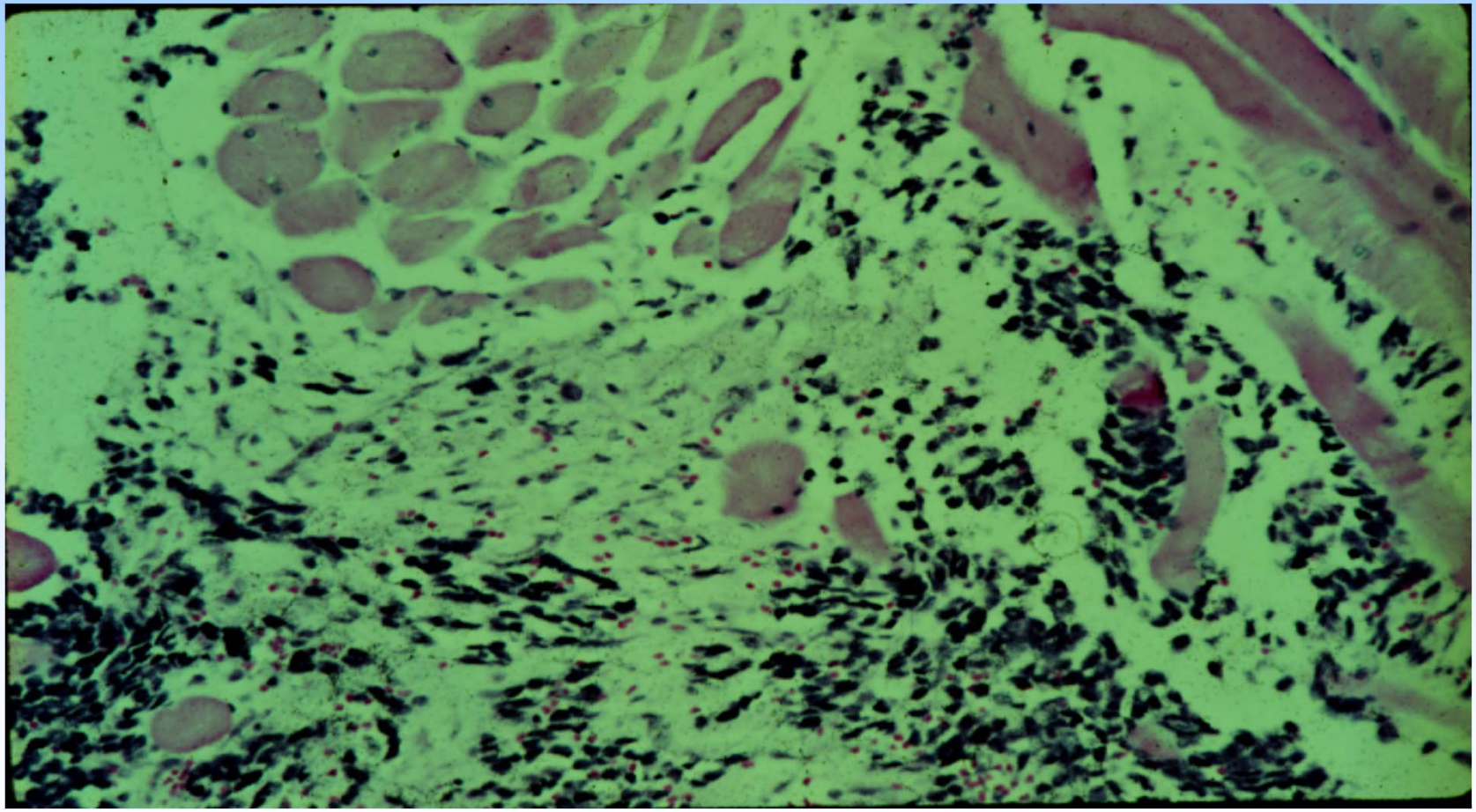
From IGF to mTOR Signaling in Pediatric Sarcomas-Opportunities for Novel Therapeutic Intervention

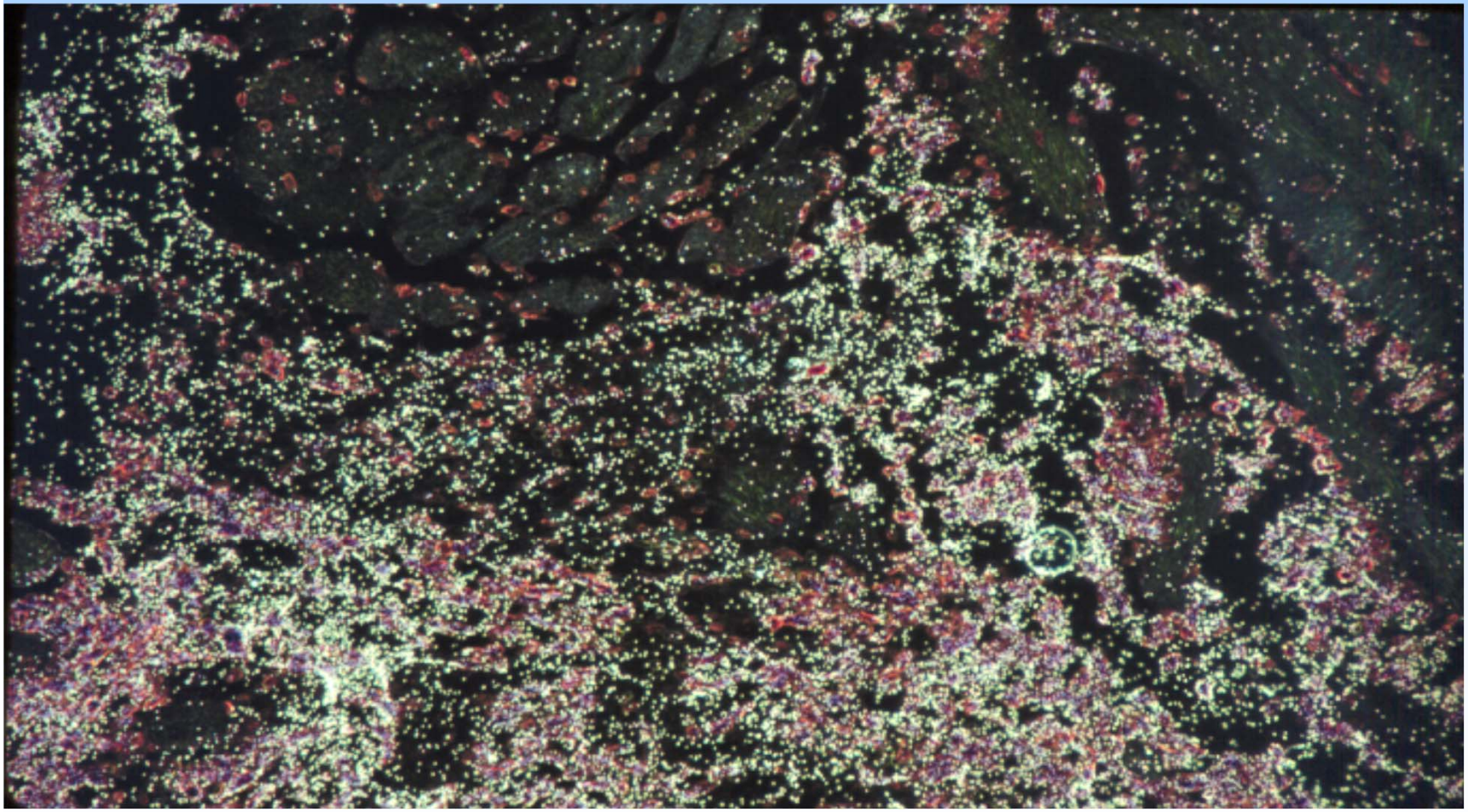
Role of IGF Signaling- Previous Studies

- IGFII is an autocrine growth and motility factor in rhabdomyosarcoma (El-Badry et al. Cell Growth and Diff 1990)
- Loss of Imprinting (LOI) of IGFII in Rhabdomyosarcomas (Zhan S, Shapiro DN, and Helman LJ JCI 1994)
- LOI of IGFII in Ewing's Sarcoma (Zhan S, Shapiro DN, and Helman LJ Oncogene 1995)
- IGFIR is required for EWS-FLI-1 transformation of fibroblastsl (Toretsky J. et al. JBC 1997)

