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ORIGINAL ARTICLE

Cytokine and hormone responses to resistance training

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Abstract This study examined the effects of heavy resistance training on physiological acute exercise-induced fatigue (5 × 10 RM leg press) changes after two loading protocols with the same relative intensity (%) (5 × 10 RM_{Rel}) and the same absolute load (kg) (5 × 10 RM_{Abs}) as in pretraining in men (n = 12). Exercise-induced neuromuscular (maximal strength and muscle power output), acute cytokine and hormonal adaptations (i.e., total and free testosterone, cortisol, growth hormone (GH), insulin-like growth factor-1 (IGF-1), IGF binding protein-3 (IGFBP-3), interleukin-1 receptor antagonist (IL-1ra), IL-1β, IL-6, and IL-10 and metabolic responses (i.e., blood lactate) were measured before and after exercise. The resistance training induced similar acute responses in serum cortisol concentration but increased responses in anabolic hormones of FT and GH, as well as inflammation-responsive cytokine IL-6 and the anti-inflammatory cytokine IL-10, when the same relative load was used. This response was balanced by a higher release of pro-inflammatory cytokines IL-1β and cytokine inhibitors (IL-1ra) when both the same relative and absolute load was used after training. This enhanced hormonal and cytokine response to strength exercise at a given relative exercise intensity after strength training occurred with greater accumulated fatigue and metabolic demand (i.e., blood lactate accumulation). The magnitude of metabolic demand or the fatigue experienced during the resistance exercise session influences the hormonal and cytokine response patterns. Similar relative intensities may elicit not only higher exercise-induced fatigue but also an increased acute hormonal and cytokine response during the initial phase of a resistance training period.

Keywords Serum hormones · Cytokines · Resistance training · Muscle power

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Introduction

Heavy resistance exercise protocols have been shown to elicit significant acute hormonal responses that are part of an integrated system of signaling to a multitude of target cells. Such complex interactions may be critical to the support and mediation of physiological adaptations to resistance training (Hakkinen et al. 1998; Kraemer and Ratamess 2005; McCall et al. 1999; Staron et al. 1994). Dramatic physiological changes in muscle have been demonstrated in the first several resistance training workouts. In a study by Staron et al. (1994), it was demonstrated that within four workouts myosin ATPase started to shift to the Type IIa isoform. While changes in the muscle fiber characteristics have been associated with the circulatory concentrations, how testosterone and cortisol help to mediate such protein adaptations remains unclear. The direct interactions in the circulation with immune cells and cytokine releases might be a primary target of such hormones. Thus, new insights into the physiological adjustments taking place may be gained by the further elucidation of such relationships and temporal changes in cytokine and immune regulation with anabolic and catabolic hormones during a resistance training program (Kraemer and Ratamess 2005; Peake et al. 2005).

Resistance exercise may cause myofibrillar disruption, especially after a fatiguing task, and trigger an inflammatory response. In fact, cytokines are increased in the circulation in response to intense concentric and eccentric muscle contractions. They are proposed to play a role in tissue remodeling, especially in response to muscle damage (Pedersen et al. 2001, 2003; Steensberg et al. 2000). While the immune response to aerobic training and susceptibility to upper respiratory tract infections has received great attention (Pedersen et al. 2001, 2003), much less is known about the acute changes in circulating cytokines induced by a single resistance training session or following a number of training sessions (Pedersen et al. 2001, 2003; Steensberg et al. 2000).

Prior studies have demonstrated that acute resistance exercise transiently elevates circulating concentrations of anabolic/catabolic hormones (i.e., testosterone, growth hormone (GH), cortisol), and cytokines including interleukin (IL) IL-1, IL-6, IL-1β, IL-1 receptor antagonist (IL-1ra) and IL-10 (Evans et al. 1986; MacIntyre et al. 2001; Takarada et al. 2000). The greatest testosterone and cortisol responses are observed during large muscle-mass exercise and high-volume sessions performed at moderate to high intensity, using short resting intervals between sets (Kraemer and Ratamess 2005). With the controversy of the role of testosterone and cortisol in circulation, one of its primary targets might be the modulating activities of immune cells in circulation. In vitro evidence demonstrates that testosterone may suppress the expression of the proinflammatory cytokines TNF α, IL-1ß, and IL-6 (D'Agostino et al. 1999) and potentiate the expression of the antiinflammatory cytokine IL-10 (Bebo et al. 1999). Furthermore, cytokine IL-6 release in response to exercise may also be partly responsible for increases plasma cortisol in a similar manner (Steensberg 2003). However, the extent to which there is an interaction between the endocrine and cytokine responses in trained and untrained individuals remains to be elucidated.

It is a well-established principle of training that progressive overload (e.g., increasing volume and intensity) is necessary to increase muscular strength and that for adaptations to occur, a stimulus exceeding a previous stimulus needs to be applied during a resistance training program (Gonzalez-Badillo et al. 2006; Izquierdo et al. 2006; Kraemer and Ratamess 2005). Conceptually, this would suggest that the stress-related overload in the context of a short-term resistance training cycle may be progressively increased in each training session (Gonzalez-Badillo et al. 2006; Izquierdo et al. 2006). However, the role played by the absolute load and the relative load in the acute hormone and cytokine response to resistance training remains to be determined. Therefore, the purposes of the present study were (1) to determine if the hormone and cytokine responses to a standardized training session are augmented after 7 weeks of heavy resistance training, (2) and to determine the role played by the relative load of the training session in the hormone and cytokine responses. As a result, we determined the *total and free testosterone, cortisol,* GH, insulin-like growth factor-1 (IGF-1), and IGF binding protein-3 (IGFBP-3), interleukin-1 receptor antagonist (IL-1ra), IL-1 β, IL-6, and IL-10 plasma responses and we related them to neuromuscular and metabolic (i.e., blood lactate) changes, after two loading protocols performed with the same relative intensity (% of 1 RM) as well as the same absolute load (kg) before and after training.

Methods

Experimental design

A longitudinal randomized research design was used during the early phase of heavy resistance training (7 weeks) to compare the neuromuscular and hormonal responses, and their recovery profiles elicited by two loading protocols with the same relative load (%) and the same absolute load (kg) as in pretraining. The experimental design consisted of three acute heavy-resistance exercise protocols (AHREP). One of them was performed in pretraining (5 × 10 RM leg press) and the other two after the 7-week experimental resistance training period. Baseline testing was completed

during the first 3 weeks of the study preceding the start of the training program. The two AHREP sessions performed after training were randomized and separated by 7 days. They were performed with the same relative load (%) (i.e., new 10 RM load) and the same absolute load (kg) as in pre-training (see Fig. 1). The volunteers were familiarized with the testing procedures about 2 weeks before the AHREP session. After a thorough familiarization session, the subjects participated in a control testing day 1 week before the AHREP to determine one repetition maximum (1 RM), maximal voluntary contraction (MVC), muscle power, and the load corresponding to 10 RM (Fig. 1).

