34th SITC
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Gaylord National Hotel & Convention Center in National Harbor, Maryland, USA

Immune Contexture and Immune Escape (Checkpoints) at Pre-Cancer Lesion Stages

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Disclosures

Co-founder and chairman of the scientific advisory board:

- *HalioDx*

Collaborative Research Agreement (grants) :

- *Perkin-Elmer, IObiotech, MedImmune, Janssen, Imcheck Therapeutics*

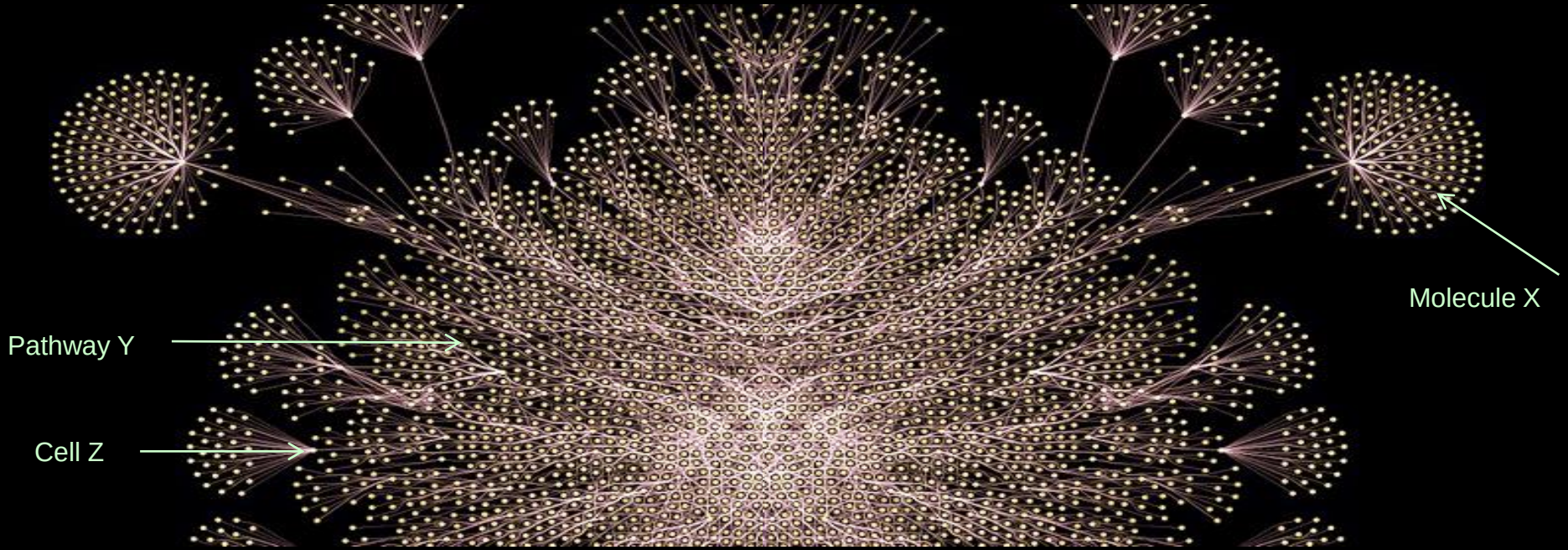
Participation to Scientific Advisory Boards:

- *BMS, MedImmune, Astra Zeneca, Novartis, Definiens, Merck Serono, IObiotech, ImmunID, Nanostring, Illumina, Northwest Biotherapeutics, Actelion, Amgen, Merck MSD*

Consultant :

- *BMS, Roche, GSK, Compugen, Mologen, Gilead, Sanofi*

Cancer is one of the most complex biological system of all



"*The whole is greater than the sum of its parts*", Aristotle

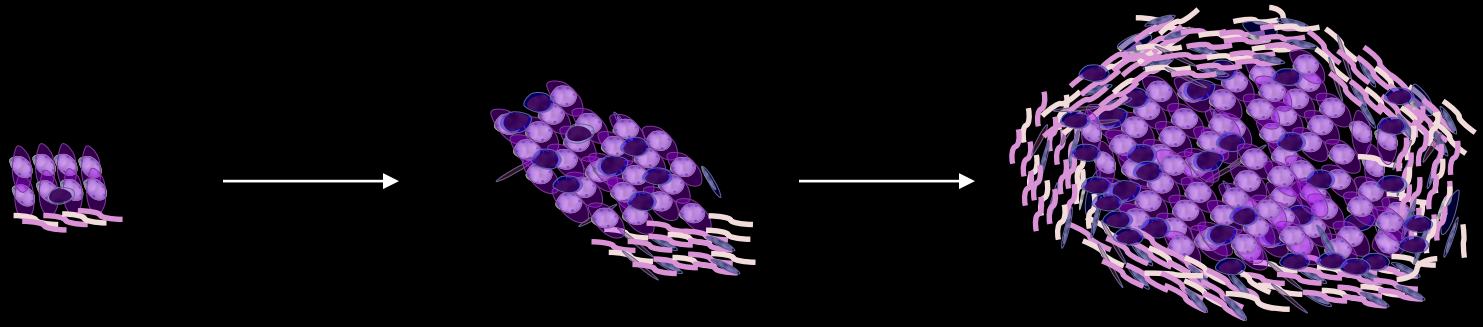
-> Systems biology in human cancer

The continuum of cancer immunosurveillance

Pre-cancerous lesions

Primary Carcinoma

Metastasis



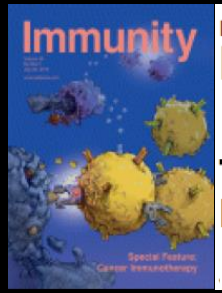
Mascaux C. … Galon J.
Nature 2019

Pagès F. … Galon J.
Lancet 2018

Phase 3 trial
Galon J. et al. Unpublished

Angelova M. … Galon J.
Cell 2018

The continuum of cancer immunosurveillance

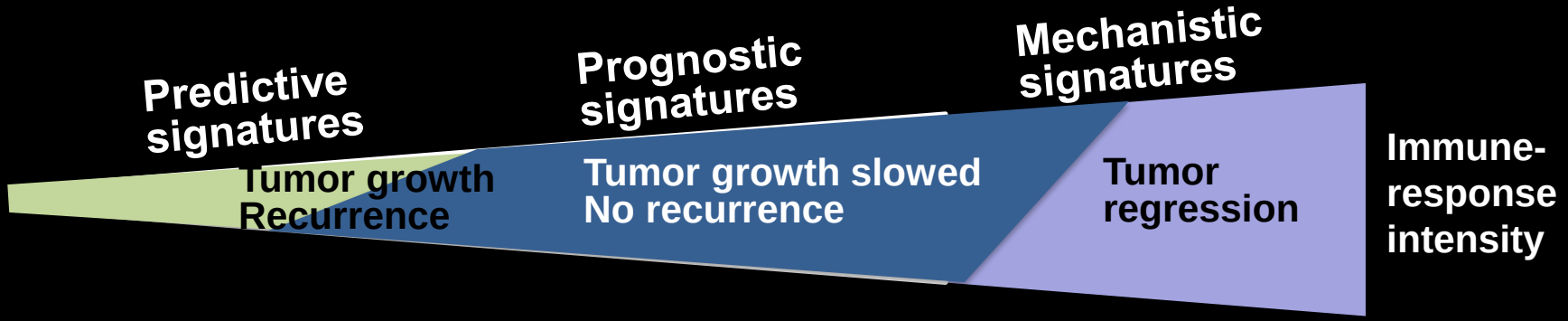


Immunity
Review

Cell PRESS

The Continuum of Cancer Immunosurveillance: Prognostic, Predictive, and Mechanistic Signatures

Jérôme Galon,[1,2,3,*] Helen K. Angell,[1,2,3] Davide Bedognetti,[4] and Francesco M. Marincola[4,5,*]



Galon J et al. ***Immunity*** 2013

What is the importance of the pre-existing immunity within tumors Does it matter

A Novel Paradigm for Cancer

Science
AAAS

Type, Density, and Location of Immune Cells Within Human Colorectal Tumors Predict Clinical Outcome

Jérôme Galon,[1*†] Anne Costes,[1] Fatima Sanchez-Cabo,[2] Amos Kirilovsky,[1] Bernhard Mlecnik,[2] Christine Lagorce-Pagès,[3] Marie Tosolini,[1] Matthieu Camus,[1] Anne Berger,[4] Philippe Wind,[4] Franck Zinzindohoué,[5] Patrick Bruneval,[6] Paul-Henri Cugnenc,[5] Zlatko Trajanoski,[2] Wolf-Herman Fridman,[1,7] Franck Pagès[1,7†]

29 SEPTEMBER 2006 VOL 313 SCIENCE www.sciencemag.org

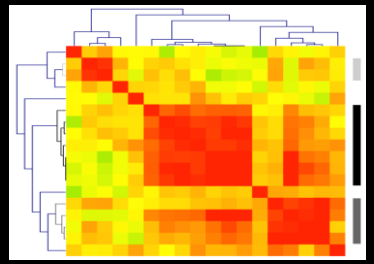
The foundation a new concept

↓

Immune contexture

- ✓ Gene expression profiling
- ✓ Qualitative immune signature

- ✓ Immunohistochemistry (IHC)
- ✓ Digital Pathology
- ✓ Quantitative immune cell infiltration