In situ bright field IGF-II



In situ dark field IGF-II

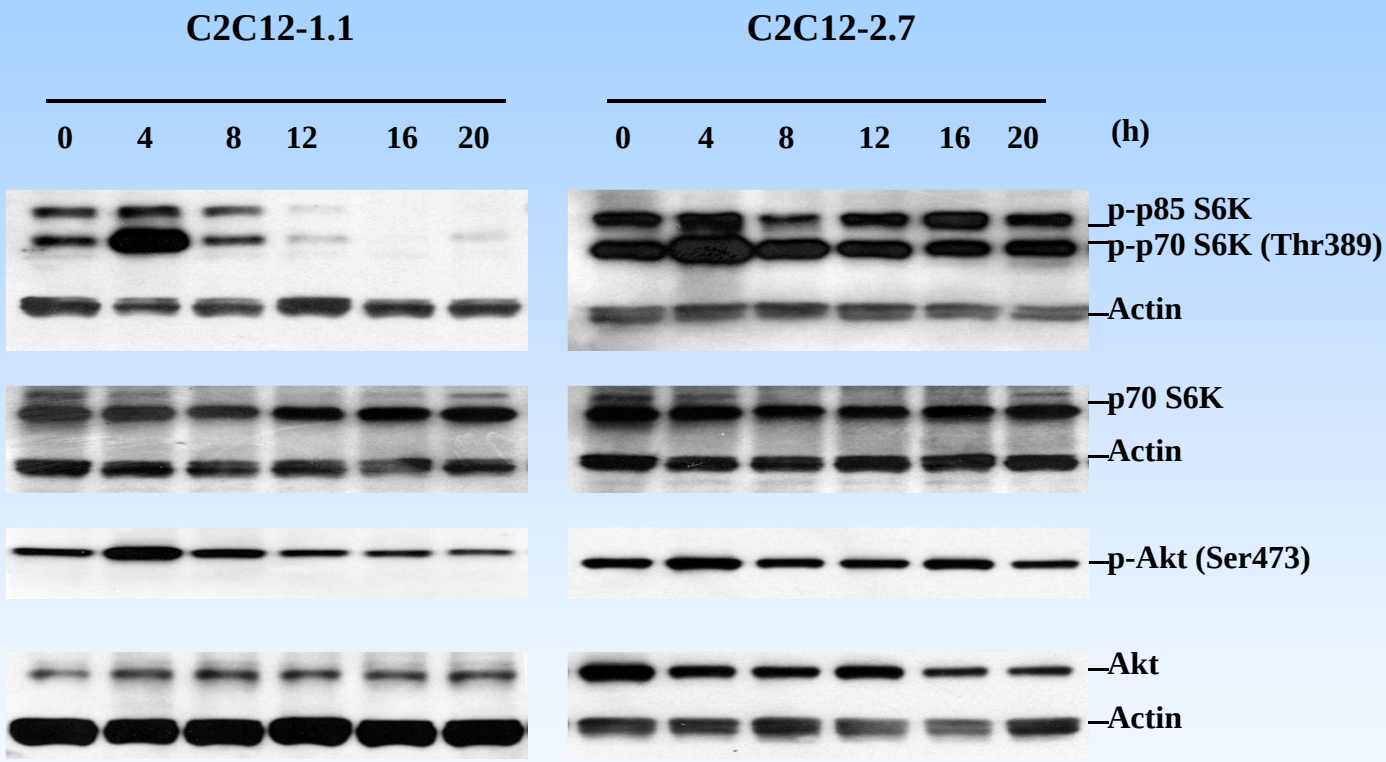


Previous Studies

- IGF-II overexpression in C2 myoblasts led to diminished G1 checkpoint (Zhang et al. JBC 1999)

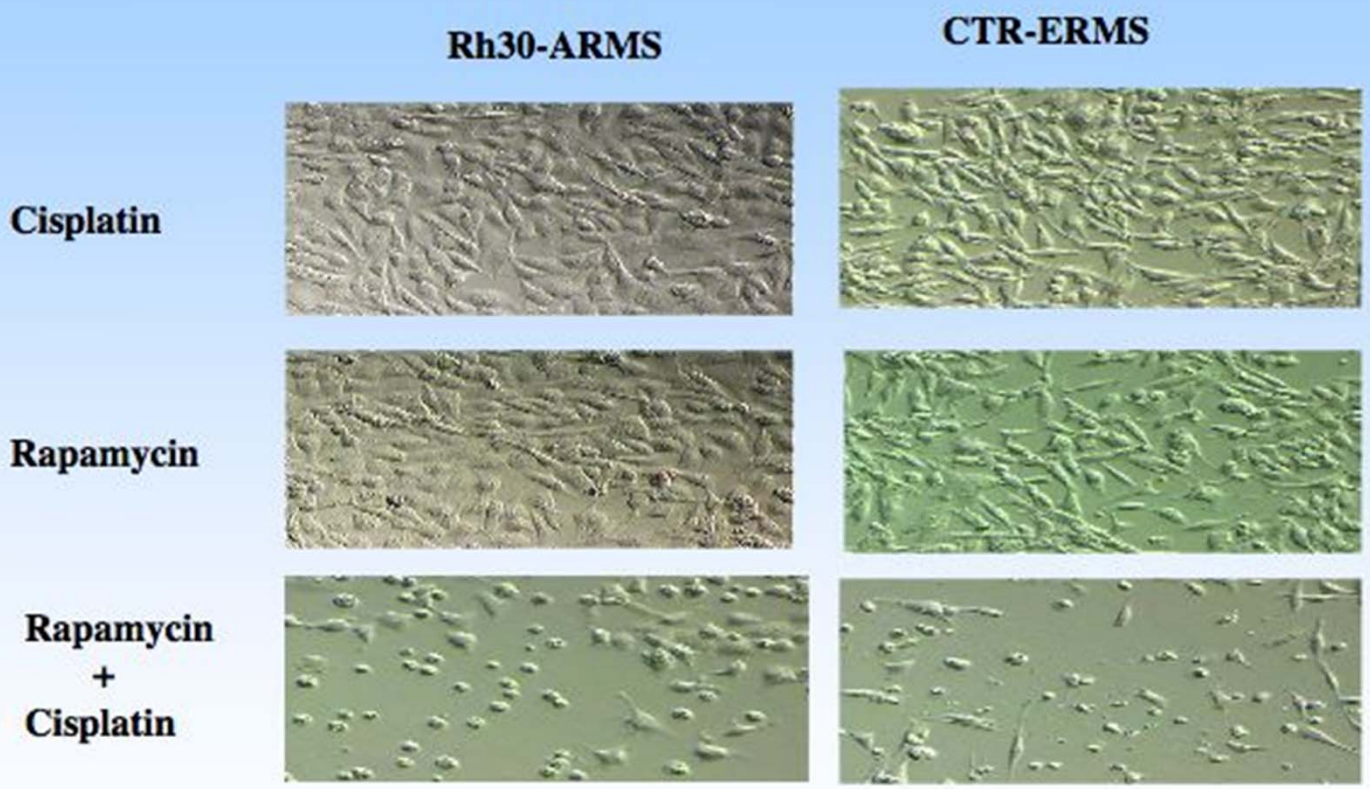
- Resistance to apoptosis most directly correlated with phosphorylation of p70S6 kinase and 4E-BP-1. Resistance to apoptosis in IGF-II overexpressing cells was reversed by rapamycin (Wan and Helman Neoplasia 2002)

Cisplatin Decreases p70S6k Phosphorylation In Wild-type but not IGFII overexpressing C2 cells



Cells treated with 25μM CDDP for indicated times

Rapamycin Abolishes the Resistance of IGF-II Driven RMS Cells to CDDP Induced Apoptosis



Conclusions

- IGF signaling provides a survival signal that contributes to tumor cell resistance to DNA-damage induced cell death

