A Basic Guide to Understanding Your Bone Marrow and Waldenstrom's Macroglobulinemia

The IWMF Vision Statement

A world without WM (Waldenstrom's macroglobulinemia).

The IWMF Mission Statement

Support and educate everyone affected by Waldenstrom's macroglobulinemia (WM) while advancing the search for a cure.

Published by the International Waldenstrom's Macroglobulinemia Foundation (IWMF)

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FOREWORD

This 2022 first edition of *A Basic Guide to Understanding Your Bone Marrow and Waldenstrom's Macroglobulinemia* is published by the International Waldenstrom's Macroglobulinemia Foundation (IWMF), a nonprofit organization founded in 1994 by Arnold Smokler. The IWMF was established to offer mutual support and encouragement to the Waldenstrom's macroglobulinemia community and others with an interest in the disease; to provide information and educational programs that address patients' concerns; and to promote and support research leading to better treatments and ultimately, a cure.

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Introduction

Waldenstrom's macroglobulinemia

Waldenstrom's macroglobulinemia (WM) is a rare, slow-growing cancer that typically involves the lymph nodes, bone marrow, and spleen. Cancers of this type are called lymphomas. WM is classified as a type of lymphoma known as non-Hodgkin lymphoma. Non-Hodgkin lymphoma is not a single disease; it refers to a group of different lymphomas that start in a type of white blood cell called a lymphocyte.

Lymphoid tissues

Lymphoid tissues are involved in helping your body fight disease and infection. Lymphoid tissue can be found in many places in the body, including:

- Lymph nodes – small, bean-shaped glands; some are found in clusters in places like your underarm, the sides of your neck, your groin, abdomen, and chest.
- Certain organs – such as your spleen, tonsils, adenoids, and thymus.
- **Bone marrow – the soft, spongy tissue in the center of most bones. This is where new blood cells are made.** There are three main types of blood cells:
    - Red blood cells (erythrocytes) carry oxygen throughout the body.
    - Platelets (thrombocytes) initiate the formation of blood clots to stop bleeding and help with wound healing.
    - White blood cells (leukocytes) help the body fight infection and some diseases.

WM is a lymphoma that affects a specific kind of white blood cell, called a B lymphocyte or B cell for short. The bone marrow produces the majority of your white blood cells, so it is helpful to know how your bone marrow works in order to understand WM.

What is bone marrow?

Basic structure and function

Bone marrow is the spongy, jelly-like material that fills the center of bones, known as the medullary cavity. It's protected by a hard, outer layer called compact bone or

cortical bone, which is wrapped in a dense fibrous membrane called the periosteum (*Figure 1*).

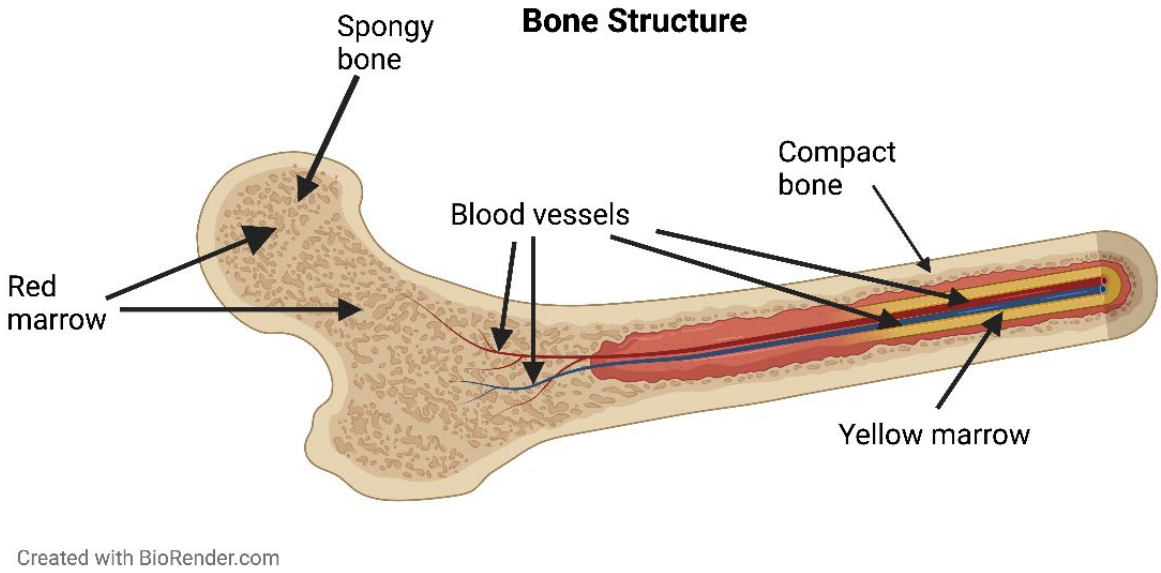

Figure 1: Bone Structure (created with BioRender.com)