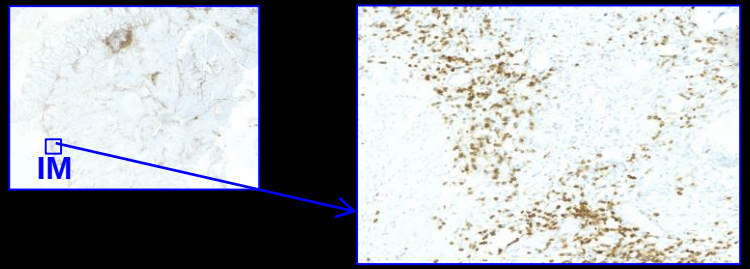
Inflammation

Adaptive immunity

Immune suppression

Survival

Quality

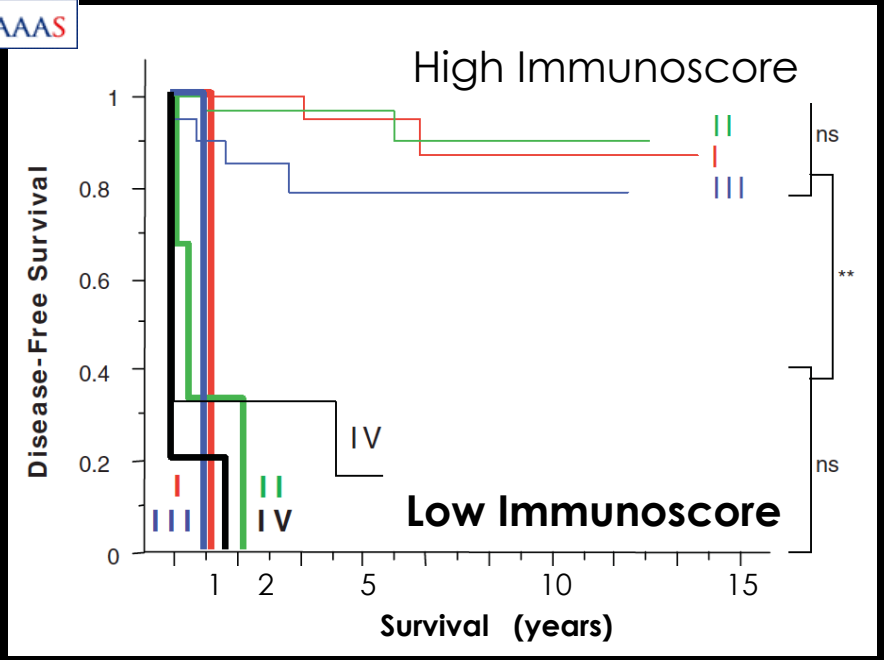


Type/Density/Location

Optimized Immunosign

Galon J et al. ***Science*** 2006

Immunoscore: a novel paradigm for cancer



- ✓ High Immunoscore
- ✓ Inflammed tumors
- ✓ Strong pre-existing adaptive immunity

- ✓ Low Immunoscore
- ✓ Non-Inflammed tumors
- ✓ Weak/absent pre-existing adaptive immunity

Coordinated adaptive immune reaction (Immunoscore) more than tumor invasion predicts clinical outcome

Galon et al. ***Science*** 2006

A Novel Paradigm for Cancer

Multivariate Cox Analysis

Parameters	*HR*	*P* value
• T-stage	1.2	0.25
• N-stage	1.4	0.15
• Differentiation	1.1	0.84
• **Immunoscore**	1.9	0.00001

"***Immune Contexture***" :

Cells -> ✓**Type**

Quantity -> ✓**Density**

Spatial -> ✓**Location**

-> Immunoscore

Quality -> ✓Immune **functional** orientation

-> Immunosign

Galon J et al. ***Science*** 2006

Implications for cancer classification and therapies ?

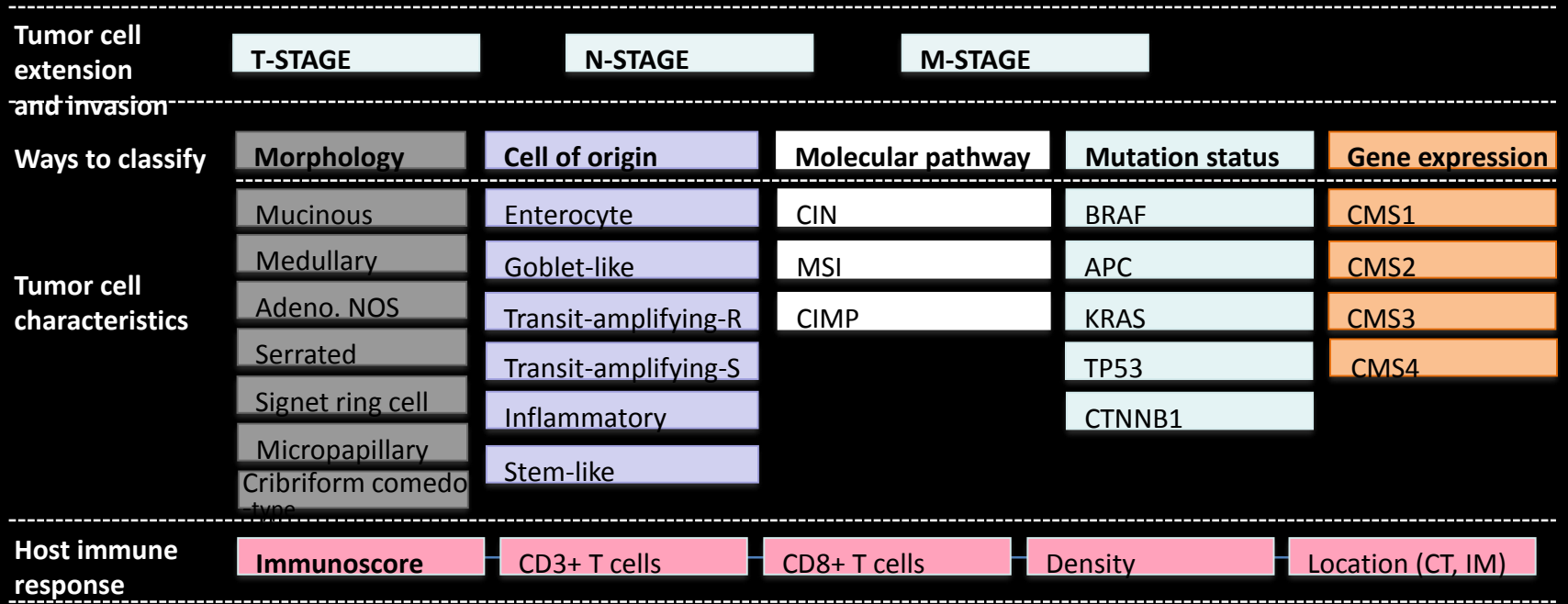
From the **Immune contexture**

(Complexity of intratumor immune reaction)

To the **Immunoscore**

(A simple and powerfull Immune Test)

Colorectal cancer classifications



Galon et al. ***J Pathol***. 2014

The Immunoscore as a New Possible Approach for the Classification of Cancer

sitc

World Immunotherapy Council inaugural meeting (Feb 2012)

Support (moral) from the World Immunotherapy Council (WIC), and support from societies including, EATI, BDA, CCIC, CIC, CRI, CIMT, CSCO, TIBT, DTIWP, ESCII, NIBIT, JACI, NCV-network, PIVAC, ATTACK, TVACT…

Worldwide Immunoscore consortium (PI: J Galon)

(17 countries: >3000 Stage I/II/III Colon cancer patients)

Assay harmonization

Immunoscore meetings :

- Feb 2012, Italy
- Dec 2012, Italy
- Nov 2013, SITC, USA
- Dec 2013, Italy
- Jan 2014, Qatar
- Jul 2014, Paris, France
- Nov 2014, SITC, USA
- Nov 2015, SITC, USA
- Dec 2015, Italy
- Feb 2016, USCAP, USA
- April 2016, USA
- Nov 2016, SITC, USA
- Dec 2016, Italy
- Feb 2017, USCAP, USA
- Dec 2017, Italy

