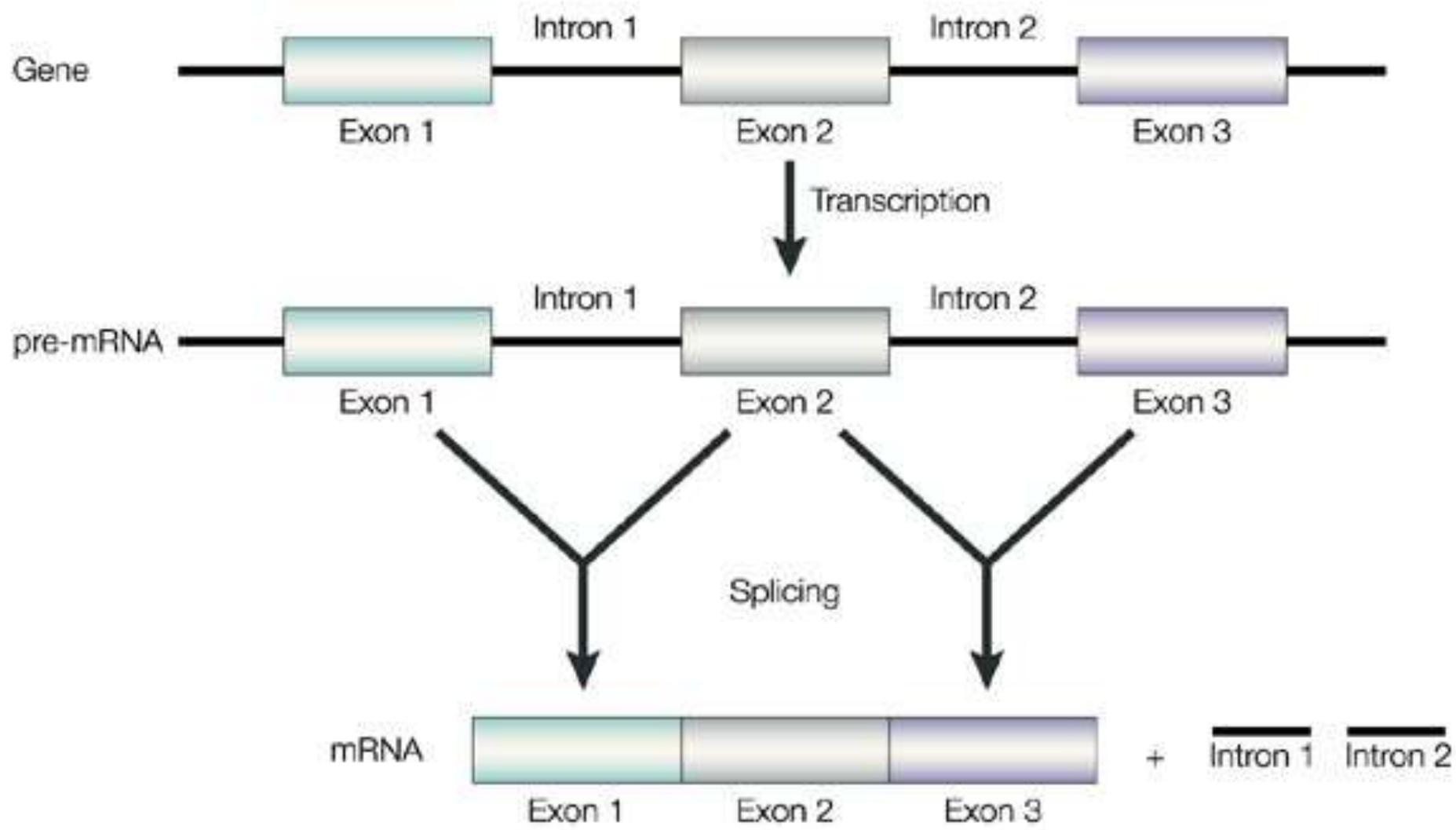


# Splicing variants.

P. Pauwels  
(UZA, UA)



## REVIEW

# Exon 14 Deleted MET Receptor as a New Biomarker and Target in Cancers

Alexis B. Cortot, Zoulika Kherrouche, Clotilde Descarpentries, Marie Wislez, Simon Baldacci, Alessandro Furlan, David Tulasne

**Affiliations of authors:** UMR 8161 - M3T - Mechanisms of Tumorigenesis and Targeted Therapies, CNRS, Institut Pasteur de Lille, Univ. Lille, Lille, France (ABC, ZK, SB, AF, DT); Thoracic Oncology Department, CHU Lille, Univ. Lille, Lille, France (ABC); Division of Biochemistry and Molecular Biology, Oncology and Molecular Genetics Laboratory, CHU Lille, Lille, France (CD); Service de Pneumologie, Hôpital Tenon, AP-HP, Paris, France (MW).

**Correspondence to:** David Tulasne, PhD, UMR 8161 - M3T - Mechanisms of Tumorigenesis and Targeted Therapies, CNRS, Institut Pasteur de Lille, Univ. Lille, F-59000 Lille, France (e-mail: david.tulasne@ibl.cnrs.fr); or Alexis B. Cortot, MD, PhD, UMR 8161 - M3T - Mechanisms of Tumorigenesis and Targeted Therapies, CNRS, Institut Pasteur de Lille, Univ. Lille, F-59000 Lille, France (e-mail: alexis.cortot@chru-lille.fr).

