Heat Shock Protein 27 Response to Wrestling Training in Relation to the Muscle Damage and Inflammation

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Abstract

Zembron-Lacny, A, Ziemann, E, Zurek, P, and Hübner-Wozniak, E. Heat shock protein 27 response to wrestling training in relation to the muscle damage and inflammation. *J Strength Cond Res* 31(5): 1221–1228, 2017—One of the unique features of an exercise is that it leads to a simultaneous increase of antagonistic mediators. On the one hand, exercise elevates catabolic proinflammatory cytokines. On the other hand, exercise stimulates anabolic components such as heat shock proteins (HSPs), which protect against stressors. Therefore, the study was designed to evaluate the blood level of HSP27 and its relationship with muscle damage and inflammatory mediators in elite Greco-Roman wrestlers during training periods differed in type and intensity exercise. Ten male wrestlers (21.2 ± 2.1 years) were observed during the conditioning camps at preseason (January), at the beginning of tournament season (April), and during tournament season (June). Twelve healthy and untrained men (19.2 ± 0.4 years) were considered a reference group. The serum levels of inflammatory mediators and HSP27 in wrestlers were significantly different from nonathletes. In wrestlers, reactive oxygen and nitrogen species H_2O_2, NO, and 3-nitro, cytokines interleukin-1β and tumor necrosis factor α, and also HSP27 reached the highest levels at preseason (January) or tournament season (June) when the special training predominated (>30% training load) over directed training (approximately 10% training load). Creatine kinase activity also demonstrated the highest level during the same training periods (January 2,315 ± 806 IU·L^{-1}; June 3,139 ± 975 IU·L^{-1}). The regression analysis revealed the relationship of HSP27 level with muscle damage (r_s = −0.613, $p < 0.001$), and also with inflammatory mediators. The results of this study show that wrestling training modulates HSP27 level, which is significantly related with skeletal muscle damage and inflammatory response, and suggest that measure of HSP27 level can be useful diagnostic tool in biochemical assessment of athletes to increase their performance.

Key Words athletes, cytokines, hydrogen peroxide, nitric oxide

Introduction

The effectiveness of physical training depends on the training load and the optimal recovery. The imbalance between the 2 may lead to under or over-training. One of the unique features of exercise is that it leads to a simultaneous increase of different antagonistic mediators. On the one hand, exercise elevates catabolic proinflammatory cytokines, such as interleukin-1β (IL-1β) and tumor necrosis factor α (TNFα). On the other hand, exercise stimulates anabolic components such as heat shock proteins (HSPs), which protect against stressors. If the anabolic response is stronger, exercise will probably ultimately lead to an increased muscle mass and exercise adaptation (21,25,27).

Heat shock proteins are constitutively expressed in many cell types under physiological conditions and function as molecular chaperones, whereas under stress conditions, HSPs protect proteins against misfolding, aggregation, and denaturation. In addition, HSPs may directly regulate specific stress-responsive signaling pathways and may antagonize signaling cascades that result in apoptosis (17,21). Exercise-induced stress is considered to be one of the stimuli that induce HSPs expression in skeletal muscles (7). Heat shock proteins increase the exercise tolerance and participate in the cellular repair processes. The persistently high HSPs synthesis may indicate a state of inadequate regeneration even after a couple of weeks of recovery from exhaustive exercise. Among the subset of stress-responsive proteins, HSP27 is considered as a new approach to monitoring exercise training and adaptive mechanisms (21). Data from

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Table 1. Anthropometric and body composition date in wrestlers ($n = 10$) and nonathletes ($n = 12$).*

Training period	BM (kg)	Height (cm)	BMI ($kg \cdot m^{-2}$)	FM (%)	FM (kg)	FFM (kg)
Preparatory: preseason training, January	78.8 ± 12.7	176.6 ± 8.1	25.4 ± 2.1	13.3 ± 3.5	11.0 ± 4.5	66.2 ± 10.4
Competition: at the beginning of tournament season, April	79.6 ± 9.1	177.0 ± 5.8	25.3 ± 1.9	15.4 ± 4.1	12.4 ± 4.0	68.1 ± 9.9
Competition: tournament season, June	79.9 ± 8.9	176.7 ± 6.0	25.6 ± 1.9	14.8 ± 2.6	12.0 ± 3.2	65.5 ± 6.8
Nonathletes	75.4 ± 8.8	179.9 ± 6.9	23.2 ± 1.7	15.4 ± 5.5	11.9 ± 5.0	63.6 ± 5.7

