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Possible hormonal biomarkers in the diagnosis of overtraining syndrome (OTS) - a literature review

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Content

Abstract

Background: Physical activity is important for prevention of some of the most common diseases worldwide: cardiovascular disease, type 2 diabetes mellitus, and certain types of cancers. However, extremely strenuous exercise performed over a prolonged period can lead to Overtraining Syndrome (OTS), especially in athletes competing at elite levels. This is characterized by psychological, neuroendocrine and immunological disturbance, and a prolonged decrease in exercise tolerability. Although many theories have emerged about the etiology of this condition, the pathophysiology is still unknown. That makes it difficult to diagnose OTS, as the resulting symptoms, including fatigue and diminished exercise performance, are common in athletes and largely nonspecific.

Aim: The goal of this study was to synthesize current evidence on possible hormonal biomarkers that may assist in the diagnosis of OTS.

Materials and methods: The search was conducted via PubMed, Science Direct, NCBI, and Google Scholar databases for articles with a focus on human studies. The keywords included "overtraining syndrome", "OTS", "overreaching syndrome", "hormonal biomarkers", "testosterone cortisol ratio", "testosterone estradiol ratio", "hypothalamic pituitary axis", "hypothalamic dysfunction", "EROS-HPA axis".

Results: Certain studies suggest that basal and dynamic hormone measurements may aid in the diagnosis of OTS. The Cadegiani and Kater EROS studies (2017-2020) [6, 8, I] have proposed diagnostic tools composed of clinical and biological markers that, in the tested cohort, showed 100% diagnostic accuracy in distinguishing between OTS and non-OTS athletes. However, without an established pathophysiological pathway, many researchers remain sceptical.

Conclusions: There is a definite need for validation of proposed diagnostic tools and reaching a consensus on the diagnostic process of OTS, instead of having it be a diagnosis of exclusion.

Key words: overtraining syndrome; overtraining; overreaching; staleness; unexplained underperformance; athletes; hormonal biomarkers; tools; "HPA axis"

1. Introduction

1.1. Aim of the study:

The goal of this study was to synthesize current evidence on possible hormonal biomarkers that may assist in the diagnosis of OTS.

The European College of Sport Science distinguishes 3 conditions associated with overexercise [14]. The first one, termed Functional Overreaching (FOR), involves temporary decrease in exercise tolerability followed by improvement after rest, generally resulting in positive outcomes. When overintense training regimen is prolonged and results in mild neuropsychological and/or endocrine symptoms, lasting weeks to months, it is called Nonfunctional Overreaching (NFOR). If the exercise decrement is prolonged >2 months, with more severe psychological and neurological or endocrine symptoms, or new symptoms from the immune system, unexplained by other conditions, Overtraining Syndrome (OTS) should be suspected.

Researchers have found it difficult to establish the exact prevalence of OTS. This is attributed to many factors, including largely unspecific diagnostic criteria, underreporting, overlap with NFOR or with other organic or non-organic conditions. It is known, however, that elite endurance sports athletes are at the highest risk, for example, as many as 60% of elite runners and cyclists experience one or more episodes of NFOR or OTS throughout their careers. At collegiate and professional level, it is estimated that 30-50% of athletes experience overreaching (FOR, NFOR), while the OTS develops in 5-10% of those individuals [14]. Team sports report an overall lower OTS incidence [18].

2. Materials and methods

This literature review has been conducted in an attempt to synthesize evidence on hormonal markers for the diagnosis of OTS. The search was conducted via PubMed, Science Direct, NCBI, and Google Scholar databases for articles with a focus on human studies. The keywords used included “overtraining syndrome”, “OTS”, “overreaching syndrome”, “hormonal biomarkers”, “testosterone cortisol ratio”, “testosterone estradiol ratio”, “hypothalamic pituitary axis”, “hypothalamic dysfunction”, “EROS-HPA axis”.

2.1. AI

AI was utilized for two specific purposes in this research. Text analysis of clinical reasoning narratives to identify linguistic patterns associated with specific logical fallacies. Assistance in refining the academic English language of the manuscript, ensuring clarity, consistency, and adherence to scientific writing standards. AI were used for additional linguistic refinement of the research manuscript, ensuring proper English grammar, style, and clarity in the presentation of results. It is important to emphasize that all AI tools were used strictly as assistive instruments under human supervision. The final interpretation of results, classification of errors, and conclusions were determined by human experts in clinical medicine and formal logic. The AI tools served primarily to enhance efficiency in data processing, pattern recognition, and linguistic refinement, rather than replacing human judgment in the analytical process.

