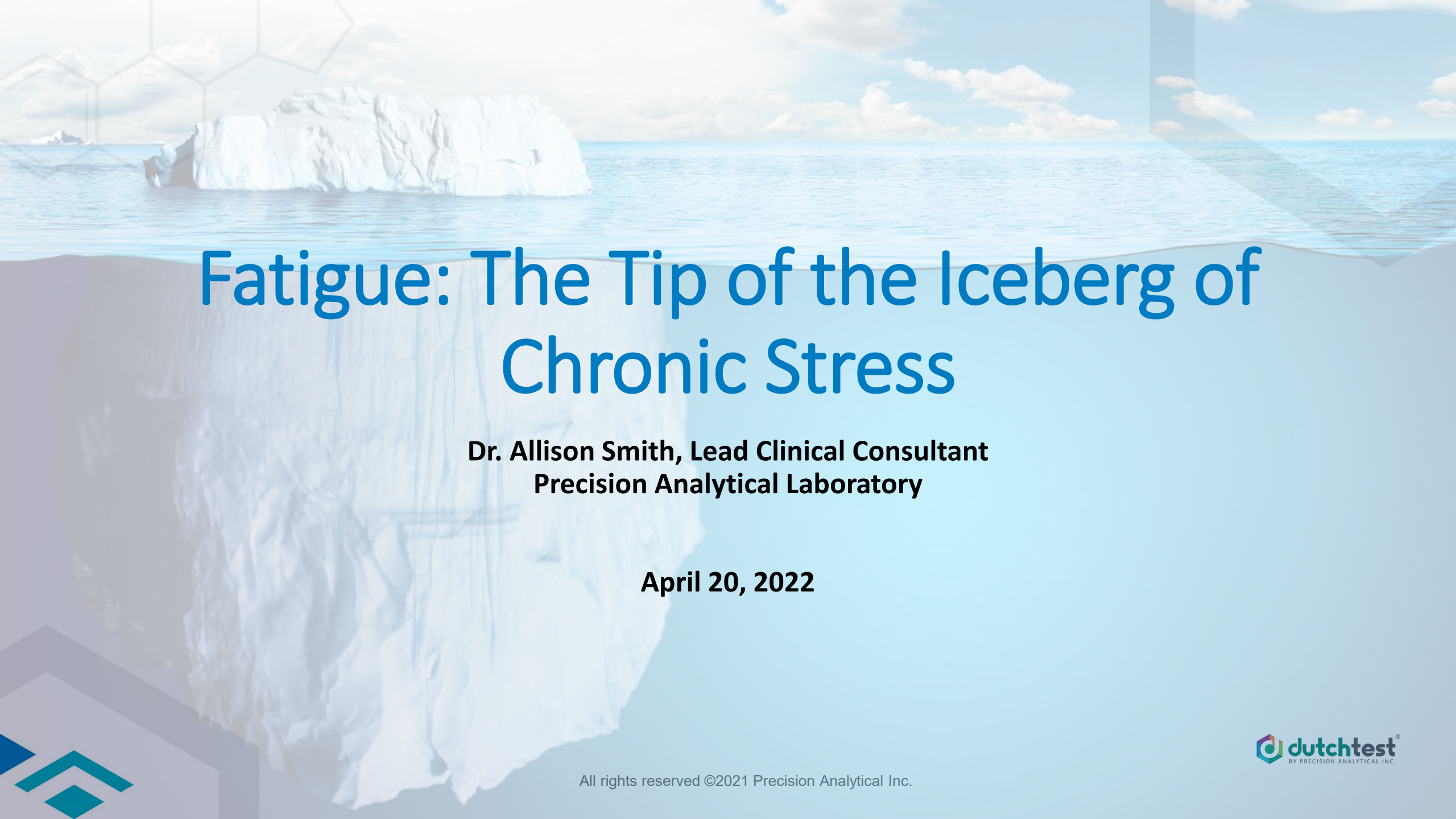


The background of the slide features a large iceberg floating in a blue ocean under a bright sky with scattered clouds. The iceberg's tip is visible above the water, while its massive, jagged base is submerged, illustrating the concept of 'the tip of the iceberg'. In the top left corner, there is a faint, light-colored hexagonal molecular structure overlay. In the bottom left corner, there is a stylized blue geometric logo consisting of several overlapping chevron-like shapes.

Fatigue: The Tip of the Iceberg of Chronic Stress

**Dr. Allison Smith, Lead Clinical Consultant
Precision Analytical Laboratory**

April 20, 2022



Meet Dr. Allison Smith



Today's Plan

Review stress physiology

Connecting chronic stress to fatigue through CORTISOL

How stress translates into cortisol labs



- Free cortisol patterns
- Metabolized cortisol

Cases and LOTS of Great Examples



Chronic Stress and its Presentation: FATIGUE

Consistently in the Top 10 primary care complaints!

STRESS IS A CHALLENGE to the body

Systems in place to deal with stressors

Mitochondrial adjustments

HPA Axis – CORTISOL

Autonomic responses – Prolactin, NE, EPI, DA, AVP, etc

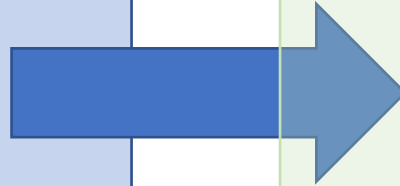
Chronic challenges can lead to physical dysfunction



Stressors: The OUTS to the INS

- Exogenous

- Work
- School
- Traffic
- Relationships
- Perceived Danger
- Crowding
- Dietary
- Water intake
- Toxic exposures
- Medications

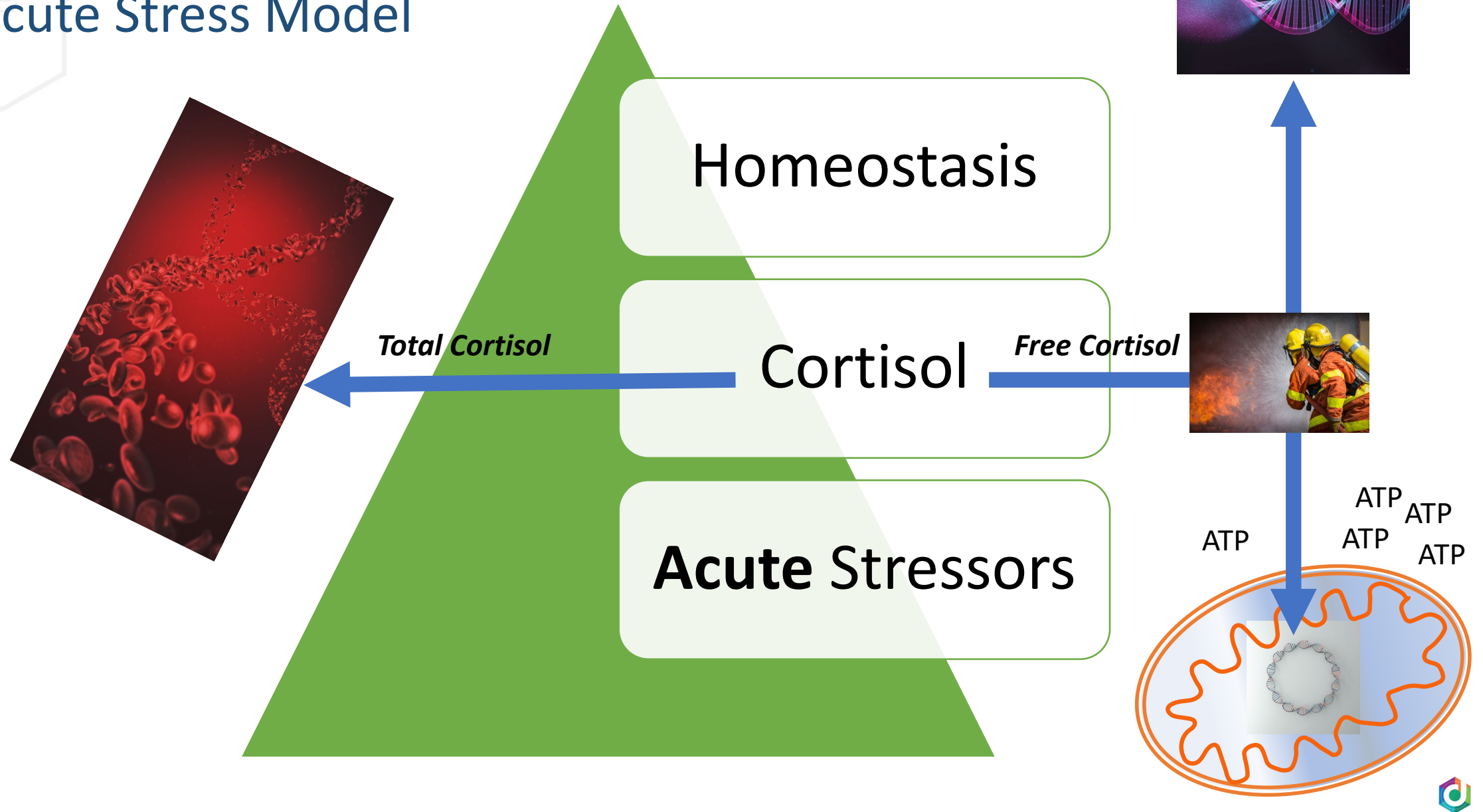


- Endogenous

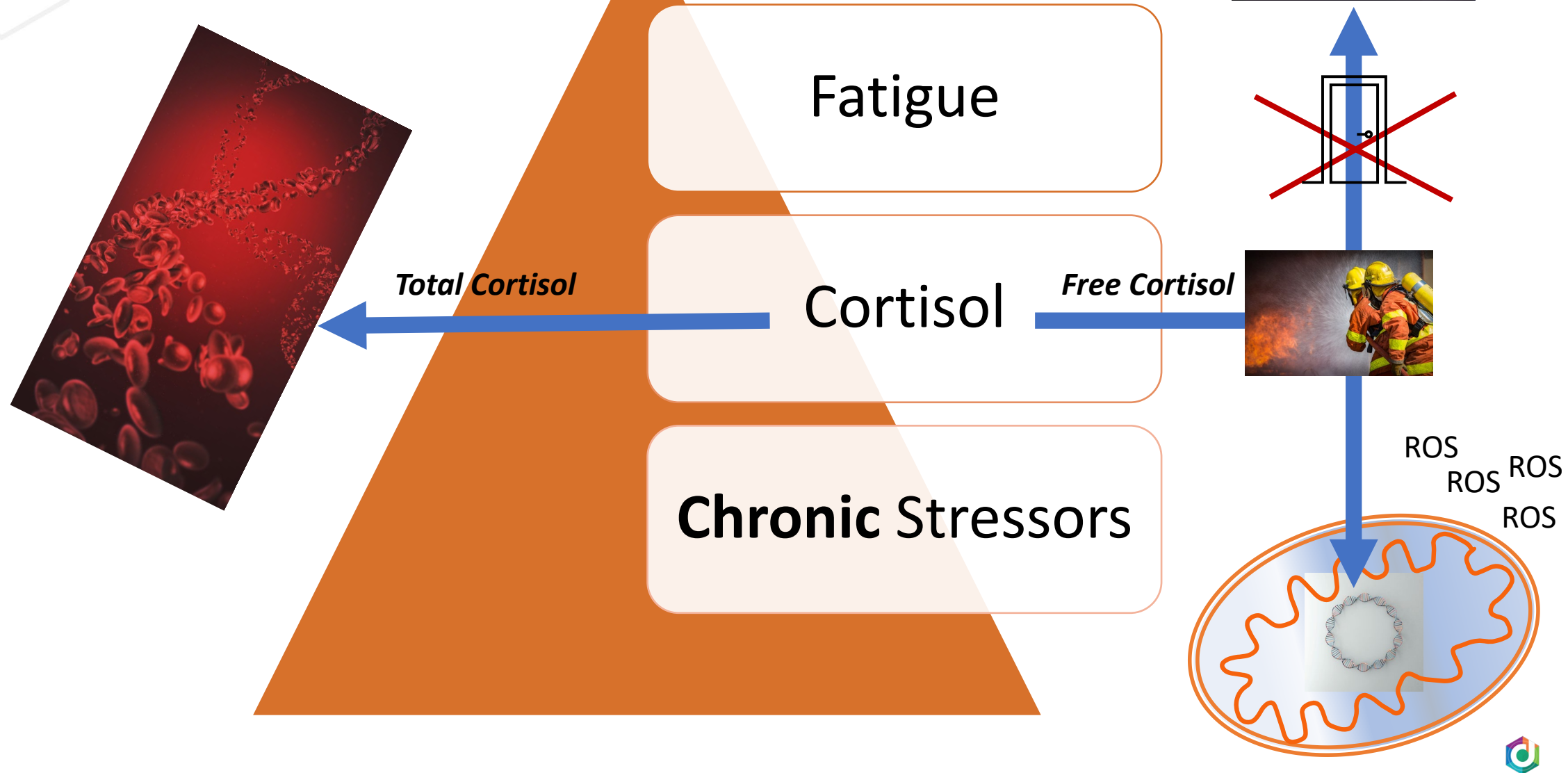
- Blood/Tissue glucose
- Pathogens/illness
- Inflammation
- Oxidative stress
- Endocrine disruption
- Nutrient Deficiencies
- Dehydration/Electrolytes
- Autoimmunity



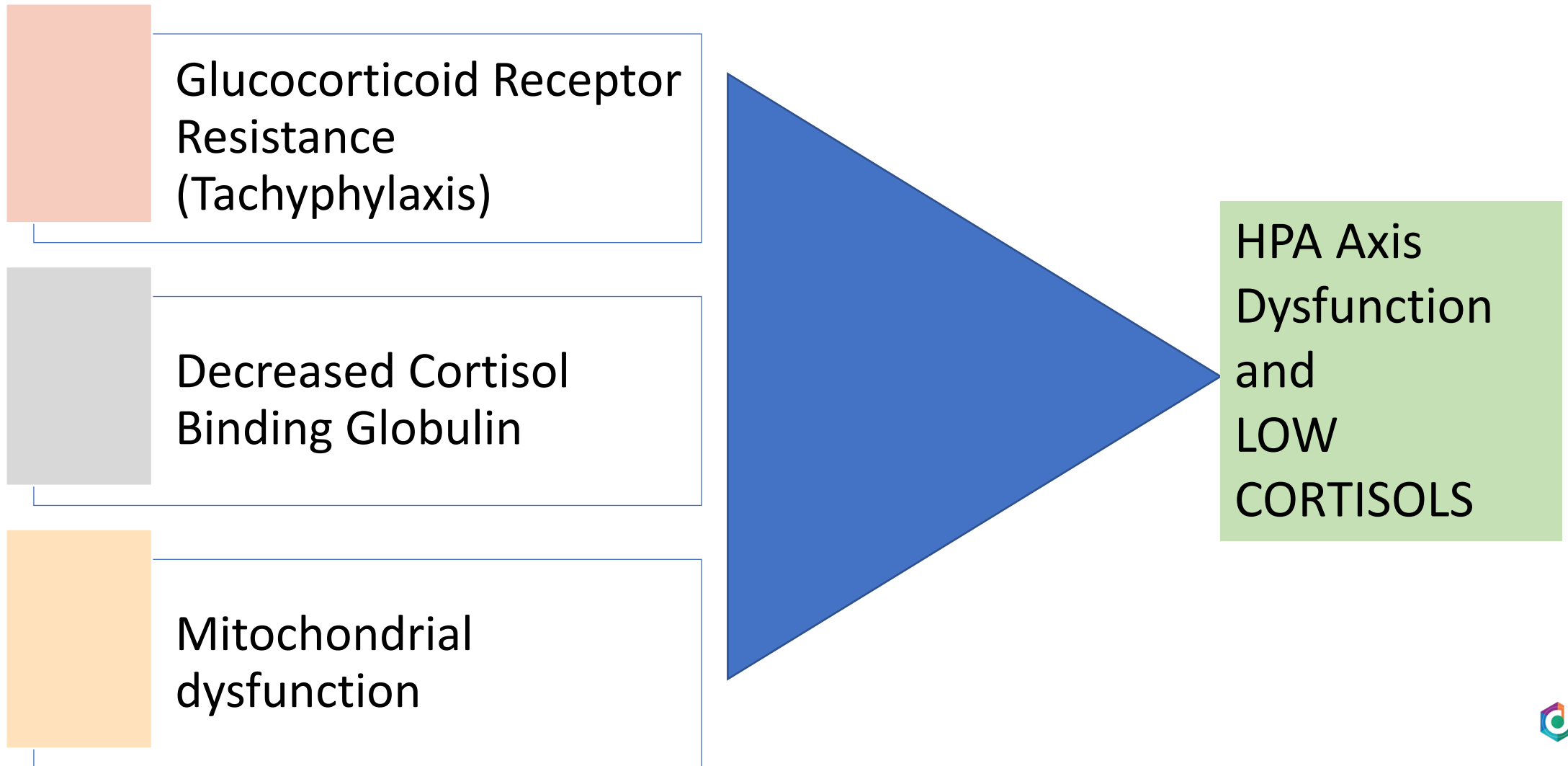
Acute Stress Model



Chronic Stress Model



Sustained Stressors - Chronically High Cortisols



When Cortisol Affects the Mitochondria

- Short Term

- ↑ Mitochondrial biogenesis
- ↑ Ox Phos enzyme activities → ATP production

MORE MITOCHONDRIA
MORE ENERGY

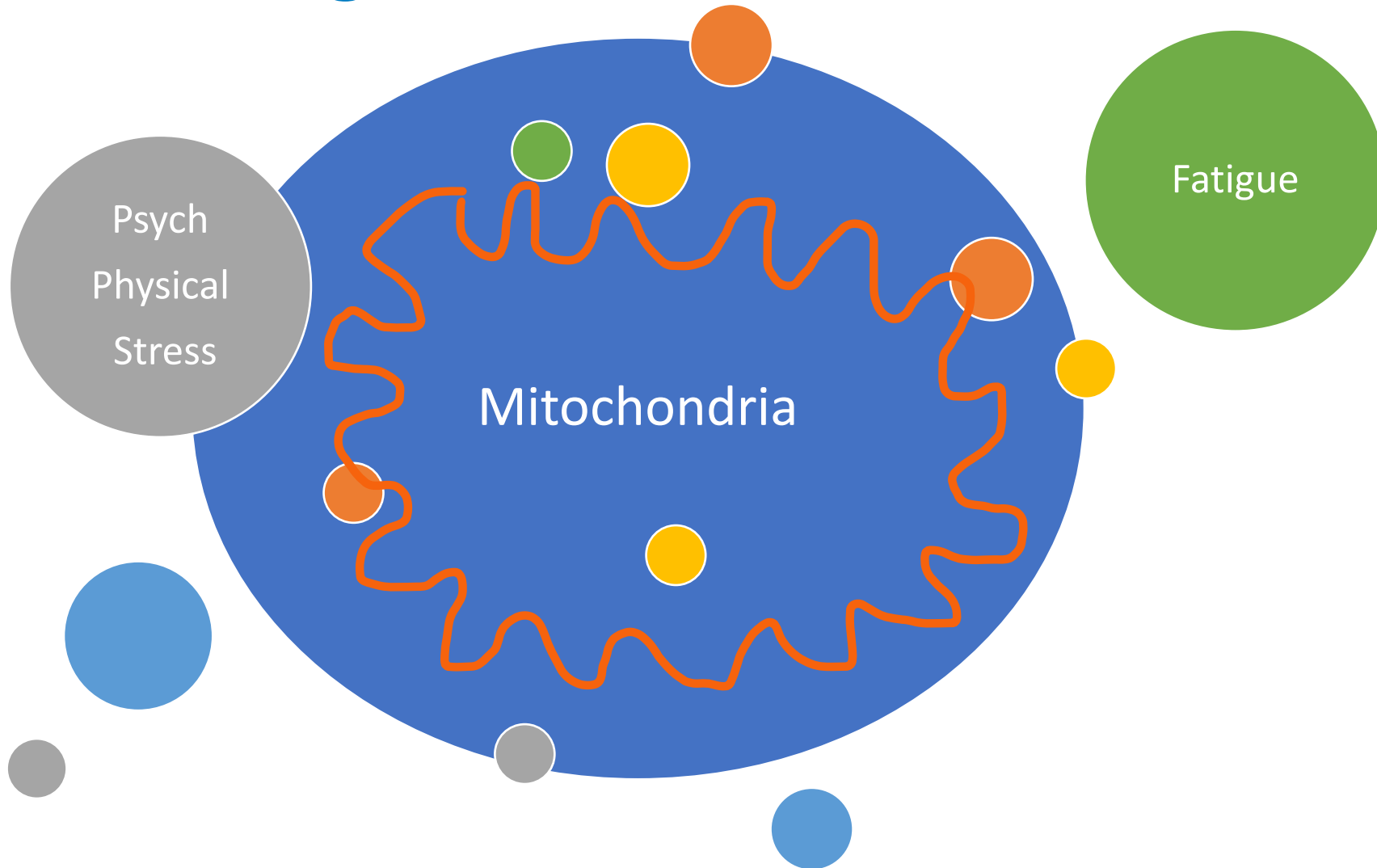
- Long Term

- Ox Phos dysfunction
- ↓ ATP production
- ↑ ROS
- Structural deformity, loss of membrane potential
- Shortened telomeres, advanced cellular aging
- ↓ Steroid hormone production
- ↓ Bile Acid synthesis

