

TEST NUMBER: G-NL-XXXXX

GENDER: XXXXXX

AGE: XX

COLLECTED: 00-XXX-2024 RECEIVED: 00-XXX-2024

TESTED:

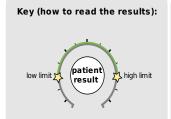
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TEST REF: GNL-NL-XXXXX

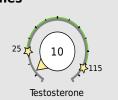
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TEST NAME: DUTCH Complete™

Hormone Testing Summary

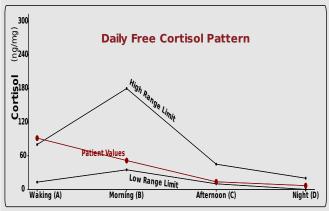


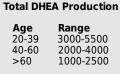




Testo	sterone
Age	Range
18-25	35-115
26-40	30-95
41-60	25-80
>60	20-60

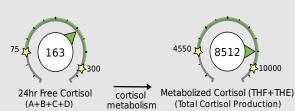
Adrenal Hormones See pages 4 and 5 for a more complete breakdown of adrenal hormones











Free cortisol best reflects tissue levels. Metabolized cortisol best reflects total cortisol production.

PLEASE BE SURE TO READ BELOW FOR ANY SPECIFIC LAB COMMENTS. More detailed comments can be found on page 7.



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Category	Test		Result	Units	Normal Range
Progesto	erone Metabolites (Urine)				
	b-Pregnanediol	Low end of range	81.6	ng/mg	75 - 400
	a-Pregnanediol	Low end of range	25.6	ng/mg	20 - 130
Estrogei	ns and Metabolites (Urine)				
	Estrone(E1)	Low end of range	4.59	ng/mg	4 - 16
	Estradiol(E2)	Within range	0.8	ng/mg	0.5 - 2.2
	Estriol(E3)	Within range	3.0	ng/mg	2 - 8
	2-OH-E1	Within range	1.95	ng/mg	0 - 5.9
	4-OH-E1	Within range	0.27	ng/mg	0 - 0.8
	16-OH-E1	Within range	0.24	ng/mg	0 - 1.2
	2-Methoxy-E1	Within range	0.99	ng/mg	0 - 2.8
	2-OH-E2	Within range	0.12	ng/mg	0 - 0.6
	4-OH-E2	Within range	0.08	ng/mg	0 - 0.3
	Total Estrogen	Low end of range	12.0	ng/mg	10 - 34
Metabol	ite Ratios				
	2-OH / 16-OH-E1 Balance	Within range	8.17	ratio	2.85 - 9.88
	2-OH / 4-OH-E1 Balance	Low end of range	7.16	ratio	6.44 - 12.6
	2-Methoxy / 2-OH Balance	Within range	0.51	ratio	0.4 - 0.7
Androge	ens and Metabolites (Urine)				
	DHEA-S	Below range	18.6	ng/mg	30 - 1500
	Androsterone	Below range	446.7	ng/mg	500 - 3000
	Etiocholanolone	Within range	657.0	ng/mg	400 - 1500
	Testosterone	Below range	10.32	ng/mg	25 - 115
	5a-DHT	Below range	3.2	ng/mg	5 - 25
	5a-Androstanediol	Below range	16.6	ng/mg	30 - 250
	5b-Androstanediol	Low end of range	40.7	ng/mg	40 - 250
	Epi-Testosterone	Within range	53.3	ng/mg	25 - 115



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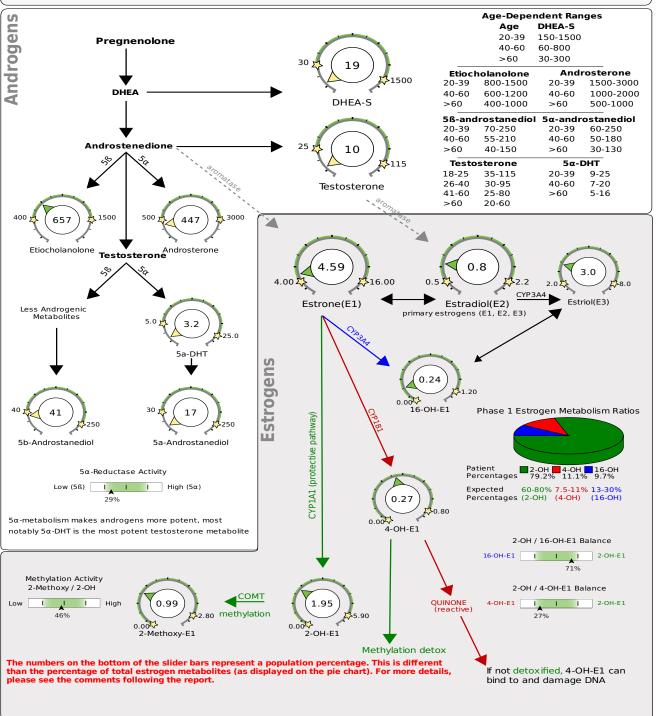
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Hormone metabolite results from the previous page are presented here as they are found in the steroid cascade. See the Provider Comments for more information on how to read the results.



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Category	Test		Result	Units	Normal Range
Creatinine	(Urine)				
	Creatinine A (Waking)	Within range	0.34	mg/ml	0.3 - 3
	Creatinine B (Morning)	Within range	0.85	mg/ml	0.3 - 3
	Creatinine C (Afternoon)	Within range	0.49	mg/ml	0.3 - 3
	Creatinine D (Night)	Within range	0.37	mg/ml	0.3 - 3
Daily Free	Cortisol and Cortisone (Urine)				
	Cortisol A (Waking)	Above range	91.5	ng/mg	13 - 80
	Cortisol B (Morning)	Low end of range	51.5	ng/mg	35 - 180
	Cortisol C (Afternoon)	Low end of range	13.6	ng/mg	10 - 45
	Cortisol D (Night)	Within range	6.8	ng/mg	0 - 20
	Cortisone A (Waking)	Within range	136.9	ng/mg	40 - 160
	Cortisone B (Morning)	Low end of range	111.5	ng/mg	80 - 240
	Cortisone C (Afternoon)	Low end of range	46.4	ng/mg	40 - 130
	Cortisone D (Night)	Within range	29.5	ng/mg	0 - 70
	24hr Free Cortisol	Within range	163.4	ng/mg	75 - 300
	24hr Free Cortisone	Within range	324.3	ng/mg	220 - 550
Cortisol Me	etabolites and DHEA-S (Urine)				
	a-Tetrahydrocortisol (a-THF)	Low end of range	219.4	ng/mg	175 - 700
	b-Tetrahydrocortisol (b-THF)	Above range	4479.7	ng/mg	1750 - 4000
	b-Tetrahydrocortisone (b-THE)	Within range	3813.3	ng/mg	2350 - 5800
	Metabolized Cortisol (THF+THE)	Within range	8512.4	ng/mg	4550 - 10000
	DHEA-S	Below range	18.6	ng/mg	30 - 1500



