Genetic Markers in Lymphomas

A review of the current role of genetic markers in six lymphoma subtypes.

Natalie Dren, Lymphoma Coalition
Lorna Warwick, Lymphoma Coalition
Jilda Lazer, Reverb Consulting

October 2019

LYMPHOMA COALITION

Worldwide Network of
Lymphoma Patient Groups

Acknowledgement

The LC wishes to sincerely thank those LC member organisations, researchers, HCPs, and other individuals who lent their time and efforts to this project. A special thank you to Professor John Seymour for his review. Each participant offered unique insight and support, and generously shared their knowledge, resources, and understanding. This thank you extends also to those who, on an ongoing basis, assisted in shaping and editing this report.

Disclaimer

The Lymphoma Coalition (LC) provides reports on information related to topics relevant to lymphoma worldwide. While LC makes every effort to ensure accuracy, the information contained in the report is taken from various public and private sources. No responsibility can be assumed by LC for the accuracy or timeliness of this information.

Warning

LC's reports should not be used for the purpose of self-diagnosis, self-treatment or as an alternative to medical care. If you have any concerns arising out of the information contained in this report, you should consult your own physician or medical advisor. If you suspect you have lymphoma, seek professional attention immediately.

Table of Contents

Overview

More is known about lymphoma today than ever before. Where it was once thought lymphoma comprised only a few diseases, there are now more than 80 known subtypes, with potential for more to be recognised in the future. This deeper understanding of the many types of lymphomas has largely come from better scientific knowledge of genes and gene mutations and the role they play in the diagnosis, prognosis and treatment of lymphomas.

Genes and their associated mutations are used to diagnose lymphoma and can also be **prognostic** about how the type of lymphoma will progress and/or **predictive** as to how the lymphoma will respond to available treatments. The presence or absence of certain mutations can help guide the type of treatment a patient receives and predict how likely the lymphoma is to respond to certain drugs.[1,2] This science of identifying disease through DNA, genes and mutations may be referred to as **molecular profiling**, genetic markers, **genetic profiling**, or driver gene-based classification.

Better understanding of gene mutations is also an important part of the development of targeted cancer treatments (also known as **precision medicine**). The Lymphoma Coalition (LC) published a report in 2013 regarding personalised medicine. The goal of personalised medicine is to select the most effective treatment for an individual patient's cancer. As more is learned about the biology of the different lymphomas, and as more selective treatments are developed, it is necessary to both identify the genetic differences as well as make the tools to measure them routinely available. Since many new targeted therapies are costly, it is imperative they are given to patients who have a high likelihood of benefiting. Six years after the release of the *Focus on Personalised Medicine* report, it is important to see what progress has been made and what gaps still exist.

This particular report looks at the following six lymphoma subtypes and explores the role of genetic mutations in diagnosis, prognosis and treatment. Other non-gene markers and tests are described where applicable to provide a more robust understanding of current knowledge.

1.   Chronic lymphocytic leukaemia (CLL)
2.   Diffuse large B-cell lymphoma (DLBCL)
3.   Follicular lymphoma (FL)
4.   Hodgkin lymphoma (HL)
5.   Mantle cell lymphoma (MCL)
6.   Splenic marginal zone lymphoma (SMZL)

Ultimately, the goal of this work is to improve the clinical care of patients.

*Words highlighted in **bold** are defined in the glossary at the end of the article.*

Cell biology basics

This report will reference 'gene mutations', 'genetic markers', and 'chromosomal abnormalities'. To contextualise and understand these concepts, it is helpful to first understand some basic cell biology.

Human bodies are made up of trillions of cells, and most cells contain a complete set of genes. Each person has thousands of genes that act like a set of instructions, controlling growth and how bodies work. Genes are made of DNA and are carried on thread-like structures called chromosomes. Chromosomes are stored in the nucleus of a cell; usually, a person has 46 chromosomes in each cell, 23 inherited from their mother and 23 from their father. These structures are described in further detail below:

Cells (1) are the basic unit of life. Cells provide structure for the body, convert nutrients from food into energy, and carry out specialised functions. Cells contain the body's hereditary material.[3]

The nucleus (2) is the cell's command and information processing centre. It has two major functions: it coordinates the cell's activities (growth, maturation, division); and it stores the cell's hereditary material which is called DNA.[3]

A chromosome (3) is a thread-like structure made up of DNA that is tightly coiled around proteins called histones. Each chromosome contains many genes and is divided into two 'arms' by a constriction point called the centromere. The short arm is called the 'p arm' and the long arm is called the 'q arm'. The centromere can be located in different places on different chromosomes, giving a chromosome its shape, and it can be used to describe the location of specific genes.[4]

DNA (4), or deoxyribonucleic acid, is the hereditary material in humans. It is a double helix structure that resembles a twisted ladder. The information in DNA is stored as a code made up of four chemical bases: adenine (A), guanine (G), cytosine (C), and thymine (T). The specific order of these bases determines the information available for building and maintaining an organism, much like how letters of the alphabet appear in different orders to form words and sentences. Each base is also attached to a sugar molecule and a phosphate molecule, and all three of these items together (base+ sugar+ phosphate) are called a nucleotide (5). Nucleotides are arranged in two long parallel strands that bind together to form the DNA double helix structure. These strands bind together through base pairing; *A* binds with *T*, and *C* binds with *G* to form the steps of the ladder, and the sugar and phosphate molecules form the vertical side pieces of the ladder.[5]

A gene (6) is the basic unit of heredity. Genes are made up of a segment DNA, and they can range in size from a few hundred DNA bases (A, C, G, T) to more than 2 million bases. Some genes act as instructions to make molecules called proteins. Genes are assigned a name and symbol; the symbol is a short combination of letters (and sometimes numbers) that represent an abbreviated version of the name.[6]
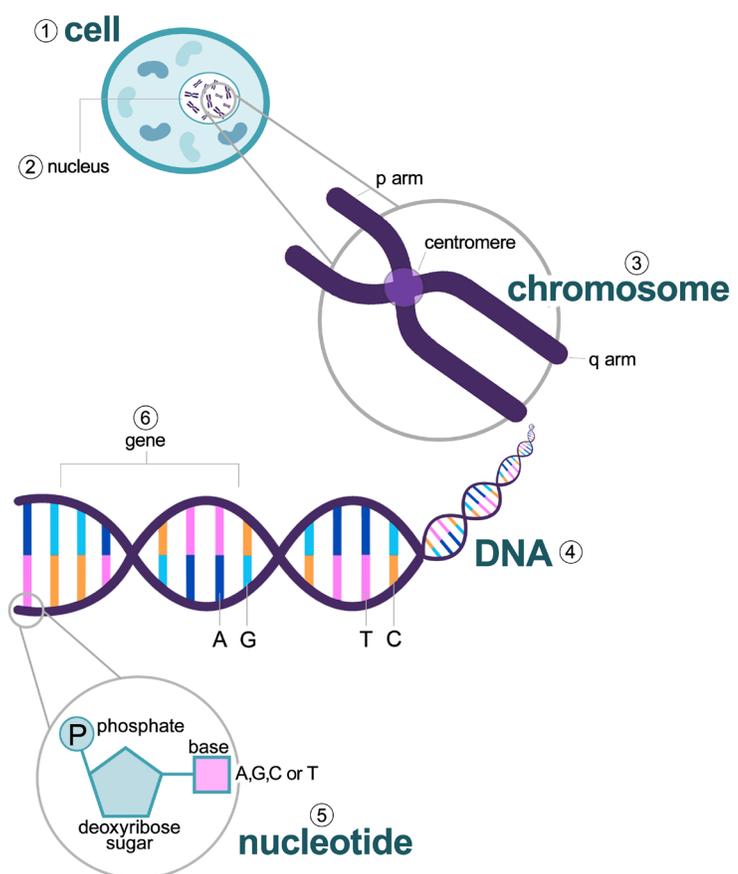
What is a gene mutation?

A gene mutation is a permanent change in the DNA sequence that makes up a gene. A gene mutation can range in size, affecting anywhere from a single DNA base pair (A&T, G&C) to a large DNA segment that involves multiple genes. Mutations can be hereditary (passed on from a parent) or acquired, meaning a change happens during a person's life that usually only affects certain cells. A person's health can be affected because mutations may lead to alterations in a protein's structure, changing the way the protein works, and affect the amount of proteins. Some mutations can change a cell from healthy to cancerous.[7]

What is a genetic marker?

