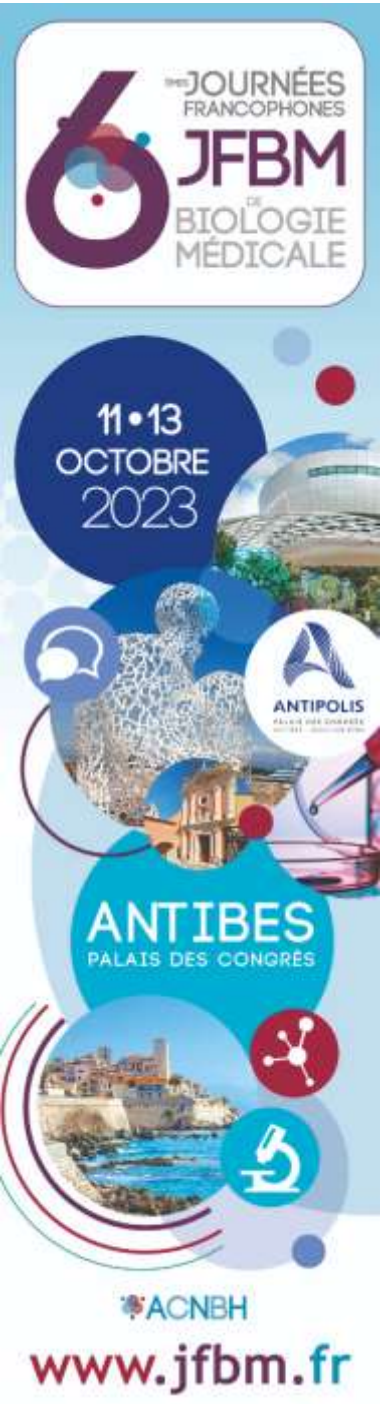



THE SHIFT PROJECT
THE SEPSIS IMMUNOLOGY TRANSITION THINK TANK

# SEPSIS: DIAGNOSTIC, SUIVI IMMUNITAIRE ET IMMUNOTHÉRAPIE

## Place des biomarqueurs

**Guillaume Monneret**

Laboratoire d'Immunologie / Hôp. E. Herriot / Hospices Civils de Lyon
EA 7426 – Immunodépression induite par les inflammations systémiques - U. Lyon 1

HCL HOSPICES CIVILS DE LYON

Flow Division
Immunology Department

Lyon 1


DECLARATION D'INTERET
DANS LE CADRE DE MISSIONS DE FORMATION
RÉALISÉES POUR LE JFBM

Pr G. Monneret déclare
**ne pas avoir d'intérêt**, direct ou indirect (financier), avec les entreprises pharmaceutiques, du diagnostic ou d'édition de logiciels susceptible de modifier mon jugement ou mes propos, **concernant le sujet et les DMDIV présentés**.

# préambule

# More than ever: septic syndromes still a serious a public health concern

2017

World Health Organization

**Recognizing Sepsis as a Global Health Priority — A WHO Resolution**

Konrad Reinhart, M.D., Ron Daniels, M.D., Niranjan Kissoon, M.D., Flavia R. Machado, M.D., Ph.D., Raymond D. Schachter, L.L.B., and Simon Finfer, M.D.

The NEW ENGLAND JOURNAL of MEDICINE

2020 (jan)

**Globally, sepsis accounts for 11 million deaths / year (Rudd et al. Lancet)**

*"By comparison, the World Health Organisation estimated that there were 9.6 million deaths from cancer in 2018".*

THE LANCET

2021

COVID-19: it's all about sepsis

Jean-Louis Vincent*,[1]

*2,5 million deaths / year (by march 2023)*

Future MICROBIOLOGY

2023

- leading cause of death in ICU
- 28-day mortality: sepsis = 20 %, septic shock = 40 %

Special Communication | CARING FOR THE CRITICALLY ILL PATIENT

# The Third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3)

JAMA The Journal of the American Medical Association

2016

**Sepsis**

Life threatening organ dysnfunction caused by a dysregulated host response to infection (*i.e,* one infection + one organ failure)

**Septic shock**

Sepsis + vasopressor therapy needed (*i.e*., cardiovascular failure) + Lactate > 2mmol/L

*Singer et al., JAMA 2016*

# Sepsis timeline

SEPSIS = ED / ICU

3

*Biomarker-guided strategy*

*Anti-inflammatoires ?*

*Monitoring altérations immunitaires*

1

*Ante-sepsis markers ?*

=> Stopper progression

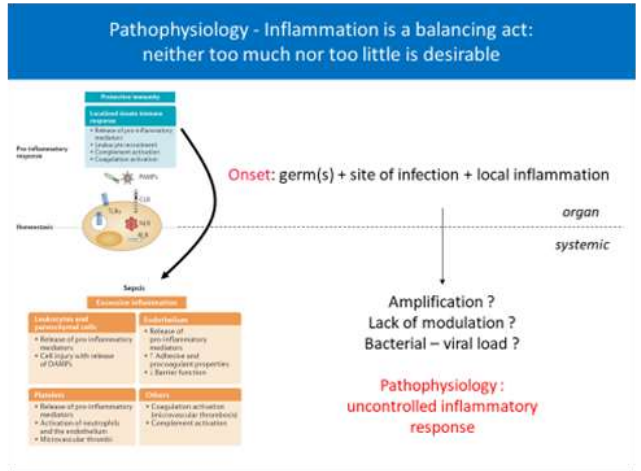
Ø

2

*Diagnostic précoce (urgence antibio)*

*Chronologie*

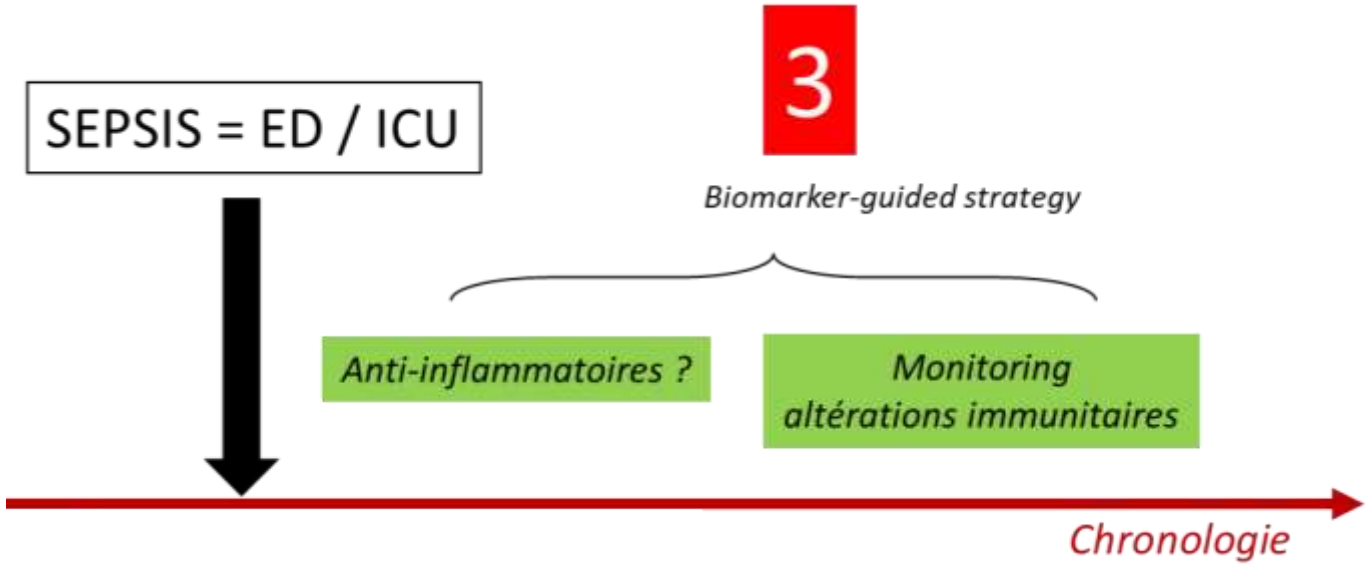
Microbiologie




PCT, CRP, MIF, cytokines, sTREM-1, pro-BNP, BNP, pro-ANP, CD64, mHLA-DR, sCD14, troponine, HMG-B1…

# Sepsis timeline

## IMMUNOMODULATION ?



# LOOKING BACK ...

- multiple negative trials over last 30 years
  - no new outcome-improving therapeutics
  - interventions likely work in some patients ... but in whom?
  - slowly learning that one size does not fit all
- wasted opportunities with COVID
  - millions of people with single infectious disease condition
  - .. but very few studies where biological impact of intervention was studied
  - 'proven' interventions (steroids, tocilizumab, baricitinib) likely work in some

    .. and likely harm others.
  - But we can't identify who to treat or not treat

*Slide courtesy : Mervyn Singer (2022)*

# Same mistakes, same consequences = COVID-19 (as a missed opportunity)

## Immunomodulators in COVID-19: Two Sides to Every Coin

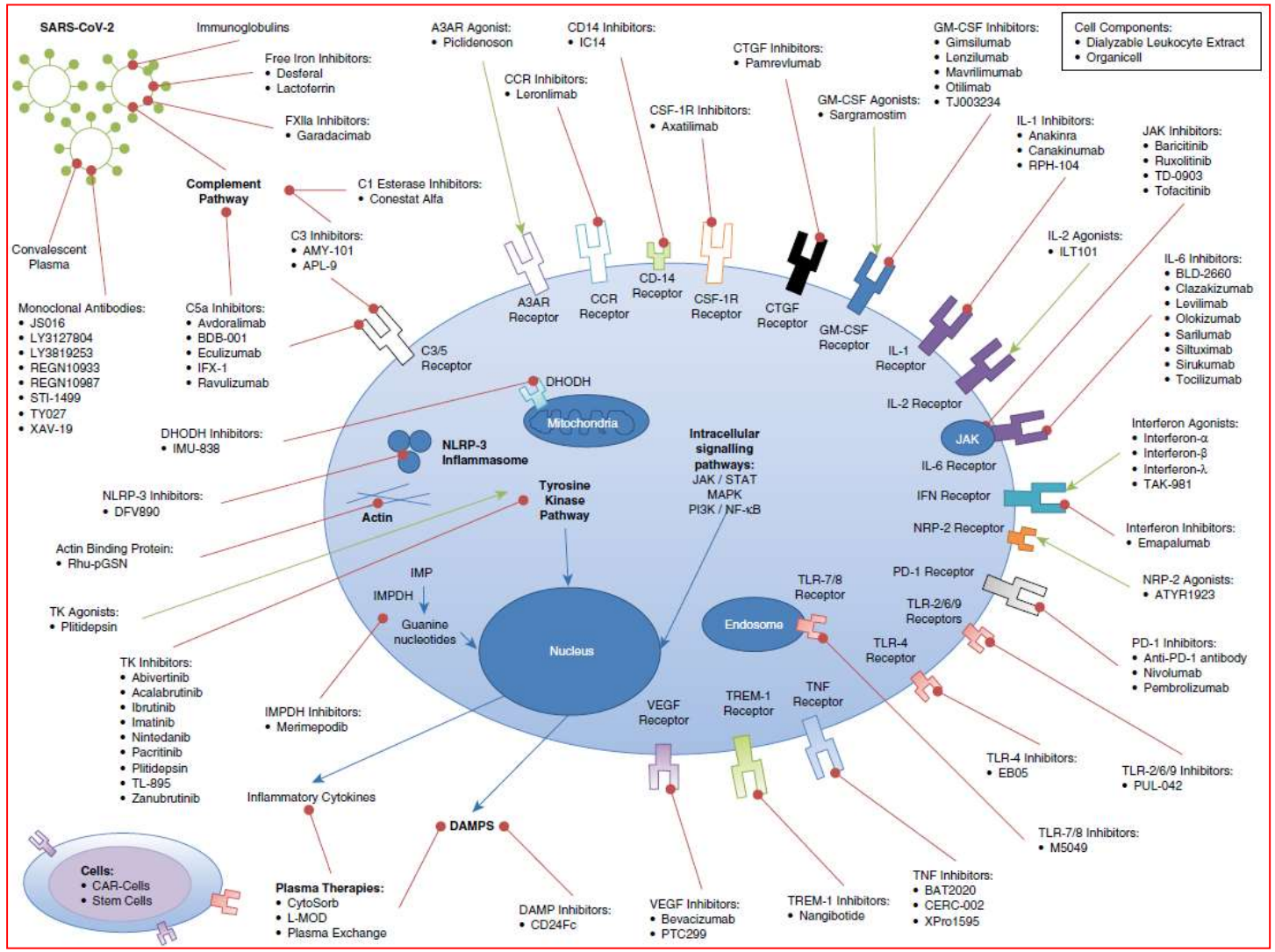
Dès la fin 2020

> 300 essais cliniques enregistrés evaluant des traitements immunomodulateurs

> 90 traitements différents

> 47 essais cliniques évaluant des traitements ciblant l'IL-6

**No stratified studies on inflammation markers**

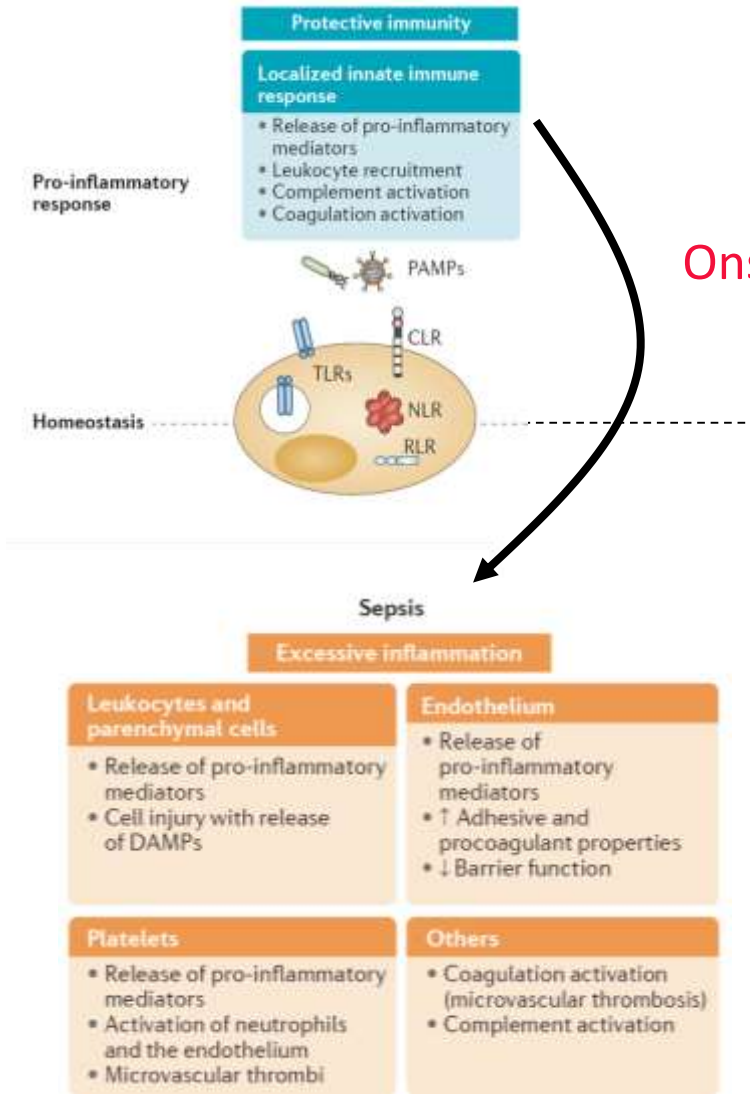



AJRCCM

Snow, Singer & Arulkumaran 2020

# Back to definitions and pathophysiology

# Pathophysiology - Inflammation is a balancing act: neither too much nor too little is desirable




Onset: germ(s) + site of infection + local inflammation

*organ*

*systemic*

Amplification ?
Lack of modulation ?
Bacterial – viral load ?

