Monocyte and Macrophage Plasticity in the Tumor Microenvironment

Jason Muhitch, PhD
Assistant Professor of Oncology
Urology Department

MIR 509
Trends in Tumor Immunology
October 21st, 2015
Know Your Enemy: Immunosuppressive Functions of Tumor-Associated Macrophages

**TAMs**

- **Impaired T cell activation**
  - B7-1/2, PDL1/2, HLA-G
  - CTLA4, PD-1, IT2
  - T cell
  - TCR
  - NO
  - T cell anergy

- **Decreased T cell viability**
  - TRAIL-R, FAS
  - TRAIL, FASL
  - TGF-β
  - T cell apoptosis

- **Treg induction and recruitment to tumor microenvironment**
  - IL-10, TGF-β, CCL5/20/22
  - FOXP3
  - Migration to tumor

- **Depletion of lymphocyte metabolites**
  - CD3ζ downregulation
  - ARG1
  - L-arginine
  - T cell
  - Cell cycle arrest

**Nonimmune-related mechanisms**

- **Induction of angiogenesis and vasculogenesis**
  - CSF1, CCL2, IL-4
  - VEGF, WNT7β, MMP9

- **Hypoxic tumor**
  - Angiogenesis and tumor vascularization

- **Promotion of metastasis**
  - TGF-β, SPARC, EGF, cathepsins
  - GM-CSF, CSF1, IL-4
  - Invasion
  - Extravasation to metastatic site

- **Promotion of tumor cell stemness**
  - CD90, Eph4A
  - CD11b, Ephrin
  - IL-6, TNF-α
  - MFG-E8, STAT3
  - Tumor cell
  - WNT-dependent stem cell genes
  - NF-κB
  - Twist
  - Chemotherapy resistance

Ugel et al JCI, 2015
Know Your Enemy: Identifying Tumor-Associated Macrophages

Murine
- CXCR9⁺
- CXCR10⁺
- CXCR11⁺

CD45⁺
CD68⁺
CD115⁺
VCAM⁺
GR-1⁻

M1 TAM
M2 TAM

Human
- CD64⁺
- CD80⁺
- CXCR10⁺

CD45⁺
CD68⁺
CD115⁺
HLA-DR⁺
CD205⁺

CD32⁺
CD163⁺
PD-L2⁺

M1 TAM
M2 TAM

Ugel et al JCI, 2015
Know Your Enemy: Tumor-Associated Macrophages Impact on Prognosis

Monocyte & Macrophage Plasticity

Ugel et al JCI, 2015
Characterization of Monocyte Subsets

Classical
- $\text{CCR2}^{\text{high}}$
- $\text{CD11B}^{\text{high}}$
- $\text{CD115}^{\text{low}}$
- $\text{CX3CR1}^{\text{low}}$

Intermediate
- $\text{CCR2}^{\text{low}}$
- $\text{CD11B}^{\text{low}}$
- $\text{CD115}^{\text{high}}$
- $\text{CX3CR1}^{\text{high}}$

Non-classical
- $\text{CCR2}^{\text{low}}$
- $\text{CD11B}^{\text{low}}$
- $\text{CD115}^{\text{high}}$
- $\text{CX3CR1}^{\text{high}}$

Less migratory *in vitro*
Higher phagocytic activity
*CCR2*-Dependent entry into metastasis

Tissue repair
Tissue “patrolling”

Geissmann *et al.* Immunity, 2003
Nahrendorf *et al.* JEM, 2007
Qian *et al.* Nature 2011
Cros *et al.* Immunity 2011
Nahrendorf *et al.* JEM, 2007
Auffrey *et al.* Science, 2007

**CCR2+ Monocyte (classical) Intratumoral Infiltration Is Associated with Macrophage Accumulation and Poor Prognosis in a Broad Tumor Types**
Characterization of Monocyte Subsets

Non-classical (Patrolling) Monocytes CX3CR1

Auffray et al. Science, 2007
Classical Monocytes Can Also Go Out on Patrol!

Classical monocytes within the liver after focal injury
Classical Monocytes Can Also Go Out on Patrol!

