

A KNOWLEDGEBASE ANALYSIS OF ROSEMARY OFFICINALIS

By: Aneeta Uppal

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Speaker & Moderator



Aneeta Uppal

Aneeta is a second year Ph.D. student at the University of North Carolina at Charlotte in Bioinformatics. She received her undergraduate degree in Biotechnology from the Rochester Institute of Technology in 2015. She later moved to Charlotte and received her masters degree in bioinformatics at UNC Charlotte in 2016. She currently spends her time flipping between wet-lab science and computational biology. Her current research focuses on essential oils for human health.



Lori Pollock

Dr. Lori Pollock is a Professor in Computer and Information Sciences at University of Delaware. She earned her Ph.D. and M.S. in CS at the University of Pittsburgh. Her current research focuses on program analysis for building better software maintenance tools, software testing, energy-efficient software and computer science education. Dr. Pollock is an ACM Distinguished Scientist and was awarded the University of Delaware's Excellence in Teaching Award and the E.A. Trabant Award for Women's Equity. She leads the Partner4CS, a projects dedicated to integrating computer science in middle and high schools in Delaware through teacher professional development for the CS10K national efforts.



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ESSENTIAL OIL (EO) INDUSTRY

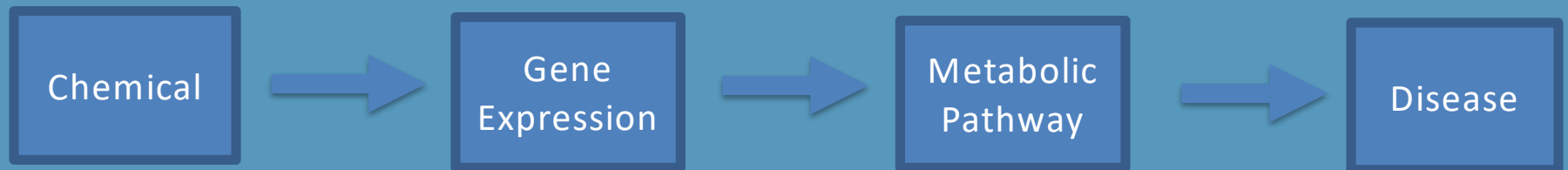
- Exceeded USD 6.0 billion in 2015
- Large growth in consumer demand for EOs and all natural based products
- Hundreds of brands of oils are now on the market
- Not FDA regulated

WHAT IS AN EO? WHAT IS IN IT?

- EOs are volatile compounds extracted from a plant
- They are extracted or produced from:
 - Steam distillation (3 methods)
 - Cold-pressed extraction (citrus rinds)
 - Solvent extraction

CURRENT RESEARCH ON EOS

- About 16,000 published studies in NCBI Pubmed on EOs – not a lot compared to popular topics
- Many studies on the most popular oils (ex. Lavender)
- Are the claims what they are said to be?
- Can oils be used to target and treat specific health conditions and disease?



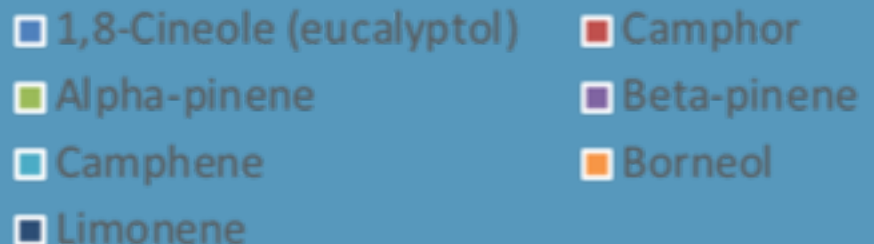
ROSEMARY APPLICATIONS ACCORDING TO A TRADITIONAL EO REFERENCE GUIDE

- For arthritis/muscle cramps (anti-inflammatory properties)
- Mental Fatigue – to enhance focus and prevent Alzheimer's disease
- Has anti-cancer properties
- **Can Rosemary do all of these things?! Maybe? But why, and how?!**

ROSEMARY EO

- Extremely commonly used essential oil and herb
- GC/mass spec shows Rosemary is primary composed of:
 - With small possible varying percentages
 - Based on chemotype, environment where it was grown, and time of harvest & distillation

Composition Of Rosemary EO



HYPOTHESIS

- Using bioinformatics computational approaches, we can learn the possible health effects of Rosemary EO based on its chemical constituents.