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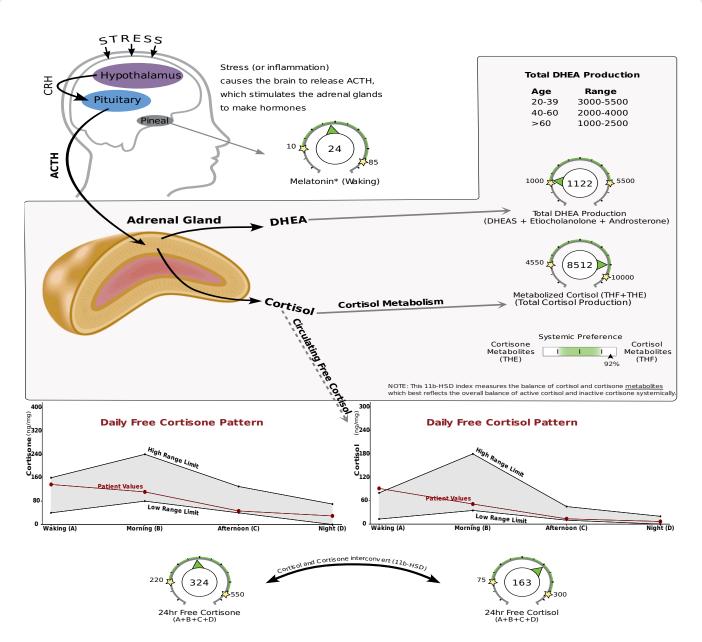
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The first value reported (Waking "A") for cortisol is intended to represent the "overnight" period. When patients sleep through the night, they collect just one sample. In this case, the patient woke during the night and collected (see the top of the report for the times collected). We call this value "A1" and the value from the sample collected at waking "A2." These values are used to create a "time-weighted average" to create the "A" value. The individual values are listed here for your use:

The middle-of-the-night "A1" sample registered a cortisol value of 12.1ng/mg.

The waking "A2" sample registered a cortisol value of 123ng/mg.

These two values are averaged together, taking into account the amount of time each one represents, to create the "A" value of approximately 91.5ng/mg that you will see on the report.

In this particular case, this A2 value is larger than the sample (collected two hours after waking) expected to have the highest cortisol value.

Cortisol levels typically rise sharply after waking thanks to the cortisol awakening response. In a case like this where the waking sample (A2) shows higher levels, this cortisol awakening response may have happened while the patient was in bed before rising.

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Category	Test		Result	Units	Normal Range
	Nu	tritional Organic Acid	ls		
Vitamin B12	Marker (may be deficient if high) - (Urine)			
	Methylmalonate (MMA)		2.1	ug/mg	0 - 3.5
Vitamin B6 M	arkers (may be deficient if high)) - (Urine)			
	Xanthurenate	Within range	0.42	ug/mg	0.2 - 1.9
	Kynurenate	Within range	2.2	ug/mg	1 - 6.6
Biotin Marker	(may be deficient if high) - (Uri	ne)			
	b-Hydroxyisovalerate	Within range	12.3	ug/mg	0 - 18
Glutathione N	Narker (may be deficient if low o	r high) - (Urine)			
	Pyroglutamate	Within range	63.6	ug/mg	38 - 83
Gut Marker (p	potential gut putrefaction or dys	biosis if high) - (Urine)			
	Indican	High end of range	126.4	ug/mg	0 - 131
	N	euro-related Markers	;		
Dopamine Me	etabolite - (Urine)				
	Homovanillate (HVA)	Above range	16.3	ug/mg	4 - 16
Norepinephri	ne/Epinephrine Metabolite - (Uri	ne)			
	Vanilmandelate (VMA)	Within range	5.7	ug/mg	2.5 - 7.5
Neuroinflamn	nation Marker - (Urine)				
	Quinolinate	Within range	10.2	ug/mg	0 - 12.5
		Additional Markers			
Melatonin (*n	neasured as 6-OH-Melatonin-Sເ	ulfate) - (Urine)			
	Melatonin* (Waking)	Low end of range	23.9	ng/mg	10 - 85
Oxidative Str	ess / DNA Damage, measured a	as 8-Hydroxy-2-deoxygu	anosine (8	-OHdG) -	(Urine)
	8-OHdG (Waking)	Within range	6.0	ng/mg	0 - 8.8



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Clinical Support Overview

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How to read the DUTCH report

This report is not intended to treat, cure or diagnose any specific diseases.

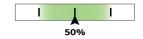
DUTCH Dials

The graphic dutch dials in this report are intended for quick and easy evaluation of which hormones are out of range. Results below the left star are shaded yellow and are below range (left). Results between the stars and shaded green are within the reference range (middle). Results beyond the second star and shaded red are above the reference range (right). Some of these hormones also change with age, and the age-dependent ranges provided should also be considered.



DUTCH Slider Bars

In a few places on the graphic pages, you will see slider bars. For adrenal hormones, you will see one to represent the balance between cortisol and cortisone metabolites. These bars indicate the relative ratio of the metabolites noted. The percentage stated is a population percentage, and so a result of 50%, as in this example (with the slider arrow in the middle of the bar) indicates that the ratio is higher than 50% of individuals tested, or right in the middle of the population's range. If the ratio between the metabolites is "low", the arrow will slide to the left and represent a smaller percentage and similarly to the right if the ratio is higher than normal.



Patient or Sample Comments

Throughout the provider comments you may find some comments specific to your situation or results. These comments will be found in this section or within another section as appropriate. Comments in other sections that are specific to your case will be in **bold**.

