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EFFECT OF ACCELERATION STRESS ON SALIVARY CORTISOL AND PLASMA CORTISOL AND TESTOSTERONE LEVELS IN CADET PILOTS

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The effects of acceleration (G_z) on changes in the levels of cortisol in saliva and of cortisol and testosterone in serum have been studied in 48 cadet pilots exposed to a linear acceleration gradient (0.2 G/s) until a loss of coordination when the mean G value was found to be 5.94 ± 0.57 . Three patterns of salivary cortisol responses were discerned based on G_z -induced significant changes: increase (I; $n = 20$), decrease (D; $n = 8$), the magnitude of changes being dependent on the pre- G_z values. Fifteen min after the G_z load, the mean salivary cortisol was significantly higher from the pre- G_z value in all subjects combined. In 19 subjects, who consented to blood sampling, significant increases in serum cortisol were observed both 3 and 15 min post- G_z (by 37 and 57% respectively) while, a significant increase in serum testosterone concentration has been observed only 3 min post- G_z . Testosterone levels 3 min post- G_z were significantly correlated with the final G_z values ($r = 0.54$; $p < 0.05$). A significant correlation was also observed between all salivary and serum cortisol values combined ($r = 0.696$; $p < 0.001$). It has been concluded that acceleration stress, although of very short duration, proved very potent in eliciting glucocorticoid and androgen responses.

Key words: *Acceleration stress, cortisol, testosterone*

INTRODUCTION

Adrenal cortex is known to respond with an increased secretion of cortisol to various forms of stress, like strong emotions, physical exercise, environmental factors, etc. (1–8) but the cumulative effect of several stimuli may be central fatigue and an impairment of functional mobilisation of the adrenal cortex in response to stress (9, 10).

The increase in cortisol concentration in body fluids depends on the kind and intensity of the stimulus and is usually less pronounced in organisms adapted to the given stressor (11–13). Cortisol measurements thus constitute a valuable diagnostic tool, and those in saliva are steadily gaining popularity due to the non-invasiveness and ease of serial sampling (14).

Stress induces also diverse changes in serum testosterone levels depending on the duration of the stimulus. Prolonged emotional stimuli (15), training overload (16, 17) or exertions associated with sport competition and lasting some hours (18, 19) decrease those levels which may represent the inhibiting effect of a high concentration of cortisol on gonadotropin secretion (20, 21). In effect, in periods of very high physical activity the concentrations of testosterone decrease and those of cortisol increase (22, 23). On the other hand, short-lasting emotional stimuli, e.g. before a laboratory exercise (24) or competition (25), augment the secretion of testosterone despite high cortisol levels (12, 25). Similar increases are also known to follow short, intense exercises (26, 27).

One of specific stressors is acceleration (G_z) to which pilots, and particularly fighter pilots, are routinely exposed (2, 28). Thus, the aim of this study was to assess the cortisol and testosterone responses observed in cadet pilots subjected to the acceleration stress.

MATERIAL AND METHODS

Students of the Military Aviation College ($n = 48$), aged 22.3 ± 0.5 years, have been subjected to the routine acceleration test on a centrifuge (radius length — 11 m). The subjects were in sitting position, the acceleration vector overlapping the body axis. A linear acceleration gradient was applied (0.2 G/s) until a loss of motovisual control (electronically monitored) when deceleration was started at a rate of 0.6 G/s. Total time of exposure to acceleration ranged from 30 to 40 s. All tests took place between 1100 and 1200 hours.

Three minutes before the test, as well as 3 and 15 min after the test had been terminated, saliva samples were collected. Nineteen subjects consented to having blood sampled simultaneously by fingertip puncture. Blood serum and saliva samples were placed in a freezer at -20° and stored until assayed.

Cortisol in saliva was determined by a specific radioimmunoassay as described elsewhere (29). Cortisol and testosterone in serum were also radioimmunoassayed by using commercial kits (Orion Diagnostica, Finland).

Individual changes in salivary cortisol concentrations were considered significant when exceeded 30% of the initial value (30). The results were analysed by using Student's t-test for independent or paired data, the level of $p < 0.05$ being considered significant.

