Exercise Induced Responses in Salivary Testosterone, Cortisol, and Their Ratios in Men: A Meta-Analysis

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Key points

- Aerobic, resistance, and power-based training modalities resulted in overall standard difference in means (SDMs) of 0.891, 1.061, and 0.509, respectively for salivary testosterone, while SDMs for salivary cortisol were 3.041, 0.773 and 1.200 respectively.
- Aerobic, resistance, power training modalities resulted in overall salivary testosterone:cortisol ratio SDMs of -2.014, 0.027 and -0.968 respectively, which were largely achieved by increases in salivary cortisol.
- Findings from this meta-analysis highlight study design and sampling regimen can impact the magnitude of change and sampling regimen can dictate the direction of observed change.
- Further randomized controlled trials are required to determine the true influence that exercise exerts on salivary hormones.

Running head: Salivary Testosterone and Cortisol Following Exercise

Abstract

Background Testosterone, cortisol, and their ratios may be indicators of anabolic status, but technical issues surrounding blood sampling has limited wider application. The advent of salivary testosterone (sal-T) analysis simplified sample acquisition resulting in a subsequent rapid increase in the number of published research articles.

Objective To undertake a meta-analysis to determine the effect of acute exercise bouts on post exercise sal-T and salivary cortisol (sal-C) concentrations and their ratio (sal-T:C).

Data Sources Relevant databases such as PubMed, Web of Science, Science Direct, and SPORTDiscus were searched up to and including 31st December 2013 for the term 'saliva AND testosterone AND exercise'

Study Selection Studies (n=21) from 933 identified included; randomized controlled trials (RCTs; n=2), uncontrolled trials (UCTs; n=18) and control trials (CTs; n=1) all of which had an exercise component characterised as either *aerobic*, *resistance* or *power training*, each with acute sal-T and sal-C measurement obtained within 30 minutes of exercise bout completion.

Study Appraisal and Synthesis Methods A meta-analysis was conducted on change in sal-T, sal-C, and the sal-T:C ratio following exercise using standardised difference in means (SDM) and a random effects model.

Results For aerobic, resistance and power exercise the overall SDMs for sal-T were 0.891, 1.061, and 0.509, respectively, for sal-C SDM were 3.041, 0.773 and 1.200 respectively. For sal-T:C SDMs were -2.014, 0.027 and -0.968 respectively. RCTs, UCTs and CTs were separated by subgroup analysis. There were significant differences in overall weighted SDM values for sal-T between RCTs, UCTs and CTs within exercise modes. When examining aerobic exercise interventions, a quantitative interaction of study design was observed. RCTs

resulted in a greater SDM compared to UCTs (1.337 versus 0.446). Power interventions displayed a qualitative interaction with study design. UCTs where baseline measures where obtained 24 hours before exercise had a SDM of -1.128 whereas UCTs where baseline was determined immediately prior to exercise had an SDM of 0.486. The single CT trial had a SDM of 2.260. Resistance exercise interventions were primarily UCTs however an observed influence of baseline sampling time whereby immediately pre and 24 h pre resulted in differing SDM. The sole resistance exercise RCTs resulted in the greatest SDM (2.500).

Conclusion The current body of evidence regarding acute responses of sal-T to exercise is weak. This meta-analysis identifies varying exercise dependent effect sizes. Each appear to be greatly influenced by study design, and sample timing. There is a need for more RCTs and a standardized methodology for the measurement of salivary hormones in order to better determine the effect of exercise modality.

1 Introduction

1.1 Rationale

Saliva collection and analysis has rapidly developed as a tool for the assessment of physiological biomarkers usually measured in blood [1- 4]. The use of saliva for monitoring steroids, peptide hormones and markers of immune function has made saliva sampling attractive to researchers and clinicians [5]. Saliva can be collected rapidly, frequently and without the stress induced by venipuncture. Furthermore, saliva collection requires minimal medical training and may be performed outside of the laboratory setting. Salivary hormones have been measured in response to psychological stress, illness, and in behavioral experiments (for a review see Groschl [2]). Recently, salivary testosterone (sal-T), in unison with cortisol

(sal-C) has been reported as a marker of anabolic status [6, 7], overtraining [8-12], training response [13-18], and training motivation [19, 20].