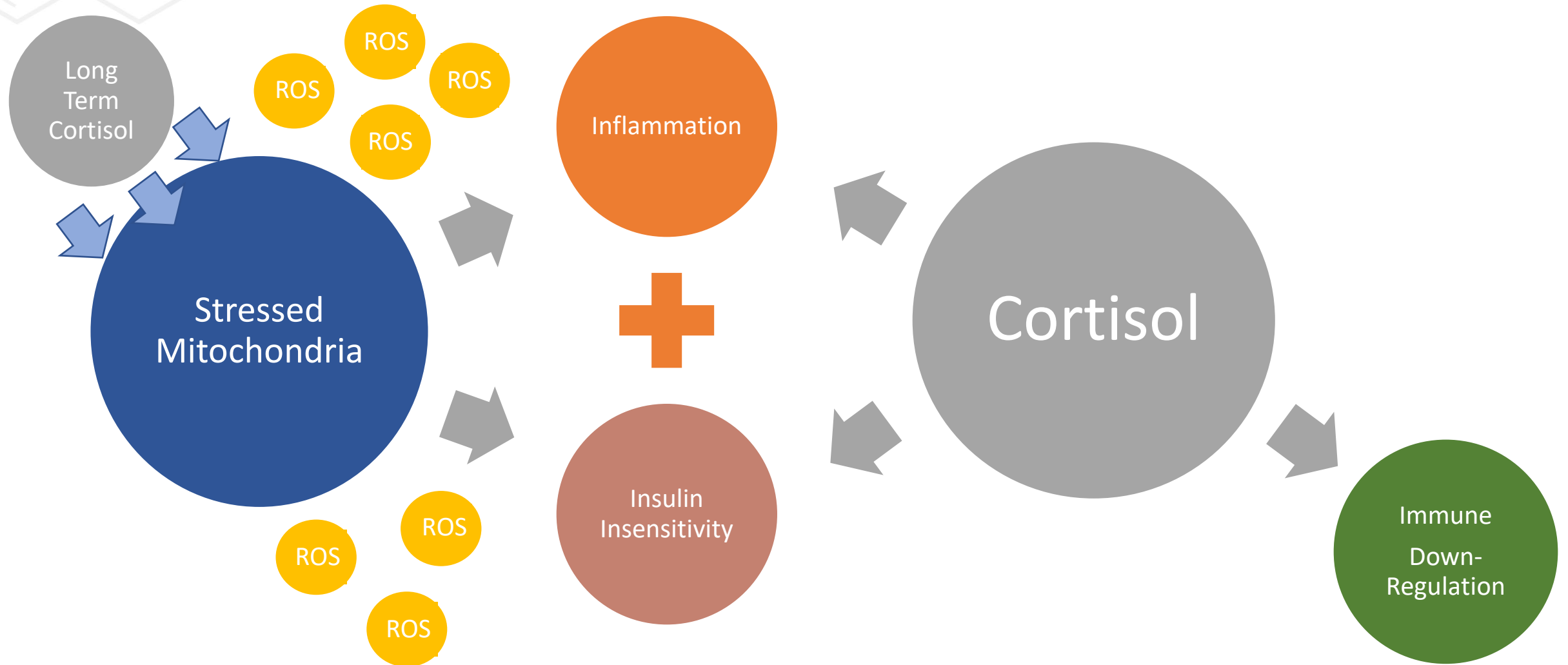




Stress Changes the Mitochondria



Chronic Stress Link to Insulin, Inflammation, Infections



Cortisol Tissue Actions for Insulin Resistance

SHORT TERM

- Fasted states:
- Increase **gluconeogenesis** and **glycogenolysis** → ↑ tissue and blood glucose levels
- Induces local insulin resistance → stabilize glucose availability in the blood stream

PRE-DIABETES

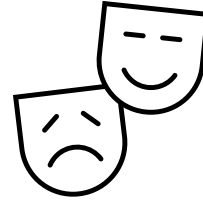
- Fasted and Fed States:
- Insulin resistance → cortisol rescues tissues by increasing gluconeogenesis
- Glycogen stores continue to be tapped by cortisol too → ↓ ability to normalize tissue glucose
- Need for cortisol ↑↑↑
- CBG gets downregulated by insulin to meet the need

TYPE 2 DM

- ↑↑↑ Gluconeogenesis but Glycogen stores tapped **out**
- ↓ success normalizing glucose availability using cortisol

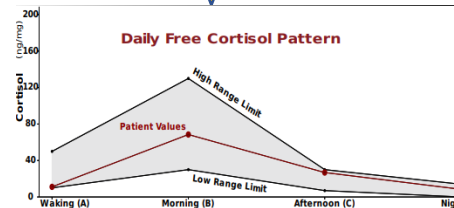
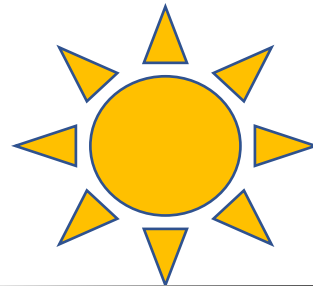


Cortisol's Many Roles



- Upregulation

- Glucose generation (GNG, glycogenolysis)
- Lipolysis (subQ brown fat only)
 - Dependent on insulin/epi
- White fat redistribution
- Protein catabolism
- Glucagon
- Norepi → Epinephrine

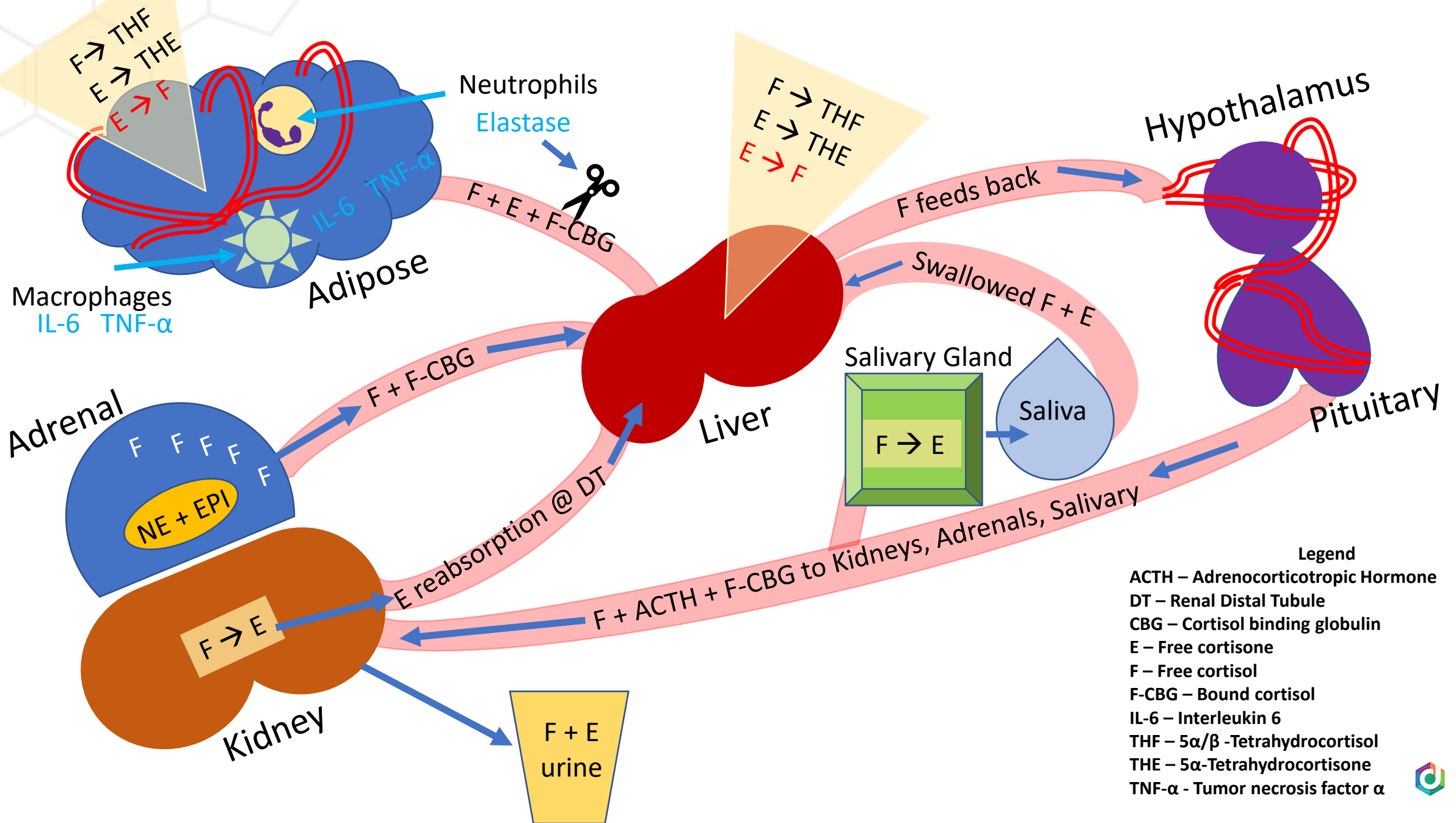


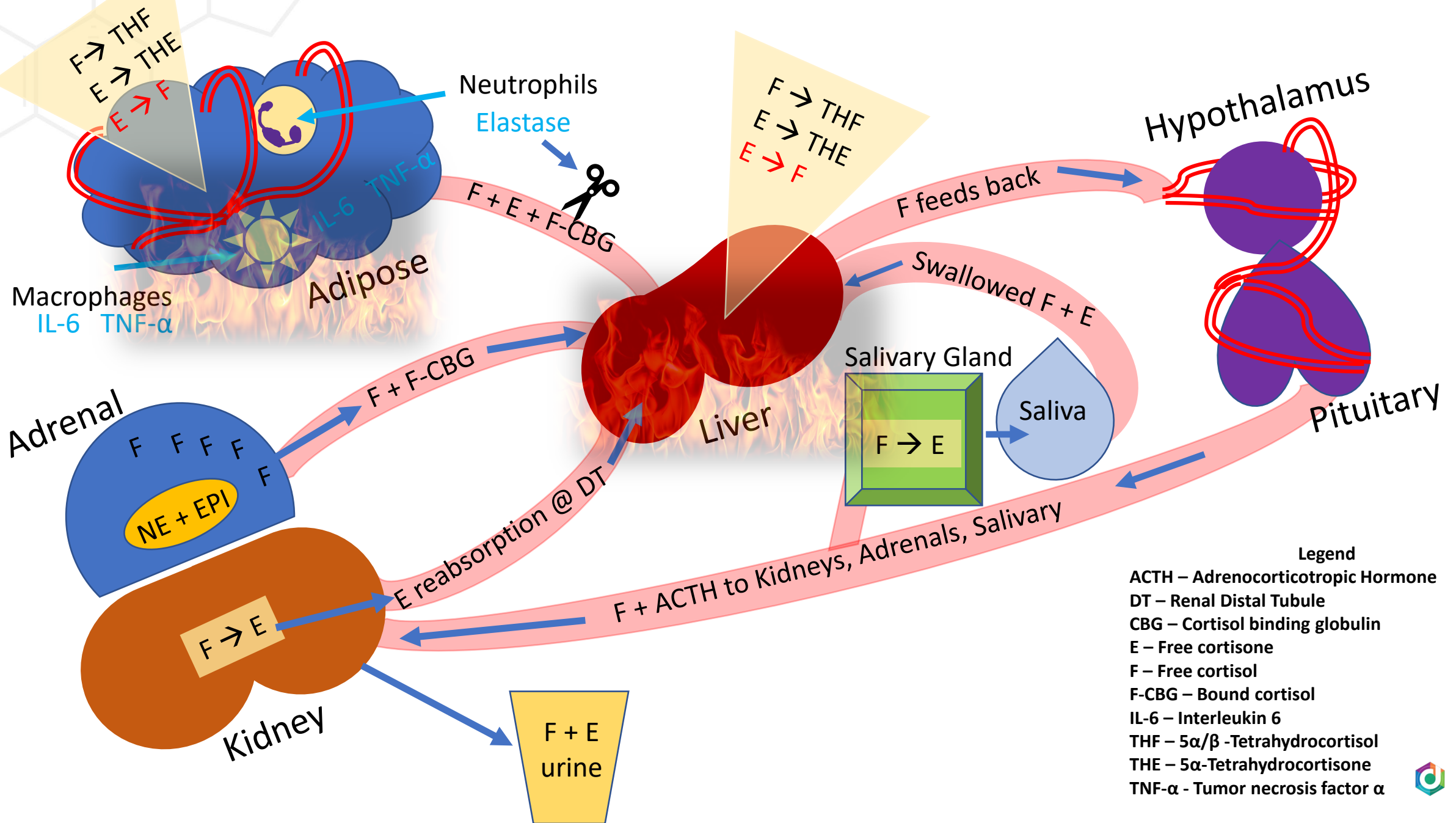
- Downregulation

- Proinflamm cytokines/chemokines (IL-6, TNF α , IL-1 β , CCL2, 3)
- Immune activity
 - Suppress B cells, T cells
 - ↓ Movement of Neutrophils
- Insulin
- fT4 → fT3, TSH
- Nrf2-mediated gene exp
- H₂O₂ detox

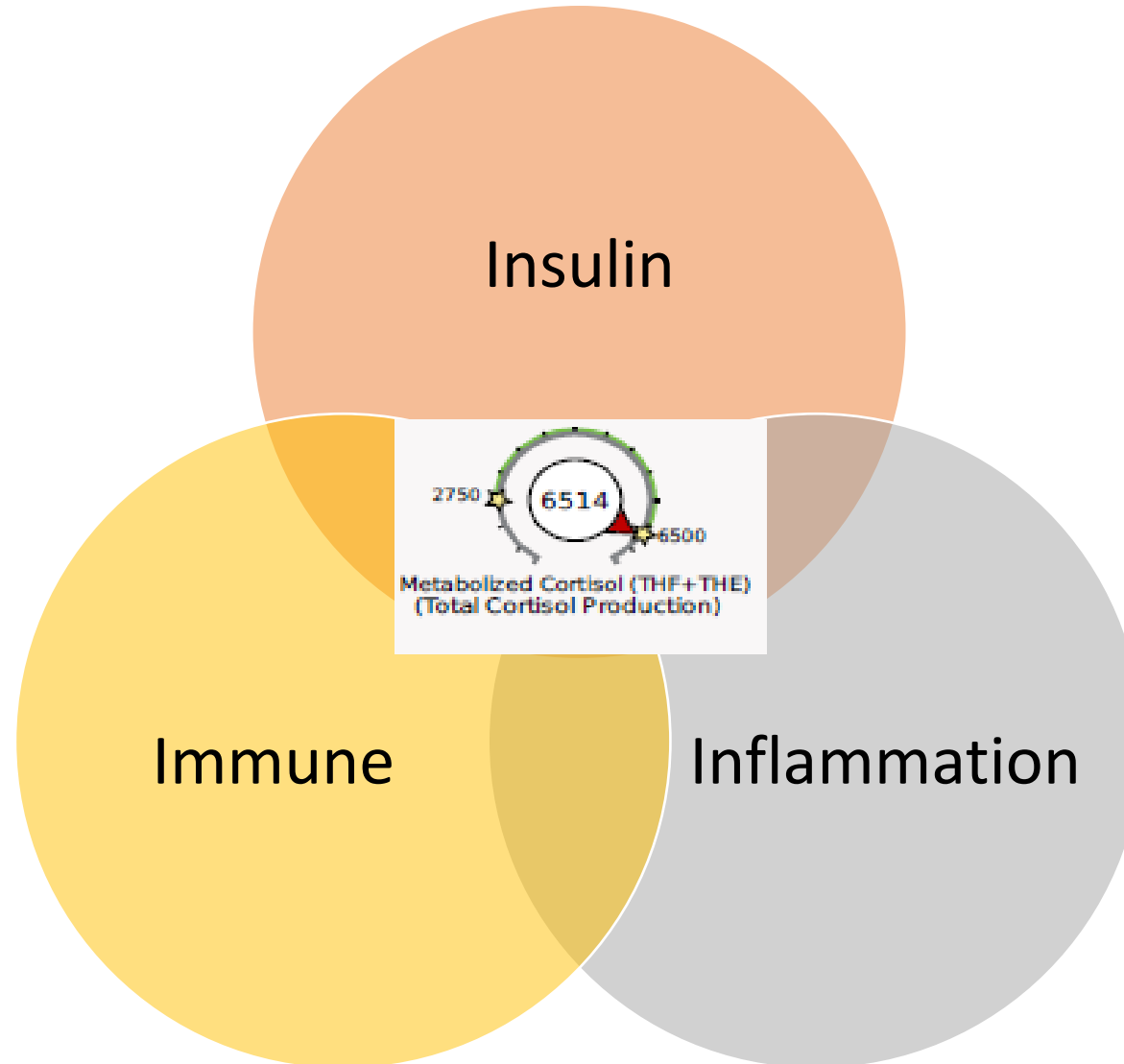
NORMAL means – with intact diurnal variation (high in the morning, low at night)







3 Main Sources of Unresolved Stress





Stress, Fatigue, and Mitochondria Studies

Filler K, Lyon D, Bennett J, et al. Association of Mitochondrial Dysfunction and Fatigue: A Review of the Literature. *BBA Clin*. 2014;1:12-23. doi:10.1016/j.bbacli.2014.04.001

Picard M, McEwen BS. Psychological Stress and Mitochondria: A Conceptual Framework. *Psychosom Med*. 2018.

Picard M, McEwen BS, Epel ES, Sandi C. An energetic view of stress: Focus on mitochondria. *Front Neuroendocrinol*. 2018.

Wood E, Hall KH, Tate W. Role of mitochondria, oxidative stress and the response to antioxidants in myalgic encephalomyelitis/chronic fatigue syndrome: A possible approach to SARS-CoV-2 'long-haulers'? *Chronic Dis Transl Med*; 2021.