*BM = body mass; BMI = body mass index; FM = fat mass; FFM = fat-free mass.

various cell types have shown that HSP27 is involved in microfilament stabilization, signal transduction, growth, differentiation, and transformation processes, and in providing protection against thermal and nitro-oxidative stress (36). Its synthesis is elevated after a high-force eccentric exercise and reduced after repeated identical bouts of exercise (33,35). Moreover, other study demonstrated that long-term endurance training may alter the specific distribution of HSP27 content in type I and type II muscle fibers, from being expressed more frequently at higher levels in type II fibers to become equally expressed in type I and type II fibers (7). Interestingly, after nondamaging treadmill running, the protein content of large HSPs increased in the human vastus lateralis, whereas HSP27 remained unchanged, suggesting that HSP27 is in fact more sensitive to structural or functional damage than the larger HSPs (20).

Cytokines play an important role in the exercise-induced immune reaction and exercise-related metabolic and cellular signal transduction; they are capable of increasing HSPs synthesis. It is possible that HSPs may act as cytokines in reaction to an exhaustive exercise and stimulate IL-1β and TNFα expression in macrophages (28). The role of IL-1β and TNFα in skeletal muscle regeneration still has not been fully explored (37). The suppression of TNFα synthesis with an anti-inflammatory drug delays muscle regeneration, but an excessive IL-1β and TNFα release may be responsible for

Table 2. Exercise intensity during training periods and creatine kinase (CK) activity.*†‡

Training period	Type of training	Training load (%)	CK initial level ($IU \cdot L^{-1}$)	CK peak level ($IU \cdot L^{-1}$)	% Increase in CK activity
Preparatory: preseason phase, January; $n = 10$	Endurance	53	553 ± 275	2,315 ± 806	407 ± 243
	Directed	9			
	Special/ wrestling	38			
Competition: at the beginning of tournament season, April; $n = 10$	Endurance	50	105 ± 21	492 ± 195	362 ± 128
	Directed	24			
	Special/ wrestling	26			
Competition: tournament season, June; $n = 10$	Endurance	55	280 ± 164	3,139 ± 975	1,213 ± 424
	Directed	10			
	Special/ wrestling	35			

*Endurance training: team games, marches and cross-country running, cross-country skiing, acrobatic exercises, climbing at ropes, pull ups, and exercises with partner.
†Directed training: intervals, toss from knees, back suples, reverse waist, and turns.
‡Special/wrestling training: elevation from the low position, keys, trolleys, throws with different amplitude of movement, and gym.