RESULTS

Mean concentrations of cortisol in saliva in all 48 subjects, as well as those of cortisol and testosterone in serum of 19 subjects together with the corresponding salivary cortisol levels are presented in *Table 1*. Cortisol concentrations increased significantly in serum, both 3 and 15

min post- G_z , the increase 3 min post- G_z being significant only in subjects who consented to blood sampling. On the other hand, testosterone significantly increased as compared with the pre- G_z value only 3 min post- G_z , after which testosterone value returned to the pre- G_z level.

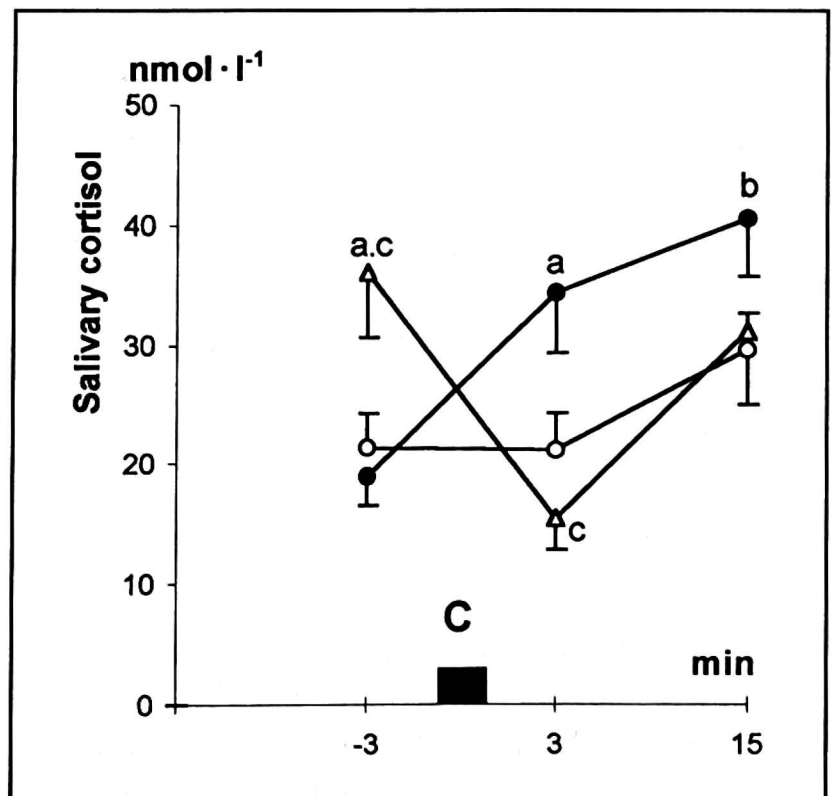
Table 1. Pre- and post- G_z (3 and 15 min) concentrations of cortisol in saliva in all subjects studied ($n = 48$) as well as in 19 subjects in whom concentrations of cortisol and testosterone in serum were determined in addition to salivary cortisol (means \pm SD).

Status	All subjects $n = 48$ Cortisol in saliva	Subjects who consented to blood sampling $n = 19$		
		Cortisol in saliva	Cortisol in serum	Testosterone in serum
3 min before G_z	22.8 ± 13.6	18.2 ± 13.0	266 ± 110	18.1 ± 5.1
3 min after G_z	26.3 ± 17.7	$21.6 \pm 14.1^*$	$363 \pm 147^{***}$	$20.2 \pm 5.7^{**}$
15 min after G_z	$34.8 \pm 17.6^{***}$	$28.7 \pm 14.0^{***}$	$417 \pm 126^{***}$	18.7 ± 6.0

The post- G_z changes vs. the initial values served as criterion of classifying the pilots into three categories: increased (I), decreased (D) or no change (N) in the salivary cortisol concentration. Mean values of salivary cortisol for those three categories are presented in Fig. 1. The pre- G_z value in Category D was significantly ($p < 0.01$) higher from those in Categories N and I, while post- G_z (3 min) values in Categories D and N were alike, differing significantly from that in Category I.

Fig. 1. Changes in salivary cortisol concentrations (means \pm SEM) in military cadet pilots subjected to G_z stress in a centrifuge (final $G = 5.94 \pm 0.57$). The subjects were divided in three classes according to the type of cortisol response to G_z : significant increase (I), decrease (D), or no change (N).