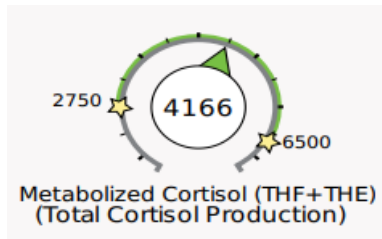
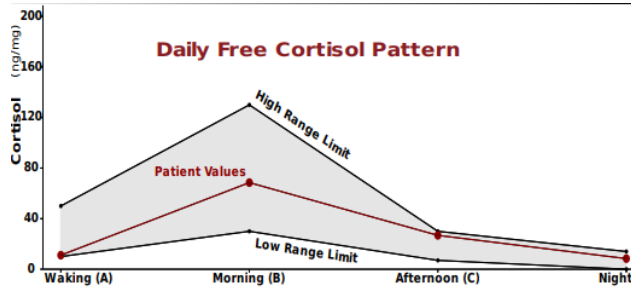




Cortisol Testing

Assessing the Fatigued Patient

Functional Cortisol Testing

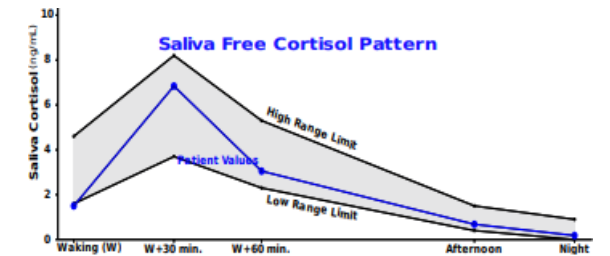
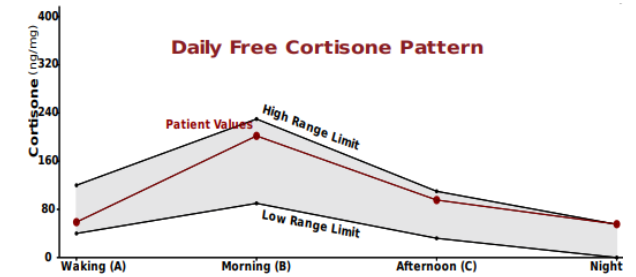


Diurnal Free
Cortisol (F)

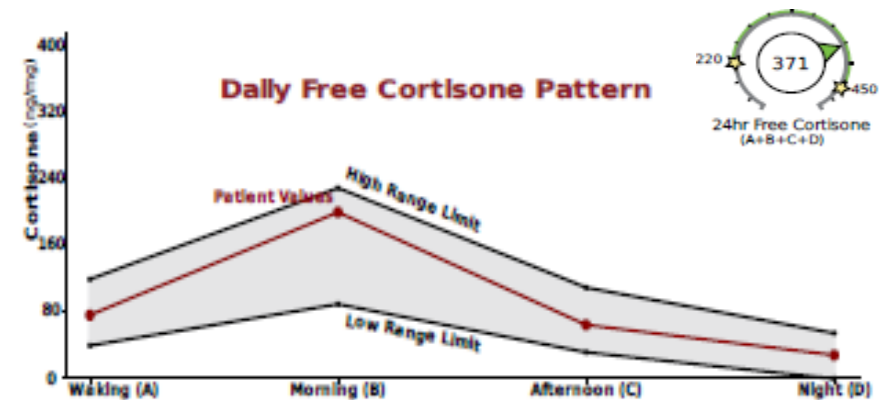
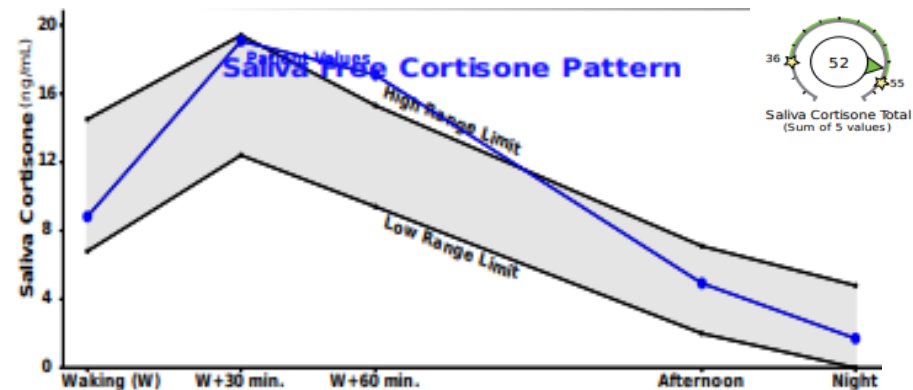
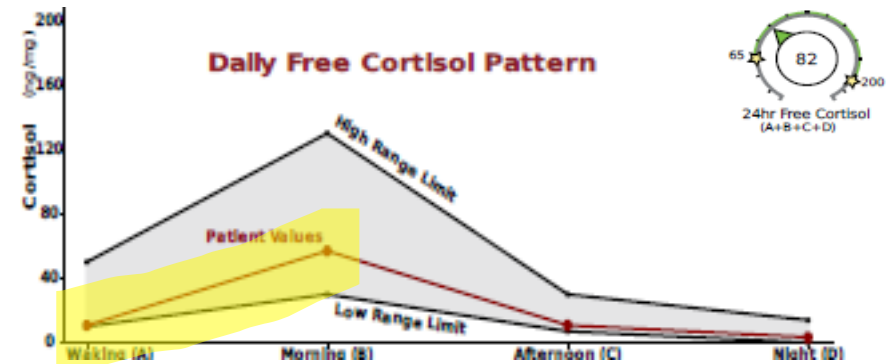
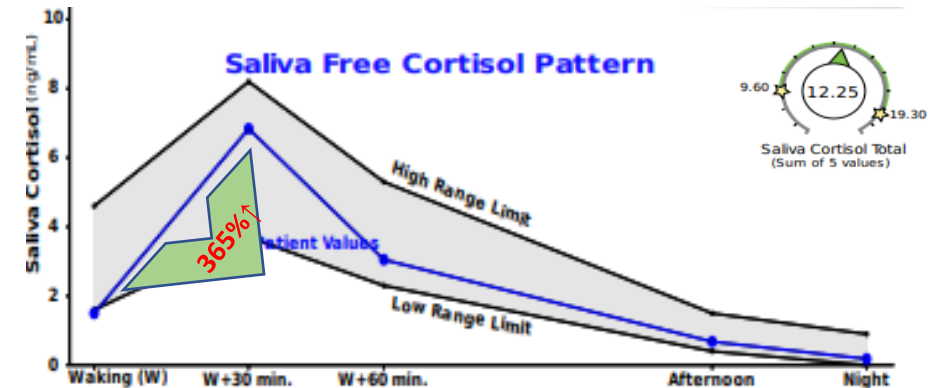
Diurnal Free
Cortisone (E)

Cortisol and
Cortisone
Tissue
Metabolites
(THF and THE)

Cortisol
Awakening
Response
(CAR)



Free Cortisol – Saliva and Urine



If cortisone is HIGHER than cortisol at each time point, then free CORTISONE is your surrogate marker for free CORTISOL.



Pattern >>> Levels in Fatigue

Flattened Free C Curves

The Flat-Line Curve

Diurnal Cortisol Slopes and Mental and Physical Health Outcomes: A Systematic Review and Meta-analysis

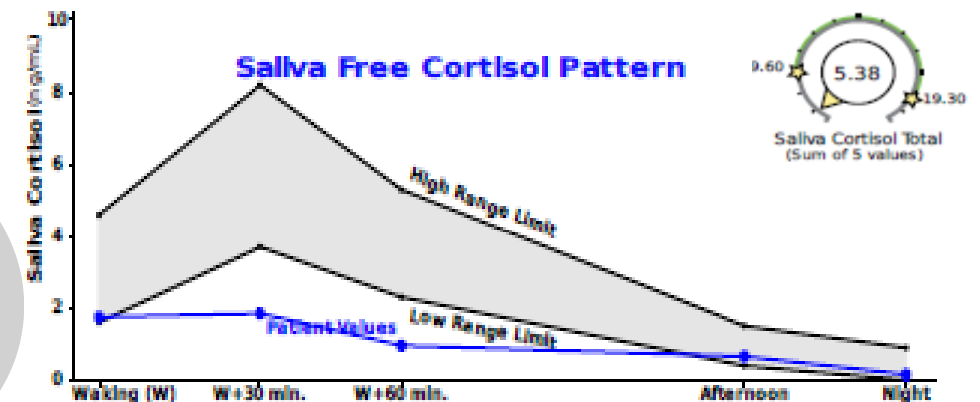
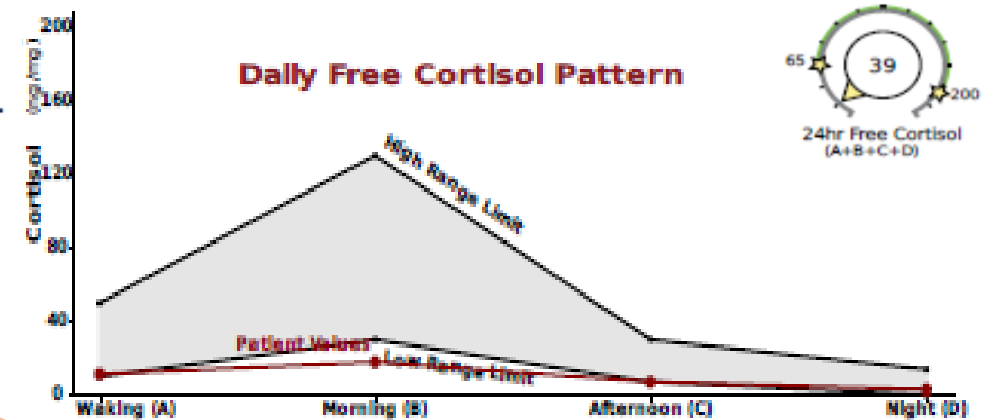
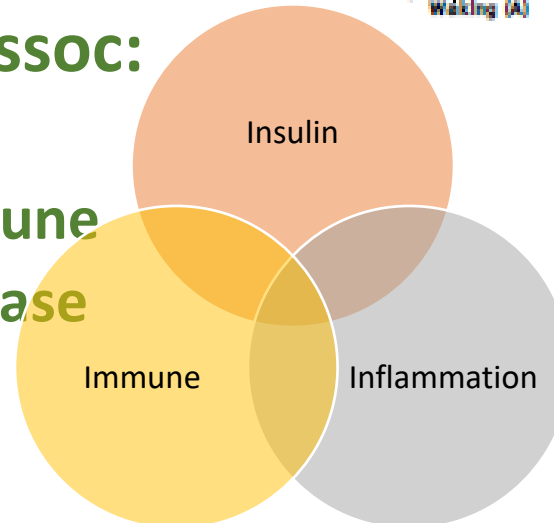
Emma K. Adam^{a,*}, Meghan E. Quinn^b, Royette Tavernier^c, Mollie T. McQuillan^d, Katie A. Dahlke^e, and Kirsten E. Gilbert^f

Meta-analysis from 2017

80 studies; 36,823 participants

Flattened diurnal slopes assoc:

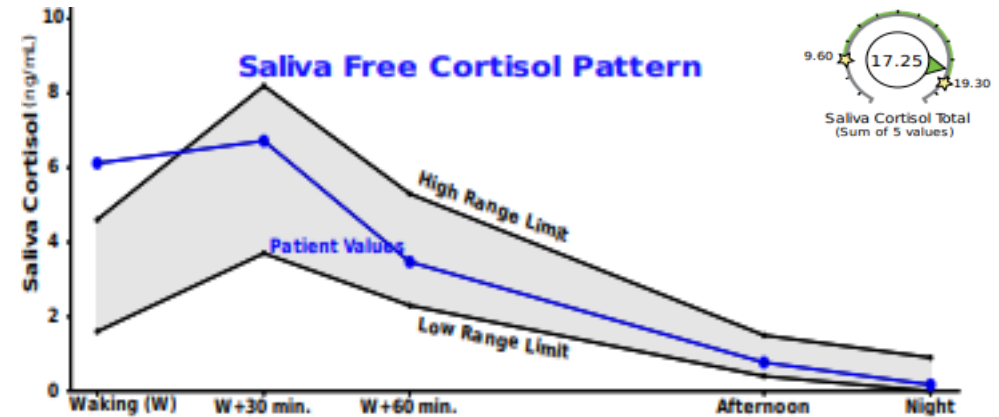
- #1 – Obesity/Adiposity
- #2 – Inflammatory/Immune
- #3 – Cardiovascular disease



The Dysglycemic Pattern

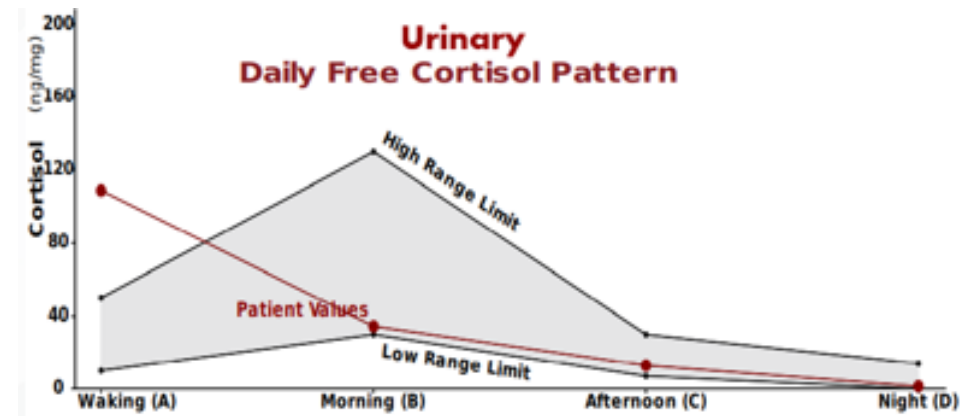
Saliva

Waking cortisol is high
CAR is low



Urine

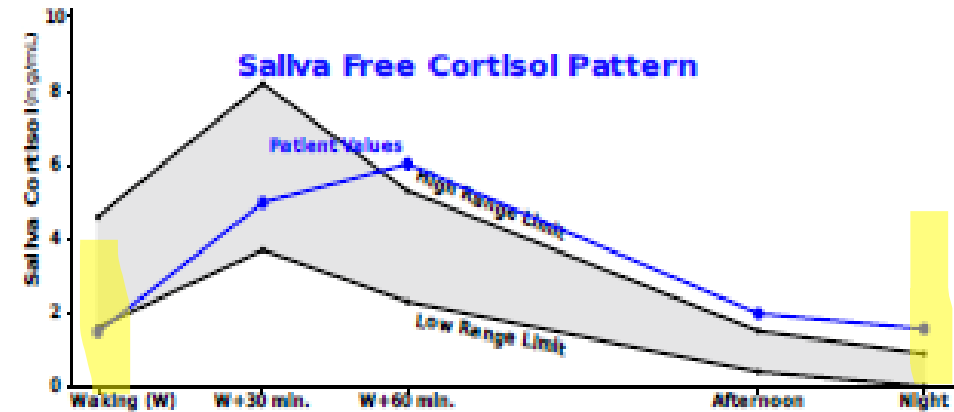
A sample cortisol
high-normal or high
(suggesting late night/early
morning C production due to
hypoglycemia)



The Flipped Curve

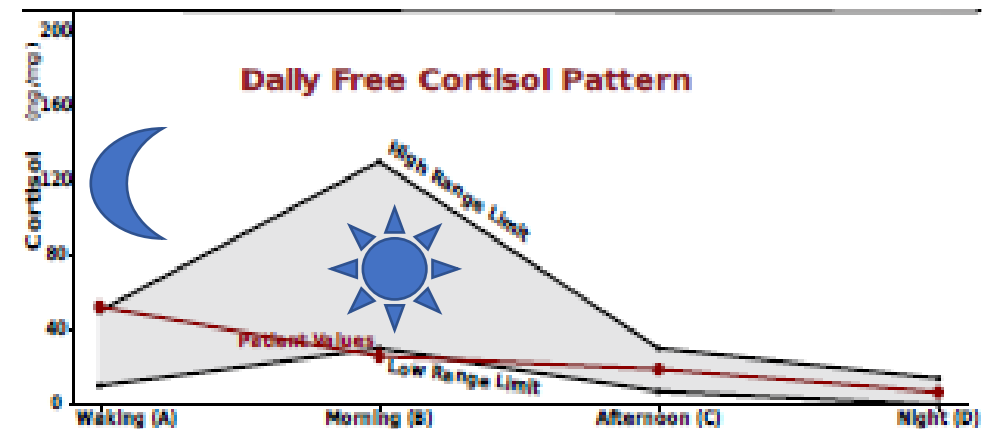
Saliva

Low morning, high night



Urine

Waking A is HIGH compared to Morning B
Morning B low point of the day



Poor Sleep Quality Pattern

Saliva

AM Cortisol is low

Afternoon C ↑

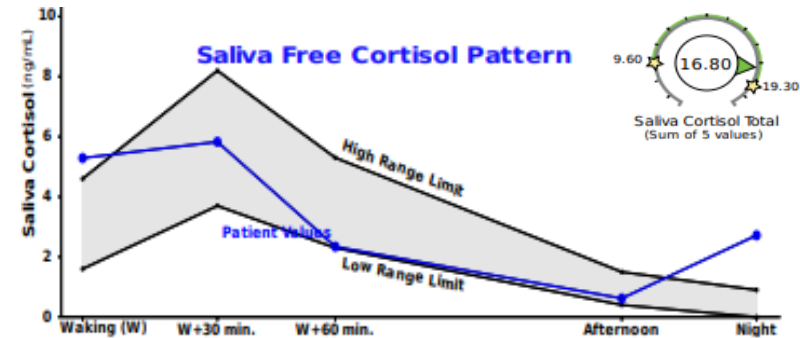
Urine

Waking cortisol high

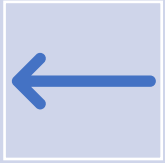
Lack of rise A → B

Afternoon C ↑

*May see bedtime rise if sleep onset part of the issue



Why Cortisol Metabolites?



Evidence of cortisol entering liver/adipose tissue



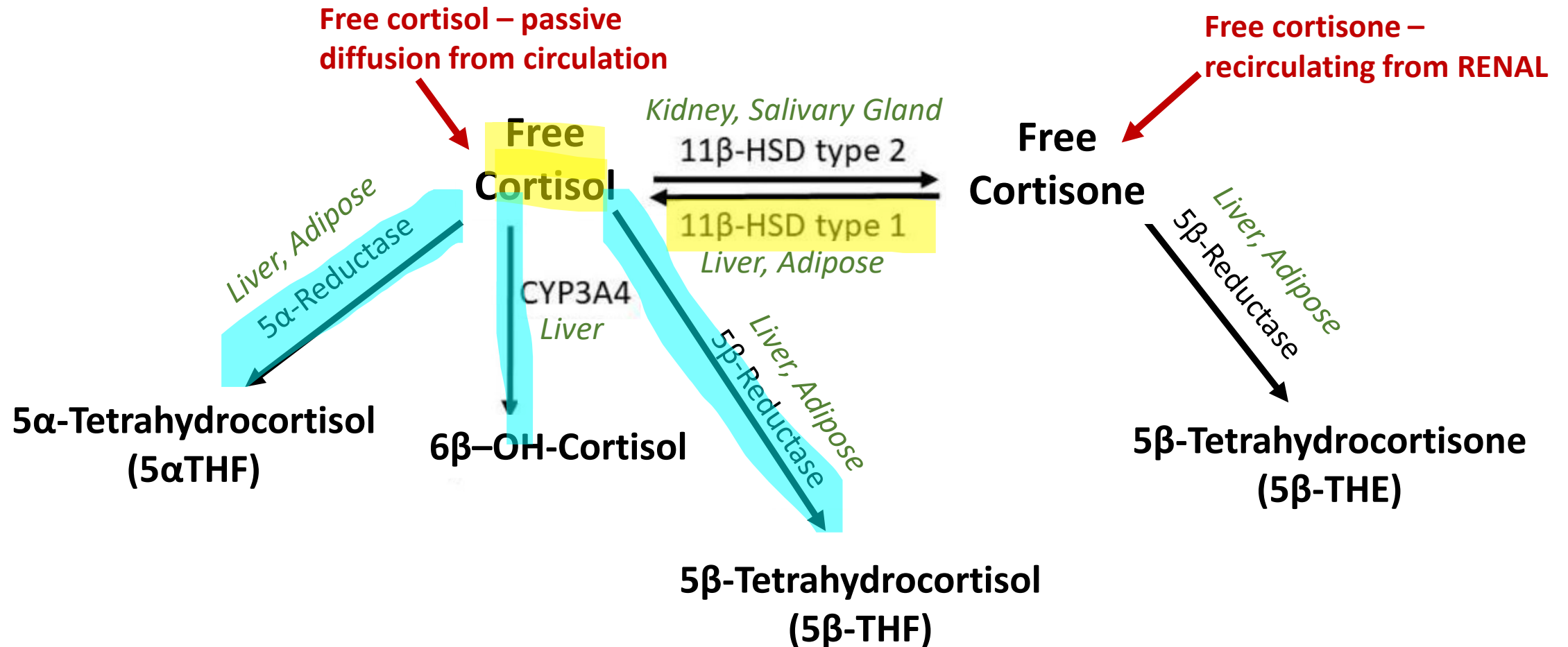
Free cortisol = potential energy (and influences ACTH prodn)



Cortisol metabolites = marker of cortisol production + evidence of presence within liver and adipose tissues

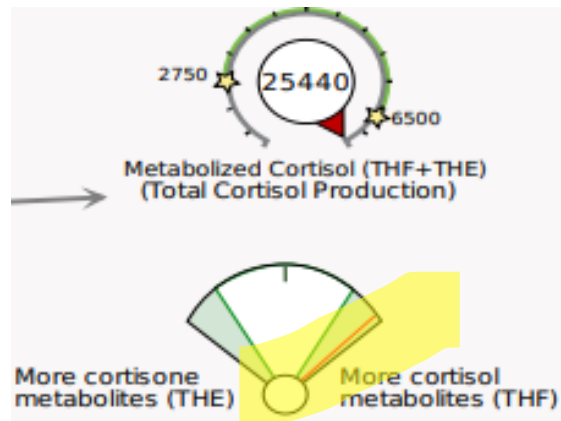


Cortisol Clearance and Metabolism

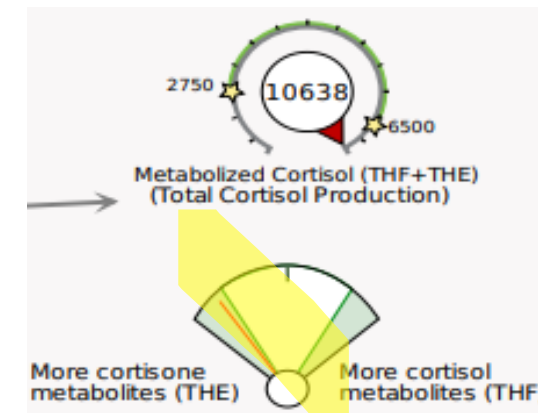


Cortisol Metabolism SHIFTS

Acute Stress



Chronic Stress*

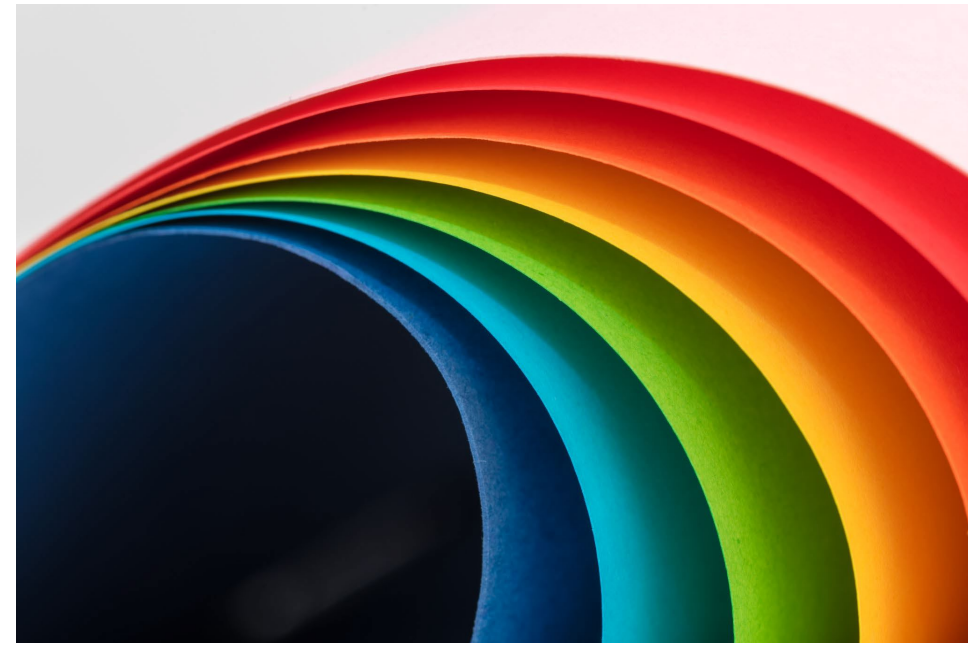


*Or hypermetabolism in HYPERTHYROIDISM



CYP3A4 Activity

Did you know.....