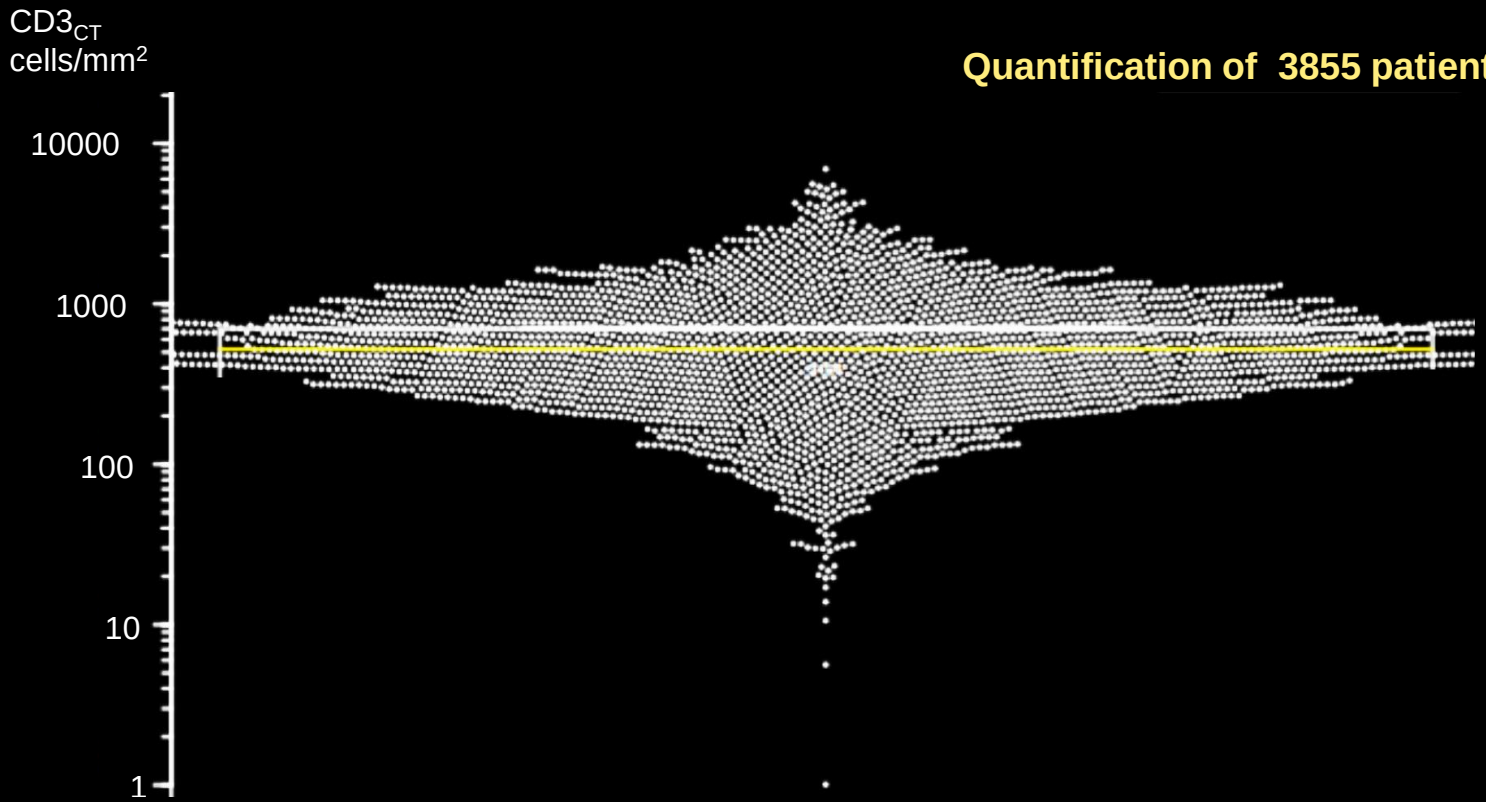
THE LANCET

International validation of the consensus Immunoscore for the classification of colon cancer: a prognostic and accuracy study

Franck Pagès, Bernhard Mlecnik, Florence Marliot, Gabriela Bindea, Fang-Shu Ou, Carlo Bifulco, Alessandro Lugli, Inti Zlobec, Tilman T Rau, Martin D Berger, Iris D Nagtegaal, Elisa Vink-Börger, Arndt Hartmann, Carol Geppert, Julie Kolwelter, Susanne Merkel, Robert Grützmann, Marc Van den Eynde, Anne Jouret-Mourin, Alex Kartheuser, Daniel Léonard, Christophe Remue, Julia Y Wang, P Bavi, Michael H A Roehrl, Pamela S Ohashi, Linh T Nguyen, SeongJun Han, Heather L MacGregor, Sara Hafezi-Bakhtiari, Bradly G Wouters, Giuseppe V Masucci, Emilia K Andersson, Eva Zavadova, Michal Vocka, Jan Spacek, Lubos Petruzelka, Bohuslav Konopasek, Pavel Dundr, Helena Skalova, Kristyna Nemejcova, Gerardo Botti, Fabiana Tatangelo, Paolo Delrio, Gennaro Ciliberto, Michele Maio, Luigi Laghi, Fabio Grizzi, Tessa Fredriksen, Bénédicte Buttard, Mihaela Angelova, Angela Vasaturo, Pauline Maby, Sarah E Church, Helen K Angell, Lucie Lafontaine, Daniela Bruni, Carine El Sissy, Nacilla Haicheur, Amos Kirilovsky, Anne Berger, Christine Lagorce, Jeffrey P Meyers, Christopher Paustian, Zipei Feng, Carmen Ballesteros-Merino, Jeroen Dijkstra, Carlijn van de Water, Shannon van Lent-van Vliet, Nikki Knijn, Ana-Maria Mușină, Dragos-Viorel Scripcariu, Boryana Popivanova, Mingli Xu, Tomonobu Fujita, Shoichi Hazama, Nobuaki Suzuki, Hiroaki Nagano, Kiyotaka Okuno, Toshihiko Torigoe, Noriyuki Sato, Tomohisa Furuhata, Ichiro Takemasa, Kyogo Itoh, Prabhu S Patel, Hemangini H Vora, Birva Shah, Jayendrakumar B Patel, Kruti N Rajvik, Shashank J Pandya, Shilin N Shukla, Yili Wang, Guanjun Zhang, Yutaka Kawakami, Francesco M Marincola, Paolo A Ascierto, Daniel J Sargent, Bernard A Fox, Jérôme Galon*

Pages et al. *The Lancet* 2018

Densities of $CD3_{CT}$ (cells/mm^2) within tumors



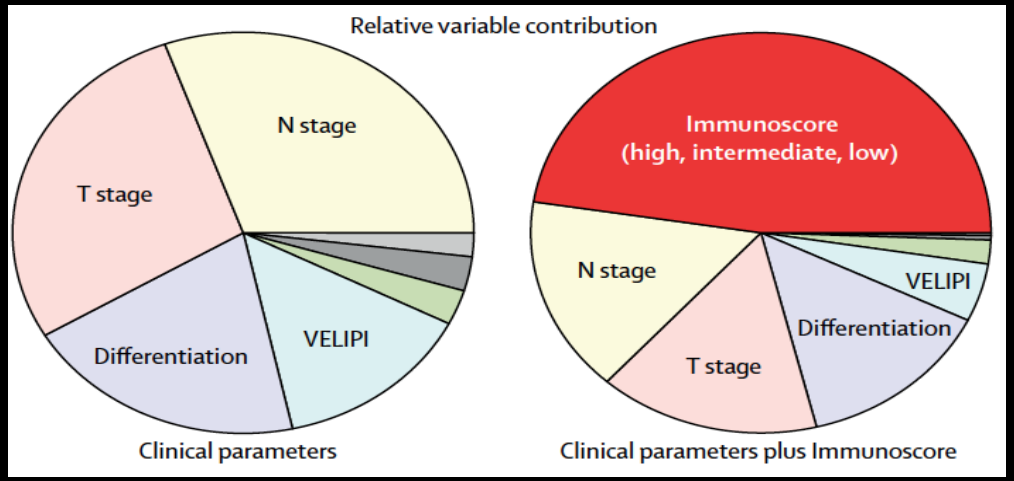
Quantification of 3855 patients

- ✓ Whole slide quantification within the CT region
- ✓ Similar quantification were performed for $CD3_{CT}$, $CD3_{IM}$, $CD8_{CT}$, $CD8_{IM}$

The Lancet 2018

Relative variable contribution to risk

Chi squared proportion (χ^2) test for clinical parameters



Multivariate Analysis for Time to Recurrence (TTR)

All patients

Immunoscore	**P-values**	***c-index***
2 groups	<0.0001	0.73 (0.66-0.80)
3 groups	<0.0001	0.73 (0.67-0.80)
5 groups	<0.0001	0.73 (0.67-0.80)

The Lancet 2018

International validation of the consensus Immunoscore for the classification of colon cancer:

Strong arguments for introducing a “I” for Immune into the classification of cancer: TNM-I

Immunoscore in locally advanced colon cancer

Stage III

Immunoscore Predictve value :

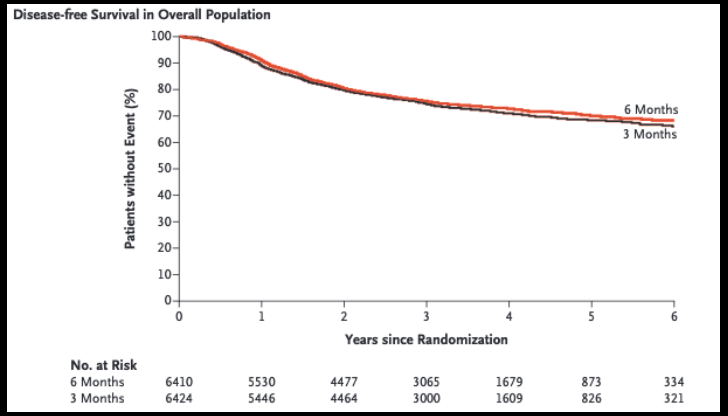
✓ IDEA, France, phase 3 clinical trial (3 months *vs* 6 months chemotherapy)