A genetic marker is a specific DNA sequence with a known physical location on a chromosome. It is described as an observable variation, which may arise because of a mutation or an alteration in the gene's location. Each chromosome has many genetic markers. While specific sequences may vary between individuals, there is enough consistency in the genetic code at that particular site on the **genome** to allow comparison between individuals.[8] A marker can have functional consequences, for example, the function or expression of a gene can contribute directly to the development of a disease. A marker can also have no functional consequences but could be located very close to a functional variant (which does alter the function of a gene), which causes both the marker and the variant to be inherited together in the population at large.[9]

How are genetic markers used, and how do they relate to gene mutations?

Genetic markers are particularly important in genetic mapping. They can be used to identify the position of different **alleles** (variant form of a given gene) located on the same chromosome that tend to be inherited together. This is called a linkage group, which can be used to identify unknown genes that influence disease risk. This is valuable for tracking inheritance of traits through generations of a family.[8]

A genetic marker can also act as a specific disease **phenotype** revealing variant on its own.[9] For example, if a large percentage of a disease population (i.e. patients with follicular lymphoma) express a specific genetic marker that those without the disease do not, then the marker can indicate a high level of probability of disease. Genetic markers can also be prognostic, meaning they are used to predict the course of disease or outcome. They can help estimate both the chance of recovery as well as disease recurrence.

Additionally, markers can be helpful for identifying gene mutations. If a mutation occurs in a genetic marker region, the mutation can be easily identified because the normal pattern of this DNA sequence is known.[9]

What is a chromosomal abnormality?

Many cancer cells have a change in their number of chromosomes; human cells normally contain 23 pairs of chromosomes, for a total of 46 chromosomes in each cell. A gain or loss of chromosomes from the normal 46 is called 'aneuploidy'. In cancer cells, these changes are not inherited but occur in somatic cells (cells other than eggs or sperm) during the formation or progression of a cancerous tumour. [10]

Changes that affect the structure of chromosomes can also cause issues with growth, development,

and the function of different body systems. These changes can affect many genes along the chromosome and can disrupt the proteins that are made from those genes.[10] A structural change occurs when pieces of DNA are rearranged within one chromosome or transferred between two or more chromosomes. The effects of these changes depend on their location, size, and if any genetic information has been gained or lost. Some changes may cause medical problems, while others have no effect on health at all.[10]

Chronic lymphocytic leukaemia (CLL)

What is CLL?

Chronic lymphocytic leukaemia (CLL) is often a progressive and persistent form of lymphoma; it presents as a high level of disease-derived B lymphocytes in the blood. Unlike other forms of lymphoma, CLL is often found in the peripheral blood rather than the tissues of the lymphatic system (such as lymph nodes). If this form of cancer is found solely in the lymphatic system, it is usually diagnosed as small lymphocytic lymphoma (SLL). CLL and SLL look identical under a microscope and are considered differing manifestations of the same disease. There is a wide range in the way CLL progresses, from an incidentally discovered asymptomatic abnormality on a routine blood test, to a slow-growing disease that does not require treatment, to a disease that progresses quickly and is resistant to treatment.[11]

Genetic markers in CLL

Several prognostic markers have been established but only a few are used in daily clinical practice.

- **IGHV (immunoglobulin heavy-chain variable):** This gene encodes **antibodies** that function in the immune response. Patients with CLL who express unmutated IGHV tend to have a more aggressive disease that will have an inferior response to chemotherapy-based treatment than those who express mutated IGHV.[11-14] IGHV status does not predict treatment response to targeted therapies like Ibrutinib or Venetoclax.
    - Patients with mutated IGHV are more likely to have a deletion on the long arm of the 13th chromosome (**DEL-13Q**), but not an expression of the antigens **CD38** or **ZAP70**. This indicates a favourable prognosis.
    - Patients with unmutated IGHV are more likely to have disease with expression of the antigens **CD38** or **ZAP70** and are likely to have an unfavourable prognosis.

- **VH3-21 gene:** Regardless of IGHV mutation status, when the CLL cells utilise this form of the immunoglobulin heavy chain gene (VH3-21), prognosis may be unfavourable and similar to those with unmutated IGVH gene status.[11-14]

- **TP53 (tumour protein 53):** Patients with CLL who express a mutation of TP53 have an inferior prognosis and the disease does not respond well to standard chemotherapy.

- **Complex karyotype:** Patients with CLL with a complex karyotype, or at least three chromosomes which are different from what is considered normal, may have poorer progression-free survival (PFS) and overall survival (OS).[15]

All of the patients mentioned above have CLL that is more appropriately treated with small molecule inhibitors (Burton tyrosine kinase, phosphoinositide 3-kinase or B-cell lymphoma-2

inhibitors).[11,13]

Approximately 80 percent of patients with CLL carry at least one or more chromosomal abnormality – an irregular, missing or extra part of the DNA. Identifying chromosomal abnormalities plays a key role in understanding the prognosis of CLL and the likelihood of the CLL responding to different treatments.

These include:

- **13q14 deletion** – Patients with CLL missing genetic material on the long arm of chromosome 13 have a more favourable prognosis if the 13q deletion is their only genetic abnormality. Detected in approximately 50 percent of all CLL cases.

- **Trisomy 12** – Patients with CLL who have an extra copy of chromosome 12 are more likely to have an unfavourable prognosis. Detected in less than 20 percent of all CLL cases.

- **11q deletion** - Patients with CLL missing genetic material on the long arm of chromosome 11 are more likely to have an unfavourable prognosis if treated with chemotherapy. Detected in approximately 10-20 percent of all CLL cases.

- **17p deletion** – Patients with CLL missing genetic material on the short arm of chromosome 17 are more likely to have an unfavourable prognosis. Detected in 5-10 percent of all CLL cases at time of first treatment but becomes increasingly common when the disease recurs after multiple episodes of therapy. Those with a 17p deletion are also less likely to respond to traditional chemotherapy.

Relevant testing suggested in clinical practice guidelines

Abnormality	Test	NCCN (2018)	ESMO (2015)	iwCLL (2018)	Marker Type*
Chromosomal Abnormality					
t(11:14)	FISH	x	x		D
t(11q;v)	FISH	x			D
11q del	FISH	x	x	x	D/P
13q del	FISH	x		x	D/P
17p del	FISH	x	x	x	P
Gene Mutation					
TP53	FISH	x	x	x	P
IGHV (mutational status)	Molecular Analysis	x	x	x	P
Protein/Antigen Expression**					
Cyclin D1	IHC	x			D
ZAP 70	Flow Cytometry	x			D/P
CD38	Flow Cytometry	x			D/P
CD49d	Flow Cytometry	x			D/P

*D= Diagnostic P= Prognostic

**Not gene tests but included due to listing in certain guidelines as diagnostic/prognostic.

See Appendix A for more information about tests used.

Continuing research to watch

There are other genetic markers that may influence prognosis of a patient with CLL, but need additional research:

- **ATM gene**
- **NOTCH1 gene**
- **SF3B1 gene**
- **BIRC3 gene**

> ### *LC recommendations*
>
> It is important to note that genetic abnormalities like deletions and additions of portions of chromosomes can change in people living with CLL due to treatment or during the course of the cancer. For that reason, it is recommended that TP53 mutations and 17p deletion be assessed before every line of new treatment to predict the course of the disease.
>
> Tests for gene mutations, markers and chromosomal abnormalities are not readily accessible in all countries. LC strongly encourages this to change to ensure all patients receive the best treatment for their type of CLL.

See Appendix B for more information about genetic marker clinical trials associated with CLL.

Diffuse large B-cell lymphoma (DLBCL)

What is DLBCL?

Diffuse large B-cell lymphoma is an aggressive, fast-growing form of lymphoma. Under a microscope, the cancerous B cells appear very large and scattered throughout the lymph nodes or tissue. This diffuse growth pattern contributes to the aggressive behaviour of DLBCL. DLBCL accounts for 30 – 40 percent of newly diagnosed lymphomas.[16]

Through **gene expression profiling**, two major subtypes of DLBCL have been identified.[12]

- **Activated B-cell Type (ABC-type):** This subtype is identified by B cells that are in the process of differentiating from germinal centre B cells to plasma cells. Patients with this type of DLBCL have a long-term survival rate of approximately 40 percent.

- **Germinal Centre B-cell Type (GCB-type):** This subtype has a gene expression profile that is characteristic of normal B cells in the germinal centre of the lymph node. Patients with this type of DLBCL have a long-term survival rate of approximately 70 percent.

Genetic markers in DLBCL

Researchers have recently further identified four genetic subtypes of DLBCL, which are seen in both ABC-type and GCB-type DLBCL, and which can help explain why some patients' lymphoma responds better to treatment than others.[17,18]

- **BN2:** Patients who have a fusion of the **BCL6** gene and/or express a mutation on the **NOTCH2**

gene. DLBCL that is identified as BN2 is more likely to respond well to treatment.

