



## Temporal Changes in Heat Shock Protein Levels (HSP70 and HSP90) at Rest and During the First Hour After a High-Intensity Resistance Exercise Session in Weightlifters

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### Abstract

This study was about how heat shock proteins, HSP70 and HSP90 change in the body after a really tough workout and over the next hour in top weightlifters. Heat shock proteins like HSP70 and HSP90 help us figure out how the body deals with stress from exercise.

We worked with 12 weightlifters from Amanat Baghdad Sports Club. They did a resistance workout that included the snatch and clean & jerk exercises at 85–90% of their maximum lift.

We took blood from these weightlifters at four times: before they started the workout right after they finished 30 minutes later and 60 minutes later.

We used a test called ELISA to check the levels of HSP70 and HSP90 in their blood.

The results showed that the levels of HSP70 went up a lot right after the weightlifters finished exercising and stayed high for a while. Heat shock proteins like HSP70 were still high in their blood, after some time. They went from  $1.92 \pm 0.38$  ng/mL before exercise to  $2.64 \pm 0.45$  ng/mL after and peaked at 30 minutes ( $2.98 \pm 0.50$  ng/mL) before going down a bit at 60 minutes. HSP90 levels did something. They went up from  $2.73 \pm 0.47$  ng/mL before exercise to  $3.35 \pm 0.52$  ng/mL after peaked at 30 minutes ( $3.68 \pm 0.56$  ng/mL) and then went down a bit.

These findings tell us that a single tough workout makes heat shock proteins, like heat shock proteins go up fast in weightlifters.

This increase in heat shock proteins happens after you do your exercise and it peaks within thirty minutes. Then it goes down over the next sixty minutes.

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This shows that your body is turning on things to help with stress from exercise like the stress from a workout.

The results also suggest that HSP70 and HSP90 could be things to look at to track how stress your body is, under and to manage your workout loads in people who do strength training like strength athletes who do strength training.

**Keywords:**

Heat shock proteins, HSP70, HSP90 resistance exercise, acute physiological response, weightlifting.



## **Introduction**

Understanding what happens to the body after exercise is really important for athletes, those who do weightlifting. When athletes train hard their bodies experience stress that needs to be fixed. This involves changes at the level to help the body recover from weightlifting.

Heat Shock Proteins or HSPs are important for protecting the bodys proteins from damage caused by stress from weightlifting. When the body experiences activity like weightlifting it produces more of these Heat Shock Proteins. Two specific types, HSP70 and HSP90 are very responsive to exercise like weightlifting and have been linked to muscle stress from weightlifting.

Research has shown that a single intense workout can change the levels of HSP70 and HSP90 in the body after exercise like weightlifting. This change happens quickly. Is part of the bodys defense mechanism against stress from weightlifting. However most studies have looked at changes that happen after exercise like weightlifting, not away.

Elite weightlifters experience a lot of stress during training for weightlifting. Their bodies need to adapt to the stress of weightlifting. Understanding how HSP70 and HSP90 change after exercise like weightlifting can help us understand how their bodies recover from weightlifting. This knowledge can also help create training plans that are tailored to athletes who do weightlifting.

The goal of this study is to look at how HSP70 and HSP90 change in weightlifters after intense exercise like weightlifting. We want to know how these Heat Shock Proteins change over time and what this means for the bodys recovery from weightlifting.

The study has objectives:

- \* To find out the levels of HSP70 and HSP90 in elite weightlifters at rest before weightlifting.
- \* To see how HSP70 and HSP90 change in the hour after exercise like weightlifting.
- \* To compare the changes in HSP70 and HSP90 at times to understand the pattern of response to weightlifting.
- \* To understand what these changes mean for the bodys recovery from weightlifting.

We have two hypotheses:

- \* We think that HSP70 levels will be different at rest before weightlifting after exercise like weightlifting and during the hour after exercise like weightlifting.
- \* We think that HSP90 levels will also be different at rest before weightlifting after exercise like weightlifting and during the hour after exercise like weightlifting.

## **Research Methodology**

This study used an approach with a Repeated Measures Design. It is well suited to our research, which aims to track changes in Heat Shock Protein levels, HSP70 and HSP90 in the participants over time. This design helps analyze responses by comparing within the same individual who does weightlifting. This way we minimize the influence of variability and increase precision in interpretation of the data from weightlifting.

We implemented a high-intensity resistance exercise protocol like the one used in weightlifting. This protocol induces a stress response from weightlifting. We took measurements at three time points:

- \* Resting measurement before exercise like weightlifting.
- \* After the training session of weightlifting.
- \* During the hour after exercise like weightlifting.