Cortisol is also metabolized and cleared down the CYP3A4 pathway?

CYP3A4 activity => Additional measure of cortisol clearance



Grapefruit juice and licorice increase cortisol availability in patients with Addison's disease

Paal Methlie^{1,2}, Eystein E S Husebye^{1,3}, Steinar Hustad¹, Ernst A Lien^{1,2} and Kristian Løvås^{1,3}

¹Institute of Medicine, University of Bergen, 5021 Bergen, Norway and ²The Hormone Laboratory and ³Department of Medicine, Haukeland University Hospital, 5021 Bergen, Norway

(Correspondence should be addressed to P Methlie at Institute of Medicine, University of Bergen; Email: paal.methlie@med.uib.no)

Abstract

Objective: Failure to mirror the diurnal cortisol profile could contribute to the impaired subjective health status in Addison's disease (AD). Some patients report benefit from the use of various nutritional compounds. The objective of this study was to investigate the impact of licorice and grapefruit juice (GFJ) on the absorption and metabolism of cortisone acetate (CA).

Design: Patients ($n=17$) with AD on stable CA replacement therapy were recruited from the outpatient clinic at Haukeland University Hospital, Norway. They were assessed on their ordinary CA medication and following two 3-day periods of co-administration of licorice or GFJ.

Methods: Time series of glucocorticoids (GCs) in serum and saliva were obtained, and GCs in 24 h urine samples were determined. The main outcome measure was the area under the curve (AUC) for serum cortisol in the first 2.6 h after orally administered CA.

Results: Compared with the ordinary treatment, the median AUC for serum cortisol increased with licorice (53 783 vs 50 882, $P<0.05$) and GFJ (60 661 vs 50 882, $P<0.05$). Median cortisol levels in serum were also elevated 2.6 h after tablet ingestion (licorice 223 vs 186 nmol/l, $P<0.05$; GFJ 337 vs 186 nmol/l, $P<0.01$). Licorice increased the median urinary cortisol/cortisone ratio (0.43 vs 0.21, $P<0.00001$), whereas GFJ increased the (allo-tetrahydrocortisol + tetrahydrocortisol)/tetrahydrocortisone ratio (0.55 vs 0.43, $P<0.05$).

Conclusion: Licorice and in particular GFJ increased cortisol available to tissues in the hours following oral CA administration. Both patients and physicians should be aware of these interactions.

European Journal of Endocrinology **165** 761–769

Licorice and Grapefruit Juice (GFJ) = **pro cortisol**

Licorice ↑ F:E ratio

GFJ ↑ cortisol : 6β-OH-cortisol (↓CYP3A4) – AND – ↑ 11BHS1 cortisol regeneration in the liver





DIM (Diindolylmethane) for Cortisol Preservation?

Pilot Study: Effect of 3,3'-Diindolylmethane Supplements on Urinary Hormone Metabolites in Postmenopausal Women With a History of Early-Stage Breast Cancer

What they did:

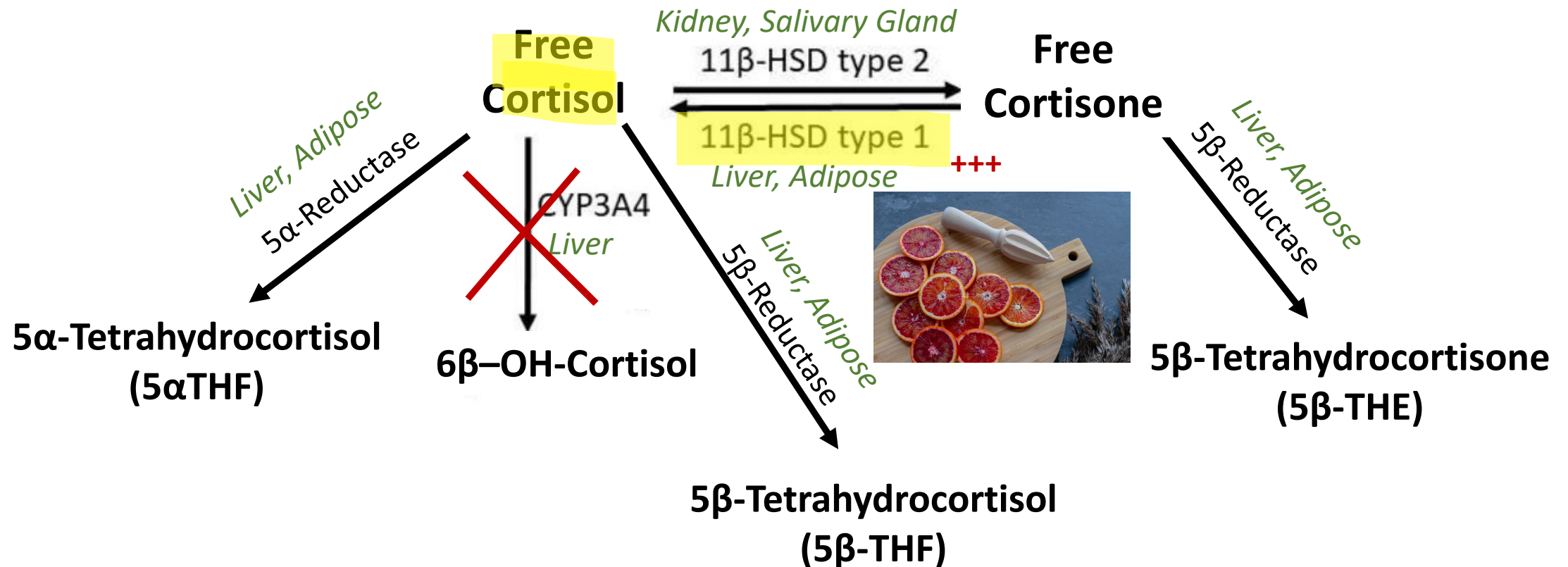
BioResponse DIM 108mg QD for 30 days and then tested urinary estrogens and metabolites (E1, E2, E3, 16OHE1), as well as cortisol and its CYP3A4 metabolite, 6BOHcortisol and DIM itself.

What they found:

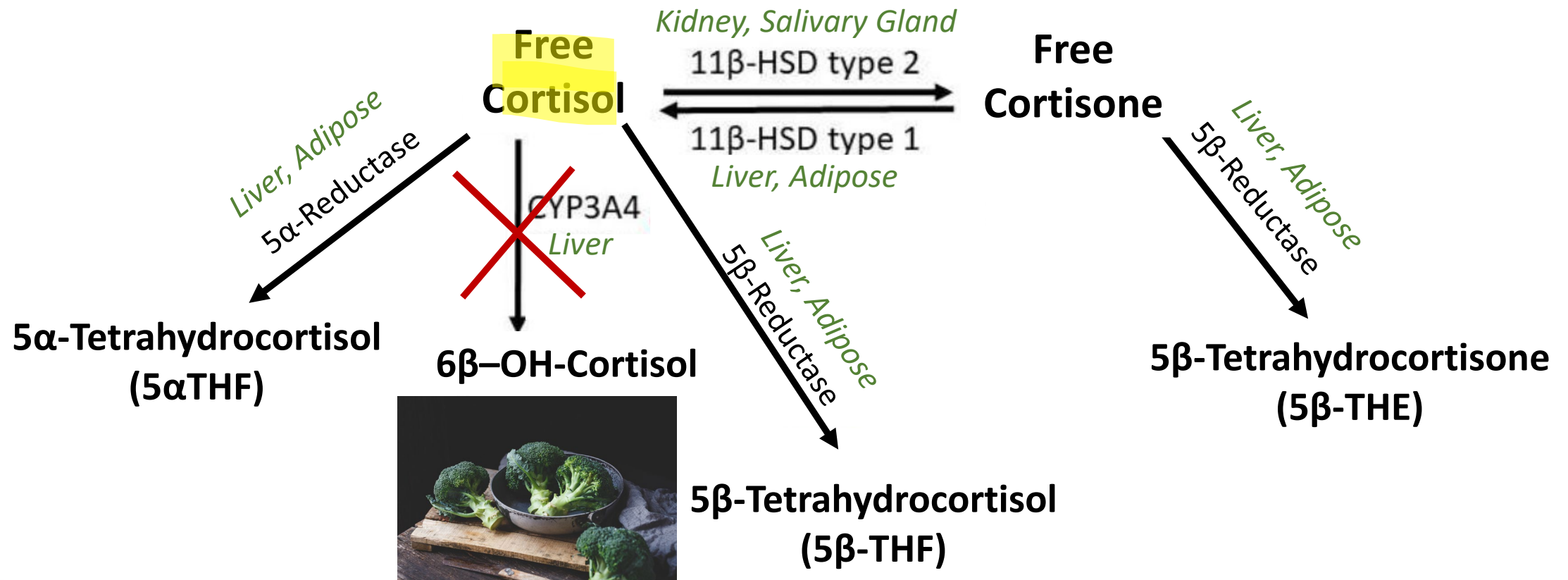
30 days of DIM not only increased 2OHE1 but it also increased the urinary free cortisol : 6 β -OH-cortisol ratio.



GFJ - Cortisol Clearance



DIM – Cortisol Clearance



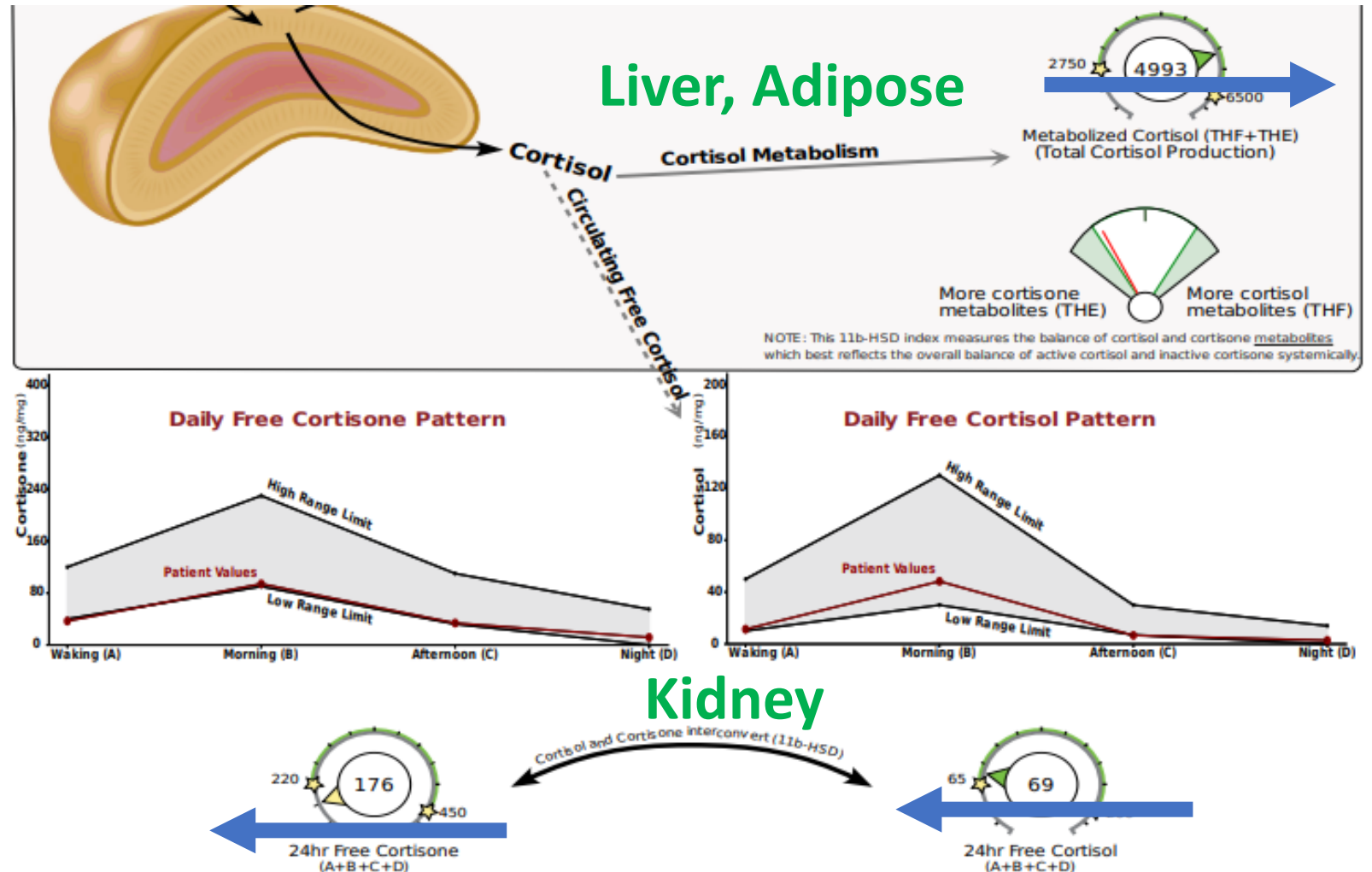
Cortisol Clearance on the DUTCH

36 year old female patient

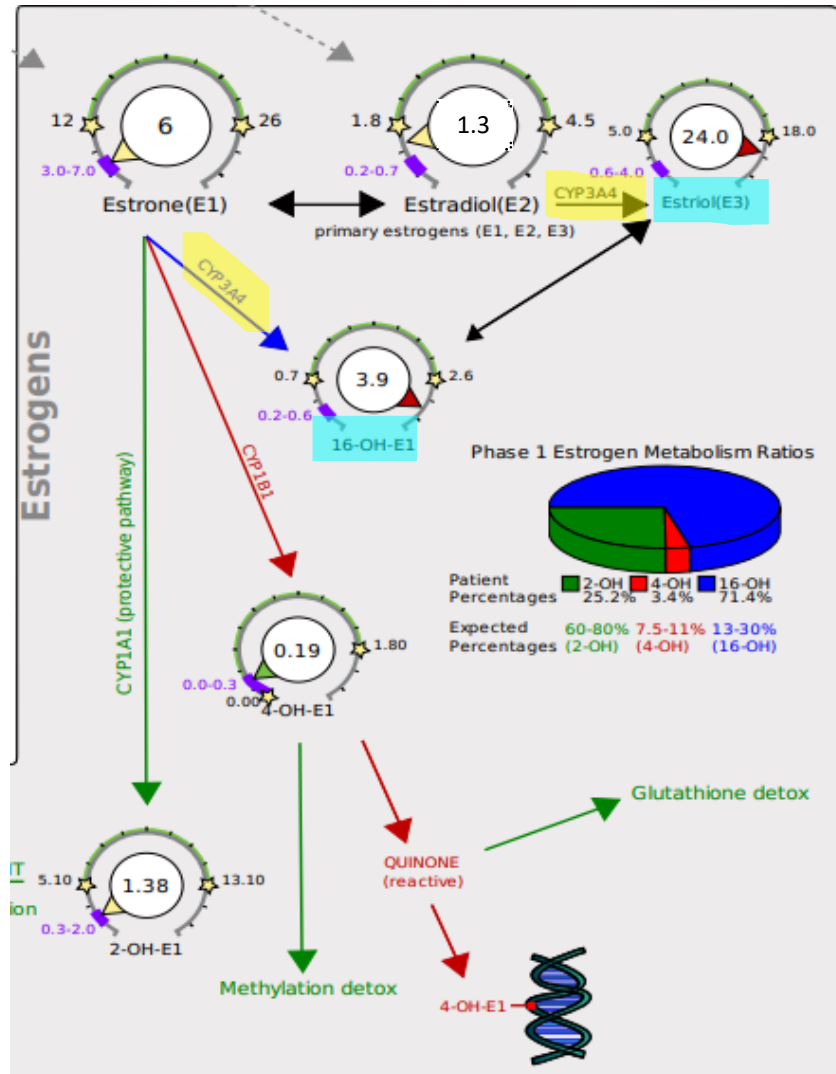
Chief complaint =
Fatigue

- 2° Mood swings
- 3° Migraines

Metab Cortisol >> Free Corts



High CYP3A4 Activity



Estrogen clearance is elevated down CYP3A4

Could she have elevated cortisol clearance down this pathway too?



High CYP3A4 Activity

Decreasing

Decreasing CYP3A4 activity can:

- Increase bioavailable cortisol
- Studies on grapefruit juice in adrenal insufficiency patients on hydrocortisone to ↑ half life



Increasing

Increasing CYP1A1 activity can:

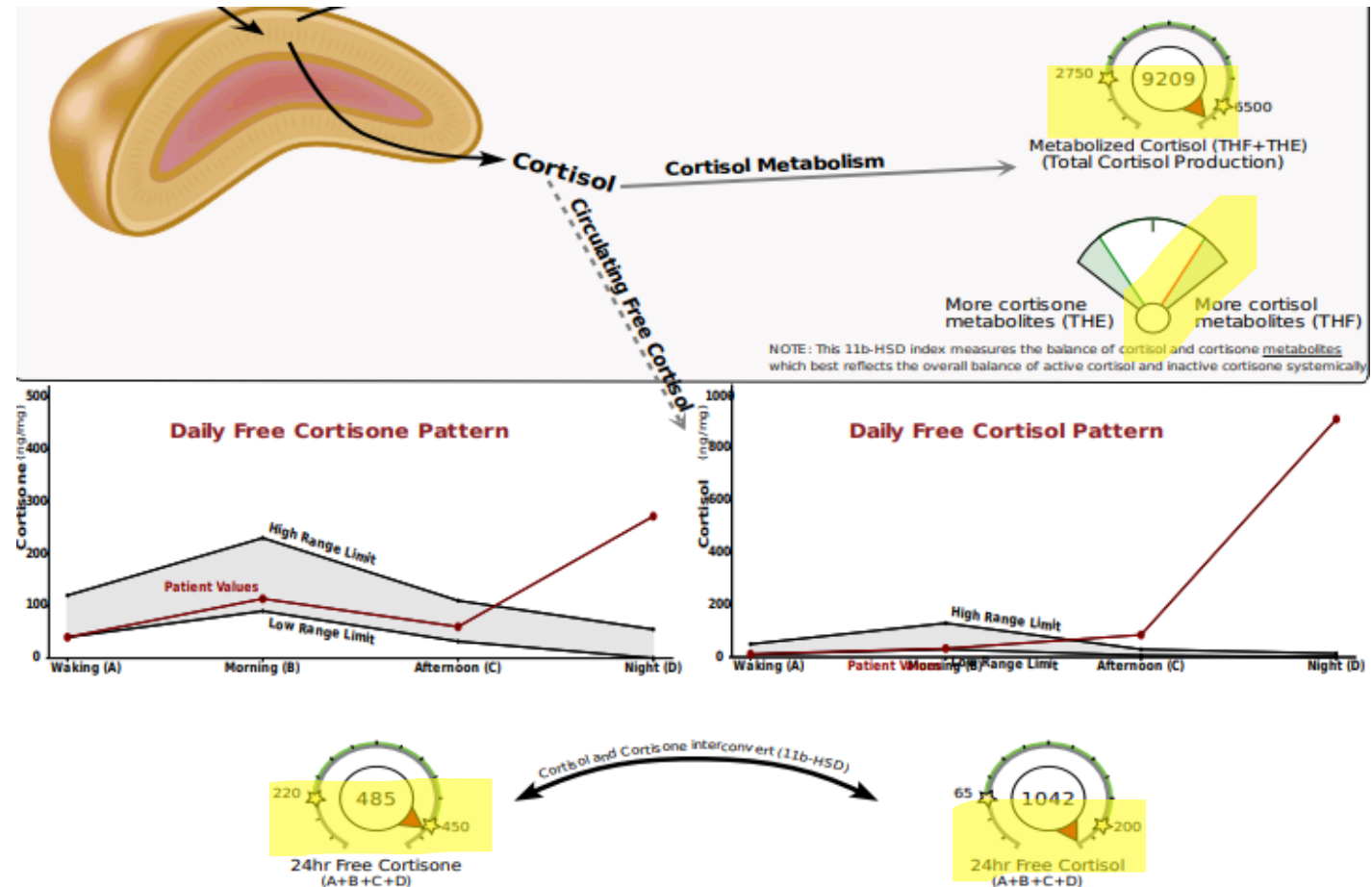
- Decrease CYP3A4 metabolism indirectly
- Increase bioavailable cortisol
- Small study involving DIM has shown this can work