There has been a near exponential increase in research investigating sal-T as a surrogate measure of serum testosterone (see Electronic Supplementary Material Figure S1) to the extent that serum and salivary measures are perceived to be equivalent. However, whether or not they are comparable, remains to be determined. More recently exercise-induced sal-T has received particular attention [1, 16, 19, 21-27], with both training effects [1, 27-31] and acute exercise-induced effects [32-34] being studied.

The acute endocrine response to acute exercise bouts has received considerable attention within recent years [3, 13-15]. Whether in saliva or serum, there are polarized opinions concerning the relative importance [35, 36], or lack thereof [37-40], of acute hormonal responses in determining chronic muscular adaptations. Despite popularity, sal-T response to acute exercise remains unclear. Inconsistencies in methodology hamper further comparisons between studies. As an example, radioimmunoassay intra- and interassay variation coefficients (CV) have been reported as below 5% [41], however enzyme linked radioimmusorbent assay intra- and interassay CVs have been reported as below 10% [14]. Furthermore, radioimmunoassay demonstrates significantly greater sensitivity than does enzyme linked immunosorbent assay in the determination of salivary hormones [42]. Additionally, it is possible that different modalities of exercise (i.e. aerobic, resistance, and power-based exercise) have different sal-T responses [43] which further limits study comparisons. For example, Wahl and colleagues [43] reported that high intensity interval training (HIIT) resulted in greater increases in cortisol, testosterone, and growth hormone compared to high volume aerobic training. These authors attributed differences to HIIT promoting anabolic adaptations. Conversely, Beaven and colleagues [14] reported a lack of change between 85%, 70%, 55%, and 40% one repetition maximum (1-RM) for acute sal-T response. As the acute sal-T and sal-

C response to exercise may influence chronic adaptations [7, 13-15], it is important to definitively determine the influence of acute exercise on sal-T and sal-C. Therefore, pooled analysis of previous investigations is warranted.

1.2 Objectives

Despite the growing popularity of the measure, there was no meta-analysis concerning acute exercise induced sal-T response. Therefore, the aim of this investigation was to conduct a meta-analysis on studies examining the effect of acute exercise on sal-T, sal-C, and the ratio between the two (sal-T:C). A secondary aim was to investigate study characteristics, namely study design and exercise mode on salivary hormone response.

2 Methods

2.1 Eligibility Criteria

Studies that met the following criteria were included in this meta-analysis: (1) published as a full-text manuscript; (2) not a review; (3) participants were apparently healthy adult males (> 18 years); (4) studies were required to employ an experimental design and include an acute exercise intervention with measurements of sal-T. Additionally, descriptive data (i.e. sample size, mean and standard deviation) were required to be reported. Where this was not possible, details were requested from authors. The primary aim was to investigate the acute sal-T response to exercise, therefore, we only included studies that collected sal-T within 30 min of exercise completion. Where an investigation took multiple samples post-exercise, we recorded the earliest sampling point. Separate figures, detailing the first two sampling time points can

be found in supplementary material (Electronic Supplementary Material Tables S1-3 and Figures S2-4).

Initially, this review aimed to consider randomized controlled trials (RCTs) and non-randomised control trials (CTs). However, due to the small number of RCTs and CTs, the study was extended to include uncontrolled trials (UCTs). For clarity, UCTs are reported separately from RCTs and CTs.

2.2 Information Sources

The following databases were identified and searched: Science Direct, PubMed, SPORTDiscus and Web of Science. The initial reporting of testosterone in saliva began in 1976 [44] and therefore, the literature search dates were set from 1st January 1976 to 31st of December 2013. The search was performed within all fields and the term was 'saliva AND testosterone AND exercise'.

2.3 Study Selection

Two of the lead authors conducted the eligibility assessment in an unblinded and standardized manner. Once each database search was completed and manuscripts were sourced, all studies were downloaded into a single reference list with duplicates removed. Titles and abstracts were then screened for eligibility and full texts were only retrieved for studies with sal-T and an acute exercise intervention incorporated. All studies retrieved as full texts were then thoroughly assessed using the complete eligibility criteria with first and second authors confirming inclusion and exclusion. Where an agreement could not be reached between the first and second

author, the third author confirmed selection. Additionally, retrieved full text articles were classified using a modified PEDro scale [45].