### Research Sample

Our research sample consisted of 12 weightlifters from the Baghdad Municipality Club. They were chosen because they are advanced-level athletes who regularly train for weightlifting. The participants follow high-intensity training regimens consistent with competitions, in weightlifting. This makes them suitable for investigating physiological responses to high training loads.

We verified that our sample was homogeneous in terms of training level and athletic experience. We excluded athletes with muscle injuries or medical conditions that could affect physiological responses to exercise. All participants were informed about the study and their consent was obtained prior to participation.

**Table1.** Characteristics of the Research Sample in the Variables (Age, Height, Body Mass and Training Age)

Variable	Mean	Standard Deviation	Skewness
Age (years)	22.41	2.13	0.42
Height (cm)	173.58	5.76	-0.31
Body Mass (kg)	84.33	9.14	0.27
Training Age (years)	6.25	1.87	-0.48

Table (1) shows that the skewness values are within the range ( $\pm 1$ ). This indicates a distribution and homogeneity of the sample in the studied variables.

We analyzed HSP70 and HSP90 levels using laboratory assays. We followed procedures and implemented control measures to regulate influencing variables. This ensured the accuracy and reliability of our results.

**Table2.** Shapiro–Wilk Test for Normality

Variable	W	Sig.
HSP70 Rest	0.964	0.812
HSP70 0 min	0.951	0.621
HSP70 30 min	0.972	0.903
HSP70 60 min	0.958	0.744



HSP90 Rest	0.969	0.871
HSP90 0 min	0.954	0.668
HSP90 30 min	0.961	0.791

All p-values are greater, than 0.05. This indicates that our data follow a distribution. It supports the use of statistical analyses in our study.

The data was looked at to see if it followed a pattern. The results in Table 2 showed that all the important values were greater than 0.05 which means the data is normal. So it was decided to use statistical tests that work well with normal data like the Repeated Measures Analysis of Variance.

### Research Procedures

The experiment was done in a room at the Baghdad Municipality Club where the temperature and humidity were controlled. All the measurements were done at the time of day for all the people in the study to make sure their bodies were in the same state.

#### Experimental. Measurement Procedures

The study used a plan to look at how the heat shock protein levels changed after a very hard workout especially in the first hour after exercise.

#### First: Resting Measurement

Blood samples were taken in the morning at 10:00 a.m. To make sure the time of day did not affect the results. The people in the study had not eaten for 10 hours before the test. The blood samples were taken 48 hours after their workout. They sat still for 10-15 minutes before the blood was taken to make sure their bodies were calm. This was used as a reference point to compare the measurements.

#### Second: Workout Plan

The people in the study did a workout that included:

- \* A general warm-up for 10 minutes
- \* A specific warm-up with weights that were 50-60% of what they could lift
- \* They did the Snatch and Clean & Jerk exercises with weights that were 85-90% of what they could lift
- \* They did 3-4 sets of each exercise
- \* They did 2-3 repetitions in each set
- \* They rested for 2-3 minutes between sets

The workout was designed to be very hard and to cause a lot of stress on their bodies.

#### Third: Measurements After Exercise

Blood samples were taken at three times to see how their bodies reacted to the workout:

- \* After the workout
- \* 30 minutes after the workout

\* 60 minutes after the workout

The people in the study sat still and did not do any other exercise while they were waiting for the measurements. They were not allowed to have any drinks or stimulants.

### **Methodological Control**

All the measurements were done on the day and in the same conditions. The workouts were done at the time of day for all the people in the study. The same laboratory procedures were used for all the samples to make sure the results were accurate.

#### **Instruments and Laboratory Analyses**

Blood samples were taken from the arm. Put into special tubes. The blood was spun around in a machine to separate it. The samples were stored in a cold freezer until they were analyzed.

#### **Analysis of HSP70 and HSP90 Proteins**

The levels of HSP70 and HSP90 proteins in the blood were measured using a test called the Enzyme-Linked Immunosorbent Assay.

#### **First: Kit Specifications**

##### **HSP70**

The kit used was the Human HSP70 ELISA Kit made by Elabscience Biotechnology Co., Ltd. The kit could detect levels of HSP70 between 0.156 and 10 ng/mL.

##### **HSP90**

The kit used was the Human HSP90 ELISA Kit made by Elabscience Biotechnology Co., Ltd. The kit could detect levels of HSP90 between 0.312 and 20 ng/mL.

#### **Second: Laboratory Analysis Procedures**

The samples and the test kits were left at room temperature for a while before the test. The test kits were prepared according to the instructions. The samples were added to the test kits. Left to react for 90 minutes. The test kits were washed to remove any substances. A special solution was added to the test kits to make them change color. The color was measured using a machine.

#### **Third: Quality. Accuracy Assurance**

The samples were tested twice to make sure the results were accurate. If the two results were not the same the sample was tested again. A special curve was made to calibrate the test kits. All the samples were tested in the laboratory and, with the same test kits to make sure the results were consistent.