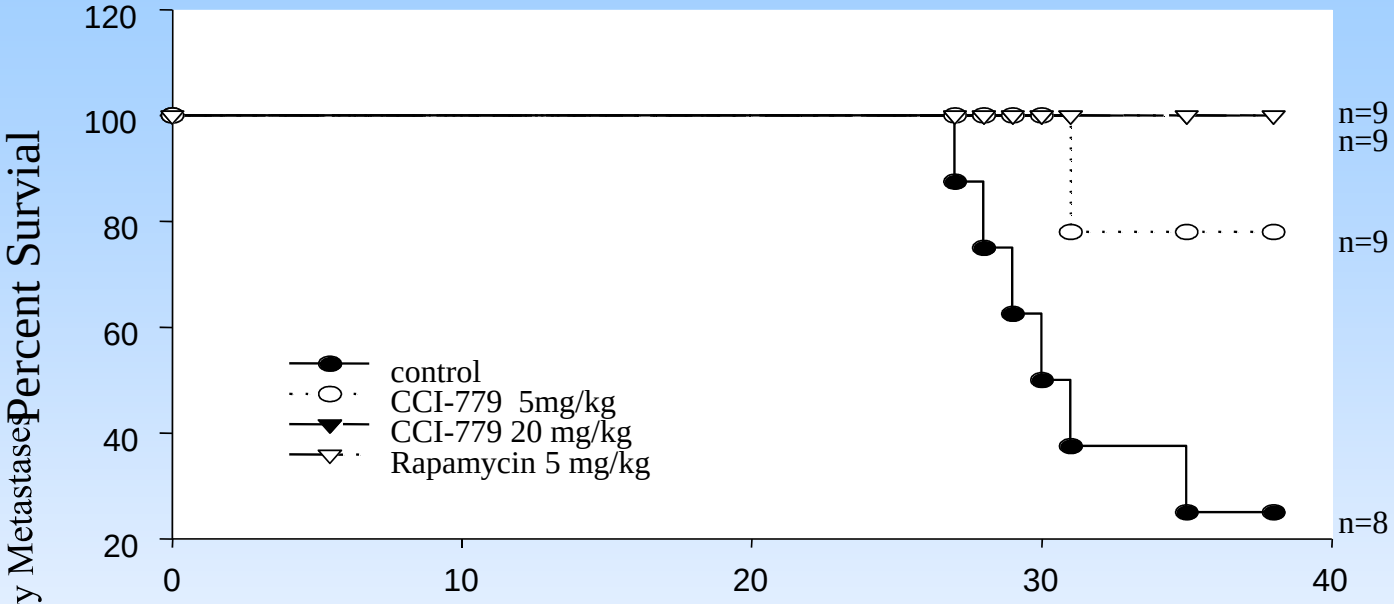
- This resistance is associated with mTOR signaling, and can be reversed with agents that block mTOR

mTOR and Sarcomas

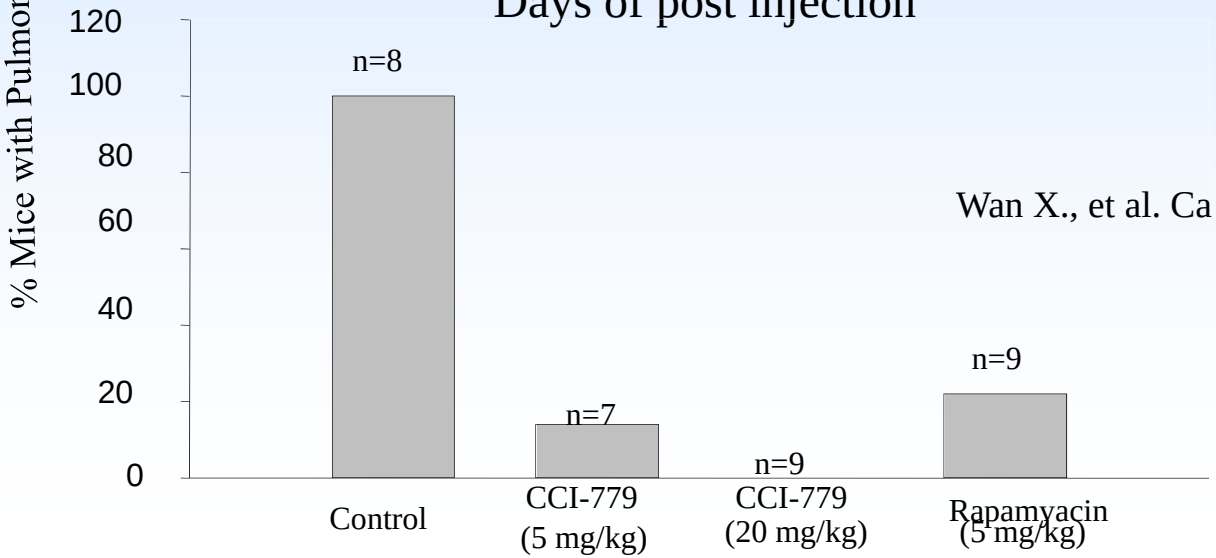
- Demonstrated that aggressive, metastatic behavior in sarcomas associated with activation of mTOR (Khanna et al. Nat Med 2004, Wan et al. Ca Res 2005)
- Demonstrated that mTOR blockade with rapamycin or analogs inhibits RMS experimental pulmonary mets (Wan Cancer Res 2005)

Rapamycin and CCI 779 prolongs survival and inhibits pulmonary metastasis of K7M2 Osteosarcoma *in vivo*