2.4 Data Collection Process

Sal-T, sal-C, and sal-T:C data were extracted for pre and post exercise intervention. In cases of missing data, authors were contacted via email and asked to provide necessary information. If no response was received, where possible means and standard deviations (SDs) were estimated from figures using computer software (Image J, Maryland, USA, Imagej.net). Information was imported into a spreadsheet, which was specifically designed for this meta-analysis (Comprehensive meta-analysis, NJ, USA). Where the sal-T:C ratio was not reported, it was calculated by dividing sal-T by sal-C.

2.5 Data Items

Heterogeneity was quantified with the I^2 statistic. An I^2 value of 25% may be interpreted as low, 50% as moderate and 75% as high between study heterogeneity. Three random-effect meta-analyses (aerobic exercise intervention, power exercise intervention, resistance exercise intervention) were conducted as each of these exercise modalities have appreciably different physiological demands and are thus considered separately. Data extracted from each study included; study sample size, intervention/control group descriptions, study design, salivary analysis method, population descriptions and outcome data (e.g. sal-T and sal-C concentrations). Furthermore, methodological quality was assessed using the modified 0-10 PEDro scale [45]. The primary outcome variables were defined as sal-T, sal-T, and sal-T:C pre- and post-intervention. Standard difference in means (SDM) were retrieved for inclusion

into the three meta-analyses; aerobic, power, and resistance exercise. Moreover, subgroup analyses were performed based on research design as a means of investigating heterogeneous results.

INSERT FIG 1 NEAR HERE.

3 Results

3.1 Study Selection

After the initial database search, 933 records were identified (see Fig. 1). Once duplicates were removed, 756 titles and abstracts were screened for inclusion by two authors resulting in 80 studies being retrieved as full text and assessed for eligibility. Of those, 53 were excluded and 27 articles remained. Authors of articles with missing details were contacted resulting in 21 studies used in the final quantitative synthesis. To assess publication bias, funnel plots for each exercise modality were computed and failsafe N was calculated.

3.2 Study Characteristics

Of the 21 studies included, 2 were RCTs [46, 47], 18 were UCTs [3, 4, 15, 22, 23, 29, 32, 34, 41, 48-56], and 1 was a CT [18] (Tables. 1-3). Where a study had multiple conditions, they were treated separately. We further divided the UCTs into studies whereby the 'pre' sample was taken immediately prior to exercise (UCT Im) and studies whereby the 'pre' sample was taken on a control non-exercising day (UCT CD) to examine the influence of sample timing on sal-T response.

INSERT TABLES 1-3 NEAR HERE

3.3 Effect of Exercise Type on SDM

SDM and 95% confidence intervals (CI) are displayed in Figs. 2-4. There were positive effects of exercise on sal-T and sal-C in each intervention type. Aerobic and resistance interventions resulted in an increase in the sal-T:C ratio, whereas power exercise resulted in a negative SDM for sal-T:C.

INSERT FIGURES 2-4 NEAR HERE

3.3.1 Aerobic Interventions

The overall sal-T SDM of aerobic exercise interventions was 0.891 (95% CI = 0.018 – 1.765). Subgroup analysis revealed a quantitative interaction of study design (i.e. the magnitude of the effect was influenced). RCTs resulted in a greater SDM than UCTs using baseline values as their control (1.337, [95% CI = 0.886 – 1.788] compared to 0.446 [95% CI = -0.002 – 0.895] respectively).

The overall sal-C SDM of aerobic exercise interventions was 1.091 (95% CI = -0.067 – 6.148). Subgroup analysis revealed a quantitative interaction of study design (i.e. the magnitude of the effect was influenced). RCTs resulted in a greater SDM than UCTs using baseline values as their control (4.624, [95% CI = 2.931 – 6.316] compared to 1.453 [95% CI = -0.257 – 3.163] respectively).