Pathophysiology :
uncontrolled inflammatory response

# Old paradigm for sepsis pathophysiology

Pathogen

Direct damage by pathogen

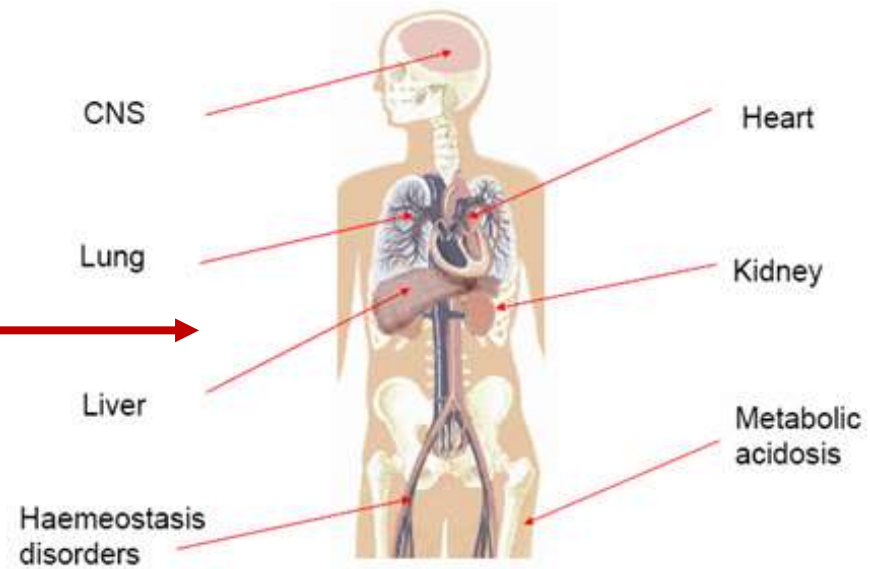
Host resistance mechanisms

Damage caused by the immune response

Host

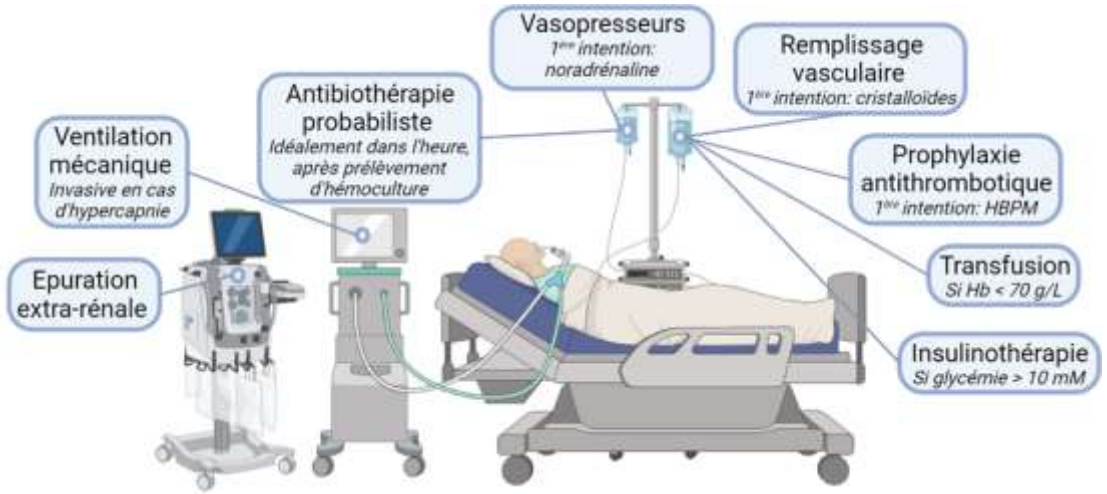
**Uncontrolled Inflammatory response**

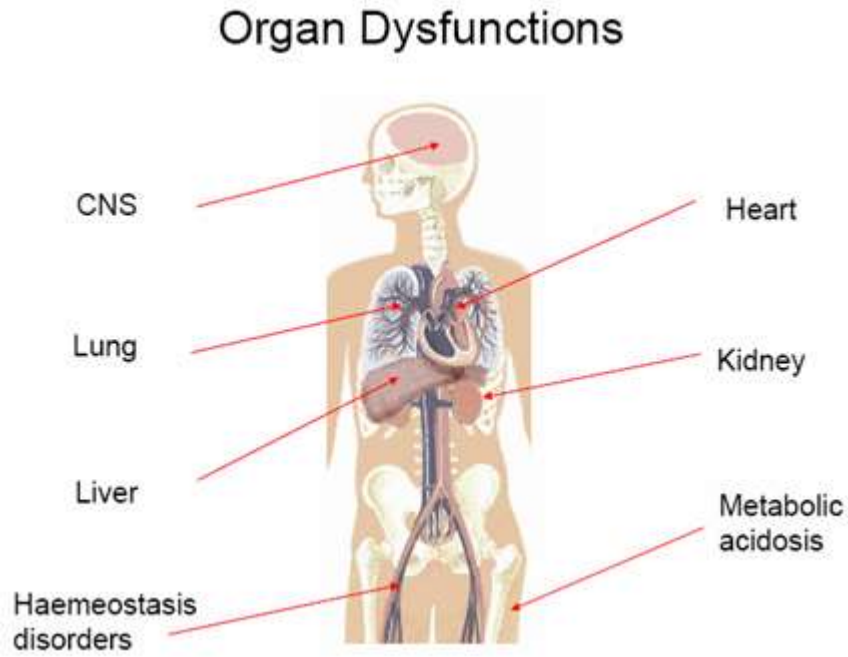
Organ Dysfunctions




Decreased arterial pressure
Shock
Multiple organ failure

# Management of septic patients

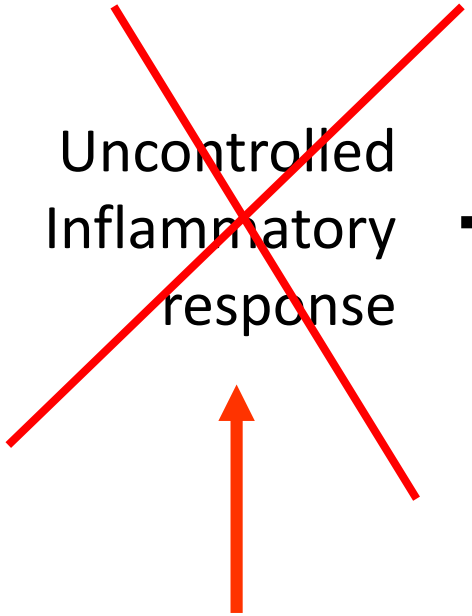


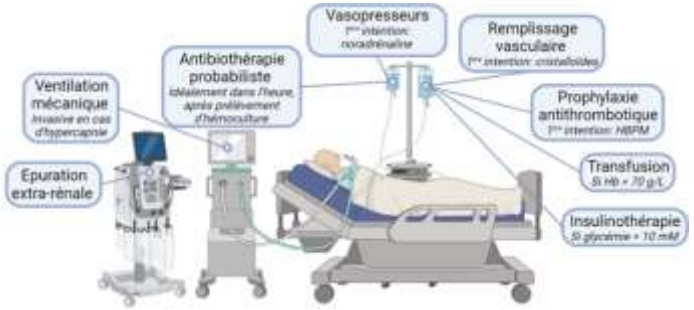

Decreased arterial pressure
Shock
Multiple organ failure

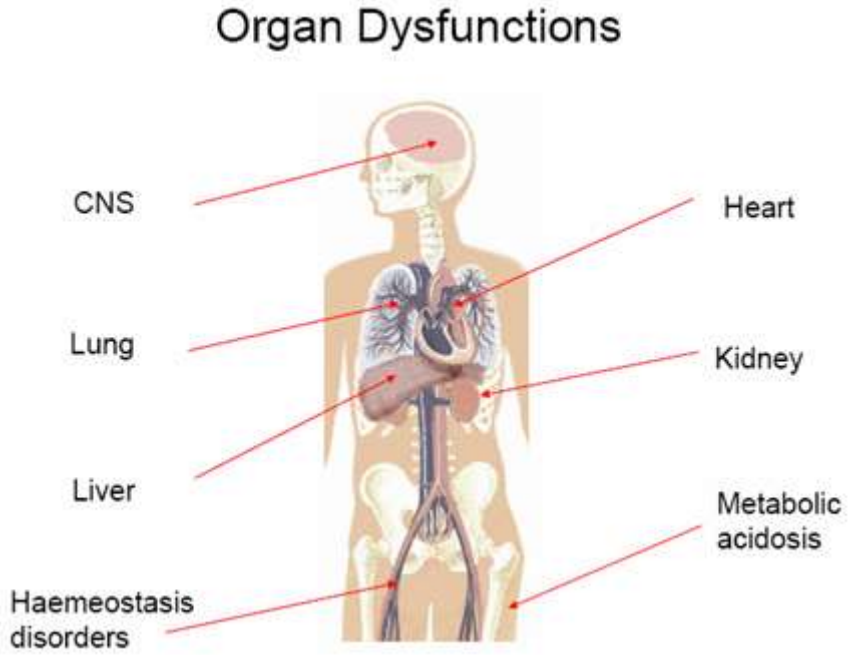
# Management of septic patients

~~Uncontrolled Inflammatory response~~



Adjunctive therapy




Decreased arterial pressure / Shock
Multiple organ failure

# Failure of clinical trials testing anti-inflammatory therapies

| Drug | Number of studies | Number of patients | Mortality (%) | |
|---|---|---|---|---|
| | | | Placebo | Drug |
| Anti-endotoxine | 4 | 2010 | 35 | 35 |
| Anti-bradykinine | 2 | 755 | 36 | 39 |
| Anti-PAF | 2 | 870 | 50 | 45 |
| Anti-TNF | 8 | 4132 | 41 | 40 |
| R solubles TNF | 2 | 688 | 38 | 40 |
| AINS | 3 | 514 | 40 | 37 |
| Steroids | 9 | 1267 | 35 | 39 |
| … | … | … | … | … |
| Total | 33 | 12034 | **38** | **38** |

Critical Care Medicine

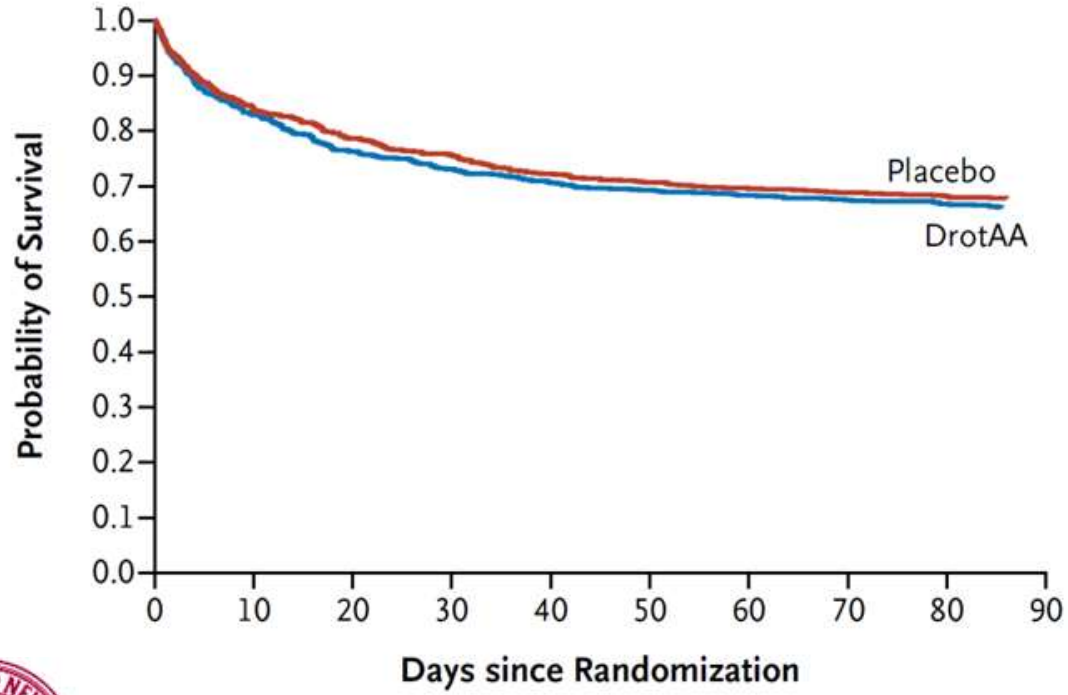
*Zeni et al, Crit Care Med, 1997*

# 40 years of failure in anti-inflammatory therapies

The NEW ENGLAND JOURNAL of MEDICINE

ESTABLISHED IN 1812 MAY 31, 2012 VOL. 366 NO. 22

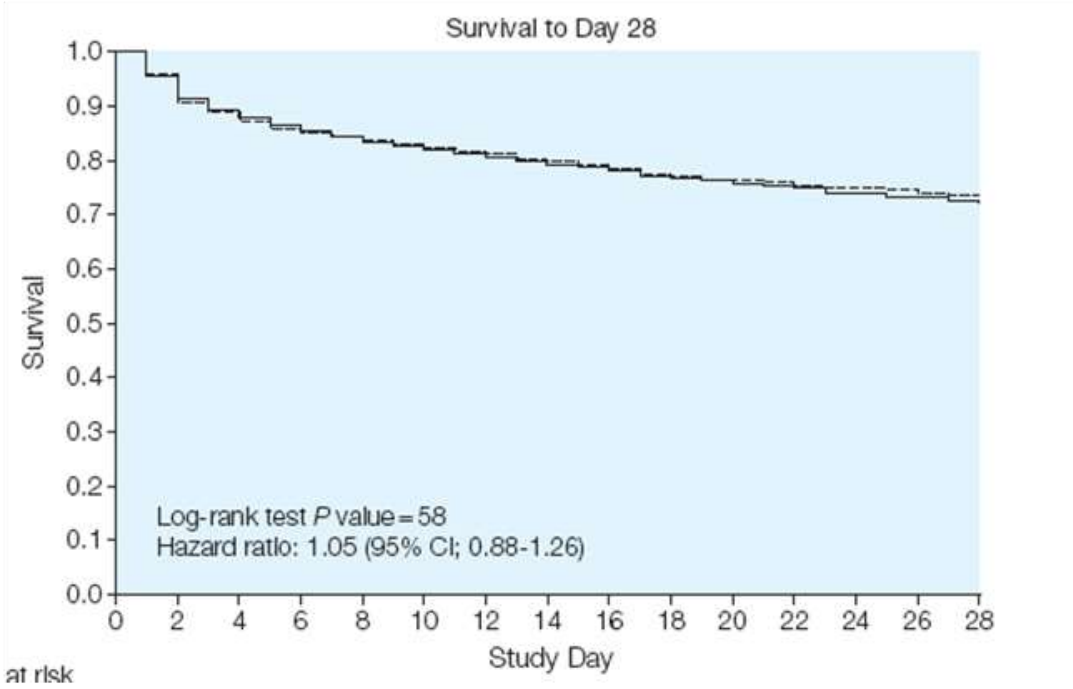
Drotrecogin Alfa (Activated) in Adults with Septic Shock




*Ranieri et al. 2012*

## Effect of Eritoran, an Antagonist of MD2-TLR4, on Mortality in Patients With Severe Sepsis

The ACCESS Randomized Trial



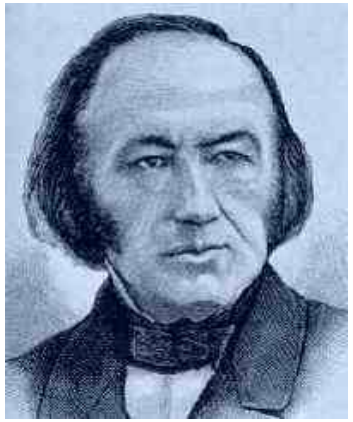

JAMA The Journal of the American Medical Association

*Opal et al. 2013*

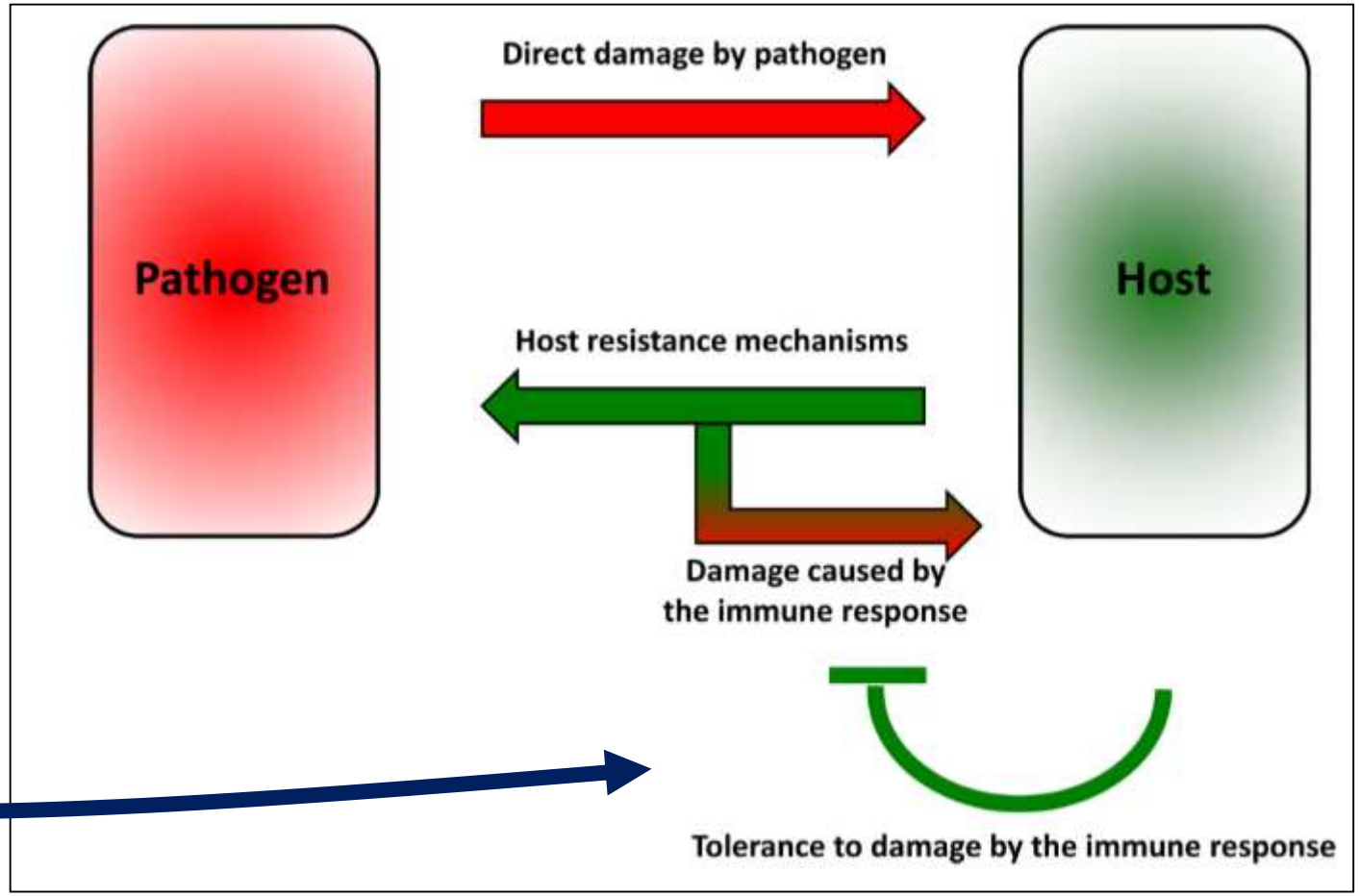
2 primary reasons to elucidate the failure of immunomodulation in sepsis

# **First cause** for the failure of anti-inflammatory strategy
# => **Neglecting the Principle of Homeostasis**

*« Only homeostasis matters »*

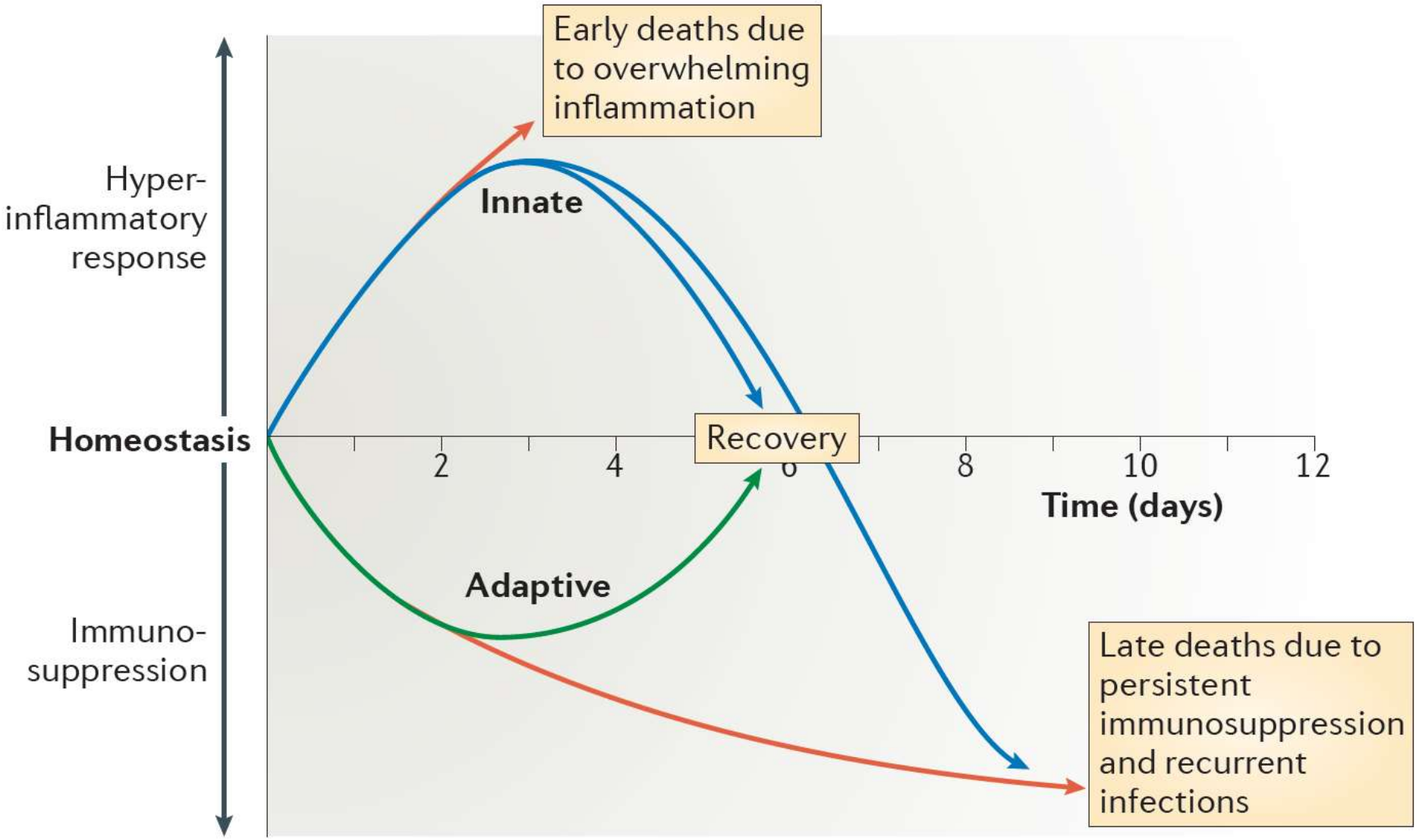


Claude Bernard (1813 – 1878)