A



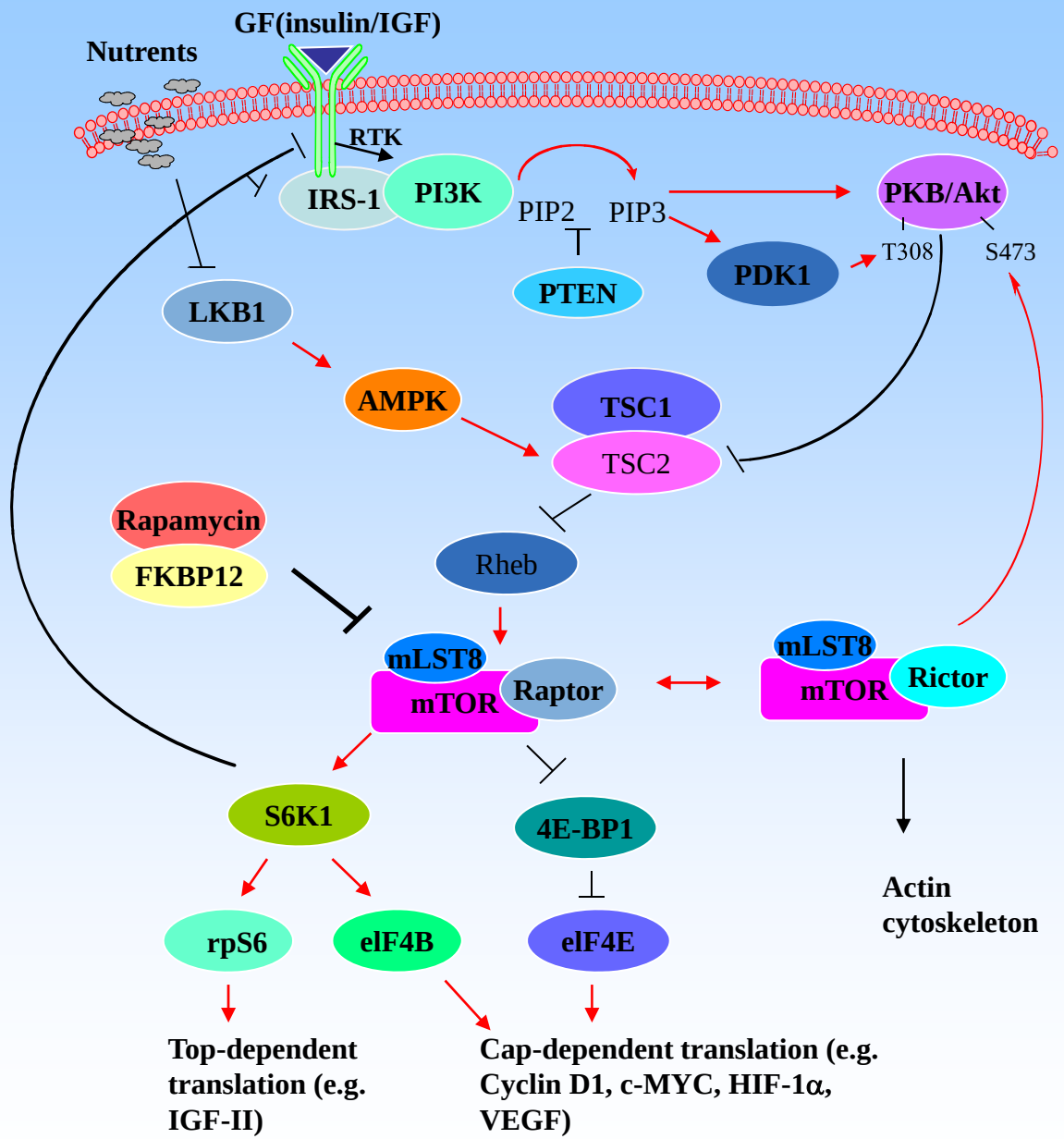
B

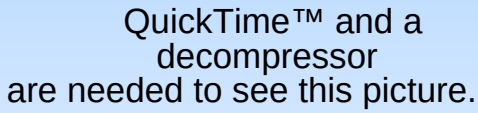


Wan X., et al. Ca Res 65:2406, 2005

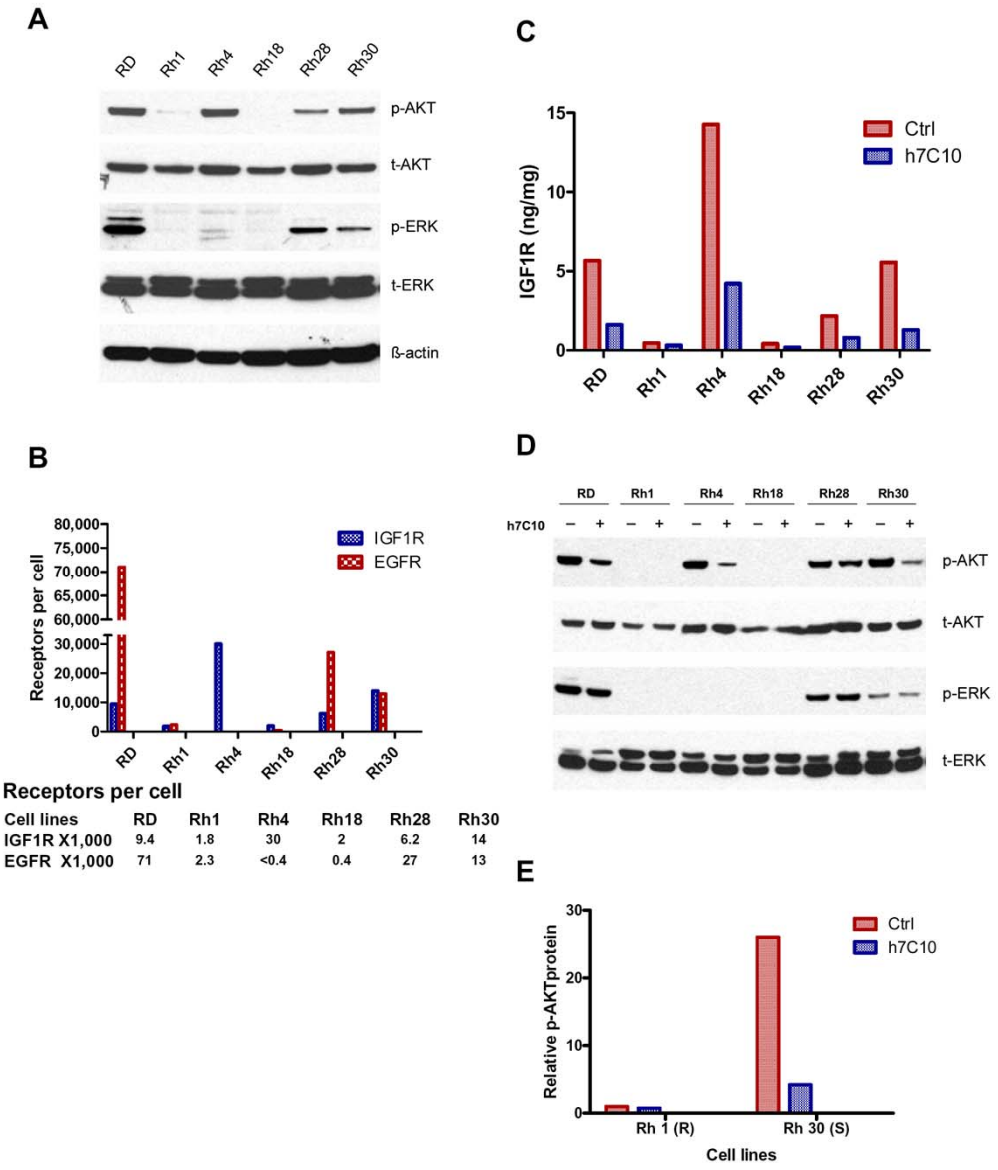
Rapalogs and IGF Signaling

- Demonstrated that rapalog treatment of RMS leads to activation of Akt *in vivo*
- Demonstrated that this activation is IGF dependent and can be blocked with IGFIR blockade (Wan et al. Oncogene 2006)


GF(insulin/IGF)
Nutrents
RTK
IRS-1
PI3K
PIP2
PIP3
PKB/Akt
T308
S473
PDK1
PTEN
LKB1
AMPK
TSC1
TSC2
Rapamycin
FKBP12
Rheb
mLST8
mTOR
Raptor
mLST8
mTOR
Rictor
S6K1
4E-BP1
Actin
cytoskeleton
rpS6
elF4B
elF4E
Top-dependent translation (e.g. IGF-II)
Cap-dependent translation (e.g. Cyclin D1, c-MYC, HIF-1α, VEGF)



In RMS, IGFIR levels directly correlate with sensitivity to IGFIR blockade *in vitro*



IGFIR is responsible for the majority of Akt activation in RMS cell lines, and IGFIR Ab specfically downregulates IGFIR and pAkt in cell lines with high IGRIR levels