The overall sal-T:C SDM of aerobic exercise interventions was -2.024 (95% CI = --4.804 – 0.756). Subgroup analysis revealed a quantitative interaction of study design (i.e. the magnitude of the effect was influenced). RCTs resulted in a greater negative SDM than UCTs using baseline values as their control (-3.441, [95% CI = -4.711 – -2.171] compared to -0.604 [95% CI = -1.884 – 0.675] respectively).

3.3.2 Power Based Interventions

The overall sal-T SDM for power based interventions was 0.509 (95% CI = -1.089 – 2.106). Power interventions displayed a qualitative interaction with study design (i.e. the direction of the effect is reversed). Two studies [29, 34] which employed a rest day as baseline sample design resulted in a SDM of -1.128 (95% CI = -1.872 - -0.384) when compared to baseline as control design (0.486 [95% CI = 0.035 – 0.937]) and control group design (2.260 [95% CI = 1.669 – 2.850]). A quantitative interaction was observed between baseline as control design and control group design. However, only one study used a control group design [18].

The overall sal-C SDM for power based interventions was 1.287 (95% CI = -0.092 – 2.666). Power interventions displayed a quantitative interaction with study design whereby two studies [29, 34] which employed a rest day as baseline sample design resulted in a SDM of 1.992 (95% CI = 1.433 – 2.550) when compared to baseline as control design (0.585 [95% CI = 0.036 – 1.134]).

The overall sal-T:C SDM for power based interventions was -0.968 (95% CI = -2.903 – 0.967). Power interventions displayed a quantitative interaction with study design whereby studies which employed a rest day as baseline sample design resulted in a SDM of -1.978 (95%

CI = -3.101 – -0.856) when compared to baseline as control design (-0.003 [95% CI = -0.959 – 0.952]).

3.3.3 Resistance Based Interventions

Resistance training interventions resulted in an overall sal-T SDM of 1.061 (95% CI = 0.174 – 1.949). We investigated the influence of baseline sampling time for UCTs on SDM and observed trivial differences between immediately pre and 24 h pre (0.771 vs. 0.851 respectively). The sole resistance exercise RCTs resulted in the greatest sal-T SDM (2.500 [95% CI = 0.515 – 4.486]).

Resistance training interventions resulted in an overall sal-C SDM of 0.773 (95% CI = -0.805 – 2.350). We investigated the influence of baseline sampling time for UCTs on SDM and observed large differences between immediately pre and 24 h pre (-0.349 vs. 1.307 respectively). The sole resistance exercise RCTs resulted in the greatest SDM (1.936 [95% CI = -0.164 – 4.037]).

Resistance training interventions resulted in an overall sal-T:C SDM of 0.190 (95% CI = -0.026 – 0.406). We investigated the influence of baseline sampling time for UCTs on SDM and observed large differences between immediately pre and 24 h pre (0.190 vs. -0.431 respectively). The sole resistance exercise RCT resulted in the greatest SDM (0.378 [95% CI = -0.507 – 1.262]).

4 Discussion

4.1 Overall SDM

The main finding from this study was that acute exercise resulted in increased both sal-T and sal-C concentrations. This was evident in each of the three exercise modalities. However, the direction and magnitude of these effects varied both with exercise modality and study design. There are opposing theories in relation to the importance of the hormonal response to acute exercise and chronic training adaptations. Given that salivary sex hormones are proposed to mirror their systemic counterparts [51], assessment of acute exercise on these salivary hormones may illuminate our understanding of the link between acute hormonal response and longer term physiological adaptations.

4.2 Aerobic Interventions

Studies displayed a positive SDM for sal-T except Doan et al. [48] who reported decreased sal-T following 36 holes of golf. It is expected that as this intervention would take considerable time and testosterone has a circadian rhythm with levels declining over a waking day, these factors may have influenced the results. Furthermore, golf is the least physiologically stressful (lowest metabolic requirement) of all the interventions examined and therefore may not have altered testosterone significantly compared to a control day. Interestingly, Doan et al. [48] reported unaltered sal-T after 36 holes compared to a baseline sample taken 24 hours prior but a lower sal-T concentration compared to immediately pre-competition. This identified how study results that utilized a control sample taken immediately prior to exercise may have been influenced by an anticipatory change to sal-T that may not have been reflective of the magnitude of true change in sal-T caused by the resultant exercise *per se*.