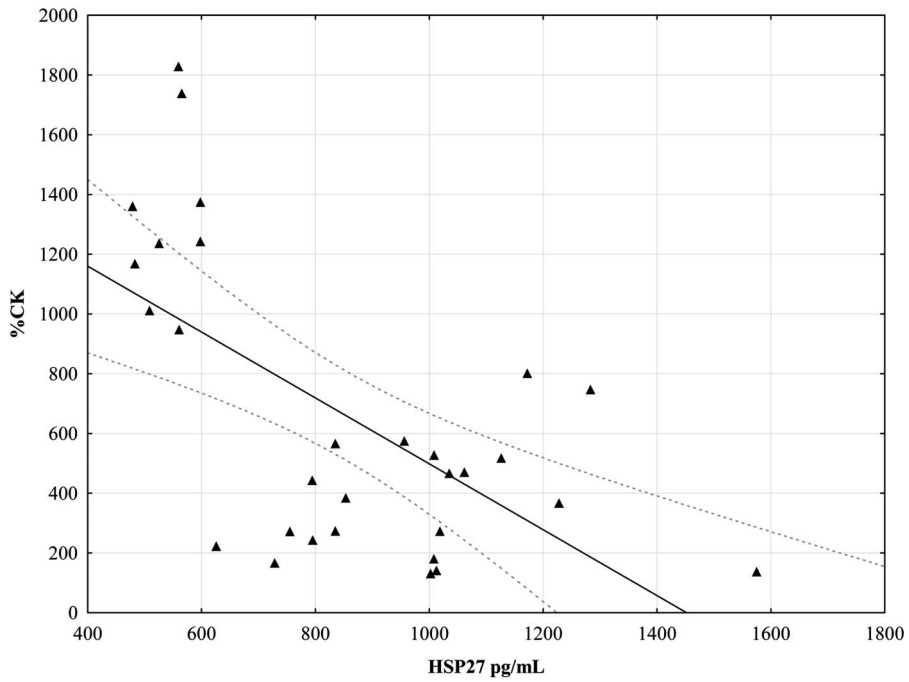

Figure 1. The significant relation between heat shock protein 27 (HSP27) concentration and percentage increase in creatine kinase (%CK) activity in wrestlers; $r_s = -0.613$, $p < 0.001$.

the overtraining syndrome (16,18,30). The measurement of proinflammatory cytokines within a population of athletes was reported in a few studies. Nowadays, it is known that proinflammatory cytokines are elevated in sport activity such as running, tennis, rowing, soccer, handball, and basketball games (3,18,19,22,26,39,41). There are some data, which indicated that appropriative designed training workload might stimulate HSP synthesis during the sport camp but stress induced by attending in competitions decreased its concentration among tennis players (41). Data about immunological response in Greco-Roman wrestlers are limited, especially in response to long-lasting training workload. Barbas et al. (1) evaluated the effects of a single-day Greco-Roman wrestling tournament on physiological responses and on performance and inflammatory status (C-reactive protein [hsCRP] and interleukin-6 [IL-6]) of elite wrestlers. They revealed that performance and inflammatory status exhibit a progressive deterioration in response to a 1-day tournament. Moreover, multiple stressors such as physical competition, fatigue, muscle damage, fluctuation in dietary intake, and psychological stress might attributed to such response (1). Yoon described physiological demands in wrestling, whereas Karnincic et al. (13) determined and compared lactate profile of 2 groups of Greco-Roman wrestlers with different competences and training experience. Still there are no reports about the levels of inflammatory mediators and HSP27 in combat sports, which consist of high-force eccentric exercises interspersed with brief periods of mild-to-moderate intensity work. Thus, we designed the study to evaluate the blood level of HSP27 and its relationship with muscle damage and inflammatory mediators in elite Greco-Roman wrestlers during training periods differed in type and intensity of exercise. We tried to find out whether the release of inflammatory mediators and HSP27 into circulation is related to training load in combat sports. The second question of our study was whether inflammatory mediators and HSP27 can be useful diagnostic tools in biochemical assessment of athletes.

Methods

Experimental Approach to the Problem

Ten male Greco-Roman wrestlers, members of national team at the age of 21.2 ± 2.1 years (Table 1), were observed during preparatory training period (preseason, January) and competition period (at the beginning of tournament season, April; in-season, June) differed in type and intensity exercise. They participated in sport camp 14 days in January, 14 days in April, and 12 days in June. All the camps were organized at the National Olympic Sport Centre in Poland. During the camps, all athletes lived in the same accommodations and followed the same training schedule and diet. Daily, energetic value of food offered in the menu did not exceed 5,200 kcal, and the protein dose varied from 1.6 to 1.8 $g \cdot kg^{-1}$ of body mass. During each camp, the wrestlers consumed an isotonic sports drink Vitargo (osmolality 317 $mOsm \cdot kg^{-1}$

Table 3. Training effect on level of hydrogen peroxide (H_2O_2), nitric oxide (NO), 3-nitrotyrosine (3-nitro), proinflammatory cytokines (interleukin-1β [IL-1β] and tumor necrosis factor α [TNFα]) and heat shock protein 27 (HSP27).