Immediately before each AHREP (pre-exercise) session each subject's 1 RM and MVC were determined. Muscle power was assessed with the load corresponding to pre-exercise 10 RM (i.e., control). After each AHREP, i.e., in the fatigued state, the MVC and muscle power with the load corresponding to 10 RM was performed immediately post-exercise (post 0) (Fig. 1) (Izquierdo et al. 2009). Each subject was required to have a 3-day food diary record for the days prior to testing and repeat the same diet before each main trial in order to minimize the variation in physiological responses. In addition, subjects did not ingest any food except water for 1 h prior to the experimental procedures. In order to exclude any residual effects of previous exercise on the experimental treatment, the subjects were also required to refrain from strenuous exercise and the consumption alcohol, tobacco or caffeine 48 h before and between the testing sessions.

Subjects

Twelve physically active healthy men volunteered to participate in the study. The subjects' mean (±SD) age, height, body mass, and percentage of body fat were 33 (±4.4) years, 1.77 (±0.06) months, 72.4 (±6.9) kg and 9.2% (±2.5), respectively. In order to document the initial physiologic changes we used subjects with no weight training experience. This enabled the observation of the acute physiologic changes that took place in a resistance training program. Each subject gave his written informed consent to participate after the risks involved in the study were explained in detail. The experimental procedures were approved by the Institutional Review Committee of the Instituto Navarro del Deporte and were in accordance with the Declaration of Helsinki. Before inclusion in the study, all subjects were medically screened by a physician and were seen to be free from any orthopedic, electrocardiographic, endocrinal, or medical problems that would contraindicate their participation or influence the results of the investigation. None of them was taking exogenous anabolic-androgenic steroids, drugs, medication, or dietary supplements with potential effects on physical performance.

Acute heavy resistance loading protocols

The experimental design comprised the examination of resistance training-induced adaptations on the acute neuromuscular, hormonal, and cytokine responses with the same absolute (kilogram) and relative load (% of 1 RM). Before training, the AHREP consisted of five sets with the maximum load possible to achieve ten repetitions (10 RM) in leg press with 120 s of rest between the sets. After training each subject performed the two AHREP in random and balanced order separated by 7 days: one with the same relative load ($5 \times 10\ RM_{Rel}$) and the other with the same absolute load ($5 \times 10_{Abs}$) as in pretraining testing. If the subject failed to reach the tenth repetition during the $5 \times 10\ RM_{Rel}$,

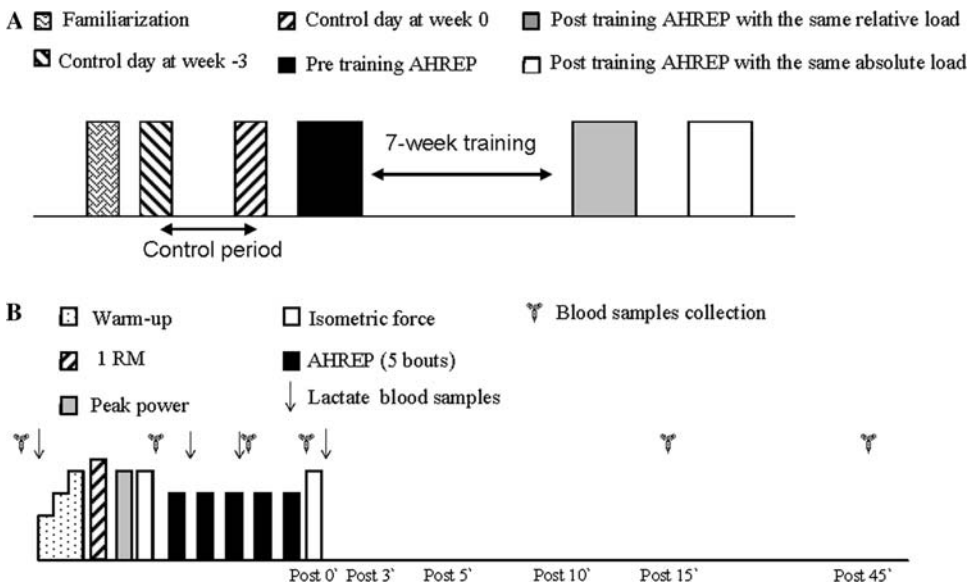

Fig. 1 Acute heavy resistance protocol design (**a**) and longitudinal experimental design (**b**)

the load was reduced and the exercise resumed allowing the completion of ten repetitions on each set. This maneuver was repeated as many times as necessary so that each set always consisted of ten repetitions. For comparison purposes, the 5 × 10 RM_{Abs} AHREP performed after the resistance training period was carried out with the same absolute loads used in pretraining testing.

A bilateral leg press exercise machine (i.e., leg press action in a sitting position) (Technogym, Gambettola, Italy) was used for all trials. The seat was individually adjusted to minimize displacement between the lower back and the backrest during muscular force exertion. Strong verbal encouragement was given to all subjects to motivate them to perform each test action as maximally and as rapidly as possible.

Maximal strength and muscle power output

1 RM (i.e., the heaviest load that could be lifted only once using the correct technique) was determined for the leg press exercise (Technogym, Gambettola, Italy). The subject was in a seated position so that the knee angle was 90º. Three to four subsequent attempts were made to determine 1 RM. The rest between maximal attempts was always 2 min.

Maximal isometric force was also measured on a modified leg press exercise machine (Technogym, Gambettola, Italy) at knee and hip angles of 90º and 45º, respectively. The exercise machine incorporated several force transducers on a foot platform located below the subject's feet. The strain gauges recorded the applied force (N) to an accuracy of 1 N at 1,000 HZ.

Muscle power output of the leg extensor muscles was measured during the concentric phase of leg press using the load corresponding to 10 RM. An optical encoder (Computer Optical Products Inc, California, USA) was attached to weight plates to record the position and direction of the displacement to an accuracy of 0.2 mm at 1,000 Hz. Customized software was used to calculate range of motion, peak power output, and average velocity for each repetition. Subjects were instructed to displace the weights as fast as possible. Two testing trials were recorded and the best trial was taken for further analysis. The test–retest intraclass correlation coefficients for all strength and power variables were greater than 0.95 and the coefficients of variation (CV) ranged from 0.9 to 2.1%.

Muscle cross-sectional area (CSA) and anthropometry

The muscle CSA of the left quadriceps femoris (QF) was assessed before and after the 7-week resistance training period using magnetic resonance imaging (MRI) (SIEMENS Magnetom Impact Expert, 1 T). Once the subject was positioned within the magnet, the thighs of both legs were kept parallel to the MRI table, and the feet were strapped together to prevent rotation. The length of the femur (Lf), taken as the distance from the intercondilar notch of the femur to the superior boundary of the femoral head, was measured on a coronal plane. CSA computation was carried out on the QF as a whole. Body mass and percent body fat (estimated from the thickness of seven skinfold sites) were taken before and after each training period (Jackson and Pollock 1978).