- **EZB:** Patients who express mutations on the **EZH2** gene and/or **translocation** (or rearrangement) of the **BCL2** gene. DLBCL that is identified as EZB is more likely to respond well to treatment.

- **MCD:** Patients who express mutations on the **MYD88** gene and the **CD79B** gene. DLBCL that is identified as MCD is less likely to respond well to treatment.

- **N1:** Patients who express mutations on the **NOTCH1** gene. DLBCL that is identified as N1 is less likely to respond well to treatment.

There can also be abnormalities in the way **antigens** are expressed in DLBCL.

- **CD5:** Approximately 10 percent of patients will have disease which expresses **CD5**. The expression of CD5 is not an independent test for prognosis, however it is associated with other poor clinical features such as advanced age, a greater number of lymph nodes affected and a more advanced stage of cancer.[12]

- **BCL2 gene:** Approximately 50 percent of patients with DLBCL will express high levels of the BCL2 protein. An increased expression of BCL2 is associated with lower event-free survival and overall survival (OS).[12]

There can also be chromosomal abnormalities in DLBCL that affect gene functioning.

- **BCL2** rearrangement: Approximately 20 percent of patients with DLBCL have a rearrangement of the BCL2 gene. While some studies show no prognostic significance to this rearrangement, others show a shorter disease-free survival and a poorer response to therapy. BCL2 rearrangement is generally associated with GCB-type DLBCL.[12,19]

- **BCL6** rearrangement: Between 30 and 70 percent of patients with DLBCL express a rearrangement of the BCL6 gene, which plays a key role in GCB-type DLBCL. A lack of BCL6 rearrangement has been associated with a poorer prognosis, while a higher expression level is related to a better prognosis.[12,19]

- **MYC oncogene:** Approximately 10 percent of patients with DLBCL carry a MYC rearrangement, which is associated with a poor prognosis.[12,19]

- **Double Hit** (MYC + BCL2): Patients that carry both MYC and BCL2 rearrangements have what is called "Double Hit" DLBCL. This type of DLBCL is more likely to follow an aggressive clinical course and patients have a poorer overall survival (OS).[12]

There can also be recurrent gene abnormalities which are expressed in DLBCL.

- **BCL6** somatic mutation: Approximately 70 percent of patients with DLBCL will have BCL6 somatic mutations (which are different from rearrangements). The presence of BCL6 somatic mutations is suggestive of a favourable prognostic marker.[12]

- **TP53** (tumour protein 53) mutation: Approximately 20 percent of patients with DLBCL express TP53 mutations. It is present in both GCB-type and ABC-type DLBCL. TP53 mutations are an

independent prognostic marker and are associated with a poor prognosis and unfavourable remission rates.[12,19]

- **MYD88 gene** mutation: Approximately 30 percent of patients with ABC DLBCL have recurrent mutation in the MYD88 gene. Extranodal involvement and **IPI** high risk is common among patients with ABC DLBCL who express this mutation. MYD88 mutation can be a predisposing factor for poorer overall survival in patients with ABC DLBCL.[19]

Other Relevant Indicators

Though not gene tests or markers, the following have also been shown to be associated with a poor prognosis.

- CD43 (cluster differentiation 43): Approximately 20 percent of patients with DLBCL express CD43, which has been shown to be associated with lower event-free survival and overall survival (OS).[12]

- Epstein-Barr Virus (EBV): In certain types of patients with DLBCL (such as the elderly), a positive test for EBV expression within the lymphoma cells is associated with a poor prognosis.[12]

Relevant testing suggested in clinical practice guidelines

Abnormality	Test	NCCN (2018)	ESMO (2015)	Marker Type*
Chromosomal Abnormality				
BCL2	IHC, Molecular Analysis	x	x	P
MYC	IHC, FISH, Karyotype	x	x	D/P
BCL6	IHC, FISH, Karyotype	x		D/P
Double hit (MYC + BCL2 or BCL6)	FISH	x	x	D/P
Cell of Origin				
GBC Variant/non-GBC Variant	Immunophenotyping	x	x	P
Gene Mutation/Rearrangement				
TCR gene rearrangement	PCR	x		D
IgH	PCR	x		D
Protein/Antigen Expression**				
IRF4	IHC	x	x	D
MUM1	IHC	x		D
Cyclin D1	IHC	x	x	D
ALK	IHC	x		D
HHV8	ISH/EBER-1 staining	x		D
SOX11	IHC	x		D
CD10	IHC	x		D
EBV	ISH/EBER-1 staining	x	x	D/P

*D= Diagnostic P= Prognostic
**Not gene tests but included due to listing in certain guidelines as diagnostic/prognostic.

See Appendix A for more information about tests used.

Due to technical challenges of using gene expression profiling in the clinical laboratory, **immunohistochemistry** (IHC) algorithms (a set of rules) are used to identify ABC-type and GCB-

type DLBCL. The most commonly used is the Hans algorithm.[12]

- **ABC-type:** the absence of **CD10** gene and **BCL6** gene expression and the presence of **MUM1** gene expression.
- **GCB-type:** the presence of **CD10** gene and **BCL6** gene expression and the absence of **MUM1** gene expression.

> ### *LC recommendations*
>
> Knowing DLBCL subtype is important but it is currently not tested for routinely. LC encourages patients to ask what their DLBCL subtype is and doctors to test and advise the patient to ensure all patients have more complete information and understanding about their disease. Improved ways to determine prognosis and treatment will be crucial to allow for more individualised and risk-adapted treatment.
>
> In addition, there continues to be research to understand the biology of DLBCL better and in turn find targeted molecular pathways and treatments. It is important to prioritise treatments that have a meaningful effect on the cancer and investigate this within the patient population that will benefit most.

See Appendix C for more information about genetic marker clinical trials associated with DLBCL.

Follicular lymphoma (FL)

What is FL?
Follicular lymphoma is a low-grade, usually slow-growing B cell lymphoma subtype. Based on the current treatments available, advanced stage FL is incurable, and the patient's disease may relapse multiple times. However, many patients have FL that responds well to treatment and they can live with the condition for many years as a chronic disease. Over time, some patient's FL may eventually transform into another form of lymphoma, which is often more aggressive and usually requires different types of treatment.[20]

Genetic markers in FL and research to watch
Currently for patients with FL, few if any **biomarkers** are used to drive treatment selection. This is despite the massive data set that has been and continues to accumulate from clinical trials and patient registries. This is in part because new and novel treatments have been brought into clinical trials faster than the ability to validate prognostic and predictive models.[21]

There are a few genetic markers which show clinical promise to improve patient care and outcomes, but nothing is currently used in practice.

- **M7FLIPI:** The first prognostic test to combine 7 genetic mutations (EZH2, ARID1A, EP300, FOXO1, MEF2B, CREBBP, and CARD11) and the **FLIPI** clinical factors (age, stage, lactate dehydrogenase, haemoglobin, and number of involved lymph nodes sites). The test helps identify if patients are high- or low-risk, and in the future the intensity of treatment may be determined accordingly.[22]

- **BCL6 gene:** BCL6 translocation in FL may constitute a subgroup of patients with a higher risk to have their FL transform into aggressive lymphoma. Of patients with FL who express translocation of the BCL6 gene, 90 percent have loss-of-function mutation of the **KMTD2**, 80 percent have mutations of **CREBBP** or **EP300**.[21]

- **EZH2 gene:** Approximately 25 percent of patients with FL express a mutation in EZH2 gene, which is associated with a favourable outcome. The EZH2 gene is also a therapeutic target.[21]

- **TP53 (tumour protein 53):** TP53 mutation has been correlated with shorter progression-free survival and overall survival among FL patients. TP53 mutation has also been associated with low expression of the immune-response 1 gene expression signature.[23]

The following chromosomal abnormalities and gene mutations have also been observed.