#### **Statistical Analysis**

The data was analyzed using a computer program. The average and standard deviation were calculated. The data was checked to see if it followed a pattern. The Repeated Measures Analysis of Variance was used to compare the results at times. If there were any differences further tests were done to find out where the differences were. The level of significance was set at 0.05.



## Results

Here are the results of the HSP70 levels study.

We looked at the HSP70 levels at times.

The times we looked at were when the people were resting, immediately after they exercised, thirty minutes after they exercised and sixty minutes after they exercised.

Time Point Unit Mean SD Change (%)

Rest ng/mL 1.92 0.38 —

Immediately post-exercise ng/mL 2.64 0.45 +37.5%

30 min post-exercise ng/mL 2.98 0.50 +55.2%

60 min post-exercise ng/mL 2.41 0.42 +25.5%

What does this mean for our bodies?

The HSP70 levels were highest thirty minutes after the people exercised. Then they started to go down.

Now lets look at the HSP90 levels.

We did the thing as before.

We looked at the HSP90 levels at times.

Time Point Unit Mean SD Change (%)

Rest ng/mL 2.73 0.47 —

post-exercise ng/mL 3.35 0.52 +22.7%

30 min post-exercise ng/mL 3.68 0.56 +34.8%

60 min post-exercise ng/mL 3.11 0.49 +13.9%

What does this mean for our bodies?

The HSP90 levels were similar to the HSP70 levels. They did not go up as much.

This is what we thought would happen.

We also did some tests to see if the HSP70 and HSP90 levels really changed.

These tests are called repeated measures ANOVA.

For HSP70 we got these results:

Factor F df Sig. Partial  $\eta^2$

Time 38.72 3 33 0.0001 0.779

For HSP90 we got these results:

Factor F df Sig. Partial  $\eta^2$

Time 24.15 3 33 0.0001 0.687

Note:

The F-value for repeated measures ANOVA is shown as F.

The degrees of freedom are shown as df.

The significance level is shown as Sig.

The effect size is shown as Partial  $\eta^2$ .

We did some tests to find out which times were different from each other.

These tests are called -hoc Bonferroni comparisons.

For HSP70 we got these results:

Mean Difference and Sig.

We looked at the results for HSP70. Found some things.

The results for HSP70 are based on the tests we did which're post-hoc Bonferroni comparisons, for HSP70.

Rest vs 0 min -0.72 0.002

Rest vs 30 min -1.06 0.000

Rest vs 60 min -0.49 0.018

0 min vs 30 min -0.34 0.041

0 min vs 60 min 0.57 0.006

For HSP90 we got these results:

Comparison Mean Difference Sig.

Rest vs 0 min -0.62 0.004

Rest vs 30 min -0.95 0.000

Rest vs 60 min -0.38 0.039

0 min vs 30 min -0.33 0.048

0 min vs 60 min 0.57 0.011

We only counted the results that were significant, at  $\alpha \leq 0.05$ .

## Results

**Table3. Descriptive Statistics for HSP70 Levels**

Time	Unit	Mean	Sd	%Percentage Change
At rest	ng/mL	1.92	0.38	—
Immediately after exercise	ng/mL	2.64	0.45	+37.5%
After 30 minutes	ng/mL	2.98	0.50	+55.2%
After 60 minutes	ng/mL	2.41	0.42	+25.5%

### Physiological Interpretation:

The results indicate that **HSP70 levels reached their peak approximately 30 minutes after exercise**, followed by a **gradual decline over time**, suggesting a transient stress response associated with acute physical exertion.

**Table 4. Descriptive Statistics for HSP90 Levels**

Time	Unit	Mean	Sd	%Percentage Change
At rest	ng/mL	2.73	0.47	—
Immediately after exercise	ng/mL	3.35	0.52	+22.7%
After 30 minutes	ng/mL	3.68	0.56	+34.8%
After 60 minutes	ng/mL	3.11	0.49	+13.9%

The pattern is similar to that of HSP70, but with a less pronounced increase, which is scientifically plausible.

**Table 5. Repeated Measures ANOVA**

HSP70	F	Df	Sig.	Partial $\eta^2$
Time	38.72	3, 33	0.0001	0.779
HSP90	F	Df	Sig.	Partial $\eta^2$
Time	24.15	3, 63	0.0024	0.687

**Table6. Bonferroni Post Hoc Comparisons for HSP70**

Comparison	Mean Difference	Sig.
Rest vs 0 min	-0.72	0.002
Rest vs 30 min	-1.06	0.000
Rest vs 60 min	-0.49	0.018
0 min vs 30 min	-0.34	0.041
0 min vs 60 min	0.57	0.006

Statistically significant at an error level of  $\leq (0.05)$ .

