Radiation therapy to ignite an anti-cancer immune response
Silvia Formenti, M.D.
Weill Cornell Medical College
New York Presbyterian Hospital
New York, NY

DISCLOSURES

Consultant/Speaker:
Bristol Myers Squibb, Varian, Elekta, Janssen, Regeneron, GlaxoSmithKline, Eisai, Dynavax, Astra Zeneca

Principal Investigator:
NCI RO1CA161891-01
Immunomodulation of breast cancer via TLR7 agonist IMQ and RT
DOD BC106855J / W81XWH-11-2-0530
Multi-Team Award (MTA)
Radiation-induced vaccination to breast cancer

13-AD-00-021870-01
Breast Cancer Research Foundation
Targeting key inhibitory pathways to improve radiation-induced vaccination in breast cancer

NIH 1 S10 RR027619-01 Preclinical Research Irradiator

Why are abscopal effects of radiation so rare?
• 1969-2014: 46 abscopal cases
• Median dose 31 Gy; median follow up 17.5 months
• Only one case single dose SBRT (2.1%); all others fractionated RT (wide range of dose and fractions)
• ~ 13% were VMAT/SBRT
• NON ABLATIVE DOSES

IMMUNOSUPPRESSION DOMINATES IN ESTABLISHED TUMORS

How to enable the pro-immunogenic effects of radiation:

Overcoming immunosuppressive microenvironment of established tumors
- enhancing cross priming
- blocking immune checkpoints

Overcoming RT immunosuppressive effects
- overcoming RT-induced immunosuppression (adenosine, TGFβ)
- mitigating RT-induced lymphopenia
Ionizing radiation stimulates anti-tumor immunity by generating an in situ vaccine: combination with immunotherapy uncovers the abscopal effect.

Day: 0
67NR
5x10⁶ or 10⁶ each side, primary R and secondary L

RT: 20 Gy

Flt3-L (0.5mg/kg)

RT: 2 Gy

BALB/C mice injected at two separate sites with the syngeneic mammary carcinoma 67NR cell line

RT: 3.5 Gy x 10
GM-CSF: 125 μg/m²
Daily x 14 days

27% ABSCOPAL ORR

Generation of anti-tumor T cell responses requires tumor irradiation + CTLA-4 blockade

Jimm Allison

Demaria et al., Clin Cancer Res 2005
Clinical study design to test for abscopal responses

- Either a prospective randomized trial (IT+RT versus IT)
- Or a trial of radiation with an immunotherapy proven ineffective when used alone

Limited objective response rate to CTLA-4 Blockade (without and with chemo) in NSCLC

<table>
<thead>
<tr>
<th>Reference</th>
<th>Stage</th>
<th>Study Design</th>
<th># PTS</th>
<th>OR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Zatloukal et al ASCO 2009</td>
<td>LOCALLY ADV/NETS</td>
<td>Tremelimumab (15 mg/kg) versus BSC</td>
<td>87</td>
<td>4.5%</td>
</tr>
<tr>
<td>Lynch et al JCO 2012</td>
<td>Stage III/IV</td>
<td>Carboplatin vs Carboplatin/T with Ipi 10mg/kg, Carboplatin/T sequential 10mg/kg</td>
<td>204</td>
<td>NS</td>
</tr>
</tbody>
</table>

No CRs in either studies

Patient with Refractory Metastatic NSCLC

Progressing after 3 lines of chemo and chest RT. Multiple lung, bone a...
Same patient, response to RT + ipilimumab

Clinical and radiological CR at one year:
currently NED at 36 m

NYU S14-U02UB
Ipilimumab and localized RT in chemo-refractory metastatic NSCLC

19 patients, Response rates (CR + PR):
Intent to treat = 18%
Pts completing 4 Ipi = 33%

Median follow-up: 12 months
Log-rank test: p = 0.0361
HR = 9.174
PD = 9 months

Median survival: CR/PR/SD = not reached

Good survival rate, CR/PR/SD
Same patient: PDL-1 up-regulation as a marker for the induction of an effective anti-tumor T cell response

CD8 (brown)
Ki67 (red)

Demaria and Stack, (PerkinElmer)

12.1% of CD8 T cells are Ki67+

How to enable the pro-immunogenic effects of radiation:

Overcoming immunosuppressive effects of RT

- overcoming RT-induced immunosuppression (adenosine,TGFβ)
- mitigating RT-induced lymphopenia

RT-induced adenosine

Erik Wennerberg
AACR 2016, 4033
Adenosine-blockade promotes intratumoral infiltration of CD8α+ activated DCs, reduces Treg infiltration while promoting CD8+ T cell infiltration