CCR2−/− Classical Monocytes within the liver 4 hours after focal sterile injury
CCR2$^+$ Monocytes can Transition to CX3CR1$^+$ Monocytes

Dal-Secco et al. JEM 2015
CCR2+ Monocytes can Transition to CX3CR1+ Monocytes

Dal-Secco et al. JEM 2015
Changes in Circulating Monocyte Populations Suggest Transition Between Subsets

Particular monocyte subpopulations after anti–M-CSF mAb infusions (for treatment of arthritis).

A. Classical

B. Intermediate

C. Non-classical

D. day 0

Changes in Circulating Monocyte Populations Suggest Transition Between Subsets

Increased % of CD16+ monocytes after rhM-CSF infusions in healthy subjects

Saleh et al. Blood 1995
Myeloid Cells Can Account for More than 50% of Tumor-infiltrating Leukocytes

Movahedi et al. Cancer Research, 2010
Monocyte / Macrophage Plasticity in the Tumor Microenvironment

Movahedi et al Cancer Research, 2010
Monocyte / Macrophage Plasticity in the Tumor Microenvironment

Tumor-Associated Macrophages

Inflammatory (Classical) Monocytes

Franklin et al Science 344, 2014
Monocytes Differentiate into TAMs In Vivo

Franklin et al Science 344, 2014
Targeting the CSF-1R to Reduce Intratumoral TAM infiltration

Ries et al Cancer Cell 25, 846-59, 2014
Targeting the CSF-1R to Reduce Intratumoral TAM infiltration

Ries et al Cancer Cell 25, 846-59, 2014
Targeting the CSF-1R to Reduce Intratumoral TAM infiltration

Molecular Profiling Reveals a Tumor-Promoting Phenotype of Monocytes and Macrophages in Human Cancer Progression
Fig. 1

A. PCA plot showing distinct clusters for Mo and RCC-Mo.

B. Dendrogram showing hierarchical clustering based on gene expression.

C. Heatmap displaying upregulated genes with GO process enrichment.

D. Heatmap illustrating gene expression levels across different staging categories for Mo and RCC-Mo.

E. Heatmap comparing gene expression levels between M1 and M2 macrophage subtypes.
**Fig. 1**

A. PCA plot showing the distribution of Mo and RCC-Mo.

B. Height distribution analysis comparing Mo and RCC-Mo.

C. Gene expression profile showing upregulated and downregulated genes in Mo and RCC-Mo.

D. Monocyte staging and expression of pro-inflammatory and anti-inflammatory markers.

Key:
- **IFN-γ**, TLR agonist IL-4, IL-13, IL-10
- **IL-12hi**, **IL-23**, **iNOS**, **ROS**
- **CD86**, **MHC II**
- **Arg1**, **CD206**, **CD163**
- **IL-10hi**, **MMPs**

References:
- Sica, Journal of Clinical Investigation, March 2012
- Gabrilovich, Nature Reviews, April 2012
- Martinez et Gordon, F1000PrimeReports, 2014

Classically Activated

Alternatively Activated

**Mo**

**RCC-Mo**
Fig. 2
Fig. 3
Fig. 4
Fig. 5
Fig. 6

A. Tumor volume (mm³)

B. Relative gene expression

C. Tumor Volume (mm³)

D. Angiogenesis and MMP activity

Fig. 6
Fig. 7

A. Box plot showing gene expression values for IL1B across stages I, II, III, and IV.

B. Scatter plots showing correlation between IL1B and IL6, PTGS2, CD163, and CD14.

C. An image showing stained tissue sections.

D. Correlation with CD163.

E. Bar chart showing relative gene expression of TNF, IL6, IL1B, VEGFA, PTGS2, MMP10, and IL8 across different cell types.

F. Number of tubes per field for different cell types.

G. Optical density measurements for different treatments.

Legend:
- Mo: Monocyte
- MDM: Macrophage-derived macrophages
- TAMs: Tumor-associated macrophages

Significance:
- *: p < 0.05
- #: p < 0.01

Note: The figures illustrate the gene expression and correlation analysis, with significant p-values indicated for each correlation and comparison.