1) Molecule binds to protein receptor

2) Receptor-molecule moves to clathrin-coated pit

3) Cell membrane folds inwards

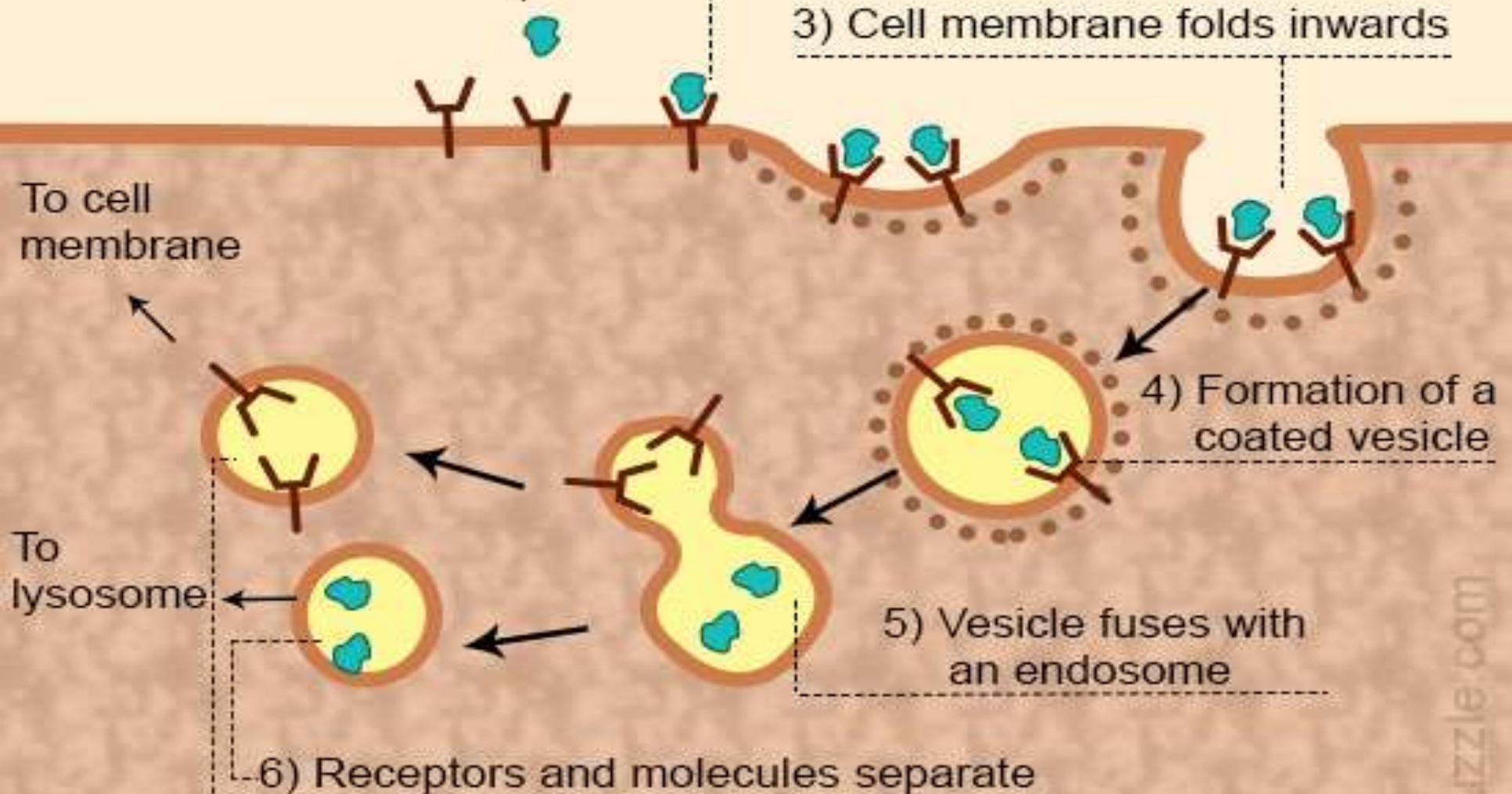
4) Formation of a coated vesicle

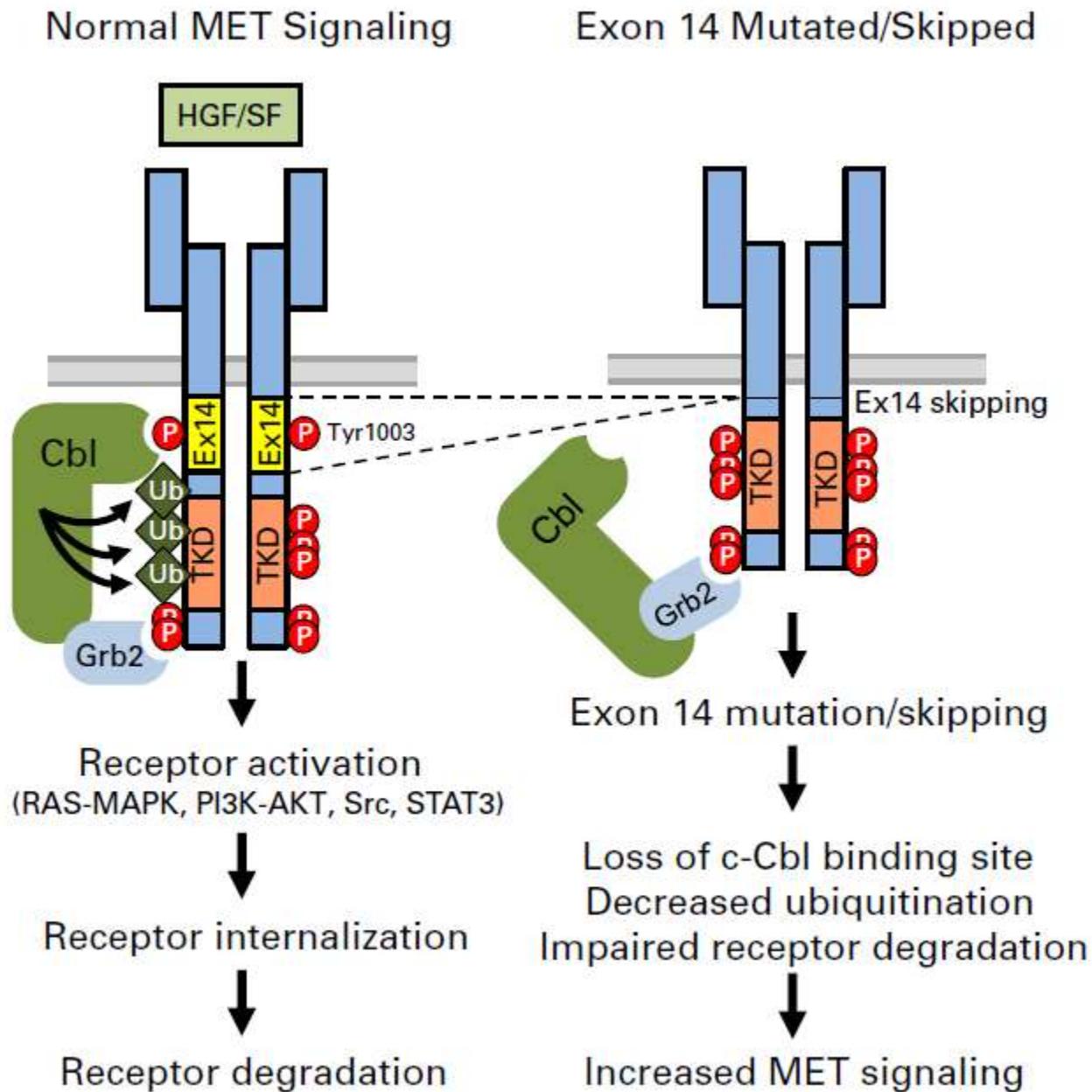
5) Vesicle fuses with an endosome

6) Receptors and molecules separate

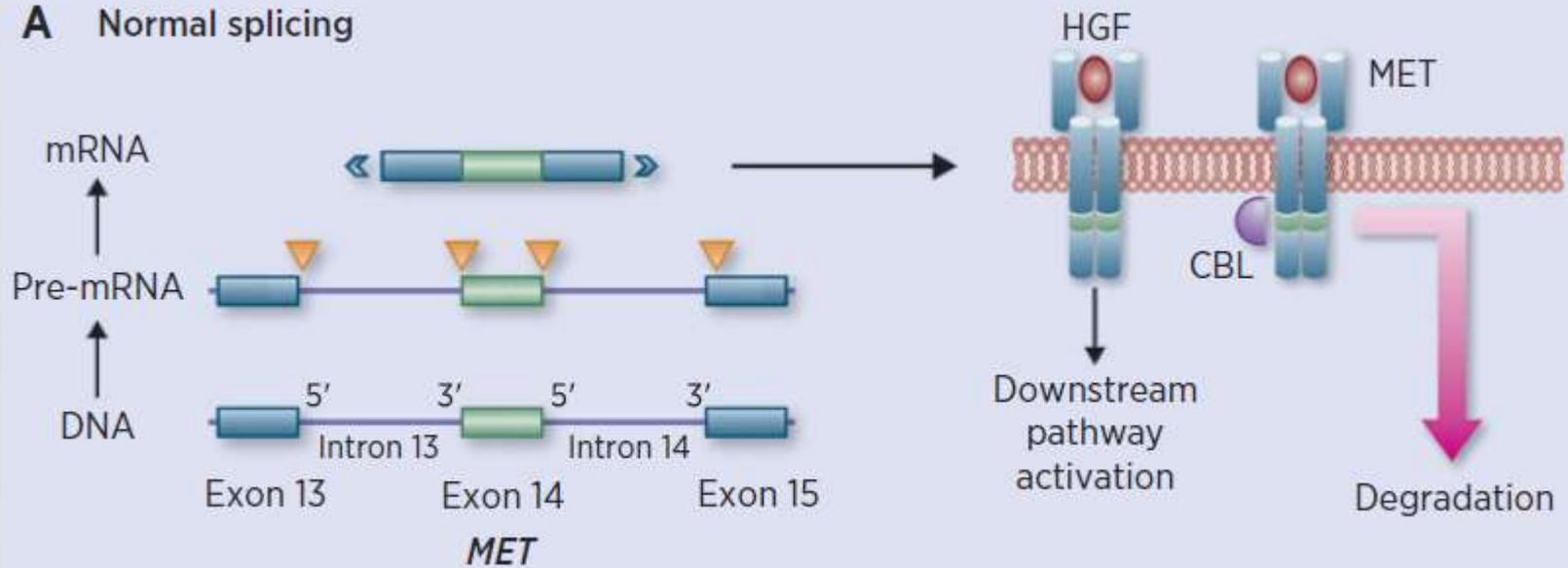