The patient reported symptoms of excess estrogen. This can be caused by excess estrogen or progesterone deficiency. Results should be carefully reviewed. We do not report a progesterone to estrogen ratio. However, you can investigate this issue by looking at the relative level of these two hormones on their respective dials.

Androgen Metabolism

• Androgen Metabolites: DHEA

DHEA and androstenedione are made almost exclusively by the adrenal gland (although a smaller amount is made in the testes). These hormones appear in urine as DHEA-S (DHEA-Sulfate), androsterone and etiocholanolone.

DHEA peaks for men in their 20's with a slow decline expected with age. DHEA mainly circulates throughout the body as DHEA-s, with interconversion to active DHEA as it reaches various tissues. DHEA is a weak androgen and will predominately convert to androstenedione, which will then convert to testosterone or aromatize to estrone. DHEA-s is made by sulfation, has a much longer half-life than DHEA and lacks a diurnal rhythm, which is why it is considered the best way to assess DHEA levels in the body. DHEA-s levels can be affected both by the total production as well as by the body's ability to sulfate DHEA.

The best way to assess the total production of DHEA is to add up these three metabolites. As DHEA production decreases quite significantly with age, we provide the age-dependent ranges.

The Total DHEA Production (page 1) was about 1,122ng/mg which is within the overall range but

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is below the range for the patient's age-dependent range. This implies that the adrenal glands are not producing appropriate DHEA levels for the patient's age. Low DHEA is associated with depression, diabetes, heart disease, inflammation and immune disorders. It can be caused by hypothyroidism. It can cause fatique, low mood and low libido. Supplementing DHEA often raises both testosterone and estrogen, which may or may not be desirable here. DHEA may increase with adaptogens such as maca and rhodiola, which improve overall adrenal output.

• Androgen Metabolites: Testosterone

The DUTCH test measures the total of testosterone glucuronide and testosterone sulfate. These conjugates of testosterone are formed mostly from bioavailable testosterone that undergoes phase 2 metabolism to make it ready for urine excretion.

Testosterone glucuronide is mostly made by the UGT2B17 enzyme, which also makes the glucuronide forms of 5a-DHT and 5b-androstanediol. Genetic variants of this enzyme reduce the urinary levels of these hormones without affecting serum levels. The genetic variants of UGT2B17 vary in the population from 7-80% (variation dependent on genetic ancestry, with the highest rates in those of Asian descent). Heterozygous individuals show milder reductions in urinary testosterone than homozygous. For this reason, low and very low levels of urinary testosterone should be confirmed with serum testing before treatment is applied. Serum testing can include free and total testosterone and SHBG.

The testes make most of the male's testosterone. Levels tend to be their highest at around 20 years of age and start to decline when men get into their 30's. Levels continue to drop as men age. Testosterone is needed for building bones and muscle mass, regulating body fat distribution and in the production of sperm and red blood cells. Testosterone is also important for libido and downstream production of modest amounts of estrogen.

Age dependent ranges are provided for all androgens as some decline is seen with age. Testosterone levels in healthy men vary widely so it is suggested that these ranges be interpreted with caution and consideration of symptoms. In addition, because estrogen also supports libido, erections and healthy weight management, estrogen levels should be considered along with the testosterone levels when assessing symptoms.

The testosterone result 10.3ng/mg is below the range for the patient's age. Review the levels of all androgens, androgenic metabolism, and patient symptoms for a complete assessment. As stated above, some patients have a genetic variant that causes low urinary testosterone, when serum levels are normal. Consider testing serum before initiating treatment.

• Andogen Metabolites: 5a-reductase versus 5b-reductase

5a-reductase converts testosterone into 5a-DHT (DHT), which is even more potent (\sim 3x) than testosterone. High levels of DHT can lead to symptoms associated with too much testosterone (thinning scalp hair, acne, etc.) and may also be associated with prostate issues in older men. However, 5aDHT plays an integral role in supporting bone, muscle and connective tissue integrity and improving brain health through the upregulation of dopamine, which can improve mood and libido.

Metabolites created down the 5b-pathway are significantly less androgenic than their 5a counterparts.

The slider bars below the hormones show the 5a or 5b preference based on the balance between etiocholanolone (5b) and androsterone (5a) as well as 5a-androstanediol and 5b-androstanediol. The slider shows the relative ratio of 5a to 5b products but does not express the absolute value of DHT or if 5a-reductase inhibition is or is not indicated. Consider symptoms and look at the total androgen levels if high androgen symptoms are a concern.

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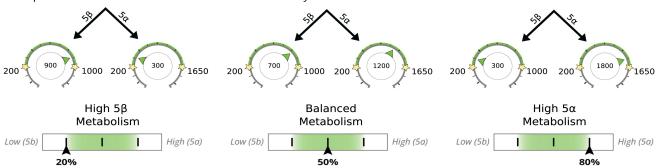
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Example of how to read sliders for 5a-reductase activity:



You will also see levels of epi-testosterone, which is not androgenic like testosterone. It happens to be produced in about the same concentrations as testosterone (this is an approximate relationship). This can be helpful when assessing the validity of urinary testosterone testing in an individual patient. If epi-testosterone is much higher than testosterone, serum testosterone assessment should considered before initiated therapy for low testosterone. Epi-testosterone is suppressed when exogenous testosterone is given, which can serve as a proxy for assessing endogenous testosterone production which can be obscured by the exogenous hormone administration.

Estrogen Metabolism

Over the past few decades research has clarified the importance of healthy estrogen levels and a balanced estrogen to testosterone ratio in men. The testes produce approximately 20% of E2 (Rochira) and the remaining 80% is aromatized from androgens in adipose (fat) tissue, muscle, breast, brain, liver and bone (Rochira). Thus, most of the estrogen in men is aromatized from testosterone, androstenedione, and DHEA in the periphery. The three estrogens (in order of strongest to weakest) are: Estradiol (E2), Estrone (E1) and Estriol (E3). E1 and E2 can interconvert and E3 is a waste product of estradiol and is the weakest of the three estrogens.

When evaluating estrogen levels, it is important to assess the following:

• The status (low, normal or high?) of estrogen production:

Levels of the primary estrogen, estradiol (the strongest estrogen), as well as "total estrogens" may be considered.