Studies displayed a positive SDM for sal-C. As aerobic exercise interventions are generally the longest in duration (Table. 1), one may have expected the greatest sal-C SDM due to energy demands of the events and therefore reduced blood glucose. The greatest SDM

was observed by Hough et al. [46] when comparing cycling to fatigue to a control condition. It is suggested that the exercise regime (primarily as it was to fatigue) and the study design caused the large SDM.

As a result of the large sal-C SDM reported by Hough et al [46], it was this investigation that exhibited the greatest negative sal-T:C SDM, which was mainly driven by sal-C rather than sal-T. All aerobic interventions displayed a negative sal-T:C SDM, with the result that aerobic exercise demonstrated the largest reduction (negative SDM) in sal-T:C compared with other exercise modalities. This was due primarily to the magnitude of the sal-C response to aerobic exercise, which was consistent between studies.

4.3 Power Interventions

Overall effect of power based exercise interventions was generally positive, with some notable exceptions. McLellan et al. [34] reported a significant fall in sal-T concentration following a rugby league match (approximately 760.2 vs. 463.11 $pmol \cdot L^{-1}$ respectively). Another intervention to report a decrease in sal-T was Elloumi et al. [29] who reported a similar fall following a rugby match. Importantly both McLellan et al. [34] and Elloumi et al. [29] compared their post exercise results against a baseline measure obtained 24 hours prior to the exercise intervention. McLellan et al. [34] and Elloumi et al. [29] observed the smallest SDM (-1.31 and -1.125). Conversely, Kilduff and colleagues [18] reported a large positive SDM when examining the response to repeated sprints. However, a greater SDM was observed when resting values taken in a non-exercising control group were compared to the baseline values (taken immediately prior to exercise) of the experimental group (2.260 vs. 0.916 respectively). Whilst some of this difference may have been accounted for by the large inter-individual variation in sal-T, both non-exercising control group and the intervention group were rugby

players. This again suggests that there may be an anticipatory effect of exercise on sal-T. Furthermore, Cook et al. [17] reported morning sal-T concentrations, prior to any intervention were lower under control conditions compared to either sprint or weight training, indicating an anticipatory response in experimental conditions.

A similar phenomenon was apparent for sal-C whereby baseline sampling immediately pre exercise may have included an anticipatory rise causing to lower SDMs compared to studies incorporating baseline on a control day. Power interventions resulted in a positive sal-C SDM. However, two studies positively weighted the overall SDM [29, 34]. These investigations examined competitive rugby matches which presumably included numerous collisions and sprints. However, McLellan et al. [57] previously reported no correlation between number of contacts and sal-C following rugby league match play. The increase in psychological stress may have exacerbated the SDM in these investigations [29, 34].

As a result of the larger sal-C SDM than sal-T S, power interventions resulted in a negative sal-T:C SDM suggesting increased catabolism. Similarly to sal-C, this was magnified in two studies which used a control day for the baseline salivary sample [29, 34].

4.4 Resistance Interventions

There is a pervasive belief that acute testosterone elevations following exercise is required to induce hypertrophy over time. If this is the case, one would expect the greatest SDM to result from hypertrophy targeting programmes. If, conversely, hypertrophy is achievable without acute testosterone elevations one would expect random sal-T response to resistance exercise. While this meta-analysis demonstrates that resistance training produces the largest SDM for sal-T, it was not a consistent finding between studies. The greatest positive and negative sal-T

SDMs were both reported from the same UCT investigation [22] which utilized a randomized cross over design of 3 loading schemes. The greatest SDM (5.80) was observed 15 min after a hypertrophy loading scheme (see Electronic Supplementary Material Figure S4). The smallest SDM was observed immediately after completion of a power training resistance scheme (-0.997). Furthermore, the protocol which produced the lowest SDM, when reassessed 15 min later resulted in a positive SDM (0.813). A paradoxical effect was observed in the strength loading scheme that produced the largest SDM whereby a negative SDM was identified 15 min later (1.369 and -0.906 respectively [see Electronic Supplementary Material Figure S4]). This suggests that sal-T is an inherently variable measure and standardized testing methodology with regards to post-exercise sampling time may benefit the research field. Studies incorporating hypertrophy style resistance training resulted in the greatest increase in sal-T [22, 47, 52]. Studies incorporating complex training [53], small muscle groups [3], middle aged men [51], and training preceding a loss [56] all resulted in no effect of exercise on sal-T. It is suggested that these factors influence the sal-T response to exercise. Trivial effect of baseline sample time was observed in the resistance exercise analysis in contradiction to power interventions and therefore, imminent exercise may produce significantly different anticipatory effects on sal-T.