To cell membrane

To lysosome

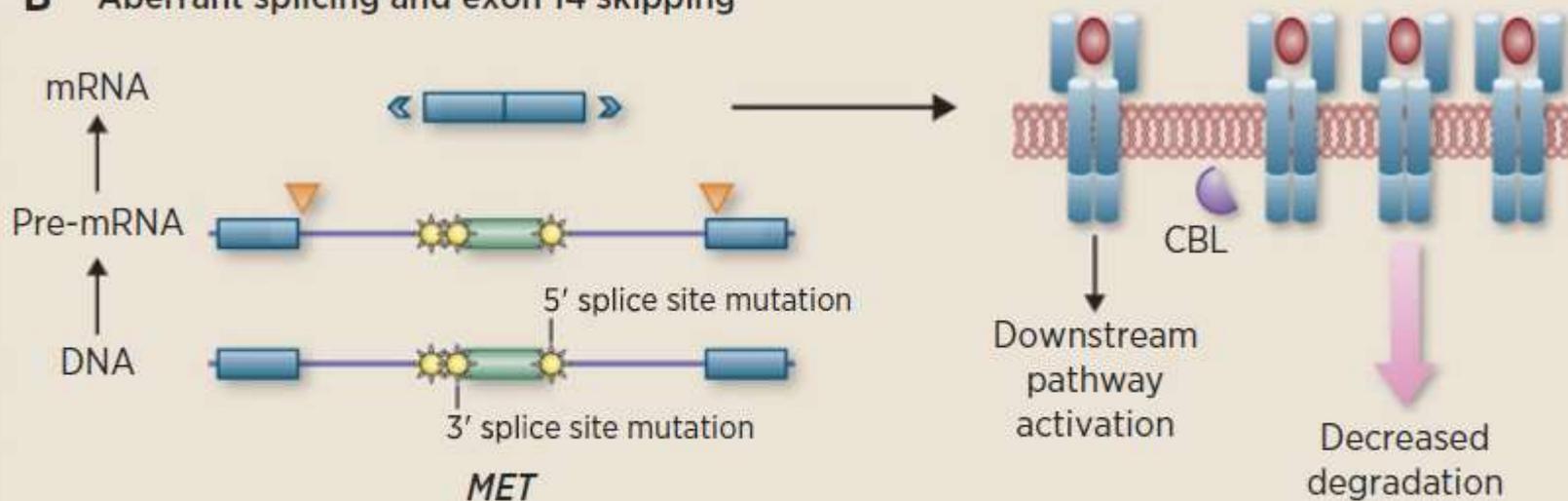




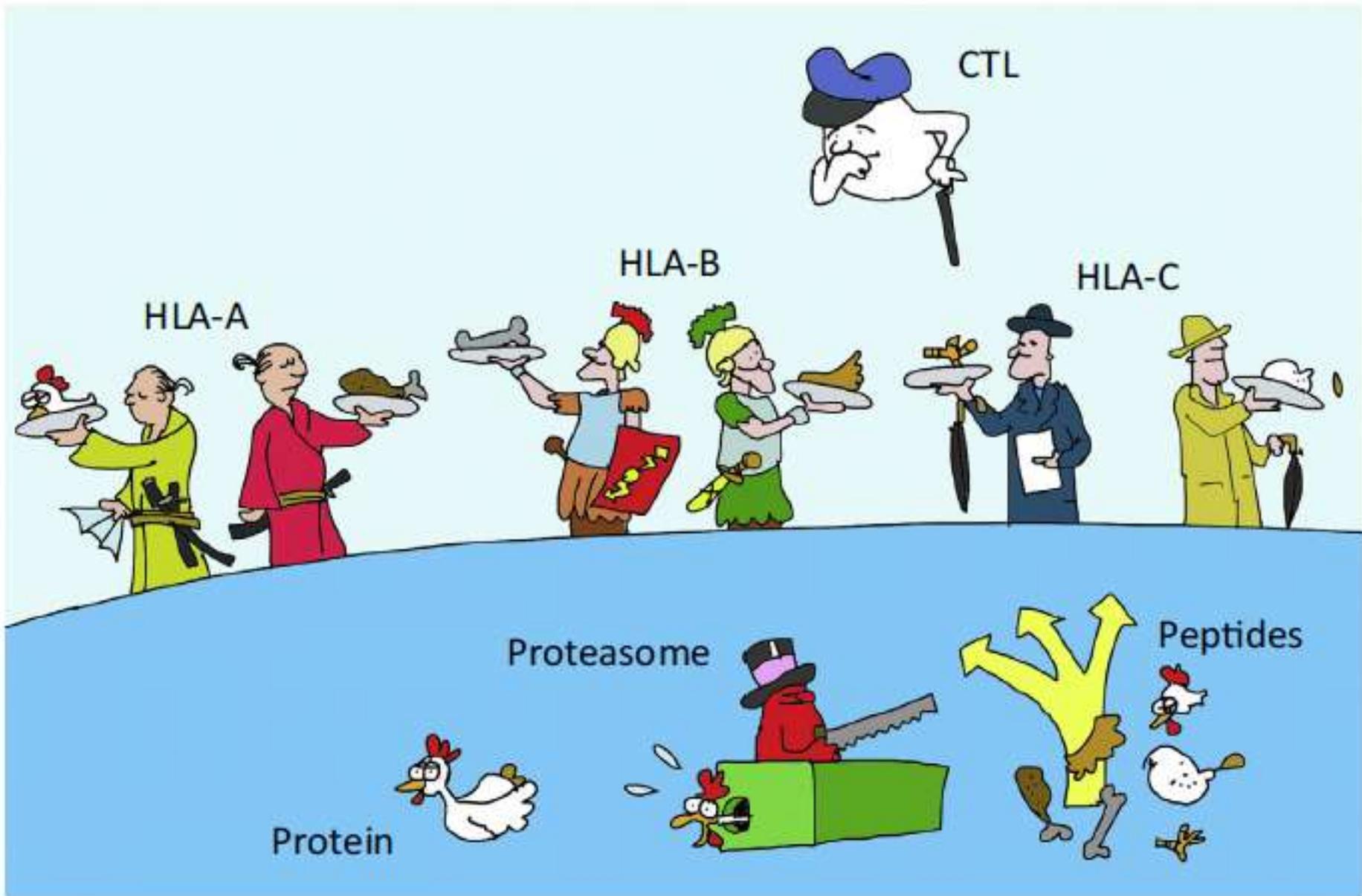
### A Normal splicing



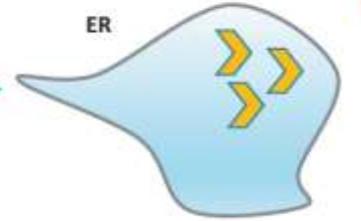
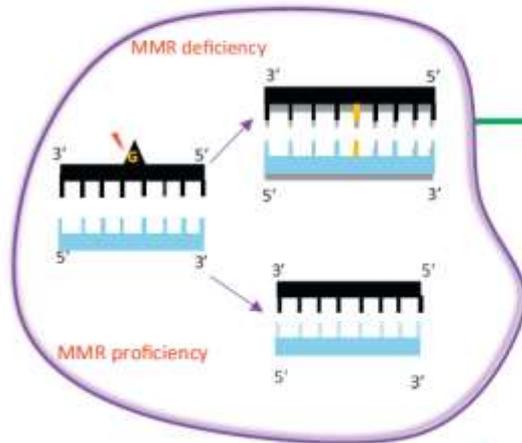
### B Aberrant splicing and exon 14 skipping



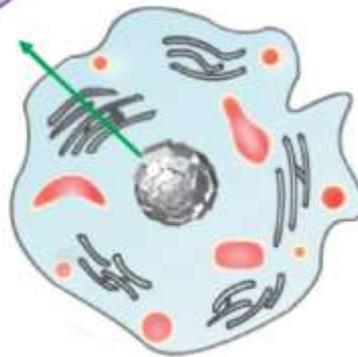
© 2016 American Association for Cancer Research