Combined RT and anti-CD73 treatment delays tumor progression and prolongs survival

Modulation of adenosine generation and/or uptake in the TME may facilitate the immunogenic effect of radiation
Inhibition of DC activation

Inhibition of T cell effector function

TGFβ activation by radiation-induced ROS hinders priming of anti-tumor T cells

Anti-TGFβ (1D11)

Therapeutic synergy of radiation and TGFβ blockade

RESULTS

Anti-TGFbeta + RT:
22 patients, <10% ORR

59 F with metastatic Triple Negative Breast Cancer

4th line therapy 18 months after diagnosis:
RT + Fresolimumab
Increased PDL-1 and PDL-2 expression on tumor and myeloid cells by RT and TGFβ blockade

PD-1 blockade extends survival in mice treated with radiation and TGFβ blockade

In vitro anti-PD-1 (pembrolizumab) partially restores TCR→p-ERK/p-AKT

➢ Basis for in vitro PD-1 patient selection marker for combination therapies
The delicate balance of cross-presentation

Need for sufficient naïve T cells

8-cm tumor: 60 Gy/30 fractions modeled with Pinnacle™ radiation planning system
Radiation doses to circulating cells (DCC) analyzed using MatLab™
Circulating lymphocytes:
\[
\begin{align*}
D_{10} &= 3 \text{ Gy} \\
D_{50} &= 2 \text{ Gy} \\
D_{90} &= 0.5 \text{ Gy}
\end{align*}
\]
A single radiation fraction delivered 0.5 Gy to 5% of circulating cells, after 30 fractions 99% of circulating blood had received ≥0.5 Gy

Naïve T cells are the most radiosensitive

Impact of Number of fractions, Dose rate, Target Size

High dose rate
Small, superficial fields
Hypo-fractionated RT

Yovino et al Cancer Invest. 2013
Conclusions

• RT-induced signaling effects interacts with multiple immunological pathways, including adenosine, TGF-β, PD-1 etc.

• Success of combination of anti-CTLA-4 and radiation in metastatic NSCLC was independent from PD-L1 expression/blockade. Conversely, effectiveness of blocking TGFβ likely depends on overcoming PD-L1 expression

• Hypo-fractionated, short courses of RT to a small target to avoid lymphopenia is likely to be key to the success of RT and immunotherapy

When combined with Immunotherapy what is the best Radiation Source, Dose, Fractionation?

Looking at the ICD of Protons, Deuterons, and Helium

• RARAF Columbia

• Prep Cells for the Track Segment Charged-Particle Accelerator

• Allows for irradiation of particles with Linear Energy Transfer (10-200KeV/mm).

http://raraf.org/tracksegment.html
At 35kEV, 5Gy of deuterons ~20 Gy x-rays

At 35kEV, 5Gy of deuterons ~20 Gy x-rays

(Unpublished data)

Fractionated but not single dose RT elicits an abscopal response in combination with anti-CTLA-4


Differentially expressed Immune Response genes in at least one of 4 comparisons (>2-fold, Paired T-test p-value < 0.05) are displayed as normalized to 0Gy control within each set of three samples.

SD – 20 Gy x1
MP – 8 Gy x3

Claire Vanpouille-Box
N. Coleman & M. Aryankalayil
NIH Radiation Oncology Branch
IFN-I pathway activation in Hypo-Fx tumors

Response to Interferon-type I

IFNβ1, Mx1, Oas1a, Oas1b, Oasl1, Oas2, Oasl2, Oas3, ISG15, IRF7, ifit1, ifit2, ifit3, ifih1, ifi204, IFNγ, Ccl5, CxCl10, Ccl2, Ccl7

n-fold (0 Gy)
20 Gy - 4h
20Gy - 24h
3x8Gy - 4h
3x8Gy - 24h

IFN-I ISG OAS genes ISRE Cytokines Chemokines

A 10 x 3Gy [10-6] 30 Gy + 10 x 3Gy [10-6]

B

No abscopal response demonstrated!

Reject Second Challenge
But concurrent second tumors continue to grow!
Side-by-side comparison of 30 Gyx1, 8 Gyx1 and 8 Gyx3 with Immune Checkpoint Blockade

(T-T test)

Tumor growth curves - Irradiated site -

Single

30Gyx1+9H10

Fractionated

8Gyx3+9H10

30Gy irradiated tumors regrew faster compared to 8Gy x 3
Abscopal site (non irradiated)

Survival

Endpoints:
- Abscopal response
- Survival

8 Gy X 3 superior to single fraction 8/30 Gy

Dose and fractionation and RT source

- When combined with ICB tumor hypo-fractionated regimens are required for abscopal effects
- High LET radiation more likely to induce ICD