Resistance training program

A trained researcher supervised each workout session carefully so that exercise prescriptions were correctly administered during each training session (e.g., number of repetitions, rest and velocity of movement). Compliance with the study was 100% of the programmed sessions.

Subjects trained two times per week for 7 weeks to perform dynamic resistance exercises from 45 to 60 min per session. A minimum of 2 days elapsed between two consecutive training sessions. During the whole training period, the core exercises were parallel-squat and bench press in addition to supplementary strengthening exercises for selected muscle groups (leg press, leg extension, shoulder press, lateral pull-down, abdominal crunch, trunk extension, and standing leg curl). The resistance training consisted of a nonlinear undulating, multi-set, multi-exercise, progressive program performed two times per week. The daily workouts were alternated by varying the resistance (intensity), and the volume (sets × repetitions × load) over the week. On Tuesday, the sets were performed at 12–15 RM with 2 min rest between sets. Finally, on Thursday the sets were performed at the 10 RM intensity with 2 min rest between sets. Three to five sets were performed during the training program. The assigned training intensities were gradually increased during the 7-week training period using a repetition maximum approach.

Blood collection and analysis

The subjects visited the laboratory and rested for 30 min before the first blood collection. During the loading session, blood samples were drawn from an antecubital forearm vein using a 20-gauge needle and vacutainers® for the determination of serum total an free testosterone, cortisol, GH, insulin-like growth factor-1 (IGF-1), and IGF binding protein-3 (IGFBP-3), interleukin-1 receptor antagonist (IL-1ra), IL-1β, IL-6, and IL-10 concentrations pre-exercise, after the third set (mid-exercise), immediately after (post), and after 15 min (post-15 min) and 45 min (post-45 min) after the loadings. During the control day, i.e., without

exercise, two blood samples were also drawn within 30 min at the same time of day when the loading protocols were carried out. Blood samples were obtained at different times of the day from subjects but at the same time of day for each subject in each loading protocol before and after the experimental training period, to minimize the influence of any diurnal variation.

Whole blood was centrifuged at 3,000 rpm (4°C) for 15 min and the resultant serum was then removed and stored at −20°C until subsequent analysis. All samples were assayed in duplicate and were decoded only after the analyses were completed (i.e., blinded analysis procedure). Circulating concentrations of total testosterone, cortisol, GH, IGF-1, IGFBP-3, IL-1ra, IL-1β, IL-6, and IL-10 were determined using commercially available enzyme-linked immunosorbent assay (ELISA) kits (DRG Diagnostics, DRG Instruments GmbH, Marburg, Germany). Because no significant differences in plasma volume changes were observed between the loading conditions before and after training, hormonal concentrations were not corrected for plasma volume changes (Dill and Costill 1974). Samples were only thawed once before the analysis for all procedures,

Capillary blood samples for the determination of blood lactate concentrations were obtained from a hyperemized earlobe pre-exercise, as well as after exercise to determine the peak blood lactate concentration post exercise (i.e., post-exercise (post 0), and 3, 5, 10, 15 min into the recovery period). Samples for whole blood lactate determination (100 µl) were deproteinized, placed in a preservative tube (YSI 2315 Blood Lactate Preservative Kit), stored at 4°C, and analyzed (YSI 1500) within 5 days of completing the test. The blood lactate analyzer was calibrated after every fifth blood sample dose with three known controls (5, 15, and 30 mmol l^{-1}).

Statistical analyses

The training-related effects were assessed using a two-way ANOVA with repeated measures (time × protocol). When a significant F value was achieved, Bonferroni post hoc procedures were performed to locate the pairwise differences between the means. Statistical comparison during the control period (from week –3 to week 0) was done by Student's paired t test. Selected absolute and relative changes (e.g., maximal strength, muscle power output, acute cytokine and hormonal adaptations) were analyzed via one-way analysis of variance. Statistical power calculations for this study ranged from 0.75 to 0.80. The $P \leq 0.05$ criterion was used to establish statistical significance. Values are reported as mean values ± and standard deviations (SDs).

Results

Anthropometry and muscle CSA

After the 7-week training period, a significant increase was observed in body mass (from 72.4 ± 6.9 to 73.4 ± 6.2 kg, $P < 0.05$), and quadriceps CSA (from 130.1 ± 10.6 to 135.7 ± 12.2 cm^2, $P < 0.001$), whereas no significant changes were observed in body fat.

Maximal bilateral isometric, 1 RM dynamic strength and muscle power output

Maximal strength and muscle power output remained unaltered during the 3-week control period (from week −3 to week 0). During the 7-week training period, significant increases of 10.8 ± 1.3% ($P < 0.05$) were recorded in maximal isometric force (from 1557 ± 211 to 1772.7 ± 304 N) and 19.7 ± 4.7% in 1 RM (from 190.6 ± 30.2 to 237.9 ± 38.8 kg).

After the 7-week training period, peak power output with the same absolute load as in pretraining increased by 16.2 ± 12.8% (from 1163.2 ± 239.9 to 1414.4 ± 372.0 W, $P < 0.01$), whereas no significant changes were observed in peak power output with the same relative load used in pretraining (from 1163 ± 239 to 1159 ± 320 W).

Acute heavy 10 RM resistance loading

After the 7-week training period, the initial load of the $5 \times 10_{Rel}$ loading protocol was increased from 160.2 ± 26.3 to 198.9 ± 33.9 kg ($P < 0.05$). In pretraining, the load used during the 10 RM loading protocol corresponded to 84.1 ± 4.8% of 1 RM (ranging from 75.1 to 90.5%) similar to that of 83.7 ± 4.8% of 1 RM (ranging from 74.9 to 91.1%) used in posttraining. After training, total work (sets × reps × load) performed with the same relative load ($5 \times 10_{Rel}$ loading) was increased by 15.5 ± 6.6% compared with that used in pretraining (from 7515.7 ± 1040.1 to 8939.6 ± 1340.6 kg, $P < 0.05$). After training, the load used during the $5 \times 10\ RM_{Abs}$ represented 67.6 ± 5.7% of the posttraining 1 RM. Total work (sets × reps × load) performed with the same absolute load ($5 \times 10\ RM_{Abs}$ loading) was similar in pre and posttraining (7515.7 ± 1040.1 kg and 7521 ± 1039.1 kg, respectively). After training, the relative decrease (%) of the load during the $5 \times 10_{Rel}$ protocol was higher ($P < 0.05$) for the second, fourth, and fifth sets compared with that recorded in pretraining. For comparison purposes, after training the absolute decrease of the load during the $5 \times 10_{Abs}$ loading protocol was matched to that recorded in pretraining.