Legend: Open triangles: Group D ($n = 8$); Full dots: Group I ($n = 20$); Open dots: Group N ($n = 20$).
a — Significantly different from the respective value in other groups;
b — Significantly different from the respective value in Group N;
c — Significantly different from the respective initial value ($p < 0.05$);
C — Centrifuge



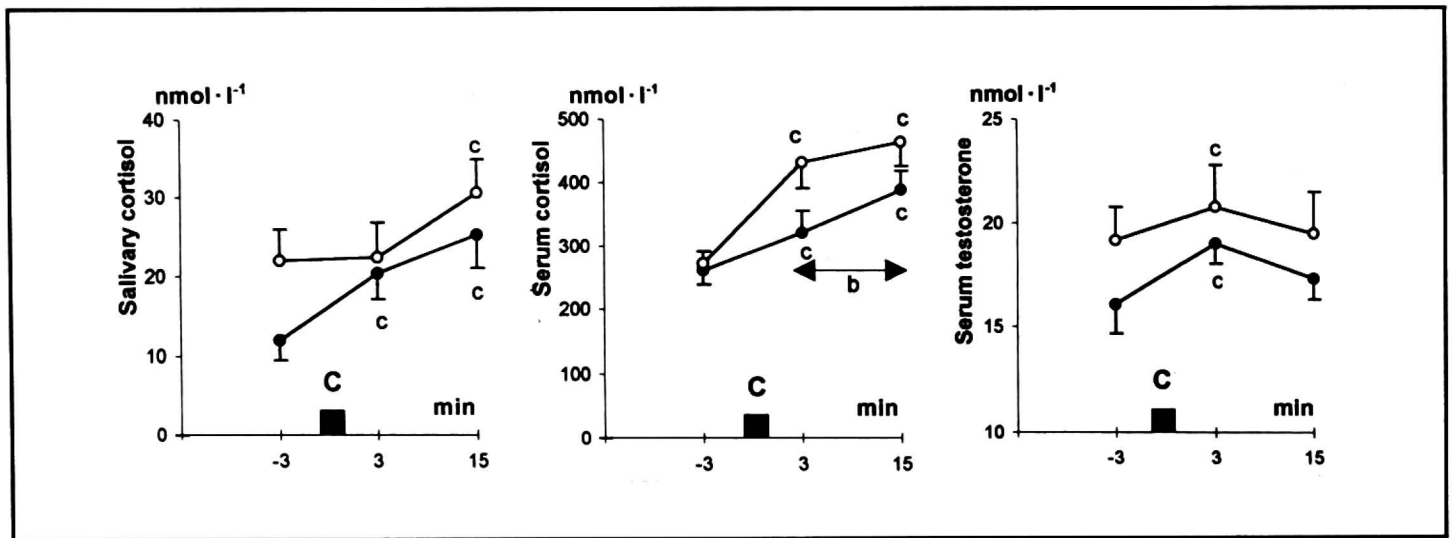


Fig. 2. Changes in salivary cortisol and in serum cortisol and testosterone concentrations (means \pm SEM) in military cadet pilots subjected to G_z stress in a centrifuge (final $G = 5.94 \pm 0.57$).

The subjects were from Groups N and I presented in Fig. 1.

Legend: Full dots: Group I ($n = 7$); Open dots: Group N ($n = 12$). b — Post- G_z values differ significantly from those in Group N ($p < 0.05$) c — Significantly different from the respective initial value ($p < 0.05$); C — Centrifuge
For other explanations see Fig. 1.

The results of cortisol and testosterone concentrations in serum from subjects who consented to blood sampling ($n = 19$), classified into the above categories, are presented in Fig. 2. Retrospectively, no subject from Category D had his blood sampled. Significant post- G_z increases in serum cortisol concentrations were noted in both categories (I and N) and those categories significantly differed from one another regarding the post- G_z cortisol levels. When all serum cortisol values were pooled and correlated with the corresponding salivary ones, the correlation coefficient was equal to 0.696 ($p < 0.001$, $n = 57$) but mean salivary and serum values were parallel to one another in Category I only ($r = 0.841$; $p < 0.01$; see Fig. 2). A significant increase in the testosterone level was observed in both categories 3 min post- G_z . In spite of a parallel shift in testosterone patterns, the difference proved non-significant. Furthermore, all mean salivary cortisol levels in Category I are clearly lower from those computed for all 48 subjects but the patterns of changes are identical (cf. Figs. 1 and 2).