• Phase I Metabolism:

Estrogen is metabolized (primarily by the liver) down three phase I pathways. The 2-OH pathway is considered the safest because of the anti-cancer properties of 2-OH metabolites. Conversely, the 4-OH pathway is considered the most genotoxic as its metabolites can create reactive products that damage DNA. The third pathway, 16-OH creates the most estrogenic of the metabolites (although still considerably less estrogenic than estradiol) - 16-OH-E1.

When evaluating phase I metabolism, it may be important to look at the ratios of the three metabolites to see which pathways are preferred relative to one another. It may also be important to compare these metabolites to the levels of the parent hormones (E1, E2). If the ratios of the three metabolites are favorable but overall levels of metabolites are much lower than E1 and E2, this may imply sluggish phase I clearance of estrogens, which can contribute to high levels of E1 and E2.

The pie chart will assist you in comparing the three pathway options of phase I metabolism compared to what is "normal." 2-OH metabolism can be increased by using products containing D.I.M. or I-3-C. These compounds are found (or created from) in cruciferous vegetables and are known for promoting this pathway.

• Methylation (part of Phase II Metabolism) of estrogens:

After phase I metabolism, both 4-OH and 2-OH (not 16-OH) estrogens can be deactivated and eliminated by methylation. The methylation-activity index shows the patient's ratio of 2-Methoxy-E1 / 2-OH-E1 compared to what is expected. Low methylation can be caused by low levels of nutrients needed for methylation and/or genetic abnormalities (COMT, MTHFR). The COMT enzyme responsible for methylation requires magnesium and methyl donors. Deficiencies in folate or vitamin B6 or B12 can cause low levels of methyl donors. MTHFR genetic defects can make it more difficult for patients to make sufficient methyl donors. Genetic defects in COMT can make methylation poor even in the presence of adequate methyl donors.

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Progesterone Metabolism

Male progesterone is synthesized in the testes and, to a lesser degree, in the adrenal glands. It's role in men's health is not well understood, although progesterone is known to be involved in sperm activation. In healthy men, progesterone is positively correlated to markers of inflammation.

Metabolites of progesterone are measured in urine, including 5b-pregnanediol and 5a-pregnanediol. 5bpregnanediol is inactive in the body but is the major metabolite of progesterone. 5a-pregnanediol is often a metabolite of more interest, as it can cross the blood brain barrier and up-regulate GABA activity and is considered neuroprotective to the brain. Both taken together represent the major metabolic end points for progesterone and can be used to represent total progesterone production.

The weighted average of the two progesterone metabolites shows progesterone is in range indicating normal production.

DUTCH Adrenal

The HPA-Axis refers to the communication and interaction between the hypothalamus (H) and pituitary (P) in the brain down to the adrenal glands (A) that sit on top of your kidneys. When cortisol is needed in the body, the hypothalamus releases cortisol releasing hormone (CRH) and the pituitary responds by releasing adrenocorticotropic releasing hormone (ACTH), which is the signal to the adrenal gland to release cortisol, DHEA and DHEA-s. It is these adrenal hormones that are assessed on the DUTCH test to understand the patient's HPA axis.

The cortisol awakening response is a complex interaction between the HPA axis and the hippocampus, where ACTH normally surges right after waking leading to the day's highest levels of cortisol. This signal is considered by researchers to be separate from the regular circadian rhythm (the smooth transition from lower cortisol at night to modestly higher cortisol in the morning) and to reflect the person's anticipation of stress during the day, some psychosocial factors such as depression or anxiety and their metabolic state. The waking surge in cortisol helps with energy, focus, morning blood sugar and immune regulation.

As the day progresses, ACTH declines and subsequent cortisol decreases throughout the day, so it is low at night for sleep. This cycle starts over the next morning.

Free cortisol provides negative feedback to CRH & ACTH. When free cortisol is too low, ACTH will surge. ACTH will

also surge when a physical or psychological stressor occurs.

Only a small fraction of cortisol is "free" and bioactive. The "free" cortisol is what the person feels in terms of energy and focus. Free cortisol is also what feeds back to the hypothalamus and pituitary gland for ACTH and cortisol regulation. The free cortisol daily pattern is very useful for understanding cortisol and its interaction with the patient's symptoms throughout the day. However, because only a fraction of the cortisol is bioactive, when considering treatments that affect the whole HPA axis, including DHEA, it is essential to measure metabolized cortisol to get a bigger picture.

In urine, we can measure both the total metabolized cortisol (THF) and total metabolized cortisone (THE) excreted throughout the day. These two components better represent the total cortisol production from the adrenal glands than the free cortisol alone. Outside of the HPA axis, metabolism of cortisol occurs with the help of thyroid hormone in the liver. A significant amount of cortisol is also metabolized in adipose tissue.

To best determine total adrenal production of cortisol throughout the day it is important to assess both metabolized cortisol and free cortisol.

When evaluating cortisol levels, it is important to assess the following:

- The daily pattern of free cortisol throughout the day, looking for low and high levels: Abnormal results should be considered along with related symptoms. Remember that with urine results, the "waking" sample reflects the night's total for free cortisol. The sample collected two hours after waking captures the cortisol awakening response, which is typically the time with the most cortisol secretion.
- The sum of the free cortisol as an expression of the overall tissue cortisol exposure: This total of four free cortisol measurements is the best way to assess the total of free cortisol throughout the day, and this result correlates reasonably well to a true 24-hour urine free cortisol. Do be aware that this measurement does not consider transitory shifts in cortisol in the late morning or early afternoon. This number is calculated from the simple addition of the 4 points, so if a single point is very high or very low, it may skew the number up or down especially if it is the morning "B" point, as it is weighted more heavily in the reference range.
- The total level of cortisol metabolites:

We call this calculation "Metabolized Cortisol" which is the sum of a-THF, b-THF and b-THE (the most abundant

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cortisol metabolites). While free cortisol is the best assessment for tissue levels of cortisol, it only represents 1-3% of the total produced. The total metabolized cortisol best represents the total glandular output of cortisol for the day, closer to 80% of the total produced.

Overall cortisol levels are appropriate as both free and metabolized cortisol levels are within range. If the diurnal pattern of the free cortisol is as expected, this implies normal HPA-Axis cortisol production.