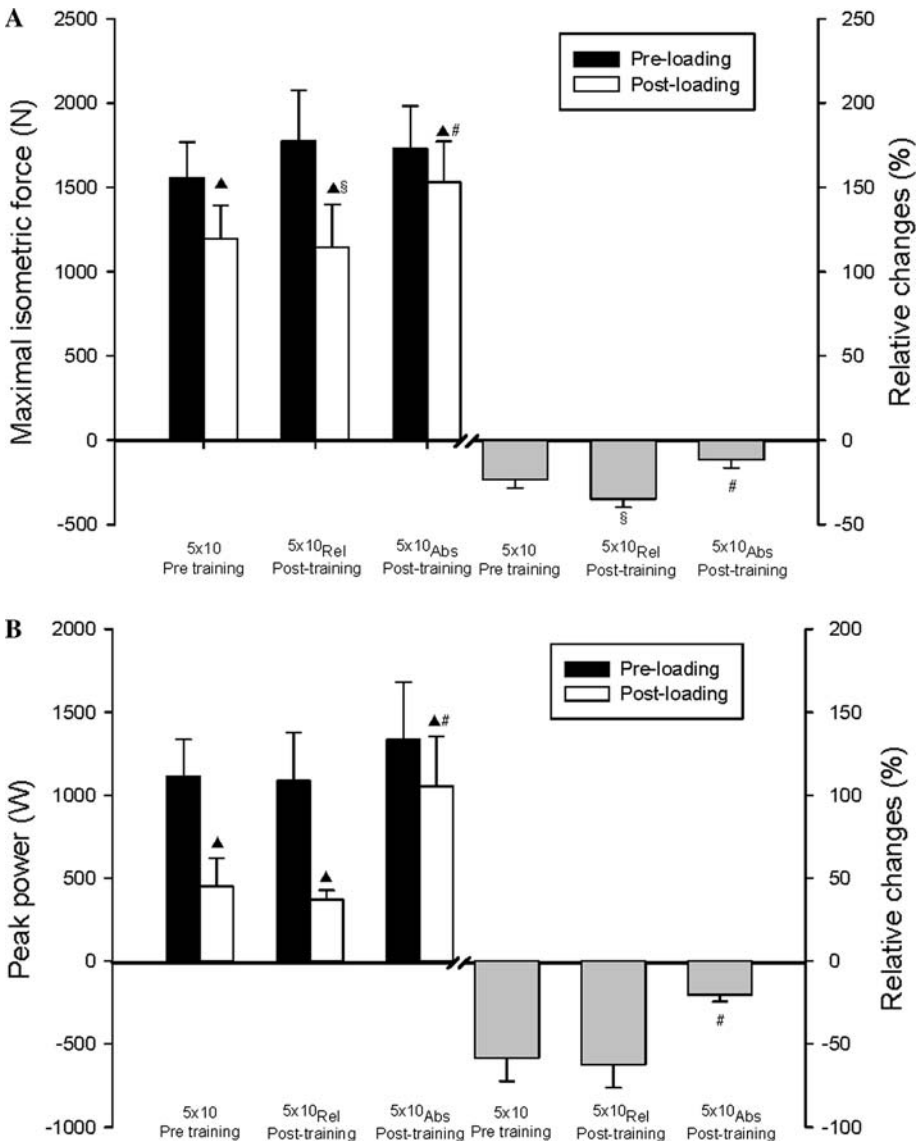

Fig. 2 Absolute and relative changes in maximal isometric force (**a**) and peak power output (**b**), pre and post loading before at pretraining (5×10 pretraining) and after 7 weeks of periodized strength training with both the same relative ($5 \times 10_{Rel}$) and the same absolute load ($5 \times 10_{Abs}$) as pretraining. *Filled triangle* significant difference ($P < 0.05$) compared to the corresponding pre-exercise value, for each protocol. $^{\#}P < 0.05$ significant differences between $5 \times 10_{Abs}$ post training and the other two protocols. $^{\S}P < 0.05$ significant differences between 5×10 pretraining and $5 \times 10_{Rel}$ post-training

Acute maximal isometric and muscle power output responses to training

Maximal isometric force immediately after the $5 \times 10_{Rel}$ loading was decreased by 23.4 ± 11.7 and $34.2 \pm 15.8\%$ (both $P < 0.05$), but with a significantly greater exercise-induced loss of MVC after training (pre vs. posttraining) (Fig. 2a). Training attenuated the losses in MVC elicited by the $5 \times 10_{Abs}$ loading protocol (11.4%) compared to those elicited by the $5 \times 10_{Rel}$ loading protocol (Fig. 2a).

Peak power output decreased in a similar manner ($P < 0.05$) immediately after the $5 \times 10_{Rel}$ loading protocols in both pretraining and posttraining (58.4 ± 14.5 and $62.3 \pm 14.4\%$, respectively) (Fig. 2b). After training, significant decreases were observed in peak power output immediately post-exercise in the $5 \times 10_{Abs}$ loading protocol (20.3%) (Fig. 2b). The $5 \times 10_{Abs}$ loading protocol elicited significant lower reductions in peak power output after training than both $5 \times 10_{Rel}$ loading protocols.

Blood lactate concentrations

After training, post exercise peak blood lactate was significantly greater in the $5 \times 10_{rel}$ compared with that observed in the 5×10 pretraining protocol. The blood lactate response was lower ($P < 0.05$) during the whole loading period in the $5 \times 10_{Abs}$ compared with the response observed in the $5 \times 10_{Rel}$ (Fig. 3).

Acute hormonal responses to training

Basal hormonal levels remained unaltered during the 3-week control period (from week −3 to week 0). There were no significant changes between the three control blood

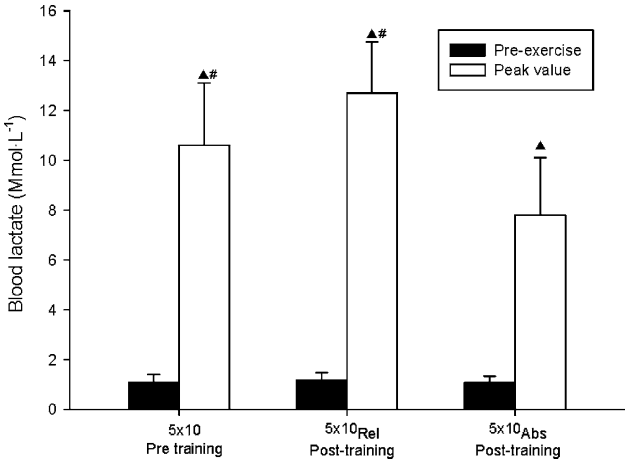

Fig. 3 Blood lactate concentrations (Mmol L^{-1}) pre-exercise and peak value at pretraining (5 × 10 pretraining) and after 7 weeks of periodized strength training with both the same relative ($5 \times 10_{Rel}$) and the same absolute load ($5 \times 10_{Abs}$) as pretraining. *Filled triangle* significant differences ($P < 0.05$) from the corresponding pre-exercise value. #Significant differences ($P < 0.05$) with the other two protocols

samples drawn within 30 min on the control day (i.e., without exercise) at the same time at which each subject had performed the two loading protocols before and after training.