	Non-athletes	*p* value*	Preparatory, January	Competition, April	Competition, June	*p* value†
H_2O_2 ($\mu mol \cdot L^{-1}$)	28.16 ± 5.86	>0.05 <0.01 <0.01	23.05 ± 6.74	15.72 ± 3.88	14.57 ± 1.97	<0.05 <0.01 >0.05
NO ($\mu mol \cdot L^{-1}$)	12.97 ± 0.80	>0.05 >0.05 <0.01	13.37 ± 0.81	13.92 ± 0.79	18.28 ± 4.07	>0.05 <0.01 <0.01
3-Nitro ($nmol \cdot L^{-1}$)	35.91 ± 1.16	<0.01 >0.05 >0.05	39.67 ± 2.94	36.45 ± 1.99	34.48 ± 2.18	<0.001 <0.001 >0.05
TNFα ($pg \cdot mL^{-1}$)	0.73 ± 0.11	<0.001 <0.001 <0.001	5.67 ± 0.46	4.43 ± 0.64	3.47 ± 0.44	<0.001 <0.001 <0.011
IL-1β ($pg \cdot mL^{-1}$)	0.50 ± 0.08	>0.05 <0.05 <0.001	0.79 ± 0.30	0.89 ± 0.35	1.58 ± 0.37	<0.001 >0.05 <0.001
HSP27 ($pg \cdot mL^{-1}$)	549 ± 61	<0.001 <0.001 >0.05	1,143 ± 186	866 ± 113	550 ± 50	<0.01 <0.001 <0.001

*p values mean significant differences between nonathlete and wrestlers in 3 consecutive training months: January, April, and June.
†The first p value means difference between January and April, the second one between January and June, and the third one between April and June.

H_2O) or plain water. The dehydration level was assessed by Osmocheck calibrated in $mOsm \cdot kg^{-1}$ H_2O from 0 to 1,500 mOsmols. None of the athletes did not demonstrated dehydration, i.e., a urine osmolality was <600 mOsmols.

The training loads were demonstrated using program Training 1.2. prepared by Department of Sport Theory at University School of Physical Education Warsaw (Table 2). Moreover, during the study, athletes participated in tournaments of Bundesliga Wrestling (Germany) and prepared for the Olympics (London 2012). Each of athletes took part average in 2/3 fights per tournament. It is worth noted that the best of them performed 4/5 during each tournament. Lately it turned out that it was Olympic bronze medalist. Twelve healthy and untrained men at the age of 19.2 ± 0.4 years were considered a reference group (Table 1).

Subjects

All subjects were informed of the aim of the study and gave their written consent for participation in the project. The protocol of the study was approved by the ethics committee at Medical University Poznan, in accordance with the Helsinki Declaration.

Body Composition

Body mass and body composition (fat-free mass [FFM] and fat mass [FM]) were estimated using a bioelectrical impedance (BIA) using Tanita Body Composition Analyzer MC-980 (Japan) calibrated before each test session in accordance with the manufacturer's guidelines. Duplicate measures were taken with the participant in a standing position; the average value was used for the final analysis. The recurrence of measurement was 98%. The measurements were taken between 7.00 and 8.00 AM before blood sampling.

Blood Sampling

Blood samples were taken from the median cubital vein between 7.00 and 8.00 AM after 15 minutes of rest (and an overnight sleep). Within 20 minutes, they were centrifuged at 3,000*g* and +4° C for 10 minutes. Aliquots of serum were stored at −80° C.

Skeletal Muscle Damage

Serum total creatine kinase (CK) activity was used as a marker of sarcolemmal disruption and was evaluated using commercially available reagents and Dr. Lange analyzer (Hach Lange; Dusseldorf, Germany) at a temperature of 20–25° C. The CK activity has been measured immediately after serum collection for the consecutive days of the conditioning camp. At 24 hours after peak serum CK activity, the levels of inflammatory mediators and HSP27 were evaluated.

Inflammatory Mediators

Serum hydrogen peroxide (H_2O_2), nitric oxide (NO), and marker of NO bioavailability (3-nitrotyrosine, 3-nitro) concentrations were determined using the Oxis Research kits (USA). H_2O_2, NO, and nitro detection limits were