Within the bone marrow are stem cells. These are immature cells that have the capacity to develop into a variety of different cell types. Bone marrow has two types of stem cells:

- **Mesenchymal stem cells**: Produce the body's connective tissue like cartilage, bone, and fat.
- **Hematopoietic stem cells**: Are responsible for the formation and development of all blood cells, a process known as hematopoiesis.

  Each type of blood cell that the bone marrow produces has a set lifespan. White blood cells last hours to days, platelets about ten days, and red blood cells about 120 days. Your bone marrow must constantly replace these cells throughout your lifetime. In addition, the bone marrow responds to the changing demands of your body. For example, white cell production increases in response to infection, red cells increase when your body needs more oxygen, and platelets increase when bleeding occurs.

Types of bone marrow

Your body has two types of bone marrow: red marrow and yellow marrow. At birth, all bone marrow is red and remains so until about the age of seven, at which point it gradually converts to the fattier yellow marrow. In a middle-aged adult, about half of the bone marrow is red and half is yellow.

Each type of bone marrow performs specific functions.

- **Red bone marrow**, called myeloid tissue, is made of highly vascular fibrous tissue that contains the hematopoietic or blood-forming stem cells. Red blood cells, platelets, and the majority of white blood cells are formed within the red bone marrow. In adults, the highest concentration of red marrow is found within the bones of the back (vertebrae), breastbone (sternum), pelvis (hip bone, where most bone marrow biopsies are done), ribs, skull, and at the ends of the upper arm bone (humerus), thigh bone (femur), and shin bone (tibia).
- **Yellow bone marrow** is fattier and contains the mesenchymal stem cells that produce the body's connective tissue. The yellow marrow also stores fats and nutrients for the red marrow to use and helps maintain the proper environment for the bone marrow to function. Yellow bone marrow can be found in the hollow cavity of long bones, like the arms and legs, and is typically surrounded by a layer of red bone marrow.

Bone marrow microenvironment

If a scientist looked closely at the red bone marrow with a microscope, they would see what is called the microenvironment. The microenvironment is an arrangement of cells and structures that allow for communication, or signaling, between the cells of the bone marrow. The microenvironment plays an important role in the growth of both cancerous (malignant) and healthy cells. Much research is currently being done to better understand the role of the microenvironment on the development and growth of malignancy, as well as the effect of the microenvironment on response to treatments.

Blood cell development

Having a basic understanding of the normal process of blood cell development, illustrated below, will help you appreciate what goes wrong when you have WM. Hematopoietic (blood-forming) stem cells can be divided into two main types of stem cells, myeloid and lymphoid (*Figure 2*).