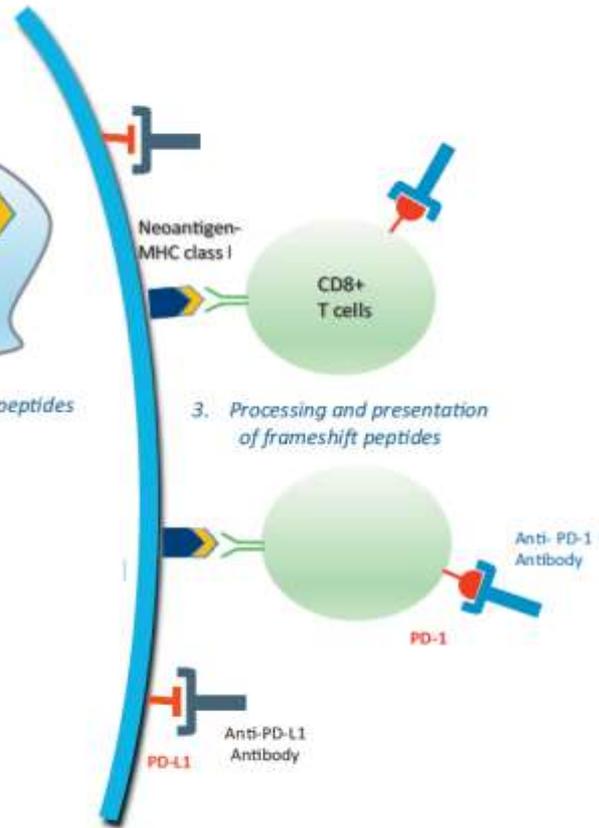
1. Insertion mutation in coding microsatellites leading to frameshift mutation