- **t(14;18) chromosomal translocation:** This is the hallmark of FL. Chromosomal translocation of the BLC2 gene to the immunoglobulin heavy chain (IGH) locus (fixed location on a chromosome). The BLC2 gene encodes a protein that inhibits apoptosis (programmed cell death); because the IGH locus is activated in B cells, the translocated BCL2 gene is overexpressed in FL which enhances the B cells survival.[24]

- **3Q27 chromosomal breakpoint:** Chromosomal translocation of the 3Q27 gene is a common genetic mutation in patients with FL.[25]

- **BCL2 gene mutations:** correlate with activation-induced cytidine deaminase (AID) expression and often alter the amino acid sequence of the protein. Mutations in the BCL2 coding sequence at FL diagnosis have been shown to be associated with shorter time to transformation and poorer survival.[24]

Relevant testing suggested in clinical practice guidelines

Abnormality	Test	NCCN (2018)	ESMO (2017)	Marker Type*
Chromosomal Abnormality				
BCL2	IHC, Molecular Analysis, PCR	x	x	D/P
BCL6	IHC, FISH, Karyotype	x		D/P
Protein/Antigen Expression**				
IRF4	IHC, FISH, Karyotype	x		D/P
MUM1	IHC, FISH, Karyotype	x		D
Cyclin D1	IHC	x		D
Ki-67	IHC	x		D

*D= Diagnostic P= Prognostic
**Not gene tests but included due to listing in certain guidelines as diagnostic/prognostic.
See Appendix A for more information about tests used.

See Appendix D for more information about genetic marker clinical trials associated with FL.

Hodgkin lymphoma (HL)

What is HL?

Hodgkin lymphoma is cancer of the lymphatic system which originates in B lymphocytes. In the case of HL, the abnormal cells are called Hodgkin cells and Reed-Sternberg (RS) cells, named after pathologist Thomas Hodgkin and two scientists, Dorothy Reed and Carl Sternberg. HL is primarily a curable cancer and with current treatments, 80 to 90 percent of patients can achieve permanent remission.[26]

Genetic markers in HL and research to watch

Defining the way genetic mutations affect HL is still a major research goal. This deeper understanding can lead to the identification of what is causing HL and springboard the development of targeted treatments.[27]

While no clear genetic markers have been identified in HL, there are certain **cytokines** and **immunologic markers** on cells in the **micro-environment** that may influence prognosis in patients with HL. Additional research is needed before these are incorporated into clinical practice.

- **CCL17 / TARC:** Chemokine (C-C motif) ligand 17 (CCL17) is also known as thymus and activation-regulated chemokine (TARC). TARC is highly elevated in patients with HL and has been proposed as a potential marker.[28] It has been proposed not only as a marker of disease, but also as a predictive marker for response to treatment.

- **PD1, PD-L1+** and **leukocytes:** PD-1 and PD-L1 are both proteins present on the surface of cells. Leukocytes are white blood cells. In one study, patients with HL with tumours with high proportions of PD1, PD-L1 and leukocytes had poorer event-free survival, while patients with HL with low proportions had better event-free survival.[29]

- **CD68+ macrophages** and **CD163+ macrophages:** Patients with HL that show high numbers of CD68+ macrophages and CD163+ macrophages often have worse overall survival. It is thought it may also be correlated with the presence of **Epstein-Barr virus** (EBV).[30]

  - A presence of EBV has also been associated with worse outcomes in older patients with HL.

  - Patients with HL who have a presence of regulatory T cells (**Treg**) and **CD4+** T cells are more likely to have a worse prognosis.

- Patients with HL who have a higher **CD4** to **CD8** ratio are more likely to have lymphoma that will fail ABVD chemotherapy treatment.

Relevant testing suggested in clinical practice guidelines

	Test	NCCN (2018)	ESMO (2018)	Marker Type*
Protein/Antigen Expression**				
CD30	Immunophenotyping	x	x	D
CD15	Immunophenotyping	x	x	D
CD79a	Immunophenotyping	x		D
EBV	ISH/EBER-1 staining	x		D/P

*D= Diagnostic P= Prognostic
**Not gene tests but included due to listing in guidelines as diagnostic/prognostic.

See Appendix A for more information about tests used.

LC recommendations

There is an untapped opportunity to better understand the biology of HL that will allow genetic markers to be used as prognostic tools and treatment predictors. This will help personalise HL treatment and allow for the intensity of treatment to be customised to the patient's specific needs. For this to become clinical reality, more research is needed.

See Appendix E for more information about biomarker clinical trials associated with HL.

Mantle cell lymphoma (MCL)

What is MCL?

Mantle cell lymphoma is a less common cancer of the lymphatic system, which arises in B-cell lymphocytes. When the abnormal B-cells are part of the mantle zone – the outer zone of a lymph node follicle – it is diagnosed as MCL. MCL is a relatively aggressive type of lymphoma; however, more recent observations have shown a subset of patients who have less aggressive forms of MCL and may not need aggressive treatment.[31]

Genetic markers in MCL

There are several genetic markers and mutations that can be used in the prognosis of MCL and help predict its response to treatment.

- **TP53 (tumour protein 53):** A mutation of TP53 is an independent prognostic marker for MCL. Additional mutations may result in a more aggressive cancer.

- **T(11;14)(q13;q32):** The most common genetic alteration in patients with MCL is the translocation in chromosomes 11 and 14, which is expressed as t(11;14)(q13;q32). This switch results in alterations in and an overexpression of **CCND1 (cyclin D1) gene**, which is a diagnostic characteristic of MCL.[19,32]

    - Patients with MCL who express alterations on the CCND1 gene are more likely to have

a poor prognosis and a reduced 5-year survival rate compared to patients who do not express alterations on the CCND1 gene.[33]

- There are rare cases of patients with MCL who do not overexpress CCND1 but may overexpress **cyclin D2** (CCND2) or **cyclin D3** (CCND3).

- **SOX11 gene:** Also known as SRY-box 11 gene, SOX11 is another marker for MCL. It is independent of CCND1, but the presence of both is helpful in diagnosing MCL.[19,34]

    - When patients with MCL do not express SOX11 it may indicate their cancer is indolent, or less aggressive.

    - When patients with MCL express SOX11 and have low expression of **Ki-67**, it is more likely their cancer is less aggressive.

    - Patients with MCL who express SOX11 and receive intensive chemotherapy usually have an improved survival.

- **MYC gene:** Patients with MCL that express translocations of the MYC gene are more likely to have a poor prognosis.[19]

- **NOTCH 1 gene:** Patients with MCL that express the NOTCH 1 gene are more likely to have a poor prognosis.[32]

- **NOTCH 2 gene:** Patients with MCL that express the NOTCH 2 gene are more likely to have a poor prognosis.[32]

- **ATM gene:** Mutations in the ATM gene are most frequently seen in patients with MCL and it is deleted in 50 percent of MCL tumours. However, on its own the ATM mutation does not impact the overall survival of patients with MCL.[32,35]

    - The correlation of the ATM gene with **TP53 gene** could be significant because both are involved in regulating the processes associated with the death of cells.

- **Complex karyotype:** Patients with MCL with a complex karyotype, or at least 3 chromosomes which are different from what is considered normal, may have poorer progression-free survival (PFS) and overall survival (OS).[36]

- **CDN2A/CDK4/RBI axis:** The genes in this pathway are deregulated in MCL, resulting in losses, gains and mutations.[32]

- **CDKN2B/MDM2/TP53 axis:** The genes in this pathway are deregulated in MCL, resulting in losses, gains and mutations.[32]

There is significant knowledge about how genetic markers play a role in MCL, yet there is still no reliable way to translate the data into improved patient care. Consequently, the treatment of MCL is still largely dependent on the clinical features of the cancer, rather than the patient's genetic profile.

Relevant testing suggested in clinical practice guidelines

Abnormality	Test	NCCN (2018)	ESMO (2017)	Marker Type*
Chromosomal Abnormality				
t(11;14)	FISH	x	x	D
t(14;18)	FISH, PCR	x		D
BCL2	IHC, Molecular Analysis	x		D
BCL6	IHC, FISH, Karyotype	x		D
17p del	FISH	x		P
Gene Mutation/Rearrangement				
IGHV	IHC	x		P
IGH rearrangement	PCR		x	D
TP53	FISH	x	x	P
Protein/Antigen Expression**				
CCND1	IHC	x	x	D
Ki-67	IHC		x	D/P
SOX11	IHC	x	x	D/P
CD200	Flow Cytometry	x		D

*D= Diagnostic P= Prognostic
**Not gene tests but included due to listing in certain guidelines as diagnostic/prognostic.

See Appendix A for more information about tests used.

> ### *LC recommendations*
> While the majority of MCL cases are aggressive, it's known that there is a subset of patients whose cancer is indolent and can benefit from a watch and wait treatment approach and delay the effects of aggressive treatment. Better understanding and use of genetic markers will help bring this theory into clinical reality.
>
> Furthermore, while there is much understanding and data that helps determine the prognosis of MCL, this knowledge is still not being used to personalise a patient's treatment to best target their genetic markers. More work in this area is needed.

See Appendix F for more information about genetic marker clinical trials associated with MCL.