The NEW ENGLAND JOURNAL of MEDICINE

ESTABLISHED IN 1812 MARCH 29, 2018 VOL. 378 NO. 13

Duration of Adjuvant Chemotherapy for Stage III Colon Cancer

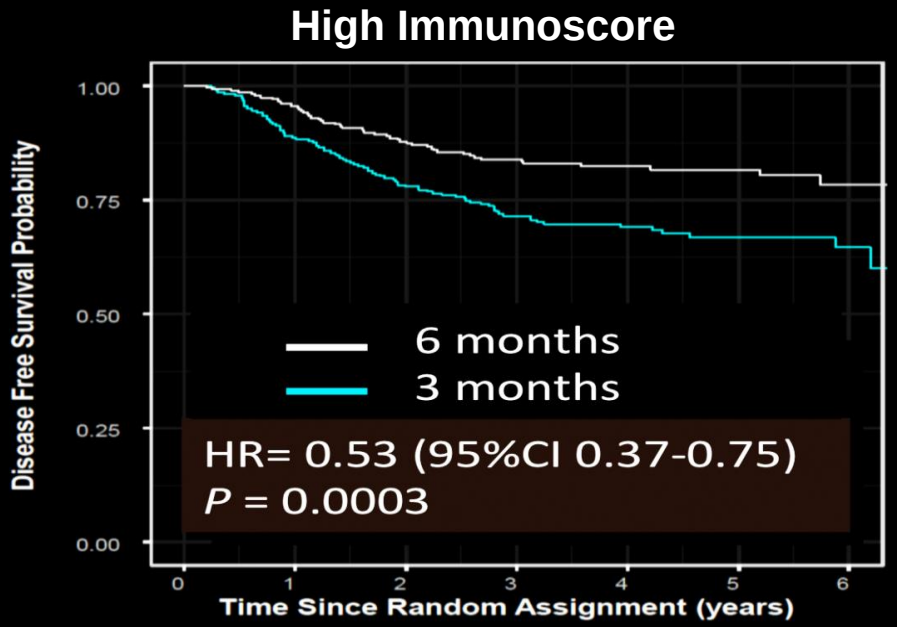
A. Grothey, A.F. Sobrero, A.F. Shields, T. Yoshino, J. Paul, J. Taieb, J. Souglakos, Q. Shi, R. Kerr, R. Labianca, J.A. Meyerhardt, D. Vernerey, T. Yamanaka, I. Boukovinas, J.P. Meyers, L.A. Renfro, D. Niedzwiecki, T. Watanabe,* V. Torri, M. Saunders, D.J. Sargent,* T. Andre, and T. Iveson

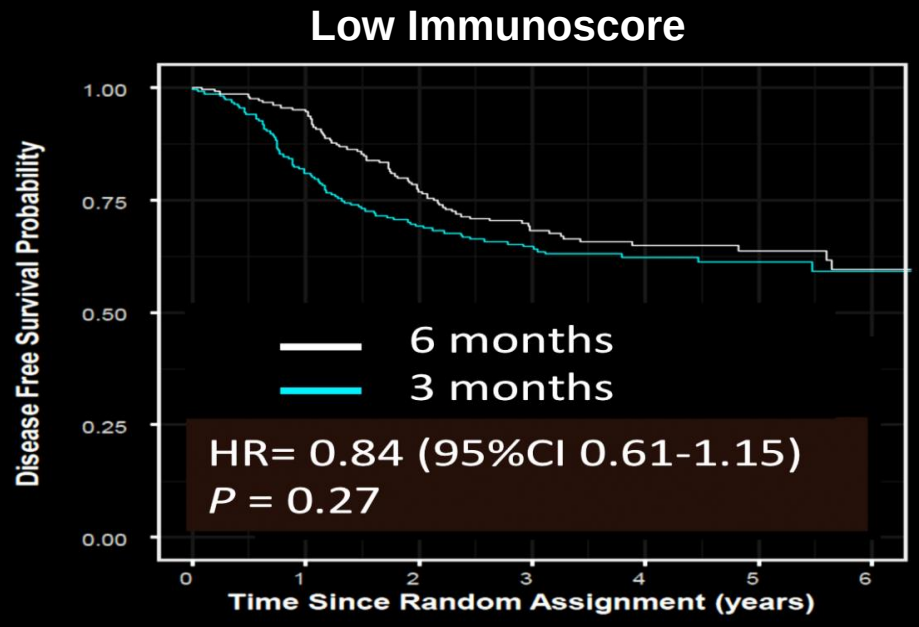


Unconclusive

Phase 3 randomized study of stage III colon cancer patients (IDEA)
3 *vs* 6 months of chemotherapy (n=1062)

All Stage III treated with FOLFOX





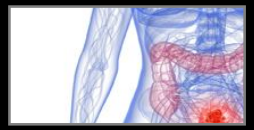
High Immunoscore significantly **predicts** response to 6 months FOLFOX chemotherapy in all Stage III patients

Is there an immune escape at the metastatic stage ?

Metastasis analysis

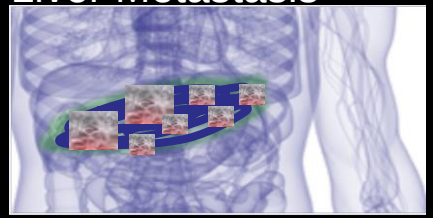
One primary tumor

Colorectal cancer



Multiple metastatic sites

Liver Metastasis



Lung Metastasis

N=603 metastases

➢ Immunoscore within multiple metastases at different sites

Mlecnik et al. ***JNCI*** 2018

Van den Eynde M. *et al*. ***Cancer Cell 2018***

What drives metastasis

What are the metastatic escape mechanisms

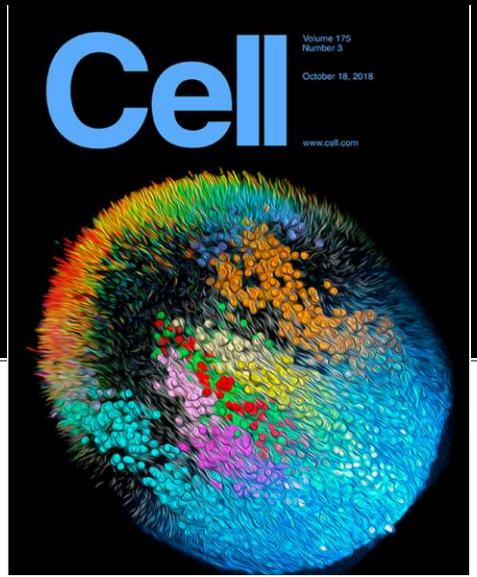
A Novel theory of cancer evolution

Current theories of cancer evolution

Models

LINEAR	NEUTRAL	BIG-BANG	BRANCHED
			
Immune pressure from Darwinian selection			
NO	**NO**	**NO**	**NO**

- The 4 proposed theories of cancer evolution
- All theories are tumor cell-centric. None involves a role of the immune system.