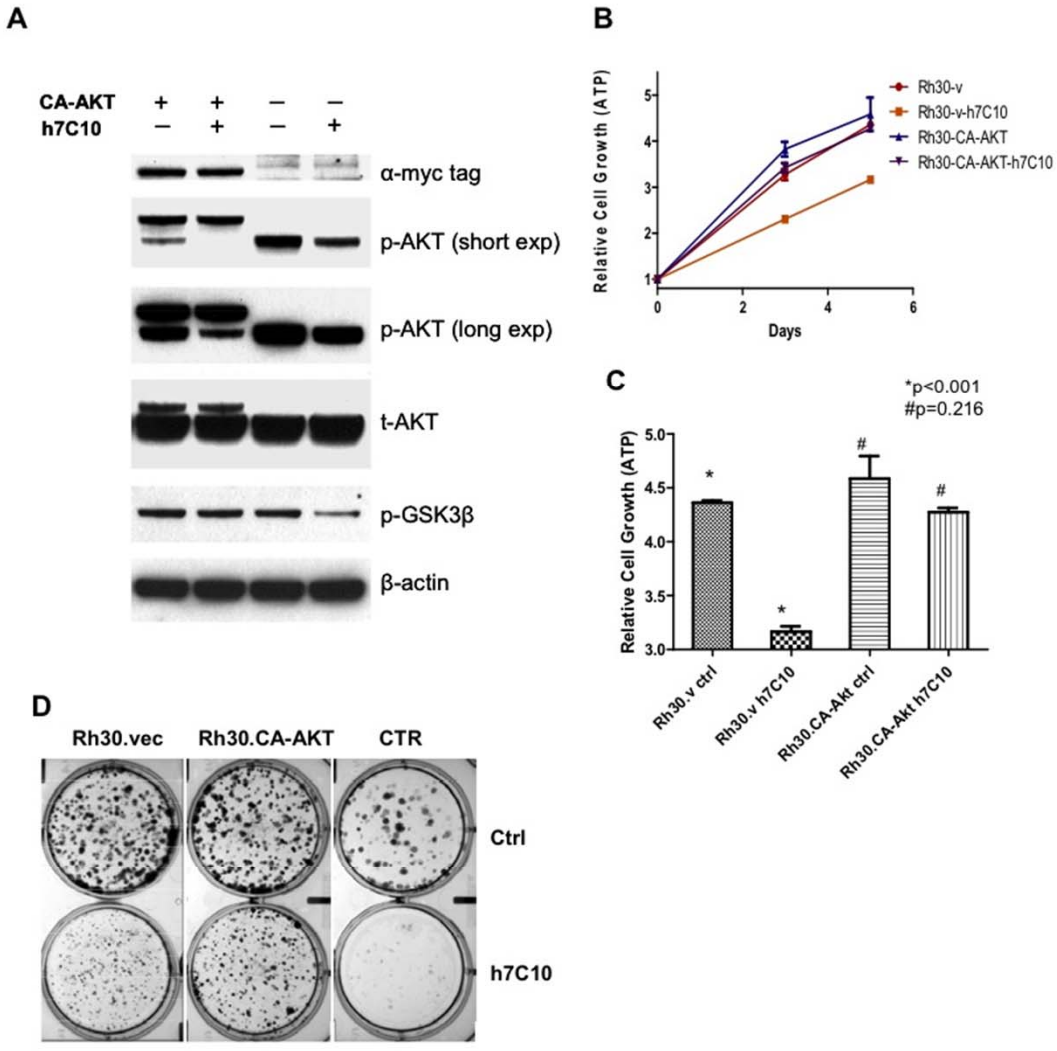


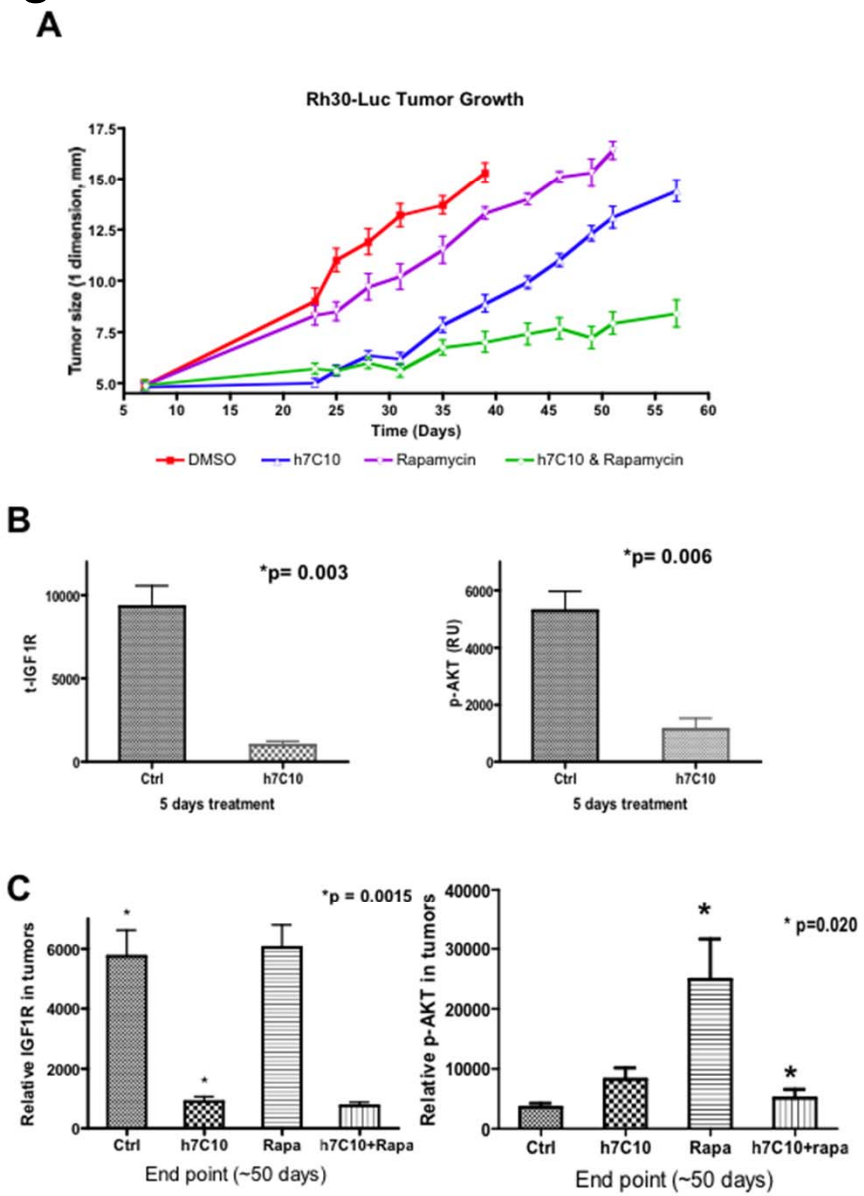
Fig. 4

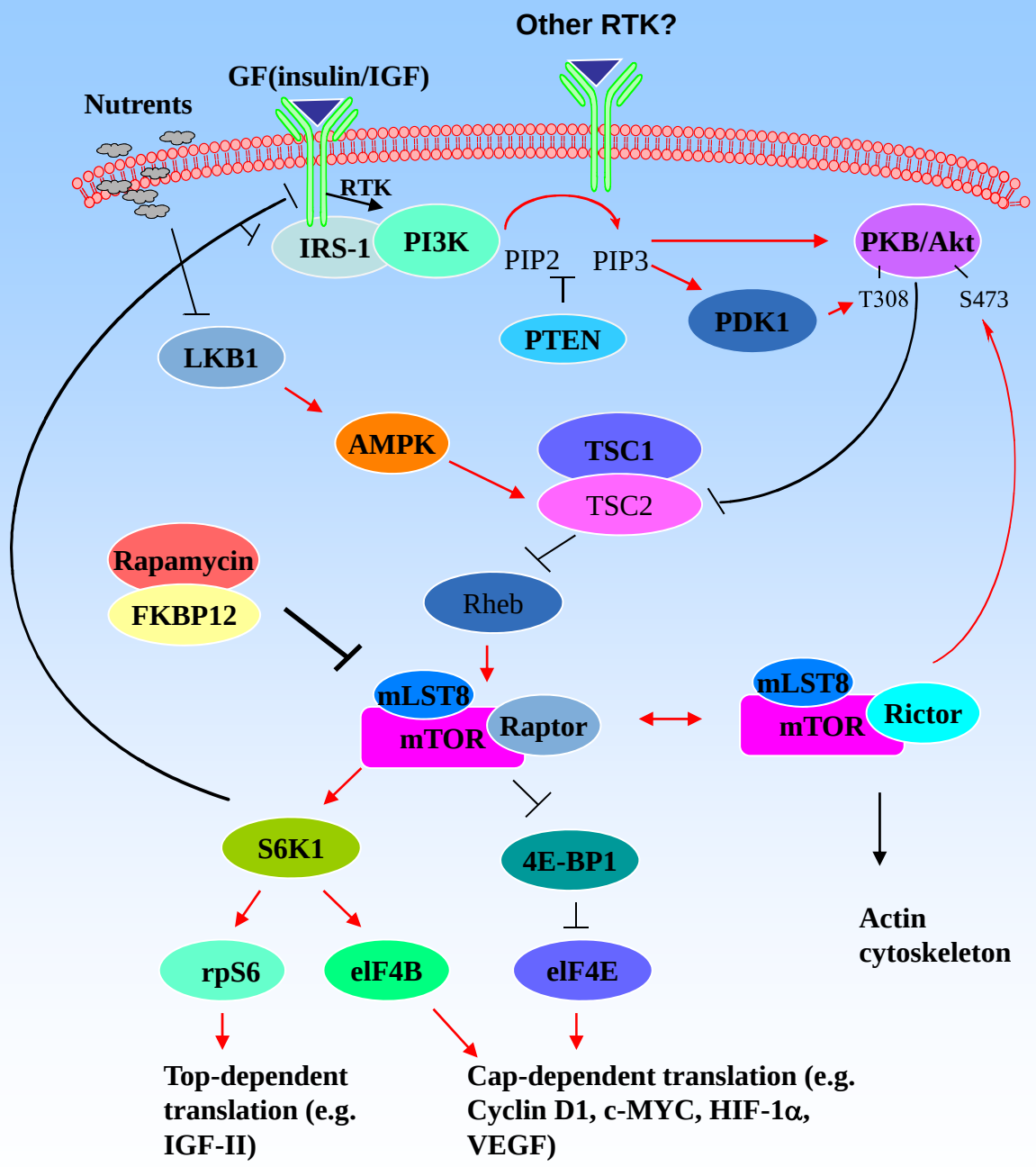
Activated Akt reversed h7C10 effect on proliferation

QuickTime™ and a
decompressor
are needed to see this picture.