Splenic marginal zone lymphoma (SMZL)

What is SMZL?
Marginal zone lymphoma (MZL) is an indolent or slow-growing cancer that is thought to originate from B cells present in the marginal zone of lymphoid follicles. MZL is further divided into three subtypes based on diagnostic criteria. Splenic marginal zone lymphoma is a rare a subtype of MZL and occurs most often in the marginal zone of the spleen. It usually affects elderly or middle-aged patients and has also been associated with Hepatitis C.[37] The median survival of patients with SMZL is 8-10 years, but approximately 30 percent of patients will have a worse prognosis, among which 5-10 percent of patients will have SMZL that will transform into DLBCL.[38]

Genetic markers in SMZL

Due to its rarity and its indolent nature, there are very few clinical trials or treatment options for patients with SMZL. This creates challenges in advancing the understanding of and the role of genetic markers in the diagnosis, prognosis and treatment of patients with SMZL.

There are a few genetic markers that are used to diagnose SMZL, as outlined in the chart below. As well, the following may help predict the course of the disease but are not used routinely in clinic.

- **NF-κ*B*:** Nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κ*B*) is a protein complex that controls DNA transcription (the copying of DNA) and cell survival. Active NF-κ*B* **signalling** is required for normal marginal zone B-cells. However, mutations of positive and negative NF-κ*B* regulators are present in about 35 percent of patients with SMZL. This implicates activation of NF-κ*B* as a major contributor in the development of SMZL.[38,39]

- **NOTCH genes:** Approximately 40 percent of patients with SMZL express mutations in the NOTCH gene pathways. This includes 10 – 25 percent of patients with SMZL expressing recurrent mutations in the **NOTCH 2 gene**.[38,39]
  - The expression of NOTCH 2 mutations in patients with SMZL helps diagnose SMZL and are also related to a poorer outcome.

- **KLF2 gene:** It's been shown that 20 – 42 percent of patients with SMZL express mutations in the KLF2 gene. While it is not used to diagnose SMZL, patients who express the mutation are more likely to receive an unfavourable prognosis.

- **TP53 (tumour protein 53):** Mutation in TP53 are present in approximately 12 percent of patients with SMZL. When patients express abnormalities in the TP53 gene, prognosis may be unfavourable.[32,38]

Relevant testing suggested in clinical practice guidelines

Abnormality	Test	NCCN (2018)	ESMO	Marker Type*
Chromosomal Abnormality				
t(11;18)	FISH, PCR	x		D
MYD88	Molecular Analysis	x		D
t(11;18)	FISH, PCR	x		D
t(11;14)	FISH	x	x	D
t(14;18)	FISH, PCR	x		D
17q isochromosome	FISH		x	D
7q del	FISH	x		D
3q del	FISH		x	D
13q del	FISH		x	D
Protein/Antigen Expression**				
Cyclin D1	IHC	x		D

*D= Diagnostic, all markers listed for SMZL are used for differential diagnosis not prognosis
**Not gene tests but included due to listing in certain guidelines as diagnostic/prognostic.
See Appendix A for more information about tests used.

See Appendix G for more information about genetic marker clinical trials associated with SMZL.

Summary

A better understanding of the role of genetics in the diagnosis, prognosis and treatment of lymphomas continues to evolve, with the goal of improving the clinical care of all patients. Genes and associated mutations can be prognostic as to how a type of lymphoma will progress and predictive as to how it will respond to treatment.

There continues to be significant study into the role of genetics in lymphomas. However, to translate the knowledge into effective clinical practice, studies must be standardised and catalogued in a way that sheds light on how the findings will best serve patients' outcomes.

Furthermore, testing for gene mutations, markers and chromosomal abnormalities is not readily accessible in all countries for many subtypes of lymphoma. While no publicly available data was found outlining which countries have access to which tests in clinical practice, anecdotal evidence from LC member organisations indicates testing is not readily available, especially the most sophisticated tests. For patients to have the best diagnosis and prognosis, and for their healthcare team to recommend the most appropriate treatment, access to testing must be increased.

Appendix A:
Tests used to detect genetic abnormalities/ evidence of cancer

FISH (fluorescence in situ hybridisation): FISH is a test that examines the genes or chromosomes in cells and tissues. It is used to identify genetic mutations and abnormalities. To perform this test, pieces of DNA containing a fluorescent dye are added to the patient's cells or tissues on a glass slide. When these pieces of DNA bind to specific genes or areas of chromosomes on the slide, they light up when viewed under a microscope with a special light.

Flow cytometry: Flow cytometry means cell ('cyto') measurement ('metry') in a fluid stream ('flow'). This test looks at blood cells, marrow cells or cells from a biopsy. Flow cytometry measures the number of cells in a sample, how fast the cells are dividing, and can determine such cell features as size, shape and presence of tumour markers. To perform the test, cells are stained with a dye that is light sensitive and passed in a stream before a laser or another type of light. How the light-sensitive dye reacts is then measured. The test can help determine if there is evidence of cancer.

Immunohistochemistry (IHC): Immunohistochemistry means the composition and properties ('chemistry') of immune ('immuno') tissue ('histo'). IHC uses antibodies to test for certain markers, also called antigens, which are on or in a cell. Antibodies are created in a lab to recognise antigens linked to cancers. When the antibodies bind to the antigen in the tissue sample, the enzyme or dye attached to the **antibody** is activated. This antigen can then be seen under a microscope. IHC helps to diagnose cancer and may also help in distinguishing between different types of cancer.

Immunophenotyping: Immunophenotyping is a process used to determine immune ('immuno') cells observable ('pheno') appearance (type). Immunophenotyping is used to diagnose certain leukaemias and lymphomas. This test uses antibodies to identify cells based on the types on antigens or markers on the cell's surface. Through immunophenotyping, cancer cells are compared with normal cells of the immune system. Immunophenotyping can also be used to separate cells into different groups. Flow cytometry is often the test used to do immunophenotyping.

ISH (in situ hybridisation): ISH or in situ (in place) hybridisation (pairing) uses either a single-stranded DNA or **RNA** sequence called a probe. This probe forms corresponding base pairs with DNA or RNA that is in the patient's tissue or chromosome sample. A chemical or radioactive label is attached to the probe allowing observation of the binding. It is used to identify genetic mutations and abnormalities.

Karyotype: A person's collection of chromosomes. It is used to look for abnormal numbers or structures of chromosome.

Molecular analysis: Complements the clinical and histopathologic tools used to diagnose and sub-classify haematologic malignancies. The presence of clonal antigen-receptor gene rearrangements can help to confirm the diagnosis of a B or T cell lymphoma.

PCR (polymerase chain reaction): PCR is used to find specific DNA sequences, like those found in lymphoma. PCR uses short DNA sequences called primers to select the portion of the genome to be amplified (increased). This technique can produce a billion copies of the target sequence in only a few hours.

Appendix B:
Clinical trials exploring genetic markers in CLL

Genetic Marker	Type	Detection Method	Prognosis	Phase II & III Clinical Trials
11q del	Chromosomal abnormality	FISH, array-based methods, karyotype	Unfavourable	3
17p del	Chromosomal abnormality	FISH, array-based methods, karyotype	Unfavourable	19
13q del	Chromosomal abnormality	FISH, array-based methods, karyotype	Favourable	2
BIRC3	Gene mutation	**NGS**	Unfavourable	1
Trisomy 12	Chromosomal abnormality	FISH, array-based methods, karyotype	Likely unfavourable	1
NOTCH1	Gene mutation	NGS	Unfavourable	2
ZAP-70	Protein expression	Flow cytometry	Unfavourable	4
CD38	Protein expression	Flow cytometry	Unfavourable	1
TP53	Gene mutation	**Sequencing**, FISH	Unfavourable	9
IGVH	Gene mutation status	Molecular analysis, sequencing	Unmutated -> Unfavourable Hypermutated -> favourable	3

As of February 18, 2019. Based on information gathered from Lymphoma Coalition's clinical trial database.

(Sources: Chastain Arch Pathol Lab Med 2015; Rosenquist Haematologica 2016; Rosenquist J Intern Med 2017; Hallek Blood 2018; NCCN Guidelines)

Appendix C:
Clinical trials exploring genetic markers in DLBCL

Genetic Marker	Type	Detection Method	Prognosis	Phase II & III Clinical Trials
MYD88 Mutation	Gene mutation	Sequencing	Likely unfavourable	1
CARD11	Gene mutation	Sequencing	Likely unfavourable	1
EBV	Antigen expression	ISH	Unfavourable	3
GBC variant	Cell of origin	IHC	Favourable	4

Non-GCB variant/ ABC variant	Cell of origin	IHC	Unfavourable	7
BCL2 rearrangement	Chromosomal abnormality	FISH	Neutral	2
BCL6 rearrangement	Chromosomal abnormality	FISH	Likely unfavourable	2
MYC rearrangement	Chromosomal abnormality	FISH, IHC	Unfavourable	4

As of February 18, 2019. Based on information gathered from Lymphoma Coalition's clinical trial database.

