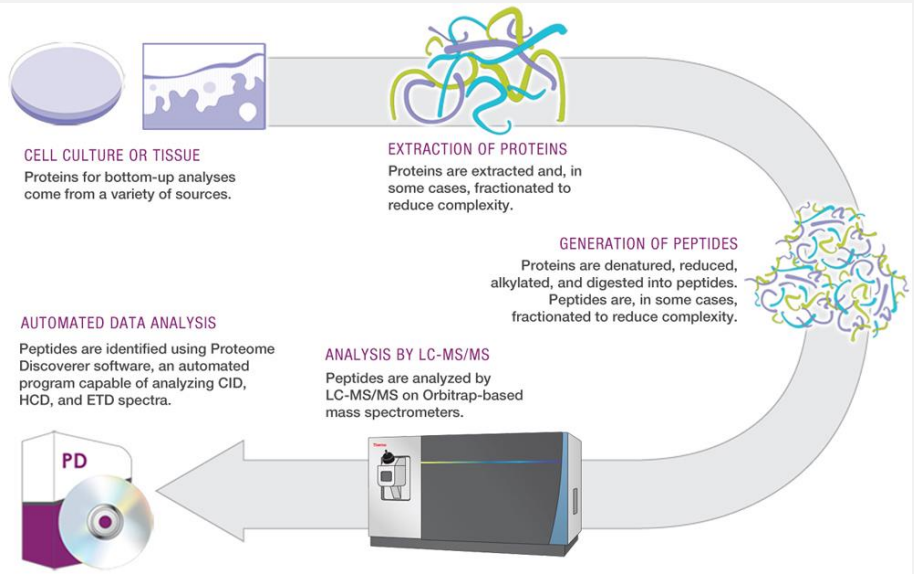
MY EXPERIENCE AT WHIRC

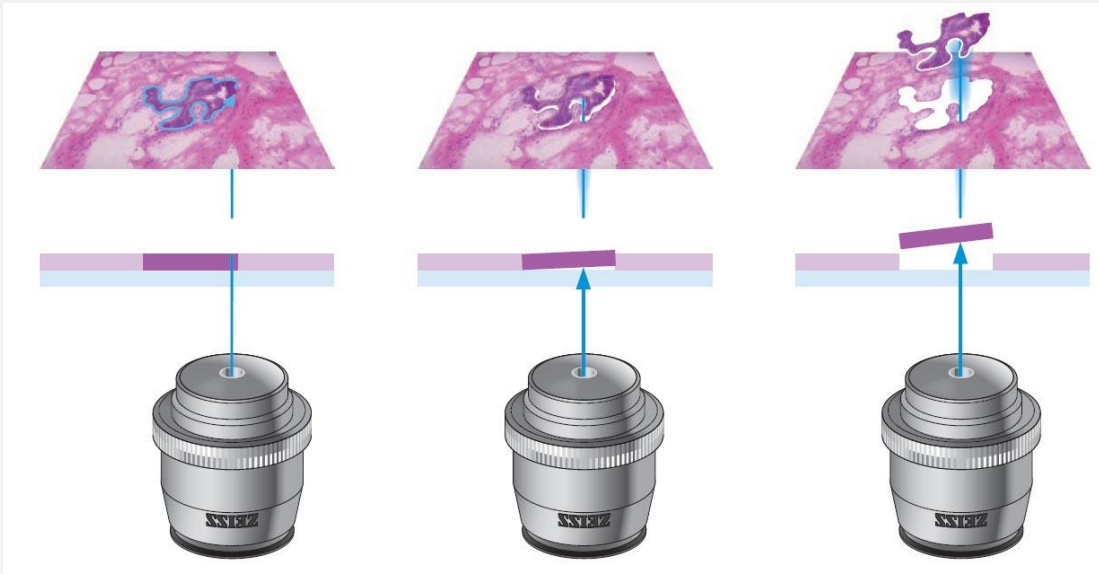
Roshini Balan

PROTEOMIC ANALYSIS

Mass Spectrometry

Laser Microdissection





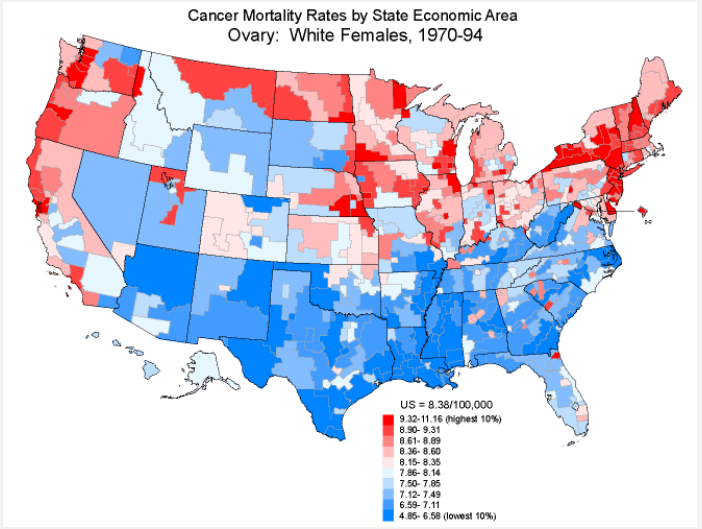
MAJOR PROJECT: VITAMIN D + PROGESTERONE

HYPOTHESIS/GOALS OF THE VITAMIN D STUDY

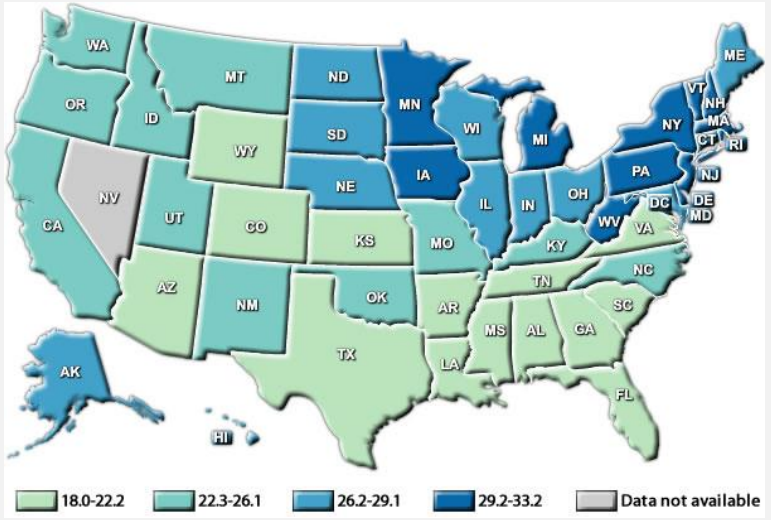
- To further characterize the effects of progestins on vitamin D metabolism in cells derived from ovarian epithelium

- To search for progestin-related biological effects of vitamin D metabolism

OVARIAN AND UTERINE CANCER STATISTICS