In RMS, IGFIR levels directly correlate with sensitivity to IGFIR blockade *in vivo*

Uncoupling of IGFIR and Akt signaling after long-term Rx-RH30 cells. Combination IGFIRAb plus rapamycin is more potent in xenograft growth inhibition



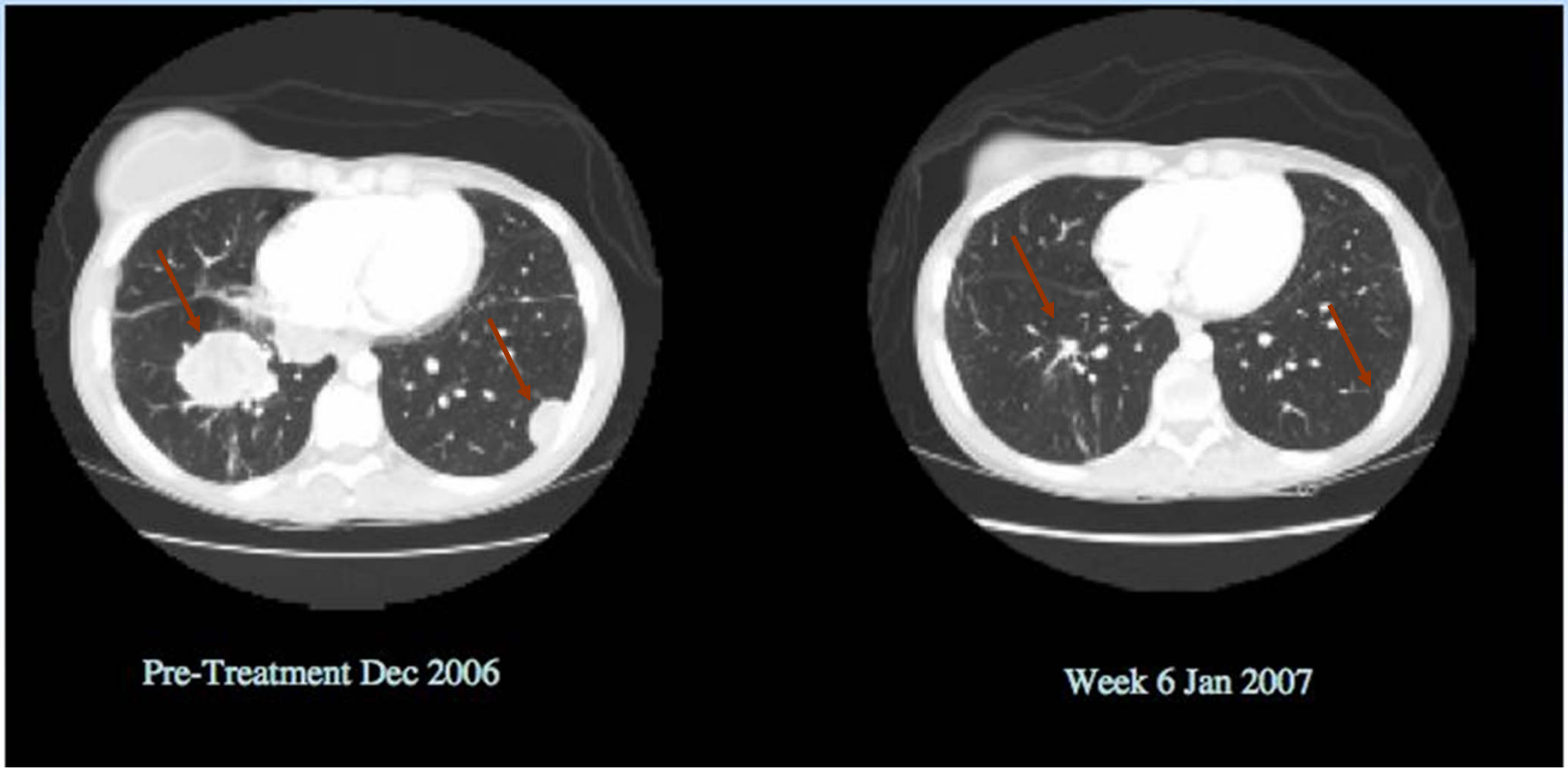

Other RTK?
GF(insulin/IGF)
Nutrents
RTK
IRS-1
PI3K
PIP2
PIP3
PKB/Akt
T308
S473
PDK1
PTEN
LKB1
AMPK
TSC1
TSC2
Rapamycin
FKBP12
Rheb
mLST8
mTOR
Raptor
mLST8
mTOR
Rictor
S6K1
4E-BP1
rpS6
elF4B
elF4E
Actin cytoskeleton
Top-dependent translation (e.g. IGF-II)
Cap-dependent translation (e.g. Cyclin D1, c-MYC, HIF-1α, VEGF)

Conclusions

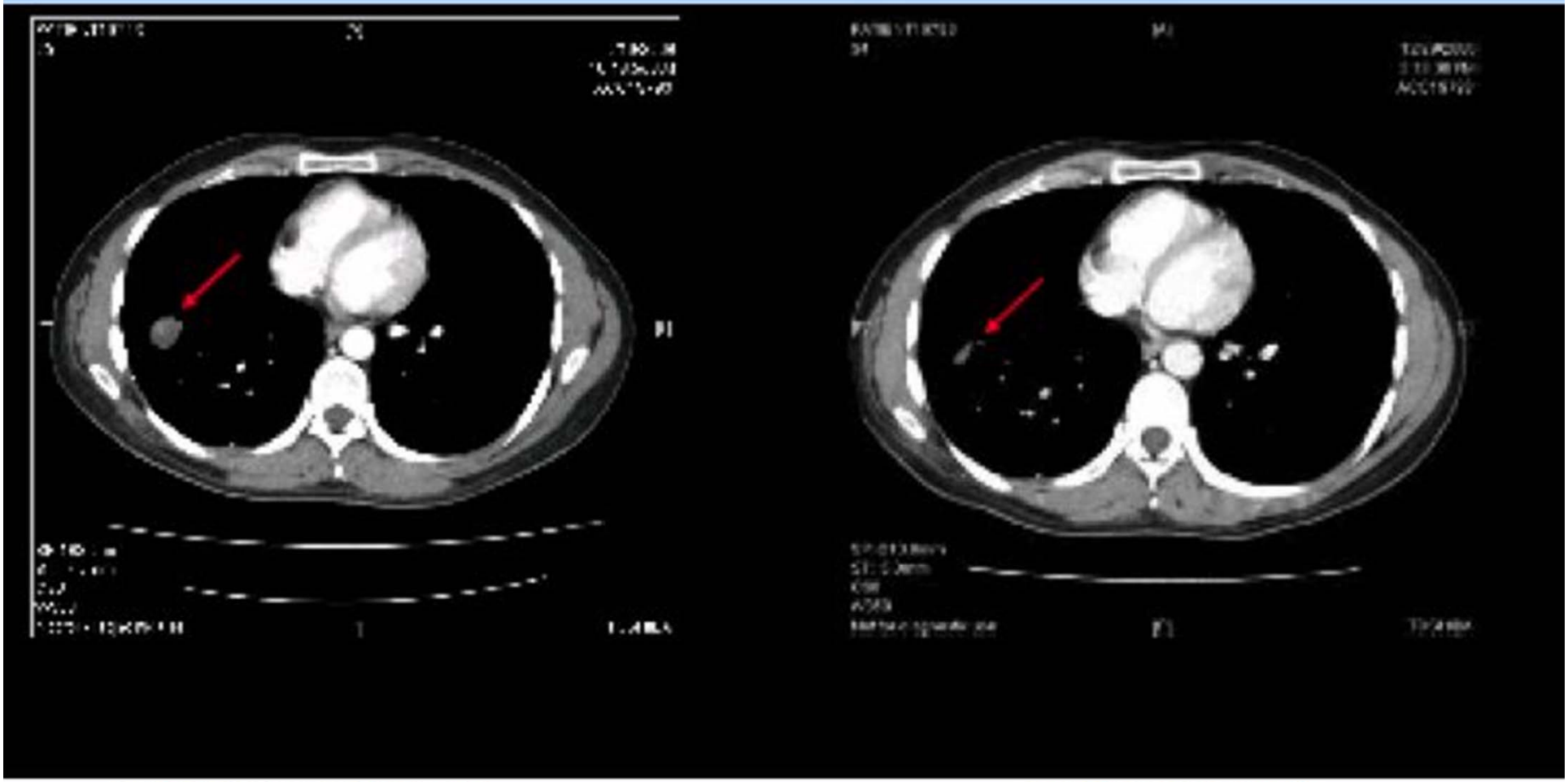
- Early evidence to suggest beneficial combination of mTOR inhibition combined with IGFIR inhibition
- Effect of IGFIR inhibition correlates with IGFIR levels
- Effect of IGFIR blockade on decrease in pAkt is lost in long-term xenografts, and this “tachphylaxis” is abrogated with mTOR inhibtion