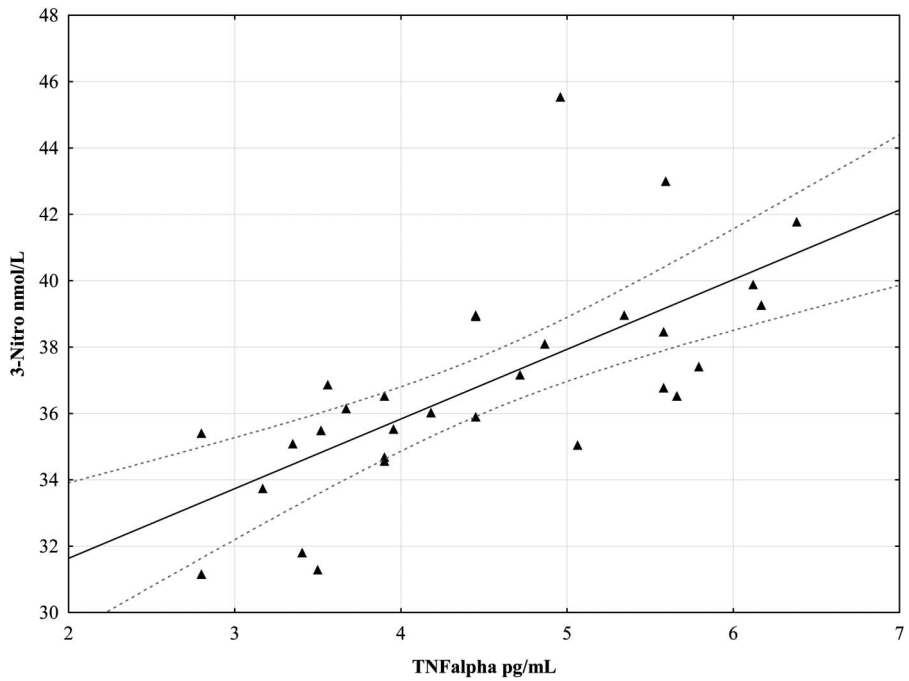

Figure 2. The significant relation between tumor necrosis factor α (TNFα) and nitrotyrosine (3-nitro) in wrestlers; $r_s = 0.694$, $p < 0.001$.

6.25 μmol·L^{-1}, 0.5 μmol·L^{-1}, and 2 nmol·L^{-1}, respectively. The intra-assay coefficient of variation for the H_2O_2, NO, and 3-nitro kits was <5%. Serum IL-1β and TNFα levels were determined by enzyme immunoassay methods using commercial kits from R&D Systems (USA). The detection limits for IL-1β and TNFα were 0.023 and 0.038 pg·mL^{-1}, respectively. The average intra-assay coefficient of variation was approximately 8.0%.

Heat Shock Protein

Serum HSP27 was measured with a Calbiochem kit (USA). The HSP27 detection limit for procedure was 0.2 ng·mL^{-1}, and intra-assay coefficient of variation was <5%.

Statistical Analyses

Statistical calculations were performed using the statistical software Statistica 10 (StatSoft Inc., Tulsa, OK, USA). All data were tested for distribution normality by the Shapiro-Wilk test. Comparisons of repeated measurements in wrestlers were assessed by the Wilcoxon signed-ranks test. The nonparametric test of Mann-Whitney was used for evaluation of significant differences between the resting values and the training-induced increases in wrestlers and nonathletes. Associations among measured parameters were analyzed using Spearman's rank correlation (r_s: Spearman rank correlation coefficient). Statistical significance was set at $p \leq 0.05$. Results are expressed as mean and *SD* (x ± *SD*).

Results

Body Composition

Wrestlers and nonathletes have shown a body weight, height, and body mass index on similar level. The significant changes concerned a percentage fat content (FM%), FM, and FFM were also on similar levels in wrestlers and nonathletes. Our results were similar to observation made by Demirkan et al. (4) in elite middle-weight wrestlers. Wrestlers have shown a body weight, height, and body mass index on similar level at preseason and in-season training period.

Skeletal Muscle Damage

As expected, CK activity was even 33-fold elevated in wrestlers compared with nonathletes (94 ± 24 IU·L^{-1}). In wrestlers, CK activity gradually increased on 5 consecutive days of conditioning camp. Creatine kinase reached the high activity >2,000 IU·L^{-1} at preseason and in-season period when the special training with elements of wrestling bout (>30% training load) predominated over directed training (approximately 10% training load). Creatine kinase activity decreased to approximately 500 IU·L^{-1} at the beginning of tournament season when directed training constituted >20% of the training load. Percentage increase in CK activity (%CK), calculated by comparison of peak with initial activity (the first day of conditioning camp), highly correlated with HSP27 (Figure 1).

Table 4. Relationships between heat shock protein 27 (HSP27), hydrogen peroxide (H_2O_2), nitric oxide (NO), 3-nitrotyrosine (3-nitro), interleukin-1β (IL-1β), and tumor necrosis factor α (TNFα) in wrestlers.