Cases and Examples

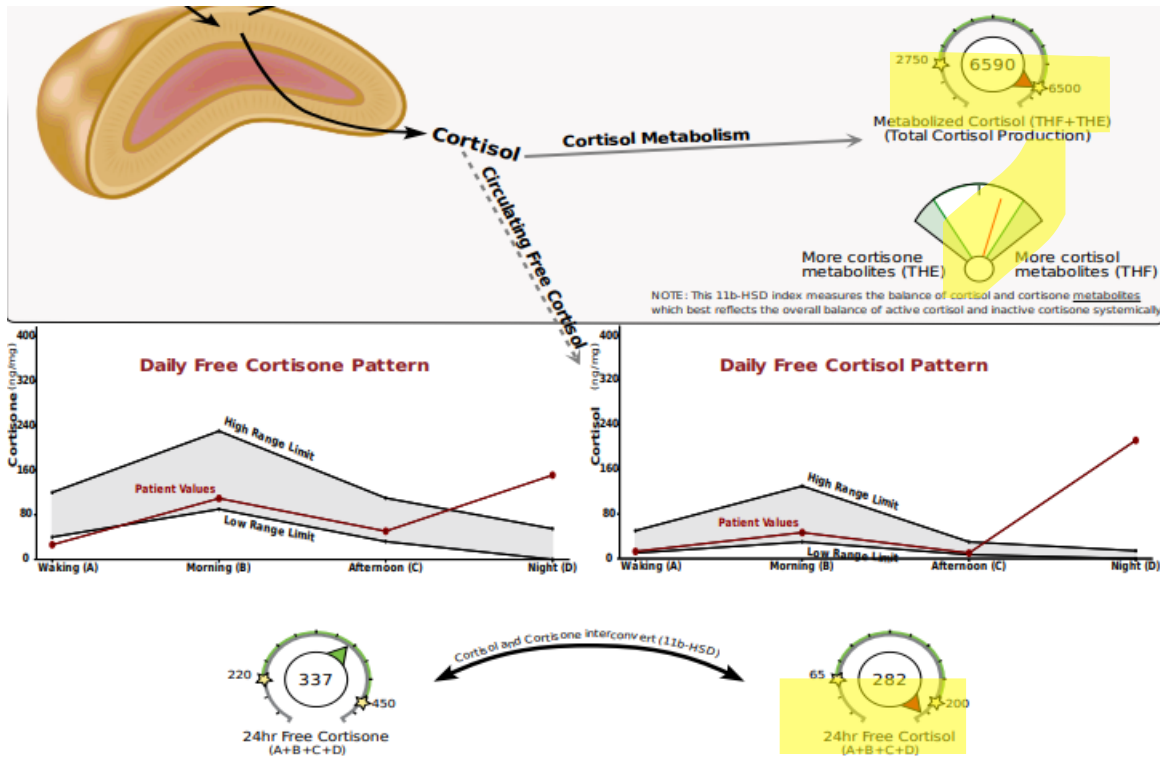
Let's Review ACUTE STRESS

Migraine onset in the afternoon

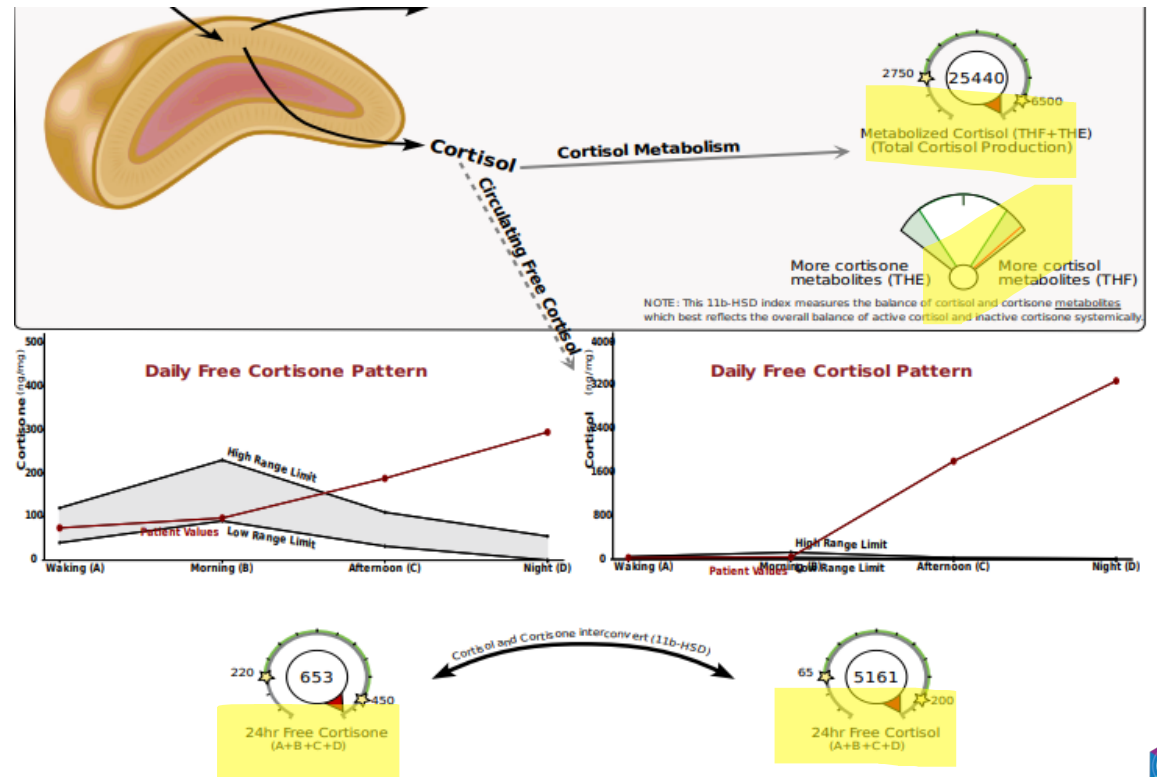


Acute Stress – More migraines

“Bad headache” on the day of testing

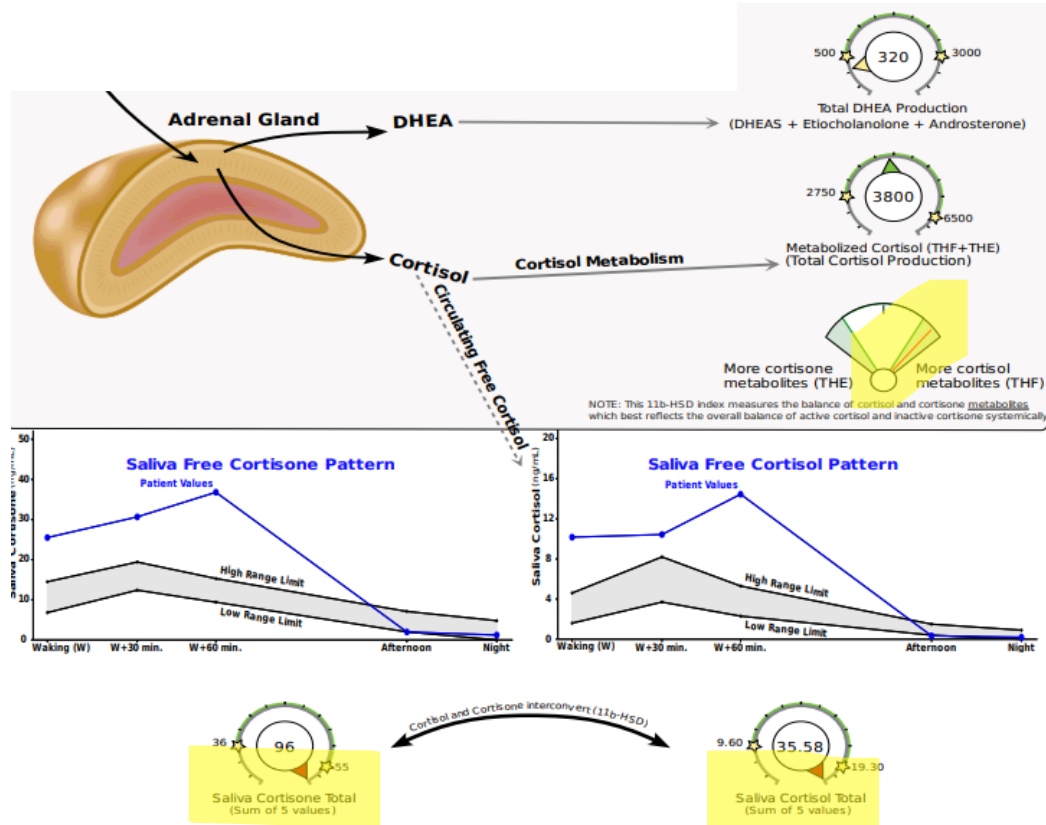


Migraine afternoon, caffeine withdrawal



Acute Stress - Illness

This person had COVID-19 during the day of testing



Treatment for Acutes

Rest

Anti-Inflammatories

Nutrition

Gentle Movement

Meditation

Hydrotherapies

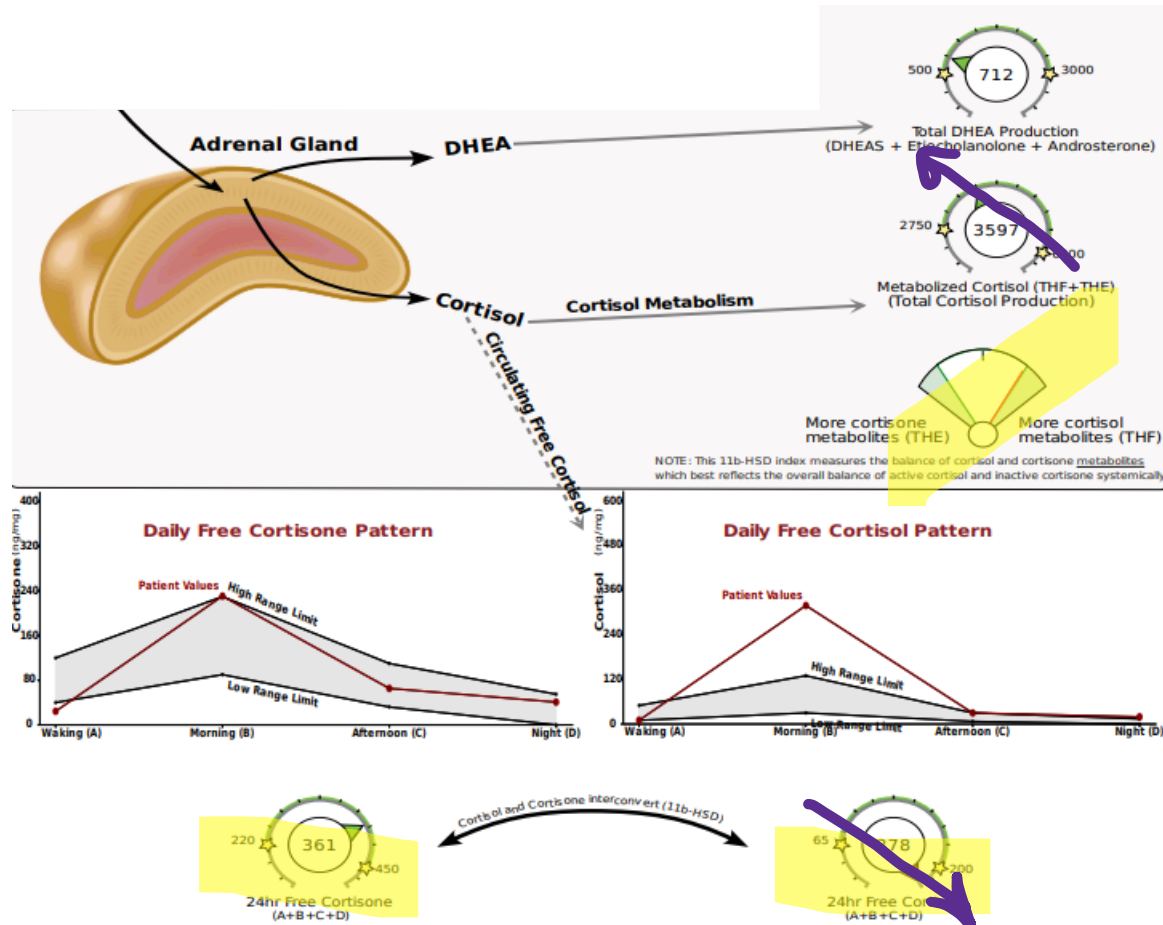




“Classic Patterns” of Chronic Stress

And Interp Considerations

The Hypometabolic Pattern

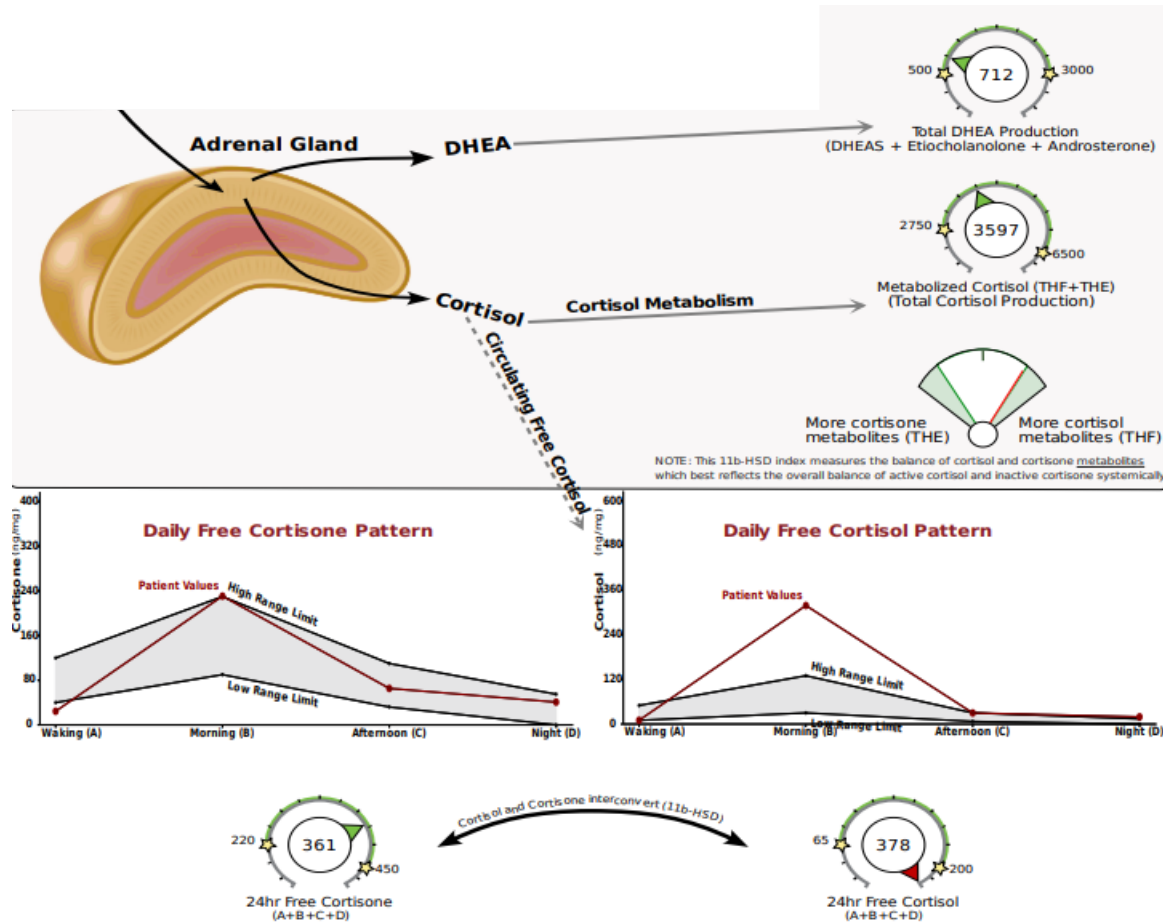


Free cortisol HIGH
(compared to free cortisone)

Free cortisol HIGH
(compared to metabolized cortisol)

Favor toward THF

The Hypometabolic Pattern



Top Considerations:



Hypothyroid (even subclinical)



Anemia (iron deficiency)