Serum total and free testosterone concentrations increased significantly after the loading both before and after the strength training period, but decreased to the pre-exercise value at 45 min post-exercise. Acute exercise-induced serum total testosterone responses to $5 \times 10_{Abs}$ or $5 \times 10_{Rel}$ loading protocols were similar in pre and post-training (Fig. 4a). After training, the mean acute free testosterone response was greater ($P < 0.05$, mid exercise and post exercise) in $5 \times 10_{Rel}$ compared with that observed in the $5 \times 10_{Abs}$ protocol and the $5 \times 10_{Rel}$ at pretraining (Fig. 4b).

Serum cortisol concentration increased significantly 15 and 45 min after $5 \times 10_{Rel}$ pretraining and $5 \times 10_{Rel}$ post-training (Fig. 5), the increment being similar in both conditions. Acute cortisol responses were significantly lower during the whole loading and recovery period in $5 \times 10_{Abs}$ compared with the responses observed in both $5 \times 10_{Rel}$ loading protocols (pre and posttraining) (Fig. 5).

Serum GH concentration was significantly increased post-exercise as well as after 15 and 45 min during the recovery period. This increase was similar during both relative loading sessions performed before ($5 \times 10_{Re}$ pretraining) and after the training period ($5 \times 10_{Rel}$ posttraining). After training, no significant changes were observed in any

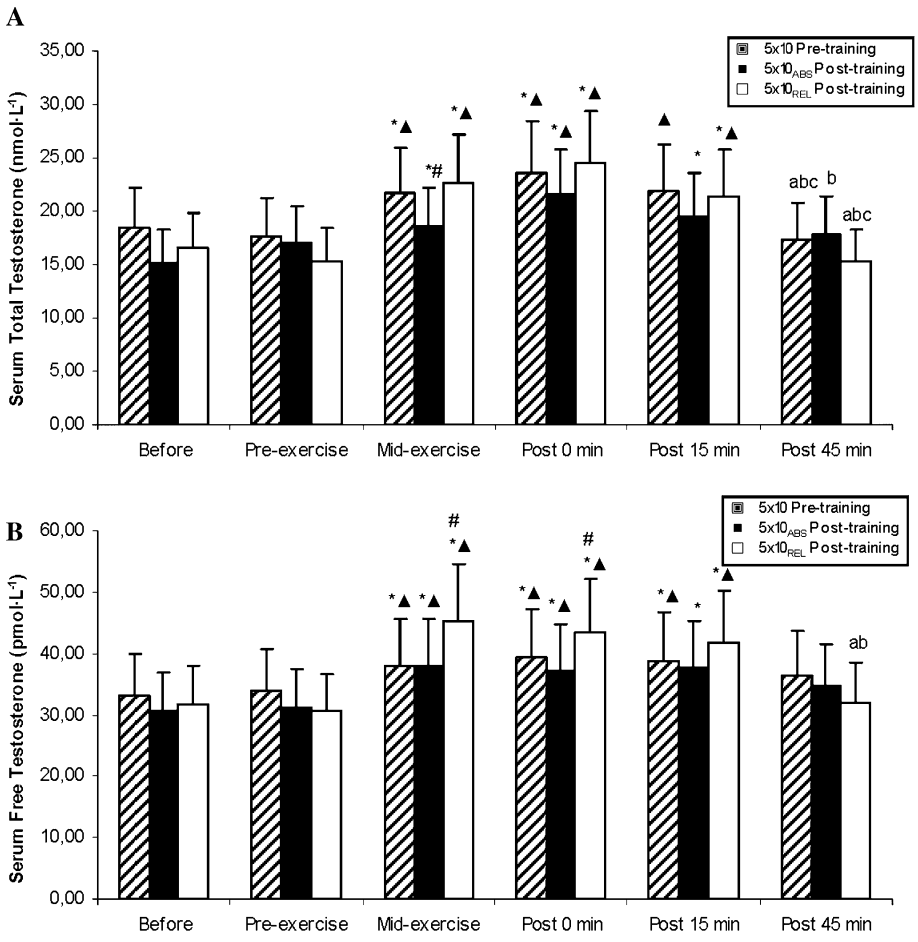

Fig. 4 Serum total testosterone (**a**) and free testosterone (**b**) concentrations before, pre-, mid-, and post-exercise and during recovery at pretraining (5 × 10 pretraining) and after 7 weeks of periodized strength training with both the same relative ($5 \times 10_{Rel}$) and the same absolute load ($5 \times 10_{Abs}$) as pretraining. *Filled triangle* significant difference ($P < 0.05$) compared to the corresponding pre-exercise value, for each protocol. [a]Significant differences ($P < 0.05$) from the corresponding post 0 value, for each protocol. [b]Significant differences ($P < 0.05$) from the corresponding post 3 min value, for each protocol. [c]Significant differences ($P < 0.05$) from the corresponding post 5 min value, for each protocol. #$P < 0.05$ significant differences between $5 \times 10_{Abs}$ post training and the other two protocols

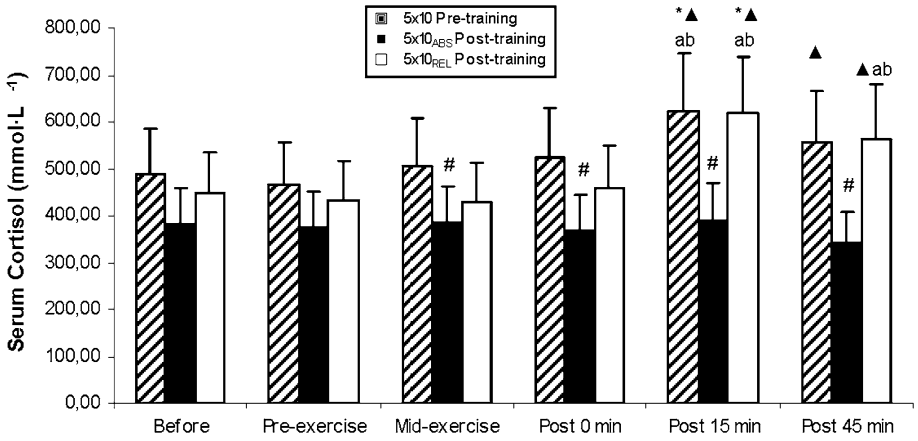

Fig. 5 Serum cortisol concentration before, pre-, mid- and post-exercise and during recovery at pretraining (5 × 10 pretraining) and after 7 weeks of periodized strength training with both the same relative ($5 \times 10_{Rel}$) and the same absolute load ($5 \times 10_{Abs}$) as pretraining. *Filled triangle* significant difference ($P < 0.05$) compared with the corresponding pre-exercise value, for each protocol. [a]Significant differences ($P < 0.05$) from the corresponding post 0 value, for each protocol. [b]Significant differences ($P < 0.05$) from the corresponding post 3 min value, for each protocol. [#]$P < 0.05$ significant differences between $5 \times 10_{Abs}$ post training and the other two protocols