(Sources: Chastain Arch Pathol Lab Med 2015; Sun Modern Pathol 2016)

Appendix D:
Clinical trials exploring genetic markers in CLL

Genetic Marker	Type	Detection Method	Prognosis	Phase II & III Clinical Trials
CREBBP	Gene mutation	Sequencing	Unfavourable	1
EZH2	Gene mutation	Sequencing, PCR	Favourable	1
EP300	Gene mutation	Sequencing	Unfavourable	1
BCL2	Gene mutation	IHC, Molecular analysis, PCR	Debated	4

As of February 18, 2019. Based on information gathered from Lymphoma Coalition's clinical trial database

(Sources: Chastain Arch Pathol Lab Med 2015; Sun Modern Pathol 2016)

Appendix E:
Clinical trials exploring biomarkers in HL

Biomarkers/Signature	Phase II & III Clinical Trials
JAK2/JMJD2C	2
CD30	23
EBV	7

As of February 18, 2019. Based on information gathered from Lymphoma Coalition's clinical trial database.

Appendix F:
Clinical trials exploring genetic markers in MCL

Genetic Marker	Type	Detection Method	Prognosis	Phase II & III Clinical Trials
BCL2	Apoptosis	IHC, Molecular analysis, PCR	Debated	1
CCND1	Cell cycle	IHC, FISH, PCR	Unfavourable	1
TP53	DNA repair and integrity	IHC, FISH, PCR	Unfavourable	1
Complex karyotype	DNA repair and integrity	Karyotyping	Unfavourable	1
MYC	Apoptosis	IHC, FISH, PCR	Unfavourable	1

As of February 18, 2019. Based on information gathered from Lymphoma Coalition's clinical trial database.

(Sources: Rosenquist J Intern Med 2017; Inamder Oncotarget 2016; Bachegowda Future Oncol 2014; Inamdar Oncotarget 2016)

Appendix G:
Clinical trials exploring genetic markers in SMZL

No clinical trials have been found studying genetic markers pertaining to SMZL, as of February 18, 2019.

Based on information gathered from Lymphoma Coalition's clinical trial database.
(Source: Rosenquist J Intern Med 2017)

Glossary: Genetic markers

7q deletion: Loss of genetic material located on the long arm (q) of chromosome 7.

13q deletion: Loss of genetic material located on the long arm (q) of chromosome 13.

17p deletion or del(17p): Loss of genetic material located on the short arm (p) of chromosome 17. The tumour suppressor gene TP53 resides on the short arm of chromosome 17. If this short arm (p) is missing, cells cannot use this pathway to prevent and control any malignant cells.

ALK gene (anaplastic lymphoma kinase receptor tyrosine kinase): This gene provides instructions for making the protein ALK receptor tyrosine kinase, which is believed to help regulate the reproduction of nerve cells.

ATM gene (ataxia-telangiectasia mutated serine/threonine kinase): Through the ATM gene, instructions are provided for making a protein that plays an important role in the normal development and activity of several body systems. The ATM protein also assists cells in recognising damaged or broken DNA strands and then coordinates DNA repair.

ARID1A gene (AT-rich interaction domain 1A): Chromatin is the network of DNA and protein that packages DNA into chromosomes. The chromatin structure can be remodelled to alter how tightly DNA is packaged. ARID1A gene provides instructions for making ARID1A protein that is involved in regulating chromatin remodelling.

BCL2 gene (B-cell lymphoma 2): This gene programmes a protein that blocks the death of some cells, such as lymphocytes.

BCL6 gene (B-cell lymphoma 6): The thousands of genes expressed in a cell determine what that cell can do. Transcription is the first step in this process. This protein acts as a repressor (negative influencer) of transcription.

CCND1 gene (cyclin D1): Cyclins are proteins that control how cells move through the cell cycle, the process by which cells divide and grow. Mutations and overexpression of the CCND1 gene are observed frequently in a variety of tumours and may contribute to tumours forming.

CD (cluster of differentiation): CD molecules are markers on the surface of cells that help identify and characterise white blood cells.

- **CD10:** A cell surface enzyme that serves as a marker for follicular lymphoma. CD10 is usually present on the surface of early lymphoma cells.

- **CD15:** A carbohydrate widely used for diagnosing Hodgkin lymphoma. It facilitates phagocytosis (the ingesting by living cells of other cells) and chemotaxis (the response of cells to chemicals).

- **CD30:** A member of the tumour **necrosis** factor family of cell surface receptors (they can induce cell death in certain tumours). Overexpression of CD30 causes expression of nuclear factor-KB (NF-κ*B*), which is considered important in the development of Hodgkin lymphoma.

- **CD38:** CD38 is a multifunctional molecule. It is a component of a molecular network which delivers growth and survival signals to CLL cells.

- **CD49d:** Relevant to tumour progression and metastasis (the development of secondary cancer growths in different sites than the primary cancer).

- **CD79a:** This gene encodes the Ig-alpha (immunoglobulin) protein, which is necessary for expression and function of the B cell antigen receptor (BCR), which then controls the activation of B cells.

- **CD200:** Various cell types, including B cells and some T cells, express this gene which plays a key role in immunosuppression (a reduction in the activity of the immune system) and regulation of anti-tumour activity.

CREBBP gene (CREB binding protein or CBP): Through this gene, instructions are provided for making CREB binding protein which regulates the activity of many genes in the body. The protein also plays an active role in controlling cell growth and division.

EP300 gene (E1A binding protein p300): This gene provides information for making p300, a protein that regulates the activity of many genes and tissues in the body. It has a key role in controlling cell growth and division and prompts cells to mature and differentiate (develop into specialised functions).

EZH2 gene (enhancer of zeste 2 polycomb repressive complex 2 subunit): This gene provides instructions for making the enzyme histone methyltransferase, which can suppress the activity of certain genes.

IgVH (immunoglobulin heavy chain variable gene): This gene encodes antibodies that function in the immune response.

IRF4 gene (interferon regulatory factor 4): The protein encoded by this gene belongs to the IRF (interferon regulatory factor) family of transcription factors which are key in the regulation of interferons. Interferons are proteins that respond to viral infections and help regulate functions of the immune system.

JAK2 gene (Janus kinase 2): This gene provides instructions for making a protein that promotes the growth and division of cells. The JAK2 protein plays a key role in controlling the production of blood cells from stem cells.

JMJD2C (also known as KDM4C/GASC1): This gene helps regulate gene expression and chromosome segregation (cell division).

MUM1 gene (melanoma associated antigen [mutated] 1): MUM1 is needed for DNA repair and cell survival following DNA damage.

MYD88 gene (myeloid differentiation primary response 88): This gene gives instructions for making MyD88, a protein involved in signalling within immune cells that control the body's

immune responses and inflammatory reactions.

MYC (V-Myc avian myelocytomatosis viral oncogene homolog): This gene plays a key role in cell cycle progression, cell death (apoptosis) and cellular transformation. Unusually high amounts of this gene can be observed in numerous cancers.

NOTCH1: This gene provides instructions for making the Notch1 protein, which plays a role in the normal development of many body tissues. For instance, it helps to determine the specialisation of cells into cell types that fulfil particular functions in the body, and it has a role in cell growth, expansion and death. Given the diverse roles of the protein produced by the NOTCH1 gene, the gene is considered both a cancer-causing gene and a tumour suppressor.

NOTCH2: This gene provides instructions for making the Notch2 protein, which is important to the normal development of many body tissues both before and after birth. Notch2 signalling appears to play a role in the development of cells that will be part of the heart, liver, kidneys, teeth, bones and other structures in a growing embryo. Following birth, Notch2 signalling is involved in immune system function, tissue repair and bone remodelling.

SOX11 gene (SRY-box 11): This gene encodes a member of the SOX (SRY-related HMG-box) family of transcription factors that are involved in the regulation of embryonic development and in the determination of the cell's fate. It may also play a role in the development of tumours.

t(1;14), t(3;14), t(11;14), t(11;18), t(14;18): t stands for translocation. All refer to chromosomal abnormalities in a person's DNA. These abnormalities occur when a portion of a chromosome is relocated or exchanges parts with another unlike chromosome. Several cancers can result from these abnormalities.