A potential preference for cortisol or cortisone (the inactive form):

To determine total systemic preference for cortisol or cortisone, it is best to look at which metabolite predominates (THF or THE?). This preference can be seen in the slider bar. This is known as the 11b-HSD index. The enzyme 11b-HSD II converts cortisol to cortisone in the kidneys, saliva gland and colon. 11b-HSD I is more active in the liver, fat cells and the periphery and is responsible for reactivating cortisone to cortisol. Both are then metabolized by 5a-reductase to become tetrahydrocortisol (THF) and tetrahydrocortisone (THE) respectively. We can see more cortisol or cortisone in different metabolic conditions. For example, a preference for cortisol indicates possible inflammation, insulin resistence or hypothyroidism. A preference for cortisone can indicate chronic stress or chronic infection (such as the later stages of a virus or common cold).

Nutritional Organic Acids

Organic acids are the metabolic byproducts of cellular activity in the body. Organic acid production varies by the individual and can be influenced by foods, environmental toxins, medications or supplements, nutrient status, genetics and more. Organic acids begin to build up when a nutrient cofactor or mineral is not present for a specific reaction to occur. As a response, byproducts (organic acids) build up and can be measured in urine. On the DUTCH test, the organic acids we measure were chosen due to their specific roles in the metabolism and function of enzymes required for hormone and adrenal health and function. As industry standard dictates, the organic acids are measured from the waking sample.

Methylmalonate (MMA)

Methylmalonic acid is a metabolic byproduct of the Citric Acid Cycle (Krebs cycle). Methylmalonic acid requires adenosylcobalamin for conversion to succinyl-CoA and onto ATP synthesis. If someone does not absorb enough B12 from their diet due to low B12-rich food consumption, low stomach acid, has an autoimmune disorder impacting Intrinsic Factor in the gut (required for B12 absorption), or has an MUT enzyme SNP (required for conversion of MMA to Succinyl coA, dependent on adenosylcobalamin) then MMA will build up. Vitamin B12 is required for COMT activity (estrogen methylation, dopamine breakdown) and PNMT activity (the enzyme that takes norepinephrine to epinephrine), but is also critical for memory, energy production (ATP synthesis), gait and more. When MMA is high, consider supporting B12 through foods, digestive support or supplementation.

Xanthurenate & Kynurenate

Xanthurenate and kynurenate are metabolic byproducts in the production of tryptophan to NAD in the liver. If either xanthurenate or kynurenate build up in the urine, it can indicate a need for vitamin B6. This need is amplified if BOTH markers are elevated, and often indicates a more severe deficiency of vitamin B6. Vitamin B6 is critical as a co-factor to over 100 important reactions that occur in the human body and is stored in the highest concentration in muscle tissue.

Tryptophan is converted to NAD by the liver and one of the steps in this pathway requires B6. When B6 is insufficient, xanthurenate is made instead. Xanthurenate can also bind to iron and create a complex that increases DNA oxidative damage resulting in higher 8-OHdG levels. If both the xanthurenate and 80hdG levels are elevated, there is likely an antioxidant insufficiency.

Kynurenate may also become elevated when patients are B6 deficient because of a different, possibly less B6 dependent pathway. While there is always some tryptophan going down the kynurenine pathway towards NAD, and possibly xanthurenate, this process is up regulated by inflammation, estrogen and cortisol elevations. If levels of estrogen or cortisol are high, it may exacerbate kynurenic acid and increase the need for vitamin B6. As the Xanthurenate and Kynurenate pathways lead to biomarkers with other influence in the body, elevations in these markers may not always agree.

b-Hydroxyisovalerate

b-Hydroxyisovalerate is made when the body is deficient in biotin. Elevated levels may indicate biotin deficiency. Biotin is an important cofactor in mitochondrial function, metabolism of fatty acids, glucose, and protein, and ROS production. Biotin deficiency has similar symptoms as other B-vitamin deficiencies but is most often associated with hair loss. Factors influencing biotin levels include inadequate dietary intake, long-term and highdose B5 supplementation, dysbiosis/gut health, antibiotic use, medications, and biotinidase deficiency. Note: If

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TEST NUMBER: G-NL-XXXXX GENDER: XXXXXX AGE: XX

COLLECTED: 00-XXX-2024 00-XXX-2024 RECEIVED: 00-XXX-2024 TESTED:

TEST REF: GNL-NL-XXXXX XXXXXXXXXXX

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TEST NAME: DUTCH Complete™

beta-hydroxy-beta-methylbutyrate (HMB), an amino acid conjugate supplement, is taken within 72 hours of testing, we can see high levels of b-hydroxyisovalerate without indicating a biotin deficiency. Please check with supplements when interpreting results.

Pyroglutamate

Pyroglutamate is an intermediate in glutathione recycling and production. Glutathione requires the amino acids cysteine, glycine and glutamate for production. If the body cannot convert pyroglutamate forward to glutathione, it will show up elevated in the urine. High pyroglutamate is an established marker for glutathione deficiency. Remember that glutathione is one of the most potent antioxidants in the human body and is especially important in getting rid of toxins including the reactive guinone species formed by 4-OH-E1 and 4-OH-E2. This reactive species can damage DNA if not detoxified by either methylation or glutathione. Some have reported that low pyroglutamate may also be indicative of a need for glutathione; however, this is not established in the scientific literature.

Note: Pyroglutamate in the urine can also be elevated with Italian cheese consumption. Italian Cheeses (parmesan, etc.) may transiently increase pyroglutamate because they use a thermophilic lactobacilli to ripen the cheese- which our gut breaks down into pyroglutamate. This is not clinically significant and only reflects that they ate this style of cheese (if applicable).

Indican

Indican is a byproduct of tryptophan putrefaction by microbes in the gut. Accumulated levels of indican in the urine suggest higher levels of tryptophan putrefaction from gastrointestinal dysbiosis or malabsorption. Production of indican occurs when tryptophan creates indoles in the colon. No other endogenous indoles are metabolized in this way, so when we see indican in the urine, it is directly related to gut production and a direct reflection of gut health. Chronic dysbiosis can impact sex hormone metabolism, cause inflammation, and influence cortisol levels and metabolism. High urinary indican suggests further testing to rule out gut dysbiosis.