Article

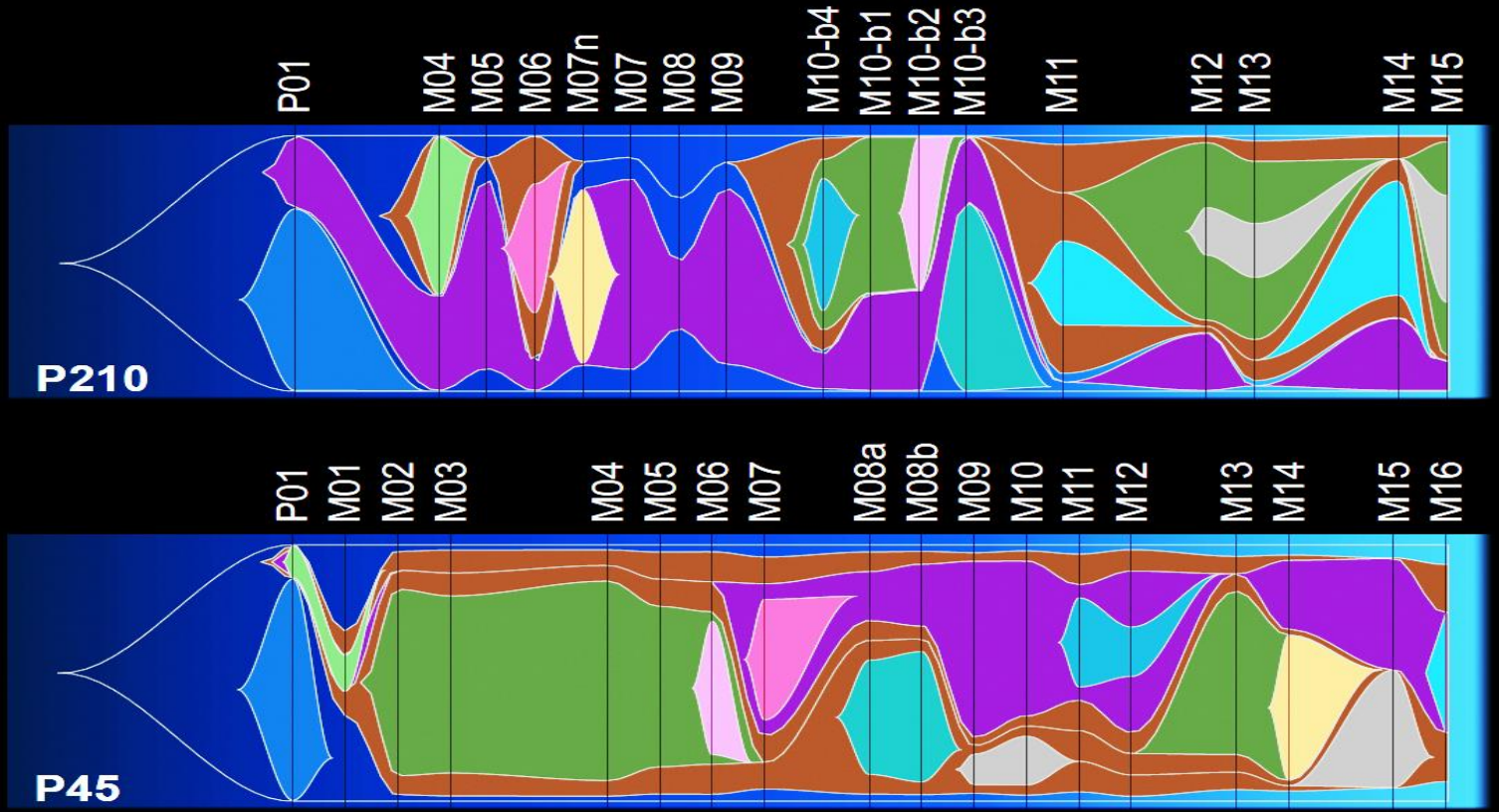
Cell

Evolution of Metastases in Space and Time under Immune Selection

Mihaela Angelova,[1] Bernhard Mlecnik,[1,2] Angela Vasaturo,[1] Gabriela Bindea,[1] Tessa Fredriksen,[1] Lucie Lafontaine,[1] Bénédicte Buttard,[1] Erwan Morgand,[1] Daniela Bruni,[1] Anne Jouret-Mourin,[3] Catherine Hubert,[3] Alex Kartheuser,[3] Yves Humblet,[3] Michele Ceccarelli,[4,5] Najeeb Syed,[6] Francesco M. Marincola,[7,8] Davide Bedognetti,[9,10] Marc Van den Eynde,[1,3,10] and Jérôme Galon[1,11,*]

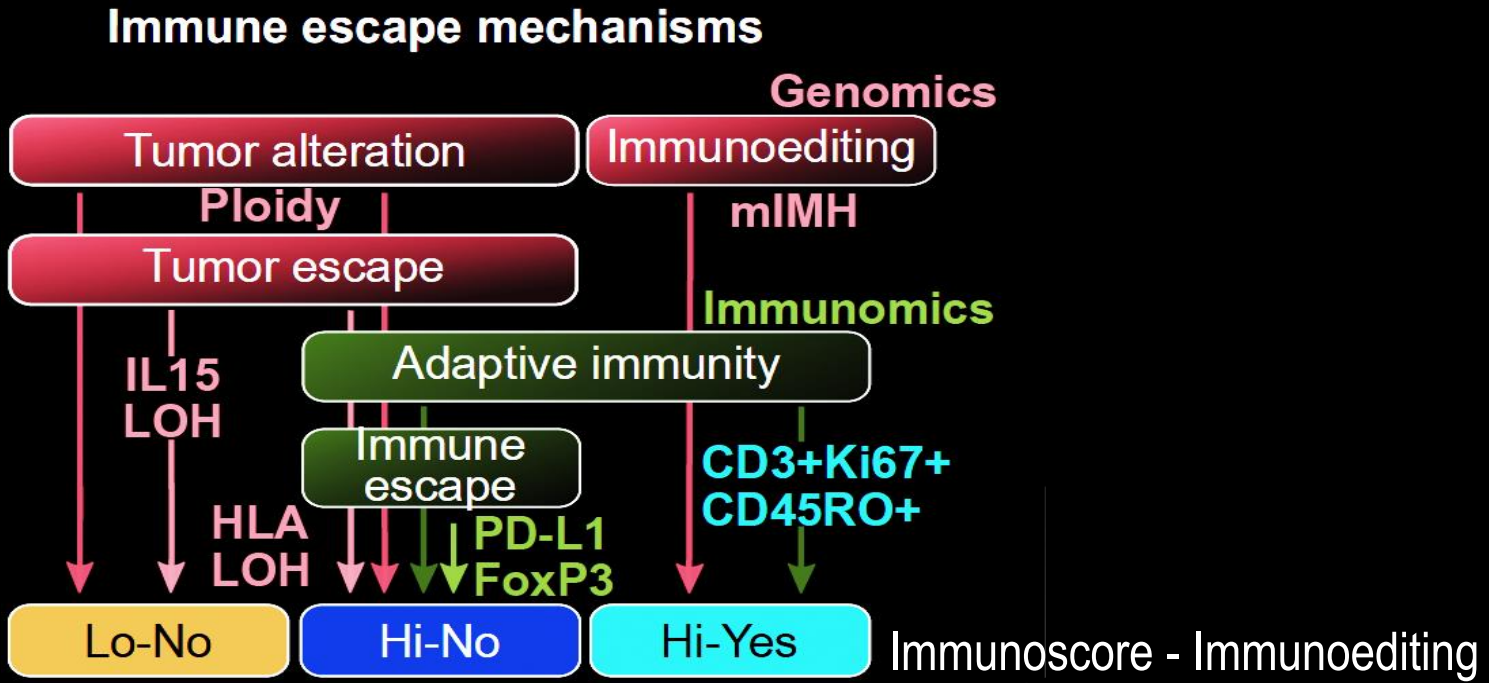
Angelova M. *et al.* ***Cell*** 2018

Evolvogram of tumor clones



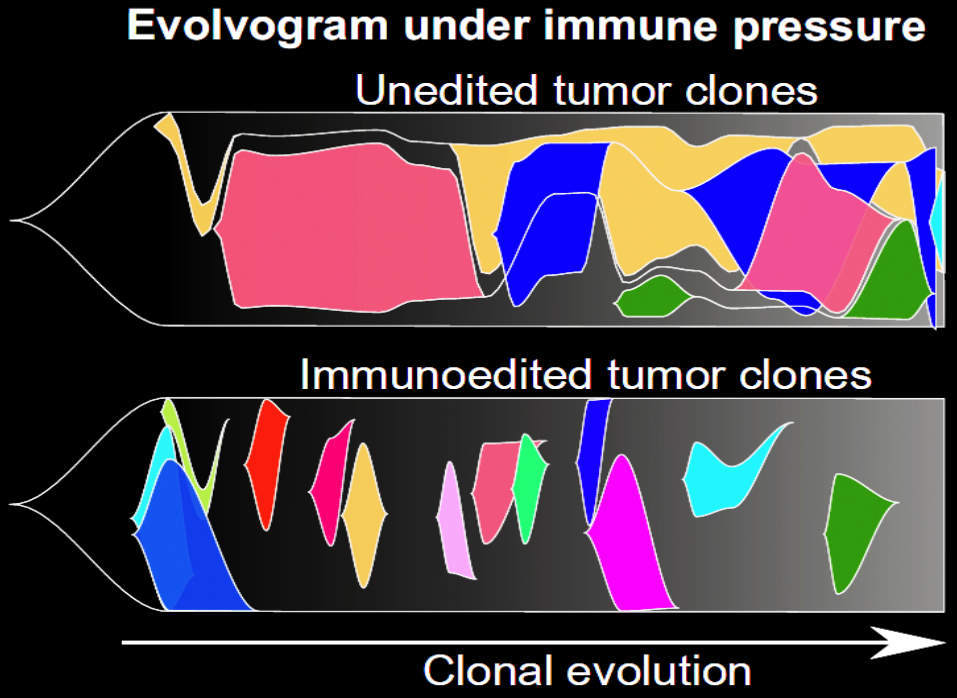
- ✓ Clonal evolution and cancer evolvogram
- ✓ Non-recurrent clones are immunoedited. Progressing clones are immune privileged

What drives metastasis? Conclusions (1)



