clonal relationship between the two karyotypes as evidenced by numerous shared structural abnormalities. The coexistence of clonally related subclones with shared chromosomal abnormalities suggests that the near-tetraploid subclone was derived from the near-diploid subclone during clonal evolution.

Karyotypic studies in patients with MGUS have been hampered by a low percentage of bone marrow plasma cells that are predominantly nonproliferating. However, in one study that combined fluorescence in situ hybridization (FISH) with cytology, chromosomes 3, 7, 11, and 18 were investigated. These chromosomes had been previously found to be aneuploid by FISH in myeloma. Three hybridization signals for one of these chromosomes were observed in 19 (52.8%) of 36 patients. Gains of chromosome 3 were most common, occurring in 39% of patients, followed by chromosome 11 (25%), 7 (16.7%), and 18 (5.6%). Among bone marrow plasma cells, the frequency of aneuploid cells was 19% for chromosome 3, 22% for chromosome 11, 23% for chromosome 7, and 6% for chromosome 18. No gain of hybridization signals was observed in normal and reactive plasma cells. Thus, the MGUS state already has the chromosomal characteristics of a plasma cell malignancy.

Immunoglobulin Translocations, Rb/p53, BCL-2, and Ras

Despite the low incidence of translocations to IgH loci by standard cytogenetics, most myeloma cell lines and primary myeloma tumors appear to exhibit IgH translocations by Southern analysis that involve mainly switch regions. These translocations involve two loci, 11q13 (bcl-1) and fibroblast growth factor receptor 3, in 50% of cases and, in the remaining 50%, a large number of other chromosome partners including 8q24 (c-myc), 18q21 (bcl-2), and many others. In another study, using double-color FISH, IgH translocations were detected in 31 of 42 patients with MM and in 3 of 5 patients with MGUS, suggesting that these translocations occur at a high frequency in patients. IgH translocations appear to be an early event in the pathogenesis of myeloma and may be the cause of MGUS.

Several oncogenes have been implicated in the pathogenesis of monoclonal gammopathies that probably serve to perpetuate the monoclonal plasma cells and may increase the plasmablastic compartment. The majority of patients with myeloma appear to have an increased production of bcl-2 protein. Since t(14;18) has been observed in a minority of these patients, the mechanism of increased bcl-2 expression is unknown. However, bcl-2 and several related proteins can inhibit (Bcl-XL) or enhance (Bax, Bad) apoptosis. Over-expression of bcl-2 or Bcl-XL can prevent apoptosis induced by IL-6 withdrawal in the IL-6-dependent cell line, B9. Therefore, increased bcl-2 production may be an important factor in the persistence of the myeloma clone.

Neri and colleagues found point mutations in both the K-ras and N-ras oncogenes in approximately 30% of myeloma patients. In another report, 17 (74%) of 23 patients with active myeloma had higher H-ras p21 protein fluorescence in aneuploid tumor cells compared with marrows from patients in remission. Seremetis et al. studied the effects of ras oncogene activation in B cells using retroviral vectors to introduce ras oncogenes into human B lymphoblasts immortalized by Epstein-Barr virus. Expression of both H-ras and N-ras led to malignant transformation and terminal differentiation into plasma cells. The introduction of an N-ras cDNA containing a glutamine to arginine amino acid substitution at codon 61 into an IL-6-dependent cell line resulted in IL-6 independent growth and a decrease in apoptosis in the absence of IL-6. Thus, mutations of ras may result in growth factor independence and an increase in the pool of proliferating plasmablasts.

Genetic abnormalities of the tumor suppressor genes p53 and Rb have been detected in a number of human cancers including myeloma. Two studies reported a mutated p53 gene in eight (80%) of 10 human myeloma cell lines and in six (20%) of 30 myeloma patients. Of the six patients with a mutated p53 gene, four were in the terminal phase of disease. Using FISH, Barlogie and coworkers found deletions in the Rb-1 gene in 12 (52%) of 23 patients. Interestingly, cytogenetics revealed an abnormal chromosome 13 in only four (17%) of 23 patients. Both p53 and Rb function as transcriptional repressors of IL-6 gene expression. Mutations of p53 and Rb could result in increased IL-6 production by the myeloma cells themselves that would stimulate their own growth (autocrine mechanism). Such patients might be expected to have an elevated IGF and a poor prognosis.

Summary and Future Therapeutic Considerations

The mechanisms involved in the multistep pathogenesis of monoclonal gammopathies are highly complex and involve cytokines such as IL-6 and IL-1, adhesion molecules, viruses, oncogenes including ras, bcl-2, Rb, p53, and probably many as yet undefined factors that act on different stages of B-cell maturation. In this report, we reviewed those changes that occur in the clonal plasma cell in the context of the clinical progression of MGUS to active myeloma. Abnormalities such as trisomy 3 may be less clinically significant since they are commonly found in the MGUS state. In contrast, IL-1 produced by plasma cells from virtually all MM patients but is not detectable in the plasma cells of most MGUS patients. IL-1 has potent OAF activity, can increase the expression of adhesion molecules, and can induce paracrine IL-6 production. The increased production of adhesion molecules could explain why myeloma cells are found predominantly in the bone marrow. Subsequently, these “fixed” monoclonal plasma cells could now stimulate osteoclasts through the production of IL-1 and paracrine generation of IL-6 resulting in osteolytic disease (Figure). With continued progression of the myeloma, the monoclonal plasma cells may later acquire the ability to produce IL-6 in an autocrine
fashion that will be manifested clinically by an elevated labeling index. Abnormalities of ras, Rb, and p53 may be later events that are important factors in the persistence of the myeloma clone. Finally, various combinations of any of the above genetic alterations are likely possible and probably explain the clinical heterogeneity of monoclonal gammapathies and the variable response to chemotherapy.

**Role of IL-1β and IL-6 in the Transition of MGUS to MM**

The above studies may suggest new innovative future therapies for myeloma. For example, a mouse anti-IL-6 antibody has already been shown to suppress cell growth in myeloma patients with aggressive disease. In the future, IL-6 inhibitors such as humanized anti-IL-6 and anti-IL-6R antibodies or a human soluble IL-6R mutant may be useful therapeutically to suppress long-term myeloma cell growth. Along similar lines, if IL-1 has potent OAF activity, antagonists of IL-1 such as human soluble IL-1R and IL-1R antagonist may play a role in controlling bone lesions in patients with active disease. New therapies to kill myeloma cells should also become more evident. IL-6 fused to a mutant form of *Pseudomonas* exotoxin was found to kill malignant plasma cells from eight of 15 myeloma patients *in vitro.* A radioimmunoconjugate approach directed against target antigens present on myeloma cells may also be a reasonable therapeutic consideration. A radioimmunoconjugate against CD20 has been shown to be successful in several patients with refractory lymphoma. Since virtually all patients with myeloma will eventually relapse on chemotherapy, new biologic therapies will likely be required in the future.

**References**


From the Division of Hematology and Internal Medicine, Mayo Clinic, Rochester, MN, 55905

Address reprint requests to John A. Lust, MD, PhD, at the Division of Hematology, Mayo Clinic, Rochester, MN 55905.

Supported by grant CA62242 from the National Institutes of Health.