Resistance training interventions resulted in an overall sal-C SDM of 0.773 however, baseline sampling time for UCTs caused large changes in SDMs when measured immediately pre or 24 h pre (-0.349 vs. 1.307 respectively), indicating an influence of sampling regimen. The sole resistance exercise RCT resulted in the greatest SDM (1.936) suggesting research design influenced subsequent results and conclusions. The overall sal-C SDM of 0.773 was the lowest of this meta-analyses which may be attributable to the different stress response and metabolic pathways of resistance exercise compared with power and aerobic exercise.

Due to the interaction between study design and sal-C, the sal-T:C ratio also demonstrated a similar interaction. Studies that took baseline samples immediately prior to exercise had, overall, an increased sal-T:C ratio indicating a more anabolic environment, while studies that took baseline measures 24 hrs prior had reduced sal-T:C changes indicative of a catabolic profile. Consequently the overall effect, when combining these two subgroups, resulted in no change in sal-T:C ratio. Therefore, due to the differences in study designs, it is not possible to determine the true effect of acute resistance exercise on salivary hormones.

4.5 Study Design

It is apparent that the timing of sampling prior to exercise may significantly affect the magnitude and direction of sal-T and sal-C response observed [18, 29, 34]. This raises an important methodological consideration and suggests an anticipatory effect on sal-T and sal-C. Investigators may be unaware of this phenomenon as the majority of investigations solely collect samples immediately prior to exercise rather than in addition to a control day [3, 4, 15, 22, 23, 29, 32, 41, 48-56]. Therefore, care should be taken when comparing exercise-induced acute changes in sal-T and sal-C with regards to the timing of baseline measurement. When reporting baseline on a rest day, considerable caution should be used when attempting to compare investigations that have utilized different methodologies (e.g. sampling regimen, study design). As with measurements comparing blood testosterone between laboratories, it may be useful for each lab to consider establishing reference ranges and variation coefficients for sal-T.

There are known appreciable difficulties for field-based practitioners attempting to study sal-T and sal-C responses (e.g. it is not usually feasible for professional athletes to act as a control and therefore not train) however, practitioners should avoid taking baseline samples immediately before exercise as analysis of the literature suggests there is an anticipatory effect on sal-T [17, 29, 34]. Further issues include the heterogeneity of study design and the timing of post-exercise samples in particular. The majority of studies within this meta-analysis collected saliva samples immediately upon finishing exercise, however, it appears that sal-T can increase and then decrease (and vice versa) within 15 min of completion of exercise [22]. Additionally, exercise interventions varied in duration, and it is not possible to determine whether sal-T varied throughout the duration of the exercise.

From the present meta-analysis, it is tempting to conclude that CTs and RCTs result in different SDMs to UCTs. However, due to the distinct lack of CTs and RCTs it is difficult to justify such a conclusion, and further RCTs are warranted to add greater credence to the field. Furthermore, it may be necessary to control for age as young and middle-aged men exhibited differences in acute sal-T response to exercise [4]. Additionally given that the hormonal response to resistance exercise is a commonly cited rationale for hypertrophy type training [13, 35, 58-61] (although controversial [37-40]), the large sal-T and sal-C SDM seen following a hypertrophic protocol [22] warrants further investigation. Finally, while it is common practice to control for variables such as diurnal variation, other factors such as auditory [62] or visual [63] cues can cause significant changes of a similar magnitude. Confounding variables such as these may contribute to the variability of response evident in the literature and is a further factor that should be considered amongst researchers that employ sal-T and sal-C measurement.

4.6 Limitations

While the literature assessment was comprehensive, it is possible that studies may have been missed from the analysis. Furthermore, in order to reduce heterogeneity, studies were classified into one of three broad categories reflecting the physiological requirements of each. There may be some overlap, where some exercise interventions could possibly include components of more than one classification. However, the classifications used are plausible, scientifically robust and produced groups of sufficient size to allow for meaningful analysis.