Vegetarian and vegan diets have less protein, therefore elevated levels with these diets are likely stronger suggestions of gut dysbiosis. The amount of indican present does not correlate to the degree of dysbiosis but merely shows that dysbiosis is present. Common causes of high indican include malabsorption of protein because of low stomach acid, poor pancreatic function, Celiac disease, the overgrowth of anaerobic bacteria in the colon, small intestinal bacterial overgrowth (SIBO), medications that reduce protein absorption (like proton pump inhibitors or other antacids or H2 blockers), and constipation. Urinary indican can increase with recent (<72 hours) tryptophan supplementation without indicating dysbiosis. Please keep supplements in mind when interpreting the result.

Neuro-related Markers

Neurotransmitters are chemical signals produced by neurons in tissues throughout the body that act as chemical messengers that influence mood, cortisol, heart rate, appetite, muscle contraction, sleep and more. Measuring neurotransmitters directly is difficult because of their instability, and their direct urinary measurements are controversial with respect to how well they reflect the body's level of these neuro-hormones.

Each of the neurotransmitters assessed on the DUTCH test (dopamine, norepinephrine/epinephrine) can be assessed indirectly by measuring their urine metabolites (HVA and VMA respectively). While these metabolites are not a perfect reflection of what is going on in the brain, the scientific literature does affirm their use for a good representation of overall levels of these neurotransmitters in the body.

Homovanillate (HVA)

Homovanillate (HVA) is the primary metabolite of dopamine, a brain and adrenal neurotransmitter that comes from tyrosine (with BH4 and iron as co-factors). Dopamine goes on to create norepinephrine and epinephrine (adrenaline).

Low levels of dopamine are associated with depression, addictions, cravings, apathy, pleasure seeking behaviors, increased sleepiness, impulsivity, tremors, low motivation fatigue and low mood. High levels of dopamine are associated with agitation, insomnia, mania, hyperactivity, hyper-focus, high stress, anxiety and addictions/cravings/pleasure seeking (to maintain high levels).

High HVA can be caused by the use of the following supplements, foods or medications within 72 hours of collecting urine samples: tyrosine, phenylalanine, mucuna, quercetin, bananas, avocados as well as parkinson's medications. If these are being used, the HVA on the DUTCH test may not accurately reflect circulating dopamine levels and should be disregarded.

Vanilmandelate (VMA)

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TEST NAME: DUTCH Complete™

Vanilmandelate (VMA) is the primary metabolite of norepinephrine and epinephrine (adrenaline). The adrenal gland makes cortisol and DHEA (from the adrenal cortex) as well as norepinephrine and epinephrine (from the adrenal medulla). When adrenal hormone output is low, VMA levels may be low. If HVA levels are significantly higher than VMA, there may be a conversion problem from dopamine to norepinephrine. This case can be caused by a copper or vitamin C deficiency.

The enzymes COMT (methylation of catechols) and MAO are needed to make HVA and VMA from dopamine and norepinephrine respectively. If these enzymes are not working properly, HVA and/or VMA may be low in urine, when circulating levels of dopamine and/or norepinephrine/epinephrine may not be low.

Low levels of norepinephrine/epinephrine are associated with addictions, cravings, fatigue, low blood pressure, low muscle tone, intolerance to exercise, depression, and loss of alertness.

High levels of norepinephrine and epinephrine are associated with feelings of stress, aggression, violence, impatience, anxiety, panic, excess worry/hypervigilance, insomnia, paranoia, increasing tingling/burning, loss of memory, pain sensitivity, high blood pressure and heart palpitations.

Quinolinate (QA)

The quinolinate is high. Quinolinate is a neurotoxin derived from tryptophan metabolism in the context of high inflammation or high cortisol. Elevated quinolinate has been seen in brain and nerve tissue damage, especially in disorders such as Alzheimer's disease, Parkinson's disease, Huntington's disease, motor neuron diseases, multiple sclerosis, epilepsy, amyotrophic lateral sclerosis, and major depressive disorder. We can also see elevated quinolinate due to low serotonin and need for vitamin B3 (niacin). If tryptophan supplements are taken within 72 hours of collecting DUTCH samples, there may be high levels of quinolinate in the urine which may not be associated with neuroinflammation. Keep supplements in mind when interpreting results.

Melatonin (measured as 6-OHMS)

Melatonin is considered one of our sleep hormones. It is made predominately by the pineal gland in response to darkness and is stimulated by melanocyte stimulating hormone (MSH). A low MSH is associated with insomnia and an increased perception of pain. Mold exposure can inhibit MSH as well. The majority of our melatonin production comes from the pineal gland, but melatonin is also made in the gut, and to a lesser extent in the bone marrow, lymphocytes, epithelial cells and mast cells.

Please note that some foods contain small amounts of melatonin that are unlikely to increase circulating levels of melatonin, but may increase metabolites in urine due to first pass metabolism. The most significant of these foods are tomatoes, walnuts, strawberries and caffeinated coffee. These foods are thought to contribute to mildly elevated urinary melatonin. Extremely high urinary melatonin is seen when melatonin is supplemented directly. This is also due to first pass metabolism and is not an accurate reflection of circulating melatonin.

The DUTCH test uses the waking (A) sample to test melatonin. The urine sample given on waking reflects overnight hormone production and metabolism. This sample can be used to assess melatonin throughout the night. When patients take a middle of the night urine sample, a large amount of data strongly suggests that the waking sample alone still correlates best to overnight melatonin production, so the waking sample is still used for the DUTCH melatonin result.

8-OHdG (8-Hydroxy-2-deoxyguanosine)

8-OHdG (8-Hydroxy-2-deoxyguanosine) is a marker for estimating DNA damage due to oxidative stress (from ROS creation). 8-OHdG is considered pro-mutagenic and is a biomarker for various cancer and degenerative disease initiation and promotion states. It can be increased by chronic inflammation, increased cell turnover, chronic stress, hypertension, hyperglycemia/pre-diabetes/diabetes, kidney disease, IBD, chronic skin conditions (psoriasis/eczema), depression, atherosclerosis, chronic liver disease, Parkinson's (increasing levels with worsening stages), Diabetic neuropathy, COPD, bladder cancer, or insomnia (to name a few). Studies have shown higher levels in patients with breast and prostate cancers. When levels are elevated it may be prudent to eliminate or reduce any causes and increase the consumption of antioxidant containing foods and/or supplements.