*Adated from Medzhitov et al., Science 2012*

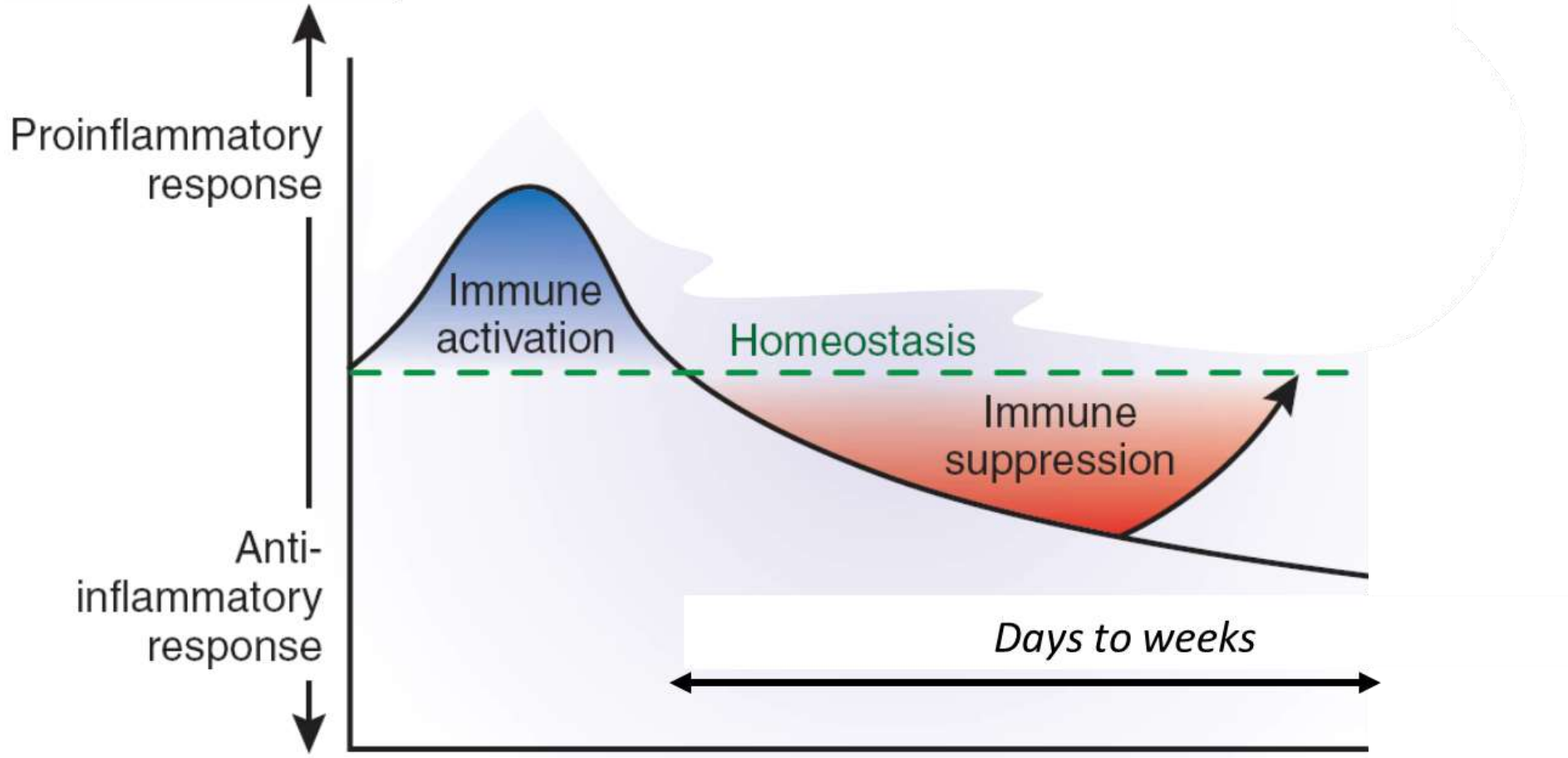
# Pro- / anti-inflammatory balance in septic shock




nature REVIEWS IMMUNOLOGY

*Hotchkiss, Monneret & Payen, 2013*

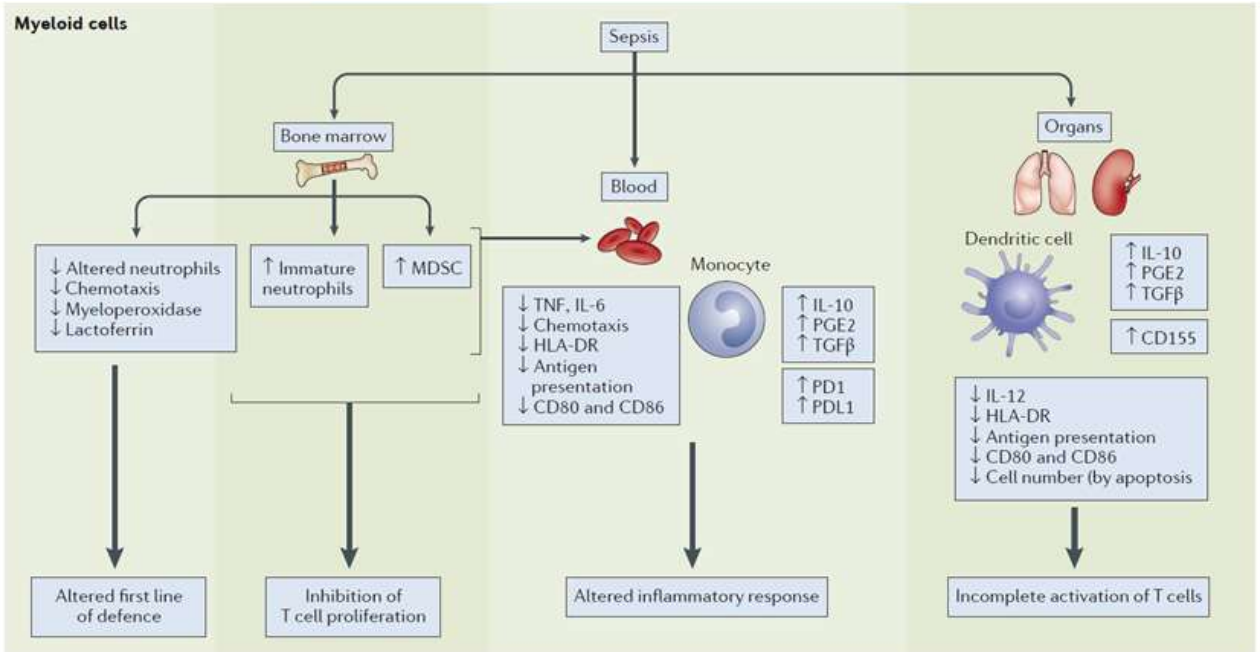
# From exacerbated inflammation to immunosuppression in severely injured patients (a simplified view of resulting forces)




nature medicine

*Adated from Hotchkiss et al., 2009*

# Myeloid cells (summary)

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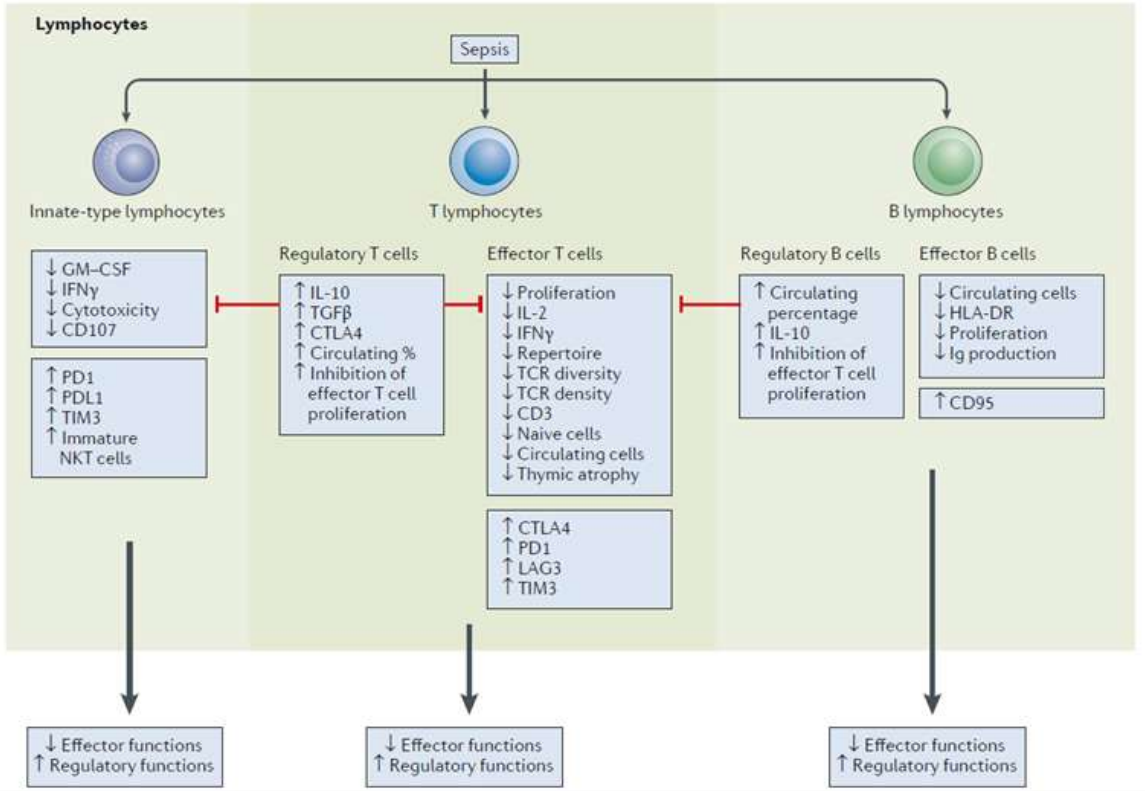



**Organs**

Thymic atrophy + ↓ Thymocytes + T diveristy

Reprogrmmation in bone marrow : ↓ Ly + ↑ MDSC

Lymphopenia in Ly nodes

nature REVIEWS

*Venet & Monneret, 2018*

## *Myeloid cells (summary)*


## *cells (summary)*




**Organs**

Thymic atrophy + ↓ Thymo

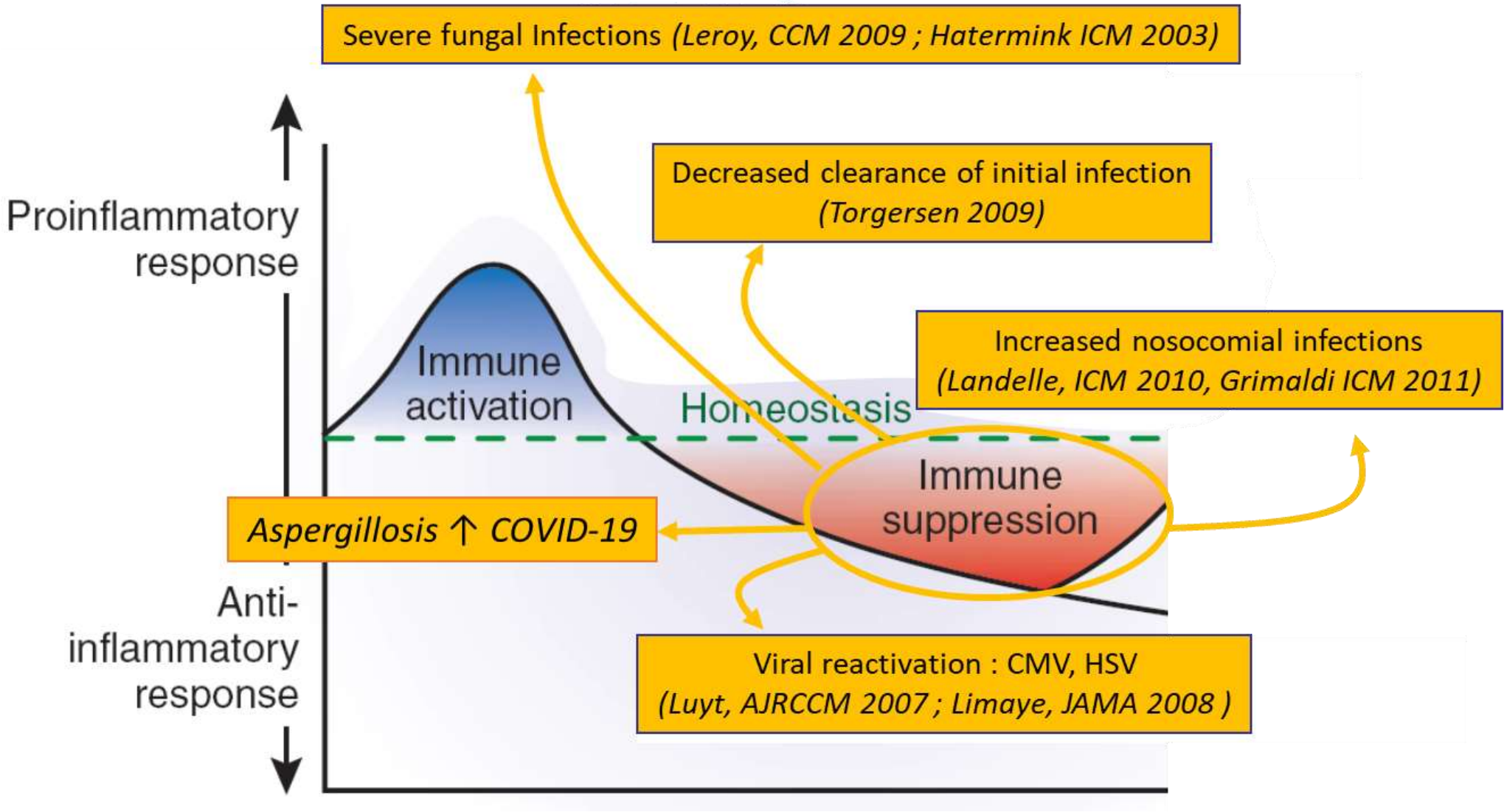
Reprogrmmation in bone ma

Lymphopenia in Ly nodes

nature REVIEWS

*Venet & Monneret, 2018*

# Increased secondary / nosocomial infections after sepsis



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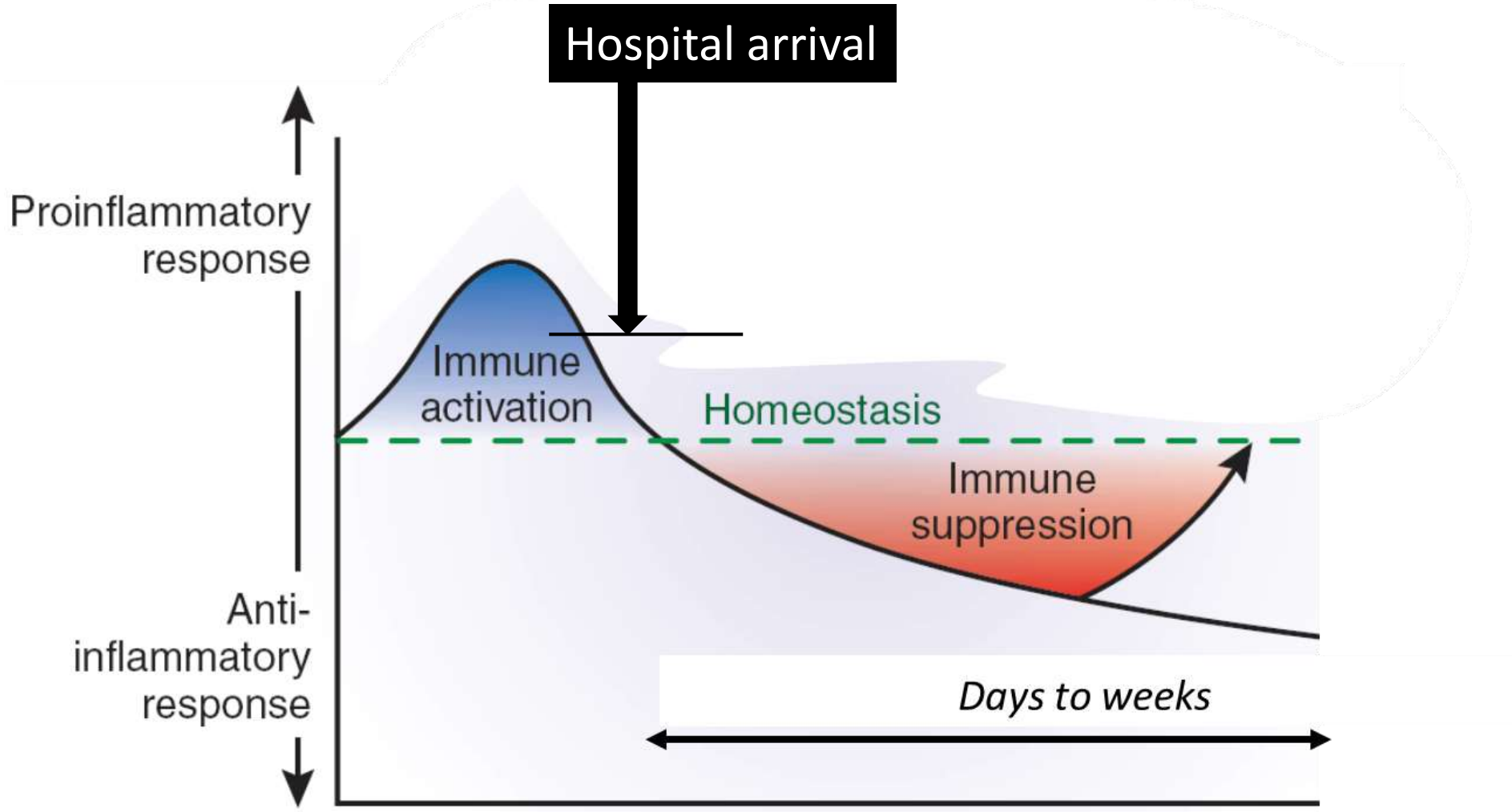



nature medicine

*Adated from Hotchkiss et al., 2009*

Second cause for the failure of anti-inflammatory strategy
=> **« one size fits all (sepsis) » approach**

one size does not fit all (at all times)