2. Translation of frameshift peptides



3. Processing and presentation of frameshift peptides

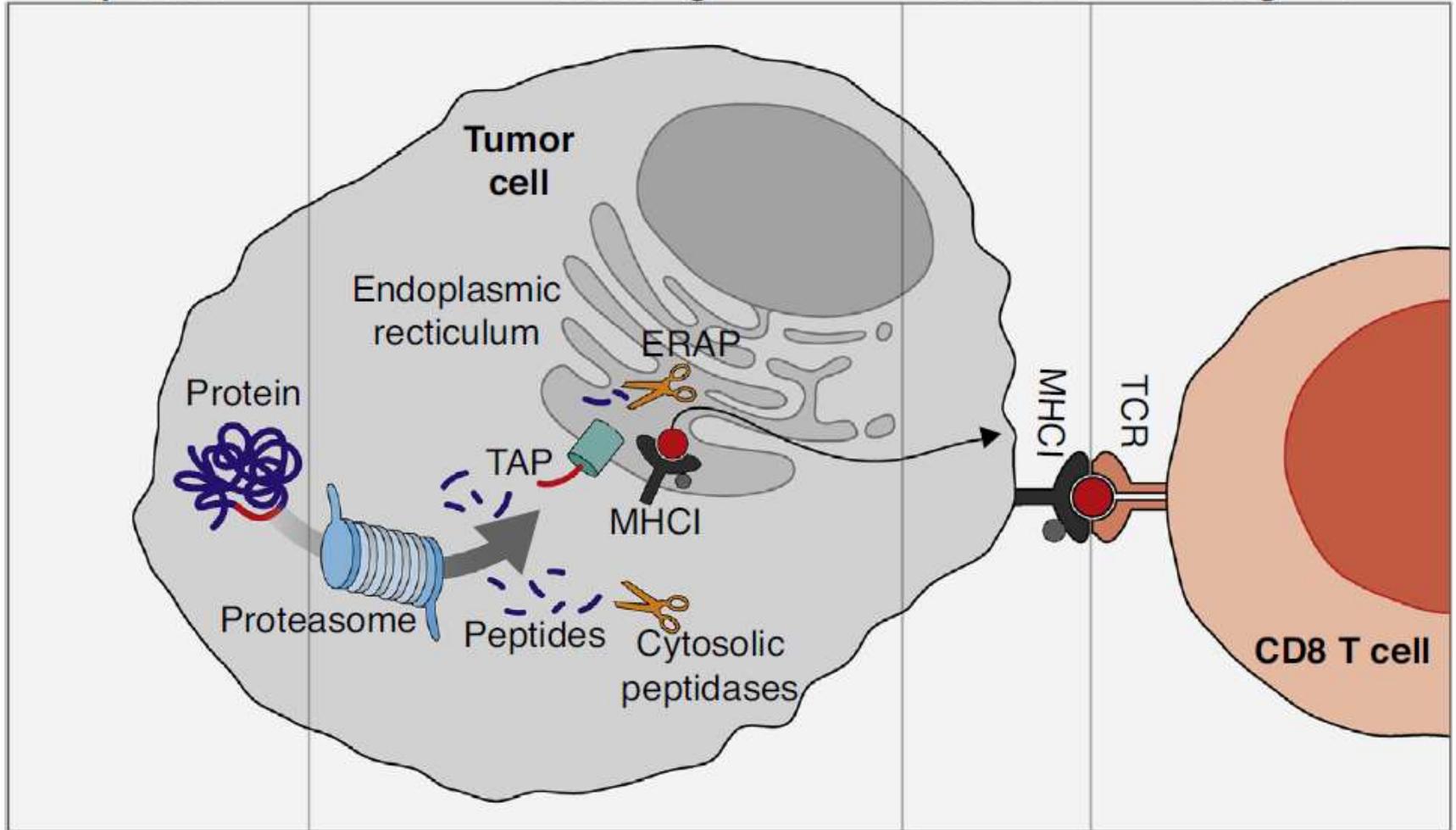


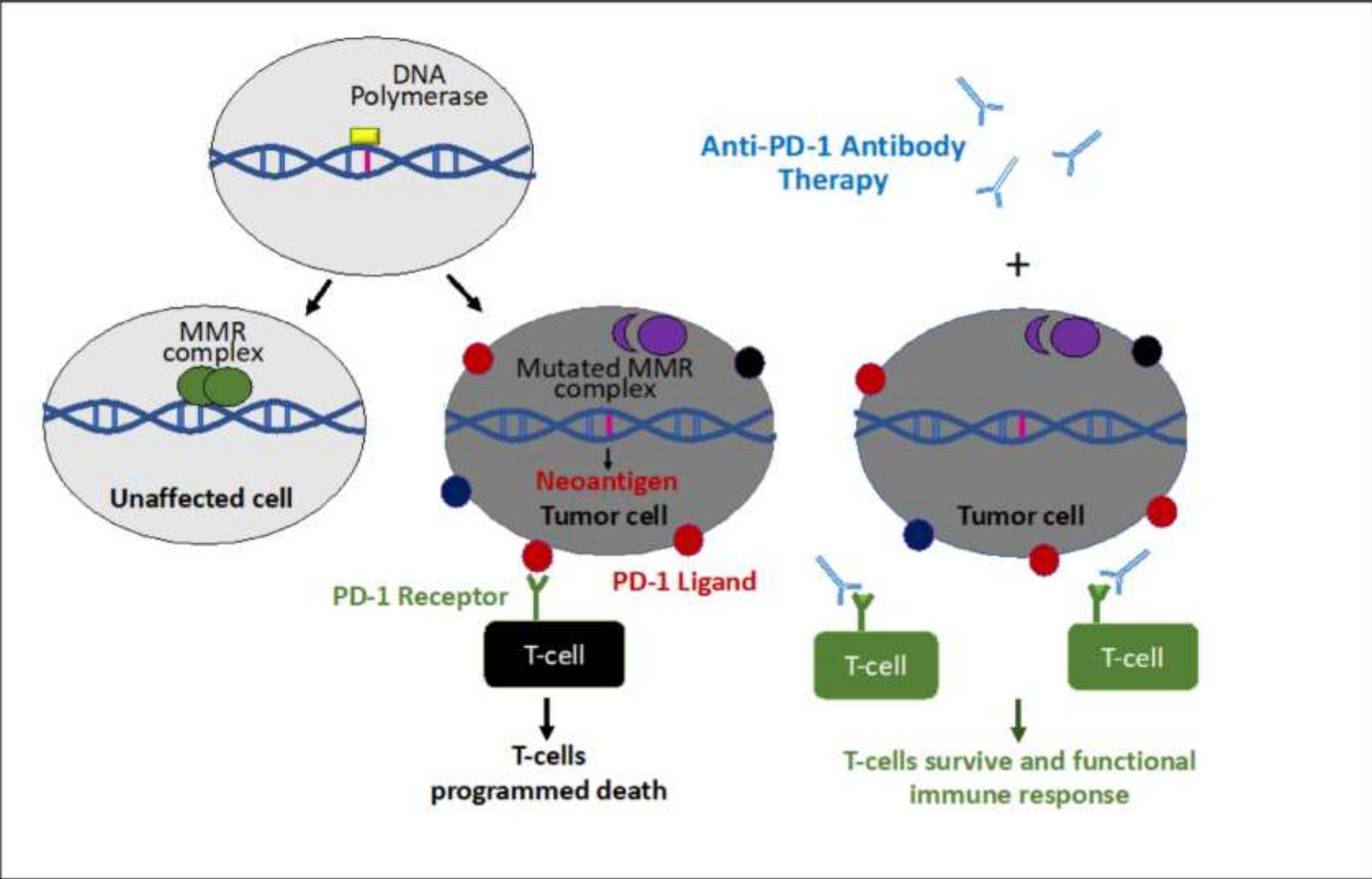
**1. Antigen Expression**

**2. Antigen Processing**

**3. Antigen Presentation**

**4. TCR Recognition**





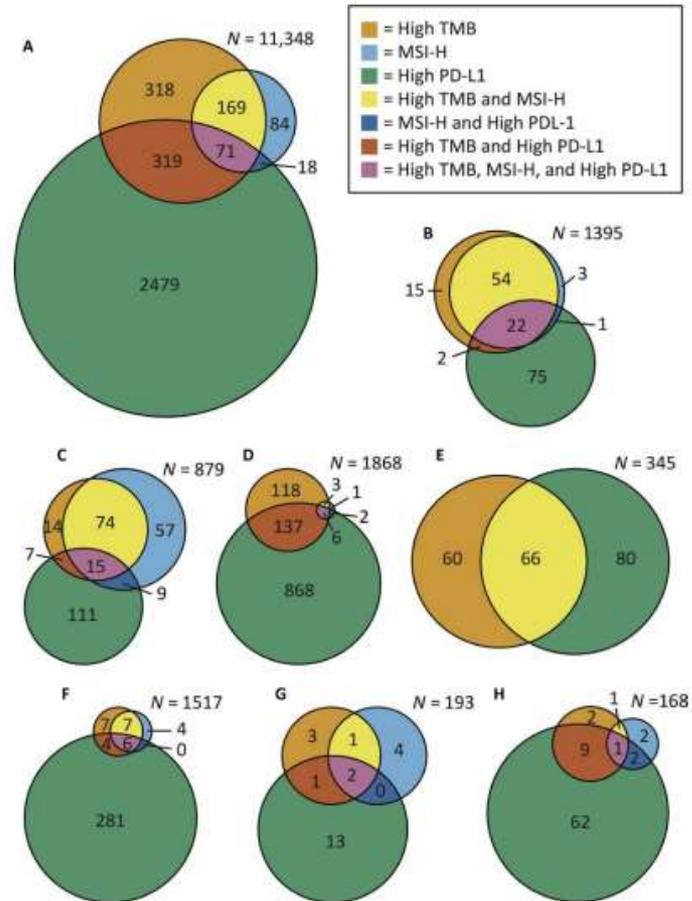
# *Tumor Mutational Burden (TMB) or Tumor Mutation Load (TML)*

---

TMB or TML: total number of somatic/acquired mutations per coding area of a tumor genome (Mut/Mb)



The number of mutations can vary across different tumor types.



# THANK YOU!!



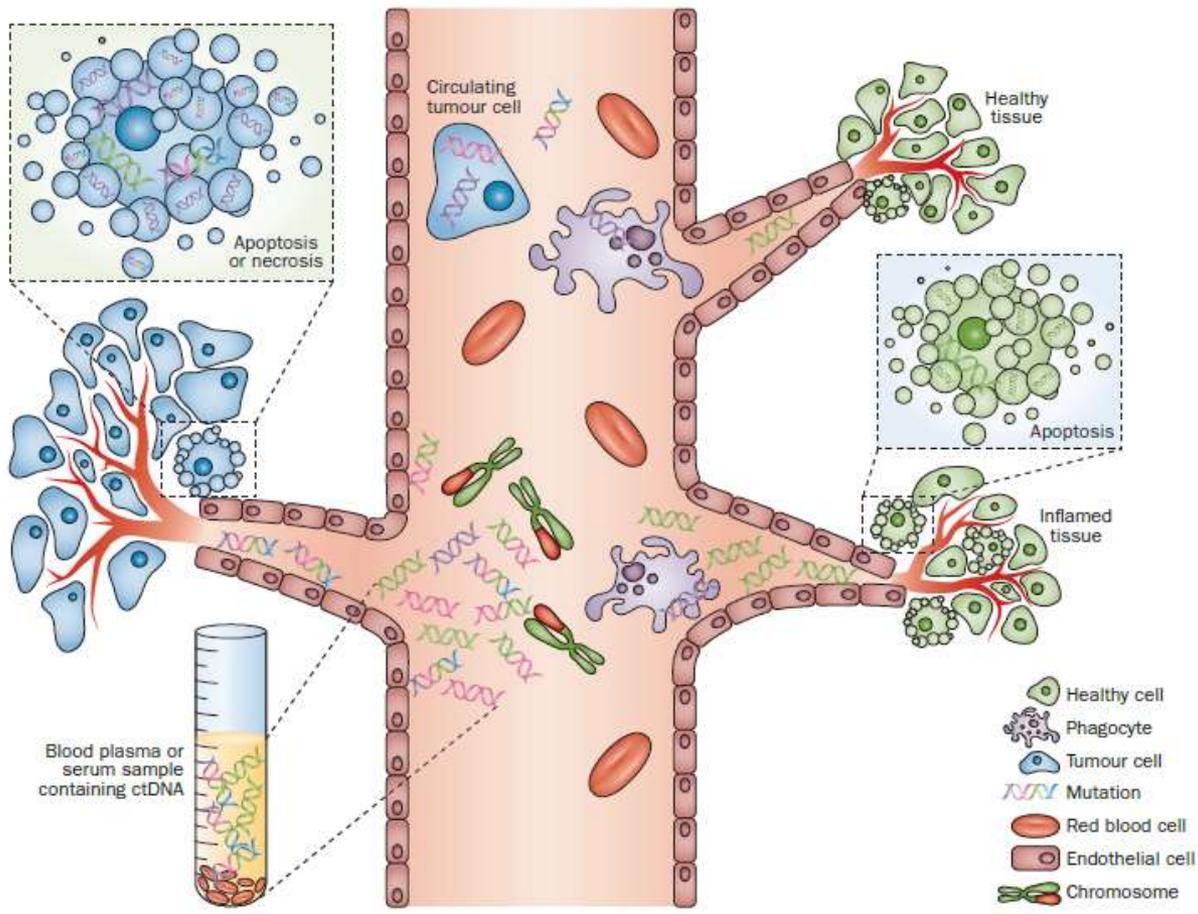
PP-XLK-NLD-0018



# Liquid biopsy: an introduction.

Prof. Dr. P. Pauwels  
(UZA, UA)