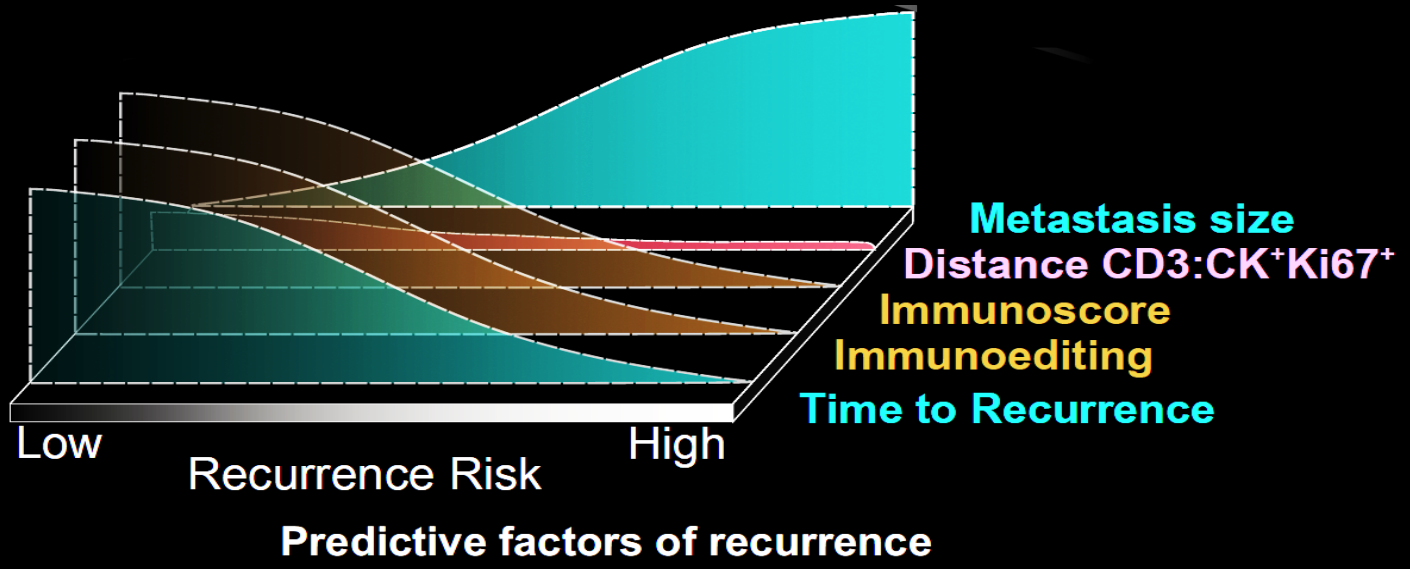
- Different escape mechanisms delineated by lack of adaptive immunity or immunoediting.
- Multiverse of metastases evolution in space and time under immune selection

What drives metastasis? Conclusions (2)



- Evolution of tumor clones is linked to the intra-metastatic immune contexture.
- Non-recurrent clones are immunoedited. Progressing clones are immune privileged.

What drives metastasis? Conclusions (2)



- Immunoediting and Immunoscore are predictive factors of metastasis recurrence.
- Distance between CD3 + cells and tumor cells Ki67+ and metastasis size are also associated metastasis recurrence.

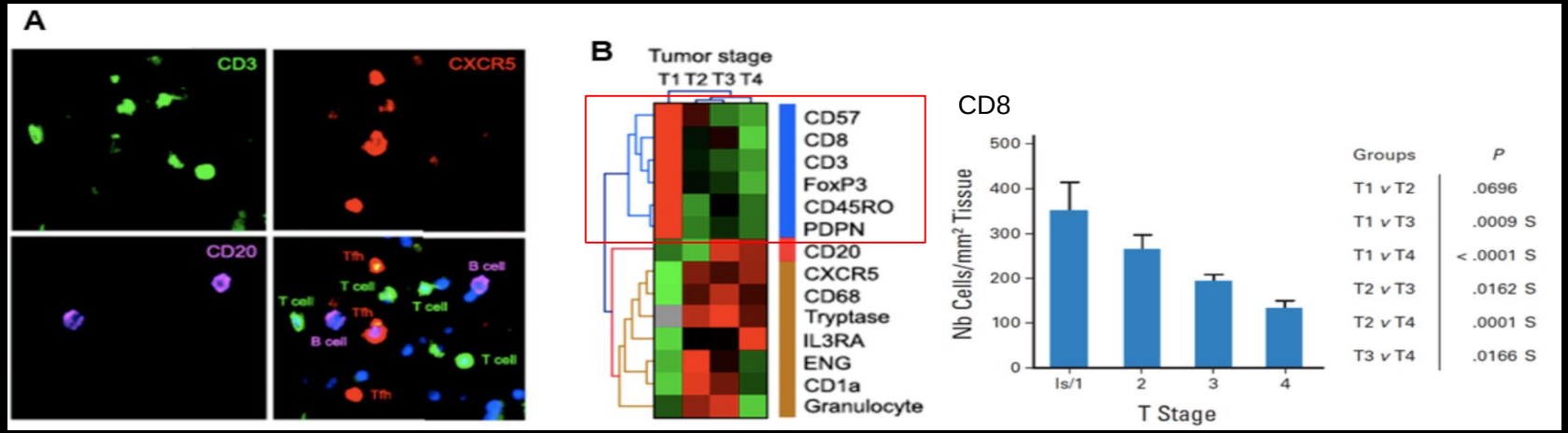
A Novel theory of cancer evolution

Models

LINEAR	NEUTRAL	BIG-BANG	BRANCHED	SELECTION
				
Immune pressure from Darwinian selection				
NO	**NO**	**NO**	**NO**	**YES**

- **Parallel immune selection model**
- Dynamic interaction of tumor-cells with immune-cells and Darwinian selection of immune escape variant, with parallel evolution and multiverse of metastases.

Adaptive immunity decreases with tumor progression



Bindea G. et al. ***Immunity*** 2013

Mlecnik B. et al. ***J Clin Oncol*** 2011

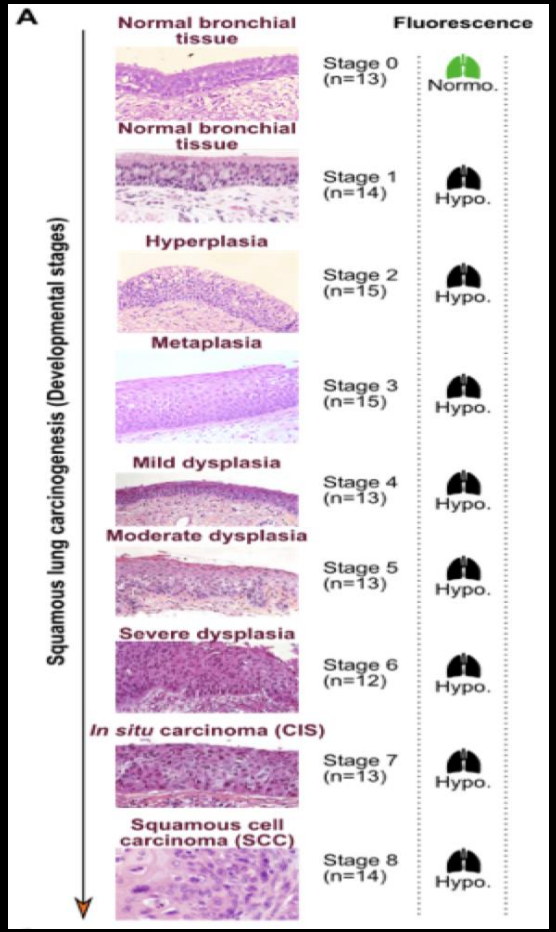
nature
International journal of science

Immune evasion before tumour invasion in early lung squamous carcinogenesis

Céline Mascaux[1,2,3,4,14,15,18]*, Mihaela Angelova[5,6,7,8,16,18], Angela Vasaturo[5,6,7,8], Jennifer Beane[2], Kahkeshan Hijazi[2], Geraldine Anthoine[1], Bénédicte Buttard[5,6,7,8], Françoise Rothe[9], Karen Willard-Gallo[10], Annick Haller[11,17], Vincent Ninane[12], Arsène Burny[13], Jean-Paul Sculier[1], Avi Spira[2] & Jérôme Galon[5,6,7,8]*

Mascaux C et al. ***Nature*** 2019

Oncogenesis of lung squamous cell carcinoma



- ✓ Analysis of 122 pre-cancer lesions across 9 developmental stages

Mascaux C et al. ***Nature*** *2019*

Main gene expression patterns across 9 developmental stages

Ascending

Ascending from High-Grade

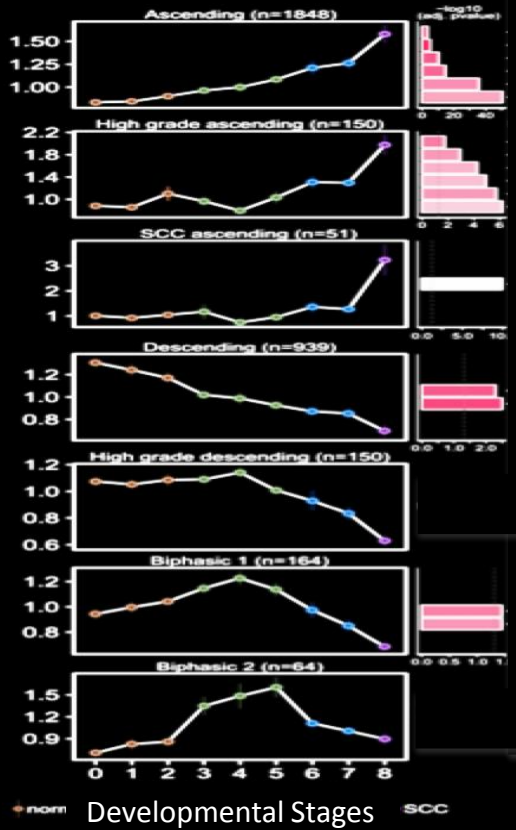
SCC Ascending

Descending

Descending from High-Grade

Biphasic 1

Biphasic 2



Proliferation

Immune

Develomental (EMT)