Mean final G_z load, at which a loss of motovisual control took place, was 5.94 ± 0.57 and no differences between the three categories were observed. That load was correlated with the pre- G_z cortisol concentration in saliva only in cases when the post- G_z value was lower (not necessarily significantly) from the pre- G_z ones ($r = 0.59$, $p < 0.05$, $n = 13$). On the other hand, the post- G_z levels of testosterone, but not the initial ones, were significantly correlated with G_z values ($r = 0.54$; $p < 0.05$).

DISCUSSION

After a short, very intense stimulus, highest salivary cortisol values have been observed by Tauri and Nakamura 15 to 30 min after the stimulus (28). These authors reported that one min-exposition to acceleration stress induced increases in salivary cortisol only at 4 G or more. Peak concentration, observed 20 min after G_z , was about twice higher at 5 than at 4 G. Those results, taken together with those presented here, suggest that the duration of exposition to high accelerations plays an important role.

In this study, a very pronounced variability in the post- G_z cortisol levels in saliva was found. The direction of G_z -induced changes depended on the pre- G_z values. A similar relationship was observed also in athletes subjected to maximal exercises (31). The magnitude of observed changes — from a mean decrease by 20 nmol/l to a mean increase by 15 nmol/l (cf. *Fig. 1*) is much higher than that resulting from natural fluctuations in the hormone concentrations in saliva or that observed following a maximal exercise (30).

Although groups of pilots, classified according to the type cortisol response to G_z , did not differ from each other with respect to the maximum bearable G value, a weak correlation between that G value and the pre-test salivary cortisol ($r = 0.588$, $p < 0.05$) was observed in those subjects only in whom the post- G_z cortisol was lower (not necessarily significantly) from the pre- G_z one. This might suggest a better tolerance of G_z by those who are better capable of mobilising their adrenocortical reserve but this is inconclusive due to a very homogenous G_z tolerance shown by the group studied.

The post- G_z changes in cortisol concentrations suggest they were induced by ACTH. The dynamics of those changes resembles that observed in healthy subjects following administration of CRF (32) or in athletes subjected to a 30 a s supramaximal exercise (13).

In subjects from Group I, who volunteered to undergo blood sampling, much lower resting values of salivary cortisol were observed compared with the mean value for all subjects from that group (11.9 ± 8.2 and 19.6 ± 11.1 nmol/l, respectively; cf. *Figs 1 and 2*). Maybe, the former ones were less stressed which was reflected in their lower adrenocortical activity prior to the test. The correlation between pooled salivary and serum cortisol value ($r = 0.696$) is relatively high and similar to that reported by McLean *et al.* (3) well as earlier by us (31).

The post- G_z increase in testosterone concentration observed about 7 min after the initial blood sampling could not have been gonadotrophin-induced because the delay in the testosterone response to LH pulses was reported to be as high as 45 min (33). Testosterone level may also respond to exogenous ACTH (34, 35) but noticeable increases take place only about 30 min later and a further increase is accompanied by a decrease in the LH level (36). The post- G_z

increase in the testosterone concentration observed in this study was most likely due to adrenergic activation. It has been reported that exercise-induced changes in testosterone levels depended on β -receptor blockade (37, 38) and that post-exercise changes in testosterone and catecholamine concentrations were parallel to one another (39). Moreover, the parallelism between the intensity of exercise and testosterone increase (39), as well as a significant, positive correlation between the post- G_z testosterone level and final G_z value observed in this study may suggest that the mechanisms of stimulation of testosterone secretion by both stressors (acceleration and physical load) are alike.

It is to be mentioned that a similar pattern of testosterone response was observed by us in judo athletes following a highly stressful, 30 s supramaximal exercise (27). However, no data on G_z -induced changes in testosterone levels have been found in the available literature.

In conclusion, our results suggest a rather complex mechanism of hormonal responses to the G_z stress and the existence of diverse response patterns to that stimulus.

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