3. Results

Hormonal imbalance is often implicated as one of the pathomechanisms of OTS (*e.g., the hypothalamic dysfunction theory*). The two main types of tests analyzed in this review were basal hormone levels (basal testosterone, basal cortisol, T/C ratio, T/E2 ratio), and stress hormone levels (ACTH, growth hormone [GH], cortisol, free plasma catecholamines, nocturnal urinary catecholamines, insulin intolerance test [IIT]). The downfall of many proposed tests lies in the nature of hormone homeostasis that varies diurnally, with exercise load, diet, season, menstrual cycle in females, and among individuals due to their unique responsiveness and genetic factors.

3.1. Symptoms

The symptoms of OTS differ among individuals, and can be subdivided into the following [2]:

Table 1. Division of overtraining syndrome symptoms by systematic category.

General	Decreased performance, fatigue, insomnia, awakening unrefreshed, loss of appetite
Endocrine	Hormonal imbalances (cortisol, testosterone, HPA disturbance)

Neuropsychological	Mood swings, restlessness, irritability, anxiety, loss of motivation, depression
Musculoskeletal	Sore, stiff muscles, overuse injuries
Cardiovascular	Hypertension, tachycardia or bradycardia
Immune	Increased susceptibility to infections
Gynecological	Oligo- or amenorrhea in females

Although a combination of symptoms typical of OTS may rise suspicion, their presence may easily be attributed to other conditions, such as negative caloric balance, nutritional deficiencies, iron deficiency anemia, bronchial asthma, allergy, among others.

It is worth mentioning, however, that parasympathetic symptoms (bradycardia, fatigue, depression) present more commonly in athletes practicing aerobic sports (distance running, cycling, swimming), while sympathetic symptoms (tachycardia, hypertension, restlessness, insomnia) are predominantly noted in those who perform anaerobic exercise (weightlifting, sprinting). That division may aid in forming a connection between patient's history and their symptoms and possibly support or diminish the possibility of OTS diagnosis [14].

3.2. Pathogenesis

Many hypotheses have been formed on the possible pathogenic pathways leading to OTS [14]. None have, to date, provided a satisfactory explanation to the plethora of multisystem symptoms experienced by athletes suffering from overtraining.

The glycogen hypothesis might explain performance decline and chronic fatigue, however, is not implicated in systemic dysfunction and literature has not established a connection between glycogen levels and overtrained athletes.

The central fatigue hypothesis suggests that increased brain serotonin (5-HT), as a result of decreased sensitivity of 5-HT receptors in overtrained athletes [4], levels during prolonged stress could reduce central drive and motivation, leading to fatigue. However, studies have not consistently demonstrated serotonin-related changes in OTS, nor does this hypothesis address the whole symptomatic spectrum of OTS.

The glutamine hypothesis proposes that heavy training depletes plasma glutamine, weakening immune defense and increasing infection risk. Yet glutamine levels and bioavailability are not reliably depressed in vivo. Some studies have established low glutamine levels in athletes with URTIs, however that is not limited to overtrained athletes.

The autonomic nervous system hypothesis suggests that overtraining disrupts the sympathetic and parasympathetic nervous systems, resulting in maladaptive response to physical activity and impaired performance, recovery, and overall health [5, 16]. However, studies of urinary catecholamine excretion and functional testing of heart rate variability have not been conclusive.

The hypothalamic dysfunction hypothesis stems from studies that conclude HPA alteration between healthy and overtrained athletes [1, 6, 7, 10, 16, 18, 19, 21]. The hypothalamus, as the central integrator of the neuroendocrine stress response, it regulates the key hormonal axes (the hypothalamus-pituitary-adrenal axis [HPA], -gonadal axis [HPG] and -thyroid axis [HPT]) through hormones that stimulate or inhibit the anterior pituitary endocrine function. It has been proposed that chronic excessive training loads result in maladaptation within the hypothalamus, producing a downstream of hormonal changes, such as blunted cortisol or testosterone responses to stress, altered nocturnal growth hormone pulsatility, as well as reproductive and metabolic irregularities. While an extent of hypothalamic dysfunction has been shown in some studies [7, 8, S], it has been widely debated among researchers as to whether the maladaptation lies within the hypothalamus, or its dysregulation may be attributed to secondary causes, such as a pituitary or adrenal level dysfunction [1], a short-term adaptive stress response [15], peripheral metabolic and adipokine-driven mechanisms [17] or, simply, a multifactorial process [18]. In light of this ongoing debate, this review aims to explore possible hormonal biomarkers of OTS, with a particular focus on hormones of the hypothalamic-pituitary axis.