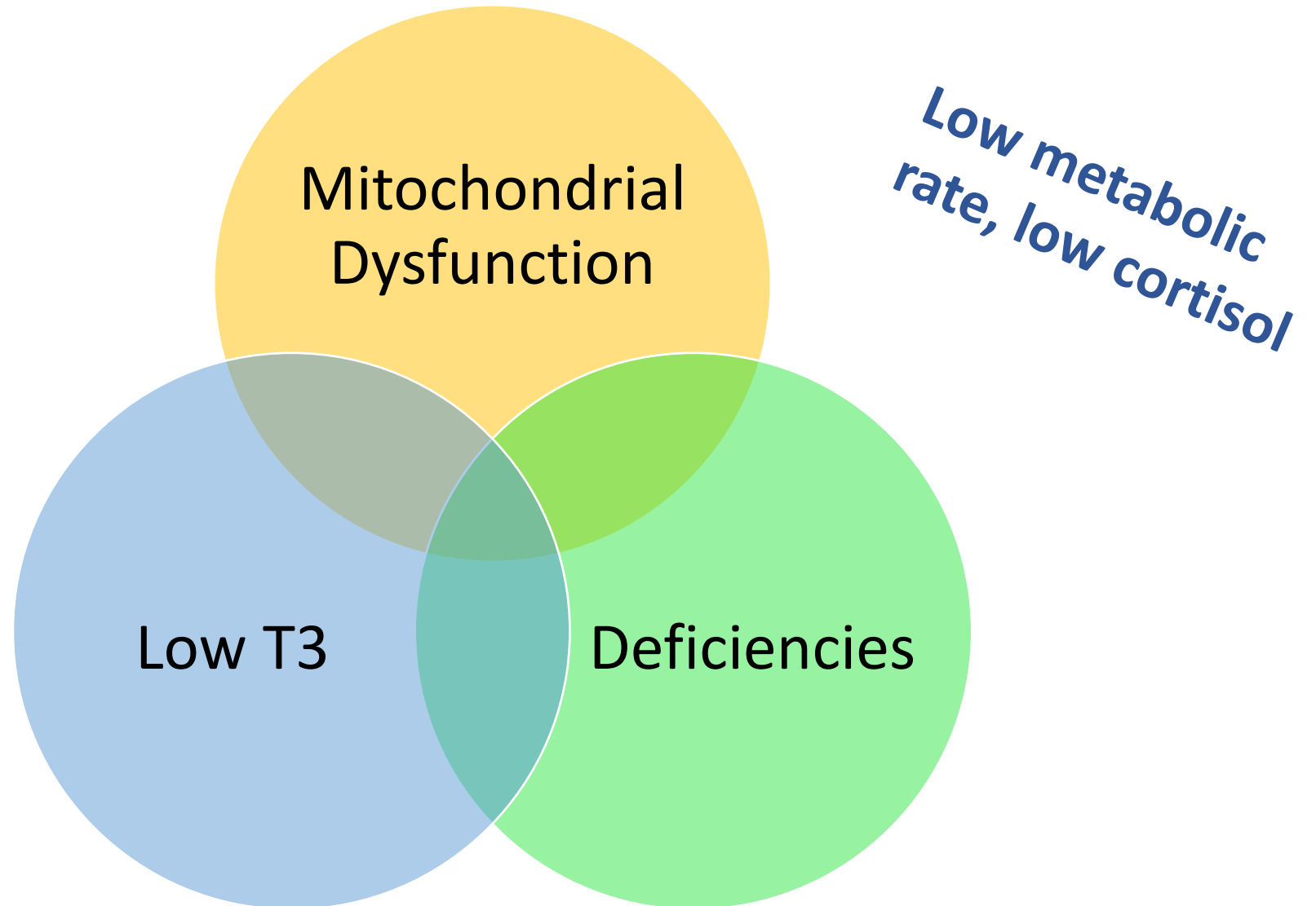
Mitochondrial dysfunction



Liver/Gallbladder Stasis



Hypometabolic Pattern

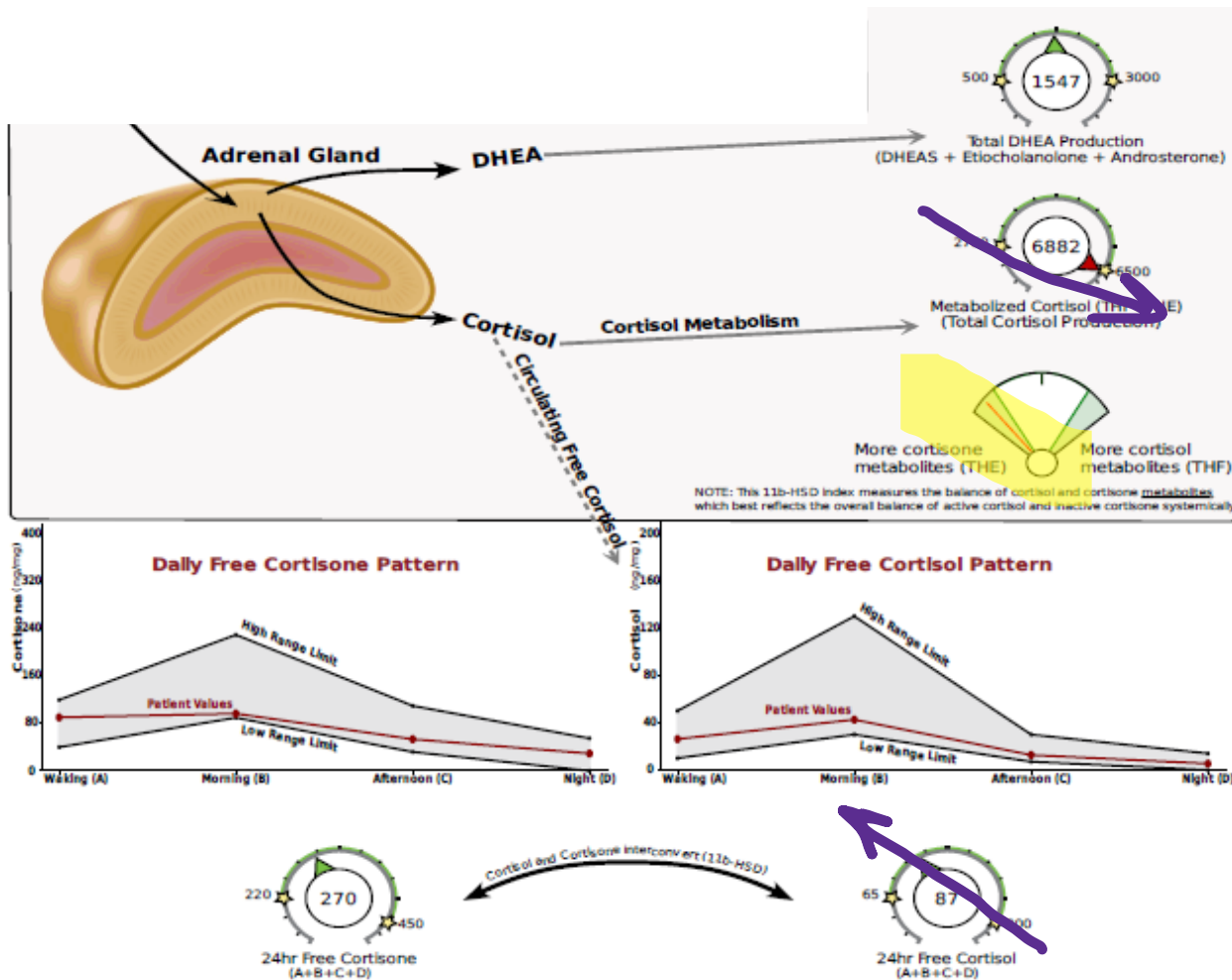


Treatment Goals – ↑ BMR

- Increase thyroid hormone activity
 - Thyroid hormone replacement
 - Iodine therapy if low dietary intake
 - T4→T3 conversion supports
 - Vitamins A, C, E, Sel, Zinc, B vits
- Increase mitochondrial function
 - CoQ10, ALA, Carnitine, Antioxidants, NADH, Taurine
 - If there's toxicity, detox and rebuild
 - Exercise (from yoga → HIIT, trial and individualize)
- Feed, feed, feed - Trophic supports
 - Targeted nutrition, glandulars, B complex, C, EFAs



The Hypermetabolic Pattern



Free cortisol and cortisone
LOW

(compared to total
metabolized cortisol)

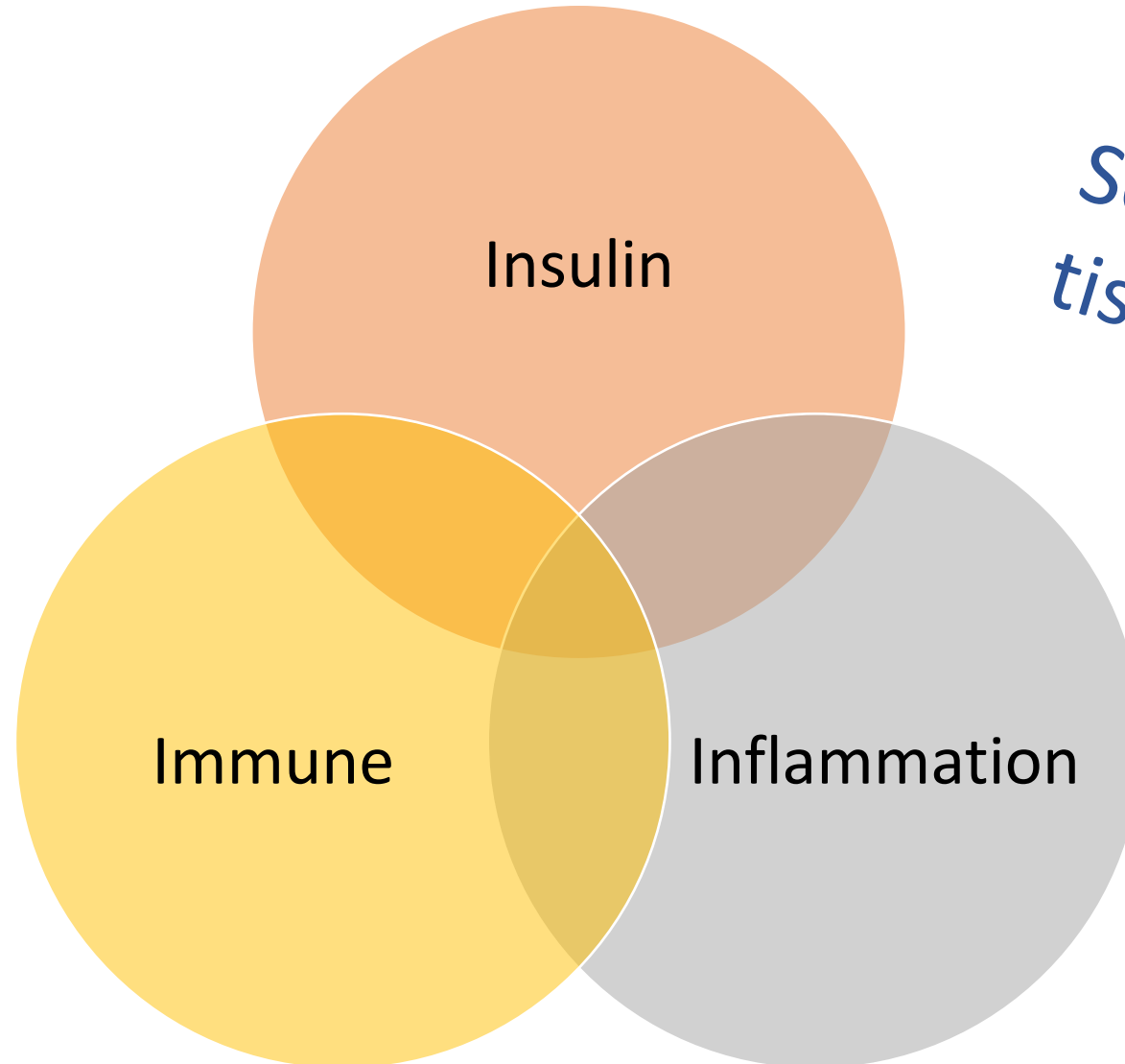
Metabolized cortisol **HIGH**
(compared to free cortisol)

Favor toward THE





The Hypermetabolic Pattern

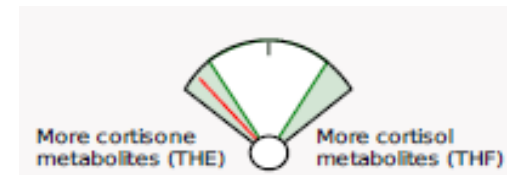
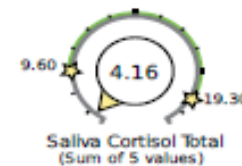
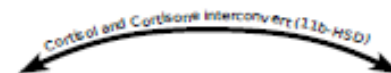
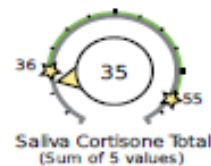
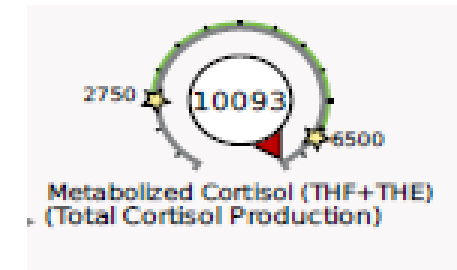
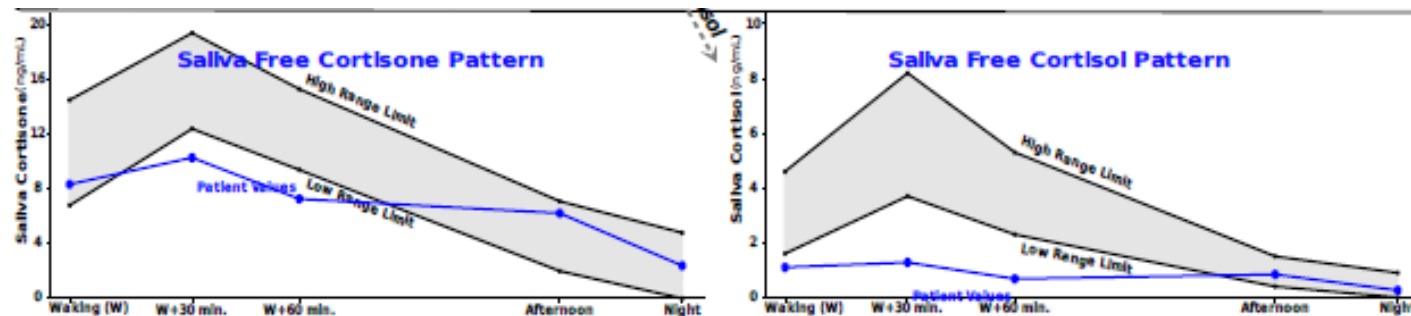
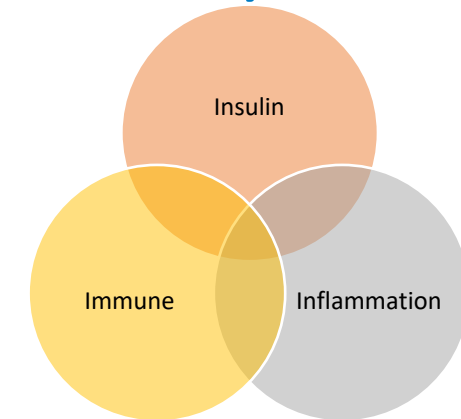


*Solve the
tissue issue!*



Hypermetabolism Assoc with Obesity

- Example: 28yoF, BMI 42
- Shift to free cortisol
- High metabolized cortisol, shift to THE

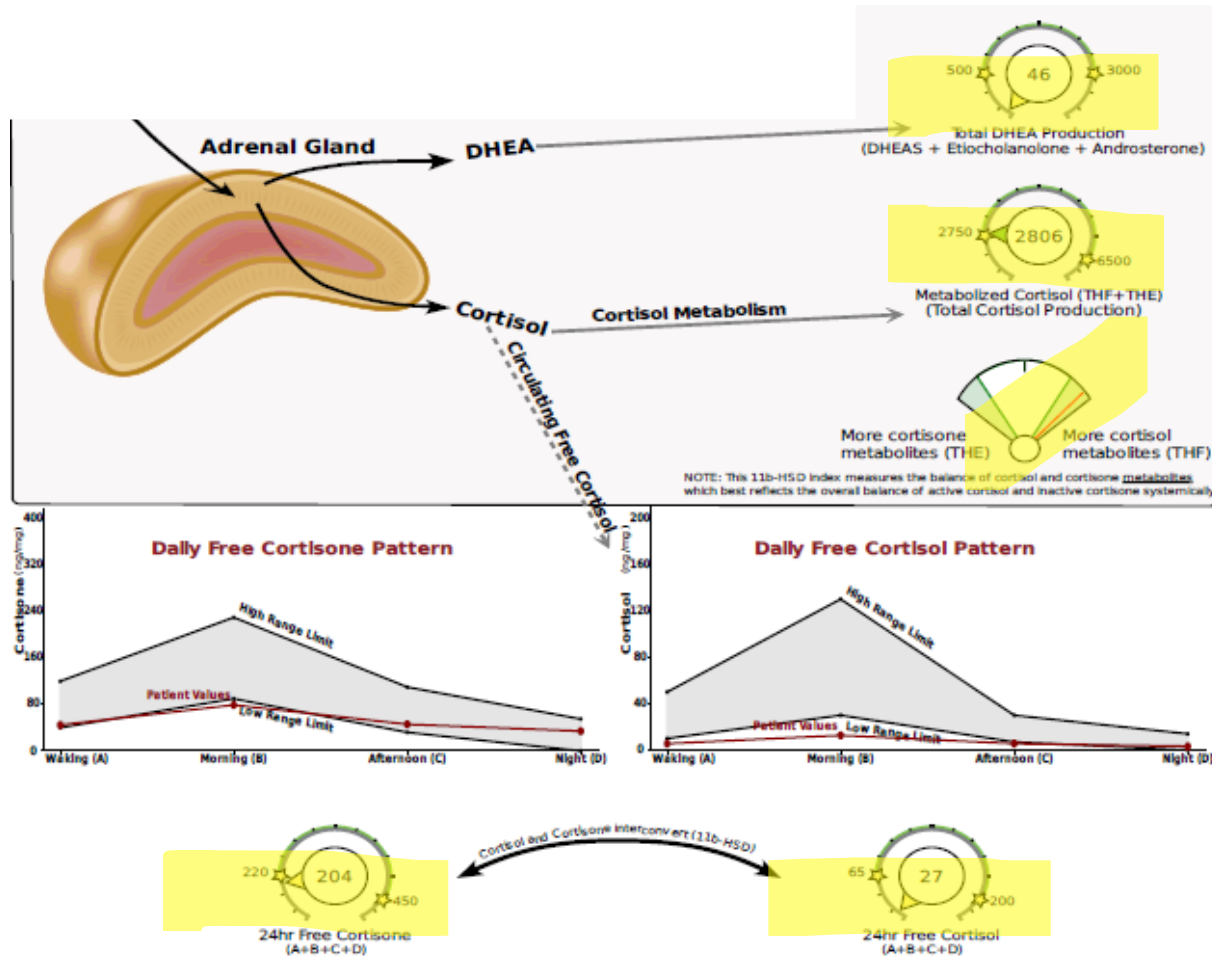


Treatment Goals – Solve the Tissue Issue

- Address inflammation
 - AIP, Exercise, systemic enzyme therapies, mushroom therapies, Curcumin, EGCG,
- Increase insulin sensitivity
 - Bitter melon, Cinnamon, Chamomile, Berberine, ALA, Myoinositol
- Modulate immune activity
 - Berberine, Sulforaphane, Scutellaria, Ginger, Plant sterols, Astragalus
- Assess for sleep apnea
 - Wearables can help, sleep study best
- Rule out hyperthyroid
- Assess for metabolic dysfunction (leptin resistant, cortisone dominant)
 - Especially if weight loss not forthcoming with best efforts → PreDM, T2DM
 - 11BHSD1 inhibiting supports: EGCG, Curcumin, Bitter Melon



Hypocortisol Pattern



LOW Free Cortisol

LOW Free Cortisone

LOW or LOW-NORMAL
metabolized cortisol

Shift to THF (if adapting)

LOW DHEA common



Treatment Goals – Increase Production

- Mitochondrial Supports
 - CoQ10, ALA, Carnitine, Antioxidants, NADH, Taurine
- Trophic Supports
 - B6, B9, B12, C, EFAs, glandulars
- Increase cortisol effects
 - Licorice Extract, Grapefruit Juice, DIM
- Energetic Field Supports
 - Gemmos – Ribes/Sequoia/Quercus, Spagyrics, Homeopathics
- Energizing Adaptogens
 - Cordyceps, Ginsengs, Epimedium, Rhodiola, Maca, Shatavari
- Physical Medicine Approaches
 - Vibration, Biofeedback/HRV training, Meditation, Breathing, Sleep Hygiene practices





Real Fatigue Cases

We made it!

64yoF – Fatigue, Anxiety

- 64yoF with mild anxiety
- Moderate fatigue through the day, resolving by evening – second wind
- Self proclaimed excellent sleeper, bed by 11pm and up at 5am daily (but with melatonin)
- Retired and has a low stress life
- No Rx meds
- Supplements: Probiotic, NAC, B vitamins, Inflammation (for intermittent back pain), Garlic, B12, melatonin, and a MVM



64yoF – Fatigue, Anxiety

- Blood Pressure 122/70
- Serum labs drawn and micronutrient testing
 - CBC w/ diff - WNL
 - CMP-14 - WNL
 - Ferritin, B12, TIBC - WNL
 - Thyroid panel - WNL
 - Urinalysis – WNL
 - Vitamin D, CoQ10, ALA, omega 3s, and magnesium all found to be low
- DUTCH Complete – sex and adrenal hormones

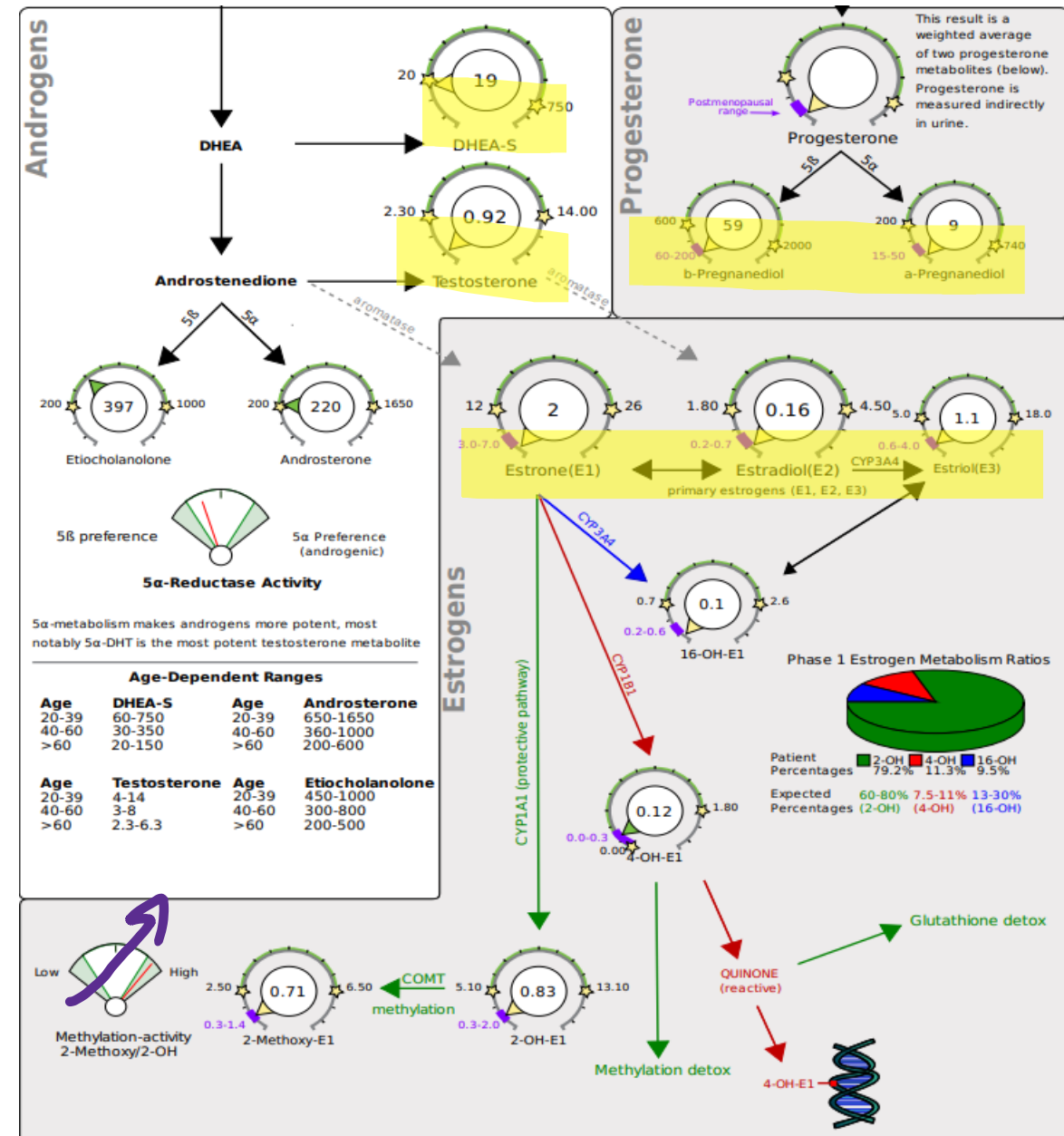


64yoF – Fatigue, Anxiety

Pan LOWS, even for
menopausal status

Mitochondrial dysfunction?