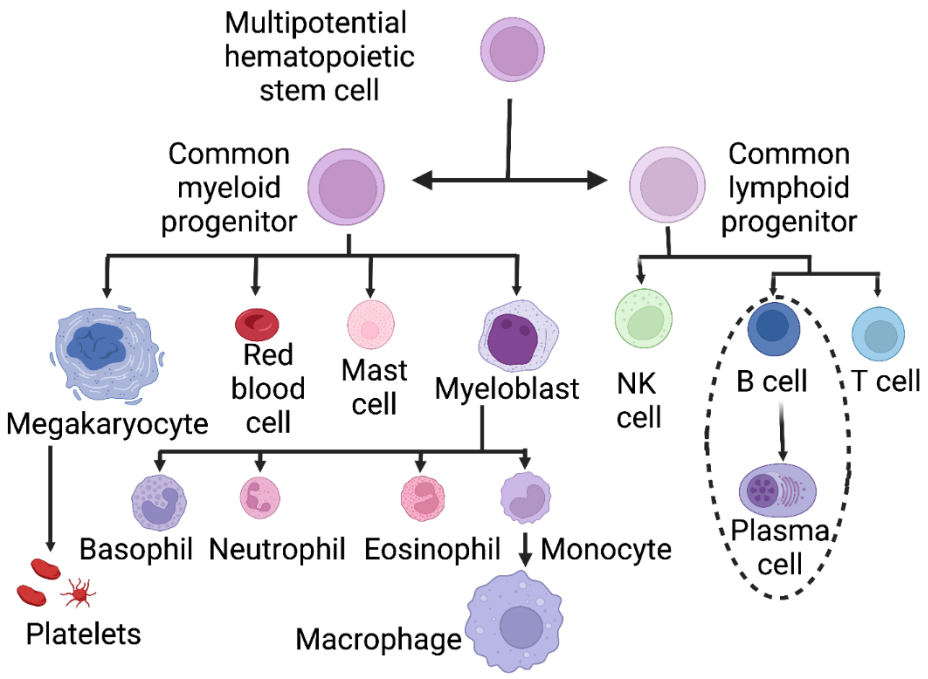

Figure 2: Blood cell development (created with Biorender.com). Malignant WM cells form from cells in the pathway between B cells and plasma cells (dotted oval).

- **Myeloid stem cells** develop into a number of different blood cells, including:
  - Red blood cells (erythrocytes), which contain a protein called hemoglobin that picks up oxygen in the lungs and transports it in the red blood cells to tissues that need it, like your brain, heart, and muscles. The lab value of hemoglobin is often used as an indicator of the amount of effective red blood cells circulating in the body.
  - Platelets (thrombocytes), which are critical to the formation of clots to stop bleeding. Tissue injury (such as a cut) triggers platelet activity at the site of a wound.
  - Granulocytes play an important role in protecting your body against infection, especially bacterial infections. – Granulocytes include three types of white blood cells: basophils, eosinophils, and neutrophils.
  - Monocytes, a cell type important in defense against a variety of infections.
- **Lymphoid stem cells** develop into a number of different white blood cells that make up a central part of the immune system, including:
  - Natural killer cells
  - T lymphocytes (T cells)
  - B lymphocytes (B cells)
  - Plasma cells

Normally, B cells help your body fight infection by maturing into plasma cells. The job of plasma cells is to make a protein called antibodies – also known as immunoglobulins or Ig for short. Antibodies help your body protect itself against disease and infection. There are five main types of antibodies, abbreviated IgA, IgD, IgE, IgG, and IgM. Although immunoglobulin M (IgM) is not the most abundant antibody, it is the largest of all the antibodies, called a macroglobulin *(Figure 3).*

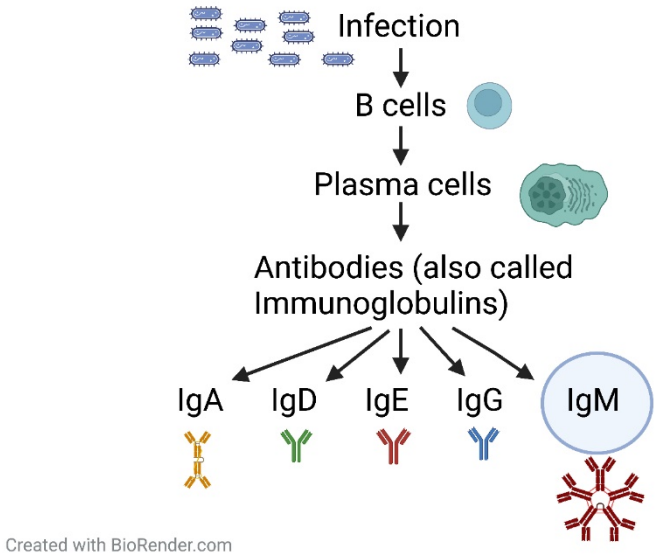

Figure 3: Antibody Development (created with BioRender.com).
Plasma cells can make different classes of antibodies (called Ig A, IgD, IgE, IgG, and IgM. In WM, the cancerous cells produce too much IgM.

How does WM affect bone marrow?