METHODS: LINGUAMATICS

Query editor Smart query

essential oil volati... main group molecular... Gene/Protein Evidence #Docs

- Join (document)
 - AI_EOs.i2qy (1 output column, 0 parameters)
 - Join (document)
 - Chemical.i2qy (1 output column, 0 parameters)
 - Join (column and document)
 - gene.i2qy (1 output column, 1 parameter)
 - Subtraction
 - Union
 - Subtraction
 - Union
 - gene_with_long_name.i2qy (2 output columns, 0 parameters)
 - gene_with_greek_letter.i2qy (2 output columns, 0 parameters)
 - gene_near_gene_term.i2qy (2 output columns, 0 parameters)
 - gene_in_apposition_with_longer_form.i2qy (2 output columns, 0 parameters)
 - gene_in_list_of_genes.i2qy (2 output columns, 0 parameters)
 - Subtraction
 - gene_with_multiple_synonyms.i2qy (2 output columns, 0 parameters)
 - gene_with_similar_synonyms.i2qy (2 output columns, 0 parameters)
 - Subtraction
 - gene_with_multiple_synonyms.i2qy (2 output columns, 0 parameters)
 - gene_with_similar_synonyms.i2qy (2 output columns, 0 parameters)
 - gene_in_apposition_with_non-gene.i2qy (2 output columns, 0 parameters)

Abstract, Other section

[1]

main group molecular entity

s0w (ordered)