TCR gene rearrangement (T-cell receptor gene rearrangement): Diverse TCR-gene rearrangement is important for normal immune functions. The development of diverse T cells protects against many kinds of antigens. If this process goes wrong and an abnormal T cell reproduces numerous copies of itself, it may result in disease.

TP53 (tumour protein 53): This TP53 gene gives instructions for making TP53, a protein which acts as a tumour suppressor by keeping cells from growing and dividing too fast or in an uncontrolled way. Dysfunction of the TP53 tumour suppressor gene is common in many blood cancers.

ZAP70 (zeta chain of T cell receptor associated protein kinase 70): This gene provides instructions for making zeta-chain-associated protein kinase (ZAP70), a protein that is part of a signalling pathway that directs development of T cells as well as activates T cells. T cells identify foreign substances and defend the body against infection.

Glossary: Terms

Allele: An allele is a variant form of a gene. Each gene lives at a specific location on a chromosome in two copies, one copy of the gene is inherited from each parent. The copies are not necessarily the same, and when they differ from each other they are known as alleles.

Antibody (also known as an immunoglobulin): A protein produced in response to and counteracting a specific antigen (e.g. bacteria, virus). The antibody attaches to the antigen and tags it for attack by another part of the immune system or directly neutralises the antigen itself, making it ineffective.

Antigen: Either a toxin or foreign substance that the immune system recognises should not be present (such as bacteria, viruses). This triggers an immune response in the body, especially the production of antibodies. An antigen can also be a normal protein on a normal cell.

Biomarker: Also called a molecular marker or a signature molecule, a biomarker is a biological molecule found in blood, other body fluids or tissues that is an indication of either a normal or abnormal process, or of a condition or a disease. It can be used to see how well the body is responding to a treatment.

Cytokines: Small secreted proteins released by cells that have a specific effect on the interactions and communications between cells.

FLIPI: The Follicular Lymphoma International Prognostic Index estimates overall survival based on clinical information. The FLIPI risk stratifies FL patients based on prognosis and helps guide when and how to treat such patients.

Genetic profiling: Gathering of information about specific genes, including variations from what is considered normal, and gene expression, in an individual or in a certain type of tissue. This information can help diagnose a disease, suggest how the disease may progress or how the disease will respond to treatment.

Gene-expression profiling (GEP): Messenger RNA (mRNA) molecules carry the genetic information needed to make proteins. GEP examines the relative abundance of mRNA from the various genes. GEP can assist in making a diagnosis; it can also determine response to treatment.

Genome: This is an organism's complete set of DNA, including all its genes. Each genome contains all the information needed to build and maintain that organism. In humans, a copy of the entire genome – more than 3 billion DNA base pairs – is contained in all cells that have a nucleus.

Immunohistochemistry (IHC): Immunohistochemistry means the composition and properties ('chemistry') of immune ('immuno') tissue ('histo'). IHC uses antibodies to test for certain markers, also called antigens, which are on or in a cell.

Immunologic marker: Markers on normal immune cells (B cells, T cells) in the microenvironment that could point towards malignancy.

International Prognostic Index (IPI): A scoring system that assesses prognosis using risk factors (age, stage, etc.). A risk factor is anything that increases the chance of an outcome or event. The IPI is a clinical tool developed by oncologists to aid in predicting the prognosis of patients with many aggressive lymphomas.

Isochromosome: An abnormal chromosome that has arms which are mirror images of each other. The chromosome will have two copies of either the long arm or the short arm. This is because there is a simultaneous duplication and deletion of genetic material when the isochromosome is formed.

Leukocyte: White ('leuko') blood cell ('cyte'). These cells are made in the bone marrow. Leukocytes help the body fight infection and other diseases. Granulocytes (neutrophils, eosinophils, and basophils), monocytes and lymphocytes (B cells and T cells) are types of leukocytes.

Micro-environment (cellular): Local surroundings with which cells interact by processing various chemical and physical signals, and by contributing their own effects to this environment.

Molecular profiling: Identifies and documents the structure of a specific DNA, RNA or protein molecule. This information helps in the diagnosis of disease.

NGS (next-generation sequencing): A DNA sequencing technology. With NGS, the human genome can be sequenced in one day. This is much faster than previously available technology.

Necrosis: Cell death caused by an external force, like an infection, trauma or poison.

Oncogene: The word means tumour ('onco') gene. Oncogenes can cause cancer cells to grow. They are a mutated form of a gene involved in normal cell growth. They can be inherited or result from exposure to substances in the environment.

Phenotype: The visible or observable expression of the results of genes. The genotype is the set of genes in human DNA which is responsible for a particular trait, the phenotype is the physical expression, or characteristics of that trait.

Precision medicine: Emerging approach for disease treatment and prevention that accounts for individual variability in genes, environment, and lifestyle for each person.

Predictive (marker): Can be used to help tell whether a person's cancer will respond to a specific treatment. They can also identify something that may increase a person's risk of developing a disease.

Prognostic (marker): Biological characteristics that can be used to predict the course of a disease or a response to a therapeutic intervention. Some examples include the presence of a particular gene variant or levels of a particular protein in body fluids.

Proteins: Proteins develop within cells and are essential for life. There are many types of proteins within a body, each with a specific role. They are necessary for cell structure, function and regulation.

Ribonucleic acid (RNA): Unlike DNA, RNA is single stranded. Within a cell different types of RNA exist: messenger RNA (mRNA), ribosomal RNA (rRNA) and transfer RNA (tRNA). Research has shown that some small RNAs are involved in regulating gene expression (a range of processes that cells use to increase or decrease the production of specific gene products, like proteins).

Sequencing (DNA): The process of determining the sequence of the nucleotide bases (A, T, G, C) along a strand of DNA. Some types of sequencing include:

- **WES (whole-exome sequencing):** This lab process determines the nucleotide sequence primarily of the exomic (protein-coding) regions of an individual's genome and related sequences. This represents approximately 1 percent of the complete DNA sequence.

- **WGS (whole-genome sequencing):** This lab process determines nearly all of the approximately 3 billion nucleotides of an individual's complete DNA sequence.

Signalling pathways: A group of molecules work together to control one or more cell functions, such as cell division or death. When the first molecule in a pathway receives a signal, it activates another molecule. This process continues until the last molecule is activated and the cell function occurs. If the activation in the signalling pathway is abnormal, it can result in cancer. Some of the signalling pathways that play a role in lymphoma are:

- B cell receptor (BCR) signalling
- NF-κ*B* signalling pathway
- Toll-like receptor signalling
- Notch signalling

Translocations (chromosomal): When a piece of one chromosome breaks off and attaches to another chromosome. This type of rearrangement is called balanced if no genetic material is gained or lost or in the cell; if there is gain or loss of genetic material it is called unbalanced.

References

1. American Association for Clinical Chemistry. Genetic tests for targeted cancer therapy. Lab Tests Online website. https://labtestsonline.org/tests/genetic-tests-targeted-cancer-therapy. Updated November 12, 2019. Accessed November 14, 2019.

2. National Cancer Institute. Targeted cancer therapies. National Institutes of Health website. https://www.cancer.gov/about-cancer/treatment/types/targeted-therapies/targeted-therapies-fact-sheet. Updated September 23, 2019. Accessed September 26, 2019.

3. U.S. National Library of Medicine. What is a cell? National Institutes of Health website. https://ghr.nlm.nih.gov/primer/basics/cell. Updated November 12, 2019. Accessed November 14, 2019.

4. U.S. National Library of Medicine. What is a chromosome? National Institutes of Health website. https://ghr.nlm.nih.gov/primer/basics/chromosome. Updated November 12, 2019. Accessed November 14, 2019.

5. U.S. National Library of Medicine. What is DNA? National Institutes of Health website. https://ghr.nlm.nih.gov/primer/basics/dna. Updated November 12, 2019. Accessed November 14, 2019.

6. U.S. National Library of Medicine. What is a gene? National Institutes of Health website. https://ghr.nlm.nih.gov/primer/basics/gene. Updated November 12, 2019. Accessed November 14, 2019.

7. National Cancer Institute. The genetics of cancer. National Institutes of Health website. https://www.cancer.gov/about-cancer/causes-prevention/genetics. Updated October 12, 2019. Accessed October 15, 2019.

8. National Human Genome Research Institute. Genetic mapping fact sheet. National Institutes of Health website. https://www.genome.gov/about-genomics/fact-sheets/Genetic-Mapping-Fact-Sheet. Updated October 21, 2015. Accessed September 6, 2019.