Clinical Studies

Refractory recurrent Ewing's patient Rx with R1507



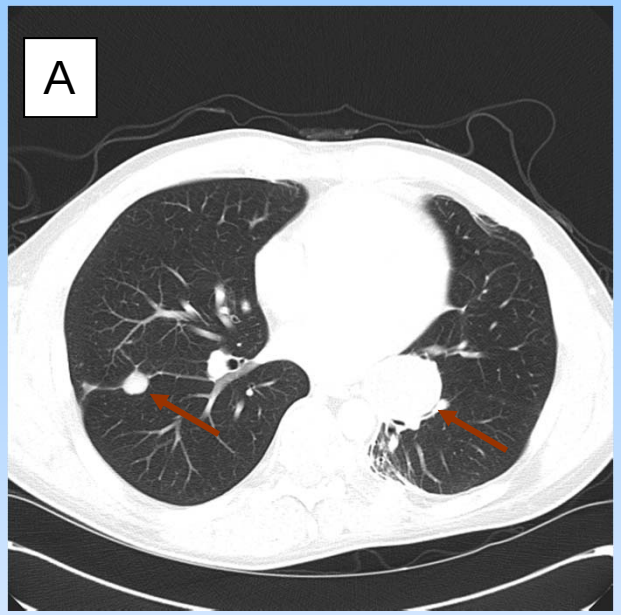
Refractory Ewing’s sarcoma patient Rx with R1507

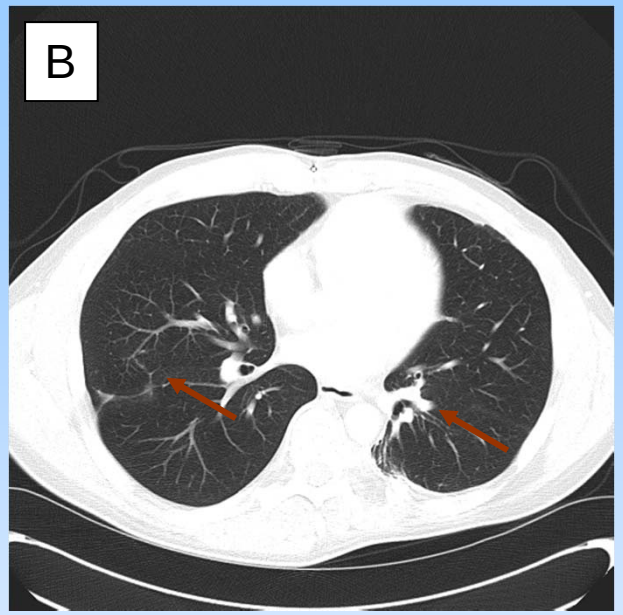


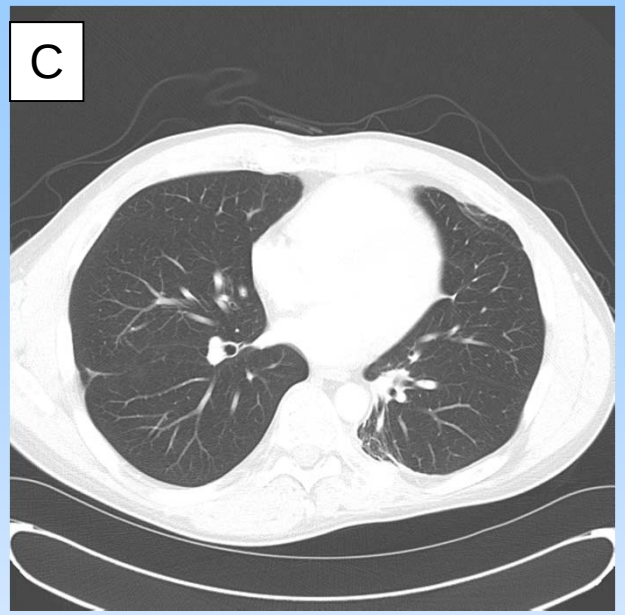
Pre-Treatment Jun 2006

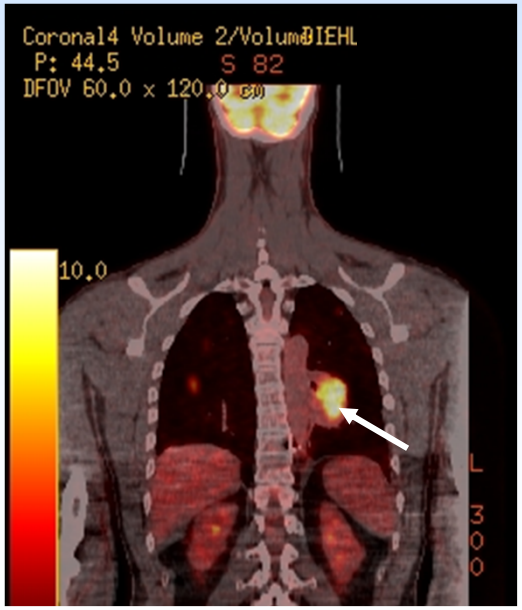
Week 25 Dec 2006

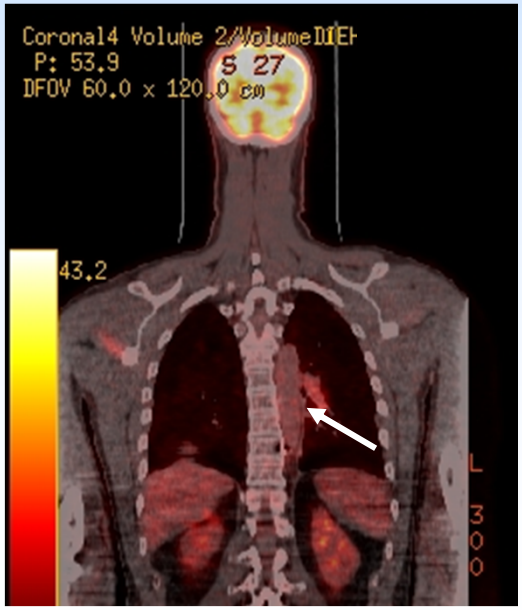
44 year old US farmer with third systemic relapse of Ewing's sarcoma

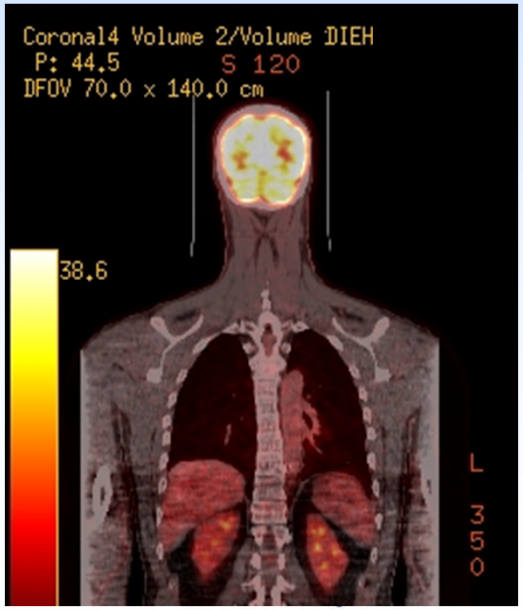












SARC Protocol #: SARC 011
Hoffmann-La Roche Protocol #: N021157

TITLE: SARC Global Collaboration*: A Phase II Trial of R1507, a Recombinant Human Monoclonal Antibody to the Insulin-Like Growth Factor-1 Receptor for the treatment of patients with recurrent or refractory Ewing's sarcoma, osteosarcoma, synovial sarcoma, rhabdomyosarcoma and other sarcomas

*SARC Global Collaboration represents collaboration among the Innovative Therapies for Children with Cancer (ITCC), Istituti Ortopedici Rizzoli, European Organization for Research (EORTC/STBG), Cooperative Osteosarcoma Study Group (COSS), EuroEwings, Euramos