essential oil

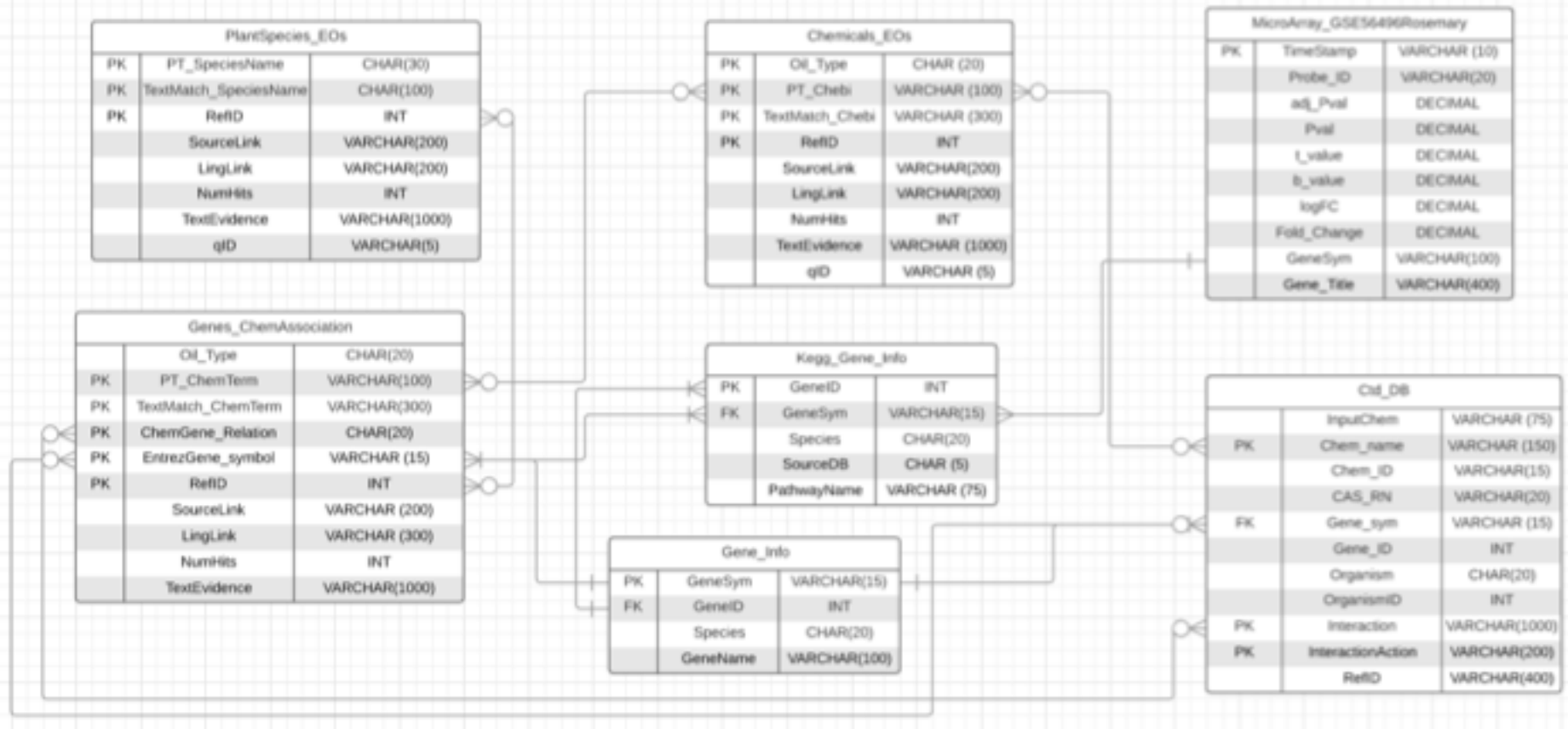
MIC protein lipid

METHODS: LINGUAMATICS

essential oil volatL	main group molecular	Gene/Protein	Chemical Gene	Evidence	#Docs	Doc#	Doc	#Hits	Hit#	Hit
essential oil	1-[5- isoquinolinesulfonyl]-2- methylpiperazine	ACSM1	activate	gene term	1	1	27322724	28	1	... ginate (LAE) is a water-soluble cationic surfactant which has antimicrobial activity against a broad spectrum of ... Some spice essential oils are effective against antimicrobials. ... Escherichia coli O157 H7, and Listeria monocytogenes, ... to the MICs of free LAE showed that binding among the LAE and Tween ...
essential oil	1-acyl-sn-glycero-3- phosphoserine	GORA5P1	activate	gene term	1	1	27387396	9	8	The activation of NF- κ B p65 in colonic mucosa was also ... addition, MQEO significantly suppressed LPS-stimulated production of TNF- α and IL-1 β Essential oil
essential oil	1-acyl-sn-glycero-3- phosphoserine	IL1B	block	greek character	1	1	27387396	27	10	... showed that the Magian fruits essential oil (MQEO) exhibited an ... the mRNA levels of IL-1 β , IL-6, IL-12p35 and ... The LPS-stimulated THP-1 cell line was used for exploring the ... LPS-stimulated production of TNF- α and IL-1 β , effectively blocked phosphorylation of IKK and I κ B...
essential oil	1-acyl-sn-glycero-3- phosphoserine	IL1B	concentrate	gene term	1	1	27387396	27	1	Protective effect of the essential oil of Zanthoxylum myriacanthum var. pubescens ... the damage of colons, the mRNA levels of IL-1 β , IL-6, IL-12p35 and TNF- α The LPS-stimulated THP-1 cell line was used for exploring the ... LPS-stimulated production of TNF- α and IL-1 β , effectively blocked phosphorylation of ... TLR4 in THP-1 cells at concentrations ranging from 0.01% to 0.05% ...

Reducing noise can be very difficult and takes most of my time...

METHODS: POSTGRESQL



Entity-Relationship Diagram

TAKING A CLOSER LOOK AT ROSEMARY

Microarray dataset of
over-expressed/ under-
expressed genes

Linguamatics chemical
– gene associations of
Rosemary constituents

Take Gene sets in
common and look at
the associated
pathways



GENE-CHEMICAL ASSOCIATIONS

```
SELECT *
FROM ChemGenesCtd JOIN ChemGenesLing ON (ChemGenesCtd."Gene_sym" = ChemGenesLing."EntrezGene_symbol" AND ChemGenesCtd."Chem_name" =
ChemGenesLing."PT_ChemTerm");
```

```
SELECT *
FROM ChemGenesLing JOIN public."Kegg_Gene_Info" ON (ChemGenesLing."EntrezGene_symbol" = "Kegg_Gene_Info"."GeneSym");
```