3.3. Diagnosis

The diagnosis of Overtraining Syndrome is one of exclusion. The list of conditions to consider is long and although many diagnostic approaches have been suggested in the past, no widely accepted guidelines have been established up to date. The differential diagnosis of OTS includes many organic diseases (e.g., thyroid dysfunction, diabetes mellitus, iron deficiency anemia, infectious diseases) and non-organic diseases, such as eating disorders (anorexia nervosa, bulimia) [3].

3.4. Basal hormonal tests

The earliest proposed objective hormonal markers of OTS were resting testosterone and resting cortisol measurements presented as testosterone-to-cortisol (T/C) ratio [11, 18].

Testosterone is considered an anabolic hormone (tissue-building), while cortisol exerts catabolic actions (tissue breakdown). A decrease in the T/C ratio would indicate a shift of metabolic homeostasis towards tissue breakdown, explaining the chronic fatigue as a sign of inadequate muscle recovery. There are imperfections, however, with the T/C ratio. A shift in the ratio does not always point directly to OTS, for example, we may observe such result in isolated testosterone decrease, with normal cortisol levels; or an increase in both, but cortisol increase being more pronounced [21]. Additionally, an altered T/C ratio cannot be interpreted as diagnostic alone, without supportive patient history and symptoms.

Isolated basal concentrations of testosterone and cortisol have also been proposed as diagnostic tools [1, 18]. Especially, it has been hypothesized that elevated basal cortisol in the morning salivary sample may be a marker of chronic overtraining, as it typically rises with physiological stress [1, 18]. However, cortisol exhibits diurnal variation, high variability between individuals, is an unspecific marker, easily influenced by acute physiological stress, infections, and sleep disturbance. Moreover, there is no clear distinction between cortisol levels in well-trained, overreaching and overtraining athletes. When it comes to basal free testosterone, it exhibits a wide inter-individual variability and can be normal even in overtrained athletes.

Another ratio that has emerged in recent years, thanks to the EROS studies [6, 8, 9, 10], is the testosterone-estradiol (T/E2) ratio. According to these studies, the T/E2 ratio was one of the strongest supportive evidences of OTS diagnosis and is included in the EROS-SIMPLIFIED and EROS-COMPLETE models, which showed 100% accuracy in discriminating OTS from non-OTS athletes. The estradiol levels on their own were found to be relatively higher in overtrained athletes, even when accompanied by normal testosterone levels, suggesting either a shift towards aromatization or reduced estradiol clearance. However, the study was performed only on a limited group of male athletes under rigorous testing, and necessitates validation, especially in female athletes, in whom sex hormone distribution differs greatly from males.

3.5. Dynamic hormonal tests

In comparison to resting (basal) hormonal tests, which are often normal or inconsistently altered due to diurnal and daily variability in OTS, dynamic tests provide more reliable information about hormonal reserve and responsiveness. The most-commonly cited studies on OTS [6, 8, 9, 16, 21] have revealed a so-called “blunted HPA response” in OTS. However, others caution about unstandardized protocols and inconsistent results, calling for more comprehensive studies [18]. Stress tests have their own limitations – in order exert stimulation, the patient needs to either perform physical exercise (e.g., on an ergometer), or be administered stimulatory substances (e.g., insulin in IIT). Many hormonal pathways are also altered in response to those stimuli, as it is virtually impossible to target just one hormone. Despite those difficulties, some stress tests have been found to possibly provide proof of hypothalamic fatigue, which is one of the proposed hypotheses of OTS.

One of the earliest studies on OTS (Barron et al., 1985; [2]) found that, following an insulin intolerance test and subsequent insulin-induced hypoglycemia, overtrained endurance athletes also experienced a decrease in cortisol, ACTH, GH and prolactin in comparison to their healthy counterparts; however, the LH, TSH, and prolactin release in response to LHRH and TRH did not differ – suggesting that the issue selectively affected the hypothalamus. However, other studies have implicated accompanying variable degree of pituitary dysfunction, too [2, 6, 8, 9]. Nearly 20 years later, another study was performed (Meeusen et al., 2004 [18]) that measured ACTH, cortisol, prolactin and GH after each of 2 bouts of endurance training. The study showed a fairly normal response after the first training bout, and a blunted hypothalamic and pituitary response after the second bout, that was performed around 6 hours from the first. The conclusion was that a two-bout exercise challenge could reveal disturbances in hormone stress responses in the recovery phase, which may serve as a tool to diagnose overreaching before it progresses into full overtraining syndrome.

3.6. EROS diagnostic tools

Fast-forward another 15 years, the EROS-Complete Tool was developed (Cadegiani et al., 2019; [8]). It includes 20 variables, each assigned 1 point, and at 11 points or more, allows for the diagnosis of Overtraining Syndrome. Among different clinical, psychological, nutritional, and physical parameters, 9 of them are assigned to hormonal disturbances. These include:

Table 2. Hormonal parameters included in the EROS-COMPLETE tool (Cadegiani et al., 2019; [8]).