In the bone marrow, the development of B lymphocytes into plasma cells is a normal process, called a pathway. When you have WM, a mutation generally occurs somewhere within this plasma cell development pathway and an abnormal WM cell develops. When a WM cell forms, it duplicates making many copies of itself, called a clone. The abnormal clone in WM may contain any of the cells in this pathway, including B lymphocytes, lymphoplasmacytic cells (cells that have characteristics of B lymphocytes and plasma cells), and plasma cells. In addition, these clonal WM cells make abnormally large amounts of the antibody IgM, resulting in higher levels of IgM in your blood.

There are normally many different kinds of IgM antibodies, each made by a small number of plasma cells and each present in very small amounts. Because WM cells arise from a single clone, all the IgM they produce is the same. Large amounts of a single, identical antibody are referred to as a "monoclonal immunoglobulin spike" or "monoclonal spike" or "M spike," for short.

Because of IgM's large size and bulky structure, the blood can become thick at very high IgM levels, a condition called hyperviscosity. Thick, or viscous, blood cannot flow easily through the body. This can lead to many of the symptoms associated with WM, including excess bleeding, vision problems, cardiovascular complications, and nervous system issues.

In addition, because the abnormal cancer cells multiply over and over again and do not undergo a normal planned cell death, they can take over the bone marrow interfering with normal blood cell production (hematopoiesis). This can lead to a deficiency of healthy blood cells in your body. As a result, you may not produce enough red blood cells to carry adequate oxygen to your body's organs, leading to the most common early symptoms of WM – weakness and fatigue. You may also have low counts of other blood cells, such as platelets – leading to bleeding problems, or neutrophils – leading to reduced ability to fight infections.

Bone marrow biopsy procedure

Since the source of healthy blood cells, including the immune cells that make IgM, originate in the bone marrow, this is where investigations of hematologic (blood-related) issues typically begin. If blood tests indicate the presence of low blood counts (cytopenias), or high monoclonal IgM, your doctor may recommend a bone marrow examination. **A diagnosis of WM can only be confirmed by testing your bone marrow,** as the symptoms of WM can be similar to those caused by other diseases or infections**.**

When a thorough investigation of the bone marrow is required, two samples are typically obtained: a bone marrow aspirate and a bone marrow biopsy. The bone marrow aspirate is a liquid sample of the bone marrow and the bone marrow biopsy is a solid sample of the spongy center of the bone marrow (*Figure 4*).

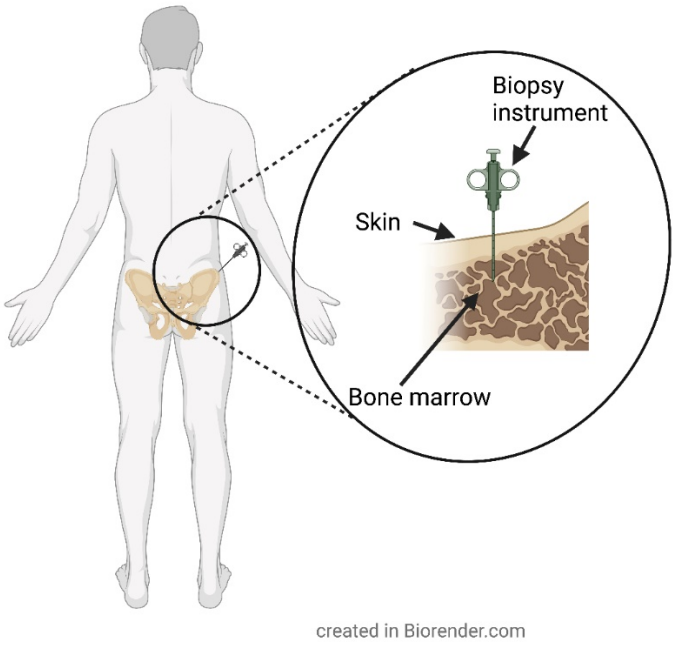

Figure 4: Bone Marrow Aspiration and Biopsy (created with BioRender.com). The bone marrow aspiration and biopsy are often drawn from the part of the pelvis called the iliac crest. The bone marrow biopsy instrument shown here is not drawn to scale.

Bone marrow aspiration and biopsy can be done at your doctor's office or in the hospital. These procedures are performed at the same time, usually on the back of your hip bone. The area will be numbed with a local anesthetic. In some cases, mild sedation may be used. Once the skin and bone have been anesthetized (numbed), a needle will be inserted to remove liquid bone marrow with a syringe (aspiration). Then another needle will be inserted in the same anesthetized location to remove a small piece of bone marrow (biopsy).