EntrezGene_symbol	PT_ChemTerm	ChemGene_Relation	GeneSym	GeneID	Species	SourceDB	PathwayName
TP53	1,8-cineole	regulate	TP53	10404	Homo sapiens	KEGG	Hepatitis C
TP53	1,8-cineole	regulate	TP53	514	Homo sapiens	KEGG	Bladder cancer
TP53	1,8-cineole	regulate	TP53	582	Homo sapiens	KEGG	Chronic myeloid leukemia
TP53	1,8-cineole	regulate	TP53	597	Homo sapiens	KEGG	p53 signaling pathway
TP53	1,8-cineole	regulate	TP53	593	Homo sapiens	KEGG	Pancreatic cancer
TP53	1,8-cineole	regulate	TP53	570	Homo sapiens	KEGG	Basal cell carcinoma
NFKBIA	(-)-beta-caryophyllene	phosphorylate	NFKBIA	4389	Homo sapiens	KEGG	Chemokine signaling pathway
NFKBIA	(-)-beta-caryophyllene	phosphorylate	NFKBIA	4397	Homo sapiens	KEGG	Pathways in cancer
NOS3	(-)-beta-elemene	increase	NOS3	462	Homo sapiens	KEGG	Arginine and proline metabolism
NOS3	(-)-beta-elemene	increase	NOS3	432	Homo sapiens	KEGG	VEGF signaling pathway
NOS3	(-)-beta-elemene	increase	NOS3	594	Homo sapiens	KEGG	Calcium signaling pathway
APOE	(-)-beta-elemene	inhibit	APOE	521	Homo sapiens	KEGG	Alzheimer's disease
NOS3	(-)-beta-elemene	maintain	NOS3	462	Homo sapiens	KEGG	Arginine and proline metabolism
NOS3	(-)-beta-elemene	maintain	NOS3	432	Homo sapiens	KEGG	VEGF signaling pathway
NOS3	(-)-beta-elemene	maintain	NOS3	594	Homo sapiens	KEGG	Calcium signaling pathway

COMPARING TO MICROARRAY DATASETS CAN PROVIDE GENES OF INTEREST

```
CREATE VIEW GSE56496OverExp AS
SELECT *
FROM GSE56496 JOIN ChemGenesLing ON (GSE56496."GeneSym" = ChemGenesLing."EntrezGene_symbol") WHERE GSE56496."Fold_change" >1.4;

SELECT * FROM GSE56496OverExp JOIN public."Kegg_Gene_Info" ON (GSE56496OverExp."GeneSym" = "Kegg_Gene_Info"."GeneSym");

CREATE VIEW GSE56496UnderExp AS
SELECT *
FROM GSE56496 JOIN ChemGenesLing ON (GSE56496."GeneSym" = ChemGenesLing."EntrezGene_symbol") WHERE GSE56496."Fold_change" < .5 ;

SELECT * FROM GSE56496UnderExp JOIN public."Kegg_Gene_Info" ON (GSE56496UnderExp."GeneSym" = "Kegg_Gene_Info"."GeneSym");
```

TimeStamp	Fold_change	EntrezGene_symbol	PT_ChemTerm	ChemGene_Relation	PathwayName
A_10018	1.720242197	HMGCR	terpenes	overexpress	Terpenoid backbone biosynthesis
A_10018	1.720242197	HMGCR	monoterpene	overexpress	Terpenoid backbone biosynthesis
A_10018	1.720242197	HMGCR	camphor	overexpress	Terpenoid backbone biosynthesis
A_10018	1.720242197	HMGCR	(R)-camphor	overexpress	Terpenoid backbone biosynthesis
A_10018	1.720242197	HMGCR	1,8-cineole	overexpress	Terpenoid backbone biosynthesis
A_10018	1.720242197	HMGCR	terpenes	overexpress	Bile secretion
A_10018	1.720242197	HMGCR	monoterpene	overexpress	Bile secretion
A_10018	1.720242197	HMGCR	camphor	overexpress	Bile secretion
A_10018	1.720242197	HMGCR	(R)-camphor	overexpress	Bile secretion
A_10018	1.720242197	HMGCR	1,8-cineole	overexpress	Bile secretion
A_14785	2.879213756	NLN	terpenes	overexpress	Renin-angiotensin system
A_14785	2.879213756	NLN	pinene	overexpress	Renin-angiotensin system