Urine Hormone Testing - General Information

What is actually measured in urine? In blood, most hormones are bound to binding proteins. A small fraction of the total hormone levels are "free" and unbound such that they are active hormones. These free hormones are not found readily in urine except for cortisol and cortisone (because they are much more water soluble than, for example, testosterone). As such, free cortisol and cortisone can be measured in urine and it is this measurement that nearly all urinary cortisol research is based upon. In the DUTCH Adrenal Profile the diurnal patterns of free cortisol and cortisone are measured by LC-MS/MS.

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TESTED: 00-XXX-2024

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TEST NAME: DUTCH Complete™

All other hormones measured (cortisol metabolites, DHEA, and all sex hormones) are excreted in urine predominately after the addition of a glucuronide or sulfate group (to increase water solubility for excretion). As an example, Tajic (Natural Sciences, 1968 publication) found that of the testosterone found in urine, 57-80% was testosterone-glucuronide, 14-42% was testosterone-sulfate, and negligible amounts (<1% for most) was free testosterone. The most likely source of free sex hormones in urine is from contamination from hormonal supplements. To eliminate this potential, we remove free hormones from conjugates. The glucuronides and sulfates are then broken off of the parent hormones, and the measurement is made. These measurements reflect the bioavailable amount of hormone in most cases as it is only the free, nonprotein-bound fraction in blood/tissue that is available for phase II metabolism (glucuronidation and sulfation) and subsequent urine excretion.

Disclaimer: the filter paper used for sample collection is designed for blood collection, so it is technically considered "research only" for urine collection. Its proper use for urine collection has been thoroughly validated.

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TEST NAME: DUTCH Complete™

Reference Range Determination (last updated 5.1.2024)

We aim to make the reference ranges for our DUTCH tests as clinically appropriate and useful as possible. This includes the testing of thousands of healthy individuals and combing through the data to exclude those that are not considered "healthy" or "normal" with respect to a particular hormone. As an example, we only use a premenopausal woman's data for estrogen range determination if the associated progesterone result is within the luteal range (days 19-21 when progesterone should be at its peak). We exclude women on birth control or with any conditions that may be related to estrogen production. Over time the database of results for reference ranges has grown quite large. This has allowed us to refine some of the ranges to optimize for clinical utility. The manner in which a metabolite's range is determined can be different depending on the nature of the metabolite. For example, it would not make clinical sense to tell a patient they are deficient in the carcinogenic estrogen metabolite, 4-OH-E1 therefore the lower range limit for this metabolite is set to zero for both men and women. Modestly elevated testosterone is associated with unwanted symptoms in women more so than in men, so the high range limit is set at the 80th percentile in women and the 90th percentile for men. Note: the 90th percentile is defined as a result higher than 90% (9 out of 10) of a healthy population.

Classic reference ranges for disease determination are usually calculated by determining the average value and adding and subtracting two standard deviations from the average, which defines 95% of the population as being "normal." When testing cortisol, for example, these types of two standard deviation ranges are effective for determining if a patient might have Addison's (very low cortisol) or Cushing's (very high cortisol) Disease. Our ranges are set more tightly to be optimally used for Functional Medicine practices.

Below you will find a description of the range for each test:

			Male Refe		nges (Updated 05.1.2024)				
	Low%	High%	Low	High		Low%	High%	Low	High
b-Pregnanediol	10%	90%	75	400	Cortisol A (waking)	20%	90%	13	80
a-Pregnanediol	10%	90%	20	130	Cortisol B (morning)	20%	90%	35	180
Estrone (E1)	10%	90%	4	16	Cortisol C (~5pm)	20%	90%	10	45
Estradiol (E2)	10%	90%	0.5	2.2	Cortisol D (bed)	0	90%	0	20
Estriol (E3)	10%	90%	2	8	Cortisone A (waking)	20%	90%	40	160
2-OH-E1	0	90%	0	5.9	Cortisone B (morning)	20%	90%	80	240
4-OH-E1	0	90%	0	0.8	Cortisone C (~5pm)	20%	90%	40	130
16-OH-E1	0	90%	0	1.2	Cortisone D (bed)	0	90%	0	70
2-Methoxy-E1	0	90%	0	2.8	Melatonin (6-OHMS)	20%	90%	10	85
2-OH-E2	0	90%	0	0.6	8-OHdG	0	90%	0	8.8
4-OH-E2	0	90%	0	0.3	Methylmalonate	0	90%	0	3.5
2-16-ratio	20%	80%	2.85	9.88	Xanthurenate	0	90%	0.2	1.9
2-4-ratio	20%	80%	6.44	12.6	Kynurenate	0	90%	1	6.6
2Me-2OH-ratio	20%	80%	0.4	0.7	b-Hydroxyisovalerate	0	90%	0	18
DHEA-S	20%	90%	30	1500	Pyroglutamate	10%	90%	38	83
Androsterone	20%	80%	500	3000	Indican	0	90%	0	131
Etiocholanolone	20%	80%	400	1500	Homovanillate	10%	95%	4	16
Testosterone	20%	90%	25	115	Vanilmandelate	10%	95%	2.5	7.5
5a-DHT	20%	90%	5	25	Quinolinate	0	90%	0	12.5
5a-Androstanediol	20%	90%	30	250					
5b-Androstanediol	20%	90%	40	250	Calculated Values				
Epi-Testosterone	20%	90%	25	115	Total DHEA Production	20%	80%	1000	5500
a-THF	20%	90%	175	700	Total Estrogens	10%	90%	10	34
b-THF	20%	90%	1750	4000	Metabolized Cortisol	20%	90%	4550	1000
b-THE	20%	90%	2350	5800	24hr Free Cortisol	20%	90%	75	300
					24hr Free Cortisone	20%	90%	220	550

e population percentile: Example - a nigh limit of 90% means results higher than 90% of the women tested for the reference range will be designated as "high."

