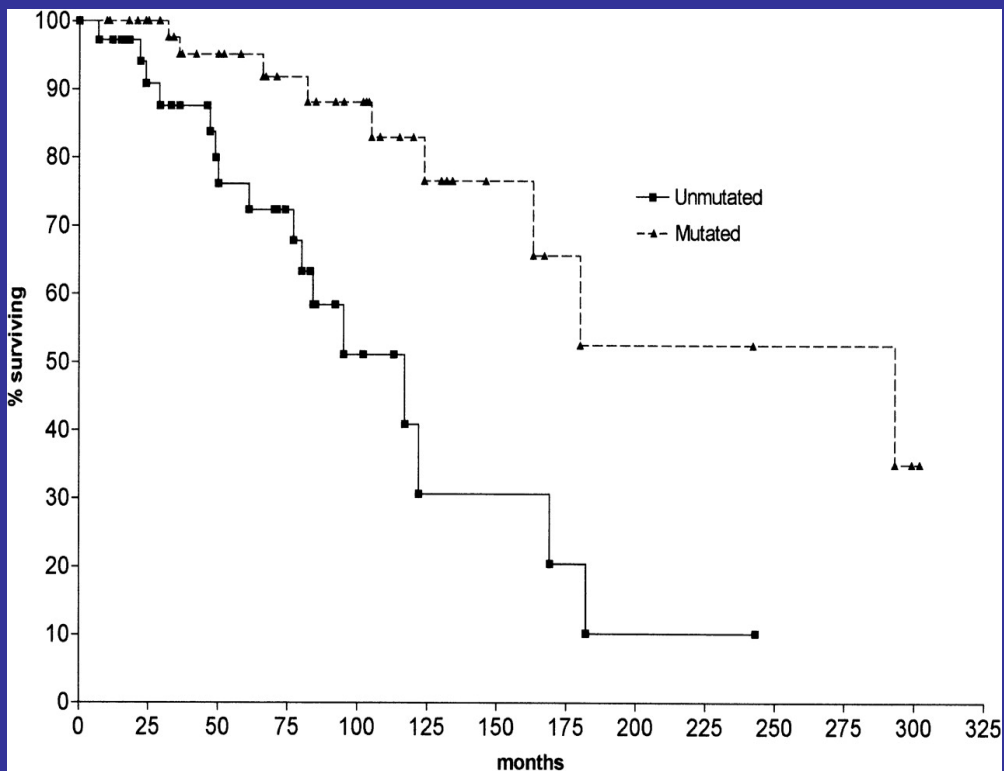


How to report Ig gene data in clinical routine?



Hamblin et al, Blood 1999

Richard Rosenquist

How to report Ig gene data in clinical routine?

- **Basic data**

- Patient data
- Tissue type
- Arrival date
- Referral dr
- etc

- **Technique**

- PCR, primer set
- Direct sequencing, subcloning

- **Result**

- IGHV, IGHD, IGHJ
- Homology
- Reading frame

- **Conclusion**

**UPPSALA UNIVERSITY HOSPITAL
UPPSALA, SWEDEN**



Molecular Pathology, Rudbeck Laboratory

Clinical Scientist: Ingrid Thörn

Senior Physician: Richard Rosenquist

Telephone: 018-611 0000

Fax: 018-554025

**Determination of immunoglobulin heavy-chain variable (IGHV)
gene mutational status in chronic lymphocytic leukemia**

Patient name: **Sven Svensson**

Date of birth: **420319-2020**

Arrival date: **07-06-05**

Sample type: **Peripheral blood**

Referral doctor: **Eva Kimby**

Technique:

PCR amplification using framework 1 BIOMED2 primers.

Direct sequencing.

Results:

IGHV: 3-72

Homology:

95.7%

IGHD: 3-16

Reading frame:

In frame

IGHJ: 6

Conclusion:

This CLL sample displays a functional **IGHV mutated** rearrangement. Mutated IGHV genes have been associated with good clinical outcome.

Dr. Richard Rosenquist

**UPPSALA UNIVERSITY HOSPITAL
UPPSALA, SWEDEN**



Molecular Pathology, Rudbeck Laboratory

Clinical Scientist: Ingrid Thörn

Senior Physician: Richard Rosenquist

Telephone: 018-611 0000

Fax : 018-554025

Determination of immunoglobulin heavy-chain variable (IGHV) gene mutational status in chronic lymphocytic leukemia

Patient name: **Sven Svensson** Date of birth: **420319-2020**
Arrival date: **07-06-05** Sample type: **Peripheral blood**
Referral doctor: **Eva Kimby**

Technique:

PCR amplification using framework 1 BIOMED2 primers.
Direct sequencing.

Results:

IGHV:	1-69	Homology:	99.5%
IGHD:	3-16	Reading frame:	In frame
IGHJ:	6		

Conclusion:

This CLL sample displays a functional **IGHV unmutated** rearrangement. Unmutated IGHV genes have been associated with poor clinical outcome.

