ANTI-TUMOR EFFECTS OF AN IMIDAZOQUINOLINE IN RENAL CELL CARCINOMA

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What are imidazoquinolines?

- Family of immune response modifiers of which imiquimod is the most commonly used.

- Act on Toll-like receptors which can increase cellular production of TNF, IFN-α, and a variety of other cytokines.

- Shown to improve antigen presentation by dendritic cells

- Imiquimod is FDA approved for topical treatment of genital condyloma and basal cell carcinoma.
Project Background

- Renal cell carcinoma (RCC) responds to immunomodulators (IL-2, IFN-α)
- Imiquimod has been used as an immune response modifier in basal cell carcinoma
- We hypothesized that imidazoquinolines may have direct in vitro and in vivo biological activity against renal cell carcinoma.
Study Design

- 4 renal cell carcinoma cell lines used
  - 1 mouse (RENCA)
  - 3 human (CAKI-1, CAKI-2, A-498)

Experimental drug: 3M-011

- Closely related to Imiquimod (3M Pharmaceuticals)
- Potential for use as an oral agent
Do renal cell carcinoma cell lines express Toll-like receptor 7?

Western blot analysis of TLR-7 expression in RCC cell lines. HELA cells (cervical cancer line) serve as a negative control.
Can 3M-011 affect cell viability?

- Evaluated by cell counts
- Cells cultured in media supplemented with 3M-011 at 100 µg/ml
- Cells harvested and counted at 24, 48, and 72 hour time points
Potential correlation between cell viability and level of TLR-7 expression

3M-011 Affect on Cell Viability Relative to Control

RENCA
CAKI-1
CAKI-2
A-498

% decrease in cell viability

Western blot analysis of TLR-7 expression in RCC cell lines. HELA cells (cervical cancer line) serve as a negative control.

TLR-7 Expression in RCC cell lines

(-) Control

Cell Line
Apoptosis in RENCA cells induced by 3M-011

Control

3M-011

Absorbance (405 nm)

Media supplement
**In vivo affects of 3M-011 on a murine metastatic RCC model**

- Tail vein injection
  - (2 x 10^5 RENCA cells)
- One week for tumor growth
- Group 1: Placebo
- Group 2: 3M-011
- Two week treatment
  - (every other day oral gavage)
- Sacrifice for pathology
**In vivo results**

- **% of mice with tumor**

  - Placebo: 100%
  - 3M-011: 70%

- **Mean # of macroscopic tumor nodules**

  - Placebo: 37.2
  - 3M-011: 12.6

- **Mean # of microscopic tumor nodules**

  - Placebo: 51.6
  - 3M-011: 22.9

- 30% of mice in 3M-011 group showed no pathologic evidence of tumor.
Summary and Conclusions

• TLR-7 is expressed in both murine and human renal cancer cell lines.

• Imidazoquinolines have direct biological effects on renal cell carcinoma cell lines by decreasing cell viability and inducing apoptosis.

• Our results suggest a potential correlation between the TLR-7 expression level and the effect of 3M-011 on cell viability.

• Initial results in an immune competent mouse model of metastatic renal cell carcinoma suggest anti-tumor effects in vivo.
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