Whilst some of these investigations have reported statistical significance, the smallest effect to be considered biologically significant remains to be fully determined. The critical difference of sal-T using radioimmunoassay (RIA) has previously been calculated as 77.7% [64], and the critical difference for sal-T using ELISA has only recently been reported as 90% [65]. Therefore, it is difficult to ascertain whether any of the sal-T responses are biologically meaningful.

While meta-analyses describe a population effect, i.e. group mean change, some authors have previously described a case study effect [13, 14, 66] whereby individuals display varying levels of sal-T response to the same exercise intervention in a homogenous group. For example, Beaven and colleagues [14] indicated professional rugby players responded maximally to different protocols when all other methodology was standardized. As such, a case study effect may have been evident and could explain large standard deviations in sal-T response. This has implications for clinicians as well as biological researchers, as salivary hormone measurement may be more relevant on an individual level, however making large cohort conclusions is not possible due to varied SDMs reported here.

5 Conclusion

Acute aerobic and resistance exercise consistently increased sal-T, although with large variations in effect size. Acute effects on power based exercise was less clear with both increased and decreased sal-T being evident. In terms of sal-C both aerobic and power exercise demonstrated consistent increases, whereas there was considerable variability following resistance exercise, dependent upon study design. Sal-T:C ratio remained largely unaffected following resistance exercise, while aerobic and power exercise demonstrate study design effects that can largely be accounted for by changes in sal-C. The timing of saliva sampling appeared to bias the direction of change in sal-T following exercise. Furthermore, there is a need for more RCTs to improve the quality of available evidence, as only two studies in the present investigation achieved a score of 5 on the PEDro scale [46, 47]. Additionally, RCTs would also overcome sample timing bias. Finally, a standardized method for the measurement of sal-T and sal-C, particularly with regards to the timing of post exercise samples and the use of standardized control conditions would help progress the field.

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FIGURE LEGENDS

Fig. 1 Schematic flow diagram describing exclusions of potential studies and final number of studies. RCT = randomized control trial. CT = controlled trial. UCT = uncontrolled trial.

Fig. 2 Summary of salivary (a) testosterone (sal-T); (b) cortisol (sal-C); and (c) ratio of testosterone to cortisol (sal-T:C) values for aerobic exercise interventions. All samples considered as baseline were taken immediately before exercise. [1,2,3] indicate separate conditions within one investigation. Filled diamond indicates overall SDM. SDM = standard difference in means. RCT = randomized controlled trial. UCT Im= uncontrolled trial; baseline immediately pre exercise.

Fig. 3 Summary of salivary (a) testosterone (sal-T); (b) cortisol (sal-C); and (c) ratio of testosterone to cortisol (sal-T:C) values for power exercise interventions. Baseline sample prior to exercise was defined as either immediately pre-exercise (immediately) or a minimum of 24 hours prior to exercise (control day). [1,2,3] indicate separate conditions within one investigation. Hollow diamonds indicate grouped mean of CT, UCT CD, and UCT Im, respectively (from highest to lowest). Filled diamond indicates overall SDM. SDM = standard difference in means. RCT = randomized controlled trial. UCT Im = uncontrolled trial; baseline sample immediately pre exercise. UCT CD = uncontrolled trial; baseline sample control day. CT = controlled trial.

Fig. 4 Summary of salivary (a) testosterone (sal-T); (b) cortisol (sal-C); and (c) ratio of testosterone to cortisol (sal-T:C) values for resistance exercise interventions. Baseline sample

prior to exercise was defined as either immediately pre-exercise (immediately) or a minimum of 24 hours prior to exercise (control day). [1,2,3] indicate separate conditions within one investigation. Hollow diamonds indicate grouped mean of RCT, UCT CD, and UCT Im respectively (from highest to lowest). Filled diamond indicates overall SDM. SDM = standard difference in means. RCT = randomized controlled trial. UCT Im = uncontrolled trial; baseline sample immediately pre exercise. UCT CD = uncontrolled trial; baseline sample control day**.**