HIGHLIGHTED RESULTS

Chemical	Relationship	Gene Symbol	Highlighted Pathways involved
1,8-cineole & limonene	Increased expression	BAX & BCL2	Colorectal, Small cell lung & Prostate cancer
1,8-cineole	Increased expression	IL10	Intestinal immune network for IgA production, Cytokine-cytokine receptor interaction, T cell receptor signaling pathway
1,8-cineole	Increased expression	MME	Alzheimer's disease
1,8-cineole & limonene	Increased expression	COL11A2	ECM-receptor interaction, Focal adhesion, Protein digestion & absorption
1,8-cineole & camphor	Increased expression	HMGCR	Bile secretion, Terpanoid backbone biosynthesis

Table 1: Highlighted results of using the knowledgebase for the chemical constituents of Rosemary EO

COMPARISONS

TimeStamp	Fold_change	GeneSym	PT_ChemTerm	ChemGene_Relation	PathwayName	Species
A_272	3.865099087	IL10	1,8-cineole	increased expression	Cytokine-cytokine receptor interaction	Homo sapiens
A_272	3.865099087	IL10	1,8-cineole	increased expression	T cell receptor signaling pathway	Homo sapiens

Oil_Type	PT_ChemTerm	TextMatch_ChemTerm	EntrezGene_sy...	ChemGene_Relation	RefID	SourceLink
essential oil	1,8-cineole	eucalyptol	IL10	increased expression	21830186	http://www.ncbi.nlm.nih.gov/pubmed/21830186

Artemisia princeps Pamp. Essential oil and its constituents eucalyptol and α -terpineol ameliorate bacterial vaginosis and vulvovaginal candidiasis in mice by inhibiting bacterial growth and NF- κ B activation.

Trinh HT¹, Lee JA, Hyun YJ, Kim DH.

⊕ Author information

Abstract

To investigate the inhibitory effects of *Artemisia princeps* Pamp. (family Asteraceae) essential oil (APEO) and its main constituents against bacterial vaginosis and vulvovaginal candidiasis, their antimicrobial activities against *Gardnerella vaginalis* and *Candida albicans* in vitro and their anti-inflammatory effects against *G. vaginalis*-induced vaginosis and vulvovaginal candidiasis were examined in mice. APEO and its constituents eucalyptol and α -terpineol were found to inhibit microbe growths. α -Terpineol most potently inhibited the growths of *G. vaginalis* and *C. albicans* with MIC values of 0.06 and 0.125% (v/v), respectively. The antimicrobial activity of α -terpineol was found to be comparable to that of clotrimazole. Intravaginal treatment with APEO, eucalyptol, or α -terpineol significantly decreased viable *G. vaginalis* and *C. albicans* numbers in the vaginal cavity and myeloperoxidase activity in mouse vaginal tissues compared with controls. These agents also inhibited the expressions of proinflammatory cytokines (IL-1 β , IL-6, TNF- α), COX-2, iNOS, and the activation of NF- κ B and increased expression of the anti-inflammatory cytokine IL-10. In addition, they inhibited the expressions of proinflammatory cytokines and the activation of NF- κ B in lipopolysaccharide-stimulated peritoneal macrophages, and α -terpineol most potently inhibited the expressions of proinflammatory cytokines and NF- κ B activation. Based on these findings, APEO and its constituents, particularly α -terpineol, ameliorate bacterial vaginosis and

CONCLUSION

- Need more data before any solid conclusions can be drawn
- Given a basic gene sets and pathways to investigate further
- Increased expression of IL10 (an anti-inflammatory cytokine)
- BAX and BCL2 are involved in P53-mediated apoptosis
- MME is a membrane metalloendopeptidase that degrades beta-amyloid

CONCLUSION

- This evidence suggests that it could be possible Rosemary EO may be effective for treating arthritis, cramps, and help as a preventative against Alzheimer's disease
- Gives researchers a candidate gene set to study further (this can save time and money)

FUTURE WORK

- Eliminate noise – look for errors in the data
- Add more expression data
- Drugbank database information on chemicals
- RNA-sequence data (my own)
- Possibility to add machine learning algorithms

ACKNOWLEDGEMENTS

- Dr. Cory Brouwer
- Dr. Jeremy Jay
- UNC Charlotte Bioinformatics Services Division

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MAKING YOUR HEALTH A PRIORITY IN GRAD SCHOOL

By: Aneeta Uppal

Bioinformatics Ph.D. Student, UNC Charlotte



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A LITTLE BIT ABOUT ME...

- Chronic Autoimmune urticaria – 2015
 - Xolair injections
 - Side effects...
- Hashimoto's disease – 2017
- 50% increased chance of developing another autoimmune disease (Multiple sclerosis, Lupus, rheumatoid arthritis) Scary.