DNA damage

Heterogeneous

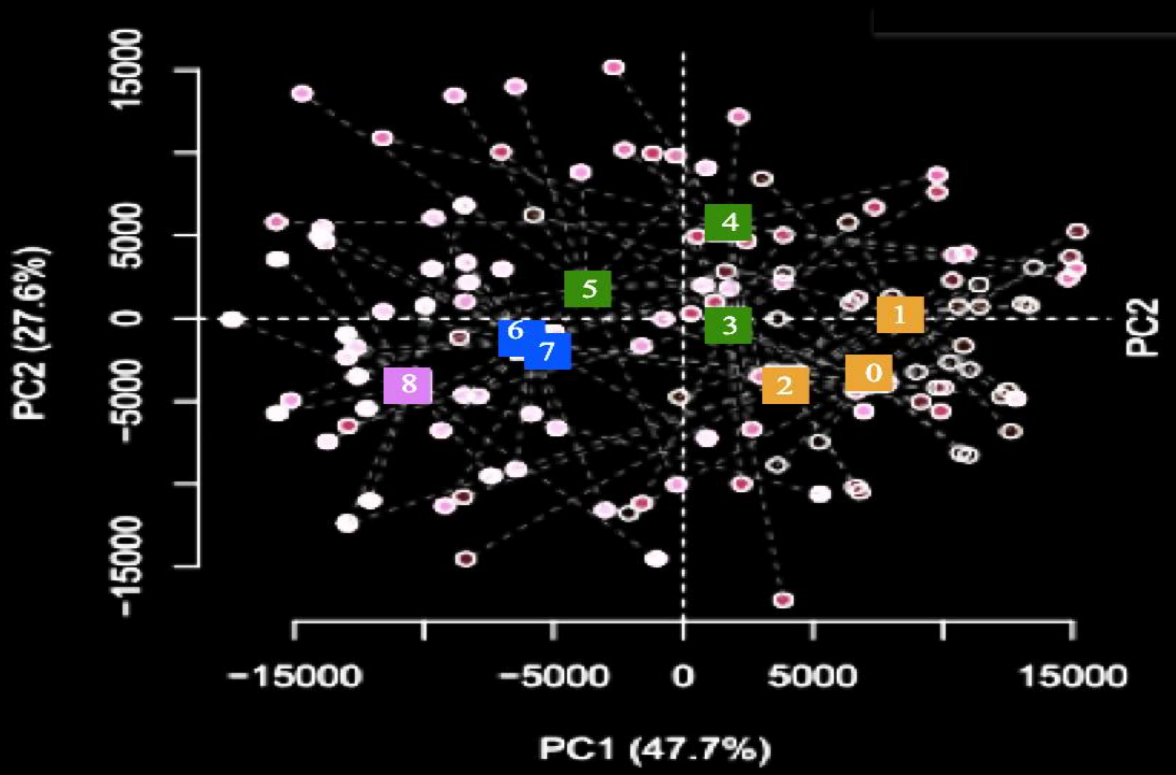
Metabolism

Heterogeneous

Immune functions mostly associated with genes ascending from high-Grade

Mascaux C et al. ***Nature*** *2019*

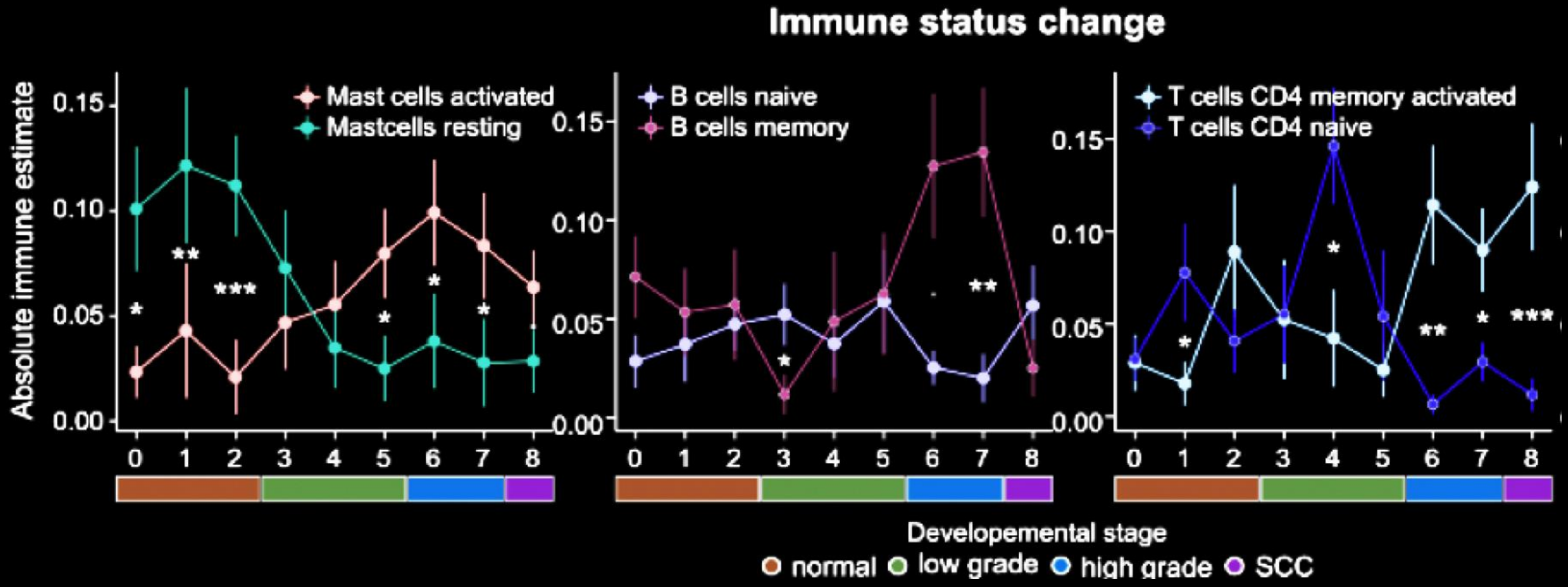
Principal components evolution of the 9 developmental stages



- Normal
- Low Grade dysplasia
- High Grade dysplasia
- Squamous cell carcinoma (SCC)

Mascaux C et al. ***Nature*** *2019*

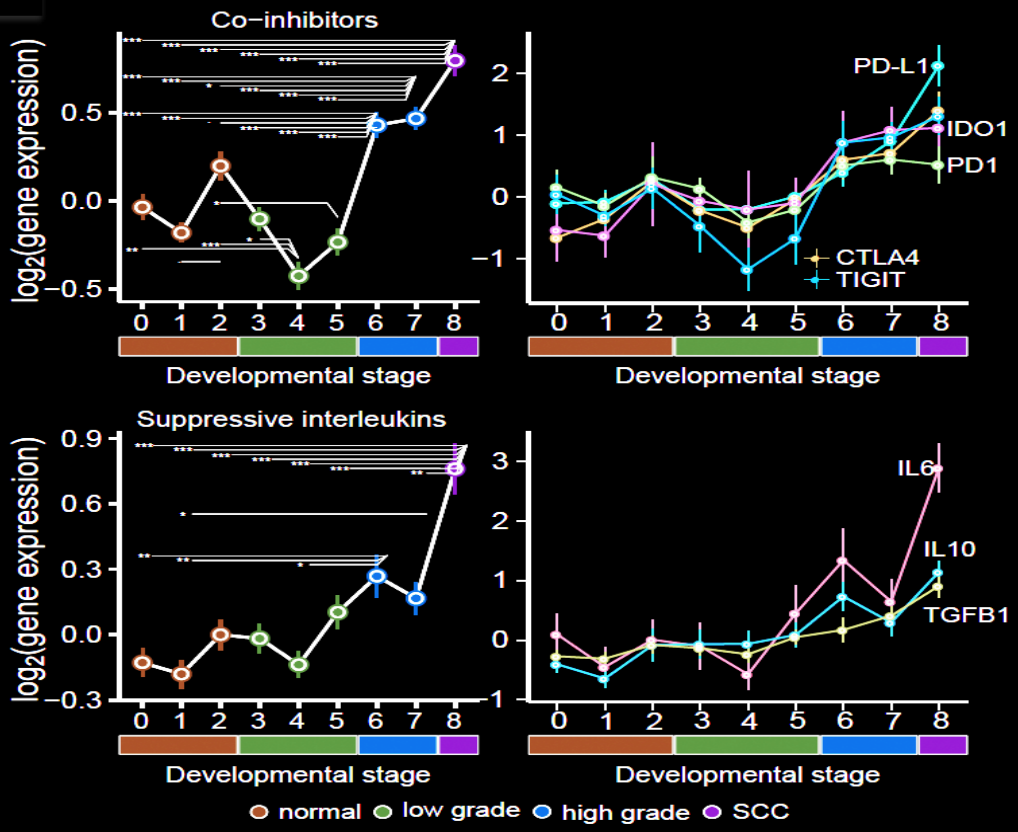
Immune activation across developmental stages



- ✓ Early Immune activation in Low-Grade dysplasia (Immune sensing)
- ✓ Adaptive immune activation and memory in High-Grade dysplasia