	H_2O_2 ($\mu mol \cdot L^{-1}$)	NO ($\mu mol \cdot L^{-1}$)	3-Nitro ($nmol \cdot L^{-1}$)	TNFα ($pg \cdot mL^{-1}$)	IL-1β ($pg \cdot mL^{-1}$)
HSP27 ($pg \cdot mL^{-1}$)	$r_s = 0.634$, $p < 0.001$	$r_s = -0.823$, $p < 0.001$	$r_s = 0.579$, $p < 0.001$	$r_s = 0.833$, $p < 0.001$	$r_s = -0.669$, $p < 0.001$

Inflammatory Mediators

The H_2O_2 concentration in wrestlers was significantly lower than nonathletes in contrast to NO, 3-nitro, TNFα, and IL-1β (Table 3). Tumor necrosis factor α concentration was even 7-fold higher in wrestlers (January). In wrestlers, the changes in H_2O_2, NO, and 3-nitro and also TNFα and IL-1β concentrations were dependent on training load and muscle damage, i.e., molecules increased when the special training dominated, and CK demonstrated activity >2,000 $IU \cdot L^{-1}$. However, the changes in inflammatory mediators did not proceed simultaneously, i.e., H_2O_2 and TNFα reached the highest level at preseason (January) whereas NO and IL-1β during the tournament season (June). This confirms an involvement of H_2O_2 and NO in proinflammatory cytokines release, and vice versa. The low NO concentration was accompanied by high 3-nitro and TNFα levels at preseason. The changes in 3-nitro highly correlated with TNFa levels during the all training periods (Figure 2).

Heat Shock Protein

The HSP27 concentration was significantly higher in wrestlers then nonathletes. The HSP27 reached the highest level during preparatory period, similarly to H_2O_2, 3-nitro, and TNFα (Table 3). The significant relationships between HSP27 and inflammatory mediators were observed (Table 4). Moreover, HSP27 inversely correlated with degree of muscle injury, i.e., athletes with high concentration of HSP27 demonstrated the smaller changes in CK activity than athletes with low concentration of HSP27 (Figure 1). This shows the protective effect of HSP27 on skeletal muscles.

Discussion

In wrestlers, skeletal muscles performing an intermittent physical exercise of variable intensity, such as sudden, explosive attacks, and counterattacks, are particularly exposed to damage because of a disruption of sarcomeres leading to an intensified CK efflux into the extracellular space. This increase in the serum CK activity strongly correlates with the degree of injury of the skeletal muscle cell structure. Therefore, CK is widely used in monitoring the training load, physical efficacy, and overtraining (10). CK reached the highest values during the tournament season (June), which was in accordance with the previous observation of Barbas et al. (1). They demonstrated that a 1-day wrestling tournament strongly induces the muscle damage, which may affect wrestlers' performance and inflammatory status especially during the later stages of the tournament. Overall, determining the levels of these 2 indicators might prove useful in the assessment of fatigue and appropriative response to physical workload.

The high CK activity was related to the H_2O_2 generation during the preparatory period and with NO generation during the tournament season. Earlier, we had demonstrated that eccentric work is an important factor enhancing the inflammatory response and production of H_2O_2 and NO (38). H_2O_2 and NO are secreted in the injured muscle by various cell types, including muscles cells, fibroblasts, neutrophils, and macrophages, depending on the time course of their recruitment to the region of damage. H_2O_2 and NO are generated at an earlier stage of exercise-induced inflammation and are involved in transcriptional control of many molecules expression, such as cytokines and HSPs (6,9,31). According to Scheele et al. (29), H_2O_2 and NO generation represents important mechanisms in muscle regeneration and training adaptation. However, an excessive H_2O_2 and NO generation results in the proapoptotic H_2O_2 and NO activities, decrease in the satellite cells number, and finally an impairment of muscle regeneration (32).

The study showed that TNFα inhibits the endothelial nitric oxide synthase (eNOS) and activates the inducible nitric oxide synthase (iNOS). Disturbance to the equilibrium between eNOS and iNOS activities results in the proapoptotic NO activity, changes in protein structure and dysfunction (40). Our study demonstrated the strong relation of TNFα with 3-nitro, which is a cytotoxic metabolite of NO. Therefore, detection and quantification of 3-nitro were used as an indicator for the participation of reactive nitrogen species in pathological processes (2,40). Despite the general lack of information in the literature on 3-nitro changes with physical training, our results agree with the study reporting higher levels of systemic markers of nitro-oxidative stress and inflammation observed in wrestlers (1).