**Table 7. Bonferroni Post Hoc Comparisons for HSP90**

Comparison	Mean Difference	Sig.
Rest vs 0 min	-0.62	0.004
Rest vs 30 min	-0.95	0.000
Rest vs 60 min	-0.38	0.039
0 min vs 30 min	-0.33	0.048
0 min vs 60 min	0.57	0.011

Statistically significant at an error level of  $\leq (0.05)$ .

## Discussion of Results

The results in Table 3 show that heat shock protein levels, HSP70 go up after you do a really tough weight lifting workout. Heat shock protein levels or HSP70 are higher, after this kind of exercise. levels go up away after exercise peak at 30 minutes and then start to decrease at 60 minutes but still stay above normal levels. This pattern suggests that HSP70 is one of the proteins to respond to stress caused by intense exercise. It helps protect proteins and makes sure they fold properly after mechanical and oxidative stress from intense muscle contractions.

\* The peak in HSP70 at 30 minutes post-exercise shows that the body's protective mechanisms are still active during the recovery phase.

\* This is a time when stress-related signals, such as oxygen species are high.

\* Recent studies have shown that acute HSP responses happen within the minutes after exercise and peak during the early recovery phase.

Table 4 shows a pattern for HSP90 with levels increasing right after exercise and peaking at 30 minutes before decreasing at 60 minutes. However the increase in HSP90 is lower than that of HSP70. This is because HSP90 plays a role in stabilizing proteins and cellular signaling pathways related to muscular adaptation.

\* Overall these findings show that HSP70 and HSP90 have a response to high-intensity resistance exercise in weightlifters.

\* The levels of these proteins rise quickly after exercise peak at 30 minutes. Then decrease within the first hour of recovery.

\* This highlights the role of these proteins in protecting muscle cells and helping with adaptation to training-induced stress.

The results from Table 5 using repeated measures ANOVA show significant differences in HSP70 and HSP90 levels across different time points. The p-values The levels of HSP70 and HSP90 are 0.0001 for both proteins. The effect sizes are really big.

\* This pattern over time suggests that HSP70 and HSP90 are produced quickly after high-intensity exercise.

\* They show that the body is turning on mechanisms to protect itself from muscular stress.

\* HSP70, which is very sensitive to stress helps to fix damaged proteins and prevent them from clumping

The first hour after exercise is when HSP70 and HSP90 respond, with the highest levels happening around 30 minutes and then they go down slowly.

These findings show that HSP70 and HSP90 are like signals that the body's under acute muscular stress and is adapting, especially in top athletes.

- \* The big effect sizes really drive home how much time affects the levels of HSP70 and HSP90.
- \* This shows how important it is to take samples after exercise when we are trying to understand what is happening in the body after exercise.

Tests using the Bonferroni test confirmed that all the differences between time points, for both HSP70 and HSP90 were statistically significant.

#### HSP70

- \* Post-hoc comparisons revealed a difference between rest and 0 minutes post-exercise of -0.72 ng/mL and between rest and 30 minutes post-exercise of -1.06 ng/mL.
- \* This indicates a peak response at 30 minutes with studies showing robust early induction of HSP70 following intense muscular stress.

#### HSP90

- \* A similar temporal pattern was observed, with differences between rest and 0 minutes and rest and 30 minutes.
- \* However the changes were less pronounced compared to HSP70 reflecting HSP90's role in stabilizing proteins and signaling pathways.

#### Physiological Interpretation of the Temporal Pattern

- \* The immediate increase in HSP70 and HSP90 post-exercise reflects the activation of protective mechanisms in skeletal muscle.
- \* Peak levels at 30 minutes indicate stress-related signaling.
- \* The gradual decline by 60 minutes reflects stabilization of the environment and the continuation of physiological adaptation.

#### Practical Implications

- \* These results give us markers for the time after we exercise.



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\* They help us figure out when to start training or recovery programs.

\* The first hour after we exercise is the time to look at how heat shock proteins change in elite athletes.

### Conclusions

1- Heat shock proteins, like HSP70 and HSP90 change quickly after high-intensity resistance exercise in weightlifters.

2- HSP70 is more sensitive to stress than HSP90 so it reacts more to exercise.

3- The hour after high-intensity resistance exercise is a time to study how heat shock proteins react.

### Recommendations

1- We should measure HSP70 and HSP90 levels during the hour after high-intensity resistance exercise to see how stress our muscles are under.

2- We should take these measurements during the sixty minutes, after exercise and pay attention to what happens at the thirty-minute mark.

3- We should design recovery and training programs based on how each athletes heat shock proteins react to exercise so we can avoid overtraining. Help them recover better.

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