# Second cause for the failure of anti-inflammatory strategy
# => « one size fits all (sepsis) » approach

Patients' heterogeneity

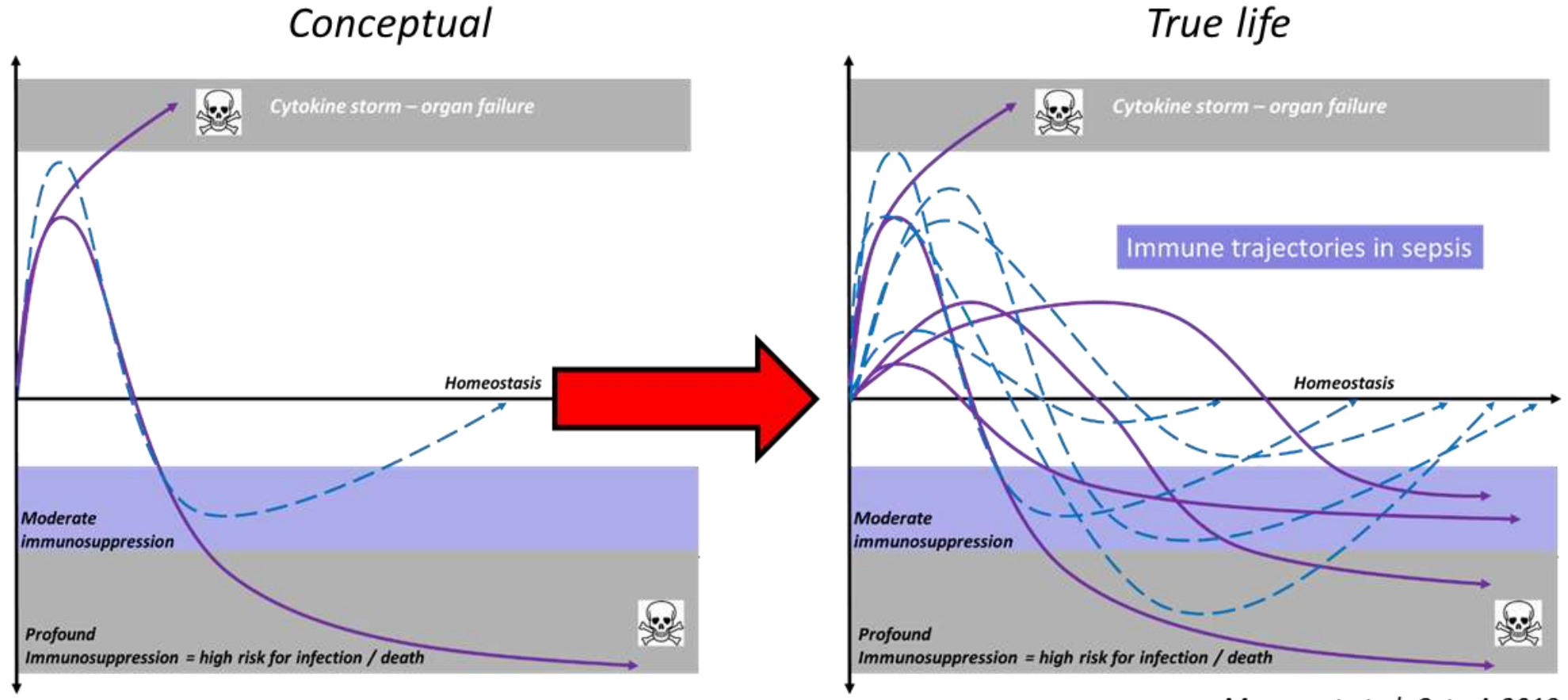
**Infection**
Exacerbated inflammation
Pathogens (PAMPS)
Injury (DAMPS)

**Patients**
Age
Comorbidities
CMV+ serology
Genetic background
Microbiote

**Central Regulation**
Sympathetic
Parasympathetic
HPA axis

**Treatments**
Antibiotics
Catecholamines
Blood transfusion
Steroids
Nutrition

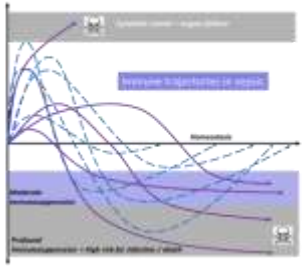
Timing of ICU admission



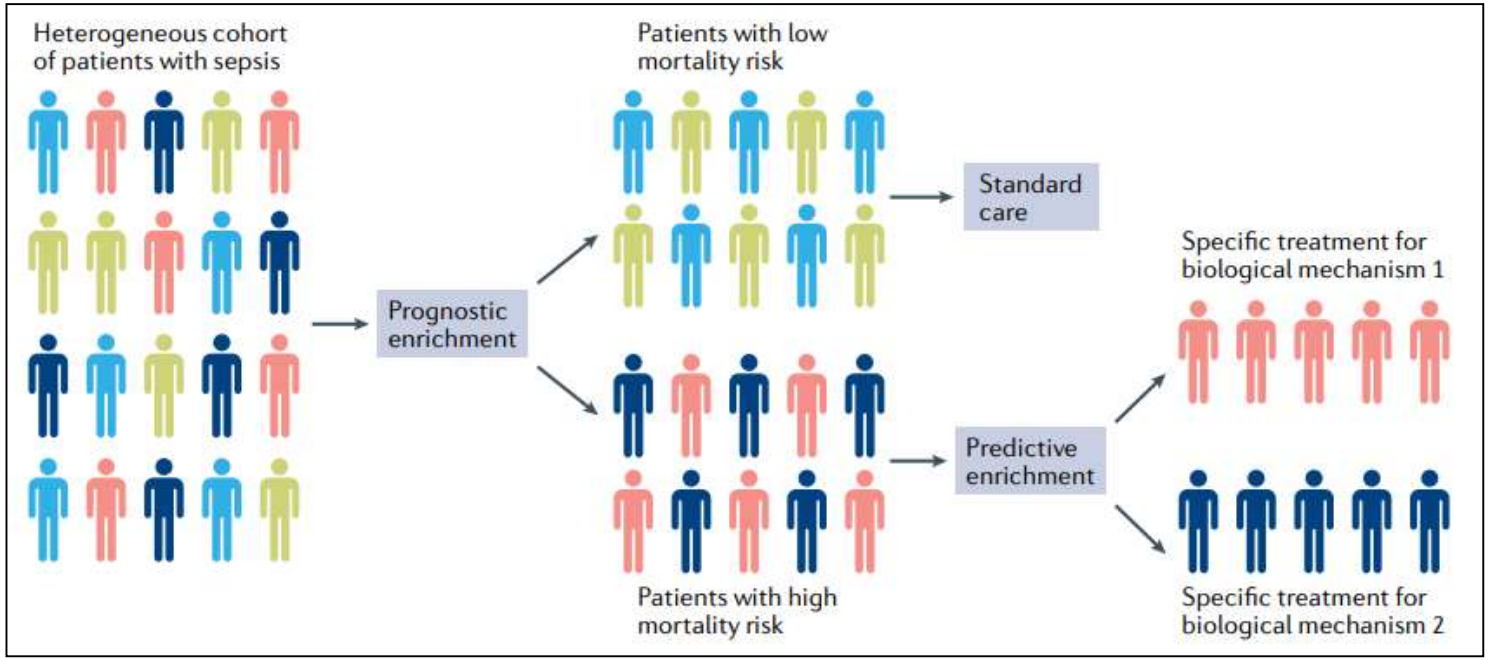

Monneret et al. Cyto A 2019

# Biomarkers needed for precision medicine / individualized therapy

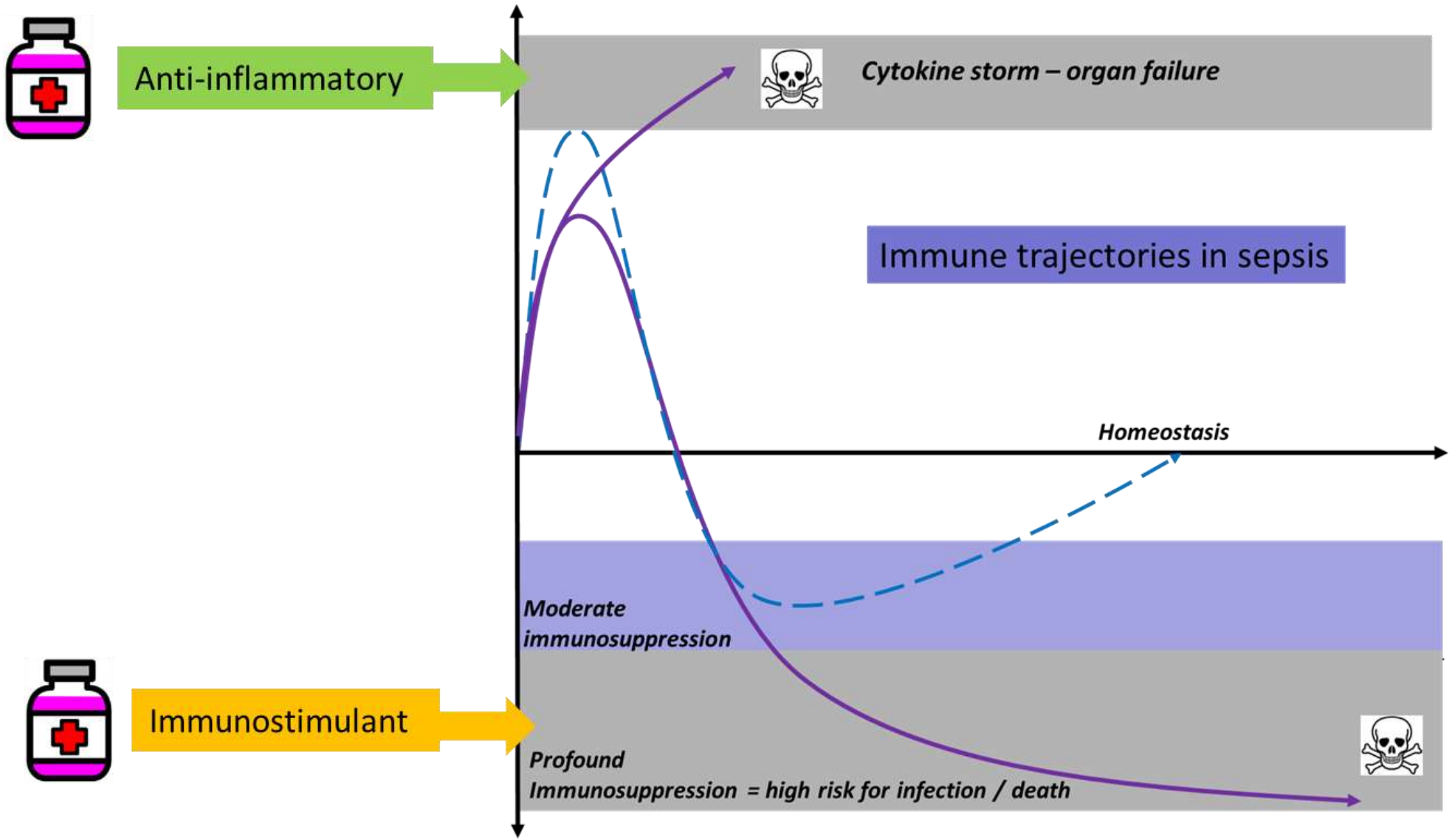
one size does not fit all (at all times)



## Prognostic and predictive enrichment in sepsis

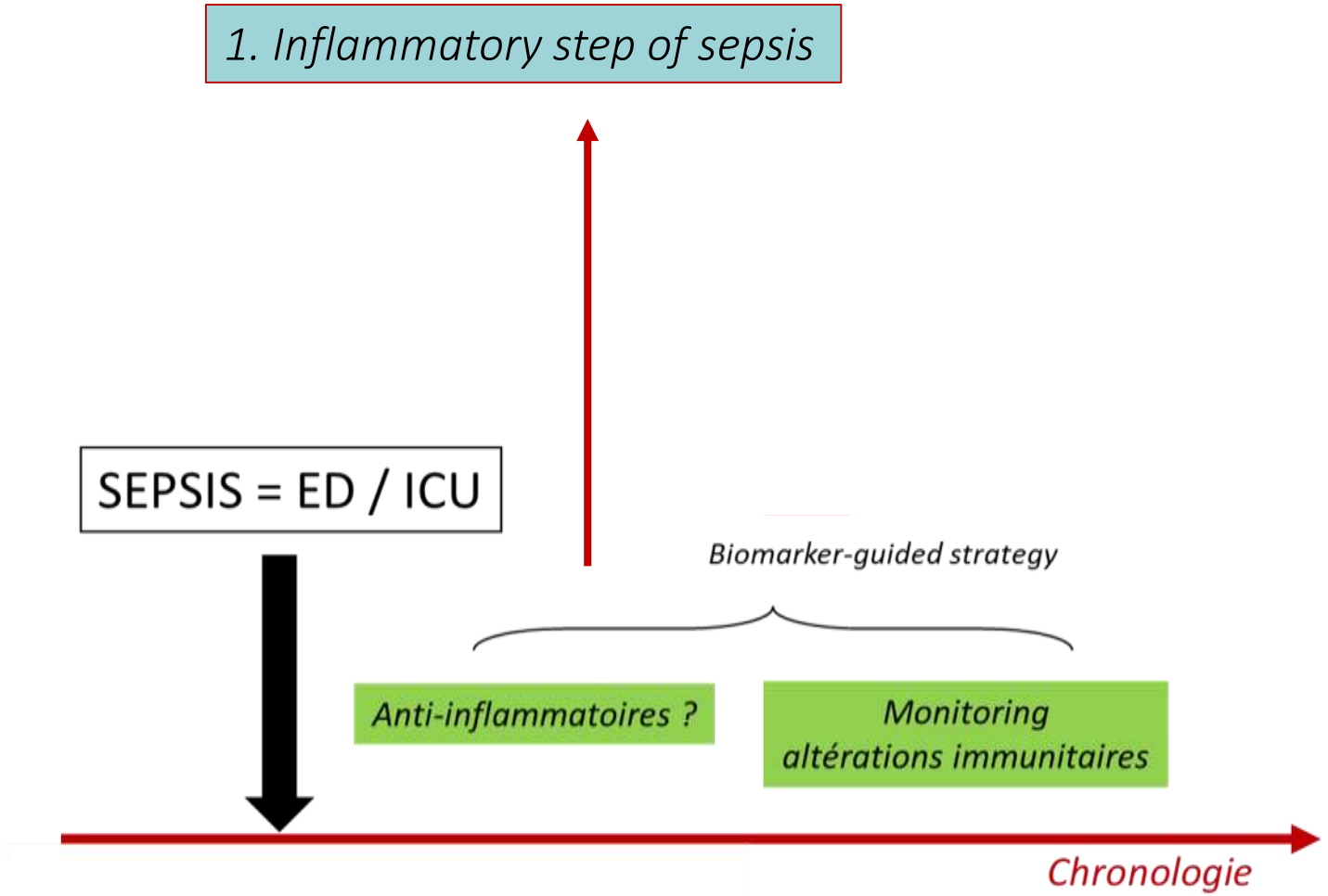



*Stanski & Wong, Nature Rev Nephrol 2020*

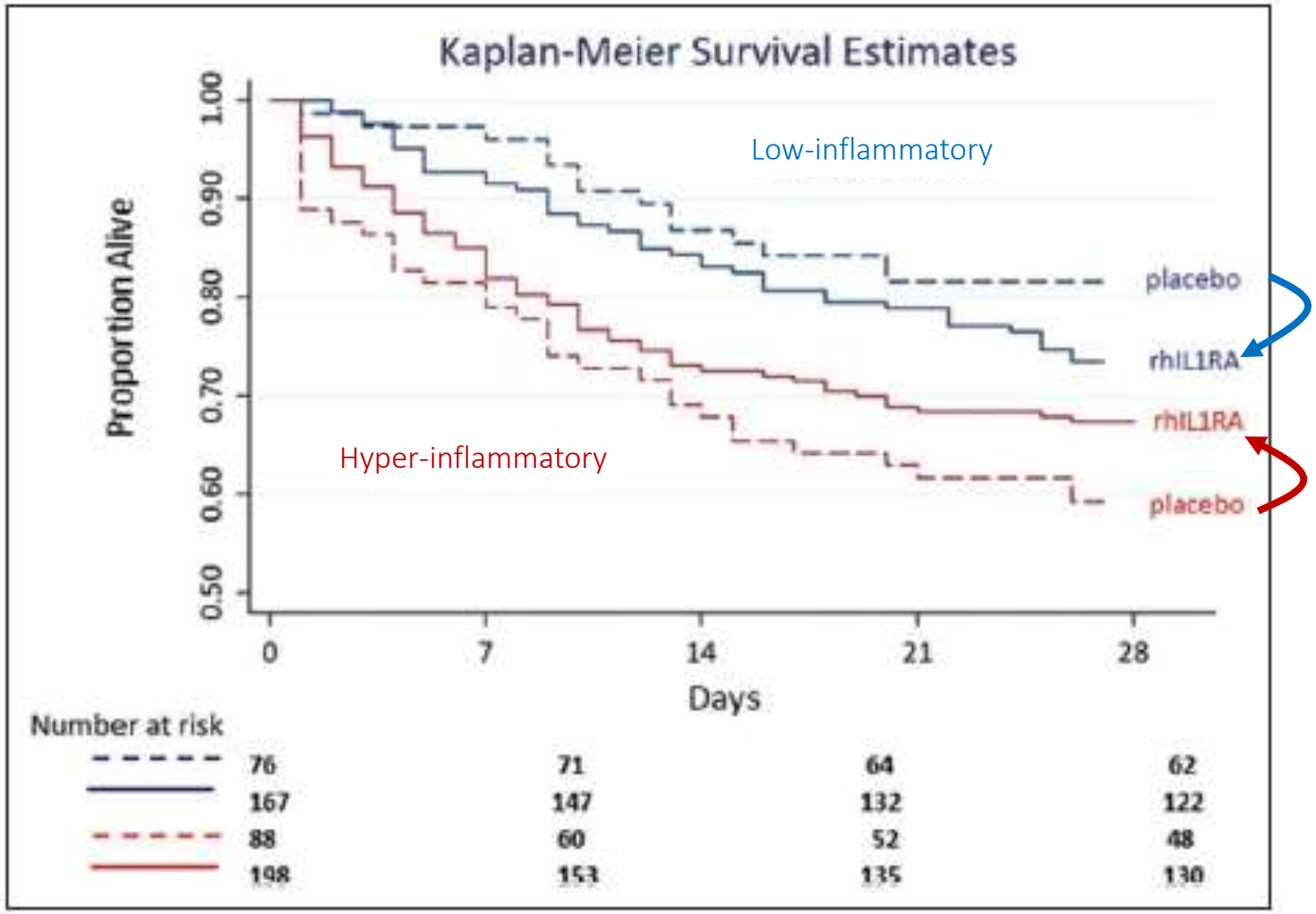



*Monneret et al. Cyto A 2019*

# Individualized immunotherapy in sepsis



# This Approach Yields Results: Post-hoc Analysis (IL1-RA)




*Meyer NL et al., CCM 2017*

# This Approach Yields Results: Post-hoc Analysis (IL-6)

## IL-6 serum levels predict severity and response to tocilizumab in COVID-19: An observational study

José María Galván-Román, MD,[a]* Sebastián C. Rodríguez-García, MD,[b]* Emilia Roy-Vallejo, MD,[a] Ana Marcos-Jiménez, MSc,[c] Santiago Sánchez-Alonso, MSc,[c] Carlos Fernández-Díaz, MD,[b] Ana Alcaraz-Serna, MD,[c] Tamara Mateu-Albero, MSc,[c] Pablo Rodríguez-Cortés, MD,[a] Ildefonso Sánchez-Cerrillo, MD,[c] Laura Esparcia, BSc,[c] Pedro Martínez-Fleta, MSc,[c] Celia López-Sanz, BSc,[c] Ligia Gabrie, MD,[c] Luciana del Campo Guerola, MD,[c] Carmen Suárez-Fernández, MD, PhD,[a] Julio Ancochea, MD, PhD,[d] Alfonso Canabal, MD, PhD,[e] Patricia Albert, MD, PhD,[e] Diego A. Rodríguez-Serrano, MD, PhD,[e] Juan Mariano Aguilar, MD, PhD,[f] Carmen del Arco, MD, PhD,[f] Ignacio de los Santos, MD, PhD,[a] Lucio García-Fraile, MD,[a] Rafael de la Cámara, MD, PhD,[g] José María Serra, Pharm,[h] Esther Ramírez, PharmD,[h] Tamara Alonso, MD,[d] Pedro Landete, MD, PhD,[d] Joan B. Soriano, MD, PhD,[d] Enrique Martín-Gayo, PhD,[c] Arturo Fraile Torres, Pharm,[i] Nelly Daniela Zurita Cruz, Pharm,[i] Rosario García-Vicuña, MD, PhD,[b] Laura Cardeñoso, MD, PhD,[f] Francisco Sánchez-Madrid, PhD,[c,j] Arantzazu Alfranca, MD, PhD,[c]‡ Cecilia Muñoz-Calleja, MD, PhD,[c]‡ and Isidoro González-Álvaro, MD, PhD,[b]‡ on behalf of the REINMUN-COVID Group§ *Madrid, Spain*

