OSU Research Progress Update: February 2012

1. Progress on AR-42 (HDAC-42) as a potential therapy for NF2-associated schwannomas and meningiomas:

Recently we (Bush et al., 2011. Neuro-Oncology 13:983-999) showed that AR-42 inhibited the growth of primary human vestibular schwannoma and *Nf2*-deficient mouse schwannoma cells with IC_{50} values of about 0.5 nM. AR-42 also inhibited primary meningioma cells and the benign meningioma cell line, Ben-Men-1, with IC_{50} values of about 1~1.5 nM. We also found that AR-42 treatment reduced AKT activation and induced cell cycle arrest at G2/M and apoptosis in both schwannoma and meningioma cells. To quantitatively measure the growth inhibitory activity of AR-42, we have established luciferase-expressing Ben-Men-1 meningioma and *Nf2*-deficient mouse schwannoma cells. Using bioluminescence imaging, we showed that AR-42 potently inhibited the growth of intracranial Ben-Men-1-LucB meningioma xenografts by as much as 90% after treatment for three months. Although we found that AR-42 was well-tolerated in mice, AR-42 also inhibited the proliferation of normal meningeal cells with a similar IC_{50} of about 1 nM. Treatment of meningeal and Ben-Men-1 meningioma cells with AR-42 increased the expression of the pro-apoptotic factor Bim while decreasing the expression of the anti-apoptotic factor Bcl_{XL}. In addition, expression of the CDK inhibitors $p16^{INK4A}$, $p21^{CIP1/WAF1}$, and $p27^{KIP1}$ increased in both meningeal and Ben-Men-1 cells. However, AR-42 induced cell cycle arrest at the G1 phase in normal meningeal cells while it arrested Ben-Men-1 meningioma cells at G2. One reason normal meningeal cells are spared from AR-42 inhibition is that G1-arrested normal cells exit the cell cycle (at G0) and remain dormant. Cells can survive in G0 for some time. In contrast, AR-42 arrests tumor cells at G2. As cells at G2 are already prepared to undergo mitosis (M phase), they cannot exit the cell cycle. Prolonged G2 arrest induces apoptotic signals and cell death in tumor cells. These results suggest that G1-arrested normal meningeal cells may exit the cell cycle and remain dormant, while meningioma cells arrested at G2 by AR-42 cannot exit the cell cycle, consequently leading to apoptotic cell death. A manuscript describing these findings is being prepared for publication.

2. Progress on the investigation of natural compounds for the treatment of NF2-associated tumors:

We have screened 19 plant-derived natural compounds and found four of them potently inhibit the growth of schwannoma and meningioma cells with sub-micromolar or nanomolar IC_{50} values. Particularly, we showed that the growth inhibitory activity of *silvestrol,* a rocaglate derivative from the tropical Asian plant *Aglaia foveolata,* in schwannoma and meningioma cells (IC_{50} of about 5-10 nM) is 1,000-fold or more potent than four anti-tumor natural compounds: curcumin (the principal curcuminoid in turmeric, which is a spice derived from the rhizomes of *Curcuma longa,* a member of the ginger family), CAPE (caffeic acid phenyl ester, an active ingredient in propolis extracts *[Bio30]), resveratrol* (a polyphenolic compound found in grapes, red wine, purple grape juice, peanuts, and some berries), and *sulforaphane* (a phytochemical belonging to the family of *isothiocyanates* found in cruciferous vegetables, such as broccoli, cauliflower, cabbage and kale). Note that *silvestrol* has been shown to possess potent *in vitro* and *in vivo* activities in multiple cancer models, including acute lymphoblastic leukemia (ALL) and is currently under pre-clinical development by NCI. Also, we found that *silvestro*l induced cell cycle arrest at G2/M in schwannoma and meningioma cells. We are presently investigating the signaling pathways that are affected by *silvestrol.*