Parameter	Range
ACTH 30 minutes after hypoglycemia (pg/mL)	<35
Cortisol response to ITT (ug/dL)	<19.1
Basal GH (ug/L)	**<0.1**
GH 30 minutes after hypoglycemia (ug/L)	<1.0
Basal prolactin (ng/mL)	**<7.1**
Prolactin during ITT (ng/mL)	<12
Prolactin 30 minutes after hypoglycemia (ng/mL)	<10
Total testosterone (ng/dL)	**<400**
Testosterone to estradiol ratio	**<13**

Note: The parameters in bold represent the variables included in the EROS-SIMPLIFIED tool.

The simplified version of the tool, the EROS-SIMPLIFIED tool, includes only 4 of those measurements: basal GH, basal prolactin, total testosterone, and the testosterone-to-prolactin ratio, thereby excluding the functional tests. Nevertheless, the list is extensive and measurements not always possible to perform. There are some individual difficulties with particular tests. For instance, basal GH measurement is challenging because of its large, irregular and mostly nocturnal pattern of secretion and between these bursts, GH may be very low or undetectable. Accordingly, prolactin levels are altered by many factors, including gender, sleep, sexual activity, alcohol consumption, among others, therefore it is difficult to obtain a reliable result.

Another version of the tool, the EROS-CLINICAL, excludes biochemical tests all together, however, according to the authors, where it did not provide 100% accuracy, EROS-SIMPLIFIED was still used.

It is necessary to point out, however, some limitations of the study. The EROS tools have only been used to assess for OTS in 51 male subjects that practiced both endurance and strength sports, aged between 18 to 50 years, with other exclusions such as weekly training time, drug or substance use, confounding diseases. In the 2020 research article by Cadegiani et al. [9] the authors themselves acknowledge that for the tools to ever become an agreed upon assessment of overtraining in athletes, more studies are needed for to strengthen and validate the evidence. It is necessary to conduct further studies that are more inclusive of female athletes,

athletes below the age of 18 and above the age of 50, and those with confounding conditions, disabilities, using medications, or presenting with other conditions. Based solely on the EROS study it is impossible to know whether the established tools would reproduce the same accuracy as in the examined cohort.

Nonetheless, the tools presented in the set of studies performed by Cadegiani et al. give hope that, in the future, a consensus on a particular diagnostic tool may be reached.

4. Discussion

Overtraining syndrome is a complex condition that is not yet fully understood. Despite multiple hypotheses, no single mechanism fully accounts for the wide spectrum of symptoms observed in affected patients. This ambiguity is further exacerbated by other physical and psychological conditions frequently seen in elite athletes, as well as difficulty in delineating chronic overreaching from a fully developed OTS. Hormonal biomarkers, particularly those involving basal testosterone, cortisol, T/C and T/E2 ratios, as well as dynamic stress tests assessing HPA responsiveness, hold promise but are hindered by inter-individual variability, diurnal fluctuations, and methodological inconsistencies. Previous studies have examined the usefulness of measuring basal and dynamic hormone levels of the HPA axis, however, it is debated whether any brought results that are reproducible. This doubt is further exacerbated by the fact that hormonal dysfunction does not account, to our current understanding, for all symptoms experienced by affected patients. In recent years, the EROS studies have enriched the conversation by introducing potential diagnostic tools that demonstrated impressive accuracy within a limited cohort. Nonetheless, a scientific consensus on diagnostic criteria cannot be reached until these tools are validated among larger, more diverse participant groups.

5. Conclusions

Currently, overtraining syndrome still remains an elusive and complex condition, lacking a definitive pathophysiological explanation and universally accepted diagnostic criteria. The EROS studies have shed a new light on the conversation, but it remains important to establish whether the tools they have come out with could truly be applied in the clinical setting. Ultimately, while hormonal biomarkers offer a possible path for understanding the pathophysiology and refining the diagnosis of OTS, continued research exploring various hypotheses is imperative to eventually transition OTS from a diagnosis of exclusion into one grounded in objective, reproducible evidence.

Disclosure

Author contributions

Conceptualization: Iga Nowicka
Methodology: Iga Nowicka, Bartosz Łuniewski, Angelika Macko, Paweł Rios Turek, Maria Łuniewska
Formal analysis: Iga Nowicka, Bartosz Łuniewski, Angelika Macko, Paweł Rios Turek, Maria Łuniewska
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Writing - review and editing: Iga Nowicka, Paweł Rios Turek, Bartosz Łuniewski
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Authors declare no conflicts of interest.

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