**Results: One hundred forty-six patients were studied, predominantly males (66%); median age was 63 years. Forty-four patients (30%) required IMV, and 58 patients (40%) received treatment with TCZ. IL-6 levels greater than 30 pg/mL was the best predictor for IMV (odds ratio, 7.1; $P < .001$). Early administration of TCZ was associated with improvement in oxygenation (arterial oxygen tension/fraction of inspired oxygen ratio) in patients with high IL-6 ($P = .048$). Patients with high IL-6 not treated with TCZ showed high mortality (hazard ratio, 4.6; $P = .003$), as well as those with low IL-6 treated with TCZ (hazard ratio, 3.6; $P = .016$). No relevant serious adverse events were observed in TCZ-treated patients.**

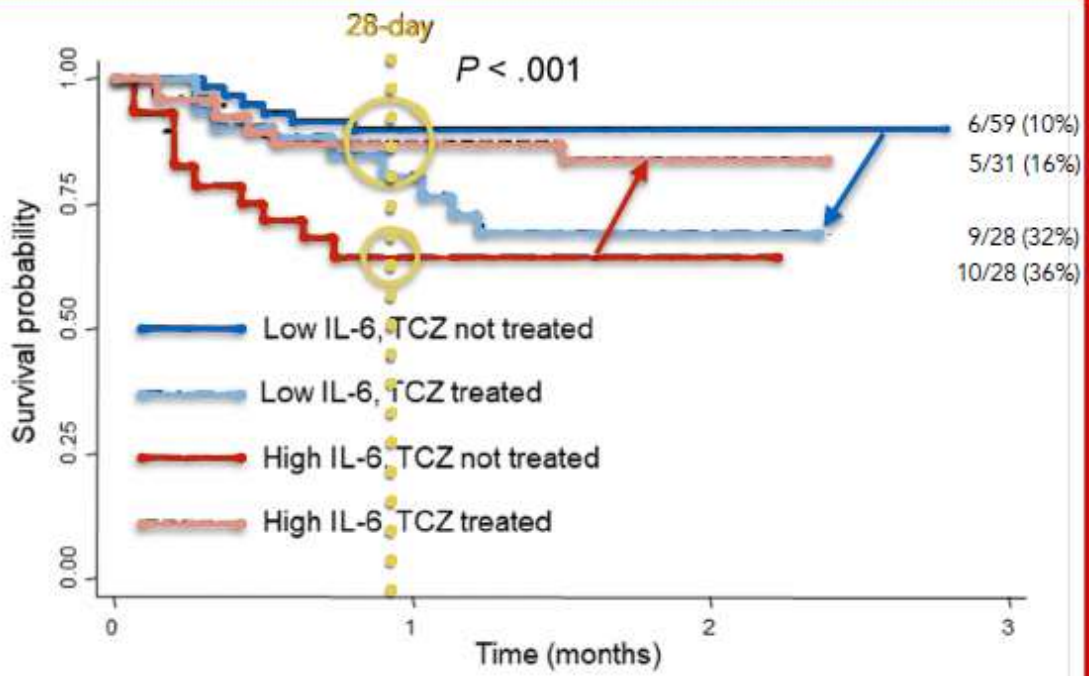


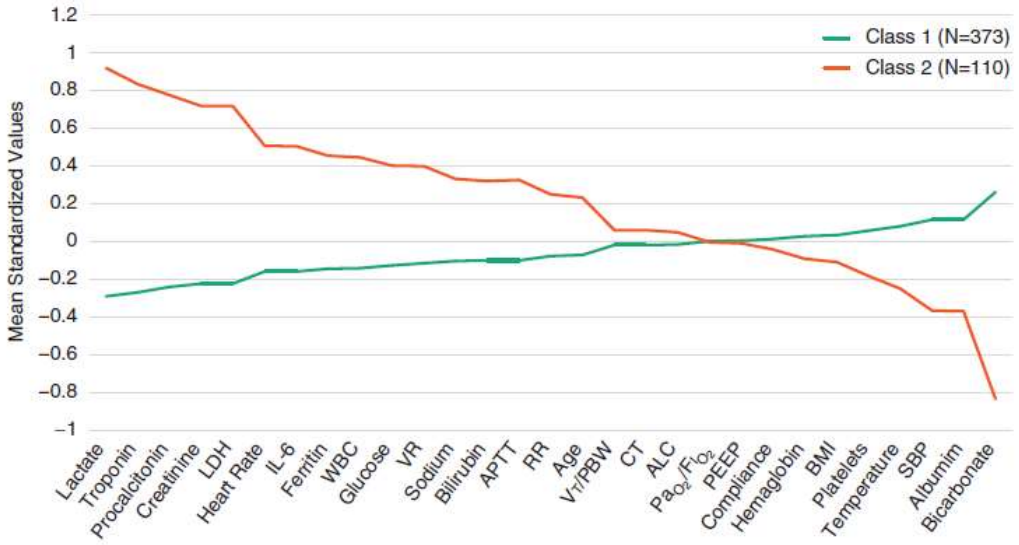

**FIG 4.** Survival curves of patients with COVID-19 grouped according to baseline IL-6 levels and TCZ treatment.

(J Allergy Clin Immunol 2021;147:72-80.)

# The « hyper-inflammatory » phenotype (latent class analysis – Calfee's group)

*These phenotypes, termed "Hyperinflammatory" and "Hypoinflammatory" based on patterns of inflammatory plasma cytokines have widely divergent clinical features, including significantly different clinical outcomes, and respond differently to therapies including mechanical ventilation, fluid therapy, simvastatin, and corticosteroids*

## Latent Class Analysis Reveals COVID-19–related Acute Respiratory Distress Syndrome Subgroups with Differential Responses to Corticosteroids



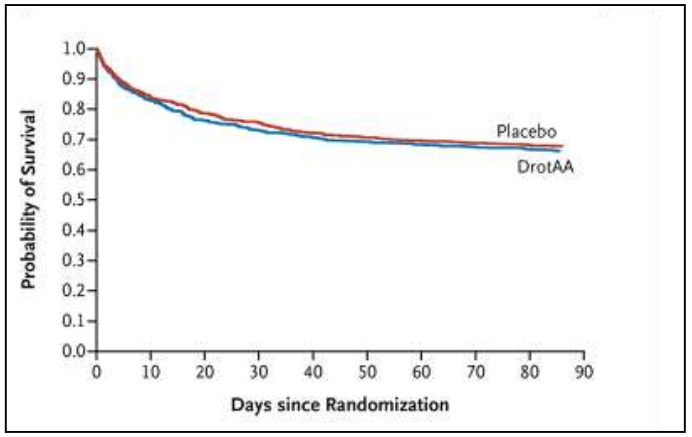

| Corticosteroid Use | COVID-19–related ARDS Class | |
|---|---|---|
| | Class 1 | Class 2 |
| Yes | 120/244 (49%) | 52/76 (68%) |
| No | 58/127 (46%) | 30/34 (88%) |

*p = 0,027*

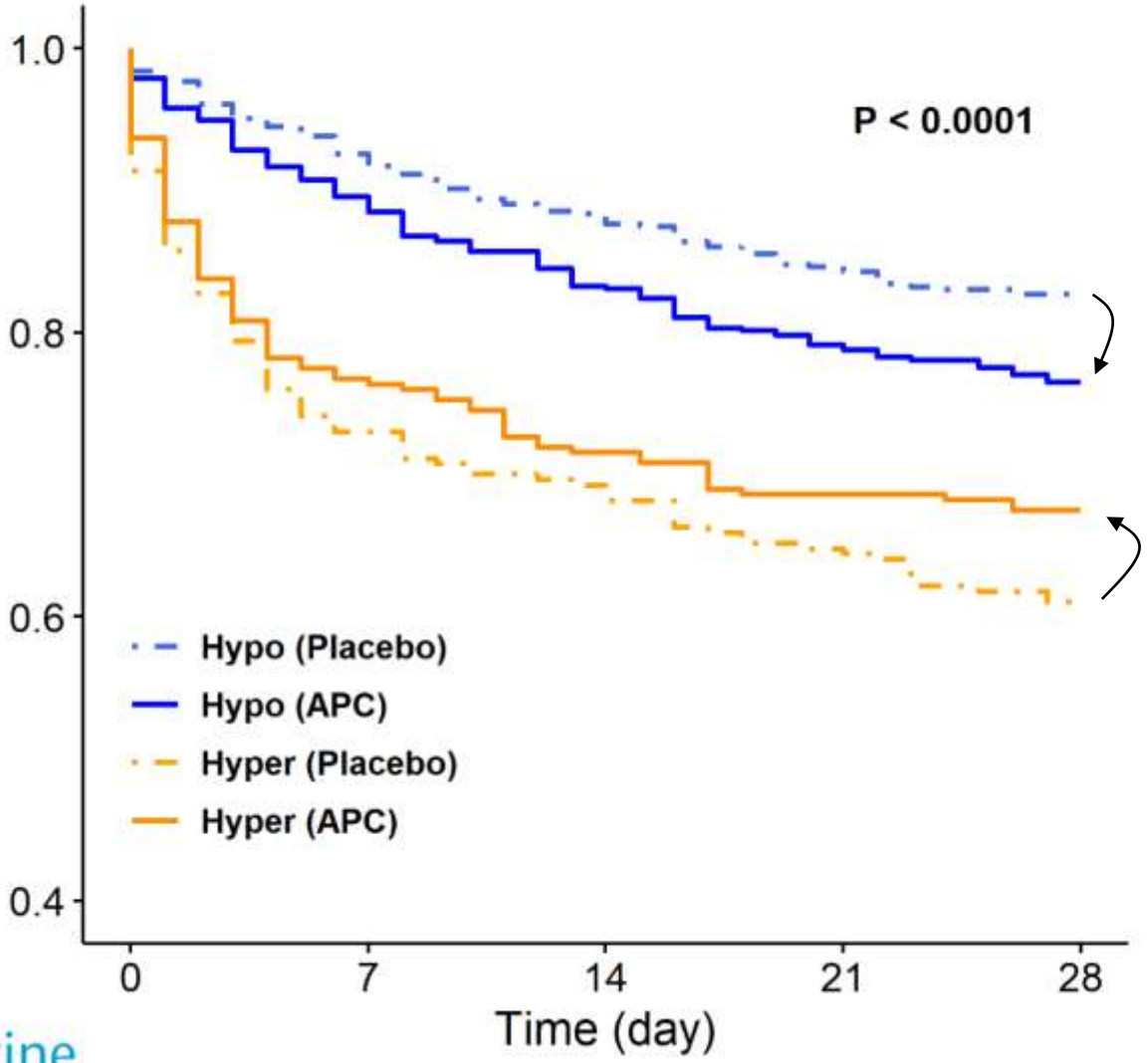
*Sinha P et al., AJRCCM 2021*

The « hyper-inflammatory » phenotype (latent class analysis – Calfee's group)

The NEW ENGLAND JOURNAL of MEDICINE 2012




Identifying molecular phenotypes in sepsis: an analysis of two prospective observational cohorts and secondary analysis of two randomised controlled trials




*Sinha et al., Aug 2023*

THE LANCET Respiratory Medicine

ARTICLES
https://doi.org/10.1038/s41591-021-01499-z
nature medicine
Check for updates

OPEN

# Early treatment of COVID-19 with anakinra guided by soluble urokinase plasminogen receptor plasma levels: a double-blind, randomized controlled phase 3 trial

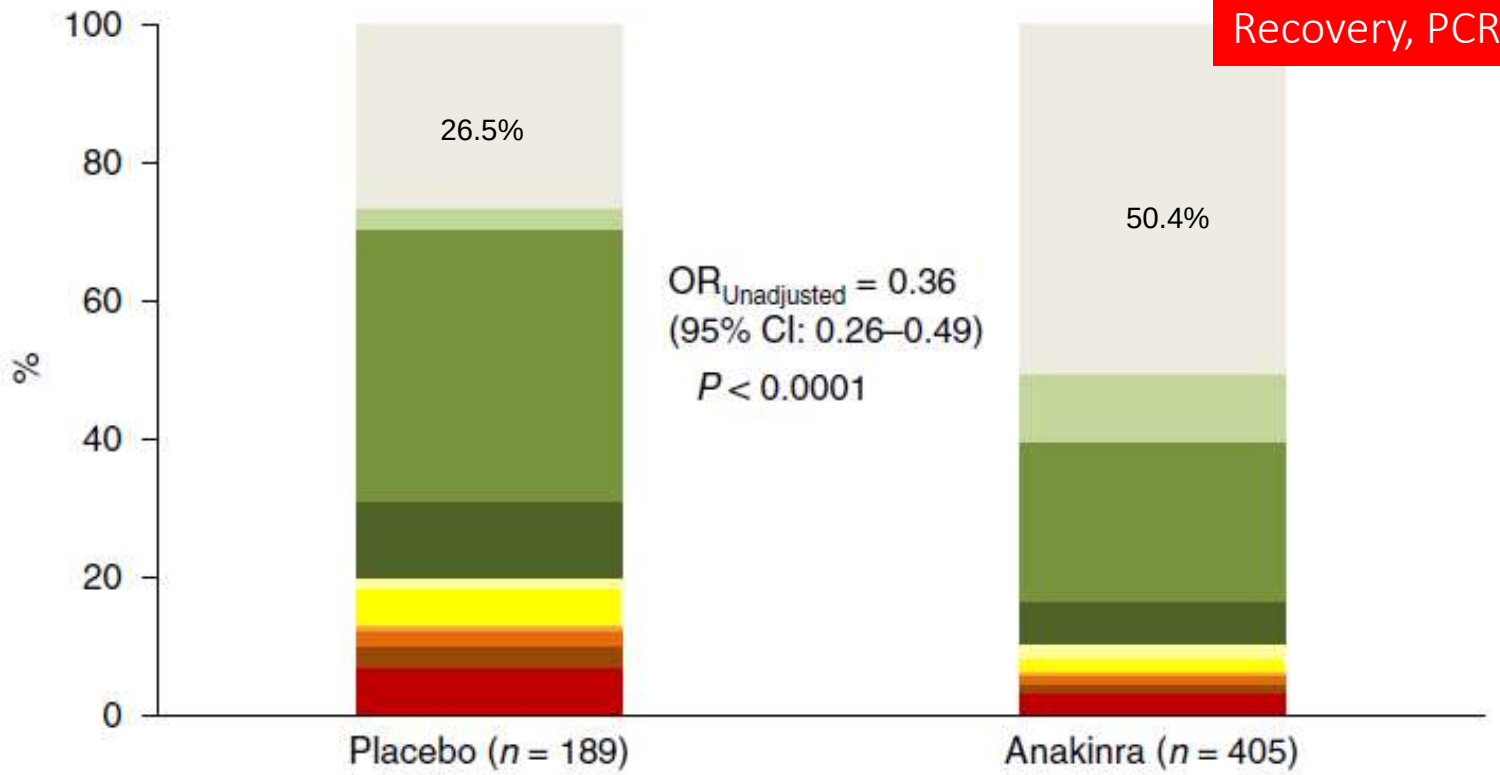
Phase 3 clinical trial

Inclusion criteria :
Hospitalized patients with COVID-19
at risk of progressing to respiratory failure
Plasma suPAR ≥6 ng/mL