After the procedure, a pressure bandage is usually applied to the site to prevent any bleeding. You may experience bruising, tenderness, or mild pain, at the biopsy site. People are generally able to return to their usual activities the same day the biopsy is done.

Bone marrow examination and tests

The bone marrow biopsy and aspirate samples are sent to a laboratory to be examined by a pathologist who will look for abnormalities.

- **Bone marrow biopsy:** The bone marrow biopsy sample is used to assess the structure of the bone marrow, the number of cells present in the bone marrow

(cellularity), the proportion of different cell types, and how the cells are maturing (differentiation). The sample is examined under a microscope.

In WM, an excess of B lymphocytes, lymphoplasmacytic cells, or plasma cells will often be present in the bone marrow. It's common for people with this disease to have a hypercellular bone marrow (bone marrow with more cells than would be expected for the person's age) due to the malignant WM cells filling the bone marrow. The amount of these cells is generally reported as a percent of the bone marrow infiltration; this is the quantity of malignant cells that is referred to when doctors discuss the results with you.

However, bone marrow infiltration with malignant cells is variable because the abnormal cells are not evenly distributed throughout the marrow. They tend to clump in the bone marrow and a specific biopsy sample may miss them. As such, if you have more than one bone marrow biopsy done, it's possible to get different results because the biopsy samples will likely not be taken from the same exact spot.

- **Bone marrow aspirate**: The bone marrow aspirate sample is used to evaluate the amount of each cell type and the shape of the cells (morphology). It's also used for detailed genetic testing. There are a number of tests that may be performed:
    - **Cytology:** The aspirate sample is smeared into a thin layer of cells on a microscope slide and examined to give a detailed assessment of the bone marrow cells. This also allows the pathologist to determine if there are other bone marrow diseases going on.
    - **Flow cytometry**: This test is used to identify specific types of cells – like plasma cells, lymphoplasmacytic cells, and B cells – based on protein markers found on the outside of the cells. With this test, the pathologist can identify if a clone of abnormal cells is present.
    - **Karyotyping:** A process performed to evaluate the size, shape, and number of chromosomes. Chromosomes are structures within the cells of the bone marrow that carry DNA (the genetic material of the human body). A karyotype can give a general idea about any additions or deletions of chromosomes, as well as any translocations (swaps of material between different chromosomes).
    - **Fluorescence in situ hybridization (FISH**): In-depth testing of the chromosomes may also include FISH testing, which looks for specific chromosomal changes that may be associated with other bone marrow disorders, such as multiple myeloma or follicular lymphoma. Although this information is not routinely used to determine treatment or confirm the

diagnosis of WM, there are some mutations found on these tests that are often seen in WM – such as a TP53 mutation and deletion 6q.

- **Genetic mutation testing:** Additional tests are usually done to evaluate for specific genetic mutations that are often found in people with WM. Most people with WM have an abnormal (mutated) version of a gene called MYD88; about 40 percent of people with WM also have a mutation of the gene CXCR4; 5% percent will not have a mutation of either of them. Thorough testing for an MYD88 mutation and a CXCR4 mutation should be performed as part of an initial comprehensive work-up for WM. Knowing if there is a MYD88 mutation or a CXCR4 mutation is important when deciding about treatment options.

More detailed information about these and other tests can be found in the IWMF booklet, *Medical Tests*, on our website at https://iwmf.com/publications/.

Depending on the results of your bone marrow biopsy and aspiration, blood tests, imaging tests, physical exam, and whether or not you're experiencing symptoms, a diagnosis of WM is further refined into one of two subtypes: symptomatic, also called active WM, or asymptomatic, meaning without symptoms.

Not all people newly diagnosed with WM need immediate treatment. If you don't have symptoms, you usually do not need to be treated. One in four people (25 percent) are asymptomatic when diagnosed. And because WM is a slow-growing lymphoma, you may not have symptoms – or need treatment – for many years. Speak with your hematologist/oncologist about what your test results indicate and how they'll be used to help develop a personalized management plan.

For a more in-depth understanding of the bone marrow and Waldenstrom's macroglobulinemia, go to https://iwmf.com/publications/ and scroll down to **The Bone Marrow and Waldenstrom's Macroglobulinemia (Beyond Basics) Fact Sheet.**