postulate that mimicking LH GABAergic activity will produce its previously
demonstrated anxiolytic effects. DISCUSSION/SIGNIFICANCE OF IMPACT:
Identifying the important role for a reward-oriented feeding center in the brain
in producing antipsychotic weight gain will allow a more comprehensive,
ethologically sound approach to behavioral modification therapy in these
patients. It will lend mechanistic credence to weight control therapies which
have used token economy, opioid antagonism, and other inhibition-promoting
therapies. This study will also increase the validity for testing further the use of
selective serotonin agonists which prevent weight gain such as lorcaserin.

Innovative 3D printed intravaginal rings for
contraception and HIV prevention
Rahima Benhabbour, Rima Janusziewicz, Sue J. Mecham,
Roopali Shrivastava and Gayane Parvayan

OBJECTIVES/SPECIFIC AIMS: The long-term goal of this project is to develop a
cost effective 3D printed multipurpose intravaginal ring (IVR) to prevent against
unintended pregnancies and infectious diseases. Our goal is to develop a female-
controlled method for prevention using innovative IVRs. METHODS/STUDY
POPULATION: In vitro and in vivo characterization. RESULTS/ANTICIPATED
RESULTS: Controlled and fine-tuned release kinetics 100% drug release from
3D printed IVRs compared with 10%–15% with traditional injection molded
IVRs cost-effective engineering of multipurpose IVR with CLIP 3D printing
technology. DISCUSSION/SIGNIFICANCE OF IMPACT: If successful, this
project will revolutionize the engineering of IVRs and will have a global impact
on human health. Not only will we help save millions of women around the
world but also millions of children that are infected by their HIV-positive
mothers through gestation or breast feeding.

Hydrogen bonding and water accessibility changes
upon expansion of PolyQ tracts in ataxin-2 and ataxin-3
Jingran Wen, Daniel Scales and Julio C. Facelli

OBJECTIVES/SPECIFIC AIMS: Polyglutamine (polyQ) neurodegenerative dis-
eases, associated with the unstable expansion of polyQ tracts, are devastating
diseases for which no treatments exist. Moreover, most drug discovery
attempts have been hindered by the lack of understanding on the relevant
pathogenic mechanisms. Here, using previously reported 3D protein predicted
structures of ataxin-2 and ataxin-3, we analyze the effect of polyQ enlargement
on hydrogen bonding and water accessibility patterns as a possible mechanism
for pathogenesis thought enhanced protein aggregation. METHODS/STUDY
POPULATION: Using the I-TASSER predicted structures of ataxin-2 and
ataxin-3 with different numbers of glutamine repeats representing polyQ
lengths characteristic of both normal and pathological tracts (Journal of
Biomolecular Structure and Dynamics, 2016: 1–16), we identified hydrogen bonds
(HBs, UCSF Chimera FindHBond module) and calculated solvent-accessible
surface areas (SASA, DSSP program) for the polyQ tracts available in the 3D
structures. RESULTS/ANTICIPATED RESULTS: The identified HBs were
analyzed as the function of the number of glutamines in the polyQ tracts and
categorized as those intra-polyQ and exter-polyQ, respectively. The SASA of
the polyQ region was also studied as the function of the polyQ tract length.
DISCUSSION/SIGNIFICANCE OF IMPACT: The results obtained here indicate
that polyQ regions increasingly prefer self-interactions, which consistently can
lead to more compact polyQ structures. The results strongly support the notion that the expansion of the polyQ region can be an intrinsic force leading
to self-aggregation of polyglutamine proteins and suggest that the modulation of
solvent-polyQ interactions could be a possible therapeutic strategy for polyQ
diseases.

Investigation of sAC signaling reveals new therapeutic
targets for cancer cell metabolism
Jenny Wang and Jonathan Zippin