Multicentric international (Greece/Italy) :
=> Anakinra vs placebo

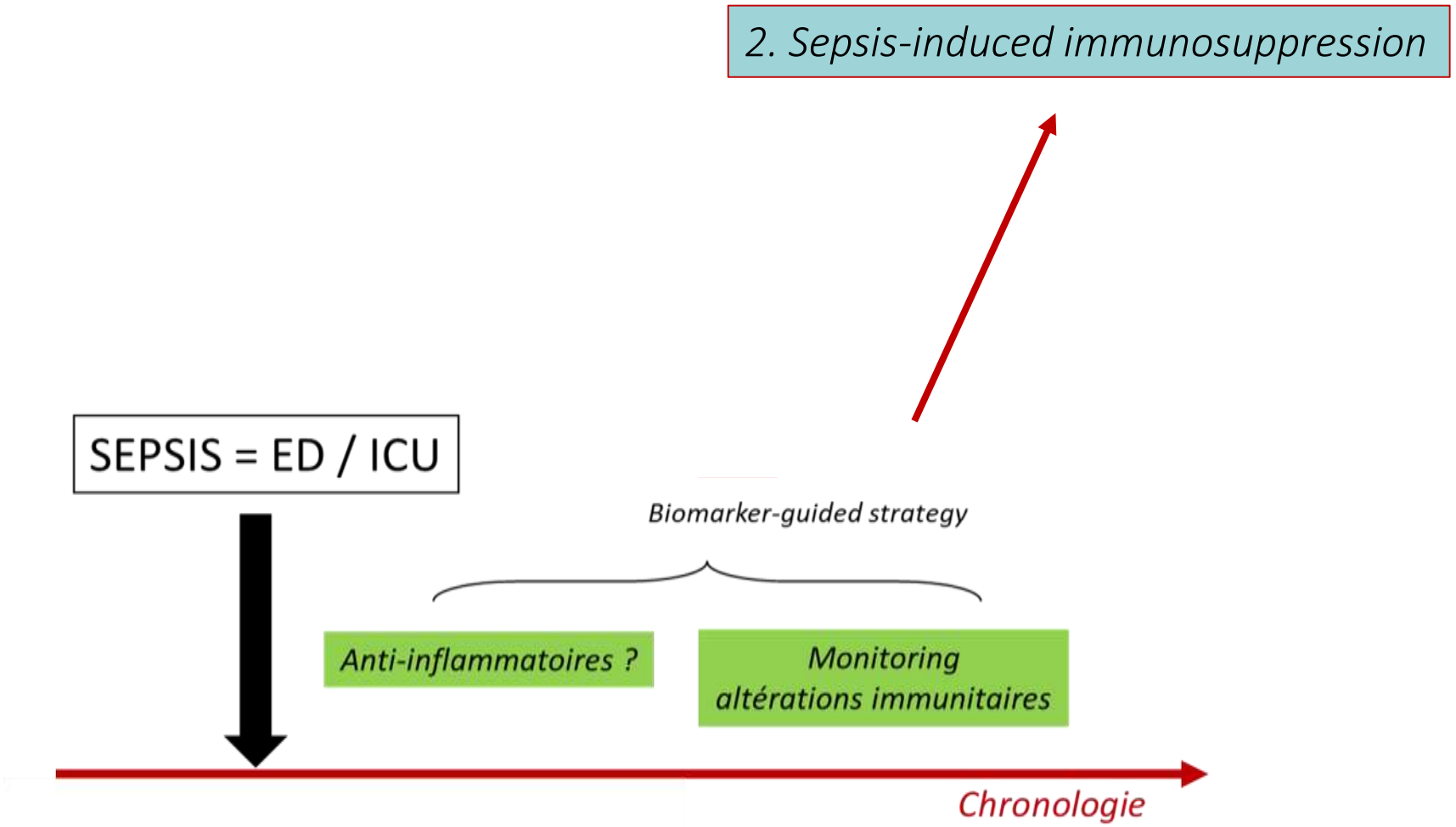
n = 594 patients

Recovery, PCR-




*Kyriazopoulou Nat Med 2021*

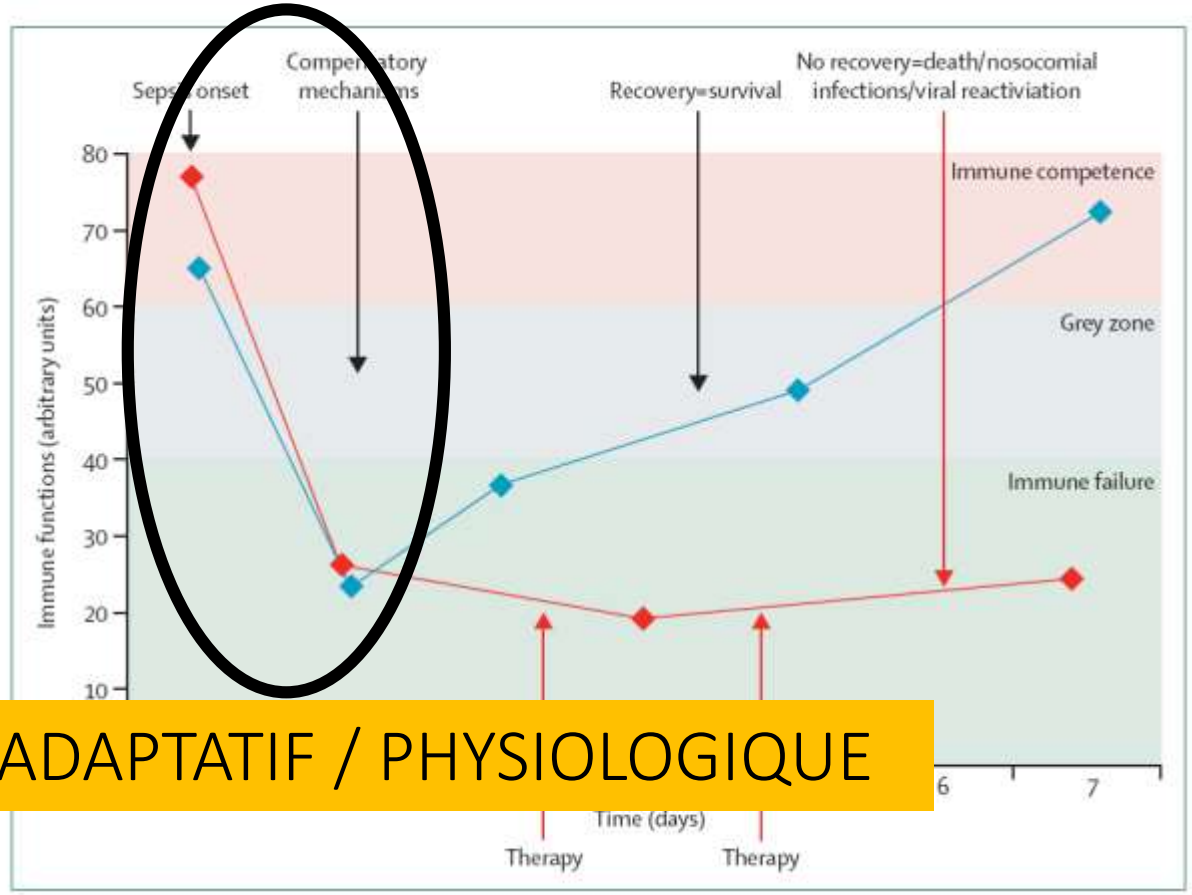
# Individualized immunotherapy in sepsis



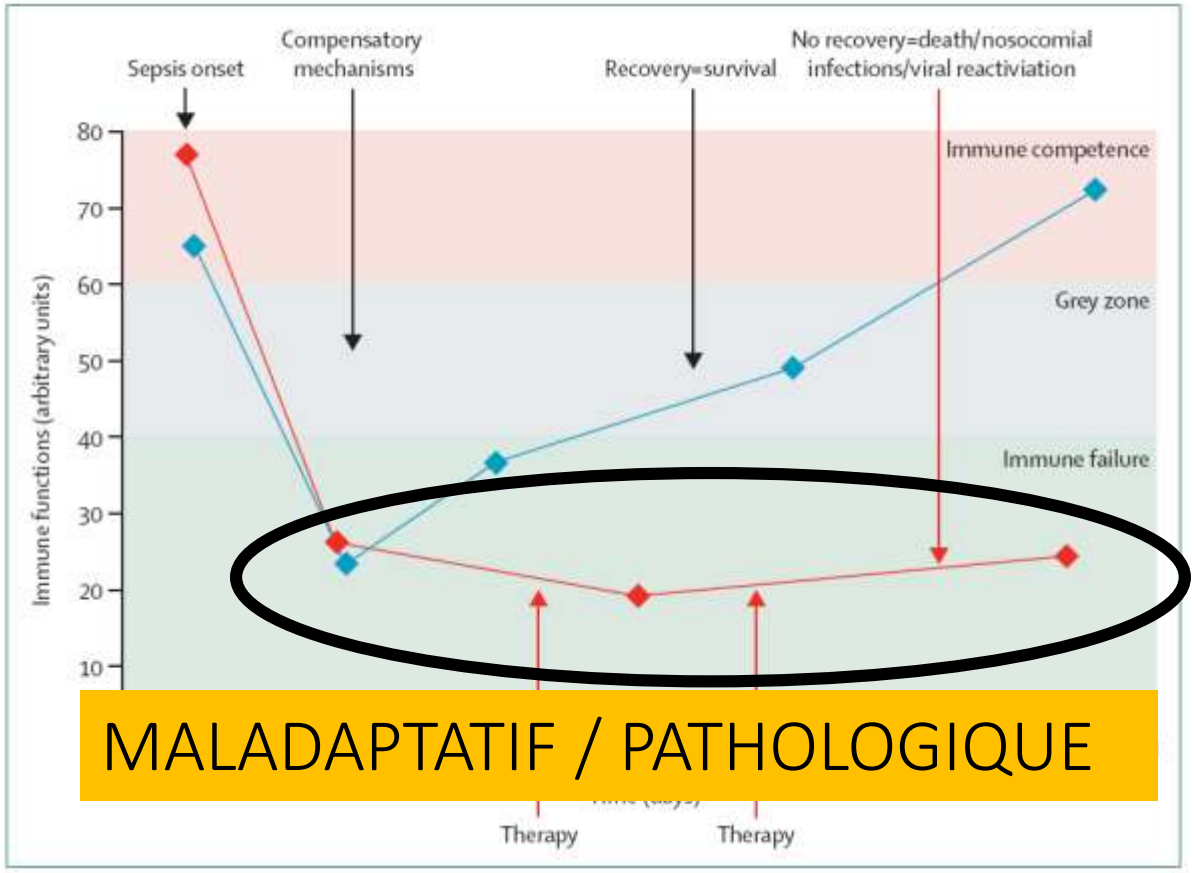
# Immunosuppression in sepsis: a novel understanding of the disorder and a new therapeutic approach

THE LANCET
Infectious Diseases
2013

*Richard S Hotchkiss, Guillaume Monneret, Didier Payen*



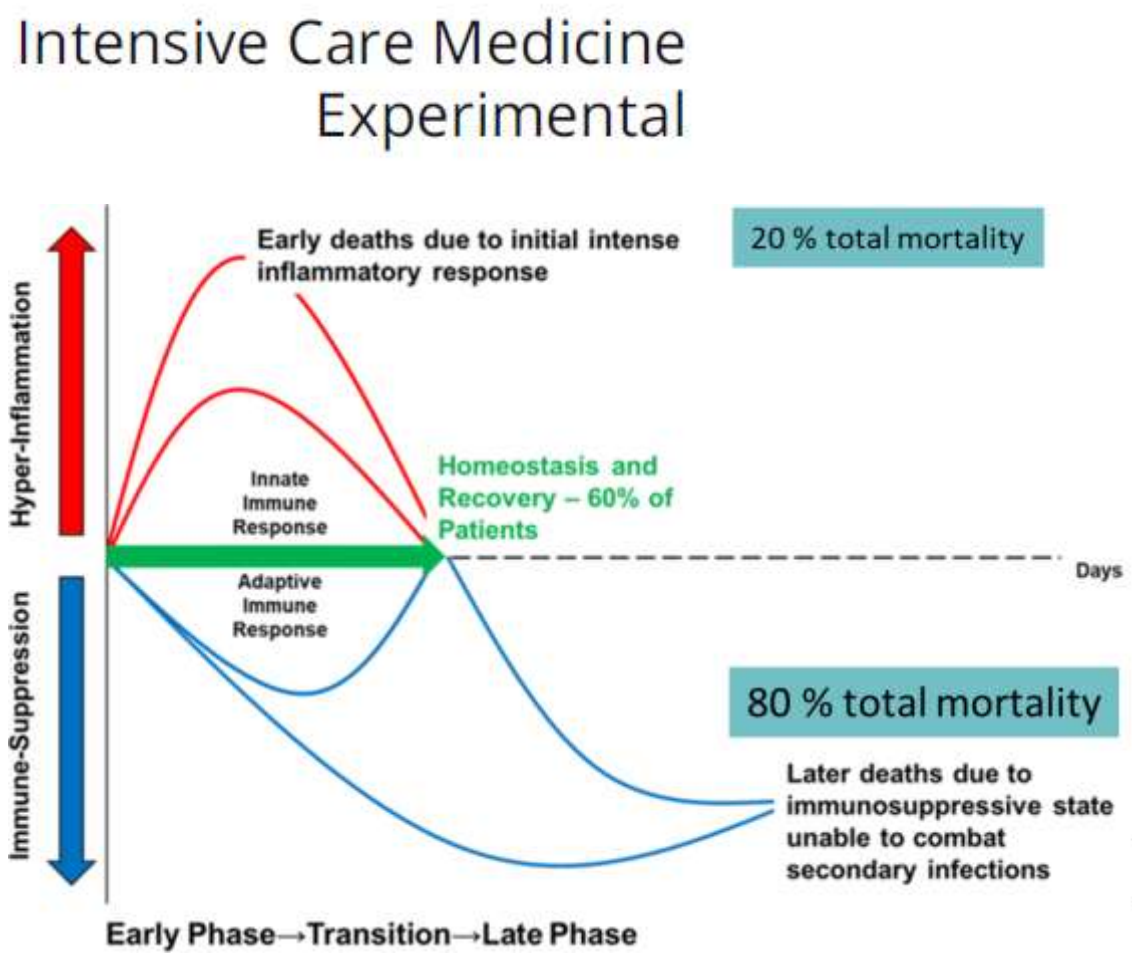

ADAPTATIF / PHYSIOLOGIQUE



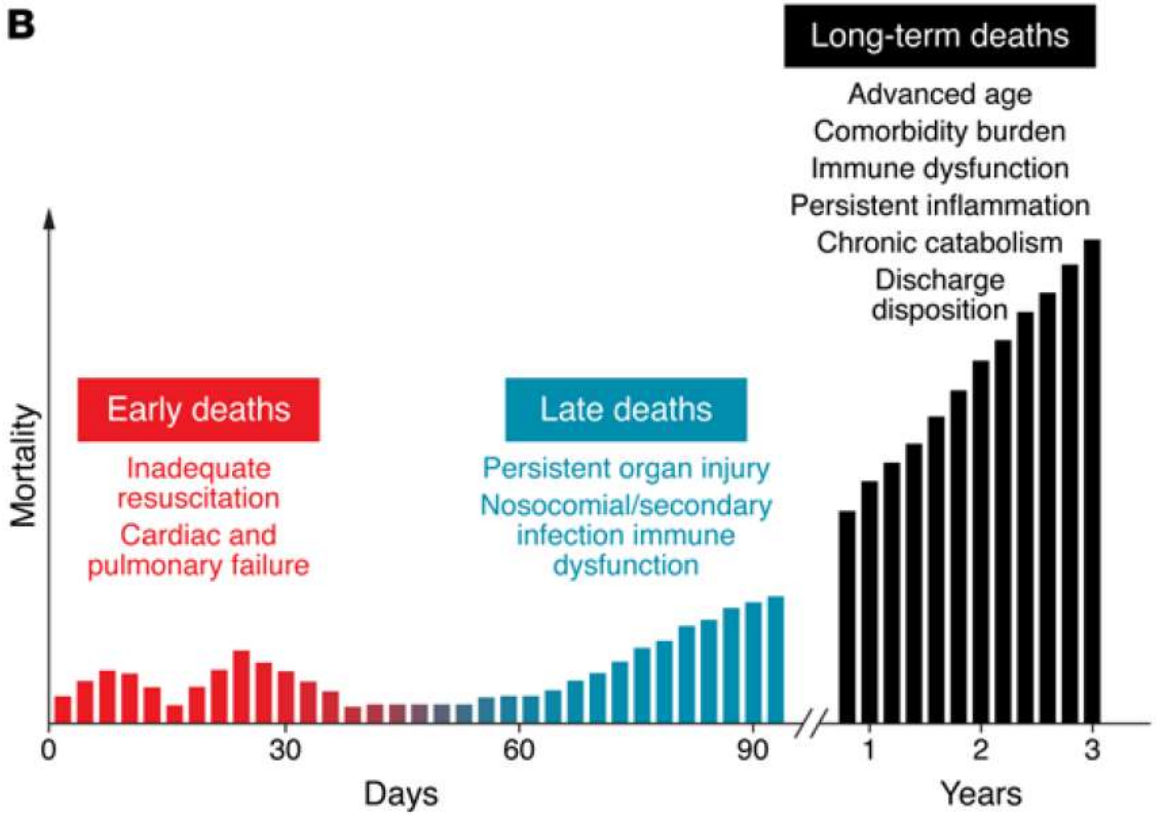

MALADAPTATIF / PATHOLOGIQUE

# Sepsis mortality overtime

2 years mortality : + 20 % / infection = first cause
Sepsis survivors : 60 % 1-year rehospitalisation
First cause : sepsis or significant unresolved infection

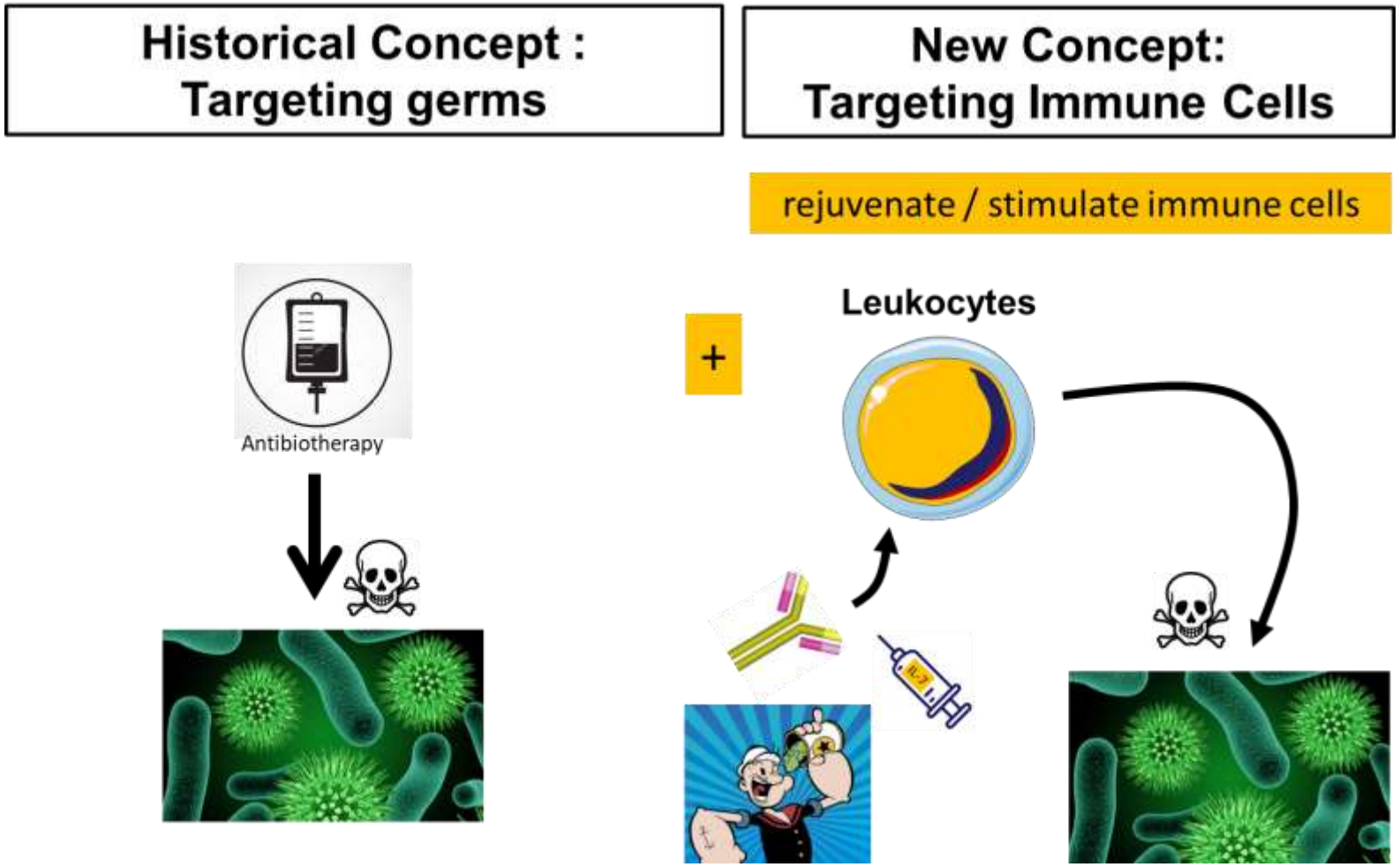



*Brady et al., 2020*




Delano and ward, J Clin Invest 2016, Shankar-Hari and Rubenfeld, Curr Infect Dis Resp 2016, Kennelly and martin-Loevhes, Ann Transl Med 2016, Kaur and Levy, Ann Transl Med 2016, Prescott et al., BMJ 2016, Sun et al., CCM 2016, Pavon et al., CCM 2014, Wabng et al BMJ 2014