COMT activity is high



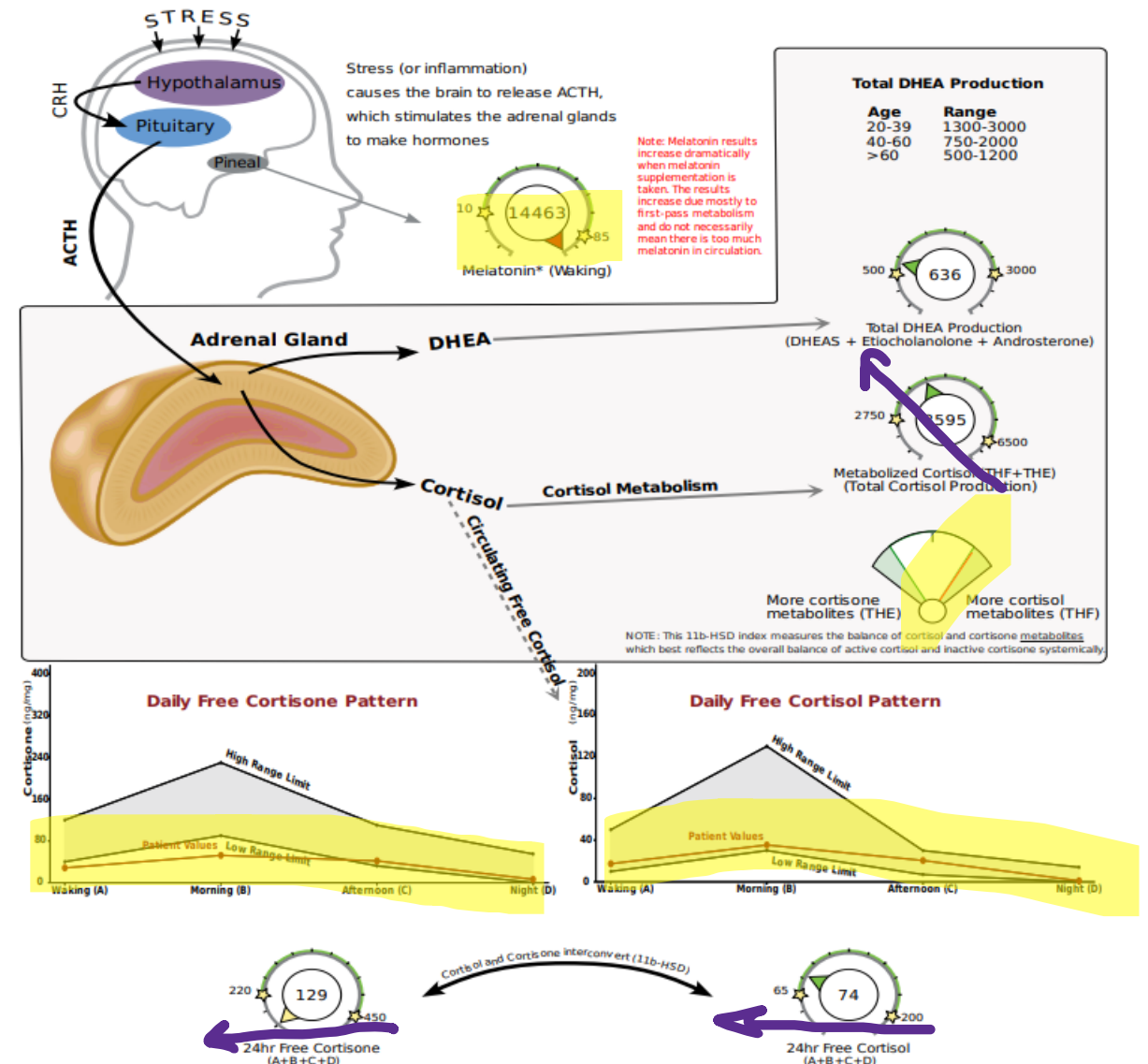
64yoF – Fatigue, Anxiety

Melatonin high (with melatonin supp)

Flattened free cortisol slopes, low-normal

Metabolized cortisol isn't super low but it's not even mid-range

Shift to THF



64yoF – Fatigue, Anxiety



- Continue with Probiotic, NAC, B vitamins, Inflammation, B12, melatonin
- Discontinue MVM, garlic
- Add: Vitamin D 5000 IU (125 mcg) QD, ALA 300 mg BID, CoQ10 100 mg QD, EPA 250 mg/DHA 750 mg QD, Mag threonate QD, “Cortisol Manager” (Phosphoserine, Theanine, Ashwagandha, Magnolia, Epimedium), and “Licorice Plus” (Licorice extract, Ashwagandha, Rehmannia, Polygonum)



64yo (now 65yoF) RTC 1 year later

- No longer complaining of fatigue
- 9-10/10 on the VAS for energy throughout the day
- She came back just to retest and see everything normalize to how she feels
- Anxiety rarer in frequency and mild when it does present
- She discontinued the vitamin D several months ago after a serum D level came back at 68 ng/mL
- Also discontinued melatonin, doesn't feel she needs anymore
- Follow-up DUTCH Complete ordered



64yo (now 65yoF)
RTC 1 year later

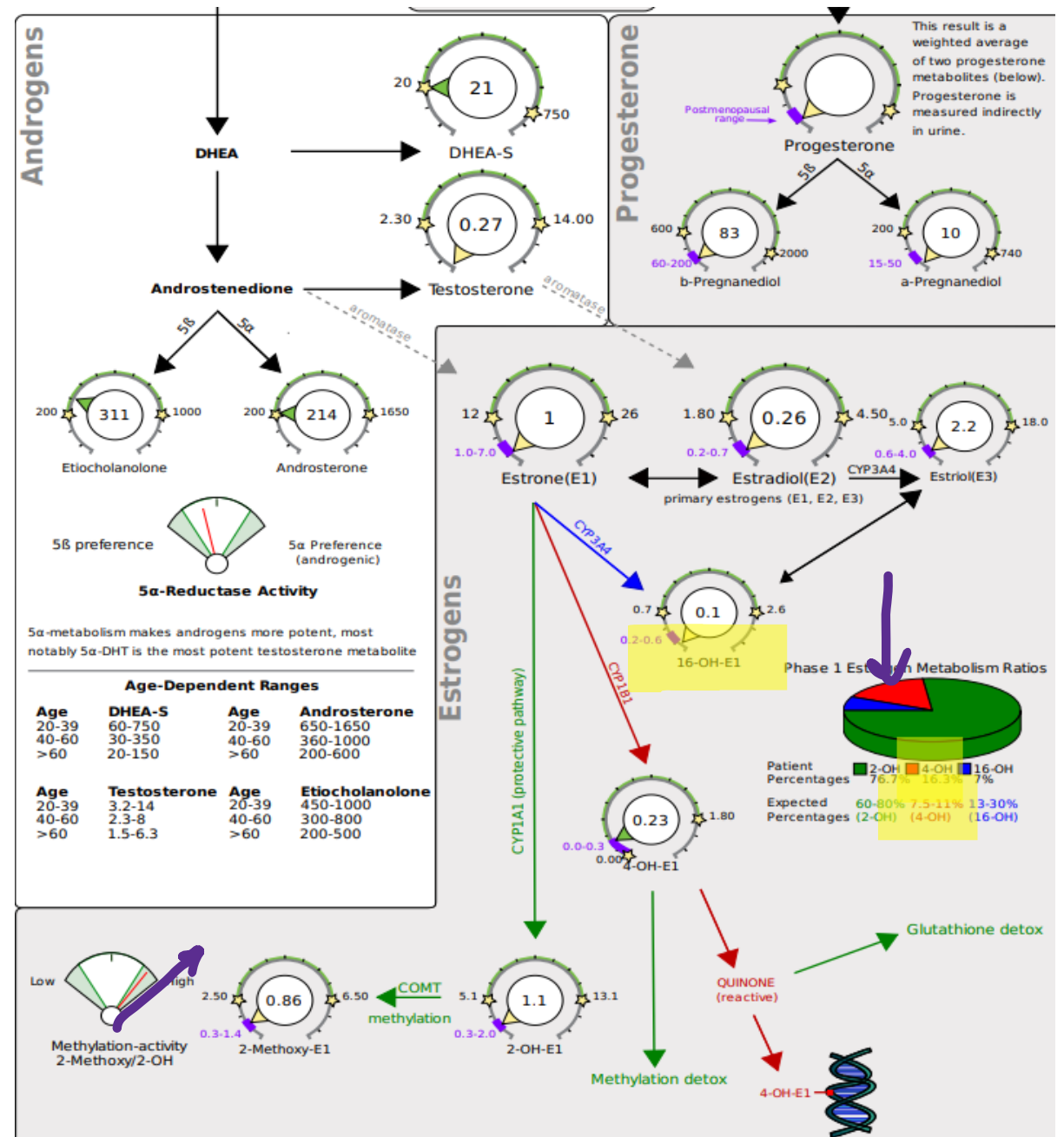
Mostly NORMAL, few lows

16OHE1 still low

Bone health?

CYP1B1 activity high –
inflammation?

COMT activity still high



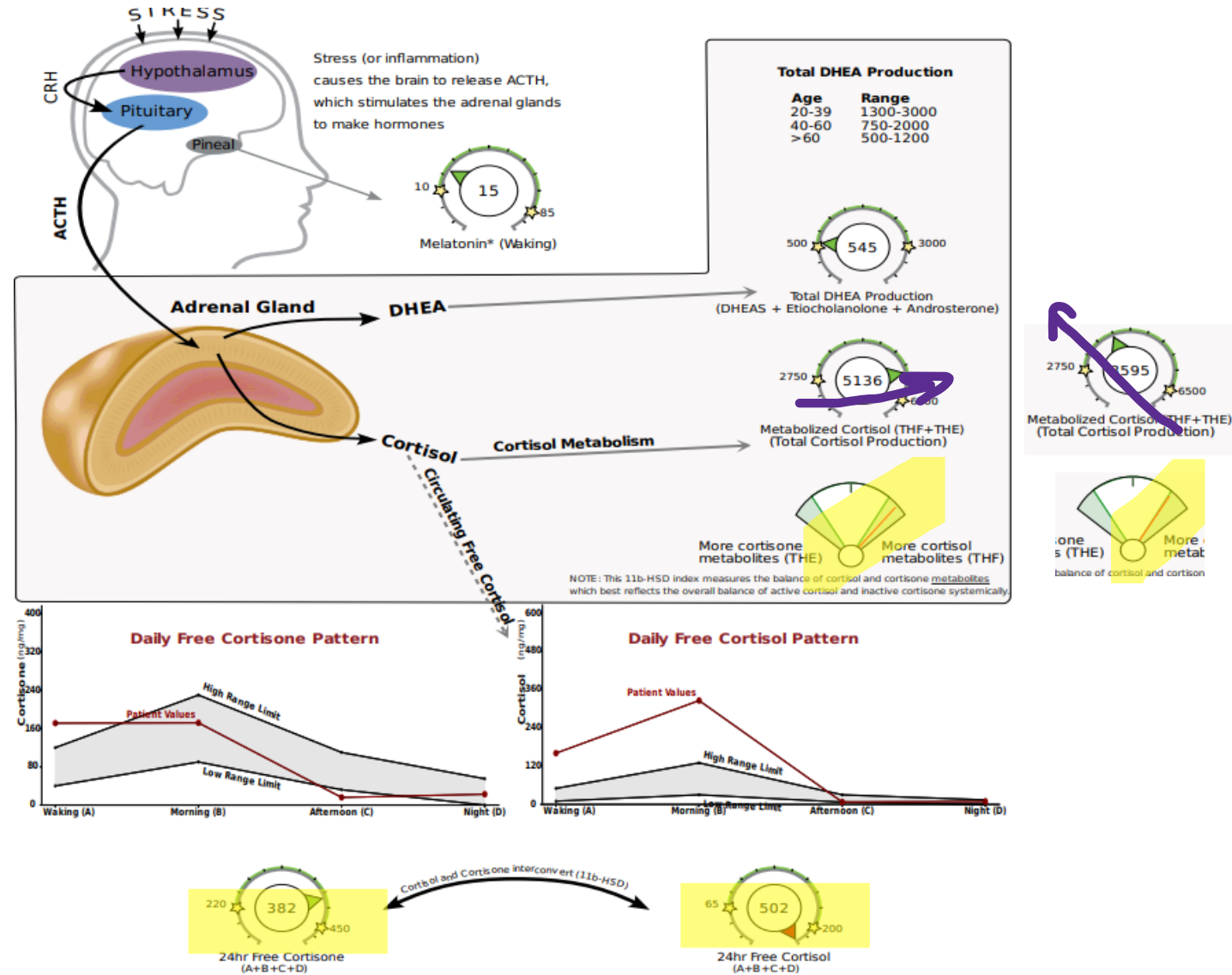
64yo (now 65yoF)
RTC 1 year later

Free Cortisols are HIGH
Licorice likely cause
Notice 11BHSD2
activity looks blocked
Note to check K levels

Metabolized cortisol much
higher, pull to THF

Inflammation?
CYP1B1 was also ↑

Melatonin output low-normal



64yo (now 65yoF) – Resolved Fatigue, Anxiety

- Future directions:
 - Draw inflammatory markers
 - Order CMP – check electrolytes if continued use of Licorice is desired
 - Time for a DEXA
 - Explore sleep



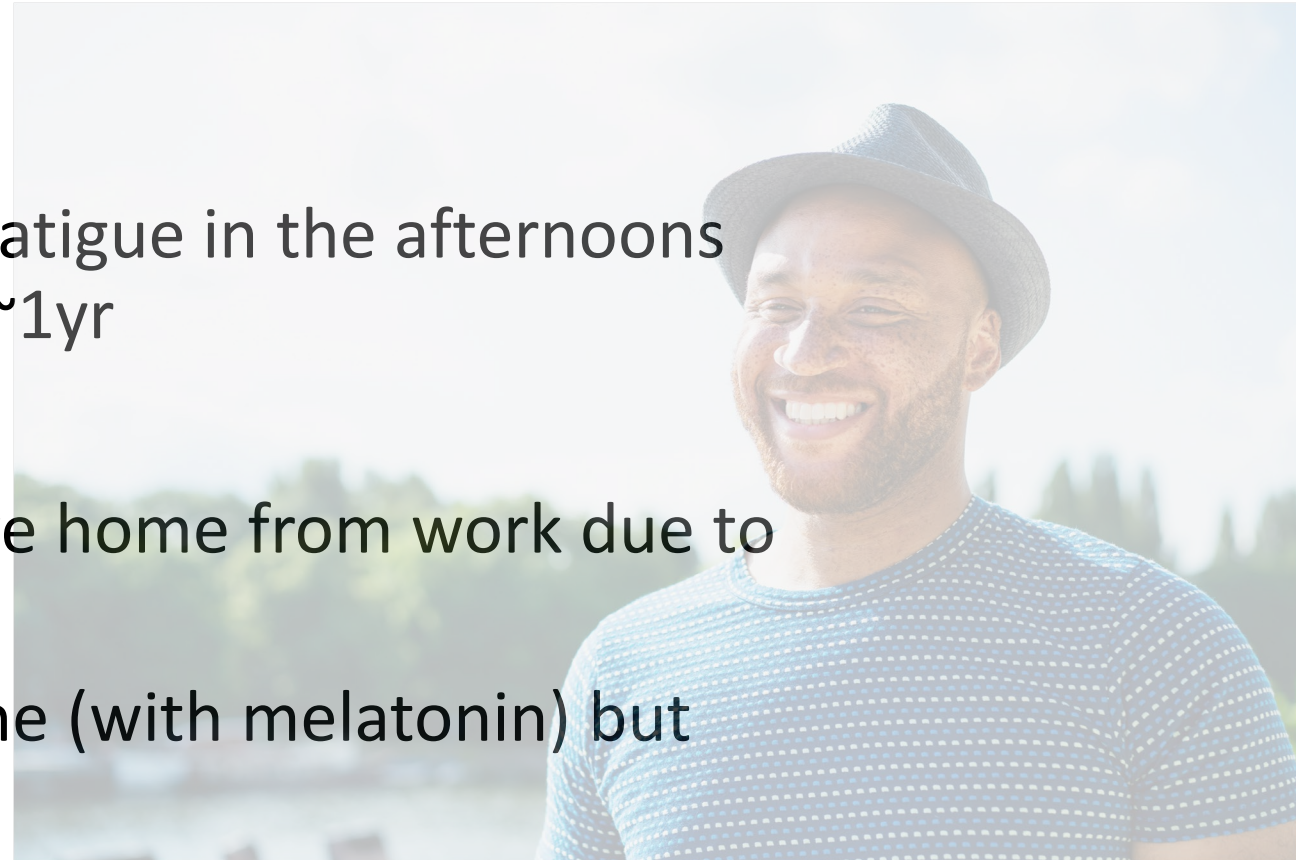


Ready for one more?

Male Case – severe fatigue

59yoM – Fatigue, Sleep (frequent waking)

- BMI 28 – overweight category
- Chief complaint is debilitating fatigue in the afternoons particularly and consistently x ~1yr
- High stress, high level career
- Concerned about ability to drive home from work due to fatigue
- Sleep disturbed, sleep onset fine (with melatonin) but frequent waking after that
- Meds: ACEI QD for blood pressure, melatonin 10mg QHS

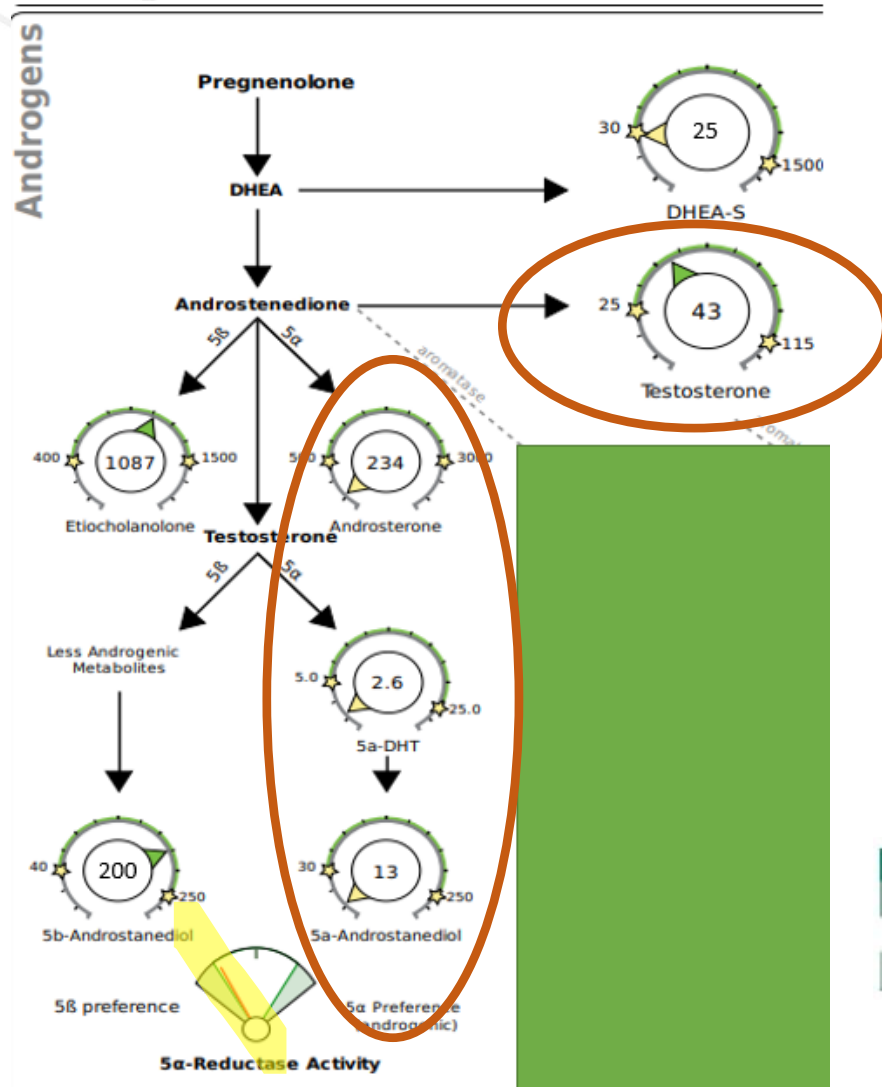


59yoM - Pertinent Findings

- Blood Pressure 130/76
- Serum labs drawn – Few remarkable findings
 - CBC w/ diff
 - CMP-14 (fasting lipids elevated)
 - Ferritin, B12, TIBC
 - Thyroid panel
 - Urinalysis
- DUTCH Complete – sex and adrenal hormones



