Tumor-Associated Macrophages Promote Invasion while Retaining Fc-Dependent Anti-Tumor Function


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Monocytes

• Innate immune system
• Produced in bone marrow
• Differentiate to
  – Dendritic cells
  – Macrophages
    • M1 subset
    • M2 (a, b and c) subset
    • TAMs

M1 Macrophages

- activated by TLR2/4 ligands, IFN-γ
- pro-inflammatory
- promote Th1 response (AG presentation)
- high Fcγ-Receptor expression
- tumor and tissue destructive

M2 Macrophages

- activated by
  - IL-4/IL-13: M2a
  - immune complexes: M2b
  - IL-10: M2c

- anti-inflammatory
- promote Th2 response
- immunoregulation, tissue remodeling

TAMs

• tumor-associated Macrophages
• M2-like
• anti-inflammatory
• secretion of growth promoting factors
• alleviate metastasis
• high incidence of TAMs → poor clinical prognosis

Monoclonal Antibodies (mAb)

- diverse anti-cancer mechanisms:
  - blocking target functionality
  - induction of apoptosis
  - Fc-dependent
    - activation of the complement system
    - recruitment of innate immune effector cells
      → CDC, ADCC, ADCP

Methods & Results

• Do TAMs from human breast tumors express Fcγ-receptors?
  – tumor tissue from 9 Patients
  – received within 24h of surgical removal
  – detection of FcγRs using flow cytometry
human breast tumor TAMs: FcγR expression

Methods & Results

• Do in vitro differentiated macrophages express Fcγ-receptors?
  – human PBMCs
    • conditioned medium from MDA-MB-231 breast cancer cells → TCMs
    • incubation with M-CSF + polarization with
      – IFN-γ → M1 macrophages
      – IL-13 → M2a macrophages

Methods & Results

In-vitro macrophages: FcγR expression

TCMs express both M1- and M2a-associated markers

Methods & Results

human breast tumor TAMs: marker expression

In-vitro macrophages: Luminex analysis of secreted cytokines

Methods & Results

In-vitro macrophages: 3D culture (Cultrex), assessment of tumor invasion after 24h coculture

mAb mediated phagocytosis of tumor cells

mAb mediated phagocystosis of tumor cells (3D culture)

Methods & Results

• In vivo mouse studies
  – SCID/beige mice
    • no B cells, no T cells, no functional neutrophils and NK cells
    • functional macrophages
  – Macrophage depletion utilizing clodronate-encapsulated liposomes (CEL)

Methods & Results

• In vivo mouse studies
  – 6 groups á 10 mice
  – 2 groups each received 100 μl CEL, EmptyLipo or PBS i.p.
  – Day 1: MDA-MB-231 tumor cells implanted into mammary fat pad
  – Day 3: mAb therapy started with anti-CD142 (control: PBS)
  – mAb injections weekly, tumor volume measured twice weekly
  – Day 42: The End

Methods & Results

In vivo TAMs enhance tumor suppression under mAb therapy

Methods & Results

In vivo TAMs enhance tumor suppression under mAb therapy

Methods & Results

Assessment of differentiating cytokines

Methods & Results

Assessment of differentiating cytokines

Discussion

• TCMs
  – promote tumor cell invasion in the absence of tumor-targeting mAbs
  – display potent anti-tumor properties in the presence of tumor-targeting mAbs
• Complex interactions between immune cells, tumor cells and tumor stroma
• Macrophages might play a more important role in mAb therapies than NK cells
• TAMs as therapeutic targets in mAb therapy