Mascaux C et al. ***Nature*** *2019*

Immune escape mechanisms in pre-cancer lesions

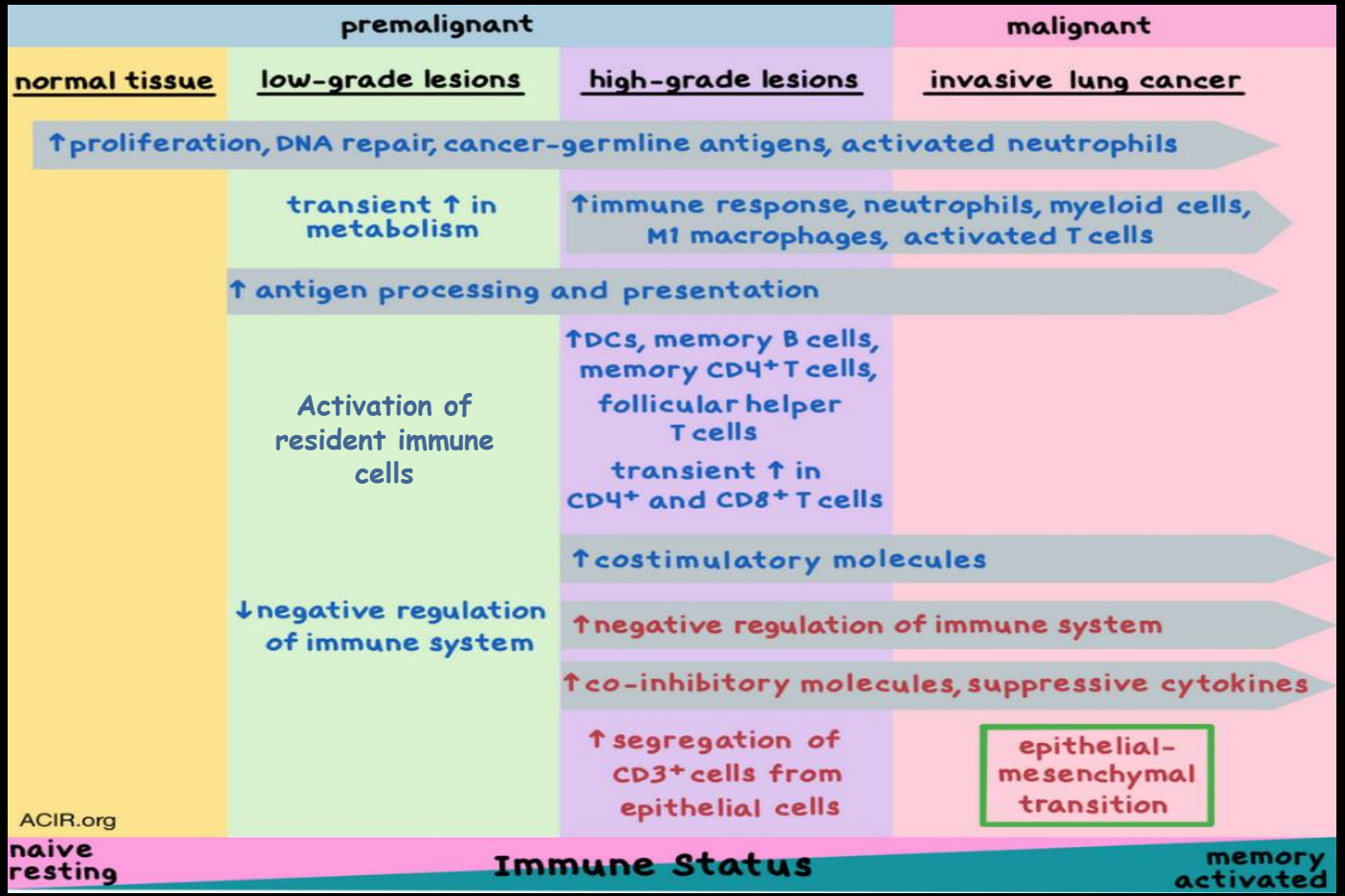


- ✓ Decreased expression of co-inhibitors in Low-Grade
- ✓ Increased expression of co-inhibitors in High-Grade
- ✓ Increased expression of suppressive cytokines in High-Grade

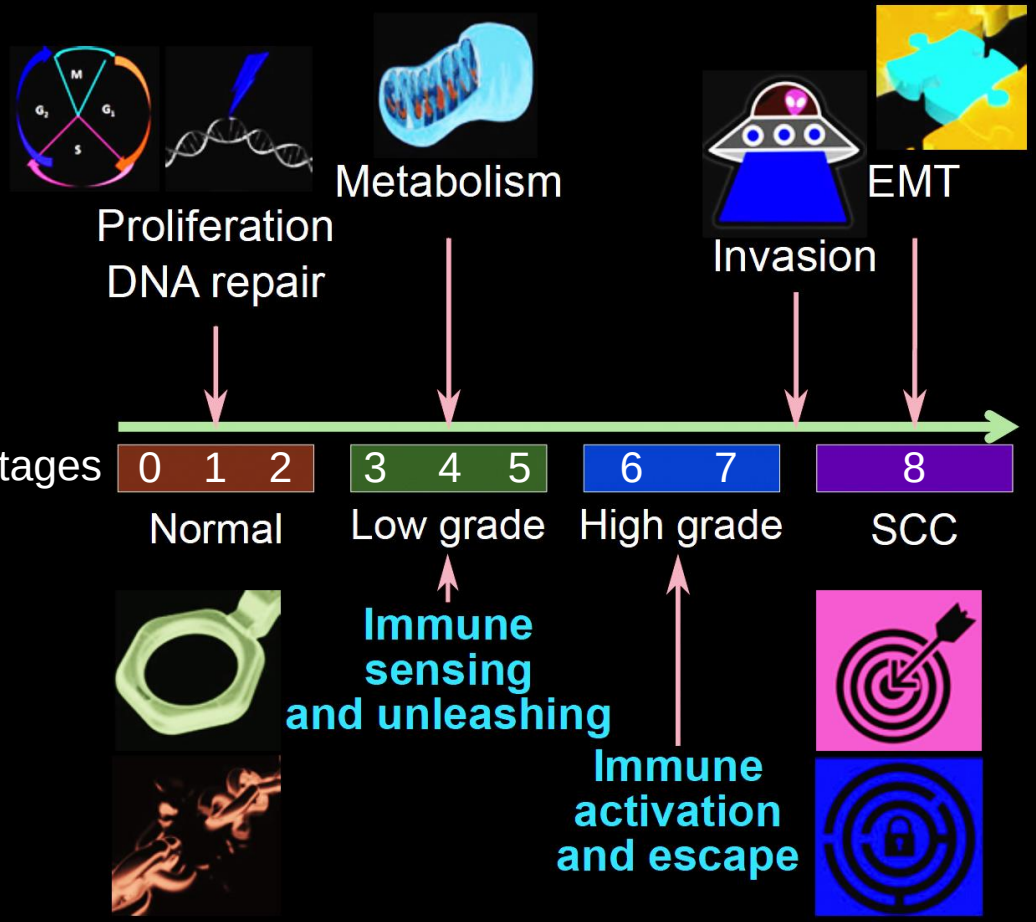
-> Immune evasion before tumor invasion (SCC)

Mascaux C et al. ***Nature*** *2019*

Immune activation and immune escape during lung oncogenesis



Pre-Neoplastic / Pre-Cancer Lesion evolution

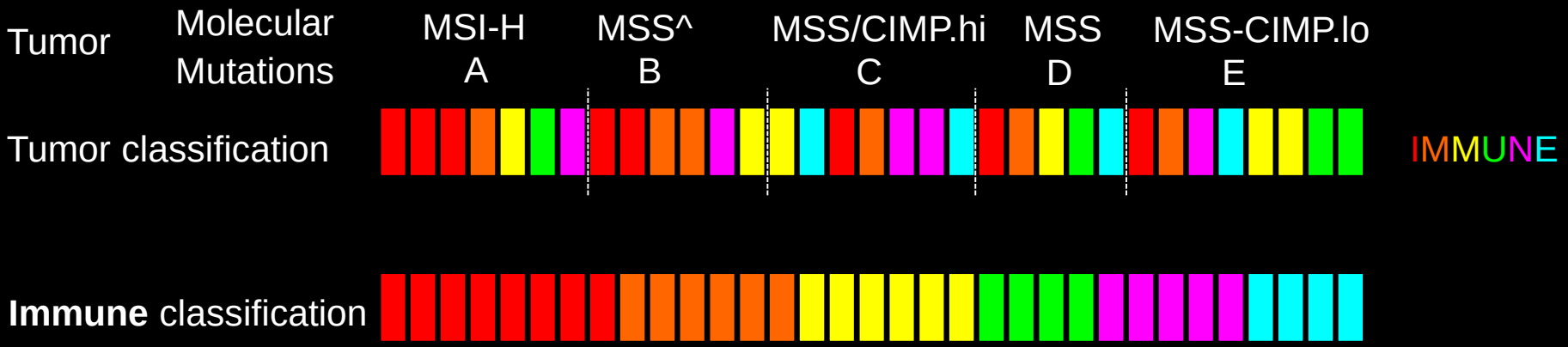


Epithelial cells
Stroma
Tumor cells

Immune microenvironment

Mascaux C. … Galon J. ***Nature*** 2019

Stratification of cancer based on the immune status



-> Importance of having standardized immune Assays

Galon lab.

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