OBJECTIVES/SPECIFIC AIMS: The soluble adenyl cyclase (sAC) is a
noncoinferenced source of cAMP in mammalian cells. sAC is an ATP/bicarbonate
ion sensor, whose activity responds to intracellular signals such as pH changes and
metabolism. Unlike the more traditionally studied transmembrane adenyllyl
cyclase, sAC is not tethered to the cell membrane and instead is found in
subcellular microdomains like the mitochondria and nucleus. In particular, sAC
localization in the mitochondria has been implicated in oxidative phosphoryla-
tion and mitochondrial metabolism. Specific changes in sAC mitochondrial
localization have diagnostic utility in a wide variety of cancers, namely
melanoma. We have recently found that loss of sAC leads to tumorigenesis
and a Warburg/cancer-like metabolic phenotype, consisting of an activated flux
through glycolysis, increased lactate production, and dependence on glucose
metabolism. In addition, computational analysis of the metabolomics profile of
sAC null cells suggests an increased flux through serine synthetic pathways.
We hypothesized that specific sAC microdomains are responsible for this cancer-
like metabolic state. METHODS/STUDY POPULATION: We have established
oncogenic SV40 large T antigen and HPV16-E6 expressing mouse embryonic
fibroblasts lacking sAC expression (SV40 KO and E6 KO, respectively). Using
these parental lines, we reintroduced sAC by targeting the protein to specific
microdomains. sAC was either driven into the mitochondria (mito-sAC) or was
driven into all possible microdomains (WT sAC). Single clones were generated and
sAC expression was confirmed by Western analysis. Stable cell lines were
evaluated for mitochondrial metabolism, glucose sensitivity, and serine
sensitivity. RESULTS/ANTICIPATED RESULTS: We found that reintroduction
of WT sAC into sAC null cells rescued sensitivity to glycolytic inhibition
compared with control cells (p < 0.01). The effect was not dependent on the
method of immortalization as it was seen in both SV40 and E6 KO cell lines. sAC
activity was not directly proportional to expression suggesting that additional
regulatory pathways exist. Interestingly, targeted delivery of sAC to the
mitochondria was not as effective in rescuing glucose sensitivity as untargeted
delivery of sAC into all sAC microdomains. Therefore, even though
mitochondrial sAC is known to influence metabolism, our data suggests that the
nonmitochondrial isoform is most important for cancer cell metabolism.
Although metabolomics analysis suggested that serine synthetic pathways are
activated in sAC null cells, there is no evidence to suggest that serine is required
for sAC null cell growth. Neither inhibition of serine synthesis nor serine
starvation differentially affected the growth of sAC null cells compared with
WT sAC. DISCUSSION/SIGNIFICANCE OF IMPACT: These data suggest that
the Warburg metabolic phenotype in sAC null cells is regulated by specific sAC
microdomains. By targeting sAC to specific microdomains, we can further
distinguish the role of sAC localization in cellular metabolism. Cancer cells
have been shown to exhibit altered metabolic circuitry of pathways like glycolysis,
which allow them to adapt to increased metabolic demands of cellular
proliferation and waning environmental resources. Beyond helping us improve
the use of sAC immunolocalization as a cancer diagnostic, a better under-
standing of sAC microdomains in transformed cells will help us understand how
this signaling pathway is important in cancer. Pharmacologic manipulation of
sAC signaling may represent a new cancer therapeutic strategy.

In silico prediction of NS1 structure and influenza A
virus pathogenesis
Joshua Klonoski and Julio C. Facelli

OBJECTIVES/SPECIFIC AIMS: This poster presents preliminary results of using
in silico approaches to predict a priori, based on sequence alone, the
pathogenesis of novel influenza A virus. METHODS/STUDY POPULATION:
Here we analyzed the structure of the NS1 protein of 11 strains of well
characterized influenza A virus with known pathogenesis, reported in the
literature as LD50 values, and published sequences. We performed homology
comparison of these sequences using the ExPasy SIM alignment tool for protein
sequences and then predicted their 3D structures using the I-TASSER method
for protein structure prediction. We retained the best 20 I-TASSER models for
the NS1 sequences considered here and compared their structures with that of
the X-ray crystallographic structure of the NS1 protein in the A/Blue-winged
teal/MN/993/1980 (H6N6). The average RMS between this experimental
structure and the best 20 I-TASSER models was used as a measure of structural
similarity between the 3D structures among the proteins. RESULTS/
ANTICIPATED RESULTS: The sequence homology shows modest correlation
between sequence and pathogenicity. Linear correlations with R values as large
as 0.6 were observed for the full sequence homology and the homology of the
RBBD domains of the proteins. The correlations with the other protein domains
were significantly lower. We did not find overall correlation between the
3D structures and pathogenesis of all the variants considered here, but the
initial results suggest that correlations do exist for different subgroups of
viruses. In future work we will use advanced data mining methods to better
understand clustering and correlation between structure and pathogenesis.
DISCUSSION/SIGNIFICANCE OF IMPACT: The results presented in this
poster demonstrate, as proof of concept, the use of in silico approaches to
determine pathogenesis of viruses with substantial impact on human health.
The ability of computationally predicting pathogenesis of rapidly mutating viruses