Sponsor: F. Hoffmann-LaRoche

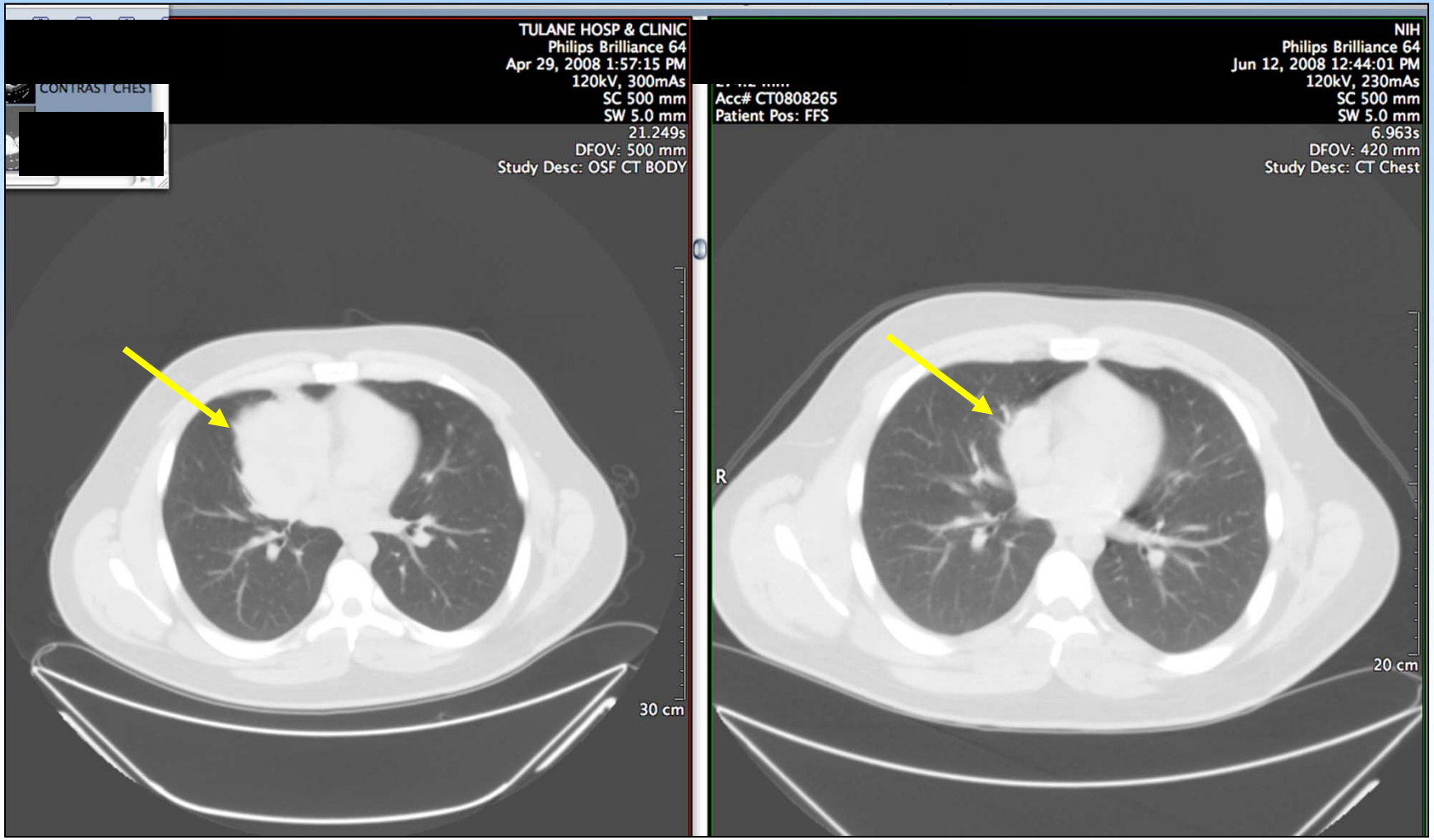
Coordinating Center: SARC (Sarcoma Alliance for Research through Collaboration)

SARC Sarcoma Alliance for Research through Collaboration
Collaborating for a Cure.

17 yo boy with multiply recurrent Ewing's sarcoma

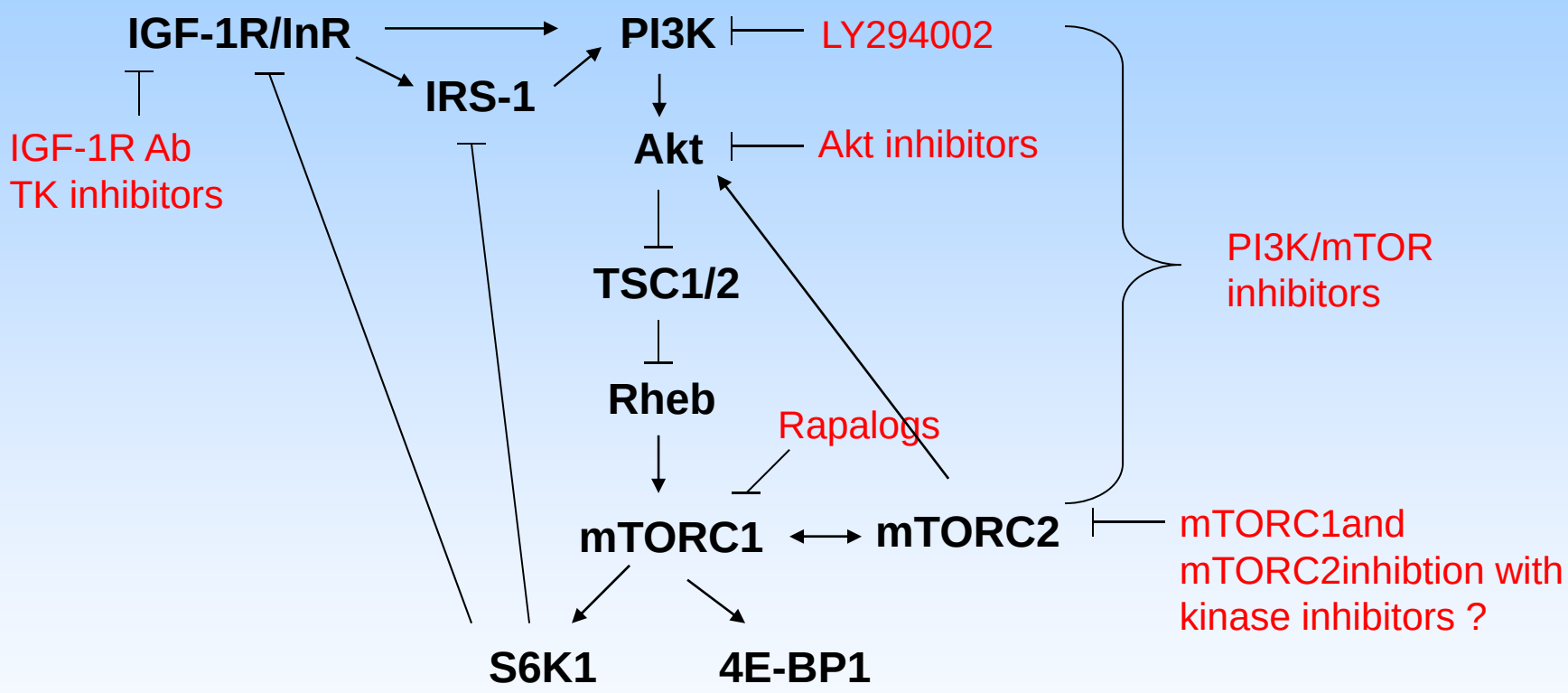
Baseline CT Chest

Week 6 CT Chest



Conclusions

- Humanized IGFIR moAb shows remarkable clinical activity in Ewing's sarcoma patients in early Phase I studies
- Early evidence to suggest beneficial combination of mTOR inhibition combined with IGFIR inhibition
- Phase II study ongoing-responses ongoing
- Planned study using mTOR inhibitor plus IGFIR moAB


IGF-1R/InR
PI3K
LY294002
IRS-1
IGF-1R Ab
TK inhibitors
Akt
Akt inhibitors
TSC1/2
PI3K/mTOR
inhibitors
Rheb
Rapalogs
mTORC1
mTORC2
mTORC1and
mTORC2inhibtion with
kinase inhibitors ?
S6K1
4E-BP1

Lee Helman

Seth Cohen Arnulfo Mendoza (not shown)

Chand Khanna Choh Yeung Issac Darko(not shown)

Sung-Hyeok Hong Brieanne Midura

Xiaolin Wan Ling Ren Patrick Grohar Kartik Krishnan Melissa Paoloni

Martin Mendoza Christine Mazcko Brian Harkavy Duane Currier (not shown)

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