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TEST NUMBER: G-NL-XXXXX
GENDER: XXXXXX
AGE: XX

COLLECTED: 00-XXX-2024

RECEIVED: 00-XXX-2024

00-XXX-2024

TESTED:

TEST REF: GNL-NL-XXXXX

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TEST NAME: DUTCH Complete™

T-NL-1433839 Name: Mario Ttakoushis DOB: 03-Mar-1966 Sex: Male DOC: 17-Sep-2024 Test: DUTCH Complete™		Pati	Urinary Hormone ent Information Question	
PATIENT INFORMATION PATIENT NAME:		akoushis		
DATE OF BIRTH (DD/MM/YY): 031031266	HEIGHT (metres):	WEIGHT (kgs):	GENDER:	
MENSTRUAL CYCLES:	OVARIES F	REMOVED: FIRST DATE O	F LAST MENSES (DD/MM/YY): PREGNANT: YES NO	
HORMONE, SUPPLEI			Pregnenolone, or cortisol (cortef, hydrocortisone	e, etc)
you are taking. "Last used" should For Route of Administration (F	be the last time you took the h	ormone before finishing the tes g: 1=oral, 2=sublingual (unde	ts. r the tongue, between cheek/gum) 3=trans	
(skin) cream, 4= transderm HORMONE BRAND			=patch, 8=pellet, 9=injection, 10=other T USED TIMES PER DAY LENGTH OF US	SE
SEX HORMONE/ METABOLISM SUPPORT:	O DIM/I-3-C OGlutathione	Saw Palmetto B-12/Methyl B-12	Aromatase Inhibitor (Chrysin, Arimid Please specify:	ex, etc);
WETABOLISM SOTT OTT.	O Calcium-d-glucura	te 7-Keto-DHEA		
	O Phosphatidylserine O St. John's Wort,	 Adaptogenic herbs (Sib Ginseng, Panax Ginseng, Rhodiola, Ashwaghanda, 	 Melatonin Anxiety/Depression media Please specify: 	cation;
ADRENAL & BRAIN SUPPORT:	C Licorice Root,	Astragalus) O High Dose Vitamin C	Adrenal Support Product;	
	○ 5-HTP,	(>1000mgday),	Please specify:	
BIRTH CONTROL:	○ SAM-e, ○ Yes ○ No; If yes	please specify:		
LIST ANY OTHER RELEVANT MEDICINES, SUPPLEMENTS OR O	THERS Ramipril	Small Rosuvi	astatin long //Levored Immuno = 17	thegra
	Chemo = 1	from Pemetiex	ed Immuno = pembrolizu	mail
SAMPLE COLLECTION	ON DATE & TIME			
SAMPLE 1 DATE: TIME:	SAMPLE 2 DATE:	TIME:	SAMPLE 3 DATE: TIME:	
	OO AM 16 /09 /2) O PM
SPM O Bed Overnight O	Morning (2hr) 5PM	Bed Overnight O Morning (2h	r) SPM Bed Overnight Mornin	
In the hours preceding the sample were you u		ading the sample were you under stress? Mild Moderate Severe	In the hours preceding the sample were you under str	Name and Address of the Owner, where
SAMPLE 4	SAMPLE 5	;	DID YOU URINATE OVERNIGHT WITHOUT COLLECTING A SAMPLE?	т
DATE: TIME: 17/09/24 07:	40 OPM 17/09/2	TIME: 9:45 %		
SAMPLE TYPE: 5PM Bed Overnight	SAMPLE TYPE:	Bed Overnight Morning (2h	ADDITIONAL COMMENTS:	
In the hours preceding the sample were you u	THE RESIDENCE AND ADDRESS OF THE PARTY OF TH	eding the sample were you under stress? Mild Moderate Severe		+

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AGE: XX

COLLECTED: 00-XXX-2024
RECEIVED: 00-XXX-2024
TESTED: 00-XXX-2024

TEST REF: GNL-NL-XXXXX

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XXXXXXXXXXXXXXXXXXXX

TEST NAME: DUTCH Complete™

SYMPTOMAT	OLOGY						
DISEASE STATES		I DO NOT SUSPE HAVE THIS	CT I I SUSPECT HAVE TH	I MAY I HAVE	E BEEN D WITH THIS	Please list ar	ny current/recent Inosis NOT listed
Addison's Disease		~/	O			elsewhere or	this form:
Adrenal Fatigue			0				
Chronic Fatigue		8	0				
High Blood Pressur	e	×	0				
Hyperthyroidism (O	veractive)		0				
Hypothyroidism (Ur	deractive)	XXXXX	0	3	×		
Kidney Disease		×	0				
Polycystic Ovary Sy	ndrome	\sim	0	(
Please rate your fa	tigue level <u>d</u>	uring the day					
FATIGUE		NEVER/NONE	SOMETIMES/MII	_D ALWAYS/	SEVERE		
Morning Fatigue			*				
Afternoon Fatigue		0					
Evening Fatigue		0	×				
Please circle sympt	oms you are	e experiencing and rate the	e overall category		1	1	
		Increased say drive box	dy or facial bair agar	NEVER/NONE	SOMETIME/MILD	OFTEN/MODERATE	ALWAYS/SEVERE
ANDROGEN	MEN	Increased sex drive, boo behaviour, acne	dy of facial flall, aggi	essive		0	0
EXCESS	-WOMEN-	Loss of scalp hair, increa	ased body or facial h	air, acne	0	0	0
	MEN	Decreased libido, erecti		0	(6)	0	0
ANDROGEN DEFICIENCY		increased belly fat, apat				_	
	WOMEN	Vaginal dryness, decrea	sed sex drive	0	0	0	0
ESTROGEN	MEN	Weight gain (breasts or	hips), prostate probl	ems	0		0
EXCESS	-WOMEN	Tender or fibrocystic bre fibroids, heavy bleeding		uterine			0
ESTROGEN							
DEFICIENCY	WOMEN	Hot flashes; night sweat	s, vaginal dryness	0	0	0	0
ADDITIONAL SYMP		NEVER/NONE	SOMETIM	E/MILD OFT	EN/MODERATE	Marie Commission Commi	S/SEVERE
Trouble falling aslee		0	0				0
Trouble staying asle	еер	0		1			0
Depression		0		7	0		0
Anxiety Migraines		0	Q		(3)		0
Migraines							
	ORMA	TION IS REQ	UIRED FOF	R A FULL A	ND ACC	JRATE RE	EPORT
Head Office: Nygade 6, 3.sa 1164 Copenha Denmark +45 33 75 10 0	l gen K	UK Office: 11 Old Factory Bui Stonegate E. Sussex TN5 7DL +44 (0)1580 201 6	ildings J	Africa Office: 9 Blackpool Rd Bryanston 2191 South Africa +27 825 725 725			
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