Higher risk



Higher risk

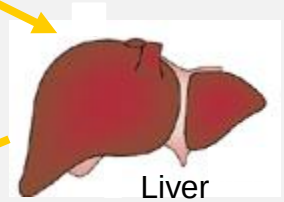
VITAMIN D

UVB irradiation

Skin

7-Dehydrocholesterol

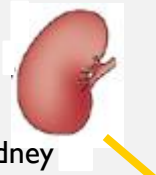
Vitamin D3 (Cholecalciferol)



CYP27A1

Liver

25-hydroxyvitamin D3 (Calcidiol or Calcifediol)



Kidney

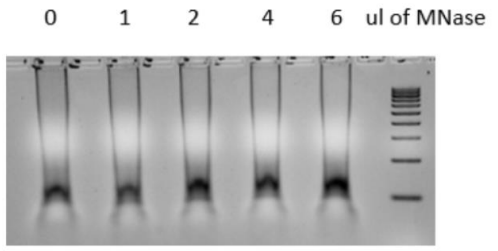
CYP27B1

1,25-dihydroxyvitamin D3 (physiologically active form)
(Calcitriol)

- Maintains Ca2+ balance
- Increases bone mineralization
- Induces differentiation of immune cells
- Inhibits angiogenesis
- Inhibits proliferation (cell cycle arrest, apoptosis)