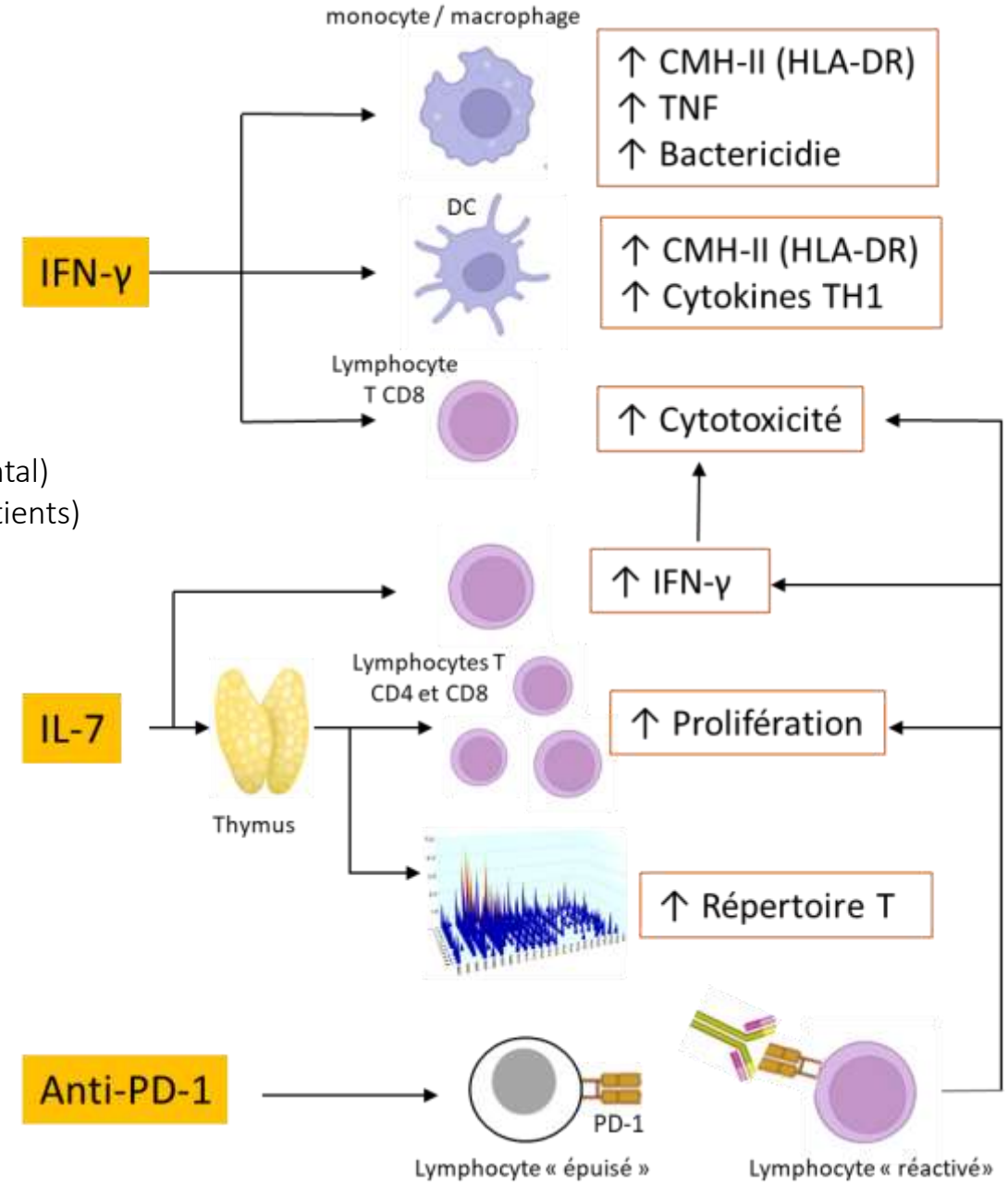
# Principles of adjuvant immunotherapy in sepsis



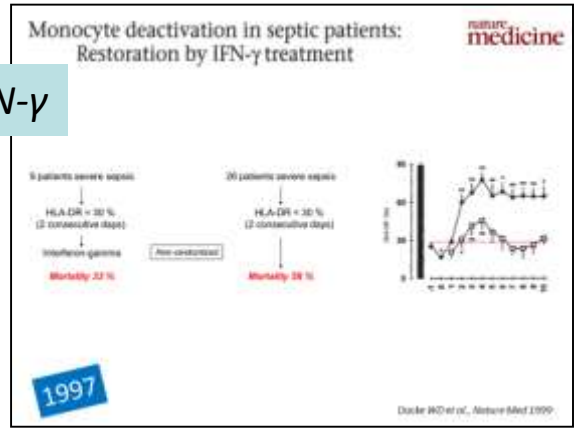
# Immunotherapy in sepsis = current candidates

RCT

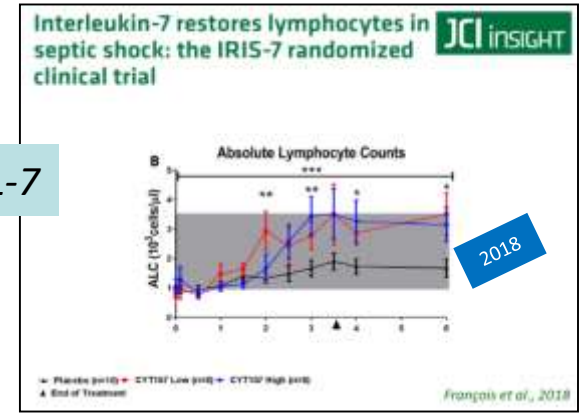
+ > 30 articles preclinical results (experimental)
+ > 20 articles preclinical results (ex vivo patients)
+ > 50 clinical cases or series (either drug)



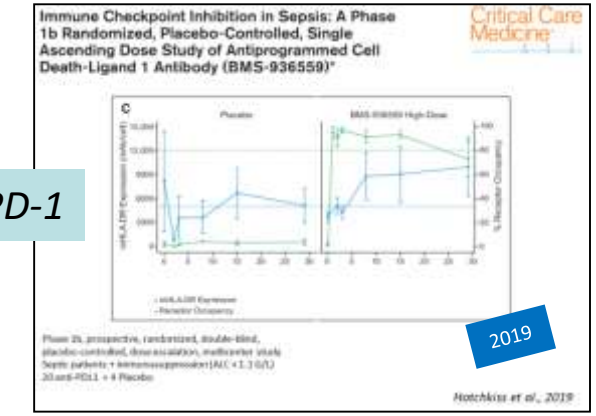

*IFN-γ*




*IL-7*




*Anti-PD-1*



# Awaiting news from large RCT => rescue therapy in the most immunosuppressed patients

Available tools at immunology lab for routine care

Severe ICU patients
Weeks in ICU
Still worsening
Lack of infection control

*Check-up of immune functions*

Innate immunity
- immature neutrophils (CD16low) = ↑
- Deactivated monocyte (mHLA-DR) = ↓

Adaptive immunity
- Lymphocyte count (+ subsets) =↓
- Check point inhibitor (PD-1) = ↑


Accredited Laboratory / Fully Certified Laboratory
(in France ISO 15189 accreditation) for routine care

# Interferon gamma as an immune modulating adjunct therapy for invasive mucormycosis after severe burn – A case report

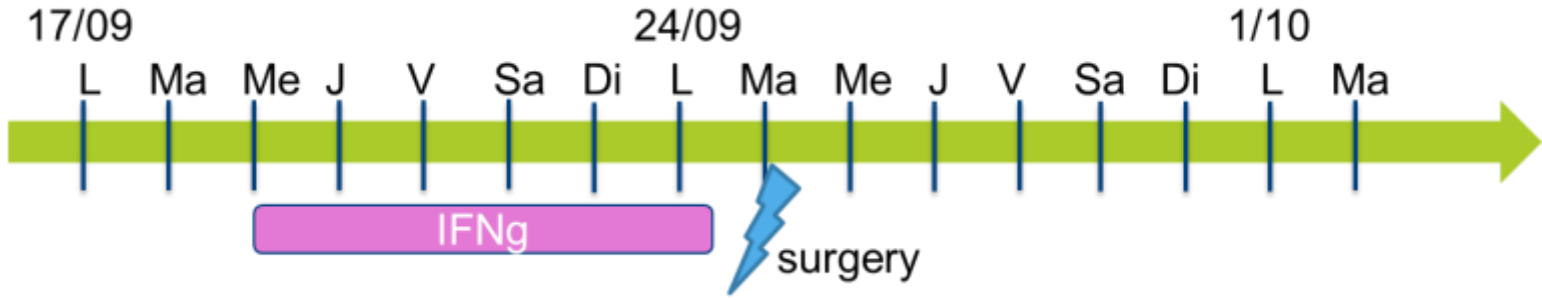
Women, 61 yo, 43% TBSA deep burns

Peri-stomial ulcerations => diagnosis of mucormycosis (infection by *Rhizopus microsporus*)

23/08 06/09 11/09

Antifungal treatment with:

- I.V. Ambisome
- p.o. posaconazole
- local instillation fluconazole




Surgical resection with carcinologic margins
standard treatment

*Tawik et al., 2022*

Frontiers in Immunology

# After IMUKIN => Surgery (day 40)


ED –

Culture –

Pan-fungal PCR –

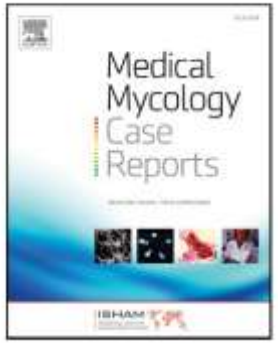
Mucorales PCR –

4 months later

*Tawik et al., 2022*

Frontiers in Immunology

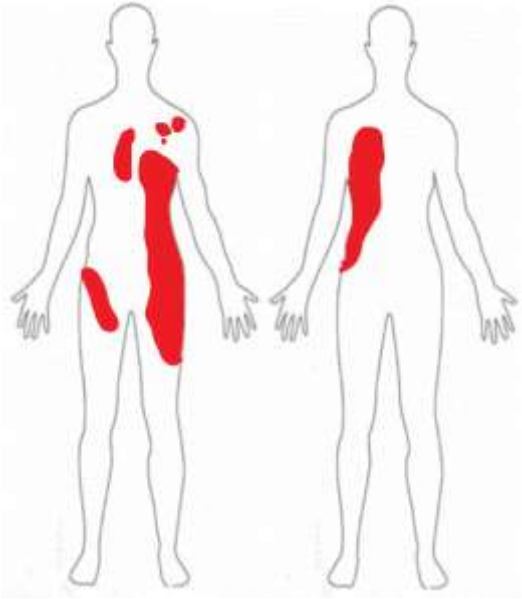
# Nivolumab and interferon-γ rescue therapy to control mixed mould and bacterial superinfection after necrotizing fasciitis and septic shock




Previously healthy 38-year-old female

The patient was admitted (day 0) to a general hospital intensive care unit (ICU) for shock associated with streptococcal necrotising fasciitis of the left chest wall, which was secondary to a minor thoracic trauma 2 days before.

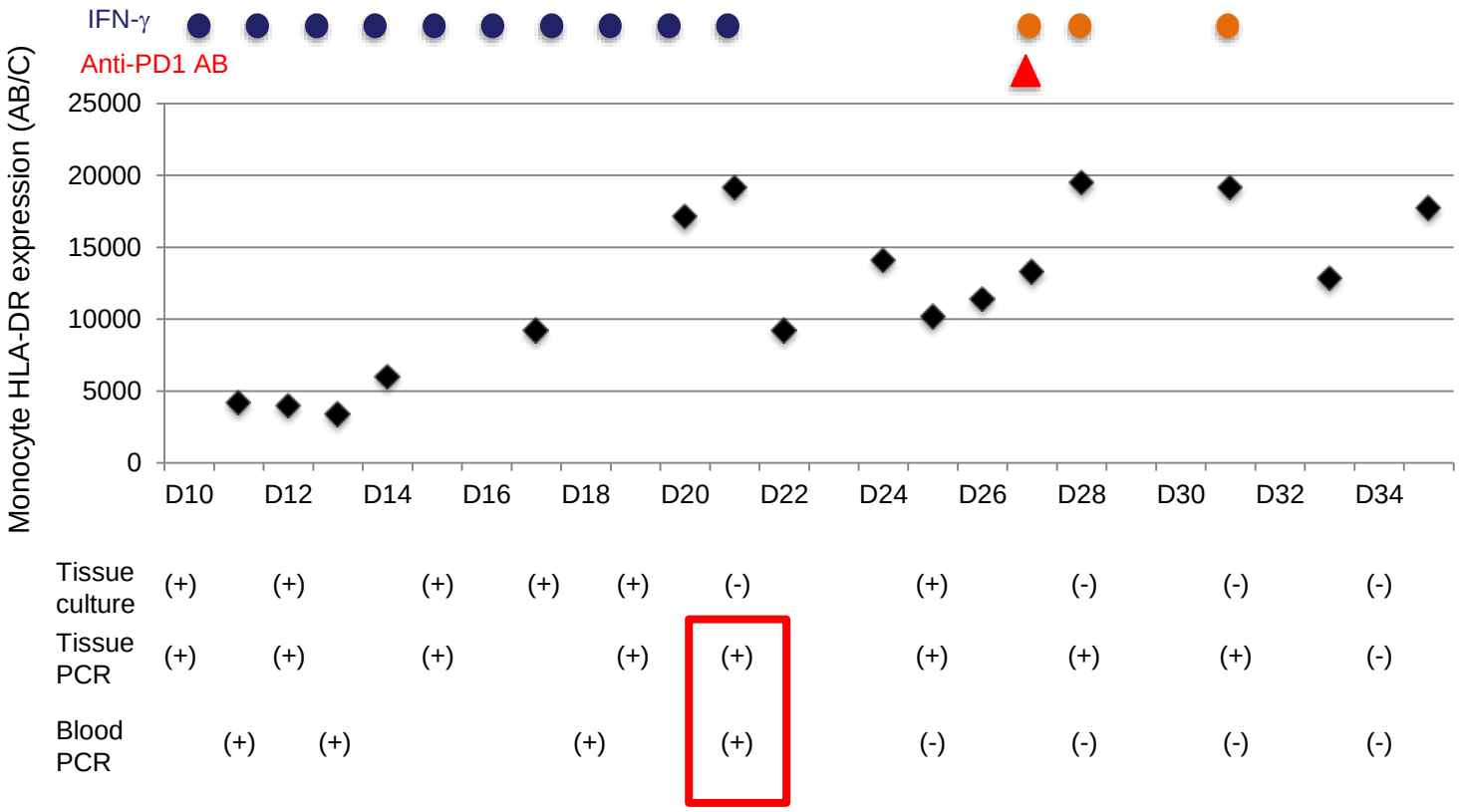
At day 9, transferred to our ICU for hyperbaric oxygenotherapy + surgery
mucormycosis was diagnosed



Lukaszewicz et al., 2022

# Nivolumab and interferon-γ rescue therapy to control mixed mould and bacterial superinfection after necrotizing fasciitis and septic shock

Immumonitoring showed severe lymphopenia and very low mHLA-DR expression
**=> severe immunosuppression**



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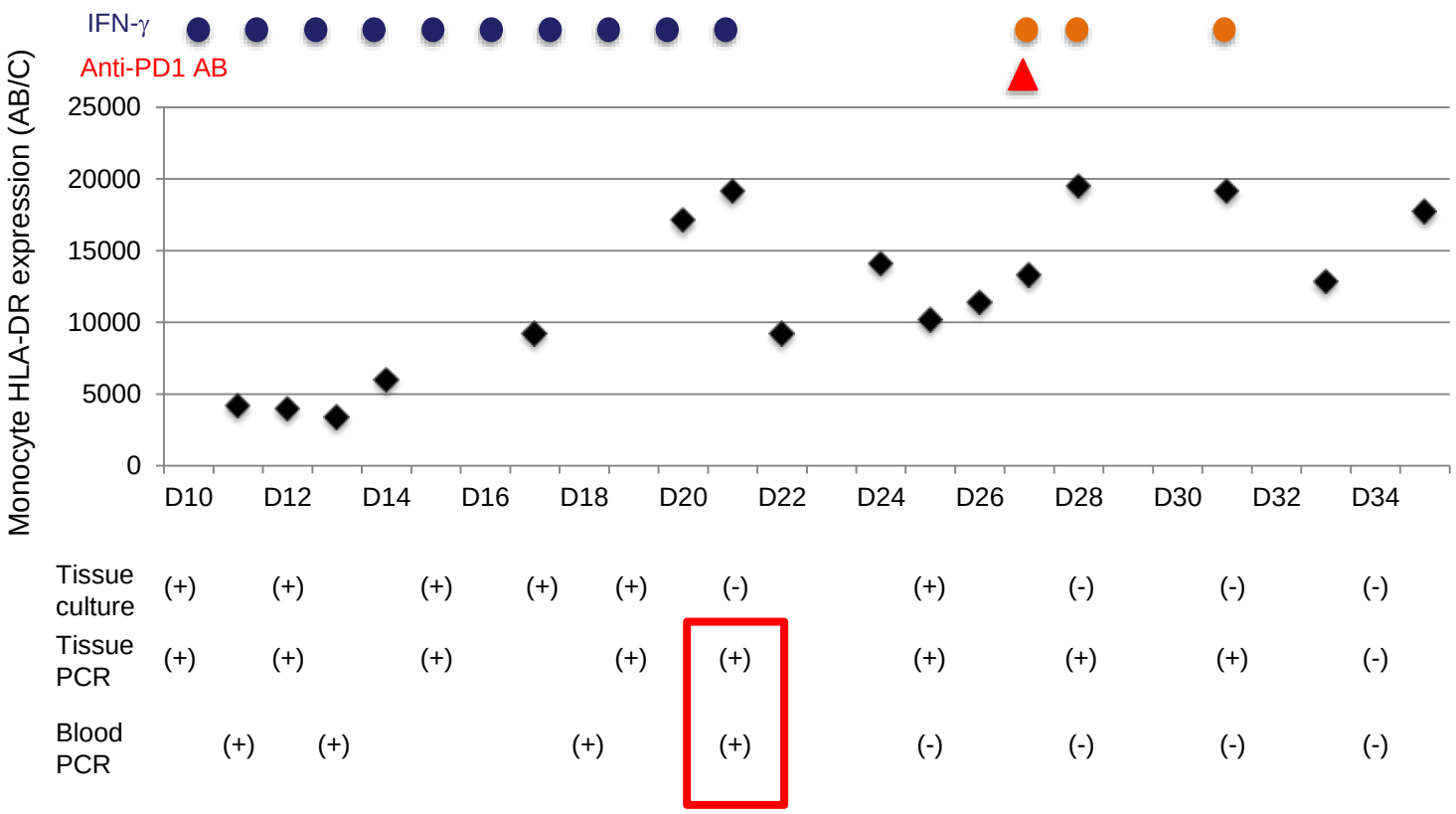
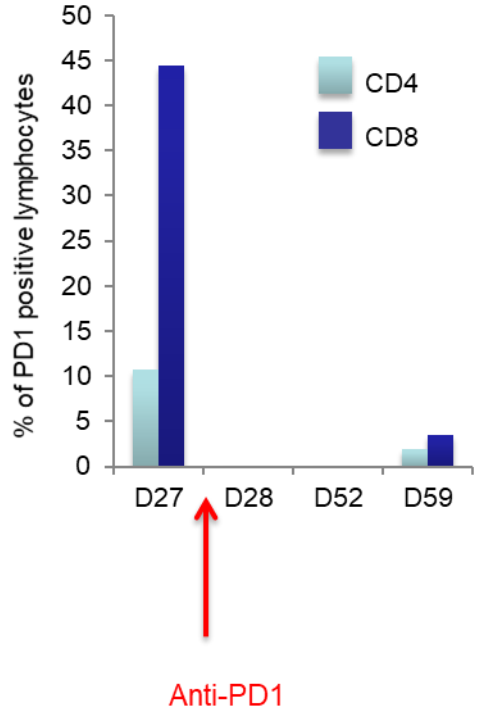
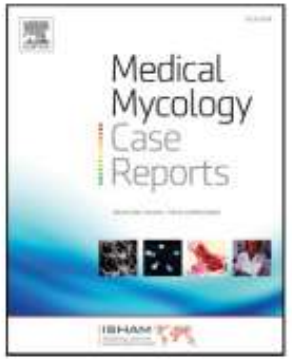