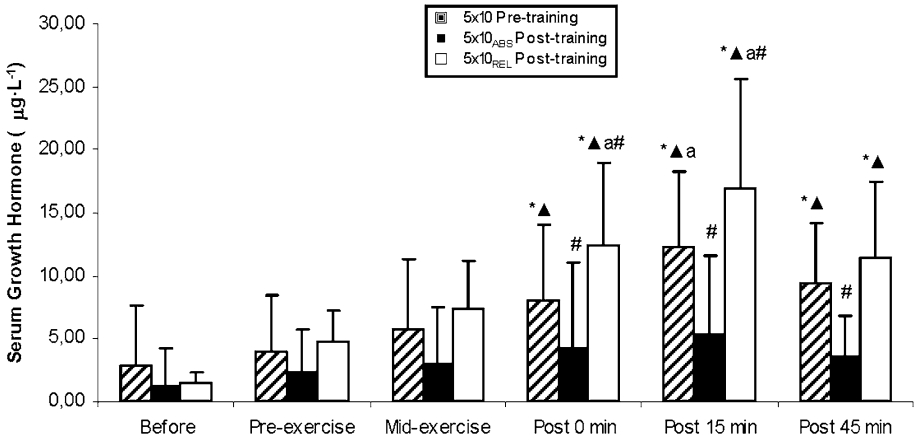

Fig. 6 Serum growth hormone concentration before, pre-, mid-, and post-exercise and during recovery at pretraining (5 × 10 pretraining) and after 7 weeks of periodized strength training with both the same relative ($5 \times 10_{Rel}$) and the same absolute load ($5 \times 10_{Abs}$) as pretraining. *Significant difference ($P < 0.05$) compared to the corresponding before value, for each protocol. *Filled triangle* significant difference ($P < 0.05$) compared to the corresponding pre-exercise value, for each protocol. [a]Significant differences ($P < 0.05$) from the corresponding post 0 value, for each protocol. [#]$P < 0.05$ significant differences between $5 \times 10_{Abs}$ post training and the other two protocols

loading or recovery points of the loading session performed with the same absolute load as in pretraining ($5 \times 10_{Abs}$ protocol). After the training period, acute GH response was significantly greater in the $5 \times 10_{Rel}$ protocol, compared with those recorded during the $5 \times 10_{Rel}$ pretraining (i.e., post exercise and post 15 min) and $5 \times 10_{Abs}$ posttraining loading sessions (i.e., post exercise, post 15 min and 45 min). Furthermore, the acute GH responses were significantly lower after the $5 \times 10_{Abs}$ protocol (i.e., post exercise, 15 and 45 min post-exercise) compared with the responses observed during the $5 \times 10_{Rel}$ loading sessions (pre and posttraining) (Fig. 6).

No significant changes were observed in the acute IGF-1 and IGFBP-3 responses at any point ($P < 0.05$, mid-exercise, post-exercise, and 45 min after recovery) during the loading or recovery period during both pretraining and posttraining loading sessions ($5 \times 10_{Abs}$ or $5 \times 10_{Rel}$). No significant differences were observed in the acute IGF-1 and IGFBP-3 responses, regardless of whether $5 \times 10_{Abs}$ or $5 \times 10_{Rel}$ protocols were performed after the training period.

Cytokines

After the training period, the acute IL-1 β response was significantly higher during the whole loading and recovery period in both $5 \times 10_{Abs}$ and $5 \times 10_{Rel}$ protocols, compared with the acute response recorded during the 5×10 RM protocol in pretraining.

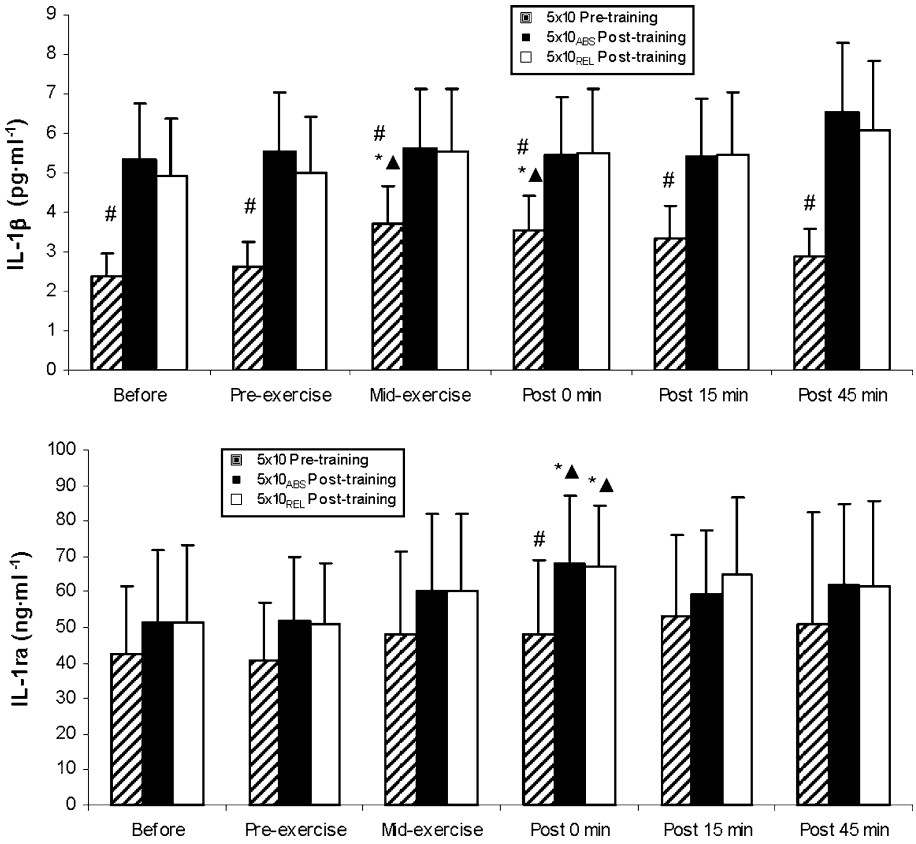

Fig. 7 Interleukins IL-1 β (**a**) and IL-ra (**b**) serum concentrations before, pre-, mid- and post-exercise and during recovery at pretraining (5×10 pretraining) and after 7 weeks of periodized strength training with both the same relative ($5 \times 10_{Rel}$) and the same absolute load ($5 \times 10_{Abs}$) as pretraining. *Significant difference ($P < 0.05$) compared to the corresponding before value, for each protocol. *Filled triangle* significant difference ($P < 0.05$) compared to the corresponding pre-exercise value, for each protocol. $^{\#}P < 0.05$ significant differences between $5 \times 10_{Abs}$ post training and the other two protocols

IL-1ra concentrations significantly increased at mid- and post-exercise point, but decreased to the pre-exercise value at 15 and 45 min after recovery during the $5 \times 10_{Rel}$ pretraining loading protocol. After training, no significant changes were observed IL-1β and IL-1ra ($P < 0.05$; for IL-1ra at Post 0 min) concentrations in both relative loading protocols (Fig. 7a, b).