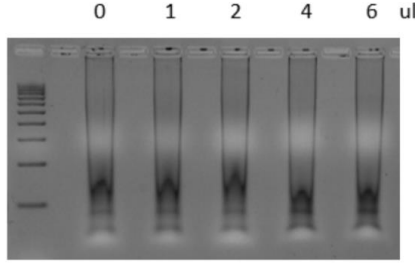
WHAT I WORKED ON: OPTIMIZATION OF MNASE DIGESTION

Round I



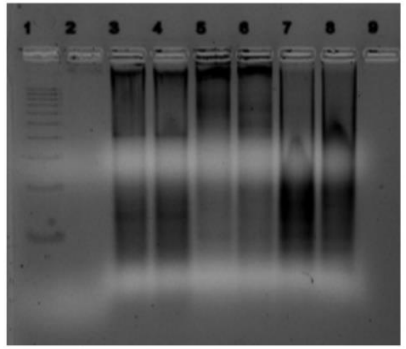
3% gel

Gel Electrophoresis Results



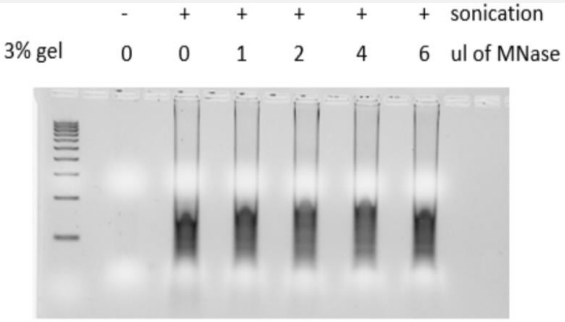
3% gel (ran longer)

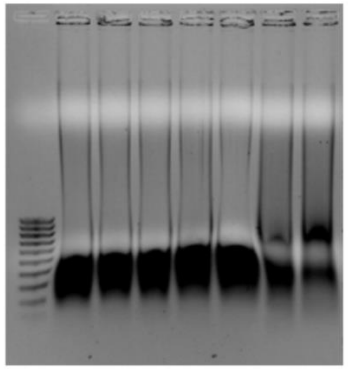
Round III



+ control (1ug of intact DNA)

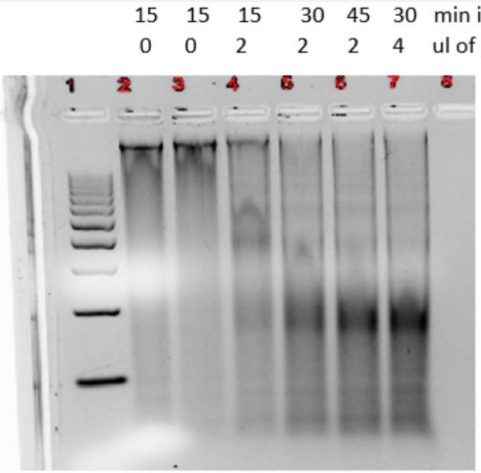
Round II





1% gel, last 2 lanes were 1:1 diluted samples

Round IV



BIOANALYZER

Nanodrop Results				
Assay Date	**Mnase (uL)**	**Time (Min)**	**Sample ID**	**[ ]**
6/17/2019	0	15	A	722.7
6/17/2019	2	15	B	966
6/17/2019	2	30	C	909.7
6/17/2019	2	45	D	1858.5
6/17/2019	4	30	E	1081.6
7/3/2019	0	15	0A	708.8
7/3/2019	0	30	0B	1014.5
7/3/2019	4	30	4A	1118
7/3/2019	4	30	4B	912

The **Bioanalyzer** is a chip-based capillary electrophoresis machine to analyse RNA, DNA, and protein.

Microchannels are used for separation of nucleic acid fragments based on their size as they are driven through it electrophoretically.

PROCEDURES LEARNED

MASS SPECTROMETRY

- Machine calibration
- Running standards
- Observed running samples

LASER MICRODISSECTION

- Cut sections of tissue for further analysis
- Scanned and labeled ~300 slides in preparation

CELL CULTURE

- Prepared cells for different experiments
- Split + trypsinized cells

MISTAKES MADE

QUESTIONS?