WHY ME?!

- I mean I was healthy, physically? I CrossFit all the time!
- I was young?!
- I ate decent, but I wasn't that strict
- It's hard to determine what triggered this cascade, but after correcting how I was managing my stress, nutrition, exercise schedule, vitamins and supplements, I've seen a huge change

AUTOIMMUNE DISEASES

- The causes of autoimmune disease are still unknown, but research suggests that it could be a number of things including:
 - Genetics
 - Viruses
 - Toxins
 - Environmental pollutants
 - Hormonal imbalances
 - Leaky gut
 - And many other factors

WHAT WAS IN MY CONTROL TO CHANGE?

- Nutrition
- Vitamins and supplements
- Stress management
- Learning to say NO
- Not overloading my schedule
- Listening to my body (over-exercise)

IMPROVEMENT AFTER 2.5 MONTHS

- Thyroid Peroxidase (TPO) Ab
 - Ref Range (0-34 IU/mL)
 - 8/02/17: 204 IU/mL
 - 10/20/17: 193 IU/mL
- Thyroglobulin Antibody
 - Ref: 0.0-0.9 IU/mL
 - 08/02/17: 40.9 IU/mL
 - 10/20/17 28.8 IU/mL
- Thyroid stimulating hormone (TSH)
 - Ref: 0.450-4.500 IU/mL
 - 08/02/17: 5.580 IU/mL
 - 10/20/17: 3.990 IU/mL
 - 6/25/18: 2.85 IU/mL !!!!

HANDLING STRESS AND FEELING BURNED OUT

- It's NOT sustainable!
- You NEED sleep and REAL foods
- Sure, you can eat processed foods, not exercise, pull all-nighters but I can promise you you're not going to feel great or healthy about yourself in the process

EXERCISE IS KEY

- Whatever it is you like – do that!
- Walk in the park
- Kicking around a soccer ball in an empty field
- Yoga
- Running
- Weight lifting
- Get active! You don't need a gym membership to be active

VOLUNTEERING

- It's free – and I promise you, you'll feel good afterwards
- Local shelter and feed meals to those in need
- Animal shelter
- Local rescues and adoption events
- Habitat for humanity
- Teaching opportunities

GET OUT MORE

- Grab a friend, go see a movie
- Go out to eat
- Explore the city you are in
- Grad school isn't mean to be ALL work NO play
- Find a new activity you've never done before
- YOU can have a healthy balance!
- Connecting with people will help you in your future careers, you never know who you will run into.

UNWIND & UNPLUG

- We spend too much time on our computers
- Read a book – PUBLIC LIBRARIES ARE GREAT!
- Take a bath
- Listen to a podcast
- Take 5 minutes, anywhere to meditate
- Whatever that interest may be, find it, do it and remove yourself from the rest of the world for a little while

NUTRITION

- The biggest lie is that healthy foods cost too much
- You don't have to stick to a strict diet but here are some ideas:
 - Meal prep
 - Crock pot
 - Cook bigger batches
 - Make a big bowl of salad
 - Cut a melon
- Fruits, grains, nuts, seeds, fish, meat, vegetables, sprouted grains. Whole, real foods.

NUTRITION

- Nutrition most importantly has a huge impact on our hormones
- Unbalanced hormones can cause you to feel tired, upset, and feel a whirlwind of different emotions
- Fatty acids & Omega-3's
- Stay hydrated
- Avoid added sugars

VITAMINS AND SUPPLEMENTS

- Vitamins and supplements should be kept to a minimum
- Ideal to get those nutrients from whole foods rather than pill form
- Targeted supplements are good for areas of need (ex. Thyroid, adrenal, etc.)
- If you have a known deficiency (iron)

MENTAL HEALTH

- You are not alone
- Your mental health may also be linked to your physical health
- Your school has resources
- You also probably have an ombudsman
 - Get unbiased guidance and opinion
- Make a friend
- National Suicide prevention lifeline: 1-800-273-8255

YOU ARE IMPORTANT!

YOU HAVE A PURPOSE!

- YOU have gotten to where you are because you are GIFTED
- YOU have the power to change the future through your research
- YOU can be a light for someone else in a struggle – someone may be looking up to YOU right now!
- No one else can take better care of you, than you

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