9. Raza S, Shoaib MW, Mubeen H. Genetic markers: Importance, uses and applications. International Journal of Scientific and Research Publications. 2016; 6(3). ISSN 2250-3153.

10. U.S. National Library of Medicine. Can changes in the structure of chromosomes affect health and development? National Institutes of Health website. https://ghr.nlm.nih.gov/primer/mutationsanddisorders/structuralchanges. Updated November 12, 2019. Accessed November 14, 2019.

11. Lymphoma Coalition. 2017 Global subtype report on chronic lymphocytic leukaemia. Lymphoma Coalition website. https://www.lymphomacoalition.org/images/subtype-reports/CLL_Global_2017_Report.pdf. Updated June 9, 2017. Accessed September 6, 2019.

12. Chastain EC, Duncavage, EJ. Clinical prognostic biomarkers in chronic lymphocytic leukemia and diffuse large b-cell lymphoma. Archives of Pathology & Laboratory Medicine. 2015. 139(5). doi: https://doi.org/10.5858/arpa.2014-0086-RA

13. Hallek M, Cheson BD, Catovsky et al. iwCLL guidelines for diagnosis, indications for treatment, response assessment, and supportive management of CLL. Blood. 2018; 131(25). doi: 10.1182/blood-2017-09-806398

14. National Comprehensive Cancer Network (NCCN). NCCN practice guidelines in oncology: Chronic lymphocytic leukemia/small lymphocytic lymphoma. NCCN website. https://www.nccn.org/professionals/physician_gls/pdf/cll.pdf Updated October 8, 2019. Accessed October 15, 2019.

15. Baliakas P, Jeromin S, Iskas M et al. Cytogenic complexity in chronic lymphocytic leukemia: Definitions, associations, and clinical impact. Blood. 2019; 133(11). doi: 10.1182/blood-2018-09-873083

16. Lymphoma Coalition. Diffuse large b-cell lymphoma subtype report. Lymphoma Coalition website. https://www.lymphomacoalition.org/images/subtype-reports/DLBCL-Report-Sept-2016---Final.pdf. Updated Sept 27, 2016. Accessed October 9, 2019.

17. Schmitz R, Wright GW, Huang DW, et al. Genetics and pathogenesis of diffuse large b-cell lymphoma. The New England Journal of Medicine. 2018. 378:1396-1407. doi: 10.1056/NEJMoa1801445

18. National Institutes of Health. NIH study revises molecular classification for most common type of lymphoma. National Institutes of Health website [news release]. https://www.nih.gov/news-events/news-releases/nih-study-revises-molecular-classification-most-common-type-lymphoma. Updated April 11, 2018. Accessed September 6, 2019.

19. Sun R, Medeiros LJ, Young KH. Diagnostic and predictive biomarkers for lymphoma diagnosis and treatment in the era of precision medicine. Modern Pathology. 2016. 29:1118-1142. doi: https://doi.org/10.1038/modpathol.2016.92

20. Lymphoma Coalition. Follicular lymphoma subtype report. Lymphoma Coalition website. https://www.lymphomacoalition.org/images/subtype-reports/FL-Report-Sept-2016---Final.pdf. Updated September 26, 2016. Accessed September 6, 2019.

21. Weigert O, Weinstock DM. The promises and challenges of using gene mutations for patient stratification in follicular lymphoma. Blood. 2017. 130(13). doi: 10.1182/blood-2017-07-73735

22. LaCasce AS. M7FLIPI: A follicular lymphoma prognostic model for the 21st century. The Hematologist. 2015.12(6).

23. O'Shea D, O'Riain C, Taylor C et al. The presence of TP53 mutation at diagnosis of follicular lymphoma identifies a high-risk group of patients with shortened time to disease progression and poorer overall survival. Blood. 2008. 112(8). doi: 10.1182/blood-2008-05-154013

24. Correira C, Schneider A, Dai H et al. BCL2 mutations are associated with increased risk of transformation and shortened survival in follicular lymphoma. Blood. 2015. 125(4). doi: https://doi.org/10.1182/blood-2014-04-571786

25. Vergilio J, Gay LM, Zhong L et al. Comprehensive genomic profiling (CGP) identifies novel BCL6 rearrangements in diverse subtypes of non-Hodgkin lymphoma (NHL) as well as known rearrangements not detected using standard of care (SOC) assays. Blood. 2017. 130(1). doi: https://doi.org/10.1182/blood.V130.Suppl_1.417.417

26. Lymphoma Coalition. 2017 Global subtype report on Hodgkin lymphoma. Lymphoma Coalition website. https://www.lymphomacoalition.org/images/subtype-reports/Hodgkin-Lymphoma_Subtype_Report-1_1.pdf. Updated May 1, 2017. Accessed September 6, 2019.

27. Mata E, Diaz-Lopez A, Martin-Moreno A et al. Analysis of the mutational landscape of classic Hodgkin lymphoma identifies disease heterogeneity and potential therapeutic targets. Oncotarget. 2017. 8(67). doi: 10.18632/oncotarget.22799

28. Sauer M, Plutschow A, Jachimowicz RD et al. Baseline serum TARC levels predict therapy outcome in patients with Hodgkin lymphoma. American Journal of Hematology. 2013. 88(2). doi: 10.1002/ajh.23361

29. Hollander P, Kamper P, Smedby KE et al. High proportions of PD-1+ and PD-L1+ leukocytes in classical Hodgkin lymphoma microenvironment are associated with inferior outcome. Blood Advances. 2017. 1(18). doi:https://doi.org/10.1182/bloodadvances.2017006346

30. Karantanos T, Politikos I, Boussiotis VA. Advances in the pathophysiology and treatment of relapsed/refractory Hodgkin's lymphoma with an emphasis on targeted therapies and transplantation strategies. 2017. Blood and Lymphatic Cancer: Targets and Therapy. 7:37-52. doi:10.2147/BLCTT.S105458

31. Lymphoma Coalition. Mantle cell lymphoma subtype report. Lymphoma Coalition website. https://www.lymphomacoalition.org/images/subtype-reports/MCL_Report_Nov_2016_-_Final.pdf. Updated November 4, 2016. Accessed September 6, 2019.

32. Rosenquist R, Bea S, Nadel B et al. Genetic landscape and deregulated pathways in B-cell lymphoid malignancies. Journal of Internal Medicine. 2017. 282(5). doi: 10.1111/joim.12633

33. Inamdar AA, Goy A, Ayoub NM et al. Mantle cell lymphoma in the era of precision medicine-diagnosis, biomarkers and therapeutic agents. Oncotarget. 2016. 7(30). doi: 10.18632/oncotarget.8961

34. Dreyling M, Campo E, Hermine O et al. Newly diagnosed and relapsed mantle cell lymphoma: ESMO clinical practice guidelines for diagnosis, treatment and follow-up. Annals of Oncology. 2017. 28(4). doi: 10.1093/annonc/mdx223

35. Ahmed M, Zhang L, Nomie K et al. Gene mutations and actionable genetic lesions in mantle cell lymphoma. Oncotarget. 2016. 6(7). doi: 10.18632/oncotarget.10716

36. Greenwell IB, Switchenko JM, Staton AD et al. Complex karyotype predicts the prognostics impact of WBC count in untreated mantle cell lymphoma and is associated with shortened survival independent of specific cytogenetic abnormalities. Blood. 2017. 130(1). doi: https://doi.org/10.1182/blood.V130.Suppl_1.1499.1499

37. Lymphoma Coalition. Marginal zone lymphoma subtype report. Lymphoma Coalition website. https://www.lymphomacoalition.org/images/subtype-reports/MZL_Report_Feb_2017_Final.pdf. Updated February 2017. Accessed September 6, 2019.

38. Arcaini L, Rossi D, Paulli M. Splenic marginal zone lymphoma: From genetics to management. Blood. 2016. 127(17). doi: https://doi.org/10.1182/blood-2015-11-624312

39. Campos-Martin Y, Martinez N, Martinez-Lopez A et al. Clinical and diagnostic relevance of NOTCH2-and KLF2-mutations in splenic marginal zone lymphoma. Haematologica. 2017. 102: e310-e312. doi: 10.3324/haematol.2016.161711

LYMPHOMA COALITION

Worldwide Network of Lymphoma Patient Groups
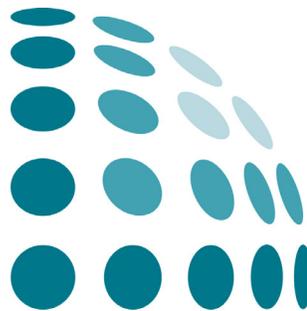
Since 2002

www.lymphomacoalition.org