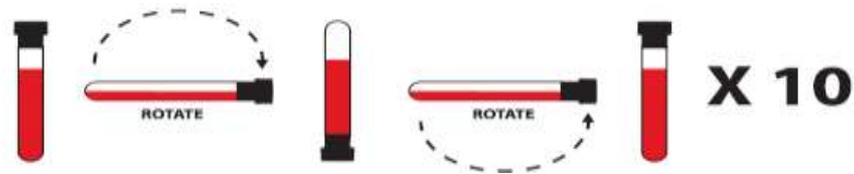
Crowley, E. et al. *Nat. Rev. Clin. Oncol.* **10**, 472–484 (2013)

# Liquid Biopsy - practical

Follow up NSCLC patients – # time points

**1. Obtaining a blood sample (10 ml – STRECK)**

→ Blood: immediately 8 – 10 inversions

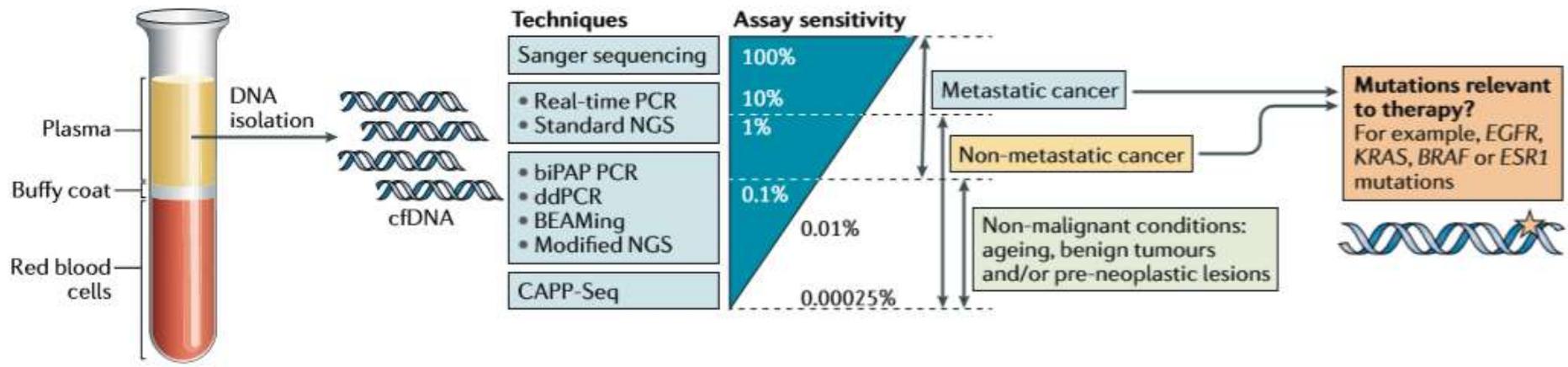


Storage & transport at ROOM TEMPERATURE (not in refrigerator)

**2. cfDNA extraction ( the MaxWell® ccfDNA Plasma Kit (Promega))**

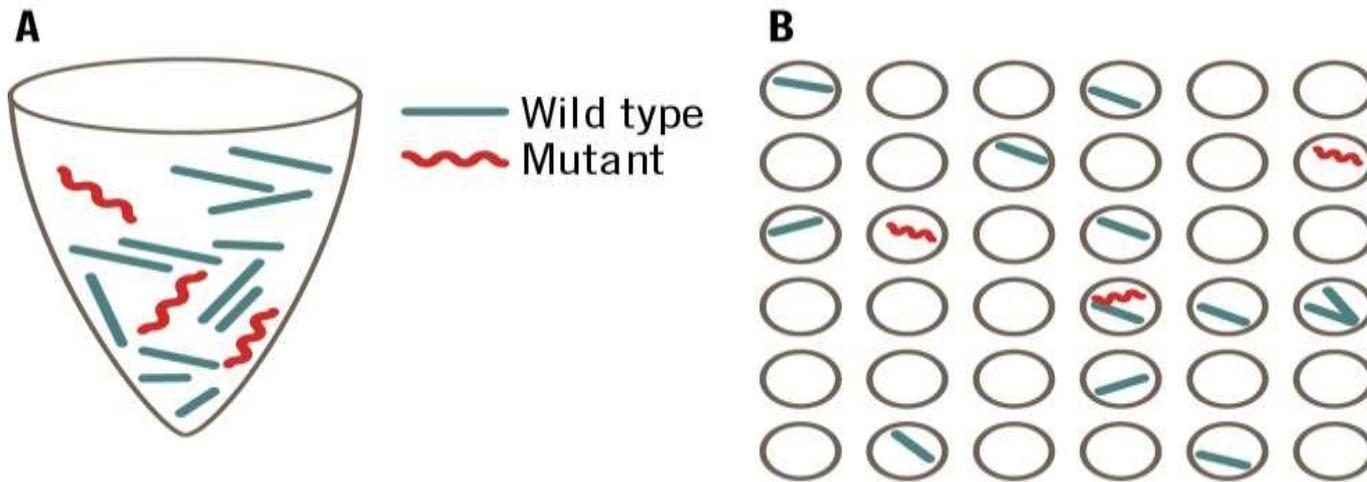
**3. ctDNA detection by digital droplet PCR (ddPCR)**





# Liquid Biopsy: ctDNA

## Digital Droplet PCR (ddPCR)



### Advantages ddPCR

- High sensitivity
- Multiplexing capacity
- Absolute quantification

- Detection: gene amplification, deletion, translocations & gene mutations



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Contents lists available at [ScienceDirect](#)

## Lung Cancer

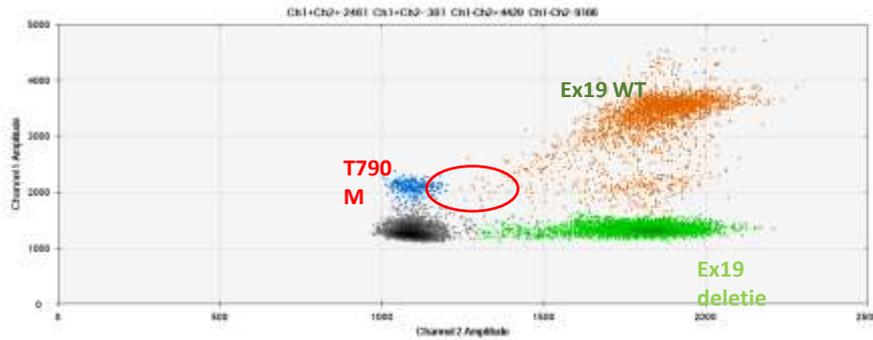
journal homepage: [www.elsevier.com/locate/lungcan](http://www.elsevier.com/locate/lungcan)



### Circulating cell-free nucleic acids and platelets as a liquid biopsy in the provision of personalized therapy for lung cancer patients

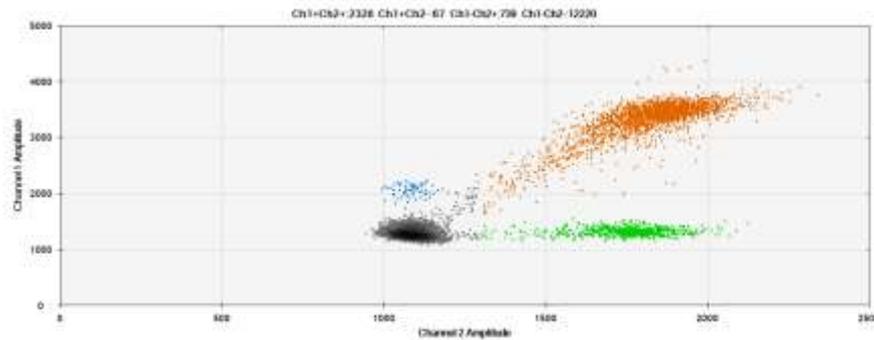
L. Sorber<sup>a,b,\*</sup>, K. Zwaenepoel<sup>a,b</sup>, V. Deschoolmeester<sup>a,b</sup>, P.E.Y. Van Schil<sup>c</sup>,  
J. Van Meerbeeck<sup>a,d</sup>, F. Lardon<sup>a</sup>, C. Rolfo<sup>e</sup>, P. Pauwels<sup>a,b</sup>