Dr. Richard Rosenquist

**UPPSALA UNIVERSITY HOSPITAL
UPPSALA, SWEDEN**



Molecular Pathology, Rudbeck Laboratory

Clinical Scientist: Ingrid Thörn

Senior Physician: Richard Rosenquist

Telephone: 018-611 0000

Fax : 018-554025

Determination of immunoglobulin heavy-chain variable (IGHV) gene mutational status in chronic lymphocytic leukemia

Patient name: **Sven Svensson** Date of birth: **420319-2020**
Arrival date: **07-06-05** Sample type: **Peripheral blood**
Referral doctor: **Eva Kimby**

Technique:

PCR amplification using framework 1 BIOMED2 primers.
Direct sequencing.

Results:

IGHV:	3-21	Homology:	96.7%
IGHD:	-	Reading frame:	In frame
IGHJ:	6		

Conclusion:

This CLL sample displays a functional **IGHV3-21 mutated** rearrangement. IGHV3-21 usage has been associated with poor clinical outcome regardless mutational status.

Dr. Richard Rosenquist

**UPPSALA UNIVERSITY HOSPITAL
UPPSALA, SWEDEN**



Molecular Pathology, Rudbeck Laboratory

Clinical Scientist: Ingrid Thörn

Senior Physician: Richard Rosenquist

Telephone: 018-611 0000

Fax : 018-554025

Determination of immunoglobulin heavy-chain variable (IGHV) gene mutational status in chronic lymphocytic leukemia

Patient name: **Sven Svensson** Date of birth: **420319-2020**
Arrival date: **07-06-05** Sample type: **Peripheral blood**
Referral doctor: **Eva Kimby**

Technique:

PCR amplification using framework 1 BIOMED2 primers.
Direct sequencing.

Results:

IGHV:	3-72	Homology:	97.6%
IGHD:	3-16	Reading frame:	In frame
IGHJ:	6		

Conclusion:

This CLL sample displays a functional **IGHV mutated** rearrangement with **borderline homology**. Caution should be made when interpreting the clinical correlation.

Dr. Richard Rosenquist

Molecular Pathology, Rudbeck Laboratory

Clinical Scientist: Ingrid Thörn

Senior Physician: Richard Rosenquist

Telephone: 018-611 0000

Fax : 018-554025

**Determination of immunoglobulin heavy-chain variable (IGHV)
gene mutational status in chronic lymphocytic leukemia**

Patient name: **Sven Svensson** Date of birth: **420319-2020**
Arrival date: **07-06-05** Sample type: **Peripheral blood**
Referral doctor: **Eva Kimby**

Technique:

PCR amplification using framework 1 BIOMED2 primers.
Direct sequencing.

Results:

Rearrangement 1

IGHV:	1-69	Homology:	99.5%
IGHD:	3-16	Reading frame:	In frame
IGHJ:	6		

Rearrangement 2:

IGHV:	3-30	Homology:	100%
IGHD:	3-3	Reading frame:	In frame
IGHJ:	4		

Conclusion:

This CLL sample displays two functional IGHV rearrangements and would be interpreted as expressing **unmutated IGHV genes**.

Dr. Richard Rosenquist

Molecular Pathology, Rudbeck Laboratory

Clinical Scientist: Ingrid Thörn

Senior Physician: Richard Rosenquist

Telephone: 018-611 0000; Fax: 018-554025

Determination of immunoglobulin heavy-chain variable (IGHV) gene mutational status in chronic lymphocytic leukemia

Patient name: **Sven Svensson**

Date of birth: **420319-2020**

Arrival date: **07-06-05**

Sample type: **Peripheral blood**

Referral doctor: **Eva Kimby**

Technique:

PCR amplification using framework 1 BIOMED2 primers.

Direct sequencing.

Results:

Rearrangement 1

IGHV: 1-69

Homology: 99.5%

IGHD: 3-16

Reading frame: In frame

IGHJ: 6

Rearrangement 2:

IGHV: 4-34

Homology: 99.5%

IGHD: 2-2

Reading frame: Stop codon in junction

IGHJ: 4

Conclusion:

This CLL sample displays a functional IGHV unmutated rearrangement and a non-functional IGHV unmutated rearrangement. Altogether, the interpretation is a case with **unmutated IGHV genes**.

Dr. Richard Rosenquist

**UPPSALA UNIVERSITY HOSPITAL
UPPSALA, SWEDEN**



Molecular Pathology, Rudbeck Laboratory

Clinical Scientist: Ingrid Thörn

Senior Physician: Richard Rosenquist

Telephone: 018-611 0000

Fax: 018-554025

Determination of immunoglobulin heavy-chain variable (IGHV) gene mutational status in chronic lymphocytic leukemia

Patient name: **Sven Svensson**

Date of birth: **420319-2020**

Arrival date: **07-06-05**

Sample type: **Peripheral blood**

Referral doctor: **Eva Kimby**

Technique:

PCR amplification using framework 1 BIOMED2 primers.

Direct sequencing.

Results:

IGHV: 1-69

Homology:

99.5%

IGHD: 3-16

Reading frame:

Out of frame

IGHJ: 6

Conclusion:

This CLL sample displays only a non-functional IGHV mutated rearrangement. At present, the clinical correlation for this case cannot be made.

Dr. Richard Rosenquist