Serum IL-6 concentration significantly increased at 45 min post-exercise in both pretraining and posttraining $5 \times 10_{Rel}$ protocols. After the training period, the acute IL-6 response in the $5 \times 10_{Rel}$ protocol in the 45-min recovery period was significantly greater compared with the acute responses recorded during 5×10 pretraining and posttraining $5 \times 10_{Abs}$ loading conditions (Fig. 8a).

No significant changes were observed in acute serum IL-10 concentrations at any loading or recovery point of the loading sessions performed in pretraining and posttraining with the same absolute load ($5 \times 10_{Abs}$ protocol). After training, acute serum IL-10 concentration significantly increased at mid- and post-exercise, as well as after 15 and 45 min during the recovery period in the $5 \times 10_{Rel}$ protocol. After the training period, the acute IL-10 response in the $5 \times 10_{Rel}$ protocol was significantly greater in mid- and post-exercise as well as during the 15 and the 45 min recovery period, compared with the acute responses recorded during pretraining 5×10 RM and posttraining $5 \times 10_{Abs}$ loading conditions (Fig. 8b).

Discussion

The mains findings of this study were that 7 weeks of heavy resistance training induced (1) greater magnitude of acute exercise-induced MVC loss than in pretraining, when exercising with the same relative intensity, (2) similar acute responses in serum cortisol concentration but increased free testosterone and GH responses, to the same relative load, (3) greater IL-6 (pro-inflammatory) and IL-10 (anti-inflammatory) responses after exercising with the same relative load, as well as (4) greater release of pro-inflammatory cytokines IL-1β and IL1-ra after exercising either at the same relative or absolute load.

Effects on fatigue

The short-term resistance training period led to higher accumulated fatigue and metabolic demand (i.e., blood lactate accumulation) after multiple sets of dynamic fatiguing contractions with the same relative load as in pretraining. As has been previously reported (Izquierdo et al. 2009), after a short-term strength training period, when the relative intensity of the fatiguing dynamic protocol was kept the same, the magnitude of exercise-induced loss of functional capacity (power and MVC) was greater than that observed before training, but lower fatigue occurred when the same absolute load was used. Previous studies have demonstrated

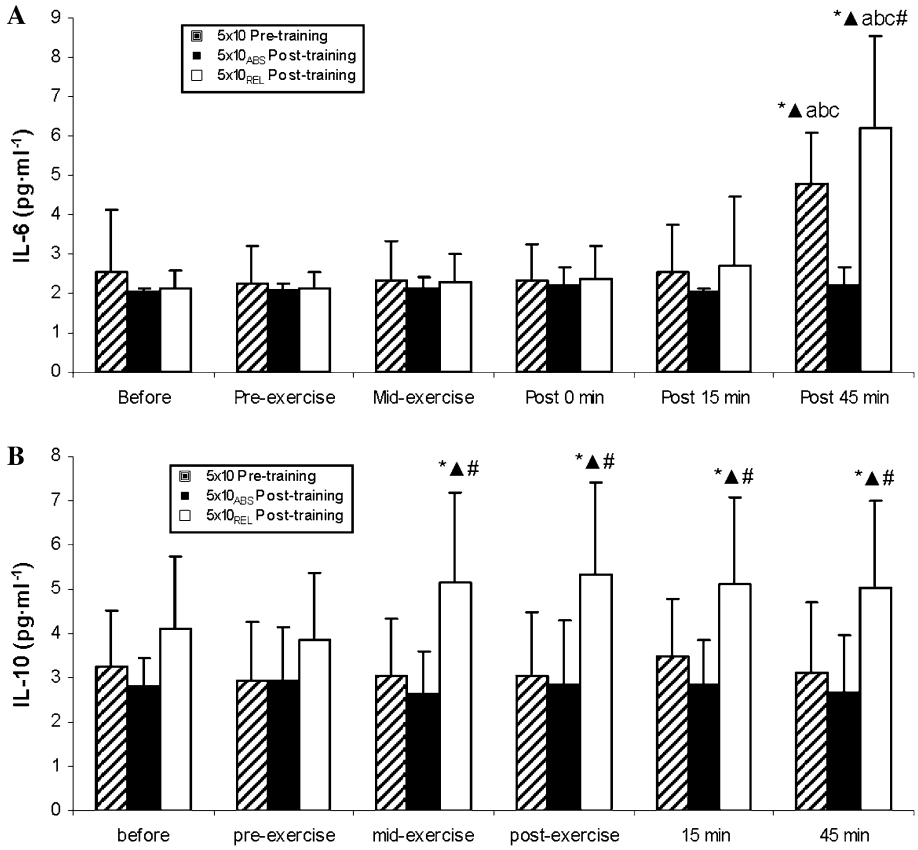

Fig. 8 Interleukins IL-6 (**a**) and IL-10 (**b**) serum concentrations before, pre-, mid- and post-exercise and during recovery at pretraining (5×10 pretraining) and after 7 weeks of periodized strength training with both the same relative ($5 \times 10_{Rel}$) and the same absolute load ($5 \times 10_{Abs}$) as pretraining. *Significant difference ($P < 0.05$) compared with the corresponding before value, for each protocol. *Filled triangle* significant difference ($P < 0.05$) compared with the corresponding pre-exercise value for each protocol. $^{\#}P < 0.05$ significant differences between $5 \times 10_{Abs}$ post training, and the other two protocols

similar decreases in maximal isometric strength in physically active and strength-trained male athletes (Ahtiainen et al. 2003), when the relative intensity of the loading was kept the same before and after a long-term strength training period (21 weeks), whereas in other studies the magnitude of exercise-induced loss in maximal strength or muscle power was not reported (Hickson et al. 1994; Kraemer et al. 1998; Kraemer et al. 1999; McCall et al. 1999). Furthermore, as expected, resistance training resulted in a remarkable increase in exercise performance (i.e., reduced fatigability) as shown by the attenuation of the degree of fatigue elicited by the same absolute load after training.

Hormonal responses

A limited number of studies have examined the effects of resistance training on acute exercise-induced hormonal responses with the same relative and absolute intensity as in pretraining. The increased free testosterone and GH response to acute exercise with the same relative load after progressive heavy resistance training is in accordance with other studies (Craig et al. 1989; Kraemer et al. 1998). Craig et al. (1989) reported that young men increased GH concentration up to fivefold in response to an acute bout of resistance exercise and this increased up to sixfold after training, whereas the acute total testosterone response to a single lifting session was the same before and after training. Kraemer et al. (1998) reported that untrained men are able to develop an acute exercise-induced increase in testosterone, but a similar GH response in pre and posttraining. However, other studies did not find any significant changes in the profiles of acute hormonal responses to resistance exercise due to long-term strength training in adult men (Ahtiainen et al. 2005; Hickson et al. 1994; Kraemer et al. 1999; McCall et al. 1999). Comparison of our results with other studies is difficult because the magnitude of exercise-induced loss in maximal strength or muscle power has not been reported systematically.