## Patiënt: NSCLC - EGFR exon19 deletie + T790M mutatie



**11/02/2015**

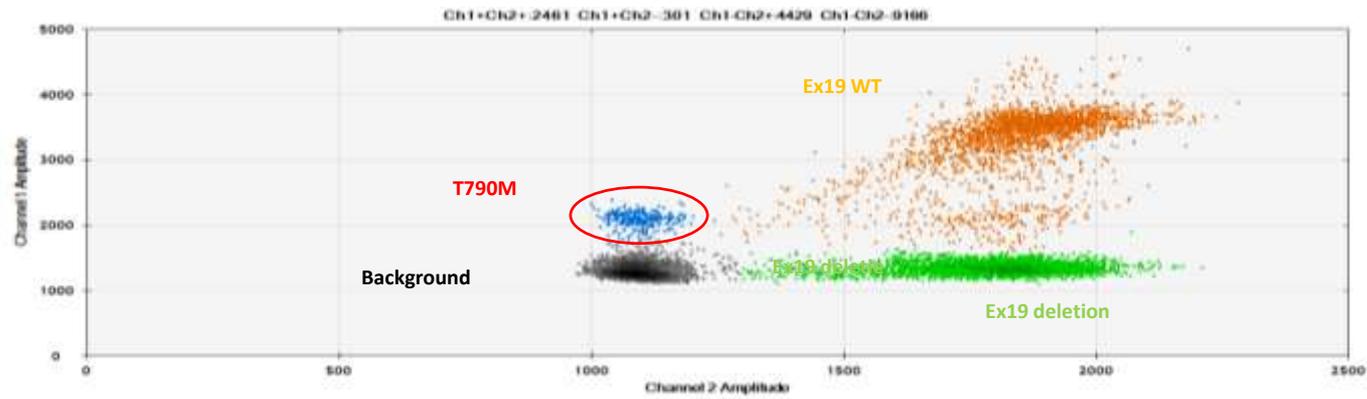
- Geen behandeling
- Ex19 del: 70% - T790M: 3.59%



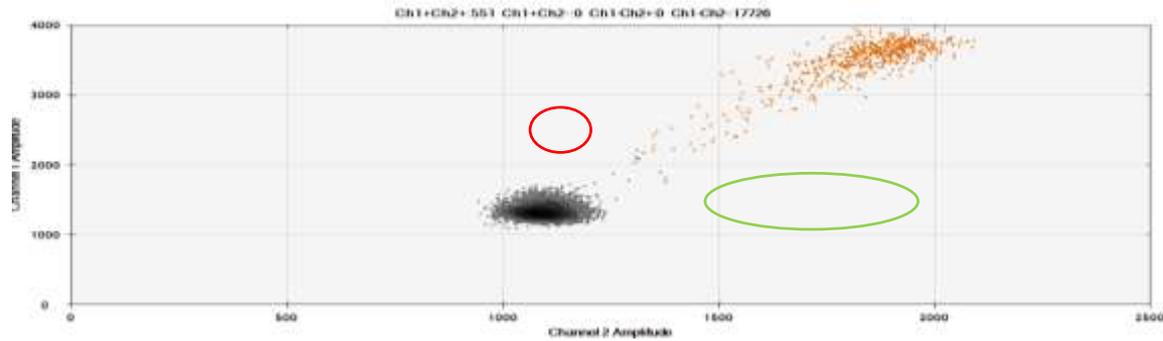
**12/03/2015**

- Behandeling AZD9291  
(start 06/03/2015)
- Ex19 del: 26.2% - T790M: 1.96%

# Patient with EGFR exon 19 del – T790M



Treatment with AZD9291

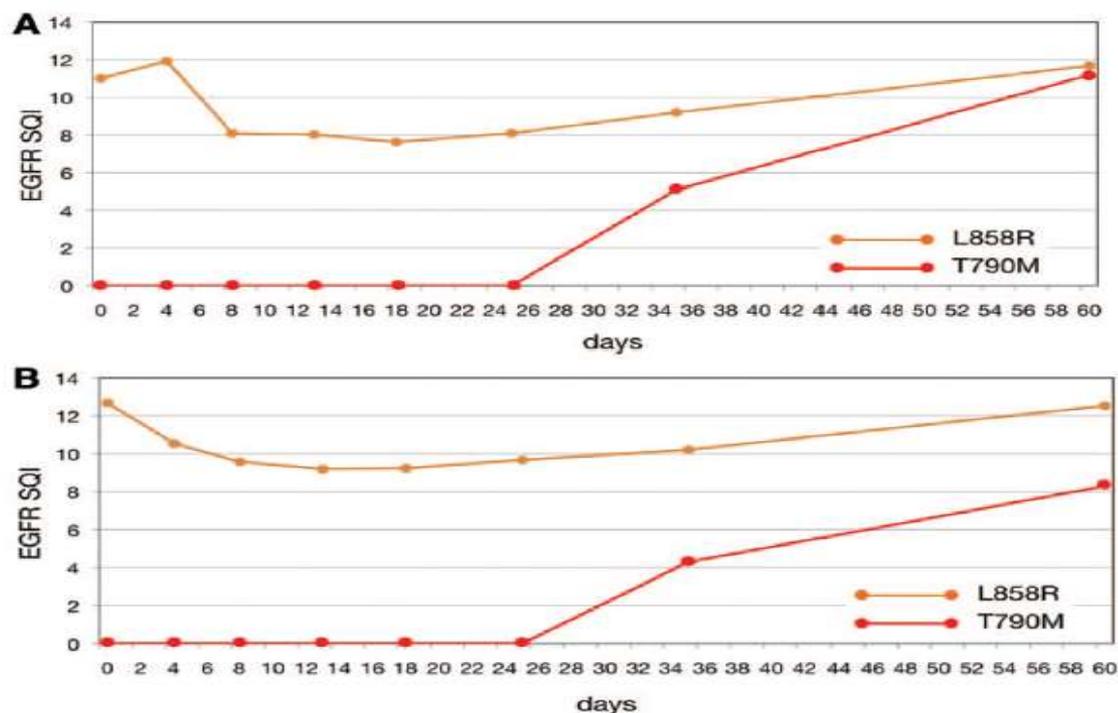


# Early Prediction of Response to Tyrosine Kinase Inhibitors by Quantification of *EGFR* Mutations in Plasma of NSCLC Patients

*Antonio Marchetti, MD, PhD,\* John F. Palma, PhD,† Lara Felicioni, PhD,‡ Tommaso M. De Pas, MD,§ Rita Chiari, MD,|| Maela Del Grammastro, PhD,\* Giampaolo Filice, PhD,\* Vienna Ludovini, PhD,|| Alba A. Brandes, MD,¶ Antonio Chella, MD,# Francesco Malorgio, MD,\*\* Flavio Guglielmi, MD,†† Michele De Tursi, MD,‡‡ Armando Santoro, MD,§§ Lucio Crinò, MD,||| and Fiamma Buttitta, MD, PhD‡*

*(J Thorac Oncol. 2015;10: 1437–1443)*





**FIGURE 3.** Quantification of mutated *EGFR* DNA from plasma of two slow responders with T790M mutation by the PCR test. The figures show the failure to clear the initial *EGFR* mutated DNA (L858R) and the emergence of T790M DNA during week 3 after initiating *EGFR* TKI treatment. PCR, polymerase chain reaction; SQI, semi-quantitative index; TKI, tyrosine kinase inhibitor.

an early increase in the circulating levels of the T790M mutation was observed. No T790M mutations were seen in serial plasma samples of the rapid responders. We therefore speculate that slow responders are more prone to develop early resistance. However, further clinical validation is required to assess the long-term impact of TKI treatment on rapid versus slow responders relative to progression-free and overall survival.