# Nivolumab and interferon-γ rescue therapy to control mixed mould and bacterial superinfection after necrotizing fasciitis and septic shock

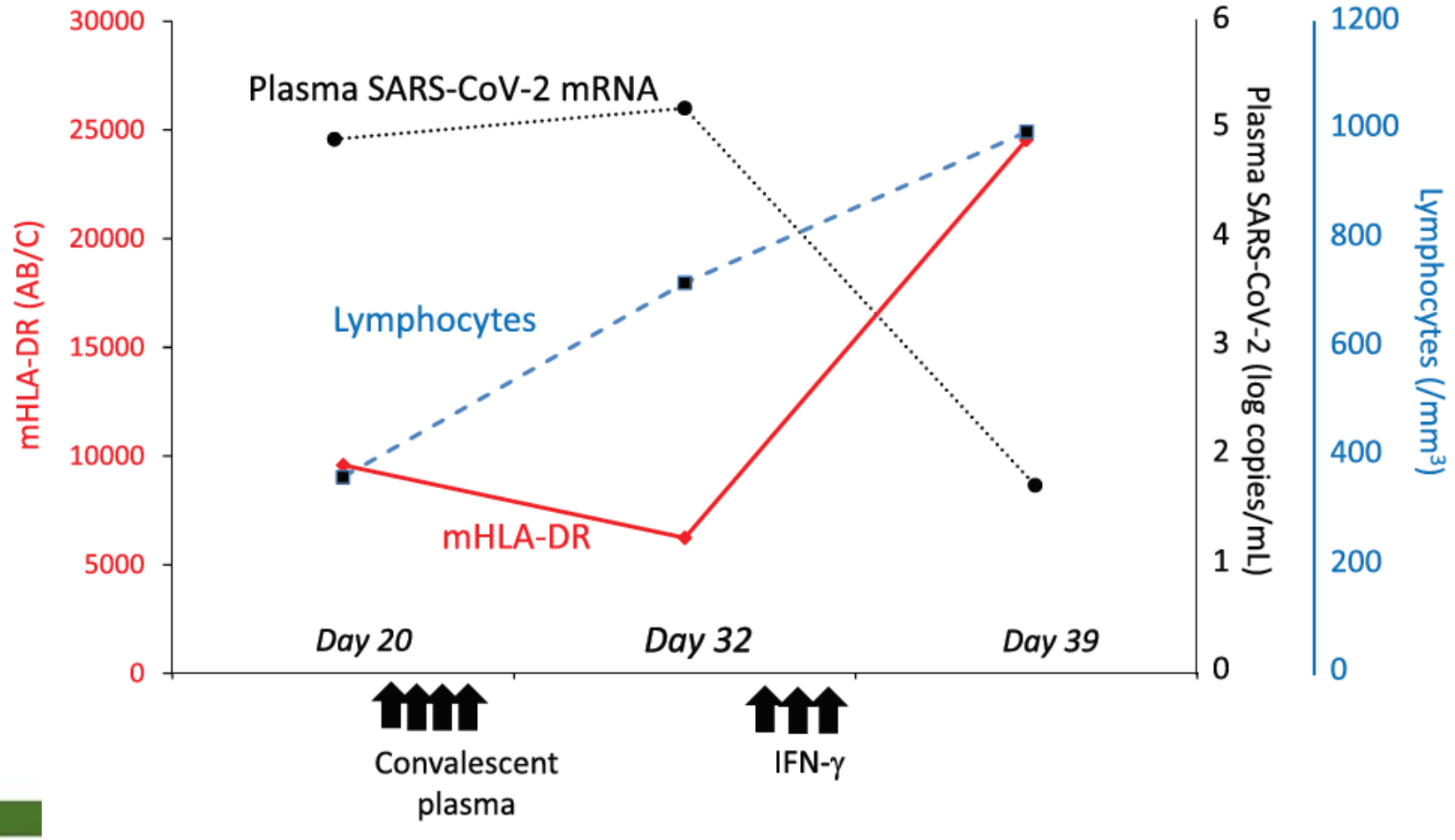
5.5 months later

4 months later




Lukaszewicz et al., 2022

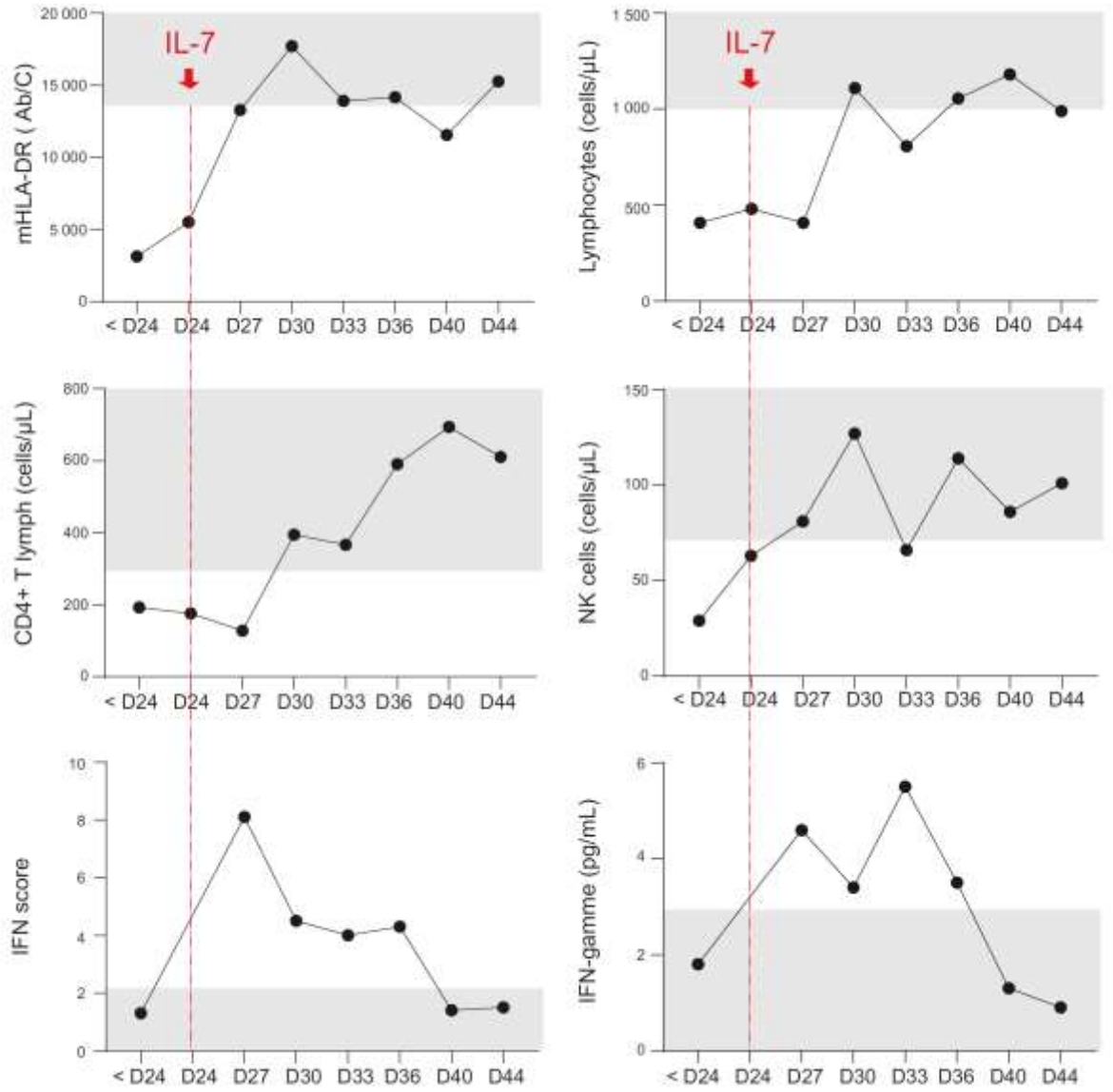
# Immunostimulation with interferon-γ in protracted SARS-CoV-2 pneumonia




*Lukaszewicz et al., 2021*

JOURNAL OF MEDICAL VIROLOGY

# Immune monitoring of interleukin-7 compassionate use in a critically ill COVID-19 patient

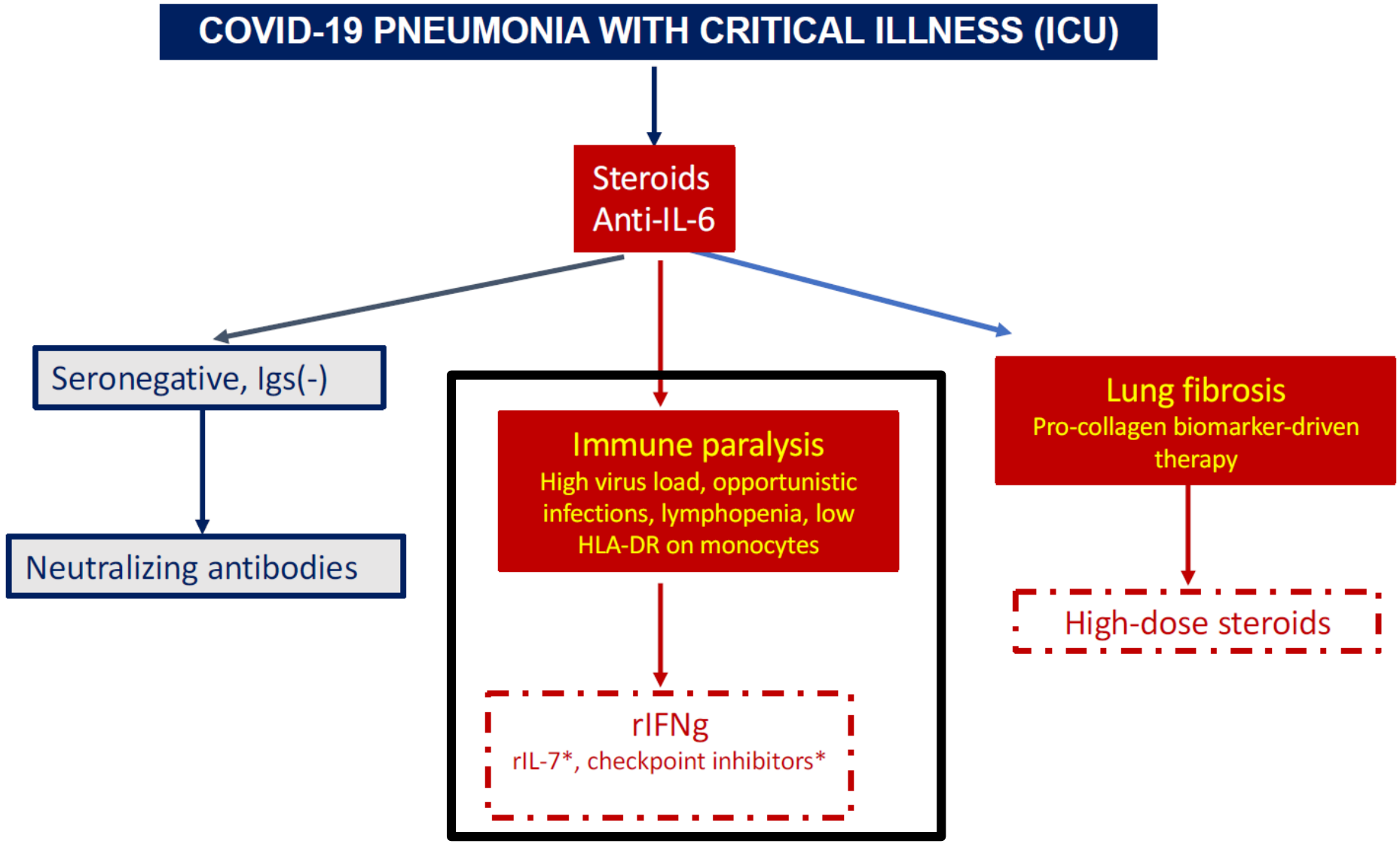



*Monneret et al., 2020*

Cellular & Molecular Immunology

# A guide to immunotherapy for COVID-19

Frank L. van de Veerdonk[1], Evangelos Giamarellos-Bourboulis[2], Peter Pickkers[3], Lennie Derde[4,9], Helen Leavis[5], Reinout van Crevel[1], Job J. Engel[1], W. Joost Wiersinga[6], Alexander P. J. Vlaar[7], Manu Shankar-Hari[8], Tom van der Poll[6], Marc Bonten[9], Derek C. Angus[10], Jos W. M. van der Meer[1] and Mihai G. Netea[1,11]




2022
nature medicine

DÉPÊCHE - Jeudi 06 janvier 2022 - 18:10

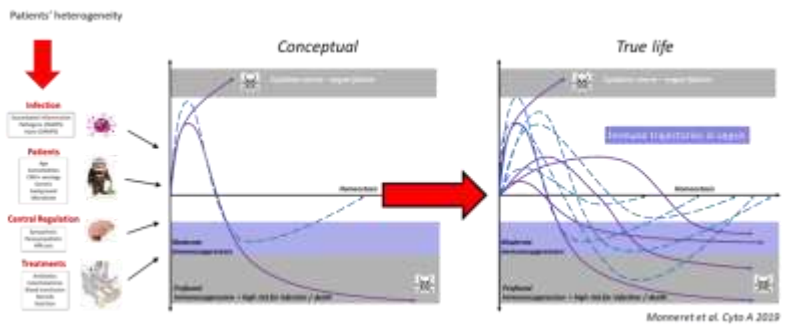
# Un effort "majeur" reste à faire pour individualiser le traitement du Covid sévère

Le Pr Timsit a rappelé lors de son audition les différentes phases de la maladie: la partie virale initiale "pas très sévère", la partie immunologique qui a un effet délétère "considérable" sur l'organisme et, chez les patients les plus sévères, la phase "d'immuno-paralysie post-agressive" caractérisée par des surinfections et des complications infectieuses qui dégradent lourdement le pronostic des malades, en particulier les plus sévères.

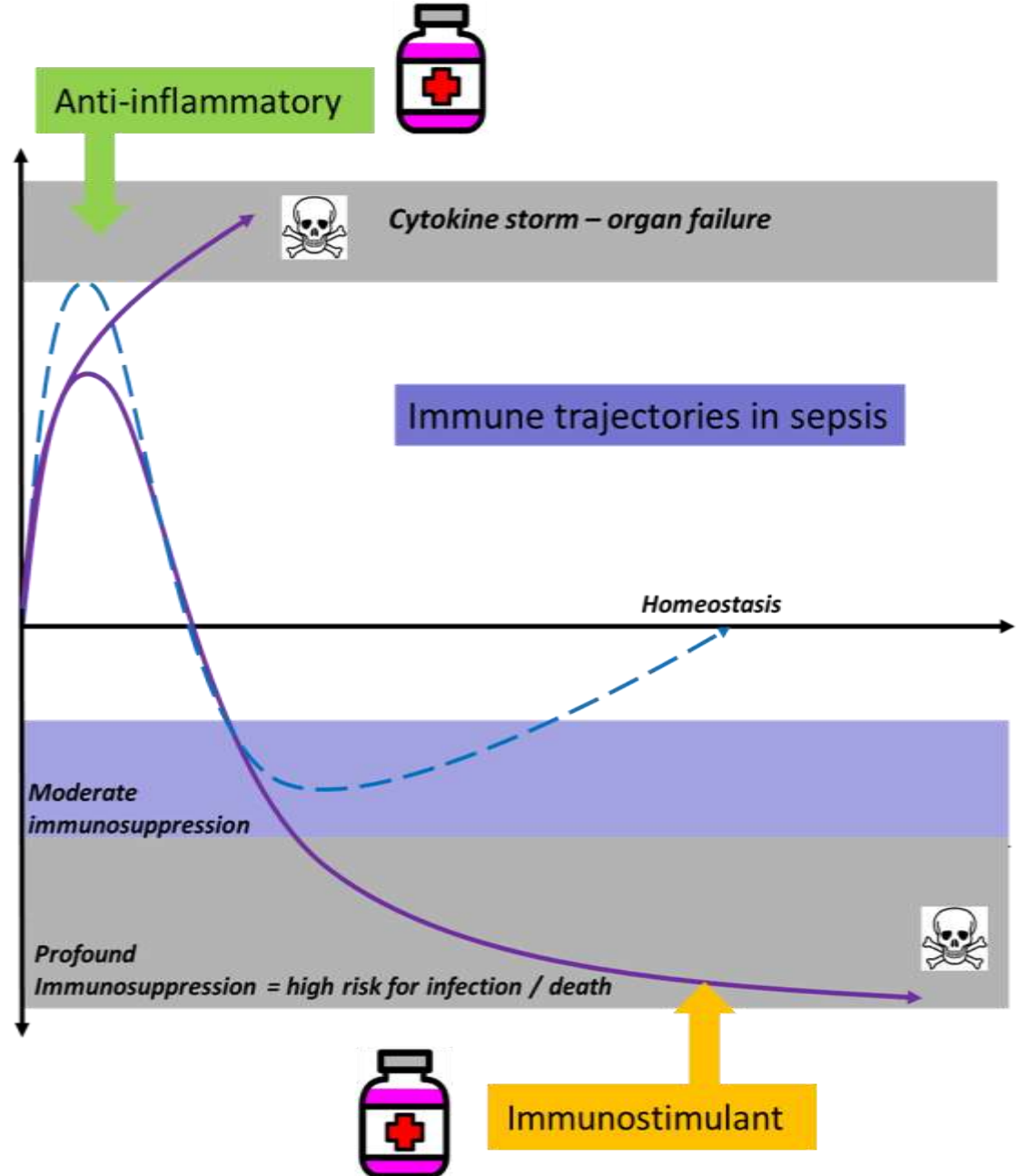
ASSEMBLÉE NATIONALE

Assemblée nationale, F, 6 janvier 2022

# CONCLUSION




**Biomarqueurs**



*CRP / PCT*
*IL-6*
*IL-18*
*Ferritine*
*suPAR*
*S100*
*Nucleosome*
*....*

**Adult and pediatric ARDS:**
Hypoinflammatory*
Hyperinflammatory*
(53–63)

Reactive and
uninflamed (63–64)

**Adult sepsis:**
Coagulation (dA, dB, dC, dD)* (43)

**Proteins**
Biomarkers
Proteomics

**RNA**
Microarray
Bulk seq
Single cell seq

**Adult sepsis:**
SRS1 and SRS2*
Phenotypes (21–23)

Mars1—Mars4 (24)

**Adult and pediatric sepsis:**
Inflammopathic, Adaptive*,
and Coagulopathic (25–27)

?

Big data

*mHLA-DR*
*CD4*
*MDSC*
*PD-1*
*....*

**Adult sepsis flow cytometry:**
Non-improvers,
decliners, improvers,
high expressors
(33–34)

**Immunology**
Immunophenotyping
Functional assay

PHENOTYPES

Patients' characteristics

Age, sex, comorbidities,
SAPS II, SOFA,
Type of germ, bacterial load,
organ, virulence factors…

Annu. Rev. Med. 2023. 74:457–71

Pratik Sinha,[1] Nuala J. Meyer,[2] and Carolyn S. Calfee[3]

The demonstration of the effectiveness of this approach is still pending

We urgently need prospective randomized trials guided by biomarkers

# Thanks for your attention

***Clinical lab***

THE SHIFT PROJECT
THE SEPSIS IMMUNOLOGY TRANSITION THINK TANK

*Pr Anne-Claire Lukaszewicz*
*Head of Anesthesia and Critical Care Medicine Department*
*Hôpital E. Herriot, Lyon, F*