To the author's knowledge only Ahtiainen et al. (2003) reported similar exercise-induced decreases in maximal isometric strength pre and posttraining after the 21-week training period. This was accompanied by no significant differences in acute cortisol, total and free testosterone responses between strength-trained and untrained men between the loading sessions, but an attenuated acute GH response in the male strength-athlete group. Resistance training may also have led to an overall reduction (Kraemer et al. 1999; Staron et al. 1994) or similar (Ahtiainen et al. 2003; Ahtiainen et al. 2005) cortisol responses to exercise loading in men. In the present study, after the 7-week strength training period, when the relative intensity of the fatiguing dynamic protocol was kept the same, the acute

cortisol response to resistance exercise was similar to that observed before training. These results may indicate that differences in GH and testosterone responses to acute exercise after training may not only be sensitive to the relative intensity but also to other factors related to the higher absolute intensity of the exercise after training, e.g., greater metabolic demand. It maybe that this metabolic demand plays a primary role in these circulating hormonal concentrations.

A unique but expected finding of this study was fewer signs of acute exercise-induced fatigue (i.e., reduced fatigability) in the protocol in posttraining with the same absolute load as in pretraining, accompanied by an attenuated release in the cortisol and GH response but interestingly similar total and free testosterone acute responses after training. This could be due to the decreased stress response and/or decreased hormone production. A typical response to resistance training is a reduction of the amount of muscle mass needed to perform a given dynamic task, as demonstrated using NMR and EMG measurements (Lewis et al. 1984; Ploutz et al. 1994). Thus, the same amount of strength and power can be developed after training using fewer muscle fibers with lesser accumulation of fatigue. These two effects can only be explained by changes in neural activity combined with local muscular functional and structural changes. Strength training leads to improvements in neuromuscular efficiency (i.e., the relationship between EMG and mechanical power output) (Behm and St-Pierre 1998), muscle mass (Aagaard et al. 2001; Hakkinen et al. 1985), selective hypertrophy of IIA muscle fibers (Staron et al. 1994) and enhanced muscle metabolism (i.e., increased activity of creatine kinase (CK), glycolytic enzymes, e.g., phosphofructokinase (PFK), decreases in lactate accumulation, and increases in muscle buffering capacity) (Hellsten et al. 1996; McKenna et al. 1993; Mohr et al. 2007; Sahlin and Henriksson 1984). Changes of this nature could have occurred in our subjects as a result of resistance training and hence contribute to the explanation of reduced fatigability and attenuated acute hormonal response to acute loading with the same absolute load as in pretraining. This finding implies that it may be necessary to adjust the load to achieve similar acute hormonal responses after a short-term training period. Therefore, similar relative intensity may induce not only higher exercise-induced fatigue but also increased acute hormonal responses during the initial phase of the resistance training period.

Cytokine responses

Resistance exercise is known to cause myofibrillar disruption, especially during eccentric muscle actions (Bruunsgaard et al. 1997; Gibala et al. 2000). This disruption results in an inflammatory response, modulated by cytokines (Pedersen et al. 2001; Pedersen et al. 2003). Interleukin-6 (IL-6) is locally produced by growing myofibers and acts as an essential regulator of satellite cell (muscle stem cell)-mediated hypertrophic muscle growth (Hiscock et al. 2004; Keller et al. 2001; Penkowa et al. 2003; Serrano et al. 2008; Vierck et al. 2000). Further, it appears that cytokines may also play a role in the repair and remodeling process of muscle (Pedersen et al. 2001, 2003; Steensberg et al. 2000). Our study shows that greater exercise-induced levels of IL-6 are only elicited in the training sessions in the initial phase of heavy resistance training when the sessions are carried out with the same relative loads. In accordance with this, IL-6 was shown to be significantly high for up to 48 hours after heavy total body eccentric resistance exercise, together with closely matched delayed onset muscle soreness (Smith et al. 2000; Suzuki et al. 2002). An earlier isokinetic eccentric-only training study also reported increases in IL-6 post exercise, but there were no significant differences between pre and posttraining levels (Croisier et al. 1999). IL-6 was elevated for up to 1 day after prolonged eccentric leg extension exercise (300 repetitions), and these elevations were significantly correlated to delayed onset muscle soreness (MacIntyre et al. 2001). Even very light eccentric exercise (20% or 1 RM leg extension) coupled with circulatory occlusion resulted in increased IL-6 response up to 3 h post exercise (Takarada et al. 2000).

Furthermore, a higher release of pro-inflammatory cytokine IL-1β in the $5 \times 10_{Abs}$ or $5 \times 10_{Rel}$ acute loading protocols was also reported after training in the present study. Previous studies reported reduced (Smith et al. 2000) or increased (Evans et al. 1986) levels of IL-1β associated with muscle damage following eccentric exercise. The discrepancies between these studies may partly result from differences in the loading regimes studied, and to the fact that IL-1β undergoes a rapid clearance rate and local accumulation at the trauma site (i.e., in this case, muscle). Furthermore, in early studies such as this one, assay did not differentiate between IL-1α and IL-1β (Evans et al. 1986). However, the concomitant greater increase of IL-1ra in response to resistance exercise (either at the same absolute or relative intensity) could have blunted the biological activities of IL-1α and IL-1β (Pedersen et al. 2001). IL-1ra is up-regulated among others by IL-6 (Pedersen et al. 2001, 2003).

The present study also shows that short-term resistance training only enhances the responsiveness of IL-10 to a resistance exercise session when exercising at the same relative load. Previous studies have shown that IL-10 is increased by catecholamines and in vivo IL-6 (Peake et al. 2005). IL-10 is increased after an acute bout of eccentric elbow flexor exercise; interestingly, this response is accentuated when the same eccentric exercise is repeated 4 weeks later (Hirose et al. 2004). The latter could contribute to an

attenuated inflammatory response to eccentric exercise and explain the known protective effect of a single session of eccentric exercise on muscle structure and function. It remains to be determined why and how the IL-10 and IL-6 responses to a resistance exercise session performed at the same relative exercise intensity are accentuated after resistance training. The interactions with circulating anabolic and catabolic hormones may provide one distinct area of research, linking these circulatory elements in an endocrine-immune cytokine cybernetic relationship. Our data reflect such a pattern of interactions.

In summary, the present resistance training program induced similar acute responses in serum cortisol concentration but increased responses in anabolic hormones of FT and GH, as well as inflammation responsive cytokine IL-6 and the anti-inflammatory cytokine IL-10, when the same relative load was used. This response was balanced by a higher release of pro-inflammatory cytokines IL-1β, cytokine inhibitors (IL-1ra) when both the same relative and absolute load was used after training. This enhanced hormonal and cytokine response to resistance exercise at a given relative exercise intensity after resistance training occurred with greater accumulated fatigue and metabolic demand (i.e., blood lactate accumulation). The magnitude of metabolic demand or the fatigue experienced during the resistance exercise session influences the hormonal and cytokine response patterns. Similar relative intensities may elicit not only higher exercise-induced fatigue but also an increased acute hormonal and cytokine response during the initial phase of the resistance training period.

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Conflict of interest statement The authors declare that they have no conflict of interest relevant to the content of this manuscript.

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