REVIEW

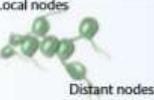
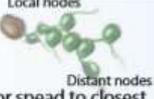
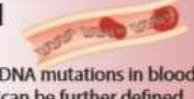
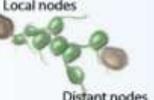
## Incorporating blood-based liquid biopsy information into cancer staging: time for a TNMB system?

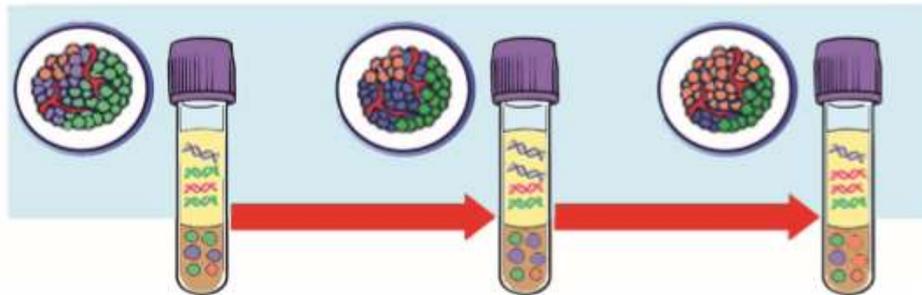
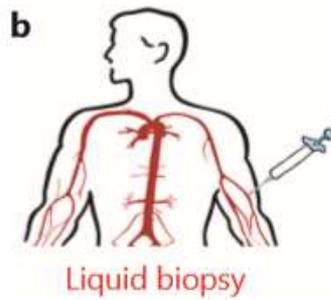
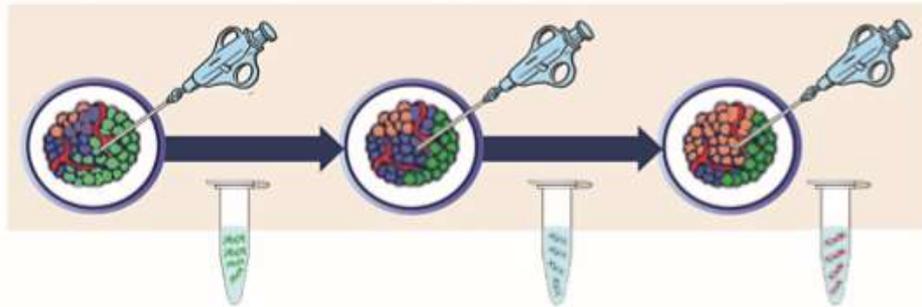
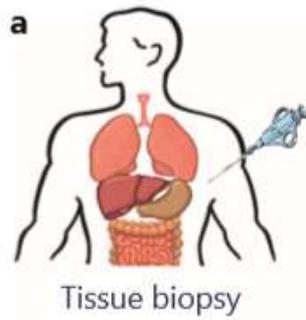
M. Yang<sup>1,2,3,4</sup>, M. E. Forbes<sup>1,2</sup>, R. L. Bitting<sup>1,5</sup>, S. S. O'Neill<sup>1,6</sup>, P.-C. Chou<sup>1,2</sup>, U. Topaloglu<sup>1,2</sup>, L. D. Miller<sup>1,2</sup>, G. A. Hawkins<sup>1,7</sup>, S. C. Grant<sup>1,5</sup>, B. R. DeYoung<sup>1,6</sup>, W. J. Petty<sup>1,5</sup>, K. Chen<sup>3,4\*</sup>, B. C. Pasche<sup>1,2,5</sup> & W. Zhang<sup>1,2\*</sup>

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Prof. Kexin Chen, Department of Epidemiology and Biostatistics, Tianjin Medical University Cancer Institute and Hospital, Huanhu Xi Road, Tiyuan Bei, Hexi District, Tianjin 300060, China. Tel: +86-22-2337-2231; E-mail: chenkesin@tjmuch.com

<p><b>T</b></p> <p><b>Tumor Size</b></p>	<p><b>N</b></p> <p><b>Lymph Node</b></p>	<p><b>M</b></p> <p><b>Metastasis</b></p>	<p><b>B</b></p> <p><b>Blood</b></p>
<p><b>T1</b></p>  <p>Tumor size/local invasion</p>	<p><b>N0</b></p>  <p>No regional lymph node invasion</p>	<p><b>M0</b></p>  <p>No distant metastasis</p>	<p><b>B0</b></p>  <p>No ctDNA mutations in blood</p>
<p><b>T2</b></p>  <p>Tumor size/local invasion</p>	<p><b>N1</b></p>  <p>Tumor spread to closest or small number of regional lymph nodes</p>	<p><b>M1</b></p>  <p>Distant metastasis</p>	<p><b>B1</b></p>  <p>ctDNA mutations in blood (can be further defined with more detailed quantification in the future)</p>
<p><b>T3</b></p>  <p>Tumor size/local invasion</p>	<p><b>N2</b></p>  <p>Tumor spread to an extent between N1 and N3</p>		
<p><b>T4</b></p>  <p>Tumor of any size that invades to other organs</p>	<p><b>N3</b></p>  <p>Tumor spread to more distant or regional numerous lymph nodes</p>		





*Review*

# Circulating Cell-Free DNA and Colorectal Cancer: A Systematic Review

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<sup>3</sup> Biomedical Centre, Faculty of Medicine in Pilsen, Charles University in Prague, 323 00 Pilsen, Czech Republic

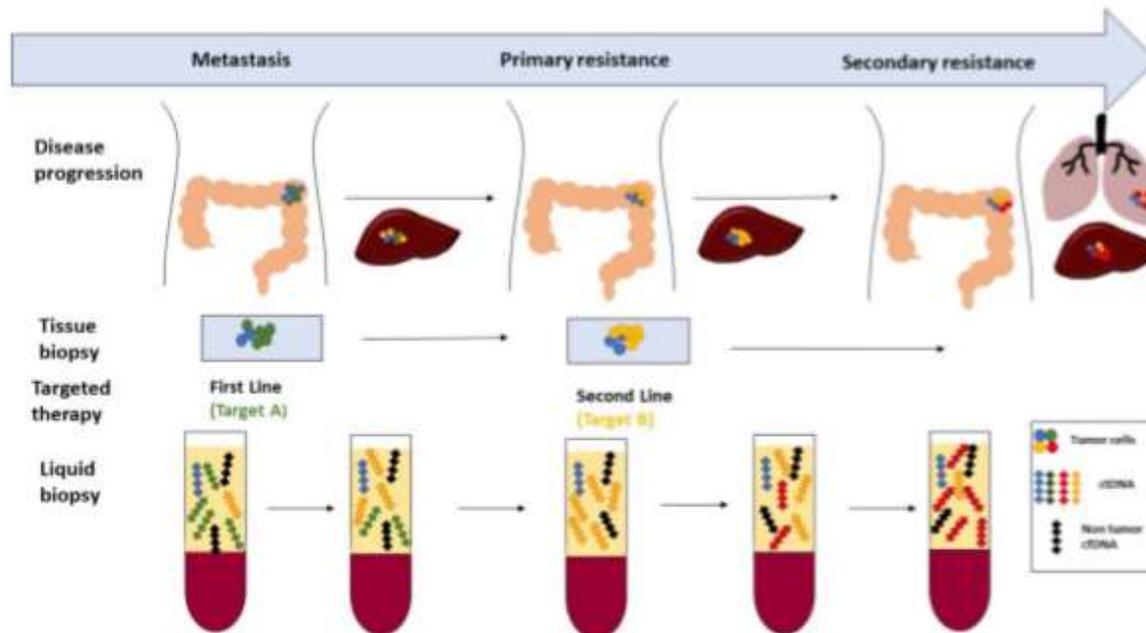
\* Correspondence: veronika.vymetalkova@iem.cas.cz

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## 2.2. Cell-Free DNA and Colorectal Cancer

Analysis of ctDNA is a promising new tool in oncology. ctDNA mutational content can provide invaluable information on the genetic background of a tumor, and assist oncologist in deciding on therapy, or in following the residual disease (Figure 2, Table 3).







**Thank  
You!!!**