DUTCH Results – Androgens Down



DHEA LOW

Testosterone NORMAL(ish)*

***5αReductase Activity LOW**

Ask patient about Finasteride use

Age-Dependent Ranges

Age	DHEA-S
20-39	150-1500
40-60	60-800
>60	30-300

Etiocholanolone	Androsterone
20-39 800-1500	20-39 1500-3000
40-60 600-1200	40-60 1000-2000
>60 400-1000	>60 500-1000

5β-androstenediol	5α-androstenediol
20-39 70-250	20-39 60-250
40-60 55-210	40-60 50-180
>60 40-150	>60 30-130

Testosterone	5α-DHT
18-25 35-115	20-39 9-25
26-40 30-95	40-60 7-20
41-60 25-80	>60 5-16
>60 20-60	

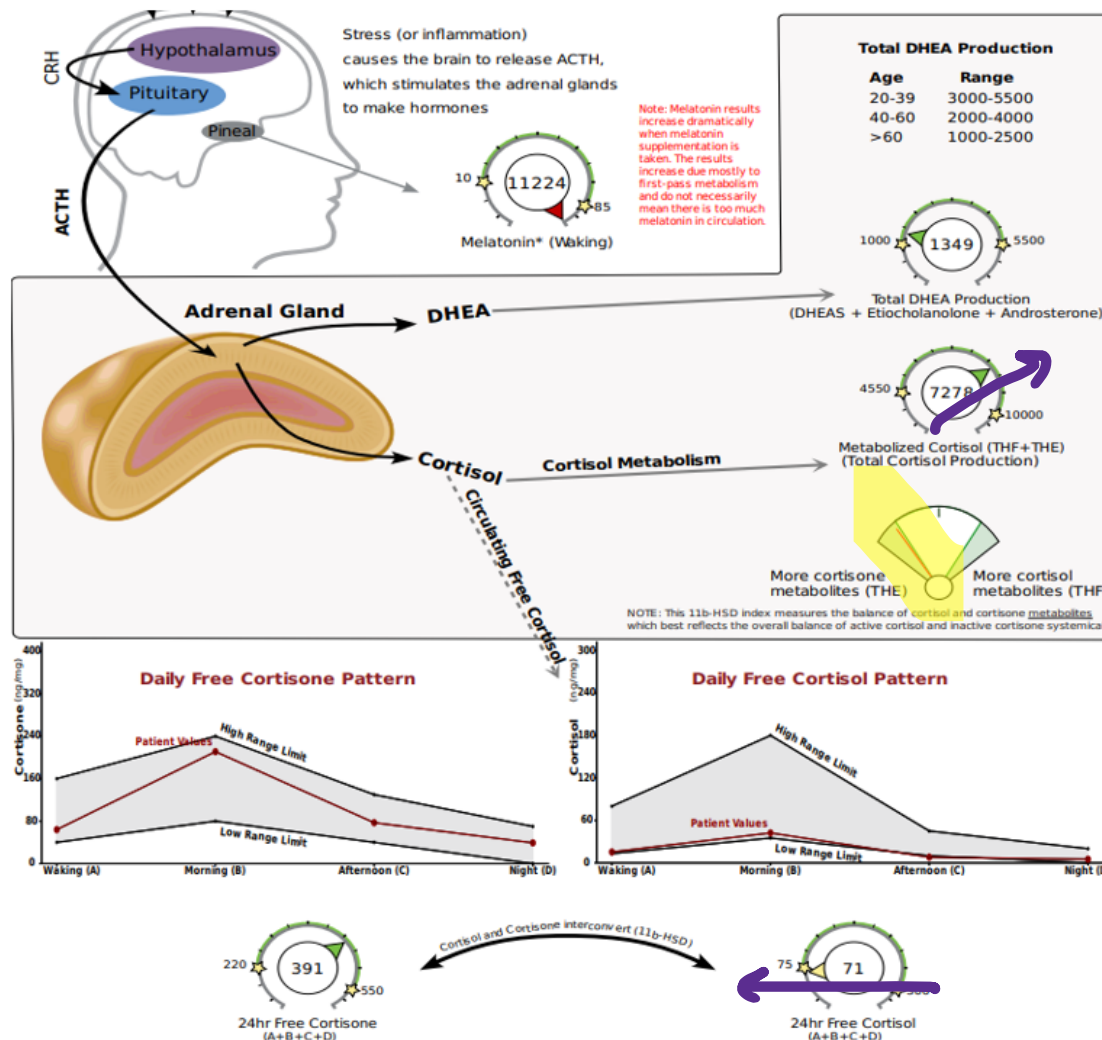
Category	Test	Result	Units	Normal Range
Progesterone Metabolites (Urine)				
	b-Pregnanediol	Within range	231.0	ng/mg 75 - 400
	a-Pregnanediol	Below range	11.8	ng/mg 20 - 130





Good for estrogen, bad for dopamine and norepi?

DUTCH Results – Adrenals + Melatonin



Melatonin high BUT these are 1st pass metabolites from oral melatonin 10mg

DHEA LOW for age

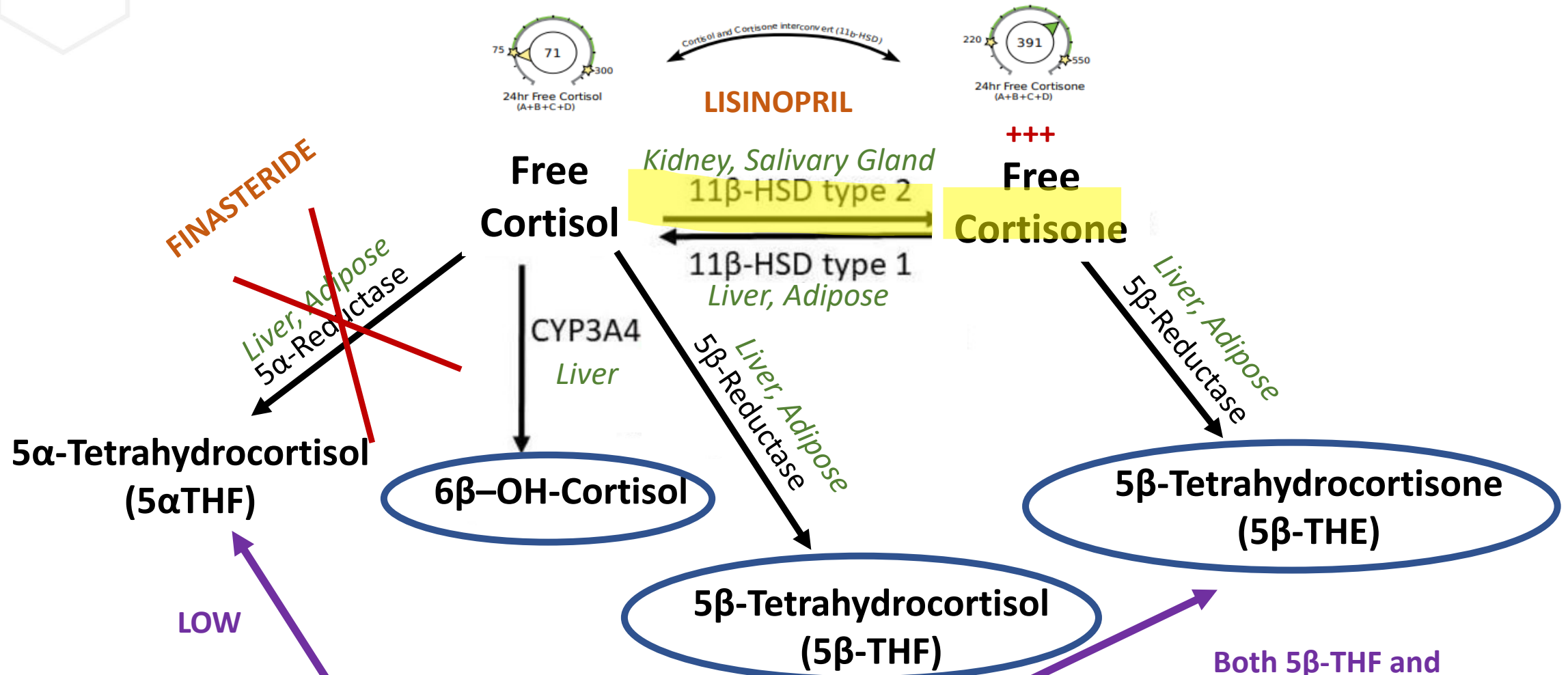
Cortisol production via 11B-HSD1 NORMAL
(total metabolized cortisol 7278)

SHIFT to THE

Little to no 5aReductase activity

Cortisol Metabolites and DHEA-S (Urine)				
a-Tetrahydrocortisol (a-THF)	Below range	12.6	ng/mg	175 - 700
b-Tetrahydrocortisol (b-THF)	Within range	2721.6	ng/mg	1750 - 4000
b-Tetrahydrocortisone (b-THE)	Within range	4544.1	ng/mg	2350 - 5800
Metabolized Cortisol (THF+THE)	Within range	7278.3	ng/mg	4550 - 10000
DHEA-S	Below range	28.0	ng/mg	30 - 1500

Cortisol Clearance and Metabolism

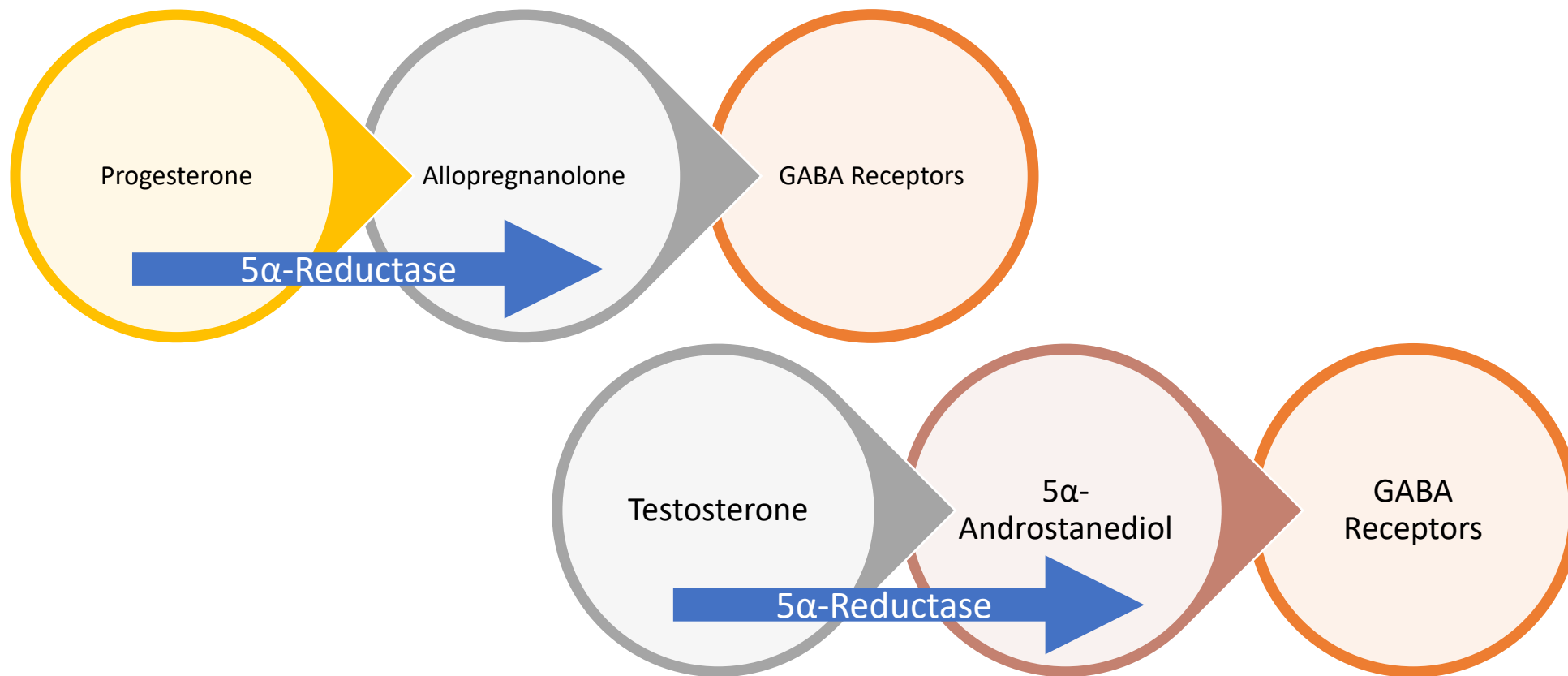


Cortisol Metabolites and DHEA-S (Urine)				
a-Tetrahydrocortisol (a-THF)	Below range	12.6	ng/mg	175 - 700
b-Tetrahydrocortisol (b-THF)	Within range	2721.6	ng/mg	1750 - 4000
b-Tetrahydrocortisone (b-THE)	Within range	4544.1	ng/mg	2350 - 5800
Metabolized Cortisol (THF+THE)	Within range	7278.3	ng/mg	4550 - 10000



59yoM - So why FATIGUE and SLEEP?

5 α Reductase creates NEUROSTEROIDS



Thoughts for Increasing Neurosteroids

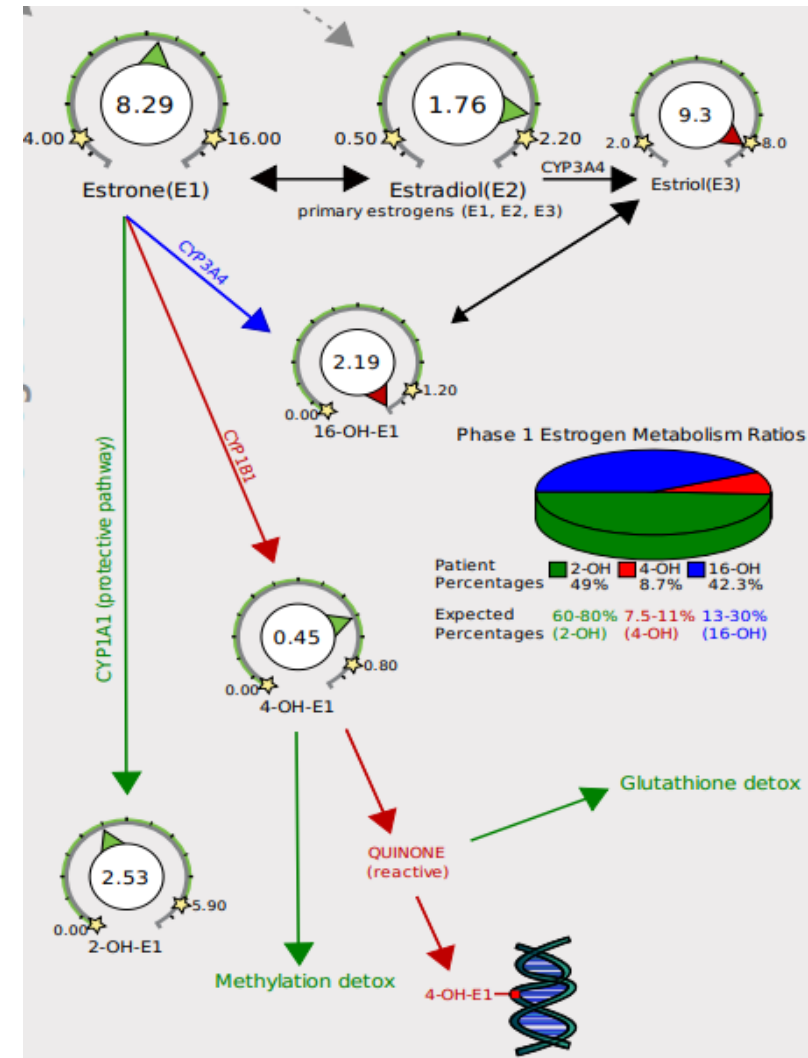
- Micronized Progesterone 25-100mg PO QHS
- Pregnenolone 30 – 300mg PO QHS
- Testosterone replacement
- DHEA oral therapy 25-100mg PO QD
- GABA-ergic Adaptogens:
 - Bacopa, Ashwagandha, Relora, Jujube, Melissa, Mimosa
- GABA signaling probiotic strains
 - L. brevis
 - L. rhamnosus



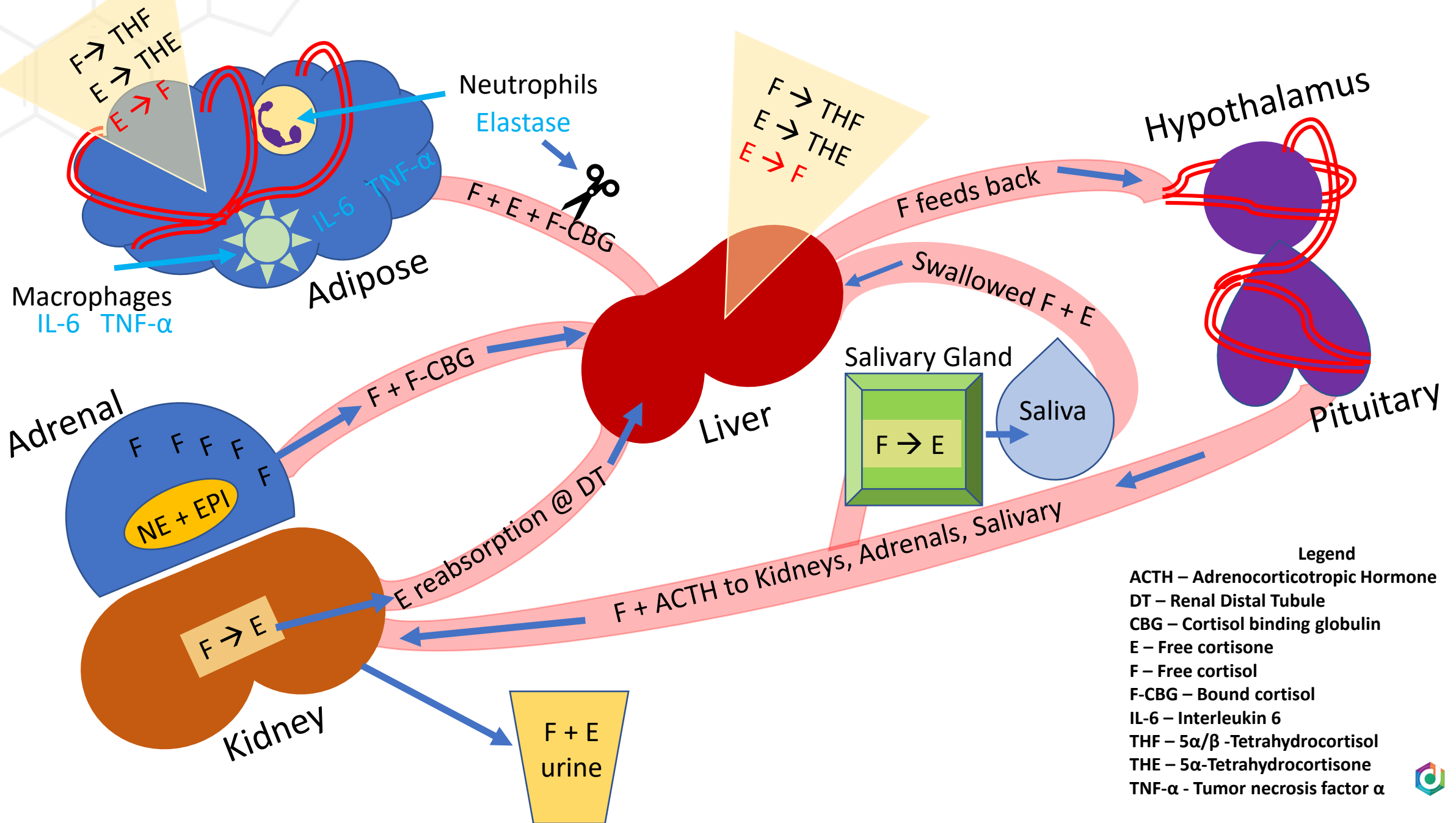
59yoM – So why FATIGUE and SLEEP?

CYP3A4 Overactivity and high Estrogens/Testosterone

- CYP1A1 Inducers:
 - DIM 50-200mg QD
 - Ground flax seeds
 - Brassica family veggies
 - Rosemary Extract
 - Soy, even soy milk
- As 3A4 activity normalizes, so should cortisol availability









New Providers Receive 50% Off Up To 5 Testing Kits

Additional Benefits Include:

- Easy At-Home Collection
- Comprehensive Reporting Results
- Drop-Ship to Your Patient's Doorstep
- Quick Turn-Around Time on Lab Results
- Dedicated Onboarding Concierge to Help Providers Get Started
- Free Clinical Consults
- Video Tutorials
- Validated Peer-Reviewed Research

Click the link below to get started today!



Questions?

Email: info@dutchtest.com

Thank you for listening!

Like Us, Follow Us, or Leave a Review!

<https://www.facebook.com/dutchtest>

<https://twitter.com/DutchTestLab>

<https://www.linkedin.com/company/10883377>

<https://www.instagram.com/dutchtest/>

<https://www.youtube.com/channel/UCauTDzKu4VS74PMiYgda-PQ>