Tumor necrosis factor α and IL-1β are expressed in muscle up to 5 days after damage. They are first involved in the degradation of the damaged tissue and then in the muscle regeneration. Both cytokines enhance H_2O_2 and NO

production whereby amplifying the signal transduction to cell nucleus (12,15). The exercise-induced TNFα and IL-1β release may have a negative effect on striated muscle. Smith (30) presented a study, where an excessive training (high-volume/intensity training) with an insufficient rest or recovery caused a chronic inflammation. This state was associated with the symptoms of overtraining. Similar situation was observed by Main et al. (18) in rowers after a 7-week training. In our study, TNFα and IL-1β were elevated in wrestlers compared with nonathletes and were dependent on the degree of muscle injury. Still, both cytokines did not reach the values observed by Main et al. (18). Tumor necrosis factor α was similar, whereas IL-1β level was even 92-fold lower in wrestlers than rowers participating in Main's study. Moreover, the assessment of immunological response after the high intensity training is needed to accurately determine the level of proinflammatory cytokines appropriate for athletes. We still have not known what level of proinflammatory cytokines is appropriate for athletes. It would facilitate differentiating what levels of cytokines indicate exercise overloading or an elevated risk of overtraining. Therefore, athletes who undergo intensive trainings should be monitored to establish the optimal level of inflammatory mediators (8).

The main finding of this study indicates that the release of HSP27 into circulation is proportional to the degree of muscle injury and inflammation. Thus, in our opinion, the quantification of HSP27 and inflammatory mediators in blood could be a marker of the regenerative ability of skeletal muscle in wrestlers. HSP27 possibly stabilizes and protects the myofibrillar structures during and after an unaccustomed eccentric exercise. In a high-force eccentric exercise, the cytoprotective role of HSP27 was indicated by its translocation from the cytosolic compartment to cytoskeletal/myofibrillar fraction (14,24,34). In our study, serum levels of HSP27 was approximately 2-fold elevated in wrestlers (during the preparatory period) compared with nonathletes. Regular physical training influences HSPs level as demonstrated by Fehrenbach et al. (5), who compared the expression of a variety of HSPs in the cytoplasm and on the surface of immune cells in marathon runners and untrained persons. The wrestling training significantly affected serum HSP27 synthesis and its release into circulation. Surprisingly, the initial CK activity (first day of the conditioning camp) influenced HSP27 level more than the peak CK activity. HSP27 was inversely correlated with the degree of muscle injury, i.e., athletes with high concentration of HSP27 (preparatory period) exhibited smaller changes in the CK activity than athletes with low concentration of HSP27 (tournament season). These data confirm the protective role of HSP27 in the repeated high-force eccentric exercises in wrestling training. Thompson et al. (34) suggested that HSP27 may be important to a long-term skeletal muscle adaptation such as hypertrophy.

The most interesting observation in our study was the relation of HSP27 with the mediators of an inflammatory response (Table 4). Extracellular HSP27 acts as a signaling molecule to activate a key transcription factor NF-κB that mediates important cell functions including survival, apoptosis, proliferation, inflammatory and stress responses. The activation of NF-κB by HSP27 is associated with the upregulated expression and secretion of proinflammatory and anti-inflammatory cytokines (23,28). The higher levels of TNFα contribute to the greater HSPs response in various cell types (11,23). In our study, serum HSP27 highly correlated with TNFα and 3-nitro concentrations, which belong to proapoptotic signals. The elevated level HSP27 reduces the cytotoxic effects of TNFα and 3-nitro and thus reveals its antiapoptotic properties. However, the elevation of TNFα contributes to the greater small HSPs response in type I and type II fibers during the early stages of muscle functional overload (11).

Practical Applications

The blood level of HSP27 is significantly higher in elite Greco-Roman wrestlers than nonathletes. The release of HSP27 into circulation is related to training load and muscle injury, and it can protect against cytotoxic effects of proinflammatory molecules. The quantification of HSP27 in blood could be useful diagnostic tool in biochemical assessment of the regenerative ability of skeletal muscle or provide evidence the early stages of muscle functional overload in wrestlers.

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